Effectiveness of promising commercial bio-fertilizers on soybean production in Bungoma county, western Kenya

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Abstract

The study was conducted to compare the performance of promising commercial bio-fertilizers that have been evaluated under the green-house conditions at TSBF-CIAT, in farmers' conditions through the use of promiscuous soybean variety (SB19). The trials were laid out on small scale farms in Bungoma County, situated in Western Kenya. The experiment was established in March 2010 during the long rains (LR) and repeated during the short rains (SR) of 2010; laid out in multi-locational and used individual farmer field as areplicate. Treatments were not replicated within each field. During LR 2010, 50 farms were researched on and 100 farms in the second season (SR 2010). A promiscuous medium-maturity soybean variety TGx1740-2E (SB 19) was inoculated with Legumefix (Rhizobia) or/and Rhizatech (mycorrhizae) inoculants. The mycorrhizae inoculum was applied to the soil in the seed furrows at the recommended rate of 30 kg ha^{-1}. Nodulation was examined at mid-podding (50% podding) by carefully uprooting all plants with their entire root system from a 1 m^2 section in each plot. Nodules were counted and weighed; the root and shoot parts separated, and fresh and dry weights assessed. Analysis of variance was conducted to determine the effects of (and interactions between) the two inoculants on plant parameters using a mixed linear model (MIXED procedure, SAS). Rhizobial inoculation resulted in significantly ($p<0.001$) higher nodule biomass (0.93 g per plant) compared to the control (0.27 g per plant) across many farms. Mycorrhizal inoculation had no significant effect on nodulation when applied solely (0.38 g per plant), but co-inoculation of Rhizobia and mycorrhizae increased nodule biomass further by 0.09 g per plant There was a significant difference ($p<0.001$) in terms of biomass yield between treatments. Rhizobial inoculated plants had the highest biomass production of 2086 kg/ha. Rhizobial inoculation resulted in higher grain yields of 1116 kg/ha above the control. Soybean inoculation increased both nitrogen and phosphorus uptake in the biomass. Rhizobial inoculant had the highest soybean N uptake of 48.6 N kg/ha which was significantly different ($p<0.05$) from control and sole application of mycorrhizae. Statistical analysis showed that soil factors (pH, P, C, N) significantly ($p<0.001$) affected soybean grain yields during both seasons. It is concluded from this study that rhizobial inoculants have a high potential as commercial bio-fertilizers. However, there is need to target these inputs to the most responsive fields. Further studies are needed to elucidate the conditions under which synergism between both inoculants may occur, with specific focus towards soil P availability and management of P inputs.

Key words: soybean, Rhizobia, Mycorrhizae, Inoculant.

Introduction

Soybean [*Glycine max* (L.) Merrill] is an annual legume that belongs to the legume family *Fabaceae*. It is a strictly self-pollinating legume. World demand for soybean has been able to absorb ever-increasing production at prices that are profiTable to producers. Since 1970, world consumption of soybean has grown at an annual rate of 4.8% on average and since the 1990s it showed an annual increase of 5.4% on the average (Okalebo *et al.,* 2005).

In Western Province, mixed cropping, with minimal nutrient inputs are the norm and crop rotation is secondary to continuous maize cropping. Few farmers recognize the benefit of improved soil fertility through nutrient recycling. Leguminous intercrops and improved short fallows contribute nitrogen (N) to the soils through litter falls and biological nitrogen fixation, but this process is not widely recognized as beneficial by farmers. On the other hand, mineral fertilizers and livestock manure are considered important inputs, but are usually in short supply (Chianu, 2009).

The high cost of chemical fertilizers and other inputs has not favored increased food production. One way of increasing food production without degrading the environment is through bio – intensive farming (Chianu, 2009). An indirect benefit of growing soybean is the change they introduce in crop rotations, by acting as break-crops to slow down the build-up of cereal pests, diseases and weeds thus reducing the need for pesticides in subsequent cereal crops (Mahasi *et al*, 2009). Due to lack of alternative crops, most farmers practice continuous cropping (mostly maize, cassava, sweet potatoes and cotton).

