Sudan University of Science and Technology

College of Studies and Scientific Research

Effect of Garlic (*Allium sativum*) aqueous extract against *Fusarium solani*

تاثير المستخلص المائي لنبات الثوم علي نمو الفطر فيوزيريوم سولاني

A thesis Submitted in Partial Fulfillment of the Requirements of the Degree for B.Sc. (Agric.) in Plant Protection

BY

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بسم الله الرحمن الرحيم

الايـــــــــــة

قال تعالى:

إِنّا قَد أوحِيَ إِلَينا أَنّ العَذابَ عَلى مَن كَذَّبَ وَتَوَلّى ﴿٤٨﴾ قالَ فَمَن رَبُّكُما يا موسى ﴿٤٩﴾ قالَ رَبُّنَا الّذي أَعطى كُلّ شَيءٍ خَلقَهُ ثُمّ هَدى ﴿٥٠﴾ قالَ فَما بالُ القُرونِ الأولى﴿٥١﴾ قالَ عِلمُها عِندَ رَبّي في كِتابٍ لا يَضِلُّ رَبّي وَلا يَنسَى﴿٥٢﴾ الّذي جَعَلَ لَكُمُ الأَرضَ مَهدًا وَسَلَكَ لَكُم فيها سُبُلًا وَأَنزَلَ مِنَ السّماءِ ماءً فَأَخرَجنا بِهِ أَزواجًا مِن نَباتٍ شَتّى﴿٥٣﴾ كُلوا وَارعَوا أَنعامَكُم إِنّ في ذلِكَ لَآياتٍ لِأُولِي النُّهى﴿٥٤﴾

صدق الله العظيم

سورة طه الآيات : (48-54)

DEDICATION

This work is dedicated to my

Father and my mother,

to my brothers and sisters

and all my family uncles and aunts

To my class mate

ACKNOWLEDGEMENTS

I would like to express my thanks to my supervisor Dr. Ekhlass Hussein Mohammed Ahmed College of Agricultural studies, Sudan University of science and Technology, who devoted most of his time to teach us on Various disciplines of scientific research.

Thanks are also extended to my dear teachers Ustaz Moda Ibrahim for their continuous unlimited helps during this study.

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ABSTRACT

This study was carried out in the laboratory of plant pathology, plant protection department, College of Agricultural Studies, Sudan University of Science and Technology in 2017.The objective of this study is to evaluate the effect of Garlic aqueous extracts and Tilt fungicides against *Fusarium solani* in potato in culture media Potato dextrose agar (PDA) *in vitro*. The fungi are an important causing significant reduction in yield. In the present study, the pathogenic fungi were isolated from infected plant parts. The causal agents were identified as; *Fusarium solani*. The identification was confirmed by visual culture characteristics, mainly the growth patterns and pigmentation.

Aqueous extract was prepared from the Garlic plant. Studies were conducted three concentrations of aqueous extract of Garlic (100%, 50%, and 25%) addition to the control treated with distilled water.

The results indicated that, the effect of the aqueous extracts of the Garlic in all concentrations was of significant effect in inhibiting the growth of the fungus compared to the control (). Effect of inhibition increases with increasing concentrations 2days (86.6, 80 and 66.6%) 4days (72.2, 61.1and 44.4%) 6 days after inculcation (65.2, 52.2 and 34.9%) respectively.

In conclusion, this study showed that the Garlic contain antifungal properties that could investigated in further studies.

ملخص الاطروحة

اجريت هذه الدراسة في مختبر علم أمراض النبات,قسم وقاية النبات,كلية الدراسات الزراعية,جامعة السودان للعلوم والتكنولوجيا 2017 لتقييم تاثير المستخلص المائي لنبات الثوم على فطر فيوزيريوم سولاني في بيئة بطاطس دكستروز أجار مقارنة بمبيد تلت (Tilt) تحت ظروف المعمل.

تم تحضير المستخلص المائي من الثوم.أستخدمت ثلاثة تركيزات من المستخلص المائي لنبات الثوم (25%,50%,100%)

النتائج التي تم الحصول عليها في اليوم الثاني والرابع والسادس توضح ان تاثير المستخلصات المائية للثوم في كل التركيزات خلال ثلاث أيام كانت زات تاثير معنوي في تثبيط نمو الفطر مقارنة بالكنترول تاثير التثبيط يزيد بزيادة التركيز في اليوم الثاني النمو كان (86.6,80,66.6 %) واليوم الثاني (44.4,61.1,72.2%) واليوم الرابع (34.9,52.2,65.2%)

نتيجة لذلك توضح هذه الدراسة بأن نبات الثوم تحتوي علي مواد ذات تاثير مضاد لنمو الفطر يمكن توضيح هذه المواد في الدراسات المستقبلية.

CHAPTER ONE

INTRODATION

Potato Plant (*Solanum tuberosum* L.) is member of the family solanaceae that includes egg plant, tobacco and tomato. the crop which was originally belived to be domesticated independently in multiple locations is an important crop worldwide and ranks fourth in production among food crops after maize (*Zea Mays* L.), rice (*Oryza sativa* L.), and wheat |(*Triticum aesitvum* L.) (FAOSTAT data, 2006).the importance of potatoes is increasing due to the rising world population. The potatoes can grow well in diverse conditions, with high nutritional value with an annule production of 3.6 x108 tons (Hamilton, 2005 and Anonymous, 2012).

