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Isolation and Identification of Fungal Species Associated with Fruits Spoilage in Bwari Market Abuja, Nigeria

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Authors' contributions

This work was carried out in collaboration between all authors. All authors read and approved the final manuscript.

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Original Research Article

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ABSTRACT

Spoilage of fruits imparts great economic loss and a potential health hazard to the general populace. A research was conducted to identify the different types of fungal flora responsible for the spoilage of fruits in Bwari market, Abuja, Nigeria. Seventy fruits that showed decayed symptomatology and thirty healthy ones were procured from Bwari market. Sections of the rotten and healthy fruits were obtained using a sterile blade, inoculated onto Potato Dextrose Agar and incubated at 27°C for five days. All other mycological techniques were carried out using standard methods. The result showed the presence of *Aspergillus niger*, *Rhizopus nigricans*, *Mucor mucedo*, and *Fusarium oxysporum*. However, *Aspergillus niger*. (58.06%) was found to be the most dominant spoilage causing organism on fruits vended in Bwari market while *Fusarium oxysporum* (9.68%) was the least abundant fungal flora. However, the pathogenicity of *Rhizopus* sp. was found to be higher than that of all the remaining fungal species. This can be attributed to the predisposal and nature of fruits handling and storage in the market. Thus, because all the fungal species identified were known to produce toxins that could impart severe food poisoning, attention of the

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authorities concerned and the health care providers is needed towards orienting the vendors on the hygienic methods of storing fruits to prolong their shelf-life and the general populace on the dangers associated with the consumption of fruits from Bwari market without thorough washing with saline water more especially those fruits that showed spoilage symptomatology.

Keywords: Aspergillus; Fusarium; Mucor; Rhizopus.

1. INTRODUCTION

Fruits play a vital role in human nutrition by supplying necessary growth factors such as vitamins and essential minerals in daily diet which help to live a healthy life [1]. Besides all these, there is an increasing concerned upon the use of fruits in diet therapy for certain clinical diseases. However, despite all these tremendous benefits of fruits to human well-being, pathogenic attacks threatened the shelf-life of fruits and degrade their economic value. Fruits are mainly exposed to microbial contamination through contact with soil, dust and water and by poor handling or during postharvest processing. This makes them to harbor a wide range of microorganisms (Ofor et al., 2009; Eni et al., 2010) especially fungi.

The principle of spread of fungal infection in fruits supports that a single infected fruit can infect other healthy fruits during storage and on transit (Jay, 2003). The fungi can be introduced into the crop on the seed itself, during crop growth in the field, during harvesting and postharvest handling, or during storage and distribution [2]. Postharvest losses and decay of fruits can be traced mostly to infections that occur during harvesting and subsequent handling and storage activities. Preharvest infections are mainly caused by fungal pathogens such as Phytophthora sp., and Colletotrichum gloeosporioides (Browning et al., 1995: El Ghaouth et al., 2002). Isolation and identification of the decay-causing fungi are desirable in order to strategize the control measures with a view to reducing losses due to spoilage or infections (Singleton et al., 1992). Thus, this study aimed at isolating and identifying fungi associated with fruits spoilage in Bwari market, Abuja, Nigeria.

2. MATERIALS AND METHODS

2.1 Sample Collection

The fruit samples used in this study were obtained from the fruits retailers randomly from Bwari market, Abuja, Nigeria. A total of one hundred samples comprising of seventy fruits that showed decay-symptomatology and thirty healthy fruits were procured. The fruits were placed in sterile plastic bags and labeled accordingly. The samples were kept in the refrigerator at 4°C till when use for mycological analysis.

2.2 Isolation of Fungal Species from Spoilt Fruits

The isolation procedure for spoilage inducing fungal flora was done according to the methodology described by Bukar et al. [3]. The infected fruits were surface sterilized with 0.1% mercury chloride for 2 minutes using cotton wool and then rinsed thrice with distilled water. A sterile razor blade was used to cut small sections of 3 mm diameter from the tissues of spoilt portions of the fruits and placed on solidified Potato Dextrose Agar (PDA) containing 30 mg/l of Chloramphenicol antibiotic to prevent bacterial growth. The inoculated plates were incubated at 30°C for seven days. The fungal isolates were sub-cultured on PDA slants to obtain pure cultures.

2.3 Identification of Fungal Isolates

Fungal isolates obtained from the slants were identified based on their Gross Morphology such as colony growth pattern, conidial morphology and pigmentation by slide culture techniques according to the protocol described by Oyeleke and Manga [4]. A small portion of the aerial mycelia from the representative culture was picked using a sterile inoculating needle and inoculated on a slide containing a fraction of a prepared solidified Potato Dextrose agar and incubated for 48 hours, after which it was viewed under the light microscope first with low resolution objective of 10x and then with high resolution objective of 40x to detect spore, hyphae and other special structures according to the methods described by Barnett (1991). The Morphological characteristics and appearance of the fungal species isolated from the fruits used in this study were confirmed and authenticated using Mycological Atlas of Robert and Ellen (1988).

2.4 Pathogenecity Test

Healthy fruits were surface sterilized with 0.1% mercury chloride using cotton wool. Cylindrical tissues were cut out from the fruits using a sterilized 3mm sized cork borer. Agar discs containing one week old fungal culture were aseptically placed in these holes, then covered and sealed off by means of petroleum jelly. The procedure was repeated separately across each of the fungal isolates. The inoculated samples and the control were placed in sterile polythene bags and incubated at 30°C for 14 days. The point of inoculation of each type of fungus was examined and recorded accordingly. The diameter of the rotten portion of the fruits was measured. The fungi were later re-isolated from the inoculated fruits and compared with the initial isolates.