Soybeans that nodulate effectively with diverse indigenous Rhizobia are considered as promiscuous (Kuneman *et al*, 1984). Hence, promiscuous genotypes of soybean form symbiotic association with available *Rhizobium* strains in the soil and thus fix atmospheric nitrogen whilst non-promiscuous genotypes need specific rhizobial strains to fix nitrogen from the air.

In the late 1970s, breeders at IITA observed that most high yielding soybean cultivars from USA have specific requirements for *Bradyrhizobium japonicum* (Pulver *et al.*, 1982) and inoculation of these varieties was found to be essential when growing them under tropical conditions of low soil nitrogen. In the early 1980s, it was assumed that most tropical countries did not have the facilities and personnel required for inoculum production, storage, and distribution and were dependent upon importation of the final product. The non abundance of commercial *Bradyrhizobium japonicum* inoculants and nitrogenous fertilizers led to the option of breeding promiscuous cultivars in IITA since soybean genotypes that do form symbiotic association with indigenous cowpea-type Rhizobia were identified. Generally, soybean varieties developed for promiscuous nodulation with the indigenous Rhizobia were considered to increase production of soybean in tropical Africa with minimum cost affordable to small-scale farmers (Giller & Wilson, 1991).

Materials and methods

Experimental site

The trials were laid out on small farms in Bungoma County, situated in Western Kenya. The district lies between latitude 00^0 34′ N and longitude 34^0 34′E. Bungoma County falls under two major agro-ecological zones: the transitional upper midland zone UM4 (referred to as the maize-sunflower zone) and the Lower Midland zones which cover a greater proportion of the district (LM1-LM3).

The district has a bimodal rainfall pattern, with the first growing season (long rains) extending from March to August, and the second (short rains) from October to January. The district has generally well-distributed annual average rainfall of 1000-1800 mm, depending on the location (TSBF, 2009). The temperature in the district ranges from about 20-22°C in the southern part of Bungoma to about 15-18°C on the slopes of Mount Elgon in the northern part of the district.

Field layout and design

Performance of soybean was tested with rhizobial and myccorhizal bio-inoculants. The experiment was established during the long rains (LR) and repeated during the short rains (SR) of 2010 laid out in a multi-locational one farmer field one replicate design. Since one of the objective was to assess the correlation between selected soils chemical properties on bio-fertilizers performance within a large geographical area in terms of soybean grain yields, treatments were not replicated within each field or farm: instead, farms and seasons were considered as replicates (Pypers, 2010), with 50 farms in the LR 2010 and 100 farms in the second season (SR 2010). Treatments were allocated in new farms each season to avoid contamination and residual effects of the inoculants. Soil characterization was done on each farm so as to determine the soil types and properties in each farm.

Land preparation

Land preparation was done in February 2010 for long rains and September 2010 for short rains using hand hoes. Fine-seedbed preparation was also done by hand prior to demarcation of plots. All the initial land preparations for the two cropping seasons were done by farmers themselves to facilitate the adoption of technologies through their participation in the experimentation.

Soil sampling

Plots of 10 x 10 m area were demarcated and zigzag method used to sample the soils giving a total of 9 sub-samples per plot. The top 0-2 cm soil layer was removed to avoid sampling excess debris and samples taken up to 15 cm depth with a soil auger. The sub-samples were thoroughly mixed and 500 g composites were packed in polythene bags for laboratory analysis. The samples were analyzed for pH, organic carbon (C), total N and available P, according to Okalebo *et al.,* (2002). Other routine analyses on cations, micronutrients were not performed due laboratory limitations.

