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Evaluation of Pathogenic Microorganisms that Cause Onion Bulb Rots in Selected Markets in Abuja, Nigeria

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Authors' contributions

This work was carried out in collaboration among all authors. Author OTO designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors IRO and ONNF managed the analyses of the study. Author EEU managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

Onion bulb rots are caused by microorganisms especially fungi and bacteria leading to economic loss. The research study evaluated the microorganisms associated with the rots of onion bulbs. The microorganisms isolated include fungi species such as *Saccaromyces cerevisiae, Aspergillus niger*, *Rhizopus stolonifera*, *Fusarium oxysporum* and *Penicillium digitatum* and bacterial species such as *Pseudomonas* spp*., Enterobacter* spp., and *Escherichia coli*. Results from the percentage distribution of the fungi in the spoilt onion bulbs about the markets showed the highest percentage distribution of 42.85% for the samples from Bwari market and the lowest percentage distribution of 28.57% for Dutse and Zuma respectively. It was observed that the percentage distribution of the bacteria about the markets were 42.85% for the samples from Bwari market and 21.42% from Zuma market. Pathogenicity tests for Fungi revealed that all the isolated fungi were pathogenic on onion bulbs, however; *Saccaromyces cerevisiae, Aspergillus niger* and *Penicillium digitatum* were the most pathogenic leading to the rapid disintegration of the infected bulbs within 14 days of inoculation

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while *Rhizopus stolonifer* was the least pathogenic. Pathogenicity tests for Bacteria revealed that all the isolated bacteria were pathogenic on onion bulbs with *Pseudomonas* spp. as the highest pathogenic and *Escherichia coli* the least.

Keywords: Bacteria; fungi; onion bulb; Abuja.

1. INTRODUCTION

Onion (*Allium cepa*) is a universally import culinary vegetable used on account of its volatile flavour components [1]. It is used mainly in its fresh or stored form or as dehydrated flakes or powder in some processed foods. Numerous benefits have been attributed to onions including; prevention of cancer and cardiovascular disorders, reduction in the blood levels of cholesterol, reduction in osteoporosis, reduction in stomach ulcers, inhibition of the proliferation of cultured ovarian, breast and colon cancer cells, inhibition of platelets-mediated thrombosis, prevention of inflammatory processes associated with asthma, treatment of fever, common cold, cough, sore throat and its use as an antimicrobial agent. In the tropics most onions are grown with irrigation during the relatively cool dry season in a semi-arid region, they are not a good crop for hot wet tropics. In Nigeria, Onion is grown exclusively in the northern part of the country, grown during the dry season (October to April). The spoilage of onion bulbs is mainly due to harvesting, post-harvest handling and marketing practices. In tropical countries, the storage is in ambient temperature (23 - 32 degree Celsius) and at a variable relative humidity depending on location and season. The onion farmers in Nigeria almost always store their onions after harvest for one to five months to ensure a continual supply through seasons when fresh produce was unavailable, as a result, some of them got rotten and spoilt. Hence, Onion bulb rots are a common cause of onion loss during storage. Furthermore, the deterioration of raw onions may result from physical factors; actions of their own enzymes, microbial or combination of these factors. Mechanical damage resulting from the action of animals, birds or insects or from brushing, wounding, bursting, cutting, freezing, desiccation or other mishandling may predispose onion towards increased enzymatic action or entrance, and growth of microorganisms. Also, previous damage by plant pathogens may make the part of the crop used as food unfit for consumption or may open the way for the growth of saprophytes and spoilage by them. The prominent storage diseases of onion bulb have been reported to be caused by fungi such as *Aspergillus niger* (black rot), *Botrytis* (neck rot) and *Fusarium oxysporum* (bulb-end rot) [2]. Equally significant are the soft rot diseases of bacterial origin, particularly those that are caused by *Pseudomonas allicola, P. marginalia and P. cepacea*. The present study investigates the microbial species involved in onion bulb spoilage in selected markets in Abuja, Nigeria.

2. MATERIALS AND METHODS

2.1 The Study Area

The study was conducted from November to May, in the Department of Biological Sciences, Veritas University, Bwari Area Council, FCT Nigeria. Bwari , Kubwa and Zuma markets were used for the sampling. The markets are located in the northeastern part of FCT and characterized by long dry season (October to April) and a short rainy season (May to September). Average monthly temperature ranges from 21 to 35ºC and is lowest in December and January. Temperature is more severe in March and April.

2.2 Collection of Samples

Onion bulbs showing signs of rotting and discolouration were randomly selected from stock of onion sold at Kubwa and Bwari Market in Abuja, for microbiological analysis.

2.3 Isolation Procedure

The method [3] was employed. The bulbs were stripped of their outer dry scales and the surface was sterilized in 1% Sodium hypochlorite solution for 60 sec. These were then rinsed in three successive changes of sterile distilled water and blotted dry with sterile filter paper. Small segments of the onion tissues, from the margins of rotted parts were cut out with a sterile scalpel and plated on potato dextrose agar (PDA). The plates were incubated at room temperature (25-28ºC) for 5 days for growth.

