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## Antifungal effect of Polar and non polar extracts of *Aframomum Sceptrum* on Two Isolates of Oil Palm

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KEY WORDS: Oil palm, Antifungal, Phytochemical, DMSO, Pathogens.

**ABSTRACT:** In different parts of the world, attention is being paid to exploitation of higher plants as biodegradable fungicides in the control of most plant pathogenic fungi. Different spices of the Zingiberaceae family have been tested for their antifungal properties, but there exists little or no information on the antifungal potential of a particular member of that family; Aframomum sceptrum on fungal pathogens of some economic important palms in Nigeria. In this study, the phytochemical composition of the seed extracts of this spice was analyzed by standard methods while the antifungal activities of polar and non polar extracts of the spice was tested on two major isolates affecting the Oil palm, Fusarium oxysporum f.sp elaeidis and Hypocrea lixii (IMI 501885) Cold extraction using Acetone, Ethanol, Hexane, Methanol, and Diethylether solvents were used in the seed extract preparation. The broad spectrum fungicide, Mancozeb (80% wettable powder) was used as the positive control while the negative control was Dimethyl sulphoxide. The Dimethyl sulphoxide was also used to reconstitute the solvent extracts by dissolving the extracts and fungicides in appropriate amount of 15 % (v/v) to obtain a concentration of 0.0624g/ml. The phytochemical screening revealed the presence of the following phytochemicals in different quantities; Alkaloids, Terpenoids, Anthraquinones, Flavonoids Tanins, Saponins. Results obtained showed that all the extracts had a significantly higher antifungal effect (p < 0.05) than the broad spectrum fungicide, Mancozeb at 2000ppm. Non polar hexane seed extract had the highest percentage inhibition of 60.26% on Hypocrea lixii (IMI 501885 while the Polar ethanolic extracts with a percentage inhibition of 52.73 % on Fusarium f.sp elaeidis. Amongst all the extracts used in this study, the seed extracts that gave a low percentage inhibition of 42.45% was the non -polar acetone seed extract on Fusarium oxysporum fsp. elaeidis and methanol extract on H.lixii with the least percentage inhibition of 42.31%. The implications of these findings are discussed. © JASEM

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Introduction: The Oil palm (Elaeis guinnensis) of the family Arecaceae is a major economic palm of the world, with their oils as their major economic products (Nair, 2003). They are perennials, monocots and are grown majorly in tropical regions of the world with heights up to 1000 meters above sea level (Nair et al., 2003). The palm is generally regarded to as the tree of life because of its diverse way in which it can be utilized both in small-scale and large-scale. It is native to West and Southwest Africa (De Marco et al., 2007) and it is classified as the most important oil crop in the world (Oil world, 2008). The palm is classified into three separate groups based on the shell thickness of its fruit: Dura (thick shell), Tenera (relatively thin shell) and Pisifera (absence of shell). Tenera, the genetically formed hybrid between Dura

and Psifera has higher oil extraction efficiency (Hartley, 1988; Harminder *et al.*, 2010) and The empty fruit bunch, the shell and fiber that remain after oil extraction are used for mulching, manuring and as fuel (Ravi Menon *et al.*, 2003). The leaves of Oil palms are used for making brooms, roofing and thatching, basket and mats. The sap tapped from the tree called "The palm wine" can be allowed to ferment and distilled into a gin known as "*Akpetesin*" in Ghana and "*Ogogoro*" in Nigeria (Simon Heap, 2008). Plant diseases play a direct role in the destruction of natural resources in agriculture and in particular, pathogens cause important losses, fungi being the most aggressive (Abou-Zeid *et al.*, 2008). Fungi often attack these palms either at the pre-nursery, nursery and adult palms stages often

