REACTION OF SNAP BEAN (*Phaseolus vulgaris L.*) CULTIVARS TO NEMATODE (*Meloidogyne incognita)*

M.S.c Thesis

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Jimma Ethiopia

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By

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M.Sc. Thesis

Submitted to the School of Graduate Studies Jimma University College of Agriculture and Veterinary Medicine In Partial Fulfillment of the Requirements for the Degree of Master of Science in Industry Based Horticulture

Advisor: Dr. Beira H. Meressa

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APPROVAL SHEET

We, the undersigned member of the Board of Examiners of the final Open Defense by Mulugeta Nigussie have read and evaluated his thesis entitled "Reaction of Snap bean (*Phaseolus vulgaris L.)* Cultivars against nematode (*Meloidogyne incognita*)" and examined the candidate. This is therefore to certify that the thesis has been accepted in partial fulfilment of the requirements of Degree of Master of Science in Industry Based Horticulture.

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DEDICATION

Dedicated to my mother w/ro Belaynesh Tadesse for her kind support that brings my life into the right track.

STATMENT OF AUTHOR

I declare that this thesis is my work and all sources of materials used have been duly acknowledged and it's not submitted to any other institution anywhere for the award of any academic degree, diploma, or certificate. Brief quotations from this thesis are allowable without special permission provided that accurate acknowledgment of sources is made.

Name: Mulugeta Nigussie

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Place: Jimma University, Ethiopia.

Date of submission: August, 2019

BIOGRAPHICAL SKETCH

The author was born on July 11, 1981 G.C in Debrezeit. He attended elementary school at Fincha (Eastern Wolega) and high school at Ambo Comprehensive Senior Secondary School, respectively. He joined Jimma University Ambo College of Agriculture in 2001 and graduated in 2004 with a Diploma in General Agriculture. Then after graduation, he worked in flower companies, for Ethiopia Cuttings Plc (Syngenta) and Florensis Ethiopia Plc as a section head and Senior Agronomist respectively. He joined Haramaya University and graduated with BSc in Agricultural Economics in 2012. While the author is working he got the opportunity to join Jimma University on location M.Sc program in 2014 through self-financial support to pursue his M.Sc. study in industry based horticulture. He is married and has a child.

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ACRONYMS AND ABBREVIATIONS

ANOVA	Analysis of Variance
a.i	Active Ingredients
CRD	Complete Randomized Design
CSA	Central Statistical Agency
FNPP	Final Nematode Population per Plant
LSD	Least Significant Difference
MARC	Melkassa Agricultural Research Center
m.a.s.l	Meter above Sea Level
MOANR	Ministry of Agriculture and Natural Resource
PLC	Private Limited Company

Table of Contents

ABSTRACT

Snap beans are an economically important commodity grain legume produced in Ethiopia for food security and to incur foreign currency. However, the production of this crop particularly in the rift valley of Ethiopia is impaired by plant parasitic nematodes. The current study aimed at evaluating the reaction of snap bean cultivars those are in the pipe line BC4.4 and Plati, and cultivars those have been in production Dwarf bean Sony, Dwarf bean Faraday, Serengeti and Amy were included in the study. Pot experiment was carried out under greenhouse condition in complete randomized design (CRD) with five replications and one level of nematode population density. Each plant was inoculated with 1000 second stage juveniles (J2) previously maintained on tomato plants while uninfected plants served as a control. Eight weeks after infection, pod number, pod weight, root fresh weight, shoot fresh weight, number of nematodes per 100gram of soil, number of nematodes per plant and the multiplication rate of the nematode were taken. All the tested cultivars reacted differently and their yield and yield components also significantly ($P<0.05$) different between the cultivars. High root fresh weight (10.7g) recorded from the control of Dwarf Sony and shoot fresh weight (26.5g) performed by infected Dwarf Sony and both lower pod number (3) and pod weight (3.14g) recorded for cultivar BC4.4. The multiplication rate of nematode population for all evaluated cultivars in $Pf/Pi > 1$ found to be susceptible for *Meloidogyne incognita*. Even though the values of the reproduction factor were numerically different, all the cultivars grouped under excellent host range. Highest yield gained by Amy and Dwarf bean Faraday under high nematode reproduction while the least yield gained by cultivar BC4.4. So, using different initial population density and other additional available cultivars would help to determining threshold level of the nematode and it will be the future line of work.

Key words:-snap bean cultivars, host to *M.incognita*, nematode reproduction factor.

1. INTRODUCTION

Legumes of family Fabaceae has more than 20,000 species those are the most economically important crops next to cereals that comprises 27% of the crop production in the world (Smykal et al., 2015). Common bean (*Phaseolus vulgaris L.)* takes the larger part next to faba bean and field pea in Ethiopia (CSA, 2011). In supplementing smallholder farm family unit's pulses have larger parts and it also uses in providing cheaper sources of protein for deprived farmers (Ferris and Kaganzi, 2008).

All Snap bean, haricot bean, French bean, kidney bean, navy bean and black bean are synonyms and derived from common bean ancestors and also referred as common bean (CIAT, 2006 and NHB, 2015). Snap bean, the strain of common bean is one of the important vegetable crop that characterized by its succulent, flavorful pod and low fiber developed through breeding (Stephen, 1998; CIAT, 2006).

It's originated from Central and South America and domesticated nearly 6000BC and 5000 BC years ago in Peru and Mexico (Wortmann, 2006). In Ethiopia the production of snap beans was started in the early 1970s for the purpose of export to different market destinations (Desalegn et al., 1994). During 2015/16 cropping season the national average yield was 14 quintals per hectare from the estimated total area of production which was 356,299.89 hectares (MoANR, 2016).

As Abate (1985) reported many pests including nematodes are the production constraints for the production of cereals, pulses and oil crops in Ethiopia. However the degradation of soil fertility, erratic rainfall, pest pressure, poor agronomic practices and poor accessibility to good quality seed attributed to the low yield and quality of snap bean (Katungi et al., 2010). Since the agro-climatic conditions and cropping practices in the country are different, many biotic and abiotic yield limiting factors affects economic advantage from bean. Among the biotic constraints plant parasitic nematodes (root knot nematodes Meloidogyne spp.) are economically important and causes significant crop losses in temperate, subtropical and tropical climates (Perry et al., 2009).

Based on 37 life sustaining crops the global average estimated annual damage caused by nematodes, is US$ 358.24 billion, which is about12.6% (9-15%) of total crop production (Abd-Elgawad,2014; Martin & Fleming, 2014). In Ethiopia the availability (spread) of these plant parasitic nematodes surveyed in some parts of the country on tomato, pepper, onion, snap bean, cabbage, beetroot, carrot and potato by Mandefro and Mekete (2002). The survey included Western (Bako, Ambo and Guder), Southern (Butajira and Alaba) and Central (Koka, Meki, Ziway, Melgaewondo, Melkassa, Upper Awash, Melkasedi and Melkawerer) parts of Ethiopia. Then most frequently found and widely distributed species reported in the study was *M. incognita* (53.3%) followed by *M. ethiopica* (14.9%) and *M. javanica* (12.8%).

From practical observations there was patchy growth on different spots of beans field, gall on the root parts of the crops and yield losses in common beans and other vegetables in Ethio Vegfru plc. The company has been trying to alleviate the problem by using the same treatments (like the same nematicides, dosage and application frequencies) for different cultivars with no clear isolations of the cultivars those could be categorized under the susceptible or resistant. On the other part, even the amount of the yield loss was not clear except knowing the problem was root knot nematode (*M. incognita*).

As Meressa et al., (2016) reported, the survey done specifically at Koka Ethio Vegefru plc's different snap bean cultivars (Faraday, Boby bean, PeaA4 and peaB5) field and Ziway rose farms on 2016 confirmed that other than M. incognita some other species of nematodes also found.

The study mainly focused on the reactions of M. incognita on different cultivar of snap beans due the distribution and availability of different nematode species around the commercial farms and some other locations in the country had been surveyed by different authors.
Beside the readiness of Melkassa Agricultural Research Center (MARC) to release two snap bean cultivars those are in the pipeline and these cultivars have high chance to be produced in these particular areas by this farm or other commercial farms and also small holder farms.

Therefore, determining cultivars reaction to *M. incognita* might give some direction on which suitable cultivars will be chosen in the future and helps to reduce the cost of production (like reducing costs of nematicides). As the reaction of these snap bean cultivars those are in the pipelines and those have been in production to the root knot nematode have not been done in this way, that initiated to know the cultivars reaction to the infection of nematode *M. incognita*. And with these rational the study was conducted to address the following general and specific objectives

1.2 Objectives

General objective

- ✓ To determine effect of *M. incognita* on susceptibility and growth performance of snap bean

Specific objectives

- ✓ To evaluate the reaction of snap bean cultivars to *M. incognita*
- ✓ To evaluate the growth performance of snap bean cultivars under nematode infection.