3. RESULTS

The result for the prevalence of fungal isolates on the fruits sold in Bwari market is presented in Table 1. The result showed that, four different fungal species were found associated with the fruits sold in Bwari market. The result indicated that *Aspergillus niger* is present in all the eight different fruits from Bwari market and is the most predominant species (58.06%) abundance. However, *Fusarium oxysporum* is present only in water melon, tomato and pepper and is the least predominant fungal species (9.68%).

However, the result for the prevalence of the fungal flora on the eight fruits vended in Bwari market is presented in Table 2. The result showed the presence of four different fungal species in the fruits vended in Bwari market. The fungal species are: *Aspergillus niger*, *Rhizopus nigricans*, *Mucor mucedo*, and *Fusarium oxysporum*. The result indicated that, *Aspergillus niger* has the highest occurrence of 58.06%; while *Fusarium oxysporum* has the least abundance of 9.68%.

Furthermore, the result of the pathenogenicity test (Table 3) shows that, *Rhizopus nigricans* induces the highest rotten diameter on the fruits (36 mm in diameter). However, *Fusarium oxysporum* was found to have the lowest rotten diameter of 22 mm in diameter.

4. DISCUSSION

The presence of four different fungal species (Aspergillus niger, Rhizopus nigricans, Mucor mucedo, and Fusarium oxysporum) identified in

the spoilt fruits obtained from Bwari market have provided an impetus upon the dangers associated with the consumption of such fruits without proper washing or treatment. More so, these fungal pathogens impart severe losses of marketable quality and posed threat to public health resulting in major economic problem in Nigeria. The species isolated in this study were previously identified in different fruits obtained in Nigeria by Amadi and Oso [5], Oyetunji et al. (2012), Amadi et al. [6], Mbajiuka et al. [7] and Ezikanyi [8]. These fruits might likely be contaminated via infestation in the field prior to harvest, handling during harvesting and methods of packaging and distribution of produce to the market as stressed by Amadi et al. [6]. The high prevalence of Aspergillus niger in the spoilt fruits vended in Bwari market is in conformity with the findings of Mailafia et al. [9] who reported high prevalence of Aspergillus niger among fruits vended in Gwagwalada market. The finding also is in consistent with that of Samuel et al. [10] who identified Aspergillus niger as the most prevalent fungal species causing spoilage of carrot (Daucus carota). This indicated the risks associated with the consumption of these fruits as Aspergillus niger was highly toxigenic known to produce several toxic metabolites such as malformins, nathopyrones as reported by Frisvad and Samson [11] and ochratoxin which are popular for their harmful effects to human and animal health as reported by Peraica et al. [12] and Petzinger and Weldenbach [13]. However, this finding contradicts the work of Amadi et al. [6] who reported Fusarium oxysporum as the most prevalent fungal species in the spoilage of guava fruits.

Proof of pathogenicity is the single most reliable criterion used in implicating associated microorganisms in the causal processes of disease development as stressed by Amadi et al. [6]. All the species proved to be pathogenic on the fruits and this is consistent with the work of Udoh et al. [14] who reported similar finding among the fungal species isolated from different fruits in Enugu, Nigeria. The study also shows that all the species isolated and identified are responsible for the fruits spoilage in Bwari market. This is in line with the work of Peter et al. [15], Amadi et al. [16] and Renu and Lal [17] that Fusarium oxysporum and Aspergillus niger are notorious causal agents of rot in many fruits and vegetables including watermelon, carrot and guava. Similarly, Mathew et al. [18] had reported Aspergillus niger and Rhizopus stolonifer in postharvest diseases of guava.

Samples	Aspergillus niger	Rhizopus nigricans	Mucor mucedo	Fusarium oxysporum
0	7	1	-	-
В	7	-	1	-
PA	4	2	2	-
PI	5	1	2	-
WM	2	2	1	2
PU	4	1	3	-
ТО	2	1	2	2
PE	5	1	-	2
Total	36 (58.06%)	9 (14.52%)	11 (17.74%)	6 (9.68%)

Table 1. Occurrence of fungal species in some fruits sold at Bwari Market, Abuja

Key: O=Orange, B=Banana, PA=Pawpaw, PI=Pinapple, WM=Water melon, PU=Pumpkin, TO=Tomato, PE=Pepper

Table 2. Percentage prevalence of fungal species in fruits sold in Bwari Market, Abuja

S/N	Fungal isolate	Frequency	Percentage (%)
1	Aspergillus niger	36	58.06
2	Rhizopus nigricans	9	14.52
3	Mucor mucedo	11	17.74
4	Fusarium oxysporum	6	9.68
	Total	62	100

Table 3. Pathogenicity of Fungal Species on the Fruits Obtained from Bwari Market, Abuja

S/N	Fungal species	Diameter of rot (mm)
1	Aspergillus niger	27
2	Rhizopus nigricans	36
3	Mucor mucedo	30
4	Fusarium oxysporum	22

Fruit spoilage can be prevented using physical methods as reported by Boyer [19] and chemical method as reported by Msagati [20], but no efficient strategy has been proposed so far to reduce the microbial growth ensuring public health safety.

5. CONCLUSION

It was concluded that *Aspergillus niger.*, *Rhizopus nigricans*, *Mucor mucedo* and *Fusarium oxysporum* were responsible for the spoilage of fruits in Bwari market which subsequently inferred a great loss to the retailers and imposed clinical threat to the consumers via severe food poisoning.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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