

# Susceptibility of White Yam (*Dioscorea rotundata* Poir) Tuber to Rot Fungi and Control with Extracts of *Zingiber officinale* Rosc. *Azadirachta indica* A. Juss. and *Piper guineense* Schumach

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## Abstract

Isolation, identification and control of fungal pathogens of *Ogoja* yam tubers collected from Benue and Nasarawa States, Nigeria were studied between December, 2015 and April, 2017. Fungi identified from the rotted samples were: *Aspergillus flavus*, *A. niger*, *A. ochraceus*, *Botryodiplodia theobromae*, *Curvularia eragrostide*, *Colletotrichum sp.*, *Fusarium moniliforme*, *F. oxysporum*, *F. solani*, *Penicillium expansum*, *Pestalotia sp.* and *P. purpurogenum*. Fungi with the highest mean frequency of occurrence were *A. niger* (21.84%), *B. theobromae* (19.10%), *A. flavus* (16.84%) and *F. oxysporum* (15.49%) while the least were *Colletotrichum sp.* (1.36%) and *P. expansum* (1.49%). Tests of pathogenicity carried out on the head and tail regions of the *Ogoja* yam tubers showed that the head was more susceptible than the tail and all the fungi elicited rot when tested on healthy tubers of yam. Fungi with the highest rot depth both at the head and tail regions were *A. niger* (23.00 mm, 27.33 mm), *A. flavus* (16.33 mm, 21.00 mm) and *B. theobromae* (9.33 mm, 11.33 mm) while the least virulent were *Colletotrichum sp.* (5.00 mm, 6.66 mm) and *P. purpurogenum* (4.00 mm, 7.66 mm), respectively. Extract application of leaves of *Carica papaya* Lam. (pawpaw), rhizomes of *Zingiber officinale* Rosc. (Ginger), *Piper guineense* Schumach. (Black pepper), *Azadirachta indica* A. Juss. (neem), and leaves of *Nicotiana tabacum* Linn. (Tobacco) on tubers before storing for five months showed high level of potency in controlling rot pathogens. These plant extracts could therefore, be applied on tubers to prolong their shelf life.

**Keywords:** Identification; Isolation; *Ogoja*; Pathogenicity tests; Plant extracts; Rot

## Introduction

Cultivated yams are members of the family *Dioscoreaceae*. African, Asian and South American countries are the major producers of yams in the world as reported by Okigbo and Ogonnaya [1], FAO [2]. Varieties of yam that are mostly produced are *Dioscorea rotundata* (white yam) and *D. alata* (water yam). Statistics have shown that Nigeria is the most consistent and largest producer of yams. Annual output in Nigeria equals 38.92 million metric tonnes [3,4]. Pathogens such as bacteria, fungi, viruses and nematodes are responsible for rots in yams at different stages of growth and storage [5-9]. The pathogens reduced growth, quantity and quality of yam [8,10-13]. Rot initiated by pathogens in yam tubers ranged from 10% to 15% after three months of storage [14-16] estimated the losses to be as high as 50% in storage in Nigeria. Pathogens commonly implicated with diseases in yam at various stages of growth included: *Aspergillus flavus*, *A. niger*, *A. ochraceus*, *Botryodiplodia theobromae*, *Colletotrichum spp.*, *Curvularia eragrostide*, *Fusarium moniliforme*, *F. oxysporum*, *F. solani*, *Rhizoctonia spp.*, *Rhizopus nodosus*, *Penicillium expansum*, *P. digitatum*, *P. oxalicum*, *P. purpurogenum* [8-10,17-23]. There are different types of pesticides that are currently in use to control plant pathogens which included chemical fungicides and plant extracts. These chemicals have been found to cause soil pollution, surface and ground water contamination as well as development of resistance by the target organisms [24]. Plant materials as well as antagonistic organisms are the popular options to the use of chemical fungicides [25-29]. Phytochemical compounds synthesized by plants are known to have inhibitory properties against plant pathogenic organisms at different stages of growth and in storage life of crops such as yam, cassava, tomato, cowpea, rice, etc [9,30,31]. Plant products are locally available, biodegradable, little toxicity and are easy to prepare by local farmers [19,32]. The study therefore, aimed at

identifying the different pathogens associated with rots in yam tubers and the use of plant materials to control these pathogens and prolong the lifespan of yam in storage.

## Materials and Methods

### Study area

The location lies between latitudes 8°30' and 9°30' N, and on longitudes 7°20' and 8°45' E in the North central region of Nigeria called 'Middle belt'. The area covers four major yam producing settlements in four local government areas that cut across two States in the derived guinea savannah of Nigeria; the locations are Tor-Donga in Katsina-Ala and Zaki-Biam in Ukum local government areas of Benue State, respectively as well as Lafia in Lafia local government area and Kadarko in Keana local government area of Nasarawa State, respectively.

