# FIELD SURVEY OF MAJOR FUNGAL DISEASES AND SCREENING FUNGICIDES FOR THE CONTROL OF FUSARIUM WILT OF SWEET POTATO (*Fusarium oxysporum* f. sp. *batatas*) IN KANO STATE

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DECLARATION

I hereby declare that this work is the product of my research efforts undertaken under the supervision of Mallam S.G. Haruna and has not been presented anywhere for the awards of degree or certificate, all sources have been duly acknowledged.

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CERTIFICATION

"This is to certify that the research work for this dissertation and the subsequent write-up" by Ali Ismaila Muhammad (SPS/15/MCP/00005) were carried out under my supervision.

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APPROVAL

This dissertation title "Field Survey of Major Fungal Diseases and Screening Fungicides for the Control of Fusarium wilt of sweet potato (*Fusarium oxysporum* f. sp. *batatas*) in Kano state" has been examined and approved for the award of Masters of Science in Crop protection (Phytomycology)

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# DEDICATION

This research dissertation is dedicated to the entire Wangara Family of Alh.Ismaila Muhammad Wangara, and my Late Mother Hajiya Binta.

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# ABSTRACT

Survey was conducted in 2016 rainy season at five local government areas (LGAs) of Kano state to assess incidence and severity of major fungal diseases in the areas The LGAs surveyed included Rimingado, Madobi, Kibiya, Bagwai and Garko which were all located Sin the Sudan savannah ecological zone of Nigeria. Screen house experiment was conducted to test the efficacy of plant extracts against Fusarium wilt (FW) of sweet potato, the most prevalent fungal disease in the surveyed areas. A2x5 factorial experiment consisting of 10 treatments was laid in a randomized complete block design. The treatments were replicated three times. Result of the survey showed prevalence of four major fungal diseases;- Fusarium wilt, Chlorotic leaf distortion, Minor leaf spot and Alternaria leaf spot on 10 breeding lines and 1 local variety of sweet potato across the locations. Higher incidences and severities of the fungal diseases were recorded in Garko (61%; 63%), followed by Bagwai (47%; 46%), Rimingado (45%;47%),Madobi (20%; 18%) and Kibiya (16%; 15%) respectively. *Danchana* had the least incidence and severity across the locations, followed by KingJ,T121, Mother delight, Ao305, Delvia, SumaiA, Melinda, Gloria, Lourdes and Centennial, respectively. Among the fungal diseases (FW) had the highest incidence and severity than the other fungal diseases. Result of the screen house experiments showed significantly ($P \leq 0.05$) lower incidence (15.7%) and severity (12.9%) on local variety *Danchana* grown on soil amended with 10% aqueous Neem leaf extracts (ANLE) compared to the other treatments. *Danchana* variety and 10% ANLE could therefore be use as an integrated disease management strategy against FW of sweet potato

# CHAPTER ONE

# INTRODUCTION

## 1.1 BACKGROUND OF THE STUDY

Sweet Potato [*Ipomoea batatas* (L.) Lam] is a dicotyledonous plant that belongs to the family *Convolvulaceae* with 45 genera and 1000 species. Sweet potato is a creeping plant and the only known economic specie of the *Convolvulaceae* family (Cobley and Steel, 1976). It is the third most important consumable root crop after potato and cassava and ranked fourth in production after rice, wheat, and corn in the developing countries (Kays, 2005). The top five producers are: China 70,526,000 m/t (68.6%), Tanzania 3,470,304m/t (3.3%), Nigeria 3,450,000m/t (3.3%), Indonesia 2,386,729m/t (2.3%), Uganda 1,810,000m/t (1.1%) (FAOSTAT, 2016). It is one of the most widely grown root crops in Sub-Saharan Africa, it is particularly in countries surrounding the Great Lakes in Eastern and Central Africa, Angola, Madagascar, Malawi and Mozambique in Southern Africa, and Nigeria in West Africa. In Africa, the crop is grown predominantly in small plots by peasant farmers (FAOSTA, 2012). Sweet potato is one of the major tuber crops in Nigeria which has been part of regular food habit of many Nigerians and major contributor to cross-substitution when other food stuffs are in short supply (FAO, 2008). The production of sweet potato has shifted since 1971 from the southern Guinea zones of Kwara, Plateau, Niger and Benue states to northern agro-ecological zones, where Kano, Kaduna and Bauchi states are leading production states (CIP, 2009).

High incidence and severity of diseases, especially those caused by fungi has been one of the major limiting factor in the production of sweet potato in Nigeria apart from soil and climatic conditions (Ilondu, 2013). Sweet potato is attacked by many fungal diseases which affect both the roots and leaves in the field (Ilondu, 2013). High yield reduction (up to 98%) caused by bacterial, fungal and viral diseases was reported (Kapinga *et al*., 2007). Fungal pathogen *Alternaria bataticola* is one of the major causes of leaf petiole and stem blight of sweet potato. Although a number of *Alternaria* species such as *A. alternata, A. brassicae* and *A. solani* have also been found causing spots on sweet potato leaves. Other fungal pathogens such as *Monilochaetes infuscans, Fusarium oxysporum, Ceratocysts fimbriata, Rhizopus stolonifer*, *Macrophomina phaseolina*, *Fusarium solani* and *Botryodiplodia theobromae* are reported to attack the crop (Clark and Hoy, 1994). Ansari (1995) reported that chemical fungicides could be effective in the management of fungal diseases; but the attendant problem of indiscriminate use of fungicides is not only hazardous to human but disrupts the natural ecological balance by killing the beneficial soil microbes. There are number of useful agrochemicals that are derived from plants and these plants compounds may provide useful templates to produce more active agrochemicals with less environmental risk. The presence of antifungal compounds in higher plants has long been recognized as an important factor to reduce disease resistance due to over dependence on synthetic fungicides (Kurucheve *et al*., 1997). Such compounds being biodegradable and selective in their toxicity are considered valuable for controlling different plant diseases (Singh and Dwivedi, 1999). Shahi *et al*. (2003) also reported antifungal activity of essential oils of some angiospermic plants, oil of *Cymbopogon flexuosus* against dominant post harvest fungal pathogens

namely; *Alternaria alternata*, *Aspergillus flavus, A. fumigatus, A. niger, A. parasiticus, Cladosporium cladosporioides, Colletotrichum capsici, C. falcatum, Curvularia lunata, Fusarium cerealis, F. culmorum, F. oxysporum, F. udum, Gloeosporium fructigenum, Penicilliumexpansum, P. italicum, P. implicatum, P. digitatum, P. minio-luteum, P. variable*, *Botrytis cinerea, Helminthosporium oryzae, H. maydis, Phoma violacea,* and *Rhizopus nigricans* causing rotting in *Malus pumilo* fruits. Therefore, use of botanicals in the control of plant pathogens is a potential means for plant disease control and can serve as an alternative to costly synthetic fungicides (Omorusi *et al*., 2007). Yield of sweet potato cultivar have gradually been on the decline as a result of fungal, bacterial, viral and genetic mutations (Clark et al 2003). An integral approach involving cultural practices is essential for effective management of fungal diseases of sweet potato like collar rot and Fusarium wilt, drenching of soil around plant with Vitavex (50 ppm), also application of biocontrol like *Trichoderma viride* to soil control Fusarium wilt and Cercospora leaf spot of sweet potato (Clark *et al.,* 2003). Mishra (2009) reported that use of resistance varieties is the best option in controlling the fungus *Fusarium oxysporum f.sp. batatas* and 16 lines of sweet potato have been developed. Dipping of seed tubers and vines in fungicide like Carbendazim, benomyl and Thiabendazole prevent the occurrence of some fungal diseases of sweet potato has also been reported (Nielson, 2000)

## 1.2 PROBLEM STATEMENTS

Fungi are responsible for more than 70% of all major crop diseases (Agrios, 2005). Many fungal diseases have been reported to attack sweet potato both in field and storage in different growing areas of the world ThanKappan (1994). Surkova (1978)

reported *Fusarium oxysporium, Fusarium trichothecoides* and *Fusarium radicicolato* as incitant of sweet potato tuber rot under different condition of temperature and humidity. Wheeler (1979) and Ameinyo and Ataga (2006) reported Rhizopus soft rot (*Rhizopus stolonifer*), Fusarium surface rot (*Fusarium oxysporum*) and black rot (*Ceratocystis fimbriata)* as some of the postharvest diseases of sweet potato. Despite the extensive work done on fungal disease of many food crops in Nigeria not much work has been done to identify the common fungal diseases of sweet potato in Kano state and it is one of the major states where the crop is produced.

## 1.3 JUSTIFICATION

The importance of sweet potato is becoming increasingly important in Nigeria's farming systems because it is easy to grow, and has enormous industrial and economic potentials (Chukwu, 1999). It has high nutritional energy qualities and the leaves are consumed as vegetables (Chukwu, 1999). Sweet potato is grown in most parts of Nigeria as a secondary crop but it reaches intense levels in some areas (Akoroda and Nwokocha, 1996). Sweet potatoes offer a particularly significant potential for increasing food production and income among Nigerian farmers. It has a significant role to play in the developing economies. Its production provides job opportunities for farmers, thus raising their income. It is consumed without much processing in most part of the tropics. It also complements other food crops and serves to bridge periods of food shortage before the next harvest of other staple crops (Jeremiah *et al*.,1999). Traditionally it is a food security item and dependable source of income for resource-poor families in Africa (Kaitisha and Gibson,1999). Studies on fungal diseases affecting the sweet potato roots is important because they affect the yield, aesthetic quality, storage life and

nutritional value of the storage roots. These diseases create local discoloration and disruption of surrounding tissues of infected tubers (Snowdon, 2010). Fungal plant diseases are generally managed with the applications of chemical fungicides which have been found to be very effective for some fungal diseases. Screening of fungicides and bio-products will provide a baseline for the control of fungal diseases of sweet potato in the study area and reduce dependence on chemical fungicides alone. There is also a dearth of information on the diseases of sweet potato in Kano state particularly those diseases caused by fungi. Survey of the diseases from different areas of production will provide baseline information for disease management strategy in the study area.

## 1.4 OBJECTIVES OF THE STUDY

i) To determine the incidence and severity of major fungal diseases of sweet potato in five selected local government areas of Kano State.

ii) To screen the efficacy of some fungicides against *Fusarium oxysporium* f. sp. *batatas* of sweet potato.

iii) To determine *in vitro* and *in vivo* efficacy of some indigenous plant extracts on *Fusarium oxysporium* f. sp. *Batatas.*

# CHAPTER TWO

# 2.0 LITERATURE REVIEW

## 2.1 INTRODUCTION

Sweet potato [*Ipomoea batatas* L. (Lam).] is a dicotyledonous plant belonging to the family *Convolvulaceae*. It is an important tuberous root crop cultivated throughout the tropical and warm temperate regions wherever there is adequate soil moisture to support growth (Demissew, 2006). Sweet potato [*Ipomea batatas* (L) Lam.] is a crop that is tolerant to a wide range of edaphic and climatic conditions. It is more tolerant to cold than other tropical root and tuber crops and can be grown at altitudes of up to 2500 mm (Luisa and Robert, 2000). There are approximately 50 genera and more than 1000 species in the family. Davison (1978) identified two species in the section *batatas*, which includes the sweet potatoes. The closely related species of the sweet potato appears to be *Ipomoea trifida* and *Ipomoea tabascan*. The starchy tuberous roots are the major source of food for millions of people; the leaves are also important sources of vegetable in some countries (Croft, 2007).

It is cultivated in many developed and developing countries and ranks among the five most important food crops in over 50 of those countries (FAOSTAT, 2012). It is one of the most widely grown root crops in Sub-Saharan Africa (SSA), it is particularly important in countries surrounding the Great Lakes in Eastern and Central Africa, in Angola, Madagascar, Malawi and Mozambique in Southern Africa, and in Nigeria in West Africa. (CSA, 2011)

## 2.2 BOTANY OF SWEET POTATO

Sweet potato is herbaceous and perennial plant although, it is grown as an annual plant by vegetative propagation using either storage roots or stems cuttings. Its growth habit prostrate with a vine system that extend rapidly horizontally on the ground. The Types of growth habits are erect, semi-erect, spreading and very spreading (Huaman 1992). The stem is cylindrical and its length depends on the growth habit of the cultivar and the availability of soil moisture. *Ipomoea batatas* is native to the Americas. The leaves are simple and arranged spirally and alternatively on the stem. The sweet potato cultivar colour can be green, yellowish-green, or purple pigmentation in part or all of the leaf blades. The storage roots are the commercial part of the sweet potato plant. The colour of the smooth skin of the root tuber ranges between yellow, orange, red, brown, purple, and beige. Flesh ranges from beige to white, red, pink, violet, yellow, orange, and purple. Sweet potato varieties with white or pale yellow flesh are less sweet than those with red, pink, or orange flesh (Loebenstein and Thottappilly, 2009).

## 2.3 ORIGIN AND SPREAD OF SWEET POTATO

The centre of origin is believed to be in tropical America from where it spread to the part of the world (Yen, 1974). Sweet potato together with the wild *Ipomoea s*pecies originated from Central or South America in the region between Yucatan peninsula of Mexico and Orinoco river in Venezuela, about 8000-6000 BC, where *I. trifida* and *I. triloba* might have crossed to produce the wild ancestor of *I. batatas* (Austin, 1988). Sweet potato cultivars are grown in almost all tropical and sub-tropical parts of the world and in warmer temperate regions such as southern USA sub-Saharan Africa (Nigeria),

West Indies, China and Japan (Horton *et al*., 1984). The secondary centres of diversity are found in Guatemala, Colombia, Ecuador and Peru (Austin, 1983).

## 2.4 CLIMATIC CONDITION

Sweet potatoes are somewhat drought-tolerant as well as hardy during summer dry spells. However, low humidity impairs growth even if the plant receives water following stress or drought (Monteiro, 1992). Sweet potatoes are cultivated wherever there is enough water to support their growth; optimal annual rainfall for growth ranges between 750 and 2000 mm. Where the rainfall is below 850 mm irrigation may be necessary, but it should be stopped before harvest in order to prevent the tubers from rotting (Monteiro, 1992). It is a warm-season annual, requiring 20-25°C average temperatures and full sunlight for optimal development. It needs a frost-free period of 110-170 days and growth may be hampered with an average day temperature below 20°C (Roullier, 2013). The crop thrives in well-drained loamy soils with a high humus content that provides a warm and moist environment for the roots. Optimal soil pH is between 5 and 7 (Roullier, 2013)

## 2.5 IMPORTANCE AND PRODUCTION OF SWEET POTATO IN NIGERIA

It is estimated that sweet potato cultivation in Nigeria has extensively increased with annual production of 3,450, 000 metric tons per year, making it the third largest producer in the world after China and Tanzania (FAO, 2015). The International Potato Centre (CIP) Lima, Peru ranked sweet potato as the seventh most important food crop in the world after wheat, rice, maize, potato, barley and cassava (Mwanga *et al*., 2001). Sweet potato is one of the major root crops in Nigeria which has been part of regular food

habit of many Nigerians and major contributor to cross-substitution when other food stuffs are in short supply (FAO, 2008). It is a food security item and dependable source of income for resource-poor families in Africa (Kaitisha and Gibson, 1999). The leaves are popularly used for livestock feeds (Madibela *et al*., 1999); agro-industrial and pharmaceutical raw materials (Chukwu, 1999).

