

**RESPONSE OF PROMISCUOUS SOYBEAN TO RHIZOBIAL INOCULATION IN  
COMBINATION WITH ORGANIC AND MINERAL FERTILIZERS IN SOME SOILS  
OF THE NIGERIAN GUINEA SAVANNA**

**BY**

**EKAETTE, Joy Etopobong**

**M.TECH/SAAT/2014/4915**

**DEPARTMENT OF SOIL SCIENCE AND LAND MANAGEMENT**

**FEDERAL UNIVERSITY OF TECHNOLOGY**

**MINNA**

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## ABSTRACT

Effective soybean rhizobia are seldom found in sufficient or large numbers in the soils that have no history of soybean cultivation. There may be a need for soybean grown in such soils to be supplied with inoculant rhizobia to ensure optimal inputs from biological nitrogen fixation. Reports have indicated that yield per hectare of soybean in farmers' fields are still very low. This scenario calls for trials that are capable of establishing some of the biophysical factors limiting the yield of soybean in Nigerian savannas. Available information in literature showed that in addition to the deficiencies of N, P and to some extent potassium, there are generally low to deficient levels of Cu, Mo and Zn in a number of soils from Nigerian savanna. Also, the organic matter content in these soils is low, thus there is need to add organic manure in order to raise the fertility thereby improving soil physical, chemical and biological properties. A trial was set up to diagnose the underlying factors responsible for variation in yield among soybean treatments at four sites in Shiroro Local Government Area of Niger State. The trial consisted of six (6) treatments in the first year, nine (9) treatments in the second year replicated four (4) times arranged in randomized complete block design (RCBD). The treatments were (i) Control, (ii) Inoculant only, (iii) Inoculant + phosphorus (30kg P<sub>2</sub>O<sub>5</sub>/ha), (iv) Inoculant + phosphorus + potassium (20kg K<sub>2</sub>O/ha), (v) Inoculant + phosphorus + potassium + micronutrients (3.3kg/ha) and (vi) Inoculant + phosphorus + potassium + micronutrients + organic manure (4tons/ha), (vii) Inoculant + organic manure, (viii) Inoculant + phosphorus + micronutrient and (ix) Organic manure only. Response to nutrients varied across sites. Treatments with organic matter were significantly greater than the control in terms of plant height, nodule number and shoot dry weight but marginally greater than the control in pod number and pod dry weight. The treatments with inoculant only had similar yield parameters as the control in terms of plant height, shoot biomass, days to 50% flowering, nodule number/weight, pods per plant, number of seeds per pod, grain yield and stover yield, also treatment with inoculant only had similar yield parameters as the mineral fertilizer treatments in terms of plant height @ 4WAS in both year, nodule number, nodule weight, number of branches, days to 50% flowering and stover yield as well as the organic matter treatments. The results indicate the benefit of using bio, mineral and organic fertilizers in improving the productivity of soybean.

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## CHAPTER ONE

### 1.0

### INTRODUCTION

#### 1.1 Background of the Study

Soybean is one of the most important crop in Nigeria. The present production of this crop in Nigeria is projected at 542,050 metric tons. Among the producing states in Nigeria, Benue State is the largest producer of soybeans its production is about 181,680MT and Niger state, which lies within the soybean production belt, produces about 14,050 metric tons of soybean NAERLS/NPAFS (2010). Soybean is one of the agricultural crop known today, as it is one of the best source of protein and oil, and it contains a good quality protein of around 42% and 19.5% oil (Wilcox *et al.*, 2001). In food industry, soybean is used for making flour, oil, cookies, candy, milk, vegetable cheese and many other products (Coskan *et al.*, 2011).

In order to attain high yield potential, Soybean must sustain high photosynthetic rates and accumulate large amounts of Nitrogen in seeds (Sinclair, 2004). Biological nitrogen fixation and mineral fertilizers are the main sources of meeting the N requirement of high-yielding soybeans. However, antagonism between nitrate concentration in the soil solution and the N<sub>2</sub> fixation process in the nodules is the main constraint the crop faces in terms of increasing N uptake (Streeter, 1988).

In general, most tropical soils have low nitrogen content therefore a starter dose of N is usually applied to support plant growth before the onset of BNF (FPDD, 1990). Legumes have a high phosphorus (P) requirement for growth and also for nodulation and N fixation (Israel, 1987).

Next to Nitrogen, Phosphorus (P) is the most commonly deficient nutrient element in tropical soils (Vladimir, 2010). The higher P concentration in nodules demonstrates the higher demand of legumes for P nutrition (Vladimir, 2010). Some studies have also established that phosphorus fertilization stimulates root growth (Singh *et al.*, 2000). Recommended P rate for soybean in soils of the Nigerian savanna is 20kg/ha – 40kg/ha (FMARD 2012). Soil fertility is ‘the state of a soil with respect to its ability to supply elements essential for growth without toxic concentration of any element. Soybean like other N<sub>2</sub> fixing legumes, can meet part of its N requirement through biological nitrogen fixation (BNF).

Micronutrients, although required for plant growth in minute quantities are as important to the plant as the macronutrients such as N and P. Application of micronutrient in combination with N,P and K has been reported to increase crop yields and break the cycle of poverty in Sub-Sahara Africa (Nube *et al.*, 2006). Recent literature reports indicate that a number of soil of the Nigerian savanna have low to deficient level of boron, copper, molybdenum and zinc (Oyinlola *et al.*, 2010)

Soybean is widely reputed to be highly specific in its rhizobial requirement, hence the need to apply rhizobial inoculants to help the crop meet its BNF needs (Vladimir, 2010). However, the TGx soybean varieties known as promiscuous soybean developed by the International Institute of Tropical Agriculture (IITA) breed freely to form nodules and fixing nitrogen with indigenous rhizobium in tropical soils (Kueneman, *et al.*, 1984; Pulver *et al.*, 1985). Nonetheless, recent studies have shown that these varieties of soybean respond to rhizobial inoculation (Osunde *et al.*, 2003, Ronner *et al.*, 2016).

The uses of organic matter for enhanced fertility and crop productivity is generally recognized providing most of the nutrient requirement by crops (Asei *et al.*, 2015). Organic manure provides a good substrate for the growth of microorganisms and maintains a favorable nutritional balance and soil physical properties. One of such strategy to maintain soil fertility for sustainable production of soybean is through judicious use of fertilizers (Bobde *et al.*,1998) coupled with organic resources that help to achieve sustainability in production as the use of organic manures alone is not sufficient (Prasad, 1996). It also have been reported that the use of organic manures in integration with fertilizers meets the need of micronutrients of soybean (Joshi *et al.*, 2000). Lourduraj (2000) also reported that the combined application of inorganic and organic manures significantly enhanced the growth attributes and yield of soybean as compared to the sole application of either of them.

## **1.2 Statement of Problem**

Soybean yield in farmer's field is still very low. Average yields are estimated at less than 500 kg/ha NPAFS, (2009) although up to 3 – 4 t/ha are obtained in some other countries (DAFF, 2010). Low soil fertility especially deficiencies of P and K and micro nutrients generally have been reported as a major factor hindering soybean productivity. Research has shown that some Nigerian soils are deficient in micronutrient such as Mo, Cu, (and Zn).

### **1.3 Justification of the Study**

Responses of crop to added micronutrients have been reported to increase yield over 100% above control with optimum rate of micronutrient application (Oyinlola *et al.*, 2010). Also, the organic matter content in these soils is low, thus there is need to add organic manure in order to raise the fertility thereby improving soil physical, chemical and biological properties (Peter *et al.*, 2000). However there is a dearth of knowledge on the use of organic matter in soybean production within the Nigerian Guinea Savanna.

Although the TGx soybean varieties nodulate freely with indigenous rhizobial populations it is not in all cases that they form effective symbiosis with the indigenous rhizobial, and thus may benefit from inoculation using highly effective rhizobial strain (Pulver *et al.*, 1985). Other studies have also reported significant responses of promiscuous soybean to rhizobial inoculation (Osunde *et al.*, 2003, Ronner *et al.*, 2016). Additionally, Asei *et al.* (2015) observed that the application of molybdenum, a micronutrient, along with rhizobial inoculation resulted in significant increase in soybean grain yield relative to the control.

### **1.4 Aim and Objectives of the Study**

The aim of this study, therefore is to determine the response of a promiscuous soybean variety to rhizobial inoculation in combination with organic and mineral fertilizers in some soils of the Nigerian Guinea Savanna.

The specific objectives for this study are

- To determine the effect of fertilizer combination on the growth of promiscuous soybean
- To measure the effect of inoculation on Nodulation of promiscuous soybean.

- To determine the effect of both mineral and organic fertilizer in combination with inoculation on yield and yield attributes of a promiscuous soybean



## CHAPTER TWO

### 2.0 LITERATURE REVIEW

#### 2.1 Botanical Description and Uses of Soybean

Soybean (*Glycine max (L) Merr.*) belongs to the leguminous family *Leguminosae*. It is a herbaceous annual legume, normally erect, bushy and leafy which is ranked as an oilseed crop, providing approximately 50 % edible oil of the world (Akparobi, 2009). It is grown for its edible bean, oil and protein around the world. Soybean is recognized as one of the prominent agricultural crops today, thus it is the best source of protein and oil and has now been recognized as a potential supplementary source of nutritious food (Wilcox *et al.*, 2001). It has been found to substitute other sources of good quality protein such as milk, meat and fish, therefore has become very suitable to some areas where other protein sources are scarce or too expensive to afford (Anwar *et al.*, 2010). Soybean contains a good quality protein of around 42% and 19.5% oil (Wilcox *et al.*, 2001). Soybean has been found to have different uses; for example in food industry, soybean is used for flour, oil, cookies, candy, milk, vegetable cheese and many other products (Coskan *et al.*, 2011).

Soybean is eaten in fresh green state and dry beans. The plant is used as feed for animals and also soil fertility improvements when used as soil cover crop. Soybean can be grown as a sole crop, intercropped, or mixed with important cereals such as maize, sorghum and millets (Mmbaga *et al.*, 2003).

##### 2.1.1 Promiscuous Soybean Variety

Report by IITA showed soybean improvement by the Institute started around 1974 (IITA, 2003). Low yield of about 0.5 tonha<sup>-1</sup> of soybean is attributed to negligence in improving the crop

generally in Africa (Hailu, 2011). Some other problems include low seed viability (Asafo-Adeji *et al.*, 2001), poor nodulation with native *rhizobium* available in the soil and high shattering score in the savanna regions (Tukamuhabwa *et al.*, 2000; Tukamuhabwa *et al.*, 2002). Low level utilization of the crop also contributes to the limitation in developing recipes for small holder farmers in Africa (Hailu, 2011). These inadequacies led IITA to exploit some of the opportunities soybean can offer to tropical agriculture for over three decades (IITA, 2000). Preliminary yield trial carried out on soybean germplasm materials in 1974 revealed that yields were high as compared to other legumes (IITA, 2003). The high yield of soybean under tropical condition was a factor leading to soybean improvement (Giller *et al.*, 2006). Apart from being a good source of protein and vegetable oil, the presence of genetic diversity to solve some of the limitations such as poor seed longevity and efficient natural nodulation were reasons to advance in soybean (Tefera *et al.*, 2010). National Agricultural Research system (NARS) in the 1970s showed giant interest and commitment in the expanding soybean production and utilization.

Breeding for promiscuous genotypes was one of the approaches IITA followed to enhance biological nitrogen fixation of tropical soybean (IITA, 1997). Soybean that nodulate effectively with indigenous rhizobia are considered as promiscuous, and the characteristic promiscuity (Kueneman, Root *et al.*, 1984; Gwata *et al.*, 2004; Hailu, 2011). Hence, promiscuous genotypes of soybean are able to form symbiotic association with *Rhizobium* strains present in the soil and fix atmospheric nitrogen but non-promiscuous genotypes always require specific rhizobial strains to fix nitrogen from the atmosphere (Alves *et al.*, 2003; Osunde *et al.*, 2003; Tefera *et al.*, 2009; 2010), and in Africa indigenous rhizobium strains for cowpea are available and in abundance (Giller *et al.*, 2006).

High yielding soybean cultivars from USA in the 1970s showed to have specific requirements for *B. Japonicum* (Pulver *et al.*, 1982; Gwata *et al.*, 2005) and inoculation of these varieties was found to be essential when growing them under tropical conditions of low soil nitrogen (IITA, 1997; 2000). In the early 1980s, facilities required for inoculum production, storage and dissemination were not available in tropical countries this led to importation of the finished product (Pulver *et al.*, 1982; Hailu, 2011). Other report show that the limited commercial *B. japonicum* inoculants and nitrogenous fertilizers brought about breeding promiscuous varieties in IITA since soybean genotypes that form symbiotic association with cowpea-type rhizobia were identified (IITA, 1997; 2003). Generally, soybean varieties developed for promiscuous nodulation were considered to increase production of soybean in tropical Africa with minimum cost affordable to small-holder farmers (Hungria *et al.*, 2000; Sanginga *et al.*, 2000; Alves *et al.*, 2003).

Nodulation and non-nodulation observation of some soybean varieties in soils not cultivated previously showed the presence of genotypic variability in soybean ability to recognize and form symbiosis with diverse species of rhizobia strains (Tukamuhabwa *et al.*, 2002; Osunde *et al.*, 2003). Gwata *et al.*, (2004) and Hailu (2011) observed a genotypic variation in six soybean genotypes in their ability to form an effective symbiosis with local *Rhizobium spp.* They also noted that local cultivars were more promiscuous as compared to improved cultivars from the USA. Screening of 400 pure soybean germplasm accessions for their compatibility with indigenous rhizobia in tropical environments and assessing their efficiency of symbiosis under greenhouse and field conditions was carried out by IITA (Pulver *et al.*, 1985; Hailu, 2011). It was observed that ten (10) out of the 400 genotypes screened for promiscuity formed effective symbiotic

relationships with the soil rhizobia at five different sites in Nigeria (Tukamuhabwa *et al.*, 2000; 2002). Tropical Africa and South East Asia are sources of the germplasm accessions. (Tefera *et al.*, 2010).

Developing high yielding, stable, tolerant or resistant to biotic and abiotic promiscuous soybean varieties was the ultimate aim of IITA. (Hailu, 2011). Combining yield potential of different cultivars with promiscuous soybeans has been IITA's focus right from the beginning (Giller *et al.*, 2006). The methodology used in developing the varieties over the years has been selection and Hybridization (Tukamuhabwa *et al.*, 2000). Pedigree method of selection is followed to advance segregating populations by raising two generations per year – one during the main growing season and the second through off-season irrigation (Gwata *et al.*, 2004). Tukamuhabwa *et al.* (2000); Tefera *et al.*, (2010) reported that selection in the F2 and F3 generations are restricted to selecting good individual plants and discarding single plants and progeny rows susceptible to diseases such as bacterial pustule, frog-eye leaf spot and rust. In F4 and F5 generations, progeny rows (families) are discarded when they are found to be susceptible to frog-eye (*Cercospora*) leaf spot or bacterial pustule, or if they are of poor seed colour or plant type. Single plant selections to establish homozygous lines are done at F5 or F6 generation during the main growing season. Seeds of selected individual plants are then multiplied during the off-season. At this stage lines are screened for pod shattering in the laboratory, seed size (10-13 g per 100 seeds), and uniform cream seed colour. Progeny rows that passed the screening procedure are harvested in bulk grouped by maturity and were promoted to preliminary variety trials. The maturity groups in IITA trials are early (less than 100 days), medium (101 – 110 days) and late (more than 110 days).

Twenty-five to 30 superior lines are normally tested under preliminary variety trials at two to three locations in three replications for one year. Better performing lines for key traits are promoted to the advanced variety trial and the rest are discarded. In the advanced variety trial, lines are evaluated in at least three locations in four replications per country. The best lines from the advanced variety trial are distributed to collaborators mainly in Africa in the form of international trials. The purpose of the international trials are to test adaptation of elite soybean lines in different countries under diverse environmental conditions so that breeders from different national programs are able to compare their local varieties with the new lines and eventually release new varieties from IITA's lines. Moreover, it helps national breeders to access new germplasm from IITA for their breeding programs. Within Nigeria, these superior lines are promoted to the National Soybean Variety Trials, which are part of the Nationally Coordinated Research Projects on soybean. While evaluating lines, all the crop management practices are similar to farmer's condition. Starter fertilizer is incorporated into the soil before planting and the rate used is 100 kg ha<sup>-1</sup> NPK (15-15-15) and 50 kg ha<sup>-1</sup> single super phosphate. No fungicides, insecticides or *Rhizobium* inoculants are used and weeds are controlled using herbicides and hoe as needed (IITA, 2000).

Report shows that a total of 21 IITA bred tropical soybean varieties have been released in Africa, and most of these varieties were released in Nigeria. In terms of maturity groups, seven varieties each was released from the early, medium and late, respectively (IITA, 1997). Grain yields ranged from 1 - 2.1 t ha<sup>-1</sup> in the early maturing varieties depending on locations. In medium maturing varieties grain yields ranged from 1 - 2.7 t ha<sup>-1</sup>. In the case of late maturing varieties grain yields ranged from 1.3 - 2.3 t ha<sup>-1</sup>. These yields were achieved through natural nodulation and only starter fertilizers were applied. In addition to grain yield and promiscuous nodulation, several other traits

were incorporated into these varieties. Some of them are fodder yield, seed longevity, resistance to shattering and lodging, suitability for processed products such as soymilk, resistance to major foliar diseases such as bacterial blight and pustule, frogeye leaf spot and tolerance to rust in recently released varieties.

In order to meet the need of nitrogen fixation by small holder farmers, promiscuous varieties were developed by IITA. Tireless effort has been put in place in the last three years to ensure the breeding of promiscuous soybean is implemented. (IITA, 2003; Tefera *et al.*, 2010). The report further shows the need for selection of traits while also developing biotic and abiotic stress resistant varieties. It is crucial that plant with high BNF capacity are selected on soils with low nitrogen fixation. Early stage selection will assist in breeding promiscuity traits. (Tefera *et al.*, 2010). The use of traits indicator of BNF will make it easy to handle large population. (Gwata *et al.* 2005).

Hungria *et al.* (2000) clearly stated out that while breeding promiscuous cultivars high yielding specific varieties with specific strains of *Bradyrhizobium* should be added. This specific strains when used could enhance the biological nitrogen fixation capacity of promiscuous genotypes (Sanginga *et al.*, 2000; Ogoke *et al.*,2003). Identification of soybean varieties with a high capacity for biological nitrogen fixation is essential when recommendation is made to farmers and determination of cultivars which can be used as parent genotypes in breeding programs (Hungria *et al.*,2000). Several scientists have confirmed that breeding soybean for boosted N<sub>2</sub> fixation can be achievable (Hungria *et al.*, 2000; Alves *et al.*, 2003).

Soybean breeders at the International Institute of Tropical Agriculture (IITA), Nigeria developed promiscuous soybean genotypes in order to take advantage of high yielding American and promiscuous Asian soybean varieties that were introduced to the country. These genotypes, known as Tropical Glycine cross (TGx) nodulate effectively with *Bradyrhizobium spp.* populations that are indigenous to African soils (Abaidoo *et al.*, 2007). Okereke *et al.* (2000) reported that promiscuous soybean may also need to be inoculated with exotic Bradyrhizobia depending on the effectiveness and population of the indigenous *bradyrhizobia* in the locality. Indigenous *Bradyrhizobium japonicum* strain IRj 2180A has been isolated from soybean since 1979 and used for inoculation of soybean (Okogun *et al.*, 2003; Muhammad, 2010; Yusuf *et al.*, 2012). The breeding programme to introduce promiscuous soybean was initiated in IITA, Ibadan Nigeria, within this period in 1977 (Sanginga *et al.*, 2000), which later manifested in the 1980s.

### **2.1.2 Response of Promiscuous Soybean Variety to Inoculation**

Soybean response to inoculation in Nigeria is influenced by several factors including high rainfall, high temperature, drought, salinity, acidity and low level of soil mineral nutrients (Freire, 1977). Some studies feature various responses of some promiscuous soybean cultivars to inoculation, phosphorus and nitrogen (Anne *et al.*, 2011). A similar study was conducted on an early maturing promiscuous cultivar (TGX 1485) inoculated with one of four rhizobial strains (R25B, IRj 2180A, IRc 461 and IRc 291) at Minna Nigeria, in the southern Guinea savanna of Nigeria. The four rhizobia inoculants increased all parameters including grain yields over those of the control and Inoculant strain IRj 2180A produced yields comparable to plots supplied with 60 kg N ha<sup>-1</sup> (Muhammad, 2010). A different study showed the response of two soybean cultivars a Malaysian (SAMSOY 2) and TGX (TGX 1448-2E) to *Bradyrhizobium* inoculation with a mixture of R25B

and IRj 2180A rhizobial strains tested in the northern Guinea savanna of Nigeria, in a research - managed on-farm trial. SAMSOY-2 was not significantly affected by the treatment in terms of vegetative parameters such as shoot and root weight, nodule number, biological nitrogen fixation and grain yield. This was attributed to possible high indigenous rhizobia population, adequate for soybean nodulation. However, significant effect of the treatment was observed in root biomass of TGX 1448-2E. The BNF and grain yield of the promiscuous cultivar was significantly higher than that of SAMSOY-2, showing that varietal differences masked the effect of inoculation (Okogun Otuyemi and Sanginga, 2004).

### **2.1.3 Influence of Phosphorus Fertilizer on Soybean**

Phosphorus is one of the most important nutrient elements that limit plant growth in the case of unavailability (Fernandez, Zalba, Gomez and Sagardoy, 2007). Phosphorus is needed in relatively large amounts by legumes for growth and has been reported to promote leaf area, biomass, yield, nodule number and nodule mass in different legumes (Berg and Lynd, 1985; Pacovsky *et al.*, 1986; Kasturikrishna and Ahlawat, 1999). Phosphorus is a vital element in crop production which performs important role for many characteristics of plant growth such as sugar and starch utilization, photosynthesis use, cell division and organization, nodule formation, root development, flower initiation and seed and fruit development (Gangasuresh *et al.*, 2010). Phosphorus being required in large quantities in young cells, particularly shoots and root tips of soybean, where metabolism is high and cell division is rapid, highest concentration of P is required in seeds of the mature soybean plants (Sanginga *et al.*, 1997). Furthermore, phosphorus has important effects on photosynthesis, root development, fruiting and improvement of crop quality (Sara *et al.*, 2013). Large amount of phosphorus applied as fertilizer enters in to the immobile pools through precipitation reaction with highly reactive Aluminium ( $Al^{3+}$ ) and Iron ( $Fe^{3+}$ ) in



acidic, and Calcium ( $\text{Ca}^{2+}$ ) in calcareous or normal soils (Gyaneshwar *et al.*, 2002; Hao *et al.*, 2002). Efficiency of P fertilizer throughout the world is around 10 - 25 % (Isherword, 1998), and concentration of bioavailable phosphorus in soil is very low reaching the level of 1.0 mg kg<sup>-1</sup> soil (Goldstein, 1994). Microbial community influences' soil fertility through soil processes such as decomposition, mineralization, storage and release of nutrients. Microorganisms enhance the phosphorus availability to plants by mineralizing organic phosphorus in soil and by solubilizing precipitated phosphates (Chen *et al.*, 2006; Khan *et al.*, 2002; Pradhan and Sukla, 2005). Have its sources in arable soils and chemical fertilizers, but 75 to 90 percent of the phosphorus combines with iron, calcium and aluminums in soil (Turan, Ataoglu and Sahin, 2006). It has been proven that P increases weight and number of root nodules and also can enhance the pod yield (Jones, Lutz and Smith, 1977). Different reports have shown that when soil P concentration is above 20 mg per kg increase in soybean yield would not be expected (Webb, Mallarino and Blackmer, 1992; Borges and Mallarino, 2003). Abdul and Saud (2012) reported the results of their experiment showing that as P- fertilization increased, growth and yields of the promiscuous soybean cultivar increased. This findings was consistent with the work of Muhammad; Response of a Promiscuous Soybean Cultivar to Rhizobia Inoculation and Phosphorus in Nigeria's Southern Guinea Savanna Alfisol. El- Ghandour, El-Sharawy and Abdel (1996) reported that fertilization with phosphorus increased nodulation, growth, grain yield and N contents in Faba-bean compared with the control. Phosphorus addition led to an increase in bacteria nodules, which was progressive until 120 kg of P /ha having 350 nodules per plant. The interaction between bacterial inoculation and different levels of phosphorus revealed significant differences at ( $P \leq 0.01$ ) in the number of nodules at all levels of phosphorus; the best interaction was the inoculated treatment with 120 kg of P /ha giving 40.50 nodules per plant. The effect of adding different levels of phosphorus increased the weight

of root nodules, compared with control. The highest increase in the weight of nodules was at 120 kg of P /ha having a nodule weight of 52.50 mg /plant. The results demonstrate the obvious interaction between bacteria inoculant and phosphorus level whereby nodule weight increased in response to higher levels of P. The role of phosphorus fertilization was obvious in increasing the dry weight whereby the dry weight increased from 13.34 g /plant for control to 16.59, 22.52 and 27.79 gm. /plant for 40, 80, 120 kg of P /ha. Ahiabo *et al* (2014) reported that the application of phosphorus fertilizer and Rhizobium inoculants showed on the growth of soybean when compared to the non-phosphorus and non-Rhizobium treatments. The plants grew taller with time within a fertilizer application regime, especially when inoculated. Irrespective of growth stage, plants fertilized with 45kg P/ha and inoculated grew tallest (51.8 cm). Legumes such as soybean need phosphorus for adequate growth and nitrogen fixation. Sufficient phosphorus levels are also required to enhance different plant organs growth and promote nodulation and early maturity (Kamara *et al.*, 2010). Studies conducted by Ndakidemi *et al.* (2006) and Shahid *et al.* (2009) provide a proof that increased phosphorus application enhances plant growth significantly. Supplementing legumes with nutrients, especially phosphorus has great potential for increasing yields, as it not only promotes plant growth but also enhances symbiotic establishment for increased N<sub>2</sub> fixation (Gangasuresh *et al.*, 2010). In soybeans, the demand for phosphorus is greatest during pod and seed development where more than 60% of phosphorus ends up in the pods and seeds (Kumar *et al.*, 2008) and (Shahid *et al*, 2009).