resulting in an indirect yield reduction by affecting fruit quality. The Vascular wilt disease of Oil palm first described by Wardlaw in 1946 is caused by Fusarium oxysporum f. sp elaeidis (Ho and Varghese, 1986) and it is the most damaging disease of Oil palm in Africa (Flood, 2006; Tengoua and Bakoumé, 2008), causing up to 70% mortality. Several Western and Central African countries are particularly affected by vascular wilt (Aderungboye, 1982; Corley and Tinker 2003; Tengoua and Bakoumé, 2008; de Franqueville et al., 2011). Some other fungal isolates from the exocarp of deteriorating palm fruit is Hypocrea lixii (anamorph Trichoderma harzianum) Chaverri and Samuels, (2002). Trichoderma harzianum as it is the most frequent Trichoderma species in majority of samples world1`wide (Zachow et al., 2009).Many of the genus are ubiquitous inhabitants of soils, decaying plant matter and debris (Harman et al., 2004). They have been found to colonize and cause infections on oil palm seeds in germinators and storage rooms (Ghosh, 2009) especially in subtropical and tropical regions like Nigeria (Cirumalla et al., 2011; Gomathi and Ambikapathy, 2011). Fungicides have been used over time by farmers to reduce the activities of these pathogens however; the extensive use of fungicides in plant protection against fungal disease generates long-term residues in food and in the environment (EFSA, 2009; Petit et al., 2008). In the annual EU report, EFSA (European Food Safety Authority), where vegetables and fruits of 27 countries were surveyed for pesticides contamination, the results highlighted that dithiocarbamates are among the most common residual contaminants (EFSA, 2009). The abusive use of such compounds in agriculture has mobilized public concern because of the harmful potential of such substances in the environment and in the food chain representing a risk for human health (EFSA, 2009). A major toxicological concern with respect to mancozeb and other dithiocarbamates is its primary metabolite, ethylene thiourea (ETU), shown to cause structural and functional alterations of gonads and thyroids of humans (Kackar et al., 1997; Baligar et al., 2001). Ethylene thiourea is very soluble and moderately mobile in the environment, it potentially could be found in drinking water from both surface water and groundwater sources close to some farm lands. It was measured at 0.21 ppb in the raw water from one public drinking-water well. (Detection limit = 0.1 ppb (EPA 2005). Studies have shown the importance of natural chemicals as a possible source of non-phytotoxic, systemic and easily biodegradable alternative fungicides (Loizzo et al., 2004). Mushin et al., 2001; Okigbo and Emoghene, 2003 reported that many plant extracts have been demonstrated to be effective in the control of several plant diseases as they exhibit a number of chemical and biological advantages as fungicides. Biological investigations carried out on several species from the genus Aframomum, of the family Zingiberaceae revealed their antiparasitic, antifungal, antibacterial and antiviral properties. (Cousins and Hoffman, 2002; Okwu et al., 2003). Further studies their analgesic, aphrodisiac. showed antiinflammatory, antioxidant, antiulcer and antiprotozoal efficacies (Singh et al., 2007; Cheikh et al., 2011) while the seeds of Aframomum species have been found to contain phytochemicals (Fasoyiro and Adegoke, 2007). Exploitation of antifungal agents from plant metabolites is considered to be an approach to identify novel fungicides which meet environmental requirements as well as help control plant pathogenic organisms. This *invitro* study seeks to investigate the possibility of using the seeds of this plant to control two economically important pathogens of the Oil palm, Fusarium oxysporum and Trichoderma sp.

### **MATERIALS AND METHODS**

Sample collection: Infected plant parts showing disease symptoms were identified from different parts of the plant, samples were taken from the mesocarp of rottening palm fruits, while other samples were collected from the roots, the foliar parts and soil area from some infected Oil palms showing symptoms of vascular wilt disease. A pre-sterilized soil probe was used to collect five grams (5g) of soil with a depth of about four inches from the perimeter base of each Oil palm sampled for this experiment. The soil samples were then placed in sterile polyethylene bags, labeled and sealed by the bags. Diseased root samples collected were cut with the use of a sterile scissors and the samples placed in separate labeled sterile sample bags and sealed.

Culture media Preparation and Sterilization of materials: The culture media used for this study is the general purpose media, the Potato dextrose agar (PDA). The media was prepared according to manufacturers' instructions and sterilized using a portable Gallenkamp autoclave at  $121^{\circ}$ C at 15 psi for 15 minutes and allowed to cool to a temperature of about 23° C before using (Obagwu *et al.*, 1997). Autoclavable materials such as McCartney bottles, beakers, and distilled water were sterilized using the same autoclave as above under the same condition of

 $121^{0}$ C at 15 psi for 15 minutes while Petri dishes and other metal apparatus such as spatula and forceps was sterilized using hot air oven at a temperature of  $160^{0}$ C for 2 hours. The wire loops were sterilized by heating in the blue flame of a Bunsen burner until red-hot and allowed to cool before using.

