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PATHOGENIC FUNGI ASSOCIATED WITH POST-HARVEST DETERIORATION OF CASSAVA

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ABSTRACT

Cassava (*Manihot esculenta*) is a valued root crop grown throughout the tropics for food, feed, biofuel, and other industrial products. Nigeria is the largest producer of Cassava, and post-harvest deterioration of Cassava is one of the major challenges of long-term storage of cassava tubers. This study aimed to isolate and characterize pathogenic fungi implicated in the deterioration of cassava tubers, using samples from Iwo, Osun State, and Ijebu-Ode, Ogun State, Nigeria. The fungi genera isolated at the end of this study and their frequency of occurrence were *Aspergillus* (45.5%), *Botryodioplodia* (9.1%), *Penicillium* (18.2%), *Rhizopus* (18.2%), and *P.sorghina* (9.1%). A total of eleven fungi species were isolated and characterized in this study, seven of which were from the Ijebu- Ode Cassava samples and 4 fungi species from the Iwo samples. The pathogenicity test showed that *Aspergillus nigricans* (60mm) and *P.sorghina* (50mm) caused extensive deterioration of Cassava tuber. However, mild deterioration was observed from samples inoculated with *Botryodiplodia* (10mm) and *Penicillium* (14mm). This study has shown that a consortium of fungi species is implicated in the Post harvested deterioration of Cassava tubers.

Contribution/Originality: The primary contribution of this study is to determine the pathogenesis and frequency of occurrence of fungi associated with post-harvest deterioration of Cassava. It shows the necessity of improving post-harvest/storage practices during Cassava production.

1. INTRODUCTION

Cassava (*Manihot esculenta*) is a major food crop in Nigeria, supplying about 70% of the daily calories for about 50 million people. It is also widely consumed around the world, providing nutrition for an estimated 500 million people daily [1]. It is low in protein and fat and especially high in carbohydrates [1]. The edible part of fresh Cassava root contains 32-35% carbohydrate, 2-3 % protein, 75-80% moisture, 0.1% fat, 1% fiber and 0.75-2.5% ash [1-4]. No continent depends on root and tuber crops, especially Cassava in feeding its population as much as Africa. Cassava plays a crucial role in Africa's agricultural sector because it can thrive in poor soils, and it offers flexibility to farmers as a subsistence or cash crop [5]. Second to Yam, Cassava is the most cultivated root crop produced in Africa. Cassava is sometimes referred to as the bread of the tropics [6]. It is used to produce alcoholic beverages, biofuel, animal feeds, food, and industrial purposes. Although Cassava is an important crop with diverse uses, it does not receive all the needed attention during its production. Cassava has a high moisture content which predisposes it to post-harvest spoilage. Fungi are one of the predominant perpetrators of post-harvest deterioration of Cassava. The high moisture content of fresh tubers, high humidity, conducive environmental temperature, coupled with the bruises received during harvesting make these microorganisms formidable [6]. The deterioration of Cassava starts as quickly as 48 hours post-harvest [7]. Several investigations report a very short period before Cassava tuber becomes completely spoilt and wasted [7]. Post-harvest deterioration of Cassava is of great concern especially in Nigeria, because she is the largest producer of Cassava in the world, producing over 200 million tonnes of cassava

annually [8]. Poor and inadequate post-harvest practices, inaccessibility to storage facilities, and poor access roads are the major contributing factors to the extensive wastage of this root crop [7]. However, Cassava remains a desirable crop because of its doggedness and economic value.

This research aims to isolate, characterize, and identify fungi species associated with the post-harvest deterioration of Cassava (*Manihot esculenta*), emphasizing their degree of pathogenicity.

2. MATERIALS AND METHODS

2.1. Sample Collection

Fourteen diseased Cassava tubers were obtained from two local farms at Ijebu-Ode and Iwo. The samples were transported to the Microbiology Laboratory of Bowen University, Iwo Osun State, Nigeria for analysis.

2.2. Isolation of Fungi Pathogens

The surface of the deteriorating samples was sterilized with 70% ethanol solution. A sterilized kitchen knife was used to cut open the samples to reveal the boundary between healthy and rotten parts. The deteriorating portions were carefully sliced into bits (about 3mm in diameter). These portions were inoculated on solidified Sabouraud dextrose agar. The plates were incubated at 27°C for 3 days. On the third day, the fungi consortium was sub-cultured repeatedly until a pure culture was obtained.

2.3. Determination of the Percentage of Fungal Occurrence

The percentage frequency of occurrence of the different fungal isolates was determined. The number of occurrences of each isolate were recorded and calculated as a ratio of the total number of occurrences and expressed in percentage. It was given by the formula: $\eta/N \times 100$

Where;

η = Total number of times the organism occurred.

N = Total number of all fungi isolates in the samples screened.

2.4. Characterization and Identification of Pathogenic Fungi

The isolated fungi were identified based on the morphological appearance on SDA medium. The microscopic features of the fungi isolates were observed under the microscope at x40 objective.