2. LITERATURE REVIEW

2.1.History and distribution of bean

Snap bean (*P. vulgaris L.*) has been grown since a period of 7000 – 8000 years from a wild ancestral form and distributed in Latin America between northern Mexico and Northern Argentina (Gepts and Debouck, 1991). Snap bean widespread and cultivated many tropical, subtropical and temperate areas of Americas, Europe, Africa and Asia as a major food crop (Wortmann, 2006). According to Ibarra (1997) from the current total production (world) around 30%was from Mexico, central and South America and higher quantities grown in Asia and Africa, but the production in Ethiopia is concentrated mainly around the rift valley (dry and warmer part).

2.2. Biology and ecology of snap bean

Bean (*P. vulgaris*), is herbaceous annual and highly polymorphic warm season plant and its edible seed or unripe fruit consumed in a worldwide (Ecocrop, 2013). The plant has two types and the first type are erect herbaceous bushes in which its stem is slender, pubescent, highly branched and 20 to 60 cm high; and the other climbing vines (twining) is the second type high from 2 to 5 m and stems are pro length and rise towards the end (Smoliak *et al*.,1990; Ecocrop,2013).

The range of the snap bean life cycle depends on the determinate and indeterminate climbing types which are from 60-90 and 250-300 days respectively. For green pods, after 25-30 days of flowering the grain can be harvested (Wortmann, 2006; Ramirez, 2013). The crop largely self-pollinated and possibly crosspollination too (Ibarra *et al*., 1997) from the determinate bush and indeterminate climbing types of the growth habit the determinate type is the most predominant bean that grown in Africa (Buruchara, 2007).

It grows in a range of 200 to 600mm annual rain fall (IFPRI, 2010) and needs high water at pod filling stage (Raemaekers, 2001). The crop adapts at an altitudes ranged 1200 to 2000 m.a.s.l (Wortmann *et al*., 1998). For most cultivars of snap beans warmer temperature between 19-27^{0}c are suitable (Rice *et al*., 1990).

2.3.Botany of Snap bean

The annual leguminous plant, snap bean belongs to genus *Phaseolus*, species *vulgaris*, family leguminosae, subfamily papilionoideae, tribe phaseoleae and sub tribe phaseolina (Nonnecke, 1989).

2.4. Production and Economic importance of snap beans

More than 90% of snap beans produced, which is exported to within Africa and to other international markets are from east Africa (CIAT, 2006). In Ethiopia 80% of the snap bean produce is contributed by the large-scale state-owned farms, but quite small shares are from small holder out growers (Wiersinga and Jager, 2007; Okello *et al*., 2007). During the years from 2003-2013 the area and production of snap bean in Ethiopia increased by 76. 5 and 77.1 % respectively (FAOSTAT, 2013). In Ethiopian as a means of incurring foreign currency, snap bean has been the highly prioritized and the most important crop (Kay, 1979; Gezahegn and Dawit, 2006) and the production for both local and export market has also increased in the country (FAOSTAT, 2014).

Snap bean become the most important vegetables in local market and used in standard hotels and for festivities, to create variety of dishes that has been considered as an important protein supplement in the low income community of the country. As green vegetables snap bean provides protein, calories, vitamins and minerals (Lemma, 2003). The bush or pole plant type of diverse pod characters like bobby or fine bean of the crop are produced in the country for export purposes. The involvement of state horticultural enterprises, local and foreign private investors and farmers resulted for the gradual augmentation of snap bean production (Dessalegn, 2003) and covers 94% which was highest share of export potential among all vegetables (Dessalegn *et al.*, 2006 ; Dessalegn, 2011).

Pod characteristics such as sieve size, percent seed weight of total pod weight, pod fiber content, smoothness and straightness, color and flavor are other determinants the degree to which snap beans are accepted by consumers and processors. Since the demand of snap beans for local consumptions has been increasing, the non-exportable grades also will be sold for the local market (Lemma *et al.,* 2006).

2.5.Production constraints of snap beans

There are several biotic and abiotic yield limiting factors that inhibit the production of snap beans. The most common factors are: moisture stress, weeds, pests (disease and insect), soil fertility (Kidane, 1987) and lack of improved seeds (Ayele, 1991), but in Ethiopia erratic rainfall and poor agronomic practices also contributes for low yield (Katungi et al.,2010). Among biotic constraints plant parasitic nematodes (root knot nematodes *Meloidogyne spp*.) are economically important and causes significant crop losses in temperate, subtropical and tropical climates (Perry *et al*, 2009).

2.5.1. Moisture stress/drought stress

Both as a part of climate change and as a seasonal phenomenon, stress is the leading threat to the worlds food supply and in common bean its more severe than other abiotic factors that made it the main problem to livelihood of the farmers in marginal and unfavorable environment. In Africa where drought is already a problem like Ethiopia the crop suffers from warmer and un-successively drier (weather) because of the climate change. As several studies confirmed bean performance can be affected by drought stress; when the stress happened during flowering and post flowering the yield may reduce from 60-90%, 25% in number of pods per plant and 20.3 and11% in number of seeds per pod and seed size respectively (Manjeru *et al*., 2007; Khaghani, *et al*., 2008; Asfaw *et al*., 2014; Ambachew *et al*., 2015)

2.5.2. Weeds

In bean production if weeds are not controlled, substantially reduce the yield up to 90% (Tilahun, 1998; Rezene and Kedir, 2008; Mengesha *et al*, 2013). Plant spacing, planting pattern as well as weeding frequencies can suppress the weed growth and development that resulted less available spacing for the development as well as higher distribution of the seedling per unit area can lead the beans better competent for nutrients and moisture than the weeds (Page and Wilenberg, 2013).

2.5.3. Lack of improved seed

According to Fikadu (2007), one of the top problems of common bean is lack of improved varieties that causes for low yield and affects the production and productivity of the crop. Even though no snap bean cultivars were registered nationally in Ethiopia only few recommended cultivars were on the way to be released by MARC for the local farmers (Hussein, 2015). The Ethiopian farmers have a limited access to grow the cultivars those are introduced and private company owned cultivars from the expensiveness of the seed cost and limited efforts have been made for the development and identifying suitable varieties both for local and export market. Generally since the cultivars were developed under intensive production systems they need high nitrogen (Hussein, 2015).

2.5.4. Soil fertility

Acidic soil is one of the major factors that affects the production and productivity of snap beans in Ethiopia. Degradation of soil fertility attributes to the low yield and quality of snap bean. Since snap bean is a heavy feeder the management of soil fertility also limited in Ethiopia the application of fertilizer by small holder farmers mainly depends on the blanket recommendation without considering production area, soil type and fertility status. The production best suited to friable, deep and well drained soils high in organic matter (Abebe, 2007; KARI, 2007; Katungi *et al*., 2010 and MoANR, 2016)

2.6 Plant parasitic nematodes

Plant parasitic nematodes(*Meloidogyne spp.)* are the economically important and causes significant crop losses in temperate, subtropical and tropical climates (Ayele, 1991; Perry *et al*., 2009).

Nematodes are microscopic worms (round) which can be found in nearly all over the environment (Dropkin, 1980). Those worms feed on the root of many common garden crops and due to their feeding system they makes galls (swelling or “knots”) on the roots of infected plants named root knot nematodes which are scientifically from genus *Meloidogyne*. In 1885, the root knot nematode, *Meloidogyne sppecies* that cause damage on cucumber first observed by Barkeley and the Greek name *Meloidogyne* mean that an apple shaped female (Mitkowski and Abawi, 2003). In regions those have high temperature and environmental factors expose crops to high stress and interfere its resistance to nematode, the indo-parasitic root knot

nematode (*Meloidogyne spp.*) are important pathogens that causing yield losses(Pedrosa *et al*.,2000).

Out of several species of nematodes that damage bean *Meloidogyne spp*.(i.e *M.incognita, M.javanica, M.arenaria and M.hapla*) are responsible for big damage (Sikora *et al*., 2005) to reduce the quality of vegetables and around 50 to 80% yield losses per annum worldwide (Siddiqi, 2000; Moens *et al*., 2009). As different authors reported depending on the crop and locality the root knot nematodes causes for 10-100% yield loss and also with other pathogens it suppresses the nodulation in legumes (Jenkins and Taylor, 1967; Taha and Samie, 1993 and Trudgill *et al*., 2001). Specifically for snap and dry beans in addition to the quality, the yield loss can reach 90% (Shree and Schwartz, 2011).

