Sudan University of Science and Technology

College of Graduate Studies

Department of Plant Protection

Management of Early Blight Disease in Tomato Plants (*Lycopersicon esculentum*) in White Nile State

إدارة مرض اللفحة المبكرة في نبات الطماطم بولاية النيل الأبيض.

A thesis submitted in partial fulfillment of the requirements for the Ph.D. Degree in Plant Protection

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DEDICATION

To my Father and Mother, my Wives, my Sons, my Daughterly, my Brothers and my Sisters for all the love and support with love.

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This study would not have been possible without the efforts of the Good Lord – Almighty. Thanks and praise be to the Lord – Almighty, for seeing me through the successful completion of this project work, Amen.

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Abstract

The presents study was carried out to investigate prevalence, spatial spread, incidence level and management of the fungus *Alternaria spp*. that causes early blight disease (EBD) in tomato crop in the White Nile State. The investigation about the spread and incidence level of disease was conducted in four locations (North, Central, Eastern and Western of South), where the disease constitutes a serious obstacle to the production of vegetable crops in general and tomato crops in particular. Accordingly, comprehensive surveys were conducted within three successive seasons (2017/2018–2018/2019–2019/2020) to investigate the spread and the level of incidence of the disease. Determination of the level of incidence of the disease was based on calculating the number of plants showing typical and apparent symptoms of the disease in selected plants as mean percentage of the total number of plant inspected in each of the three sites selected in each of the four locations surveyed in the State. This was in addition to isolation and microscopic identification of the pathogen. To manage the disease, the study involved the investigation of the effect of different concentrations of natural products formulations and fungicide, Seed Star 42, on the incidence of the disease under natural infection conditions for two successive seasons. Three concentrations were tested from each of the natural products e.g. Neem oil, aqueous extracts of plants leaves of argel, Neem and usher plus mesquite plant fruits extract, as follow; 2.5, 5 and 10 ml/l and 25, 50 and 100% for each of the extracts respectively. This in addition to argel 5gm powder per hole and the fungicide seed star 42 at the rate of 5 gm/Kg seeds as seed dressing plus the control. The assessment of the treatments effects on the pathogen was recorded as percentage disease incidence. Likewise, six varieties of tomato were screened for their resistance to early blight disease under natural infection conditions namely, Castle Rock, Strain B, B286, Hiraihry, Goal, Domestic (Local one)). The results of the surveys showed the prevalence of

the disease in all the surveyed locations but at variable levels. The data showed invariable differences in the levels of incidence of the disease between the locations rather than the seasons. In fact, there was high rate of disease infection in the North of the State with an average of (15.11), followed (7.78), (6.0) and (0.89) in the South East, Center and South West of the State respectively. Moreover, within the South of the State, There was remarkable increase in percent disease incidence in the Eastern location of South (7.78) than in the Western location of it (0.89). The results of the investigation of the effect of natural products and fungicide on disease incidence reflected the positive effect in controlling early blight disease of tomato but their effects were variable. The result showed an invariably high effect on disease incidence obtained by the concentrations of Neem oil at 5ml/l, argel at 5gm/hole and seed star 42 at 5 gm/Kg seeds which gave almost 100% disease control where the percent was 0.0, 0.0, 1.0 at the forth count respectively compared to control which was 16.33. It is noteworthy that those encouraging results were confirmed by the same experiment repeated the season after. However, the effect of Neem leaves extracts at the three concentrations, 25, 50 and 100% was the lowest in disease control, at the forth counts; 8.67, 9.67 and 10.33% respectively, compared to the other treatments and this was an indication of inefficacy. Beneficial effects expressed as reduction in percent of fruits infected were also recorded due to application of these three treatments. Apparently, there was also obtained a high yield gains in this study upon treatments of tomato with those three products. The yield was almost doubled by application of the forgoing products giving a total of 41.6, 46.0 and 36.0 Kg per treatment respectively compared to control 23.1 Kg. The results of the screening of tomato varieties for resistance to early blight disease under natural infection attest the high level of resistance of the domestic variety to the disease (33.3%) followed by strain B (55.6%) and B286 (77.8%) compared to other varieties where the percent of disease incidence range from 88.9 to 100% infection. Nevertheless,

this comparatively low level of disease incidence in these two varieties was coupled with high productivity in comparison to other varieties. The current results were considered promising and encouraging to carry out a photochemical analysis of Argel plant using different solvents so as to determine the bioactive ingredient in the plant. Obviously, the present study is presenting for the first in Sudan the highly effective method for control of early blight disease in tomato using argel powder at planting time.

الـخلاصـــــــــــــة

أجريت هذه الدراسة للتحقق من الإنتشار المكاني، ومستوى حدوث المرض وإدارة الفطر نوع الالترناريا المسبب لمرض اللفحة المبكرة لمحصول الطماطم. بولاية النيل الابيض. البحث عن إنتشار ومستوى حدوث المرض تم إجراءه في أربعة أجزاء من الولاية (شمال، وسط، الجنوب الشرقي والجنوب الغربي للولاية) حيث يمثل المرض عائقا خطيرا لإنتاج محاصيل الخضر بشكل عام ومحصول الطماطم بشكل خاص. ومن ثم تم أجراء مسح شامل خلال ثلاثة مواسم متتالية (٢٠١٧/ ٢٠١٨– ٢٠١٨/ ٢٠١٩ – ٢٠١٩/ ٢٠٢٠). للتحقق من إنتشار ومستوى حدوث المرض.تحديد درجة حدوث المرض تم علي أساس حساب عدد النباتات التي أظهرت بوضوح أعراض المرض الحقيقية من بين النباتات التي تم إختيارها كنسبة مئوية من مجموع النباتات التي تم فحصها فى كلمن المواقع التي تم إختيارها في الولاية. هذا بالاضافة الي عزل وتعريف الكائن الممرض. من أجل إدارة المرض، تضمنت الدراسة التقصي عن تأثير تراكيز مختلفة لتركيبات من نباتات طبيعية ومضاد حيوي (Seed Star 42) علي حدوث المرض تحت حالة العدوى الطبيعية لموسمين متتاليين. تم إختبار ثلاثة تراكيز من كل من المنتجات الطبيعية وهى زيت النيم، والمستخلصات المائية لكل من أوراق نباتات النيم، العشر، الحرجل و ثمار نبات المسكيت؛ كالآتي ٢.٥ و ٥ و ١٠ مل للتر ماء و ٢٥، ٥٠، و١٠٠% لكل من المستخلصات. هذا بالإضافة الي بدرة مسحوق الحرجل والمبيد الفطري (seed star 42)بمعدل.٥ جرام للكيلو كمعفر للبذور إضافة للشاهد. لقد تم تقييم تأثير المعالجات علي الكائن الممرض بتسجيل نسبة الاصابة بالمرض. أيضا لقد تم إختبار مدي مقاومة مرض اللفحة المبكرة تحت حالة العدوى الطبيعية لدي ستة أصناف من الطماطم تحديدا، Castle Rock, Strain B, B286, Hiraihry, Domestic, Goal, (صنف محلي). نتائج المسوحات اوضحت إنتشار المرض في كل المواقع التي تم مسحها ولكن بمستويات مختلفة. كما اظهرت أيضأ فروقات ثابتة في مستويات المرض بين هذه المواقع عوضأ عن المواسم. حقيقة هنالك معدل عالي للإصابة بالمرض فى شمال الولاية وبنسبة ١٥.١١% يليها ٧.٧٨، ٦.٠٠ و ٠.٨٩% في الجنوب الشرقي، الوسط و الجنوب الغربي للولاية علي التوالي. إضافة إلn ذلك، فيما بين جنوب الولاية هنالك توجد زيادة ملحوظة في نسبة الإصابة بالمرض في الموقع الجنوبي الشرقي للولاية (٧.٧٨) مما في موقع الجنوب الغربي (٠.٨٩%). نتائج دراسة تأثير المنتجات الطبيعية والفطر علي مستوى حدوث المرض عكست التأثير الإيجابي في مكافحة مرض اللفحة المبكرة في الطماطم ولكن تأثيرها متفاوت. أوضحت النتائج بثبوتية التأثير العالي علي مستوى حدوث المرض الناجم عن تراكيز زيت النيم بمعدل ٥ مل للتر الواحد و الحرجل، ٥جرام للحفرة، والمبيد الفطري بمعدل ٥ جرام للكيلو والتي نجم عنها مكافحة مائة بالمائة حيث ان النسب كانت ٠.٠، ٠.٠و ١.٠عند التعداد الرابع والأخير علي التوالي مقارنة مع الشاهد الذي

كان ١٦.٣٣%. الجدير بالملاحظة هو ان هذه النتائج المشجعة قد تم تأكيدها في التجربة مثيلتها التي كررت العام التالي. مع ذلك، فإن تأثير مستخلص أوراق شجرة النيم للثلاثة تراكيز ٢٥، ٥٠، و ١٠٠% كانت الأقل تأثيرا في مكافحة المرض عند التعداد الرابع؛٨.٦٨، ٩.٦٧، و ١٠.٣٣ غلي التوالي مقارنة بالمعاملات الأخرى وهذا يدل علي اللافعالية. التأثير المفيد الذي ظهر في نقص نسبة الثمار المصابة أيضا تم تدوينة نتيجة لاستعمال هذه المعاملات الثلاثة.من الواضح، ان هنالك ايضاً مكسب إنتاجية عالي تم الحصول عليه في هذه الدراسة نتيجة معاملة محصول الطماطم بهذه المنتجات الثلاثة (زيت النيم بمعدل ٥ مل للتر الواحد و الحرجل، ٥جرام للحفرة، والمبيد الفطري بمعدل ٥ جرام للكيلو). كانت الإنتاجية مضاعفة عند استعمال هذه المعاملات أنفت الذكر حيث أعطت ٤٦.٠، ٤١.٦و ٣٦.٠ كيلو مقارنة مع الشاهد ٢٣.١ كيلو.النتائج التي ابرزها إختبار مدي مقاومة عدد من أصناف الطماطم لمرض اللفحة المبكرة تحت حالة العدوي الطبيعية اكدت مستوي المقومة العالي للصنف المحلي للمرض (٣٣.٣%) يليه الصنف (strain B, 55.6%) ثم الصنف (B2-86, 77.8%) مقارنة بالأصناف الأخري حيث ان نسبة الإصابة بالمرض تتراوح بين ٨٨.٩ الي ١٠٠%. ومع ذلك، فإن هذا المستوي القليل نسبياً من الإصابة بالمرض في بعض الأصناف مصحوب بإنتاج عالي مقارنة بالأصناف الأخري. تعتبر هذه النتائج واعدة ومشجعة لإجراء تحاليل كيميائية لنبات الحرجل باستعمال عدة مذيبات لتحديد المكونات البيولوجية النشطة في النبات. من الواضح، ان الدراسة الحالية قدمت للمرة الأولى في السودان طريقة ذات فعالية عالية لمكافحة مرض اللفحة المبكرة في محصول الطماطم باستعمال بدرة نبات الحرجل في وقت البذور.

CHAPTER ONE

INTRODUCTION

1.1 Tomato

Tomato (*Solanum lycopersicum*L.(*Synonoum Lycopersicon esculentum* (Mill) (Peralta *et al*., 2005), which is believed to have originated in the coastal strip of western America (Papadopoulos,1991), is belonging to the important fruit vegetables for human nutrition ; Thus cultivated across all continents (Anon, 2009).

In the Sudan the tomato is considered as one of the major vegetable crops due to its economics and nutritional value and being one of the main cash vegetable crops. Total production in Sudan reached 529,200 tons in 2012 produced from 37,044 hectares and increased to 617,400 tons in 2016 produced from 46,746 hectares ((FAO, 2006, 2018). The crop is grown mainly to be used either fresh or cooked mixed with other vegetables. Tomato *Lycopersicon esculentum*L is a member of the family Solanaceae (Shread, 1966). It was described by Hansen (2000) as an ancient vegetable crop which originated in Peru, South America. It was taken to Europe by the earlier invaders and from there to North and South America and the rest of the world. In the Sudan it is the second most important vegetable crop of Onion it is produced in a wide area around large cities along the Nile and on seasonally flooded plains.

1.2 Tomato Production in Sudan:

Winters are the major seasons for production where both productivity and quality are at their best. Summer production is faced with harsh hot-dry conditions especially in Central Sudan.The main production areas of Tomato in Sudan are Gezira and Managel Scheme, Khartoum, Blue Nile, White Nile ,

Kassala States and Western State. The crop is also produced in Jabel Marra and some parts of the main rain fed areas around villages in central clay plains and utilized as sun dried slices (FAO, 1999), Summer production of tomato is produced in limited areas in Blue and White Nile and Khartoum state, Northern State. It ensures high profitability because of the scarcity of the crop at that time. It is recently produced under controlled greenhouses during summer season and this practice is extending rapidly every year.

One of the major constraints facing the production of tomato is the losses caused by fungal diseases, insects, nematodes and parasitic weeds. Among these early blight of tomato caused by *Alternaria alternata*.is considered as the most important fungal disease of tomato plants (A 2005).The disease becomes wide spread and serious in Sudan, causing large economic loss to the growers in all tomato growing areas.

The epidemic disease occurs annually across all seasons wherever tomatoes are grown. In spite of its name, the disease may occur any time during the growing season; the disease is particularly destructive during summer production. The fungus attacks leaves, stems and fruits and is known to attack on potato, pepper and eggplant and Datura sp.

Furthermore, the nature of damage and survival ability of the fungus which can survive in soil and plant debris in the absence of susceptible host (Delahat and Sterenson, 2004, 2014) render the management of Early Blight of tomato more difficult. In fact, the problem of the disease control was even more complicated by controversy around the geographical distribution and seasonal occurrence of the two species of the genus Alternaria (*Alternaria alternata, A. solani and A. tenuis*) causing early blight in tomato (Giha, 1973: Pandey *et. al*., 2003 and Reni and Roeland 2006).

However, the disease has been managed primarily by the use of resistant varieties (Jalali and Chand, 1992) but breakdown in resistance of these

varieties due to evolution of virulent races of the pathogen have undermined their importance in recent years (Haware and Nene, 1982). In most cases chemical control methods are in practice. However, although the use of chemicals has helped increasing yields obtained (Ali, 1996), but the world wide trend to world environmentally-safe methods of plant diseases control have initiated the exploration of safe alternate products.

Apparently, insecticides were considered indispensable for sustainable agriculture production but, their increasing and irrational use has become a source of great concern because of their possible effect on human health and non-target components of the environment. This concern is heightened by the non-specificity and high toxicity of some pesticides and development of resistant strains of microorganisms against other ones.

The foregoing has initiated the exploration of safe alternate antimicrobial agents (Ali, 1996).Historically, more than 1000 species of plants have been reported to have chemicals in leaves, stems, flowers, seeds and roots which have insecticidal properties. Still only a few of them have been used for insect control on commercial scale.The best candidate of these plants is Argall. The bioactivity of this plant were mainly attributed to the presence of varieties of bioactive organic substances mainly terrenes peregrine glycosides alkaloids and sterols (El-kamali 2001; and Sidahmed *et al*,. 2009. Thus the chemical poisons of Argall plant (*Solenostemma argall* Del Hyne) are mostly alkaloids which are nitrogenous in nature and they are heterocyclic compounds having strong effects on the nervous system of organisms and causing death .

In an attempt to achieve these objectives, some alternative methods of control have been adopted. This included, bio fungicide or natural products which emerged as promising alternatives, e.g. Biological agents, Neem, Garlic, and few other plants proved to inhibit Early Blight in tomato and other plants

diseases (Schmutterer, 2002; Prasad and Naik, 2003; Adandonon *et al*., 2006 and Anjorin *et. al*., 2010)

Obviously, no single approach for Early blight disease control was proved to be effective and without drawback. Therefore, integrated management strategies are the only solution to maintain plant health. These strategies should include minimum use of chemicals for checking the pathogen population, optimization of cultural practices to reduce pathogen inoculum, modification of cultural practices and safe alternate antimicrobial compounds of higher plants.

Based on the foregoing, the rational of this study will focus on occurrence, distribution, identity of the pathogen, quantification of losses caused by early blight and assessment of different components for management of the disease in order to develop an integrated disease control strategy.

1.3 Objectives

1.3.1 Main objective:

The main objective of this study is to improve tomato productivity by developing a package of integrated control measures that offer several options for farmers to manage early blight disease of tomato in White Nile State production areas.

1.3.2 Specific objectives are to:

1. Conduct a field survey to determine relative occurrence of *Alternaria alternata*. and quantify the damage and level of disease incidence caused by early blight'
2. Isolate and identify the genus Alternaria.
3. Explore the antifungal potentials of different formulations of some higher plants and fungicide against infection of tomato plant under field conditions.
4. Screen and evaluate some tomato varieties and hybrids for resistance to early blight disease.
5. Identify reliable sources of resistance to early blight disease.

