Efficacy of *Jatropha curcas* Linn. as fungicides in the control of *Ceratocystis paradoxa* (*Chalara anamorph)* IMI 501775 associated with bole rot of *Cocos nucifera* Linn. seedlings

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Abstract: The study was aimed at evaluating the antifungal properties of the ethanolic extracts of *Jatropha curcas* seeds and various fungicides; benlate, captan, difolatan and dithane M-45 against the causative agent of bole rot of coconut seedlings; *Ceratocystis paradoxa.* Five different concentrations of *J. curcas* seed; 0%, 25%, 50%, 75% and 100% were tested against *C. paradoxa* using the pour plate method. The extract of ethanol of the *Jatropha* seed showed a maximum zone of inhibitory potentials at 100% (0.5mm) and 75% (0.5mm) compared to others which did not produce zone of inhibition. The various fungicides were also tested for antifungal properties at different concentrations; 500ppm, 1000ppm, 1500ppm and 2000ppm against *C. paradoxa*. Amongst the fungicides tested, benlate and captan had higher zones of inhibition of 0.5mm and 0.5mm respectively for all the concentrations. This result therefore confirms the efficacy of benlate (0.5mm) and captan (0.5mm) as well as 75% and 100% concentration of ethanolic extract of *Jatropha curcas* seeds as the best selective control measure in management of *Ceratocystis paradoxa*.

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1. INTRODUCTION

The Coconut palm (*Cocos nucifera* Linn.) is a member of the palm family *Arecaceae*. It is undoubtedly the most economically important plant in the family *Arecaceae*. (Nair *et al*., 2003); and the most useful palm in the agrarian economy of many nations' worldwide; providing food, drink, shelter and raw materials for industries (Nair *et al*., 2003). It is also used as ornamental and food crops, providing oil, coconut milk, fibre from the husk, palm wine and timber for furniture and construction (Uwubanmwen *et al.,* 2011).

Coconut water serves as juice drink. The white meat (copra) is processed to produce coconut milk, dessicated coconut, coconut powder, cosmetic and pharmaceutical (MARDI, 2000). Coconuts are propagated solely by seed. Apart from copra and coconut oil, the finished products obtained from the coconut plant serves as a major means of export, such as desiccated coconut, copra meal, coco-chemicals (fatty acids, fatty alcohol, methyl ether) shell charcoal and activated carbon, fibre products, coconut cream and coconut milk powder (Punchihewa and Arancon, 2000). The seed-nut has no dormancy and requires no special treatment to germinate (Ojomo *et al.*, 1987).

Fungi that have been found to be associated with bole rot in Nigeria are: *Phytophthora palmivora, Ceratocystis paradoxa,* and *Microphomina phaseolina;* the disease is often fatal to palms up to eight years old. It spreads readily through the soil by root contact between palms and probably by air-borne basidiospores (Ojomo *et al.*, 1987). In Nigeria, bole rot is commonly caused by the fungus *Ceratocystis paradoxa.* It affect seedlings in ground bed or poly bags in the nursery after germination from plumule sheath from fourth to sixth leaf stage that lack adequate maintenance (Ojomo *et al.*,1986). The disease symptoms have been observed to manifest during the dry season, becoming acute in prolonged dry spells. The symptoms are similar to that of the blast of oil palm seedlings (Ojomo *et al.*, 1991).

Ceratocystis paradoxa is a pathogen of tropical crops notably palms like *Borassus flabellifera*, *Cocos nucifera*, *Elaeis guineesis* and crops like *Carica papaya* and *Ipomea batatas* (Nag Raj, and Kendrick, 1975). *Ceratocystis paradoxa* is known by several names such as *Thielaviopsis paradoxa*, *Chalara paradoxa* (Chase and Broschat, 1991). It belongs to the Ascomycetes group. *Ceratocystis paradoxa* is mainly soil borne. The pathogen can be spread from soils to coconut stems by splashing rain or irrigation water. Growth cracks on the coconut trunk, severe downpours, water stagnation, imbalances in nutrition, excess salinity, and plant stress can act as predisposing and aggravating factors (Elliot *et al.*, 2004).

Jatropha curcas Linn. (physic nut, purging nut) is a shrub or a small tree belonging to the family

Euphorbiaceae. Jatropha, a drought-resistant shrub or tree, which is widely distributed in the wild or semi-cultivated areas in Central and South America, Africa, India and South East Asia (Cano-Asseleih, 1986; Cano-Asseleih *et al*., 1989). Linnaeus (1753) was the first to name the physic nut *Jatropha* Linn. in "Species Plantarum". The genus name *Jatropha* derives from the Greek word "*jatros"* (doctor) and "*trophe"* (food), which implies medicinal uses. Jatropha grows to about 5-6 m height. It is a monoecious plant, meaning that the same plant bears both male and female flowers in the same cluster.

The first commercial applications of *Jatropha* were reported from Lisbon, where the oil imported from Cape Verde was used for soap production and for lamps. In addition to being a source of oil, Jatropha also provides a meal that serves as a highly nutritious and economic protein supplement in animal feed, if the toxins are removed (Becker and Makkar, 1998). The plant can be used to prevent soil erosion, to reclaim land, grown as a live fence, especially to exclude farm animals and also planted as a commercial crop (Heller, 1996). Various parts of the plant are of medicinal value, its bark contains tannin, the flowers attract bees and thus the plant has a honey production potential. Its wood and fruit can be used for numerous purposes including fuel. It is easy to establish and it grows faster.

2. MATERIALS AND METHODS

2.1 Source and collection of plant materials / fungicides

Infected coconut seedlings used in this study were collected aseptically in a sterile polyethene bag from Nigerian Institute for Oil Palm Research (NIFOR) nursery in

2.2 Sources of Fungicides: Benlate, Captan, Difolatan and Dithane M-45; were collected from chemical store of plant pathology division of the Nigerian institute for Oil Palm Research (NIFOR), Benin city.