### Collection of decayed yam tubers

Decayed tubers of *Ogoja* white yam variety with characteristic rot symptoms were got from yam farmers' storage barns at four locations between December, 2015 and April, 2017. Decayed tubers were sampled and neatly collected and assembled in sterilized polyethylene bags

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and conveyed to the University laboratory after which isolation and identification of the fungal pathogens were made.

### Identification of fungal pathogens

Decayed yam tissues were cut about 2 mm diameter at the edge of infected portions with sterile scalpel. Cut tissues were surface sterilized in 5% Sodium hypochlorite (Commercial bleach) solution for few minutes. Cut sections were removed and dipped in sterile water to remove the chemical [9]. The sterilized sections were dried on sterile Whatman No.42 filter paper (Sigma-Aldrich) for 2 minutes before inoculation of the plates. Four sections of the sterilized tissue pieces were plated out in the solidified Potato Dextrose Agar. Incubated plates were kept at ambient room temperature ( $30^{\circ}\text{C} \pm 5^{\circ}\text{C}$ ) for a period of five days for adequate growth of fungal mycelial [19]. Different fungal colonies that grew on the plates were carefully sub cultured on new sterile PDA plates for pure culture to grow. Identified fungi were purified and subsequently multiplied on PDA. Morphological identification was done to ascertain the identities of the fungi with the aid of compound microscope and identification books [33-35].

### Frequency of occurrence of fungi

Frequencies of occurrence of isolates were determined periodically. Fungi organisms were isolated and identified at monthly interval, record of fungi isolated were kept periodically and the number of times of occurrence of a particular fungus was calculated as a percentage of all fungi organisms isolated at a particular time [34], hence expressed as:

$$\text{Percentage frequency of occurrence} = \frac{Y}{Z} \times \frac{100}{1}$$

Where,

Y=Number of times of occurrence of each fungal isolate at a particular time.

Z=Total number of times of occurrence of all fungi organisms isolated over a given period.

### Pathogenicity test

Good looking *Ogoja* varieties of yam tubers (*Dioscorea rotundata*) were collected and neatly washed using clean tap water to get rid of dirt. 5% Sodium hypochlorite solution was used to further disinfect the tubers for up to 2 minutes before the tubers were rinsed in four successive changes of sterile water to remove chemicals [22,23]. Sterile filter papers were used to dry the tubers for about 20 minutes in the Laminar Air flow cabinet. A five day old culture of the fungal isolates grown on potato dextrose agar collected for deteriorated tubers was used as inoculum for pathogenicity tests. A 5 mm diameter cork borer was used to remove 4 mm tissue from the healthy tuber surfaces aseptically from the "head" and "tail" regions of each yam tuber [36]. A cork borer of 5 mm diameter size which was sterilized by dipping in alcohol followed by flaming, the cork borer was allowed to cool before it was used to obtain inoculum from the five-day old cultures of the fungi. Fungal plugs that were cut were inserted into the holes created in the tubers. A portion of the tuber flesh removed earlier was cut off to give way for inoculum size and the remaining tuber flesh was used to plug the remaining parts of the hole made in the yam tubers. Sterile petroleum jelly was used to cover the edges of the inoculated yam tissues. This process prevented any external influence on the positioned inoculum. The procedure was repeated for the control but instead of inserting inoculum in the holes created, uninoculated discs of PDA were inserted in the holes made in the yam tubers [37]. The treatments comprised twelve isolates from rotted yam tubers and a

control inoculated at the two regions of *Ogoja* yam. The experimental design was a  $2 \times 13$  factorial which was replicated three times for all the pathogens. Inoculated yam tubers were completely randomized [38] and placed at room temperature under sterile condition for 14 days for rot to establish. The extent of rot was calculated by using a clean knife to cut open the inoculated yam tubers at the point of inoculation. A sterilized transparent ruler was used to measure the depth of rot. Actual depth was calculated with the formula of Ezeibekwe and Ibe [39] with some modifications:

$$X = Y - Z$$

Where,

X=Actual depth

Y=Final depth

Z=Initial depth

The formula was modified to read as:

$$\text{Percentage rot (\%)} = \frac{\text{Final Depth (Y)} - \text{initial Depth (Z)}}{\text{Final Depth (Y)}} \times \frac{100}{1}$$

### Preparation of plant materials

The preparations of plant materials were done using the methods of Gwa and Akombo [19] Seeds of *Piper guineense* Linn., leaves of *Azadirachta indica* A. Juss. and rhizomes of *Zingiber officinale* Rosc. were properly washed in clean water, dried and ground using mortar and pestle. About 30 g, 60 g and 90 g of the ground plant materials were each added to 1litre of sterile hot water ( $100^{\circ}\text{C}$ ) in 1500 ml Pyrex flask. Mixtures were stirred and left for 24 hours after which they were filtered using four-fold sterile cheese cloth. Concentrations of 30 g/L, 60 g/L and 90 g/L, respectively were made from each of the mixtures of plant extract. Mancozeb (synthetic chemical) was prepared by dissolving 4 g of the chemical in 1L of sterile cold distilled water to obtain 4 g/L concentration. Efficacies of the plant extracts as well as mancozeb were compared for their abilities to inhibit growth of fungal pathogens in storage.