CHAPTER TWO

LITRATURE REVIEW

2.1. History of Early Blight Disease:

Early blight is the major fungal disease of tomato caused by the fungus *Alternaria alternata*. (Ellis and Martin, 1971). This disease, which in severe cases can lead to complete defoliation, is most damaging on tomato (Peralta *et al*., 2005)in regions with heavy rainfall, high humidity, and fairly high temperatures (24°–29°C). Epidemics can also occur in semi-arid climates where frequent and prolonged nightly dews occur (Rotem, 1994).

The disease was first described in New Jersey (U.S.A) in 1882 and later by. The most critical early work was that of Jones during the period 1891 – 1903 Until 1945 where extensive research on the disease was done Walker (1952). After that, many aspects of the disease were studied and showed that early blight is a very common disease of both Tomato and Potato. Fruits, roots and stems lesion on tomato were observed by Walker (1952). Paul grow, (2000) reported that Early blight pathogen was first described by Ellis and Martin in 1994 from dying Potato leaves and was called at that time *Macrosporium solani.* In spite of its name the disease may occur at any time during the growing season (Hansen, 2000). In contrast to name of early the blight could appear on maturity stage (Paul grow, 2000) .

2.2. *Alternaria* Diseases:

The diseases caused by genus *Alternaria* are among common diseases of many kinds of plants throughout the world. They affect primarily the leaves, stems, flowers and fruits of annual plants especially vegetables, ornamentals and trees such as citruses and Apples. *Alternaria* diseases appear usually as leaf spots and blights, but they may also cause damping off for seedlings and

also rots of stem, tuber and fruits are taking place (Agrios, 1997). Some of diseases caused by Alternaria include early blight of potato and tomato blight of carrot, leaf spot and fruit spot in many plant species throughout the world (Agrios, 1997).Some species of Alternaria produce toxins which are not host specific whereas others are host specific (Agrios, 1997).

2 .2.1 Pathogen:

Neergaard (1945) reported that *Alternaria spp*. has large spores producing group of fungus and characterized by separate conidia borne singly on simple conidiophores..

Joly (1959) studied the morphological variations of Alternaria species and later during 1964 divided them in three sections and proposed a simple Key for identification and determination of the most common species. Furthermore, noticed that the conidia of *Alternaria solani* are uniform, beaked, dark uniform, pale golden or olivaceous brown and smooth and usually 150-300 um in length and 15-19 um thick in the broadest part, with 9-11transverse septa and 1-4 longitudinal or oblique septa, sometimes branched 2.5-5 um thick tapering gradually

Bose and Som (1986) observed septets and the branched, light brown hyphae which turned darker with age. The conidiophores were short measuring 50-90 um long and dark color. The conidia were 120-296*12-20 um in size, beaked, uniform, dark color and borne singly. However, in culture they formed short chains. Singh (1987) reported that the conidia contained 5-10 transverse septa and 1-5 longitudinal septa. The mycelium was septets, branched, light brown hyphae which turned darker with age.

2.2.2. Geographical Distribution:

Many *Alternaria spp.* are recorded mostly in all countries around the world, whereas the presence of others is restricted to specific areas. *Alternaria spp.*

on potato and tomato exemplifies the world wide distribution of species, which spread from Iceland to Equatorial areas in South America and Africa and further south to cool parts of Chile and Argentina. Other pathogens of worldwide distribution include *Alternaria brassicheekas* (Anon., 1983) and *Alternaria brassicicola* (Millar and Pollard, 1976).

2.2.3 Symptoms

Stems**,** leaves and fruit of tomato are all are subject to infection by *Alternaria*. It may girdle seedling and causing damping off in the seedbed. On the leaves, brown circular spots are often surrounded by yellow area (Dillard, 1995). Leaf spots have characteristic dark concentric rings. Leaf spots usually appear on the oldest leaves first and progress to the upper parts of the plant (Castano Zacata,1994).

The first symptoms usually appear on older leaves start with small irregular dark brown to black spot. As the spot enlarge, concentric rings may form as a result of irregular growth patterns of the pathogen. This gives the lesion a characteristic shape such as “Target spots” or “Bull’s eye” appearance. There is often a narrow yellow halo around each spot (Pscheit, 1985).

Leaf symptoms are circular to oval spots appearing first on lower leaves. They may cause a collar rot of young tomato seedling, sunken spots or cankers on older stems, leads to blossom drop of young fruit (Westcott, 1971). Walker (1952) reported that in plant grown from infected seed, stem lesions are frequently occur and are elongated, sunken dark and zonated up to 2cm in length.

Tomato plants were found susceptible to *Alternaria spp.* during all growth stages (Vloutoglou and Calogerakis, 2000). If infected seeds are used to start tomato, transplanted seedling might damp- off soon after emergence. Large lesions also develop at the ground level on stem of transplant or seedling. The plant may become girdled, a condition known as “Color rot”. Such plants may

die when set in the field or the stems are weakened and may break early in the season. On older fruits early blight also causes dark leathery, sunken spots usually at the point of stem attachment. These spots may enlarge to cover the whole fruit, often showing concentric marking like those on leaves. Fruits can also be infected while they are green or during ripening stage through growth cracks and other wounds, often drop before reaching maturity (Dillard , 1995).

2.2.4. Disease cycle:

The fungus can survive in soil and in infected crop residue and weed residues. Thus, it may be soil or seed borne or can be carried by wind,water, insects, workers and farm equipments. The spores that land on tomato plants will germinate and infect the leaves when they are wet. Spores can enter the leaves, stem or fruit directly throw cuticle or through natural opening. The fungus is most active during mild to warm temperatures and wet weather (Castano Zacata, 1994). The disease is worse during the rainy season.

Early blight is very severe on plants stressed by a heavy fruit load, nematode attack or low nitrogen content (Dillard and Wilkinson, 1995).

2.2.5. Damage:

Alternaria cause damage to susceptible plant and infection result in loss of yield due to early leaf death.The infection causes direct losses when attacking the fruits and indirect losses by reducing plant vigor (Dore Zhkhin and Laanyuk , 1979)

Poulgrow (2004) reported that the disease can be very destructive if left uncontrolled, as this will lead to complete defoliation of plant. (Foolad *et,al*., (2002) found that the disease causes plant defoliation which reduces yield and fruit quality, and contributes to significant crop losses. The epidemic of the disease is common under cooler and warmer areas. Doolittle (1948) and

Nancy Pataky (1999) reported that early blight disease appears on tomato as they start to set fruit. The high level of humidity and presence of dew are favorable for early bight and cool temperature may favor disease development whereas dry weather is not favorable for development of early blight.

2.2.6 Losses due to Early blight disease:

Yield losses up to 79% due to early blight were reported from Canada, India, USA, and Nigeria (Dator and Mayee 1981). Collar rot caused by *Alternaria alternata* can cause seedling losses in the field from 20 to 40 %. (Meitei *et,a*l., (2012)reported the loss in yield due to the early blight disease was 2.15% in highly resistant genotype and 42.75% in highly susceptible ones.

Saha and Dos (2012), conducted experiment to assess the crop loss in relation to disease severity due to early blight in the year 2007 – 2008 and 2008 – 2009 in West Bengal and revealed that loss in yield was 0.76 ton/ha for every 1% increase in disease severity.

2.2.7 Host plant:

The most important host plants of *Alternaria alternata* are tomato, potato, eggplant and pepper. Other hosts include non Solanaceous hosts such as Cabbage, Cucumber, (Rand, 1917, Neergard 1945 and Westcott1971). Temperature may favor disease development contrary to dry weather which is not favorable for development the disease.

2.2.8. Etiology:

Alternaria alternate .spores are overwintering and over summering as chlamydospores on crop debris and in the soil carried on tubers and seeds. In warmer climates it can also survive on volunteer plants as well as weeds. Spores germinate when temperature ranging between 34 – 50ºC (42 - 48ºC is optimum) when prevailing during wet weather whereas winds and rains dislodge spore (Soltanpour and Harrison 1974). Conidia serve as primary

inoculum and infect plants directly through the cuticle. Then, these conidia serve as secondary inoculums and are disseminated by wind, running water, insects, and field workers. The pathogen spores also can be transmitted by seeds from area to another (Anon, 1983).

(Anon, 1983). Reported that the mycelium of the fungus remains dormant in dry infected leaves for a year or more and conidia found to retain viability for 17 months at room temperature. Tiny wounds caused by blowing sand favor disease development, especially if followed by dew fog, or rain (Rotem ,1994). High nitrogen low phosphorus fertilizers decrease resistance to *Alternaira alternata* (Barchay *et, al*., 1973).

2.2.9. The occurrence of Early Blight:

Giha (1987) reported that two species of Alternaria were found in Sudan *Alternaria Solani* and *Alternaria tenuis*. They are attacking a variety of vegetable crops including tomato, potato, eggplant and Onion. *Alternaria solani* is the main species responsible of these attacks particularly in the wet parts of the Sudan. The diseases caused by this fungus of common occurrence wherever potatoes and tomato are grown in the world (Rich, 1983 and Singh, 1983).

2.2.10. Effect of pH on growth of *Alternariasolani*

Samuel and Govindas wany (1972) demonstrated that good mycelia growth and sporulation of *Alternaria solani* was between pH 4.0 to 8.0 and pH 5.0 was the best for mycelia growth and pH 7.0 for sporulation. Whereas, Gemawat and Gohosh (1980) observed that the *Alternaria alternata* was capable to grow on wide range of pH (4.0 to 9.5), and maximum growth and sporulation were observed at 6.3 pH.

Alhussaen (2012) observed that the optimum pH level for the growth of *Alternaria alternate* grown in vitro was 6 to 7. Maximum growth of *Alternari*

alternate was recorded at 6.5 pH level on PDA medium under continuous light condition by Chohan *et.al*.,(2015).

2.2.11. Effect of temperature on growth *Alternaria alternata*

Kemmitt (2002) reported that warm, humid (24-29 C^0) environmental conditions are conducive to infection in tomato in the presence of free moisture at optimum temperature range of 28 to 30 C^0 where conidia germinate in approximately 40 min.

Arunakumara (2006) postulated that the *Alternaria alternata* produced maximum growth at 25 to 30 C^0temperature followed by 25C^0, 20C^0, 35C^0, 15C^0, 40C^0, Rodrigues*et.al*., (2010) studied conidial production and reveal that the fungal colonies maximum growth in V8 medium at 25c in the dark with agitation for seven days at 25+ 2C^0 under near ultraviolet light and 12 h photoperiod .

2.2.12. Control Measures

Early blight can be controlled by using different methods including cultural, breeding of resistant varieties and chemical methods or through using several measures together e.g. cultural control with the least amount of fungicides. Although genetic resistance to *Alternaria solani* has been reported the disease is mainly controlled by chemical sprays.

2.2.13.1. Cultural Control:

Alternaria disease on potato and tomato can be controlled thought crop rotation or by burning of infected plant debris, as this will help in reducing the amount of primary inoculum. Agrios (1997) reported, that adequate nitrogen fertilizer and resistant varieties generally reduce the rate of infection by Alternaria. Crop rotation, removal and burning of infected plants debris and eradication of weed hosts help to reduce the inoculums for subsequent plantings of the crop.

Management of early blight by the use of crop rotation which include Potato or tomato once every three or four years to allow infected plant debris to decompose in the soil, was proposed by Rowe, *et.al*., (1996). One of the most important components of early blight disease management is selection of cultivars that have lower susceptibility to disease (Rowe *et. al*., 1996)

2.2.13.2. Biological Control

Sowing coated tomato seeds with spores of the bacteria *Streptomyces spp* as antagonist before sowing proved high efficiency in controlling early blight disease (Elobyad *et. al*., 1993). Biological strategies (biofungicide) can reduce early blight disease below the level achieved with commercial fungicides (Steventet.al. 1998).

However, seed treatment with fungal Trichoderma *virids* was found to be the most effective control measure for early blight disease in comparison with other treatments (Sawant, *et.al*., 1999).

2.2.13.3. Chemical Control

George (1978) reported that the application of fungicides (foliar spraying) should begin soon after transplanting or after seedlings have emerged which provided good control. However, fungicides should be applied when first early blight lesion were observed and continued at ten days intervals till harvesting (Hawisonet.al. 1956; Abusin 1994) found that chlorthalonil was effective in the controlling early blight. Also the disease can be controlled though the use of chemical spraying with fungicides such as Chloroth, Maneb, Captafol and Mancozeb (Agrios, 1997) and reported also Bordeaux mixture and insoluble copper as are only moderately effective but Zineb and Ziarm are more effective.

Difenoconazole and mancozeb at the rate of 125g/ha at the interval of 14 days proved effective in controlling early blight disease (Follas *et.al*., 1992).

Moreover, Paul grow (2000) showed that applications of fungicides in tomato usually started at 2-3 weeks following emergence or soon after transplanting if a calendar schedule is followed . chlorthalonil applied at the rate of 3 litter/ h with irrigation as conventional spraying method proved efficient in combating early blight disease (Brandao, *et.al*., 1996).

2 .3. The role of Natural Products:

2.3.1. Neem (*Azadirachta indica* A. Juss):

2.3.1.1Origin and Characteristics of Neem tree:

Neem tree was introduced to Sudan from India. The tree showed promising results in reducing as well as controlling some insects and disease. The most effective part of Neem is seeds and leaves from which powder is extracted and applied in various ways to infected plants (Ruskin, 1991).

2.3.1.2. Chemistry of Neem Tree:

All parts of *A. Indica* tree have been examined by many chemists who showed that Neem trees contain a number of chemicals and showed that Neem compound called "triter penes" or " Limonoids". There are nearly about 100 proto limonoids or tertranor titer, pentanor titer penoids hexanor titer and some none titer penoids (Jones *et. al*., 1989).

Limonoids occurring in Neem are related to nine different basic structure groups such as the azadiron, ammorastanin, vepinin and vilasinin, and seco system related to gedunin, nimbin, nimbdinin and salanin and azadirachtin group that in fact belong basically to the Azadirachtin which is naturally found in Neem seed kernel depending on the method of extraction (Schumtterer 1995 and Anon, 1996).

2.3.1.3. Bio-activities of Neem products

Neem has been used as an effective post-harvest protect ant against different insects for many crops. Neem is especially effective against the cowpea Weevil (David *et.al*. (2003). Neem oil and its isolates inhibit fungal growth on humans and animals. (Schumtterer,*et. al*., 1984 and Anon, 1997).

2.3.2. Neem oil:

Neem oil is a vegetable oil pressed from the fruits and seeds of the Neem. (*Azadirachta indica*) is an ever green tree which is endemic to the Indian sub continent and has been introduced to many other areas in the tropics. It is the most important of the commercially available products of Neem for organic farming and medicines.

2.3.2.1 Toxicity of Neem oil:

The ingestion of Neem oil, even in small doses, is severely toxic and can induce metabolic acidosis seizures. This can also be associate d withallergic contact dermatitis.Formulations made of Neem oil also find wide usage as a biopesticides for organic farming , as it repels a wide variety of pests including the Mealy bug , beet army, worm, aphids,thrips, white flies, Locust and the Japanese beetle. Neem Oil also controls of black spots, powdery mildew, anthracnose, rust and Alternaria.

2.3.3 Mesquite (*Prosopis juliflora*):

Prosopis juliflora (SW) DC is an evergreen tree native to South America, Central America, and the Caribbean. In the United State of America, it is well known as Mesquite (Anderson, 2005). It is fast growing, Nitrogen- fixing, and tolerant to arid conditions and saline soils. In some circumstances *Prosopis juliflora* can provide a variety of valuable goods and services: fuel wood. Charcoal, animal feed, construction materials, soil conservation and rehabilitation of degraded and saline soils (Pasiecznik, 1999 and Pasiecznik

et,al., 2001). In the dry land of India, *Prosopisjuliflora* is considered as one of the most valuable tree species (Pasiecznik, *et.al*. .2001).

Prosopis spp is ever green leguminous trees or sharps.The genus comprises 44 species of which 40 are native to the Americas, of the remaining species 1929kdeziana, PAfricana is indigenous to Africa ,where is P kodzina , P farcta and P. cineraria are native to middle east and Pakistan [Borun and Messey ,1929 and Bukarat, 1976].Prosopis species grow in arrays of environment and are not restricted by soil type, pH, salinity or fertility [Sid Ahmed, 2005 and Babiker 2006]

The name Prosopis was selected by Linnaeus to describe the only species he was aware of *P.spicigera* in 1776. Felker, et. Al., (2003). Stated that genus *Prosopis Linnaeus* Burkat is in the family Leguminous [Fabaceae], Sub family Mimosoidae. The placing of Prosopis in the wider taxonomy classification system given below based on.

2.3.3.1Allelopathy

The leaves of *P.juliflora* contain variouse chemicals inclouding tanni , flavonoid , steroids,hydrocarbons,waxes, and alkaloids.These are known to affect palatability to livestock but also have effect on the germenation and growth of Prospis, weeds and other trees.