## 2.2 Inoculant and Inoculation

Inoculation means passing appropriate rhizobia in interaction with legume seeds or roots. Inoculant is a rhizobium bacterium that is applied to legume seed before planting. Soybean rhizobium

inoculation is the process of applying rhizobium inoculants to the soybean seed before planting in order to increase the nitrogen fixation and nodulation of the soybean roots. Inoculating soybean provides adequate number of bacteria in the soybean root zone, so that effective nodulation will take place (Lamprey *et al.*, 2014). Rhizobium inoculation is a significant technology for the manipulation of rhizobia for improving crop productivity and soil fertility. Rhizobium inoculation can lead to establishment of large rhizobia in the rhizosphere and improved nodulation and nitrogen fixation even under adverse soil conditions (Peoples *et al.*, 1995). Modern inoculants comprise living rhizobia that are developed in the laboratory and transported to farmers in solids or liquid form. These modern inoculants are smeared as seed coatings or integrated into the soil.

The detection of Biological Nitrogen Fixation has been since late 19th century, other works have reported the potentiality of BNF as alternative to inorganic N-fertilizer in agriculture (Bala, 2011; N'cho *et al.*, 2013). This knowledge has led to the practice of inoculation, with first acceptance achieved by moving soil from field to field, or soil to seed before planting. However, this was quickly replaced by the use of pure cultures on agar slants, and later as broths (Bala, 2011). Hence, rhizobia inoculants - used to deliver nitrogen fixing bacteria (collectively termed rhizobia), have been on the commercial market for over 100 years in many developed countries (Nelson, 2004; Giller, 2008; GRDC, 2013). Million tons of soybean are inoculated annually with *Bradyrhizobium japonicum* in the USA in addition to 34 and 53 million tonne in Argentina and Brazil, respectively (Bala, 2011). Inoculation benefits and application of deficient nutrients, mostly phosphorus varies with location and soils. Inoculation of legumes with rhizobia strains has brought large increase in legume nodulation, grain and biomass yield, nitrogen fixation and post-crop soil nitrate levels over this period. These increases are usually highest when the inoculated legumes are grown in zero-

rhizobia or low-rhizobia soils, but minimal in soils already having high number of compatible rhizobia (GRDC, 2013). Abdul and Saud (2012) reported that Un-inoculated plants produced fewer nodules and had lower dry matter than inoculated plants, which confirmed the result obtained by Hafeez *et al.*, (1988). Accordingly, Hafeez *et al* (1988) observed that applying bacterial inoculation and using saline water with suitable concentration had obvious effect on both bacteria and plant and suggested that salinity had an indirect effect on biological nitrogen. Lamptey *et al.* (2014) also reported that Rhizobium inoculated soybean plots produced significantly higher growth characters especially high number of nodules (245), fresh shoot weight (1906 g) and dry shoot weight (52.94 g) while in un-inoculated check plots, the number of nodules, fresh shoot weight and dry shoot weight recorded 152, 1201g and 33.37 g respectively. Zoundji *et al.*, (2015) reported that inoculation with different Bradyrhizobia strains improved significantly ( $p < 0.001$  to  $p < 0.05$ ) height, grain, biomass yield, nodulation, and nitrogen uptake of soybean but less than treatments where phosphorus application was combined to rhizobial inoculation.

The use of rhizobia inoculants in the establishment of legumes is now widely recognized, especially in areas where indigenous nodulation has been found to be inadequate. (Zarin Fatima *et al.*, 2007) It has been reported that soybean inoculated with different rhizobial strains react differently in the growth, yield and nitrogen fixation (Mmbaga *et al.*, 2014). Soybean is mainly nodulated by Bradyrhizobium japonicum and B el-Kanii. (Barros de carvalles, 2013) but most soils usually lack these specific strains. Therefore inoculation of seeds with relevant strains of bacteria before sowing is important especially if the crop is to be grown for the first time on the land.

Inoculation of soybean seeds with appropriate rhizobia strains provides high number of viable effective rhizobia around the root region which allows rapid colonization and nodulation. (Deaker *et al.*, 2012; Lemus 2012). Some experiment have justly shown the positive effect of inoculation on soybean nodulation and consequently on growth. (Kadiata *et al.*, 2012; Solomon *et al.*, 2012; Yamakawa *et al.*, 2013).

Fitsum *et al.*, (2016) reported that the two season experiments analysis of variance indicated that there was significant difference ( $P < 0.05$ ) among treatments in biomass and grain yields. The 1st experiment, he reported had the highest biomass and grain yields respectively were rhizobial strain legume fix was used (2804.7 and 1245.9 kg ha<sup>-1</sup>) followed by MAR-1495 (2628.7 and 1182.5 kg ha<sup>-1</sup>).

## **2.3 Factors Affecting Inoculation**

### **2.3.1 Inoculant Quality**

The quality of an inoculant depends on the number of live and infective rhizobia in it. Enumeration methods require that the inoculant be diluted serially. Several dilutions are then selected for counting. For inoculants based on sterile carriers, aliquots of these dilutions can be spread onto plates containing solid growth medium. The resulting rhizobia colonies can then be counted. For inoculants based on non-sterile carriers, this method is not practical because other microorganisms present interfere with the plate count. Aliquots of the serial dilution are therefore pipetted onto the roots of seedlings which have been grown aseptically. The nodulation ability of these dilutions will then give information for an estimate of the number of rhizobia present.

Just how important are rhizobia numbers? Roughley *et al.* (1993), in a field study of the narrow-leaved lupin, reported that increasing the numbers of rhizobia applied to the seed from  $1.9 \times 10^4$  to  $1.9 \times 10^6$  increased nodule number from 8 to 26 per plant; nodule weight from 65 to 393 mg/plant; % plants nodulated from 89 to 98%; shoot dry matter from 7.8 to 9.0 t/ha and, most importantly, grain yield from 1.9 to 2.1 t/ha (i.e. a 10 % increase). The responses to increasing numbers of inoculant rhizobia were almost linear through a range of just 2 per seed to the highest rate of  $1.9 \times 10^6$  per seed.

### **2.3.2 The Population and Symbiotic Effectiveness of Indigenous Rhizobia**

When rhizobia live in the soil, they are called saprophytes. Many soils contain rhizobia that live on soil organic matter, without legume partners. These are called native rhizobia, while those that farmers add as inoculants are called introduced rhizobia. The population of native rhizobia in any soil can be very diverse, including several species, and many distinct strains within each species. Numbers can range from zero to more than a million rhizobia per gram (g) of soil. Several factors affect the number of rhizobia in the soil. These include vegetation, cropping history and environmental and soil conditions.

### **2.3.3 Rhizobium Symbiosis of Legume**

Obviously the requirement for a legume to form an effective N<sub>2</sub> fixating symbiosis is the ability to form nodules with necessary organization and ancillary machinery for N<sub>2</sub> fixation (Gibson, 1988) number of genes for which formation of N<sub>2</sub> fixing nodules has been identified (Nap *et al.*, 1990) and limited understanding of genetics of legumes nodulation has largely been gained by the study of plant genotypes which have lost the ability to form a nodule is not enough and many other plant characters will markedly influence the amount of Nitrogen fixed in the symbiosis. It

has been suggested that increasing the amount of Nitrogen fixed in Soybean and the proportion of N derived from fixation may only be achieved with concomitant yield increase. (Herridge *et al.*, 1988).

#### **2.3.4 Legumes Nodulation and Symbiosis Estimation**

The problem faced by farmers in the moist savanna in West Africa is the small capacity of the soil to supply the quantities of Nitrogen required for food production. Lal (1989) indicated the rapid decline of available N once cropping commences. Van *et al.*, (2008) reported that moist savanna soils must supply 15Kg N ha and 2Kg P ha for each of ton of maize grain produced. However since Nitrogen fertilizer are expensive and inaccessible to subsistence tropical farmers, the emphasis now is on biological Nitrogen fixation by leguminous plants as the best option to the maintenance of soil fertility level. Sanginga *et al.* (1996) reported a large variability in nodulation and growth of mucuna pruriens grown in farmers' fields in derived savanna in Benin, west Africa. Nodulation did not occur in 40% of the field, indicating that some of these legumes might behave as non- fixers depending solely on available N, if they are not effectively nodulated. Soybeans obtain up to 70 percent of their total nitrogen requirement from biological nitrogen fixation conducted by Rhizobia bacteria colonies (nodules) living on soybean roots. If the nodules fail to form, the plants will become deficient in nitrogen and significant yield reductions can occur. (Afolabi *et al.*, 2014).

### **2.3.5 Effect of Phosphorus and Inoculation on Growth Yield and N<sub>2</sub> Fixation by Legumes**

Nitrogen is not always the primary limiting factor and when it is not, there will not be a response to inoculation (Eagle Sham, 1989). Apart of nitrogen, phosphorus (P) is the second major plant growth-limiting nutrients in most agriculture soils (Shahid *et al.*, 2009). Phosphorus is quite abundant in soil but it reacts readily with iron, aluminium and calcium to form insoluble compounds, resulting in very low phosphorus availability (Zarrin *et al.*, 2006). It plays an important role in the plant's energy transfer system since its deficiency retards growth (Shahid *et al.*, 2009). Symbiotic nitrogen fixation needed high phosphorus as large amounts of energy being consumed during the process of photosynthesis, or energy generating metabolism depends on the availability of phosphorus (Schulze *et al.*, 2006). Through its basic functions in plants as an energy source, phosphorus affects nodule development, production of protein, phospholipids and phytin in grains legume (Rahman *et al.*, 2008). Inadequate P restricts root growth, the process of photosynthesis, translocation of sugars and other such functions which directly influence N fixation by legume plants (Abdul-Aziz, 2013). P-supplementation can enhance plant growth by increasing the efficiency of biological nitrogen fixation, enhancing the availability of macronutrients in legumes (Makoi *et al.*, 2013). There were several reports on the interaction between Rhizobia inoculation and P supply. Ndakidemi *et al.* (2006) and Akpalu *et al.* (2014) had reported that combination of beneficial bacteria of soil and phosphorus in legume plants significantly increased nodulation, pod formation and development, and a subsequent grain yield comparatively to the single use of phosphorus or beneficial bacteria. The inoculation with Rhizobium and phosphorus supplementation improved the macronutrient uptake (N, P, and K) in different organs of the whole plant of soybean (Tairo *et al.*, 2014). Highly effective and competitive Rhizobium strains and a supply of appropriate amount of phosphorus (Scherer *et al.*,



2008) could markedly increase legume growth and nitrogen fixation. Cassman *et al.* (1981) found out that field grown to soybean has a higher P requirement when it is dependent on BNF for its N supply as compared to mineral N dependency. Soybean depends on BNF but not supplied with P attained only 28% of the maximum yield obtained at optimum P level (Keyser *et al.*, 1992). The addition of P and Inoculation improved yields in the Dominican Republic, neither different tillage practice nor phosphorus fertilizer influenced the response to inoculation. Yield of inoculated treatment were not statistically different from controls and ranged from 30 – 8% of those obtained on N fertilized plot (Huntington *et al.*, 1986). In Nigeria, Olufayo *et al.*, (1992) while investigating the effect of inoculation and phosphorus application on the growth and biological nitrogen fixation of soybean, reported that nodule formation was low when no P was recorded for nodules dry weight at flowering even though the response did not follow a consistent trend.

### **2.3.6 Vegetation and Cropping History**

In most agricultural soils, the presence of vegetation, legume or non-legume encourages large number of rhizobia. Rhizobia are found in large numbers in the region close to the roots of plants, known as the rhizosphere. Non-legume vegetation also can encourage native rhizobia, the largest numbers of rhizobia are generally found in areas with wild or cultivated legumes. The particular species of native rhizobia present in an area depends on the legume grown in the soil. When a new crop is introduced to an area, it may take some time to build up the soil rhizobia population. A survey in Lampung and Sumatera Barat, Indonesia provides an example (Singleton *et al.*, unpublished data). Among fields where soybeans had grown for only one year, none had more than 100 soybean rhizobia per gram of soil. After two or more years of soybean cropping, 30 % of the fields tested still had fewer than 100 soybean rhizobia per gram soil. Where numbers of native

rhizobia are this low, farmers are advised to apply rhizobia inoculant to benefit fully from biological nitrogen fixation.

### **2.3.7 Soil Fertility Challenges**

Soil fertility challenges can be very serious, severe soil degradation can lead to such strong constraints that even legumes perform poorly and are unable to produce sufficient biomass. (Jonas *et al.*, 2011). Africa exhibit the lowest rate of fertilizer use in the world with an average consumption estimated at 8.3Kg ha<sup>-1</sup> (Morris *et al.*, 2012; Sanginga *et al.*, 2009).

### **2.3.8 Level of Nitrogen (N) in the Soil**

Nitrogen fertilizer severely depressed nodulation in this cultivar. This is however consistent with the work of Eaglesham (1989) who reported that the increasing levels of mineral N in the rhizosphere inhibits soybean nodule formation and functioning. Increasing levels of N application reduced nodulation of vegetable soybean. Pod/plant and green pod yields were the highest at 25 kg N/ha, application of 100 kg N/ha produced the lowest pod number and pod yield while pod yields decreased slightly at 50 kg N/ha.

Soybean can fix atmospheric nitrogen for the restoration and maintenance of soil fertility in a sustainable way and consequently could improve crop yields. In fact, soybean is estimated to fix 80% of its nitrogen needs (Smaling *et al.*, 2008). Soybean N<sub>2</sub> fixation is beneficial as it provides necessary N to the plant from the atmosphere which otherwise would proceed from soil and/or manure (Chianu *et al.*, 2011). The fixation of soybean as much as 300 kg of N ha<sup>-1</sup> in addition

to the release in the soil, of 20 - 30 kg N ha<sup>-1</sup> for the following crop had been estimated (Hungria *et al.*, 2006).

### **2.3.9 Soil Temperature, pH and Soil Moisture**

Soil temperature and acidity (pH) are also important. Rhizobia prefer a soil temperature of 25° to 30 °C and a pH of 6.0 to 6.8. Rhizobia are sensitive to low pH (acid soils). One of the most important factors is rainfall. Areas with adequate rainfall often have large numbers of native rhizobia because the rhizobia themselves survive well in moist soils and also because more rainfall usually means that there are more legumes and other plants. Rhizobia prefer soils that are moist but not waterlogged.

## **2.4 Influence of Potassium Fertilizer on Soybean**

Potassium (K) is one of the three major essential nutrient elements required by plants. Unlike nitrogen and phosphorus, potassium does not form bonds with carbon or oxygen, so it never becomes a part of protein and other organic compounds (Hoefl *et al.*, 2000). Although K is not a constituent of any plant structures or compounds, it is involved in nearly all processes needed to sustain the plant life. Potassium in cell sap is involved in enzyme activation, photosynthesis, transport of sugars, protein and starch synthesis. It is known to help crop to perform better under water stress through the regulation of the rate at which plant stomata open and close. It is also known for its role to provide lodging resistance and insect/disease resistance to plants. Since potassium is involved in many metabolic pathways that affect crop quality, it is often called as “the quality element” (Dev, 1995). Potassium (K) as a nutrient affects most of the biochemical and physiological processes of plant growth and metabolism (Wang *et al.*, 2013). Its importance in the

formation of crop production and its quality is well known. Potassium plays essential roles in enzyme activation, protein synthesis, photosynthesis, osmoregulation, stomata movement, energy transfer, phloem transport, cation-anion balance and stress resistance (Marschner, 2012). The availability of potassium to the plant is highly variable, due to complex soil dynamics, which are strongly influenced by root–soil interactions. The molecules that signal low K status in plants include reactive oxygen species and phytohormones, such as auxin, ethylene and jasmonic acid. Potassium deprivation, triggers developmental responses in plant root system. All these acclimation strategies enable plants to survive and compete for nutrients in a dynamic environment with a variable availability of potassium (Ashley *et al.*, 2006). Potassium fertilization also causes responses in soybean plants. Vyas *et al.*, (2005) reported that Potassium application had significant positive effect on growth and nodulation in soybean. They reported that it favorably influenced the quality parameters of soybean as well. And there was significant reduction in insect-pests infestation and disease incidence in soybean receiving potassium dressings. Application of 25 kg basal + 25 kg K<sub>2</sub>O/ha at flowering (R2 stage) gave maximum productivity of soybean-wheat cropping system while application of 25 kg K<sub>2</sub>O/ha as basal was most economical in terms of IBCR indicating the utility of K applications for resource poor farmers (Vyas *et al.*, 2005).

Hanway *et al.*, (1971) reported that soybean takes up and accumulates K throughout the growing season. However, young seedlings of soybean do not use much potassium, but the rate of uptake climbs to a peak during the period of rapid vegetative growth. The potassium in vegetative parts is transferred to seed during pod fill process. The mature soybean seed contains nearly 60 per cent of the total K in plant (Hoeft *et al.*, 2000).

## **2.5 Impact of micronutrient and its mode of application.**

Poor availability of plant nutrients limits yield in many of the world's crop production areas (Hanway *et al.*, 1971). Application of micronutrients to crops in combination with macro fertilizers (N, P, K) was found to help break the cycle of low yields, poverty and poor human nutrition in sub-Saharan Africa (Nube *et al.*, 2006). Metals such as Zinc, iron and manganese have vital roles in plant's life cycle and very important for normal growth plants (Fageria, 2009). Zinc is considered as the most limiting factor in producing crops in different parts of the world (Fageria *et al.*, 2005). Zn is an essential catalytic component of over 300 enzymes, including alkaline phosphatase, alcohol dehydrogenase, Cu-Zn superoxide dismutase, and carbonic anhydrase (Khaleel *et al.* (1981). Zinc plays an important role in synthesizing proteins, RNA, DNA and precursor of auxin which is essential for cell elongation (Bobde *et al.* 1998). Also role in metabolism of nitrogen, synthesis of amino acid tryptophan, metabolism of starch, plants flowering and fruit set, increasing plant resistance to fungal disease and expanding plants roots (Fageria, 2009). For most plants, optimal Zn concentration is between 20 and 100 mg kg<sup>-1</sup> tissue (Fageria, 2009). Fe is the most limiting to agricultural production in the world (Bobde *et al.* 1998). Fe deficiency as chlorosis is a wide spread problem for soybean grown on alkaline, calcareous soils (Caliskan *et al.*, 2008). Iron plays an important role in nitrogen fixation and photosynthesis (Vahedi, 2011).

Synthesis of chlorophyll, thylakoid, and many ferrous proteins is dependent on this element (Imsande, 1998). Iron deficiency in plants is caused by factors that either inhibit its absorption and translocation or impair its utilization in metabolic processes (Vahedi, 2011). Application of 2.5 mg Fe kg<sup>-1</sup> soil not only increased top dry weight but also increased mean uptake of by 35.4 % in different genotypes of soybean (Ghasemi-Fasaei *et al.*, 2003). Foliar spraying is a new method for

crop feeding, which micronutrients in form of liquid are used into leaves (Nasiri *et al.*, 2010). Foliar application of microelements is more beneficial than soil application. Since application rates are lesser as compared to soil application, same application could be obtained easily and crop reacts to nutrient application immediately (Zayed *et al.*, 2011). Undoubtedly, soybean higher yield and quality as well as its oil will be obtained by microelements foliar spraying (Vahedi, 2011). Foliar spraying of microelements is very helpful when the roots cannot provide necessary nutrients (Kinaci *et al.*, 2007; Babaeian *et al.*, 2011). Narimani *et al.* (2010) reported that microelements foliar application improve the effectiveness of macronutrients. It has been found that microelements foliar application is in the same level and even more influential as compared to soil application. It was suggested that micronutrients could be applied successfully to compensate shortage of those elements (Arif *et al.*, 2006). These authors found that based on soil properties, foliar spraying could be effective 6 to 20 times as compared to soil application. Resistance to different stress will be increased by foliar application of micronutrients (Ghasemian *et al.*, 2010). Since in field situation, soil features and environmental factors which affect nutrients absorption are extremely changeable, foliar application could be an advantage for crop growth (Seifi Nadergholi *et al.*, 2011). Also, effectiveness of foliar spraying is higher and the cost of foliar application is lower as compared to soil application (Yassen *et al.*, 2010). Symptoms associated with most micronutrients deficiency in plants include inter-veinal chlorosis, necrotic spots on leaves, rosetting of leaves and stunting of plants due to disturbances in metabolism of auxins, especially indole-acetic acid, which is a growth hormone (Alloway, 2008) .

## **2.6 Influence of Organic Matter on Soybean**

A number of research studies showed that organic farming ensures better yields at low cost and fetches more income (Mitchell *et al.*, 2007). Taie *et al.* (2010) confirmed that application of organic fertilizer using different compost levels with bio-fertilizer to soybean plants resulted to the highest values of total phenolics and flavonoids as compared to chemically grown counterpart. Few studies have been conducted on the evaluation of the influence of the application of the various soil organic amendments on the antioxidant components in broccoli. (Ali *et al.*, 2014)

Organic manures provide a good substrate for the growth of microorganisms and maintain a favorable nutritional balance and soil physical properties. One of such strategy to maintain soil fertility for sustainable production of soybean is through judicious use of fertilizers (Bobde *et al.*, 1998) coupled with organic resources that to achieve sustainability in production, the use of organic manures alone is not sufficient (Prasad, 1996). It has also been brought out that the use of organic manures in integration with fertilizers meets the need of micronutrients of soybean (Joshi *et al.*, 2000). Lourduraj (2000) has also reported that the combined application of inorganic and organic manures significantly enhanced the growth attributes and yield of soybean as compared to the sole application of either of them.

## **2.7 Functions of Organic Matter**

### **2.7.1 Soil Structure and Aggregate Stability**

Soil structural stability refers to the resistance of soil to structural rearrangement of pores and particles when exposed to different stresses (e.g. cultivation, trampling/compaction, and irrigation). The interrelationship between SOC and soil structure and other physical properties has been extensively studied, and excellent reviews can be found in Tisdall *et al.*, (1982), Oades (1984), and Carter *et al.*, (1996). It is well established that addition of SOM can not only reduce bulk density (Db) and increase water holding capacity, but also effectively increase soil aggregate stability. Angers *et al.*, (1996) noted that the amount of water-stable aggregates (WSA) was often associated with SOC content, and that particularly labile carbon was often positively related to macro-aggregate stability. Kay *et al.*, (1999) reported that a minimum of 2 % SOC was necessary to maintain structural stability and observed that if SOC content was between 1.2-1.5 %, stability declined rapidly. Boix-Fayos *et al.*, (2003) showed that a threshold of 3-3.5 % SOC had to be attained to achieve increases in aggregate stability; no effects on aggregate stability were observed in soils below this threshold. However, there is no general agreement as to the type of organic matter essential for aggregation. This is most likely due to the fact that different types of organic matter perform different functions at different times during the aggregate formation and conservation process. In fact, Kay *et al.*, (1999) suggested that most or all SOC fractions were involved to different degrees in aggregate formation and stabilization. Manure as well as fertilizer (Nitrogen-Phosphorus-Potassium = NPK) applications are often used to maintain or even enhance the ability of soil to produce arable crops. In the long-term, increased crop yields and SOM returns with regular application result in higher SOM content and biological activity (Haynes *et al.*, 1998). The effects of different SOM and fertilizer treatments on soil structure and organic matter content



have been investigated in various studies. Aoyama *et al.* (1999) followed the effects of long-term (18years) application of manure and NPK fertilizer on organic matter fractions and water stable aggregates in the 0-10 cm of a humic gleysol. Manure application (20 Mg ha<sup>-1</sup> yr<sup>-1</sup>) led to an increase in carbon content in most fractions and increased the pools of protected carbon (x3) and nitrogen (x4), located in small macro-aggregates (250-1000  $\mu$ m). In contrast, NPK fertilizer only increased the pool of macro-aggregate protected nitrogen (x2.5) but not that of carbon. They concluded that manure application, compared with sole application of NPK, contributed to the accumulation of macro-aggregate protected carbon and nitrogen and provided a mechanism for protection of labile soil organic matter in annually tilled cropping systems. The effect of organic amendments on SOM and clay dispersibility was investigated by Debosz *et al.* (2002). Sandy loam was amended with anaerobically digested sewage sludge and household compost and incubated for 11 months at constant temperature (10 °C). They found that clay dispersibility of the un-amended soil increased, indicating progressive destabilization of soil structure, whereas waste-amended soil remained at initial levels. Biomass C increased by only 0.2 % (sludge) and 1 % (compost), suggesting that the effects of organic matter on soil properties are more likely to be related to quality not quantity.

