Effect of Cobalt, Molybdenum and Rhizobium on Yield of Soya Beans in Kisii County, Kenya.

Ombuna Dorice[1] , Mworia Mugambi[2], Muchiri John[3]

[123] Kenya Methodist University.

Department of Agriculture and Rural Development

Email: dorisombuna1@gmail.com, mworia. mugambi @kemu.ac.ke, john.muchiri @kemu.ac.ke

Correspondence: Dorice Omwocha,

: Email: dorisombuna1@gmail.com

Abstract.

Introduction: Soybean is a leading source of protein and oil used for human consumption and livestock feed, also considered a valuable crop globally. The leading producers worldwide are United States of America, Brazil and China that produce as high as 89.9 Million tonnes annually. In Kenya, the beans are mainly grown in the western and central regions of Kenya, and the average yield is 363 $kgha^{-1}$. This is low compared to the other countries, the poor yield in Kisii County is associated to low soil fertility, poor agronomic practices and acidified soil. Additionally, insufficient nitrogen and phosphorus in soil, use of unimproved seed varieties and poor management practices are causes. **Objectives**: The study aimed at evaluating the effects of molybdenum, cobalt and rhizobium inoculants on yield of soya beans in Kisii County. **Research methodology**: A randomised complete block design experiment was carried out at Kisii Agricultural Training Centre. Seeds were coated with ttreatments that included: Control (WT), DAP, Rhizoliq Top Soya 3m/kg, Wuxal Extra COMO1.5 at 1m/kg of seed, Wuxal Extra COMO 1.5 at 1.5 mls/kg of seed, (Wuxal Extra COMO 1.5 at 1.5 mls/kg / Rhilizic top 3mls) and (Waxul Extra15 COMO1m/Rhizoliq top 3ml/kg) of soya seed. Parameters collected were manipulated using SPSS Version 24.0, and tested using ANOVA (95% CI). Statistically significant results were further subjected to DMRT post hoc test. **Results:** Soil analysis showed moderately acidic (pH 5.5) soil low nitrogen (0.12%), moderate Phosphorus (20ppm) and low potassium levels (40%). The highest effect on six plants pod weight was shown by COMO1ml+ Rhizobium treatment. The treatments were statistically significant (p=0.004) and COMO1ml+Rhizob was highest on DMRT. Highest effect on 100 seeds weight was also seen with COMO1.5ml+Rhizob and p value (<0.001).DMRT showed como1.5 + Rhizob to be the highest. Further, COMO1ml+Rhizob indicated the highest effect on yield Kg per ha, (p=0.006). Weight of seeds of the plot was shown to be influenced highly by COMO1ml+Rhizob with (p=0.005). COMO1ml+Rhizob had the highest mean on DMRT. **Conclusion:** Use of inoculant in combination in improving low yield (COMO+Rhizob) at different graded levels is recommended. Research to evaluate the effects of nitrogen and phosphorus levels on soya bean production in Kisii is highly recommended.

Key words: *Agronomic*, *Combination, Nitrogen, Phosphorus, Fixation, Production, Treatments, Soil.*

Introduction

Soya bean is one of the most popular leguminous plants grown worldwide (Ntambo et al., 2019).). Alongside other grain legumes, soya beans grow in diverse environmental conditions(Mathenge et al., 2019). It is recognized to be a rich source of oil and protein, widely used for human consumption and animal feed. The protein content in soya beans is as high as 35-40%, contributing 58% to the world's oil seed production. It is a valuable cash crop in the manufacturing industry (Ohyama et. al., 2017). Historically, soybeans originated from East Asia (Sedivy et al., 2017) and the largest producers now are the United States of America (USA), Brazil, India and Argentina, contributing 82% of total world production (Ohyama et al., 2017). According to Food and Agriculture Organization [FAO], cultivation and yield of soya beans worldwide improved since 1961 to more than 340 million metric tonnes in the year 2016 (Mbebe et al., 2019).

