CHAPTER ONE

INTRODUCTION

Soybean *Glycine max* L. Merrill is regarded as one of the most nutritions among all crops. The grain; on dry weight basis, contains an average 21% oil and 40% protein. The soybean oil ranks first among all vegetable oils in the international trade. And its protein is considered as the most balanced among al non animal sources (Fehr, 1987). Hauck *et al* (1972) summarized soybean utilization into 42 different edible and industrial forms. They showed that these forms are derived from the whole bean products. In Sudan, soybean is regarded as a promising crop (Ageep and Khalifa, 1981). The country currently is importing vegetable oils for the local consumption, thus the introduction of a new oil crop will cut these imports and help in the self sufficiency of such vital commodity. Moreover, the protein balanced meal will be of great significance for the dairy and poultry industries.

Mycor" rhiza" literally means "fungus" - "root" and defines the mutually beneficial relationship between the plant root and fungus. These specialized fungi colonize plant roots and extend far into the soil. Mycorrhizal fungal filaments in the soil truly extensions of root systems and more effective in nutrient and water absorption than the roots themselves (Amaranthus, 2004). Kawai and Yamamot, (1986) reported that mycorrhizal plants absorbed more P, Ca and Mg and had higher P, Ca and Mg contents in their stems or leaves than non-mycorrhizal plants. Phosphorus concentration was also higher in the nodules of mycorrhizal plants. VA mycorrhizae increased nodule number, nodule weight and acetylene reduction activity of nodules. Concomitantly seed production and N content of leaves were enhanced. Mahadi (1993) reviewed the use of mycorrhizal fungi as biofertilzers in Sudan. He suggested that mycorrhizal fungi have a great potentiality to be used as biofertilzers in this country whose soil was found to be very poor in available phospours. Nodulation and plant growth were significantly enhanced by mycorrhiza and by p fertilization at Kenana (Mahadi, 2004). Mycorrhizal inoculation could remove the deficiency effect of P on plant growth, particularly in soil of low nutrient content (Elghandour *e t al*, 1996).

Diammonium phosphate (DAP) is one of world's most widely used phosphorus (P) fertilizer, it contains 18% N and 46% P2O5, Soybean is known to be highly nitrogen demanding crop, since the seeds are very rich in protein. The main source of nitrogen for the crop, so as to achieve high yield, is symbiotic N2-fixation. However, it is reported that applying moderate amounts of mineral nitrogen as starter dose was found to boost the nodule formation (Sigh and Saxena, 1972). For the development of the nodules, phosphorous plays a functional rote, and for their activity iron is very essential since it is involved in the synthesis of the nitrogenase enzyme (Elsheikh Hiatti, 1993). Both phosphorous and iron are characterized by very low availability to plants grown on alkaline clay soils. Accordingly, poor nodulation and N2 – fixation was proved to be very poor under such conditions (Hamid, 2005; and Rabih, 1999).

The objectives of the study were:

1- To determine the effects of rhizobium on growth and yield of soybean
2- 1- To determine the effects of mycorrhizal fungi on growth and yield of soybean

3- To investigate the effects of the interactions between rhizobium, mycorrhiza and DAP on growth and yield of soybean

CHAPTER TWO

REVIEW OF LITERATURE

2.1Botany

Soybean (*Glycine max* Merrill) is a member of family Fabaceae. The cultivated soybean has never been found in the wild. It is an annual plant generally exhibiting an erect, sparsely branched, bush type growth habit with pinnately trifoliolate leaves. The leaflets are broadly ovate, oval to elliptic- lanceolate. The purple or white flowers are borne on short axillary racemes on reduced peduncles. The pods are usually ovoid to subspherical. The seed coats range in color from light yellow, olive green, and brown to reddish black (Hymowitz and Singh, 1987).

2.2 Economic importance

The demand for soybean derives mainly from the oil and meal products, and to only a small extent from whole bean products. Soybean meal accounts for an even higher share in the high protein international trade. Soybean meal is used mainly in developed countries and some higher income developing countries. Soybean meal is a major protein component in livestock feed. Livestock and poultry feeds containing soybean meal as their major protein ingredient are used worldwide. The relative importance of soybean meal in the international market has increased over the years as seen by comparing percentage of soybean meal production. Soybean oil accounts 20 to 25% of total world fats and oil production and 30 to 35% of total edible vegetable oil production. (Smith and Huyser 1987).

2.3 Chemical composition

Martin and Leanard (1964) showed that the oil content of the seeds range from 14 to 24 percent or more while the protein may range from 30 to 50 percent. Soybeans with a low fat content are high in protein and vice versa. Soybean protein contents all the essential amino acids for animal feed and human food. Soybeans are a valuable source of calcium and phosphorus. Like other edible legume seeds, they are high in thiamin (vitamin B1). Hucheson *et al* (1936) reported that of the entire soybean plant 25.24 % is stems, 40.18 % leaves, and 34.37 % pods. The seed consists of cotyledons, 90 %; embryo, 2% ; and seed coat, 8%. The straw analyses show 2.8% protein; 38.5% carbohydrates; and 1% fat. Wolf *et al* (1971) demonstrated that commercial soybean seeds approximately 8% hull, 90% cotyledon, and 2% hypocotyl and plumule. The constituents of major interest – oil and protein make up about 60% of the bean, and about one third consists of carbohydrates including polysaccharides, stachyose (3.8%), raffinose (1.1%), and sucrose (5%). Phosphatides, sterols, ash, and other minor constituents are also present. Oil and protein contents depend on variety, soil fertility, and environmental conditions.

2.4 Main producer

The U.S., Brazil, Argentina, China and India are the world's largest soybean producers and represent more than 90% of global soybean production. The U.S. produced 75 million tons of soybeans in 2000, of which more than one-third was exported. Other leading producers are Brazil, Argentina, China Paraguay, and India (wekipidia web site, 2011).

According to Richard *et al* (1984) during the mid- to late- 1970s the leading importers of raw soybeans were Japan, Germany, Netherland, and Spain. Exports continue to be an important market for the crop but percentage varies from year to year depending upon world supply and demand. Brazil has become an important soybean producer and now competes with United State for world sales. Soybeans also are increasing in importance in Argentina.

2.5 Soybean in Sudan

Sudan has great potential for growing soybeans. The country is a large exporter of oil seeds. Soybeans are looked on by planners as a crop for the future when diversifying crop production for export. Soybean trials started as early as 1925 at Gezira Research Farm where a low yield was obtained. This low yield was attributed to lack of cultivars adaptable to the Sudan agro-ecological conditions. Lack of adaptable cultivars to the Sudan agro-ecological conditions has enormously contributed to the existing information gap on association of traits with seed yield (Tony *et al*, 2013) Kaltenbach, and Legros(1936) reported that soya growing was first introduced at the Gezira Station in 1931-1932. No native genotypes are grown; all have been introduced either from the United States, the

Union of South Africa or India. In general, the Indian types of soya grow better than the American or South African types. Faisal (1986) illustrated that research on soybean in the Sudan started more than fifty years ago with introduction of genotypes from USA, but there are no released cultivars adapted to Sudanese local conditions. Records of some introduced cultivars tested under both rain fed and irrigation conditions at Agadi experimental plots showed that seed yield produced ranged from 500 to 1000 kg/ha.

Soybean genotypes were tested at the Gezira Research Station, Wad Medani, between 1973 and 1977. Yields of over 2,700 kg/ha were obtained from the Hardee LS variety in 1978. (Ageep and Khalifa, 1981). According to of ARC, Sudan web site (2012) in Sudan, soybean is a new crop, struggling to be introduced in irrigated and rainfed farming to increase oil production, improve human nutrition, improve soil fertility and provide a high-protein feed source for livestock. Recently, the feed market has great potential in Sudan due to increase and expansion in the poultry industry. The ongoing soybean research program at the Agricultural Research Corporation (ARC) is currently focusing on developing improved soybeans cultivars and suitable agronomic practices. Soybean varieties introduced from USA and International Institute of Tropical Agriculture (IITA), Nigeria were tested at multi-locations and over many years. In irrigated farming, grain yield of 2.5 ton/ha was obtained in research and on-farm trials.

2.6 Adaptation

2.61Temperature

Martin *et al* (1964) showed that the climatic requirements for soybean are about the same as those for corn. Soybeans will withstand short period of drought after the plants are well established. In general, combination of high temperature and low precipitation are unfavorable. Soybean seed produced under high temperature conditions tends to be low in oil and oil quality. A wet season dose not seriously retard plant growth, but soybeans are sensitive to over irrigation. Chapman and Carter (1976) reported that soybeans are a warm – season crop. Soil temperature of 60° F or above favor rapid Germination and vigorous seedling growth, which is essential for successful competition with weeds and therefore for weed control. However, very warm midsummer temperatures (above 90° F or

32° C) reduced yield and oil quality. The minimum temperature for effective growth is about 50° F (10° C). Soybeans are produced in areas with a minimum frost – free period of 120 days and a mean summer temperature above 70° F (21° C) cultivars differ with respect to the minimum growing season required for maturation. Mike (2012) reported that very high soil temperatures (90° degrees F or 32° C) can cause decreased nodulation and nitrogen fixation to occur in soybeans.Samia (2013) reported that in Sudan soybean crop can successfully be grown in the summer season.

2.6.2 Rain fall requirements

Chapman and Carter (1976) illustrated that to produce optimum yields, soybean require between 20 and 30 inches (51-76 cm) of water – either natural precipitation, irrigation, or a combination of the two. Soybean crops are generally produced in regions where annual rainfall is adequate, but the availability of irrigation is insurance for producers. Soybeans are sensitive to both the amount and distribution of moisture. Germination is reduced by either excessive or insufficient soil moisture is also critical during flowering and immediately thereafter, during pod filling and during seed development.

2.6.3 Photoperiod

Soybean plants are the most sensitive of all crop plants to light duration (photoperiod) and are sensitive to light quantity. They are short- day plants but cultivars differ markedly with respect to the minimum dark period required to induce flowering. In addition to controlling the initiation of flowering, the photoperiod (which varies with changes in latitude) affects the development of soybean plants. Plants require high light intensity for vigorous growth. They suffer from shading and competition for light from taller- growing weeds; thus for highest yields, strict weed control is essential. To produce optimum yields, soybean requires between 20 and 30 inches (51-76 cm) of water – either natural precipitation, irrigation, or a combination of the two (Chapman and Carter (1976).

2.6.4 Soil

Soybean can be grown on a wide range of soils with pH ranging from 4.5 to 8.5. Soybean should not be planted in sandy, gravelly, or shallow soils to avoid drought stress. It should not be grown in waterlogged soils or soils with surfaces that can crust, as this will lead to poor seedling emergence (Dugje *et al* 2009).

