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STUDIES ON THE EFFECTS OF PLANT EXTRACTS ON PAWPAW FRUIT (*Carica papaya* L.) ROT FUNGI IN GIREI AND YOLA SOUTH LOCAL GOVERNMENT AREAS OF ADAMAWA STATE

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ABSTRACT

A study for fungal post-harvest rot pathogens of pawpaw fruit rot was investigated in three markets in Adamawa state, (Sabon Gari, Voniklang and Yola town market) with objectives to identify the fungal pathogens of pawpaw, to determine the incidence and the severity of the pathogens and to determine the in-vitro and in-vivo control of the three plant extracts (garlic oil, neem oil and ash) on the pathogens. Thirty six pawpaw fruits were randomly collected from the three markets and taken to laboratory of Plant Science, Modibbo Adama University of Technology Yola. Industrial potato dextrose agar (PDA) was used for the isolation of fungi. Completely randomized design (CRD) was used in

Introduction:

Papaya is a useful plant with nutritional, medicinal and health benefits (Danielone, 1997). In spite of all these benefits, the plant is besieged by lots of pathogens both in the field and post-harvest storage and these diseases result in yield losses thus making its valuable components unavailable (Danielone,1997). These parts of the plant are however, infested with pathogens and result in huge yield losses making the desired raw materials unavailable (Morton, 1987). The use of synthetic chemicals is now known to be

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three replicates for each of the treatment. Data collected were subjected to Analysis of Variance (ANOVA) and least significant difference (LSD) separated means that were significant. The symptoms of the disease were observed and associated organisms isolated and identified through pathogenicity test and the following pathogens were identified; Aspergillus niger, Aspergillus flavus and Mucor spp. A severity test on the pathogens was carried out with the sample size of twelve pawpaw fruits by measuring the level rot of the pathogens. Result showed (p=0.0001) that Mucor spp was the most severe. The incidence pawpaw fruit rot in three markets revealed a high percentage rot in Sabongari market, Aspergillus niger (38.4), Aspergillus flavus (18.2) and Mucor spp (47.5). Control trials both in-vitro and in-vivo with garlic oil, neem oil and ash were found promising, and their different concentrations (1ml, 2ml and 3ml) were also found promising and the effectiveness of the control increased as the oils and quantities of ash increased. Essential oil of garlic, neem oil and ash extracts proved effective in the control of pawpaw fruit fungal rot and are recommended as an alternative to synthetic fungicides which are often hazardous and costly.

Keyword: *carica papaya, Aspergillus niger, flavus, mucor, garlic, neem, ash concentration, invivo and invitro.*

dangerous to the environment. While use of plant extracts or plant oil is safer and cheaper to be used and handled by farmers (Olowo *et al.,* 2003). There is need to employ the use of plant oil and plant extract in controlling the disease of plant.

Garlic oil was reported by (Milner, 2001) to have antimicrobial substance such as volatile oil; these volatile compounds are generally considered to be responsible for most of the pharmacological properties and contain at least 33 sulphur compounds like aliin, alicin, ajone, dially, sulphide, and others. Neem crude oil has been used by small scale farmers to protect seeds, grains and legumes (Suleiman *et al.*, 2013). Ash has been used to control fungal pathogens of plantain (Channya, 1991). Few studies have been done by different researchers in part of Nigeria, Otuonye and Adedeji

(2006 and 2010) recommend the use of Botanicals such as garlic and neem leaf extract against storage rot pathogens of different fruits.

Several postharvest technologies have been used to control pawpaw fruit fungal rot (Nneka, 2006). The following methods are used to control fungal rot of pawpaw fruits.

Sanitation: This involves any measures that can be taken to reduce the spore load and is regarded as most desirable such as removal or destruction of diseased fruit part is an obvious first step. Removal or destruction of infected parts reduces sources of primary and secondary infection (Wills *et al*., 1998). Environmental manipulation: Environmental factors such as temperature, humidity and atmosphere through various manipulations can be reduced to 0^0C which will inhibit the growth of fungi. (Opeke, 1992).