### Effect of some plant extracts and mancozeb on storage pathogens

Potency of leaves of *A. indica* (neem), seeds of *P. guineense* (black pepper), rhizomes of *Z. officinale* (ginger) and mancozeb (chemical fungicides) were applied on yam tubers in storage to see their effects on rot pathogens. Plant extracts were prepared as described above and were sprayed on *Ogoja* white yam tubers using a hand sprayer at concentrations of 30 g/L, 60 g/L and 90 g/L, respectively. Application of mannose on the yam tubers was at 4 g/L. Tubers were left to dry at ambient room temperature for 1 hour and were taken to storage barns and stored for a period of five months. Three tubers of yam made up a treatment and were replicated three times giving a sum of 9 tubers. The 11 treatments comprised of three plant extracts at three concentrations each, mancozeb and a control. The experiment was made of a total of 99 *Ogoja* yam tubers. Completely randomized design was used for the treatments but the control experiment had only sterile distilled water in place of plant extract or chemical which was used to sprinkle on the yam tubers before storing them. Potencies of the extracts and mancozeb in controlling rot causing microbes throughout the study period was determined using the data got for the period of five months. Tubers of yam that were not rotted and those that were rotted in each treatment were determined. Efficacy of plant extracts and mannose at their various levels of concentrations in controlling rot pathogens

of yam in store were tested using the Decay Reduction Index formula developed by Amadioha [40] as:

$$\text{Decay Reduction Index (DRI)} = \frac{\% \text{ decay in control} - \% \text{ decay in treated tubers}}{\% \text{ decay in control}}$$

### Data analysis

Data collected were subjected to Analysis of Variance (ANOVA) using GenStat Discovery Edition 12 for mean separation. Graph Pad Prism 6 was used for trend graphs and means were separated using Fisher's least significant differences (F-LSD) at  $p \leq 0.05$  [41].

## Results

### Identification of yam pathogens

*Aspergillus flavus*, *A. niger*, *A. ochraceus*, *Botryodiplodia theobromae*, *Curvularia eragrostide*, *Colletotrichum sp Fusarium moniliforme*, *F. oxysporum*, *F. solani*, *Pestalotia sp*, *Penicillium purpurogenum* and *P. expansum* were got from rotted *Ogoja* tubers from Nasarawa and Benue States of Nigeria.

### Determination of frequency of pathogens

Table 1 shows the occurrence of yam pathogens across the locations. The most frequently occurred pathogens are *A. niger* (21.84%) followed by *B. theobromae* (19.10%), *A. flavus* (16.84%), *F. oxysporum* (15.49%) and so on while the least frequently occurred pathogenic fungi are *Colletotrichum sp.* (1.36%) followed by *P. expansum* (1.49%) and *C. eragrostide* (2.47%).

### Pathogenicity tests

Tests of pathogenicity of some identified fungi are presented in Figure 1. Fungal organisms tested on the two regions (head and tail) of healthy *Ogoja* yam tubers included *A. flavus*, *A. niger*, *B. theobromae*, *C. eragrostide*, *F. oxysporum* and *P. expansum*. Differences were observed in the pathogenic potency of the different fungal isolates tested at different regions of yams. The tests showed that all the isolates induced rot at both regions of the *Ogoja* tubers 14 days after inoculation of the tubers with the test pathogens. Yams were observed to show symptoms of infection. Tubers that were not inoculated with rot causing fungi did not produced symptoms of rot (Figure 1).

### Effect of the fungal pathogens on rot development at different regions of yam tubers

The fungi that proved most potent in causing rot both at the head and tail regions were *A. niger* followed by *A. flavus*, *B. theobromae* and *F. solani* while the least virulent were *Colletotrichum sp.*, *A. ochraceus* and *P. purpurogenum* (Table 2). Significant differences ( $p \leq 0.05$ ) were observed in rot development and spread between head and control and between tail and control regions for all the fungal pathogens tested on *Ogoja* yam tubers. Data obtained and analysed revealed that rot depths were higher at the tail regions of the yam tubers in comparison with the head regions for any of the tested pathogen (Table 3). Each of the isolates induced rot on the head region but there was no rot observed on the head region where no organism was inoculated (control). Similarly, the tail regions inoculated with different isolates