2.7 Cultural practices

2.7 .1 Land preparation

The soil should be worked before planting by harrowing or disking lightly to destroy weeds. Soybeans cannot compete successfully with heavy infestations of weeds, and yields usually are reduced under such condition. Furthermore, thorough seedbed preparation make planting easier and makes soil condition more favorable for fertilizer and herbicide application, good germination, plant growth, and harvesting. (Richard *et al*, 1984). Dugje *et al* (2009) recommended that clear all vegetation before land preparation. The seedbed may be prepared manually with a hoe or animal-drawn implement or tractor. Well-prepared land ensures good germination and reduces weed infestation.

2.7 .2 Cultivar and Seeds

During the second half of the seventies, Kenana Research Station at Abu Naama released cultivar Davis to be grown under rainfed areas within the Central Clay Plain of the Sudan, and cultivar Semmis under irrigation (Khalifa, 1987).

2.7 .3 Sowing depth

Ideal sowing depth for soybeans is between 2.5 and 4 cm; shallow sowing is recommended for cool soils. Deeper sowing exposes the seedling to greater risk of damage from soil-borne pathogens (Upfold and Olechowski, 1994). Richard *et al* (1984) demonstrated that soybean should not planted more than one inch deep on clay and other heavy soils and two inches deep on light or sandy soils.

2.7 .4 Sowing time and rate

Faisal (1986) and Ibrahim (2012) recommended that the optimum sowing date for irrigated soybean in central Sudan is mid June. Dugje *et al*, (2009) showed that about 50–70 kg are required to obtain a population of 444,444 plants/ha for soybean varieties. Plant 3 to 4 seeds/hole at a spacing of 75 cm between rows and 10 cm between stands.

2.7 .5 Weed control

Soybean is a poor competitor with weeds when cool soil temperatures cause slow germination and growth, but soybean does compete effectively in warm soils when germination and growth are rapid. Soybean production requires good cultural practices. Prepare the seedbed prior to planting to kill germinating weeds. Management practices such as thorough seedbed preparation, adequate soil fertility, and choice of a well-adapted variety and use of good-quality seed all contribute to conditions of good competition with weeds (Zollinger 2011).

2.7 .6 Harvesting

Soybean matures within 3–4 months after planting and requires timely harvesting to check excessive yield losses. At maturity, the pod is straw colored. It is recommended that soybean be harvested when about 85% of the pods have turned brown for a non-shattering variety and 80% for shattering genotypes. (Dugje *et al*, 2009).

2.8 Nutrition

2.8.1 Mineral Nutrition

2.8.1.1 Nitrogen

Nitrogen is one of the most important nutrient elements affecting the yield of soybean. Soybean is known to be highly nitrogen demanding crop, since the end product is very rich in protein. The main source of nitrogen for the crop so as to achieve high yield, is the symbiotic N2-fixation. Nevertheless soybean can obtain a large portion of this nitrogen from atmosphere through its symbiotic relationship with rhizobium bacteria. (Sigh and Saxena, 1972). Nitrogen fixation begin fourteen days after planting only when plants were grown under optimum moisture and temperature conditions, thus a small amount of nitrogen at planting could be beneficial to early growth, given that nodules were not present until at least 9 days after soybean emergence (Boroomandan, *et al* 2009). Darrel *et al* (1980) stated that large amounts of nitrogen are not applied because of the ability to fix atmospheric nitrogen, but in some areas a complete starter fertilizer such as 6 -24– 24 containing a small quantity of nitrogen is used at planting. The nitrogen can stimulate early vegetative growth.

According to Pioneer web site (2011) soybean is a high-protein crop and requires a large quantity of nitrogen to synthesize amino acids and proteins. As a legume crop, however, soybeans supply most of their own N needs by fixation of atmospheric N2 into ammonia (NH3) a form that is readily available to the plant. Additional N is scavenged from the soil through organic matter cycling and rainfall deposition to supply N needs not met by N2 fixtion. Research has shown that if ammonium or nitrate is available to be absorbed from the soil when nodules are present, N-fixation will decrease proportionally. For this reason, N fertilization in soybeans rarely results in agronomic or economic yield increases when nodulation is normal, and is generally not recommended. However, research in some irrigated, high-yield environments has demonstrated that N applied during the pod or seed set stages of soybean may increase yield.

Young *et al* (1989) found that soybean yield response to N application (40 kg N/ ha) was not significant. Akbari *et al* (2002) showed that seed yield increased with increasing rates of N. Papastylianou (1986) found that as N fertilizer rate increased the number of nodules and their DW/plant declined. Grain yield of inoculated plants was not statistically different from that of plants receiving 180 kg N/ha applied as 3 equal split applications (at sowing, at 30 and at 60 days after sowing): grain yield for control, inoculated and fertilized plots were 3.7, 4.8 and 4.8 t/ha respectively. Control plots gave a relatively high yield due to high soil mineral N supply during the growing season. Campso *et al* (2001) demonstrated that nitrogen fertilizer decreased nodulation and did not increase soyabean grain yield. Novo *et al* (2001) showed that inoculation of the seeds did not provide the amount of nitrogen required to maximize soyabean production. Nitrogen fertilization reduced nodulation but increased nitrogen concentration in the seeds and the yield. Manral and Saxena (2003) showed that nitrogen rate and plant density influenced plant height of soybean significantly over control. As nitrogen rate increased, plant height increased

Christmas (2002) found that nitrogen rate affected branch number. Nitrogen rate of 90 kg /ha had the highest branch number (7.0/ plant), and control had the lowest (6.0 / plant). Olsen *et al* (1957) reported that nitrogen fertilizer applied to the surface of the soil in the field did not increase the yield of soybeans, and at higher levels reduced it, but may or may not significantly reduce soybean nodulation and plant height. Mehmet (2008) tested four different nitrogen doses (0, 30, 60 and 90 kg/ ha), and observed that increased nitrogen rate increased plant height, reduced pod height, harvest index and seed yield. Otherwise, the number of branches per plant, pod number per plant, yield per plant and 100 seed weight decreased as nitrogen rate increased. Anderson (2007) found that as nitrogen rate increased from 30 up to 90 kg/ ha, pod number per plant increased. Nitrogen produced 100 seed weight higher than the non-nitrogen application condition. Valinejad et al (2013) demonstrated that applying nitrogen as starter dose has the potential to increase soybean yield

Eman (2002) reported that protein content in leaves and seed, as well as the seed and straw yields increased with increasing N levels. Oil percentage decreased with increasing N. Osborne and Riedel (2006) found that oil concentration decreased as soybean N concentration increased due to N fertilization, applying N as starter dose has the potential to increase soybean yield and improve quality. Wang *et al* (2002) stated that soybeans seed protein content was initially low in the early stages of seed formation, increased steadily in the mid-stages and rapidly increased at the latter part of the seed formation stage. Nitrogen content in the plant was highest during the flowering stage and continuously decreased from the seed formation stage up to the ripening stage. Nitrogen distribution was influenced by dry matter and nitrogen content on a soil with no previous history of soybean cultivation. Mrkovacki (2008) studied the effects of different nitrogen rates (0, 30, 60, 90 kg N/ha) and reported that at flowering, the largest mass and length of the above-ground plant parts were recorded in the treatment with 60 kg N/ha, while the largest nodule number, mass and N content were obtained with 30 kg N/ha. Wood (1993) showed that nitrogen fertilization increased seed oil and protein concentrations

2.8.1.2 Phosphorous

Phosphorus is one of the essential elements in the plant growth. Phosphorus availability for soybean growth is frequently low because P reacts with iron, aluminum and calcium in soil to form insoluble phosphates. The use of phosphate fertilizer can produce substantial increases in soybean yields if soil test values for phosphorus are in the low and very low ranges (Rehm, 2001**)**. Maurya and Rathi (2000) found that the application of phosphorus 75 kg P/ ha enhanced the plant height.

Fahmina et al (2013) tested four level of P_2O_5 (viz. 0, 15, 30, 50 kg P_2O_5/ ha) and reported that application of different levels of phosphorus showed that plant height and number of branch/ plant increased significantly up to 50 kg P_2O_5/ ha. On the other hand, number of pods/ plant, thousand seed weight, grain yield and biological yield increased significantly up to 30 kg P/ha. Mahamood *et al.,* (2009) observed that phosphorus application increased plant height, leaf area, number of branches, crop growth rate, relative growth rate, number of pods per plant of soybean. Mahamood *et al* (2009) found that application of 30 kg P/ ha produced significantly higher number of branches than at 0 kg P/ ha**.** Jaidee1 *et al* (2013) showed that phosphorus application increased the root length density, shoot dry weight per plant, leaf area index, number of pods per plant, and 100-seed weight of soybean**.** Abdul-Aziz (2013) showed that phosphorus rate significantly increased both grain yield and 100 seed weight. Kazi (2002) found that a seed yield reduction of 10% was recorded when the P rate was reduced from 90 to 60 kg/ha. Majumdar (2001) stated that P significantly increased the grain and straw yields, pods per plant, 100-seed weight, oil and protein content and their yields and N, P and S uptake by soyabean.

Costa (2003) showed that application of P_2O_5 up to 80 kg/ha significantly increased the yield of soyabean. Rezende (2001) observed that the fresh matter, dry matter and hay yield and quality increased as phosphorus rates increased. Phosphorus rates did not alter soybean grain yield and other characteristics. Rehm *et al* (2000) reported that soybean yield increased resulting from the use of phosphate fertilizer varied from 268 to 940 kg /ha when soil test values were in the low and very low range, respectively. No yield increase was measured when the soil test level for P was in the high and very high range.

Thirumurugan (2002) found that the highest protein content due to P application was obtained with 60 kg/ha. P treatment had no significant effect on oil content. Sharma *et al* (2002) tested three levels of phosphorus (30, 60 and 90 kg/ha) and found that 60 kg/ha produced better yield, dry matter and yield components compared to 30 kg/ha. Maurya et al (2000) stated that application of P up to 90 kg/ha showed a positive effect on height of soyabean, dry weight, number of nodes per plant and other yield contributing. In Pakistan,Malik *et al* (2000) tested four levels of phosphorus(30, 60, 90 and 120 kg/ha) and The results revealed that phosphorus 90 kg per hectare with recommended application of N(30 kg per hectare) gave higher seed yield (1911.12 kg/ha) than control (1274.07kg).

Bulg *et al* (2003) found that Phosphorus deficiency decreased the whole plant fresh and dry mass, nodule weight, number and functioning. Under conditions of phosphorus oversupply the decrease in plant growth, nodulation and acetylene reduction was stronger. Phosphorus deficiency significantly affected all phosphorus metabolites. Contents of different phosphorus fractions were decreased under the conditions of phosphorus deficiency. Mahmoodi *et al* (2013) stated that the increased of phosphorous and sulfur fertilizer caused significant reduction in the protein concentration of the seed in comparison with control treatment. In contrast, reduction in the application of phosphorous and sulfur fertilizer, caused significant increase of the protein concentration of soybean seed. Tomar *et al*. (2004) observed that inoculation and phosphorus increased oil contents of soybean

2.8.2 Biological Nutrition

2.8.2.1 Mycorrhizal Symbiosis

According to Orlando, (2003) Vesicular-arbuscular mycorrhiza fungi are associated with the majority of the terrestrial plants. Their function ranges from stress alleviation to bioremediation in soils polluted with heavy metals. However, knowledge about this symbiosis is still limited. For the semi-arid tropics, there is a great possibility of using mycorrhizas as a biological tool for sustainable agriculture. Menge *et al.*, (1978) illustrated that the reduction in mycorrhizal infection under high phosphorus levels may be due to robust root cell walls which are less liable to infection.