Isolation and Identification of Test organisms: Thin sections (2mm diameter) were cut from the peripheral areas of diseased lesions of the samples using a sterile blade and sterilized for one minute in already prepared 30% v/v sodium hypochlorite solution and rinsed in three changes of sterile distilled water (Kinkel and Andrews, 1988) and placed on sterile filter papers in order to blot out traces of water. In the sterile Laminar flow chamber, the samples were transferred aseptically using a sterile wire loop into pre-sterilized 9cm diameter Petri plates containing solidified Potato Dextrose agar (PDA) supplemented with streptomycin. The plates were incubated at ambient conditions of light and temperature of 25  $\pm$ 2°C (Abou-Zeid et al., 2004). The cultures were monitored on a daily basis until growths were observed. Individual fungal colonies were purified by aseptically sub culturing into fresh PDA plates. The plates were further allowed to stand for another seven to ten days to allow for proper sporulation and consequent identification while some cultures were stored in PDA slants and refrigerated at 4°c for future use. Isolates of Fusarium oxysporum were identified based to their cultural, morphological and microscopical characteristics as described by Nelson et al., (1994) and compared with texts from Common wealth Mycological Institutes identification Manual while the Trichoderma sp. was identified by CABI, as Hypocrea lixii (anamorph Surrey, UK Trichoderma harzianum) with IMI No. 501885. Snap shots of the spores of these organisms as shown on plate 1.0 were captured using a motic camera attached to a microscope.

*Extract Preparation:* Fresh seeds of *Aframomum sceptrum* were obtained from a market in Benin City, Edo State, Nigeria and identified by a botanist at University of Benin, Benin City. The seeds were harvested from the pods by hand picking. They were washed thoroughly with sterile distilled water; oven dried at a temperature of  $45^{\circ}$ C and pulverized using a Lexus mixer grinder with model No: MG 2053 to a fine powder form according to the method of Wokocha and Okereke, 2005. Thirty grams (30 g) of the fine powder was weighed using an analytical weigh balance with model NO: ALC 201.3 15038730. Five different extracting solvents of

analytical grade were used to prepare the seed extracts (Bautista et al., 2003). The solvents include Ethanol, Diethyl ether, Hexane, Acetone and Methanol. Fifty (50mls) of each solvent was measured using a clean calibrated cylinder and poured carefully into sterilized 250ml Erlenmeyer flask each. The already weighed powder (30g) of the seed powder was then dissolved in the respective solvents to give 60% w/v. The flasks were sealed with a sterile foil paper and held tightly with rubber bands. The labeled extracts were then placed in an orbit shaker with model No: 3521 for 24 hour duration to allow for uniform extraction of the active ingredients. The solvents were recovered using a buchii rotary evaporator Manufactured by Bibby Sterlin LTD., England with model No: RE 100 at 40 °C under vacuum and the extracts reconstituted by dissolving in appropriate amount of 15 % (v/v) dimethyl sulphoxide to obtain a concentration of 0.0624g/ml. Dimethyl sulphoxide an organosulphur compound which is colorless with a molecular formula of (CH<sub>3</sub>)<sub>2</sub>SO dissolves polar and non polar compounds (Novak, 2002). This dissolution in DMSO did not affect the mycelia growth of the test microorganisms negatively; this was observed during the preliminary studies and confirmed by the control experiments.

Antifungal activity assay using Pour Plate Method A micropipette was used to measure 1ml of each of the solvent extracts into already prepared 10 ml of PDA medium in a glass test tubes, the mixture was shaken thoroughly and poured into a labeled sterile Petri plate in a laminar flow chamber .This procedure; food-poisoned technique was carried out according to Nene et al., 2000. The amended medium was swirled gently to ensure thorough mixing of the contents before solidification of the medium. Three (3) mm disc of three-day-old culture of the fungus, Fusarium oxysporum and Hypocrea lixii IMI No. 501885 was harvested using a 3mm sterile cork borer and each organism was aseptically inoculated unto the centre of the labeled Petri plates in an inverted position in order for the mycelium to achieve greater contact with the culture medium. The position of the disc was marked on the base of the dish with a marker pen and two perpendicular lines were drawn passing through the marked position, this line was used measuring the radial growth of the organism. The experiment was carried out in triplicates and the plates were incubated at temperature of  $28\pm 2^{\circ}$ C. Two controls were set up, the PDA medium modified with the 1ml of DMSO4 served as the negative control and the