In Sudan, although potato cultivation depends mainly on exotic advanced cultivars but an old introduced material is still produced in Jebel Marra in the far west and it is locally known as Zalingei potato. The crop is cultivated in small area around large cities along the Nile and on seasonally flooded plains (FAO, 1999). However, the area around Khartoum accounts over 70 percent of the country's potato production (Geneif, 1986).

The losses caused by diseases and insects constitute the major constraints that facing the production of potato worldwide and among these, the most important wide spread and important are pathogenic fungi, affecting tubers and vegetative parts. One of the main fungal pathogenic that attack potato is Fusarium dry rot (*Fusarium solani*) which is a worldwide economic problem. There are many species of fusarium reported to cause dry rot of

potato worldwide (Nielson, 1981) of which *Fusarium solani* has been

reported as the most pathogenic fusarium species causing potato dry rot (sharifi *et al*.,2009 Soheili-Moghadam and Hosseinzadeh,2013).

The disease affects tubers in storage and seed potato pieces after planting.

Hanson *et al.,* (1996) reported that fusarium dry rot of feed tubers can cause crop losses up to 25%, while more than 60% of tubers can be infected in storage (Hilo, 1974). Indiscriminate use of chemical pesticides to control various pests and pathogenic microorganism of crops plants is causing health hazard both in terrestrial and aquatic lives through their residual toxicity (Viana *et al,* 1996; Nikan and Morowati, 2013). Much attention is being focused on the alternative methods of pest control (Ali, 1996).

Natural plant extracts have been recommended as suitable alternative choices to synthetic chemicals (Suhr & Nielsen, 2003; Babu *et al*., 2008; Ownagh *et al*.,2010; Bahraminejad *et al*., 2010; Man gang and Chetry, 2012;Jafarpour *et al*.,2013) to control diseases and pests of crops . In Sudan the bioactivity of many cultivates and wild plants are demonstrated by many researchers (El-kamali 2001; Al-Doghairi *et al*., 2004; and sidahmed *et al*., 2009).

Based on the foredoing and considering the adverse and alarming effects of synthetic pesticide on environment and natural habitats, this study was under taken to find out a later native and nontoxic biological control agentes to control the dry rot fusarium in potatoes. Its aimed at investigating the

antifungal activity of some higher plant extracts and fungicide (Tilt) against fusarium dry rot of potato under laboratory conditions with following

objectives:-

1/ To explore the antifungal potentials of some higher plant crude extract against fusarium dry rot of potato.

2/ To evaluate the efficacy of systemic fungicide of fungal growth.

CHAPTER TWO

LITRUTER REVEW

2-1.Potato (*Solanum tuberosum* L.)

The potato plant which belongs to the family Solanaceae includes, among 2000 other species, tomato (*Lycopersicum esculentum* L.), sweet pepper (*Capsicum annuum* L.), egg plant (*Solanum*. *Melongena* ver.) , Esculentum L.), tobacco (*Nicotiana tabacum* L.), and petunia (*Petunia hybrid* L.) (Fernald, 1970).

2.1.1. Scientific classification

Kingdom: Plantae

Class: Magnoliopsid

Order: Solanales

Family: Solanaceae

Genus: Solanum

Species: tuberosum

(Hopes and Plaisted, 1987).

(Binomial name: Solanum tuberosum L.)

The genus solanum is a polymorphous and largely tropical and subtropical genus containing more than 1000 species .The origin agreed to be the high

elevation of South America and the area of first domestication was reasoned to be the area where wild diploids are still found and where the greatest diversity of cultivated forms can still be found, and is identified as the high plateau of Bolivia and Peru, in the general region of Lake Titicaca (Hopes and Plaisted, 1987).

Potato is one of the major vegetable crops grown worldwide following wheat, maize, and rice, with a production estimates of 368 million tons (FAOSTAT, 2015). It is the staple Food of many cultures and civilizations past and present. The term Potato is used to refer both to the plant, and the vegetable itself (Howard *et al.*, 1970).

In Sudan, the Potato is grown mainly as winter crop and the main area of production are along the Nile bank in both Khartoum and Northern Estates. Although potato cultivation in Sudan depends mainly on exotic advanced cultivars but an old introduced material is still produced in Jebel Marra in the far west and it is locally known as Zalingei potato (Abdelgadir, 2005).

Potatoes in Sudan are an important cash crop for small-scale growers, and have the potential to increase in comes in periurban areas, improve living standards and create employment opportunities. Potato production is steadily increasing in Khartoum; the acreage devoted to this crop has more than tripled in the last ten years (Ahmed, 1985).

The total acreage under potato cultivation in the Khartoum region amounts to about 6.500 hectares, with yields of 17 to 25 ton/ha .However, crops. Seed potatoes have to be imported and account for more than half of the total

production cost of potatoes (Elsir, 2005). This is a major constraint to further expansion of potato production (Elrasheed and Ballal, 2009). The estimated total potatoes production in Sudan is about 616,000 tons in a cultivated area of about 88,000 feddans (Hind and Mohamed, 2010).

