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# Role of Fungal Rots in Post-harvest Storage Losses in Some Nigerian Varieties of *Dioscorea* Species


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***Authors' contributions***

*Author COF supervisor of this project, and author CAK Student carried out project as component of requirements for M.Sc. of Nnamdi Azikiwe University. All authors read and approved the final manuscript.*


***Original Research Article***

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## ABSTRACT


**Aims:** To study storage rots in yam varieties cultivated in South East Nigeria and to determine under conditions of experimental storage, the influence of fungal rot on their post harvest storage losses.
**Place and Duration of Study:** Laboratories, Department of Applied Microbiology & Brewing, Nnamdi Azikiwe University, Awka, Nigeria between January 2012 and July 2013.
**Methodology:** Five yam varieties; *Dioscorea dumentorum*, two varieties each of *D. alata* and *D. rotundata,* obtained immediately after harvest were stored in an experimental barn (30ºC and 95% RH) and examined at intervals for storage rots. Fungal causative agents of rots were isolated and identified using the partial ITS rDNA sequencing analysis and a BLAST search using the GenBank sequence database. Post harvest storage losses in terms of weight loss and reduction of shelf life among the varieties were determined.
**Results:** All varieties of yams studied suffered fungal rots, predominantly, dry rots during storage. Seven distinct fungal isolates, which caused these rots, were fully characterized. The species were *Aspergillus tamari*, *Fusarium solani*, *Lasiodiplodia theobromae, Aspergillus niger, Mucor circinelloides, Aspergillus flavus* and *Aspergillus* sp. In all the yams, storage rots reduced shelf life and aggravated weight loss. Post harvest storage losses varied among the different varieties of yams.
**Conclusion:** The varieties of yams studied suffer rots from various fungi, which are similar



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to those reported in other parts of the world. Severity of post harvest losses resulting from fungal rots varies among different varieties of yams. This should be taken into consideration in the development of storage techniques.

*Keywords: Dioscorea sp; Fungi; Storage rots; Post-harvest losses.*

## 1. INTRODUCTION

Yams are second to cassava as the most important tropical root crop and are a staple crop in many parts of Africa and Southeast Asia. In 2005, 48.7 million tones of yam were produced world wide. West and Central Africa account for about 94% of world production, Nigeria being the major producer [1]. Beyond their importance to the tropical and subtropical world, yams are becoming increasingly well-known in Europe, where in addition to imports, they have recently been cultivated experimentally in Greece [2]. In addition to their importance as food source, yams also play a significant role in the socio-cultural lives of many African societies. Thus, yams also serve as indices of wealth and essential components of many traditional rites [1].

Several methods have been developed for the storage of yams [3]. However, the traditional yam barn, in spite of its inadequacies, remains the most popular among farmers. As a consequence, post harvest storage losses remain an impediment in the production of this important crop. Losses during storage are known to be high and, depending on the species and the storage environment, may be of the order of 30-60% during the course of 3-6 months [4]. Amusa et al. [5] have estimated losses of up to 50% of the fresh matter. The causes of yam tuber loss during storage are associated with mechanical damage, probably caused by insects, nematodes and poor handling before, during and after harvest. Physiological changes within the tuber as well as storage rots due to microorganisms are also very important contributors. Yam tuber loss during storage manifests as weight loss, mechanical damage, discolorations and shriveling, making it unfit for consumption.

Fungi are the primary causal agents of storage rots, which contribute to post harvest storage losses of yam. Amusa and Baiyewu [6] have categorized storage rots of yams into three groups namely: dry rot, caused by several fungi including *Botryodiplodia theobromae, Aspergillus tamari* and *Penicillium oxalicum*; soft rot, usually involving *Rhizopus nigricans, Sclerotium rolfsii* and *Mucor circinelloides* and wet rot, which results from secondary infection by *Erwinia carotovora.* There are several reports in the literature about the fungal causal agents of yam rots [7-9]. However, yam varieties cultivated in different regions of the world vary among themselves. It is also known that different varieties of yams may vary in their intrinsic properties. The combined effects of intrinsic and extrinsic properties of foods determine the types of microorganisms that may spoil them. Data on causative agents of rots specific to different varieties will help improve techniques adopted for their storage.

The objectives of this work are to contribute to knowledge on storage rots with respect to yam varieties cultivated in South East Nigeria and then, to determine under conditions of experimental storage, the influence of fungal rot on post harvest storage losses in terms of weight loss and reduction of shelf life among these varieties.