Leaf extract were also noted to kill some insect, bacteriaand fungi . However there is some depate as to the importance of allelopathy in tree ,crop interaction and the applicability of results from pot trials to field conditions . Alkaloidsand flavonoids are known to degrade rabidly folowing Leaf senescence but other chemicals may accumulate under tree crowns .

Most studies have utilised leaf extract or dry leaves incorporated into soil for analysis in pot trials.This often exaggerates the concenteration of chemicals leading to misleading results. However, reduction in crop seed germination

due to chemicals inhibition was noted with *P.juliflora* leaves concenteration of more than 3 % but it was thought that this would not be noticable under field conditions.These effects of those allelochemicals my be indirect,upon the seed and seedling ,or my be in directly italics effects on other soil organisms.

Extracts from plant parts of *P. juliflora* decreased germination and growth of almost all plant tested in several studies, indicating that allelopathic effects. Therefore effects are important in the ecology of the *P. Juliflora al* .However Sen and Chawan ([1970), assessed the effects of *P.juliflora* extracts on germination of Euphorbia spp. and concluded that the phytotoxicity was without ecological significant ,thought that the accumulation of steroids hydro carbons and waxes in P. *ruscifolia* leaves letter affected hydrophlic constituents and soil moisture capacity.

Whereas all other authers discussed only allelochemicals effects Auotutoxicity of *P. juliflora* which has been observed on seed germination and growh of crop plant. found decreased shoot and particularly,roots growth of orange of plant following treatment with *P. juliflora* leaf extracts. Fresh leaf extracts of *P. juliflora* were found to have greater negative effects on germination than extracts from stems dry litter or fruits (Sen and chwan, 1970).

However, Noor *et. al.* , (1995), observed agreater affect from fruit and seed extracts than from root or leaf or flower extracts. Bark extracts have also proved effective in inhibiting germination .

2.3.3.2 Benefit uses of mesquite

The tree has some benefits that include combating desertification,nitrogen fixation as legumenous plants increasing the global green coverage and using its timber for furniture,fencing and fuel, also as animal feed.However

,resently it was realized that the problems caused by the plants are more than benifits derived from them (Sidahmed,2005).

2.3.4 Usher (*Caltropis procera*)

Sadana and Didwania (2015) studied the bio-efficacy of *Calotropis procera* and *Eucalyptus oblique* extracts against *Alternaria solani* under in vitro conditions. They found that fresh aqueous extract of at 15% was most effective which gave 88% percent inhibition of mycelia growth of *Alternaria solani* strain A1 followed by *Calotropis procera*.

2.4. Fungicide (Seed Star 42 WS)

Apron Star 42 WS is a new fungicide-insecticide that combines two active ingredients, namely thiamethoxam, metalaxylam (mefenoxam) azoxietrobin and difenoconazole (www.syngenta.com, 2006). The trade names include Cruiser and Actara (Horii *et.al*. 2007).

CHAPTER THREE

MATERIALS AND METHODS

This study which conducted at White Nile State, that situated South of Khartoum State, between Latitudes (13 30 12 N) and Longitudes (33 30 31 E) aiming to improve tomato productivity at this State by developing a package of integrated control measures that offer several options for farmers to manage early blight disease of tomato.

3.1 Survey for incidence and prevance of *Alterlenaria spp.* in tomato in White Nile State

In this surveillance, three field surveys were carried out during the winter seasons (December - January) for three successive years (2017 / 2018 ,2018/2019 and 2019 / 2020) in order to assess the incidence and extent of prevalence of early blight disease in commercial tomato fields under natural infection pressure. The surveys were conducted when the crop in the different locations was at its vegetative to early flowering stage. The surveillance structure consisted of two levels; sites within localities. Throughout the State, four locations were selected (North, Center, and West , East on South of the State) to carry out the surveys. Each location was divided into three sites and 5 commercial fields, each of eight feddans, were chosen randomly from each site. A total of twenty five plants were selected randomly and inspected from each site to give a total of seventy five plants from each location.

The plants that showed typical early blight symptom were calculated as percentage incidence of the disease from the total number of plants inspected. Infected plants were further used to isolate and identify the causal agent.

3.1.1. Isolation and identification of *Alternaria spp.*

Alternaria spp. were isolated from diseased tomato leaves collected from different sites of the White Nile State during the surveys using single spore isolation method. Pieces of lesion tissue surface were disinfected with 70% ethanol for one minute rinsed with sterilized distilled water and then air-dried. Dried samples were placed on water agar 3% concentration, and single spore then collected under microscope using a hand-made glass needle under laminar flow unit were placed on potato dextrose agar (PDA) according to Agrios (1997).

After seven days of incubation at, plates were then examined for fungal growth under stereo microscope. The identification of the fungus was based on visual culture characteristics, mainly the growth patterns. Furthermore, microscopic examinations were carried out for mycelial and conidia structure based on the methods of. This was supplemented by microscopic examination of spores using a compound microscope. Other identification aids were; Ellis, and Ellis, (1985);; Agarwal, *et. al.*, (1989); Burgess*et al.*, (1994); Mathur, and Jorgensen, (1998); and Mathur and Kongsdal 2003).

3.2 Effect of aqueous extracts of natural products and fungicide on early blight disease:

3.2.1 Preparation of aqueous extracts:

The objective of this experiment was to study the antifungal activities of plant extracts of leaves of Neem, Usher, Argel and the fruit of mesquite on the incidence of early blight disease on tomato under field conditions. The extracts from the four plants were tested for their effects on the incidence of the fungus. Aqueous extracts of each of the plant materials were prepared as recommended by Okigbo (2006). The leaves of Neem, Usher, Argel and the fruit of mesquite were first washed carefully, shade dried, ground into powder

and stored in tightly covered glass jars wrapped with Aluminum foil until needed for preparation of extracts.

The obtained fine powder form different products were weighted separately and added to it equal amount of sterilized distilled water by volume into conical flask 250 ml and then placed in a shaker for 24 hrs. The mixture was then strained thorough a light cloth and then filtered through a WattmanNo1filter paper (24 cm). The stock solution was kept in the refrigerator at 4˚c for further work. Three concentrations (v/v) 100%, 50%, 25% were prepared by serial dilution with distilled water.

3.3 Field experiments:

The field experiment was conducted twice during seasons (2017/18 and 2018/19), in two different locations. The experiments which were carried out during winter season were conducted in an area infested with early blight of tomato to ensure presence of high inoculums pressure. Land was prepared by proper plaguing and then divided in to 54 plots allocated to three replications each of 18 plots. The plot size was 3* 2.5 m containing two Mastaba each of 240*80 cm. Plots were arranged in a complete Randomized Block Design (CRBD) with three replicates.

3.3.1 Sowing of seeds

Seeds of the tomato variety Strain B were sown directly in holes on the side of Mastaba, 20 cm between holes giving a total of thirty plants per plots. After irrigation, the plants were fertilized once by using Urea after 21 days after seeds germination at the rate of 40 kg urea / feddan.

3.3.2 Treatments:

Eighteen treatments were tested in this experiment which included; seed dressing by Seed Star 42 fungicide, Argel powder per holes at time of seeds sowing, Neem oil and aqueous extracts of leaves of, Neem, Usher, and Argel

in addition to aqueous extract of fruits of mesquite and control. The treatments were assayed for their bioactivity against *Alternaria spp*. in tomato winter production season under field conditions.

With the exception of Argel powder which was applied at time of sowing at the rate of 5g/ hole, the treatments were applied after appearance of first symptoms of Early Blight. The Neem oil was used at the concentrations of 10, 5 and 2.5 ml/ liter of water whereas aqueous extracts of leaves of Argel, Neem and Usher plus fruits of mesquite of 100, 50 and 25%. This was in addition to the Apron star fungicide at its standard rates of 5gm/1kg, seed dressing. Knapsack sprayer, 20 liter size was used for spraying. Application of treatments which started with the appearance of first symptom was repeated five times at ten days interval.

3.3.3 Data Collection:

Disease symptoms were observed starting from appearance of the first symptom on leaves before each application of treatments and continued until the downfall of diseased leaves. In each count ten plants were randomly selected from the middle of each plot and the number of plants showing *Alternaria* leaf symptoms were counted and expressed as a percentage of the plants inspected. Similar data about percentage of diseased fruits was also obtained according to;

$$\textbf{Disease incedence}\ (\%) = \frac{\textbf{Number of diseased leaves or fruits}}{\textbf{Total number of leaves or fruits inspected}}\textbf{x100}$$

At harvest time, the total yield was calculated for each treatment including the untreated control (Sallam and Kamal, 2012)underfield conditions.

3.4 Screening of tomato varieties for resistance to *Alternaria spp.*

Six commercial varieties of tomato, namely Castle Rock, Strain B, B2 (86), Hiraihry (Local variety), Goal and Domestic one (local name, Alla Kareem,

as resistant check) were used in this experiment. The land preparation and cultural practices were done as described before. Thirty plants of each variety were assigned to each plot of 3m x 2.5m size. Plots were arranged in a randomized complete block design (RCBD) with three replications under field conditions for natural infection where natural inoculums pressure was high.

3.4.1 Collection of data

A total of 9 tomato plants were randomly selected from the centre of the three plots, three from each replication and visited each count to assess the disease incidence till 100% infection was reached by any one of the test varieties. The assessment started with appearance of first symptom of early blight disease (Agrios, 1997). At the end of five count visit the number of plants showed early blight symptoms among the nine plants were calculated and expressed as a percentage of the regularly inspected ten plants. Similar counts for percentage disease incidence on fruits were done. At harvest time, the mean total yield was calculated for each variety , based on mean of four harvests of the crop during the season.

3.5 Data analysis

The obtained data were subject to analysis of variance for the randomized complete block design, using M Stat C computer program. Means were separated by Duncan's multiple range test at P = 0.05.

CHAPTER FOUR

RESULTS

The results of these studies are presented under the different parameters investigated. The results cover surveys of tomato commercial fields for occurrence and spread of early blight disease, isolation and identification of *Alternaria alternata.*, effect of natural plants formulations and fungicide on control of the pathogen and evaluation of tomato varieties for resistance to the disease.

4.1 Occurrence of *Alternaria spp.*

This was accomplished by conducting statewide tomato surveys for Early Blight disease in the cultivation sites for three successive growing seasons (2017/18, 2018/19 and 2019/20), covering four different locations in the White Nile State (North, Center, Eastern and Western South). The surveys were designed to collect data and qualitative information with which to assess the occurrence of *Alternaria spp*. and distribution as to be use as road map for future cultivation of tomato in the State.

The results of the three surveys were presented in Table 1 and figure 1. Regarding the survey of the first season and among the four locations the mean incidence of early blight disease was the highest in the North of the State of 14.67% followed by 10.33 %, 7.00%, and 1.67% in the Eastern South, Centreand Western South respectively. Obviously, the intensity of the disease as percent was less towards the South of the State. As for the South of the State, the data of the first season survey showed that the percentage of the disease incidence was relatively high in the Eastern South (10.33%) than in the Western South which had the lowest mean incidence of 1.76% percent.

As for the survey of the second season (2018/2019), the data showed similar trend of disease incidence to season 2017/2018 where in this season the highest incidence of *Alternaria spp*. was recorded from the North of the State 11.67% followed by 7.33%, 4.76% and 0.33%in Eastern South, Central and Western South respectively. During this survey season, the data also showed the high variation in early blight disease incidence between Eastern location of the South of the State 7.33% and the Western location 0.33%.

In the third survey season as well the data obtained showed that the North location of the State ranked the highest in the level of *Alternaria spp*. incidence, 19.00% followed by 6.33%, 5.67% and 0.67 % in the Central, Eastern and Western South of the State respectively. The data also revealed the high variation in early blight disease incidence between the two locations, Eastern (5.67%) and Western (0.67%), within the South of the State.

Obviously, the data obtained from the three successive surveys showed the consistent of high level of early blight disease in commercial field of tomato at the North part of the White Nile State. Likewise, the results of the three successive year's surveys highlighted the continued high difference in the level of incidence of *Alternaria spp*. between the Eastern South where the occurrence of the disease was relatively high compared to the Western side of the South of the State. As for the Centre of the State, the results of the three successive surveys showed relatively consistent level of disease incidence, 7.00%, 4.67% and 6.33% during the three years, 2017/2018, 2018/19 and 2019/2020 respectively and which ranking after North of the State in the level of disease prevalence.

Table 1: Mean percentage incidence of early blight disease in commercial field of tomato crop at different locations of White Nile State surveyed for three successive growing seasons

Locations	Seasons			Mean	Percentage incidence
	2017-18	2018-19	2019-20		
North of State	14.67 (58.67)	11.67 (46.67)	19.00 (76.00)	15.11 (60.44)	60.44%
Center of State	7.00 (28.00)	4.67 (18.66)	6.33 (25.33)	6.00 (23.99)	23.99%
Eastern South	10.33 (41.33)	7.33 (29.33)	5.67 (22.67)	7.78 (31.11)	31.11%
Western South	1.67 (6.67)	0.33 (1.33)	0.67 (2.67)	0.89 (3.56)	3.56%
Seasons mean	8.42 (33.67)	6.00 (23.99)	7.92 (31.67)	7.45 (29.78)	

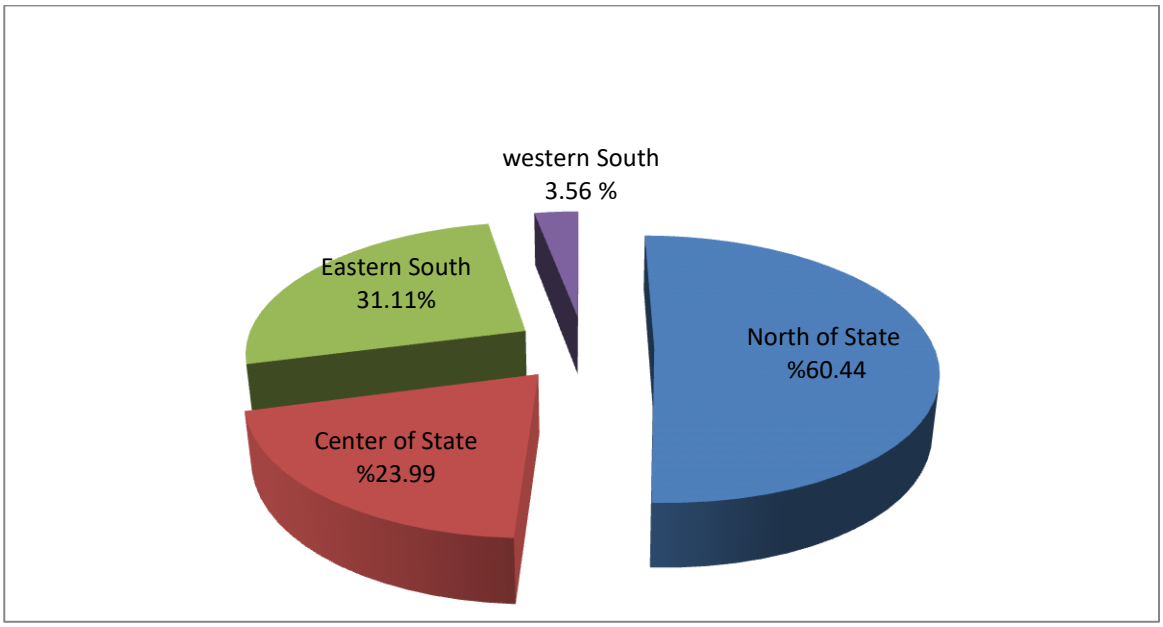

Fig. 1: Mean percentage incidence of early blight disease in randomly selected commercial fields of tomato crop at different locations of White Nile State surveyed for three successive growing seasons

4.2 The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in tomato plants under natural infection (first count, season2017 – 2018)

Table 2, presents the first count of percent disease incidence after ten days of the first spray of treatments that done at time of appearance of first symptoms of the disease.

The data showed that, among all treatments, six ones namely, Neem oil at 5ml/l, Argall aqueous extract at all three concentrations, Neem leaves extract at 100% concentration and Argall powder per hole, controlled the disease to zero was compared to other treatments. The effect of other treatments on percent disease incidence was variable, ranging from 0.33 to 2.67 compared to control 1.33. It is noteworthy that the extract of Argall at all concentrations decreased the disease incidence to zero (0.0) level. The disease incidence when Apron Star was use was 0.67% .

4.3: The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in tomato plants under natural infection (second count, season2017 – 2018)

Generally, in the second count all treatment except mesquite extract at 100 concentration, gave control on disease incidence. Their influences on disease incidence were significantly high at $P< 0.05$ compared to control which was 5.67 percent. The zero disease incidences were given by the treatments of Neem oil 5ml/l, Argall *leaves* extracts 25% and Argall powder per hole (table, 3). The variability in disease control among treatments was also observed during this second count. Their effects on disease incidence range between 0.33 percent by treatments of extracts of Argall 50%, Usher 100%, and Neem extracts at 100% and 4.00 percent of that of mesquite at 25%. Notably, the best treatments of Argall leaves extract 25 and 50% ranked second in the second count instead of zero percent in the first count.