### **2.7.2 Water-Holding Capacity**

An important indicator of soil physical fertility is the capacity of soil to store and supply water and air for plant growth. The ability of soil to retain water is termed water holding capacity (WHC). The amount of plant-available water in relation to air-filled porosity at field capacity (FC) is often used to assess soil physical fertility (Peverill *et al.*, 1999). Total plant available water (PAW) is the amount of water held between the wettest drained condition (FC, at matric suction of -10 kPa) and the water content at which plants are unable to extract water (permanent wilting point (PWP), at

matric suction of -1500 kPa). However, some studies use -10 kPa for coarse textured soils only and use -33 kPa for fine-textured soils (Bauer *et al.*, 1992). WHC of soils is controlled primarily by the number of pores and pore-size distribution of soils, and by the specific surface area of soils. In turn, this means that with an increase in SOC content, there is increased aggregation and decreased Db, which tend to increase the total pore space as well as the number of small pore sizes (e.g. Khaleel *et al.*, 1981; Haynes *et al.*, 1998). These relationships highlight the interconnectivity between soil structure, Db and WHC.

Increases in SOC can be achieved by adding organic matter (as manure, plant residues or sewage sludge) to the soil and positive effects of organic amendments on WHC have been reported by Khaleel *et al.* (1981) and Haynes and Naidu (1998). Similar to the results obtained from Toma *et al.* (2004), Khaleel *et al.* (1981) found that the relative increase in WHC became smaller as the amount of organic matter from amendments increased.

### **2.7.3 Soil Color**

Soil colour is often used as the highest categorical level in many soil classification systems, e.g. the concept of the Russian chernozem was centered around the thick dark soils of the Russian steppe and the Mollisol order of the US soil taxonomy is specifically defined to include most soils with relatively thick, dark surface horizons (Schulze *et al.*, 1993). Generally good soil conditions are associated with dark brown colours near the soil surface, which is associated with relatively high organic matter levels, good soil aggregation and high nutrient levels (Peverill *et al.* 1999). Schulze *et al.* (1993) found that within similar landscapes and soil texture classes, there was a good linear correlation between Munsell soil colour and SOM for Ap horizons from Indiana and Illinois.

The effect of usually dark brown or black SOM on soil colour is important not only for soil classification purposes, but also for ensuring good thermal properties, which in turn contribute to soil warming and promote biological processes (Baldock *et al.*, 1999). Only about 10 % of the solar energy reaching the earth's surface is actually absorbed by the soil, which can be in turn used to warm the soil. Naturally, dark-coloured soils absorb more energy than light-coloured ones. However, this does not imply that dark-coloured soils are always warmer: since dark-coloured soils usually have a higher amount of organic matter, which holds comparatively larger amounts of water, a greater amount of energy is required to warm darker soils than lighter-coloured ones (Brady, 1990). Thus, the thermal property of soil is to a large degree influenced by water content, Db, soil texture (fine versus coarse) and soil colour. In addition, the surface cover of soil affects the heat transfer in and out of a soil as bare soils warm up and cool off more quickly than those with a vegetation or mulch cover.

#### **2.7.4 Cation Exchange Capacity**

Cation exchange capacity (CEC) is defined as the measure of the total capacity of a soil to hold exchangeable cations and indicates the negative charge present per unit mass of soil (Peverill *et al.*, 1999). A high CEC is regarded as favourable as it contributes to the capacity of soils to retain plant nutrient cations. CEC is most commonly expressed as centimols of positive charge per kilogram of soil (cmol/kg), which provides values that are numerically equivalent to the previous conventional unit of mequiv./100g. Soils can have permanent and variable charge. Permanent charge is derived from certain clay minerals (e.g. smectite) when Mg is replaced by Al or Si is replaced by Al. The strength of variable charge (provided by clay minerals and organic matter)

depends on ionic strength and pH and is therefore influenced by the chemical environment of the soil.

Soil buffering is considered to be an important aspect of soil health, as it assures reasonable stability in soil pH (preventing large fluctuations) and influences the amount of chemicals (lime or sulfur) needed to change the soil pH. The BC of a soil is defined as its resistance to changes in pH when an acid or base is added. Buffering at intermediate pH values (5-7.5) is mainly governed by exchange reactions where clays and functional groups of SOM act as sinks for H<sup>+</sup> and OH<sup>-</sup>. The relationship of pH to percent base saturation varies from substance to substance. For example, different types of clay will affect the pH-base saturation to different degrees, and Al and Fe compounds are known to affect the BC of soils. At low pH values, Al<sup>3+</sup> and hydroxyl aluminum tend to block exchange sites in silicate clays and humus, thereby reducing the CEC of the colloids. As a consequence, liming is required to raise the pH and increase the CEC (Brady, 1990)

### **2.7.5 Soil Organic Matter as a Source of Energy**

Baldock *et al.*, (1999) stressed that one of the most fundamental functions of SOM was the provision of metabolic energy which drives soil biological processes. In essence, it is the transformation of carbon by plant, micro- and macro-biological processes that provides energy and results in the establishment of a cycle that connects above- and belowground energy transformations. Anderson (1995) suggested that the proposition “that natural systems have a tendency to self-organise in order to maximise useful power (i.e. store more energy that in turn can be fed back to catalyse the inflow of additional energy) (Veizer, 1988)” can be applied to soil.

Ecosystems. Thus, the transformation from relatively labile SOM into increasingly complex, stabilized SOM can be viewed as a form of energy conservation as the largest pool of SOM consists of the humus pool which is recalcitrant enough to endure in an edaphic environment for longer periods of time but still allows for decomposition and nutrient release to take place.

### **2.7.6 Soil Organic Matter as a Source of Nutrients**

SOM is an important source of nutrients for plants in general and crops in particular. Nitrogen, phosphorus and sulphur are considered macronutrients, essential micronutrients are iron, manganese, zinc, copper, boron, molybdenum, and chlorine and beneficial but not essential elements are silicon, vanadium, cobalt and nickel. Particular emphasis will be placed here on the role of macronutrients. Most of the nutrients in SOM are derived from the mineralization of SOM and become available for plant uptake during decomposition and for this reason, the particulate organic matter fraction is often considered the most important proportion of SOM in providing nutrients to plants (Wolf *et al.*, 2003). Losses of nutrients might occur via leaching or conversion to gaseous forms or are the result of immobilization. Due to the conversion of energy from primary sources to heterotrophic organisms, mineralization of complex organic molecules by primarily microbial processes is possible. Some soil nutrients are used in the synthesis of new biomass, some are immobilized and another portion is mineralized and released as plant-available forms into the soil mineral nutrient pool. With the exception of fertilizers, SOM provides the largest pool of macro-nutrients with >95% of N and S and 20-75% of P found in SOM (Duxbury *et al.*, 1989; Baldock *et al.*, 1999). A systematic review of the contents, chemical structures and transformations of N, P and S can be found in Baldock and Nelson (1999). Only about 40-50 % of organic N is identifiable and quantifiable as amino acids and amino sugars and the remaining portion consists

of unidentifiable structures. The principal form of organic S added to soil is amino acid S (methionine, cysteine, cystine), which accounts for up to 30% of the total organic S pool. About 30-80 % of extractable organic P is in the form of monoesters, and phosphate esters of inositol are the most abundant identifiable P compound class (5-80 %). Inositol may form insoluble precipitates with Fe, Al and C and adsorb onto Fe and Al oxide surfaces. At low soil pH (<4.5-5), precipitates of Al and Fe phosphates may form while at higher pH values (>6-6.5), Ca phosphates form; however, mostly, specific adsorption reactions control P concentrations in soil solution (Duxbury et al., 1989). While C is required as a primary source of energy for the mineralization of N and C-bonded S, biochemical mineralization is necessary for the release of phosphate and sulfate via enzymatic hydrolysis. As a result, cycles of P and S are often decoupled from the C and N cycles, leading to large variations in the ratios of the respective macro-nutrients. Furthermore, the type and amount of P is often a function of inorganic parameters such as parent material and degree of weathering.

Ratios of C/N/S in agricultural soils (130:10:1) differ from those under indigenous vegetation (200:10:1). These differences could either imply that there is preferential mineralization of C in cultivated soils, or that differences exist with respect to retention of nutrients in the soil-plant systems, or that there is a higher nutrient concentration in arable soils due to fertilizer input (Duxbury et al., 1989). The N/S ratio usually varies between 6-8, and the C/N ratio of SOM, depending on the C/N ratio of the vegetation and degree of decomposition, can vary between 12-16 but may be much higher in plant litter or in environments where SOC decomposition is restricted (e.g. peats) (Baldock *et al.*, 1999; Baldock *et al.*, 2000)

## CHAPTER THREE

### 3.0 MATERIALS AND METHODS

#### 3.1 Experimental Sites

Farmers' fields located at four different sites, Kuta, (latitude 09° 53' 43" N Longitude 006° 42' 345" E) Gupe, (Latitude 09° 48' 234" N and longitude 006° 41' 868" E) Webo, (Latitude 09° 49' 591" N and Longitude 006° 45' 860" E) and Kukulu ( Latitude 09° 40' 792" N and Longitude 006° 44' 564" E) in Shiroro LGA of Niger state, Nigeria were used for this trial in 2015 cropping season, and Kukulu ( Latitude 09° 40' 794" N and Longitude 006° 44' 522" E), Gyigy ( Latitude 09° 44' 681" N and Longitude 006° 46' 659" E), Kuta ( Latitude 09° 53' 114" N and Longitude 006° 42' 330" E) and Agbolo ( Latitude 09° 52' 016" N and Longitude 006° 43' 864" E) were used for the trial in 2016 cropping season.

This zone is characterized by distinct rainy and dry seasons in a year. The rainy season lasts from April to October and the dry season between Novembers and March during the study. The mean annual rainfall in the area is about 1338mm, with the highest mean of about 300mm being in August. The temperature rarely falls below 22 °C and peaks at 40 °C between February and March with the mean being about 35 °C (Osunde and Alkassoum, 1998).

Previously before the commencement of this study, farmers in these areas cultivate their lands to maize, yam and sorghum etc. most farmers in these areas use farmyard manure while some others do not apply fertilizer as a result of the increase in price of mineral fertilizer.

### **3.2 Land Preparation**

The land area used for this experiment was cleared by slashing, marked, measured, demarcated and ridged manually. The size of each plot being 10 m x 10 m containing (14) fourteen ridges per treatment. The inter row spacing between two ridges was 0.75 m, and the spacing between each plot was 0.5m.

### **3.3 Source of Seeds, Inoculant and Fertilizers**

Seeds, inoculant and fertilizers were all sourced from IITA Ibadan. The soybean variety used was TGx 1951-3F. Legume fix, a peat based inoculant containing *Bradyrhizobium sp.* Strain 532c was manufactured by legume technology Ltd, UK. The Agrolyser, a micronutrient blend fertilizers manufactured by Cybernetic Nig. Ltd. containing Ca 20.14%, Na 1.04%, Zn 0.11%, Mg 0.19%, Cu 0.19%, S 2.72%, Fe trace, Mn trace, Bo trace, and Mo trace was used to supply micronutrient.

### **3.4 Seed Dressing**

Soybean seeds were inoculated using Legume fix inoculant. Inoculation was done by simply mixing 1 g of inoculant with 250 g of seed.

### **3.5 Agronomic Practices**

The planting method was by dibbling. Two (2) seeds per hole were planted at intra row spacing of 10 cm. Fertilizer application was done at planting using drill method. The micro nutrient (Agrolyser) was applied by foliar application, three weeks after planting, immediately after the first weeding. The first weeding was done 3 weeks after planting, the second was done 6 weeks after planting.



### **3.6 Treatments**

The experiment was laid out in a Randomized Complete Block Design. The trial consisted of six (6) treatments in the first year and nine (9) treatment in the second year with each site serving as replicate. The treatments were

- (i) Control, ( no input)
- (ii) Inoculant only,
- (iii) Inoculant + phosphorus (30 kg P<sub>2</sub>O<sub>5</sub>/ha),
- (iv) Inoculant + phosphorus + potassium (20 kg K<sub>2</sub>O/ha),
- (v) Inoculant + phosphorus + potassium + micronutrients (3.3 kg/ha) and
- (vi) Inoculant + phosphorus + potassium + micronutrients + organic manure (4 tons/ha).
- (vii) Inoculant to organic matter
- (viii) Inoculant + phosphorus +Micronutrient
- (ix) Organic matter only

In the first year of the experiment, the first six (6) treatments were applied. Following the trend of the results obtained in the first year, there was a need to investigate more options by adding three (3) more treatments to determine their different effects on the farmer's field.

### **3.7 Soil Sampling and Preparation**

Soil samples were collected randomly at 5 different points within the field at a depth range of 0 – 20 cm. the samples were bulked in a bucket, a portion of the bulked soil was taken and put in a plastic bag, labeled and taken to the laboratory. The soil at the laboratory was air dried at room temperature by spreading it out in a well-ventilated room, protected from rain and contamination. When the soil is dusty it is dry enough for use. Soil lumps were gently crushed so that the gravel

and roots are separated from the mineral soil. The soil was sieved through a 2mm and 0.5mm sieve leaving the gravel and roots etc. in the sieve and stored inside plastic bag in a cool dry place. Soil pH was determined using glass electrode (pH meter) method in water, Nitrogen was determined using micro-Kjeldahl method, phosphorus was determined by Mehlich method, Organic carbon by Walkley Black wet oxidation method, Particle size analysis was determined by Bouyoucos hydrometer method, exchangeable acidity was determined by KCL extraction method, and Cation Exchange Capacity. The analyses were conducted following the procedures written in Laboratory methods of soil and plant analysis by Okalebo *et al.* (2002).

### **3.8 Observation and Data Collection**

The following data were collected during this trial.

i. **Plant Height (cm)**

Plant height was taken with the aid of a meter rule, measured from the ground level of the plant to the first flag leaf measured in centimeters at 4, 6 8 10 and 12 weeks after sowing.

ii. **Biomass Weight (g)**

The shoot of soybean was cut at mid-flowering from the ground level using a sharp knife and put into an envelope, labeled and taken to the laboratory for weighing and oven drying. Fresh weight was taken by the use of a sensitive weighing balance, and afterward oven dried at a constant temperature of 70-72 degrees Celsius for 72 hours to obtain the dry weight.

**iii. Determination of Nodule Number/ Weight**

Five plants were carefully uprooted with the aid of shovel from the soil. The nodule numbers were determined at mid-flowering by picking out the fresh nodules after gently washing the sand off the roots. The number of nodules was determined by physically counting the visible nodules. The nodules per each plot was kept in labelled paper envelopes and the fresh weight determined in grams (g) using the electronic weighing scale. It was then oven dried at the temperature of 45 °C for 12 hours to a constant weight for dry weight determination

**iv. Number of pods**

The number of pod was determined at the maturity stage of the plant (at harvest), the pods were carefully plucked off from the branches of the soybean plant, counted, put into a medium sized envelope and labelled. .

**v. Number of Branches**

The operation was carried out by counting the number of branches present on the randomly selected plant stands.

**vi. Days to 50% flowering**

This was an observation, when the flower on the plants generally reaches 50 %, that is to say, when half of the soybean plant in each plot has developed flowers. The dates were recorded and calculated by counting from the date of sowing up to the date 50 % flowering was observed, that gave the total number of days to 50% flowering.

**vii. 100 seed weight (g)**

After harvest and threshing, 100 grain of soybean were counted out and weighed separately using the sensitive scale.

**viii. Grain yield**

This was determined by weighing the total quantity of grains obtained from the net plot of the harvested area of each treatment.

**Statistical Analysis**

Data obtained were subjected to analysis of variance (ANOVA) using the Statistical Analysis System (SAS) version 9.2 software (SAS, 2009). The least significant difference (LSD) test was used to separate the treatment means at 5% level of significance.

## CHAPTER FOUR

### 4.0 RESULTS

#### 4.1 Soil Characteristics of the Study Area 2015

The results of the initial soil characteristics of the study area for 2015 and 2016 are presented in Table 4.1 and 4.2. The soil textural class for Kukulu, Webwo, Kuta and Gupe were loam, silty loam, sandy loam and sandy loam respectively. The pH (in water) ranged from 5.6 in Kuta to 7.0 in Webwo i.e moderately acidic to neutral. Total carbon ranged from 0.35 g kg<sup>-1</sup> in Gupe to 10.6 g kg<sup>-1</sup> in Webwo which is very low to moderate. Total nitrogen ranged from 0.27 g kg<sup>-1</sup> in Gupe to 1.65 g kg<sup>-1</sup> in Webwo i.e very low to medium, Phosphorus ranged from 2.88 to 5.33 i.e very low to low. Potassium ranged from 0.06 cmol/kg to 0.57 cmol/kg i.e low to high. Sodium ranged from 0.58 cmol/kg to 4.52 cmol/kg i.e high. Magnesium ranged from 0.02 cmol/kg to 0.49 cmol/kg i.e low to medium. Zinc ranged from 0.68 mg kg<sup>-1</sup> to 11.66 mg kg<sup>-1</sup> i.e very low to high. Copper (Cu) ranged from 0.28 mg kg<sup>-1</sup> to 1.35 mg kg<sup>-1</sup> i.e low to high. Manganese (Mn) ranged from 24.09 mg kg<sup>-1</sup> to 97.41 mg kg<sup>-1</sup> i.e high. Iron Fe ranged from 58.88 at Kukulu to 98.16 mg kg<sup>-1</sup> i.e high

#### 4.2 Soil Characteristics of the Study Area 2016

Textural class for Kukulu, Gwada, Kuta and Agbolo was sandy clay loam across sites. P<sup>H</sup> (in water) for ranged from 5.1 in Kukulu to 5.9 in Gwada i.e strongly acidic to moderately acidic. Total carbon range was from 5.0 (gkg<sup>-1</sup>) in Kuta to 7.9 (gkg<sup>-1</sup>) Agbolo indicating that the carbon content was low in the soil. Total nitrogen ranged from 0.539 (gkg<sup>-1</sup>) in Kukulu to 0.939 (gkg<sup>-1</sup>) in Agbolo i.e very low to low. Phosphorus ranged from 2.28 to 6.38 i.e very low to low. Potassium ranged from 0.02 in Agbolo cmol/kg to 0.07 (cmol/kg) in Gyigy i.e low. Sodium ranged from

0.07 (cmol/kg) in Kukululu to 0.09 (cmol/kg) in Kuta i.e low. Magnesium ranged from 0.22 (cmol/kg) in Kukululu to 1.07 (cmol/kg) in Kuta i.e low to high. Zinc ranged from 5.35 mg kg<sup>-1</sup> in Kuta to 7.70 (mg kg<sup>-1</sup>) in Gyigyí i.e high. Cu ranged from 2.04 mg kg<sup>-1</sup> in Gyigyí to 3.01 mg (kg<sup>-1</sup>) in Kuta i.e high. Mn ranged from 28.16 mg kg<sup>-1</sup> to 86.09 (mg kg<sup>-1</sup>) i.e high. Iron (Fe) ranged from 50.24(mg kg<sup>-1</sup>) in Gyigyí to 76.17(mg kg<sup>-1</sup>) in Agbólo which was high.

### **4.3 Plant Height**

The response of promiscuous soybeans to rhizobia inoculation in combination with mineral fertilizer and organic manure on plant height in 2015 and 2016 cropping season is presented in Fig. 1 and Fig. 2. Nutrient combination had no significant ( $p \leq 0.05$ ) effect on plant height of soybean at 4WAS in 2015 but significant at 6, 8, and 10WAS respectively. The application of I+P+K+M+OM consistently produced the tallest plant at 6, 8 and 10 WAS compared to the other treatment and the control

In 2016, at 4WAS similar taller plants were observed in plots given all the nutrients except that with OM which produced the shortest plants. At 8WAS application of I+P+K+M+OM resulted in taller plants, which were in turn similar to all other plots except those given I only and I+OM. At 12WAS, plots given I+P+K+M recorded the tallest plants than plots only given I only and OM respectively.

**Table 4.1 Physical and Chemical Properties of the Soil Sample Before Planting 2015 Cropping Season**

Soil parameters	Location			
	Kukulu	Webwo	Kuta	Gupe
Sand (g/kg)	660	440	780	640
Silt (g/kg)	220	380	100	180
Clay (g/kg)	120	180	120	180
Textural class	loam	Silty loam	sandy loam	sandy loam
pH H <sub>2</sub> O	6.7	7	5.6	5.8
Total Carbon (g/kg)	5.8	10.6	5.1	3.5
Total Nitrogen (g/kg)	0.59	1.65	0.31	0.27
P (Mehlich) (mg/kg)	4.72	5.33	5.13	2.88
ECEC (cmol/kg)	2.92	5.98	0.73	0.96
Exch. K (cmol/kg)	0.16	0.57	0.06	0.07
Exch. Na (cmol/kg)	2.25	4.52	0.58	0.77
Exch. Mg (cmol/kg)	0.49	0.03	0.02	0.02
Zn (mg/kg)	1.26	11.66	0.89	0.68
Cu (mg/kg)	1.35	1.12	0.43	0.28
Mn (mg/kg)	24.09	97.41	74.15	58.09
Fe (mg/kg)	58.88	94.14	98.16	60.63

**Table 4.2 Physical and Chemical Properties of the Soil Sample Before Planting 2016 Cropping Season**

Soil parameters	Location			
	Kukulu	Gyigyi	Kuta	Agbolo
Sand (g/kg)	700	580	700	700
Silt (g/kg)	50	50	70	70
Clay (g/kg)	250	370	230	230
Textural class	SCL	SCL	SCL	SCL
pH H <sub>2</sub> O	5.1	5.9	5.4	5.2
Total Carbon (g/kg)	6.00	5.90	5.00	7.90
Total Nitrogen (g/kg)	0.52	0.64	0.65	0.93
P (Mehlich) (mg/kg)	4.53	6.38	2.28	3.51
ECEC (cmol/kg)	1.55	2.90	2.99	3.22
Exch. K (cmol/kg)	0.05	0.07	0.03	0.02
Exch. Na (cmol/kg)	0.07	0.07	0.09	0.08
Exch. Mg (cmol/kg)	0.22	0.97	1.07	1.04
Zn (mg/kg)	5.35	7.70	5.35	5.75
Cu (mg/kg)	3.01	2.04	3.01	3.01
Mn (mg/kg)	86.09	44.03	28.16	53.90
Fe (mg/kg)	57.06	50.24	67.30	76.17

**SCL = Sandy clay loam**



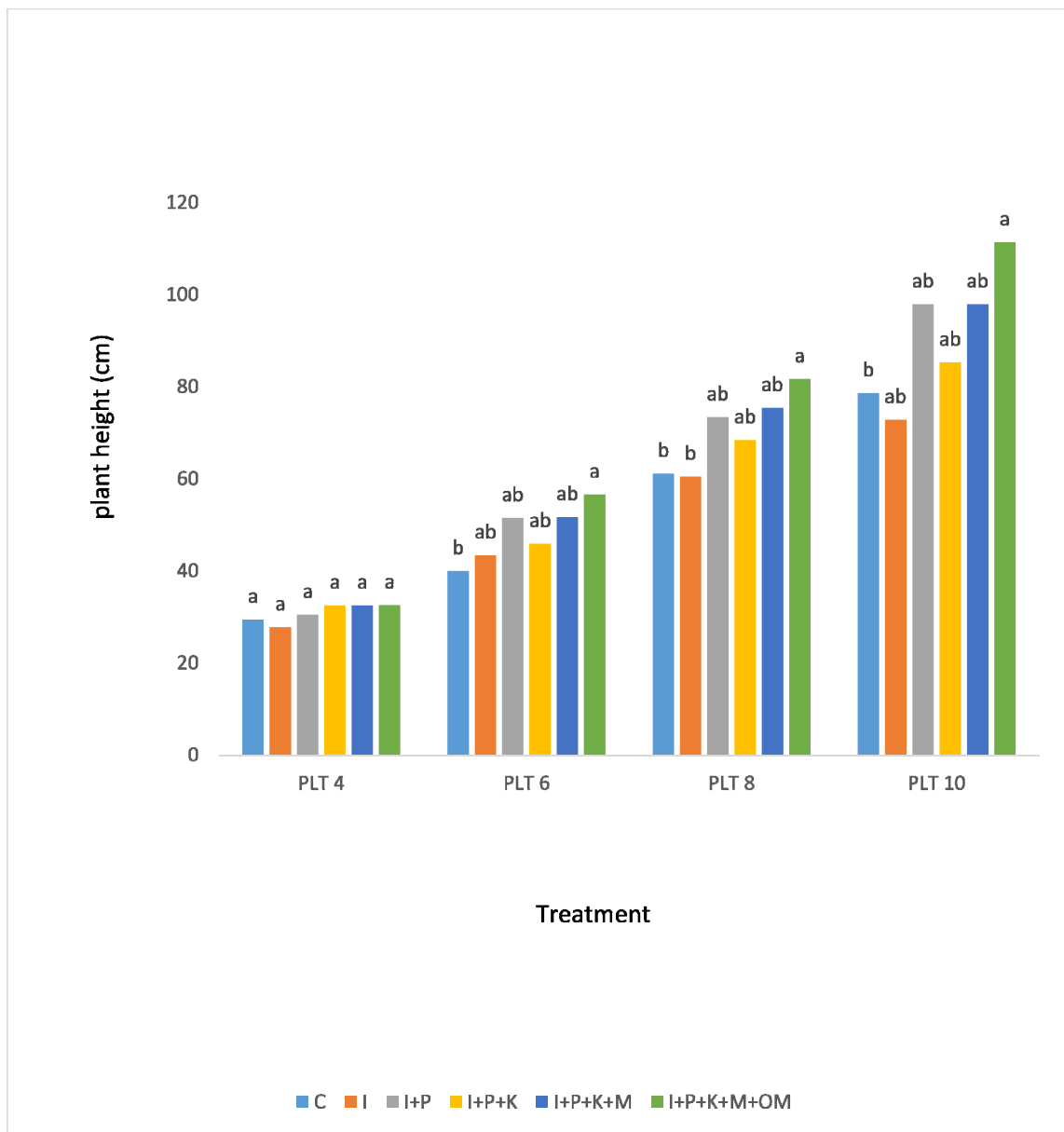


Figure 1: Effect of treatment combination on plant height at different growth stage in 2015  
 C = control, I = Inoculant only, P= phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.