The world soybean production in the 2017/2018 crop season was approximately 346.9 million Mg (Soymeal Info source, 2019). According to FAO the Sub-Saharan Africa region has remained a sole importer of soybean and soybean products, except for Uganda which is self-reliant in soybean production (Mbebe et al., 2019) . Kenya is among the lowest soya producers with 6000 - 7000 Mt, its domestic consumption is 150,000 (Mathenge et al., 2019). This means that the country continues to spend earnings on soybean imports to supplement local production. According to (Mbebe et al., 2019), statistics from FAO, indicate that in 2018, Kenya imported 68,000 tons of soy meal; this figure had tripled in 10 years**.** Kenya also spent USD 12.8 million on importation of soy oil alone in 2016. In Kenya, cultivation of soya beans is commonly in the maize growing areas and by small-scale farmers. Soya beans is grown mainly in the Kakamega, Vihiga, Busia, Bungoma, Transnzoia, Siaya, Homabay, Migori, Kisii and Nyamira counties. Kirinyaga, Embu, Meru and TharakaNithi Counties are among other soybean producing zones (Mathenge et al., 2019). There is paucity of actual data on the yield and productivity of soya bean in Kisii County.

Soya beans is cultivated from the Equator to latitude 55degN or 55degS, and 2000 m below sea level. In Kenya where the altitude is from 0 to 2200m above sea level. The rainfall required for favorable cultivation is usually 300 to 1200 mm per annum and pH between 6 to 6.5. Soya bean also thrives in warm climates that range from low to medium altitudes (21degC to 30 degC). (Infonet Biovision, n.d.). It is important to note that high altitude zones negatively affect the flowering and maturity of soybeans, high temperature affects the initiation of flowering. Such conditions renders the plant to a vegetative state. The nitrogen-fixing bacteria do not also flourish in low soil pH of 4.2 (Carlos Felipe Cordeiro & Echer, 2019).

Nitrogen and phosphorus are essential micronutrients for plant development and yield. Although approximately 78% of nitrogen is available in the atmosphere, it is inaccessible to plants. Normally, the atmospheric nitrogen is changed into forms that can be accessible to crops by a process referred to biological nitrogen fixation (BNF), nitrogen (N_2)is changed into NH_3 by N_2-fixing microorganisms (Loboa et al., 2018). Phosphorus and nitrogen micronutrient deficiency is known to limit the growth and yield of soybean (Bulgarelli et al., 2017). Due to the mutual symbiotic relationship with some microbials in soil, legumes can increase nitrogen (N) amount through biological N-fixation (BNF). However, to optimally achieve that, phosphorus (P) is needed is high amounts because of the required energy conversion in nodules. Moreover, P is vital in root development, nutrient uptake and development of crops (Mitran et al., 2018). Therefore, P levels is always maintained above respective critical levels. The critical level for phosphorus is 15 ppm and the maintenance range for soybeans is 15 ppm, so P soil test levels has to be maintained between 15 and 30 ppm (Abdulaha-Al Baquy et al., 2017).

In one of the studies on the effects of Mg combined with N on soybean nodulation was observed, the authors emphasized on the importance of N and Mg in regulating root system development and nodule establishment (Egamberdieva et al., 2018). Enhancing rhizobial nitrogen fixation in soybean is anticipated to be of value to small scale farmers through better production and improved integrated soil fertility for consequent crops (Heerwaarden et al., 2018). Biological nitrogen fixation of crops is strictly dependent on proper growth, development and the leghaemoglobin content of the root nodules, which is the means that crops meet their required nitrogen amount. BNF is a technology is a game changer only if farmers can afford (Loboa et al., 2018). Soybean is rated the top most inoculant-using crop globally, the plant carries bacteria belonging to the genus *Bradyrhizobium* (Santos et al., 2019)

Use of inoculants such as *Brady rhizobium japonicum, bradyrhizo-biumelkani and sinorhizobium fredi* are known to produce root nitrogen fixing nodules. Due to the limited availability of *Bradyrhizobium* in agricultural soils, application in the form of bacterial preparation is recommended when planting soya beans (Filho et al., 2017). Combined strains of inoculants is noted to be more effective than single strains. The use of two strains increases the chances that at least one would nodulate and perform well with the legume. For example, in Brazil, the farmers preferentially used the combination of two *Bradyrhizobium* strains for the soybean crop since the 1950s. Particularly in the last decade, the use of inoculants containing microorganisms of “different type” has expanded (Santos et al., 2019). The thought is that, combination of strains or species acting in different microbial processes, results in combined benefits, resulting in higher yields. Available inoculants in the market that have been tested and proven to increase yield include molybdenum, cobalt and rhizobium among others (Meena et al., 2018).