## 2.6 FUNGAL DISEASES OF SWEET POTATO

Attack by fungi is one of the major constraints to sweet potato cultivation resulting in both pre- and post harvest losses, sweet potato is affected by many fungal diseases and more than 40 pathogens have been reported to attack sweet potato both in field and storage in different sweet potato growing areas all over the world (ThanKappan, 1994). Surkova (1978) reported *Fusarium oxysporium F. trichothecoides* and *F. radicicolato* as incitant of sweet potato tuber rot under different condition of temperature and humidity. Ameinyo and Ataga (2006) reported that *Rhizopus oryzae, R.stolonifer, Aspergillus niger, Fusarium oxysporum* and *Ceratocystis fimbriata)* as pathogen causing sweet potato rot.

### .2.6.1 Leaf Spot of Sweet Potato

The leaf Spot of sweet potato caused by Alternaria spp. the fungus survives in the soil and in plant debris. The airborne spores are spread through infected planting material, wind, rain splash and water. Infection by the disease on sweet potato is aggravated by high relative humidity. The disease incidence and severity increase with increase relative humidity and altitude. (Stathers *et,al* 2005). The disease first reported from subsistence food garden in the Nebilyer valley of the Western Highlands province

in Papua New Guinea in early 1987 (Lenne, 1991). *Alternaria* leaf and stem blight of sweet potato was reported in sweet potato producing countries including Nigeria (Arene and Nwankiti, 1978).

The first symptoms of the disease appears as small, brown/grey/black oval lesions with a typical bulls' eye appearance of concentric rings, on leaves, stems and petioles. On the lower side of the leaf, blackened veins are observed. As the disease progresses, the lesions become necrotic usually surrounded by a wide yellow halo; soon after the whole blade turns chlorotic and drops. Death of vines can occur under severe infection. (Stathers, *et,al* 2005). *Alternaria* species (e.g. *A. alternata, A. brassicae* and *A. solani*) have been found causing spots on sweet potato leaves, and *A. bataticola* attacks the whole vine (leaves, petioles and stems) throughout the crop cycle. Surveys in Africa, for instance, showed the predominant species were *A. bataticola* (55% of isolates) and *A. alternata*(40%). Increased infection of sweet potato by *Alternaria* spp was reported to be due to stress factors such as drought, inadequate nutrients, infestations by insect pests or other pathogens, and wounds inflicted by nonspecific agents (Lenne, 1991).

### 2.6.2 Black Rot of Sweet Potato: *Ceratocystis fimbriata*

*Ceratocystis fimbriata* causes black rot diseases of sweet potato the fungus survives in soil, water, and on decaying organic matter such as sweet potato debris. It can survive for several years in the soil (Lenne, 1991). The fungal spores are spread by wind, water, soil, farm machinery, insects, humans (clothing), contaminated tools. Wounds on the sweet potato skin are important entry points for infection by the fungus. Sweet potato roots and stems are equally susceptible to infection.

Early symptoms begin with small, circular, slightly sunken, dark brown or grey spots on the sweet potato surface. Advanced symptoms results in to a large, circular, sunken, dark brown to black spots on the sweet potato root surface Anon (1988). The brownish colored rot usually remains shallow, but can extend into the inner part of the root, leading to secondary infection by other organisms which can destroy the entire roots. (Curir *et al.* (2005).

2.6.3 Scurf (Monilochaetes infuscans)

It is a very slow growing fungus, and has been reported in Brazil, USA, Japan, and Australia (Onwueme, 1978). It causes brownish blotches on the tubers and other subterranean parts of the plant. It does not directly affect the cortex or under lying tissues of the storage root.

2.6.4 Soft rot (Rhizopus stolonifer)

The disease is commonly called *Rhizopus* soft rot caused by *Rhizopus stolonifer*. It is a serious post-harvest disease of sweet potato. It is the most prevalent in temperate and sub-tropical growing regions, but also common in tropical growing regions. The fungus could not penetrate through the intact periderm of the tuber; but normally gains access through wounds. The disease causes a general rotting of the tuber during storage (Moyer, 1981). The disease is widely found in all sweet potato growing countries (Ray *et al.,* 2010)

### 2.6.5 Fusarium Wilt of Sweet Potato *Fusarium oxysporum* f.s.p *batatas*

Fusarium wilt or stem rot caused by *Fusarium oxysporum* f. s.p *batatas* has been reported in Africa, USA, and Japan, and many other countries including Nigeria, although primarily caused by *F. oxysporum* f.sp. *batatas*, sometimes strains of the tobacco pathogen (*F. oxysporum* f.sp *nicotianae*) can cause wilt in susceptible potato cultivar (Loebenstein and Thottappilly, 2009). Yellowing and wilting of the lower, older leaves and the vines turning tan to light-brown are the typical symptoms. The vascular tissues of affected plants turn dark brown or black, especially close to the soil level. The dying vines shows pinkish fruiting bodies of the fungus. The tubers produced by an infected plant have discolored vascular tissues which may rot upon storage. Fields are commonly infected through contaminated cuttings. Once in the field, the fungus penetrates healthy plants through open wounds. Yield losses may be up to 50 percent, and are more likely under warm weather and in dry soils. Plants normally die within a few days after visible symptoms appear in the plant (Gunua, 2010).

Control method include disease resistance plants crop rotations, planting diseases free plants and fungicides applications (Clark and Moyer, 1988). It enters the plant mostly through wounds, and attacks the vascular tissue, especially the xylem. Growth of the plant is stunted, and leaves are wrinkled and turns yellow (Onwueme, 1978). *Fusarium oxysporum* Schlecht causes vascular wilt diseases in a wide variety of economically important crops (Beckman 1987).

The general symptoms of wilts caused by *Fusarium* are stunted growth, yellowing and wilting of the leaves, reddish discoloration of the xylem vessels, visible inside the

stem as lines (if the stem is cut open lengthways) or dots (if it is cut across). Others are white, pink or orange fungal growth on the outside of affected stems, particularly in wet conditions, root or stem decay (Miller *et al*., 2011). Severity of leaf damage in the field depend on a number of factors, including the distribution of inoculums of the fungal organism in the field, the degree to which the vascular tissue in stems has been damaged by the fungus as it grows within plant tissue, the duration of the fungal "attack" and ability of the plant to re-grow or retain leaves. At the seedling stage or in young plants, cotyledons and leaves wilt and drop, leading to bare stems. For example, symptoms are easily confused with those of crown or root rot, stem cankers, pest injury, drought, nutrient deficiency, bacterial and *Verticillium* wilts (Hutmacher *et al*., 2003) There are many important pathogenic species of the fungus which are able to infect different plants causing wilts in crops of economic importance. Some of the formae speciales are identified on the basis of virulence to a set of differential cultivars within the same plant species (Armstrong and Armstrong, 1981).

Currently, there are over 100 different formae speciales described, causing disease in a wide range of dicot and monocot plant species. For example, Banana Fusarium wilt or Panama wilt (*F. oxysporum* f.sp. *cubense*), *Fusarium* wilt of cotton (*F. oxysporum* f.sp. *vasinfectum)*, *Fusarium* wilt of sweet potato (*F. oxysporum* f.sp. *batatas), Fusarium* wilt of *Callistephus* (*F. oxysporum* f.sp. *allistephi); Fusarium* wilt of tomato (*F. oxysporum* f.sp. *lycopersici*), *Fusarium* wilt of date palm (*F. oxysporum* f.sp. *albedinis*) and *Fusarium* yellows of common beans (*F. oxysporum* f.sp. *phaseoli*). Sweet potato and other solanaceous crops, like tomato, legumes, cucurbits and banana are the most

susceptible plants (Miller *et al*., 2011), though it will also infect other herbaceous plants as well as cotton, ornamentals and palms. Other hosts are

C*allistephus*(Chinaaster), *Dianthus* (carnations, pinks), French/runner beans, hebe, peas. However, individual pathogenic strains within the species have a limited host range, and strains with similar or identical host ranges are assigned to the same *formae speciales* (Armstrong and Armstrong, 1981). Approximately 1000 Fusarium species had been described by 1900, based largely on examination of fruiting structures (sporodochia) on plant material. This large number of species was reduced by Wollenweber and Reinking, (1935) to 65 species, 55 varieties and 22 forms, in 16 sections, and all taxonomic systems proposed since then have been based on this system (Burgess *et al*., 1994). Gerlach *et al* (2000) recognized over 90 species in Fusarium. They proposed delimitation and further emphasized the importance of analyzing variation in a large number of cultures from a wide range of substrates and geographic sources (Burgess *et al*., 1994).

## 2.7 BIOLOGY OF *FUSARIUM OXYSPORUM*

### 2.7.1 Taxanomy

Based on the structure in or on which conidiogenous hyphae are borne, *Fusarium spp.* are classified under the Hyphomycetidae subclass of the Deuteromycetes. *Fusarium oxysporum*, as emended by Snyder & Hansen (1940), comprises all the species, varieties and forms recognised by Wollenweber & Reinking (1935) within an intragenic grouping called section Elegans. Booth (1971) described *F. oxysporum a*s a cosmopolitan soil-borne filamentous fungus. It is an anamorphic species that includes numerous plant

pathogenic strains causing wilt diseases of a broad range of agricultural and ornamental host plant species (Appel and Gordon1996)

2.7.2 Lifecycle

The life cycle of *F. oxysporum* starts with a saprophytic phase when the fungus survives in soil as chlamydospores (Beckman and Roberts 1995). Chlamydospores remain dormant and immobile in the remains of decayed plant tissue until stimulated to germinate by utilizing nutrients that are released from extending roots of a variety of plants (Beckman and Roberts 1995). Following germination, a thallus is produced from which conidia form in 6-8 hours and chlamydospores in 2-3 days if conditions are unfavorable. Invasion of the roots is followed by the penetration of the epidermal cells of a host (Beckman and Roberts 1995) and the development of a systemic vascular disease in host plants (Stover 1970). The fungus grows out of the vascular system into adjacent parenchyma cells, producing vast quantities of conidia and chlamydospores. The pathogen survives in infected plant debris in the soil as mycelium and in spore forms, but most commonly as chlamydospores in the cooler temperate regions (Agrios,1997).

*Fusarium* species are soil borne pathogens, which attack the water conducting vessels of host plants. In soil, fungal colonization of plant roots has been traditionally studied by indirect methods such as microbial isolation that do not enable direct observation of infection sites or of interactions between fungal pathogens and their antagonists (Olivain *et al*., 2006). The disease cycles of most *Fusarium* wilts are similar and resemble that of the *Fusarium* wilt of sweet potato (Agrios, 2005). Like other vascular pathogens, such as *Verticillium*, the life cycle of *Fusarium* species can be

divided into dormant, parasitic and saprophytic stages (Beckman, 1987). The dormant stage comprises inhibition and germination of resting structures in soil. The parasitic stage comprises penetration of roots, colonization of the root cortex and endodermis, movement to the xylem; colonization of the xylem of stems and leaves, symptom expression and, finally death of the host.

The saprophytic stage of the pathogen started with the formation of resting structures in the dead host (Schnathorst, 1981). In the dormant phase, mycelia, chlamydospores, macroconidia and microconidia (propagules) present in infested soil are inhibited from germinating because of mycostasis or microbiostasis (Huisman, 1982). Reversal of inhibition of resting structures from germination in non-host specific could be reversed when propagules come in contact with *Fusarium* wilts Root exudates released in the rhizosphere of a host plant (e.g. sweet potato), a non-host plant (e.g grass) or contact with pieces of fresh non-colonised plant remains (Steinkellner *et al*.,2008).The exudates serve as rich source of carbon (sugar), nitrogen (amino acids) and organic acids, which are generally known to stimulate spore germination(Nelson,1991). *Fusarium* species enter the parasitic phase when any of the propagules or germ tube of spore, penetrates the host through cracks formed by emerging lateral roots, wounds or at the root cap, root hairs or branch roots (Mandeel, 2007).

The penetration process is likely enhanced by certain hydrolyzing enzymes secreted by *Fusarium* (Walter *et al*., 2009). There has been report of an association with *Fusarium* wilt and nematode colonization, where the nematodes provide a potential entry point (wound) for the fungus (Morrell and Bloom, 1981). Penetration is usually intercellular. Inside the root, the cortex is colonized by emerging mycelia (Bishop and

Cooper, 1983). From the cortex, the hyphae penetrate the endodermis and invade the xylem vessels through the pits. The mycelia remain in the vessels where they invade the plant in an upward direction (colonization), through the stem and crown of the plant (Rodríguez and Gálvez, 1995). Effective colonization is genetically controlled (Inoue *et al*., 2001). Inside the xylem vessels, the mycelia produce microconidia, which are released to travel upward in the transpiration stream, until trapped in pit cavities or at vessel end walls. They germinate into new hyphae and penetrate adjacent vessel elements to continue colonization and increase infection (Schnathorst, 1981). At this stage, *Fusarium* hyphae spread within the cell apoplast, which leads to significant cytological alterations resulting in symptom expression (Walter *et al*., 2009). A combination of vessel clogging by mycelia, spores (from the fungus) and gels, gums, tyloses and crushing of the vessels by proliferating adjacent parenchyma cells from the host plant in an attempt to defense itself plug vessels and is responsible for the breakdown of the water conducting system of infected plant(Agrios, 2005).

This gives rise to wilting of lower branches, followed by the entire plant, which eventually leads to death. The new spores can either be returned to the soil when the plant decomposes or disseminated to new plants or areas by wind or water. In the process, conidia are also formed in sporodochia on dead leaves, and hyphae and chlamydospores are also produced extensively. The chlamydospores are returned to the soil, when the diseased plant residues decay. They can remain viable in the soil in their dormant state for several years and grow upon germination by parasitic or saprophytic colonisation of a new host. Certain weeds are symptom less carriers of *Fusarium* (Fassihiani, 2000).

### 2.7.3 Disease Development

Fusarium wilt is most likely caused by a combination of pathogen activities. These include accumulation of fungal mycelium in the xylem and/or toxin production, host defense responses, including production of gels, gums and tyloses, and vessel crushing by proliferation of adjacent parenchyma cells (Beckman 1987).

### 2.7.4 Macroscopic and Microscopic Features

The genetic structure of Fusarium species is variable and the morphology of the species is influenced by environmental factors. For many of the species, specific conditions are required for optimal morphology manifestations and the tendency to mutate causes difficulties in identification. Most Fusarium species grow rapidly on Sabouraud dextrose agar at 25°C and produce woolly to cottony, flat, spreading colonies. The only slow-growing species is *Fusarium dimerum.* The colour of the colony may be white, cream, tan, salmon, cinnamon, yellow, red, violet, pink or purple and on the reverse, it may be colorless, tan, red, dark purple, or brown (Kontoyiannis *et al*., 2000).

### 2.7.5 Isolation and Ecology

*Fusarium* species can be found in soil, water and on seeds, roots and leaves of most plants. Several selective media have been developed for the isolation, growth and sporulation of Fusarium species, including selective *Fusarium* Agar (SFA), Dichloran Chloramphenicol Peptone Agar (DCPA), Spezieller Nahrst of farmer Agar (SNA) and Modified Potato Dextrose Agar (MPDA). The isolation of Fusarium species from plants is affected by the nature of the source material, method of surface sterilization, plating procedures, medium and incubation conditions (Burgess *et al*., 1994).The choice of

medium depends largely on the nature of the tissue involved in the isolation exercise. Selective media are normally used for the isolation of Fusarium species from diseased crown or root samples. There are several other techniques for recovering Fusarium species, directly or indirectly, from plant samples, which do not involve plating tissue segments on agar media. Some species produce sporodochia on the surface of the diseased tissue. Macroconidia can be taken from these sites and used to prepare a conidial suspension which is plated on Water Agar containing antibiotics. Germinated single conidia are later taken to initiate pure cultures for identification of Fusarium species (Burgess *et al.,* 1994). Several species produce airborne conidia and are common colonizers of stems, leaves and floral parts of plants (Burgess, 1994).