2.3 Isolation and identification of the pathogen

Isolation was done according to the method of Narayanasamy (2011). Small portions of rotted and healthy parts, measuring 5mm in diameter were cut with sterilized scalpel from the infected bole, and point of emergence. These were sterilized in 0.1% mercuric chloride solution for 2 mins and rinsed in three changes of sterile distilled water, dried with sterile filter paper and teased before plating in Petri dishes containing potato dextrose agar (PDA) medium. Petri dishes were incubated in an incubator at room temperature of 25°C ± 2°C for 3-7 days. After the period of incubation, different colonies of fungal associated with bole rot were aseptically subcultured using flamed inoculating loop into a sterile plate containing PDA. The spores of the pathogen were taken from the pure culture and temporary slide mounts were prepared in lactophenol. Pathogens were identified based on their mycelia orientation and production of pigments on potato dextrose agar; production of sporodochia, microconidia, conidia, and phialides aided by the magnification from the scanning electron photomicrograph and description given by Narayanasamy (2011), manual of Commonwealth Mycological Institute (CMI) on Identification of Fungi and Bacteria. Cultured sample was also sent to the Commonwealth Mycological Institute England for confirmation.

2.4 Soil analysis

Analysis of soil samples was carried out to determine the difference in microbial load of the soil samples. 1g of both sterilized and unsterilized soil sample were collected and labeled appropriately. The collected samples were serially diluted and plated in potato dextrose agar (for fungal growth) and nutrient agar (for Bacteria growth). The microbial load was there after determined using Gram staining techniques for Bacteria and lactophenol in cotton blue staining method for fungi.

2.5 Preparation of spore suspension

Spore suspension were prepared in two ways depending on the purposes, either for inoculation or intended for germination. For inoculation experiments on plant materials, spore suspension of each isolates was prepared according to the method of Yadahalli (2005). Four plates of 4-14days old culture of the isolate, was blended using waring blender in 1000ml of sterile distilled water while for the germination process, depending on the amount of inoculum load required, 75mls of prepared liquid broth was dispensed into ten (10) 100mls conical flasks each and was inoculated with the test organisms and incubated at room temperature for about 10 -14days to allow sporulation of the test organism before being used for inoculation of plant materials. The spore suspension was filtered through two layers of cheese cloth and a millipores filter into a conical flask. A range of spore load from 4.1×10^5per ml was used as standard for pathogenicity inoculations.

2.6 Pathogenicity test

A Completely Randomized Design (CRD) with four replicates was used for the field experiment. The most occurring and pathogenic fungi *Ceratocystis paradoxa* in bole rot were tested for

pathogenicity using the method of Narayanasamy, (2011). Thirty two (32) healthy coconut seedlings of 5months old of different cultivars (WAT, DG, YD and DR) planted in sterilized soil were used for this study. Sixteen healthy coconut seedlings (composition of the different cultivars) were inoculated by surgically exposing the young tender spear leaf base and injecting 20ml of inoculum suspension of 4.1×10^5 spores/ml into the bud and outer tissues were then repositioned and tightly wrapped with string to prevent drying which served as the treatment and the other sixteen (16) coconut seedlings (same composition as above) used for the control were inoculated with sterile distilled water. To confirm the presence of the test fungus in the inoculated coconut seedlings, healthy and infected coconut seedlings were taken to the laboratory for diagnosis. Both seedlings were cut into two equal halves with the aid of a sterile knife and symptoms were looked out for in both the healthy and infected seedlings. Samples were collected from the healthy and infected seedlings and surface sterilized. The tissues were then plated on sterilized PDA and incubated at room temperature. Observations were recorded starting from five weeks after inoculation for symptoms such as wilting of the leaves, failure of germination of the central spear leaf etc.

Re-isolation of the pathogen was made from the infected bole. The culture obtained was compared with the original culture for confirmation.

2.7 IN-VITRO EVALUATION OF PLANT EXTRACTS

2.7.1 Ethanol Extract

The method of extraction used was according to the procedure described by Arekemase *et al.,* (2011). Five grams of the Jatropha seed grinded into a powdered form was measured into a conical flask and 20ml of 95% ethanol was added, covered with a cork, mixed together and left on the shaker at 100 r.p.m. for 24 hours after which the extract was filtered and squeezed through four layers of muslin cloth. The filtrate was then centrifuged at 2,000 r.m.p. for 5 minutes after which it was decanted. The pellet was discarded and the supernatant was sterilized by using the membrane filtration unit with type HC filters. The filtrate obtained was stored in sterile McCartney bottles and kept in the refrigerator at 4^oC and later used for the antifungal tests.

2.8 Phytochemical screening of the extracts of *Jatropha curcas* seeds

The methods described by Odebiyi and Sofowora (1978) were used to test for the presence of saponins, tannins, phenolics and alkaloids, Lieberman Burchad reaction as described by Harborne (1973) was used to test for steroids, while the Salkowski test was used to test for the presence of glycosides.

- **Testing for saponins:** Each extract (0.5g) was mixed with water in test tube. Foaming which persisted on warming was taken as an evidence for the presence of saponins.
- **Testing for tannins and phenolics:** Each extract (0.5g) was separately stirred with 10mL of distilled water and then filtered. Few drops of 5% $FeCl_3$ reagent was added to the filtrate. Blue-black or blue-green colouration or precipitation was taken as an indication of the presence of phenolics and tannins.
- **Testing for alkaloids:** Each extract (0.5g) was stirred with 5mL of 1% HCl on a steam bath. The solution obtained was filtered and one 1mL of the filtrate was treated with a few drops of Mayer's reagent. The turbidity of the extract filtrate on addition of Mayer's reagent was taken as evidence of the presence of alkaloids in the extracts.
- **Testing for steroids:** 0.5g of each extract was separately added with 5 drops of acetic anhydride and then a drop of concentrated H_2SO_4. The mixture was steamed for 1 hour and neutralized with sodium hydroxide (NaOH), followed by the addition of chloroform. The appearance of a blue-green colour indicated the presence of steroid.
- **Testing for glycosides:** 0.5g of each extract was dissolved in 2ml of chloroform. Tetraoxosulphate VI acid (H_2SO_4) was carefully added to form a lower layer. A reddish brown colour at the interface indicates the presence of a steroidal ring, that is, a glycone portion of the cardiac glycosides.

