

POST HARVEST FUNGAL DETERIORATION OF SWEET POTATO (*Ipomoea batatas*(L)Lam) ELITE VARIETIES IN SOUTH EASTERN NIGERIA

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ABSTRACT: Assessments of the pathogenicity of the postharvest spoilage of tubers of elite cultivars of sweet potato (*Ipomoea batatas* (L)Lam) from different locations in the South eastern Nigeria agroecological zone were carried out between October 2020 – March 2021. Isolated fungi include *Botryodiplodia theobromae*, *Rhizopus stolonifer*, *Fusarium oxysporum*, *Penicillium citrinum*, *Aspergillus niger*, and *Trichoderma viride*. All the isolates proved pathogenic except *T. viride*. Of all the isolated fungal pathogens, *B. theobromae* had the highest percentage occurrence of 36.07% followed by *R. stolonifer* at 29.02%. Percentage occurrence varied ($P < 0.05$) significantly within the locations surveyed. The more humid location - Umudike (Abia State) indicated higher incidence while the drier location – Ezillo (Ebonyi State) recorded lower fungal growth and disease incidence. The local sweet potato variety (Ex-Igbariam), gave consistently the highest disease severities when inoculated with the three major pathogens – *B. theobromae*, *F. oxysporum*, and *R. stolonifer*. The trend of susceptibility of the cultivars was Ex-Igbariam > UMUSP/2 > UMUSPO/3. The deep orange fleshed cultivar (UMUSPO/3) showed the least mean internal lesion diameter (MILD) of lengths: 10.20mm, 8.60mm, 10.20mm and depths 5.20mm, 5.50mm, 6.20mm on *B. theobromae*, *F. oxysporum*, *R.*

stolonifer respectively. Further research is needed to explore opportunities of scaling up selection of sweet potato lines with vitamin B6 carotenoids ready to provide disease resistance in stored sweet potato in the Southeastern Nigeria.

Keywords: *Pathogens, disease severity, percentage occurrence, mean internal lesion diameter, orange fleshed, sweet potato.*

INTRODUCTION

Sweet potato (*Ipomoea batatas* (L) Lam), (Convolvulaceae) is one of the world most important food crops -the 7th after wheat, rice, maize, potato, barley and cassava (Dayal and Sharma 1991; Ray and Ravi 2005; FAO 2016). The world annual production of sweet potato stands at 106,569 million tons covering about 8.0 million hectares of arable land area. Nigeria is the second highest producer of sweet potato with 3.49 million tons after China at 102.44 million tons (Egeonu and Akoroda 2010). Presently due to the release and adoption of improved varieties of sweet potato, average yield could get up to higher than 5t/ha. The storage roots and leaves are edible and so can be used as staple foods, animal feed as well as industrial raw material for starch production etc (Bevell- Benjamin 2007). As food, it's a source of important nutrients such as potassium, calcium, iron, vitamins and minerals. Sweet potato is a rich source of beta carotene and anthocyanin which are important natural health promoting compound (Paul et al. 2021, Onwueme and Charles 1994). As a crop with medicinal value, it has anticancer, antidiabetic, antiinflammatory properties (Afuape 2014). Sweet potato roots have short storage shelf life of less than 4 weeks in the tropics a major challenge in the production and utilisation of this crop. The high moisture content of the organ in the high temperatures of the tropics predisposes the storage organ to the pathological and physiological deterioration after harvest (Ray and Ravi 2005; Arinze and Smith 1982). Because the periderm of the root tubers is thin, it increases the disease vulnerability to numerous fungal pathogens leading to many diseases in storage e.g dry rot, soft rot, wet rot, Java black rot, blue mold etc (Onuegbu 2002; Chandrasokaran and Kumar 2016). Commonly associated fungal pathogens include *Aspergillus* spp, *Penicillium*, *Rhizopus* spp, *Fusarium* spp, and even *Botryodiplodia theobromae* (Ray and Pati 2001., Rees et al. 2003, Clark and Moyer, 2013). Some of these pathogens enter the storage organs from the farm

as crop residues and infected soils while others enter through wounds created during harvest, impact during transportation, processing and storage (Tomlins et al. 2000). Management of postharvest/storage diseases of sweet potato includes the use of fungicides as well as cultural methods (Ray and Pati, 2001, Onuegbu, 2002). Breeding for resistance to field diseases of sweet potato varieties has led to the release and adoption of some improved varieties of sweet potato in Nigeria. Cultivars with high yielding, disease resistant and possession of desirable organoleptic properties are widely gaining acceptance among farmers and consumers. A typical example is the orange fleshed sweet potatoes (OFSP) which are nutritionally superior to the white fleshed varieties currently in use in most parts of Nigeria. The orange fleshed varieties have the potential to prevent vitamin A deficiency due to their marginal higher beta carotene contents (Afuape 2014, Low et al. 2007, Agble 2004). The aims of this study therefore were to:-