Planting

The treatments were administered into plot sizes of 10 m by 10 m. A promiscuous medium-maturity soybean variety TGx1740-2E (SB 19) that was recommended across locations in Western Kenya by Mahasi *et al.,* (2009) was inoculated with either or both inoculants and planted at 50 cm between rows and 7.5 cm between plants in the rows to give a soybean population of 266,667 per hectare. Each experimental plot had nine rows. Rhizobial inoculation (Legumefix, Legume Technologies, UK, containing 532c strain of *Bradyrhizobium japonicum)* was done by thoroughly mixing 125 g of damp seed with 2 g of inoculum (1×10^{-9} CFU g^{-1}) as per the manufacture's recommendation. The mycorrhizal inoculum (Rhizatech, Dudutech Ltd., Kenya, containing spores and mycelial fragments of *Glomus intraradices* (50 propagules/cm^3)) was applied to the soil in the seed furrows at the recommended rate of 30 kg ha^{-1} by the manufacture. The germination and emergence were uniform in all the treatments and there was no visual observation on detrimental effects from the treatments. Apart from the technical operations such as treatment application and data collection; all the other operations were managed by the individual farmers.

Plant sampling for biomass and tissue N analysis

Nodulation was examined at mid-podding (50% podding) by carefully uprooting all plants with their entire root system from a 1 m^2 section in each plot. Nodules were washed, counted, put in zip lock bags and weighed. The root and shoot parts were separated, and fresh and dry weights assessed. Pods were also separated from the shoots, fresh and dry weights assessed.

Harvesting

Soybean was harvested at physiological maturity when the pods were dry but not yet shattered in August 2010 for the first crop and second crop was harvested in January 2011. All the plants in the entire plot were harvested and grain yields measured by weighing the dry soybean grain yields produced from each plot.

Statistical analysis

Analysis of variance was conducted to determine the effects of the inoculants and their interactions on plant parameters using a mixed linear model (MIXED procedure, SAS Institute Inc., 2003). The effects of different treatments were compared by computing least square means and standard error of difference (SED): significance of difference was evaluated at $p<0.05$ level of probability. In the mixed model analysis, farmer group nested within site and season were considered as random factors (Pypers, 2010) while the treatment effects (biofertilizers) were evaluated as fixed factors as shown in the SAS model below.

$$Y = X\beta + Z\gamma + \varepsilon$$

Where: Y = Yield (observation), β = treatment (biofertilizer) effect with known design matrix X, γ = denotes the farmer group within site and season which are considered as a random - effects parameters with known design matrix Z, and ε is an unknown random error vector whose elements are no longer required to be independent and homogeneous (MIXED procedure, SAS Institute Inc., 2003). Pearson correlation analysis was done to determine the effect of selected soil chemical properties on soybean grain yields.

Results

Initial soil characterization

The major soil type in the experimental sites was Haplic Ferralsols. These soils are characterized by deep yellowish or reddish colour, highly weathered, high permeability and sTable micro-structure, with very low CEC. They are also chemically poor, with low pH and nutrient reserves, high P fixation, easily depleted by agricultural practices (TSBF, 2009), thus the pH of the soils in surface (0 - 15 cm) ranged from 4.4 to 7.8 in the 44 farms with a mean of 5.46 during Long rains of 2010 and a pH of 5.39 in 63 farms in second rains of 2010. Available phosphorus in surface soils (0-15 cm) by the Olsen *et al.,* (1954) sodium bicarbonate extraction, ranged from 1.31-34.64 mg Pkg^{-1} during LR 2010 and from 1.1 to 40 mg Pkg^{-1} during SR 2010. The total N content in soils was low to moderate (0.05 to 0.25 %N) as per Okalebo *et al.,* (2002) in Bungoma farms during both seasons. The carbon (or organic matter) contents of soils were low to moderate (0.5-2 %C) according to Okalebo *et al.,* (2002) with a mean of 1.32%C during LR and 1.42%C during SR seasons of 2010.