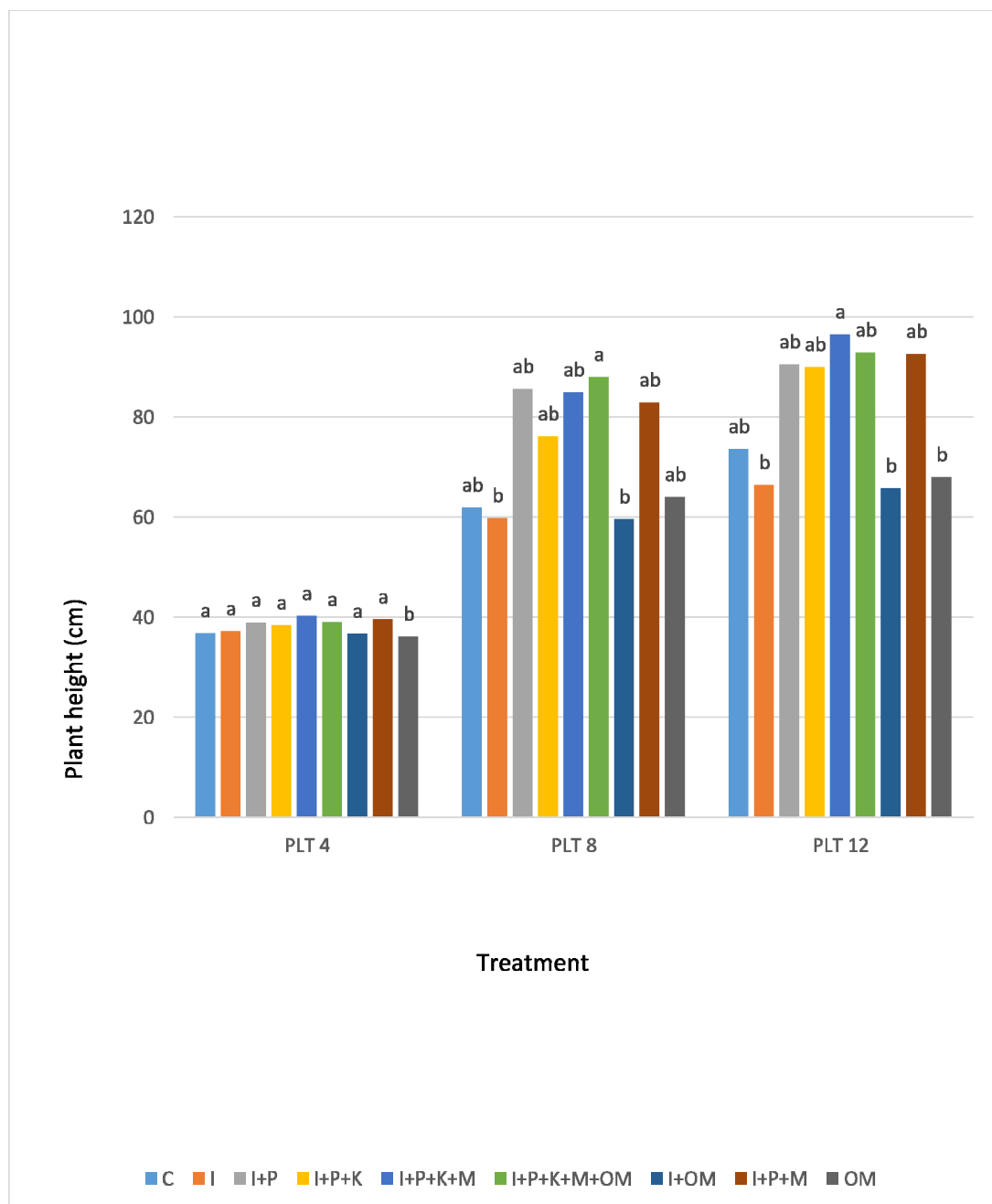


Figure 2: Effect of treatment combination on plant height of promiscuous soybean in 2016

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

#### **4.4 Shoot Biomass and 50% Flowering**

The response of promiscuous soybean to rhizobia inoculation in combination with organic and mineral fertilizers on shoot biomass of soybean is shown in Fig. 3 and 4. Nutrient combination had a significant effect ( $p \leq 0.05$ ) on soybean shoot biomass in 2015. The application of I+P+K+M produced the heaviest shoot biomass this was similar with other nutrient plots but differed to plots only given inoculant only and control which produced the lightest shoot biomass. In 2016 nutrient management had no significant effect ( $p \leq 0.05$ ) on soybean shoot biomass in this study.

The response of promiscuous soybean to rhizobial inoculation in combination with organic and mineral fertilizer on days to 50% flowering in 2015 and 2016 is shown in Fig. 5 and 6. Nutrient combination had a significant ( $p \leq 0.05$ ) effect on days to 50% flowering of soybean. In 2015, delay was observed in flowering of soybean plant in plot given I+P, I+P+K, I+P+K+M and I+P+K+M+OM, than plot given C and I only. That was not the case in 2016 plot given I only had delay in flowering than plots given I+P+M and OM.

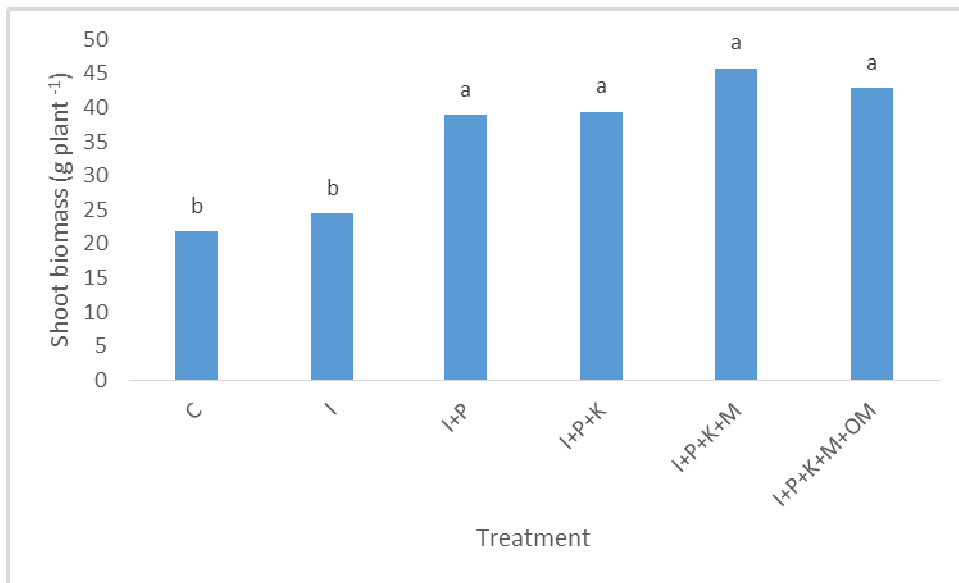


Figure 3: Effect of treatment combination on shoot biomass of promiscuous soybean at mid- flowering in 2015

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

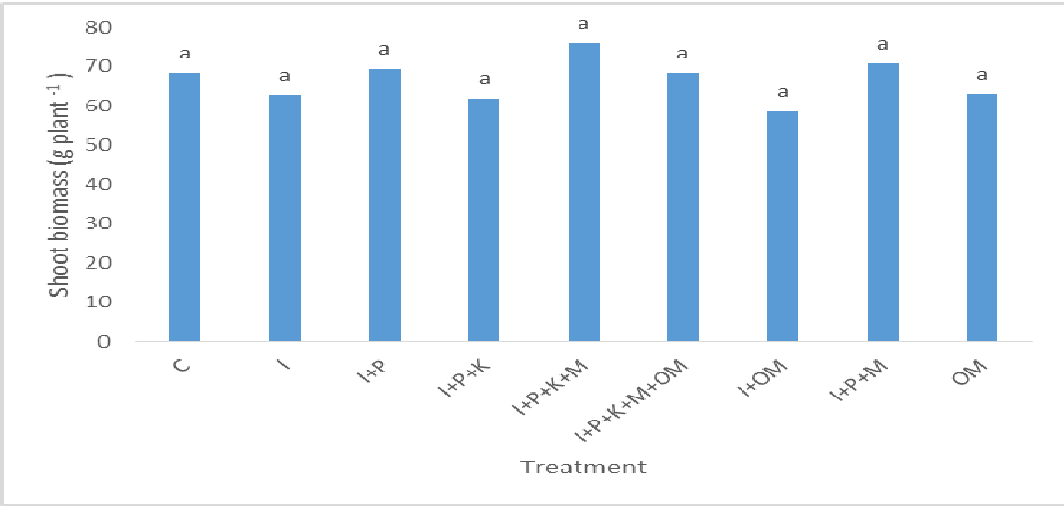


Figure 4: Effect of treatment combination on shoot biomass of promiscuous soybean at mid- flowering in 2016

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

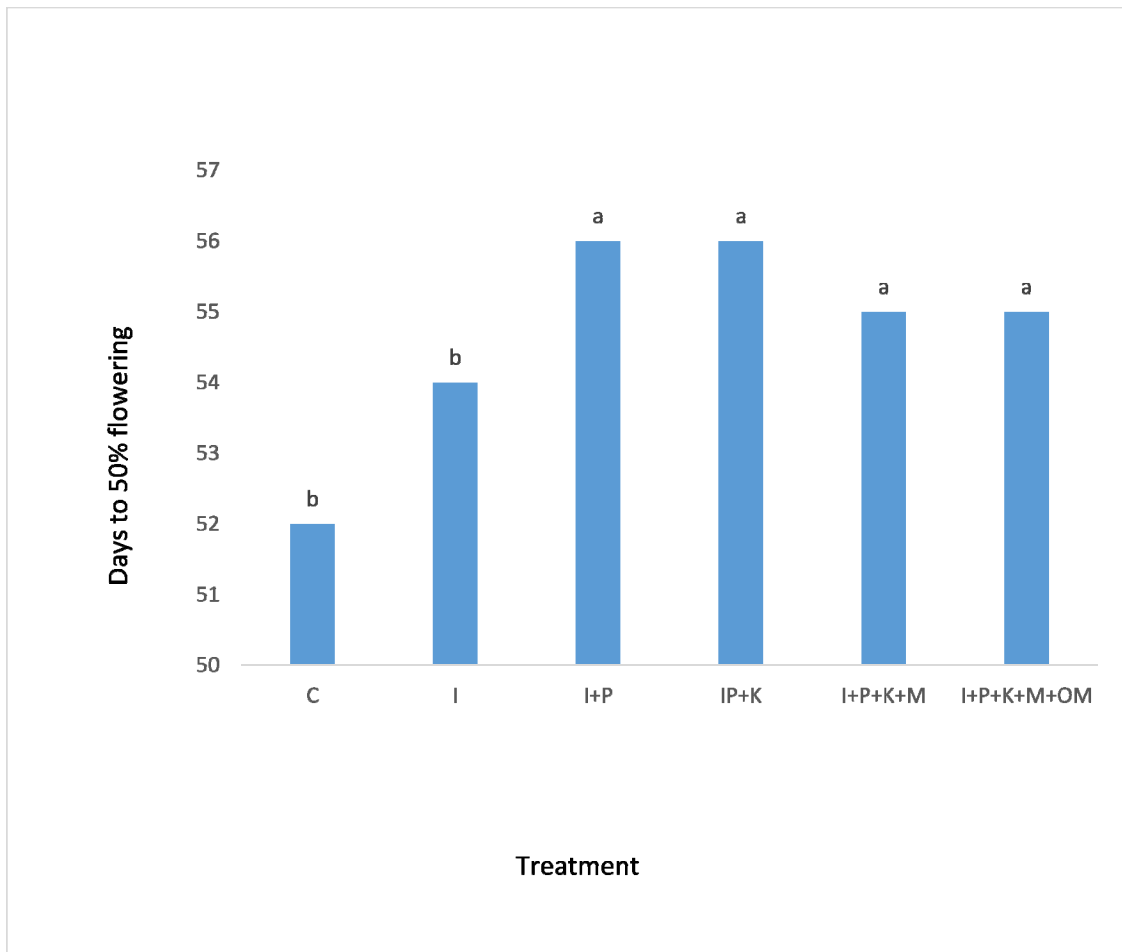


Figure 5: Effect of treatment combination on day to 50% flowering of promiscuous soybean in 2015

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

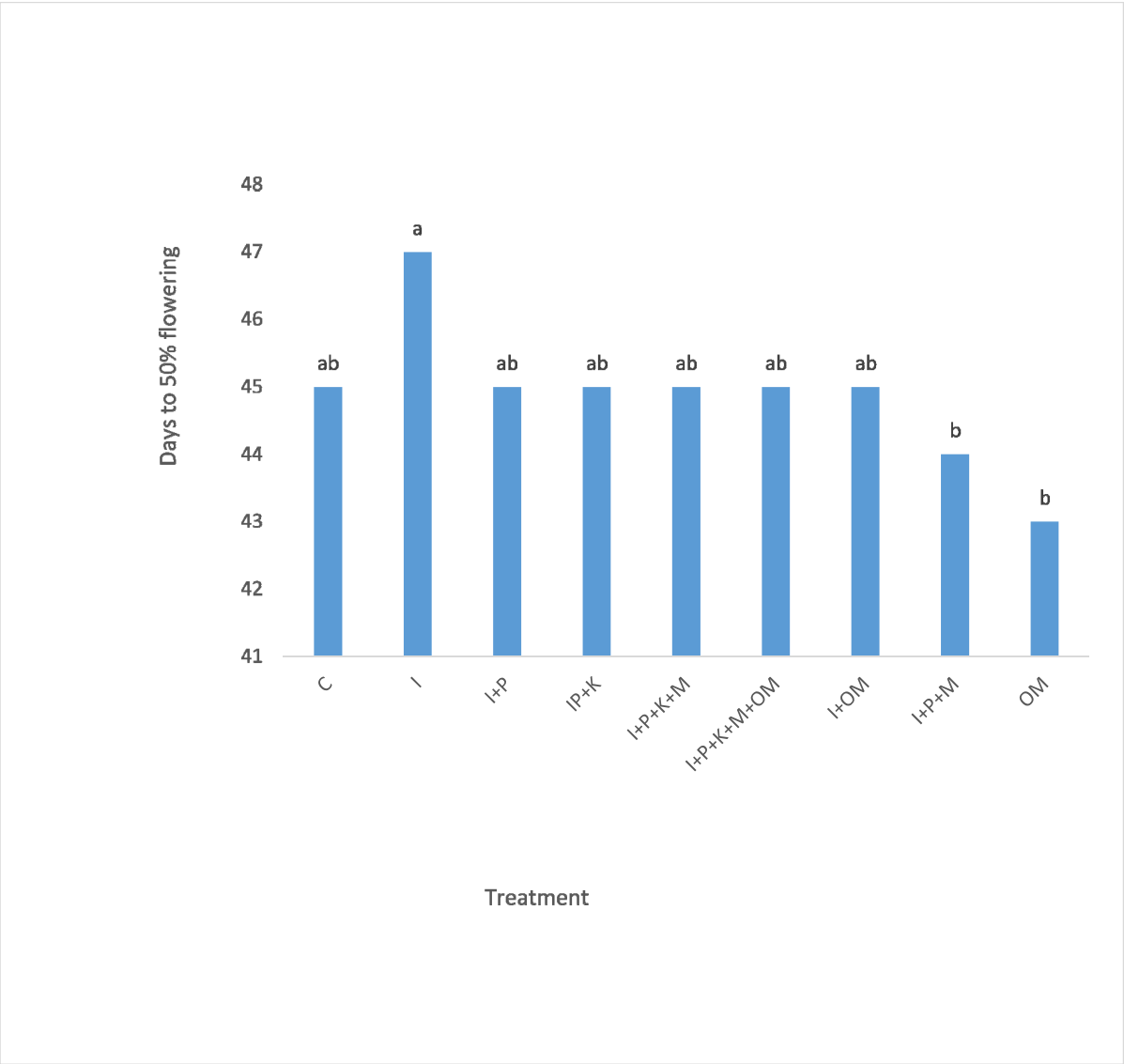


Figure 6: Effect of treatment combination on day to 50% flowering of promiscuous soybean in 2016

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.



#### **4.5 Nodule Number and Nodule Weight**

The response of promiscuous soybeans to rhizobial inoculation in combination with mineral fertilizer and organic manure on soybean nodule number is presented in Fig 7 and 8. Nutrient combination had a significant ( $p \leq 0.05$ ) effect on nodule number. Application of I+P, I+P+K and I+P+K+M+OM recorded the higher number of nodules which were similar to other nutrient plot except Control plot. In 2016, plots given I+OM and I+P+K+M produced highest number of nodules than plots with treatment I only and control plot in this study.

The response of promiscuous soybeans to rhizobial inoculation in combination with mineral fertilizer and organic manure on nodule weight is shown on Fig 9 and 10. Nutrient combination had no significant ( $p \leq 0.05$ ) effect on nodule weight in this study in 2015, but significantly differed in 2016. Application of I+P+K+M and I+OM produced the heaviest nodule than plots given control, I only and OM only which produced the lightest nodule weight.

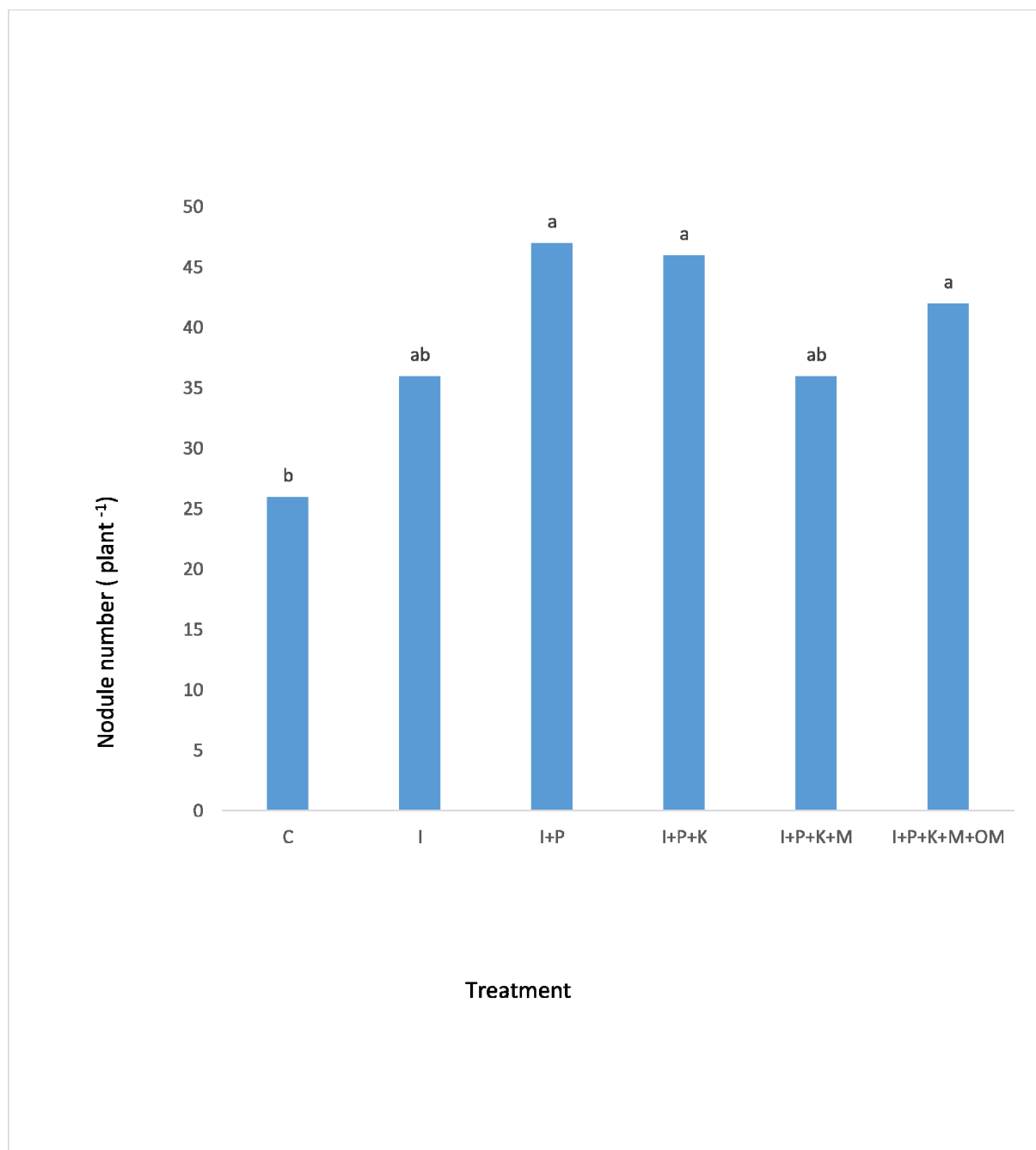


Figure 7: Effect of nutrient combination on nodule number of promiscuous soybean at mid-flowering in 2015

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

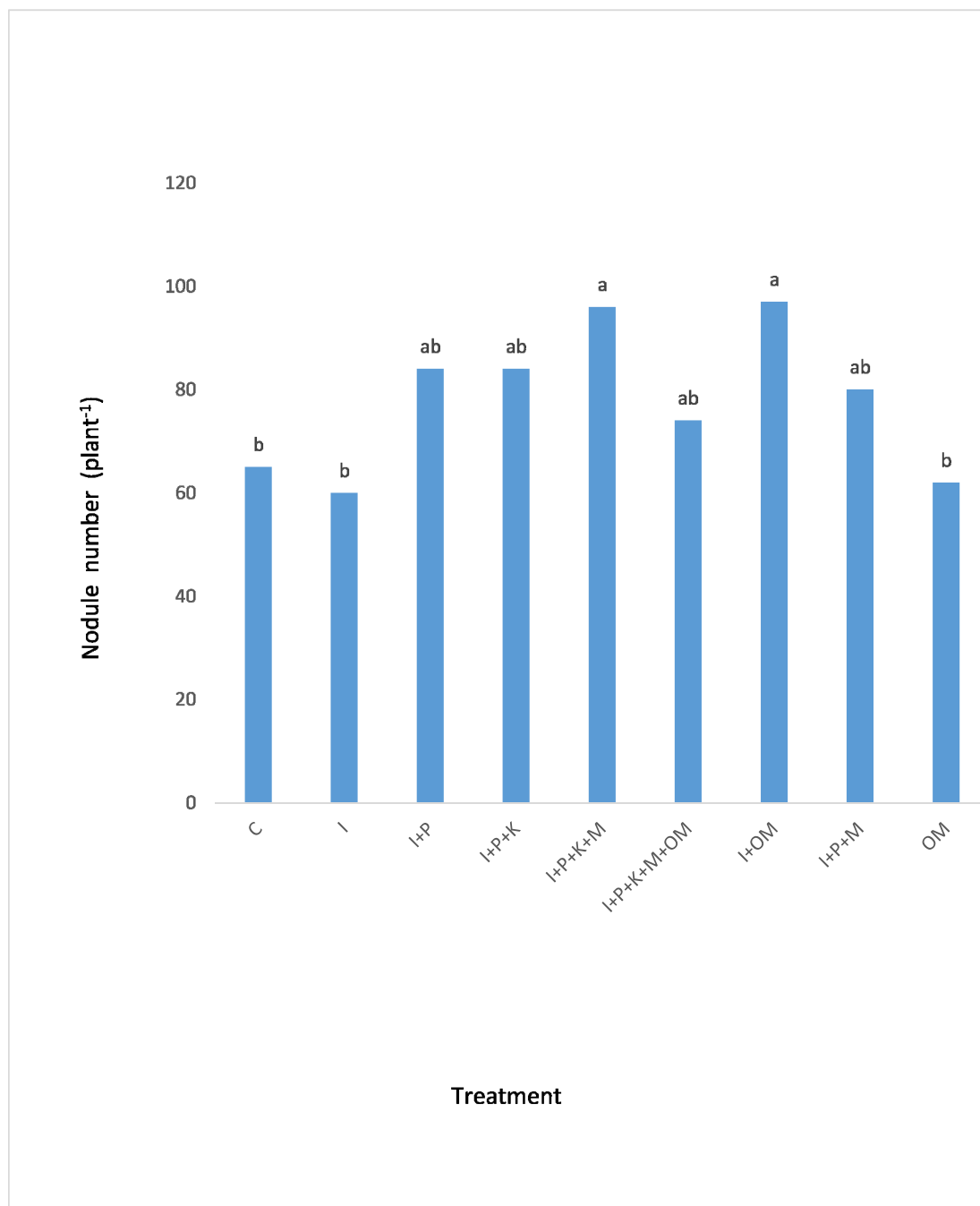


Figure 8: Effect of treatment combination on nodule number of promiscuous soybean at mid-flowering in 2016