4.4 The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in tomato plants under natural infection (third count, season2017 – 2018)

The results of the analysis of the third count data are shown in table 4. Showed that all treatments proved to be effective in controlling the disease but their effects on percent disease incidence were highly variable and inconsistent in some of them. It was observed that this variability in efficacy of treatments in controlling the disease increased with the advancing in number of count and age of the plant. However, the effects of all treatments on disease incidence were significantly high at $P < 0.05$ compared to control which was 14.33 percent. Moreover, the effects of Neem oil at 5ml/l and Argall powder at 5g/hole on disease incidence ranked top in controlling early blight disease in tomato. They consistently maintained highly significant effect on percent disease incidence recording zero percent disease for the three successive counts compared to the control which was 14.33 percent. It was observed the high level of disease control by Neem oil concentrations was sustained during the three counts.

Likewise, the effect of the treatments of argel leave extract 100%, Neem oil at 2.5ml/l, Argall leaves extract 100%, usher extract at 25%and 100% and the fungicide excelled the remaining treatments and control and gave 0.33, 1.0, 1.0 and 1.0respectively.

However, the data revealed that, although the efficacy of all concentrations of extracts of mesquite and Neem leaves on disease incidence was significant compared to control, but showed a decreased incidence with counts. Their low level of disease control ranging from 2.33 to 8.67 percent compared to control 14.33.

Table 2: The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in tomato plants tested under natural infection(first count, season2017 – 2018).

Treatments		Mean
Neem oil	**10 ml/l water**	0.67^{BC}
	5.0 ml/l water	0^{C}
	2.5ml/l water	0.33^{C}
Mesquite extract	100%	0.33^{C}
	50%	1.0^{BC}
	25%	2.67^{A}
Argall leaves extract	100%	0^{C}
	50%	0^{C}
	25%	0^{C}
Usher leaves extract	100%	0.33^{C}
	50%	0.33^{C}
	25%	2.00^{AB}
Neem leaves extract	100%	0.0^{C}
	50%	0.33^{C}
	25%	1.33^{ABC}
Argall Powder	5gm/hole	0.0^{C}
Seed star 42	5gm/Kg	0.67^{BC}
Control	0	1.33^{ABC}
$LSD_{0.05}$	**1.48**	
SE±	**0.72**	
CV%	7.8	

No significant differences between means with the same letter(s) within column at P= 0.05

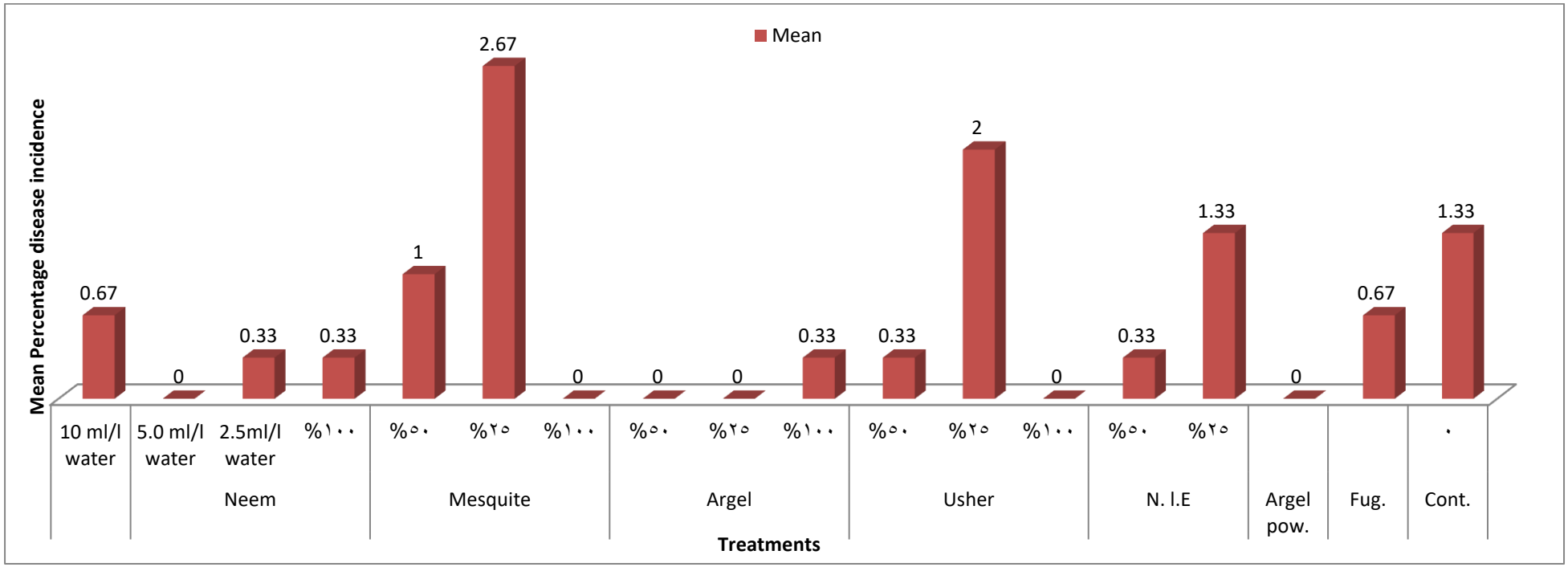

Fig. 2: The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in tomato plants under natural infection (First count, season 2017 – 2018).

Table 3: The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease on tomato plants tested under natural infection (second count, season 2017 – 2018).

Treatments		Mean
Neem oil	**10 ml/l water**	2.00^{BCDE}
	5.0 ml/l water	0.00^{E}
	2.5ml/l water	0.67^{DE}
Mesquite extract	100%	1.00^{DE}
	50%	2.33^{BCD}
	25%	4.00^{AB}
Argall leaves extract	100%	0.00^{E}
	50%	0.33^{DE}
	25%	1.00^{DE}
Usher leaves extract	100%	0.33^{DE}
	50%	1.00^{DE}
	25%	2.00^{BCDE}
Neem leaves extract	100%	0.33^{DE}
	50%	1.33^{CDE}
	25%	3.33^{BC}
Argall Powder	5gm/hole	0.00^{E}
Seed star 42	5gm/Kg	1.00^{DE}
Control	0	5.67^{A}
$LSD_{0.05}$	**2.12**	
SE±	**1.04**	
Cv%	8.75	

No significant differences between means with the same letter(s) within column at P= 0.05.

Table 4: The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in tomato plants tested under natural infection (third count, season2017 – 2018).

Treatments		Mean
Neem oil	**10 ml/l water**	2.00^{EF}
	5.0 ml/l water	0.00^{F}
	2.5ml/l water	1.00^{F}
Mesquite extract	100%	5.00^{CDE}
	50%	2.33^{EF}
	25%	8.67^{B}
Argall leaves extract	100%	0.33^{F}
	50%	2.00^{EF}
	25%	2.00^{EF}
Usher leaves extract	100%	1.00^{F}
	50%	3.00^{DEF}
	25%	1.00^{F}
Neem leaves extract	100%	5.67^{BCD}
	50%	7.67^{BC}
	25%	6.00^{BCD}
Argall Powder	5gm/hole	0.00^{F}
Seed star 42	5gm/Kg	1.00^{F}
Control	0	14.33^{A}
$LSD_{0.05}$	**3.09**	
SE±	**1.52**	
Cv%	3.45	

No significant differences between means with the same letter(s) within column at P= 0.05

4.5 The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in tomato plants under natural infection (forth count, season2017 – 2018)

Considering the past three counts, the data of the forth count sustained the same pattern where all treatments had reduced the disease incidence to a level lower than that of the control. Their scores on disease incidence were significantly high at $P< 0.05$ compared to control which was 16.33 percent. Among these treatments, that of Neem oil at 5 ml/l, Argall powder per hole, Argall leaves extracts 100% and fungicide ranked top in control of the disease. However, the data highlighted the remarkable control of early blight disease given by these treatments. Their score of disease control range from 0.0 percent with Neem oil at 5ml/l and Argall powder per hole to 0.33 and 1.0 percent with Argall leaves extract at 100%and fungicide respectively.

It is worthnoty that the efficacy of these four treatments in sustaining this very low disease incidence (0.0 to 1.0 percent) throughout the counting periods. However, the effect of other treatments e.g. that of mesquite and Neem leaves exhibited a successively reducing efficacy of disease control and buildup of early blight with plant again each count. The record of their disease incidence was ranging from 4.67 to 11.33 percent.

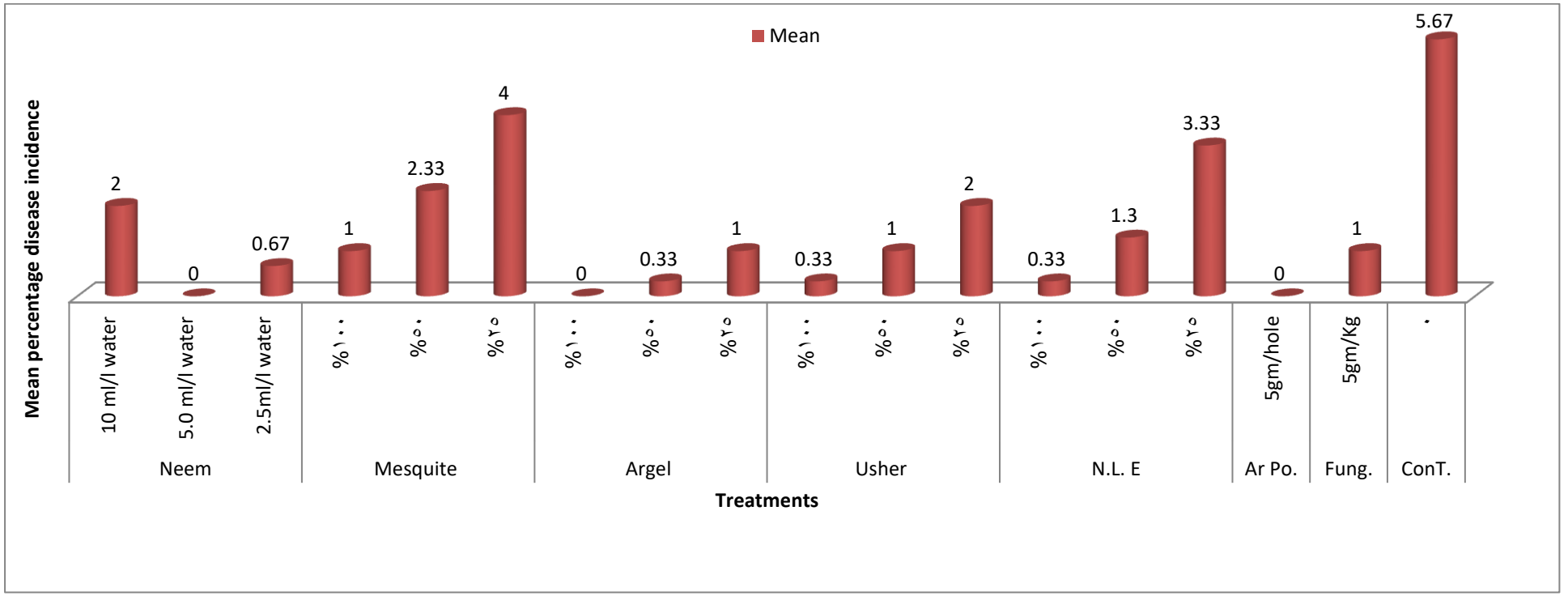

Fig. 3: The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in tomato plants under natural infection (Second count, Season 2017 – 2018)

Table 5: The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in tomato plants tested under natural infection (forth count, season2017 – 2018).

Treatments		Mean
Neem oil	**10 ml/l water**	2.00^{EFG}
	5.0 ml/l water	0.00^{G}
	2.5ml/l water	1.33^{FG}
Mesquite extract	100%	6.00^{CDE}
	50%	4.67^{DEF}
	25%	11.33^{B}
Argall leaves extract	100%	0.33^{G}
	50%	2.33^{EFG}
	25%	2.33^{EFG}
Usher leaves extract	100%	2.00^{EFG}
	50%	3.00^{EFG}
	25%	1.33^{FG}
Neem leaves extract	100%	8.67^{BCD}
	50%	9.67^{BC}
	25%	10.33^{B}
Argall Powder	5gm/hole	0.00^{G}
Seed star 42	5gm/Kg	1.00^{FG}
Control	0	16.33^{A}
$LSD_{0.05}$	**4.19**	
SE±	**2.06**	
CV%	5.20	

No significant differences between means with the same letter(s) within column at P= 0.05

4.6 The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease on tomato fruits under natural infection (first count, season2017 – 2018)

Table, 6 presents the incidence of early blight diseases on tomato fruits at harvest time. The data indicated clearly the influence of the four top ranked treatments; Neem oil at 5 ml/l, Argall powder per hole, Argall leaves extracts 100% and fungicide, in controlling the early blight disease in plant leaves was also continued to express their effect on control disease incidence on fruits of tomato at harvest. They significantly reduced the incidence of the disease giving 0.33 percent with Neem oil at 5 ml/l, Argall powder per hole, fungicide and 0.67 with Argall leaves extracts at 100% compared to control 3.67 %. The other treatments have had variable level of disease control ranging between 1.0 to 5.0 percent.

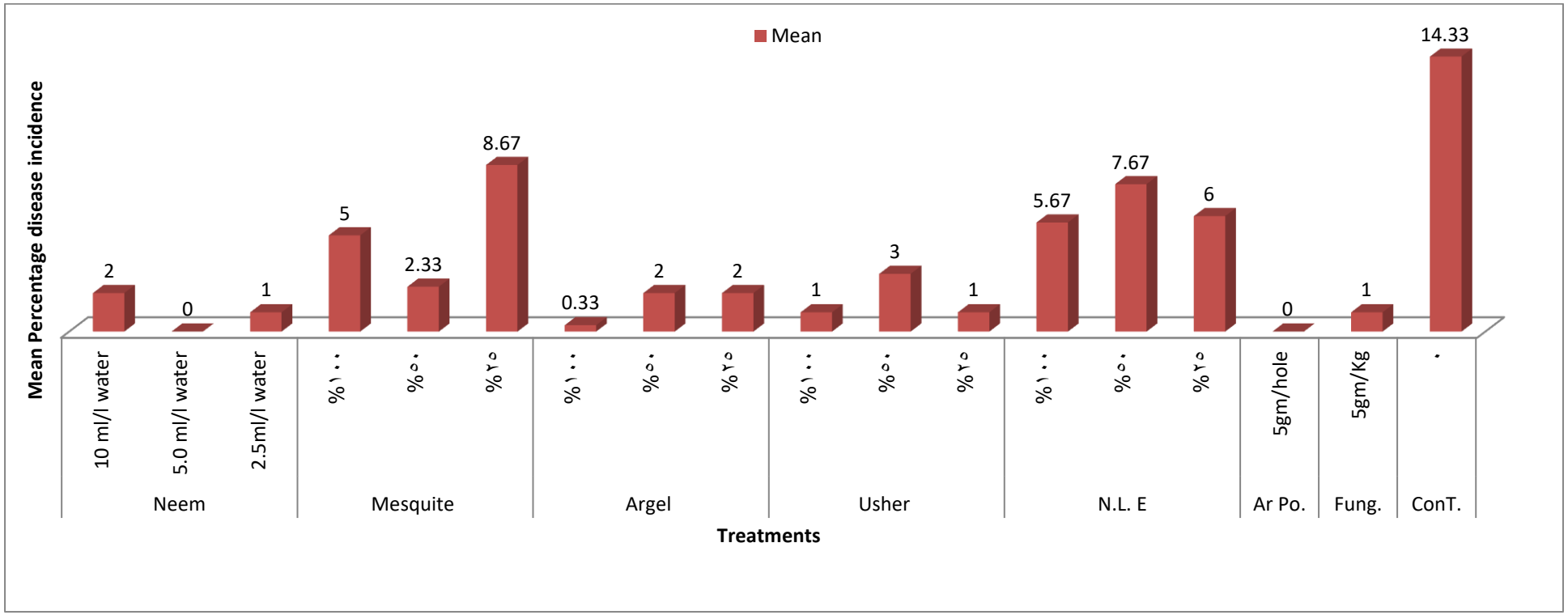

Fig. 4: The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in tomato plants tested under natural infection (third count, season 2017 – 2018).