2.8.3 Antifungal activity assay

The antifungal testing was carried out using two methods the pour plate and the direct plate methods. The antifungal activity was determined by measuring the mycelia growth of *C. paradoxa* (colony diameter) on each of the PDA plate by drawing two perpendicular lines which meets at a right angle at the centre of the plates. After each day of growth, the diameter of growth was measured using a metre rule along the lines of growth. Antifungal action of the extract was calculated using the formula below;

$$\text{Growth inhibition (\%)} = \frac{\text{colony diameter of (control-treatment)}}{\text{Colony diameter of control}} \times 100$$

2.8.4 Preparation of concentration of the crude extract

The concentration of the crude extract was prepared by dispensing 0mls, 2.5mls, 5mls, 7.5mls and 10mls of distilled water into a McCartney bottle. This was then sterilized in an autoclave for 121^{0}C for 15minutes. After sterilization, each of 10mls, 7.5mls, 5mls, 2.5mls and 0ml of the crude extracts were added to obtain 100%, 75%, 50% 25% and 0% of the crude extract respectively.

2.8.5 Pour plate method

Percentage concentration of the crude extract of *Jatropha curcas* in five McCartney bottles containing 100%, 75%, 50%, 25% and 0%(control) were set up. With the aid of a sterilize syringe, 1ml each from the prepared concentration of the crude extract was added into labelled sterilized Petri dishes of 100%, 75%, 50%, 25% and 0% (control) in an inoculating chamber, and 9mls of PDA was poured into these Petri dishes. After the medium has gelled, with the aid of a sterilized inoculating loop, the fungal mycelial disc was then used to transfer the fungal disc onto the centre of the plates.

2.8.6 In-vitro evaluation of fungicides on the test pathogen

The fungicides used are; Benlate, captan, difolatan and dithane M-45.

10mls of each fungicide obtained at different concentrations (i.e. dissolved solution) was introduced aseptically into 90mls of sterilized potato dextrose agar at room temperature after the introduction of chloramphenicol to inhibit the growth of bacteria. This was carefully swirled to attain homogenization status. Thereafter, 10mls of the resulting mixtures were aseptically dispensed into sterile Petri dishes. A sterile cork borer measuring 5mm in diameter was used to introduce the test pathogen unto the control medium (without fungicide) and treatment medium (with fungicide) and the cultures were incubated at room temperature for 7days after which radial growth measurements were recorded daily.

2.8.7 Statistical analysis

The data collected were analysed using the Minitab statistical package while means were separated using the least significant difference (LSD) test at 5% level of significance.

3. Results

Seven (7) different pathogenic fungi isolated from the infected coconut seedlings and soils rhizosphere of infected seedlings. Fungi isolated include; *Ceratocystis paradoxa, Botryodiplodia theobromae, Aspergillus niger, Aspergillus flavus, Penicillium sp., Fusarium sp* and *Aspergillus ochraceus* whereby *Ceratocystis paradoxa* was found to be the most occurring isolates. *paradoxa* on potato dextrose agar was initially greyish white later turned to dark olivaceous green with smooth margin. The fungus sporulated heavily on it.

Table 4.1, shows the level of sporulation of each isolates on potato dextrose agar (PDA). Result obtained from this table shows that *Ceratocystis paradoxa, Botryodiplodia theobromae* and *Aspergillus niger* had a good sporulation compared to *Aspergillus flavus* and *Fusarium sp.* which had a moderate sporulation followed by *Penicillium sp* and *Aspergillus ochraceus* which had a scanty sporulation on potato dextrose agar after 7days incubation.

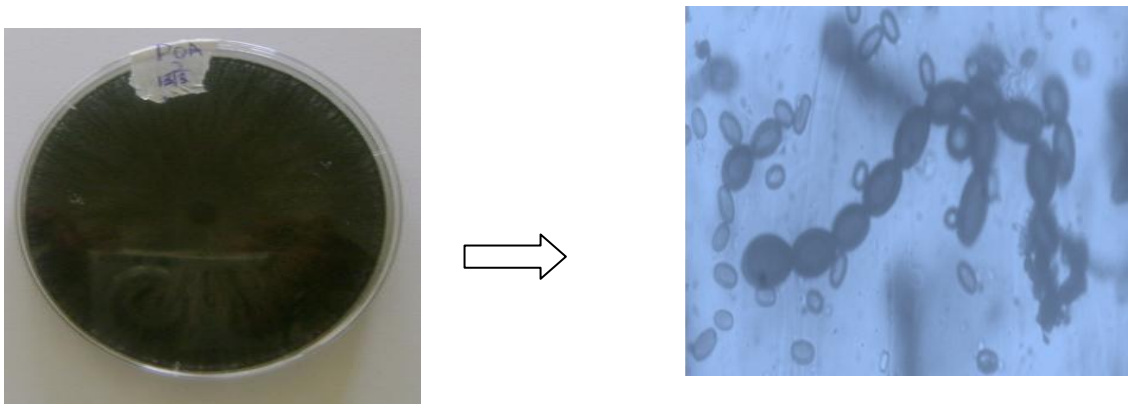

Plate 4.1a: Appearance of *Ceratocystis paradoxa* on PDA after 3-7days incubation.
b: Photomicrography appearance of *C. paradoxa* on scanning electron photomicroscope showing the microconidia and macroconidia spores.