M. incognita is the single most destructive plant parasitic nematode and its host ranges around 3000 plant species. *It is* also known as the southern root knot nematode which is one of the species out of the several *Meloidogyne spp.*. Tomato, pepper, okra, water melon, cantaloupe, onion, pumpkin squash, sweet potato, sweet corn, carrot, eggplant bean and pea are some of the crop species those may be severely damaged by root knot nematodes(Trudgill and Block, 2001; Ehlers *et al*., 2002).

2.7 Biology and ecology of nematodes

The life cycle contains egg, four juvenile stages and adult which starts when the only infective and moveable stage, j2 that move in to the growing media water phase searching a host crop and entered to the root . It penetrates the root through two ways just behind the root tip which are mechanical (stylet thrusts) and chemical means (cellulase and pectinase). It turns at meristem by migrating through intracellular space of the cortex and to stablish permanent feeding site it gets back to zone of cellular differentiation of the vascular cylinder. Then the giant cell formed by differentiation of five to seven cells induced by the establishment of the feeding sites which is adjacent to its head of Proto xylem and Proto phloem cells (Karsen and Moens, 2006).

Among the types of nematodes, the indo-parasitic nematodes live with in plant roots, root hairs and soil around plant roots. Even if one of this nematode feed inside the plants tissues, it may kill the crop and reduce its productivity. But the ecto-parasitic nematodes feed on a plants root surface without affecting its production (Dropkin, 1980).

2.8 Damage symptoms of plant parasitic nematodes

As the plant parasitic nematode mainly affects the root parts of the crop through formation of giant cells and causes for sever nutritional deficiency that may result declining of the plant. When the crop is attacked by nematodes the plant could show different symptoms mainly uneven plant growth, wilting during the hottest part of the day, dwarfism, reduction of the total shoot and root (total mass), lower number of reproductive buds, pods and seeds that can result drop the production (Ferraz and Monteiro,1995; Baida *et al* 2011). Since legumes produce nodules which are normally round, small and attached to outside of the roots, but the root knot nematode swelling (symptom) are different and found within the body of the root (www.infonet-biovision.org Jul, 2019)

2.9 Factors increasing incidences of nematodes

Plant-parasitic nematodes are soil-borne pathogens and their growths and pathogenicity are influenced by the soil conditions like soil texture, moisture, aeration and osmotic potential in field soils (Van Gundy, 1985). Since nematodes are motile animals, most of them can move no more than a meter through the soil in their lifetime. However, this does not mean that nematodes cannot rapidly spread from field to field. While farm equipment and even muddy shoes can rapidly disperse nematodes, floods and irrigation also disperses nematodes over long distances (Lambert and Bekal, 2002). Out of so many factors those causes for high incidence of root knot nematode, using susceptible cultivar, infected planting material, untreated water (irrigation), continuous use of the same field for 3-4years is the major reasons (Wahundeniya and Kurukularachchi, 1999).

2.10 Nematode control methods

Among different pest control methods, the highly important and eco-friendly method that helps in sustainable production is an integrated Pest Management (IPM). It is the selection, integration and implementation of available pest control method. As Food and Agriculture Organization (FAO) described, the system is utilizing all suitable techniques in a compatible manner to reduce pest populations and maintain them at levels below those causing economic injury (Smith RF and Reynolds HT 1966). The common and major components of IPM are cultural, physical, chemical and biological control methods. To control plant parasitic

nematodes there are different methods that used to reduce effectively, but cultural, chemical and biological are the three main methods.

2.10.1 Cultural methods

Cultural strategies are applied in both commercial and small-scale farmers in developing countries (Madulu *et al*., 1994) uses various integrated farming practices, like:

(i) Prevention of spreading plant-parasitic nematodes (nematode-free planting Material) is successful nematode controlling method in preventing the establishment of these parasites (Jensen, 1972; Bridge, 1996) Since infected planting material with plant-parasitic nematodes resulting in poor quality seedlings or tubers (Sikora & Femandez, 2005).

(ii) The use of direct, non-chemical, cultural and physical control methods:

Fallow method often used to reduce populations of plant-parasitic nematode and unless food residues available for a given period of time nematode populations would decline rapidly to levels below the damage threshold for crop (Ferraz & Brown, 2002).

Flooding in areas where water is abundant and fields are level (Johnson & Fassuliotis, 1984) flooding to a depth of 10 cm of water or more for several months can control the plant parasitic nematodes (Johnson & Fassuliotis, 1984), but this method is not economically feasible (sustainable subsistence-agriculture) as abundant water supply is not always available specially in resource-poor areas (Ferraz & Brown, 2002).

Trap crops controls particularly endo-parasitic nematodes (Keetch & Milne, 1982) which needs a highly susceptible, quick-growing crop to plant on a field and allow growing for a short time, then plowed or destroyed before the endo-parasitic nematodes become sedentary and before they are able to reproduce. The disadvantage of this method has additional expense and needs careful timing for destroying the crop otherwise the nematode populations might increase and reproduction occurs (Keetch & Milne, 1982; Johnson & Fassuliotis, 1984).

Crop rotation is very effective to limit nematode growth if non-host plant is rotated. Typically, a cropping system is devised that selects plants that nematodes can and cannot

grow on. When non host plant grown in alternate years, problematic nematode population will decreases dramatically in the years that the non-host is grown, and the damage threshold will be below the level. But the effectiveness of this method depends on the producers' choice if several different crops can be grown and when the host range of the problematic nematode is not broad or survives in the soil in acryptobiotic state for long periods of time (Lambert and Bekal, 2002).

(iii) Encouragement of naturally- occurring agents like bacteria, fungi, arthropods, protozoa and nematodes are abundant in most soils (Ferraz & Brown, 2002) those can be used as integrated controlling system in conducive environments (Webster, 1972) by modifying the ecological environment just to restrict its activities below damage threshold levels (Webster, 1972). Plant growth, nutrient and water holding capacity of the soil can be improved by soil amendments like cow dung , chicken manure and oilseed cake, etc. (Keetch & Milne, 1982). The microbial activity can be stimulated by higher organic matter content amendments that increase the activity of beneficial micro-organisms (i.e. fungi, bacteria, etc.) which reacts antagonistically to nematodes (Bridge, 1996a).

2.10.2. Biological methods

Biological control agents kill nematodes in controlled laboratory settings which have difficulties in implementing of growing large amounts of nematode pathogens in the field due to expense (Lambert and Bekal, 2002). Certain biocontrol agents (fungal) were found that grew on roots and provided physical barrier against the nematode and boosted plant growth by colonizing near the plant roots (Wickramaarachchi and Ranaweera, 2008). For example as Liu, (2007) reported Trichoderma sp., a fungus that showed antagonistic effect towards the nematodes and fungal pathogens (soil borne). For encouraging sustainable agriculture biocontrol strategies are environmentally safe and ecologically feasible option for plant protection and they also help beneficial microorganisms in the soil. The efficiency of the biocontrol depends on the other crops rotated, nematode species, plant host and their root exudates (Hallman et al., 2009).

2.10.3 Use of resistant cultivars

In addition to the cultural, chemical and biological methods using nematode-resistant plants is the most practical form (Lambert, and Bekal, 2002). This method is when the naturally nematode resistant gene crossed by breeders to cultivated plant species to improve the resistance of the crop to nematodes. Its advantage is that, it is cheap way for growers to control their nematode problems, but it takes years for screening of resistant plant varieties and needs more time for breeding the resistance traits into commercial varieties. Some species of nematodes are able to grow on resistant plants, for all cultivated species naturally nematode resistance source do not exist which is the other complication (Lambert, and Bekal, 2002).

2.10.4. Chemical methods

Since 1900's, nematicides have been used extensively (Ferraz & Brown, 2002) in high-value crops such as vegetables and legumes to reduce the number plant-parasitic nematodes (Netscher & Sikora, 1993). While using nematicides are declining (Ferraz & Brown, 2002) particularly for subsistence farming systems, in contrary environmentally-friendly and cost-effective nematode control methods are becoming increasingly important (Bridge, 1996). However, due to the maximum chemical residue level requirements set by the European markets application of pesticides affect the market of the produce (makes less marketable) (Kimani, 2002).

The two types of nematicides are soil fumigants (gas) and non-fumigants (liquid or solid). Soil fumigants are cost effective for most crops and becoming popular because they drastically reduce nematode population in the soil and do not rely on alternative host crops for rotation (Lambert and Bekal, 2002). Except 1,3 dichloropropene (Telone II), chloropicrin (tear gas), and dazomet (Basamid) most fumigant nematicides have been banned because of environmental toxins. Even though methyl bromide was largely discontinued in 2005 it had been providing excellent reduction of soil nematode populations and was multipurpose soil fumigant (Lambert and Bekal, 2002).