- i. investigate the possible causative agents of postharvest fungal spoilage of tubers of some of these elite cultivars in Southeastern Nigeria.
- ii. ascertain their pathogenicities and disease severities at different locations in South eastern Nigeria.
- iii. make recommendations necessary for improvement on postharvest spoilage resistance so as to enhance sweet potato value chain.

MATERIALS AND METHODS

Sweet potato survey locations in South eastern Nigeria.

The three (3) locations in South eastern Nigeria include:

- a) Umudike (Latitude $5^{\circ} 28' 33''$ N and Longitude $7^{\circ} 32' 56''$ E, Mean annual rainfall 1800mm-2400mm, vegetation of humid tropical rainforest. In semi urban area in Ikwuano LGA of Abia State.
- b) Enugu (Lat $6^{\circ} 21' N$ and Long $7^{\circ} 26' E$, lies between two vegetation zones of the Tropical Rainforest and the tall woodland and tall grass Savanna (Derived

Savanna), Poor soil fertility. The Urban administrative Capital of Eastern Region and present Enugu State.

- c) Ezillo (Lat $6^{\circ} 25' 38''$ and Long $7^{\circ} 50' 9''$, Mean annual Rainfall of 1500mm, Temperature 27° c, vegetation is Derived Savanna. Ezillo is purely an agrarian community and headquarter of Ishielu LGA of Ebonyi State.

SWEET POTATO CULTIVARS

Sweet potato cultivar roots assayed as described in Afuafe, (2014)

- a) UMUSPO/1 – Light orange fleshed sweet potato, widely adapted across agroecologies from the humid forest in southern Nigeria to the Northern Guinea Savanna, resistant to sweet potato virus disease (SPVD) tolerant to *Cylas* spp weevil, high yielding, high dry matter (30-32%) total carotene 7.12 ug/g FW, root shape is long elliptic.
- b) UMUSPO/3 (CIP 440293)- deep orange fleshed, widely adapted but better in low SPVD pressure ecologies of southern Guinea Savanna to the Northern Sudan Savanna, high yielding high carotenoids (66.3 ug/g FW), shape round elliptic.
- c) UMUSP/2 – white fleshed sweet potato, broadly adapted from the humid forest to the Northern Guinea Savanna high yield, moderate dry matter (<30%), low carotene (2.41ug/g FW) shape=ovate.
- d) Ex-IGBARIAM(local cultivar)-yellow root fleshed, adapted to all agroecologies, high dry matter, moderate resistance to SPVD, big tubers and vigorous.

Sampling of sweet potato tubers

Five equal sized (1kg) of the four sweet potato cultivars – UMUSPO/1, UMUSPO/3, UMUSP/2 and Ex-IGBARIAM identifiable by their skin colour and shape were randomly selected and bought from various market stalls at the various locations described above. Diseased samples were put separately inside sterile polythene bags and brought to the Plant Pathology Laboratory of the National Root Crop Research

Institute Umudike for further analysis. These were temporarily preserved in the refrigerator at 5⁰c before use.

Isolation and identification of associated spoilage fungi of sweet potato

The sweet potato samples were first washed under running tap water to remove surface dirt and enable clearer view of disease symptoms. Various observed disease symptoms were identified, described and classified. The roots were surface sterilized using sterile non absorbent cotton wool soaked in 70% ethanol and left to dry with a flamed sterile scalpel, 3-4 pieces about 5cm long were cut at the intersection of the diseased and healthy portions of the root samples. The root pieces were later immersed in a solution of sodium hypochlorite (0.5%) for one minute before being washed in three changes of sterile distilled water and air dried. Each root piece was plated onto Potato Dextrose Agar in a Petri dish and incubation was at 28+₂⁰c for 48 hours and examined for fungal growth. Re-isolation of grown fungi onto freshly prepared sterile PDA plate and incubated at 28+₂⁰c for 3 days were carried out. The cultural, macroscopic and microscopic characteristics of the fungal isolates were recorded and described by making reference to standard laboratory manuals (Barnett and Hunter, 1998).