Pathogen	Location and frequency of occurrence (%)				Mean
	Kadarko	Lafia	Tor-Donga	Zaki-Biam	
<i>A. niger</i>	20.08 ± 0.90 <sup>b</sup>	22.90 ± 0.92 <sup>a</sup>	22.93 ± 0.57 <sup>a</sup>	21.43 ± 0.71 <sup>ab</sup>	21.84 ± 0.43
<i>B. theobromae</i>	18.34 ± 0.62 <sup>b</sup>	20.37 ± 0.60 <sup>ab</sup>	15.46 ± 0.74 <sup>c</sup>	22.22 ± 1.10 <sup>a</sup>	19.10 ± 0.58
<i>A. flavus</i>	16.44 ± 0.35 <sup>a</sup>	19.06 ± 1.11 <sup>a</sup>	15.72 ± 0.77 <sup>b</sup>	16.15 ± 1.52 <sup>a</sup>	16.84 ± 0.54
<i>F. oxysporum</i>	16.33 ± 0.58 <sup>a</sup>	16.15 ± 0.70 <sup>a</sup>	15.22 ± 0.80 <sup>a</sup>	14.24 ± 0.61 <sup>b</sup>	15.49 ± 0.35
<i>F. moniliforme</i>	10.35 ± 0.56 <sup>a</sup>	11.26 ± 0.64 <sup>a</sup>	11.66 ± 0.57 <sup>a</sup>	6.24 ± 1.62 <sup>b</sup>	9.87 ± 0.59
<i>P. purpurogenum</i>	NE	4.73 ± 1.81 <sup>ns</sup>	6.71 ± 0.43 <sup>ns</sup>	4.23 ± 1.18 <sup>ns</sup>	3.92 ± 0.68
<i>Pestalotia sp</i>	NE	NE	7.19 <sup>ns</sup>	3.47 <sup>ns</sup>	2.66 ± 0.58
<i>A. ochraceus</i>	3.42 ± 0.99 <sup>b</sup>	NE	6.69 ± 0.35 <sup>a</sup>	NE	2.59 ± 0.54
<i>F. solani</i>	10.01 ± 0.53	NE	NE	NE	2.50 ± 0.25
<i>C. eragrostide</i>	5.53 ± 0.60 <sup>b</sup>	4.37 ± 0.44 <sup>a</sup>	NE	NE	2.47 ± 0.53
<i>P. expansum</i>	NE	NE	NE	5.99 ± 0.60 <sup>ns</sup>	1.49 ± 0.48
<i>Colletotrichum sp</i>	4.32 ± 0.70	1.14 ± 1.61	NE	NE	1.36 ± 0.26

Means occurring on same row but with unlike superscript <sup>a, b</sup> and <sup>c</sup> are significantly different ( $p \leq 0.05$ ); NE=Not Encountered; ns=Not Significant

**Table 1:** Mean percentage frequency of isolated fungal pathogens from *Ogoja* white yam tuber at different locations for four months.

Pathogens	Head	Mean Rot Depth (mm) and Percentage Rot (%)		
		Tail	% Head	% Tail
<i>A. flavus</i>	16.33 ± 0.88 <sup>b</sup>	21.00 ± 1.53 <sup>b</sup>	15.76	15.52
<i>A. niger</i>	23.00 ± 1.53 <sup>a</sup>	27.33 ± 2.85 <sup>a</sup>	22.19	20.2
<i>A. ochraceus</i>	4.33 ± 0.66 <sup>e</sup>	7.00 ± 0.57 <sup>d</sup>	4.18	5.17
<i>B. theobromae</i>	9.33 ± 1.76 <sup>c</sup>	11.33 ± 1.67 <sup>c</sup>	9	8.37
<i>C. eragrostide</i>	7.66 ± 0.88 <sup>cd</sup>	9.33 ± 1.45 <sup>cd</sup>	7.39	6.9
<i>Colletotrichum sp</i>	5.00 ± 0.57 <sup>de</sup>	6.66 ± 0.88 <sup>d</sup>	4.82	4.92
<i>F. moniliforme</i>	6.33 ± 1.76 <sup>cdde</sup>	8.33 ± 1.45 <sup>cd</sup>	6.11	6.16
<i>F. oxysporum</i>	5.66 ± 0.33 <sup>de</sup>	8.00 ± 1.53 <sup>cd</sup>	5.46	5.91
<i>F. solani</i>	9.00 ± 0.57 <sup>c</sup>	10.67 ± 0.88 <sup>cd</sup>	8.68	7.89
<i>P. expansum</i>	6.66 ± 0.33 <sup>cdde</sup>	9.66 ± 0.88 <sup>cd</sup>	6.43	7.14
<i>P. purpurogenum</i>	4.00 ± 0.57 <sup>e</sup>	7.66 ± 0.88 <sup>cd</sup>	3.86	5.66
<i>Pestalotia sp</i>	6.33 ± 1.45 <sup>cdde</sup>	8.33 ± 1.45 <sup>cd</sup>	6.11	6.16
Control	0.00 ± 0.00 <sup>f</sup>	0.00 ± 0.00 <sup>e</sup>	-	-