Fenamiphos (Nemacur) and aldicarb (Temik) are non-fumigant nematicides those can be applied in liquid or granular formulations that based upon the same kinds of active ingredients as many insecticides (i.e. nerve poisons). Comparing the fumigant and non-fumigant

nematicides, the non-fumigant reduces nematode populations and its effectiveness is not as consistent as that of fumigant (Lambert and Bekal, 2002).

Nematicides like Vyadate SL (a.i oxamyl), Rugby100g/l ME (a.i Cadusafos) and Sesamine EC (a,i Sesame oil) are the most commonly used in Ethiovegfru plc and other commercial farms. On the other hand Botanicals, plant-based pesticide chemicals have found favor as alternatives to pesticides in recent times (Marahatta et al.,2010). For example Neem, *Azadirachta indica*, is known to possess potential nematicidal compounds. Azadirachtin is the main nematotoxic compound in neem and all other nematotoxic compounds. These compounds are released through volatilization, exudation, leaching and decomposing of the plant parts (Nanjegowda et al., 1998). The leaf extracts of Garlic (*Allium sativum*) has been successfully used in laboratory condition to increase Tylenchulus semipenetrans mortality at high concentrations. Garlic has nematicidal effect and disrupts the mobility, food absorption and reproduction of nematodes. Its oil has been shown to offer significant protection against free living soil inhabiting nematodes (Block, 2010).

2.11. Types of Host-plant

To reduce and maintain the pest damage below threshold levels principles and practices of root-knot nematode management is essential that result in increasing and or maintaining the quantity as well as the quality of vegetable crops (Johnson & Fassuliotis, 1984). Host-plant resistance is one of the most popular way(strategy) in both commercial and subsistence farming systems by environmentally friendly and cost- effectiveness (Bridge, 1996a; Starr *et al*., 2002).

A susceptible host plant has a complex of features that are encouraging nematodes for reproduction and development which means that plants can not impede the growth and development of the nematode (Bos & Parlevliet, 1995). But, host-plant resistance has the capacity to resist nematode penetration, reproduction, establishment and spread of a nematode within a host (Bos & Parlevliet, 1995).

A cultivar that supports little nematode reproduction (< 10 % compared to a susceptible cultivar) is a highly resistant and a cultivar supports in intermediate level of reproduction relative to a Susceptible host, its moderately resistant (Hussey & Janssen, 2002).Tolerant a host crop that has the ability to withstand nematode infection and support nematode populations and crop yield which would otherwise severely damage susceptible plants (Oostenbrink, 1972; Roberts, 2002).

3. MATERIALS AND METHODS

3.1.Description of the study area

The study was conducted at Jimma University college of Agriculture and Veterinary Medicine from May 2018 to July 2018 under greenhouse condition.

3.2.Experimental materials and design

The experimental set up was in a completely randomized design (CRD) with five replications and six snap bean cultivars as well as one levels of nematode inoculations which is a population density of 1000 second stage juvenile(J2) .

In addition to personal observations at work place for the nematode infested plants and galls on the roots of some vegetable plants like tomato and beans the report of the survey collected by (Meress *et al*.,2016) for availability conformation of *M. incognita* on snap beans field of Ethio Vegfru plc was used.

Furthermore the report of (Mandefro and Mekete, 2002) also used as an input, for conforming *M. incognita* was highly distributed *(*53%) than other species around the rift valley and other locations.

Table 1 Description of snap bean cultivars used for the experiment

Cultivars under production	Cultivars in pipelines
1. Amy	**BC4.4**
2. **Dwarf bean Faraday**	**Plati**
3. **Dwarf bean Sony**	
4. **Serengeti**	

Amy has white flower, slender, round, white pods and medium length.

Dwarf bean Faraday has 13-14cm pod length, 50cm plant height and very uniform and very high yielding.

Dwarf bean Sony has 12-13 cm pod length, 50cm plant height and remarkable yields throughout the season.

Serengeti is a fine bean with beautiful pods,14-16 cm pod length, 6-8mm pod diameter, circular and straight pod shape and has medium erect bush plant habit that needs 55 days for maturity.

These four commercial snap bean seeds Dwarf bean Faraday lot no 160278-1Sep 2016, Dwarf bean Sony lot no 160711-p Sep 2016, Serengeti lot no16-12512 and Amy were treated by chlorpyrifos + thiram and chosen based on the market demand and its availability on the production of the private sectors like Ethio Vegfru plc. The two cultivars Plati and BC4.4 are in the pipelines, and chosen due to their reaction for *M. incognita* infection was not known.

3.3.Nematode source

M. incognita second stage juvenile was taken from the soil sample previously taken from infected snap bean field (Ethio Vegfru plc located at Koka which is in the North latitude of 8^0 26' 27.56" and altitude of 390 01' 54.45" East and 93 Km far from Addis Ababa). To get sufficient pure culture of *M. incognita* (j2) from a single egg mass it was maintained on tomato plants grown in the green house of Jimma University college of Agriculture and Veterinary of Medicine. Then second stage juveniles extracted from those infested tomato plants using the modified Baermann funnel technique (Hooper, 1986).

Second-stage juveniles(J2) is the infective stage that pierce the root to enter the vascular cylinder in order to induce the formation of a feeding site (Escobar *et al.*, 2015).

3.4.Plant growth condition and nematode inoculation

The crops were grown under the greenhouse condition on sterilized sand and field soil in 1:1 proportion and in 1lit plastic pot on a raised bench. Plants were watered daily.

The six cultivar seeds were seeded on May, 2018. After germination of five days, while one seedling thinned out the left grown for inoculation. On the tenth day of sowing three holes were prepared around the plant stem 4cm deep then all the six cultivars were individually inoculated with the suspension of second stage juvenile (J2) as a population density of 1000 (j2) per plant.

3.5. Sample collection and Nematode extraction

The data collection was carried at the maturity stage of the crop or the time reference of the crop to be harvested which was eight weeks after inoculation of *M. incognita (*J2). Plant roots were carefully uprooted and washed in running water to remove adhering soil.

Sample soil (infected) was collected and taken to laboratory and homogenized for nematode extraction. From the homogenized soil sample using measuring cup 100 ml was taken. The extraction setup was kept for 48 hrs. to ensure the migration of nematodes from soil to the water. After 48 hours nematode suspension was collected in a beaker and concentrated by passing through 20 µm stainless steel sieve.

Following the collection of nematodes, 1 ml nematode suspension was taken in to a counting chamber for enumeration. Nematode number obtained in 1 ml suspension then converted to the whole volume (100 ml). Reproduction factor was calculated following Osei (2010) as

Rf = Pf/Pi was used, where: Rf = reproduction factor, Pf=final population and Pi=the primary or initial nematode population used for inoculation .Then the multiplication rate from the total amount of the soil used as an indicator for host susceptibility.

Table-2 Reproduction factors and host plant category or classification according to Windham and William (1988)

Category	**Reproduction factor(Pf/Pi)**	**Host status**
1	<1	Resistant
2	1 to 5	Good host
3	>5	Excellent host/

Where: Pf=final nematode population, Pi= initial nematode population

Pod number per plant: counting the whole number of pods per plant since the low grades of snaps were also marketable there was no need to separate pod size and shape.

Pod weight (g): the weight of total pods per plant that was taken/ counted.

Shoot fresh weight (g): the weight of above ground part of the crop excluding the root part was taken.

Root fresh weight (g): the root part was washed gently with water and blotted with tissue paper and weighed using digital sensitive balance (EX-2000).

3.6.Statistical analysis

The collected data were subjected to analysis of variance (ANOVA) using SAS version 9.3 and means were separated using post hoc Tukey range test at the $P<0.05$ significance level. For each cultivar, the reproduction rate, RI=Pf/Pi, was calculated, where Pf= total number of nematodes extracted from the soil and Pi=initial population of nematodes inoculated per pot (Ferris and Noling, 1987; Osei *et al*.,2010).

4. Results and Discussion

4.1.Pod number and Pod weight

Analysis of variance (ANOVA) (table. 1) showed that there is a significant difference among snap beans cultivars infected by nematode *M. incognita.* Over all mean value of pod number 6.38 and pod weight 10.55g was recorded. Among infected bean cultivars the lowest value were recorded for pod number (3) and pod weight (3.14) in BC44 cultivar.

Table.1 Overall mean value for pod number, total shoot weight, root fresh weight, pod weight, final nematode population per plant, and final nematode population per 100 gram of soil and reproduction factor for nematode infected.