Chemical treatments: Use of chemical treatments for post-harvest disease control requires knowledge of the fungus and of the mode of action of the chemical. In general, chemicals are most effective during the lag phase before infections become firmly established in the tissues. Post-harvest treatment of Captan 5000ppm is effective against watery rot of papaya (Pathak *et al*., 1976). Field spray of mancozeb reducing *Rhizopus* soft rot incidence by reducing field initiated fruit diseases (Nishijima *et al*., 1990). Use of Bordeaux mixture for *Colletotrichum gloeosporioides* at post-harvest period can reduce the fungal rot of pawpaw.

Hot water treatments: Effect of hot water treatment between 50 - 60°C for a few minutes can kill fungi but can be tolerated by the plant hence care must be taken to ensure good temperature control and duration as injury to the product could result (Morton, 1987) Use of hot water treatment combined with the vapour heat treatment provides adequate control of rot diseases (Morton, 1987).

Biological Control Agents: Use of micro-organisms to control pathogen which may cause plant to produce toxins that inhibit the pathogen and also out compete pathogen for nutritional resources (Emmechebe, 1985). Two products registered in 1995 namely; *Pseudomonas syringae* (Bio-Save) and *Candidia oleophila* (Aspire) can be applied as dips, drenches or sprays on the plant. Planting of disease-resistant cultivars is also a bio- control measure of reducing infection rates (Emechebe, 1985).

Medicinal plant materials have been successfully used for the treatment of fungal and bacterial infections in humans (Akinyosoye and Oladummoye, 2000), suggesting that some plant materials may also possess antifungal and antibacterial constituents that are useful in controlling plant diseases (Amadioha, 1998). Previous reports (Akpomedaye and Ejechi, 1998; Ejechi and Ilondu, 1999) show that spices, herbs and other plant materials possess antifungal activity.

Materials and methods

The study was conducted in the Department of Plant Science Laboratory of Modibbo Adama University of Technology, Yola, in 2019. Yola is located on latitude 7^0 and 11^0 N of the equator and between longitude 11^0 and 14^0 E of the Greenwich meridian (Adebayo, 1999). The area has tropical climate marked by dry and raining seasons. The rainy season commences around May and ends in the middle or late October (Adebayo, 1999).

Source of Samples

Twelve pawpaw fruits samples were purchased/ collected using Completely Randomized Design (CRD) from three different markets in Adamawa state and making the total number of thirty-six. (Yola town,Voniklang and Sabon Gari Sangere). Both the diseased and the healthy papaya fruits were collected in a sterile polythene bag and then taken into the laboratory for further studies. In the laboratory, the diseased fruits were incubated in the sterilized desiccators for the development of rots, to study the characteristics of the rots and observation of spoilage periods for the complete rotting of the fruits.

Incidence of Pawpaw Fruit Rot

Survey on the papaya rot was done in three different markets in Girei and Yola South local Government areas of Adamawa state (Voniklang market, Sabon gari market and yola town market) during the period of February, March and April, 2019. Two shops were taken randomly from each market and twelve pawpaw fruits in each market making the total of 36 pawpaw fruits. Each of the selected areas was surveyed. Diseases were identified by

observing different symptoms of papaya rot described by (Alveraz and Nishijima, 1987) and (Singh *et al.*, 2012). These symptoms were spot, fruit rot, necrosis, water soaked lesion, corky lesion etc. Diseased fruit samples were collected in polythene bags for further studies of the symptoms and confirmation of the respective fungal pathogens. Beside these, total number of fruits, number of diseased fruits, types and number of disease symptoms shown on the fruits surface were taken into consideration. The data were collected and calculated in terms of percentage fungal pathogen incidence by the following formula given by (Singh *et al.*, 2012).

$$\% \text{ Disease Incidence} = \frac{\text{Loss number of diseased fruits affected by particular disease}}{\text{Total number of fruits sampled}} \text{ X } 100$$

Isolation and Identification of Causal Organisms

The infected pawpaw fruit tissues were sectioned into 3mm disc surface sterile using 0.1% sodium hypochloride, for 30 seconds and then rinsed in three changes of sterile distilled water and between sterile filter papers and plated on potato dextrose Agar (PDA).