other, PDA amended medium with 1ml of the fungicide, mancozeb (80% wettable powder) served as the positive control using the recommended dosage of 2 g/l (2000ppm) for antifungal activity. Daily measurements for seven days of the mycelia extension of the individual cultures were taken with the aid of a meter rule by measuring colony diameters along the two perpendicular lines drawn on the reverse side of the plates and the average means were calculated for each duplicate and recorded. The percentage inhibition was calculated as percentage of the difference between the radial growth of the isolate when inhibited and the radial growth when uninhibited, for each isolate using the formula:% Inhibition = 100 (Control - Treatment)/Control (Singh and Tripathi 1999).

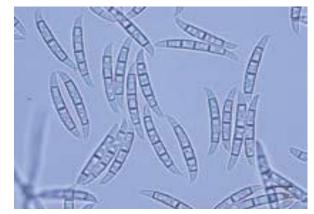
The values of percentage inhibition of the test and controls were compared for each test sample. The difference in their percentage inhibition gives a reflection of the extent of inhibition by the extracts on each isolate.

Phytochemical Screening Extracts: Qualitative and quantitative phytochemical analysis was carried out on the five different extracts for Six (6) phytochemicals bringing each the total phytochemicals screened to thirty. The screening was carried out in the Biochemistry laboratory of Institute for Oil Palm Nigerian Research (NIFOR.)Benin City and in the Chemistry laboratory of the Nigerian Institute for Science Laboratory technology (NISLT) Ibadan, Nigeria. The extracts were evaluated for the presence of Alkaloids, Flavonoids, Saponins, Anthraquinone, Tannins, and Terpenoids using standard procedures as described by Sofowora 1993, Trease and Evans, 1989, Harborne, 1998 and Ghani 1998).

*Statistical analysis:* Data obtained was subjected to biostatistical analysis using the one-way ANOVA, while significant differences among the means were determined by using Duncan's New Multiple Range (DMR) Test as outlined by Obi, 2002. The statistical analysis was conducted with SPSS software (SPSS 17, USA set at significant levels of 0.05).

### **RESULTS AND DISCUSSION**

Different fungi were isolated from the diseases leaf lesions of oil palm leaves, roots, soil area as well from the mesocarp of oil palm fruit showing symptoms of deterioration. Amongst these fungi isolated and tested for this invitro antifungal investigation were Fusarium oxysporum f.sp eleaidis (Summerell et al., 2003) and Trichoderma sp. further identified as Hypocrea lixii based on: CABI Identification Report; Hypocrea lixii. (IMI number 501885): The sample was identified using partial ITS DNA sequencing analysis. The sequence obtained showed 100% identity to multiple ITS sequences reported from Hypocrea lixii including a number cited in peer-reviewed publications (Chaverri and Samuels, 2002). The isolate showed the anamorphic state of Trichoderma harzianum. Plates 1 and 2 below, shows the pictomicrograph capture of isolates



**Plate 1:** Pictomicrograph capture *F. oxysporum* showing slightly sickle-shaped macroconidia .



Plate 2: Pictomicrogh capture of Hypocrea lixii IMI 501885

oxvsporum.

Results on tables 1 and 2 respectively shows the effect of the five seed extracts of *A. sceptrum* on the mycelia growth within a period of seven days of monitoring and incubation under temperature of  $28\pm 2^{\circ}$ C. **Table 1** Average mycelia measurement in cm for each extract during the period of seven days on *Fusarium* 