One of the major constraints facing the quantity, quality and availability of

Healthy crop worldwide are the losses and contamination caused by post-harvest diseases .The major groups of postharvest disease are those which arise from infections initiated during and after harvest.

The threat to potatoes from fungal infections has now reached a level that outstrips that posed by bacterial and viral diseases (Berger, 1977). One of the main fungal pathogens that attack potatoes is *Fusarium spp* which are a worldwide economic problem (Nielson, 1981).

2-1-2 Economic importance;

The potato is a starchy tuberous crop from the perennial Solanum tuberosum of the solanaceae family (also known as the nightshades). The word potato may refer to the plant itself as the edible tuber. In the region of the Andes, there are some other closely related cultivated potato species. Potatoes are the world's fourth largest food crop, following rice, wheat, and maize. Longs –term storage of potatoes requires specialized care in cold warehouses and such warehouses are among the oldest and largest storage facilities for perishable goods in the world.

Once established in Europe, the potato soon became an important food

staple and field crop. The annual diet on of an average global citizen in the first decade of the twenty-first century included about 33Kg (or 73 IB) of potato. However, the local importance of potato is extremely variable and rapidly changing. It remains an essential crop in Europe, where per capita production is still the highest in the world, but the most rapid expansion over the past few decades has occurred in southern and eastern Asia.

China is now the world's largest potato-producing country, and nearly a third of the world's potatoes are harvested in China and India (Thompson and Morgan, 1855).

2-1-3 Geography and Production Zones

Ares of concentrated production include: Khartoum: the area around Khartoum, the capital of the Sudan ,accounts for over 70percent of the country's potato production (Geneif,1986).Located at the confluence of the Blue and White Niles ,Khartoum receives less than 300mm of rain annually, practically all of it from May to October.(El baz,1974).

Jebel Marra: the area around jebel marra, in the western part of the country, is reported to be the second most important potato production area of Sudan, although production figures are not available.

Situated between 1,500 and 3,000 meters above sea level (masl), jebel marra has acool, wet climate well suited to potato production. Soils are generally fertile, derived from volcanic materials in the uplands and from fluvial

deposits in the valleys. (Geneif, 1986 El baz, 1974).

Kassala: the Gash Delta area in kassala province is often mentioned as a zone of high potential for potato production, though figures on actual production in the area are lacking.

The general environmental conditions are roughly similar to those around Khartoum. (El baz, 1974).

2-1-4 Marketing:

The general absence of reliable storage facilities and transport has a substantial effect on distribution and consumption patterns.

Potatoes are available in the market on a seasonal basis, mainly February, March, and April. In the harvest season prices are high, leading many farmers to harvest the tubers before they reach maturity. During and immediately after, harvest prices plummet as farmers try to unload their crop before spoilage occurring, those farmers with access to modern storage facilities can subsequently take advantage of premium prices in the post.

Harvest season (Ahmed, 1981). Potatoes are apparently marketed with little or no grading (Accatino, nd).

2-2 Fusarium dry rot of potato

Is a devastating post-harvest disease affecting both seed potatoes and potatoes for human consumption. In fact, Fusarium dry rot of potatoes is a

worldwide economic problem. There are many species of Fusarium reported to cause dry rot of potato worldwide (Nielson, 1981). The disease may cause greater losses of potatoes than any other-post harvest disease. Crop losses attributed to dry grot have been estimated to an average of 6 to 25 % (Powel son *et al*, 1993).

Fusarium species which cause dry rot are also important to the consumer because some, Fusarium which cause dry rots also produce mycotoxins, one of such toxins is trichothecene which is an inhibitor of eukaryotic protein synthesis and can pose serious health problem to man and animal (Beremaid *et al*, 1991).

This fungus which prefers warmer climates causes a variety of colored rots in potatoes (Rowe *et al*, 2013). There are many species of Fusarium reported to cause dry rot of potato worldwide of which Fusarium solani has been reported as the most pathogenic Fusarium species causing potato dry rot (Sharifi *et al*, 2009).

2-2-1 Scientific Classification of Fusarium dry rot:

Classification:

Kingdom: Fungi

Phylum: Ascomycota

Class: Sordariomycetes

Subclass: Hypocreomycetidae

Order: Hypocreales

Family: Nectriaceae

Genus: Fusarium

Species: solani

Host range and distribute:

The predominant hosts for Fusarium solani are potato, pea, bean, an members of the cucurbit Family such melon, cucumber, and pumpkin. Some strains may cause infections in human.

Fusarium damping-off, corn rot, fruit rot, root rot and surface rot are caused by F. solani f.sp.eumartii (Aoki *et al.*, 2003).

Mart, Sacccand found in most states in the United States. The fungus has a worldwide distribution, but its frequency as an important plant pathogen is well known and hence remains the most common disease-causing fungus in its genus (Aoki *et al.*, 2003).

2-2-2 Phylogeny:

The phylogeny of isolates from potato and tomato was determined based on sequences of two DNA fragments: DNA internal transcribed spacer regions and partial sequences of elongation factor 1- anal isolates of Solani .f.sp.eumartii from tomato and potato formed a single monophyletic clade distinct from other formal specials and mating population of *F.solani*.*F.eumartii*, the results of this study demonstrates that Emeriti wilt and tomato foot rot in California both both are caused by *F.solani*.*f.sp.eumartii*,(Romberg and Davis, 2007).