## 2.8 CONTROL OF THE FUNGUS

*Fusarium* wilts are generally presumed to be polycyclic that is, the disease does exhibit plant-to-plant spread during the season this is primarily because there are no propagules capable of dissemination to other plants to cause secondary infections that form above ground until very late in the season. (Egel and Martyn,2007). The control of *Fusarium* wilt of Sweet potato is important in maintaining plant vigour and root quality and quantity. However, attempts to control the disease have experienced limited success due mainly to emergence of new pathogenic races ,Documented methods that are used in the control of the disease include cultural, biological, use of resistance, and chemical (L'Haridon *et al*. (2011).

### 2.8.1 Chemical Control

Several chemical fungicides such as prochloraz and carbendazim (Song *et al.*, 2004), Bavistin (Alam *et al*., 2010) and salicylic acid (Amel *et al*., 2010), were used in managing diseases. Fungicides such as Dichloronitroanline was also reported to be use in protecting tubers against Rhizopus soft rot (Clark and Moyer, 1988). Rhoda *et al*., 2007 reported the use of Mancozeb (dithiocarbamate),Carbendazim (benzimidazole) on soil borne fungi. Nel *et al*. (2007) reported partial effectiveness of benomyl against *F. oxysporum* f.sp *batatas* using the root dip treatment method. The use of carbendazime on sweet potato vines infected with *Fusarium* wilt led to about 24 % increase in yield (Khan and Khan, 2002). Moreover, pesticides generally are more effective against aerial plant pathogens than their soil-borne counterparts (Recycled Organics Unit, 2006). Generally, the use of fungicide in conventional sweet potato production is common among farmers because they give immediate remedy to most diseases. However, environmental and health hazards coupled with increase in production cost delimit the use of fungicides on sweet potato (Lobato *et al*., 2010).

### 2.8.2 Resistance Variety

Cultivation of resistant varieties (the best option) is commonly practiced as well as maintaining good hygiene (such as avoidance of contamination, during planting and vines transplanting, irrigation, and clearing of debris from previous year's planting) (Jones and Woltz, 1981). The use of resistant varieties is the best strategy for the disease control (Sheu *et al*., 2006) and also one of the most effective alternative approaches to controlling wilt disease (Singh, 2005). But, due to breakdown of resistance in the face of

high pathogenic variability in the pathogen population, the usefulness of many resistant cultivars is restricted to only a few years (Kutama *et al*., 2013). Modification of the soil pH and fertilizer composition significantly reduced disease development (Borrero *et,al.,*2011).

2.8.3 Biological/Botanicals Control

Despite the present trend to discourage the use of chemical fungicides to control post harvest diseases of produce, they are still extensively used in many developing countries (Champ *et al.,* 1994).Recent studies on the use of plant extracts have opened a new avenue for the control of plant diseases. These plants extracts have been reported to be safe, non-toxic to man, but effective against plant pathogens (Shivpuri *et al*., 1997). The use of plant products for the control of *Fusarium* wilt in crops is limited (Agbenin and Marley, 2006). In Nigeria plant extracts have been used to control fungal diseases of plants such as cowpea (Amadioha and Obi, 1998), banana (Okigbo and Emoghene, 2004) and Yam (Okigbo and Nmeka, 2005).

The reports of Okungbowa (2011) have indicated the possible use of plant extracts for the control of *F. oxysporum*. Plant products have generally been used in the control of insect pests. However, extracts of ginger rhizomes, garlic bulb and aloevera were successfully used in the control of fungal pathogens (Ahmad and Beg, 2001). The limonoids in Neem products (*Azadirachtin*) though extensively used in insect pest management have been reported to have some fungicidal effect (Stoll, 1998) and bactericidal properties ( Bdliya and Dahiru, 2006) and have been used in plant disease

management. Limonoids in Mahogany are reported to possess some antiviral, antifungal and bactericidal properties ( Ademola *et al*., 2004).

Research conducted by Kimaru *et al*. (2004) revealed that neem cake powder contains ingredients that have fungistatic effects against *Fusarium* wilt of sweet potato. Hanaa *et al*. (2011) investigated the effect of *Azadirachta indica* (Neem) and *Salix babylonica* (Willow)10% aqueous extracts on *Fusarium* wilt disease in sweet potato and revealed that the percentage of disease incidence was reduced to the level of 25.5 % and 27.8 % after 6 weeks of infection respectively. Also, Agbenin and Marley (2006) reported that crude extracts of neem (*Azadirachta indica*) and garlic (*Allium sativum*) at concentrations ranging from 5 % to 30 % of the material in 100 ml of potato Dextrose Agar inhibited mycelial growth of *Fusarium oxysporum* f. sp. *lycopersici* at various levels. Dry neem seed extract gave 100 % inhibition of *Geotrichum spp in vitro*. Fresh neem leaf extract reduced mycelial growth with increasing concentration while in garlic, there is no difference in growth inhibition among the various concentrations used. (Umar *et al,* 2013)

Several researchers (Ejechi *et al*. 1999 ; Jasso de *Rodriguez et al*., 2005; Curir *et al*. 2005) have reported the fungitoxic effect of some plants extracts in the managements of crop diseases. Some extracts from neem plant have shown to be toxic to fungal pathogens, such as *Poria monticolad* infecting wood , *Aspergillus flavus* on soybean seeds (Krishnamurthy *et al*., 2008), *Pyricularia oryzae* infecting rice plant in field and the harvested rice (Amadioha, 2000).

Little is known about the antimicrobial activities of *Calotropis procera* except for their activities against a small range of microorganisms (Kareem *et al.,* 2001). Freitas *et, al,* (2011) recently reported an antifungal protein purification from the latex of *Calotropis procera.* Leaf extracts, chopped leaves and latex of *C. procera* have shown great promise as a nematicide *in vitro* and *in vivo* (Khirstova and Tissot, 1995).. Investigations have also shown inhibitory properties of garlic against *Penicillium digitatum*. (Kanan and Al-Najar 2008). Whilst many studies have highlighted the antimicrobial action of garlic on plant pathogens, little research has been done relating to postharvest plant pathogens, (Obagwu and Korsten, 2003).

### 2.8.4 Soil Solarization

Soil Solarization is a simple, safe, and effective alternative to the toxic, costly soil Fungicides to control many damaging soilborne fungi. ( Austin 2018) Radiant heat from the sun is the lethal agent involved in soil solarization. A clear polyethylene mulch or tarp is used to trap solar heat in the soil. Over a period of several weeks to a few months, soil temperatures become high enough to kill many of the soilborne pathogens depth of nearly 8 inches ( Austin 2018). This is done by spreading a clear plastic sheet over the soil for several weeks. This helps to trap solar energy which in turn inhibits soilborne diseases, nematodes, insects and many weed seeds. This is usually done during the summer when the air temperature is high and there is intense radiation, Combination of soil Solarization and two layers of mulching decrease the rate of fungal infection in plants (Pottorf, 2006). Thermotolerant and thermophyllic soil-borne microorganisms usually survive Solarization, but become weaker and more sensitive than other microorganisms-antagonists (Stapleton and DeVay, 2000). This method is effective for the control of soil-

borne pathogens (*Verticillium dahliae, Rhizoctonia solani, Fusarium* sp., *Pythium* sp. and others), nematodes and weeds. Transparent polyethylene is the ideal film for soil heating since it allows solar radiation to penetrate moist soil. Thicker polyethylene (50.100 and 150 microns) can also be used, but thinner polyethylene (25 and 40 microns) is more effective for soil heating and more cost-efficient per unit area (Stapleton and De Vay,2001). Soil moisture is essential to favour good soil solarization performance. Heat transferred by sun rays is well trapped by the soil, while soil-borne pests are well controlled. The higher the soil moisture the higher the temperature (Mahrer *et al*., 1999).

2.8.5 Soil Disinfection using Heating and Steaming

Steaming the soil (disinfecting the soil with steam) is the most effective soil pest control method. For over a hundred years, this has been the most efficient method for disinfecting soils. When applied, it must be considered that nematodes and Oomycetes die at temperatures above $5000^0$C bacteria and many fungi die at temperatures above 600–$7000^0$C; weeds, bacteria, viruses and insects, at above $8200^0$C it is applied where soil is strongly infected by *Verticillium, Fusarium*, or when there is debris of plant residues. It must be pointed out, however, that excessive and continuous high temperature has a negative effect, because it may kill the beneficial saprophyte microflora in the soil, creating an "ecological vacuum". Such a condition makes it easy for pathogens to re-colonize the soil, causing a "boomerang effect". (Mahrer *et al*.,1984). Hot water can also be used and can keep the soil sterilized for up to three years. Steam can also be used especially in greenhouse conditions ,Soil disinfection helps to keep the soil sterile and free from disease causing pathogens (Pottorf, 2006).

# CHAPTER THREE

## 3.0 MATERIALS AND METHODS

### 3.1 SURVEY OF THE MAJOR FUNGAL DISEASES OF SWEET POTATO IN THE STUDY AREAS.

The survey was conducted in five selected local government of Kano state namely: Rimin Gado [Gulu village, ($11^0$ 57'54"N $8^0$ 15'0"E)], Madobi [Gara village ($11^0$ 46'18"N $8^0$ 17'18"E)], kibiya [Fagam village ($11^0$ 32'N $8^0$ 40"E)], Bagwai ($12^0$ 09'28"N $8^0$ 08'09"E), and Garko [Sarina village($11^0$ 39'N $8^0$ 50'E)] located within Sudan savannah of Nigeria. Survey on fungal diseases of sweet potato was conducted on ten (10) Breeding lines (Mother Delight, Lourdes, Delvia, Centennial, Gloria, A0305, T121, King J, Melinda and SumaiA) and Local variety (*Danchana*) grown in five selected local governments of Kano state. The survey was carried out between the months of July to September, 2016 rainy season. Each production centre was visited every three weeks; incidence and severity were assessed on the major fungal diseases of sweet potato. Samples of infected plants were collected; photographs and pictures of diseased plants were taken from the fields for laboratory diagnosis and identification of pathogens. The samples were collected in a quadrant of $3m^2$ in an X pattern across each field as recommended by Agrios (1997) and the plant samples were pressed and kept fresh under refrigeration for laboratory investigation.

The disease incidence was calculated using the formula: Number of infected plants/ Total number of plant assessed x100. Disease severity (DS) was also calculated using the formula:

S = {(∑N) / (N X Highest Score of Severity)} * 100 (Singh, 2006). Where ∑N = Summation of Individual Ratings, N = Total Number of Plants Assessed.

Diseases severity rating scale: For Alternaria Leaf Spot, and Minor Leaf Spot are: 0= No Diseases, 1=Small Traces of Lesion, 2-3=Slight Infection (Lesion on Upper and Lower Leaves), 4 =Moderate infection (Advanced Lesion on Upper and lower leaves with or without new infection on Petiole, 5=Severe infection (Advanced Lesion on Upper and lower leaves backs and petiole dark to dark Brown. For Fusarium wilt and Chlorotic Diseases 1= No Symptoms on healthy heaves, 2 = Wilted leaves without Chlorosis, 3 =One or Several Slightly Chlorotic Diseases, 4= Chlorotic bands over The Entire Surface of the leaves or Chlorotic bands with necrotic centre, 5= Complete necrosis or death of the leaves.

## 3.2 ISOLATION OF FUNGAL PATHOGENS OF SWEET POTATO

Isolation of causal agents was carried out in the laboratory from sample collected in the field from crop roots, stems, leaves and the soil.

### 3.2.1 Isolation from Roots, Stems and Leaves

Infected plant roots, stems and leaves were washed under tap water and cut with sterilized laboratory knife to small pieces (2cm) and were surface sterilized for two minutes using 0.1% sodium hypochlorite. The pieces of sample were rinsed five times with sterilized distilled water (SDW), dried on sterile filter paper and placed on PDA amended with streptomycin. The Petri dishes were then incubated at 28°C for about 10 days for the fungi to grow and sporulate, after which they were placed on slide for examination under the microscope for identification.

3.2.2 Isolation from Soil

Soil samples from each of the five locations were taken randomly from infected plant at depth of 5cm using soil auger and were put in different plastic bags which were labeled accordingly (location). The samples were taken to the laboratory and later subjected to laboratory analysis for isolating soil borne pathogens associated with fungal diseases of sweet potatoes using the procedure as suggested by Fox (1993). The method used was a soil dilusion plate method (this method is used for the isolation of Fusarium species and some other soil borne fungi from dry soil). The procedure used is as follows:

1. The air dry soil samples were grinded with a mortar and pestle and mixed thoroughly.

2. 10 g of soil sample was taken as subsample and put into 100ml distilled water which gave a dilution ratio of 1:10

3. 10ml was transferred to second container with 90ml of distilled water which gave a dilution ratio 1:100. Similar step was repeated upto a ratio of 1:1000 which is satisfactory for isolation of the Fungi.

4. 1ml of the soil suspension was spread across the PDA in a 90mm Petri dish.

5. The isolation plate was then incubated under light for 5-7 days until colonies developed.

6. The colonies of the Fungi were sub cultured and purified using the single spore technique on the PDA

7. A single spore was taken and placed on the slide to view the morphological feature of the different fungus.

## 3.3 PATHOGENICITY TEST

Pathogenicity test was carried out in the screen house to confirm that the isolated pathogen is responsible for the disease. Local variety *Danchana* and King J (breeding line) were inoculated with ten milliliters (10 ml) spores suspension of the fungus (*Fusarium oxysporum* f. sp *batatas*) which was isolated from diseased leaves of the sweet potato by simply adding the spore suspension to the plastic bucket (20 L) filled with 6 kg sterile soil. Sweet potato vines of two varieties *Danchana* (local variety) and King J (breeding line) were obtained from Department of Agronomy farm, Bayero University Kano. The vines were transplanted on to the inoculated soil in plastic buckets. Un-inoculated soil plastic buckets served as control. At 6 weeks after transplanting, the symptoms of the Fusarium wilt on the sweet potato were compared with those that developed naturally in the field to confirm if they are similar to the diseases recorded in the field. The fungus was re-isolated and observed under microscope to find out if the morphology was similar to one isolated from field.

## 3.4 IDENTIFICATION OF ISOLATED FUNGAL PATHOGENS

The Identification of the isolated fungi was done in the Plant pathology laboratory in the department of crop protection, Faculty of Agriculture, Bayero University Kano. Identification was conducted through charts, descriptions of microscopic images of the pathogenic fungi according to Barnett and Hunter (1998) and Watanabe (2010). The procedure is as follows:-

Fungal culture growing on potato dextrose agar were taken using heat sterilized needle and placed on microscope slide, with a drop of water on glass slide, cover slip was then

placed at 45 degree angle (to minimize air bubbles in the mount) over the glass slide, these were then viewed using the morphological features and the spores produced by the fungus for identification.