Means occurring on same column with unlike superscript a, b, c, d, e and f are significantly different ( $p \leq 0.05$ )

**Table 2:** Mean rot depth and percentage rot caused by different fungal pathogens in different regions of *Ogoja* white yam tuber after 14 days of incubation.

also induced varying degrees of rots but no rots were observed on the control tubers where no organism was inoculated. At the head region, *A. niger* (22.19%) was more virulent followed by *A. flavus* (15.76%), *B. theobromae* (9.00%) and *F. solani* (8.68%) while the least virulent isolates were *P. purpurogenum* (3.86%) and *A. ochraceus* (4.18%). The tail region of yam tubers showed a similar trend of susceptibility to the pathogens but the least virulent isolates were *Colletotrichum sp.* (4.92%) and *A. ochraceus* (5.17%). Comparison of the virulence of the isolates on the head and tail regions of the *Ogoja* tubers inoculated with

different pathogenic fungal showed significant differences ( $p \leq 0.05$ ) only in *A. ochraceus* and *P. purpurogenum* (Table 3).

### Effect of extracts of plant and mancozeb on stored *Ogoja* yam tubers

Results presented in Table 4 show the activity of 30 g/L, 60 g/L and 90 g/L of plant extracts and 4 g/L of chemical fungicide on rot pathogens of stored *Ogoja* yam tubers for five months in two years. Findings showed average decay reduction index (DRI) of 0.00

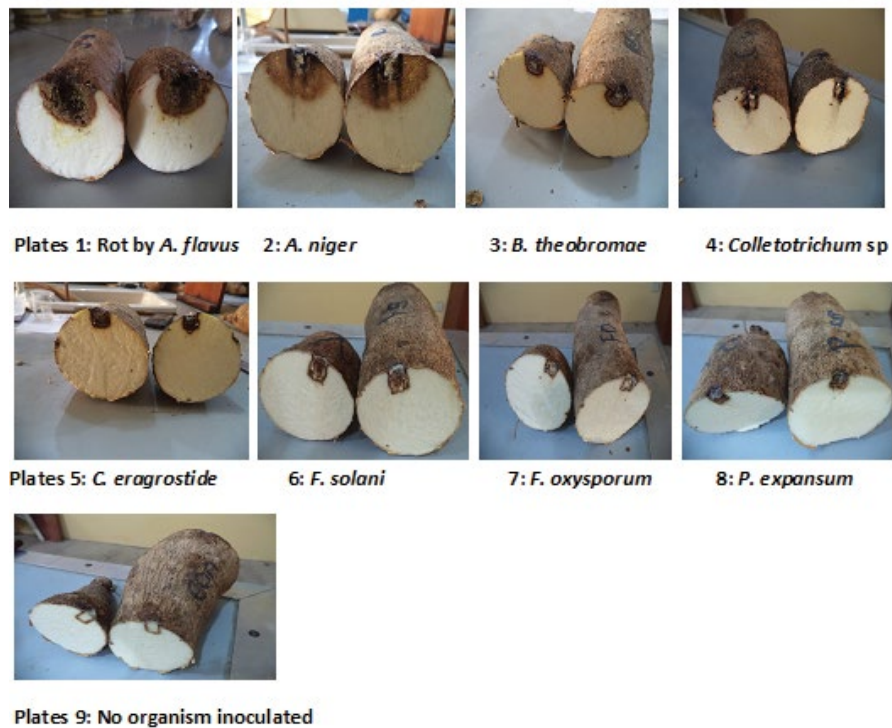


Figure 1: Effect of the fungal pathogens on rot development at different regions of yam tubers.