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

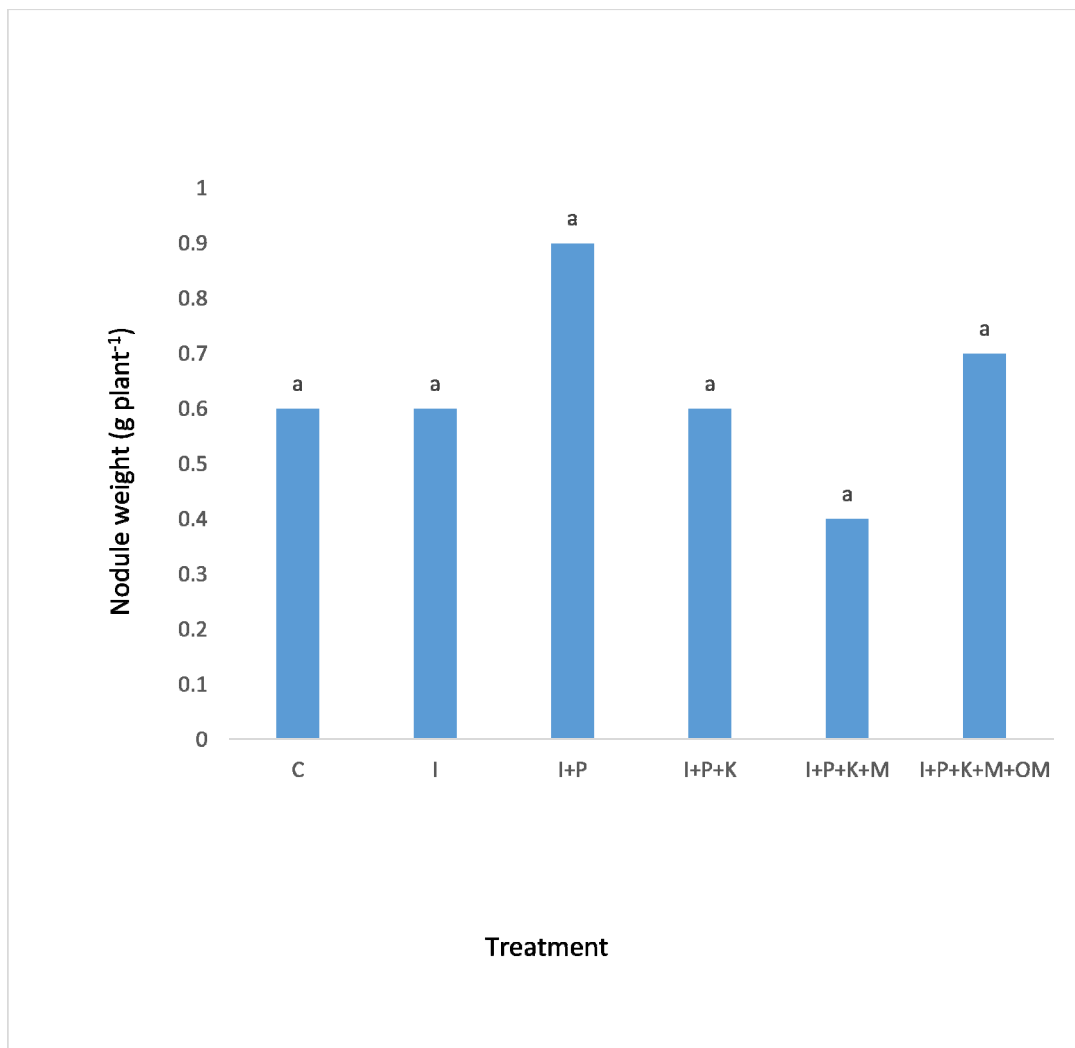


Figure 9 : Effect of treatment combination on nodule weight of promiscuous soybean at mid-flowering in 2015

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

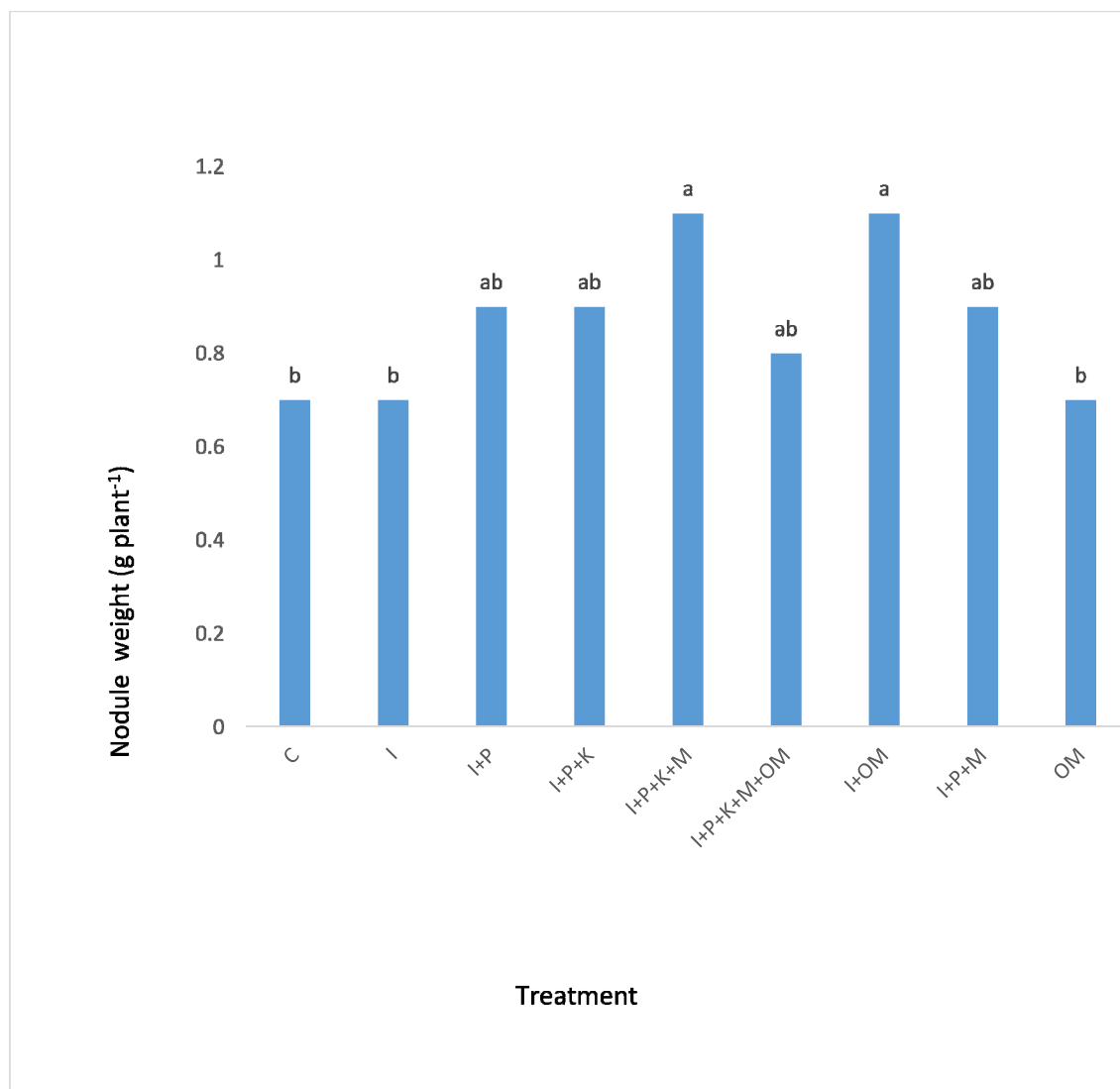


Figure 10 : Effect of treatment combination on nodule weight of promiscuous soybean at mid-flowering in 2016

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

#### **4.6 Number of Branches and Pods/plant**

Figure 11 and 12 show the response of promiscuous soybean to rhizobial inoculation in combination with mineral fertilizer and organic manure on number of branches, shoot biomass and of soybean in 2015 and 2016. Nutrient combination had a significant ( $p \leq 0.05$ ) effect on number of branches of soybean at mid-flowering in both year. The application of I+P+K+M+OM produced higher number of branches which was similar to all other plots except control plot in 2015. Similarly in 2016, application of I+P+K+M+OM produced the highest number of branches though statistically similar with plot given I+P+K and I+P+M but significantly higher than control produced the smallest number of branches in this study respectively.

The response of promiscuous soybean to rhizobial inoculation in combination with mineral fertilizer and organic manure on pods per plant is shown in Fig.13 and 14. Nutrient combination had a significant ( $p \leq 0.05$ ) effect on pod number per plant. Plots given all nutrient recorded the highest number of pod which was similar to other plots but control plot produced the smallest number of pod which was similar to plot given I only.

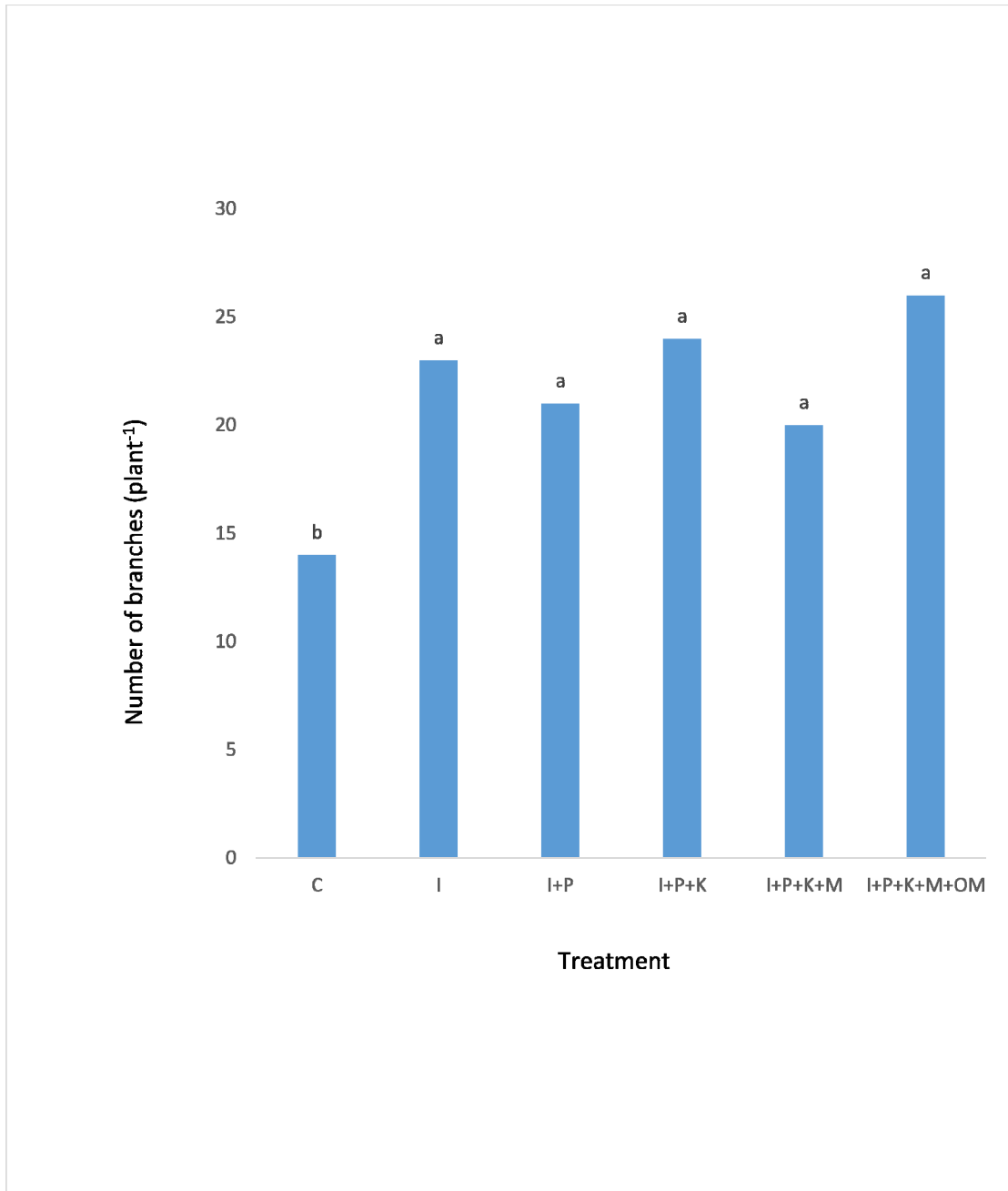


Figure 11: Effect of treatment combination on branch number of promiscuous soybean at mid- flowering in 2015

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

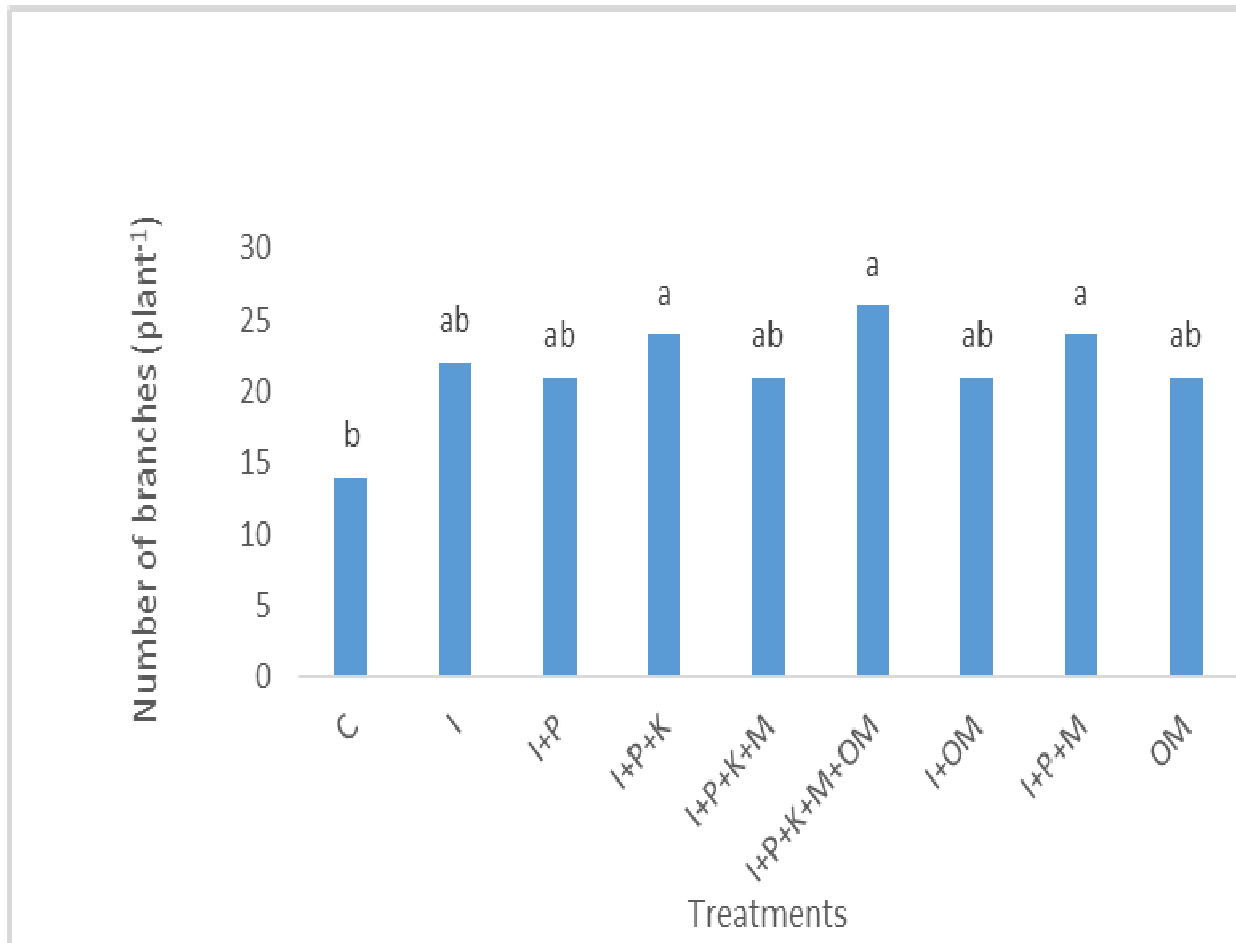


Figure 12: Effect of treatment combination on branch number of promiscuous soybean at mid- flowering in 2016

C = control, I = Inoculant only, P= phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.



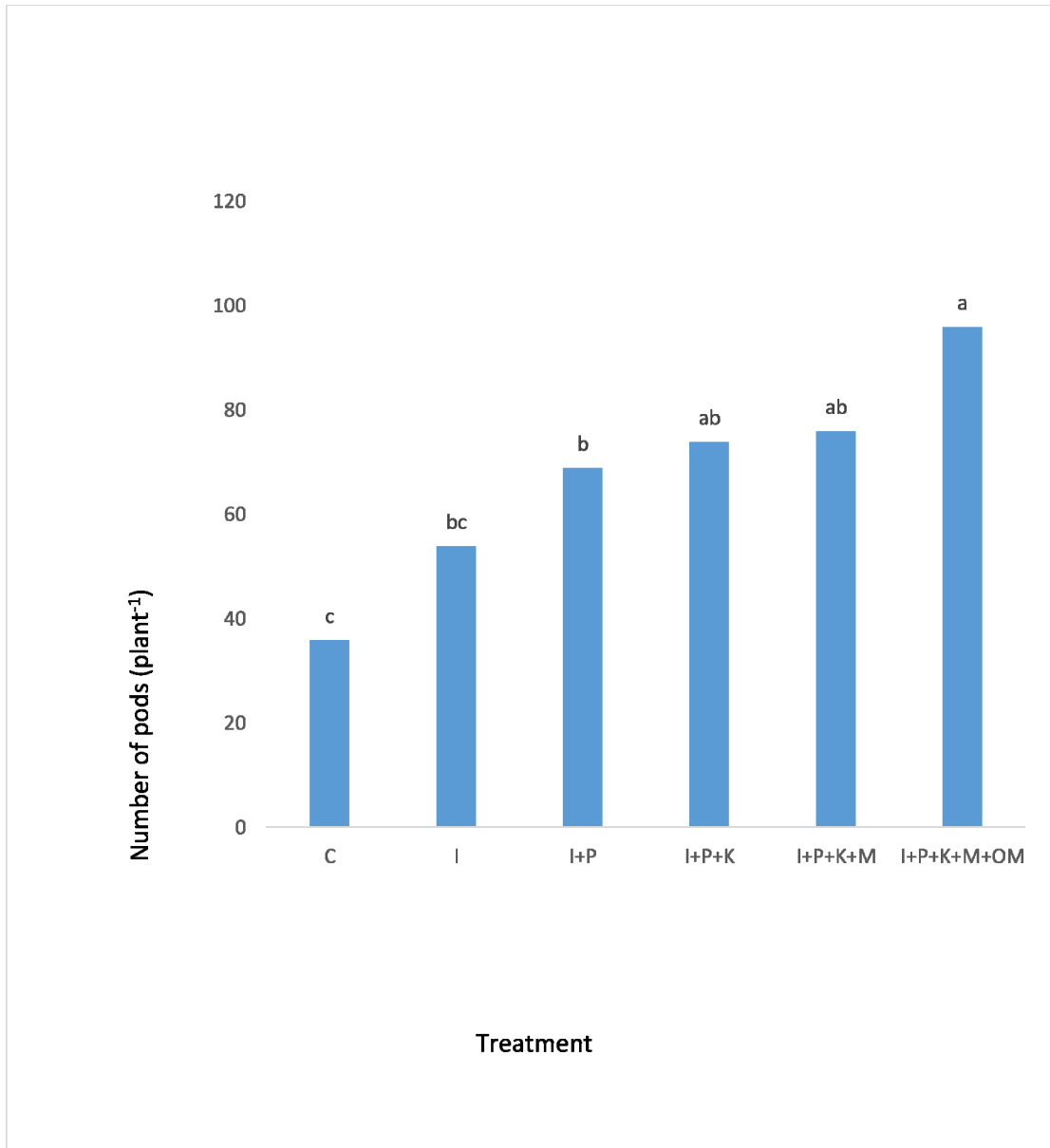


Figure 13: Effect of treatment combination on number of pods per plant of promiscuous soybean at harvest in 2015

C = control, I = Inoculant only, P= phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

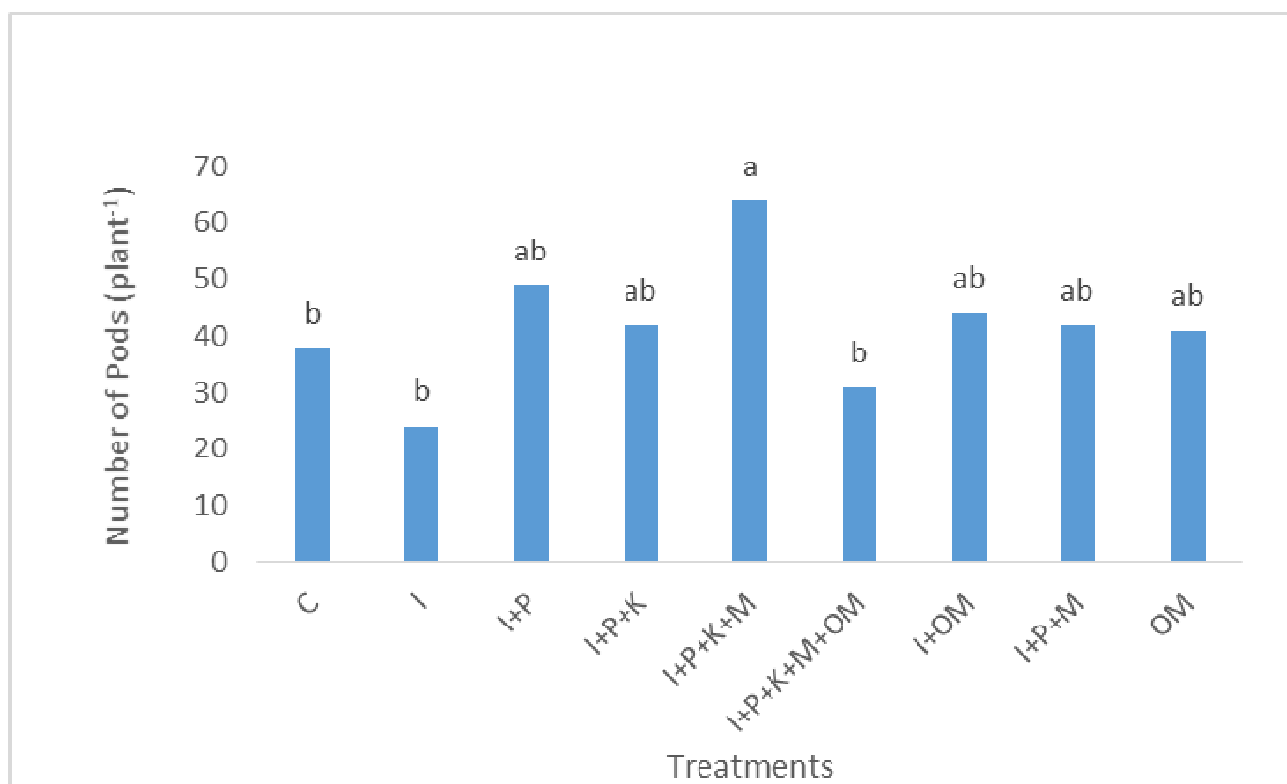


Figure 14: Effect of treatment combination on number of pods per plant of promiscuous soybean at harvest in 2016

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

#### **4.7 Number of Seeds per Pod and 100 Seed Weight**

The response of promiscuous soybean to rhizobial inoculation in combination with mineral fertilizer and organic manure on number of seeds /pod is shown on Figure. 15 and 16. Nutrient combination had a significant effect ( $p \leq 0.05$ ) on number of seed per pod in 2015. Application of I+P, I+P+K+M and I+P+K+M+OM produced higher number of seed per pod which was significantly higher than the plots treated with C, I and I+P+K.

The response of promiscuous soybean to rhizobial inoculation in combination with mineral fertilizer and organic manure on 100 seed weight is shown on Fig 17 and 18. Nutrient combination had no significant ( $p \leq 0.05$ ) effect on 100 seed weight in this study in 2015 and 2016.