The plates were incubated at 25ºC for 24 hrs to 7 days and observed for any growth. The stock culture was stored at agar slants in Mc Cartney bottles at 0-4^{0}C. The organisms to be isolated were stained with lactophenol cotton blue viewed under the microscope and subsequently identified, by comparing the morphological characteristics of the organisms using method of Alexopoulus and Mins (1966).

Pathogenicity Test

The healthy pawpaw fruits (*Carica Papaya* L.*)* were used in this experiment. Semi-ripe fruits that appeared healthy and uniform in size were selected and washed to remove any dirt, surface sterilized in 0.1% sodium hypochloride solution for 30 seconds and rinse in three changes of sterile distilled water and then air dried (Liu and Kushalappa, 2002). The fruits were wounded with a cork borer with a diameter of 5mm to a depth of 5mm and bored tissue and were removed. (Choiseul *et al.,* 2007). The wound was sealed with sterile vesper prepared from wax and Vaseline. The controls were set up in the same manner except that 0.2mm of sterile

distilled water was used instead of the inocolum. All the wounded pawpaw fruits were wrapped in black polythene bags (Manici and Cerato, 1994). Inoculated fruits were placed in desiccators at 30°C. All these were carried out under aseptic condition. Regular observations were made and re-isolation of pathogenic organisms was done for comparison with the original isolate, and completely randomized block designed was used and 3 replicates were used.

Preparation of Garlic Oil

Garlic (*Allium sativum*) were collected and weighed up to 50gm. The shell is separated from the bulb manually and the bulb grounded into paste using mortar and pestle and paste mixed with 250mls of water stirred for 24 hours using muslin clothes (Amuchi, 1999).

Preparation of Neem Seed Extract

Dried neem (*Azadirachta indica*) seeds collected and the shells cracked using pestle and mortar to separate the shell from the kernels were grounded into paste, 50gm of the paste was weighed and 250mls of water added and stirred for 10mins and allowed to stand for 24 hours before filtering with muslin cloth (Amuchi, 1999).

Extraction of Powdered Ash and Liquid Ash for Control.

The ash was obtained by burning a desired maize comb (*Zea mays* L). The 200 grams of the powdered ash were placed in a beaker and then sterilized in an oven for three hours. The 100g portion of the powdered ash was mixed with 120mls of distilled water and then filtered using a cheese-cloth, gauze and Whatman filter paper to obtain a liquid ash.

The filtrate was collected in a conical flask. The filtrate ash was used for the disease control treatment (Channya, 1991).

In-vitro control using garlic oil, neem oil and ash

PDA media 500ml glass flask was autoclaved for 15 minutes, after autoclaving, the flask was cooled to about 45^0C. Approximately 5ml of each of the oil were taken and pipetted in the 500ml flask each. A quantity of

250mg of chlorophenicol were dissolved in 2ml of distilled water and then added to 500ml of PDA to prevent bacterial growth. The media containing the oil and chlorophenicol were gently agitated by hand for 2 minutes for proper mixing of the content. Up to 20ml of the mixed media were dispensed into 9cm Petri dishes. Approximately 1ml from one of the colony of the spire of fungal pathogen suspensions (Conc. 1x106 spores/ml) were pipetted on the center of the amended PDA media. The inoculated plates were then sealed with masking tape and then incubated at 28± 2^0C for 24 to72 hours in the month of June 2015. The Petri dishes without the oil served as the control. The experiments were performed under aseptic conditions and replicated three times. Colony diameters of the radial growths were measured from day 3 to day 7. The inhibition zones (p) were calculated using the formula of (Francisco, 2010): p = c – t/c x 100.Where C was the colony diameter of the control and T the treatment (three replicates).