Staehelin (1995) demonstrated that rhizobium enhances mycorrhizal colonization of soybean. Spokes (2015) reported that high phosphorus and low nitrogen had little influence an mycorrhizal infection, the combination of high phosphorus and high nitrogen decreased mycorrhizal infection. Jose et al (2009) stated that mycorrhiza increase relative growth rate of palm. Mahadi (2004) showed that nodulation and plant growth were significantly enhanced by mycorrhiza and by p fertilization at Kenana region. Hayman (1970) observed that high levels of nitrogen and phosphorus fertilizer decreased mycorrhizal infection of wheat at intermediate P levels, infection increased with increased nitrogen application. Faisal *et al* (2014) showed that mycorrhizal colonization was reduced by the addition of phosphorus and nitrogen fertilizers of faba bean and maize. A number of workers reported positive effects of mycorrhiza on plant growth (Habibzadeh and Abedi**,** 2014; Hoeksema *et al*, 2010; Shrestha *et al*, 2009) reported that mycorrhiza showed more plant height, number of branches and leaf per plant than control.

Tholkappian *et al* (2001) showed that pod number per plant increased in mycorrhizal soyabean than the non-mycorrhizal plants

Fredeen *et al* (1989) demonstrated that in plants grown in low-P soil, mycorrhizal infection resulted in higher foliar P concentration compared with non-mycorrhizal treatment, and greater shoot and nodule dry weight. In plants grown in high-P soil, mycorrhizas had no effect on shoot or root dry weight or P concentration. Photosynthetic rate was not affected by mycorrhizal infection or P treatment. El – Shourbagy *et al* (1989) found that inoculation increased soyabean shoot fresh weight and dry weight.

Karunaratne *et al* (1987) showed that urea-treated nodulated plants showed increased growth due to mycorrhizae. Urea-treated or ammonium-treated nonnodulated plants showed growth increases due to mycorrhiza. Mycorrhizal infection was greatest with urea nutrition, and the infection increased the tissue N content of these plants relative to nonmycorrhizal plants. Mobasser and Moradgholi (2012) reported that application of mycorrhiza had a significant effect on plant height, 100 seed weight and seed yield of soybean.

Bader- El – Din *et al* (1989) found that VA mycorrhizal inoculation significantly increased plant dry weight and phosphorus content of the plants, as did fertilization with superphosphate. The addition of rock phosphate in combination with VA mycorrhizal inoculation significantly increased plant dry weight and P uptake of the plants. Karunaratne (1989) stated that non-nodulating plants benefitted more from mycorrhizas than nodulating plants. High levels of P applied to soil or to leaves inhibited vesicular-arbuscular mycorrhizas. Mycorrhizal roots depleted culture solution P faster than non-mycorrhizal roots. Young *et al* (1989) found that inoculation with VAM fungi alone did not have a significant effect on nodulation and nitrogenase activity. Azcon *et al* (1987) stated that mycorrhizal inoculation increased plant growth and nutrition and improved plant utilization of tricalcium phosphate. However, the proportion of P in shoots derived from the fertilizer was lower in mycorrhizal plants. Abdel-Fattah (2002) showed that mycorrhizal inoculation significantly increased plant growth responses, P and N contents and total soluble protein of soyabean. Taiwo *et al* (2001) found that mycorrhizal inoculation led to an increase in percentage of roots infected by mycorrhiza as against the lower infection recorded in the uninoculated treatment. Arbuscular mycorrhiza inoculation and inorganic P application generally and significantly led to increase in the percent P in plant tissue.

Mala *et al* (2000) showed that mycorrhizal species showed high potential to enhance soyabean growth. Shirani *et al* (2000) found that application of VAM on soyabean caused a significant increase in potassium uptake efficiency. Tholkappian *et al* (2001) showed that the nitrogen content and pod number per plant increased in mycorrhizal soyabean than the non-mycorrhizal plants at 25% soil moisture Bressan *et al* (2001) demonstrated that mycorrhizal inoculation contributed substantially to the increase in shoot and grain dry weight, and N, P, K, Zn and Cu shoot contents. These responses varied with soil P levels and mycorrhizal species. Ezawa *et al* (2000) found that very high-P conditions may influence the species composition of the mycorrhizal fungi. Gabor and Bethlenfalvay (1988) showed significant differences in plant response to colonization were found in dry mass, leaf K, N and P concentrations, and in root/shoot, nodule/root, and root length/leaf area and root length/root mass ratios. Photosynthesis and nodule activity were higher in vesicular arbuscular mycorrhiza (VAM) treatments than the controls. Amiri et al (2013) found that mycorrhiza and rhizobium significantly increased oil content of soybean

2.8.2.2 Rhizobium

Although the atmosphere is 78% N_2, plants cannot use it directly. Plants can use only ammonium-N, or nitrate-N and some extent NH3-N. Soybean is a legume and should normally provide its N through a symbiotic relationship with N-fixing bacteria of the species *Bradyrhizobium japonicum*. In this symbiotic relationship, carbohydrates and minerals are supplied to the bacteria by the plant, and the bacteria transform nitrogen gas from the atmosphere into ammonia-N for use by the plant. (Franzen**,** 1999). Hutcheson *et al* (1936) reported that inoculated soybean as a rule contain more protein and produce more dry matter than uninoculated plants. Koivisto (1980) showed that rhizobial inoculation of seed with *Bradyrhizobium japonicum* is beneficial to nodulation, plant growth and nitrogen fixation on soils where soybeans have not been previously grown. Ibrahim *et al* (2011) stated that at Shambat inoculation significantly improved the dry weight of shoots and roots, nodulation, yield and yield components. Islam *et al* (1987) found that rhizobium inoculation gave greater number of nodules/ plant and plant dry matter content of soybean. Malik *et al* (2006) found that seed inoculation with *Rhizobium* significantly increased plant height, LAI, number of pods per plant, number of seeds per pod, 1 000 seed weight, total dry matter, seed yield and harvest index in soybean. Tairo (2013) showed that *Bradyrhizobium japonicum* inoculation significantly increased plant height and number of branches per plant of soybean. Togay *et al* (2008) reported that inoculation with *Rhizobium* significantly increased the plant height, first pod height, number of branches, pods and seeds per plant, grain and dry matter yield in chickpea.

Gulati (1989) illustrated that soybean nodulation ability, nodule dry weight, plant dry weight and seed yields increased and N status of soil improved, with increasing inoculant load during culture preparation, while number and dry weight of nodules was not correlated with seed yield or soil N status. Hernandez and Cuevas (2003) reported that inoculation increased the plant height of soybean. Faizah *et al* (1989) indicated that inoculation of soybean or N application increased dry matter yield and plant N content. Bader- El – Din *et al* (1989) demonstrated that inoculation with Rhizobium significantly increased plant dry weight and N content of soybean. Papastylianou (1986) reveal that nodules were only formed on inoculated plants and their number varied with inoculant. Aslam *et al* (2010) found that rhizobium increased 100 seed weight of soybean. Zaidi (2001) reported that inoculation with *B. japonicum* significantly increased plant growth, nodulation, nutrient uptake of N, P, Zn, Fe and Mn and yield of soybean over control. Akhtar and Siddiqui (2009) showed that rhizobium significantly increased 100 seed weight and yield over uninoculated soybean. Shirani *et al* (2000) reveal that inoculation with *B. japonicum* significantly increased the soyabean phosphorus uptake efficiency. Elsheikh and Elzidany, (1997) reported that inoculation has been reported to increase seed protein content of soybean. Mirzakhani, (2008) reported that seed inoculation with rhizobium bacteria increased oil percent.

2.8.3 Interaction between mineral and biological nutrition

Majid (2009) tested the effect of *Rhizobium* inoculation and phosphorous fertilization on nodulation, growth and yield characteristics of soybean grown in the presence of starter N fertilizer and concluded that *Rhizobium* inoculation and phosphorous increased number of nodules over un-inoculated control. Starter N fertilizer supply did not affect the number of nodules; however, combination of *Rhizobium* inoculation and phosphorous with 25 kg N ha/produced the highest number of nodules. *Rhizobium* inoculation, phosphorous fertilization and starter N fertilizer and their combinations increased shoot and root biomass and seed yield over control. Application of *Rhizobium* inoculation, phosphorous fertilization and their combinations increased oil and protein contents

Taiwo *et al* (2001) reveal that dual inoculation with VAM and Bradyrhizobium significantly increased nodule number of soybean when compared with control. Kaurtany *et al* (2005) reported that soybean seed yield were increased significantly by inoculation with mycorrhiza, the increase in seed yield for inoculated treatment over the non inoculated treatment was 181%. The inoculated treatment without phosphorus had higher seed yield than non inoculated treatment with high level of phosphorus. The phosphorus use efficiency was increased clearly by the inoculation with VAM fungus. Ali (2011) found that application of N and *Rhizobium* inoculation continued to have positive effect on growth indices and yield components of chickpea. Lower levels of nitrogen application and non-inoculated plants showed less growth indices including total dry matter (TDM), leaf area index (LAI) and relative growth rate (RGR) while the highest values of these indices were observed at the high levels of nitrogen application and inoculated plants. The greatest plant height, number of primary and secondary branches, number of pods per plant and number of grains per plant were obtained from the highest level of nitrogen fertilizer (100 kg urea /ha) and *Rhizobium* inoculation

Hicks *et al* (1987) showed that even though P reduced mycorrhizal infection from 25.4 to 5.2 vesicles/4 cm root length, there was no effect on the number of nodules/plant or on the resulting seed yields. Umale et al (2002) demonstrated that the maximum plant height was recorded in the of seeds inoculated with phosphate solubilizing bacteria (PSB) and 75 kg P2O5/ha. The highest number of branches, number of leaves and dry matter weight were also obtained with the same level of P. The grain yield per plant increased due to inoculation and higher level of P. Yanni *et al* (1987) found that Plants in non-inoculated plots with *Rhizobium japonicum* failed to nodulate, while nodule number was highest in inoculated plots receiving 72 kg N/ha one month after planting. Plots inoculated with *R. japonicum* and fertilized with 48, 96, 144 or 192 kg N/ha had higher yields than non-inoculated plots receiving corresponding nitrogen levels.