To isolate bacterial pathogens from the rotting onion bulb, the Plate Count Agar was used. Sample was picked from the rotting part of the onion bulb and inoculated onto the solid medium by streak method. Plates were incubated at 37ºC for 24 hours.

2.4 Preparation of Pure Cultures

Pure cultures of the bacterial isolates were prepared by subculturing on nutrient agar medium.

2.5 Identification of Isolates

Morphological characteristics were observed while Gram's staining and biochemical tests were also carried out for further identification of the bacterial isolates. For fungal Isolates, the colonial morphology was studied and observations were recorded. The fungi plates were examined by flooding lactophenol cotton blue on a slide and fungus placed from the fungus plate by cutting part of the agar along with the fungus, teasing on the slide, covered with a cover slip and placed under the microscope. Samples were examined using the X40 objective lens of the microscope and the observations were recorded. The plates were kept in the refrigerator for further analysis.

3. RESULTS AND DISCUSSION

The fungal species were identified as *Penicillium digitatum*, *Fusarium oxysporum*, *Rhizopus stolonifer, Aspergillus niger* and *Saccharomyces cerevisiae* while bacteria species includes; *Escherichia coli*, *Enterobacter* spp., and *Pseudomonas* spp (Table 1). *Penicillium digitatum* was isolated from the samples from Zuma market only, *Fusarium oxysporum* from the samples from Dutshe and Bwari markets, *Rhizopus stolonifer* from the samples from Bwari market only, from the samples from Bwari and Zuma markets, *Aspergillus niger* from the samples from all markets while *Saccharomyces cerevisiae* was isolated from the samples from Dutshe market only. *Enterobacter* spp was isolated from the samples from Bwari, Dutshe and Zuma Markets, *Escherichia coli* was isolated from the samples from Bwari market only While *Pseudomonas aeruginosa* from the samples from Dutse and Zuma markets.

3.1 Distribution of the Fungi Associated with Onion Spoilt in the Markets

The distribution of the fungi in relation to the markets is presented in Table 5. The percentage distribution was 28.57%, 42.85% and 28.57% for the samples from Dutshe, Bwari and Zuma Markets respectively.

Table 1. Fungal and bacterial species isolated from the rotted onion bulbs

Market	Fungal species	Bacterial species
Dutse	*Aspergillus fusarium oxysporium and Sacharomyces cerevisiae*	*Enterobacter* spp *and Pseudomonas aeruginosa*
Bwari Central	*Aspergillus fusarium Oxysporium and Rhizopus tolonifer*	*Escherichia coli and Enterobacter* sp
Zuma village	*Aspergillus niger and Penicillium digitatum*	*Pseudomonas aeruginosa and Enterobacter* sp

Table 2. Colony morphology of bacterial isolates

Isolates	Form	Size	Margin	Elevation	Colour of colony
Escherichia coli	circular	large	undulate	flat	cream
Enterobacter sp	round	small	entire	Slightly raised	Yellowish-white
Pseudomonas sp	circular	small	entire	raised	Yellowish-white

Table 3. Grams and Biochemical reactions of bacterial isolates

Cell Morphology	Grams reaction	Catalase	Citrate	Coagulase	Indole	motility	Possible organism
Rods	+	+	-	-	+	+	*Escherichia coli*
Cocci	-	-	-	-	+	-	*Enterobacter sp*
Rods	-	+	+	-	-	-	*Pseudomonas sp*

(+) positive (-) negative

Table 4. The physical and microscopic characteristics of fungi isolated from onion bulbs

Physical characteristics	Microscopic characteristics	Fungi isolated
Greenish-grey colony	septate aerial conidiophore that is perpendicular	*Penicillium digitatum*
Pink in centre with white edges	Mucor conidia, ovoid to ellipsoidal, slightly curled and pointed at both ends	*Fusarum oxysporium*
Grey, smooth, moist colony	Septate mycelia with branching sporongiospores	*Saccharomyces cerevisiae*
Pale-white colony later turned brown-black	Non-septate mycelia with ovoid shape	*Rhizopus stolonifer*
Pink-like black wide colony with white edges	Large conidia heads, dark brown becoming radiat and split to columns	*Aspergillus niger*

Table 5. Distribution of the fungi in the markets

Market	*Aspergillus niger*	*Penicillium digitatum*	*Fusarium oxysporium*	*Rhizopus stolonifer*	*Saccharomyces cerevisiae*	% distribution
Dutse	1	0	1	0	2	28.57
Bwari	3	0	1	2	0	42.85
Zuma	1	2	0	0	1	28.57

3.2 The Percentage Distribution of the Fungi in the Rotted Onion

The percentage distribution of the fungi in the rotted onion bulbs is shown in Table 6. The percentage distribution was 18.2%, 18.2%, 18.2%, 36.4% and 9.1% for *Penicillium digitatum, Fusarium oxysporum, Rhizopus stolonifer, Aspergillus niger* and *Saccharomyces cerevisiae* respectively.