4.7 The effect of different concentrations of natural products formulations and fungicide on total weight of tomato fruits tested under natural infection (season2017 – 2018)

The effects of treatments on the total weight of tomato after four successive harvests are presented in Table 7. Treatments of Neem oil at 5 ml/l, argel powder per hole, extracts of leaves of Argall at all concentrations and fungicide scored the highest total weights of tomato, in evaluation of effect of treatments on yield, with significant difference from the control. Concerning the impact of other treatments on total weight, three of them were ranked second to the above mentioned treatments for total yield although the difference was not significant compared to those four treatments? It is noteworthy that in this investigation, all Argall treatments resulted in highest total weight compared to the control. However, the remaining treatments effect on yield was not significant where they gave total weight of tomato ranging from the lowest yield of 18.4 to 23.0 Kg per treatment compared to control 23.1kg.

Table 6: The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease on tomato fruits tested under natural infection (fife count, season2017 – 2018).

Treatments		Mean
Neem oil	**10 ml/l water**	1.00^{CDE}
	5.0 ml/l water	0.33^{E}
	2.5ml/l water	1.67^{BCDE}
Mesquite extract	100%	2.33^{ABCDE}
	50%	2.67^{ABCDE}
	25%	3.67^{ABC}
Argall leaves extract	100%	0.67^{DE}
	50%	1.67^{BCDE}
	25%	2.33^{ABCDE}
Usher leaves extract	100%	3.33^{ABCD}
	50%	3.00^{ABCDE}
	25%	4.67^{A}
Neem leaves extract	100%	4.00^{AB}
	50%	4.33^{AB}
	25%	5.00^{A}
Argall Powder	5gm/hole	0.33^{E}
Seed star 42	5gm/Kg	0.33^{E}
Control	0	3.67^{ABC}
$LSD_{0.05}$	**2.77**	
SE±	**1.36**	
CV%	7.11	

No significant differences between means with the same letter(s) within column at P= 0.05

Table 7: The effect of different concentrations of natural products formulations and fungicide on total weight of tomato fruits tested under natural infection after four harvests(first season 2017 – 2018)

Treatments	Weights (kg) per treatment				Total count	Mean	S.d	C.V
	Harvest 1	Harvest 2	Harvest 3	Harvest 4				
Neem oil 10 ml/l water	0.5	3.5	16	5	25.0	6.3	2.16	٠.34
Neem oil 5.0 ml/l water	2.3	8.5	22.5	8	41.3	10.3	2.78	٠.26
Neem oil 2.5ml/l water	2.1	9.5	14	5.5	31.3	7.8	2.42	٠.31
Mesquite extract 100%	0.5	6.5	12	4	23.0	5.8	2.07	٠.35
Mesquite extract 50%	2.3	6	8	3.5	19.8	5.0	1.92	٠.38
Mesquite extract 25%	2.1	8	6	3	19.1	4.8	1.89	٠.39
Argall leaves extract 100%	3.3	7.5	17	7	34.8	8.7	2.55	٠.29
Argall leaves extract 50%	2.3	9	16	8	35.3	8.8	2.57	٠.29
Argall leaves extract 25%	2.0	6.5	16	6	30.5	7.6	2.39	٠.31
Usher leaves extract 100%	0.6	4	9	6	19.6	4.9	1.92	٠.39
Usher leaves extract 50%	2.6	6.5	11	2	22.1	5.5	2.03	٠.36
Usher leaves extract 25%	0.6	3.5	13	4	21.1	5.3	1.98	٠.37
Neem leaves extract 100%	1.0	6	9	3	19.0	4.8	1.88	٠.39
Neem leaves extract 50%	1.2	8	8	4	21.2	5.3	1.99	٠.37
Neem leaves extract 25%	0.4	3	10	5	18.4	4.6	1.86	٠.40
Argall Powder 5gm/hole	4.0	6	26	10	46.0	11.5	2.93	٠.25
Seed star 42 5gm/kg	0.8	6	23	7	36.8	9.2	2.62	٠.28
Control	2.6	8.5	10	2	23.1	5.8	2.07	٠.35

No significant differences between means with the same letter(s) within column at P= 0.05

4.8 Mean percent incidence of early blight disease on some tomato varieties screened for their resistance to the disease under natural infection

The results of the interaction of tested tomato varieties with early blight disease under natural infection were presented in table 8. All tomato varieties screened for evaluation of their resistance to the disease were infected but at variable level. Nevertheless, the domestic variety (Allah Kareem) showed low percent of disease incidence with 33.3% followed by the hybrid Strain B compared to other tested varieties. The other varieties exhibited high level of disease incidence ranging from 77.8 % with B2-86, 88.9 with varieties Castle rock and Hiraihry and 100 % with Goal.

4.9 The effect of early blight disease on yield of different tomato varieties tested under natural infection

Table, 9 presents the results of the effect of early blight disease on total yield of four harvests of different tomato varieties under natural infection. Generally, the tested varieties which exposed to natural infection were given variable yield performance. Once again, the domestic variety and Strain B ranked the top in yield compared to other ones. The total yield obtained was significantly excelled that of other at P= 0.05. They yielded a total of 20.8 Kg and 20.4 Kg for variety Domestic and Strain B respectively after four harvests followed by 17.2, 14.1, 11.9 and 7.5 Kg for Castle rock, B2-86, Hiraihry and Goal respectively. It is worth mentioning that the Goal variety which scored the lowest yield total (7.5 Kg) gave the highest percent disease incidence under natural infection (100%).

Table 8: Mean percent incidence of early blight disease on some tomato varieties screened for resistance to early blight disease under natural infection

Varieties	Number of plants of positive infection among nine ones									Total Plants infected	Percentage Incidence
	1	2	3	4	5	6	7	8	9		
Castle rock	+	+	+	+	+	+	+	0	+	8.0	88.9%
Strain B	0	+	+	+	0	0	0	+	+	5.0	55.60%
B2-86	+	+	+	0	0	+	+	+	+	7.0	77.8%
Hiraihry	+	+	+	+	0	+	+	+	+	8.0	88.9%
Goal	+	+	+	+	+	+	+	+	+	9.0	100%
Domestic (control)	0	+	0	0	0	0	+	+	0	3.0	33.3%

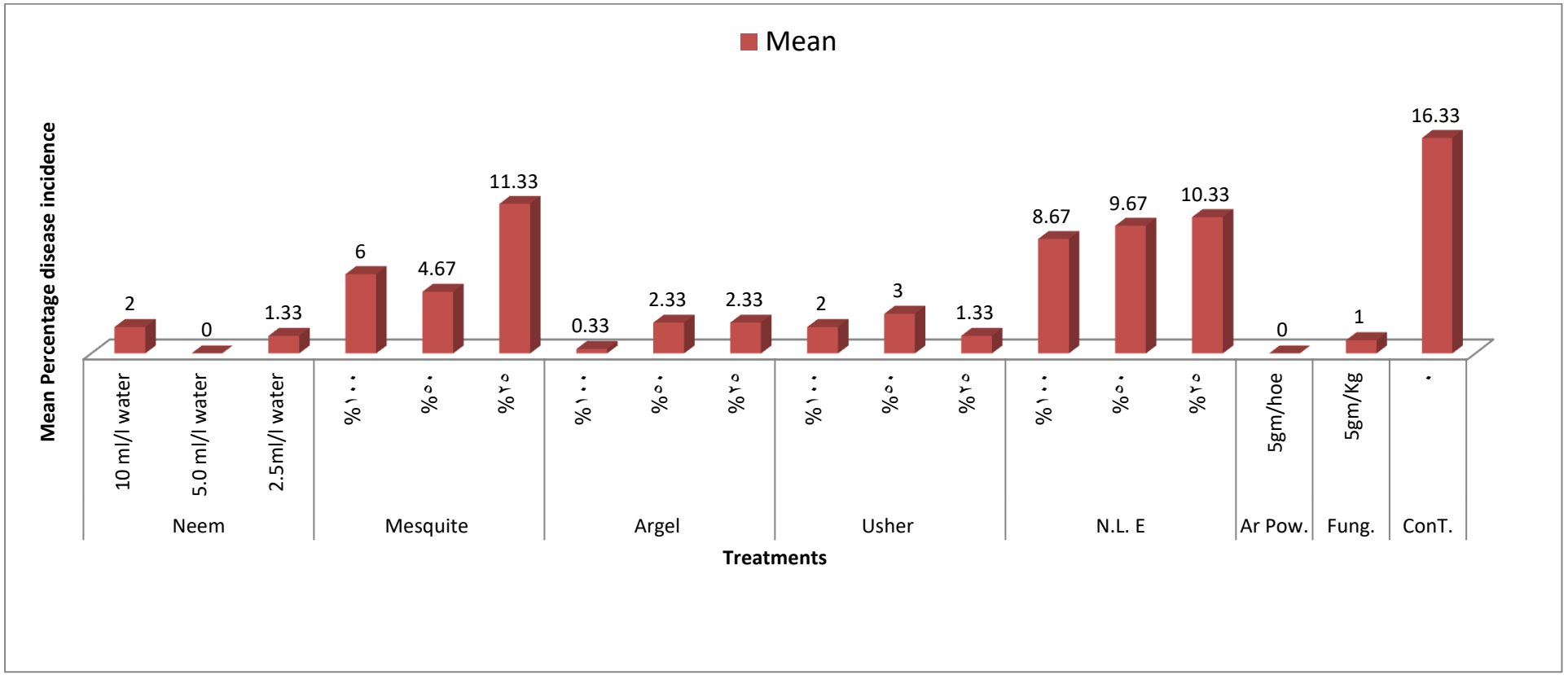

Fig. 5: The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in tomato plant tested under natural infection (forth count, season 2017 – 2018).

Table 9: The effect of early blight disease on yield of different tomato varieties tested under natural infection.

Varieties	Weight(kg)				Total	Mean
	Harvest 1	Harvest 2	Harvest 3	Harvest 4		
Castle rock	3.1	6.4	5.0	2.7	17.2	4.3
Strain B	4.6	7.1	5.3	3.4	20.4	5.1
B2 -86	2.1	4.9	5.4	1.7	14.1	3.5
Hiraihry	2.0	4.3	4.6	1.0	11.9	3.0
Goal	1.0	2.6	3.0	0.9	7.50	1.9
Domestic	4.8	6.7	6.1	3.2	20.8	5.2

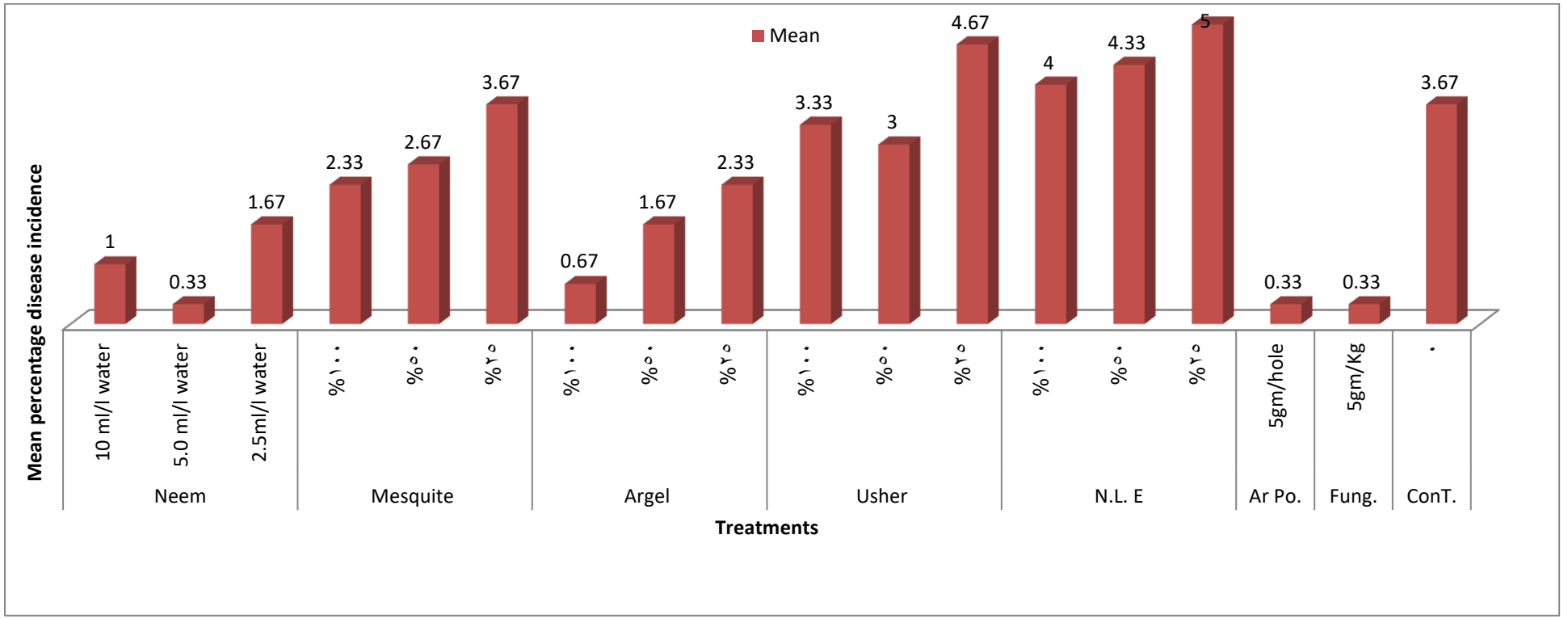

Fig. 6: The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease on tomato fruits tested under natural infection (first count, season 2017 – 2018).

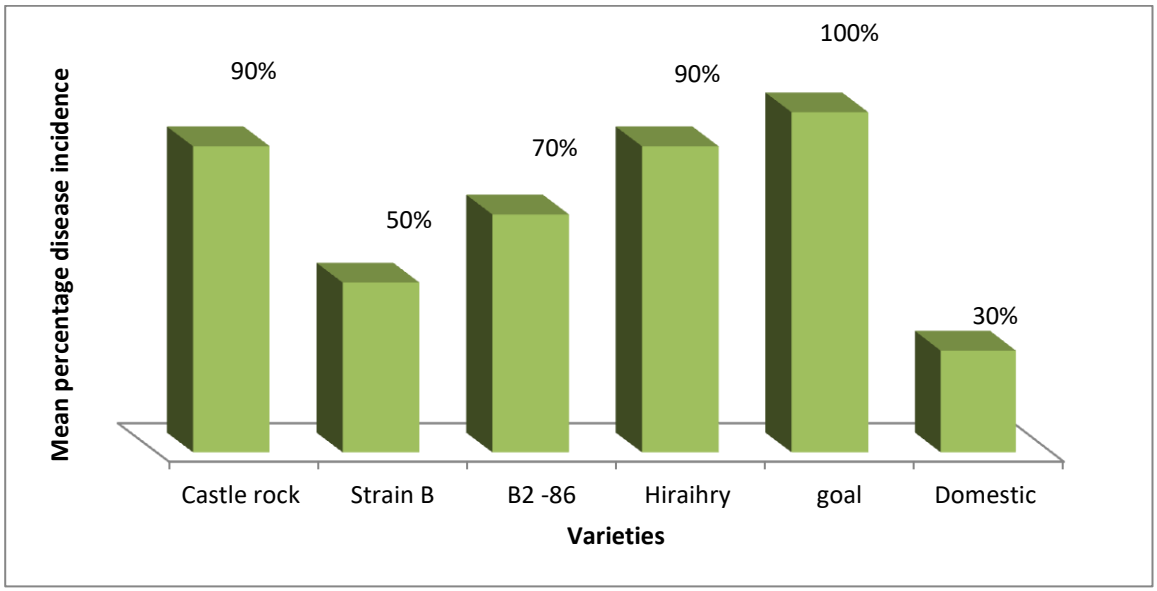

Fig. 7: Mean percent incidence of early blight disease on some tomato varieties tested under natural infection

Table 10: The effect of early blight disease on yield of different tomato varieties tested under natural infection.