In-vivo control using garlic and neem oil and ash extracts

A healthy semi ripe pawpaw fruit was punctured with a sterile dissecting needle and spores of the most isolated fungi were subjected to it, 1ml, 2ml, and 3ml of garlic oil, neem oil as well as the ash extract were injected into the injured part of the fruit. Some controls were set with garlic oil, neem oil, and ash extract free pawpaw. Three replicates for each treatment were used and inoculated at the centre of the pawpaw fruits. The culture was kept for seven (7) days at room temperature 33±2C and radial growth of the mycelium in each fruit was recorded, (Moshood *et al.*, 2010)

Experimental Design and Data Analysis

The experimental layout was completely randomized design of three treatments. The experiments were replicated three times. All the data were analyzed using analysis of variance (ANOVA) according to (Gomez and Gomez, 1984) and the least significant difference (LSD) at $p<0.0001$) according to (Scheffer,1953) were used in separating significant means. The statistical package used to analyze the result was Statistical Analysis System (SAS) version 7 statistical programme.

RESULT AND DISCUSSION

Incidence of fungal pathogens

The three different fungal pathogens had frequencies which vary, with *Mucor* spp showing the highest incidence of (47.5% in Sabon Gari Market, 45.5% in Voniklang Market and 43.5% in Yola Town market) followed by *Aspergillus niger* (38.4% in Sabon gari market,(36.4%) in Voniklang market and (34.4%) in Yola Town market) and lastly the *Aspergillus Flavus* showing the least incidence (of 18.2% in Sabon gari market, 20.2%) in Voniklang market and 16.2% in Yola Town market) as shown in (Table 1).

Severity of pathogens on pawpaw

The severity of the fungal pathogens responsible for pawpaw fruits rots varied significantly at p=0.0001, with *Mucor* spp having the highest radial growth rot of pawpaw (20.40mm) and *Aspergillus niger* with (18.60mm) and lastly *Aspergillus flavus* with (18.30mm) as shown in (Table 2)

In-vitro trials

Analysis of variance among the three treatments (garlic oil, neem oil and ash) and the control gave a significant difference at p= 0.001. The aqueous extracts and the ash inhibited the growth of the pathogens. However there was no significant difference among garlic oil, neem oil and the ash in (Table 3).

The analysis of variance at p= 0.0001 revealed significant difference among the concentrations of the garlic on the fungal pathogens on *in-vitro* trials (Table 5), 3ml was more effective at controlling *Aspergillus niger* (1.10mm) followed by 2ml (1.47mm) and lastly 1ml (2.47mm), 3ml has the highest inhibition on *Aspergillus flavus* with (1.27mm) followed by 2ml with (1.57mm) and lastly 1m with (2.30mm). 3ml also inhibited the *Mucor* spp with (1.21mm), 2ml (1.58mm) and lastly 1ml (2.29mm).

The analysis of variance at p= 0.0001 revealed significant difference among the concentrations of the neem oil on the fungal pathogens on *in-vitro* trials (Table 7), 3ml was more effective at controlling *Aspergillus niger* (1.24mm) followed by 2ml (1.61mm) and lastly 1ml (2.58mm), 3ml

has the highest inhibition on *Aspergillus flavus* with (1.30mm) followed by 2ml with (1.50mm) and lastly 1ml with (2.44mm). 3ml also inhibited the *Mucor* spp with (1.32mm), 2ml (1.72mm) and lastly 1ml (2.42mm).

The analysis of variance at p= 0.0001 revealed significant difference among the concentrations of the ash on the fungal pathogens on *in-vitro* trials (Table 9), 3ml was more effective at controlling *Aspergillus niger* (1.25mm) followed by 2ml (1.55mm) and lastly 1ml (2.76mm), 3ml has the highest inhibition on *Aspergillus flavus* with (1.18mm) followed by 2ml with (1.40mm) and lastly 1m with (2.64mm). 3ml also inhibited the *Mucor* spp with (1.27mm), 2ml (1.67mm) and lastly 1ml (2.30mm).