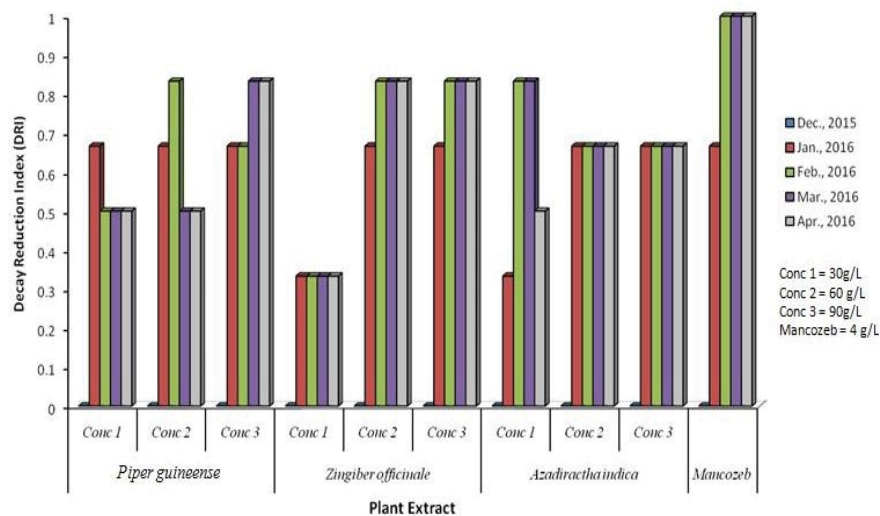


Figure 2: Decay reduction index of *P. guineense*, *Z. officinale*, *A. indica* and mancozeb in controlling rot fungi of *Ogoja* white yam tuber after five months of storage.

Pathogens	Mean Rot Development (mm)		p-Value
	Head	Tail	
<i>A. flavus</i>	16.33 ± 0.88	21.00 ± 1.53	0.07
<i>A. niger</i>	23.00 ± 1.53	27.33 ± 2.85	0.27
<i>A. ochraceus</i>	4.33 ± 0.66	7.00 ± 0.57	0.05*
<i>B. theobromae</i>	9.33 ± 1.76	11.33 ± 1.67	0.47
<i>C. eragrostide</i>	7.66 ± 0.88	9.33 ± 1.45	0.39
<i>Colletotrichum sp</i>	5.00 ± 0.57	6.66 ± 0.88	0.21
<i>F. moniliforme</i>	6.33 ± 1.76	8.33 ± 1.45	0.41
<i>F. oxysporum</i>	5.66 ± 0.33	8.00 ± 1.53	0.27
<i>F. solani</i>	9.00 ± 0.57	10.67 ± 0.88	0.21
<i>P. expansum</i>	6.66 ± 0.33	9.66 ± 0.88	0.08
<i>P. purpurogenum</i>	4.00 ± 0.57	7.66 ± 0.88	0.04*
<i>Pestalotia sp</i>	6.33 ± 1.45	8.33 ± 1.45	0.38
Control	0.00 ± 0.00	0.00 ± 0.00	--

\*Significantly different (0.05%)

**Table 3:** Pathogenicity tests of some fungal pathogens showing rot development (mm) at head and tail regions of *Ogoja* tuber after 14 days of incubation.

Period of Storage	Plant Extract				LSD
	Mancozeb®	<i>N. tabacum</i>	<i>P. guineense</i>	<i>Z. officinale</i>	
<b>1<sup>st</sup> Year</b>					
December, 2015	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	--
January, 2016	0.66 ± 0.33	0.55 ± 0.17	0.66 ± 0.16	0.55 ± 0.17	0.61 <sup>ns</sup>
February, 2016	1.00 ± 0.00	0.72 ± 0.14	0.66 ± 0.14	0.66 ± 0.14	0.48 <sup>ns</sup>
March, 2016	1.00 ± 0.00	0.72 ± 0.14	0.61 ± 0.13	0.66 ± 0.14	0.48 <sup>ns</sup>
April, 2016	1.00 ± 0.00	0.61 ± 0.16	0.61 ± 0.13	0.66 ± 0.14	0.50 <sup>ns</sup>
<b>Mean (1<sup>st</sup> Year)</b>	<b>0.73 ± 0.06</b>	<b>0.52 ± 0.08</b>	<b>0.51 ± 0.09</b>	<b>0.51 ± 0.10</b>	0.32 <sup>ns</sup>
<b>2<sup>nd</sup> Year</b>					
December, 2016	0.66 ± 0.33	0.66 ± 0.16	0.55 ± 0.17	0.66 ± 0.16	0.60 <sup>ns</sup>
January, 2017	0.66 ± 0.33	0.33 ± 0.16	0.44 ± 0.17	0.55 ± 0.17	0.61 <sup>ns</sup>
February, 2017	1.00 ± 0.00	0.55 ± 0.17	0.77 ± 0.14	0.77 ± 0.14	0.52 <sup>ns</sup>
March, 2017	1.00 ± 0.00	0.55 ± 0.17	0.66 ± 0.16	0.77 ± 0.14	0.55 <sup>ns</sup>
April, 2017	1.00 ± 0.00	0.61 ± 0.13	0.77 ± 0.12	0.83 ± 0.08	0.39 <sup>ns</sup>
<b>Mean (2<sup>nd</sup> Year)</b>	<b>0.86 ± 0.13</b>	<b>0.54 ± 0.12</b>	<b>0.64 ± 0.11</b>	<b>0.72 ± 0.08</b>	0.37 <sup>ns</sup>
<b>Mean (both Years)</b>	<b>0.80 ± 0.05<sup>a</sup></b>	<b>0.53 ± 0.08<sup>b</sup></b>	<b>0.57 ± 0.08<sup>ab</sup></b>	<b>0.61 ± 0.05<sup>ab</sup></b>	<b>0.26</b>
<b>Difference between Years</b>					
1 <sup>st</sup> Year	0.73 ± 0.06 <sup>b</sup>	0.52 ± 0.08	0.51 ± 0.09	0.51 ± 0.10 <sup>b</sup>	--
2 <sup>nd</sup> Year	0.86 ± 0.13 <sup>a</sup>	0.54 ± 0.12	0.64 ± 0.11	0.72 ± 0.08 <sup>a</sup>	
LSD	0.12	0.15 <sup>ns</sup>	0.20 <sup>ns</sup>	0.12	