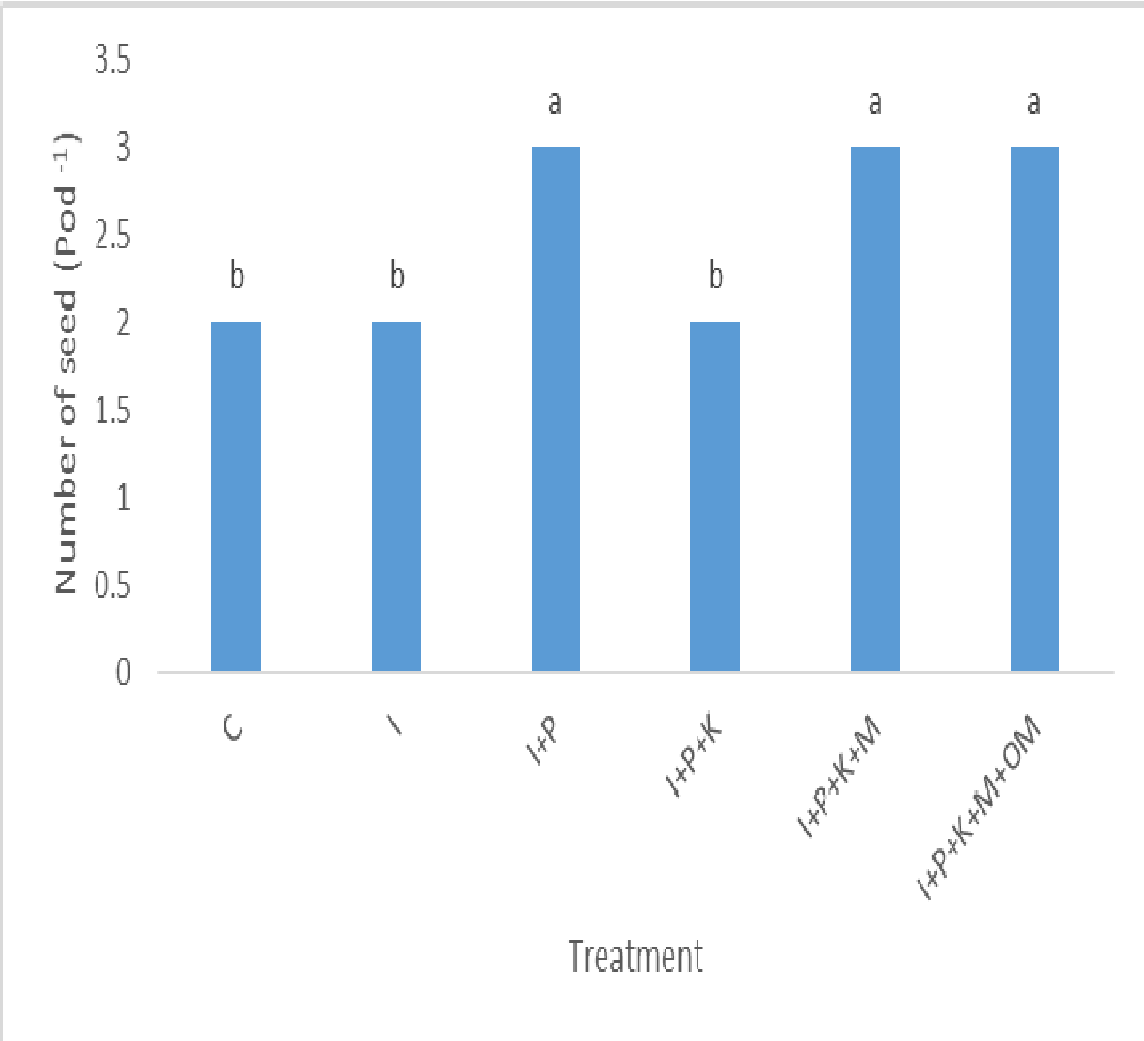


Figure 15: Effect of treatment combination on number of seeds per pod of promiscuous soybean at harvest in 2015

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

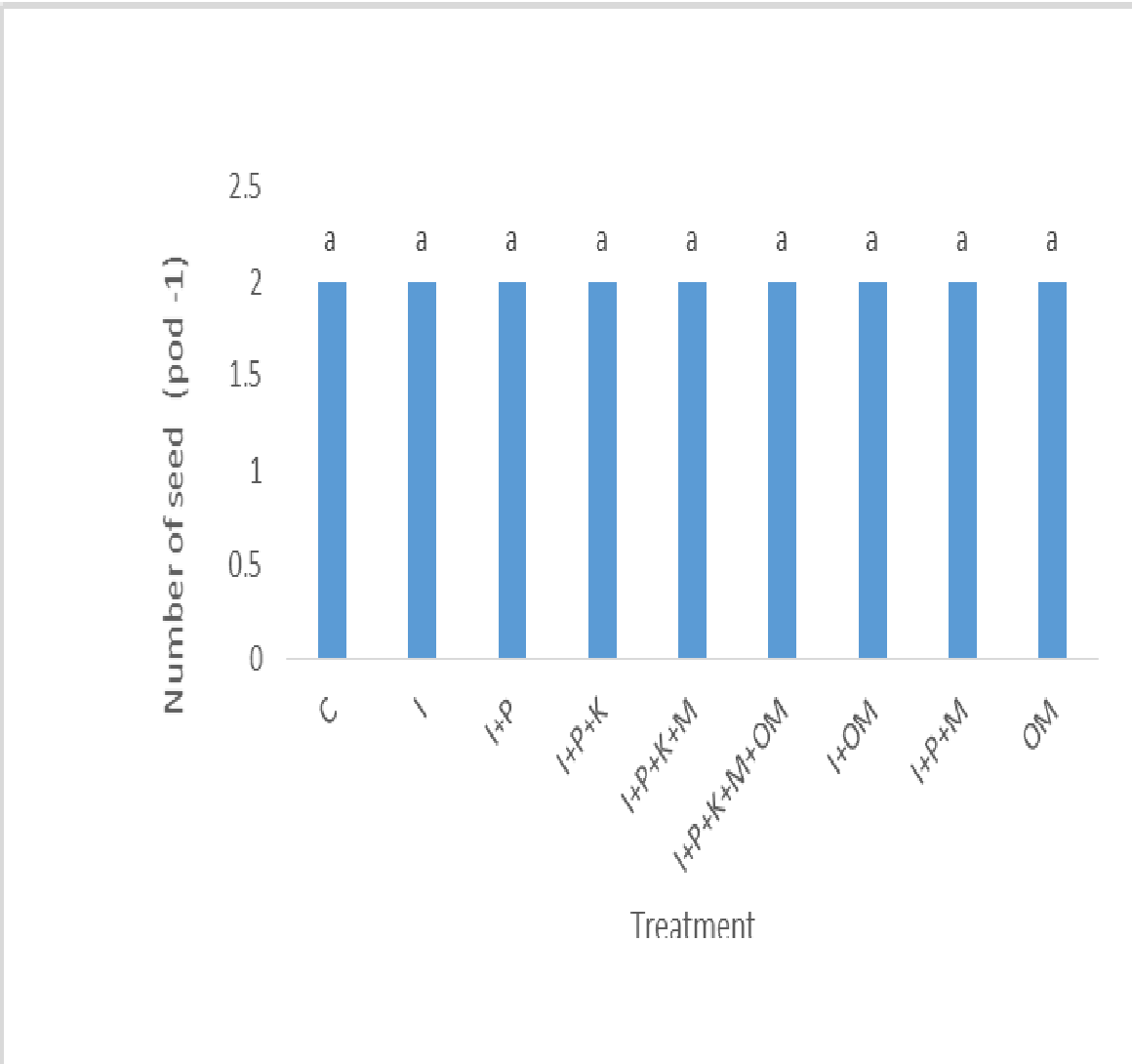


Figure 16 Effect of treatment combination on number of seed per pod of promiscuous soybean at harvest in 2016

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

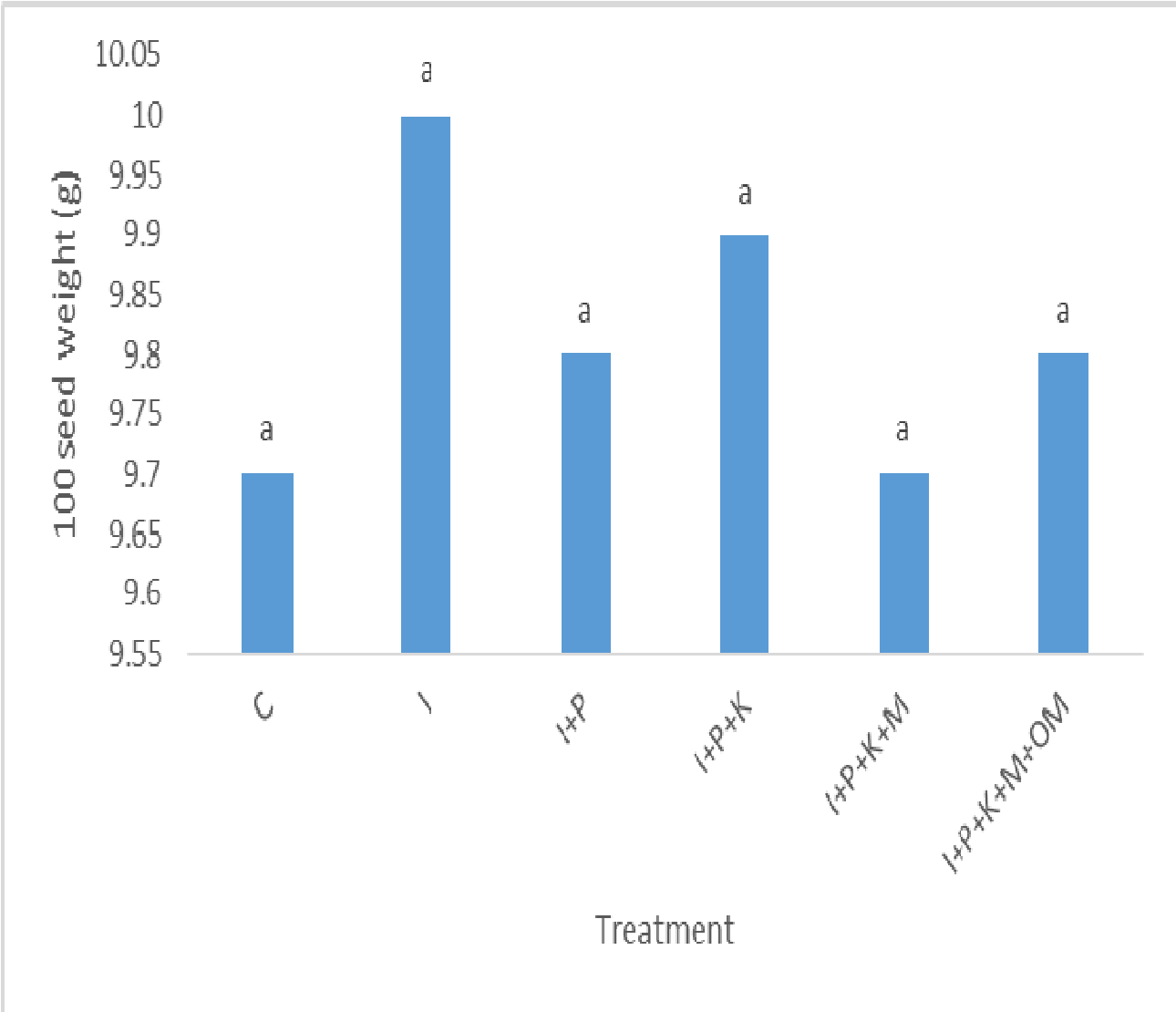


Figure 17: Effect of treatment combination on 100 seed weight of promiscuous soybean in 2015

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

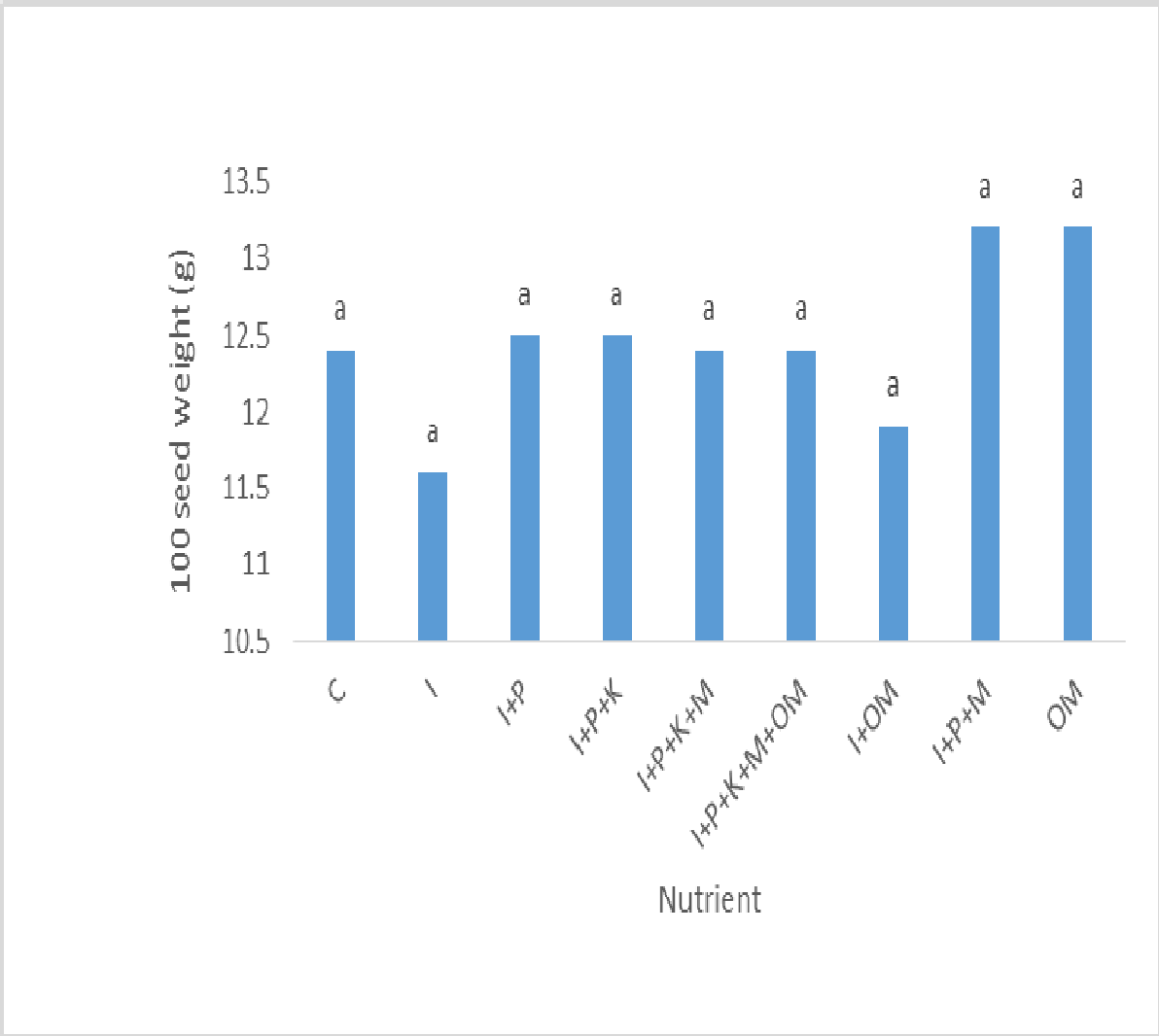


Figure 18: Effect of treatment combination on 100 seed weight of promiscuous soybean in 2016

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

#### **4.8 Grain Yield and Stover Yield**

The response of promiscuous soybean to rhizobial inoculation in combination with mineral fertilizers and organic manure on grain yield in 2015 and 2016 is shown on figure 19 and 20. Treatment combination had a significant ( $p \leq 0.05$ ) effect on grain yield of soybean. Application of I+P+K+M+OM produced the highest grain yield which was similar to other treatment combination but significantly higher than Control and I only plots in 2015. In 2016, plot given I+P produced the highest grain yield which was similar to plots treated with I+P+K, I+P+K+M+O and I+P+M but Control and I only treated plots produced significantly lowest grain yield which was similar to plots treated with I+OM and OM in this study.

The response of promiscuous soybean to rhizobial inoculation in combination with mineral fertilizers and organic manure on stover yield is shown on figure 21 and 22. Treatment combination in 2015 had a significant ( $p \leq 0.05$ ) effect on stover yield. The application of I+P+K+M+OM produced the heaviest stover while plot given I only produced the lightest stover. In 2016, plots given I+P+K+M recorded the heaviest stover while Control and I only plots produced similar lightest stover.



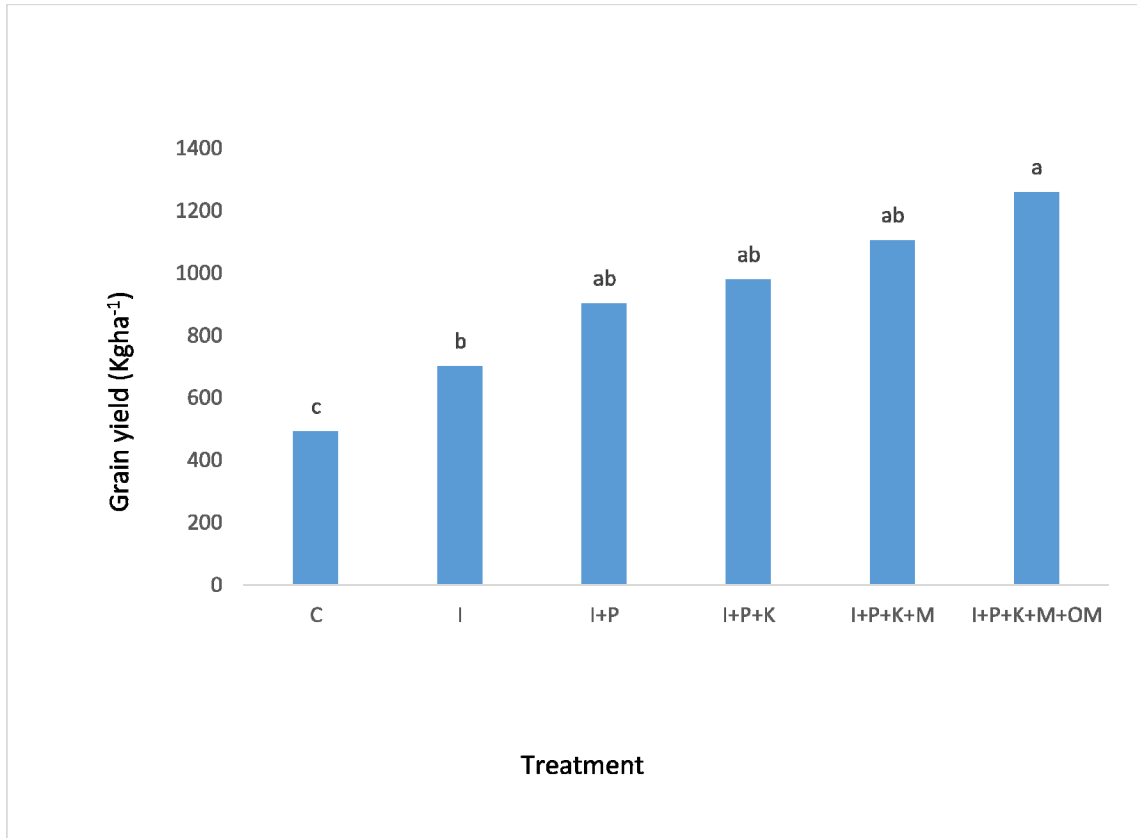


Figure 19: Effect of treatment combination on grain yield of promiscuous soybean in 2015

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

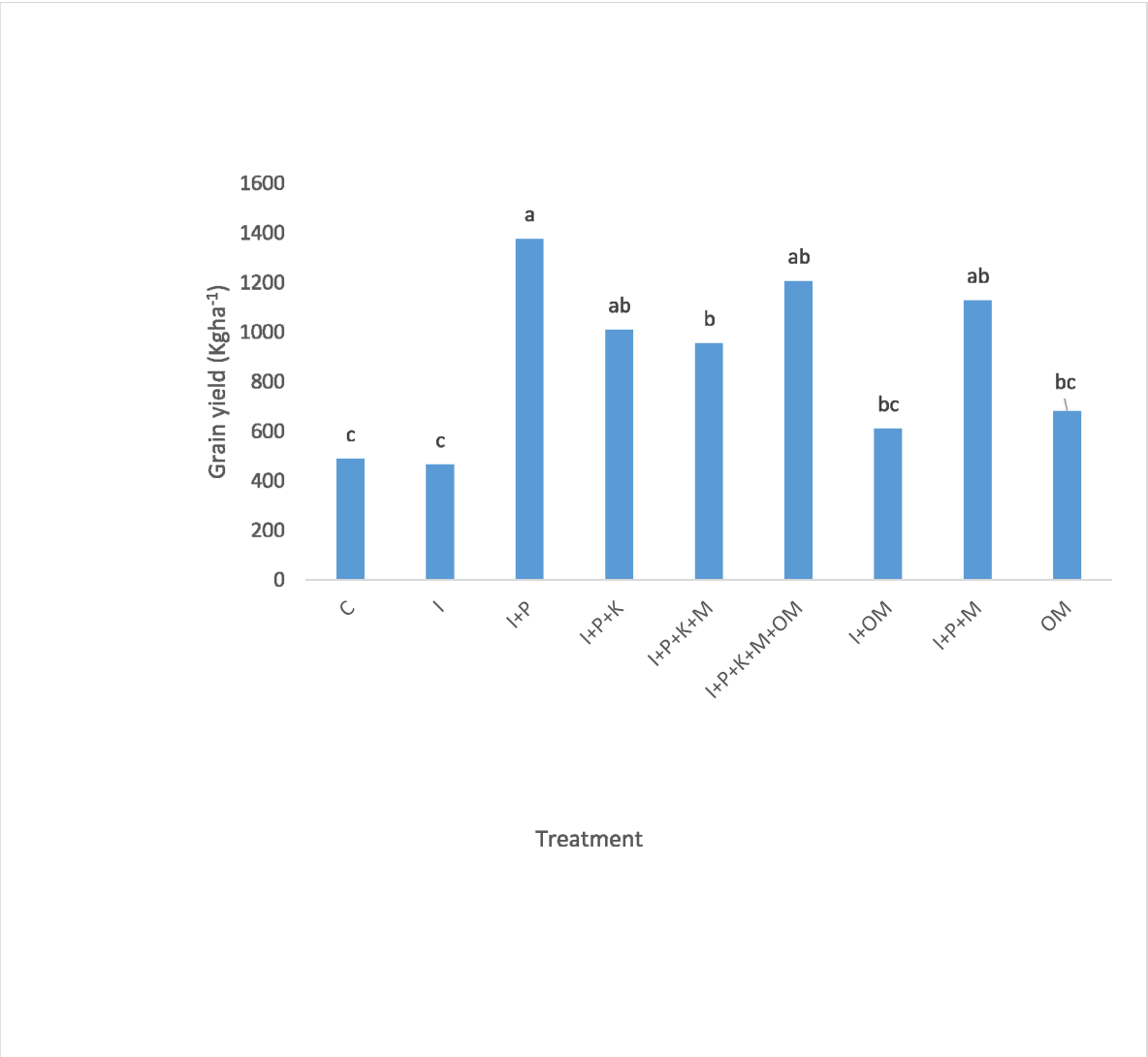


Figure 20: Effect of treatment combination on grain yield of promiscuous soybean in 2016

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

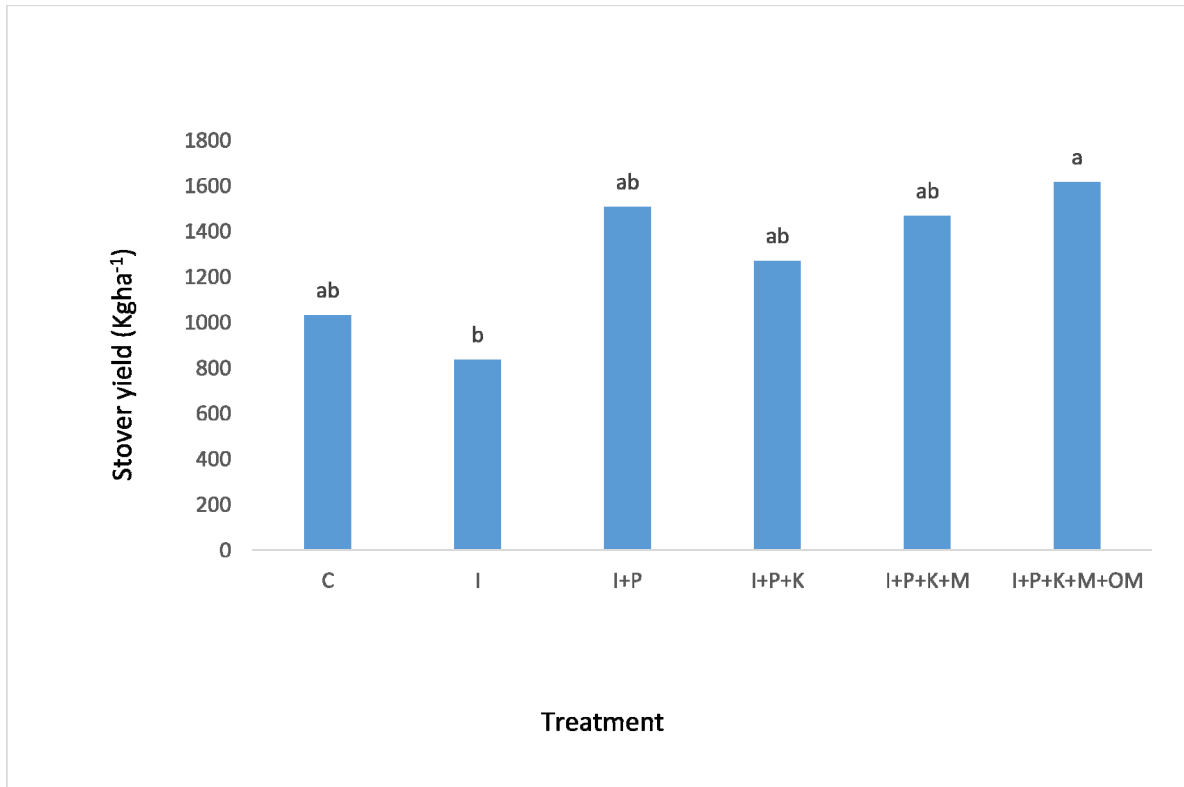


Figure 21: Effect of treatment combination on stover yield of promiscuous soybean in 2015

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

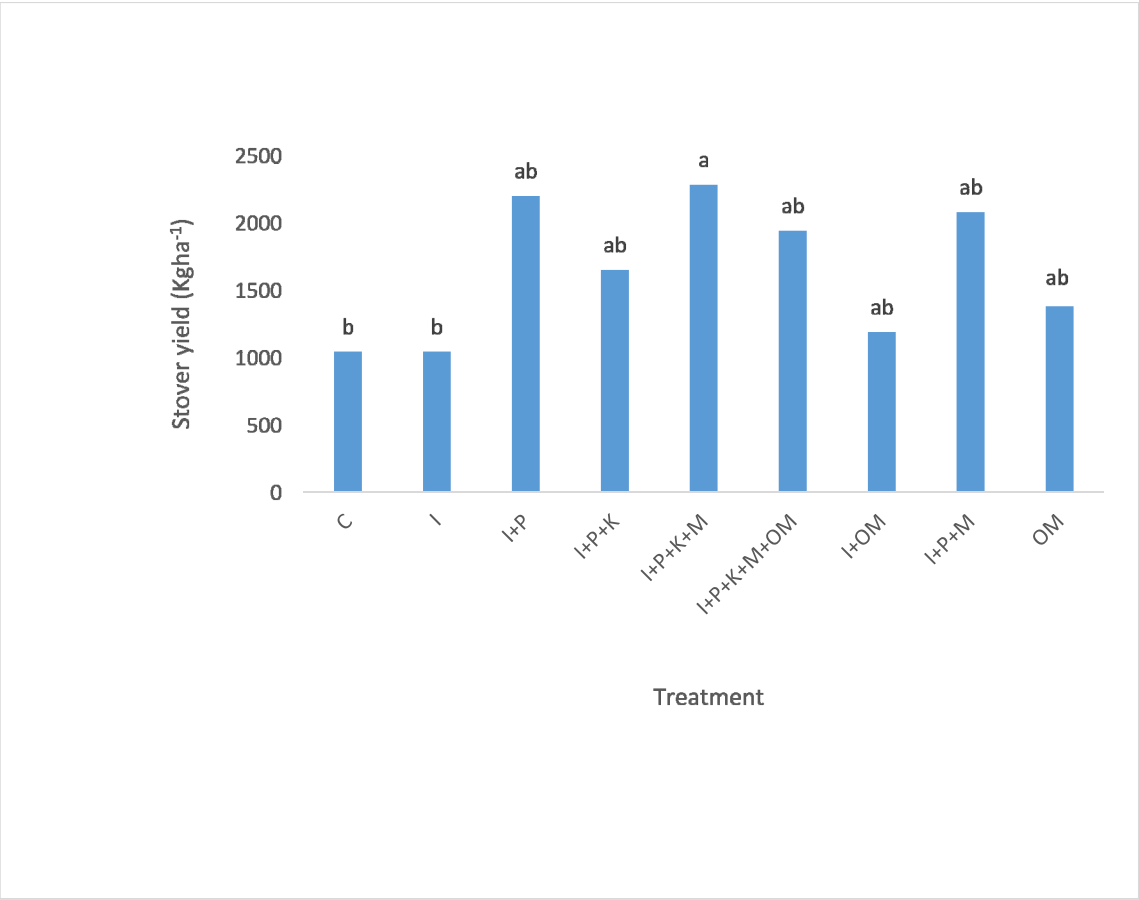


Figure 22: Effect of treatment combination on stover yield of promiscuous soybean in 2016

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

## 4.9 Combine Result for 2015 and 2016 Cropping Seasons

### 4.10 Plant Height

The response of promiscuous soybean to rhizobia inoculation in combination with mineral fertilizer and organic manure on plant height is shown in Fig.23 and 24. Nutrient combination had a significant effect ( $p \leq 0.05$ ) on plant height at 4 and 8WAS. The application of I+P+K+M produced significantly taller plants than the plots with the application of inoculant only and control produced the shorter plants at 4 WAS but at 8WAS the combine application of I+P+K+M+OM produced significantly taller plants while plots with inoculant only produced the shorter plants.