The low soybean yield in SSA is attributed to the use of poor-performing varieties, inadequate nitrogen, phosphorous, organic matter and acidified soil acidity (Mathenge et al., 2019). Additionally, farmers in Kenya also face numerous challenges ranging from production, including biotic, abiotic and socio-economic factors among others. Competition and marketing from cheap imports of soybeans are also factors that limit domestic production. Additionally, due to the high cost of inputs farmers are not able to access inoculants, and end up using varieties’ that have limited ability to fix nitrogen in the soil (Mbebe et al., 2019). Soybean is fundamental in development planning of countries of SSA considering its economic importance in nutrition, food security and industrial use (Mbebe et al., 2019). To increase soybean production in Kenya, integrated soil fertility management using soya beans is an option towards sustainable intensification of smallholder agriculture (Mathenge et al., 2019). Use of BNF technology in soybean cultivation is anticipated to be a game changer to farmers and a cost-effective option to increase yield in Kenya.

In general, to ensure sustainability of soya beans production in countries, an in depth understanding of the interaction effects of different combination of inoculant is of paramount importance in years to come. Effective application of co-inoculants is anticipated to increase soya beans growth and productivity (Meena et al., 2018). The hypothesis of this research was that interaction of cobalt, molybdenum and rhizobium does not significantly affect yield of soybean. Therefore, the study aimed to evaluate the effects of cobalt, molybdenum, and rhizobium on yield of soya beans in Kisii County.

Materials and Methods

a) Study site description.

Four replications in four blocks randomised complete block design (RCBD) experiments were conducted at Kisii Agricultural Training Centre. The agro ecological zones, lies between 1000m- 1500m above sea, humid with an average temperature of 21-30 ^{0}C, the annual rainfall averages 1500mm**.** The study area is characterized by a hilly topography with red volcanic soils (75%), rich in organic matter. The experiment was carried during the short rain season of September-December 2018 (Kisii County, 2020).

b) Source of seeds and inoculants

SC SAGA soybean cultivar and inoculant were obtained from Bayer Company Kenya. The cultivar variety is certified by KEPHIS (*Infonet Biovision*, n.d.)

c) Soil sampling, analysis and planting.

The soil samples were collected from uniformly distributed point with an auger at the experimental site using the grid sampling method. It was then analysed using dried chemistry process at CROP NUTS and KARLO Laboratories in Kisii. Tests performed organic carbon, total N, extractable P, and K, soil pH (water), electrical conductivity and particle size distribution. Soil pH (1:2 soil/water), organic carbon (%), Textural class, N (kg ha), P (kg ha^{-1}), K (kg ha^{-1}), S, Zn, Mg, Cu, Fe, mn, Co and Mo. After planting, soil nutrients (NPK) and levels of Cobalt and molybdenum were also analysed. Seeds were also for viability then inoculated before planting. Further, a sample of treated seed was taken for physical assessment of damage. The cultivars were then planted at a spacing of 45 cm by 10 cm at the onset of the rainy season giving a total population of 444,444 plants/ha. All blocks were planted at a rate of two seeds per hill, and on the 21st day thinned to one seedling per hill. Due to the use of biological products, all storage conditions and handling maintained.

d) Treatment and experimental layout.

The experimental blocks (28) were prepared, each measuring 3 x 2 m. Plots and replications were interspaced with footpaths of 2m spacing to prevent contamination. A total of seven treatments were used consisting of; Control (Without Treatment), DAP, Rhizoliq Top Soya 3m/kg, Wuxal Extra COMO 1.5mls at 1m/kg of seed, Wuxal Extra COMO 1.5 at 1.5 mls/kg of seed, (Wuxal Extra COMO 1.5 at 1.5 mls/kg / Rhilizic top 3mls) and (Waxul Extra15 COMO1m/Rhizoliq top 3ml/kg) of soya seed. DAP fertilizer was applied to all treatments at the rate of 100 kg $ha^{-1.}$ The seeds were inoculated prior to sowing with the specific strain of Rhizobium at the rate of 20 kg/acre. A sample of treated seed was assessment for damage during the treatment.

Data collection and analysis

Six plants (6) were randomly selected per plot, were away from the edge of the plot and from the third plant inside the plot. Six plant pod weight, 100 seed weight, yield yield in kilograms per hectare and weight of seed whole plot were recorded.

Data collected was analysed using SPSS Version 24.0 statistical package. Analysis of Variance (ANOVA was performed to determine the treatment effects on harvest parameters mentioned above. All statistical comparisons undertaken at $\alpha = 0.05$ probability level, statistically significant results were further subjected to Duncan multiple range (DMRT) test- post hoc test for separation of mean.. Data was then visualised using tables and charts. Ethical clearance and permit to conduct the study was sought from NACOSTI and principal KATC through the Kisii County Director of Agriculture

Results.