Means occurring on same row (comparing plant extracts) and column (comparing year of storage) with unlike superscript a, b and c are significantly different ( $p \leq 0.05$ ); ns=Not Significant

**Table 4:** Mean decay reduction index (DRI) of mancozeb and plant extracts on *Ogoja* yam tubers after two years of storage.

(control), 0.73 (mancozeb), 0.52 (*A. indica*), 0.51 (*P. guineense*) and 0.51 (*Z. officinale*) were recorded from December, 2015 to April, 2016. Mean DRI of 0.00 (control), 0.86 (mancozeb), 0.54 (*A. indica*), 0.64 (*P. guineense*) and 0.72 (*Z. officinale*) were recorded from December, 2016 to April, 2017. Significant differences ( $p \leq 0.05$ ) were not observed between first and second years of storage in all the plant extracts for each month of storage of the tubers. On the other hand, significant differences in means were observed for both years. Mean differences between the first year and second year of storage for mancozeb and the different plant extracts were significantly difference ( $p \leq 0.05$ ) only in mancozeb and *Z. officinale*.

### Effect of mancozeb and plant extracts in controlling pathogens on *Ogoja* tuber

Figure 2 presents the effect of concentrations of *P. guineense*, *A. indica*, *Z. officinale* and mancozeb in controlling rot fungi of *Ogoja* yam tubers in storage for five months. Results of extract of *P. guineense* at 30 g/L reviewed the decay reduction index from lowest in December, 2015

to highest in January, 2016, respectively. At concentration of 60 g/L, DRI was 0.83 for February, 2016 and 0.50 each for March and April, 2016. Concentration of 90 g/L produced the highest effect in March, 2016 and April, 2016 but was however, low in January and February, 2016. The activity of *Z. officinale* at 30 g/L was the same throughout the period of storage except December, 2015 but differed significantly when 60 g/L and 90 g/L concentrations were used. The performance of *A. indica* at 30 g/L was comparatively better in February and March, 2016 to 60 g/L and 90 g/L with a DRI of 0.66 during the period of storage. Mancozeb had the least DRI in January, 2016 but became better in inhibiting rot pathogens in February, March and April, 2016. Rots were not recorded in December, 2015 in spite of the different concentrations of plant extracts tested.

### Discussion

Pathogenic fungi found inciting rot on yam tubers at the various locations were *Penicillium expansum*, *P. purpurogenum*, *Pestalotia sp*, *Botryodiplodia theobromae*, *Curvularia eragrostide*, *Aspergillus flavus*,

