



## Shelf Life Assessment of Fungal Contaminants Associated with Selected Bakery Products (Bread, Meat Pie and Cake) in Dutse Metropolis

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### ABSTRACT

Bakery products such as bread, meat pie, cake etc are staple foods consumed by both poor and rich are subject to fungal contamination. This research was aimed at isolating and identifying the fungal species associated with selected bakery products (bread, meat pie and cake) sold within Dutse, Jigawa State. A total of 27 samples, 9 from each baked product were used in the study, with Potato Dextrose agar and Sabouraud Dextrose agar as culture media while pour plate technique was employed in the fungal isolation. The inoculated samples were incubated at room temperature for 3-5days and were isolated and identified on the basis of their colonial and morphological features as the species of *Penicillium spp*, *Mucor spp*, *Rhizopus spp*, *Aspergillus niger* and *Aspergillus flavus* occurred in all samples at different frequency of occurrence studied with *Aspergillus flavus* having the highest occurrence (31.3%) from cake while *Aspergillus niger* having the highest occurrence (30.0%) from meat pie and *Mucor spp* having the highest occurrence (36.4%) from bread.

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### INTRODUCTION

Valuable sources of nutrients in our diets is providing us with most of our food calories and approximately half of our protein requirements (Baur, 2001). Cereals have been a basic food since prehistoric times and were consumed long before bread making was developed, varieties of bread and other bakery products have increased in sales volume within the past decades (Carla and Mariaines, 2009). Bakery industry in India is the largest of the food industries with an annual turnover of about 3000 crores (1 crore = ten million rupees). Breads and biscuits form the major baked foods accounting for over 80 percent of the total bakery products produced in the country and their quantities produced are more or less the same (Hickey, 2000). Bakery products once considered sick man's diet have now become essential food items of the vast majority of population. Bakery products are an important part of a balanced diet

and a wide variety of such products can be found on supermarket shelves, the contributing factors were urbanization, resulting in increased demand for ready to eat products at reasonable costs. According to Baur (2001) many industrially produced baked goods emerge from the baking process with a surface that is essentially sterile but post bake handling can quickly lead to fungal, microbial surface contamination as a result of exposure to airborne contaminants as well as equipment contact. Importance of bakery products has expanded especially the use of natural and whole grains and other natural ingredients (Oluwajoba *et al.*, 2012).

In Nigeria, some bakery products has become the second most widely consumed non indigenous food product after rice and their consumption cut across socio-cultural and religious barriers and is a food choice for both the rich and poor. Microorganisms especially fungi are associated in variety of ways with all food we



eat. They may influence the quality, availability and quantity of our food. Ehavald (2009), reported that despite the awareness created and efforts to ensure that quality is available for citizens, there are several routes through which bakery products get contaminated such as in the process chain, especially during packaging. Filamentous fungi that are responsible for the spoilage of bakery products include *Rhizopus* spp, *Aspergillus* spp, *Mucor* spp, *Penicillium* spp etc as they have the ability to produce spores which are transmitted via air and contaminate surfaces of materials or equipment (Emeje *et al.*, 2010).

In the last few years, the bakery products and flour confectionary sector has witnessed particularly intense technological progress which has brought clear and tangible changes, not only in terms of commercial and qualitatively characteristics of the products, but also in terms of process innovation (Anon, 2000).

## MATERIALS AND METHODS

### Study Area

The study area was Dutse metropolis located in Northern Nigeria. It is a capital city of Jigawa State and a home to Federal University Dutse among others at latitude 11°42'04"N 9°20'31"E with an estimated population of 153,000 (2009). The State was created in 1991 during the military regime of General Ibrahim Badamasi Babangida. Mostly igneous in nature, the rocky town got its name from these naturally endowed resources, Dutse (Hausa term for rock). The populations of Dutse are predominantly Hausa and Fulani which are mostly farmers, other occupations typical to rural area are also available among the populace. The site for sample collection were Hasina confectionary and catering service, Hannee bakery, Stone castle bakery, Kowanaka bakery, Zuma and Lautai bakery which are all located within Dutse metropolis.

### Sample Size

A total of twenty seven samples of meat pie, bread and cake were randomly collected from different bakeries of within the study area, where nine samples of each baked product were collected. Sample size was

determined by an epidemiological formula put forward by Lwanga and Lemeshow (1991) using local base study done in Enugu State by (Nimala *et al.*, 2016).

### Sample Collection

The samples were aseptically collected using clean leather bags and disposable hand gloves and taken immediately to the Biology laboratory and stored in the drawer for five days for spoilage observation.

### Sample preparation

#### Serial Dilution

The contaminated portions of the samples were picked using a sterilized forceps and transferred aseptically into 9ml of normal saline in a sterilized test tube which was mixed thoroughly and 1ml from the stock solution was picked using a syringe and transferred into another test tube also containing 9ml of normal saline. This procedure was repeated until a 10<sup>-9</sup> dilution was obtained.