2.5. Pathogenicity Testing

Each fungus isolate obtained from the diseased cassava tubers was tested for its ability to cause the same disease conditions in a healthy cassava tuber. The tubers were washed with sterile distilled water, thereafter, disinfected with 70% ethanol solution. Cylindrical discs were removed from the disinfected tubers with a sterile 5mm cork borer, fungi isolates were inoculated into the holes and the cylindrical discs were replaced. The inoculation points were sealed with petroleum jelly. The test was carried out for 5 days. A healthy piece of cassava tuber was used as a negative control. Another piece inoculated with the consortium of isolates was used as a positive control.

3. RESULTS

A total of 11 molds were isolated from the Ijebu and Iwo samples. The molds and their features are presented in Table 1.

The Figure 1 and Figure 2 are the results of the pathogenicity test after 5 days.

4. DISCUSSION AND CONCLUSION

Cassava (*Manihot esculenta*) is an economically important crop in the tropical and subtropical regions of the world, most especially in South America and Sub- Saharan Africa. It is particularly of importance to the economy of Nigeria, which is the largest producer and exporter of Cassava [8]. Cassava is a rugged crop that grows in unfavorable soil conditions, requiring little use of fertilizers. Fungi associated post- harvest deterioration of Cassava is one the major challenges facing cassava productions in Nigeria. According to the research carried out by Agu, et al. [7] of the department of Microbiology, Nnamdi Azikiwe University Nigeria, the fungi associated with post-harvest deterioration of cassava were: *Fusarium solani*, *Lasiodiplodia theobromae*, *Aspergillus terreus*, *Aspergillus tamarii*, *Aspergillus flavus and Fusarium oxysporum*. In this study, some similar fungi genera such as *Aspergillus* and *Lasiodiplodia* were isolated from our samples. However, contrary to the results of Agu, et al. [7], fungi belonging to the genus *Penicillium* and *Rhizopus* were isolated from our samples while fungi of the genus *Fusarium* were not isolated. According to Table 2, Fungi of the genus *Aspergillus* had the highest frequency of occurrence, while *P. sorghina* was isolated only from the Iwo samples. This shows that these genera are common spoilage fungi of Cassava, irrespective of which location the samples were obtained. This study also corresponds to the research carried out by Ogaraku and Usman [2] which reported that *Aspergillus niger*, *Aspergillus flavus*, *Fusarium oxysporum*, *Rhizopus*, *Scelerottium rolfstii* were associated with the deterioration of Yam tubers. This study further elucidates that

fungi deterioration is indeed a menace not only to Cassava production, but also the production of other tuber crops and similar fungi genera are implicated.

Table-1. Macroscopic and Microscopic morphology of Fungal isolates.

Isolates	Macroscopic Morphology	Microscopic Morphology	Identity
Ijb 1	Dark spores, white cottony hyphae, slow growing with a spreading pattern. It was incubated at 27°C for 5 days.	It retains the color of the dye, dark spores, coenocytic septation	Botryodiplodia
Ijb 2	Short whitish conidiophore with greyish- green spores. Contoured growth and slow growing. It was incubated at 27°C for 5 days.	It retains the color of the dye, greyish-green spores, septate with brush like ends	Penicillium sp
Ijb 3	White sporangiophores with dark spores. It is fast growing with a spreading growth pattern. Incubated at 27°C for 5 days.	It retains the color of the dye, dark spores, aseptate with rhizoids.	Rhizopus sp
Ijb 4	Black spores with white cottony hyphae. It has a spreading and a profuse growth pattern. Incubated at 27°C for 5 days.	It does not retain the color of the dye, dark spores, septate with foot cell	Aspergillus niger
Ijb 5	Dark spores with yellowish hyphae. It has a spreading and profuse growth pattern. Incubated at 27°C for 5 days.	It does not retain the colour of the dye, septate with a foot cell.	Aspergillus nigricans
Ijb 6	Yellow brown pigmentation, whitish conidiophore.	It does not retain the color of the dye, septate with a foot cell	Aspergillus terreus
Ijb 7	Blue- green spores with a white hypha. Contoured and slow growing. Incubated at 27°C for 5 days.	It retains the color of the dye, greenish spore, and septate.	Penicillium chrysogenum
Iwo 1	Greyish-white woolly hyphae with dark spores. On the third day, it turned SDA pinkish red. Incubated at 27°C for 5 days.	It retains the color of the dye, dark spores and septate hyphae	P.sorghina
Iwo 2	Greyish-white wooly hyphae with dark spores, profuse and spreading growth pattern. Incubated at 27°C for 5 days.	It does not retain the color of the dye, dark spores, septate with a foot cell.	Aspergillus niger
Iwo 3	Dark spores with whitish hyphae, spreading and profuse growth pattern. Incubated at 27°C for 5 days.	It retains the color of the dye, dark spores and non-septate	Rhizopus sp
Iwo 4	Dark spores with whitish hyphae, spreading and profuse growth pattern. Incubated at 27°C for 5 days.	It does not retain the color of the dye, dark spore, septate with a foot cell.	Aspergillus sp

Table-2. Frequency of occurrence of each isolate.