2-2-3 Description:

Fusarium solani is a filamentous fungus in the genus Fusarium, and the anamorph of haematonectriaaematococca. *Fusarium solani* (mart.) Sacc. (2008). is a name that has been applied broadly to what is now known as the F. solani species complex (FSSC; O Donnell 2000). Members of the FSSC, which includes several additional named species and currently corresponds to approximately 50 phylogenetic species (Zhang *et al*., 2006; O,Donnelletal., 2008), are ubiquitous in soil, plant debris and in other plant and animal substrata and can be serious plant and human pathogens (Booth, 1971). The FCCS contains both heterothallic and homothallic strains and species, as well as strains that have no known sexual stage. The fungus produces three types of asexual spores, micro conidia and Chlamydia spores.

The macro conidiophores by division. They are important in secondary infection (Agrios, 2005).

The chlamydispores are globes and have thick walls. It is formed from hyphae or alternatively by the modification of micro cells. Conidia considered as endurance organs in primary infection. The telemorph or sexual reproductive stage of *F. oxysporum* is unknown. (Booth, (1977) stated that the chromosome number of the fungus is (12) and the perithecial state is Gibberella but not confirmed (Agrios, 2005).

2-2-4 Causal organism:

Several fusarium spp., including *F.sambucinum*, *F.solanivar F* .coeruleumand, *F. avvenaceum*, can cause dry rot. These fungi survive on refuse and live in soil. Infections can originate from infected seed tubers. Tuber rot usually does occur unless the tuber is injured during harvest. Wound provides a way for the fungus associated with soil to enter the tuber. Dry rot is one of the common storage diseases in Idaho. Fusarium dry rot lead to secondry infections by soft rot bacteria (Ocamb et al., 2006).

2-2-5 Symptoms:

The fungus can be soil borne, airborne or carried in plant residue and can be recovered from any part of the plant from the deepest root to the highest flower (Summeral *et al*., 2003).

Fusarium dry rot of potato is a devastating post-harvest losses disease affecting both seed potatoes and for human consumption (Loria and

Rosemary, 1993). Dry rot causes the skin of the tuber to wrinkle. The rotted areas of the potato may be brown, grey, or black and the rot creates depressions in the surface of the tuber. Seed pieces may rot completely before they have the chance to be planted. Sings of a pathogenic Fusarium species can be seen on an infected potato, and include white or pink mycelia(masses of vegetative fungal tissue) and very colorful spores that can be blue, black, purple, grey, white, yellow, or pink (Loria and Rosemary, 1993).

2-2-6 Disease Cycle:

Fusarium dry rot is caused by several fungal species in genus *Fusarium. Sambucinum* (teleomorphGiberellapulicaris) is the most common causing dry rot of stored tubers in North America, but other Fusarium species are also known to cause dry rot, particularly. *F.solani* var. coeruleum and F. avenaceum. (Philip and William, 2008).

2-2-7 Environment (Ecology):

Fusarium solani produces asexual spores (micro conidia and macro conidia). It is sexual state is Nectriahaematococca (Ascomycete), and overwinters as mycelium or spores in infected or dead tissues or seed. It can be spread by air, equipment, and water. (Vincent and Jean, 1971)

Warmer climates are preferred. (Warton *et al*., 2013). However, different species of Fusarium may be more prevalent in different areas. (Rowe *et al*., 2013). The fungus can persist in the soil for several years. The spores and

mycelium are carried into the soil tools. They my also be splashed by rain or carried by floods. The chhlamydospore is the survival structure in the absence of a host plant. (Vincent and Jean, 1971).

2-2-8 Importance:

Dry rot is not just a cosmetic problem like many other pathogens. It destroy tubers and leaves them completely inedible or unusable as seed in the future. Long-term storage losses have been reported to be as high as 60% while annual dry rot losses can range from 6 to 25% (Gachango and Hanson, (2012) . In Michigan, over 50% of seed lots have reported having variable levels of dry rot. (Gachango and Hanson, (2012). Fusarium spp ., are among the most important plant pathogens in the world and are highly variable because of their genetic makeup and changes in environment in which they grow causing morphological changes (Nelson, 1983).

2-2-9 Management:

There are many ways to manage dry rot. This includes application of fungicides, cultural practices, sanitation, biological control and botanical pesticides. However, most techniques for managing dry rot are aimed at preventing injury to the tubers, either seed or the harvested crop .preventing bruises will greatly aid in avoiding infection (Warton *et al* . 2007).

2-2-9-1 Cultural practices:

Cultural practices can also limit the spread of dry rot .plant high quality seed free from Fusarium dry rot pathogens into soils without a history of Fusarium dry rot. Varieties vary in their reaction to dry rot, and highly susceptible varieties should be avoided. Harvest tubers at least 14 days after vine kill to promote good skin set and reduce skinning injury that can increase storage dry rot. Avoid harvesting cold tubers that are more susceptible to injury .provide conditions that promote rapid wound healing early in storage, including high humidity, good aeration, and temperatures of 55 to 64, F for 14to 21 days. Since Fusarium dry rot increases with length in storage, short-term storage is advisable for fields where severe infection is expected (Howard *et al*., 2005).