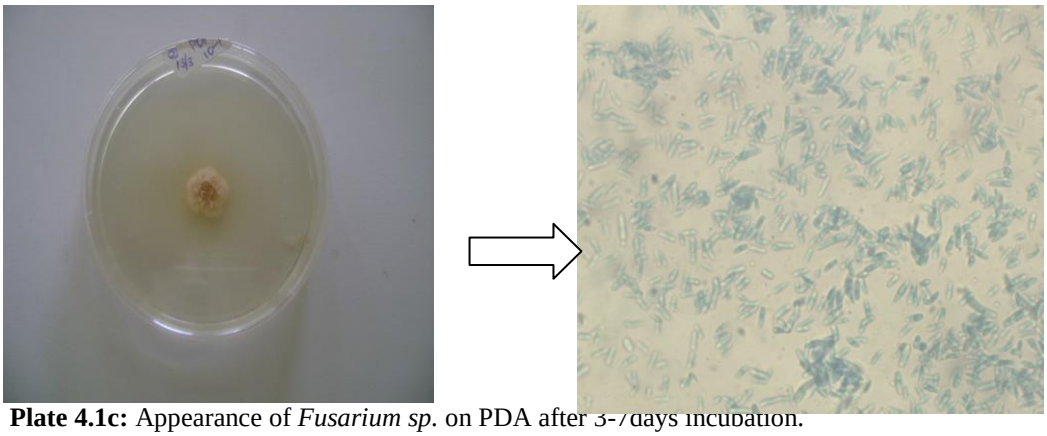

Plate 4.1c: Appearance of *Fusarium sp.* on PDA after 3-7days incubation.
d: Photomicrography appearance of *Fusarium sp* on scanning electron photomicroscope showing the microconidia and macroconidia spores.

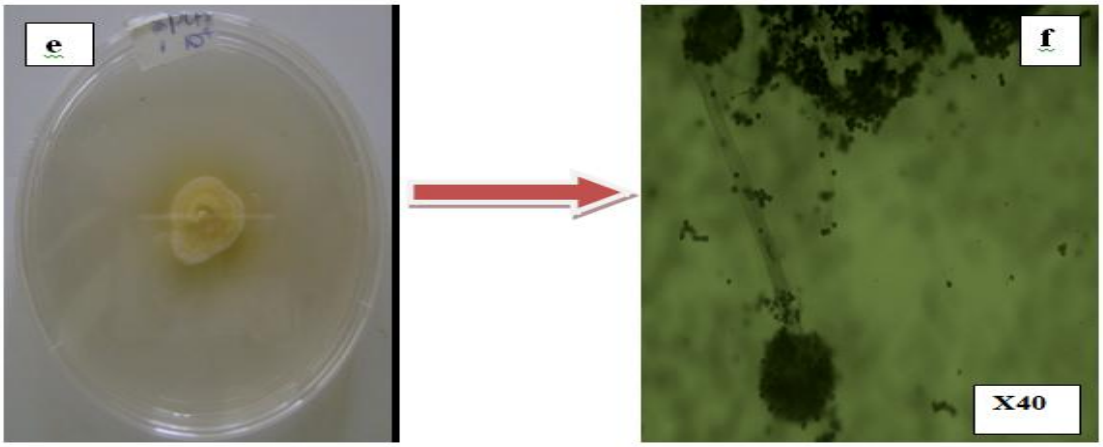

Plate 4.1e: Appearance of *Aspergillus ochraceus* on PDA after 3-7days incubation.
f: Photomicrography appearance of *Aspergillus ochraceus* on scanning electron photomicroscope showing the microconidia and macroconidia spores.

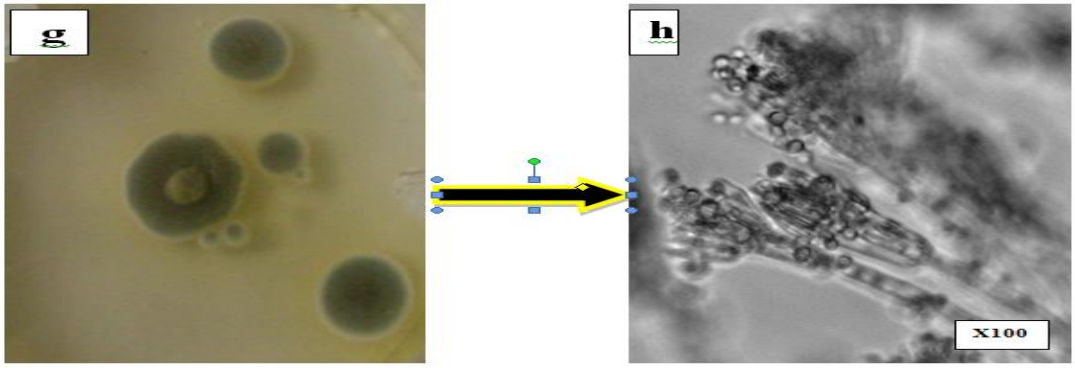

Plate 4.1g: Appearance of *Penicillium sp.* on PDA after 3-7days incubation.
h: Photomicrography appearance of *Penicillium sp* on scanning electron photomicroscope showing the hyphae, phialides and conidiophores.

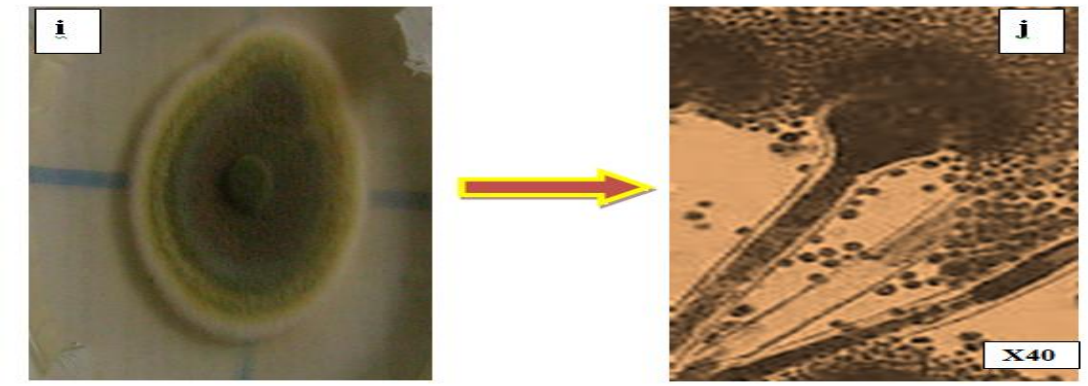

Plate 4.1i: Appearance of *Aspergillus flavus* on PDA after 3-7days incubation.
j: Photomicrography appearance of *Aspergillus flavus* on scanning electron photomicroscope showing the hyphae, phialides and conidiophores.