The antimicrobial activity of garlic is believed to be due to the effect of allicin, the main ingredient in garlic, generated by the phosphopyridoxal enzyme allinase (Ankri and Mirelman, 1999), and ajoene (Yoshida *et al.*,1987). Garlic oil was reported by (Milner, 2001) to have antimicrobial substance such as volatile oil; these volatile compounds are generally considered to be responsible for most of the pharmacological properties and contain at least 33 sulphur compounds like aliin, alicin, ajone, dially, sulphide, and others. Garlic (*Allium sativum*) is a spice with global recognition. In the present study, it has been shown to inhibit the growth of fungi when *in vitro* tested.

(Azadirachta indica) neem oil have been found to be effective in fungal pathogens by this study are capable of controlling the colony growth of fungal pathogens and has inhibitory effect on microorganisms (Nneka, 2006). Investigation into the antifungal properties of neem crude extracts possesses some inhibitory components which cause significant reduction in mycelial growth of the fungi. This agrees with the results of Amadioha (1998) and (Adejumo *et al.,* 2000) who reported the efficacy of extracts from pesticides. Benlate and Ridomil. Akpa *et al.,* (1991) reported a significant inhibitory property of neem (*Azadirachta indica*) extracts on mycelial growth to reduce the radial growth of *Mucor* spp.

In-vivo trials

Analysis of variance at p= 0.0001 among the three different treatments (garlic oil, neem oil and ash) and the control gave a significant difference.

The aqueous extracts and the ash inhibited growth of the fungi. However there was no significant difference among garlic oil, neem oil and the ash in (Table 4).

There was also a significant difference at p=0.0001 among the levels of concentration of garlic on in-vivo trials (Table 6), 3ml was more effective with (2.13mm) on the control of *Aspergillus niger* followed by 2ml with (2.59mm) and lastly 1ml with (3.43mm). At the *Aspergillus flavus*, 3ml had the highest zone of inhibition with (2.07mm) followed by 2ml with (2.64mm) and lastly 1ml with (3.50mm). At the *Mucor spp*, 3ml had the highest zone of inhibition with (2.53mm) followed by 2ml with (3.13mm) and lastly 1ml with (3.35mm).

The analysis of variance at p= 0.0001 revealed significant difference among the concentrations of the neem oil on the fungal pathogens on *in-vivo* trials (Table 8), 3ml was more effective at controlling *Aspergillus niger* (2.24mm) followed by 2ml (2.76mm) and lastly 1ml (3.76mm), 3ml has the highest inhibition on *Aspergillus flavus* with (2.16mm) followed by 2ml with (2.64mm) and lastly 1ml with (3.48mm). 3ml also inhibit the *Mucor* spp with (2.46mm), 2ml (3.28mm) and lastly 1ml (3.68mm).

The analysis of variance at p= 0.0001 revealed significant difference among the concentrations of the ash on the fungal pathogens on *in-vivo* trials (Table 10), 3ml was more effective at controlling *Aspergillus niger* (2.06 mm) followed by 2ml (2.87 mm) and lastly 1ml (3.63mm), 3ml has the highest inhibition on *Aspergillus flavus* with (2.27mm) followed by 2ml with (2.78mm) and lastly 1ml with (3.39mm). 3ml also inhibited the *Mucor* spp with (1.27mm), 2ml (1.67mm) and lastly 1ml (2.30mm).

Several reports have been made on the fungicidal properties of neem oil (Singh *et al.,* 1980; Kazmi *et al.,* 1995). Locke (1995) reported that in field *Alternaria alternata, Aspergillus niger and Fusarium oxysporum* have been completely controlled by using 2-10% neem oil. According to (Niaz and Kazmi, 2005) neem oil was quite effective on *Aspergillus* spp. Vir and Sharma (1985) found antifungal activity in neem oil against *Alternaria alternata* and *Aspergillus* spp. Sinnah *et al.*, (1983) also studied the toxicity of neem oil on *Aspergillus* spp.The legendary medicinal qualities of the neem tree have been known for a long time and their aqueous leaf extracts have systemic action (Egunjob and Onoyemi, 1981).