2-2-9-2 Botanical controls:

The antifungal effect of certain medicinal and aromatic plants extracts have been investigated by many workers (Singh and Dwivedi, 1987, Henrique and Singh 1990). Thus, the development of new and different antimicrobial agents more safe has been a very important step (Agrafotis, 2002). However, the step of validation of traditional uses of antimicrobial compound in higher plants was studied by a number of researchers. Accordingly, the effect of different plants extracts on the germination and growth of many fungal pathogens have been reported (Agrafotis, 2002).

The use of plant extracts for controlling Fusarium wilt, cultural practices and the use of other methods are the most common strategies. However, they are

either not available or effective. The uses of natural products for the control of fungal diseases in plant are considered as an interesting alternative to synthetic fungicides due to their less negative impacts on the environment. Plant extracts or plant essential oils have been tested against *F.oxysporum* species for inhibitor effect and control efficacy under greenhouse condition (Bowers, and Locke, 2000). If natural plant products can reduce populations of soil borne pathogens and control disease development, that these plant extracts have potential as environmentally safe alternatives and as component in integrated pest management programs.

2-2-9-3 Biological control:

Biological control of dry rot is an intriguing concept, but currently nothing is available commercially. Researchers at Michigan State University are investigating the efficacy of *Bacillus subtitles* and *Bacillus pugilism* and *Trichoderma harzianum* in controlling Fusarium dry rot. (Warton and Phillip, 2013).

2-2-9-4 Chemical control:

Effective chemical control of dry rot can be achieved with chemicals like Tops MZ, Maxim MZ, and Moncoat MZ. These chemicals protect not only against dry rot, but also against other potato diseases like Rhizoctonia, silver scurf, and black rot. This chemical treatment can delay emergence of the

young plant, but this doesn’t mean these chemicals shouldn't be used. Many fungicides, including thiabendazole, work best when they are applied to tuber8s they are cut into seed pieces. (Schwartz, *et al*, 2005).

2-3 Garlic:

Kingdom: Plantae

Class: Monocots

Order: Asparagales

Family: Amaryllidaceae

Genus: Allium

Species: Sativum

Garlic is the oldest know

N medicinal plant variety or spice in existence. It belongs to the genus allium and is native to Central Asia. Mankind recognized the curative qualities of this magic herb over

3,000 years ago. Sir Louis Pasteur, the scientist who discovered pasteurization, effectively utilized the anti-bacterial qualities of garlic all the way back in 1858.

2-3-1 Garlic Nutrition Facts:

World War medical surgeons used the health benefits of garlic juice as an

antiseptic for treating war wounds. It contains useful minerals such as phosphorous, calcium, and iron, as well as trace minerals like iodine, sulfurs (Salunkhe, *et-al*, (1998).

CHAPTER THREE

MATRIAL AND METHOD

This study which conducted under laboratory conditions was carried out at Plant pathology laboratory of plant protection department college of Agricultural studies Sudan University of science and technology to evaluate the inhibitory effect of Garlic aqueous extracts against *Fusarium solani*.

The materials and equipment's used in this study are mentioned below

3-1 Equipment's

Needle laminar

Petri dishes

Autoclave

Conical flasks

Incubator

Measuring cylinder

Carbora

Centrifuge

Filter paper

3-2 Materials:

Potato dextrose agar

Tilt Fungicide

Infected plant

Garlic

Distilled water

3-3 Collection of plant samples:

Random samples in fected potato field were collected from central market for vegetables/bahry area brought to the laboratory for isolation.

3-4 Isolation:

Isolation of *Fusarium solani* the causal agent of potato wilt

3-4-1 Isolation from plant materials

Infected potato (tubers) showing symptoms of the disease were obtained From sick blots from Shambat Research station in August, 2017 the tuber Were cut into small sections(0 .4-1.0 cm),washed thoroughly with tap water, Surface sterilized with Clorox(NaOCL) for 5 minutes rinsed three time in changes of sterilized distilled water and dried on sterilized filter papers the Sterilized roots sections were plated at the rate of five sections / plate on to

potato dextrose agar (PDA) medium supplemented with chloramphenicol(0.05g / L) in 9-cm Petri dishes .The Petri dishes were incubated at 25^{Co}.After incubation for 7 days, isolated fungi were sub cultured on PDA. When free from contamination; Isolates were maintained on PDA slants and examined visually for their growth patterns and pigmentation on the adverse side of the agar. Further microscopic examinations were carried out for mycelia and conidia structure using pure culture of *F.solani* was obtained by using Hyphal tip technique. Pure culture of the isolated fungi was transferred to PDA slants and kept in refrigerator at 40^{o} C For further use. Sample of the obtained colonies were sub cultured by transferring small mycelia from the colony margins. Pure cultures were obtained by sub-culturing three times and slides were prepared and examined microscopically to confirm identity(x: 40).

3.4.3 Identification of the pathogens

The identification of the fungus was based on visual culture characteristics, mainly the growth passerns and pigmentation.

Furthermore, microscopic examinations were carried out for mycelia and Conidia structure based on the methods of Booth's key (1977).

3.4.4 Growth Rate of the pathogen

The pure cultures of *F.solani* were prepared using 7days old mycelia.

