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By Akwaji, Patrick Ishoro, Okon, Ekeng Ita, Markson, Aniedi-Abasi Akpan & Iso, Usang Ekong
University of Calabar, Nigeria

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Abstract- This study was carried out to investigate the fungi associated with pre-harvest fruit rot of eggplant *(Solanummelongena* L.*)*, their effect on fruit nutritional content and their control using fruit extracts of *Tetrapleuratetrapterain-vitro*. The fungal pathogens isolated as the causative agents of fruit rot in this study were *Phomopsismelongenae* and *Collectotrichummelongenae.* The result of proximate analysis of fungal infected and non-infected eggplant carried out showed that there was an increase in the moisture and protein content of the fungal infected eggplant as compared to healthy ones (control), while there was a decrease in the crude fibre, fat, ash, and carbohydrate contents of the fungal infected eggplant fruits as compared to the healthy ones (control). It was observed that moisture content increased from 42.90±0.12 in the non-infected eggplant fruit to 58.03±0.20 in the eggplant fruit infected with *Phomopsismelongenae* and 42.90±0.12 in the non-infected eggplant to 60.12±0.40 when infected with *Collectotrichummelongenae* while the following parameters viz; protein 5.60±0.10, fat 5.37±0.17, crude fibre5.12±0.09, carbohydrate 3.64±0.19 and ash content 0.50±0.15 of non-infected eggplant were depleted when infected with *Phomopsismelongenae* to 1.40±0.18, 5.60±0.10, 2.98±0.27, 5.37±0.17, and0.25± 0.16 and protein 1.60±1.11, fat 3.0±0.31, crude fibre 3.30±0.7, carbohydrate 1.09±1.21, and ash content 0.30±0.21 of non-infected eggplant were depleted to 5.60±0.10, 5.37±0.17,5.12 ±0.09, 3.64±0.19 respectively when infected with C*ollectotrichum-melongenae.* Results of the *in vitro* antifungal assay carried out showed that the ethanolic extracts of *Tetrapleuratetraptera* had a significant effect ($p<0.05$) in inhibiting the redial growth of the fungal pathogens at the different concentrations (5g/100ml, 10g/100ml and 15g/100ml) tested than the aqueous extracts. Phytochemical test carried out showed that tannin, flavonoid, saponin and cyanogenic glycosides were present in aqueous extract while alkaloids, steroids, triterpens, tannin, flavonoid and saponin were present in the ethanolic extracts.

I. Introduction

Eggplant (*Solanummelongena*L.), family *Solanaceae* is a popular vegetable also known as African eggplant which contains numerous small soft seeds which although edible, taste better because, the plant related to tobacco, and contains nicotinoids and alkaloids and commonly served and eaten as desert mostly with groundnut in this part of the world (Giuliani and Smale, 2000).

It is one of the top ten vegetables in the world and it is grown on more than two million hectare with a production of nearly thirty three million tones (FAO, 2007). The plant had been cultivated in India for the past four thousand years and is one of the most important vegetable of *Solanaceae* family. The global area under eggplant cultivation has been estimated to be at 1.85million hectare with total production of about 32 million metric tons, it is grown on nearly 550,000 hectares in India, making the country as the second largest producers after China. India accounts for about 8.7 million metric tons with an area of about 0.53 million hectares under cultivation (Anon, 1998, Sidhu, 1998).

The northern and southern parts of Nigeria are involved in the cultivation of eggplant which comes in different varieties and they vary in fruit colour, shapes and sizes (Chinedu *et al.,* 2011).

Egg plant is well adapted to high rainfall and high temperatures, and is among the few vegetables capable of high yields in hot- wet environments (Hanson *et al*., 2006). Eggplant contains nutrients such as dietary fiber, folate, ascorbic acids, vitamin K, niacin, vitamin B_6, pantothenic acids, potassium, iron, magnesium, manganese, phosphorus and copper (USDA, 2009); and the nutrients contribute to the diet of the poor and mostly important during time when other vegetables are in short supply.

Eggplant is a delicate tropical perennial, often cultivated as a tender or half-hardy annual in temperate climates (Grubben and Denton, 2004). It grows 40 – 150 cm tall, with large coarsely lobed leaves that are 10 – 20 cm long and 5 – 10 cm broad (Grubben and Denton, 2004.) semi wide types can grow much larger, up to 255 cm with larger leaves, over 30 cm long and 15 cm broad, the stem is often spiny and the flowers are white to purple with a five lobed corolla, yellow stamens and the egg shaped glossy purple fruit has white fleshy with a meaty texture, and the cut surface of the flesh rapidly turns brown when the fruit is cut open (Hanson *et al*., 2006).

Author α σ ρ ω: Department of Botany, University of Calabar, Calabar, Cross River State, Nigeria. e-mail: akwajiisnever@yahoo.com

Eggplant fruit and leaves are both eaten as vegetables or used in traditional medicine (Bonsu*et al.,* 2008). Eggplants are highly valued constituents of Nigerian foods and indigenous medicine that are either eaten raw or cooked. Also very popular in rice dishes such as stew and soup of different kind, and also prepared as sauces that are consumed with yam and plantain (Edem*etal*., 2009).

In most advanced countries, such as China, India, Philippines eggplant has been used in their indigenous medicine, the medicinal uses ranges from weight reduction to treatment of several ailments which include asthma, constipation and skin infections, diabetes, leprosy, gonorrhea, dysuria, dysentery, asthenia and hemorrhoids (Gill, 1992).