Variety	PN	TSFWT	TRFWT	PD WT	FNPPP	FNP100gr	RF
Faraday	9.8^{ab}	22.23^{ab}	8.314^{ab}	13.16a	90702^{a}	9070.2^{a}	90.7^{a}
Sony	8^{abc}	26.58^{a}	8.27^{ab}	13.16^{a}	12160^{b}	1216^{a}	12.6^{b}
Serengeti	8^{abc}	13.8^{abc}	4.56^{ab}	11.42^{a}	27800^{b}	2780^{b}	27.8^{b}
Amy	10^{a}	21.66^{ab}	7.156^{ab}	11.5^{a}	83990^{a}	8399^{a}	83.9^{a}
Plati	7^{bcd}	21.44^{ab}	4.346^{ab}	14.47^{a}	38400^{b}	3840.0^{b}	38.4^{b}
BC4.4	3^{ef}	5.792^{d}	1.956ab	3.14^{b}	68000^{a}	6800.0^{a}	68^{a}
Lsd	4.877	4.87	4.8	4.87	4.44	4.445	4.42
Cv(%)	21	21.47	35	23.5	27.2	27.2	27.2

Where; Mean values sharing the same letters under the same column showed significantly not different, CV=coefficient of variation, PN= Pod number, PDWT = Pod weight, TSFWT =total shoot fresh weight and TRFWT= total root fresh weight, Faraday=Dwarf Bean faraday, Sony= Dwarf Bean Sony

Highest pod number was recorded from nematode infected cultivar snap beans as shown Figure.1B While the lowest value for this variable was recorded from nematode uninfected cultivar of snap beans. As the mean values of the pod weight recorded revealed (table.1) variety Plati infected (14.47) were highest; significantly ($P<0.05$) different from cultivar BC44 infected (3.14g) which was lowest. The mean values of all the cultivars of snap beans

were statically different from the infected cultivar BC4.4, and except this BC44 infected the others mean values recorded for the pod weight were statistically not different.

Among infected bean cultivars the lowest value were recorded for pod number (3) and pod weight (3.14) for BC4.4 cultivar. While highest pod number (10) were recorded from infected Amy cultivar and the lower pod number (2.64) were recorded from un-infected (control) of Plati. Dwarf bean Faraday and Dwarf bean Sony infected were the second and third highest pod number while these two both were performed second in pod weight. As the pod numbers mean value of both cultivars Amy control (8.6) and Dwarf Sony control were statistically similar ; Except BC4.4 cultivar all the rest infected cultivars were statistically not different in pod weight recoded.

The reason for highest value of pod number and pod weight were recorded from the infected cultivars of snap beans comparing with the control could be due to that the infection stimulates the growth performance of the bean cultivars. Possibly for these differences may be that their difference in genetic makeup makes them to respond different performance for the infection of nematode. This finding was in agreement with Christie and Peacock, (1959) who found that the nematode life cycle highly different in respect to the relationships of individual species host to parasitized and physiological characteristics.
But this was not in agreement with the report Elliot and Bird (1985) who found that the root lesion nematode (penetrants) infection was not affect the relative growth rate of some beans.

Overall high yield and growth performance from nematode infected snap bean cultivars except BC4.4 at which lower value were recorded for all collected variables. The possible reason could be food source that the host crop provides for the nematode and the nematodes population densities ratio can be responsible to what degree the host responds for the infection. These finding was in agreement with Walker and Melin (1998b) who found that better plant growth where plant parasitic nematode population than nothing or low. Similarly, Barker and Olthof (1976) reported as plant stimulated to growth under certain condition of nematodes.

According to Olthof and Peterson (1974) who found that in some case different nematodes not only stimulating growth, but also can lead the yield to be increased. Even Pepper grow better at low level of *M. javanica* than the healthy crop, on peanut also better growth at moderate level of *M.javanica* and *M.incognita* nematodes than the healthy crop (Madamba *et al.*, 1965). Similarly *M. javanica* in watermelon didn't cause disease and instead of decreasing the plant growth parameters increased comparing with non-inoculated plants like vine 19 to 33% and dry top weight 40%. The inoculated plants grew faster than that of the control, this shows the nematode population density influences the plant growth and stimulation of shoot weight also detected at low initial population (Wallace, 1971).

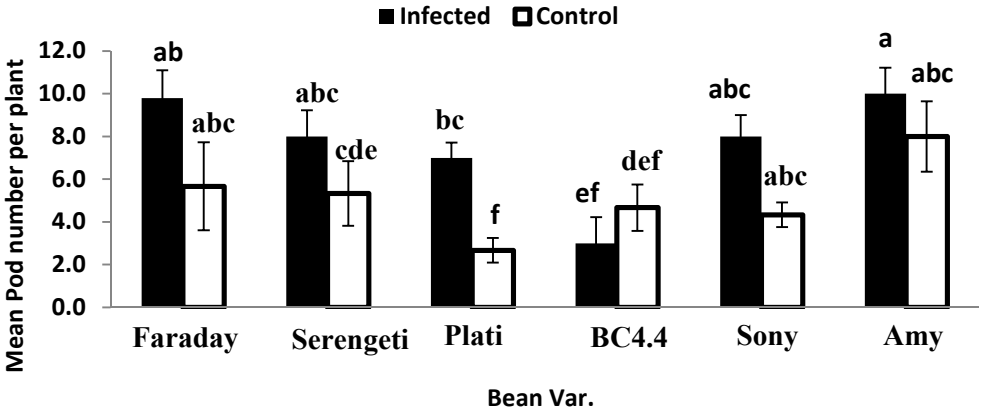

Figure.1A Pod number for infected and control snap bean cultivars.

Where; Faraday= Dwarf bean Faraday, Sony= Dwarf bean Sony

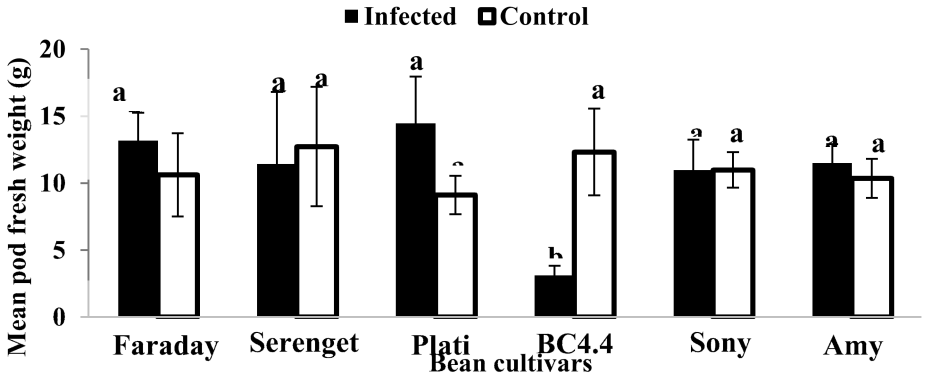

Figure.1B Pod weight of infected and control snap bean cultivars

Where Faraday= Dwarf bean Faraday, Sony= Dwarf bean Sony,

4.2.Shoot fresh weight and root fresh weight

Analysis of variance (ANOVA) (table.1) revealed that there is a significant ($P<0.05$) difference among snap beans cultivars infected by nematode *M. incognita. As* Fig.2A and 2B shown over all mean value of root fresh weight (6.59g) and shoot fresh weight (18.29g) were recorded respectively. From infected bean cultivars the lowest value were recorded for shoot fresh weight (5.792g) and root fresh weight of (1.956) for BC4.4 cultivar.

As the mean value (Figure 1) above revealed highest values for total fresh shoot weight recorded from the infected Dwarf bean Sony (26.58g) and the lowest was (5.792) for infected cultivar BC4.4. The values of total shoot weight for Plati infected (21.44g), Faraday infected (22.23g) and Amy infected (21.66) were statistically similar. While cultivars Dwarf bean Sony control (20.39), BC44 control (19.4g) and Serengeti infected (18.3g) were also statistically not different. Mean value of cultivars Dwarf bean Faraday control (16.49g) and Amy control (15.3g) were statistically not different and the mean value of cultivar Plati control (12.3g) was the second lowest value recorded which was statistically different from the others recorded value of shoot fresh weight.

From the mean values of root fresh weight recorded, higher value was cultivar Dwarf bean Sony control (10.712g) while the lower was BC4.4 (1.96g).