Extracts	Day 0	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
N	0.60±0.00	1.05±0.05	2.00±0.00	2.80±0.10	3.45±0.05	4.25±0.05	4.95±0.25	5.5±0.10
Control								
Р	$0.60 \pm 0.00$	$0.60 \pm 0.00$	$0.70 \pm 0.00$	$1.30\pm0.10$	$1.90\pm0.20$	$2.50\pm0.20$	3.00±0.30	$3.60 \pm 0.30$
Control								
ACE	$0.60 \pm 0.00$	$0.70 \pm 0.00$	$0.95 \pm 0.05$	$1.40\pm0.00$	$2.00\pm0.10$	$2.35 \pm 0.15$	$2.65 \pm 0.05$	$3.00 \pm 0.00$
ETH	$0.60 \pm 0.00$	$0.65 \pm 0.05$	$1.00\pm0.00$		$1.85 \pm 0.05$	2.20±0.10	2.30±0.00	$2.60 \pm 0.00$
				1.35±0.05				
MET	$0.60 \pm 0.00$	$0.70 \pm 0.00$	$0.80 \pm 0.00$	$1.20\pm0.00$	$1.70\pm0.05$	$2.05 \pm 0.05$	$2.45 \pm 0.05$	$2.85 \pm 0.05$
HEX	$0.60 \pm 0.00$	$0.70 \pm 0.00$	$0.85 \pm 0.05$	$1.45 \pm 0.15$	$1.80 \pm 0.05$	$2.05 \pm 0.05$	2.30±0.10	$2.75 \pm 0.05$
DEE	$0.60 \pm 0.00$	$0.65 \pm 0.05$	$0.85 \pm 0.05$	$1.30 \pm 0.00$	$1.70 \pm 0.00$	$2.10\pm0.00$	$2.40 \pm 0.00$	$2.7 \pm 0.00$

\*Data are given as mean of radial growth for three readings (triplicates), values  $\pm$  are standard errors of mean (SEM) of triplicates

**Table 2** shows the average mycelia measurements in cm for each extract during the period of seven days on

 *Hypocrea lixii* (501885)

Extracts	Day 0	Day 1	Day2	Day 3	Day 4	Day 5	Day 6	Day 7
N Control	$0.60\pm0.00$	1.20±0.00	2.65±0.05	3.00±0.00	3.40±0.10	3.70±0.10	3.80±0.10	3.90±0.10
P Control	$0.60\pm0.00$	0.70±0.10	1.50±0.20	$1.80\pm0.10$	2.05±0.15	2.25±0.25	2.35±0.15	$2.50\pm0.20$
ACE	$0.60\pm0.00$	0.65±0.05	$1.50\pm0.10$	$1.65 \pm 0.15$	$1.85 \pm 0.25$	1.95±0.15	2.15±0.25	2.15±0.25
ETH	$0.60\pm0.00$	$0.60 \pm 0.00$	$1.00\pm0.00$	$1.25 \pm 0.05$	$1.30\pm0.00$	$1.40\pm0.00$	$1.55 \pm 0.15$	$1.70\pm0.00$
MET	$0.60\pm0.00$	0.70±0.00	$1.20\pm0.10$	$1.45 \pm 0.15$	$1.65 \pm 0.25$	$1.80\pm0.20$	$2.10\pm0.30$	$2.25 \pm 0.35$
HEX	$0.60\pm0.00$	$0.60\pm0.00$	$0.80 \pm 0.00$	$1.00\pm0.00$	$1.10\pm0.00$	$1.10\pm0.00$	$1.40\pm0.00$	$1.55 \pm 0.05$
DEE	$0.60 \pm 0.00$	$0.60 \pm 0.00$	$0.85 \pm 0.05$	$1.05 \pm 0.05$	$1.40\pm0.10$	1.45±0.05	$1.65 \pm 0.05$	$1.80 \pm 0.00$

\*Data are given as mean of radial growth for three readings (triplicates), values  $\pm$  are standard errors of mean (SEM) of triplicates

#### Key:

N Control; Negative control (Dimethyl sulphoxide) P Control; Positive control. Fungicide Ace; Acetone Eth; Ethanol Met; Methanol Hex; Hexane Dee; Diethyl ether

On table 1 above, the extracts and fungicide had significant (P<0.05) antifungal effect on *Fusarium* oxysporum. Ethanol extract is seen to have the highest antifungal activity as it was able to inhibit the mycelia growth of *Fusarium oxyporum* (f.sp *eleaidis*) after the seven day period of observation. The extract with the closest inhibitory activity to Ethanol extract is Diethyl ether extract. When the activities of the two extracts were compared individually to that of the fungicide, the results were significantly better at p< 0.05 than fungicide. However, all extracts tested had better mycelia growth inhibition of *F.oxysporum* f.sp *eleaidis* than the positive control, Mancozeb.