Pathogenicity test

Out of the fungal isolates from each location, representative identified fungi were used in performing the pathogenicity test. Samples of each of the 3 cultivars of sweet potato storage roots from the National Root Crops Research Institute, Umudike barn were washed under running tap water before being surface sterilized by dipping in 1% sodium hypochlorite solution for 5 minutes, washed in three changes of sterilized distilled water and dried in a laminar airflow chamber. A 5mm sterile cork borer was used to collect 5mm plug of hyphal disc of each isolate and placed in a wound aseptically made in each root using a 5mm cork borer. A sterile blank 5mm PDA discs were used as control treatments. After inoculation, each treated root was sealed with sterilized paraffin wax to prevent microbial contamination. The storage roots were then kept in moistened clean boxes of about 70% RH and temperature of 28+₂⁰c in the laboratory. After one week of inoculation, the artificially inoculated

organs were observed daily for lesion development re-isolation of grown fungi or the diseased sweet potato and initiation of similar disease symptoms according to Koch postulate would confirm pathogenicity.

Measurement of disease development severity

Measurement of disease development on the sweet potato was carried out daily for two weeks from 48 hours after inoculation. To do this each inoculated potato root tuber was removed from the inoculation bag, cut open longitudinally with a sharp knife through the inoculation wound. The diameter and depth of the developing internal lesion, shown by the extent of the root tissue degradation, was measured in millimeter using the meter rule. The Mean of the diameter and depth was calculated to give the Internal Lesion Dimension (MILD) as in Daurte and Clarke (1993).

This experiment was set in a completely randomized design (CRD) replicated thrice.

DATA ANALYSIS

Data from results collected were subjected to analysis of variance (ANOVA) using Genstat Release Software 10.3 Discovery Edition. With Fischers Least Significant Difference (F-LSD) treatment means were compared where f-test was significant at 5.00% probability level.

RESULTS

Isolation and Identification of associated fungi

The list of isolated fungi and their percentage frequencies across the locations from major cultivars is shown in Tables I, ii, & iii. The fungi include *Botrydiplochia theobromae*, *Rhizopus stolonifer*, *Penicillium citrium*, *Fusarium oxysporium*, *Aspergillus niger* and *Trichoderma viride*. All cultivars showed disease symptoms such as rot with UMUSPO/3 having the least number of fungal isolates viz: 86 at Umudike, 58 at Enugu, and 47 at Ezillo. The local sweet potato (Ex-IGBARIAM) gave the highest yield of associated fungal isolates at 126 at Umudike, 84 at Enugu and 54 from Ezillo. Of the fungi, *B. theobromae* constantly gave the highest percentage of occurrence at the three locations and from each cultivar (36.07%,

35.60%, and 33.16%), seconded by *Rhizopus stolonifer* (25.65%, 29.02% and 23.47%) while *Trichoderma viride* was the least having just 2.33% and 1.53% occurrence on UMUSP/2 and Ex-Igbariam respectively.

Pathogenicity test.

Of all the six (6) different fungal isolates from the sweet potato cultivars, five (5) were pathogenic following the protocols of Koch postulate. The result is shown in Table IV. The organism *T.viride* did not initiate disease symptoms when inoculated on sweet potato cultivars, hence non-pathogenic.

Measurement of disease development severity

The results of internal lesion development by the three key fungal pathogens on the two elite sweet potato cultivars and a local check are shown in Table 4. Their effects as assayed from the disease lengths and depths two weeks after inoculation of sweet potato roots are given. There was significant ($P < 0.05$) difference among the diameter of fungal infections of the sweet potato cultivars assayed. The local variety (Ex Igbariam) gave the highest mean length and depth values of 17.53mm and 7.23mm respectively followed immediately by UMUSP/2 with 16.29mm and 6.47mm while the deep orange variety gave the least values of 9.57mm and 5.66mm length and depth respectively. Significant ($P < 0.05$) difference occurred among the various fungal infectivity values with *R. stolonifer* showing the highest mean internal lesion in length of 15.22mm followed by *B. theobromae* 14.61mm and *F. oxysporum* at 13.56mm. However in depth, *B. theobromae* was highest with 7.05mm, *F. oxysporum* at 6.64mm before *R. stolonifer* with 6.17mm. In the interaction effect, the *B. theobromae* on Ex Igbariam pathosystem gave the highest length and depth values of 18.31mm and 8.60mm followed by *R. stolonifer* on Ex Igbariam while the least lesion diameter values were shown in *F. oxysporum* on UMUSPO/3 pathosystem with 8.47mm and 5.48mm length and depth respectively.