Varieties	Weight(kg)
Castle rock	4.3^{A}
Strain B	5.1^{A}
B2 -86	3.5^{AB}
Hiraihry	3.0^{AB}
Goal	1.9^{B}
Domestic	5.2^{A}
$LSD_{0.05}$	2.39
$SE\pm$	1.14
CV%	4.12

No significant differences between means with the same letter(s) within column at P= 0.05

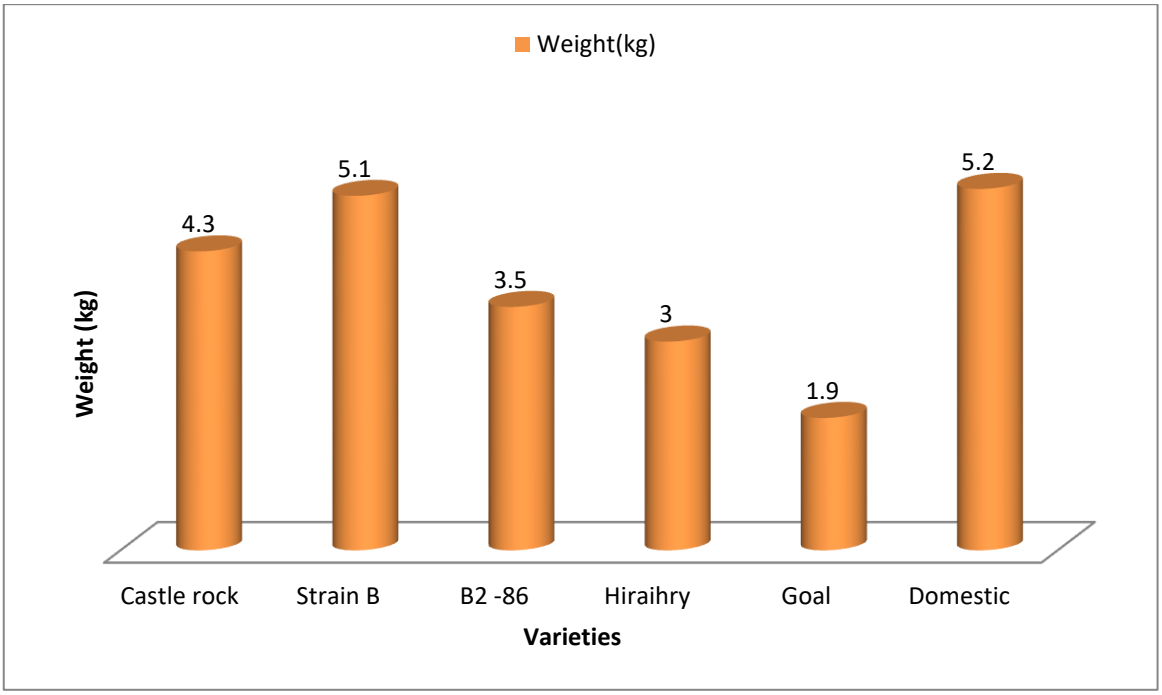

Fig. 8: The effect of early blight disease on yield of different tomato varieties tested under natural infection.

4.10 The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in tomato plants tested under natural infection (first count, second season 2018-2019)

Results of the experiment set up next season to determine the effect of different concentrations of natural products formulations and fungicide on incidence of early blight diseases in tomato plants under natural infection are presented in Table 11. Generally, the overall records of the disease incidence were low in all treatments. Accordingly, the percent disease given by all treatments was ranging between 0.0% to 1.0% percent. Although the percent disease incidence was low but the analysis of variance revealed that some of the treatments recorded 0.0% disease incidence which were significantly different at P=0.05 compared to control. However, among those treatments with 0.0% disease incidence are Argall powders per hole, Neem leaves extract 100%, and Argall leaves extract 100% and fungicide.

It is noteworthy that the efficacy of these four treatments in sustaining this very low disease incidence (0.0%) throughout the counting periods in the first year experiment (2017/2018) and in the second experiment repeated the year (2018/2019).

Season (2)

Table 11: The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in tomato plants tested under natural infection (first count, second season 2018-2019)

Treatments		Mean
Neem oil	**10 ml/l water**	0.33^{BC}
	5.0 ml/l water	0.33^{BC}
	2.5ml/l water	0.67^{ABC}
Mesquite extract	100%	0.33^{BC}
	50%	0.67^{ABC}
	25%	1.00^{AB}
Argall leaves extract	100%	0.00^{C}
	50%	0.33^{BC}
	25%	1.00^{AB}
Usher leaves extract	100%	0.33^{BC}
	50%	1.33^{A}
	25%	1.00^{AB}
Neem leaves extract	100%	0.00^{C}
	50%	0.33^{BC}
	25%	1.00^{AB}
Argall Powder	5gm/hole	0.00^{C}
Seed star 42	5gm/Kg	0.00^{C}
Control	0	1.00^{AB}
$LSD_{0.05}$	**0.95**	
SE±	**0.47**	
Cv%	10.5	

No significant differences between means with the same letter(s) within column at P= 0.05

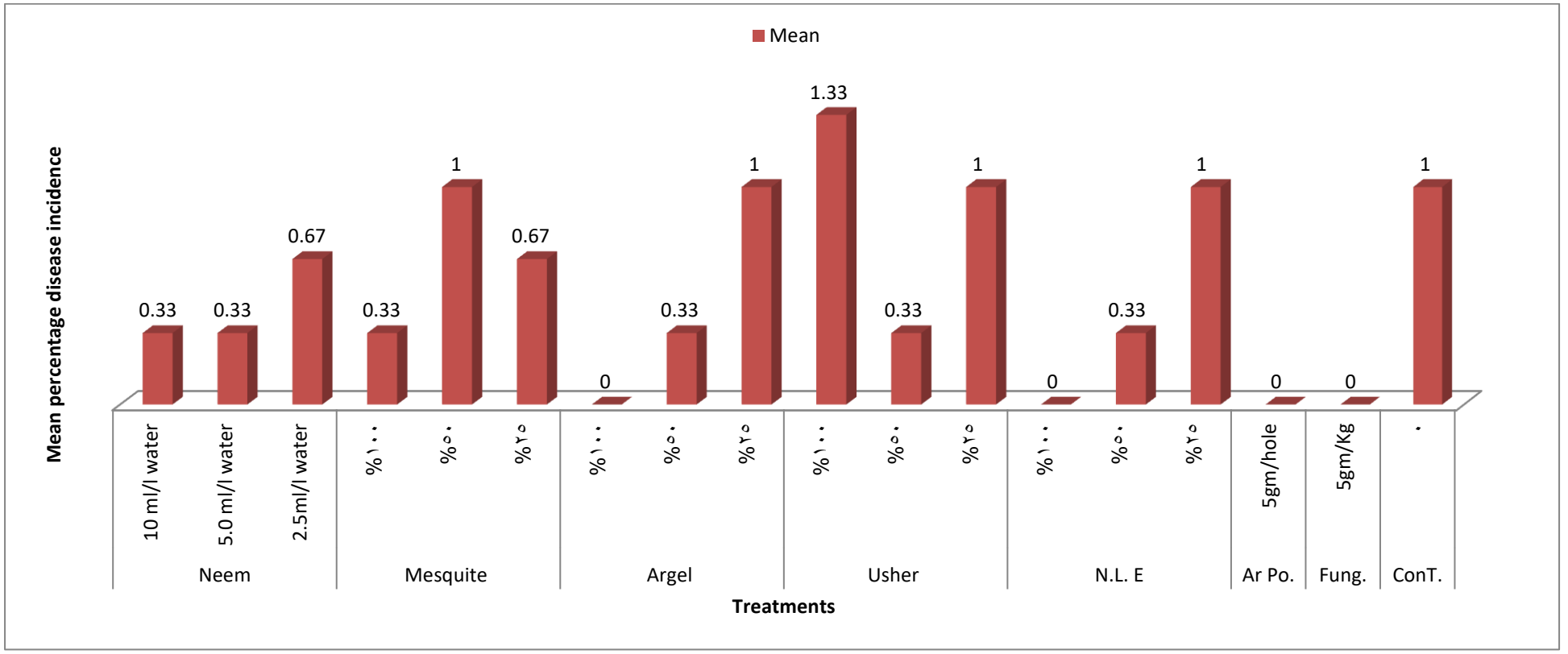

Fig. 9: The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in tomato plants tested under natural infection (first count, second season 2018-2019)

4.11 The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in tomato plants tested under natural infection (First count, second season 2018-2019)

In the second count the pattern of low disease incidence continued but with slight increase in disease incidence from that of the first count (table, 12). However, among treatments, although there is an overall increase in the percent of disease incidence but that of fungicide and Argall powder per hole remained the lowest, 0.0 and 0.33% respectively compared to control 1.67. This low level of disease incidence given by those two treatments was significantly different at P=0.05 compared to other treatments and control. Moreover, there is no great variability in disease control among other treatments during this second count. Their influence on disease incidence was ranging between 1.0 and 2.0 %.

Table 12: The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in tomato plant tested under natural infection(Second count, second season 2018-2019)

Treatments		Mean
Neem oil	**10 ml/l water**	1.33^{AB}
	5.0 ml/l water	1.00^{ABC}
	2.5ml/l water	1.67^{A}
Mesquite extract	100%	1.33^{AB}
	50%	2.00^{A}
	25%	1.67^{A}
Argall leaves extract	100%	1.00^{ABC}
	50%	1.00^{ABC}
	25%	1.33^{AB}
Usher leaves extract	100%	2.00^{A}
	50%	1.67^{A}
	25%	2.00^{A}
Neem leaves extract	100%	2.00^{A}
	50%	1.33^{AB}
	25%	1.67^{A}
Argall Powder	5gm/hole	0.33^{BC}
Seed star 42	5gm/hole	0.00^{C}
Control	0	1.67^{A}
$LSD_{0.05}$	**1.31**	
SE±	**0.65**	
CV%	7.13	

No significant differences between means with the same letter(s) within column at P= 0.05

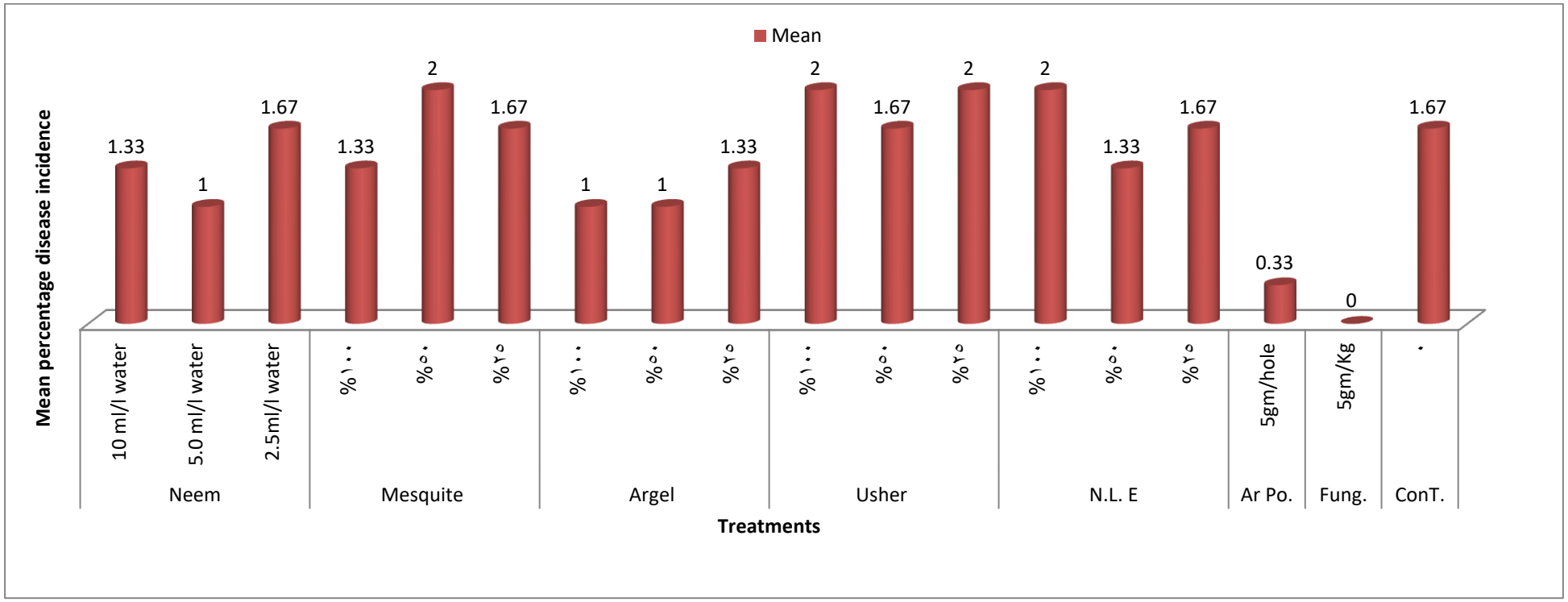

Fig. 10: The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in tomato plant tested under natural infection (Second count, second season 2018-2019)

4.12 The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in tomato tested under natural infection(third count, second season 2018-2019)

The results of the analysis of the data of the third count are shown in table 13. Generally, all treatments affect the incidence of early blight disease on tomato plants but their influence was variable and inconsistent in some of them, especially among concentrations. It was observed that this variability in efficacy of treatments in controlling the disease increased with the advancing in number of count and age of the plant. Nevertheless, the effects of treatments of Argall powder at 5g/hole and fungicide on disease incidence ranked top in controlling early blight on tomato. In fact, they consistently maintained highly significant effect at P = 0.005 on disease incidence recording the lowest one 0.33 and 0.67% for the three successive counts compared to the control which was 2.33 percent. It was observed that the relatively low level of disease control by Argall extracts at all concentrations was sustained during this count between 1.33 and 1.67. This in addition to that of Neem oil at 5ml/l of water which gave also 1.33% control compared to other treatments.

However, the data revealed that the efficacy of other treatments was decreased with counts. Their low level of disease control range from 2.0 to 3.0 percent compared to control which is 2.33 percent.

Table 13: The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in tomato tested under natural infection(third count in second season 2018-2019)

Treatments		Mean
Neem oil	**10 ml/l water**	2.00^{ABCD}
	5.0 ml/l water	1.33^{CDE}
	2.5ml/l water	2.67^{ABC}
Mesquite extract	100%	2.67^{ABC}
	50%	3.00^{AB}
	25%	3.33^{A}
Argall leaves extract	100%	1.67^{BCDE}
	50%	1.33^{CDE}
	25%	1.67^{BCDE}
Usher leaves extract	100%	2.33^{ABC}
	50%	2.33^{ABC}
	25%	2.67^{ABC}
Neem leaves extract	100%	3.00^{AB}
	50%	2.67^{ABC}
	25%	2.67^{ABC}
Argall Powder	5gm/hole	0.67^{DE}
Seed star 42	5gm/Kg	0.33^{E}
Control	0	2.33^{ABC}
$LSD_{0.05}$	**1.40**	
SE±	**0.69**	
Cv%	9.56	

No significant differences between means with the same letter(s) within column at P= 0.05

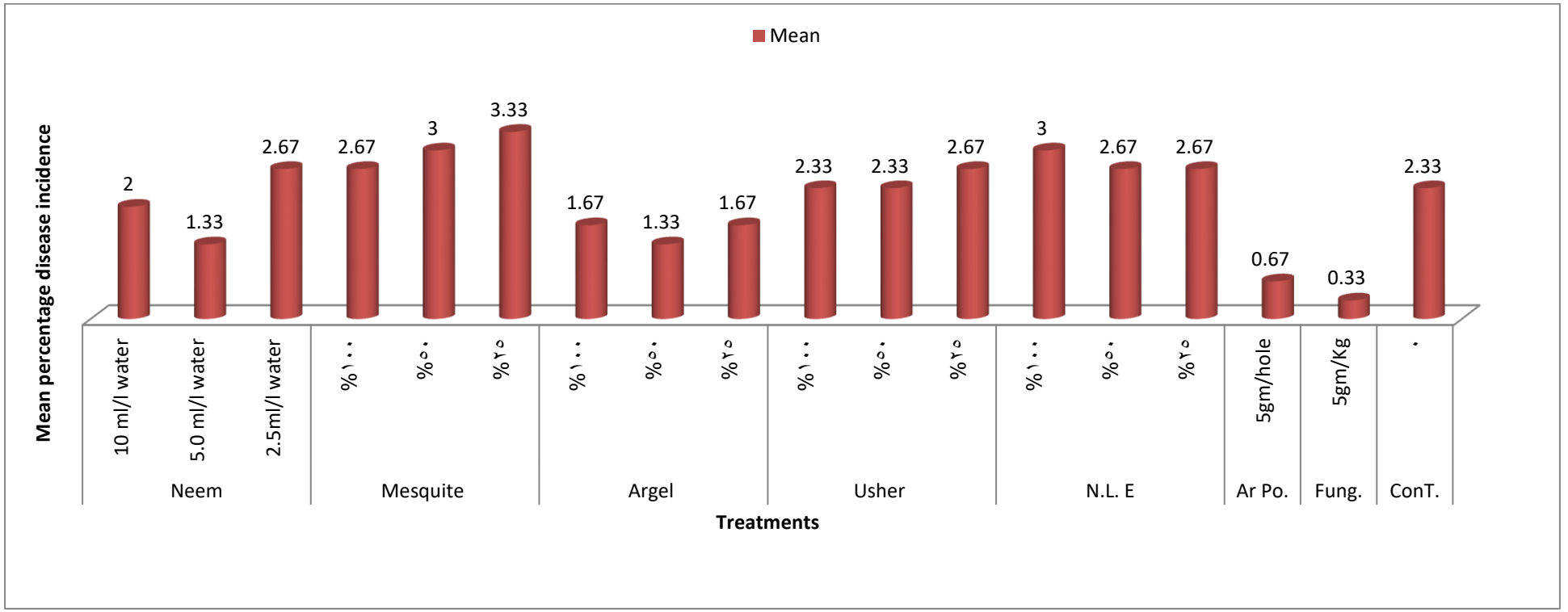

Fig. 11: The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in tomato tested under natural infection (third count in second season 2018-2019)

4.13 The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in tomato tested under natural infection (forth count in second season 2018-2019)

Results of the experiment set up next season to determine the effect of different concentrations of natural products formulations and fungicide on incidence of early blight diseases in tomato under natural infection are presented in Table 14. Although generally, the level of disease incidence was low during the last three counts but the data during this count highlighted the buildup of early blight in most of the treatments where their influence on it was variable ranging from 1.67 to 4.0 %.