Bader- El – Din *et al* (1989) stated that the dual inoculation with both rhizobia and mycorrhiza produced more significant increases in plant dry weight, N and P content and seed yield of soyabean, than inoculation with either VA mycorrhiza or Rhizobium alone. Young *et al* (1989) found that mixed inoculation with Rhizobium and mycorrhiza affected yields in the range -8% to +145. These results suggest that single or mixed inoculation of rhizobia can increase soyabean grain yield and may replace N fertilizers in soybean/rice rotations. Landege *et al* (2002) reported that an increase in the rates of N and P with Rhizobium culture improved the quality parameters of soyabean (protein oil percentage and test weight of seed). Maximum number of root nodules and their dry weight was recorded under the same treatment. Haleem *et al* (2001) found that the addition of arbuscular mycorrhizal fungi and Rhizobium as biofertilzers different concentrations of Zn have a clear effect on Fe, Zn, Cu, Pb and Cd status in soil and their distribution in plants (roots, stem, leaves and seeds). Taiwo *et al* (2001) revealed that dual inoculation with VAM and Bradyrhizobium significantly increased nodule number of soybean when compared with control. Raverkar (2002) showed that improved VAM Mikhaeel *et al* (2000) colonization due to introduced bradyrhizobia enhanced the P uptake substantially in soybean, thus complementing the process of N2-fixation and dry biomass production. Dev *et al* (1997) showed that maximum colonization (94%) of soybean roots was observed at half the recommended dose of P (30 kg/ha). showed that dually inoculated plants with VA-mycorrhizal fungi and *B. japonicum* supported the highest increases in mycorrhizal colonization rates; shoot dry weight, nitrogen and phosphorus contents of plant shoots, nitrogen fixing capacity. Mehasen and Saeed (2005) reported that 100-seed weight, oil percentage and seed, biological and oil yields/fed were significantly increased by N fertilization and manuring only or combined with inoculation as compared with the control treatment. Elsheikh and Elzidany (1997) showed that nitrogen applications and rhizobium inoculation increased 100 seed weight of faba been. Bagyaraj *et al* (1979) stated that the number, dry weight and nitrogen content of the root nodules in plants inoculated with mycorrhiza and rhizobium were significantly higher compared to plants inoculated with only Rhizobium.

Alam *et al* (1989) revealed that inoculation + urea increased plant height, nodulation, number of pods/plant, 100-seed wt and gave seed yields of 1.43, 1.41, 1.59 t/ha, respectively, compared with 1.41 t without inoculation or urea. Chetti et al (1995) reported that nitrogen and rhizobium inoculation increased 100 seed weight of groundnut. Son (2006) showed that application of *Bradyrhizobium japonicum* and phosphate solubilizing bacteria (*Pseudomonas spp*.) can enhance the number of nodules, dry weight of nodules, yield components, grain yield, soil nutrient availability and nutrient uptake in soybean crop. Badran (2003) showed that higher oil yield was produced when inoculation and higher doses of phosphorus were applied. Bhattarai et al (2011) found that rhizobium inoculated plants showed increased dry biomass, greater root and shoot length, higher number of pods and maximum nodule formation of red kidney bean. Jahangir et al (2009) reported that oil content of soybean was influenced by increasing level of nitrogen and phosphorus. The percent of oil and yield of soybean were maximized by the application of 40kg N / ha + 30kg P_2O_5 / ha.

CHAPTER THREE

MATERIALS AND METHODS

3.1 Experimental site

A field experiment was conducted for two consecutive summer and winter seasons, (2010/11) and (2011/12), at Demonstration Farm of College of Agricultural Studies, Sudan University of Science and Technology at Shambat, to study the effect of rhizobium, mycorrhiza inoculation and diammonium phosphate (DAP) on growth, nodulation and yield of soybean *Glycine max* (L.) Merrill. Shambat is Located 23°35 E, longitude 15°31'N, and altitude 288m above sea level, within the semi dry climate described by Adam (2003) Appendix (1). The soil of the site is clay with pH 8.2 .Appendix (2) shows some of the soil physical and chemical propertied described by Adelgader (2010)

3.2Materials

3.2.1Plant material

Soybean *Glycine max* L. Merrill cultivar (E01) obtained form local market.

3.2.2 Bacterial strain:

Bradyrihzobium janponicum, strain USDA 122 was obtained from National Central for Researches, Khartoum – Sudan.

3.2.3 Mycorrhizal fungi (vesicular arbuscular mycorrhizal) VAM:

Inoculum of *Glomus fasciculatum* strain was obtained from Gaara Establishment for Import and Export, Egypt.

3.2.4 Fertilizer:

A commercial Diammonium Phosphate fertilizer DAP (18% N + 46% P2O5) was used.

3.3 Methods

3.3.1 The experimental design and treatments:

The experiment was arranged in a randomized complete block design with four replications. Each replication was divided into five plots to the following treatments:

1- C = control

2- R = Bradyrihzobium *janponicum* strain.

3- RM = strains of *Bradyrihzobium janponicum* + *Glomus fasciculatum*.

4-RMD1= strains of *Bradyrihzobium janponicum* + *Glomus fasciculatum* + 100 kg (DAP)/ ha.

5-RMD2= strains of *Bradyrihzobium janponicum* + *Glomus fasciculatum* + 150 kg (DAP)/ ha.

3.3.2 Land preparation:

The land was ploughed by disc plough, harrowed, leveled and ridged. Spacing between ridges was 70 cm, and then it was divided into plots 4x3.5 meters with four ridges each 3.5 meters long.

3.3.3 Husbandry:

The sowing dates were on 15 June in summer and on 25 December in winter for each season. Inter-row spacing was 5 cm with one plant per hole on the top of the ridge and gaps were filled by replanting after germination. Irrigation was applied at an interval of 7 and 10 days during summer and winter seasons, respectively. Weeding was done by hand whenever it was necessary to avoid weed competition

3.3.4 Rhizobium, mycorrhizal and fertilizer application:

Seeds were inoculated by *Bradyrihzobium janponicum* at the rate of 100 grams per kilogram of seeds for treatment (R) and by mycorrhizal *Glomus fasciculatum* at rate of 2.5 grams per hole at sowing (RM). Bradyrhizobium inocolum was mixed with gum Arabic and water to coat the seed in shade just before sowing. 100 and 150 kg/ha of diammonium phosphate (DAP) were added after 3 days from germination.

3.3.5 Mycorrhizal infection

Mycorrhizal infection was carried out according to Phillips and Hayman method (1970). The root samples were washed with tap water. They were put in 10% KOH at 90°C for two hours, and then washed by tap water four times and placed 2% HCL for five minutes, stained by cotton blue, and boiled for three minutes. Lactic acid was used and left to stand overnight to distain the host tissues.

3.4 Data Collection:

Five plants were taken randomly from the tow inner ridges, leaving one meter at each end of the plot to determine the following character, then the mean was recorded for each character.

3.4.1 Nodule number /plant: all nodules on roots samples were counted 38 days after germination.

3.4.2 Mycorrhizal infection%: The root hairs were cut to one cm pieces. Ten pieces of root hairs were put in slide and examined for the infection under the microscope. Two replicates were used to determine the percentage of mycorrhizal infection.

3.4.3 Vegetative Growth.

3.4.3.1 Relative growth rate (g/ m^2/day): samples were taken after 28 and 38 days from germination (DW1) and (DW2) respectively from area of 3.5m^2 and dried for 48 hours at 70°C then weighted. RGR was counted according to formula dw2 – dw2 /time x area

3.4.3.2 Plant height (cm): Height main stem was measured once before harvesting for the samples selected.

3.4.3.3 Number of branches/plant: Branches were counted once before harvesting in each sample.

3.4. 4 Yield components

3.4. 4 .1 Number of pods/plant: Pods in each plot were counted.

3.4. 4 .2 Number of seeds/pod: after pods were shelled then the seeds counted and seeds number per pods was calculated.

3.4. 4 .3 100- seed weight (g): 100 seed in each were selected randomly and weighed.

3.4.5 Protein and oil content %: 30 (g) of seeds from seed sample of each plot from two replicates was used for determining protein and oil according to method of AOAC (1970).

3.4.6 Grain yield /ha (kg):

Grain yield/ha (kg) was calculated by dividing the plot yield by plot area and multiplied by 10.000m².

3.5 Statistic analysis:

Data obtained was subjected to statistical analysis system “MSTAT-C software. Analysis of variance was carried out for treatments and means were tested and separated using Duncan Multiple Range Test (DMRT) (Steel et *al*. (1997).

CHAPTER FOUR

RESULTS

4.1 Summer seasons (2010) and (2011)

4.1 .1 Nodule number/plant

In the present investigation as shown in table (1) rhizobium inoculum (R) significantly increased nodule number/ plant compared to control(C) in both summers. Combination of rhizobium and mycorrhiza (RM) produced significantly the highest nodule number/plant over all treatments. The interaction between rhizobium, mycorrhizal, 100 and 150 kg/ha of diammonium phosphate, (RMD1) and (RMD2) respectively, increased number of nodules per plant over control in both seasons. Nodulation failed in control treatment in the first season. (Table 1).

4.1.2 Mycorrhizal infection%

Rhizobium inoculum (R) significantly increased mycorrhizal infection% over control (C) in both seasons. The combination of rhizobium and mycorrhiza (RM) produced significantly higher mycorrhizal infection% in comparison to other treatments in both seasons. Mycorrhizal infection% increased significantly when seeds inoculated with rhizobium inoculum + mycorrhiza + 100 and 150 kg/ha DAP (RMD1) and (RMD2) respectively as compared to control.

Table (1). Effect of rhizobium and mycorrhizal inoculation and diammonium phosphate (DAP) on nodulation number/plant and mycorrhizal infection% of soybean. (Summers season 2010/11 – 2011/12)

Treatments	Nodulation number/plant		Mycorrhizal infection%	
	(2010/11)	(2011/12)	(2010/11)	(2011/12)
C	0.0 [e]	0.8 [e]	17.0 [e]	16.5 [e]
R	8.3 [c]	9.0 [c]	26.5 [d]	23.0 [d]
RM	19.3 [a]	24.5 [a]	53.5 [a]	64.5 [a]
RMD1	11.3 [b]	18.0 [b]	49.0 [b]	51.0 [b]
RMD2	5.3 [d]	3.8 [d]	45.0 [c]	45.0 [c]
Means	8.9	11.2	38.2	40.0
S.E=	±0.6	±0.8	±1.2	±0.7
C.V%	13.0	14.9	0.4	2.0

Means with same letter in the same column are not significantly different at1% level, according Duncan Multiple Range Test.

4.1.3 Vegetative Growth.

4.1.3.1 Relative growth rate (g/m²/day)

Rhizobium inoculum (R) significantly increased relative growth rate over control (C), in the summer season of (2011). The combination of rhizobium and mycorrhiza (RM) significantly increased relative growth rate in both seasons. Combination of rhizobium + mycorrhiza + 150 kg/ha DAP (RMD2) registered significantly higher relative growth rate (RGR) as compared to control. The highest relative growth rate was recorded in the combination of rhizobium + mycorrhiza + 100 kg/ha DAP (RMD1). (Table 2)

4.1.3.2 Plant height (cm)

Rhizobium inoculum (R) significantly increased plant height over control (C). The combination of rhizobium inoculum and mycorrhizal (RM) significantly increased plant height as compared to control. The interaction between rhizobium + mycorrhiza + 100 kg/ha DAP (RMD1) gave increased plant height over (RM) but the differences were not significant. The maximum plant height was recorded by combination of rhizobium with mycorrhiza and 150kg/ha DAP (RMD2).(Table 2).