Diseases and fungal pathogens of eggplant include damping-off caused by (*Pythiumspp*, *Phytophthoraspp*: and *Fusariumspp*) root rot (*Rhizoctoniaspp* and *Sclerotiumspp*). Blight (*Phomopsis-spp*); fruit rot (*Phomopsisvexans* and *Rhizopusstolonifor*) and wilt (*Verticillumspp*; *Fusariumspp*) are noteworthy and take a considerable proportion of the produce annually. Eggplant wilt complex is known to be caused by number of fungi genera such as *Fusarium*, *Verticillum, Rhizoctonia*, Sclerotium and Phytophthora in different parts of the world (Rangas-wami 2000).

In view of the adverse effect of fungi infection on eggplant fruits as observed in the University of Calabar Farm, University of Calabar, Calabar, Cross River State, Nigeria and environs it became necessary to isolate and identify the fungal pathogens associated with the eggplant fruits in the field, determine the effect of the pathogens on the nutritional content of the eggplant fruits through proximate analysis as well as evaluate the phytochemical and antifungal effect of *Tetrapleura-tetraptera* fruit extracts on the isolated fungi *in vitro*.

II. Materials and Methods

a) Collection of Samples

Healthy and infected eggplant fruits were obtained from the University of Calabar Farm, University of Calabar, Calabar Cross Rivers State, Nigeria. Proximate (nutrient) analysis of infected and non-infected eggplant fruits was carried out in the Department of Biochemistry, University of Calabar, Calabar, Nigeria.

b) Source of fungal pathogens and morphological identification

The fungal pathogens used in this research work were isolated from diseased eggplant fruits collected from the University of Calabar Farm, Calabar, Cross River State, Nigeria. Cut sections of the diseased assay fruits were surface sterilized with 70% sodium hypochlorite (bleach) solution for 1min and rinsed quickly in 3 changes of sterile distilled water, blotted dry on Whatman's No. 1filter paper and placed on Potato Dextrose Agar (PDA) in Petri dishes. Four (4) sections were inoculated per Petri dish. The plates were incubated at 28 ± 1°C until fungal growth was noticed. After 5 days, the different isolates were sub-cultured on freshly prepared PDA to obtain their pure culture. Isolated fungi were microscopically (Olympus optical, Phillipines) identified as far as possible using the identification guides of the International Mycological Institute, Kew and of Barnett and Hunter (1998), Alexopolous and Mins (1989).

c) Pathogenicity Test

Pathogenicity tests were carried out using the techniques of Okigbo and Nmeka,(2005). Healthy eggplant fruits were washed in distilled water and surface sterilized with 1% Sodium hypochlorite solution. A5mm diameter cork borer was used to cut discs from the fruits (three discs per fruit) and cultures of the isolated discs were introduced into holes and replaced with the discs. They were kept for 24-48hours. The inoculated fruits established symptoms on the second day and tissue segments from the infected fruits were cultured.

d) Effect of Fungal Infection on Proximate Composition of Eggplant fruit

To ascertain the effects of fungal infection on nutritional composition of egg plants, matured eggplant fruits showing signs of fungal infection were obtained and the moisture content of the eggplant fruits were determined. The plant fruit were then oven dried at 60°C for 24 hrs and grounded into fine powder using mortar and pestle. The powdered samples were stored in plastic container for laboratory analysis. Most of the methods adopted in this research work were those recommended by Association of Official Analytical Chemist (AOAC 2002).

e) Proximate Analysis of Moisture Content

A clean 100 ml beaker was dried in an oven to constant weight (a). A known amount of the 5 g sample was introduced in the beaker and weighed (b). The samples were then fried in a ventilated electrically heated atmosphere oven at 75°C for about 24hrs and cooled in a desiccators until constant weight was obtained (c). The Percentage Moisture content was calculated from the formula:

$$\% \text{ Moisture content} = \frac{\text{Weight loss of sample} \times 100}{\text{Weigh of the original sample 1}}$$

The experiment was carried out in triplicates.

f) Ash content

5 kg sample were accurately weighed into the crucible. This was ignited at 55°C for about 24 hrs in desiccators and weighted. This step was repeated until a constant weight was obtained. The percentage ash content was calculated from:

$$\% \text{ Ash content} = \frac{\text{Weight of ash x 100}}{\text{Weight of sample 1}}$$

Determination was made in triplicate.

g) Crude fat or ether extract

5 g samples were accurately weighed into a thimble. About 120 ml petroleum ether was poured into a previously dried and weighted round bottom flask. The Soxhlet extractor into which the thimble with content had been introduced was then filled into the round bottom flask and the condenser and extraction apparatus set up with a cramp and stand. Gentle heat had been applied then the heater evaporated and as it condensed, it dropped into the thimble where it extracted ether soluble constituents into the round bottomed flask. The extraction then continued for about 8 hours. The thimble was then removed and air-dried(later far free extract was used for fibre determination). The petroleum ether in the flask was distilled off and collected in the Soxhlet extractor tube. The flask was then fried in an air circulating desiccators for 8 hours. The round bottom flask and the lipid extract were then weighted. The flask and content was again dried and weighed till a constant weight was obtained. The amount of lipid extracted was obtained from the difference between the weight of the flask before and after extraction.