The fungus was cultured on PDA then transferred, aseptically, to the centre of Petri dishes containing PDA medium and incubated at 25^{oC}.

The linear growth of the fungus was assessed in cm after 48h.

3-5 collection preparing of plant materials

Garlic were collected from shambat area and brought to the laboratory where They were shade dried. After complete dryness plant samples were crushed separately to obtain fine powder for extraction.

3-6 Extraction process

The obtained fine powder from each plant was weighted (70 gm.) and placed in conical flask containing 70 ml distilled water and it was placed in a shaker for 4hrs. The extracts were filtered overnight to obtain 25% 50% and 100% concentrations.

3-7 Fungicide process

The chemical tested were Fulldazin fungicides 10ml dissolved in 100ml of sterilized distilled water to give 5, 10, 15 ppm respectively. For this solution 5, 10, 15 were completed to 100ml by adding sterilized potato dextrose agar medium to give final concentration.

3-8 Procedures.

Inhibition zone technique was used in this study to evaluate the inhibitory effect of both tested plant as well as fungicide Fulldazin previously prepared

Concentrations were add to PDA media at ratio of 10% and then 5mm in diameter of fungal mycelium disc from pure culture were placed in a center of treated PDA media in 9cm in diameter Petri-dishes.

Test procedcedure

Inhibition zone technique was used in this study (Rao and Srivastava,1994)Thefungs spore suspension was prepared from previously prepared pure culture by allowing the spores to grow on PDA media (Ramprasad,2005)treated was a desired concentration of

The PDA media was amended with the required concentration(5ml,2.50and 1.25) berore being solidifified in a conical flask of 250 ml, agitated before pouring it into sterilized perit dishes. Three plates were assigned for each concentration and left to solidify .the other three plates with PDA medium were served as control.

The Petri-dishes of each concentration were inoculated using

Sterilized filter paper disc dipped in a fresh culture suspension of corresponding fungus and placed at the centre of the plate. In case of the control the disc was treated with sterilized distilled water and placed at the centre of Petri-dishes.

Inoculated Petri-dishes were then incubated at 28^{co} for 3days. The growth of the fungus was calculated every day.

CHAPTER FOUR

RESULTS

4.1 Laboratory Experiments

This study was conducted at the laboratory of plant pathology,

Department of plant protection, college of Agricultural stuties, Sudan University of Science and Technology during August-September, 2017.

The aim of this study is to investigate the antifungal activities of (*Allium sativum*) Garlic aqueous extract on the liner growth of *Fusarium solani* in culture media under laboratory conditions where temperature around 25C^{o}

4.2Isolation and Identification of the Pathogens

The causal agent of the potato wilt disease was identified as Fusarium solani. Identification was performed depending on the culture characteristics and conidial shapes as described by Booth (1977).

4.3 Effect of Garlic aqueous extract on the liner growth of *Fusarium solani* two days after inculcation

The effect of Garlic aqueous extracts on the liner growth of the *Fusarium solani* compared to control. The three concentrations from the extract showed spectrum of fungicide activity. The results (Table 1) showed that extracts of Garlic aqueous tested had negative effects on fungal growth. In general the antifungal activity increased with increasing concentration.

Among all concentration 100% extracts tested Garlic caused 86.6% reduction, the concentration 50% Garlic aqueous extracts caused 80% reduction, followed in descending order by the concentration 25% Garlic aqueous extracts tested Garlic caused 66.6% reduction.

4.4 Effect of Garlic aqueous extract on the liner growth of *Fusarium solani* four days after inculcation

The effect of Garlic aqueous extracts on the liner growth of the *Fusarium solani* compared to control. The three concentrations from the extract showed spectrum of fungicide activity. The results (Table 2) showed that extracts of Garlic plant tested had negative effects on fungal growth. In general the antifungal activity increased with extract concentration. Among all concentration 100% extracts tested Garlic caused 72.2% reduction, the dose 50% extracts tested Garlic caused 61.1% reduction, followed in descending order by the dose 25% extracts tested Garlic caused 44.4% reduction.

Table1. Effect of Garlic aqueous extract on the liner growth of *Fusarium solani* Tow days after inculcation

Concentration		Mean	Redaction groth (%)
Control(Fusarium solani)		1.5a	0.0
Fungicide(Fulldazin)		0.0d	
Allium sativum	100%	0.2c	86.6
	50%	0.3bc	80
	25%	0.5b	66.6
CV%		36.4	
LSD		0.1	

Data average of three replicates

$R = \frac{A-B}{A} \times 100$

R=Percent reduction of *Fusarium solani*

A= *Fusarium solani* growth Control

B= *Fusarium solani* growth of Treatment

Table2. Effect of Garlic aqueous extract on the liner growth of *Fusarium solani* four days after inculcation

Concentration		Mean	Redaction groth (%)
Control(*Fusarium solani)*		1.8[a]	0.0
Fungicides(Fulldazin)		0.0[d]	
Allium sativum	100%	0.5[c]	72.2
	50%	0.7[bc]	61.1
	25%	1.0[b]	44.4
C.V		27.1	
LSD		0.1	

Data average of three replicates

$$R = \frac{A-B}{A} \times 100$$

R=Percent reduction of Fusarium solani

A= Fusarium solani growth Control

B= Fusarium solani growth of Treatment

4.5 Effect of Garlic aqueous extract on the liner growth of *Fusarium solani* six days after inculcation

The effect of Garlic aqueous extracts on the liner growth of the *Fusarium solani* compared to control. The three concentrations from the extract showed spectrum of fungicide activity. The results (Table 3) showed that extracts of Garlic plant tested had negative effects on fungal growth. In general the antifungal activity increased with extract concentration. Among all concentration 100% extracts tested Garlic caused 65.2% reduction, the dose 50% extracts tested Garlic caused 52.2% reduction, followed in descending order by the dose 25% extracts tested Garlic caused 34.9% reduction.