## 3.5 FUNGICIDES SCREENING AGAINST *Fusarium oxysporum* f. sp. *batatas* INCITANT OF FUSARIUM WILT OF SWEET POTATO

*In vitro* fungicides screening against *Fusarium oxysporum f. sp. batatas* the causal agent of Fusarium wilt of sweet potato was conducted in the plant pathology laboratory of the Department of Crop Protection, Faculty of Agriculture, Bayero University Kano. Three fungicides HATRICK (Hexaconazole 5%SC), RAKSHA (Mancozeb 80%WP), BLUE BOLTS (Cuprous Oxide 5%+Metalaxyl 12%) used at 5 % and 10 % of the recommended, were used on the fungus (*Fusarium oxysporum)* isolated from diseased samples. Potato dextrose agar (PDA) was autoclaved at 121°C for 15 minutes, 15 ml of the molten PDA medium were poured on to 15 Petri dishes (9-cm diameter) amended with 0.1 ml from each of the respective fungicides excluding the control which was left unamended. After solidification, each plate was inoculated with a 5 mm mycelial disc mat of *Fusarium oxysporum* in the centre of the plates and incubated at room temperature ($28 \pm 2^{o}$ C) for ten days. The plates without the fungicides served as control. Treatments were arranged in a completely randomized design (CRD) with four repetitions.

Food poisoning technique was used in which hyphal growth inhibition on the growth medium incorporated with fungicide were determined according to method described by Ngono *et al.* (2000). This technique involves the poisoning of the fungal growth using antifungal agent and then measuring the reduction of growth of the organism on the

medium. The decrease in mycelial growth indicates the inhibition of fungal growth by the antifungal substance. The radial growth (mm) of the pathogen was measured using meter rule seven days after inoculation and the percent inhibition of the pathogen was calculated according to the formula used by Pandey *et al*. (1995) as $(D_c - D_t) / D_c \times 100$, where $D_c$ is the average diameter increase of fungal colony with control and $D_t$ is the average diameter increase of fungal colony with treatment. Data collected were analyzed using Genstat 17$^{th}$ Edition and treatment means were separated using LSD at 5% level of significance.

## 3.5 PREPARATION OF PLANT EXTRACTS

Calotrophis and Neem leaves were sourced from Bayero University premises while the Garlic bulbs was purchased from Dawanau market in Kano. The plants parts were separately washed under tap water; air dried and ground into powder using a hammer mill. Cold water extraction was obtained by adding 10 and 20 g of the plants powder into 100 ml distilled water in 250 ml beaker. This was left for 24 hours and there after filtered through four fold of sterile muslin cloth to obtain 5 and 10% crude aqueous extract of aqueous neem leaf extract (ANLE), *Calotrophis procera* leaf extracts (CALE) and garlic bulbs extracts (GABE) according to the method used by Taiga (2011)

## 3.6 *IN VITRO* EFFICACY OF NEEM, CALOTROPHIS AND GARLIC BULBS EXTRACTS ON *Fusarium oxysporum* OF SWEET POTATO

*In vitro* screening of some plants extracts against Fusarium wilt of sweet potato was conducted in the teaching and research laboratory of Crop Protection Department, Faculty of Agriculture, Bayero University Kano. The treatments consisted of two

different concentrations (5 and 10%) of Calotrophis leaves, neem leaves and garlic bulb extracts, Mancozeb at 5% of the recommended rate was used as check. The treatments were arranged in a completely randomized design (CRD) and repeated three times. Potato Dextrose Agar (PDA) was amended separately with the fungicide as well as 0.2 ml of the three plants extracts while the control was unamended. The radial growth of the pathogen was measured (using meter rule) was taken separately after seven days of inoculation and the percent inhibition by the plant extracts were calculated according to the formula used by Pandey *et al*. (1995) as $(D_c - D_t) / D_c \times 100$, where $D_c$ is the average diameter increase of fungal colony with control and $D_t$ is the average diameter increase of fungal colony with treatment. Data collected was analyzed using Gens tat 17th Edition, and treatment means were separated using LSD at 5% level of significance.

### 3.7 *IN VIVO* EFFICACY OF SOME PLANT EXTRACTS ON *Fusarium oxysporum* f. sp. *batatas* OF SWEET POTATO

Efficacy of some indigenous plant extracts were tested on two varieties of sweet potato; *Dan Chana* (local variety) and King J (breeding line) against *Fusarium oxysporum f. sp batatas.* The experiment was conducted in screen house, Faculty of Agriculture, Bayero University, Kano. The experiment consisted of ten treatments i.e one breeding line (King J) and local variety (*Danchana*), three plant extracts, one fungicide (Mancozeb). The treatments were laid in a randomized complete block design with four replicates. Three plant extracts were prepared as Aqueous Neem leaf extract (ANLE) 10%, *Calotrophis procera* leaf extracts (CALE) 10% and Garlic bulbs extracts (GABE)10% and control.

Healthy sweet potato vines were surface sterilized in 0.5% sodium hypochlorite solution for three minutes and rinsed in sterile distilled water. The vines were transplanted in plastic buckets containing 6 kg sterile soil. The soil was inoculated by adding 10ml spore suspension of the fungus. Control plants were similarly treated with sterile distilled water and inoculated with pathogen (without plant extracts). The fungicide (mancozeb) and the plant extracts, (neem, calotrophis and garlic) were applied one day before inoculation (as preventive). After 50 days, disease infection was assessed as a total percentage of plants showing any symptoms of Fusarium wilt (yellowing and dropping of leaves, vascular discoloration. Ten percent (10%) of the plant extracts were applied soil to at the rate of 10 ml/plant one day before transplanting. Disease incidence (DI) was computed using the formula used by Anita and Rabeeth (2009): number of plant infected) / total number of plants) X 100. Percent disease severity (S) was calculated using the formula: $S = \{(\sum n) / (N \times \text{highest scale})\} * 100$ (Singh, 2006). Where $\sum n$ = summation of individual ratings, 4 = highest score of severity and N = total number of plant assessed.

Disease severity was taken to assess the disease infection, 50 days after inoculation by using the following scale (Grattidge and O'Brien 1982): 0, (0–24%) of leaves yellowed and wilted; 1, (25–49%) of leaves yellowed and wilted; 2, (50–74%) of leaves yellowed and wilted; 3, (75– 99%) of leaves yellowed and wilted; 4, (100%) dead plant.

## 3.8 DATA ANALYSIS

Disease incidence and severity, vine length, number of leaves and yield were subjected to analysis of variance using GenStat 17$^{th}$ Edition, and treatment means were separated using LSD at 5% level of significance. Data on disease incidence and severity were arc sine transformed before analysis.

# CHAPTER FOUR

## 4.0 RESULTS AND DISCUSSIONS

## 4.1 RESULTS

### 4.1.1 Incidence and Severity of Major Fungal Diseases of Sweet Potato in Kano state.

Table 1 showed the major fungal diseases affecting sweet potato in five selected local governments in Kano state, Nigeria. The major fungal diseases that were identified across the location include Fusarium Wilt, Chlorotic leaf distortions, and Minor leaf spot and Alternaria leaf spot.

Fusarium wilt had the highest incidence score across the location with incidence ranges between 19-61%. Incidence of Fusarium wilt was higher (61%) in Garko. Bagwai gave 47% fusarium wilt incidence, Rimingado gave 45% fusarium wilt incidence, and Madobi had less incidence of Fusarium wilt of 20%. Lowest incidence was recorded in Kibiya with 18% incidence of Fusarium wilt. For the Fusarium wilt disease severity across the four locations, Garko had severity scale of four, Bagwai three, Rimin gado three, Madobi one and Kibiya one.

Incidence of Minor leaf spot across the locations showed Garko with highest incidence (53%), followed by Bagwai (35%), Rimingado (27%) and Madobi (14%), respectively. There was less record of minor leaf spot in Kibiya. For the disease severity scale Garko had a scale of four, Bagwai three, Rimingado three, Madobi two and Kibiya one respectively.

In terms of Alternaria leaf spot of sweet potato on the surveyed locations, Garko recorded the highest disease incidence (49%) compared to Bagwai (35%), Rimingado (28% ) and Madobi (13% ), respectively. Lowest incidence (6%) of Alternaria leaf spot was found in Kibiya compared to the other locations. Disease severity for Garko had severity scale of four, Bagwai had three, Rimingado had two, Madobi one and Kibiya had one.

Incidence and severity of chlorotic leaf distortions in all the surveyed farms across the locations were found to be higher in Garko (33%), followed by Bagwai which had 31% disease incidence. This is greater than the incidence of the disease recorded in Rimin gado (19%), Madobi (1.4%) and Kibiya (0.4%). Disease severity showed that Garko had severity scale of four, Bagwai had four, Rimingado with three, Madobi two and Kibiya had two. Results indicated least occurrence of these fungal diseases in Kibiya followed by Madobi, Rimingado, and Bagwai. Highest incidence and severity was recorded in Garko farms.

Table 1: Incidence and severity of the major fungal diseases of sweet potato in the five surveyed locations in Kano.

| Location | Altenaria leaf spot | | Chlorotic leaf distortion | | Minor leaf spot | | Fusarium wilt | |
|---|---|---|---|---|---|---|---|---|
| | DSI (%) | DSS | DSI (%) | DSS | DSI (%) | DSS | DSI (%) | DSS |
| Garko | 41 | 4 | 33 | 4 | 53 | 4 | 61 | 5 |
| Kibiya | 6 | 1 | 1 | 1 | 1 | 1 | 19 | 2 |
| Bagwai | 34.5 | 2 | 31 | 3 | 35 | 3 | 47 | 4 |
| Rimin gado | 28 | 2 | 19 | 3 | 27 | 2 | 45 | 4 |
| Madobi | 13 | 2 | 1.4 | 1 | 14 | 1 | 20 | 2 |
| SD | 13.53 | 16.456 | 15.61 | 17.25 | 19.56 | 21.48 | 18.32 | 20.23 |

DSI =Diseases incidence DSS = Diseases severity , for Alternaria leaf spot, Minor leaf spot, 0= no diseases, 1 =small traces of lesion, 2 -3 =slight infection( lesion on upper and lower leaves, 4 =moderate infection (advanced lesion on upper and lower leaves with or without new infection on petiole, 5 =severe infection (advanced lesion on upper and lower leaves backs and petiole dark to dark brown. For Fusarium wilt and Chlorotic disease: 1= no symptoms on healthy leaves, 2 = wilted leaves without chlorosis, 3 =one or several slightly chlorotic disease, 4 = chlorotic bands over the entire surface of the leaves or chlorotic bands with necrotic centre, 5= complete necrosis or death of the leaves

4.1.2. Incidence and Severity of Fungal Diseases on Eleven sweet potato Breeding Lines

4.1.2.1 Incidence

Table 2 shows incidence of the four major fungal diseases namely Fusarium wilt, chlorotic disease, Alternaria leaf spot and Minor leaf spot affecting sweet potato in Kano state. *Danchana* (a local variety) had the lowest disease incidence of all the four fungal diseases. It had 12.5% incidence of Alternaria leaf spot, Chlorotic leaf distortion 1.25% , Danchana also had less minor leaf Spot with 9% incidence, Fusarium wilt incidence was 15.3% followed by King J an improved breeding line with score at 14%. The incidence of Alternaria leaf Spot and chlorotic leaf distortion in King J was 5% respectively. Minor leaf Spot was 10% while Fusarium wilt incidence was 18%.

Breeding lines T121, Mother Delight, Gloria, and Delvia had moderate occurrence of these fungal diseases with incidence of 17% - 33 %. Lourdes and Gloria also showed some level of infection by the fungal diseases. Higher disease incidence was recorded on Centennial with incidence ranging from 30%-55% respectively compared to other ten breeding lines.

4.1.2.2 Severity

Disease severity of the four major diseases on the breeding lines showed that Danchana had a severity score of one for Alternaria leaf spot, one for chlorotic leaf distortion, and also minor leaf spot had one while Fusarium wilt severity on Danchana had score of two. King J recorded a severity score of one for Alternaria leaf spot, Chlorotic leaf distortion was also one, Minor leaf spot had one and Fusarium wilt had two. Breeding lines T121, Mother Delight, Gloria, and Delvia had higher severity with

score ranging from 2-4 , Lourdes recorded a higher level of disease severity ranging between 3-4 as compared with Breeding lines T121, Mother Delight, Gloria, and Delvia, while highest disease severity was recorded on Centennial with severity score ranging from 4-5 respectively.

Table 2: Incidence and Severity of the Four (4) Major Fungal diseases on Eleven Sweet potato breeding lines Kano

| Variety | Altenaria leaf spot | | Chlorotic leaf distortion | | Minor leaf spot | | Fusarium wilt | |
|---|---|---|---|---|---|---|---|---|
| | DSI (%) | DSS | DSI (%) | DSS | DSI (%) | DSS | DSI (%) | DSS |
| *Dan chana* | 12.5 | 1 | 1.2 | 2 | 9 | 1 | 15.3 | 2 |
| Centennial | 45.0 | 3 | 30.0 | 3 | 29 | 2 | 55 | 4 |
| Gloria | 35 | 3 | 23 | 2 | 23.5 | 2 | 36 | 3 |
| T121 | 17 | 2 | 8 | 2 | 13 | 1 | 20 | 2 |
| Mother D | 17 | 2 | 9 | 2 | 15 | 2 | 22 | 2 |
| AO305 | 22 | 2 | 11 | 2 | 15.2 | 1 | 23 | 2 |
| Delvia | 23 | 2 | 12.5 | 2 | 17 | 2 | 23 | 2 |
| SumaiA | 25 | 2 | 18 | 2 | 2 | 2 | 30 | 3 |
| Melinda | 33 | 3 | 21 | 2 | 23 | 2 | 33 | 3 |
| Lourdes | 37 | 3 | 25 | 3 | 27 | 2 | 40 | 4 |
| King J | 14 | 1 | 5 | 2 | 10 | 1 | 18 | 2 |
| SD | 10.59 | 10.63 | 9.19 | 9.603 | 8.26 | 6.63 | 11.71 | 11.94 |

DSI =Diseases incidence DSS = Diseases severity, for Alternaria leaf spot, Minor leaf spot, 0= no diseases, 1 =small traces of lesion, 2 -3 =slight infection( lesion on upper and lower leaves, 4 =moderate infection (advanced lesion on upper and lower leaves with or without new infection on petiole, 5 =severe infection (advanced lesion on upper and lower leaves backs and petiole dark to dark brown. For Fusarium wilt and Chlorotic diseases 1= no symptoms on healthy leaves, 2 = wilted leaves with chlorosis, 3 =two or more chlorotic diseases, 4 = chlorotic bands over the entire surface of the leaves or chlorotic bands with necrotic centre, 5= complete necrosis or death of the leaves.

4.1.3 Fungal isolates Associated with infected sweet potato

The isolate of fungi from the soil, leaves and vines of sweet potato grown in the surveyed locations is present in Table 3. The genera *Fusarium* and *Alternaria* were isolated in all the five locations studied. *Aspergillus* and *Macrophimina* were present in Rimingado, Bagwai and Garko but absent in Kibiya and Madobi, while *Geotrichum* was isolated in only two locations (Bagwai and Garko) and was absent in the other three locations.

4.1.4 Yield

Sweet potato yield from the surveyed locations showed that higher yield (12.8t/ha) was obtained in Kibiya followed by Madobi (7.8t/ha), Rimin Gado (4.5t/ha), Bagwai recorded 3.7t/ha, lowest yield was recorded in Garko (3.1t/ha) farm (Figure 1). Figure 2 shows the total yield of each variety from five different locations, *Danchana* gave the highest yield across all the five locations, followed by breeding lines King J, T121, Mother delight, A0305, Delvia and SumaiA. Low yields were recorded in Melinda, Gloria, Lourdes, and Centennial.

4.1.5 *In Vitro* Effect of Synthetic Fungicides on Mycelial Growth and Inhibition of *Fusarium oxysporium f. sp. batatas*

Table 4 shows effects of three commercially available fungicides RAKSHA (Mancozeb), BLUE BOLT (Copper oxide+Metalaxyl) and HATRICK (Hexaconazole) on mycelial growth and inhibition of *Fusarium oxysporium f.sp batatas*, RAKSHA (Mancozeb) was more effective against *Fusarium oxysporum f.s.p batatas* compared to BLUE BOLT (Copper oxide 2%+Metalaxyl 5%) and HATRICK (Hexaconazole12%).