## 2. MATERIALS AND METHODS

### 2.1 Yam Varieties

Five varieties of yams cultivated in the South Eastern parts of Nigeria were used in this study. The yams, their local names and a brief description of their characteristics are given below.

*Dioscorea dumentorum*, also called trifoliate yam (var. *ona*), bitter yam or cluster yam is considered inferior in quality and has not been studied extensively [10]. *Dioscorea alata* also called water yam, winged yam or greater yam is characterized by possession of a loose watery texture [11]. The interior of the water yam tuber is white or reddish, on the basis of which farmers differentiate them into two varieties, var. *abana ọcha* and var. *abana mmei* respectively. *Dioscorea rotundata* also called Guinea yam or white yam are species, which the interior of the tuber is white and firm in texture. Two varieties of white yam used in this study were var. *adaka*, which is rough-skinned and var. *abị* with a relatively smooth skin and containing a green pigment in its mesoderm [11].

The yam varieties were obtained from farmers in Agwbu, Orumba North Local Government of Anambra State, Nigeria about two weeks after their harvest. Yams were selected on the basis of their good health, absence of cuts on their skin and fair uniformity of weight. Each tuber was given a specific code for purposes of identification.

### 2.2 Storage Conditions

Fifty tubers of each variety of yam were stored in a partially shaded experimental yam barn consisting of separate wooden shelves [12]. The barn was constructed near our laboratories at the Department of Applied Microbiology & Brewing, Nnamdi Azikiwe University, Awka, Nigeria and employed for this study between January 2012 and July 2013. The barn was well ventilated and protected from insect pests using wire netting. Temperature and relative humidity of the barn were averages of 30ºC and 95% RH respectively, during the period of storage. Yams were de-sprouted on observation to minimize the effects of this process on deterioration of tubers.

### 2.3 Examination of Tubers for Rot

This was done by hand feel and visual examination, followed by examination of the interior of suggestive tubers on a bi-weekly basis. Rots were categorized using the scheme of Amusa et al. [5] as follows: Dry rot; the infected tissues became hard and dry with varying discolourations depending on the causative agent. Soft rot; the infected tissues became soft and ramified by the fungal mycelium. Wet rot; this was typified by the exudation of whitish fluid from the infected yam tissue when pressed between the fingers. All tubers, which were rotted were removed from the barn and kept separately.

### 2.4 Determination of Shelf Life of Yams

Yam tubers, which did not become susceptible to storage rot were deemed to have reached the end of their shelf lives when they became shriveled and cooked samples became unacceptable to tasters accustomed to yam meals using acceptance tests on the 7-point hedonic scale [13]. This type of shelf life was designated "Shelf life-Auto spoilage". Shelf life influenced by storage rots, termed “Shelf life-Fungal rot” was considered to have been attained for any variety when up to 10 out of 50 tubers stored showed obvious symptoms of storage rot. Results reported are means (weeks) of measurements for all rotted tubers. Reduction of shelf life (%) due to storage rots was calculated from the ratio of Shelf life-Fungal rot to Shelf life-Auto spoilage.

### 2.5 Isolation and Characterization of Storage Rot Fungi

A slight modification of the method described by Ogaraku and Usman [8] was used for this study. Small portions of the rotted yam tissues contiguous to healthy tissue were surface sterilized by cleaning with cotton wool soaked in 0.1% silver nitrate solution. The excess sterilant was then blotted out with cotton wool. With the aid of a sterile knife and forceps the interior of the rotted tuber portion was obtained and plated asceptically on Saboraud’s dextrose agar (SDA). All samples were plated in triplicate. Incubation was done at room temperature until there was visible growth. The isolates were further purified by repeated subculture on SDA plates and then subjected to pathogenicity tests (see below). Isolates confirmed to be pathogenic to yams were characterized by the slide culture technique and reference to Fungal Atlases [14,15] in our laboratory. The identities of isolates were confirmed by the Microbial Identification Service, CABI E-UK, Bakeham Lane, Egham, Surrey, TW20 9TY, England, UK. using the partial ITS rDNA sequencing analysis and a BLAST search using the GenBank sequence database.

### 2.6 Pathogenicity Test

Pathogenicity test was carried out by inoculating healthy yams with pure cultures of each fungal isolate according to the method described by Okafor [16]. An isolate was confirmed pathogenic if it caused rot similar to that observed on the diseased yam from where it was isolated.