$$\% \text{ Fat} = \frac{\text{Weight loss of sample (extracted fat) x 100}}{\text{Weight of sample 1}}$$

h) Crude fibre

5 g far free material was weighted and quantitatively transferred into 400 ml beaker, which had been previously marked at 200 ml level. 50 ml of 1.25% sulphuric acid were added and the mixture was made up to 200 ml mark with distilled water. The contents of the beaker were heated to boiling point for 30 minutes.

i) Crude Protein (Micro Kjedahl Method)

40% Sodium hydroxide (NaOH) pellets (40 g pellets carbonates free were dissolved in 100 ml distilled water). Concentrated sulphuricacid(H2SO4), Selenium Kjedahl Catalyst (each tablet containing 1g Sodium sulphate, and 0.05 g Copper sulphate(CuSO4)was dissolved in 0.1% hydrochloric acid (HCL). Methyl red-ethylene blue indicator was prepared by mixing the equal volume of 0.2% twice recrystallized methyl red and 0.0% methylene blue made up in absolute ethanol. This sample was then stored in a dark brown bottle in a refrigerator.

j) Digestion (Micro Kjedahl)

1 g sample was weighed out into a 50 ml Kjedahl digestion flask. 20 ml of antidumping chips were added. The mixture was incinerated to gentle boiling on a digestion rack and then heated strongly until the digest became clear. The digest was removed, cooled and quantitatively transferred to a 100 ml volumetric flask and made up to mark. An Erlenmeyer flask containing 10 ml of boric acid indicator solution was placed at the tip of the condenser extended below the surface of the solution. 10 ml of the sample digest was introduced into quick fit micro Kjedahl flask and steam heated. 10 ml of 40% Sodium hydroxide(NaOH) solution was added to the digest and the digested steam distilled into the Erlenmeyer's flask until the contents become more than double of its original volume as the ammonia (NH3) changed to green. A blank determination was carried out in a similar manner as described above except 1 g digestion sample was replaced by 1 ml of distilled water.

k) Titration

The content of the Erlenmeyer flask was titrated with 0.1% hydrochloric acid to a pink end point.

Calculation

1 ml of HCl (Test) – ml of HCl (Blank) × NX × 100 % Protein = 1000 × 10 × 1

N = Normality of the acid

10 = Ml of digest use

I = Gram of sample used

III. Preparation of Plant Materials

Dried fruits used in the study were separately washed thoroughly using distilled water and surface sterilized with 70% ethanol and sun-dried for 3 days. The dried plant fruits were blended separately using a sterile electric blender to obtain 200grams of fine powder of each fruit. Aqueous extracts of fruits were obtained by adding the dried powder (blended) of plant material to distilled water at room temperature 28±1˚C. Three levels of concentrations were obtained by dissolving 5g, 10g and 15g of each sample with 100ml of distilled water. This was vigorously stirred and allowed for 24 hours. The solution was then filtered through four-folds of sterile cheese cloth for all the plant materials. The filtrates obtained were used as aqueous extracts of the test plants and stored in reagent bottles for further use. Ethanolic extracts of plant materials were obtained by adding the powdered sample at different concentrations, 5g, 10g and 15g to 100mls of ethanol. This was stirred vigorously and allowed for 24 hours at room temperature 28±1˚C. The solution was then filtered through four-folds of sterile cheese cloth for all the plant materials. The filtrates obtained were used as ethanolic extracts of the test plants and stored in reagent bottles for further use.

a) Susceptibility test

The extracts percentage concentrations were prepared at 5g/100ml, 10g/100ml and 15g/100ml with ethanol and water as solvent.

b) In vitro antifungal assay

5ml of each concentration of both the aqueous and ethanol extracts was first poured into different Petri-

dishes using sterile syringe. The sterile potato dextrose agar (PDA) was also poured into the plates containing the solvent extracts after which the plates containing the solvent extracts were gently swirled to ensure mixing. The media was allowed to solidify and with a sterilized cork borer (5mm in diameter), a disc of the matured culture was punched out from advancing margin of a four- day old pure culture and inoculated at the center of plates and incubated at room temperature (28±10c) for 7 days. The experiment was replicated thrice. Area of inhibition was measured daily for 7days using a meter rule and recorded.

c) Phytochemical Screening

Phychemical screening of the aqueous and ethanolic fruit extract of *Tetrapleuratetraptera* was carried out using the method of (Harborn, 1973). Phytochemical screening was carried out in the Department of Biochemistry, University of Calabar, Calabar, Cross River State, Nigeria.

d) Statistical analysis

Data obtained in this study were analyzed using Student T-test and a one way Analysis of Variance (ANOVA) at 5% probability level ($p<0.05$).

IV. Results

a) Isolated fungal pathogens

The fungal pathogens isolated and identified as the causative agents of pre-harvest eggplant fruit rot from this study were: *Collectotrichummelongenae* and *Phomopsismelongenae.*

b) Pathogenicity Test

Symptoms observed on the fruits inoculated with *Collectotrichummelongenae* and *Phomopsis-melongenae* were similar to those observed on the rotted eggplant fruit obtained from the field. Symptoms such as soft rots and lesions were observed on the fruits.