4.1.3.3 Branches number/ plant

Rhizobium inoculum (R) significantly increased branches number/plant over control (C). The combination of rhizobium inoculum and mycorrhizal (RM) significantly increased branches number/plant as compared to control. Although the combination of rhizobium + VAM mycorrhiza + 100 kg/ha DAP (RMD1) gave more branches number/plant than (RM) but it was statically at par. The highest branches number/plant was recorded by combination of rhizobium with mycorrhiza and 150kg/ha DAP (RMD2). (Table 2).

Table (2) Effect of rhizobium and mycorrhizal inoculation and diammonium phosphate (DAP) on vegetative growth of soybean. (Summers season 2010/11 – 2011/12).

Treatments	RGR		Plant height (cm)		Branches number/plant	
	(2010/11)	(2011/12)	(2010/11)	(2011/12)	(2010/11)	(2011/12)
C	0.3 [c]	0.4 [d]	21.0 [d]	22.1 [d]	0.7 [d]	0.9 [d]
R	0.2 [c]	0.6 [c]	22.4 [c]	25.1 [c]	1.0 [c]	1.1 [c]
RM	0.4 [b]	1.3 [b]	25.7 [b]	27.2 [b]	1.3 [b]	1.2 [b]
RMD1	0.6 [a]	1.6 [a]	26.2 [b]	27. 5 [b]	1.4 [b]	1.3 [b]
RMD2	0.4 [b]	1.3 [b]	27.9 [a]	29.6 [a]	1.6 [a]	1.4 [a]
Means	0.4	1.0	24.6	26.3	1.2	1.2
S.E	±0.3	±0.1	±0.6	±1.0	±0.1	±0.1
C.V	27.9	13.0	4.9	7.2	11.3	11.2

Means with same letter in the same column are not significantly different at5% level, according Duncan Multiple Range Test.

4.1.3. Yield components

4.1.3.1 Number of pods/plant

Rhizobium (R) significantly increased pods number/plant over control(C). Combination of rhizobium and mycorrhiza (RM) significantly increased pods number/plant. The highest number of pod/ plant was obtained at the combinations of rhizobium + mycorrhiza + 100 kg/ha

DAP (RMD1) and the combination of rhizobium + mycorrhiza + 150 kg/ha DAP (RMD2) treatments.(Table 3).

4.1.3.2 Number of seeds/pod

From the table, it was clear that there were no significant effect of any of treatments on number of seeds/pod as shown in table (3)

4.1.3.3 100 seed weight (g)

rhizobium (R) had no significant effect on 100 seed weight as compared to control (C).The interaction between rhizobium and mycorrhiza (RM) significantly increased100 seed weight. The combination of rhizobium + mycorrhiza +100kg/ha DAP (RMD) gave significantly higher100 seed weight than control. The highest 100 seed weight was obtained with the combinations of rhizobium, mycorrhiza and 150kg/ha DAP (RMD2). (Table 3)

Table (3) Effect of rhizobium and mycorrhizal inoculation and diammonium phosphate (DAP) on yield components of soybean. (Summers season 2010/11 – 2011/12).

treatments	Pods number/plant		Seeds number/pod		100 seed weight(g)	
	(2010/11)	(2011/12)	(2010/11)	(2011/12)	(2010/11)	(2011/12)
C	9.2 d	14.3 d	1.8 a	2.2 a	14.2 d	13.7 c
R	11.4 c	16. 5 c	1.8 a	2.2 a	14.2 d	13.7 c
RM	13.5 b	18.4 b	1.8 a	2.1 a	14.4 c	13.8 c
RMD1	16.6 a	26.4 a	1.9 a	2.1 a	15.0 b	14.4 b
RMD2	17.4 a	28.3 a	1.9 a	2.2 a	16.4 a	15.9 a
Means	13.6	20.8	1.8	2.2	14.8	27.3
S.E	±1.2	±1.4	±0.1	±0.1	±0.3	±0.4
C.V	17.3	13.4	17.3	11.4	4.2	5.3

Means with same letter in the same column are not significantly different at5% level, Duncan according Multiple Range Test.

4.1.4 Chemical composition and grain yield.

4.1.4.1 Oil%

The results showed that rhizobium inoculum (R) significantly increased oil% over control (C). The combination of inoculum and Mycorrhiza (RM) gave significantly higher oil% than control. Combination of rhizobium inoculum, mycorrhiza and 100kg/ha DAP (RMD1) and the combination of inoculum, mycorrhiza and 150kg/ha DAP (RMD2) increased oil% as compared to control. (Table 4)

4.1.4.2 Protein%

As shown in table (4) rhizobium (R) significantly increased protein percentage over control (C). The interaction between rhizobium and mycorrhiza (RM) significantly increased protein percentage. The combination of rhizobium + mycorrhiza +100kg/ha DAP (RMD1) also increased protein percentage over control. The highest protein content was recorded by the combinations of rhizobium, mycorrhiza and 150kg/ha DAP (RMD2).

4.1.4.3 Grain yield (kg/ha)

The data presented in table (4), shows that rhizobium (R) significantly increased grain yield, over control (C) in the first season 2010/2011 only. The interaction between rhizobium and mycorrhiza (RM) significantly increased grain yield as compared to control. The grain yield obtained from combinations of rhizobium, mycorrhiza and 150kg/ha DAP (RMD2) was found to be at par with that of combination of rhizobium + mycorrhiza +100kg/ha DAP (RMD1) and they produced significantly the highest grain yield /ha.

Table NO (4) Effect of rhizobium and mycorrhizal inoculation and diammonium phosphate (DAP) on chemical composition and grain yield of soybean. (Summers season 2010/11 – 2011/12)

Treatments	Oil%		Protein%		Grain yield kg/ha	
	(2010/11)	(2011/12)	(2010/11)	(2011/12)	(2010/11)	(2011/12)
C	14.4 e	15.0 e	24.0 e	34.0 e	145.7 d	202.3 c
R	15.1 d	15.4 d	25.4 d	32.4 d	164.2 c	215.6 c
RM	17.1 a	17.3 a	32.0 c	33.0 c	180.4 b	316.6 b
RMD1	16.5 b	16.8 b	33.5 b	34.5 b	195.6 a	350.3 a
RMD2	15.7 c	16.1 c	35.7 a	36.7 a	197.1 a	356.5 a
Means	15.8	16.1	30.1	34.1	176.6	288.3
S.E	±0.1	±0.1	±0.4	±0.4	±8.1	±19.1
C.V	1.0	0.7	1.7	1.7	9.2	11.4

Means with same letter in the same column are not significantly different at5% level, Duncan according Multiple Range Test.

4.2 Winter seasons (2010-2011) and (2011-2012)

4.2 .1 Nodule number/plant

Table (5) shows that rhizobium inoculum (R) significantly increased nodule number/ plant over control (C). Combination of rhizobium and mycorrhiza (RM) produced significantly the highest nodule number/plant. The interaction between rhizobium, mycorrhizal, 100 or 150 kg/ha of diammonium phosphate (RMD1) and (RMD2) significantly increased number of nodules per plant over control. Nodulation failed in control treatment in the first season.

4.2.2 Mycorrhizal infection%

Rhizobium inoculum (R) significantly increased mycorrhizal infection% as compared to control (C). The combination of rhizobium and mycorrhiza (RM) produced significantly higher mycorrhizal infection% over all treatments. The combination of rhizobium inoculum + mycorrhiza + 100 and 150 kg/ha DAP (RMD1) and (RMD2) increased mycorrhizal infection as compared to control (C).

Table (5). Effect of rhizobium and mycorrhizal inoculation and diammonium phosphate (DAP) on nodulation number/plant and mycorrhizal infection% of soybean. (Winter season 2010/11- 2011/12)

Treatments	Nodulation number/plant		Mycorrhizal infection%	
	(2010/2011)	**(2011/2012)**	**(2010/2011)**	**(2011/2012)**
C	0.0 e	0.8 e	20.5 e	19.0 e
R	24.5 c	17.3 c	30.5 d	27.5 d
RM	40.0 a	32. a	77.5 a	60.0 a
RMD1	28.3 b	23.5 b	65.0 b	57.0 b
RMD2	16. d	11.3 d	57.0 c	50.5 c
Means	21.8	17	50.1	42.7
S.E=	±0.3	±0.7	±0.8	±0.6
C.V%	3.1	8.7	2.2	1.7

Means with same letter in the same column are not significantly different at5% level, Duncan according Multiple Range Test.

4.2.3 Vegetative Growth.

4.2.3.1 Relative growth rate ($g/m^2/day$)

As shown in table (6), rhizobium inoculum (R) significantly increased relative growth rate over control (C). The combination of rhizobium and mycorrhiza (RM) significantly increased relative growth rate. Combination of rhizobium + mycorrhiza + 150 kg/ha DAP (RMD2) registered significantly higher relative growth rate (RGR) as compared to control. The highest relative growth rate was recorded by the combination of rhizobium + mycorrhiza + 100 kg/ha DAP (RMD1).

4.2.3.2 Plant height (cm)

Rhizobium inoculum (R) significantly increased plant height over control (C). The interaction between rhizobium inoculum and mycorrhizal (RM) significantly increased plant height as compared to control(C). The combination of rhizobium + mycorrhiza + 100 kg/ha DAP (RMD1) gave taller plants than (RM) but it the differences was not significant. The greatest plant height was given by combination of rhizobium with VAM mycorrhiza and 150kg/ha DAP (RMD2). (Table 6).

4.2.3.3 Branches number/ plant

Rhizobium inoculum (R) significantly increased branches number/plant over control (C). The interaction between inoculum and mycorrhizal (RM) significantly increased branches number/plant as compared to control. Although the combination of rhizobium + VAM mycorrhiza + 100 kg/ha DAP (RMD1) gave more branches /plant than (RM) but it was statically at par during winter of (2010/12). The highest branches number/plant was recorded in combination of rhizobium with mycorrhiza and 150kg/ha DAP (RMD2). (Table 6).