Table3. Effect of Garlic aqueous extract on the liner growth of *Fusarium solani* six days after inculcation

Concentration		Mean	Redaction groth (%)
Fusarium solani (Control)		2.3a	0.0
Fungicides (Fulldazin)		0.0d	
	100%	0.8c	65.2
Allium sativum	50%	1.1bc	52.2
	25%	1.5b	34.9
C.V		32.1	
S.E		0.2	

Data average of three replicates

$R = \frac{A-B}{A} \times 100$

R=Percent reduction of Fusarium solani

A= Fusarium solani growth Control

B= Fusarium solani growth of Treatment

PLATES

Plate (1): Control

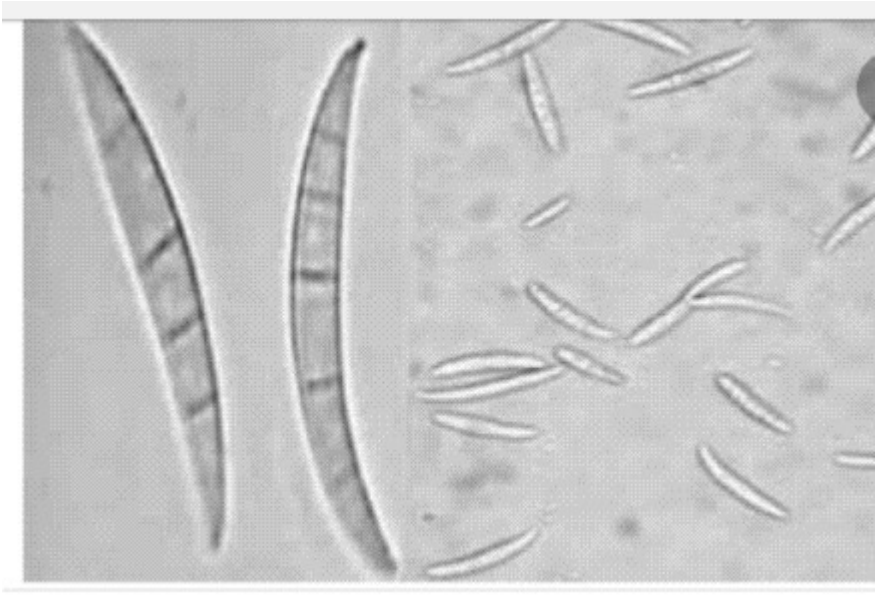
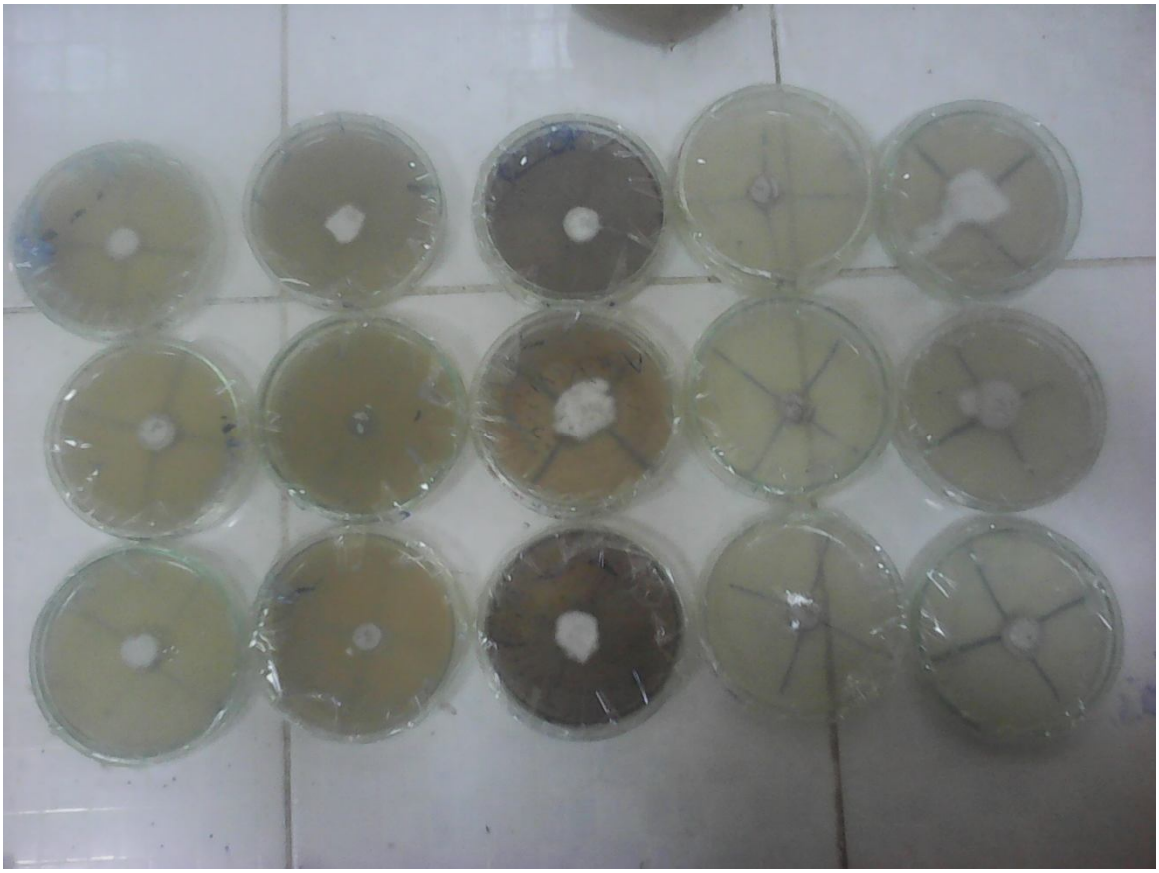