### Media Preparation and Inoculation

#### Media Preparation

Potato Dextrose Agar (PDA) and Sabouraud Dextrose Agar (SDA) were weighed, prepared and sterilized according to the manufacturer's instruction. 19.5g of PDA and 10.0g of SDA were dissolved in 250ml of distilled water.

#### Inoculation

0.1ml from the 10<sup>-4</sup> and 10<sup>-7</sup> were transferred using a syringe onto a surface of a separate labelled petri dish each containing Potato Dextrose agar using spread plate technique as described by Akintobi *et al.*, (2011).

#### Incubation of the inoculated Plates

The inoculated plates were incubated at room temperature for 5-7 days under light to enhance fungal growth and sporulation using (Okigbo and Osuinde, 2003) method.

#### Purification of Fungal Isolates

After the appearance of a mixed growth, each colony was sub-cultured in a fresh Sabouraud Dextrose agar in order to obtain a



pure culture using streak method as described by Cheesbrough (2006).

### Identification and Characterization of fungal Isolates

Identification and characterization of the various isolates were based on macroscopic and microscopic examination (Akintobi *et al.*, 2011).

### Macroscopic Examination

This was done on the basis of the nature of appearance of colony and morphological characteristics of the isolated fungi using Atlas of food Microbiology by Hart and Shears (2004).

### Microscopic Examination

Slides of the colony observed from different isolates were prepared as follows: A few drop of lactophenol cotton blue solution was placed at the centre of clean glass slides. A small portion of the unidentified fungal isolates were picked with sterile wire loop. The portions were placed in the lactophenol cotton blue droplet on the slides and emulsified out with a sterile wire loop. Cover slips were placed at the centre of the slides for viewing and examining the structure of the mycelia and fruiting bodies were identified with the help of Atlas of food Microbiology by Hart and Shears (2004).

### Determination of Percentage Frequency of Occurrence

The frequency of occurrence of different types of isolated fungal contaminants responsible for the spoilage of selected bakery products were determined. The percentage of occurrence was calculated using the formula (Morsy *et al.*, 2009).

$$\% \text{ Frequency} = \frac{\text{Number of isolated fungi} \times 100}{\text{Total number of isolates}}$$

### RESULTS AND DISCUSSIONS

The fungi responsible for the spoilage of cake, meat pie and bread were identified based on colony appearance, morphology and cellular characteristics as shown table 4.1. The percentage frequency of occurrence of fungi responsible for the spoilage of cake were shown in table 2 where *A. niger* (18.8%), *A. flavus* (31.3%), *Rhizopus spp* (6.3%), *Mucorspp* (25.0%) and *Penicillium spp* (18.6%). Table 3 shows the percentage frequency of occurrence of fungi responsible for the spoilage of meat pie where *A. niger* (30.0%), *A. flavus* (25.0%), *Rhizopus spp* (10.0%), *Mucor spp* (15.0%) and *Penicillium spp* (20.0%). Finally, *A. niger* (13.6%), *A. flavus* (9.1%), *Rhizopusspp* (22.7%) *Mucorspp* (36.4%) and *Pencillium spp* (18.2%) were found to be responsible for the spoilage of bread as shown in table 4.

However, all the control plates showed no any fungal growth.

**Table 4.1:** Colonial and morphological characteristics of fungal contaminants responsible for spoilage of cake, meat pie and bread.

Isolated fungi	Colony appearance on agar plate	Morphological and cellular characteristics
<i>Mucor spp</i>	Cotton like white growth spotted with black color	Sporangia contain spores, do not have rhizoids
<i>Rhizopus spp</i>	Cotton like white growth spotted with black color	Sporangia contain spores, have rhizoids
<i>Penicillium spp</i>	Green or green-greyish colonies	Brush like conidiophore carries conidia
<i>Aspergillus niger</i>	Pin-like black growth	Non-branched conidiophore with bulb end carries conidia like sun ray
<i>Aspergillus flavus</i>	Pin like yellow growth	Non-branched conidiophore with bulb end, carries conidia



**Table 4.2:** Determination of Frequency of occurrence

Identified fungal isolates	Number of isolates	Frequency of occurrence (%)
<i>Mucorspp</i>	4	25.0
<i>Rhizopusspp</i>	1	6.3
<i>Penicillium spp</i>	3	18.6
<i>Aspergillus niger</i>	3	18.8
<i>Aspergillus flavus</i>	5	31.3
<b>TOTAL</b>	<b>16</b>	<b>100</b>

**Table 4.3:** The determination of the frequency of occurrence of identified fungal contaminants responsible for the spoilage of meat pie

Identified fungal isolates	Number of isolates	Frequency of occurrence (%)
<i>Mucor spp</i>	3	15.0
<i>Rhizopus spp</i>	2	10.0
<i>Penicillium spp</i>	4	20.0
<i>Aspergillus niger</i>	6	30.0
<i>Aspergillus flavus</i>	5	25.0
<b>TOTAL</b>	<b>20</b>	<b>100</b>

**Table 4.4:** The determination of frequency of occurrence of identified fungal contaminants responsible for the spoilage of bread