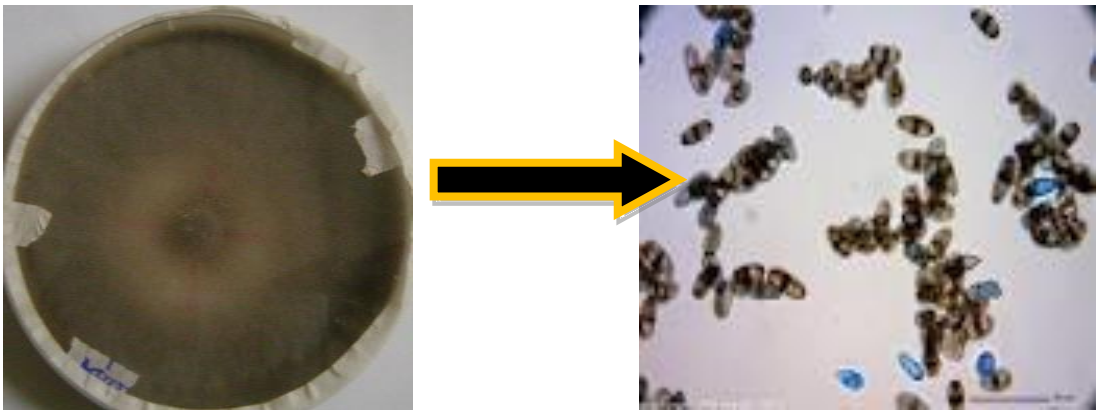

Plate 4.1k: Appearance of *Botryodiplodia theobromae* on PDA after 3-7days incubation.
l: Photomicrography appearance of *Botryodiplodia theobromae* on scanning electron photomicroscope showing the broken mycelium/hyphae.

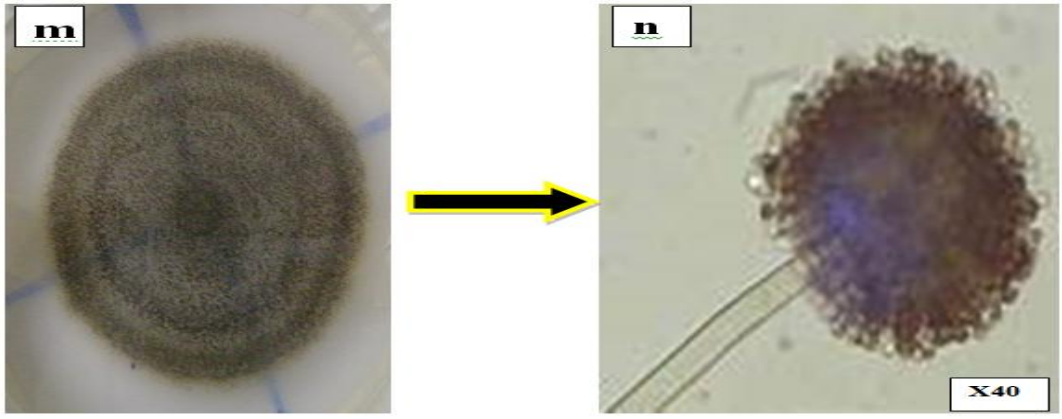

Plate 4.1m: Appearance of *Aspergillus niger* on PDA after 3-7days incubation.
n: Photomicrography appearance of *Aspergillus niger* on scanning electron photomicroscope showing the conidia.

Table 4.1: level of sporulation of fungal isolates obtained from infected coconut seedlings on pda incubated at room temperature.

S/No.	ISOLATES	LEVEL OF SPORULATION
1.	*Ceratocystis paradoxa*	+++
2.	*Fusarium sp.*	++
3.	*Penicillium sp.*	+
4.	*Aspergillus flavus*	++
5.	*Botryodiplodia theobromae*	+
6.	*Aspergillus niger*	+++
7.	*Aspergillus ochraceus*	++

KEYNOTES

+++ - Good sporulation (More than 75 spores per microscopic field)

++ - Moderate sporulation (More than 25 to 74 spores per microscopic field)

\+ - Scanty sporulation (More than 1 to 24 spores per microscopic field)

The results obtained from the pathogenicity test shown in plate 4.2 shows that the test fungus *Ceratocystis paradoxa* was able to initiate the same disease symptoms on the treated seedlings after eight (8) weeks of inoculation. Although, the level of variations of disease incidence varied amongst the four different cultivars when assessed morphologically. Results obtained from re – isolation shows that the fungus (*Ceratocystis paradoxa*) obtained initially were re-isolated only in the inoculated (treated with the test fungus) seedlings and there was absence of the fungus in the healthy seedlings.

Plate 4.2a-c: Pathogenicity of *Ceratocystis paradoxa* in West African Tall coconut seedlings at 5weeks and 8weeks after inoculation.

a.) Control
b.) Inoculated seedling at 5weeks after inoculation.
c.) Inoculated seedling at 8weeks after inoculation.

Plate 4.2d-f: Pathogenicity of *Ceratocystis paradoxa* in Dwarf red coconut seedlings 5weeks and 8weeks after inoculation.

d.) Control
e.) Inoculated seedling at 5weeks after inoculation.
f.) Inoculated seedling at 8weeks after inoculation.

Plate 4.2g-i: Pathogenicity of *Ceratocystis paradoxa* in Dwarf green coconut seedlings 5 weeks and 8weeks after inoculation.

g.) Control
h.) Inoculated seedling at 5weeks after inoculation.
i.) Inoculated seedling at 8weeks after inoculation.

Plate 4.2j-l: Pathogenicity of *Ceratocystis paradoxa* in Dwarf yellow coconut seedlings 5weeks and 8weeks after inoculation.

j.) Control
k.) Inoculated seedling at 5weeks after inoculation.
l.) Inoculated seedling at 8weeks after inoculation.

Plates 4.2m-o: Longitudinal sections of the nuts of coconut seedlings showing symptoms of the bole rot.

Tables 4.2 show the effect of phytochemical screening of *J. curcas* seeds on the growth of *Ceratocystis paradoxa* after three (3) days of incubation. The ethanolic extract was strongly positive for the presence of Alkaloids, Saponins, Tannins, Steroids, while it was slightly positive for Glycosides (Table 4.2). The presence of these phytochemicals is indicative of antifungal activity.