All the fungicides used significantly slowed the mycelia growth of *Fusarium oxysporium f.sp batatas* more than the control. All three fungicides used differ in their effectiveness in slowing down the mycelia growth of the test fungi. The order of effectiveness is as follows: RAKSHA > HATRICK > BLUE BOLT. Similarly, the percentage inhibition of the fungus growth by the fungicides followed similar trend as in the result of inhibition of mycelial growth.

Table 3: Occurrence of Fungal Pathogens from Infected sweet potato across five surveyed locations

| Fungus | Kibiya | Madobi | Rimin Gado | Bagwai | Garko |
|---|---|---|---|---|---|
| *Fusarium oxysporum* | + | + | + | + | + |
| *Altenaria* sp | + | + | + | + | + |
| *Aspergillus* sp | - | - | + | + | + |
| *Macrophomina phaseolina* | - | - | + | + | + |
| *Fusarium solani* | + | + | + | + | + |
| *Geotrichum* sp | - | - | - | + | + |

- + = Present; - = Absent

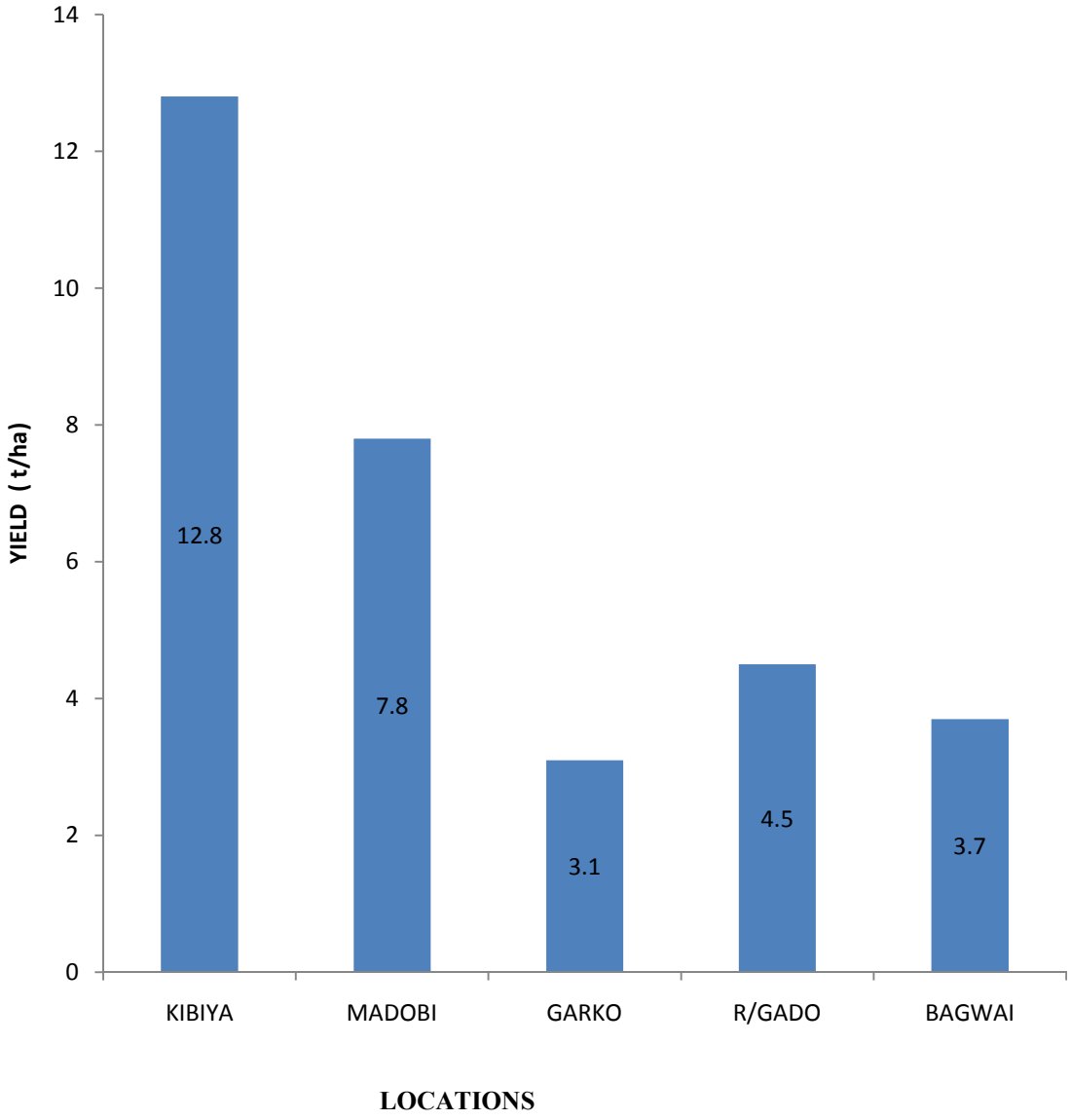


Figure 1: Yield of Sweet Potato in Five Local Government Areas of Kano State

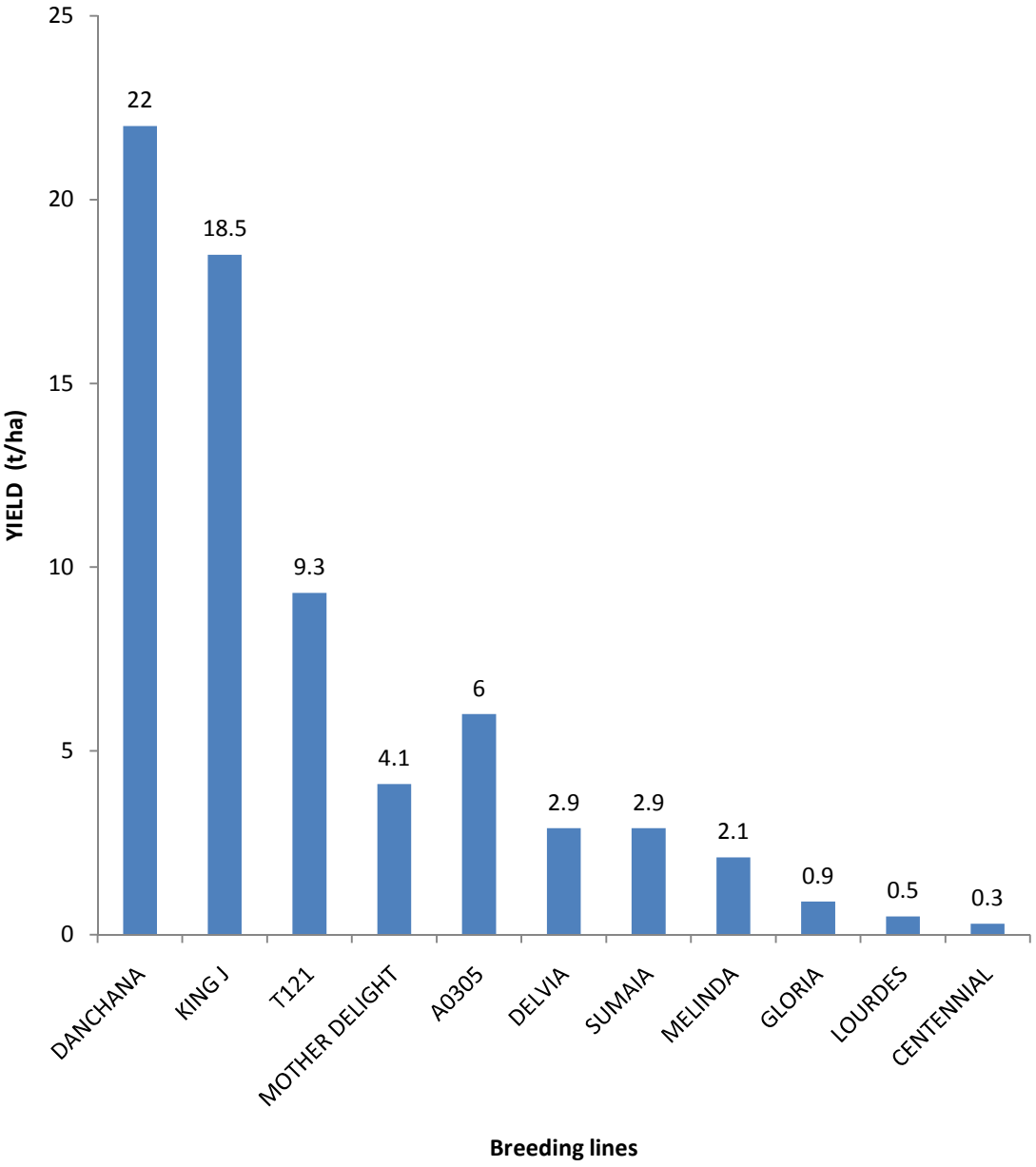


Figure 2: Yield of 11Breeding Lines

Table 4: *In vitro* effects of Fungicides on mycelial growth (mm) and inhibition percentage of *Fusarium oxysporum* f. sp. *batatas*

| Treatment | Mycelia growth (mm) | Mycelia inhibition (%) |
|---|---|---|
| Mancozeb | 13[d] | 71.1[a] |
| Hexaconazole | 26.7[b] | 36.7[c] |
| Metalaxyl +coprous oxide | 21.0[c] | 50.7[b] |
| Control | 42.7[a] | 0.0[d] |
| SE± | 0.91 | 1.88 |
| LS | ** | ** |

Means followed by different letter( s) in a column differ significantly at P≤0.05 using Fishers protected (SI) t ** Highly significant

4.1.6 *In vitro* Effect of Plant Extract on Mycelial Growth and inhibition of *Fusarium oxysporium* f. sp *batatas*

Table 5 shows the effect of plant extracts on mycelial growth and inhibition percentage of *Fusarium oxysporium f.sp batatas.* The result obtained shows inhibitory effect of neem leaf extracts, garlic bulb extracts and calotrophis leaf extracts against *Fusarium oxysporium f.sp batatas* than the control. Neem leaf extracts inhibited 35% on the mycelia disc at 5% concentration and 40% inhibitory action at 10% concentration. This was followed by calotrophis leaf extracts which gave 17.7% at concentration of 5% and 22.7% at 10% concentration, low level of inhibition was observed in garlic bulb extracts with 5% inhibition at 10% concentration and 5% inhibition at 10% concentration. The synthetic fungicide Mancozeb which was used as check inhibited the growth of the fungus up to 45%. The control had zero inhibition.

Table 5: *In vitro* effect of plant extract on mycilial growth and inhibition of *Fusarium oxysporium* f.sp *batatas*

| Treatment | Mycelial Growth (%) | Mycelial Inhibition |
|---|---|---|
| ANLE 5% | $39.3^{d}$ | $35^{b}$ |
| ANLE 10% | $36.7^{d}$ | $40.0^{ab}$ |
| GABE 5% | $58.0^{b}$ | $11.0^{de}$ |
| GABE 10% | $56.3^{b}$ | $5.7^{ef}$ |
| CALE 5% | $53.3^{bc}$ | $17.7^{cd}$ |
| CALE 10% | $49.3^{c}$ | $22.7^{c}$ |
| Mancozeb | $27.0^{e}$ | $42.0^{a}$ |
| Control | $65^{a}$ | $0.00^{f}$ |
| SE± | 2.06 | 2.2 |

Means followed by different letter(s) in a column differ significantly at P≤0.05 using Fishers protected (SI) , ANLE= neem leaf extracts, GABE=garlic bulb extracts, CALE= Callotrophis leaf extracts.

4.1.7 *In vivo* Effect of Botanicals on Incidence of Fusarium wilts

Table 6 shows *in vivo* antifungal effect of three plant extracts. At 6 weeks after transplanting (6WAT) incidence of Fusarium wilt in the inoculated plants treated with Neem leaf extracts at 10% concentration had lower wilting (9.5%) as compared with garlic bulb extracts which had 10% incidence and calotrophis leaf extracts 13.5% incidence while plants treated with Mancozeb had the least incidence (1.5%) of wilting. Statistically there were significant differences between plants treated with Mancozeb and the botanicals (Neem leaf, garlic bulb and calotrophis leaf), while the highest incidence (43%) was recorded in control plants and was significantly different from the botanicals and Mancozeb treated plants.

At 8 (WAT) the result showed that Mancozeb recorded a low Fusarium wilt incidence of 9.5% followed by Neem leaf extract which recorded 12.0% wilting incidence, Garlic also recorded wilting incidence of 17.0% and calotrophis had 31% (10% concentration). The highest record of Fusarium wilt incidence was observed in the control which had 50% incidence. At 10 (WAT) Mancozeb also showed low Fusarium wilt 9.0% incidence and was at par with Neem leaf extract which also had 9.0% incidence. Incidence was much higher in Garlic 43.8% and calotrophis 42% incidence and the highest incidence was in the control which had 63% incidence. Results obtained at 12WAT showed that Mancozeb treated plants had lowest incidence 20% followed by Neem 22.5%, then garlic 37.5% while calotrophis 45.2% which was highest when compared with the other two botanicals. Control plants had 51.7% disease incidence and was the highest when compared with the other treatment.

Table 6: *In vivo* Effect of botanical extracts on Incidences of Fusarium wilt of sweet potato

| Treatments | Weeks after Transplanting (WAT) | | | |
|---|---|---|---|---|
| Varieties | 6 | 8 | 10 | 12 |
| Danchana | 9.0(9.20) | 14.4(19.7) | 32.8(30.4) | 27.3(30.69) |
| King J | 20.7(25.7) | 27.8(28.7) | 40(37.4) | 45.2(42.16) |
| SE± | 2.16(0.78) | 0.8(0.5) | 1.11(0.65) | 0.80(0.518) |
| Plants extracts | | | | |
| NEMLE | 9.5(9.7$^{d}$) | 12.0(12.7$^{d}$) | 9.0(12.9$^{C}$) | 22.5(30.36$^{d}$) |
| CALE | 13.5(14.7$^{c}$) | 31.0(33.03$^{b}$) | 42(47.4$^{b}$) | 45.2(42.18$^{b}$) |
| GABE | 10.0(15.3$^{c}$) | 17.0(21.3$^{c}$) | 43.8(48$^{b}$) | 37.5(37.59$^{c}$) |
| Mancozeb | 1.5(5.0$^{b}$) | 9.5(9.05$^{d}$) | 9(9.8$^{c}$) | 20.0(26.01$^{e}$) |
| Control | 43.7(41.4$^{a}$) | 50.0(45.0$^{a}$) | 63(54$^{a}$) | 51.7(45.96$^{a}$) |
| SE± | 1.37(1.25) | 1.26(0.8) | 1.75(1.02) | 1.79(1.159) |
| Interaction V x BS | ** | ** | ** | ** |

Means followed by different letter (s) significantly at P≤ 0.05 using Fishers protected (SI), Significant, ** =highly significant, WAT = Weeks after Transplanting

Table 7 show disease incidences as affected by interaction between botanical and variety at (6WAT). Disease incidence on the variety King J treated with NEMLE, CALE and Mancozeb were statistically the same (0.0%). these were followed by King J treated with GABE (4.05%), then Danchana treated with Mancozeb (9.9%) and Danchana treated with NEMLE (19.37%), GABE (26.57%) and CALE (31.3%) respectively. For both varieties the control treatments had the highest incidence of (40.78%) Danchana and (41.93%) King J.