Isolates	Frequency in Ijebu samples	Frequency in Iwo samples
Aspergillus sp	42.9%	50%
Penicillium sp	28.6%	0%
Rhizopus sp	14.3%	25%
Botryodiplodia sp	14.3%	0%
P. sorghina	0%	50%

The result of the pathogenicity test (as shown in Figures 1 and 2) indicates that *Aspergillus nigricans* and *P. sorghina* caused extensive deterioration of Cassava tubers while *Botryodiplodia* and *Penicillium* only caused mild deterioration of the tubers. *Neurospora* sp was also isolated from all the samples but were eliminated at the early stages of the experiment because of its rapid growth rate and propensity to disperse its spores, thereby contaminating other fungi culture.

In conclusion, fungi associated with post-harvest deterioration is a menace to Cassava production. Therefore, research efforts focused on improving post-harvest practices will be beneficial in reducing the incidence of spoilage and waste, thereby improving Cassava production and output.

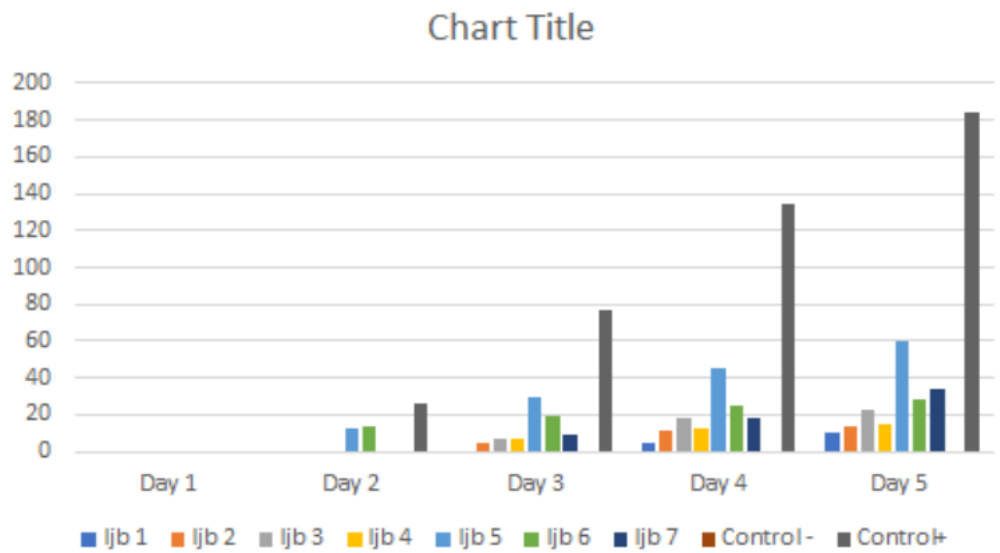

Figure-1. The growth of fungi isolated from the Ijebu samples measured in millimeters during pathogenicity test.

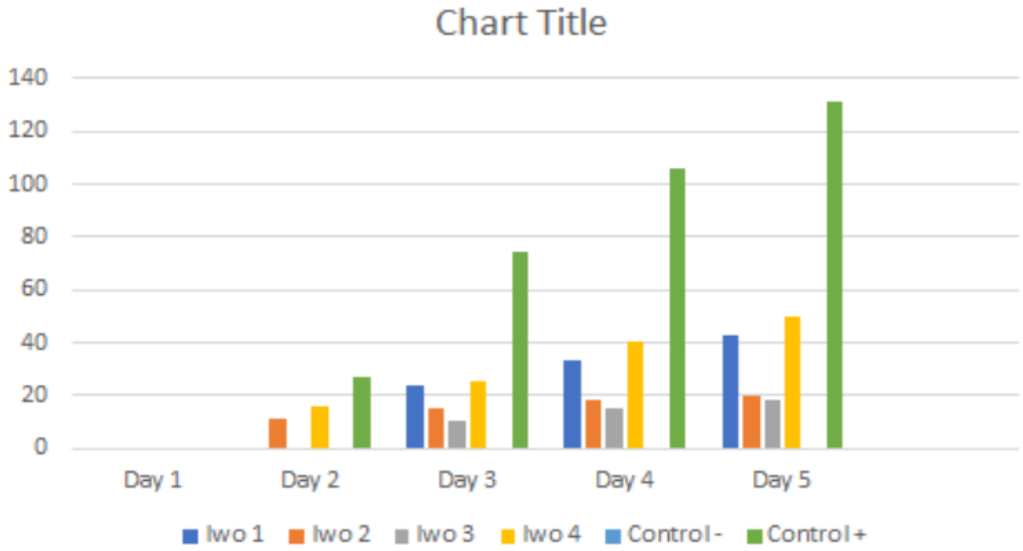

Figure-2. The growth of the fungi isolated from the Iwo samples measured in millimeters during pathogenicity test.

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