Table 4.2: Phytochemical screening of *Jatropha curcas* seeds

S/n	Phytochemicals	Indicator level
1.	Alkaloids	++
2.	Saponins	++
3.	Tannins	++
4.	Steroids	++
5.	Glycoside	+

Key: + = Slightly Positive
++ = Strongly Positive

Results obtained from table 4.3, indicated a positive significant difference (P≤0.05) amongst the various levels of concentration tested using crude extracts from *Jatropha curcas* seeds on the growth of *C. paradoxa.* The effect of the crude extract showed a remarkable influence on the growth of the fungus at 100% and 75% concentration. A less significant effect of the crude extract was observed at 25% concentration followed by 50% concentration (P≤0.05) which had a minimal inhibitory effect at 5% level of significance (P≤0.05).

Table 4.3: Effect of the crude extract of *J.curcas* seeds on the mycelia growth of *C. paradoxa* after three days incubation on PDA at room temperature 25°C±2^{0}C.

Days	100% concentration	75% concentration	50% concentration	25% concentration	Control experiment
1	0.50 ± 0.00^a	0.50 ± 0.00^a	0.78 ± 0.03^b	1.53 ± 0.03^c	3.15 ± 0.26^d
2	0.50 ± 0.00^a	0.50 ± 0.00^a	1.83 ± 0.25^b	3.33 ± 0.00^c	7.07 ± 1.10^d
3	0.50 ± 0.00^a	0.50 ± 0.00^a	1.88 ± 0.33^b	8.00 ± 0.00^c	8.50 ± 0.00^d

Keynotes: * Values in the same row with different letters in superscripts are significantly different according student t-test (p≤0.05). Each value is a mean of three replicates ± S.D.

Table 4.4, shows the percentage (%) growth inhibition of the fungus at different levels of concentration. Complete zone of inhibition of the fungus by the crude extract of *Jatropha curcas* seeds was noticed at 100% and 75% while at 50% concentration, moderate inhibition of the growth of the fungus was observed. However, least inhibition of the fungus was noticed at 25% concentration (P≤0.05). Growth of isolate at different concentrations on PDA at 25%, 50%, 75%, 100% and control are shown in plate 4.4.

Table 4.4: The percentage (%) growth inhibition of the crude extract of *J.curcas* seed on *C. paradoxa* at room temperature 25°C±2^{0}C.

Days	100% concentration	75% concentration	50% concentration	25% concentration
1	100%	100%	65.56%	32.22%
2	100%	100%	88.14%	25.93%
3	100%	100%	88.14%	5.88%

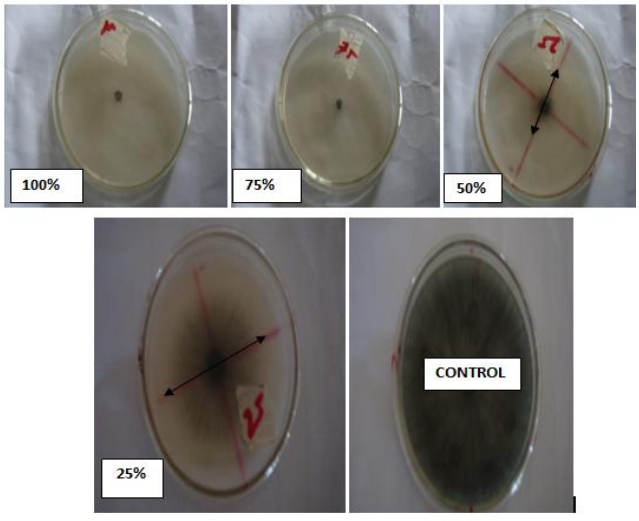

Plate 4.3: Percentage (%) growth inhibition of *C. paradoxa* after three days incubation at room temperature 25°C±2°C

From figures 3.1-3.4, two systemic and two non-systemic fungicides were evaluated for their efficacy against *Ceratocystis paradoxa* in the laboratory. The data obtained from this study revealed that there was significant difference among the systemic fungicides in inhibiting the mycelia growth of *Ceratocystis paradoxa*. Systemic fungicides viz; Benlate and Captan were effective in complete inhibition (100%) of the fungus at the different concentrations tested (P≤0.05); whereas, Difolatan and Dithane M-45 were not effective (P≤0.05) at the different concentrations tested (Figures 3.1a-d). At the increasing order of 500ppm-2000ppm, benlate and captan were highly effective in inhibiting the mycelia growth followed by Difolatan at the decreasing order of concentration 2000ppm,1500ppm,1000ppm and 500ppm respectively (P≤0.05). Least inhibition of the fungus growth was noticed in Dithane M-45 at a concentration of 2000ppm, 1000ppm, 1500ppm and 5000ppm (P≤0.05).

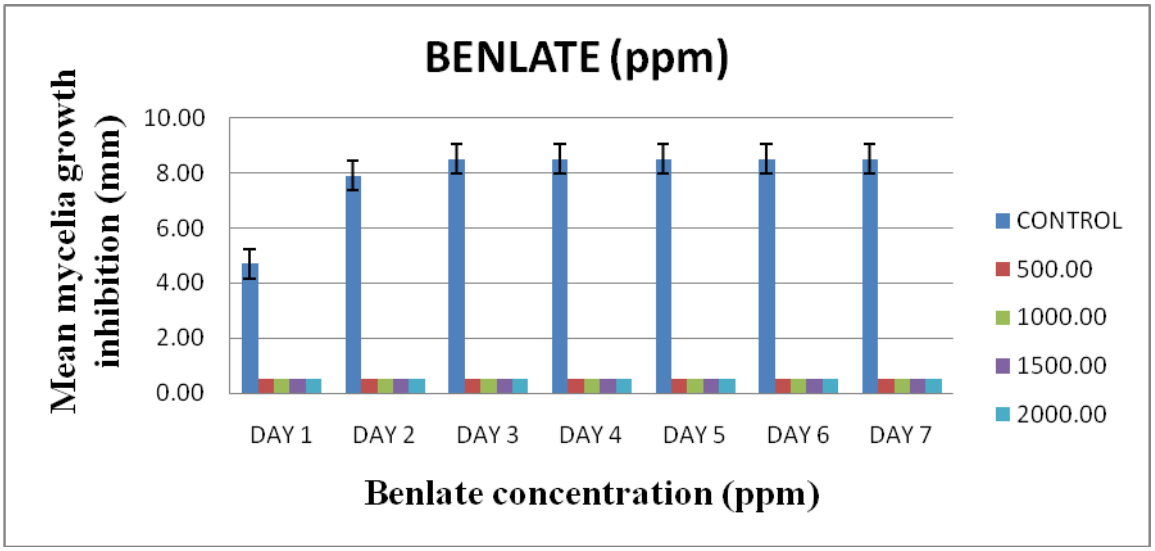

Fig. 3.1: Effect of benlate on the growth of *C. paradoxa* after 7days incubation at room temperature 25°C ± 2°C.