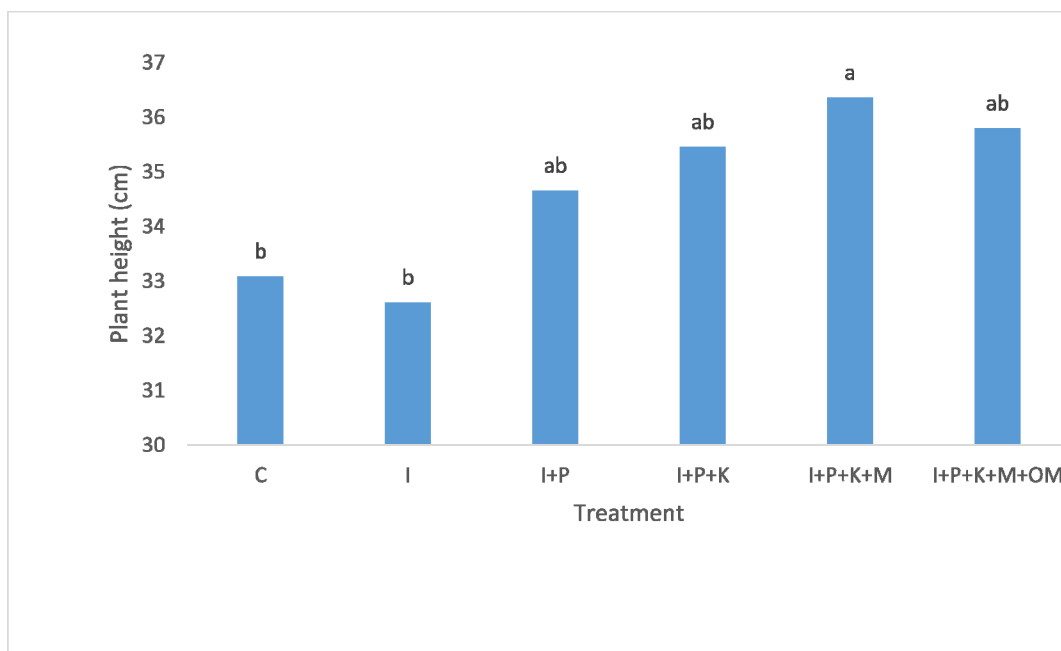


Figure 23: Effect of treatment combination on plant height 4WAS of promiscuous soybean in 2015 and 2016

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure WAS = weeks after sowing

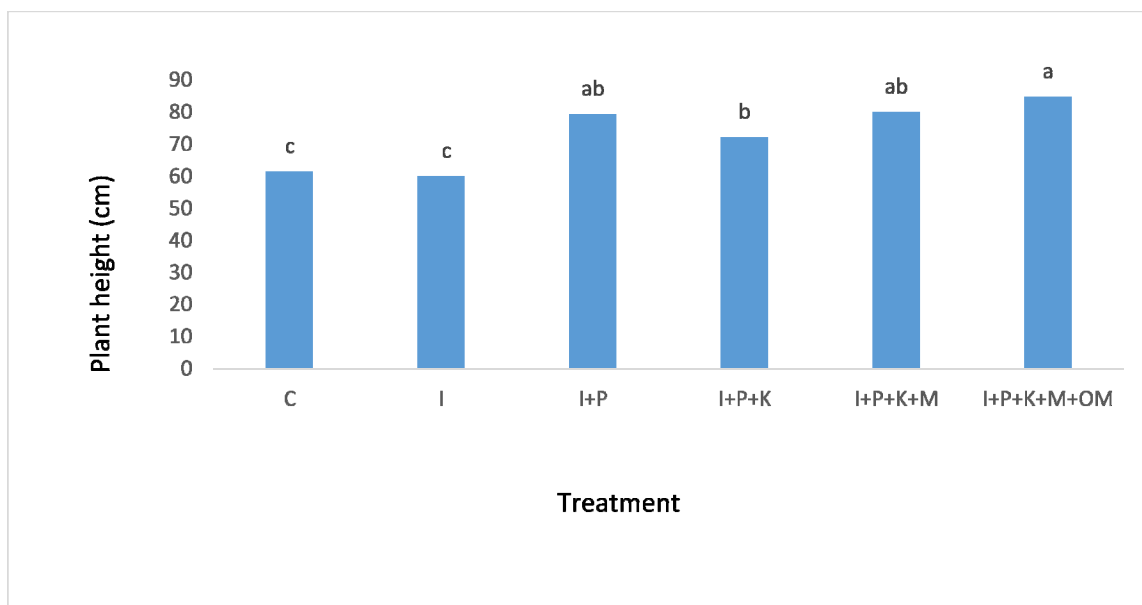


Figure 24: Effect of treatment combination on plant height at 8WAS of promiscuous soybean in 2015 and 2016

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure. WAS = weeks after sowing

#### 4.11 Shoot Biomass and Days to 50% Flowering

The response of promiscuous soybean to inoculation in combination with mineral and organic fertilizer on shoot biomass and days to 50 % percent flowering is shown on Fig.25 and 26. Nutrient combination had a significant effect on shoot biomass and days to 50% flowering. The application of I+P+K+M produced the heaviest shoot biomass which was statistically similar to I+P, I+P+K and I+P+K+M+OM but significantly higher than I only and control plots that produced the lightest shoot biomass.

The response of promiscuous soybean to inoculation in combination with mineral and organic fertilizer is presented in Fig. 26. Nutrient combination had significant effect on soybean days to

50% flowering. The application of Inoculant only, I+P, I+P+K, I+P+K+M and I+P+K+M+OM took higher number of days to attain 50% flowering while control attained 50% flowering in a shorter number of days compared to the combination plots.

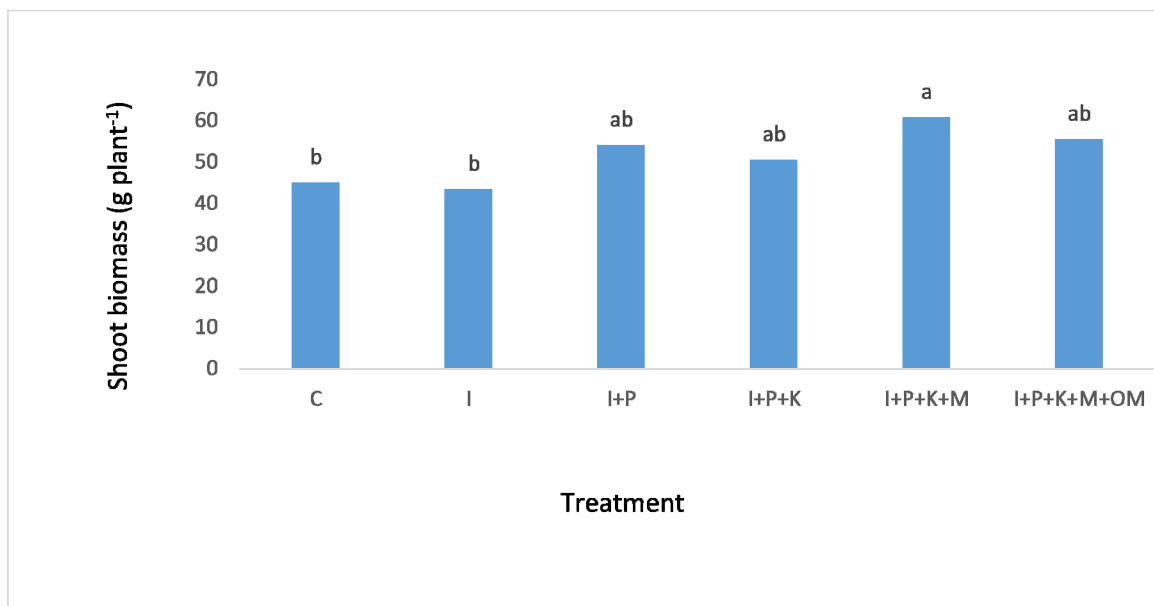


Figure 25: Effect of treatment combination on shoot biomass of promiscuous soybean in 2015 and 2016

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

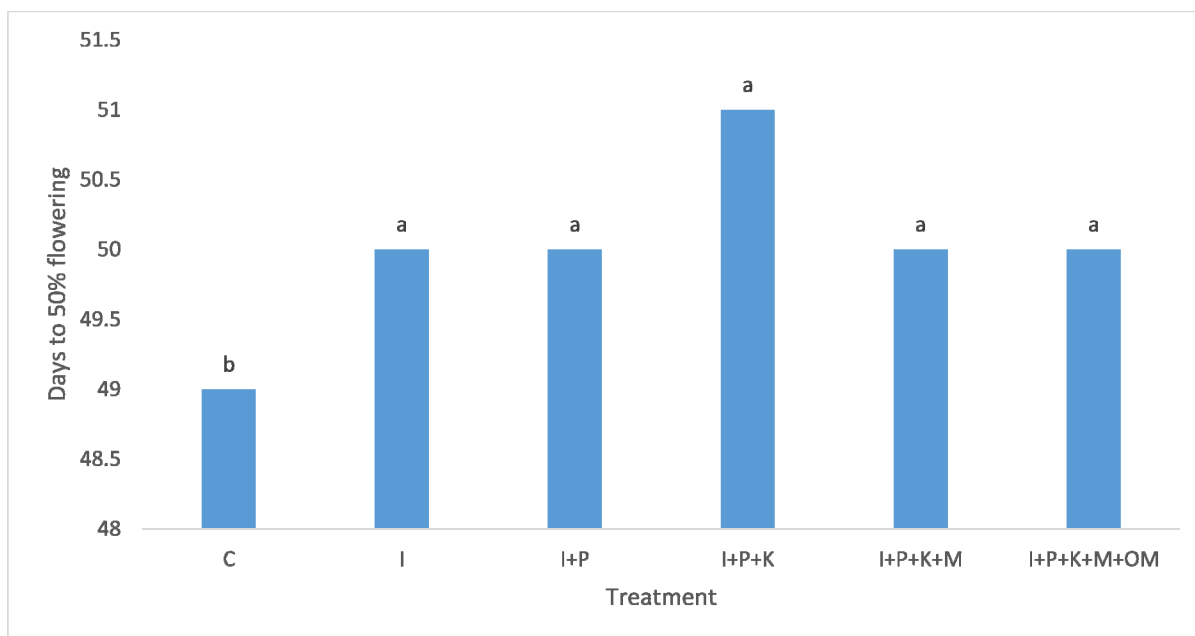


Figure 26: Effect of treatment combination on days to 50% flowering of promiscuous soybean in 2015 and 2016

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

#### 4.12 Number of Branches and Pods/Plant

The response of promiscuous soybean to inoculation in combination with mineral fertilizer and organic manure on number of branches and pods per plant is shown in Fig. 27 and 28. Nutrient combination had a significant effect ( $p \leq 0.05$ ) on number of branches and pods per plant. The application of I+P+K+M produced the highest number of branches which was not statistically different from I+P, I+P+K, and I+P+K+M+OM but significantly different from plots with Inoculant only and control gave the lowest number of branches.



Nutrient combination had significant difference on pods per plant. The application of I+P+K+M, I+P+K+M+OM, I+P+K and I+P produced the higher number of pods per plant while control and Inoculant only produced lower number of pods per plant.

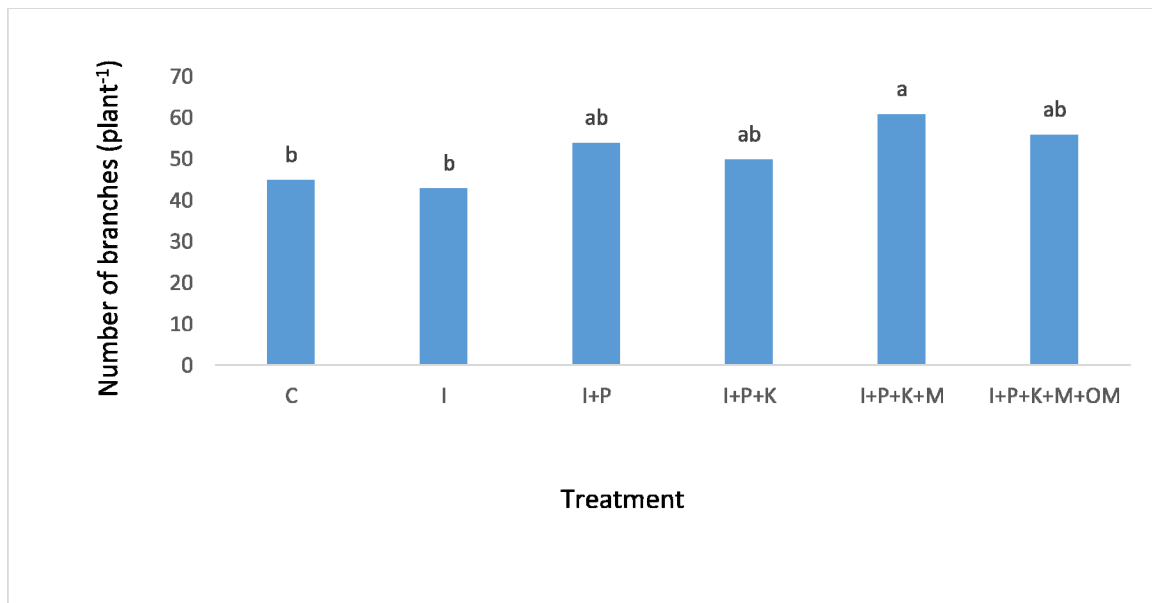


Figure 27: Effect of treatment combination on number of branches of promiscuous soybean in 2015 and 2016

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

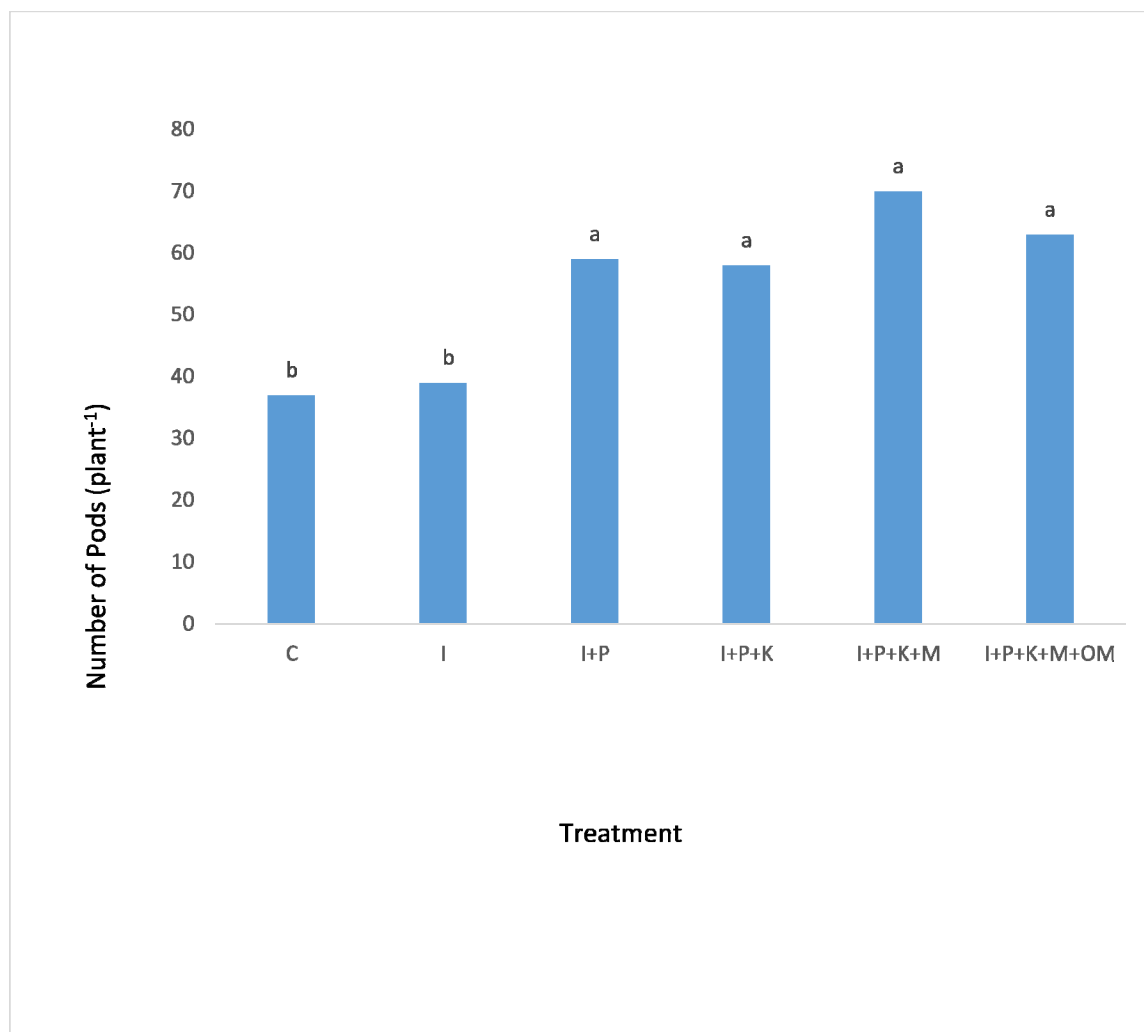


Figure 28: Effect of treatment combination on number of pods per plant of promiscuous soybean 2015 and 2016

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

#### 4.13 Nodule Number and Nodule Weight

The response of promiscuous soybean to inoculation in combination with mineral fertilizer and organic manure on nodule number and nodule weight is presented in Fig. 29 and 30. Nutrient combination had a significant difference on nodule number and nodule weight. The application of I+P, I+P+K and I+P+K+M produced the highest nodule number though statistically similar to I+P+K+M+OM while control and inoculant only produced the lower nodule number. On the other hand I+P produced the heaviest nodule which was similar statistically to I+P+K, I+P+K+M and I+P+K+M+OM while control and inoculant only produced the lightest nodule.

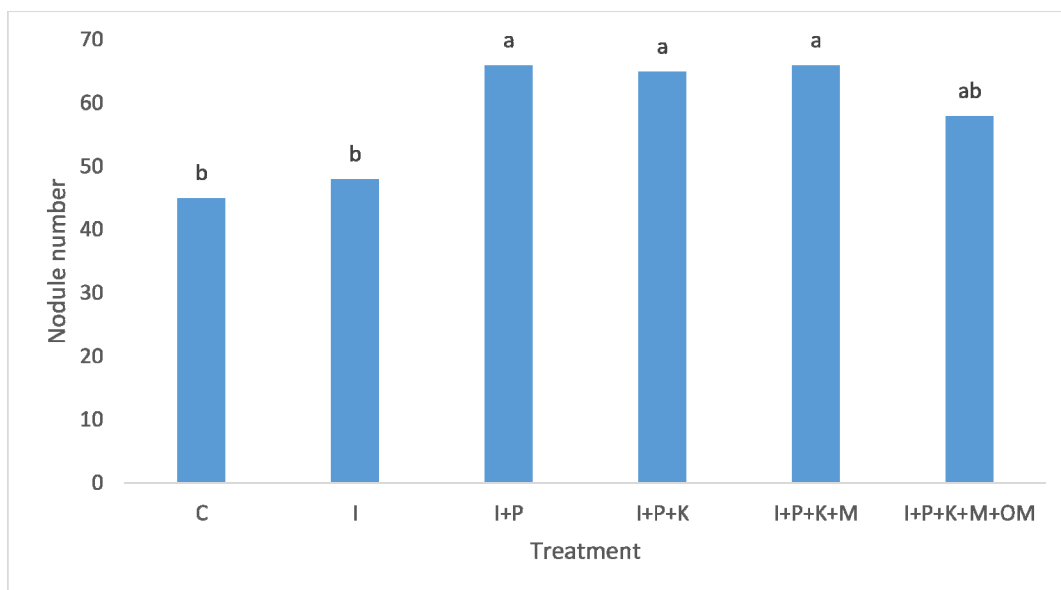


Figure 29: Effect of treatment combination on nodule number of promiscuous soybean I 2015 and 2016

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

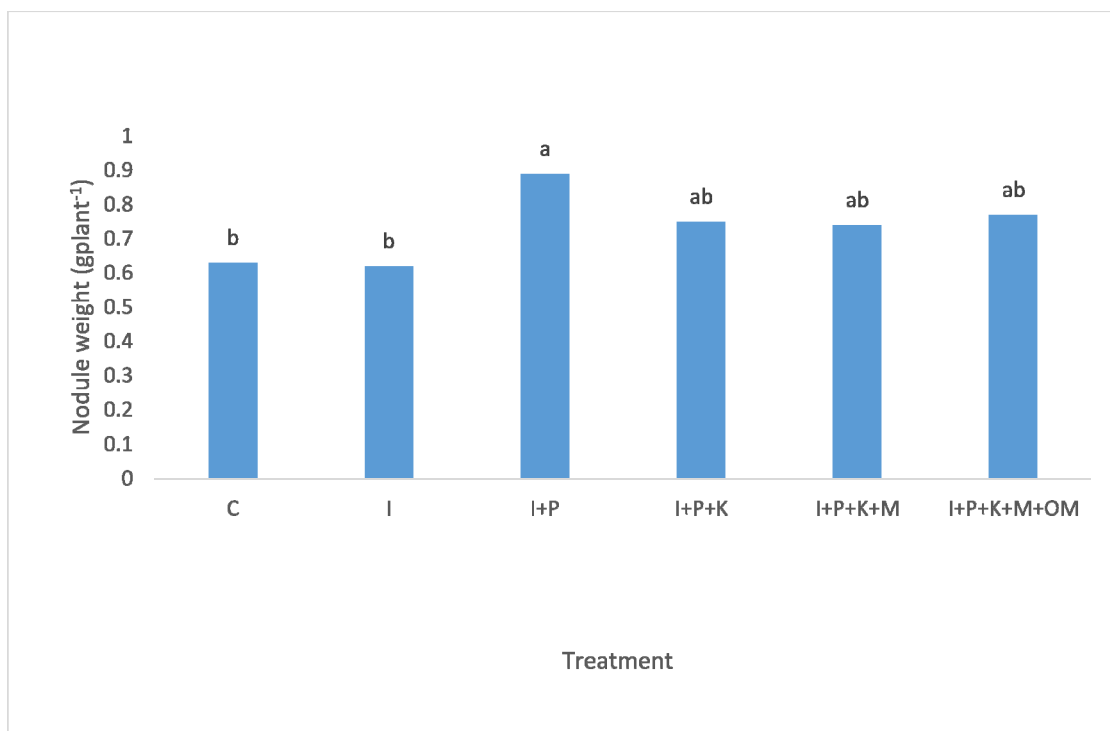


Figure 30: Effect of treatment combination on nodule weight of promiscuous soybean in 2015 and 2016

C = control, I = Inoculant only, P= phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

4.14

### Number of Seed per Pod and 100 Seed Weight

The response of promiscuous soybean to inoculation in combination with mineral fertilizer and organic manure on number of seed per pod and 100 seed weight is shown on Fig. 31 and 32. Nutrient combination had no significant effect on both number of seeds per pod and 100 seed weight.