The highest mean value for root fresh weight (10.7g) and total shoot fresh weight (26.58g) were recorded from nematode non-infected (control) and infected Dwarf bean Sony respectively. While both lowest total root and shoot fresh weight mean values were recorded from nematode infected Bc4.4 cultivar. The BC4.4 value of lower mean root fresh weight could be from the infection of the nematode. Similar result was reported by Wallace, (1971) when there is high nematode population, the development of the root hair will be less and the reduction of water and mineral absorption and also translocation in the crop will be changed as a result root growth severely affected. Other report also found by Silva dos Santos (2012) that the mean root weight values of infected (nematode) cultivar was lighter than non-infected. On the other hand the higher value recorded from the infected cultivars could be that the nematodes damage on the host crop can be reduced by interaction of phyto-nematodes

with some organisms (Davis *et al*., 2006). As Nemic and Strubble, (1968) reported in examining the effect of root-knot nematode pathogenicity on shoot growth, no difference found between infected and non-infected plants after eight weeks in a greenhouse.

For all bean cultivars there were positive association in between nematode infection and total shoot weight as well as root fresh weight. For the variables the value recorded for respective bean cultivars revealed were different, even there were cultivars those showed increased yield performance these could be from the difference of the level of susceptibility or the difference of cultivars reaction to nematodes infection is different. These finding was in agreement with SYDENHAM, (1996) who found the resistance to *M. incognita* race 2 and *M. arenaria* race1in germplasm, which contain gene system1 and gene system 2.

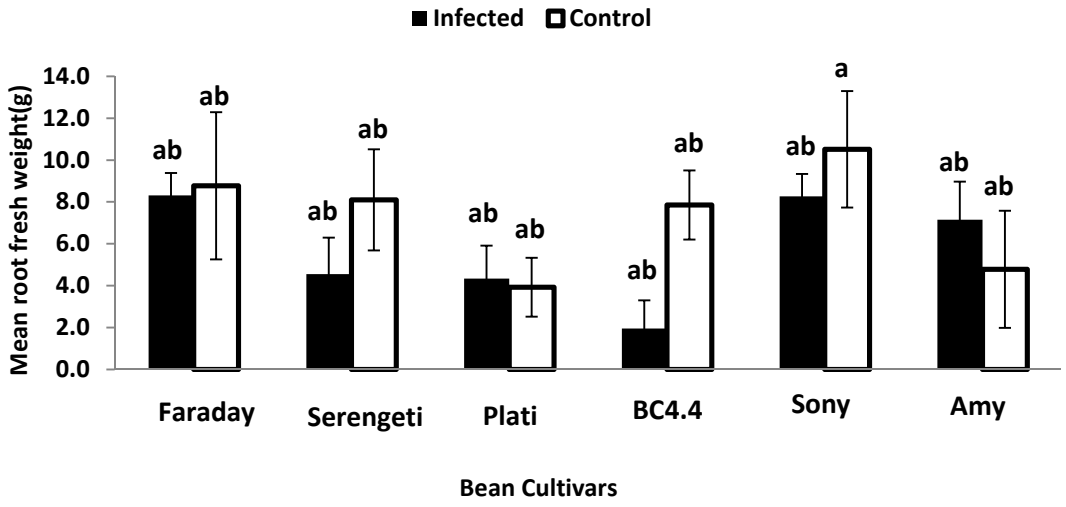

Figure 2A Root fresh weight of nematode infected and control snap bean cultivars

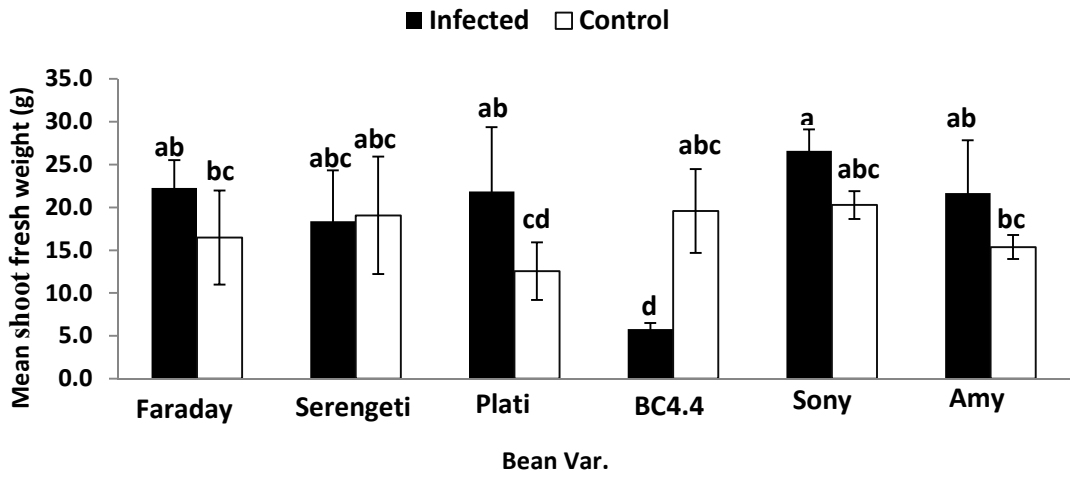

Figure 2B Shoot fresh weight of infected and control snap bean cultivars.
Where Faraday= Dwarf bean Faraday, Sony= Dwarf bean Sony

4.3. Nematode reproduction factor

As indicated on (table. 3) significantly higher values of nematode infected Dwarf bean Faraday recorded for variable FNPPP (90702), FNP100gr (9070.2), and Rf (90.7) while the lowest values recorded for these variables by Dwarf bean Sony. However for infected cultivars Plati and Serengeti lowest values for variables FNPPP, FNP100gr, and Rf were recorded.

As Nemic and Morrison (1972) reported the reason to get high values on some cultivars and low values in the other could be hypersensitivity reaction of the cultivars for the nematode, the delayed maturation of juveniles as well as lower in in number of cells (giant) and reducing in cortical hypertrophy, these mechanisms could involve for the host plant resistance to *Meloidogyne spp*.

As the (table 4) shown for the multiplication of nematode, the snap bean cultivars Dwarf bean Faraday, Amy and BC4.4 were significantly ($P<0.05$) different from cultivars Dwarf bean Sony, Plati and Serengeti. The values of reproduction factor for Dwarf bean Faraday was high

(90.7) and while the least was for Dwarf bean Sony (12.2). Followed by variety Amy, BC4.4, Plati, and Serengeti were 83.9, 68.0, 38.4 and 27.8 respectively.

As this finding revealed that except cultivar BC4.4 all the other infected cultivars Amy, Dwarf bean Faraday, Dwarf bean Sony and Plati were statistically similar with yield and some variables comparing with the non-inoculated. These could be as Baron (1939) reported the nematode juvenile penetration to the root was similar both for the susceptible and resistant cultivars and the same result was found Sydenham (1996) that for both the inoculated and non -inoculated plants shown the same measurements like leaf area and dry weight.

Table.3 Analysis of variance for final nematode population per pot, final nematode population per 100 gram of soil and reproduction factor of different snap bean cultivars infected by *M. incognita*

SV	**DF**	**FNPPP**	**FNP100gr**	**RF**
Rep	4	274,279,378ns	2,742,793.8ns	274.40ns
Cultivar	5	5,121,374,053**	51,213,740.5**	5,121.30**
Error	20	211,969,628	2,119,696.3	211.97
CV (%)		27	27	

Where: **=significantly different, ns= non-significant, SV = source of variation, CV =coefficient of variance, DF= degree of freedom, FNPPP = final nematode population per plant, FNP100gr = final nematode population per 100gram of soil, RF = reproduction factor, Rep= replication.

Susceptibility or resistance level of a crop is the capability of a crop that inhibit the multiplication or the reproduction of the nematodes. Karssen and Moens (2006) reported that allowing the juvenile to penetrate the root and reach maturity is the behavior of the highly susceptible crop or host. As different authors reported when the ratio of final population to initial population was higher (FP/IP>1) the crop or the host categorized under the good host

and poor host if the ratio was lower (FP/IP<1) and that could be influenced by the environmental conditions (Oostenbrink, 1966, Seinhorst, 1967, Cook and Evans, 1987; Turdgill, 1991).

Table. 4 Population dynamics of *M. incognita* on snap bean cultivars

Cultivars	**Pi**	**Pf**	**Rf=Pf/Pi**	**HS**
DBF	1000	90702	90.7	ES
Ser	1000	27800	27.8	ES
Plati	1000	38400	38.4	ES
BC4.4	1000	68000	68.0	ES
DBS	1000	12160	12.2	ES
Amy	1000	83990	83.9	ES

Where; Pi = initial nematode population per, Pf= Final nematode population, RF = reproduction factor, HS = Host status and ES = Excellent susceptible.