Treatments effect on soybean biomass yield at 50% podding

Biomass yield is an important measure of plant vigor and health. There was a significant difference ($p<0.001$) in terms of biomass yield between treatments in this study. Bradyrhizobium inoculant treated plants had the highest biomass production at 2086 kg/ha. Therefore, the N biofertilizer (Legumefix) can be used as an alternate or as a supplement to N fertilizer to increase agricultural production with less input capital and energy. There was no significant difference ($p>0.05$) between sole rhizobial inoculation and co-inoculation at 2048 kg/ha biomass yield but there was a significant difference ($p<0.001$) between rhizobial and control yielding 1572 kg/ha and/or mycorrhizal inoculation at 1673 kg/ha. Rhizobium inoculant produced higher quantities of biomass, and likely made highest contributions from N fixation.

Treatment effects on soybean grain yield

Rhizobial inoculation resulted in higher grain yields than control. There was no significant difference between sole application of rhizobial (1116 kg/ha) and the co-inoculation (1027 kg/ha) at ($p>0.05$). Low yield in mycorrhizal treatment could be due to the mycorrhizal strain rather than the conditions of the soil and can be attributed to the quality of the strain that might be inferior. Low soybean grain yields in Bungoma even after inoculation (less than 2 t/ha) could be attributed to high soil acidity within the farms (4.5-5.9). Soybeans are very sensitive to soil acidity and prefer a soil pH between 5.8 and 7.8. Rhizobial inoculation also increased the average grain yield by 21% over control treatment. This is because N fixed due to inoculation resulted into high biomass yield. The high biomass implies increase in the rate of photosynthesis due to high leaf number and leaf area. The photosynthates are transported via phloem and used in grain yield production (Majengo *et al.,* 2011). Control plots gave poor results, as well as the mycorrhizal product, though that was not expected, given the good performances observed under greenhouse conditions.

Accumulation of plant N and P by soybean biomass and grains

Rhizobial inoculation increased both N and P uptake in the biomass and grains. Rhizobial inoculant contributed to the highest soybean N accumulation of 48.6 N kg/ha and was significantly different ($p<0.05$) from control treatment and sole application of Mycorrhizae. This is because the rhizobial inoculum contains the strains of *Bradyrhizobium japonicum* which are able to fix N through the BNF process hence high N accumulation.

Correlations between selected soil parameters and soybean grain yields

The initial soil pH was positively but weakly correlated ($p<0.05$, $r = 0.19$) with grain yields during LR 2010 season (Table 4.6). This could be attributed to low soil pH that induced deficiency in some essential nutrients, for example P and Mo, thereby leading to a reduction in the number of nodules and BNF (Marschner, 1995 Insert reference in list). There was significant correlation ($p<0.001$, $r=0.42$) between soil organic carbon and soybean grain yields. Since the quantity of humus in soil is influenced by the quantity of carbon compounds added, the availabilities of N, S and P compounds is increased in soils with high organic C. At moderate level of carbon, the soil is able to supply the plant with essential plant nutrients hence a high significant correlation with grain yields ($p<0.001$) found in this study. Highly significant correlation ($p<0.001$) between soil available P and soybean grain yields could be attributed to the fact that P bioavailability is a major factor limiting N fixation.

Conclusions, recommendations and further studies

Bradyrhizobium inoculants have a high potential as commercial biofertilizers and can partially substitute the need for mineral N fertilizer in legume farming systems. However, there is need to target these inputs to the most responsive fields. Legumefix inoculant (Bradyrhizobium) was more effective compared to Rhizatech inoculant (mycorrhizae) under local field conditions in Bungoma at moderate soil N and P. Co-inoculation of Bradyrhizobium with mycorrhizae did not result in increased nodulation or soybean yield compared to sole rhizobia inoculation. Selected soil chemical properties (pH, Olsen P, N and C) and nodule weights had significant effect on soybean yields during both seasons. 5.2 Further studies Further studies are suggested to elucidate the conditions under which synergism between both inoculants may occur, with specific focus towards soil P availability and management of P possible inputs in the low P soils and also to determine the contribution of environment and plant interactions in soybean production, especially as soybean yields are still disappointingly low in most Kenyan conditions. Screening of other cultivars is also suggested.

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