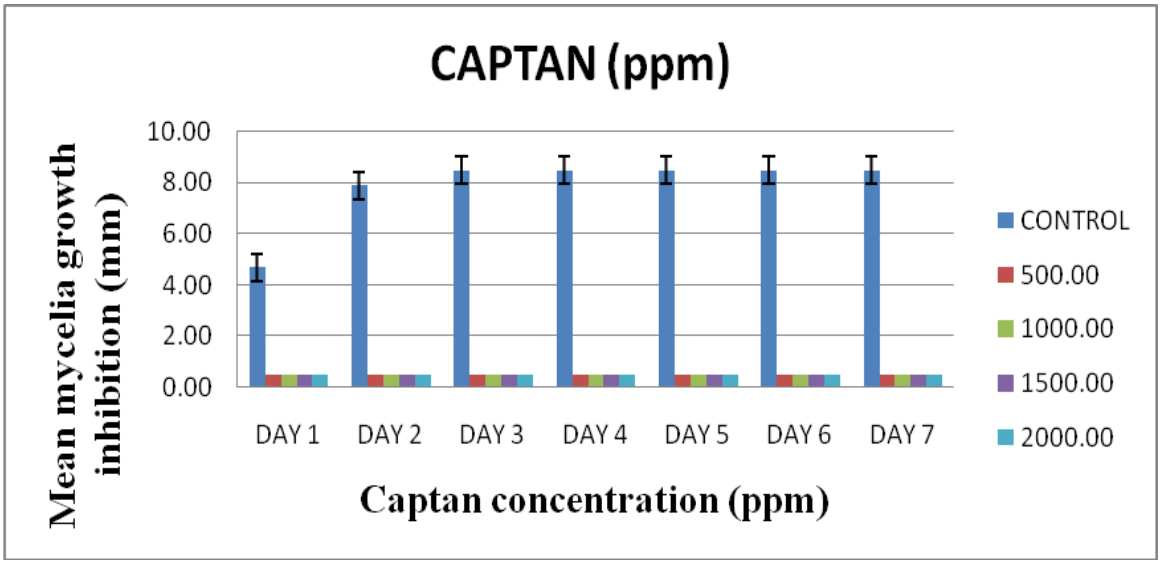

Fig.3.2: Effect of captan on the growth of *C. paradoxa* after 7days incubation at room temperature 25°C ± 2°C.

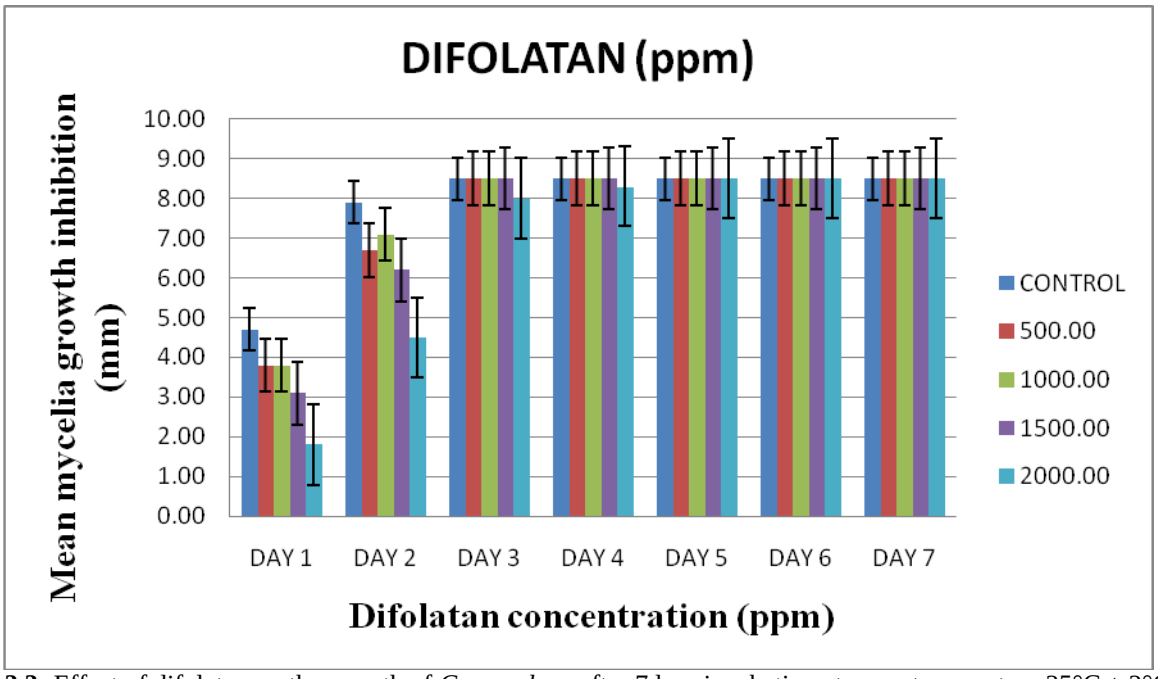

Fig. 3.3: Effect of difolatan on the growth of *C. paradoxa* after 7days incubation at room temperature 25°C ± 2°C.