*A. niger*, *A. ochraceus*, *Colletotrichum sp.*, *Fusarium moniliforme*, *F. oxysporum* and *F. solan*. Reports have shown that these fungi constitute the major treat to post harvest deterioration of yam and other crops in different parts of Nigeria [8,18,21,22,24,42]. Fungi with consistent higher frequency of occurrence in the locations included; *A. niger*, followed by *B. theobromae*, *A. flavus*, *F. oxysporum*, *F. moniliforme*. This finding is in agreement with results obtained by Ogaraku and Usman [42], Okigbo et al. [12], Ogunleye and Ayansola [18], Okigbo et al. [7], Gwa and Nwankiti [22], Gwa et al. [23] who reported these fungi as major cause of rot in yam tuber in different parts of Nigeria. Findings revealed that the rot-causing fungi that appeared less included *C. eragrostide*, *P. expansum* and *Colletotrichum sp.* which correspond with findings obtained by Ogunleye and Ayansola [18] and Okigbo et al., [7]. Result got by Ogunleye and Ayansola [18] on fungi pathogens infecting *Dioscorea sp.* in Bodija, Ibadan, Oyo State, revealed that *A. ochraceus* and *Penicillium sp.* were the least occurred fungi with percentage frequencies of 2.14% and 6.43%, respectively which agreed with our findings. Conversely, Amusa [43] showed that *C. gloeosporioides* had higher percentage of occurrence of 98% on white yam leaves compared to just 4.31% got from this finding on yam tubers. Pathogenicity test revealed that *A. niger*, *A. flavus*, *B. theobromae* and *F. solani* are the most virulent fungi causing rot in the healthy Ogoja yam tubers both at the head and tail regions compared with the less virulent *P. purpurogenum*, *F. oxysporum* and *P. expansum*. Tests suggest that these fungi pathogens could be the leading cause of postharvest decay of yam tubers in various parts of Nigeria especially in Benue and Nasarawa States from where all the rotted yam tubers were collected. The results have demonstrated that susceptibility of healthy tubers of yam to rot fungi increased from the head to the tail regions of the tubers; significant differences ( $p \leq 0.05$ ) were not observed in rot between head and tail regions caused by *A. flavus*, *A. niger*, *B. theobromae*, *Colletotrichum sp.*, *C. eragrostide*, *F. moniliforme*, *F. oxysporum*, *F. solani*, *P. expansum* and *Pestalotia sp.* but significant differences ( $p \leq 0.05$ ) were however, observed in rot between head and tail in the least virulent *A. ochraceus* and *P. purpurogenum*. This study is in consonance with results got by Awuah and Akraasi [44] who studied the suppression of yam tuber rot caused by *A. niger* by yam rhizobacterium and reported that the rot development was higher at the tail region compared with the head region when inoculated with the fungus. The study is similar to the work of Taiga [36] who studied the differential rate of dry rot in white yam along the tuber length caused by rot pathogens and found out that the depth of rot caused by *A. niger*, *P. oxalicum* and *Rhizopus stolonifer* were higher in tail followed by middle and least in the head regions of yam tuber and none of the regions of yam for any of the pathogen tested significantly differed from one another. According to Arinze [45] differences in the susceptibility of the various yam regions to pathogens may be due to variable distribution of phytoalexins within the head, middle and tail regions of the tuber. Emehute et al. [46] on the other hand, suggested that variation in the water contents of the various tuber regions, with the head having the least, could be responsible for these differences in the severity of rot caused by the pathogens. It is likely that the low rot severity recorded by some of the pathogens on Ogoja tuber may be caused by the production of phytoalexins in the yam which resisted the advancement of these pathogens. Efficacy of *Z. officinale*, *P. guineense* and *A. indica* as well as mancozeb in controlling rot fungi of stored Ogoja tubers for five months demonstrated that both mancozeb and the plant extracts possess antimicrobial activities capable of inhibiting the growth of pathogens at different concentrations. The differences in potency of *Z. officinale*, *P. guineense* and *A. indica* may be attributed to a build-up of phytochemical nutrients in the various plant extracts [47]. A record of 0.51 decay reduction index indicates that

at least 51% control was achieved using some selected plant extracts and chemical fungicide in managing yam pathogens in storage for five months. The result supports the work of Okigbo et al. [12] who found rot reduction as high as 62.80% in yam tubers with *A. sativum*. The work also agreed with result obtained by Gwa and Akombo [19], Gwa and Nwankiti [9], Gwa [23], Gwa and Nwankiti [22] and Gwa et al. [23] on inhibition of *A. flavus*, *Colletotrichum sp.*, *B. theobromae*, *F. oxysporum*, *C. eragrostide* and *P. expansum*, respectively using extract derived from *P. guineense*, *Z. officinale*, *A. indica*, *N. tabacum* and *C. papaya*. Hycenth [48] reported that *A. indica* had inhibitory effect on *Rhizopus stolonifer* which is also responsible for causing rot of yam tubers. Bonire [49] showed that postharvest losses in yam could go as high as 40%. In the views of Okigbo and Ikediugwu [50] from 20 to 39.5% of stored tubers are lost to pathogens in storage. Arinze [15] and Okigbo et al. [16] showed that about 50% of the yam tubers harvested in Nigeria is lost in storage due to pathogenic infection. The result showed that increase in concentration of plant extracts resulted to decrease in rot indicating an increase in decay reduction index. This increase in concentrations of the extracts implied an increase in the active ingredients of the extracts which acted on the rot pathogens thereby affecting its physiological processes and consequently lowering the infective capability of the pathogens. This study has also confirmed and established the antifungal activity of these plant extracts, which are interestingly systemic in action and can be used or applied as post-harvest tuber treatment against rot causing pathogens of yams.

## Conclusion

The study reviewed that rot of Ogoja yam tubers is caused by different fungal pathogens in storage. Results have also demonstrated that *P. guineense*, *Z. officinale* and *A. indica* may contain phytochemical compounds capable of inhibiting pathogens of yam in storage and comparing with the conventional fungicide (mancozeb); have proved effective against rot inducing pathogens of stored yam tubers. In conclusion, *P. guineense*, *Z. officinale* and *A. indica* are good plant that can be used to inhibit the growth of yam pathogens in storage and extend longevity of the tubers.

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