3.3 Distribution of the Bacteria Associated with Spoilt Onion in Relation to the Markets

The distribution of the bacteria in relation to the markets is presented in Table 7. The percentage distribution was 35.71%, 42.85% and 21.42% for the samples from Dutshe, Bwari and Zuma Markets respectively.

3.4 The Percentage Distribution of the Bacteria in the Rotted Onion

The percentage distribution of the bacteria in the spoilt onion bulbs is shown in Table 8. The percentage distribution was 64.28%, 21.42% and 14.28% *Enterobacter sp*ecies, *Pseudomonas species* and *Escherichia coli* respectively.

3.5 Discussion

The presence of microorganisms (fungi and bacteria) in the onion bulbs is attributable to the environmental conditions, state of handling and processing, state of the storage facility of the onions and the quality of the onion bulbs. These microorganisms have been known to cause diseases of humans and animals. They may come from the air, water, soil and even the handlers. They are the sources of highly potent mycotoxins which are hazardous to health. The presence of these fungi in significant numbers in onion bulbs is, therefore, a public health risk.

Fungal and bacterial isolates from the study are similar to those of [4], who also isolated *Aspergillus niger*, *Rhizopus stolonifer* and *Fusarium oxysporum* from the rotten onion bulbs sold at five different markets in Sokoto, Nigeria, [3,5] also implicated fungi as contaminants of many agricultural commodities including onions. Also [6] reported *Aspergillus niger* as the causative agent of black mold rot of onions while [7], isolated *Enterobacter species*, *Pseudomonas species*, *Escherichia coli* from rotten onion bulbs sold in Uyo Metropolis, Akwa Ibom, Nigeria.

Table 6. Percentage distribution of the fungi from the spoilt onion bulbs

Fungi	Number of isolates	% distribution
Penicillium digitatum	2	18.2
Fusarium oxysporum	2	18.2
Rhizopus stolonifer	2	18.2
Aspergillus niger	4	36.4
Saccharomyces cerevisiae	1	9.1

Table 7. Percentage distribution of the bacteria in the markets

Market	*Enterobacter* sp(n)	*Pseudomonas* sp (n)	*Escherichia coli* (n)	% distribution
Dutse	3	3	0	35.71
Bwari	4	0	2	42.85
Zuma	2	1	0	21.42

Table 8. Percentage distribution of bacteria isolates from the spoilt onion bulbs

Bacteria	Number of isolates	% distribution
Enterobacter sp	9	64.28
Pseudomonas sp	3	21.42
Escherichia coli	2	14.28

The percentage distribution of the fungi in the spoilt onion bulbs in relation to the markets showed the highest percentage distribution of 42.85% for the samples from Bwari market and the lowest percentage distribution of 28.57% for the samples from Dutshe and Zume respectively while the percentage distribution of the bacteria in relation to the markets showed the highest percentage distribution of 42.85% for the samples from Bwari market and the lowest percentage distribution of 21.42% from Zuma market. It was also observed that *Aspergillus niger* had the highest distribution of fungi in the spoilt onion bulb (36.4%) (Table 2) while *Enterobacter species* had the highest distribution of bacteria in the spoilt onion bulbs of 64.28%, (Table 3).

In Table 1. Findings showed that all the isolated fungi were found to be pathogenic to the onion bulbs, where *Saccaromyces cerevisiae, Aspergillus niger and Penicillium digitatum were* more pathogenic than the other fungi leading to the rapid disintegration of the infected bulbs within 14 days of inoculation. The least pathogneic among the fungi is *Rhizopus stolonifer* with 22 mm in terms of the size of rot on the affected bulb. However, the present investigation indicates that the aforementioned fungi were associated with deterioration of onion bulbs. This agrees with the findings of [3,4] that fungi constitute a menace in the storage of many agricultural commodities including fruits, vegetables and nuts. Finding also showed that all the isolated bacteria were also found to be pathogenic, the highest among of the bacteria in terms of the size of rot is *Pseudomonas* spp. with 41 mm leading the rapid disintegration of the infected bulbs within 14 days on inoculation. It was observed that *Escherichia coli* was the least pathogenic bacteria.

4. CONCLUSIONS

The fungi isolated from the spoilt onion bulbs were identified as *Penicillium digitatum*, *Fusarium oxysporum, Rhizopus stolonifer*, *Aspergillus niger* and *Saccharomyces cerevisiae* (Table 1) while the bacterial isolates from the rotted onion bulbs were identified as *Enterobacter* species*, Pseudomonas* species*, and Escherichia coli*. The high occurrence of fungi in the rotted onion bulbs is a public health risk. Appropriate control measures must be employed during the harvesting, processing, transportation and handling of the fruits. Good storage facilities should also be put in place to protect the onion bulbs from attacks by these fungi and bacteria, thereby minimizing wastes due to deterioration.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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