c) Effect of fungi infection on biochemical composition of the Eggplant

The results of proximate analysis in mg/100 g of *P. melongenae* infected eggplant fruits showed an increase in the moisture content of the fungal infected fruits of eggplant as compared to the healthy ones (control), whereas there was a decrease in the carbohydrate, fat, fibre and ash contents of the fungal infected fruits relative to the healthy ones (control). Moisture content increased from 42.90 ± 0.12 in the non-infected eggplant to 58.03±0.20 in the infected eggplant fruit with *Phomopsismelongenae* and protein content increased from 1.40±0.18 in the Post-infected eggplant to 5.60±0.10 in the non-infected fruits while the following parameters were found to decrease in the infected than in the non-infected fruits viz carbohydrate content 1.02± 0.29, fat 2.98±0.25, crude fibre 3.09±0.06, protein 1.40± 0.10 and ash content 0.25±0.16 as presented in (Table 1).The results of proximate analysis in mg/100 g of *C.melongenae* infected eggplant fruits showed that there was an increase in the moisture content from (42.90 ± 0.12 to 60.12±0.40) when infected with *Collectotrichum-melongenae* and from (1.60±0.11) in the Cs-infected garden egg to (5.60±0.10) in the non-infected eggplant when infected with *Colletotrichummelongenae* while the following parameters were found to decrease when infected with *Colletotrichummelongenae* than in the non-infected fruits viz carbohydrate content (1.09± 0.21), fat (3.0±0.31), crude fibre (3.30±0.7), and ash content (0.30±0.21) as presented in (Table 2).

Table 1 : Proximate composition of *Phomopsismelongenae* infected and non-infected eggplant mg/100g (dry matter)

Sample	Moisture	C/F	Fat	Protein	Ash	CHO
Post-infected	58.03±0.20[a]	3.09±0.06[b]	2.98±0.27 [b]	1.40±0.18[a]	0.25±0.16[b]	1.02±0.29[b]
Non-infected	42.90±0.12[b]	5.12±0.09[b]	5.37±0.17[b]	5.60±0.10[b]	0.50±0.15[b]	3.64±0.19[b]
T-value	3.54					

Note: C/F = Crude fibre, CHO = Carbohydrate, Ps = *Phomopsismelongenae*

Table 2 : Proximate composition of *Colletotrichummelongenae* infected and non-infected eggplant mg/100g (dry matter).

Sample	Moisture	C/F	Fat	Protein	Ash	CHO
Cs-infected	60.12±0.40[a]	3.30±0.7[a]	3.0±0.31[a]	1.60±1.11[a]	0.30±0.21[a]	1.09±1.21[b]
Non-infected	42.90±0.12[b]	5.12±0.09[b]	5.37±0.17[b]	5.60±0.10[b]	0.50±0.15[b]	3.64±0.19[b]
T-value	3.53					

Note: C/F = Crude fibre, CHO = Carbohydrate, Cs = *Colletotrichummelongenae.*

d) Phytochemical screening

Phytochemical screening of the aqueous and ethanolic extract of *Tetrapleuratetraptera* showed that cardiac or cyanogenic glycosides, flavonoid, saponin and tannin were present in the aqueous extract while saponin, flavonoid, alkaloids, steroids, triterpens and tannin were present in the ethanolic extracts as presented in (Table 3).

Table 3 : Phytochemical screening of ethanolic and aqueous extract of *Tetrapleuratetraptera*

Phytochemical constituents	Ethanol extract	Water extract
Flavonoid	+	+
saponin	+	+
Tannin	+	+
Alkaloid	+	+
Triterpens	+	-
Cyanogenic glycosides	-	+
Steroids	+	-

Note: (+) = Present, (-) = Absent

e) In vitro effect of ethanolic extract of Tetrapleuratetraptera on Colletotrichummelongenae and Phomopsismelongenae at the different concentrations.

The *in vitro* effect of the ethanolic plant extract at different concentrations on the radial growth of the fungal pathogens is presented in (Tables 4 and 5). Results from the study showed that extract of *Tetrapleuratetraptera* had a significant effect on the isolated fungal pathogens at all levels of concentration (5g/100ml, 10g/100ml and 15g/100ml) tested as compared with the aqueous extracts. Results (Table 4 and 5) showed that *Tetrapleuratetraptera* extract at 5g/100ml concentration completely inhibited the radial growth of *Colletotrichummelongenae* and *Phomopsismelongenae* in the first to fourth day of incubation and at 10g/100ml concentration on the first to fifth day of incubation respectively while at 15g/100ml concentration, the radial growth of *Colletotrichummelongenae* and *Phomopsismelongenae* was completely inhibited throughout the incubation period as compared with the aqueous extracts.

Table 4 : In vitro effect of ethanolic *Tetrapleuratetraptera* extract on *Colletotrichummelongenae.*

Concentrations	Days of incubation and radial growth (cm)						
	1	2	3	4	5	6	7
5g/100ml	0.0	0.0	0.0	0.0	0.1	0.2	0.3
10g/100ml	0.0	0.0	0.0	0.0	0.0	0.1	0.2
15g/100ml	0.0	0.0	0.0	0.0	0.0	0.0	0.0
LSD	0.7*						

Note: Values are means of three replicates

Table 5 : In vitro effect of ethanolic*Tetrapleuratetraptera* extract on *Phomopsismelongenae.*

Concentrations	Days of incubation and radial growth (cm)						
	1	2	3	4	5	6	7
5g/100ml	0.0	0.0	0.0	0.0	0.1	0.3	0.3
10g/100ml	0.0	0.0	0.0	0.0	0.0	0.1	0.2
15g/100ml	0.0	0.0	0.0	0.0	0.0	0.0	0.0
LSD	0.8*						

Note: Values are means of three replicates

f) In vitro effect of aqueous extract of Tetrapleuratetraptera on Colletotrichummelongenae and Phomopsismelongenae at the different concentrations

The *invitro* effect of the aqueous plant extracts at the different levels of concentration on the radial growth of the fungal isolates is presented in (Tables 6 and 7). Results from the study showed that aqueous extract of *Tetrapleuratetraptera* had little or no significant inhibitory effect on the isolated fungi (*Colletotrichummelongenae* and *Phomopsismelongenae*) at all levels of concentration (5g/100ml, 10g/100ml and 15g/100ml) tested when compared with the ethanolic extracts.