Table 8 shows the interaction between botanical and variety on the incidence at (8WAT), Danchana treated with Mancozeb (3.97%), followed by Danchana treated with GABE (8.13%) and also King J treated with Mancozeb which had (8.13%) incidence respectively and were statistically at par with the Danchana treated with Mancozeb (3.97%). These were followed by Danchana treated NEMLE which recorded (15.43%), then King J treated with CALE (22.8%) then King J treated with GABE (34.45%) and then King J in control plants (42.13%). The highest incidence were recorded on Danchana treated with CALE (43.27%) and Danchana in control plants (47.87%) which were significantly not different from one another.

Table 9 shows the interaction between botanical and variety (10WAT), Danchana treated with Mancozeb had the lowest incidence (0.00%), followed by King J treated with NEMLE (11.54%) and Danchana treated with NEMLE (14.18%) which were statistically the same. These were followed by King J treated with Mancozeb (19.66%), then followed by Danchana treated with GABE (43.09%), followed by Danchana treated with CALE (46.72%), and then King J treated with CALE (48.08%), followed by Danchana in control plants (49.82%), and the highest incidence were also recorded in King J treated with GABE (52.54%), and King J in control plants (54.94%) respectively.

Table 7: Interactions between variety and botanicals on the disease incidence of Fusarium wilt of sweet potato At 6 Weeks after Transplanting (WAT)

| | VARIETY | |
|---|---|---|
| TREATMENT | DAN CHANA | KING J |
| CALE | $31.3^{b}$ | $0.00^{g}$ |
| GABE | $26.57^{c}$ | $28.05^{C}$ |
| NEMLE | $19.37^{d}$ | 0.00 |
| Mancozeb (Fungicide) | $9.97^{e}$ | $0.00^{g}$ |
| Control | $40.78^{a}$ | $41.93^{a}$ |
| SE± | 4.13 | |

Means followed by different letter (s) differ significantly at P≤ 0.05 using Fishers protected (SI)

Table 8 Interactions between variety and botanicals on the disease incidence of Fusarium wilt of sweet potato At 8 Weeks After transplanting

| TREATMENT | VARIETY | |
|---|---|---|
| | DAN CHANA | KING J |
| CALE | $43.27^{a}$ | $22.8^{c}$ |
| GABE | $8.13^{d}$ | $34.45^{b}$ |
| NEMLE | $15.34^{e}$ | $19.97^{e}$ |
| Mancozeb (Fungicide) | $3.97^{f}$ | $8.13^{d}$ |
| Control | $47.87^{a}$ | $42.13^{a}$ |
| SE± | 1.028 | |

Means followed by different letter (s) significantly at P≤ 0.05 using Fishers protected (SI)

Table 9: Interactions between variety and botanicals on disease incidence of Fusarium wilt at 10 Weeks after Transplanting

| TREATMENT | VARIETY | |
|---|---|---|
| Plant Extracts | DAN CHANA | KING J |
| CALE | 46.72$^{d}$ | 48.08$^{cd}$ |
| GABE | 43.09$^{e}$ | 52.54$^{ab}$ |
| NEMLE | 14.18$^{g}$ | 11.54$^{g}$ |
| Mancozeb (Fungicide) | 0.00$^{h}$ | 19.66$^{f}$ |
| Control | 49.82$^{bc}$ | 54.94$^{a}$ |
| SE± | 1.445 | |

Means followed by different letter (s) significantly at P≤ 0.05 using Fishers protected (SI)

4.1.8 *In Vivo* Effect of botanicals on severity of Fusarium wilt on Sweet Potato

Table 10 shows the severity of Fusarium wilt as influenced by botanicals extracts, At 6 weeks after transplanting (6WAT) severity of Fusarium wilt in the inoculated plants shows that there is low diseases severity in plant treated with Mancozeb 11.75% followed by Neem leaf extracts with 13.9% which was statistically not different from Mancozeb, calotrophis recorded 34.4% and plant treated with Garlic had 37.5%, higher Fusarium wilt severity was recorded in control plant (65%). At 8WAT the plants treated with Mancozeb had 9.1% Fusarium wilt severity which is the lowest followed by Neem leaf extracts at 12.7% severity, Garlic 27.9%, calotrophis recorded 34.6%. Higher severity was also recorded in control plants with 49%. From the result plant treated with Mancozeb and Neem leaves extracts had low severity.

At 10 and 12 WAT, plants treated with Mancozeb gave 9.6% Fusarium wilt severity which is still the lowest then followed by Neem with 12.9% severity; plant treated with garlic recorded 47.3%, calotrophis 51% and control 65%. At 10 and 12 WAT there was higher Fusarium wilt severity in plant treated with garlic, calotrophis and control plants.

Table 10: *In vivo* Effect of botanical extracts on severity of Fusarium wilt of sweet potato

| Treatment | Weeks after Transplanting | | | |
|---|---|---|---|---|
| Variety | 6 | 8 | 10 | 12 |
| Dan chana | 25.5(28.4) | 18.7(23.0) | 34.7(32.7) | 25.5(29.25) |
| King J | 33.3(32.4) | 30.0(30.1) | 41.7(38.5) | 40.6(39.45) |
| SE± | 2.72(1.80) | 3.25(2.40) | 2.55(1.90) | 0.87(0.581) |
| Plants extracts | | | | |
| Neem | 6.5(13.9$^{c}$) | 11.0(12.7$^{bc}$) | 7.8(12.9$^{c}$) | 22.5(27.33$^{d}$) |
| Calotrophis | 32.2(34.4$^{b}$) | 33.2(34.6$^{b}$) | 59.5(51$^{b}$) | 39.8(39.08$^{b}$) |
| Garlic | 38(37.5$^{b}$) | 25(27.9$^{c}$) | 54(57.3$^{b}$) | 34.2(35.55$^{c}$) |
| Mancozeb | 4.7(11.75$^{c}$) | 2.5(9.1$^{d}$) | 4.3(9.6$^{c}$) | 17.5(24.10$^{e}$) |
| Control | 65(54$^{a}$) | 56.2(49$^{a}$) | 65(54.4$^{a}$) | 51.2(45.67$^{a}$) |
| SE± | 4.3(3.7) | 5.14(2.40) | 4.02(4.9) | 1.94(1.300) |
| Interaction V x BS | ** | ** | ** | ** |

Means followed by different letter (s) differ significantly at P≤ 0.05 using Fishers protected (SI)** =highly significant

4.1.9 Effects of variety and Plant Extracts on Vine Length (cm) of Sweet Potato Plants infected with Fusarium Wilt

Table 11 shows that both Danchana and King J varieties were statistically the same in respect of the vine length parameter. However, there were significant differences in the effects of plant extracts and fungicide on vine length with the exception of the first week. Plants treated with Mancozeb produced higher vine length at 6,8,10 and 12 WAT ( 37.3cm, 55.2cm, 77.0cm and 102.0cm respectively), next after Mancozeb is Neem leaf extracts which also produced higher vine length (35.8cm, 44.3cm, 66.0cm and 84.3cm) across the weeks next is calotrophis (21.7cm, 24.8cm, 27.7cm and 21.1cm) and Garlic leaf extracts (19.3cm, 24.5cm, 26.0cm and 28.1cm). Shorter vines were observed in control sweet potato across the weeks. This shows that Mancozeb and Neem leaf extracts are at par statistically and significantly differently from calotrophis, Garlic leaf extracts and control.

Table 11: Effect of variety/breeding lines and Plant Extracts on Vine Length (cm) of Sweet Potato infected with *Fusarium oxysporum* f .sp *batatas*

| Treatments | Weeks After Transplanting | | | | |
|---|---|---|---|---|---|
| Varieties | 5 | 6 | 8 | 10 | 12 |
| Dan chana | 13.9 | 23.5 | 31.1 | 39.3 | 48 |
| King J | 15.6 | 28.0 | 35.7 | 45.7 | 53 |
| SE± | 1.8 | 2.2 | 2.9 | 3.24 | 5 |
| LSD | NS | NS | NS | NS | NS |
| Plant extracts | | | | | |
| Neem | 17.1 | $35.8^{a}$ | $44.3^{b}$ | $66.0^{b}$ | $84.3^{b}$ |
| Calotrophis | 14.8 | $21.7^{b}$ | $24.8^{a}$ | $27.7^{c}$ | $21.1^{a}$ |
| Garlic | 14.9 | $19.3^{b}$ | $24.5^{a}$ | $26^{c}$ | $28.1^{ab}$ |
| Mancozeb | 16.2 | $37.3^{a}$ | $55.2^{b}$ | $77.0^{a}$ | $102^{b}$ |
| Control | 10.5 | $14.5^{c}$ | $18.2^{a}$ | $16^{d}$ | $14^{a}$ |
| SE± | 2.76 | 3.5 | 4.51 | 5.00 | 7.8 |
| LSD | NS | * | * | * | * |

Means followed by different letter (s) differ significantly at P≤ 0.05 using Fishers protected (SI) NS =Not Significant, ** =highly significant

4.1.10 Effect of Plant Extracts on Number of Leaves of Sweet Potato infected with *Fusarium oxysporum* f.sp *batatas*

Table 12 shows the effects of plant extracts and fungicide on the number of leaves. The result obtained shows that plants extracts and Mancozeb had significant effects on number of leaves as it relate to occurrence of wilting per plants per treatments at 5-WAT plants treated with Mancozeb had higher number of leaves then followed by Neem leaf extracts, Garlic leaf extracts and Calotrophis had little number of leaves but statistically similar. Control plants without had least number of leaves but statistically at par with garlic leaf extracts and calotrophis leaf extracts. At 6-WAT plants treated with Mancozeb were also significantly higher than Neem in the number of leaves but neem was significantly higher than garlic leaf extracts and calotrophis in the number of leaves. Control plants recorded low number of leaves and significantly lower than garlic leaf extracts and calotrophis in the number of leaves. Similar results were obtained at 8,10 and12WAT with the Mancozeb treated plants having higher number of leaves than Neem leaf extracts garlic leaf extracts and calotrophis leaf extracts but statistically similar with Neem leaf extracts. At 12WAT mancozeb is significantly higher than the rest of the treatments but statistically similar with Neem leaf extracts then followed by garlic leaf extracts and calotrophis in the number of leaves.

Table 12: Effect of Plant Extracts on Number of Leaves of Sweet Potato infected with *Fusarium oxysporum* f.sp *batatas*

| Treatments | Weeks After Transplanting | | | | |
|---|---|---|---|---|---|
| Varieties | 5 | 6 | 8 | 10 | 12 |
| Dan chana | 22.5 | 29.6 | 36.4 | 47 | 50.3 |
| King J | 28 | 35.3 | 42.3 | 51.3 | 58 |
| SE± | 9.31 | 10.52 | 9.28 | 7.43 | 7.23 |
| LSD | NS | NS | NS | NS | NS |
| Plants Extracts | | | | | |
| Neem | 37.0ab | 51.0$^{ab}$ | 68.0$^{a}$ | 84.5$^{b}$ | 96.2$^{b}$ |
| Calotrophis | 12.3b | 18.0$^{bc}$ | 18.3$^{b}$ | 19.0$^{b}$ | 18.8$^{a}$ |
| Garlic | 16.2b | 18.5$^{bc}$ | 17.7$^{b}$ | 18.2$^{a}$ | 17.7$^{a}$ |
| Mancozeb | 53.5a | 65.3$^{a}$ | 80.8$^{a}$ | 110$^{b}$ | 124$^{b}$ |
| Control | 6.8b | 9.5$^{c}$ | 11.8$^{b}$ | 12.5$^{a}$ | 13.3$^{a}$ |
| SE± | 9.31 | 10.52 | 9.28 | 7.43 | 7.23 |
| LSD | * | * | * | * | * |

Means followed by different letter (s) differ significantly at P≤ 0.05 using Fishers protected (SI) NS =Not Significant, ** =highly significant

4.1.11 Effect of Plant Extracts on the Yield (Kg) of Sweet Potato infected with Fusarium Wilt

Table 13 shows effect of botanicals on the yield (kg) of sweet potato infected with Fusarium wilt. The result obtained indicated that *Danchana* local variety gave higher yield than King J. From the result obtained on the effects of plants extracts and mancozeb on the yield shows there was a significant difference among the treatments on yield. *Danchana* treated with Mancozeb had higher yield than King J. Yield of *Danchana* treated with Mancozeb was statistically similar with *Danchana* treated with Neem. Significant difference was observed in King J treated with Neem. Low yields were obtained in *Danchana* treated with garlic leaf extracts and calotrophis leaf extracts but significantly differ with king J treated with garlic leaf extracts and calotrophis leaf extracts. No significant difference was observed in *Danchana* and King J in control plants. Lowest yield were obtained in control *Danchana* and King J breeding lines i.e. those without any treatments.

Table 13: Effect of Plants extracts on the Yield (Kg) of Sweet Potato in the Control of Fusarium Wilt

| PLANT EXTRACTS | VARIETY | |
|---|---|---|
| | DAN CHANA | KING J |
| Neem leaf extracts | 1.9[a] | 1.3[b] |
| Calotrophis Leaf Extracts | 1.0[e] | 0.7[f] |
| Garlic Bulb Extracts | 0.9[e] | 0.6[f] |
| Mancozeb (Fungicide) | 2.2[a] | 1.7[c] |
| Control | 0.3g | 0.2g |
| SE± | 0.10 | |

Means followed by different letter (s) differ significantly at P≤ 0.05 using Fishers protected (SI)

4.1.12 Correlation Analysis:

Correlation matrix between disease incidence and severity with growth parameters and yield is presented in Table 14. There was a negative and highly significant difference between disease incidence and severity with vine length, number of leaves and yield. Yield had significantly negative correlation with disease incidence (-0.888**) and severity (-0.876**) and had significant positive correlation with vine length (0.783**) and number of leaves (0.827**). Diseases incidence exhibited highly significant negative correlation with vine length (-0.878**) and number of leaves (-0.894**), while disease severity also had negative correlation with vine lengh(-0.886**) and number of leaves (-0.898**).

Table 14: Correlation coefficient between Fusarium Wilt Disease and Agronomic Components/Yield parameter of sweet potato infected with *Fusarium oxysporum* f.sp *batatas.*

| | DI | DS | VL | NL | YD |
|---|---|---|---|---|---|
| DI | 1.000 | | | | |
| DS | 0.970** | 1.000 | | | |
| VL | -0.878** | -0.886** | 1.000 | | |
| NL | -0.894** | -0.898** | 0.890** | 1.000 | |
| YD | -0.898** | -0.876** | 0.783** | 0.827** | 1.000 |

DI = Diseases incidence, DS = Disease severity, NL = Number of Leaves, VL =Vine length, YD = Yield

## 4.2 DISCUSSION

### 4.2.1 Incidences and Severity of the Major Fungal Diseases of Sweet Potato in Location in Kano state.

Result of the survey indicated that low occurrence of fungal diseases were found in Kibiya then followed by Madobi, Rimingado, and Bagwai respectively. The highest incidence and disease severity was recorded in Garko. The incidence and severity of the fungal diseases of sweet potato are bound to be influenced by differences in climatic conditions, because the survival and spread of different fungi varies with temperature and humidity. However disease management practices by farmers also played a major role in determining incidence and severity across the sweet potato surveyed locations. The diverse cropping pattern like in Gulu, village Rimingado mono-cropping system of sweet potato may facilitate the survival and propagation of these pathogens, since fungi are able to survive as saprophyte.