DISCUSSION

The occurrence of six different fungal isolates on postharvest sweet potato tubers in south eastern Nigeria is quite significant thus making sweet potato production in this

area challenging. Out of the six fungi, five of them were found pathogenic on the tested sweet potato cultivars while one (*T. viride*) was not. The significant difference ($P < 0.05$) in the number of isolates from the various locations could be taken to be the influence of varying environmental conditions during post-harvest storage. Umudike in the humid rainforest zone may possibly have encouraged the highest microbial infections thus higher disease incidence compared to drier Enugu and Ezillo locations. Ezillo in the dry Derived Savanna may provide the right condition necessary for storage roots curing effects immediately after harvest. This similar result was found out at both tropical and temperate study locations by other researchers. Paul et al (2021) isolated a total of 68 fungal isolates from three locations in South Korea, Pati and Ray (2000), Ray and Ravi (2014) in India, and Rees et al (2003) in East Africa. Arinze and Smith (1982), Arinze and Nwankiti (1978) have also reported the presence of some of the fungal causative agents such as *B. theobromae*, *F. oxysporum* as also found out in this investigation as responsible for the post harvest spoilage of sweet potato. Onuegbu (2002) reported *Penicillium* spp, *A. niger*, *A. flavus* as among other fungi responsible for the post harvest decay of sweet potato. Ray and Ravi (2005) observed that *B. theobromae*, *Ceratocystis fimbriata*, *Fusarium* spp and *R. oryzae* as the most important fungal isolates. According to Ray and Ravi (2014), wounding is the most important predisposing factor for *B. theobromae* and *Fusarium* infections. The physiology of postharvest deterioration of sweet potato by fungal organisms such as *R. stolonifer* and *B. theobromae* as reported by Samsatley et al (2018) occur by fungal hyphae from the host epidermis penetrating the inter and intracellular tissues. By colonizing the host tissues, its breakdown by means of hydrolytic enzymes leads to browning and necrosis to ultimate tissue death. Optimum temperature ($25-35^{\circ}\text{C}$) and relative humidity (85-95%) are the enabling environmental factors for fungal growth and pathogenesis. The region of South eastern Nigeria favoring the production of sweet potato is unfortunately unfavorable for the post harvest storage of the tubers due to bacterial and fungal deterioration. Sweet potato is reputed for high moisture content (60-70%), thin and delicate skin, high dry matter including the elite varieties (Afuape 2014). The high internal lesion diameter of the sweet potato cultivars both local and

elite varieties on infection with the storage fungi during this trial shows that they are susceptible to postharvest fungal attacks. However, infection levels differed. There was an indication of a probable resistance/ tolerance by the orange fleshed varieties of UMUSPO/3 and UMUSPO/1. There was comparatively lesser mean internal lesion diameter among the deep orange fleshed sweet potato roots (more resistant) than other cultivars and the trend is in this order UMUSPO/3 (deep orange fleshed) <UMUSPO/1 (light orange fleshed)<UMUSP/2 (white fleshed)< Ex-Igbariam (local check). This trend agrees with those of other tropical studies such as Rees et al (1998) in India and Van Qirschot (2000) in Kenya, East Africa where orange fleshed color was associated with pathogen resistance. The observed varietal differences in this investigation could be dependent on the amount of available carotenoids and anthocyanins as reported by Onwueme and Charles (1994). The production of Vitamin B6 in orange fleshed sweet potato roots has been opined as having a dual role as a micro nutrient and stress protectant such as fungal infection (Samsetly et al 2018, Vanderscuren et al 2013). These antioxidants have been said to stop the production of reactive oxygen species (ROS) among necrotropic fungi. Uritani (1998) has also identified the presence of Ipomeamarone _ a major furansterpenoid compound induced by microbial infection such as *B. theobromae*, *Rhizopus* spp as likely implicated in post harvest disease resistance. Selecting sweet potato cultivars with further qualities like low furanoterpene inducing potential is as well as higher carotenoids and anthocyanins is hereby recognized. Also the inclusion of breeding sweet potatoes with much thicker and tougher skins able to resist mechanical damage and fungal penetration is highlighted as plausible sources of resistance of postharvest disease fungal deterioration.