Nevertheless, the treatments of Argall powder at 5g/hole and fungicide on disease incidence consistently maintained highly significant effect at P = 0.005 on disease incidence recording the lowest ones 0.33 and 0.67% for four successive counts compared to the control which was 2.67 percent.

4.14 The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in fruits of tomato plant tested under natural infection (season 2018 – 2019)

The results of the effect of different concentrations of natural products formulations and fungicide on incidence of early blight diseases in fruits of tomato at harvest time under natural infection were presented in table 15.Generally; all treatments influenced the incidence of early blight disease on fruits of tomato but at variable level. Obviously, the treatments of Argall powder per hole and fungicide ranked toping controlling the early blight disease in fruits of tomato at harvest. They significantly reduced the incidence of the disease giving 0.00 and 0.33% compared to control 5033 %.

Moreover, the leaves extracts of argel and mesquite at 100% and Neem oil at 5ml/l of water also recorded to reduce the disease incidence to the minimum giving 0.67%. The other treatments have had variable level of disease control ranging between 1.0 to 5.33 percent.

Table 14: The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in tomato tested under natural infection (forth count in second season 2018-2019)

Treatments		Mean
Neem oil	**10 ml/l water**	2.33^{BC}
	5.0 ml/l water	1.67^{CD}
	2.5ml/l water	3.33^{AB}
Mesquite extract	100%	3.00^{ABC}
	50%	3.33^{AB}
	25%	4.00^{A}
Argall leaves extract	100%	2.67^{ABC}
	50%	2.67^{ABC}
	25%	3.33^{AB}
Usher leaves extract	100%	4.00^{A}
	50%	3.00^{ABC}
	25%	3.33^{AB}
Neem leaves extract	100%	3.33^{AB}
	50%	3.33^{AB}
	25%	3.33^{AB}
Argall Powder	5gm/hole	0.67^{D}
Seed star 42	5gm/hole	0.33^{D}
Control	0	2.67^{ABC}
$LSD_{0.05}$	**1.44**	
SE±	**0.71**	
Cv%	3.96	

No significant differences between means with the same letter(s) within column at P= 0.05

Table 15: The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in fruits of tomato plant tested under natural infection(season, 2018 – 2019)

Treatments		Mean
Neem oil	**10 ml/l water**	0.33^{FG}
	5.0 ml/l water	0.67^{FG}
	2.5ml/l water	0.33^{FG}
Mesquite extract	100%	0.67^{FG}
	50%	1.00^{EFG}
	25%	1.33^{EFG}
Argall leaves extract	100%	0.67^{FG}
	50%	1.00^{EFG}
	25%	1.67^{DEFG}
Usher leaves extract	100%	2.00^{CDEF}
	50%	2.00^{CDEF}
	25%	2.67^{BCDE}
Neem leaves extract	100%	3.33^{BCD}
	50%	3.67^{ABC}
	25%	4.33^{AB}
Argall Powder	5gm/hole	0.00^{G}
Seed star 42	5gm/hole	0.33^{FG}
Control	0	5.33^{A}
$LSD_{0.05}$	**1.92**	
SE±	**0.94**	
Cv%	6.79	

No significant differences between means with the same letter(s) within column at P= 0.05

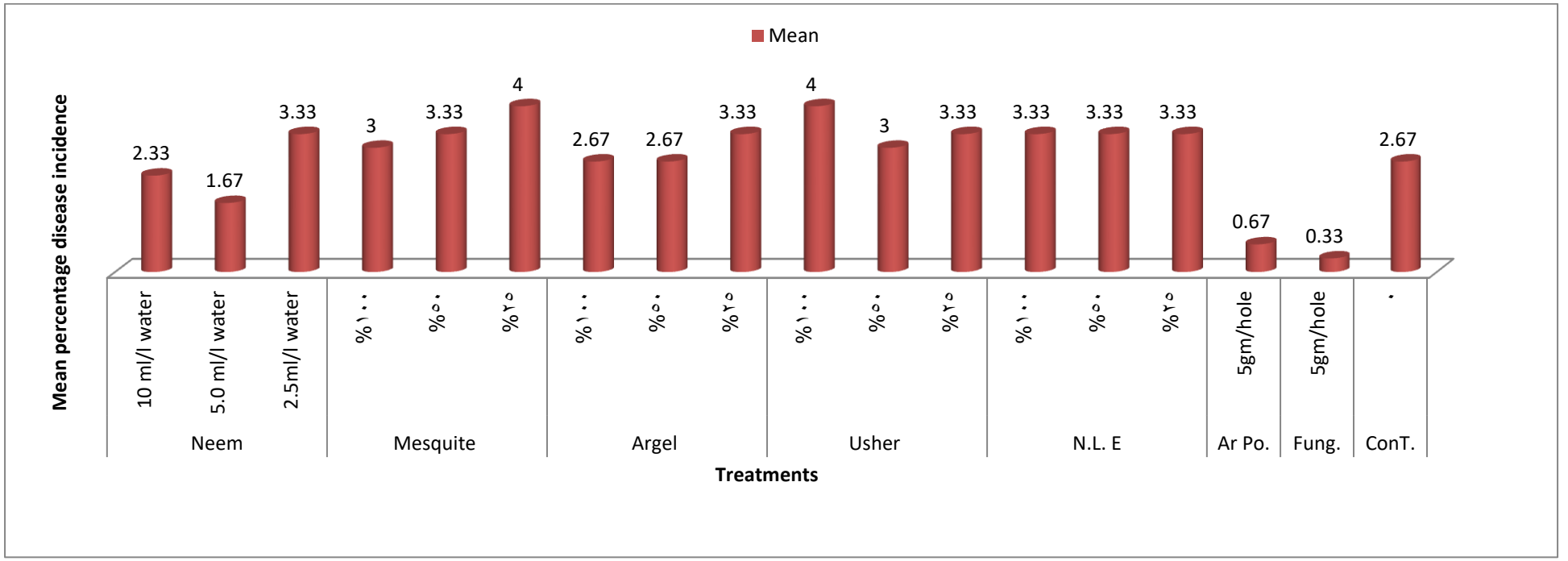

Fig. 12: The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in tomato tested under natural infection (forth count in second season 2018-2019)

Table 16: The effect of different concentrations of natural products formulations and fungicide on total weight of tomato plant tested under natural infection (Second season, 2018 – 2019)

Treatments	Weights (kg)				Total count	Mean	S.d	C.V
	w(1)	w(2)	w(3)	w(4)				
Neem oil 5.0 ml/l water	1.5	4.0	6.5	3.5	15.5	5.2	1.60	٠.30
Neem oil 2.5 ml/l water	1.0	5.5	6.5	2.0	15.0	5.0	1.58	٠.31
Neem oil1.0 ml/l water	1.0	3.5	4.5	4	13.0	4.3	1.47	٠.34
Mesquite fruit extract100%	0.5	2.7	3.9	1.5	8.60	2.9	1.19	٠.41
Mesquite fruit extract 50%	0.5	3.1	4.0	1.0	8.60	2.9	1.19	٠.41
Mesquite fruit extract 25%	0.7	2.2	5.0	1.0	9.40	3.1	1.25	٠.40
Argall leaves extract 100%	1.9	7.0	5.2	3.5	17.6	5.9	1.71	٠.28
Argall leaves extract 50%	1.6	6.7	4.8	2.0	15.1	5.3	1.56	٠.29
Argall leaves extract 25%	0.8	7.0	3.8	0.6	12.2	4.7	1.36	٠.28
Usher leaves extract 100%	0.7	4.0	2.7	0.8	8.20	2.7	1.17	٠.43
Usher leaves extract 50%	0.5	5.0	3.6	0.5	9.60	3.2	1.26	٠.3
Usher leaves extract 25%	1.0	4.9	1	0.7	7.60	2.5	1.13	٠.45
Neem leaves extract 100%	5.0	2.1	1.3	1.4	9.80	3.3	1.27	٠.38
Neem leaves extract 50%	3.1	3.5	3.o	0.4	10.0	3.3	1.29	٠.39
Neem leaves extract 25%	0.6	2.9	3.7	0.5	7.70	2.6	1.13	٠.43
Argall powder 5g/ hole	3.6	6.9	5.8	3.5	19.8	6.6	1.81	٠.27
Seed star 42 5g / kg	2.9	6.7	6.0	3	18.6	6.2	1.76	٠.28
Control	0.5	4.2	4.0	0.3	9.6	3.2	1.26	٠.39

4.15 The effect of different concentrations of natural products formulations and fungicide on total weight of tomato plant tested under natural infection(second season, 2018 – 2019)

Results of the experiment set up next season to determine the effect of different concentrations of natural products formulations and fungicide on total weight of tomato fruits under natural infection at harvest (Second season, 2018–2019)are presented in Table 15.

The data showed the variability of the influence of treatments on the total weight of tomato fruits at harvest. Obviously the extracts of leaves of Argall and Neem oil at all of their concentrations, Argall powder per hole, and fungicide scored the highest total weight of tomato with significant difference from the control at P = 0.05. They respectively yielded 15.5, 15.0, 13.0, 17.6, 15.1, 12.2, 19.8 and 18.6 Kg compared to control 9.6 Kg. It is noteworthy that the superiority of these treatments in yield performance over other treatments was also demonstrated in the experiment of season 2017-2018.

As for the impact of the remaining treatments on total of tomato fruits weight; they gave total of tomato fruits weight ranging from the lowest total one 7.6 to 10.0 Kg per treatment which was not significant at P = 0.05 compared to control 9.6 kg.

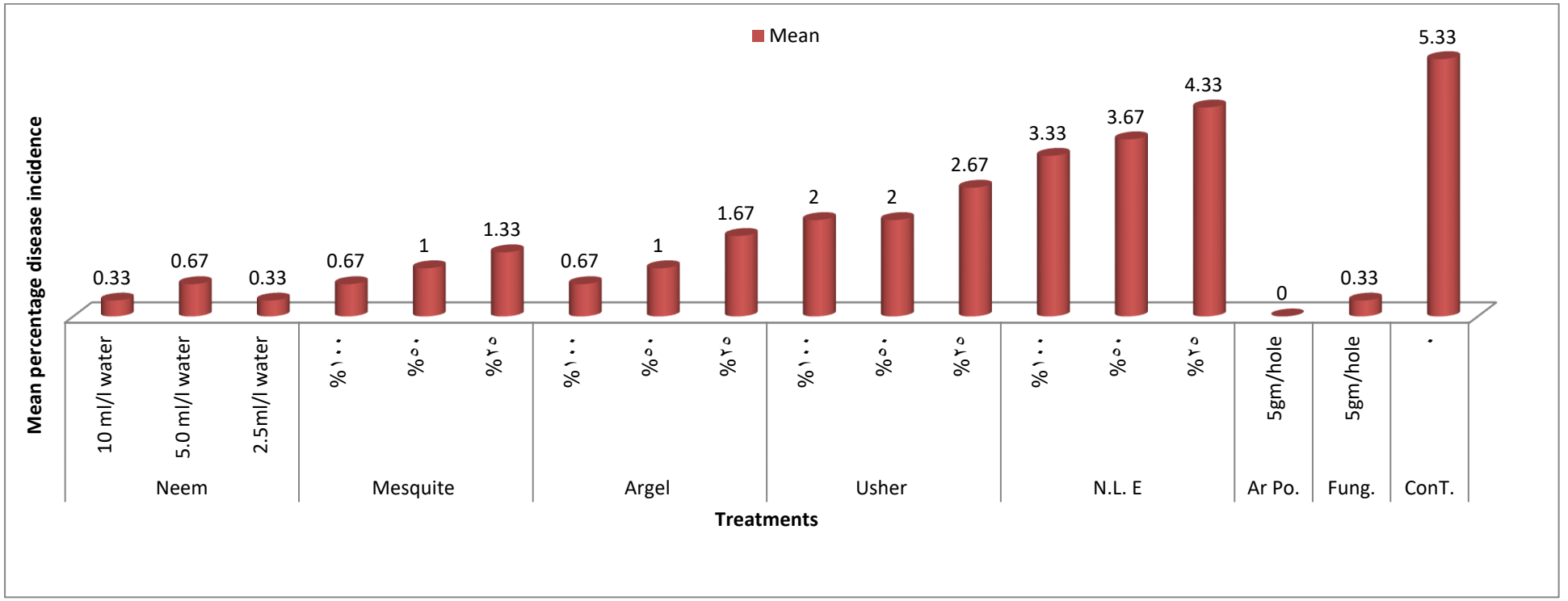

Fig. 13: The effect of different concentrations of natural products formulations and fungicide on incidence of early blight disease in fruits of tomato plant tested under natural infection (season, 2018 – 2019)

Discussion

The problem of the early blight disease control in tomato was seemed to be complicated by controversy around the geographical distribution and seasonal occurrence of the species of the genus Alternaria causing early blight in tomato (Giha, 1973: Pandey *et. al*., 2003 and Reni and Roeland, 2006).Based on the foregoing, the rational of this study focused on the identity of the pathogen, prevalence of the fungus, quantification of losses caused by early blight disease and assessment of different components for management of the disease in order to develop an integrated disease control strategy.

The results of the surveys conducted for three successive seasons (2017/18-2018/19- 2019/20), disclosed categorically, the common occurrence of *Alternaria alternata* that cause early blight disease on tomato crop in White Nile State. The data highlighted the higher incidence of early blight disease that noticed in the north of the State (15.11%), compared to southeast by south, center, south west of the state (7.78%, 6%, 0.89% respectively).

Moreover, the prevalence of the fungus was observed to be highly variable among the locations surveyed. This variability was found to be consistent during the three successive seasons. This took place in absence of link of interaction between locations. These differences in the infection levels of Early blight disease among the different surveyed locations could be attributed to various epidemiological factors prevailing in each location, such as the cropping pattern in the particular location, availability of alternative hosts, crop variety, age of the plant, level of resistance or susceptibility of cultivated hybrids, abundance of vector populations and activity, level of virulence of the pathogen and prevalence of favorable environmental conditions. This explanations could be fortified the striking variation in the percent of disease incidences between the Eastern location of South where the percent is high, and Western locations of South which of low percent

incidence. In fact, the majority of the cultivable land in Eastern location was traditionally cultivated is summer cereal crops e.g. sorghum, sesame, millet and sugarcane, leaving tomato winter crop as the only host for *Alternaria alternata*. The contrary was in the Western location were the land is rich with natural vegetation and varieties of winter crops which were cultivated with alternative host for the pathogen and hence minimize the load on tomato plants. However, these results were in line with Pandey *et, al*., (2003) who were investigated the early blight of tomato with respect to various parameters of disease epidemics. This is in addition to the traditional methods of disease control adopted in those rural areas.

However, the high percentage of disease incidence recorded at the North of the State could be probably due to intensive cultivation of tomato in that location which bordering the capital Khartoum. This is beside the irrational use of pesticides which could lead to surge of new strains of the pathogen resistant to them.

Nevertheless, the nature of damage and survival ability of the fungus which can survive in soil and plant debris in the absence of susceptible host (Delahat and Sterenson, 2004 , Agrios, 2005) render the management of Early Blight of tomato more difficult. In fact, the disease has been managed primarily by the use of resistant varieties (Jalali and Chand, 1992) but breakdown in resistance of these varieties due to evolution of virulent races of the pathogen have undermined their importance in recent years (Haware and Nene, 1982). In most cases chemical control methods are in practice. However, although the use of chemicals has helped increase of yields obtained (Ali, 1996), but the worldwide trend towards environmentally-safe methods of plant diseases control have initiated the exploration of safe alternate products. However, Plants-derived compounds (phytochemicals) have been attracting much interest as natural alternatives to synthetic compounds.

In response to this, biofungicide or natural products emerged as promising alternatives in an attempt to modify this condition where some alternative methods of control have been adopted. This included, biofungicide or natural products which emerged as alternatives, e.g. Biological agents, Neem, Garlic, and few other plants proved to inhibit Early Blight in tomato and other plants diseases (Schmutterer, 2002; Prasad and Naik, 2003; Adandonon *et al*., 2006 and Anjorin *et. al*., 2010)

Obviously, no single approach for Early blight disease control was proved to be effective and without drawback. Therefore, integrated management strategies are the only solution to maintain plant health.