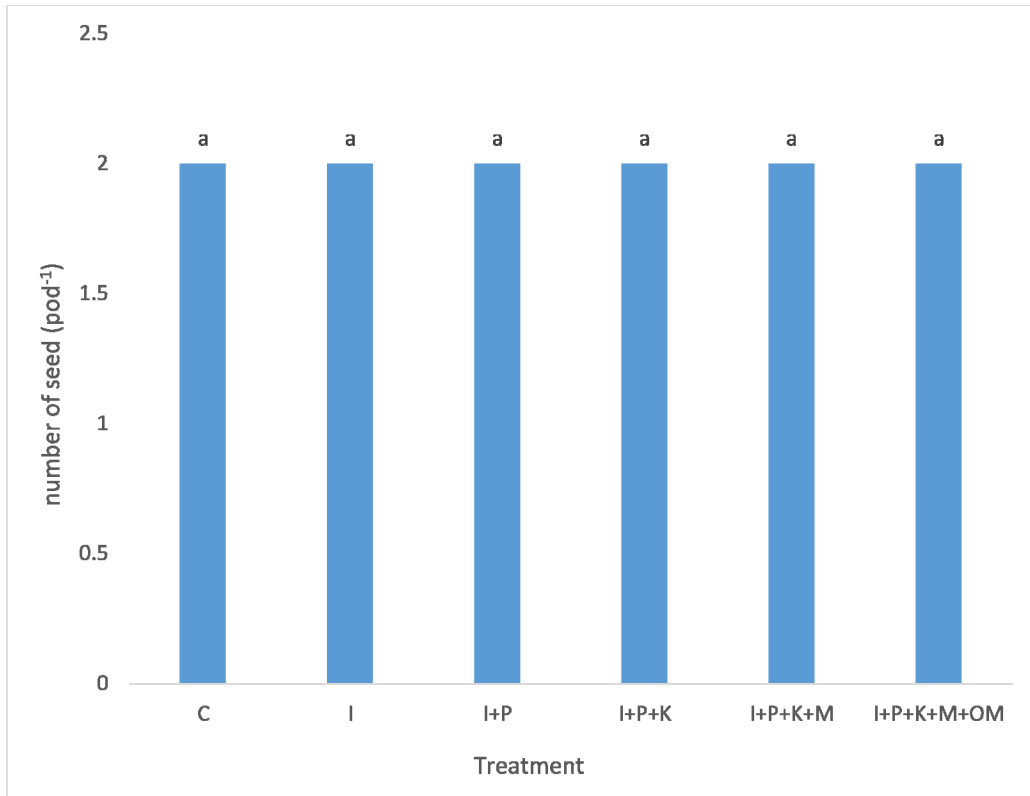


Figure 31: Effect of treatment combination on number of seeds per pod of promiscuous soybean in 2015 and 2016

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

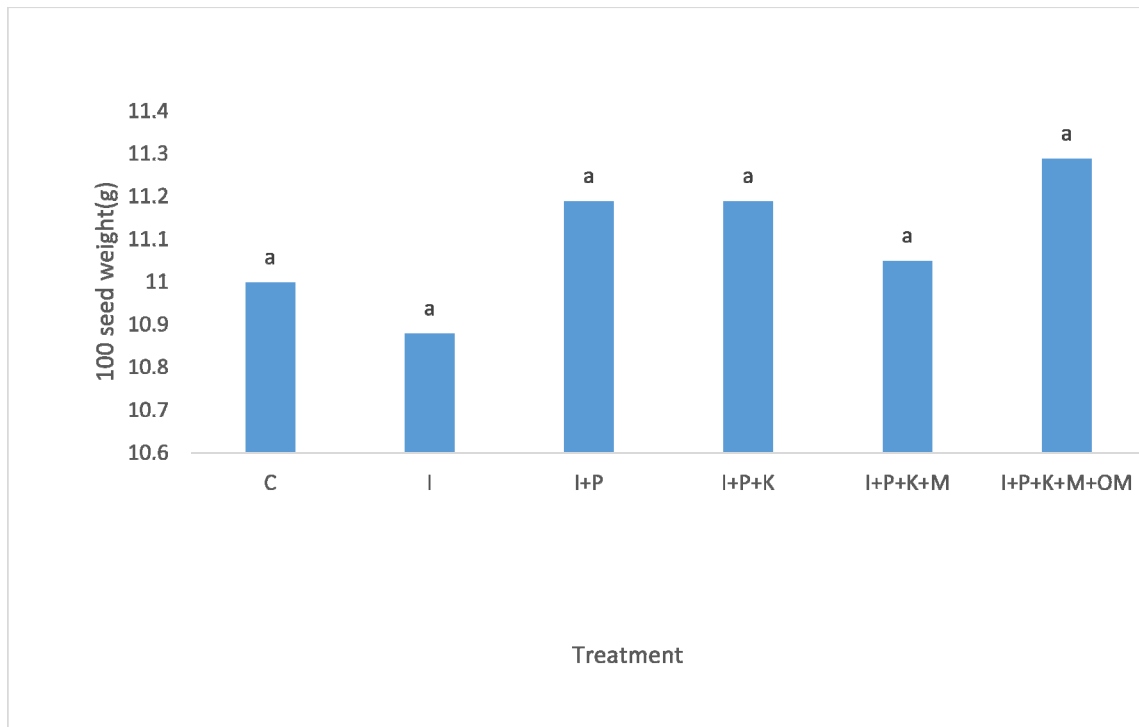


Figure 32: Effect of treatment combination on 100 seed weight of promiscuous soybean in 2015 and 2016

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

#### **4.15 Grain Yield and Stover Yield**

The response of promiscuous soybean to inoculation in combination with mineral fertilizer and organic manure on grain yield and stover yield is shown on Fig. 33 and 34. Nutrient combination had a significant effect on grain yield and stover yield. The application of I+P, I+P+K, I+P+K+M and I+P+K+M+OM produced similar grain yield which was significantly higher than the control and inoculant only that produced low grain yield. Also the application of I+P+K+M, I+P+K+M+OM and I+P resulted to similar weight of stover which differed significantly with Control and Inoculant only treated plots

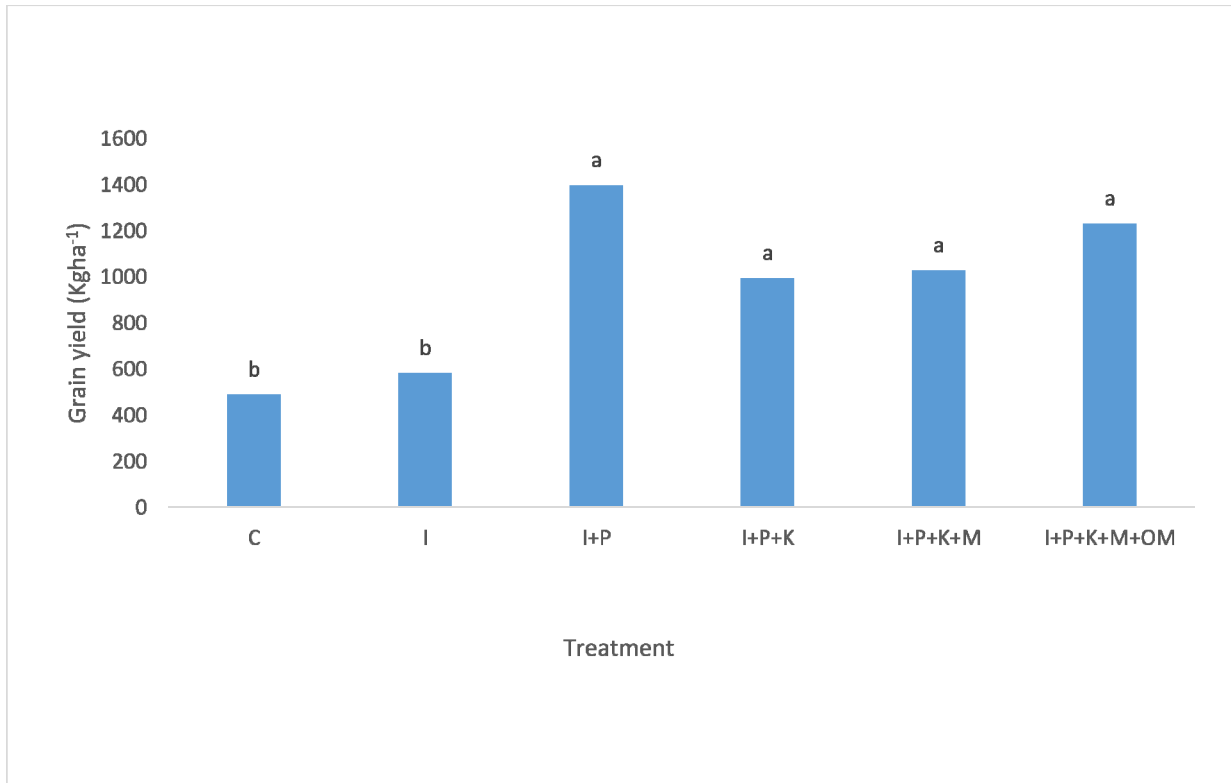


Figure 33: Effect of treatment combination on Grain yield of promiscuous soybean in 2015 and 2016

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.



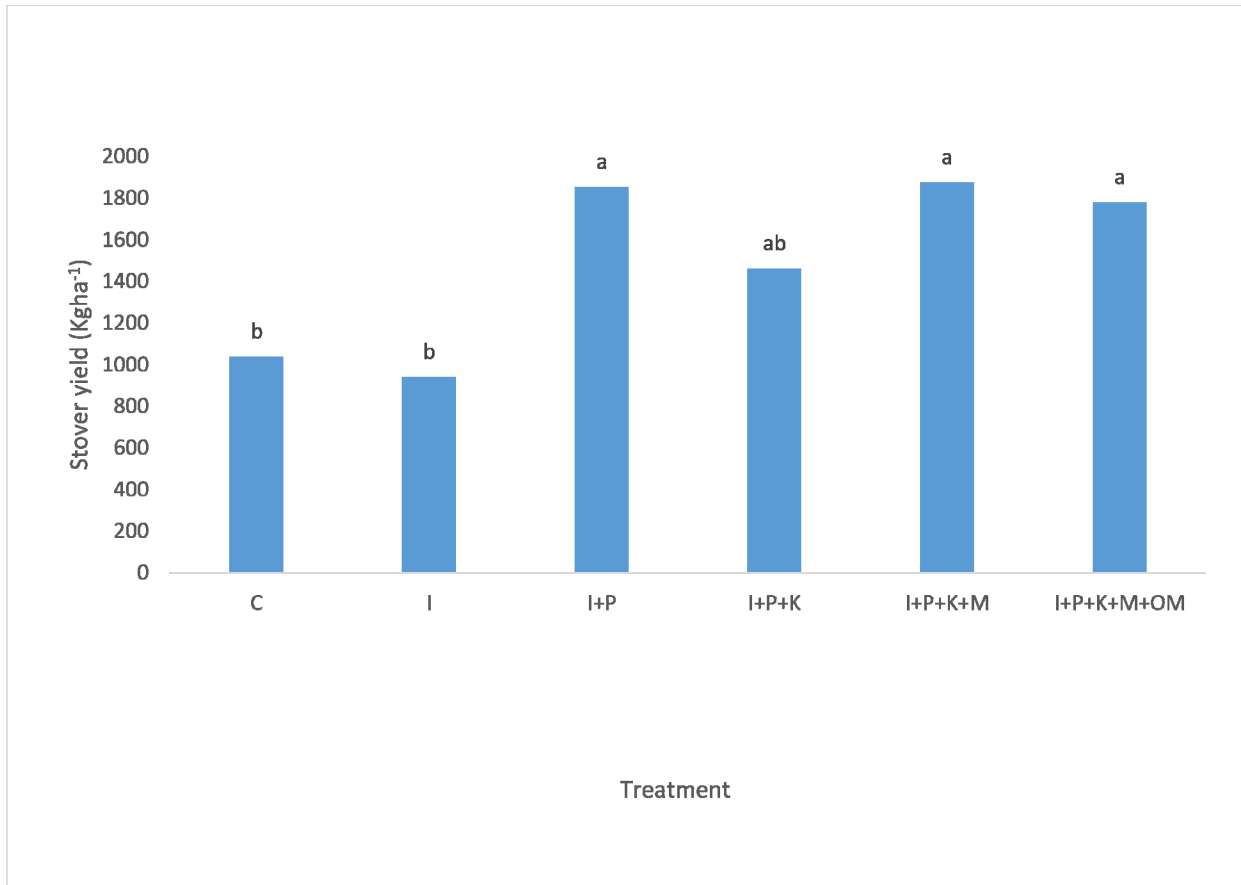


Figure 34: Effect of treatment combination on stover yield of promiscuous soybean in 2015 and 2016

C = control, I = Inoculant only, P = phosphorus, K = Potassium, M = micronutrient, and OM = organic manure.

## CHAPTER FIVE

### 5.0 DISCUSSION, CONCLUSION AND RECOMMENDATION

#### 5.1 Initial Soil Physical and Chemical Properties

The result of the initial soil of the study area shows the characteristics of a savanna soils which are usually low in organic carbon and total N contents (Okalebo, *et al.*, 1993; Aliyu *et al.*, 2013). The extractable P content of the soil was very low in the fertility class (Ikeogu *et al.*, (2013); Kamara *et al.* (2008); Enwezor, *et al.*, 1990). The effective cation exchange capacity (ECEC) of the soil was generally very low and contains low amounts of exchangeable cations this means low fertility status of the soil (Kamara *et al.* (2008); Ikeogu *et al.*, (2013); Mallarino 2013 and Ronner *et al.*, 2016 ). The micronutrient level was acceptable. The soil has loam, silty loam, sandy loam and sandy clay loam texture across sites, and is moderately acidic to neutral with the pH of (5.6 to 7.0).

##### 5.1.1 Effect of Sole or Combine Mineral and Organic Fertilizers with Rhizobium on Soybean in 2015/2016

The experiment consisted of six (6) treatment in 2015 and nine (9) treatment in 2016 and the result are discussed here.

Observation of tall plants at 6, 8 and 10 weeks after sowing when I+P was applied in 2015 implies that inoculation and phosphorus promotes early growth in soybean. This finding can be supported by Zerpa *et al.* (2013) who reported that, rhizobium inoculation favors shoot height of soybean. Mehmet (2008) also reported that mineral fertilizer applied to soybean plots had significant effect on plant height compared with control plots without nutrient addition. The tallest plants produced by I+P+K+M + OM in both years could be attributed to the presence of inoculant, organic and

inorganic nutrient in the treatment which might have helped the plant to produce higher number of nodules, firm rooting system and conducive environment for microbial activities. Khahim *et al* (2013) reported that application of organic manures in combination with recommended chemical fertilizer produced taller plants compared to the control.

Number of branches produced by plot treated with all nutrient combination was marginally greater but not different from the plot treated with I + P. This implies that application P in combination with Rhizobium inoculation can produce similar number of branches as other nutrients combinations, thus making P nutrition to be critical to production of soybean branches in the presence of rhizobium inoculant. Abdul and Saud (2012) have earlier reported that application of P leads to high number of soybean branches. The highest number of branches produced in plot treated I+P+K+M+OM in both years could be attributed to its ability to give more support to the fixation of atmospheric nitrogen, increase microbial activities and tallest plants produced which enhanced the utilization of solar radiation and in turn increased the number of branches. It has been observed earlier that, a combination of inorganic and organic manures can enhance growth and yield attributes of soybean (Lourduraj, 2000).

Similar effects observed when I+P+K+M+OM and I + P were applied on Shoot biomass is consistent with that of the number of branches produced and may be point to the fact that application of phosphorus in the presence of rhizobium inoculants may be sufficient to produce the optimum or economic yield of soybean. The interactions between bacterial inoculation and different levels of phosphorus had obvious effect in increasing the weight of the dry matter. The heaviest shoot biomass produced by I+P+K+M+OM in 2015 could be attributed to the tallest plants

and highest number of branches produced by the treatment in the same year and this may be due to the presence of rhizobium and P in the combination which was further enhanced by other nutrients applied. Yusuf *et al.* (2012) also reported that the use of inoculant increased biomass. Osunde *et al.* (2013) also reported increase in shoot biomass on inoculated compared to the uninoculated plots. It has also been observed that, response of soybean to inoculation is sometimes massive, more often with at least 20% increase in biomass (Giller, 2010).

The highest nodule numbers produced by I+P+K+M+OM in 2015 and I+P+K+M in 2016 could be attributed to the presence of inoculant in combination with organic or inorganic nutrient especially P which might have allowed more conducive environment for plant microbe interaction and enhanced nodule production. This finding is in line with Zerpa *et al.*, (2013) who reported that inoculated plots produced more nodules than un-inoculated plots. Muhammad 2010 also reported that inoculant increases all parameters including grain yields over those of the control. Increased nodulation observed in the current study could be due to high competitive ability of inoculant *bradyrhizobia* as noted by Okereke *et al.* (2000). The heaviest nodule weight produced by I+P+K+M could be attributed not only to the presence of K in the treatment, but also to the numerous nodule produced by the treatment. Yusuf *et al.* (2012) reported that the use of inoculant increased nodulation compared with the uninoculated. Whereas, Vyas *et al.*, (2005) observed that, Potassium application had significant positive effect on growth and nodulation in soybean and favorably influenced the quality parameters of soybean as well.

The highest number of pod per plant produced by I+P+K+M+OM in 2015 and I+P+K+M in 2016 could be attributed to their ability to produce tallest plants, highest number of branches and highest

nodule number that enhanced utilization of growth factors which in turn translated into highest number of pods per plant. It has been observed that, use of organic manures in combination with mineral fertilizers could meet the need of micronutrients of soybean (Joshi *et al.*, 2000). Lourduraj (2000) has also reported that the combined application of inorganic and organic manures significantly enhanced the growth attributes and yield of soybean as compared to the sole application of either of them. Khahim et al (2013) reported that application of organic manures in combination with recommended chemical fertilizer significantly increased pod number compared with the control.

The highest grain yield produced by I+P+K+M+OM may be attributed to its ability to increase nodule number, plant height, number of branches and number of pods per plant but was statistically similar to the plot where I + P alone was applied, pointing to the fact that there may be no need applying other nutrient if I+P is already applied. Ronner *et al.* (2016) reported an increase in grain yield in plots containing I and P. It was observed that, P fertilization increased growth and yields of promiscuous soybean cultivar (Abdul and Saud, 2012). Aliyu *et al.* (2013) reported that inoculation has the ability to improve the productivity of soybean. Whereas, in another related experiment, plants from the uninoculated plot (control) produced lower grain yield than plots from inoculated plots (Manchehr *et al.* (2013). Rashid et al. (1999) stated that rhizobium inoculation significantly increased pod yield. Khahim et al. (2013) reported that application of organic manures in combination with recommended chemical fertilizer significantly influenced grain yield of soybean over the control. Khalid *et al.* (2011) also observed that inoculation with *Bradyrhizobium*, chicken manure and Sulphur significantly ( $P < 0.05$ ) increased the grain yield (g/plant) and yield (kg/ha) compared to the uninoculated and untreated control

The heaviest stover yield produced by I+P+K+M+OM and I+P+K+M could be attributed to the ability of the plants to produce tall plants, high number of branches, high number of pods. Number of branches and plant height is a major determinant of stover yield. Yusuf *et al.* (2012) also reported that the use of inoculant and P source increased haulm yield of groundnut compared with the uninoculated. The highest haulm yield obtained with to cattle manure may be attributed to increase soil fertility as results of added manure resulting in increasing the plant growth and therefore accumulating biomass. Decomposition increase the total organic carbon content of the soil most of which are retained in the macro aggregation fraction (Tisdall and Oades 1982). Agbenin and Goladi (1997) recorded higher organic matter content with an addition of farmyard manure. The inclusion of Agrolyser supply micro nutrients that are probably enhanced nutrients assimilation and dry matter accumulation. Agrolyser contains micronutrients such as boron and zinc that enhanced the production of hormones (Adjeil *et al.*, 2002). Khahim *et al.* (2013) reported that application of organic manures in combination with recommended chemical fertilizer significantly influenced stover yield of soybean.

### **5.1.2 Effects of Sole or Combined Mineral and Organic Fertilizers with Rhizobium on Soybean; Average over the Two Year Planting Period**

The effects of combining rhizobium inoculation with both mineral and organic fertilizers were examined on average over 2015 and 2016 planting seasons. This assessment considered only the treatments levels common to the two seasons. Average over 2015 and 2016 planting seasons, shorter plants characterized the control plots which was similar to that of Inoculation (I) alone both at 4WAS and 8WAS. This may be due to the fact that inoculation alone is not enough to support plant growth and that inoculation should be combined with either mineral and/or organic fertilizers.

Tall plants characterized plots treated with I+P+K+M and I+P+K+M+OM at 4WAS and 8WAS which was significantly similar to plots that received I+P. this implies that phosphorus fertilizer can support plant growth confidently with or without the addition of any other fertilizer or organic manure. Abdul *et al.*, (2012) reported that increase in phosphorus application significantly increased plant height. Ahiabor *et al.*,(2014) also observed that the application of phosphorus fertilizer and rhizobium inoculant impacted on the growth of soybean. Plant height increased within the fertilizer application regime. Lamptey *et al.*, (2014) reported that the application of TSP and rhizobium inoculant increased plant height and ascribed the increase to pronounced vegetative growth. Plant height increase with increase P application could be due to the fact that P being essential constituent of plant tissue significantly influences the plant height of crop (Kumar and Chandra, 2008; Shahid et al., 2009) were also observed significant improvement in plant height of soybean by P-fertilization.

Heavier shoot biomass observed in plots treated with I+P+K+M at mid- flowering was similar to weights obtained from plots treated with I+P, I+P+K & I+P+K+M+OM. This could be attributed to the fact that phosphorus support early growth. On the other hand the light weight was recorded in plots treated with control and I only which indicates that inoculation has no effect on shoot biomass. Asia *et al.*, (2005) observed that phosphorus application increased biomass yield by 20.7% greater than the control. Lamptey *et al.*, observed that rhizobium inoculation with P increased shoot weight.

Observation of high number of pods per plant in both years in plot treated with I+P+K+M which was similar to plots treated with I+P and I+P+K+M +OM but significantly different from plot treat with inoculation only and control plots. Abdul *et al.*, (2012) reported that increase in Phosphorus level along with bacteria inoculation increased number of pods.

Higher number of nodule was observed in plots treated with I+P but was statistically similar to other treated plots except control and inoculant only this means soybean planted in soil without native rhizobia would have difficulty in fixing nitrogen through BNF. Similarly plots treated with I+P had heavier nodules. Abdul and Saud (2012) observed that bacteria inoculation and phosphorus increased nodule per plant. Other reports showed that nodule weight increased in response to phosphorus level in interaction with bacteria inoculation, (Abdul *et al.*,2012). Solaiman *et al.*, (1990) reported that the application of phosphorus along with rhizobium inoculant influenced nodulation.



High Grain yield characterized plot treated with I+P which was significantly not different from I+P+K+M+OM this means that nutrient element I+P can consistently improve grain yield compared to treatment where inoculation or control was applied this showed that inoculation alone cannot improve grain yield. Ronner *et al.* (2016) reported an increase in grain yield in plots containing I and P. It was observed that, P fertilization increased growth and yields of promiscuous soybean cultivar.

Heavy Stover also characterized plot treated with I+P and I+P+K+M but was not significantly different from treatment containing all the nutrient element, this means that without P, or K or Om being combined together, I +P alone can produce maximum stover yield compared to Inoculation plot alone which was not different from the control where low yield stover was obtain. This showed that inoculation alone cannot increase stover yield.

## **5.2 CONCLUSION**

Growth and yield parameters of the soybean measured improved with treatment combination of both mineral and organic fertilizer coupled with Rhizobium inoculation. Generally, plants that received fertilizer performed better than plants that were not fertilized as well as those plants that received inoculation only. However, the positive effects of P fertilizer application on growth and grain yield of soybean in this study were significantly boosted in the presence of Rhizobium inoculant and organic matter. The growth and grain yield enhancements observed in this investigation may also be attributed to an increased symbiotic relationship of rhizobia (bacteria) with the roots of leguminous crops resulting in the possible fixation of atmospheric nitrogen into the roots of soybean which was favored by P nutrition.,

### **5.2.1 RECOMMENDTION**

This study therefore advices that farmers to combine inoculation with low levels of mineral and organic fertilizer and / or micronutrient for improved soybean production.

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