Table 6 : Invitro effect of aqueous *Tetrapleuratetraptera* extract on *Colletotrichummelongenae*

Concentrations	Days of incubation and radial growth (cm)						
	1	2	3	4	5	6	7
5g/100ml	2.4	3.4	4.5	4.5	4.5	4.5	4.5
10g/100ml	2.1	3.6	4.5	4.5	4.5	4.5	4.5
15g/100ml	2.3	3.2	4.0	4.5	4.5	4.5	4.5
LSD	0.96						

Note: Values are means of three replicates

Table 7 : Invitro effect of aqueous *Tetrapleuratetraptera* extract on *Phomopsismelongenae*

Concentrations	Days of incubation and radial growth (cm)						
	1	2	3	4	5	6	7
5g/100ml	2.7	3.0	4.5	4.5	4.5	4.5	4.5
10g/100ml	2.4	2.9	4.5	4.5	4.5	4.5	4.5
15g/100ml	1.7	2.9	4.2	4.3	4.4	4.5	4.5
LSD	1.07						

Note: Values are means of three replicates

V. Discussion

In this study, two fungal pathogens were isolated as causative agents of fruit rot of eggplant (*Solanummelongena*), namely *Colletotricummelongenae* and *Phomopsismelongenae* in the infected fruits. Most of the fungal isolates have been found to be associated with the rots of most fruits and vegetables in Kano (Musa and Buashir, 2013), Ibadan (Adio *et al.,* 2013), Port Harcourt (Chuku and Emelike, 2013, Chuku and Barber, 2013), South Eastern States (Iwuagwu *et al.,* 2013) and in Delta state (Taiga and Eyegbagharen, 2013). Pathogenicity test carried out in this study showed that these fungi actually caused the fruit diseases earlier observed on the fruits in the field. The result of proximate analysis carried out showed that moisture content of infected eggplant fruits increase from 42.90 ± 0.12 – 58.03 ± 0.20 when infected with *Phomopsismelongenae* and from 42.90 ± 1.12 – 60.12 ± 0.40 when infected with *Collectrichummelongenae*. The result of proximate analysis (mg/100 g) of non-infected and infected eggplant revealed that moisture content increased in the infected garden egg fruits, to 5.60 ± 0.10 in the infected. This result is in agreement with the findings of Falaye and Fagbohun (2012) who reported an increase in moisture content from 5.09 in the non-infected to 6.13 in the infected and carbohydrate 5.01 to 5.53 of groundnut (*Arachis hypogea*) infected with *Phomospsismelongenae.* Similarly, Nweke and Ibiam (2012) reported an increase in the moisture and protein content of *Anoniamuricata* fruits infected by *Colletotrichumgloeosporoides* and *Rhizopusstolonifer.* Omokolo *et al.,* (1996) have also found that moisture content of non-infected pods (91.0) have decreased to (13.2) in infected cocoa beans. Onifade and Jeff-Agboola (2003) reported that moisture decreased from (36.49 to 10.4) g/100 g in infected samples. The decrease in fat (12.93 ± 0.26), crude fibre (4.67 ± 6.09), ash content (2.30 ± 0.12) and carbohydrate (1.02 0.29) in the infected sample is in agreement with the findings of Shehu and Aliero (2010) have also reported that the infected onion leaf showed a significant decrease in the quantity of the crude protein, fat, fibre and ash content. Opayemi (2012) reported that ash content of non-infected pods (10.7) and beans (8.0) were depleted when infected with *Phytophthora. Palmivora* to (9.3) and (7.8) in cocoa pods and beans, respectively. It could therefore be deduced that the relative increase of moisture in the infected eggplant fruits may be caused by the digestion, degradation and dissolution of the fruit tissue into a mush (water rot) by the pathogens. These degradation activities by pathogens might have also resulted in the relative reduction in the protein, fat, fibre, and ash contents of the infected fruits. The protein, fat, fibre and ash might have been broken down by the fungi into smaller molecules that they absorbed (Nweke and Ibiam 2012). Bonner (1997) reported that complex molecules such as polysaccharide and protein are required by fungi to build the hyphal wall (chitin, glucan and cellulose) and for respiration to obtain energy. Ward and Diener (1991) obtained similar results on groundnut seeds. A decrease in vitamin content of fruits could be due to the increase in moisture content, causing the vitamin to dissolve in it, since it is a water-soluble vitamin. The result of this study shows that the fungi caused deterioration of garden egg fruits and altered the nutritional value of the fruits. In a study conducted by Amer*et al*., (2007), the nutritional contents of garden egg fruits and seeds were greatly affected by the presence of the fungal pathogens *Aspergillusflavus* and *Phomopsis-melongenae,* This suggests that these pathogens might have denied man of these essential nutrients upon consumption through their degradation activities, thereby causing some great damaging effects to human health. Van Duyn and Pivanka (2000) stated that the deficiency of fibre in our diet leads to diverticular diseases and intestinal cancer. However Fanny *et al*., (2000) who also reported a decrease in fat, ash and protein content of maize infected by fungi, stated that the nutrient depletion in entire test plant sample might have been as a result of the internal defense system of the host tissue.