Table (6). Effect of rhizobium and mycorrhizal inoculation and diammonium phosphate (DAP) on vegetative growth of soybean. (Winter season 2010/11- 2011/12)

treatments	RGR		Plant height (cm)		Branches number/plant	
	(2010/11)	(2011/12)	(2010/11)	(2011/12)	(2010/11)	(2011/12)
C	0.3 d	0.4 d	34.2 d	30. 7 d	2.0 e	1.5 d
R	0.6 c	0.6 c	37.1 c	32.3 c	2.2 d	1.9 c
RM	1.3 b	0.8 b	40.2 b	36.2 b	2.4 c	2.2 b
RMD1	1.6 a	1.5 a	40. 7 b	36.4 b	2.6 b	2.1 b
RMD2	1.4 b	0.9 b	43.7 a	38.3 a	2.9 a	2.4 a
Means	1.0	0.8	39.2	34.8	2.4	2.0
S.E	±0.3	±0.1	±1.7	±0.7	±0.1	±0.1
C.V	23.2	17.6	8.6	4.2	17.4	8.2

Means with same letter in the same column are not significantly different at5% level, Duncan according Multiple Range Test.

4.٢.4 Yield components

4.٢.4.1 Number of pods/plant

Rhizobium (R) produced significantly more pods /plant over control(C). Combination of rhizobium and mycorrhiza (RM) significantly increased pods number/plant over the control. The highest number of pod/ plant was achieved by the combinations of rhizobium + mycorrhiza + 100 kg/ha DAP (RMD1) and the combination of rhizobium + mycorrhiza + 150 kg/ha DAP (RMD2) treatments. (Table 7).

4.2.4.2 Number of seeds/pod

Table (7), shows that there was no significant effect of any of treatments on number of seeds/pod

4.2.4.3 100 seed weight (g)

Rhizobium (R) significantly increased 100 seed weight over control (C). The interaction between rhizobium and mycorrhiza (RM) significantly increased 100 seed weight. The combination of rhizobium + mycorrhiza +100kg/ha DAP (RMD1) significantly increased 100 seed weight. The highest 100 seed weight was obtained with the combinations of rhizobium, mycorrhiza and 150kg/ha DAP (RMD2). (Table 7).

Table NO (7) Effect of rhizobium and mycorrhizal inoculation and diammonium phosphate (DAP) on yield components of soybean. (Winter season 2010/11- 2011/12)

treatments	Pods number/plant		Seeds number/pod		100 seed weight(g)	
	(2010/11)	(2011/12)	(2010/11)	(2011/12)	(2010/11)	(2011/12)
C	23.2 d	16.8 d	2.2 a	2.3 a	15.5 e	14.0 e
R	25.8 c	19. 5 c	2.3 a	2.3 a	15.7 d	14.3 d
RM	30.6 b	22.4 b	2.3 a	2.3 a	15.8 c	14.5 c
RMD1	42.2 a	32.4 a	2.2 a	2.3 a	17.0 b	14.8 b
RMD2	43. 5 a	33.3 a	2.3 a	2.3 a	17.9 a	15.8 a
Means	33.1	24.9	2.3	2.3	16.4	14.7
S.E	±1.9	±1.7	±0.1	±0.1	±0.4	±0.3
C.V	11.3	19.4	7.7	4.6	3.0	3.9

Means with same letter in the same column are not significantly different at5% level, Duncan according Multiple Range Test.

4.2.5 Chemical composition and grain yield.

4.2.5.1 Oil%

Inoculum (R) significantly increased oil% over control (C). The combination of inoculum and Mycorrhiza (RM) produced significantly higher oil% than control. Combination of rhizobium inoculum, mycorrhiza and 100kg/ha DAP (RMD1) and the combination of inoculum, mycorrhiza and 150kg/ha DAP (RMD2) increased oil% as compared to control. (Table 8).

4.2.5.2 Protein%

Rhizobium (R) increased significantly protein percentage of soybean over control. The interaction between rhizobium and mycorrhiza (RM) significantly increased protein percentage. The combination of rhizobium + mycorrhiza +100kg/ha DAP (RMD1) significantly increased protein percentage of soybean. The highest protein% was recorded by the combinations of rhizobium, mycorrhiza and 150kg/ha DAP (RMD2). (Table 8).

4.2.5.3 Grain yield kg/ha

Rhizobium (R) significantly increased grain yield over control (C). The interaction between rhizobium and mycorrhiza (RM) significantly increased grain yield as compared to control. The grain yield obtained from combinations of rhizobium, mycorrhiza and 150kg/ha DAP (RMD2) was found to be at par with that of combination of rhizobium + mycorrhiza +100kg/ha DAP (RMD1) and they produced significantly the highest grain yield /ha. (Table 8).

Table (8). Effect of rhizobium and mycorrhizal inoculation and diammonium phosphate (DAP) on Chemical composition and grain yield of soybean. (Winter season 2010/11- 2011/12).

treatments	Oil%		Protein%		Grain yield kg/ha	
	(2010/11)	(2011/12)	(2010/11)	(2011/12)	(2010/11)	(2011/12)
C	16.0^{e}	15.3^{e}	35.7^{e}	33.6^{e}	576.9^{d}	236.4^{d}
R	16.8^{d}	16.3^{d}	36.4^{d}	34.5^{d}	760.2^{c}	274.1^{c}
RM	18.0^{a}	17.7^{a}	36.8^{c}	35.5^{c}	804.1^{b}	304.8^{b}
RMD1	17.6^{b}	17.2^{b}	37.3^{b}	36.9^{b}	941.2^{a}	537.5^{a}
RMD2	17.1^{c}	16.7^{c}	38.5^{a}	38.4^{a}	956.6^{a}	542.9^{a}
Means	17.2	16.6	36.9	35.8	807.8	379.1
S.E	±0.2	±0.3	±0.3	±0.1	±79.0	±11.7
C.V	1.4	2.4	0.2	0.5	19. 9	15.6

Means with same letter in the same column are not significantly different at5% level, Duncan according Multiple Range Test.

4.3 Comparison between summer and winter seasons of (2010/11)

4.3.1 Nodule number/plant, mycorrhizal infection%, RGR, plant height and branches umber/plant.

As shown in table (9) winter season produced more nodule /plant and higher mycorrhizal infection% by about 59% and 24% respectively compared to summer season. With regard to relative growth rate winter season resulted in higher relative growth rate, plant height and branches number/plant by about 60%, 37% and 50% respectively

Table (9). Comparison between means of nodule number/plant, mycorrhizal infection%, RGR, plant height and branches number/plant, during summer and winter season of (2010/11).

treatments	Nodule number/plant	Mycorrhizal infection%	RGR	Plant height (cm)	Branches number/plant
summer	8.9	38.2	0.4	24.6	1.2
winter	21.8	50.1	1.0	39.2	2.4

4.3.2 Pods number/plant, seeds number/pod, 100 seed weight, oil%, protein % and grain yield

As shown in table (10) winter season gave higher pods /plant, seeds number/pod and 100 seed weight than summer season by about 59%, 22%, and 6% respectively. The oil%, protein% and grain yield kg/ha were higher in winter season than summer by about 4%, 18% and 78% respectively

Table (10). Comparison between means of Pods number/plant, seeds number/pod, 100 seed weight, oil%, protein % and grain yield during summer and winter season of (2010/11).

treatments	Pods number/plant	Seeds number/pod	100 seed weight(g)	Oil%	Protein%	Grain yield kg/ha
summer	13.6	1.8	14.8	15.8	30.1	176.6
winter	33.1	2.3	16.4	17.2	36.9	807.8

4.4 Comparison between summer and winter seasons of (2011/12)

4.4.1 Nodule number/plant, mycorrhizal infection%, RGR, plant height and branches umber/plant.

Nodule number/plant and mycorrhizal infection% were higher in winter season than summer by about 34% and 6%.With regard to the vegetative growth, an increase of 20% relative growth rate was decreased in summer as compared to winter season, but winter season produced greater plant height and branches number/plant by about 24% and 40% respectively than summer season. (Table 11).

Table (11) Comparison between nodule number/plant, mycorrhizal infection%, RGR, plant height and branches number/plant during summer and winter season of (2011/12)

treatments	Nodule number/plant	Mycorrhizal infection%	RGR	Plant height (cm)	Branches number/plant
summer	11.2	40.0	1.0	26.3	1.2
winter	17	42.7	0.8	34.8	2.0

4.4.2 Pods number/plant, seeds number/pod, 100 seed weight, oil%, protein % and grain yield

As shown in table (12), winter season resulted in higher pod number/plant, seeds number/pod and 100 seed weight as compared to summer season by about 16.4%, 4%, and 2% respectively. The oil%, protein% and grain yield kg/ha were also higher in winter season than summer by about 2%, 5% and 24% respectively.

Table (12) Comparison between pods number/plant, seeds number/pod, 100 seed weight, oil%, protein % and grain yield during summer and winter season of (2011/12)

treatments	Pods number/plant	Seeds number/pod	100 seed weight(g)	Oil%	Protein%	Grain yield kg/ha
summer	20.8	2.2	14.3	16.1	34.1	288.3
winter	24.9	2.3	14.7	16.6	35.8	379.1

CHAPTER FIVE

DISCUSSION

5.1 Nodule number/plant and Mycorrhizal infection%

In all seasons of the experiment, rhizobium (R) significantly increased nodule number/ plant table. Similar results were reported by Majid *et al.* (2009) and Faisal (1986). Combination of rhizobium and mycorrhiza (RM) produced significantly the highest nodule number/plant. This agreed with Taiwo *et al*. (2001) and Mahadi (2004) who reported in general that dual inoculation with mycorrhiza and Bradyrhizobium significantly increased nodule number of soybean when compared with control. The interaction between rhizobium, mycorrhizal, 100 and 150 kg/ha of diammonium phosphate increased number of nodules per plant over control. This positive result could be related to increase nutrient absorption by the rhizobium and mycorrhiza of nitrogen and phosphorus content in DAP fertilizer. This result agreed with those of Maurya and Rathi (2000) Gan *et al.* (2002), and Mrkovacki, (2008) who reported in general that phosphorus, starter nitrogen, rhizobium and mycorrhiza increased nodulation of soybean. Nodulation failed in control treatment in the first season which could be related to absence of specific or efficient rhizobium strain in the field. Similar result was reported by Papastylianou (1986) and Yanni *et al* (1987) who reported that soybean in non-inoculated plots with *Rhizobium japonicum* failed to nodulate. With regards to mycorrhizal infection, rhizobium increased mycorrhizal infection over control. Similar results reported by Staehelin (1995) who demonstrated that rhizobium enhanced mycorrhizal colonization of soybean. Dual mycorrhizal and rhizobium inoculation increased mycorrhizal infection. This agrees with Mikhaeel *et al* (2000) who reported that combined inoculation of the mycorrhizal fungi and *Bradyrhizobium japonicum* produced further increase in mycorrhizal infection. The combination of rhizobium, mycorrhiza, 100 and 150 kg/ha of diammonium phosphate (RMD1) and (RMD2) increased mycorrhizal infection as compared to control. Similarly, Baath and Spokes (2015) reported that high phosphorus and low nitrogen had little an influence mycorrhizal infection, but the combination of high phosphorus and high nitrogen decreased mycorrhizal infection. Also Faisal *et al* (2014) and Phillips and Hayman (1970) observed that high levels of nitrogen and phosphorus fertilizer decreased mycorrhizal infection of wheat. At intermediate P levels, infection increased with increased nitrogen application. Menge *et al.*, (1978) illustrated that the reduction in mycorrhizal infection under high phosphorus levels may be due to robust root cell walls which are less liable to infection.