The result of this study showed that, ash effectively controlled fungal pathogens of pawpaw fruit rot and significantly decreased the mycelia growth of the fungal isolates and to some extent depending on the concentration of the ash, this confirmed the report of (Sibounnavoung *et al.,*2009) that both fresh and dried mycelia pellets are significantly influenced by pH of the medium. Filamentous fungi are generally known to be tolerant to acidic pH and most of them have an optimum pH of 5.0 and 6.0 (Rosfarizan *et al.,*2000). The significant inhibition and slowing rate of growth of pathogens have also been reported by Channya (1991) and (Oduro *et al.,* 1997). Channya (1991) adduced the effectiveness of ash as mold control to the unsuitable alkaline of pH of ash.

Table 1: Frequency of Occurrence of Pathogen Isolates from Papaya Infected Fruits in three Different Markets of Adamawa State

	Frequency (%)		
Fungal isolates	Sabon Gari market	Voniklang market	Yola town market
Aspergillus niger	38.4	36.4	34.4
Aspergillus flavus	18.2	20.2	16.2
Mucor spp	47.5	45.5	43.5

Table 2: Radial Growth in (mm) of Pathogens of Papaya Infected Fruits

Fungal pathogens	Virulence (mm)
Aspergillus niger	18.60
Aspergillus flavus	18.30
Mucor spp	20.40
LSD (0.0001)	0.18

Table 3: Mean Effect of Garlic Oil, Neem Oil and Ash on Fungal Pathogens (*In-vitro*)

	Fungal pathogens		
Plant extracts	*Aspergillus niger*	*Aspergillus flavus*	*Mucor* spp
Ash	1.85	1.74	1.74
Neem	1.81	1.82	1.75
Garlic	1.68	1.69	1.68

Control	3.38	3.33	3.31
LSD (0.0001)	0.21	0.21	0.18

Table 4: Mean Effect of Garlic Oil, Neem Oil and Ash on the Fungal Isolates (*In-vivo*)

	Fungal pathogens		
Plant extracts	*Aspergillus niger*	*Aspergillus flavus*	*Mucor* spp
Garlic	2.72	2.74	3.07
Ash	2.85	2.81	3.23
Neem	2.92	2.76	3.14
Control	4.91	4.78	5.35
LSD (0.0001)	0.22	0.23	0.25

Table 5: Effect of Garlic Oil on the Fungal Pathogens (*In-vitro*)

Conc. (ml)	*Aspergillus niger*	*Aspergillus flavus*	Mucor spp
1	2.47	2.30	2.29
2	1.47	1.57	1.58
3	1.10	1.27	1.21
0	3.38	3.38	3.33
LSD (0.0001)	0.21	0.21	0.18

Table 6: Effect of Garlic Oil on the Fungal Pathogens (*In-vivo*)

Conc. (ml)	*Aspergillus niger*	*Aspergillus flavus*	*Mucor* spp
1	3.43	3.50	3.35
2	2.59	2.64	3.13
3	2.13	2.07	2.53
0	4.91	4.78	5.35
LSD (0.0001)	0.22	0.23	0.25

Table 7: Effect of Neem Oil on the Fungal Pathogens (*In-vitro*)

Conc. (ml)	*Aspergillus niger*	*Aspergillus flavus*	*Mucor* spp
1	2.58	2.44	2.42
2	1.61	1.50	1.72
3	1.24	1.30	1.32
0	3.38	3.38	3.33
LSD (0.0001)	0.21	0.21	0.18

Table 8: Effect of Neem Oil on the Fungal Pathogens (*In-vivo*)

Conc. (ml)	*Aspergillus niger*	*Aspergillus flavus*	*Mucor* spp
1	3.78	3.48	3.68
2	2.76	2.64	3.28
3	2.24	2.16	2.46
0	4.91	4.78	5.35
LSD (0.0001)	0.22	0.23	0.25