### 2.7 Determination of Weight Loss

Weight loss (%) was calculated from a comparison of the weight of individual yam samples at the commencement and at the end of storage. End of storage was defined by either type of shelf life as appropriate.  Results reported are means of measurements from ten individual tubers.

### 2.8 Statistical Analyses

The completely randomized experimental design and appropriate replicates were adopted in all measurements. Data were subjected to Analyses of Variance (ANOVA).

## 3. RESULTS AND DISCUSSION

### 3.1 Characterization and Pathogenicity of Storage Rot Fungi

Twelve distinct fungal colony types were isolated from the yam samples and seven (58.3 %) of these considered pathogenic to the yams were fully characterized. The report of molecular identification by CABI E-UK showed that five of the pathogenic species, *Aspergillus tamari*, *Fusarium solani*, *Lasiodiplodia theobromae, Aspergillus niger* and *Mucor circinelloides* showed 100% identity to multiple ITS sequences reported from their respective species using the GenBank sequence database. *Aspergillus flavus* shared 100% identity to multiple ITS sequences also reported from *Aspergillus oryzae.* This isolate was distinguished from *A. oryzae* on the basis of its smaller conidia (up to 6µm, whereas those of *A. oryzae* are larger, up to 8µm). The seventh isolate *Aspergillus* sp., was identified as a member of the genus *Aspergillus* from examination of morphology because the sample consistently failed to amplify probably due to impurity in our original sample.

### 3.2 Fungal Rot of Tubers

All the varieties of yams studied suffered fungal rots, which were caused by the different fungal isolates as shown in Table 1. The storage rot fungi isolated during this study are the same with those reported previously [7-9]. It is interesting to note that the types of rots caused by our isolates are in complete agreement with the categorization of Amusa and Baiyewu [6]. Dry rot is the predominant category of rot observed during this study, while wet rot was not observed at all. Wet rot arises from secondary infection with *Erwinia carotovora* [6], chances of which may have been very low in our experimental barn. Ogaraku and Usman [8] also reported the predominance of dry rot (54.2%) over soft rot (36.1%) and a low incidence (9.6%) of wet rot in similar studies on yams collected from farmers barns.

**Table 1. Categories of rots observed on the yam varieties during storage and their fungal causative agents**

| Yam variety | Category of rot | Isolate |
|---|---|---|
| *D. dumetorum* var. *ona* | Dry rot | *Aspergillus tamarii* |
| *D. alata* var. *abana mmei* | Dry rot | *Fusarium solani* |
| | Dry rot | *Aspergillus* sp. |
| *D. alata* var. *abana ocha* | Dry rot | *Lasiodiplodia theobromae* |
| | Dry rot | *Aspergillus tamarii* |
| *D. rotundata* var. *adaka* | Dry rot | *Aspergillus niger* |
| | Dry rot | *Aspergillus flavus* |
| *D. rotundata* var. *abi* | Soft rot | *Mucor circinelloides* |

Besides the causation of rots, the presence of isolates such as *A. flavus*, many strains of which produce both B and G aflatoxins, and *A. tamarrii*, which produces the mycotoxin cyclopiazonic acid, may pose health risks to handlers and consumers of yams [17].

### 3.3 Effect of Fungal Rots on Shelf Life of Yams

Shelf life is the length of time that a commodity may be stored without becoming unfit for use or consumption (Oxford English Dictionary, 2nd ed.). Table 2 reveals that the shelf life of yams studied varied significantly ($P$= 0.05) according to their varieties, even in the absence of fungal rots. This is consistent with the report of FAO [3]. Storage rots drastically reduced shelf life in all the yams but at significantly ($P$= 0.05) different rates in different varieties. The *D. dumentorum* and *D. alata* species, particularly var. *abana mmei* suffered the highest rate of reduction in their shelf lives due to fungal rots. *Dioscorea alata* varieties of yams are usually characterized by high moisture composition and a loose watery texture while the *D. rotundata* species have lower moisture contents and are firm in texture [11]. The higher moisture content and watery texture of the tissues of *D. alata* may have enhanced fungal growth and hence, the severity of accompanying rots. The mechanism of fungal rot development involves the production of a variety of extracellular enzymes and a host of metabolites by which they degrade cell wall polymers, resulting in maceration of parenchymatous tissues [18].