5.2 Vegetative Growth.

Rhizobium (R) significantly increased relative growth rate over control (C). In this connection, Ali (2011) found similar result in chickpea. This effect was not significant during summer season of (2010). The combination of rhizobium and mycorrhiza (RM) significantly increased relative growth rate. Jose *et al*. (2009) reported that mycorrhiza increased relative growth rate of palm which agreed with the present study. Combination of rhizobium + mycorrhiza + 150 kg/ha DAP (RMD2) registered significantly higher relative growth rate (RGR) as compared to control. The best relative growth rate was recorded in the combination of rhizobium + mycorrhiza + 100 kg/ha DAP (RMD1). This may be related to DAP nutrient contents and high nutrient supply to the rhizobium and mycorrhiza. Similar results were reported by Goswami and Soni (2001), Abdel-Fattah (2002), Mahamood *et al*. (2009) and Ali (2011). They reported in general that the interaction between chemical and bio-fertilizers significantly increased relative growth rate of soybean.

With regard to plant height and branches number/plant, the results during all seasons was similar; where rhizobium (R) significantly increased plant height and branches number/plant. This agrees with Tairo (2013) who showed that *Bradyrhizobium japonicum* inoculation significantly increased plant height and number of branches per plant of soybean. The combination of rhizobium and mycorrhiza (RM) significantly increased plant height Shrestha *et al.,* (2009) and Moradgholi (2013) found similar result on plant height of soybean and Habibzadeh and Abedi (2014) on branches number of mung bean. The combination of rhizobium + mycorrhiza + 100 kg/ha DAP (RMD1) was statistically at par with exception of winter season of (2010- 2011) on branches number/plant. Treatment (RMD1) produced greater plant height and branches number than (RM). The greatest plant height and branches number/plant were recorded with the combination of rhizobium + mycorrhiza + 150 kg/ha DAP (RMD2) as compared to the other treatments. This result may be due to the high nutrients supply to rhizobium and mycorrhiza, particularly nitrogen and phosphorus. Similar results were reported by Saxena (2003), Mehmet (2008), Jahangir et al (2009) Shahid (2009) Fahmina et al (2013) and Habibzadeh and Abedi (2014).

5.3 Yield components.

Rhizobium (R) significantly increased pods number/plant over control(C). This is in agreement with observations of Hernandez and Cuevas (2003) and Solomon et al (2012) who reported that the inoculation with *Bradyrhizobium japonicum* resulted in significantly higher number of pods per plant. Combination of rhizobium and mycorrhiza (RM) gave significantly higher pods number/plant than rhizobium (R) treatment alone. In this connection Tholkappian *et al* (2001) showed that pod number per plant increased in mycorrhizal soyabean than the non-mycorrhizal plants which agree with this result. The highest number of pods/ plant was achieved with the combination of rhizobium + mycorrhiza + 100 kg/ha DAP (RMD1) and the combination of rhizobium + mycorrhiza + 150 kg/ha DAP (RMD2)) treatments This might be due to the combined effect of phosphorus and nitrogen as well as enhancement of phosphorus and nitrogen availability to plants by rhizobium and mycorrhiza. Similar findings were reported by Alam *et al* (1989), Anderson (2007) , Shahid *et al* (2009) and Fahmina *et al* (2013). With respect to number of seeds/pod, the treatments had no significant effect on this character in all seasons. With regard to100 seed weight, rhizobium (R) significantly increased 100 seed weight over control (C). In this connection Akhtar and Siddiqui (2009) and Aslam et al (2010) found that rhizobium increased 100 seed weight of soybean, which agrees with this study. The differences during summer seasons were not statistically significant. The interaction between rhizobium and mycorrhiza (RM) significantly increased 100 seed weight. Mobasser *et al.,* (2012) found similar result. The combinations of rhizobium, mycorrhiza and 150kg/ha DAP (RMD2) and combination of rhizobium + mycorrhiza +100kg/ha DAP (RMD1) significantly increased 100 seed weight over control. This may be due to higher nutrient supply due to DAP, rhizobium and mycorrhiza. Other studies have also shown that nitrogen, phosphorus applications, rhizobium and mycorrhiza increased 100 seed weight. (Chetti *et al* 1995; Elsheikh and Elzidany 1997; Mehasen and Saeed 2005; Abdul-Aziz 2013; and Jaidee1 *et al* 2013)

5.3 Chemical composition and grain yield kg/ha

Rhizobium (R) significantly increased oil% as compared to control (C). Mirzakhani, (2008) reported that seed inoculated with bacteria increased oil percent which agrees with this study. The combination of rhizobium and mycorrhiza resulted in higher oil%. Similar result was reported by Amiri et al (2013) who found that mycorrhiza and rhizobium significantly increased oil content of soybean. The combinations of rhizobium, mycorrhiza and 150kg/ha DAP (RMD2) and combination of rhizobium + mycorrhiza +100kg/ha DAP (RMD1) increased oil% over control. Other workers supported this finding (Badran 2003; Tomar *et al*. 2004; Mehasen and Saeed 2005; Jahangir *et al* 2009).

With respect to protein%, rhizobium (R) increased protein percentage of soybean over control (C). Similar result was obtained by Sharma (2006) who reported that soybean plants inoculated with Rhizobium bacteria showed increased protein of soybean. The interaction between rhizobium and mycorrhiza (RM) significantly increased protein percentage. This agree with Amiri *et al* (2013) who found that mycorrhiza and rhizobium significantly increased protein of soybean. The combination of rhizobium + mycorrhiza +100kg/ha DAP (RMD1) increased protein percentage of soybean. The highest protein% was recorded in the combination of rhizobium, mycorrhiza and 150kg/ha DAP (RMD2). This may be related to higher nutrient supply by DAP, rhizobium and mycorrhiza. Other studies have shown that protein content of soybean was increased by rhizobium, mycorrhiza, nitrogen and phosphorus**.** **(**Wood *et al* 1993; Bardan 2003, and Tomar *et al*. 2004).

The results of this study showed that rhizobium (R) significantly increased grain yield over control (C), but this result was not significant during summer of (2011). Ibrahim *et al*. (2011) observed that at Shambat inoculation significantly improved yield, which agreed with the study. The interaction between rhizobium and mycorrhiza (RM) significantly increased grain yield as compared to control. Similar result was found by Young *et al.* (1989) who reported that mixed inoculation with rhizobium and mycorrhiza increased yields of soybean. The grain yield/ha obtained from combinations of rhizobium, mycorrhiza and 150kg/ha DAP (RMD2) was found to be at par with that of combination of rhizobium + mycorrhiza +100kg/ha DAP (RMD1) and they produced significantly the highest grain yield /ha. This might be due to the combined effect of DAP, which contains phosphorus and nitrogen as well as enhancement of phosphorus and nitrogen availability to plants by rhizobium and mycorrhiza. Results reported by Yanni *et al*. (1987)**,** Malik *et al*. (2006), Majumdar et al (2001), Rezende (2001), Kazi (2002), and Mehmet (2008) Valinejad *et al* (2013) indicated that mycorrhiza, rhizobium, phosphorus and nitrogen increased grain yield of soybean.

5.4 Comparison between summer and winter seasons

In the present investigation winter season produced higher nodule number /plant and mycorrhizal infection %compared to summer season. Mike (2012) reported that very high soil temperatures (90° degrees F or 32° C) can cause decreased nodulation and nitrogen fixation in soybeans, which agrees with this study. Winter season recorded higher mycorrhizal infection % than summer season. In this connection Hayman (1974) reported that the optimum temperature for the infection of mycorrhiza is reported to be in the range of 20- 25°C, while Bowen (1987) stated that mycorrhizal colonization usually increases up to about 30°C**,** which may help explain higher mycorrhizal infection in winter than in summer. Higher relative growth rate was achieved in winter season that summer. Similar result was reported by Rufty *et al* (1981) who showed that cool temperature increased relative growth rate of soybean. Plant height was higher in winter season than summer season. In this connection Singh (2014) showed that temperature of 36/32°C decreased plant height and branches number/plant of soybean which agrees with this study.

Winter season gave higher pods number/plant, seeds number/pod, 100 seed weight, oil%, protein% and grain yield than summer season. Lindsey and Thomson (2012) reported that temperature exceeding 20°C can result in a decreased number of pods/plant while temperature above 37° C severely limits pod formation which agrees with this study. Seeds number/pod was higher during winter season than summer. Prasad *et al.* (2001) reported similar observations in soybean. Lindsey and Thomson (2012) reported that the optimum temperature range for soybean was 25-29°C and pod treatment was seriously affected at temperatures above 37°C. Winter season produced higher100 seed weight than summer season. Drew and Brocklehurst (1990) and Khalil *et al* (2010) found similar result. They reported that seed weight decreased with increased temperature above 30/20°C day/night. Winter season gave higher oil% than summer season. Kipps (1959) and Rotundo and Westgate (2009) reported that high temperature above 26°C significantly decreased the oil concentration which agrees with this study. Protein% was higher in winter season than summer. In this connection Bellaloui *et al.* (2011) suggested that lower temperature during seed filling might be a possible reason for increased protein content. Grain yield kg/ha seem to be higher in winter season than summer. This result disagreed with finding of Samia *et al* (2013) and Ibrahim (2012) who recommended that soybean could be grown in Sudan as summer crop, This may be due to the cultivar habits, but this agrees with the findings of Wolfram *et al*, 2009 and Mike (2012) who reported in general that temperatures above 95°F (35°C) have been shown to significantly decrease yield.

CHAPTER SIX

SUMMARY, CONCLUSION and RECOMMENDATIONS

Summary:

A field experiment was conducted for two consecutive summer and winter seasons (2010/11 and 2011/12), at alkaline clay soil to study the effect of rhizobium and mycorrhiza inoculation and diammonium phosphate (DAP) on growth, nodulation and yield of soybean *Glycine max* (L.) Merrill, (cultivar E01). The treatments consist of , control, inoculation with Bradyrihzobium alone, combinations of Bradyrihzobium + mycorrhiza, combination of Bradyrihzobium + mycorrhiza + 100 kg /ha DAP, and combination of Bradyrihzobium + mycorrhiza + 150 kg /ha DAP. The results showed that winter season and combination of Bradyrihzobium + mycorrhiza + 150 kg /ha DAP gave higher growth and yield of soybean than summer season and other treatments.

Conclusion:

1- During winter season cultivar (E01) of soybean produced significantly higher nodulation, growth, yield attributes and yield of soybean when combination of rhizobium, mycorrhiza and diammonium phosphate was used.
2- Rhizobium increased growth and yield over control, but alone can not achieved higher grain yield of soybean.
3- Combination of mycorrhiza and rhizobium inoculation increased growth and yield of soybean in the soil of Sudan which contents low phosphorus and nitrogen quantity, because of their proved effective in enhancing absorption of nitrogen and phosphorus.
4- Cultivar (E01) of irrigated soybean can be used as winter crop under Sudan conditions, but more research is highly recommended.