Table 9: Effect of Ash on the Fungal Pathogens (*In-vitro*)

Conc. (ml)	*Aspergillus niger*	*Aspergillus flavus*	*Mucor* spp
1	2.76	2.64	2.30
2	1.55	1.40	1.67
3	1.25	1.18	1.27
0	3.38	3.38	3.33
LSD (0.0001)	0.21	0.21	0.18

Table 10: Effect of Ash on the Fungal Pathogens (*In-vivo*)

Conc. (ml)	*Aspergillus niger*	*Aspergillus flavus*	*Mucor* spp
1	3.63	3.39	2.30
2	2.87	2.78	1.67
3	2.06	2.27	1.27
0	4.91	4.78	3.33

LSD (0.0001)	0.22	0.23	0.25

REFERENCES

Adebayo, A.A (1999). Climate: sunshine, temperature and relative humidity. In Adebayo A.A and Tukur A.L (eds) Adamawa state maps. Paraclete publisher's yola - Nigeria. Pp.4-6.

Adejumo T.O,.Ikotun. T and FIorinD.A (2000). In-vitro anti-fungal properties of some botanicals on Protomycopsis phaseoli p. 13 in: *Book of Abstract, 28 Annual Conference of Nigerian Society for Plant Protection at Cocoa Research Institute of Nigeria (CRIN), Idi-Ayunre Ibadan*, 24th-28th Sept, 2000, 33pp.

Akpa, A.D., Musa, B. And Paswall, A. T (1991). Effect of neem extracts and mycelial growth of the sorghum anthracnose Pathogen. *Collectotrichum graminicola*. *Proceedings of 21st Annual Conference of Nigeria Society of Plant Protection*. **10**-13 March 1991 p.47.

Alexopoulos, C. O., Mims, C.W. and Blackwell, M. (1966). Introductory Mycology3rd Edition, John Wiley and Sans New York Pp. 204-340.

Alvarez, A.M., Nishijima, W.T. (1987) Postharvest diseases of papaya. *Plant Dis*.**71**:681-686.

Amadioha, AC. (1998). Evaluation of some plant leaves extract against *collectotrichum lindemuthiamumin* cowpea. *Journal of phytopathology and entomology*. **38**: 259 – 265.

Amuchi, R. T. (1999). Fungitoxic effects of extracts from African plants. *Annual applied Biotechnology*. 115, 451-452.

Ankri S, Mirelman D. Antimicrobial properties of allicin from garlic. Microbes Infect 1999; (2):125-9.

Channya, F.K. (1991). Pathogens of post harvest fruits rot of plantain (*Musa paradisiaca* L.) in south western Nigeria. *Journal of tropical Bioscience* **2**: 12 – 15.

Choiseul, J., Allen, L., and Carnegie, F. (2007). Fungi causing dry tuber rot of seeds potatoes in storage in Scotland. *Journal of Potatoes Resources*. 6(1):241-253.

Danielone, H. Echeverri, F. Torres, F. Duinones, W. cardona, G. Archbold, R. Roldon, J. Brito I., Lius, J.G. and L aulou, E.H (1997). Phytoaloxin from papaya fruits. *Phytochemistry*. **44** (2): 255 – 258.

Egunjobi, O.A. and SO. Onoyerni (1981). The efficacy of water extract of neem (Azadirachta indica L.) leaves as a systemic *nematicide. Nigerian Journal Plant Protection* 5: 70-74.

Francisco, D, H (2010). *Lippia graveolens and carya illionoensis* organic extracts and their *in vitro* effects against *Rhizoctonia solani Kuhn*. *American Journal of agricultural and biological sciences* 5 (3): 380-384.

Gomez, K. A. and Gomez A. A. (1984). Statistical procedures for Agricultural Research (2nd ed). John Wiley and sons, p. 680.