Plate (2): Chlamydospores

Plate (3): The growth of fungus



100% 50% 25% Fungicide Control

CHAPTER FIVE

DISCUSSION

The antifungal effects of crude medicinal plant extracts Garlic was determined by *in vitro* study using water as solvents. Three concentrations of Garlic plants extracts were used (25, 50, and 100%) as antifungal activity against *Fusarium solani.* The results of the experiment revealed that the Garlic extract was more effective this findind corroborates the notion plant plants are one of the most important sources of medicine. Plants-derived compounds (photochemical) have been attracting much interest as natural a ltematives to synthetic compounds the present investigations revealed that *in vitro* growth of *F. solani* was significantly checked by aqueous extracts of Garlic at all concentration results showed thad,Garlic had the highest antifungal activity against *F. solani* as it inhabited 55.6 % the radial fungus growth (100%). Our result is in agreement with the study of (Abdel Moneim Suleiman ------), (Wigdan M.Elzobair -------) and (Abdelrahim.------).

REFERECES

Accatino, p.Nd: Agricultural Research in sudan.MS, International potato Center. Agricultural Research Corporation (ARC), Republic of Sudan 1985. Improving Potato Production in the Sudan.

Ahmed,A.H.A.1985.Potato Production in Sudan (and the possibilities of its introduction in the Eastern Region).International potato Course: production, Storage, and Seed technology. Report of Participants. International Agricultural Center.

Hilo,A.andM.Hussein.1974.potato production in Sudan

IN the potato in Arab Countries. International

Potato Center.

FAOSTAT data, 2006 important crop worldwide and ranks Fourth in production potato among food crops after Maize (*Zea Mays* L.), rice (*Oryza sativa* L.), And wheat |(*Tritium aesitvum* L.)

Geneif, 1986.). However, the area around Khartoum accounts over 70 Fercent of the country's potato production

sharifi ,K,P;.(2009) Soheili-Moghadam,G and Hosseinzadeh,R 2013. reported as the Most Pathogenic Fusarium species causing potato dry rot

Ciegler,A,Burmeister,H.R.Vesonder, R.F. (1983).P Poisonous Fungi: Mycotoxins and Mycotoxicosees.

Pearlman,E;Leal,s;Tarabishy,A;Sun,Y;Szczotka-Flynn,L;Imamura,Y;(2011).Pathogenesis of fungal Keratitis.

Brett A.; Laurence, Matthew H.; Liew, Edward C.Y.; Leslie, John F (14September 2010).Biogeography and phylogeography Of Fusarium review.fungalDiversity.44

Zhang , O,Y;. (2006); Donnelletal. (2008). which includes several Additional Named Species and Currently Corresponds to Approximately 50 Phylogenetic Species

Summeral,A.T;. (2003). The Fungus can be Soil Borne, Airborne or Carried in plant Residue and can be recovered from any part of the plant from the Deepest root to the highest

Block, E. (2010).Garlic and other Alliums the Lore and The Science.

Meredith,Y,F;. Ted Jordan and Ducker, Avram.Garlic nalecta.Retrieved May (24, 2014).

Beremaid, P,(1991). Fusarium species which cause dry rot are also important to the consumer because some, Fusarium which cause dry rots also produce mycotoxins, one of such toxins is trichothecene which is an inhibitor of eukaryotic protein synthesis and can pose serious health problem to man and animal

Aoki ,L,G;.(2003). Fusarium damping-off, corn rot, fruit rot, root rot and surface rot are caused by F. solani f.sp.eumartii

Romberg ,A,W;. and Davis, F,U;.(2007) *F.solani*.*F.eumartii*, the results of this study demonstrates that Emeriti wilt and tomato foot rot in California both both are caused by *F.solani.f.sp.eumartii*,

Thompson ,k,H;. and Morgan, N,M;.(1855). production is still the highest in the world, but the most rapid expansion over the past few decades has occurred in southern and eastern Asia.

China is now the world's largest potato-producing country, and nearly a third of the world's potatoes are harvested in China and India