Accordingly, this study investigated the minimum use of chemicals for checking the pathogen population, and safe alternate antimicrobial compounds of higher plants.

Generally, the results of the experiments set up to determine the effect of different concentrations of natural products formulations and fungicide on total weight of tomato fruits under natural infection in this study revealed that all treatments (natural products and fungicide) have had positive effect in controlling early blight disease of tomato plant but their effects were variable. The variable effects of natural products and Apron star 42 were also clearly observed on the different parts of the plants where the effects on the leaves were considerably higher as compared with those of the fruits. However, the most pronouncing effects on disease incidence were given by the natural products (Argall powder, Neem oil) and fungicide Seed star 42.

By far, the Argall powder at sowing time and Neem oil were the most predominant treatments among natural products. Results showed its highest significant antifungal activity against *Alternaria alternata* compared with other treatments this positive effect of the two components was observed on the different parts of the tomato crop. Similar results of Argel and Neem were

also reported by *Rous et al.,* (1980), Elhadi *et, al.,* (1994), Abdel Moniem E. *et, al.*, (2009). Likewise, results obtained, agreed that the treatment of tomato plants with Neem aqueous extracts reduced the percentage of Fusarium wilt disease incidence to the level of 25.5% and 27.8% after 6 weeks of infection respectively. Moreover, the promising effect of Neem products in controlling plant disease were also demonstrated by Schmutterer, (2002); Prasad and Naik, (2003); Adandonon *et, al*., (2006) and Anjorin *et. al*., (2010)who reported the inhibitory effect of Neem as biofungicide. Nevertheless, the results of testing natural products showed that there was increase in disease incidence and loss in yield with successive counts.

This could probably be due buildup of endemic with age of the plant or loss of efficacy resulting in loss in yield. These results were in line with the study of who reported that the early blight epidemics initially progress slowly but accelerate as plants mature, resulting in a typical sigmoid disease progress curve. Pandey, *et*. *al*., also mentioned in their research that, the disease curve is occasionally bimodal which could be due to the emergence of new healthy leaves after the first cycle of infection. Also yield losses up to 79% due to early blight damage were reported from Canada, India, USA, and Nigeria (Chaerani, and Voorrips, 2006).

However, based on the results of this study, the minor insignificant difference between, natural products treatments (Argel and Neem oil) in controlling early blight disease in tomato plant, and chemical control (fungicide), in addition to the public attitude and environmental concerns towards the use of synthetic pesticides as well as the development of early blight disease strains resistant to different fungicides could reduce the appeal of chemicals and lead to the search of alternatively safe control methods.

The study also demonstrated clearly the variation of resistance to early blight disease among the different tomato varieties tested. Obviously, the domestic

variety and strain B ones, showed highly significant resistance to the disease compared to the others where the disease incidence was relatively high. These results draw the attention towards the involvement of varietal resistance within the different management components in order to develop an integrated disease control strategy. This approach was also supported by Meitei, *et*, *al*., (2012.).

Conclusions

The study was carried out to assess the occurrence of early blight pathogenic fungi on tomato commercial field by surveying different tomato producing areas of the White Nile State and the possibilities of developing control measures

In Sudan, the tomato crop is considered as one of the major vegetable crops and widely used fresh in salad or in the processed forms as paste, ketchup, sauce and dry tomato slices. Moreover, the crop presents one of the main cash vegetable crops in almost every part of the Sudan during the winter and summer season and in close system farming. The crop is subject to a large number of pests and diseases from time of emergence to harvest. Among these; Early Blight disease caused by the fungus *Alternariaspp*. is one of the most common diseases of tomatoes in Sudan. Furthermore, the nature of damage and survival ability of the fungus which can survive in soil and plant debris in the absence of susceptible host render the management of Early Blight of tomato more difficult.

- The outcome of this study from the three successive years of surveys in White Nile State revealed that the epidemiological factors in this State imposed clear geographical distribution of early blight disease of tomato where the incidence of the disease was found relatively high in the North part of the State and in Eastern South part of the Nile as compared to the Western South and Centre of the State. These results are of real importance for tomato farming in the State as it could guide the investment policies in the White Nile State.
- Among all natural products and fungicide tested for controlling early blight disease in tomato, Argel and Neem products plus fungicide proved to be very effective in reducing the incidence of the disease.

- The results indicated that the minor insignificant difference between natural products treatments (Argel and Neem oil) in controlling early blight disease in tomato in addition to environmental concerns and human health towards the use of synthetic pesticides could reduce the appeal of chemicals and lead to the search of alternatively safe control methods.
- The results highlighted the importance of the varietal resistance as part of an integrated management approach to control early blight disease in tomato.
- High yield gains were obtained in this study upon application of low quantities of non-costly argel leaves to the soil of the tomato (5gm/hole). The yield increment is of practical value for tomato growers in the White Nile State. Beside the low cost. Argel and Neem oil are natural products devoid of safety hazards associated with synthetic pesticides and its use is a step towards organic farming. This study attests a practical potential of argel and Neem oil which might be extended to other horticultural crops. However, although encouraging results were obtained from application of argel to crops but still the product did not receive agronomic research attention; such a move might be needed in the near future.

Recommendations

Based on the promising results obtained from application of natural product in addition to their safety the following investigations were recommended:-

1- More research and investigation should be carried out to find the actual role of natural products on plants in relation to this early blight disease and other diseases.
2- More emphasis on by-products is required particularly (Neem, Argall, Usher, and Mesquite) and others to find out the correlation between natural products and diseases control.
3- The variability in early blight disease incidence of tomato due to epidemiological factors within White Nile State suggest detailed surveys in different States of the Sudan to determine safe area(s) for commercial production of crops.

4- Although encouraging results were obtained from application of argel to crops but still the product did not receive enough agronomic research attention, such a move might be needed in the near future.

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APPENDICES

Plate 1: Early Blight disease symptoms on infected tomato plant leaves

Plate 2: Healthy tomato leaves

Plate 3: Early Blight diseases symptoms on Tomato fruit

Plate 4: Healthy Tomato fruit

Plate 5: Weight Tomato fruit

Plate 6: Early Blight diseases symptoms on infected Tomato Plant

Plate 7: Healthy tomato plants

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LSD All-Pairwise Comparisons Test of TABLE by TREATMENT

TREAT	Mean	Homogeneous Groups
D	2.6667	A
K	2.0000	AB
N	1.3333	ABC
R	1.3333	ABC
F	1.0000	BC
A	0.6667	BC
Q	0.6667	BC
C	0.3333	C
E	0.3333	C
J	0.3333	C
L	0.3333	C
O	0.3333	C
B	0.0000	C
G	0.0000	C
H	0.0000	C
I	0.0000	C
M	0.0000	C
P	0.0000	C

Alpha 0.05 Standard Error for Comparison 0.7286
Critical T Value 2.028 Critical Value for Comparison 1.4777
There are 3 groups (A, B, etc.) in which the means
are not significantly different from one another.

Statistix 8.0 8/1/2022, 11:19:13 PM

LSD All-Pairwise Comparisons Test of WEG by TRET

TRET	Mean	Homogeneous Groups
F	5.2000	A
B	5.1000	A
A	4.3000	A
C	3.5250	AB
D	2.9750	AB
E	1.8750	B

Alpha 0.05 Standard Error for Comparison 1.1404
Critical T Value 2.101 Critical Value for Comparison 2.3960
There are 2 groups (A and B) in which the means
are not significantly different from one another.

Statistix 8.0 8/1/2022, 11:10:51 PM

LSD All-Pairwise Comparisons Test of TABLE12 by TREATMENTS

TRE	Mean	Homogeneous Groups
P	4.9500	A
Q	4.6500	AB
G	4.4000	AB
A	3.8750	AB
H	3.7750	AB
B	3.7500	AB
C	3.2500	AB
I	3.0500	AB
N	2.5000	AB
M	2.4500	AB
K	2.4000	AB
R	2.2500	AB
F	2.2250	AB
D	2.1500	AB
E	2.1500	AB
J	2.0500	B
O	1.9250	B
L	1.9000	B

Alpha 0.05 Standard Error for Comparison 1.4234 Critical T Value 2.005 Critical Value for Comparison 2.8537 There are 2 groups (A and B) in which the means are not significantly different from one another.

Statistix 8.0 8/1/2022, 11:10:32 PM

Completely Randomized AOV for TABLE12

Source	DF	SS	MS	F	P
TRE	17	69.050	4.06176	1.00	0.4702
Error	54	218.810	4.05204		
Total	71	287.860			

Grand Mean 2.9833 CV 7.47

	Chi-Sq	DF	P
Bartlett's Test of Equal Variances	4.19	17	0.9993
Cochran's Q	0.1244		
Largest Var / Smallest Var	4.5233		

Component of variance for between groups 0.00243
Effective cell size 4.0

TRE	Mean	TRE	Mean
A	3.8750	J	2.0500
B	3.7500	K	2.4000
C	3.2500	L	1.9000
D	2.1500	M	2.4500
E	2.1500	N	2.5000
F	2.2250	O	1.9250
G	4.4000	P	4.9500
H	3.7750	Q	4.6500
I	3.0500	R	2.2500

Observations per Mean 4
Standard Error of a Mean 1.0065
Std Error (Diff of 2 Means) 1.4234

LSD All-Pairwise Comparisons Test of TABLE10 by TREAT

TREAT	Mean	Homogeneous Groups
F	4.0000	A
J	4.0000	A
I	3.6667	AB
C	3.3333	AB
E	3.3333	AB
L	3.3333	AB
M	3.3333	AB
N	3.3333	AB
O	3.3333	AB
D	3.0000	ABC
K	3.0000	ABC
G	2.6667	ABC
H	2.6667	ABC
R	2.6667	ABC
A	2.3333	BC
B	1.6667	CD
P	0.6667	D
Q	0.3333	D

Alpha 0.05 Standard Error for Comparison 0.7115
Critical T Value 2.028 Critical Value for Comparison 1.4429
There are 4 groups (A, B, etc.) in which the means
are not significantly different from one another.

LSD All-Pairwise Comparisons Test of TABLE11 by TREAT

TREAT	Mean	Homogeneous Groups
R	5.3333	A
O	4.3333	AB
N	3.6667	ABC
M	3.3333	BCD
L	2.6667	BCDE
J	2.0000	CDEF
K	2.0000	CDEF
I	1.6667	DEFG
F	1.3333	EFG
E	1.0000	EFG
H	1.0000	EFG
B	0.6667	FG
D	0.6667	FG
G	0.6667	FG
A	0.3333	FG
C	0.3333	FG
Q	0.3333	FG
P	0.0000	G

Alpha 0.05 Standard Error for Comparison 0.9493
Critical T Value 2.028 Critical Value for Comparison 1.9253
There are 7 groups (A, B, etc.) in which the means
are not significantly different from one another.

LSD All-Pairwise Comparisons Test of TABLE2 by TREAT

TREAT	Mean	Homogeneous Groups
R	5.6667	A
D	4.0000	AB
N	3.3333	BC

F	2.3333	BCD
A	2.0000	BCDE
K	2.0000	BCDE
M	1.3333	CDE
E	1.0000	DE
H	1.0000	DE
J	1.0000	DE
Q	1.0000	DE
C	0.6667	DE
G	0.3333	DE
L	0.3333	DE
O	0.3333	DE
B	0.0000	E
I	0.0000	E
P	0.0000	E

Alpha 0.05 Standard Error for Comparison 1.0482
Critical T Value 2.028 Critical Value for Comparison 2.1259
There are 5 groups (A, B, etc.) in which the means
are not significantly different from one another.

LSD All-Pairwise Comparisons Test of TABLE3 by TREAT

TREAT	**Mean**	**Homogeneous Groups**
R	14.333	A
F	8.6667	B
N	7.6667	BC
O	6.0000	BCD
M	5.6667	BCD
D	5.0000	CDE
K	3.0000	DEF
E	2.3333	EF
A	2.0000	EF
H	2.0000	EF
I	2.0000	EF
C	1.0000	F
J	1.0000	F
L	1.0000	F
Q	1.0000	F
G	0.3333	F
B	0.0000	F
P	0.0000	F

Alpha 0.05 Standard Error for Comparison 1.5275
Critical T Value 2.028 Critical Value for Comparison 3.0980
There are 6 groups (A, B, etc.) in which the means
are not significantly different from one another.

LSD All-Pairwise Comparisons Test of TABLE4 by TREAT

TREAT	**Mean**	**Homogeneous Groups**
R	16.333	A
F	11.333	B
O	10.333	B
N	9.6667	BC
M	8.6667	BCD
D	6.0000	CDE
E	4.6667	DEF
K	3.0000	EFG
H	2.3333	EFG
I	2.3333	EFG
A	2.0000	EFG
J	2.0000	EFG

C	1.3333	FG
L	1.3333	FG
Q	1.0000	FG
G	0.3333	G
B	0.0000	G
P	0.0000	G

Alpha 0.05 Standard Error for Comparison 2.0698
Critical T Value 2.028 Critical Value for Comparison 4.1977
There are 7 groups (A, B, etc.) in which the means
are not significantly different from one another.

LSD All-Pairwise Comparisons Test of TABLE5 by TREAT

TREAT	Mean	Homogeneous Groups
O	5.0000	A
L	4.6667	A
N	4.3333	AB
M	4.0000	AB
F	3.6667	ABC
R	3.6667	ABC
J	3.3333	ABCD
K	3.0000	ABCDE
E	2.6667	ABCDE
D	2.3333	ABCDE
I	2.3333	ABCDE
C	1.6667	BCDE
H	1.6667	BCDE
A	1.0000	CDE
G	0.6667	DE
B	0.3333	E
P	0.3333	E
Q	0.3333	E

Alpha 0.05 Standard Error for Comparison 1.3699
Critical T Value 2.028 Critical Value for Comparison 2.7782
There are 5 groups (A, B, etc.) in which the means
are not significantly different from one another.

LSD All-Pairwise Comparisons Test of TABLE6 by TREAT

TREAT	Mean	Homogeneous Groups
P	12.000	A
B	11.100	A
Q	9.9333	A
G	9.2667	A
H	9.1000	A
C	8.5333	A
I	8.1667	A
R	7.0333	A
K	6.7000	A
A	6.6667	A
D	6.3333	A
N	5.7333	A
L	5.7000	A
E	5.4333	A
F	5.3667	A
M	5.3333	A
J	4.5333	A
O	4.4667	A

Alpha 0.05 Standard Error for Comparison 5.5797
Critical T Value 2.028 Critical Value for Comparison 11.316

There are no significant pairwise differences among the means.

LSD All-Pairwise Comparisons Test of TABLE7 by TREAT

TREAT	Mean	Homogeneous Groups
J	1.3333	A
E	1.0000	AB
I	1.0000	AB
L	1.0000	AB
M	1.0000	AB
R	1.0000	AB
B	0.6667	ABC
F	0.6667	ABC
A	0.3333	BC
C	0.3333	BC
D	0.3333	BC
G	0.3333	BC
K	0.3333	BC
N	0.3333	BC
H	0.0000	C
O	0.0000	C
P	0.0000	C
Q	0.0000	C

Alpha 0.05 Standard Error for Comparison 0.4714
Critical T Value 2.028 Critical Value for Comparison 0.9561
There are 3 groups (A, B, etc.) in which the means
are not significantly different from one another.

LSD All-Pairwise Comparisons Test of TABLE8 by TREAT

TREAT	Mean	Homogeneous Groups
E	2.0000	A
J	2.0000	A
L	2.0000	A
M	2.0000	A
C	1.6667	A
F	1.6667	A
K	1.6667	A
O	1.6667	A
R	1.6667	A
A	1.3333	AB
D	1.3333	AB
I	1.3333	AB
N	1.3333	AB
B	1.0000	ABC
G	1.0000	ABC
H	1.0000	ABC
P	0.3333	BC
Q	0.0000	C

Alpha 0.05 Standard Error for Comparison 0.6479
Critical T Value 2.028 Critical Value for Comparison 1.3140
There are 3 groups (A, B, etc.) in which the means
are not significantly different from one another.

LSD All-Pairwise Comparisons Test of TABLE9 by TREAT

TREAT	Mean	Homogeneous Groups
F	3.3333	A
E	3.0000	AB
M	3.0000	AB
C	2.6667	ABC

D	2.6667	ABC
L	2.6667	ABC
N	2.6667	ABC
O	2.6667	ABC
J	2.3333	ABC
K	2.3333	ABC
R	2.3333	ABC
A	2.0000	ABCD
G	1.6667	BCDE
I	1.6667	BCDE
B	1.3333	CDE
H	1.3333	CDE
P	0.6667	DE
Q	0.3333	E

Alpha 0.05 Standard Error for Comparison 0.6939
Critical T Value 2.028 Critical Value for Comparison 1.4073
There are 5 groups (A, B, etc.) in which the means
are not significantly different from one another.