Identified fungal isolates	Number of isolates	Frequency of occurrence (%)
<i>Mucor spp</i>	8	36.4
<i>Rhizopus spp</i>	5	22.7
<i>Penicillium spp</i>	4	18.2
<i>Aspergillus niger</i>	3	13.6
<i>Aspergillus flavus</i>	2	9.1
<b>TOTAL</b>	<b>22</b>	<b>100</b>

## DISCUSSION

Bakery products are important staple food that can get infected at the point of processing, handling, storage or at the point of sell. This has caused an economic impact on the consumers and the society at large. Many factors such as moisture content, temperature, relative

humidity, nutrient content of the food etc has contributed to the spoilage of staple food. Dutse and indeed Jigawa State has Sahel Savannah vegetation with high temperature and humidity that supports the growth and proliferation of these fungal species. The study demonstrated that these bakery products were contaminated by



fungi. Table 1 shows the fungi responsible for the spoilage of bakery products which were *Aspergillus niger*, *Aspergillus flavus*, *Penicillium spp*, *Mucor spp*, and *Rhizopus spp*. This is in agreement with the findings of Nimala *et al.*, (2016). Findings from this research revealed that cake, meat pie and bread had the highest contamination from *Aspergillus flavus*, *Aspergillus niger* and *Mucor spp* respectively. These fungi are widely spread in nature and are commonly found in bakery products and other substrate since they can tolerate high salt and sugar concentrations. This agrees with the findings of Chozhavendhan (2014) who earlier reported that these fungi produce mycotoxins which can cause food intoxication in humans and animals.

### CONCLUSION

Bakery products sold at some confectioneries in Dutse Metropolis were found to be contaminated with different species of fungi which are responsible for the spoilage of these staple food which could be due to the ability of the fungi to produce spores that are easily transmitted via air or poor handling and storage conditions which could provide infection sites for spoilage fungi.

### RECOMMENDATIONS

1. To reduce the incidence of bakery products spoilage, bakeries and other food industries should maintain adequate cleaning and sanitation of equipment.
2. Another research should be carried out on other microbial species such as bacteria, virus and protozoa responsible for causing spoilage.
3. People should avoid eating spoiled bakery products because of their effect to human health.
4. The government and other regulatory agencies such as (NAFDAC) should assist in ensuring the production and selling of unspoiled staple food. This could be done by setting up microbiological standard to ensure that the finished products produced will not be hazardous to human health and population at large.

### REFERENCE:

- Akintobi, A. O, S. O. Agunbiade, O. R. Akano and O. Onianwa (2011). Isolation and identification of fungi associated with the spoilage of some selected fruits in Ibadan South Western, Nigeria. *Academia Arena*, 3: 1-7
- Anon. (2000). House hold consumption and expenditure on cereal-based foods. *Home Grown Cereals Authority Weekly Diges*. 18: 2-3.
- Baur, J. (2001). *Theory of bakery and confectionary*, ISBN 978-81-203-3954-5.
- Carla Luciana Gerez, Maria ines Torino. (2009). Prevention of bread mold spoilage by using lactic acid bacteria with antifungal properties. *Journal of Food Science*, 20: 144-148.
- Cheesbrough M, (2006). *District Laboratory Practice in tropical Countries*. 13:978-0-511-349355.
- Chozhavendhen, S. (2014). Prevention of bread spoilage to enhance the quality of bread by using lactic acid bacteria. *International journal of Chemtech Research*. 2014;6(9):4161-4165.
- Ehavalid H (2009). Food safe risk management in bakeries. Presentation at 3rd SAFOODNET seminar St Olav's Hotel, Tallin, Estonia; May 4-6, 2009.
- Hart, C. A. and Shears, P (2004). *Colour atlas of medical microbiology*, 2<sup>nd</sup> ed, mosby. P. 3
- Hickey, C.S. (2000). Sorbate spray application for protecting yeast-raised bakery products. *Baker's Digest*, 54: 4-7.
- Lwanga, S. K. and Lemeshow, S. (1991). Sample size determination in health studies: A practical manual. WHO, Geneva, 15.
- Morsy, S. M., Dorgan, E. A. and Mohamed, G.M. (2009). Effect of garlic and onion extracts or their intercropping on suppressing damping-off and powdery mildew diseases and growth characteristics of cucumber. *Egyptian journal of phytopathology*, 37, 35-46.
- Nimala R, Pathmanathan S, Selvarratnam S. (2016). Study on fungi associated with spoilage of bread. *International journal*



- of Advanced Research in Biological Sciences*. 2016;3(4):165-167.
- Okigbo, Ralph N; Maria I. (2003). Plant protection science; *Prague* vol. 39, Iss. 2, 70-77.
- S. O. Oluwajoba, O. Malomo, O. A. B. Ogunmoyela, O. E. O. Dudu and A. Odeyemi. (2012). Microbiological and nutritional quality of warakashi enriched bread. *Journal of microbiology, Biotechnology and food sciences*. 2, (1), pp.42-68, 2012.