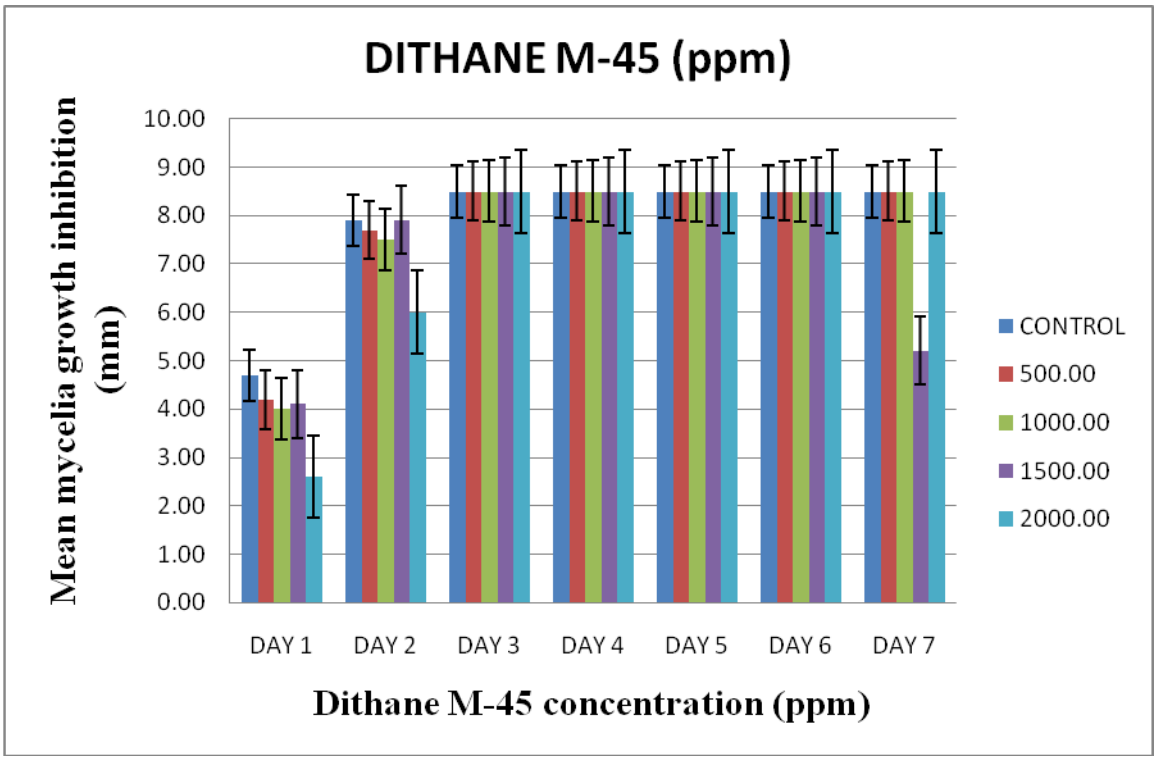

Fig.3.4: Effect of dithane M-45 on the growth of *C. paradoxa* after 7days incubation at room temperature 25°C±2°C.

4. Discussion

The microscopy of the pathogen (*Ceratocystis paradoxa*) revealed the presence of mycelium, conidiophores, microconidia, and macroconidia; that are dark greenish black, thick walled and in chains from short lateral hyphal branches. These characters are in agreement with those described by Singh *et al*., (1991). Hence, affirming the identity of the fungus.

Symptoms of the disease were observed after eight weeks of transplanting during the pathogenecity test conducted in-vivo. The symptoms of the disease observed include; failure of the spear leaf to germinate, wilting of the young seedlings, rotting and blackening of the bud as well as basal portion of the stem were. These findings were in accordance with those reported by Agnihotri (1983), Singh *et al*., (1991) and Rao *et al*., (1995).

The in-vitro control of *Ceratocystis paradoxa* using chemical based fungicides showed that Benlate and Captan significantly reduced the effects of the Fungus ($p \leq 0.05$), from the very first day of the experiment; showing 100% level of mycelia inhibition by these chemicals and 0% level of tolerance by the organism. Difolatan and Dithane M-45, were not significantly effective in the reduction of the pathogenic activities of *C. paradoxa* ($p \leq 0.05$), compared to the control experimental set up; as 100% level of tolerance to the chemical based fungicides was recorded on potato dextrose agar treated with these fungicides. The findings from this report was synonymous to the report of Pandu *et al*.,(1986) who reported a reduction in the radial mycelia growth of *C. paradoxa* using benlate and captan. The result obtained for Difolatan and Dithane M-45 was in contrast with the findings of Ojomo *et al.,* (1993), who stated that these chemicals inhibited the mycelia growth of the fungus.

The introduction of phytofungicides (plant based extracts) obtained from *Jatropha curcas* effectively reduced the pathogenic activities of *C. paradoxa;* with 100% mycelia inhibition at day 3 ($p \leq 0.05$), significantly distinct from the control experiment. This was in line with the findings of (Tarun Agarwal *et al.,* 2012).

It was observed from the experiment conducted that the continuous use of chemicals such as; fungicides in the management of the diseases also poses toxic and hazardous effect to the soil, water and air as well as development of resistance of the pathogen against chemicals and harmful effects on non-target organisms. This has brought about the under development of highly polluted environment.

Hence, the use of non-pollutants, renewable, indigenously available, easily accessible, non-phytotoxic and relatively economical methods of plant protection is needed in integrated disease management strategies. Researchers have extensively studied the biological properties of *Jatropha curcas* and their results showed that this plant is ethno-medically valuable (Tarun Agarwal *et al.,* 2012). The presence of phytochemical constituents could be attributed to antifungal potentials of the extracts of *Jatropha curcas* seed.

Biological control is an effective, Eco-friendly and alternative approach for any disease management practice. Hence, with the increased awareness of the adverse effects of plant protection chemicals on the environment, particularly in coconut plantations, the need to manage the disease using biological approaches has assumed more significance.

In spite of certain limitations, there are specific merits in adopting biocontrol practice, for the management of coconut diseases; due to the fact that it is one of the important commercial crops grown throughout the year in a large areas on a plantation scale, it enables easy application of phytofungicides which renders them self sustainable without interruption. Thus, any investment would be suitably rewarded in terms of higher production and productivity.

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