Phytochemical screening of the test plant (*Tetrapleuratetraptera*) extract used in this study was carried to determine its exact phytochemical contents. Results of the phytochemical screening carried out showed thatcardiac or cyanogenic glycosides, flavonoid, saponin and tannin were present in the aqueous extract while saponin, flavonoid, alkaloids, steroids, triterpens and tannin were present in the ethanolic extracts as presented in (Table 3). Phytochemicals, as compounds which occur naturally in plants, form part of plants defense mechanisms against diseases (Eleazu *et al.,* 2012). They are classified into primary and secondary, based on their activity in plant

metabolism. The primary ones comprise of sugars, amino acids, proteins and chlorophyll (Krishnaiah *et al.,* 2007), while secondary ones include the phenolic compounds such as tannins, flavonoids, alkaloids, saponins, anthraquinones, phlobatannins, proanthocyanidins, etc. (Eleazu *et al.,* 2013). These phenolic compounds have been reported to possess considerable antimicrobial properties, which is attributed to their redox properties (Molan and Faraj, 2010, Zongo*et al.,* 2011). Thus the antimicrobial properties of plants have been attributed to the presence of these secondary metabolites (Prakash and Hosetti, 2010).

In this study, the antifungal activity of *Tetrapleuratetraptera* fruit extract was tested *in vitro* on fungi isolated from infected fruits of *S. melongenae*. Results showed that the ethanolic and aqueous extracts of the fruit of *T. tetraptera* investigated, exhibited various antifungal activities against the species of fungi isolated. The antifungal activity of the ethanolic and aqueous extracts of *T. tetraptera* on the isolated fungal pathogens is presented in (Tables 4- 7) respectively. The results showed that, the ethanolic extract had a significant ($P < 0.05$) effect on the radial growth of the fungal pathogens than the aqueous extracts and the rate of antifungal activity differed from one concentration to the other. The differences in the fungitoxic potentials between these plant extracts may be attributed to the susceptibility of each of the fungal pathogens to the different plant extracts. This agrees with the results of some workers like Amadioha, (2000) and Okigbo and Nmeka, (2005). Ilondu *et al.,* (2001) reported that some plants contain phenolic substances and essential oils, which are inhibitory to micro-organisms. The presence of these compounds in these extracts has been reported to be responsible for their antifungal properties (Ahmed and Stoll, 1996).It is noteworthy that of all the tested plant extracts(aqueous and ethanolic), ethanolic extracts of *T. tetraptera* had a more significant effect than the aqueous extracts and the level of inhibition increased with a corresponding increase in the concentration of the extracts. Complete inhibition was observed with ethanolic extract of *T. tetraptera* at the highest concentration of 15g/100ml. The inhibitory potency of the plant extracts may be attributed to the phytochemical compounds like tannins, alkaloids, flavonoids and saponins in them as reported by Chiejina and Ukeh (2013). This is also in agreement with the works of Amadioha and Obi, (1999) and Umana *et al.,* 2014, Umana*et al.,*2015) who reported that the high potency of plant extracts containing the same bio-active compounds could be used for the control of fungal pathogens of plants.

VI. Conclusion

The fungal pathogens isolated and identified from this study as the causative agents of fruit rot of eggplant were *Collectotrichummelongenae* and *Phomopsismelongenae*. The results of proximate analysis of fungal infected and non-infected eggplant fruits showed that there was an increase in the moisture content of the fungal infected fruits of eggplant relative to the healthy ones (control), while there was a decrease in the carbohydrate, fat, fibre, protein and ash contents of the fungal infected fruits relative to the healthy ones (control). It is possible that the isolated fungal pathogens might be resident on the leaves and roots of the plant from where they were dispersed into the fruits to initiate infection spore during rainfall. Results of the *in vitro* antifungal assay carried out showed that the ethanolic extracts of *Tetrapleuratetraptera* were effective against the fruit rot fungal pathogens of eggplant at the different concentrations tested. To this effect, use of resistant seed and timely spraying of egg plant crops with extracts of *Tetrapleuratetraptera* prepared at higher concentrations during flowering and fruiting will reduce the damaging activities of the fungal pathogens and contamination with mycotoxins and other related fungal metabolites that might be hazardous to human health.

References Références Referencias

1. Adio, S. O; Arowolo, O; T, Adelani, A.S, Ogundeji, B.A and Adeji, A. O. (2013).Postharvest fungal Rot of yam (Dioscorearotundata and Dioscoreaalata) in Bodija, market, Ibadan, Oyo state. Programme of events and book of abstract. 6th annual conference of Mycological Society of Nigeria (MYCOSON). 10TH-13TH June. ChikeOkoli Centre, Nnamdi Azikiwe University, Awka. P. 25.
2. Ahmed, S. and Stoll, G. (1996). Biopesticides. In: Biotechnology; Building on Farmers' Knowledge. Macmillan Education Ltd, Bunders, J., B. Haverkort and W. Hiemstra (Eds.). London, pp: 52-79.
3. Alexopolous, C. J and Mins, C. W. (1989). Introductory Mycology, London, John Wiley and sons. Pp 224-228.
4. Amadioha, A. C. (2000). Fungitoxic effects of some leaf extracts against *Rhizopusoryzae* causing tuber rot of potato. *Archives of Phytopatholpflanzo,* 34: 1-9.
5. Amadioha, A. C. and Obi, V. I. (1999).Control of Anthracnose diseases of Cowpea by *Cymbopogon cunitus* and *Ocimumgratissimum.ActoPhytopathology and Entomology*, 85: 89-94.
6. Amer, H., Sahi, S.T., Ghazanfar, M. U. and Ali, S. (2007). Location of seed-borne mycoflora of eggplant (*Solanummelongena* L.) in different seed components and its impact on seed germinability. *International Journal of Agriculture and Biology*, 9 (3): 514-516.
7. Anonymous, (1998). Current status of vegetable Research in India. *World Conference Research on Horticultural Research,* Rome, Italy.