4.4.Correlation analysis

Pod numbers, were highly significant and positively correlated with pod weight, total shoot weight and pod weight were significant and positively correlated with total shoot weight .The possible reason could be as number of pod increase, proportionally pod weight increase. On the other hand total shoot weight were correlated with number of pods as well as pod weight these increases in total shoot weight as shown (Table 5).

While the number of pod and its weight increases, the biomass and or biological yield above ground also increase which can leads to positive correlation among these growth and yield components. This finding was in agreement with Mukeshimana *et al*., (2014) and Shakouri *et al*., (2015) who found that strong correlation existed for single plant seed weight and seed number as well as with its total volume.

Table 5 Correlations among the response variables of snap beans

Parameters*	PDN	PDW	TFSW	RFW	FNP100	FNPPP	RF
PDN	1						
PDW	0.47**	1					
TFSW	0.57**	0.79**	1				
RFW	0.09ns	0.23653ns	0.341ns	1			
FNP100gr	0.1982ns	-0.056ns	-0.21ns	0.1527ns	1		
FNPPP	0.19ns	-0.058ns	-0.21ns	0.15ns	1**	1	
RF	0.2ns	-0.057ns	-0.209ns	0.15ns	1**	1**	1

Where **= significant, ns= non-significant, PDN= Pod number, PDW =Pod weight, TFSW= Total fresh shoot weight, RFW=Root fresh weight, FNP100gr= Final nematode population per 100gram, FNPPP= Final nematode population per plant, RF =Reproduction factor

5. Summary and conclusion

All the infected cultivars allowed the nematode *M. incognita* for high reproduction and grouped to the excellent host crop. From all the infected cultivars Dwarf bean Faraday produced high pod number , high total shoot weight and total root fresh weight under high nematode reproduction.

Dwarf bean Sony produced high pod number, high total shoot weight and total root fresh weight under low nematode production (12). Cultivar Amy also allowed the reproduction of the nematode *M .incognita* at the highest level next to Dwarf bean Faraday, but high pod number even better than Dwarf bean Faraday.

Both cultivar Serengeti and Plati were allowed the reproduction of nematode *M. incognita* to the medium level comparing with the highest reproduction rate of the other cultivars. The total root fresh weights of these cultivars were highly affected by the nematode *M. incognita* next to cultivar BC4.4 which was severely affected than all the cultivars.

Cultivar BC4.4 was the least in pod number, pod fresh weight, total shoot fresh weight and root fresh weight than all the cultivars under the medium to high level of nematode reproduction.

Environmental conditions, previous cropping history, specific nematode species and race present, soil type, nematode distribution, prevailing nematode distribution pattern rate of multiplication crop has been and will be cultivated are the factors that inflicts the crop damage and yield loss or reduction (Brown, 1987, Schomaker and Bean, 2006; Khan, 2008)

The plant parasitic nematode *M. incognita* penetrates the root system and induce the formation of galls, obstructing the absorption of water and nutrients by the plants, but these symptoms are not always appear on all the crops because of the plant parasitic nematodes sometimes doesn't show symptoms and can reduce yield. In contrast the mere presence of plant parasitic nematodes in the soil does not mean (guarantee) that cause crop damage or yield loss unless the nematode population is higher than the damage threshold level for specific field (Brown, 1987; Schomaker and Been, 2006; Khan, 2008).

This initial susceptibility level information obtained in this study can be valuable for further breeding programs in order to find resistance and tolerance to nematodes subsequently its production. Further study should be repeated over different initial population, different sowing or planting season, environmental and soil conditions to screen and to study the damage and threshold level of the *M .incognita*.

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7. APPENDIXES

Table. 1 (ANOVA) Overall mean values for pod number, total shoot weight, root fresh weight, pod weight of control snap bean cultivars

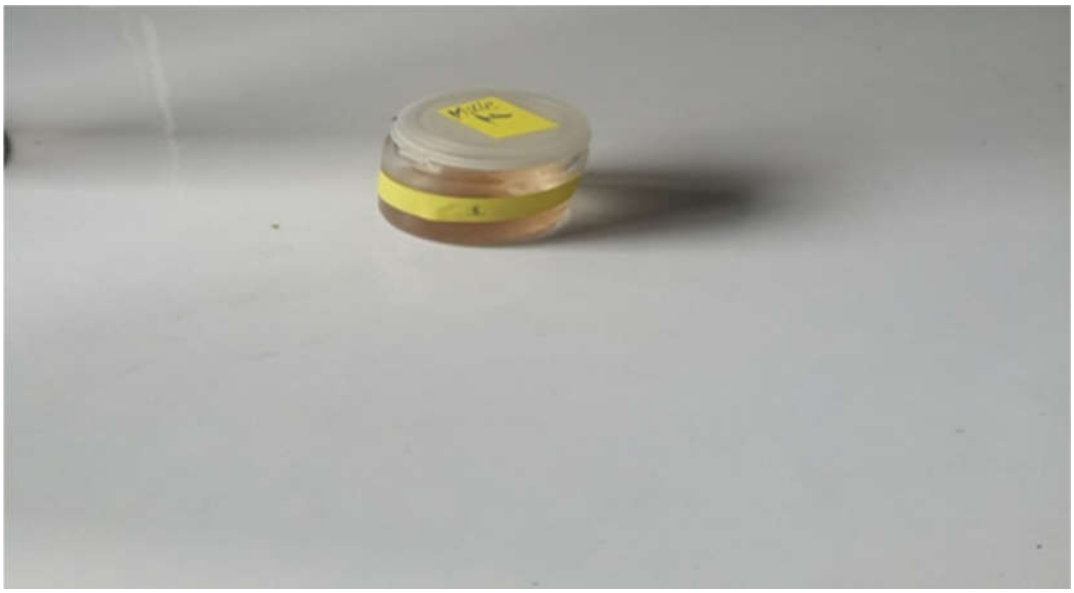
Cultivars	PDN	TSFWT	TRFWT	PDWT
Faraday	8.06^{abc}	16.49^{bc}	8.767^{ab}	10.6^{a}
DwarfSony	5.67^{abc}	20.39^{abc}	10.712^{a}	11.08^{a}
Serengeti	5.33^{cde}	19.1^{abc}	8.1^{ab}	12.7^{a}
Amy	8.06^{abc}	15.3^{bc}	4.896^{ab}	10.34^{a}
Plati	2.64^{f}	12.3^{cd}	4.03^{ab}	9.01^{a}
BC44	4.7^{def}	19.4^{abc}	8.06^{ab}	12.19^{a}
Lsd	4.877	4.87	4.8	4.87
Cv(%)	21	21.47	35	23.5

Where; Mean values sharing the same letters under the same column showed significantly not different, CV=coefficient of variation, PN= Pod number, PDWT = Pod weight, TSFWT =total shoot fresh weight and TRFWT= total root fresh weight

Table . 2 ANOVA for pod number, pod weight, total shoot and root fresh weight of Snap bean cultivars

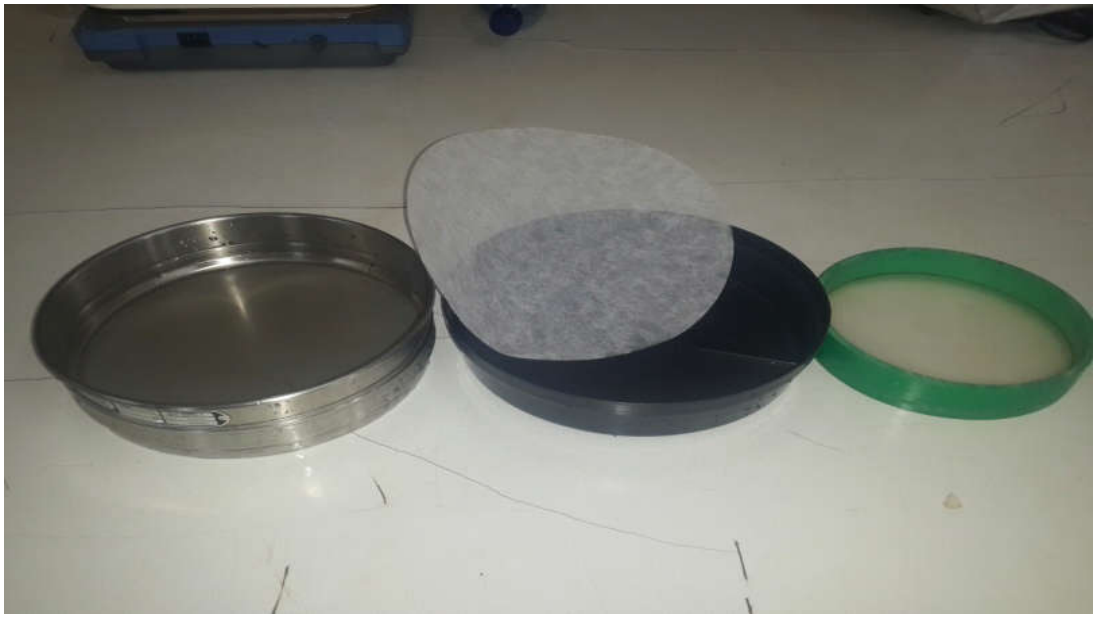
Source of variation	DF	PDN	PDWT (g)	TSFWT (g)	TRFWT (g)
Rep	4	1.8^{ns}	34.11ns	36.4^{ns}	2.0^{ns}
Cultivar	5	30.52**	40.0**	144.8**	32.8**
Error	20	1.79	6.55	15.4	5.6
CV (%)		21	23.5	21.4	35

Where **= significantly different, CV=coefficient of variation, ns =not significant, Rep= Replication, DF= Degree of freedom , CV=coefficient of variation, PN= Pod number, PDWT = Pod weight, TSFWT =total shoot fresh weight and TRFWT= total root fresh weight.