Results on table 2 indicates that Hexane seed extract gave the best results with an inhibition of mycelia growth measurement of  $1.55 \text{cm} \pm 0.05$  (SEM) on *Hypocrea lixii* (501885) than all the extracts after seven days of observation. This is followed by Ethanol extract with a mycelia growth measurement of  $1.70 \text{cm} \pm 0.00$  (SEM). The antifungal activity of all the seed extracts was higher than the two controls. The isolate that responded best to all the seed extracts is *Hypocrea lixii* (IMI 501885) as it had the least mycelia growth rate.

<b>Table 3</b> shows the results of the percentage inhibition of each extract on the two isolates after seven days.							
ISOLATES	ACETONE	ETHANOL	METHANOL	HEXANE	DIETHLETHER	FUNGICIDE	
F. oxysporum	45.45	52.73	48.18	50.00	50.91	34.55	

*1/ -1		din managements.	~~~ 0/			
H. lixii	44.87	56.41	42.31	60.26	53.85	
r. oxysporum	45.45	52.75	40.10	50.00	50.91	

\*Values are represented in percentages %

From the results on table 3, it can be clearly observed that generally, all the extracts had a significantly higher antifungal effect (p< 0.05) than the broad spectrum fungicide, Mancozeb. However, the extract that gave the highest percentage inhibition for *Fusarium oxysporum* (*f.sp elaeidis*) was the polar solvent of Ethanol seed extract with a percentage inhibition of 52.73 % while the highest percentage inhibition for *Hypocrea lixii* was non polar Hexane seed extract with a percentage inhibition of 60.26%. Amongst all the extracts used in this study, the seed extracts that gave the least percentage inhibitions are the Non polar Acetone seed extract on *F. oxporum* (*f.sp elaeidis*) with a low percentage activity of 42.45% and Methanol extract on *H. lixii* with a low percentage inhibition of 42.31%.

35.90

Figures 1.0a and 1.0b shows the graphical representation of the growth pattern of each isolate (mean values were used for the plots) over the period of seven (7) days in response to treatment with each extract as well as the positive control, fungicide and that of dimethyl sulphoxide, the negative control

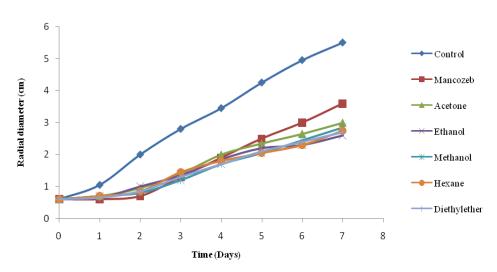
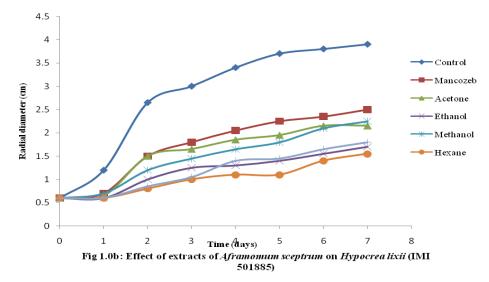


Fig 1.0a: Effect of extracts of Aframomum sceptrum on Fusarium oxysporum

From the growth pattern on the graph above (fig 1.0a), the two controls had little inhibition on the mycelia growth of the pathogen. This inhibition pattern was closely followed by acetone extract, then

Methanol and hexane extracts and Diethylether extracts. The extract with the best inhibition on F. *oxysporum* f.sp *elaeidis* was ethanol extract

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On fig 1.0b, the activities of all the extracts can be clearly seen as inhibitory although at different rates. All extracts had lower inhibitory growth pattern than the fungicide. It is also observed that the negative control and protic solvent did not have any inhibitory effect on the growth rate of the fungus as shown by the uninhibited growth rate of the isolate. *Phytochemical screening*: The results for the phytochemical screening are represented on table 3 below