8. AOAC, (2002). Official Method of Analysis, 4th Ed, Association of Official Analytical Chemist, Washington DC.P.3.
9. Barnett H. L., Hunter B. B. (1998).Illustrated genera of imperfect fungi 4th edition, St. Paul Minnesota. P. 32
10. Bonner J (1997).Vitamin B1, a growth factor for plant. Science 85:183184.
11. Bonsu, K. O., Fontem D. A., Nkansah, G. O., Iruome R. N., Owusu, E. O. and Schippers, R. R. (2008). Diversity within the Gboma eggplant (*Solanum-marcocarpon*), an indigenous vegetable from West Africa. *Ghana Journal of Horticulture,* 1:50-58.
12. Chiejina, N. V. and Ukeh, J. A. (2013). Efficacy of *Afromomummelegueta* and *Zingiberofficinale* extracts on fungal pathogens of tomato fruit. *IOSR Journal of Pharmacy and Biological Sciences,* volume 4, issue 6, 13-16.
13. Chinedu, S. N., Olasumbo, A. C., Eboji, O .K., O. C., Arinola, O . K. and Dania, D .I. (2011) Proximate and phytochemical analysis of *Solanumaethiopicum* L and *Solanummacrocarpon* C. fruith. *Research Journal of Chemical Sciences*, 1: 67-71
14. Chuku, E .C and Barber, L. (2013). Pineapple fruit rot fungi and their effects on the proximate Composition. Programme of events and book of abstract, 6th annual conference of Mycological Society of Nigeria (MYCOSON), 10TH-13TH of June. Chike Okoli Centre, NnamdiAzikiwe University Awka. P.3.
15. Chuku, E. and Emelike, N. J. T (2013).Comparative studies on the nutrient composition of water melon (*Citrulluslunatus*) and cucumber (*Cucumissativus*) and associated fungi. Programme and Events and book of abstract, 6th annual conference of Mycological Society of Nigeria (MYCOSON) 10TH-13TH June. Chike Okoli Centre, NnamdiAzikiwe University, Awka.P.3.
16. Edem, C. A., Dounmu, M. I., Bassey, F. I., Wilson, C and Umoren, P. (2009).A comparative assessment of the proximate composition of Ascorbic Acid and heavy metals content of the two species of garden egg (*solanum silo* and *aubergaine*). *Pakistan Journal of Nutrition*, 8(5): 582 – 584.
17. Eke-Ejiofor, J., Kiin–Kabari, D. B. and Chukwu, E. C. (*2012*).Effect of processing method on the proximate, mineral and fungi properties of groundnut (*arachishypogea*) seed. *Journal of Agriculture and Biological Sciences,*(1):. 257–261.
18. Eleazu, C. O., Eleazu, K. C., Awa, E. and Chukwuma, S. C. (2012) Comparative study of the phytochemical composition of the leaves of five Nigerian medicinal plants. *Journal of Biotechnology and Pharmaceutical Research*, 3: 42-46.
19. Eleazu, C. O., Iroaganachi, M. A. and Okoronkwo, J. O. (2013) Determination of the physicochemical composition, microbial quality and free radical scavenging activities of some commercially sold honey samples in Aba, Nigeria: 'The effect of varying colours'. *International Journal of Biomedical Research*, 4: 1-6.
20. Falaye, O. S. and Fagbohun, E. D. (2012). Effects of storage on the proximate, mineral composition and mycoflora of "tinco" Dried meat sold in Oshodi, market Lagos State. *Nigerian Global Journal of Biological Science and Biotechnology,* 1(1):54-58.
21. Fanny, C. P., Rigel. L., and Agricia, Q. (2000). Characterization of cocoa butter extracted from cultivars of Theobroma cacao. L. *Archive Latino America de Nutrition*, 4:4-10.
22. FAO, (2007). Food and agriculture organization FAOSTAT. (www. Fastat. Fao. Org) (assessed 3 April 2009).
23. Gill, L. S. (1992). Ethnomedical uses of plants in Nigeria. University of Benin Press. Benin, Nigeria. P. 215.
24. Giuliani, M. R. and Smale, J. L. (2000). Eggplant, In: Vegetables 7th edition of National Trust. India. Pp. 50-56.
25. Grubben, A. Y. and Denton, I. U. (2004).History of eggplant. Manual on Microbiology and Nutritional Merits of eggplant. AVI Publishing Co. West Port, Connecticut, Pp. 230-240.
26. Harborn, J. B. (1973) Phytochemical methods: A guide to modern techniques of plants analysis. Chapman and Hall Press, U.S.A Pp.29-42,23-26,31-36
27. Hanson, P. M. Yang, R. T., Isou, S. C. S, Redesma, D. Engle, L. and Lee, T. C. (2006). Diversity in eggplant *solanummelongena ascorbic acid. Journal of food composition and analysis,* 19 (6-7) 594-600.
28. Ilondu, E. M., Ejechi, B. O. and Souzey, J. A. (2001). Microbial stability of jam prepared from velvet tamarind and preserved by combined processes. *Nigerian Journal of Microbiology*, 5: 93-96.
29. Iwuagwu, C. C., Umechuruba, C. I. Ononuju, C. C. and Emeka, A. N. (2013): Studies on the mycoflora of Rice seeds from rice growing areas of South-Eastern Nigeria and impact on seed germination. Programme and Events and book of abstract, 6th annual conference of Mycological Society of Nigeria (MYCOSON) 10TH-13TH June. Chike Okoli Centre, NnamdiAzikiwe University, Awka. P40.
30. Krishnaiah, D. R. and Sarbatly, B. A. (2007). Phytochemical antioxidants for health and medicine-A move toward nature. *Biotechnology and Molecular Biology Review,* 1: 97-104.
31. Molan, A. L., Faraj, A. M. (2010) The effects of condensed tannins extracted from different plant species on egg hatching and larval development of *Teladorsagiacircumcincta* (Nematoda: Trichostrongylidae). *Folia Parasitological*, 57: 62-68.
32. Musa, H. and Buashir, R. (2013). Fungi isolates of some rotten fruits and vegetables collected at