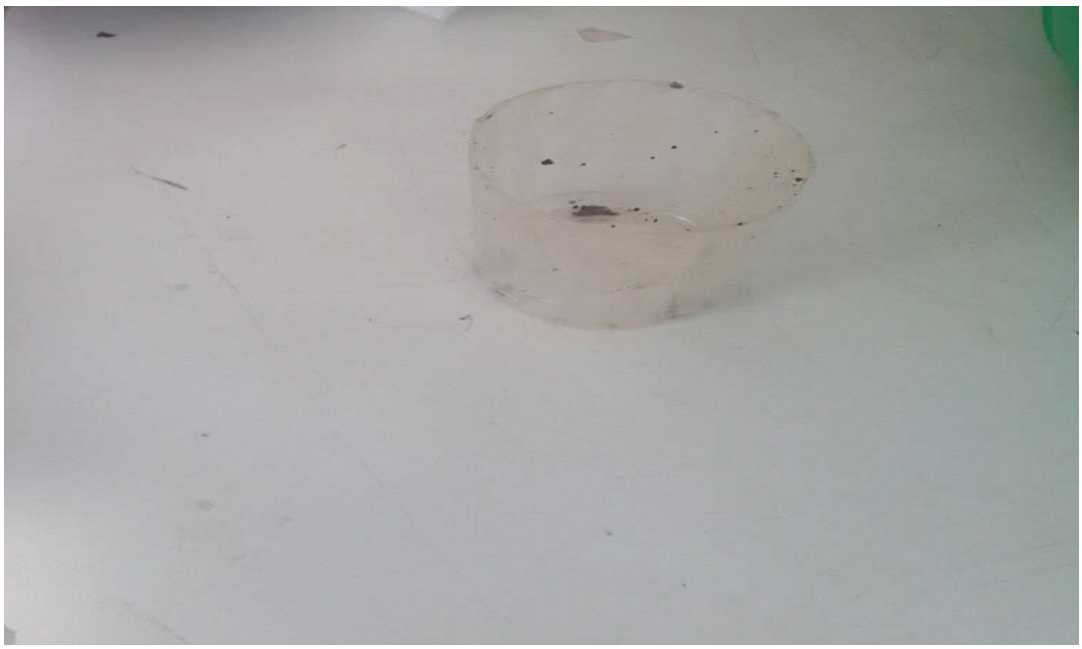


Appendix 1. Labeled *M. incognita* juvenile 2 ready to be counted

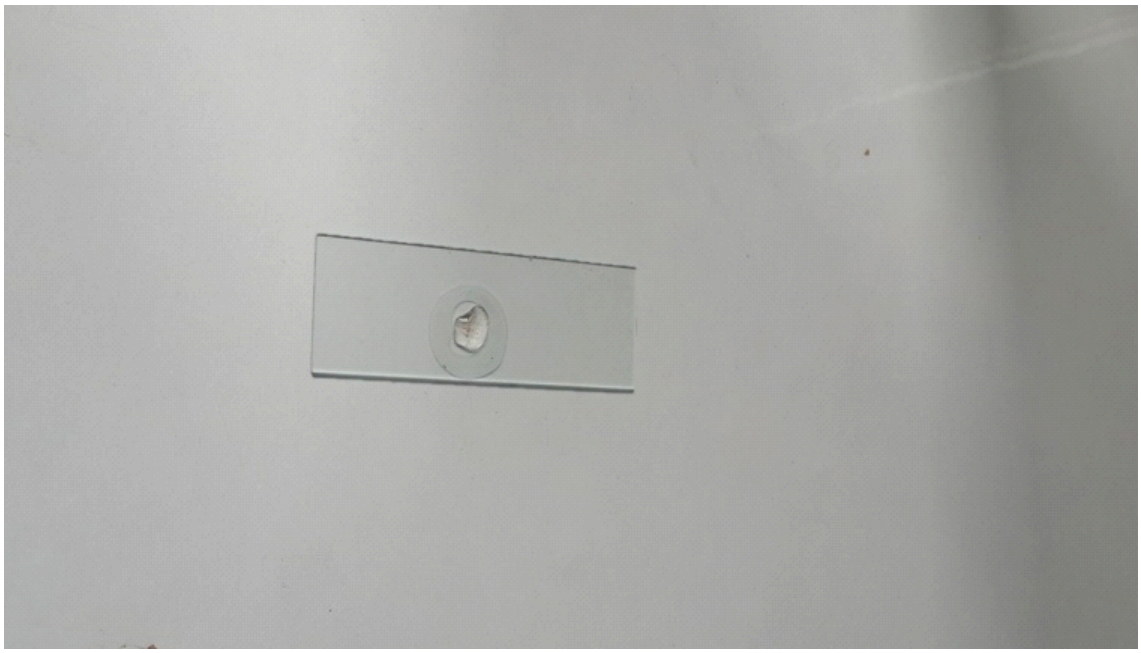
Appendix figure 2. 20µm sieve



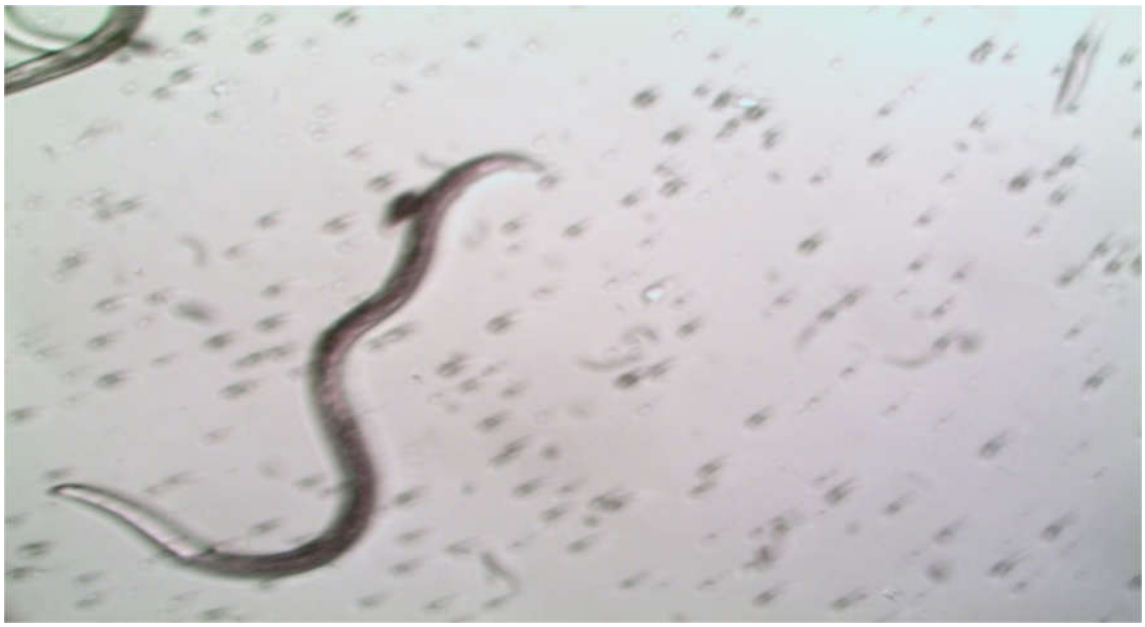
Appendix Figure. 3 20micro m sieve, petridish, white –paper



Appendix Figure 4. 100 ml measuring cup



Appendix Figure. 5 Slide (glass) used



Appendix 5 *M. incognita* j2

Appendix 6 Patchy growth (nematode damage symptom) in snap bean field