Recommendations:

1- The release of adaptable soybean cultivars is highly recommended for both irrigated and rainfed conditions.

2- For optimum nodulation of soybean grown on alkaline clay soils, the following are recommended:

   a. The use of proper strain of *Bradyrhizobium japonicum*.

   b. Inoculation of vesicular – arbuscular mycorrhiza.

   c. The application of diammonium phosphate at the rate 150kg/ha at planting.

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APPENDICES

Appendix (1): Climate

Sun- shine duration	3650 hours/year
Soloar radiation	22.7 MJ/m²/day
Maximum temperature	42°c (May)
Minimum temperature	12° c (January)
Temperature range	32°c
Rainfall	100-250 mm/annum
Evaporation	2400 mm/annum

Appendix (2): Chemical and physical properties of the field soil

PH	**8.2**
ECCds/m	**0.5**
SAR	**4.6**
Soluble cation(meg/1)	
Ca+ Mg	**0.9**
Na	**3.1**
K	**0,3**
Cl meg/L	**10.3**
Na%	**0,04**
Pp.p.m	**3.1**
CaCo%	**2.0**
Sand %	**15**
Silt%	**23**
Clay%	**62**

Appendix (3): Analysis of variation tables

Summer season (2010)

Nodulation number/plant

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	21.200	7.067	5.3671	0.0141
٢	Factor A	4	822.200	205.550	156.1139	0.0000
٣-	Error	12	15.800	1.317		
	Total	19	859.200			

Mycorrhizal infection %

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	1	0.000	0.000	0.0000	
٢	Factor A	4	3067.000	766.750	3067.0000	0.0000
٣-	Error	4	1.000	0.250		
	Total	9	3068.000			

Relative growth rate

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	0.029	0.010	0.9344	
٢	Factor A	4	0.499	0.125	11.9683	0.0004
٣-	Error	12	0.125	0.010		
	Total	19	0.653			

Plant height

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	1.949	0.650	0.4451	
٢	Factor A	4	129.370	32.343	22.1549	0.0000
٣-	Error	12	17.518	1.460		
	Total	19	148.838			

Branches number/plant

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	0.076	0.025	1.5276	0.2579
٢	Factor A	4	1.273	0.318	19.1910	0.0000
٣-	Error	12	0.199	0.017		
	Total	19	1.548			

Pods number/plant

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	10.397	3.466	0.6275	
٢	Factor A	4	190.108	47.527	8.6048	0.0016
٣-	Error	12	66.280	5.523		
	Total	19	266.785			

Seeds number/pod

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	0.138	0.046	0.7340	
٢	Factor A	4	0.072	0.018	0.2872	
٣-	Error	12	0.752	0.063		
	Total	19	0.962			

100 seed weight

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	0.378	0.126	0.2932	
٢	Factor A	4	14.207	3.552	8.2647	0.0019
٣-	Error	12	5.157	0.430		
	Total	19	19.742			

Oil%

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	1	0.001	0.001	0.0385	
٢	Factor A	4	4.256	1.064	40.9229	0.0017
٣-	Error	4	0.104	0.026		
	Total	9	4.361			

Protein%

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	1	0.025	0.025	0.1695	
٢	Factor A	4	20.666	5.166	35.0272	0.0023
٣-	Error	4	0.590	0.147		
	Total	9	21.281			

Grain yield

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	1150.502	383.501	1.4533	0.2764
٢	Factor A	4	7624.854	1906.214	7.2237	0.0033
٣-	Error	12	3166.583	263.882		
	Total	19	11941.939			

Summer season (2011)

Nodulation number/plant

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	5.200	1.733	0.6246	
٢	Factor A	4	1570.700	392.675	141.5045	0.0000
٣-	Error	12	33.300	2.775		
	Total	19	1609.200			

Mycorrhizal infection %

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	1	0.400	0.400	0.6154	
٢	Factor A	4	3783.400	945.850	1455.1538	0.0000
٣-	Error	4	2.600	0.650		
	Total	9	3786.400			

Relative growth rate

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	1.026	0.342	2.8520	0.0818
٢	Factor A	4	1.377	0.344	2.8707	0.0700
٣-	Error	12	1.439	0.120		
	Total	19	3.842			

Plant height

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	169.286	56.429	5.0001	0.0178
٢	Factor A	4	210.353	52.588	4.6598	0.0168
٣-	Error	12	135.427	11.286		
	Total	19	515.066			

Branches number/plant

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	0.046	0.015	0.9010	
٢	Factor A	4	0.682	0.170	10.1287	0.0008
٣-	Error	12	0.202	0.017		
	Total	19	0.929			

Pods number/plant

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	59.910	19.970	2.5879	0.1015
٢	Factor A	4	618.152	154.538	20.0265	0.0000
٣-	Error	12	92.600	7.717		
	Total	19	770.662			

Seeds number/pod

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	0.358	0.119	1.9834	0.1703
٢	Factor A	4	0.070	0.018	0.2909	
٣-	Error	12	0.722	0.060		
	Total	19	1.150			

100 seed weight

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	4.236	1.412	1.7707	0.2062
٢	Factor A	4	14.027	3.507	4.3976	0.0203
٣-	Error	12	9.569	0.797		
	Total	19	27.832			

Oil%

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	1	0.000	0.000	0.0000	
٢	Factor A	4	5.750	1.438	115.0005	0.0002
٣-	Error	4	0.050	0.012		
	Total	9	5.800			

Protein%

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	1	0.016	0.016	0.0440	
٢	Factor A	4	29.874	7.468	20.5461	0.0063
٣-	Error	4	1.454	0.364		
	Total	9	31.344			

Grain yield

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	741.930	247.310	0.3740	
٢	Factor A	4	31936.977	7984.244	12.0750	0.0004
٣-	Error	12	7934.666	661.222		
	Total	19	40613.573			

Winter season (2010/11)

Nodulation number/plant

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	4.150	1.383	2.9643	0.0748
٢	Factor A	4	3556.000	889.000	1905.0000	0.0000
٣-	Error	12	5.600	0.467		
	Total	19	3565.750			

Mycorrhizal infection %

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	1	4.900	4.900	4.2609	0.1079
٢	Factor A	4	5060.600	1265.150	1100.1304	0.0000
٣-	Error	4	4.600	1.150		
	Total	9	5070.100			

Relative growth rate

Value	Source	Freedom	Squares	Square	Value	Prob
١	Replication	3	3.366	1.122	7.1341	0.0052
٢	Factor A	4	4.137	1.034	6.5771	0.0048
٣-	Error	12	1.887	0.157		
	Total	19	9.390			

Plant height

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	9.113	3.038	1.4530	0.2765
٢	Factor A	4	160.515	40.129	19.1935	0.0000
٣-	Error	12	25.089	2.091		
	Total	19	194.717			

Branches number/plant

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	0.122	0.041	0.8668	
٢	Factor A	4	1.853	0.463	9.8739	0.0009
٣-	Error	12	0.563	0.047		
	Total	19	2.538			

Pods number/plant

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	306.728	102.243	7.3226	0.0048
٢	Factor A	4	1393.072	348.268	24.9428	0.0000
٣-	Error	12	167.552	13.963		
	Total	19	1867.352			

Seeds number/pod

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	0.066	0.022	0.7154	
٢	Factor A	4	0.067	0.017	0.5447	
-٣	Error	12	0.369	0.031		
	Total	19	0.502			

100 seed weight

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	20.765	6.922	5.1729	0.0159
٢	Factor A	4	16.807	4.202	3.1401	0.0553
-٣	Error	12	16.057	1.338		
	Total	19	53.629			

Oil%

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	1	0.100	0.100	1.4815	0.2904
٢	Factor A	4	6.686	1.672	24.7630	0.0044
-٣	Error	4	0.270	0.067		
	Total	9	7.056			

Protein%

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	1	0.036	0.036	1.5319	0.2835
٢	Factor A	4	80.246	20.061	853.6771	0.0000
-٣	Error	4	0.094	0.024		
	Total	9	80.376			

Grain yield

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	762941.097	254313.699	10.2031	0.0013
٢	Factor A	4	425216.618	106304.155	4.2649	0.0224
-٣	Error	12	299101.351	24925.113		
	Total	19	1487259.066			

Winter season (2011/12)

Nodulation number/plant

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	2.950	0.983	0.4487	
٢	Factor A	4	2257.700	564.425	257.5323	0.0000
٣-	Error	12	26.300	2.192		
	Total	19	2286.950			

Mycorrhizal infection %

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	1	1.600	1.600	1.8824	0.2420
٢	Factor A	4	3332.600	833.150	980.1765	0.0000
٣-	Error	4	3.400	0.850		
	Total	9	3337.600			

Relative growth rate

Value	Source	Freedom	Squares	Square	Value	Prob
١	Replication	3	0.045	0.015	0.1546	
٢	Factor A	4	0.467	0.117	1.1903	0.3642
٣-	Error	12	1.177	0.098		
	Total	19	1.690			

Branches number/plant

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	0.126	0.042	1.5590	0.2504
٢	Factor A	4	1.802	0.450	16.7888	0.0001
٣-	Error	12	0.322	0.027		
	Total	19	2.250			

Plant height

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	18.265	6.088	1.6758	0.2248
٢	Factor A	4	131.683	32.921	9.0614	0.0013
٣-	Error	12	43.597	3.633		
	Total	19	193.546			

Pods number/plant

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	138.088	46.029	2.0691	0.1579
٢	Factor A	4	592.328	148.082	6.6566	0.0046
٣-	Error	12	266.952	22.246		
	Total	19	997.368			

Seeds number/pod

Value	Source	Freedom	Squares	Square	Value	Prob
١	Replication	3	0.018	0.006	0.5255	
٢	Factor A	4	0.003	0.001	0.0657	
٣-	Error	12	0.137	0.011		
	Total	19	0.158			

100 seed weight

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	1.200	0.400	1.2632	0.3309
٢	Factor A	4	7.800	1.950	6.1579	0.0062
٣-	Error	12	3.800	0.317		
	Total	19	12.800			

Oil%

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	1	0.009	0.009	0.0584	
٢	Factor A	4	16.196	4.049	26.2922	0.0039
٣-	Error	4	0.616	0.154		
	Total	9	16.821			

Protein%

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	1	0.064	0.064	0.4089	
٢	Factor A	4	6.574	1.644	10.5016	0.0214
٣-	Error	4	0.626	0.157		
	Total	9	7.264			

Grain yield

K Value	Source	Degrees of Freedom	Sum of Squares	Mean Square	F Value	Prob
١	Replication	3	2214.413	738.138	0.5058	
٢	Factor A	4	215841.317	53960.329	36.9739	0.0000
٣-	Error	12	17513.007	1459.417		
	Total	19	235568.736			