Kazmi, S.A., Shahzad,S.and I. Niaz. 1995. Effect of neem oil on *in vitro* growth of root infecting fungi. *Pakistan Journal Botany*., 27(1): 217-220.

Liu, L.H and Kushalappa, A.C (2002). Response Surface Models to predict Potato Tubers Infection by F. *sumbucinum* from duration of wetness and Temperature and, Dry Rot lesion Expansion from Storage Time and Temperature. *International Journal of food Microbial*. 76:19-25.

Locke, J.E. (1995). Fungi. In: *The Neem Tree, source of Unique National Products for Integrated pest Management, Medicine, Industry and Other proposes.* (Ed.): H. Schmutterer. V.C.H,Weinheim, Germany. p 118-125.

Mainici , L.M and Cerato, C. (1994). Pathogenicity of *fusarium oxysporium f. sp tubererosi* isolates from tubers and potato plant. *Journal of phytopathology* 37:129-134.

Milner J.A (2001). A historical perspective on garlic and cancer. *Journal of nutrition* 131:1027s-1031s.

Masood, A., Saeed, S., Iqbal, N., Malik, M. T. and Kazmi, M. R. (2010). Methodology for the evaluation of symptoms severity of mango sudden death syndrome in Pakistan. *Pakistan Journal of Botany,* 42(2): 1289-1291.

Morton, J.F. (1987). Papaya in: fruits in worm climate. pp.336-346.

Niaz, I. and S.A.R. Kazmi. (2005). Neem seed coal oil fractions on stored grain fungi. *INT. Journal of Biological Biotechnology*., 2(3): 705-706.

Nneka, V.C. (2006). Potential of leaf extracts of *Azadirachta indica* and *ocimum gratissium* for the control of some potato *(solanum tuberosum*) fungal disease. *Nigerian journal of Botany*. 19: 68 – 73.

Oduro K. A, Damtey H. B and Adimora L. O, (1997). Evaluation of traditional control measure for disease of stored white yam*, Dioscorea rotundata* poirvar Gboko*. Ghana journal* of *science* 36:3-10.

Olowo, T., Dina, S.O., Oladiran, A.O. and Olunuga B.A.(2003). The control of weeds, pest and disease complex in cowpea (*Vigna unguiculata* L.) Walp. By the application of pesticide single and in combination. *Crop protection* **6**: 222-225).

Oluma, H.O.A. (1992). Fungi associated with root rot of pawpaw in southern and central Nigeria. *A Ph.D. thesis in the Department of Botany and Microbiology, University of Ibadan, Ibadan, Nigeria.* P. 34-36.

Rosfarizan, M., Ariff, A. B., Hassan, M. A and Karim, M. I. (2000) on kojic acid fermentation by *Aspergillus flavus*. *Pakistan Journal of Biological Science* **3**:977-982.

Scheffer H. (1953).A method of judging all contrast in the analysis of variance. Biometrica vol, .40:87-104.

Sibounnavoung, P Kalaw, S.P Divina, C. C and Soytong, K. (2009) Mycelia growth and sporulation of Emericella nidulans, A new. Fungal antagonist on two culture media. *Journal of Agricultural Technology*. **5**(2):317-324

Sinnah, D., G. Varghese and G. Baskaran. (1983). *Fungal flora of neem oil toxicity*. Malaysian Applied Biology, 12: 1-4.

Singh, P., Mishara, A.K. and Tripathi, N.N. 2012. Assessment of *mycoflora* associated with post-harvest losses of papaya fruits. *Journal of Agricultural Technology*. 8(3) 961-968.

Singh, V.P., H.B. Singh and R.B. Singh 1980. The fungicidal effect of neem extracts on some soil borne pathogens of gram (*Cicer arietinum*). *Mycologia.*, 72: 1077-1093.

Yoshida S, Kasuga S, Hayashi N, Ushiroguchi T, Matsuura H, Nakagawa S.(1987). Antifungal activity of ajoene derived from garlic. *Applied Environmental Microbiology*. 1987;53(3):615–617.

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