## 3.4 Effect of Fungal Rots on Weight Loss

It is well known that yams undergo weight loss during storage. This arises from water loss by transpiration and loss of dry matter through the physiological changes of respiration and sprouting [19]. The overall effects of these changes are the shriveling, as well as loss of the food quality of the tuber. Different varieties of yams suffered significantly (*P*= 0.05) different rates of weight loss during storage (Table 3). Among tubers which did not suffer fungal storage rot, *D. dumentorum* (var. *ona*) lost the greatest while *D. rotundata* (var. *abi*) the least weight at the end of storage. Fungal rots aggravated weight loss in all the yams with *D. dumentorum* (var. *ona*) and *Dioscorea alata* (var. *abana mmei*), suffering significant effects (*P*= 0.1). These differences are perhaps attributable to the differences in moisture content and texture of the tissues of the different varieties. Furthermore, major differences occur in the alteration of cell wall carbohydrates in *D. dumentorum* compared with *Dioscorea rotundata* during storage. The *D. dumentorum* tuber undergoes hardening during storage, characterized by the deposition in its cell walls, of a xylose-containing polymer and additional cellulose and by the lignification of the tubers [20].

**Table 2. Mean shelf life of yams in the absence and under the influence of fungal rot**

| | ***Dioscorea umentorum* (var. *ona*)** | ***Dioscorea alata* (var. *abana mmei*)** | ***Dioscorea alata* (var. *abana ocha*)** | ***Dioscorea otundata* (var. *adaka*)** | ***Dioscorea rotundata* (var. *abi*)** |
|---|---|---|---|---|---|
| Shelf life-Auto spoilage (weeks) | 31.12± 1.35 $^{a\,\alpha}$ | 26.83 ± 1.74$^{b\,\alpha}$ | 31.64±1.28 $^{a\,\alpha}$ | 35.93 ±1.24 $^{c\,\alpha}$ | 39.73 ±1.34 $^{d\,\alpha}$ |
| Shelf life-Fungal rot (weeks) | 11.30±1.0 $^{a\,\beta}$ | 9.00 ± 1.3 $^{b\,\beta}$ | 11.00 ± 1.7 $^{a\,\beta}$ | 14.8 ± 1.3 $^{c\,\beta}$ | 16.42 ± 0.1 $^{d\,\beta}$ |
| % Reduction in shelf life | 63.7 | 66.5 | 63.4 | 58.8 | 58.7 |

*Means on the same row (a – d) and column (α & β) with different suffixes are significantly different (Tukey HSD Test), at P = 0.05.*

**Table 3. Mean percentage weight loss in yam tubers at the end of their shelf lives**

| | ***Dioscorea dumentorum* (var. *ona*)** | ***Dioscorea alata* (var. *abana mmei*)** | ***Dioscorea alata* (var. *abana ocha*)** | ***Dioscorea rotundata* (var. *adaka*)** | ***Dioscorea rotundata* (var. *abi*)** |
|---|---|---|---|---|---|
| Tubers, which did not suffer fungal storage rot (%) | 52.09 ± 13.94$^{a\,\alpha}$ | 40.97 ± 4.29$^{b\,\alpha}$ | 39.30 ± 3.84 $^{b\,\alpha}$ | 43.04 ± 4.13 $^{c\,\alpha}$ | 35.60 ± 6.73 $^{d\,\alpha}$ |
| Tubers, which suffered fungal storage rot (%) | 64.38 ± 6.19$^{a\,\beta}$ | 56.00 ± 8.21$^{b\,\beta}$ | 42.24 ± 3.55$^{c\,\alpha}$ | 44.28 ± 4.96$^{c\,\alpha}$ | 42.49 ± 17.05$^{c\,\alpha}$ |

*Means on the same row with different suffixes are significantly different (Tukey HSD Test), at P= 0.05*
*Means on the same column with different suffixes are significantly different (Tukey HSD Test), at P= 0.1*

## 4. CONCLUSION

The varieties of yams cultivated in South Eastern Nigeria suffer rots from various fungi, which are similar to those reported in other parts of the world. Fungal rots exacerbate post harvest storage losses in yams by reducing their shelf lives and accelerating weight loss. Severity of post harvest loss appears to vary among the different varieties of yams studied. Data from this work suggests that *D. rotundata* (var. *abi*) would store better than all the other varieties studied. Development of storage techniques should in the future take the chemical and physical differences among yam varieties into consideration.

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## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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