Four major fungal diseases were isolated and identified in Kano state. The Major fungal diseases identified include Fusarium Wilt, Chlorotic leaf distortions, Minor leaf spot and Alternaria leaf spot. All the four fungal diseases affecting sweet potato breeding lines were found almost in all the surveyed locations. Fusarium wilt recorded highest incidence and severity across the location with incidence ranges between 20-61%. Nelson *et al*, (1994) reported that the wide spread distribution of *Fusarium* species has been attributed to the ability of these fungi to grow and survive even in the absent of the host environments on a wide range of soil substrates and their efficient mechanisms for spore dispersal. Gunua,(2010) reported that past research has shown that impacts of *Fusarium oxysporum* can also be worse when infections are combined with some types of plant stress that compromise plant health

and growth, The prevalence of Fusarium may be due to plant stress as a result of high temperatures and severe water deficiency

All the four fungal diseases were found infecting the breeding lines of sweet potato. *Danchana* a local variety had the lowest disease incidence and severity irrespective of fungal disease. This may be due to the fact that the variety is well adapted in the area and have developed a kind of resistance to these fungal diseases compared to the other breeding lines. This was followed by King j which had significantly lower disease incidence compared with T121. Gloria infection was lower than that of centennial. The occurrence of fungal diseases on the breeding lines may be attributed to the fact that they are new in the area and prone to the attack by these fungal diseases.

4.2.3 *In vitro* Effect of Synthetic Fungicides on Mycilial Growth and Inhibition of *Fusarium oxysporum f.sp. batatas*

The fungus mycelial growth and percent inhibition was significantly inhibited (71.1%) by RAKSHA (Mancozeb) compared to BLUEBOLT (cuprous oxide +metalxyl) 21.0%, HATRICK (Hexaconazole)26.7% and, control plant which had low zone of inhibition. This was in agreement with the findings of Obagwu (1997) who reported Mancozeb effectiveness in the control of brown blotch of Bambara nut caused by *Colletotrichum capsici*. Osunlaja and Alamata (1999) also reported that Mancozeb significantly inhibited *Physoderma maydis* on maize (Brown spot). BLUE BOLT (Copper oxide+Metalaxyl) depicted its low level efficacy with inhibition percentage of 21.0%. However, it reduced the mycelia growth and to certain extent higher percent inhibitory action when compared with control which had zero inhibition.

Alam *et al*. (1999) also found out that the effectiveness of copper oxide in the control of *Alternaria tenuis* in field experiments. HATRICK (Hexaconazole) had 26.7% growth inhibition with little reduction in mycelia growth and was significantly higher than control. Contrary to *in vitro* evaluation of Hexaconazole, Pampanagouda (2000) reported 100% inhibition against *Elsinoe ampelina* causative agent of Anthracnose of grapes. Many *in vitro* studies have demonstrated that some fungicides restrict or prevent the growth of fungal pathogens (Marley and Gbenga 2004). In the literature there are only few reports about the influence of fungicides such as mancozeb, metalaxyl+copper oxide and hexaconazole on mycelial growth of *Fusarium oxysporum fsp batatas* of sweet potato.

The results indicated that among the plants extracts evaluated *in vitro* against *Fusarium oxysporium f.sp batatas* Neem was significantly superior (35% inhibition) at 5% concentration and (40%inhibition) at 10% concentration and all the other plants extracts followed by calotrophis (18% inhibition) at 5%conc and(23%inhibition)at 10% concentration, which was at par with Garlic (5% inhibition) at 5% concentration and (11% inhibition) at 10% concentrations. Garlic was found to be the least effective among the plant extracts. This is in agreement with the results obtained by many researchers. Amadioha, (1999) found that Neem seeds and leaf extracts have reduced the growth of the fungi *Pyricularia oxyzae* in rice. In another study, Joseph and Kumar (2008) found Neem leaf extract (20% concentration) as the most effective among five plant extracts, in the control of *Fusarium* wilt. Dubey *et al*. (2009) also found that different extracts of Neem plant parts including leaf, bark, oil cake and Neem oil effective against mycelial growth of *Mycosphaerella phaseolina* isolated from charcoal rot of soybean, with highest effectiveness of 10% concentration, Agbenin and Marley (2006), reported that the dry neem seed extract completely

suppressed the mycelial growth of *F. oxysporum* at all concentrations, while extracts of fresh neem leaves reduced mycelial growth of fungus with increasing concentrations. Neem extract was also found by Da-Costa *et al*. (2010) to have inhibited the fungal growth (*i.e.* mycelia dry weight, diameter of colony and growth rate) of *Aspergillus flavus* on solid media at concentrations from 0.5 to 5.0% v/v,

The effect of *Calotropis procera* extracts against *Fusarium oxysporium f.sp batatas* agrees with the work of Abdulmoniem *et al*. (2012) who evaluated the antimicrobial activity of extracts of leaf and latex of *Calotropis procera,* on Fusarium wilt of cucurbits. Wilson *et al.* (1997) worked on Plant extracts of *Allium extracts* and showed that Garlic completely (100%) inhibited spore germination of *Botrytis cinerea, and* Afzal *et al*. (2010) reported *Allium sativum* to have a wide antifungal spectrum that effected 60-82% inhibition in the growth of seed borne *Aspergillus* and *Penicillium* fungi which differs with this present result.

Mancozeb inhibited the growth of the fungi with 45% inhibition. This result was similar with the result of Pampanagouda, (2000) who found out Mancozeb among the non systemic fungicides to be superior in inhibiting some fungal growth *in vitro*. The result shows that the inhibition increased as the concentrations of all extracts increased. Statistically, inhibitory activity of Neem leaf extracts, and the Mancozeb are the same.

4.2.4 Effect of Plant Extracts on Incidence and Severity of Fusarium wilt of Sweet Potato in the Screen House.

The present result showed *in vivo* antifungal test of three plant extracts on the control of Fusarium wilt of sweet potato. Disease incidence of Fusarium wilt in the inoculated plants which were recorded at 6, 8 and 10 weeks after transplanting

showed low disease incidence in plant treated with mancozeb at 6WAT then followed by Neem leaf extracts. Also, there was low wilting incidences recorded with the plants treated with Neem as compared with garlic bulb extracts and calotrophis leaf extracts. From the result Mancozeb treated plants recorded low incidence of fusarium wilt of sweet potato and this is in full agreement with the work of Vawdrey *et al*. (2004) who reported that disease assessment conducted 2 weeks before harvest in the screen house showed that mancozeb applied at the rate of 100g a.i. $ha^{-1}$ was effective than higher rates of 1760, 2000, or 2500g a.i.$ha^{-1}$in controlling yellow Sigatoka disease of banana. James *et al*. (2005) also reported that, in field, Mancozeb was the most effective among other fungicides tested in the control of anthracnose on three cultivars of *Euonymus fortune.*

There was also low level of Fusarium wilt incidence in plants treated with Neem leaf extracts and this finding is similar to result obtained by Amadioha, (2000) where *Neem* extract significantly reduced *in vitro* mycelial growth (83.6%) of *Pyricularia oryzae* (causing rice blast) while, *in vivo* application (through spray) two days before and after inoculation reduced the disease incidence by 10.2% to 19.5%, respectively Hanaa *et al*. (2011) investigated the effect of *Azadirachta indica* (Neem)10% aqueous extracts on *Fusarium* wilt disease in tomato and revealed that the percentage of disease incidence was reduced to the level of 25.5 % and 27.8 % after 6 weeks of infection respectively.

Disease incidence is more pronounced in plants treated with garlic and calotrophis leaf extracts but the highest incidence of wilting was in the control. Obagwu and korsten (2003) showed the antifungal effect of garlic on plant pathogens for the control of *Fusarium oxysporum* f.sp *phaseoli*. Calotrophis leaves extracts which has less effect in controlling Fusarium wilt had higher incidence, this agrees

with the work of Mossa *et al.*,(1991) who earlier reported that *C. procera* extract is devoid of any antibacterial and antifungal activity.

Plants treated with Mancozeb showed less severity of Fusarium wilting and this agrees with the work of Naab *et al*., 2005) that shows efficacy of mancozeb in reducing the incidence and severity of *Cercospora* leaf spot disease of groundnut. Neem also reduced severity of Fusarium wilt this is also similar with the result of Alabi and Olorunju, (2004) that the Neem seeds and leaves extract at the rate of 30:70 (v:v ) had significantly inhibited the growth of the fungi *Rhizoctonia solani and Fusarium solani* at percentages of 52.4% and 37.5% and reducing the aggressiveness of the diseases respectively. Mercado and Rodriguez (2001) treated two soil borne phytopathogens (*Ralstonia solani* and *Myrothecium roridum*) with garlic compound called ‘‘Garlic Barrier’’ and found little effectiveness in reducing populations of either pathogen in screen house.

4.2.5 Effect of Plant Extracts on Number of Leaves and Vine Length of Sweet Potato infected with Fusarium Wilt

Plants treated with Mancozeb produced higher vine length and number of leaves than other treatments. Neem leaves extracts also produced lengthy vines and reduced the Fusarium wilting of leaves increasing the number of leaves. This finding is similar to the finding of Enikuomehin, (2005) who reported that Neem leaf extracts substantially reduced the number of infected leaves and number of lesion on foliage of jute plant which in turn protected flowers and increased number of leaves on Cercospora leaves spot.

Calotrophis and Garlic leaf extracts that produced short vines with decreasing number of leaves due to effects of Fusarium wilt incidences but at par with control

sweet potato that was characterized by serious wilting which adversely affected vines length and number of leaves across 6,8,10,and 12 WAT.

4.2.6 Effect of Plant Extracts on yield (kg) of sweet potato infected with Fusarium wilt in screen house

From the result obtained on the effects of plants extracts and synthetic fungicide (mancozeb) on the yield there was a significant difference among the treatments on the sweet potato varieties. Local variety *Danchina* treated with Mancozeb had higher yield (2.2kg) and significantly higher than King J treated with Mancozeb with (1.7kg).This finding is similar to the report of (RMRDC, 2004) that mancozeb provided satisfactory control of *Cercospora* leaf spot disease of groundnut in northern Nigeria and thus increased yield.

*Danchana* treated with Neem which had (1.9kg) and King J treated with Neem had (1.3kg). This is similar to finding of Hosna *et al.,* (2003) who reported that Neem extract controlled *Alternaria* blight (*Alternria brassicicola* and *Alternaria. brassica*) of cauliflower seed increasing number of leaves and yield. Aboellil (2007) also reported that trilogy, a natural product from Neem tree, significantly retarded growth of cucumber powdery mildew, and induced resistance in cucumber plants thereby increasing yield. Low yield were obtained from the plants varieties treated with garlic leaf extracts 0.6kg-0.9kg and calotrophis leaf extracts (0.7-1.0kg). Report by Pathak and Zaidi (2013), shows that latex and leaves of *Calotrophis procera* has been found quite effective in controlling the seed-borne mycoflora of wheat and promoting growth leading to higher yield.

Tohamy *et al.* (2002) reported effectiveness of Garlic bulb extracts in reducing the activity of cucumber soil-borne pathogens, Similarly, weekly foliar

sprays of the extract reduced the incidence of both powdery and downy mildews. Sallam *et al* (2012), tested the antimicrobial activities of Garlic for controlling early blight (*Alternaria solani) in vivo*, in greenhouse experiments the highest reduction of disease severity was achieved by the extracts of garlic at 5% concentration. Garlic extract at 5% concentration increased the fruit yield by 76.2% and 66.7% compared to the control plants.

# CHAPTER FIVE

## 5.0 SUMMARY, CONCLUSION AND RECOMMENDATIONS

### 5.1 SUMMARY

The survey was conducted in five selected local government areas of Kano state. The local government are; Rimin Gado (Gulu village), ($11^0$ 57'54"N $8^0$ 15'0"E), Madobi (Gara village) ($11^0$ 46'18"N $8^0$ 17'18"E), Kibiya (Fagam village) ($11^0$ 32'N $8^0$ 40"E), Bagwai ($12^0$ 09'28"N $8^0$ 08'09"E), and Garko (Sarina village) ($11^0$ 39'N $8^0$ 50'E) located within Sudan savannah ecological zone of Nigeria. Result of the surveyed locations showed higher occurrence of Fusarium wilt incidence and severity at Garko while lowest incidence and severity was recorded in Kibiya. Fusarium wilt caused by *Fusarium oxysporum* f.s.p *batatas* was the most occurring fungal disease compared to the other diseases identified. Mancozeb had the highest efficacy against the fungus *Fusarium oxysporum* f.s.p. *batatas* in comparison with other botanicals . Aqueous Neem leaf extract recorded the highest percent inhibition and lowest mycelia growth of the pathogen (*Fusarium oxysporum f.sp batatas)* than garlic bulb and Calotrophis leaf extracts. Result showed that effective reduction of disease was obtained when *Danchana* was grown on soil amended with Aqueous Neem leaf extract than other leaf extracts.

### 5.2 CONCLUSION

Based on the results obtained, It is concluded that Kibiya had the lowest incidence and severity of the fungal diseases. Mancozeb is the most effective fungicide that controlled Fusarium wilt pathogen, growing *Danchana* on soil amended with Aqueous Neem leaf extracts was the most effective botanical in the

control of Fusarium wilt diseases of sweet potato. Neem leaf extracts could therefore be used as bio fungicide for the management of Fusarium wilt of sweet potato.

## 5.3 RECOMMENDATIONS

The following recommendations could be made as ways of managing Fusarium wilt of sweet potato:

1. Mancozeb should be use as fungicide for managing Fusarium wilt pathogen *in vivo*.
2. Farmers should use Danchana (local variety) and King J (breeding line) with 10% aqueous Neem leaf extract as integrated management of Fusarium wilt of sweet potato.
3. Due to the prevalence of Fusarium wilt across the locations farmers should select disease resistance varieties and adopt other agronomic practices such as crop rotation as most of the surveyed locations rely on mono-cropping system of sweet potato.