CONCLUSION AND RECOMMENDATION

The potentials of sweet potato production as cheap but nutritionally rich staple food and industrial raw material make it a veritable tool for ensuring food security. The elite cultivars being used by farmers in South eastern Nigeria are resistant to major field diseases but susceptible to post harvest spoilage. Because of the high susceptibility to fungal infection and poor storability farmers sell off their produce so

early resulting in low profit and food insecurity especially in the lean season. More efforts are needed towards scaling up selection and production of more marketable quality sweet potato varieties resistant to these postharvest fungal pathogens.

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TABLE 1:Percentage occurrence of fungi isolated from post harvest sweet potato cultivar (UMUSP/2) in three locations of South eastern Nigeria between October 2020 - March 2021

Location	Number of isolated fungi					
	B.T*	R.S*	F.O*	P.C*	A.S*	T. V*
Umudike	30	20	17	10	10	3
Enugu	20	14	14	8	6	0
Ezillo	15	12	8	6	3	0
Total No. of Fungi	65	46	39	34	19	3
Grand Total	196					
Percentage	33.16	23.47	19.90	12.24	9.70	1.53

B.T* =Botryodiplodia theobromae, R.S* =Rhizopus stolonifer, F.O* = Fusarium ysporium,
P.C*=Pencillium citrinum, A.N* =Aspergillus niger, T.V* =Trichoderma viride

TABLE 2: Percentage occurrence of fungi isolated from post harvest sweet potato cultivar (UMUSPO/3) in three locations of South eastern Nigeria between October 2020 - March 2021

Location	Number of isolated fungi					
	B.T*	R.S*	F.O*	P.C*	A.N*	T.V*
Umudike	30	25	15	8	8	0
Enugu	20	14	10	8	6	0
Ezillo	18	10	10	6	3	0
Total No. of Fungi	68	49	35	22	17	0
	191					
Percentage	35.60	25.65	18.32	11.51	8.90	0

Occurrence (%)

B.T* =Botryodiplodia theobromae, R.S* =Rhizopus stolonifer, F.O* = Fusarium oxysporium, P.C*
=Pencillium citrinum, A.N* =Aspergillus niger, T.V* =Trichoderma viride

TABLE 3: Percentage occurrence of fungi isolated from post harvest sweet potato cultivar (Ex-Igbariam) in three locations of South eastern Nigeria between October 2020 - March 2021

Fungal Isolation in three locations of South eastern Nigeria between October 2020 – March 2021								
Location			Number of isolated fungi					
B.T*	R.S*	F.O*	P.C*	A.N*	T.V*			
Umudike			40	36	24	10	10	6
Enugu			32	26	12	6	8	0
Ezillo			20	12	10	8	4	0
Total No. of Fungi			92	74	37	24	22	6
Grand Total			255					
Percentage Occurrence (%)			36.07	29.02	14.51	9.41	8.63	2.35

B.T* =Botryodiplodia theobromae, R.S* =Rhizopus stolonifer, F.O* = Fusarium oxysporium, P.C* =Pencillium citrinum, A.N* =Aspergillus niger, T.V* =Trichoderma viride

TABLE 4: Effect of three postharvest spoilage fungi on the mean internal lesion diameter (MILD), two weeks after inoculation of sweet potato cultivars

1 s				
B.theobromae	14.60889a	3.674954	7.046667a	1.4713514
F.oxysporum	13.55556b	3.824955	6.643333b	1.2309651
R.stolonifer	15.21556c	3.74901	6.171111c	0.2805104
Ex-Igbariam	17.525556a	1.0410705	7.7a	1.0196936
UMUSP/2	16.285556b	0.8652328	6.465556b	0.6732776
UMUSPO/3	9.568889c	0.8430368	5.655556c	0.4619554