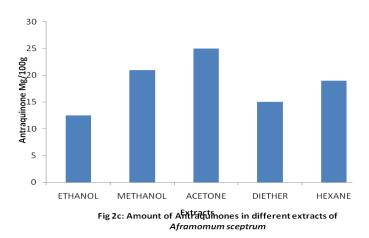
Table 3: Quantitative planation	hytochemical	screening of	polar and non	polar extracts of A.scep	otrum
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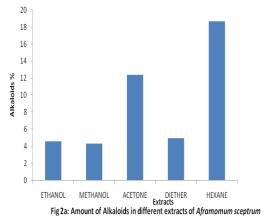
Extracts	Alka (%)	Terp (Mg/100g)	Anthra (Mg/100g)	Flav (%)	Tanins (%)	Saponins (Mg/100g)
ETH	4.59	17.50	12.50	2.15	7.00	7.00
MET	4.32	15.00	21.00	1.94	4.05	22.00
ACE	12.39	10.50	25.00	1.50	1.95	18.00
DEE	4.97	20.00	15.00	1.80	0.945	2.00
HEX	18.69	25.50	19.00	1.14	9.39	4.50

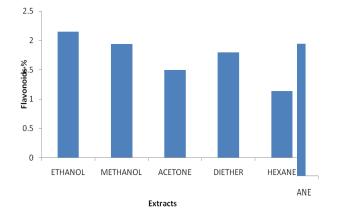
KEY: ETH, Ethanol; MET, Methanol; ACE, Acetone; DEE, Diethylether and HEX, Hexane. Alka, Alkaloids; Terp., Terpenoids, Anthra, Anthraquinones, Tanins, Saponins.

From the results on table 3, the following is observed; the highest percentage of Alkaloids was present in Hexane seed extract with a percentage of 18.69 % with the least amount of alkaloids (4.32%) present in Methanol seed extract. Terpenoids had highest concentration of 25.50% in Hexane extract and the least concentration in acetone extract with 10.50%. Amount of Anthraquinones in the seeds of A.sceptrum was highest in acetone extract with an amount of 25.00mg/100g while concentration of Anthraquinones in Ethanol extract was 12.5mg/100g. Ethanol solvent extracted 7.00% of Tannins from the seeds of this spice while hexane extract gave a percentage of Tanins as 9.39% and this was the highest concentration of tannins in all the extracts used as diethylether gave the least amount of tannins with a percentage 0.945%. The percentage of Flavonoids was highest in ethanol extract with a concentration of 2.15% which was higher than the rest extracts and a concentration of 1.14% was observed in hexane extract which gave the lowest concentration of Favonoids. Lastly, the extract of methanol was highest with Saponins at 22.00mg/100g concentration and Diethylether the least whose amount of Saponins was analysed to be 2.00mg/100g.

The graphs on Fig 2a-2f below shows the types and amounts of phytochemicals present in the Five polar and non polar extracts used for this study.







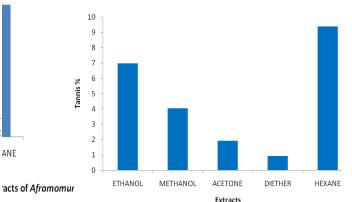
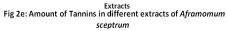


Fig 2d: Amount of Flavonoids in different extracts of Aframomum sceptrum



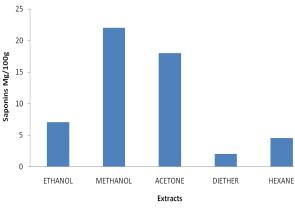


Fig 2f: Amount of Saponins in different extracts of Aframomum sceptrum

OKOGBENIN, OB; EMOGHENE AO; OKOGBENIN EA; AIREDE CE;

Conclusion: This study clearly shows the antifungal potentials of Aframomum sceptrum on two economic important pathogens of the Oil palm. The presence of various active ingredients (secondary plant metabolites) Okigbo and Emoghene, A.O. (2003) as revealed by the phytochemical screening (table 3.0) supports the resourcefulness of plant extracts of Aframomum sceptrum against some plant isolates (Sofowora, 1993). The variations in performance of the extract on each test organism may be as a result of possible synergistic interactions between the active components in the extracts as well biodiversity of the isolates. There is therefore need to harness the potential of this common spice which is eco friendly and biologically degradable to control plant pathogens in nursery fields and on a large scale as it would help ameliorate the negative effects of continuous use of synthetic fungicides.

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