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APPENDIX

APPENDIX 1: Analysis of variance of the effect of Fungicides on percent inhibition of *Fusarium oxysporum* f. sp. *batatas*

| Source of variation | d.f. | s.s. | m.s. | v.r. | F pr. |
|---|---|---|---|---|---|
| REP stratum | 2 | 35.23 | 17.61 | 1.67 | |
| REP.*Units* stratum | | | | | |
| FUNGICIDE | 3 | 8080.76 | 2693.59 | 254.86 | <.001 |
| Residual | 6 | 63.41 | 10.57 | | |
| Total | 11 | 8179.40 | | | |

APPENDIX 2: Analysis of variance of the effect of Fungicides on myceclial growth of *Fusarium oxysporum* f. sp. *batatas*

| Source of variation | d.f. | s.s. | m.s. | v.r. | F pr. |
|---|---|---|---|---|---|
| REP stratum | 2 | 5.167 | 2.583 | 1.04 | |
| REP.*Units* stratum | | | | | |
| FUNGICIDE | 3 | 1390.917 | 463.639 | 187.54 | <.001 |
| Residual | 6 | 14.833 | 2.472 | | |
| Total | 11 | 1410.917 | | | |

APPENDIX 3: Analysis of variance of the effect of plant extracts on percent inhibition of *Fusarium oxysporum* f. sp. *batatas*

| Source of variation | d.f. | s.s. | m.s. | v.r. | F pr. |
|---|---|---|---|---|---|
| REP stratum | 2 | 143.58 | 71.79 | 6.74 | |
| REP.*Units* stratum | | | | | |
| EXTRACTS | 7 | 4208.67 | 601.24 | 56.46 | <.001 |
| Residual | 14 | 149.08 | 10.65 | | |
| Total | 23 | 4501.33 | | | |

APPENDIX 4: Analysis of variance of the effect of plant extracts on myceclial growth of *Fusarium oxysporum* f. sp. *batatas*

| Source of variation | d.f. | s.s. | m.s. | v.r. | F pr. |
|---|---|---|---|---|---|
| REP stratum | 2 | 6.333 | 3.167 | 0.40 | |
| REP.*Units* stratum | | | | | |
| EXTRACTS | 7 | 3868.958 | 552.708 | 70.56 | <.001 |
| Residual | 14 | 109.667 | 7.833 | | |
| Total | 23 | 3984.958 | | | |

APPENDIX 5: Analysis of variance on the effect of plant extracts on disease incidence at 6 weeks after transplanting

| Source of variation | d.f. | s.s. | m.s. | v.r. | F pr. |
|---|---|---|---|---|---|
| REP stratum | 2 | 8.854 | 4.427 | 2.58 | |
| REP.*Units* stratum | | | | | |
| TREAT | 4 | 4474.766 | 1118.691 | 653.08 | <.001 |
| VARIETY | 1 | 2417.442 | 2417.442 | 1411.28 | <.001 |
| TREAT.VARIETY | 4 | 625.873 | 156.468 | 91.34 | <.001 |
| Residual | 18 | 30.833 | 1.713 | | |
| Total | 29 | 7557.768 | | | |

APPENDIX 6: Analysis of variance on the effect of plant extracts on disease incidence at 8 weeks after transplanting

| Source of variation | d.f. | s.s. | m.s. | v.r. | F pr. |
|---|---|---|---|---|---|
| REP stratum | 2 | 6.114 | 3.057 | 1.03 | |
| REP.*Units* stratum | | | | | |
| TREAT | 4 | 3670.582 | 917.646 | 308.97 | <.001 |
| VARIETY | 1 | 1395.106 | 1395.106 | 469.74 | <.001 |
| TREAT.VARIETY | 4 | 273.275 | 68.319 | 23.00 | <.001 |
| Residual | 18 | 53.460 | 2.970 | | |
| Total | 29 | 5398.538 | | | |

APPENDIX 7: Analysis of variance on the effect of plant extracts on disease incidence at 10 weeks after transplanting

| Source of variation | d.f. | s.s. | m.s. | v.r. | F pr. |
|---|---|---|---|---|---|
| REP stratum | 2 | 39.010 | 19.505 | 10.49 | |
| REP.*Units* stratum | | | | | |
| TREAT | 4 | 1723.334 | 430.834 | 231.69 | <.001 |
| VARIETY | 1 | 1095.751 | 1095.751 | 589.26 | <.001 |
| TREAT.VARIETY | 4 | 246.457 | 61.614 | 33.13 | <.001 |
| Residual | 18 | 33.472 | 1.860 | | |
| Total | 29 | 3138.024 | | | |

APPENDIX 8: Analysis of variance on the effect of plant extracts on disease incidence at 12 weeks after transplanting

| Source of variation | d.f. | s.s. | m.s. | v.r. | F pr. |
|---|---|---|---|---|---|
| REP stratum | 2 | 37.901 | 18.950 | 9.41 | |
| REP.*Units* stratum | | | | | |
| TREAT | 4 | 1623.436 | 405.859 | 201.51 | <.001 |
| VARIETY | 1 | 986.990 | 986.990 | 490.04 | <.001 |
| TREAT.VARIETY | 4 | 128.711 | 32.178 | 15.98 | <.001 |
| Residual | 18 | 36.254 | 2.014 | | |
| Total | 29 | 2813.291 | | | |

APPENDIX 9: Analysis of variance on the effect of plant extracts on disease Severity at 6 weeks after transplanting

| Source of variation | d.f. | s.s. | m.s. | v.r. | F pr. |
|---|---|---|---|---|---|
| REP stratum | 2 | 31.515 | 15.758 | 3.65 | |
| REP.*Units* stratum | | | | | |
| TREAT | 4 | 3284.168 | 821.042 | 190.30 | <.001 |
| VARIETY | 1 | 3358.757 | 3358.757 | 778.50 | <.001 |
| TREAT.VARIETY | 4 | 1091.230 | 272.807 | 63.23 | <.001 |
| Residual | 18 | 77.659 | 4.314 | | |
| Total | 29 | 7843.329 | | | |

APPENDIX 10: Analysis of variance on the effect of plant extracts on disease Severity at 8 weeks after transplanting

| Source of variation | d.f. | s.s. | m.s. | v.r. | F pr. |
|---|---|---|---|---|---|
| REP stratum | 2 | 23.962 | 11.981 | 3.49 | |
| REP.*Units* stratum | | | | | |
| TREAT | 4 | 1779.023 | 444.756 | 129.74 | <.001 |
| VARIETY | 1 | 1422.651 | 1422.651 | 414.99 | <.001 |
| TREAT.VARIETY | 4 | 394.085 | 98.521 | 28.74 | <.001 |
| Residual | 18 | 61.707 | 3.428 | | |
| Total | 29 | 3681.428 | | | |

APPENDIX 11: Analysis of variance on the effect of plant extracts on disease Severity at 10 weeks after transplanting

| Source of variation | d.f. | s.s. | m.s. | v.r. | F pr. |
|---|---|---|---|---|---|
| REP stratum | 2 | 36.682 | 18.341 | 10.68 | |
| REP.*Units* stratum | | | | | |
| TREAT | 4 | 1622.710 | 405.678 | 236.31 | <.001 |
| VARIETY | 1 | 1026.059 | 1026.059 | 597.68 | <.001 |
| TREAT.VARIETY | 4 | 205.539 | 51.385 | 29.93 | <.001 |
| Residual | 18 | 30.901 | 1.717 | | |
| Total | 29 | 2921.891 | | | |

APPENDIX 11: Analysis of variance on the effect of plant extracts on disease Severity at 12 weeks after transplanting

| Source of variation | d.f. | s.s. | m.s. | v.r. | F pr. |
|---|---|---|---|---|---|
| REP stratum | 2 | 29.245 | 14.623 | 5.77 | |
| REP.*Units* stratum | | | | | |
| TREAT | 4 | 1837.053 | 459.263 | 181.25 | <.001 |
| VARIETY | 1 | 780.762 | 780.762 | 308.12 | <.001 |
| TREAT.VARIETY | 4 | 188.544 | 47.136 | 18.60 | <.001 |
| Residual | 18 | 45.610 | 2.534 | | |
| Total | 29 | 2881.214 | | | |

APPENDIX 12:Analysis of variance on the effect of plant extracts on yield of sweet potato

| Source of variation | d.f. | s.s. | m.s. | v.r. | F pr. |
|---|---|---|---|---|---|
| REP stratum | 2 | 0.01867 | 0.00933 | 0.44 | |
| REP.*Units* stratum | | | | | |
| TREAT | 4 | 8.41133 | 2.10283 | 99.26 | <.001 |
| VARIETY | 1 | 0.86700 | 0.86700 | 40.92 | <.001 |
| TREAT.VARIETY | 4 | 0.25133 | 0.06283 | 2.97 | 0.048 |
| Residual | 18 | 0.38133 | 0.02119 | | |
| Total | 29 | 9.92967 | | | |

APPENDIX 13: Analysis of variance on the effect of plant extracts on vine length of sweet at 1 week after transplanting

| Source of variation | d.f. | s.s. | m.s. | v.r. | F pr. |
|---|---|---|---|---|---|
| REP stratum | 2 | 1.35 | 0.68 | 0.03 | |
| REP.*Units* stratum | | | | | |
| TREAT | 4 | 156.61 | 39.15 | 1.71 | 0.191 |
| VARIETY | 1 | 20.83 | 20.83 | 0.91 | 0.352 |
| TREAT.VARIETY | 4 | 52.30 | 13.08 | 0.57 | 0.686 |
| Residual | 18 | 411.53 | 22.86 | | |
| Total | 29 | 642.63 | | | |

APPENDIX 13:Analysis of variance on the effect of plant extracts on vine length of sweet at 2 week after transplanting

| Source of variation | d.f. | s.s. | m.s. | v.r. | F pr. |
|---|---|---|---|---|---|
| REP stratum | 2 | 7.800 | 3.900 | 1.21 | |
| REP.*Units* stratum | | | | | |
| TREAT | 4 | 1497.133 | 374.283 | 115.76 | <.001 |
| VARIETY | 1 | 80.033 | 80.033 | 24.75 | <.001 |
| TREAT.VARIETY | 4 | 107.133 | 26.783 | 8.28 | <.001 |
| Residual | 18 | 58.200 | 3.233 | | |
| Total | 29 | 1750.300 | | | |

APPENDIX 14:Analysis of variance on the effect of plant extracts on vine length of sweet at 3 week after transplanting

| Source of variation | d.f. | s.s. | m.s. | v.r. | F pr. |
|---|---|---|---|---|---|
| REP stratum | 2 | 7.800 | 3.900 | 1.21 | |
| REP.*Units* stratum | | | | | |
| TREAT | 4 | 1497.133 | 374.283 | 115.76 | <.001 |
| VARIETY | 1 | 80.033 | 80.033 | 24.75 | <.001 |
| TREAT.VARIETY | 4 | 107.133 | 26.783 | 8.28 | <.001 |
| Residual | 18 | 58.200 | 3.233 | | |
| Total | 29 | 1750.300 | | | |

APPENDIX 15:Analysis of variance on the effect of plant extracts on vine length of sweet at 4 week after transplanting

| Source of variation | d.f. | s.s. | m.s. | v.r. | F pr. |
|---|---|---|---|---|---|
| REP stratum | 2 | 74.024 | 37.012 | 4.12 | |
| REP.*Units* stratum | | | | | |
| TREAT | 4 | 13164.835 | 3291.209 | 366.71 | <.001 |
| VARIETY | 1 | 958.805 | 958.805 | 106.83 | <.001 |
| TREAT.VARIETY | 4 | 114.355 | 28.589 | 3.19 | 0.038 |
| Residual | 18 | 161.549 | 8.975 | | |
| Total | 29 | 14473.568 | | | |

APPENDIX 16:Analysis of variance on the effect of plant extracts on vine length of sweet at 5 week after transplanting

| Source of variation | d.f. | s.s. | m.s. | v.r. | F pr. |
|---|---|---|---|---|---|
| REP stratum | 2 | 80.67 | 40.34 | 2.19 | |
| REP.*Units* stratum | | | | | |
| TREAT | 4 | 18564.00 | 4641.00 | 251.41 | <.001 |
| VARIETY | 1 | 1865.99 | 1865.99 | 101.09 | <.001 |
| TREAT.VARIETY | 4 | 608.06 | 152.01 | 8.24 | <.001 |
| Residual | 18 | 332.27 | 18.46 | | |
| Total | 29 | 21450.99 | | | |

APPENDIX 17:Analysis of variance on the effect of plant extracts on number leaves of sweet at 1 week after transplanting

| Source of variation | d.f. | s.s. | m.s. | v.r. | F pr. |
|---|---|---|---|---|---|
| REP stratum | 2 | 597.07 | 298.53 | 5.07 | |
| REP.*Units* stratum | | | | | |
| TREAT | 4 | 1125.20 | 281.30 | 4.78 | 0.008 |
| VARIETY | 1 | 10.80 | 10.80 | 0.18 | 0.673 |
| TREAT.VARIETY | 4 | 95.20 | 23.80 | 0.40 | 0.803 |
| Residual | 18 | 1059.60 | 58.87 | | |
| Total | 29 | 2887.87 | | | |

APPENDIX 18:Analysis of variance on the effect of plant extracts on number leaves of sweet at 2 week after transplanting

| Source of variation | d.f. | s.s. | m.s. | v.r. | F pr. |
|---|---|---|---|---|---|
| REP stratum | 2 | 7.075 | 3.537 | 1.76 | |
| REP.*Units* stratum | | | | | |
| TREAT | 4 | 1129.790 | 282.447 | 140.24 | <.001 |
| VARIETY | 1 | 191.016 | 191.016 | 94.84 | <.001 |
| TREAT.VARIETY | 4 | 49.969 | 12.492 | 6.20 | 0.003 |
| Residual | 18 | 36.252 | 2.014 | | |
| Total | 29 | 1414.102 | | | |

APPENDIX 19:Analysis of variance on the effect of plant extracts on number leaves of sweet at 3 week after transplanting

| Source of variation | d.f. | s.s. | m.s. | v.r. | F pr. |
|---|---|---|---|---|---|
| REP stratum | 2 | 7.075 | 3.537 | 1.76 | |
| REP.*Units* stratum | | | | | |
| TREAT | 4 | 1129.790 | 282.447 | 140.24 | <.001 |
| VARIETY | 1 | 191.016 | 191.016 | 94.84 | <.001 |
| TREAT.VARIETY | 4 | 49.969 | 12.492 | 6.20 | 0.003 |
| Residual | 18 | 36.252 | 2.014 | | |
| Total | 29 | 1414.102 | | | |

s

APPENDIX 20:Analysis of variance on the effect of plant extracts on number leaves of sweet at 4 week after transplanting

| Source of variation | d.f. | s.s. | m.s. | v.r. | F pr. |
|---|---|---|---|---|---|
| REP stratum | 2 | 302.114 | 151.057 | 56.63 | |
| REP.*Units* stratum | | | | | |
| TREAT | 4 | 1142.915 | 285.729 | 107.12 | <.001 |
| VARIETY | 1 | 202.800 | 202.800 | 76.03 | <.001 |
| TREAT.VARIETY | 4 | 53.710 | 13.428 | 5.03 | 0.007 |
| Residual | 18 | 48.013 | 2.667 | | |
| Total | 29 | 1749.552 | | | |

APPENDIX 21: Analysis of variance on the effect of plant extracts on number leaves of sweet at 5 week after transplanting

| Source of variation | d.f. | s.s. | m.s. | v.r. | F pr. |
|---|---|---|---|---|---|
| REP stratum | 2 | 378.242 | 189.121 | 44.64 | |
| REP.*Units* stratum | | | | | |
| TREAT | 4 | 1252.715 | 313.179 | 73.93 | <.001 |
| VARIETY | 1 | 222.496 | 222.496 | 52.52 | <.001 |
| TREAT.VARIETY | 4 | 56.902 | 14.225 | 3.36 | 0.032 |
| Residual | 18 | 76.251 | 4.236 | | |
| Total | 29 | 1986.607 | | | |

APPENDIX 22

A B

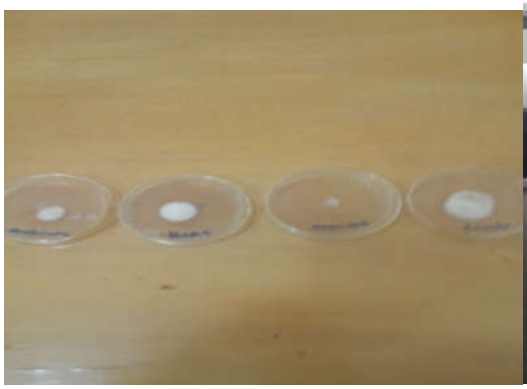
C D

Plate 1: Disease Symptoms of Sweet Potato from Field Survey, A. Fusarium Wilt B. Alternaria Leaf Spot C. Chlorotic Leaf Distortion and D. Minor Leaf Spot.

APPENDIX 23

A

B



C

D

E

F

Plate 2: A and B. Symptoms Of Fusarium Wilt after Inoculation C. Mycilial Growth of Fusarium Oxysporum on PDA Amended With Plant Extracts D. Prepared Plant Extracts E .Isolate of Fusarium Oxysporum F. Sweet Potato Growing in the Screen House.