Yankaba market in Kano state. Program of events and book of abstract of the 6th annual Conference of the Mycological society of Nigeria (MYCOSON), 10TH-13TH JUNE 2013 ChikeOkolicentre, Nnamdi Azikiwe University, Awka. P. 16.

33. Nweke, C. N., and Ibiam, O. F. A. (2012).Studies on pre and post-harvest fungi associated with the soft rot of the fruit *Anonamuricata,* and their effects on the nutrient content of the pulp. *American .Journal. Food and Nutrition,* 2(4):78-85.
34. Okigbo, R. N. and Nmeka, I. A. (2005). Control of yam tuber with leaf extracts of *Xylopiaaethiopica* and *Zingiberofficinale.African Journal of Biotechnology*, 4: 804-807.
35. Omokolo, N. D., Tsala, N. G., Djoigoue, P. F. (1996). Changes in carbohydrate, amino acid and phenol content in cocoa pods from three clones after infection with *PhytophthoraMegarkaya* Bra and Grif. *Annals of Botany*, 77:153-158.
36. Onifade, A. K., Jeff-Agboola, Y. A. (2003). Effect of fungal infection on proximate nutrient composition of coconut (*Cocosnucifeira* Linn) Fruit. *Journal of Food, AgricultureandEnvironment,* 10(2):30-33.
37. Opayemi, U. L. (2012). Effect of storage on the nutrient composition and the mycobiota of sundried water melon (*Citrilluslanatus*) seed. *Journal of Microbiology, Biotechnology and Food Science*, 1(3): 267-276.
38. Prakash, G. and Hosetti, B. B. (2010).Investigation of antimicrobial properties of *Dioscoreapentaphylla* from mid-Western Ghats, India. *Scientific World*, 8: 91- 96.
39. Rangas-wami, G. (2000). Disease of crop plant in India (2nd edition) Prentice Hall of India. PVT Limited, New Delhi. Pp.298-302.
40. Shehu K, and Aliero, A. A. (2010).Effects of purple Blotch infection on the proximate and mineral contents of onion leaf. *International.Journal. Pharmacentical.Scientific.Research*. 1(2):131-133.
41. Taiga, A. and Eyegbagharen, T.P. (2013): Fungi associated with stored maize (*Zea mays*) grains in Abraka Market, Delta State. Programme and Events and book of abstract, 6th annual conference of Mycological Society of Nigeria (MYCOSON). 10TH-13TH June. Chike Okoli Centre, Nnamdi Azikiwe University, Awka. P. 40.
42. Umana, E. J., Akwaji, P. I., Markson, A. A., Udo, S. E. and Orok, E. E. (2014). Phytochemical composition, Antimicrobial effect of Azadirachta indica and Carica papaya extracts on fungi isolated from Gmelina arborea seedlings. *International Journal of Phytopathology*, 03 (03): 109 – 115.
43. Umana, E. J., Akwaji, P. I., Markson, A. A., and Udo, S. E. (2014). Phytochemical constituents and *in vitro* inhibitory effect of ten plant extracts on fungi associated with *Gmelinaarborea*Roxb in Cross River State, Nigeria. *Journal of Biopesticides and Environment*, 1: 63 -73.
44. USDA, (2008). Eggplant (raw) Nutrient values and Weight for edible portion. United State Department of Agriculture (NDB No: 11209).
45. Van Duyn, M. and Pivonka, E. (2000).Overview of the health benefits of fruit and vegetable consumption for diabetes. *Journal of American Dietetic Association*, 100 (12): 1511-1521.
46. Zongo C, Savadogo K, Somda JM, Koudou J, Traore AS (2011) *In vitro* evaluation of the antimicrobial and antioxidant properties of extracts from whole plant of Alternantherapungens H.B. & K. and leaves of *Combretumsericeum* G. Don. *International Journal of Phytomedicine,* 3: 182-191.

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