Soils analysis done before planting showed moderate electrical conductivity (0.06 µs/cm), adequate levels of organic carbon (2.4%), manganese (1.5%) and phosphorus ppm (20). However, the nitrogen and potassium level was found to be low (0.12) and (40%) respectively. The soil was moderately acidic (5.5). Low soil pH and aluminum (Al) toxicity caused by soil acidification is one of the leading hindrances to soybean production in the world (Baquy et al., 2018). Therefore, test of soil PH is a very important component prior to planting and inoculation. According to Li et al., 2019, on soil analysis, liming rate was important in enhancing changes in soil pH and crop yield.

a) Effect of different treatments on six plants pod weight.

In this experiment, results on six plants pod weight showed that COMO1ml+ Rhizobium had the highest effect while WT (Control) had the lowest. ANOVA indicated a statistically significant difference among treatments (p=0.004). Further post hoc analysis (DMRT) showed COMO1ml+Rhizob as the highest.

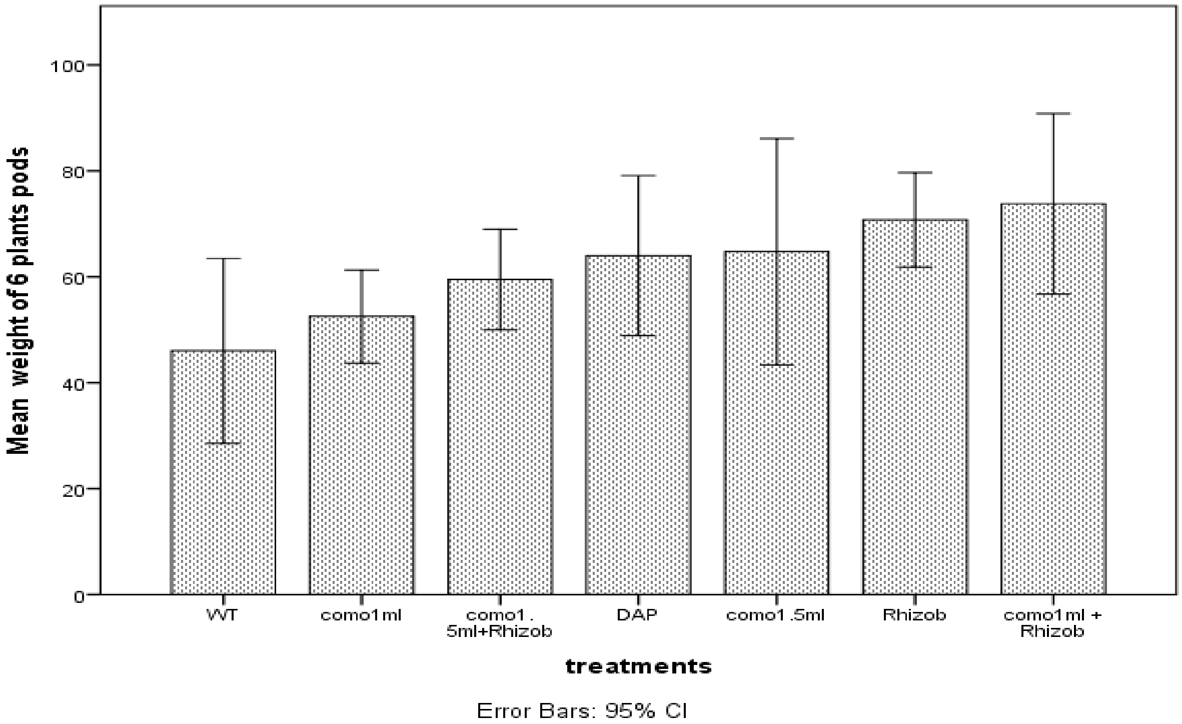

Figure 1: Mean weight of six plants pod.

b) Effect of treatments on 100 seeds weight

Effect of treatment on 100 seed weight revealed that COMO1.5ml+Rhizob treatment exhibited the highest effect, while WT (control) was the least. A strong statistical significant difference was also exhibited (<0.001), further, post hoc test analysis confirmed COMO 1.5 + Rhizob which was highest in 100 seed weight.

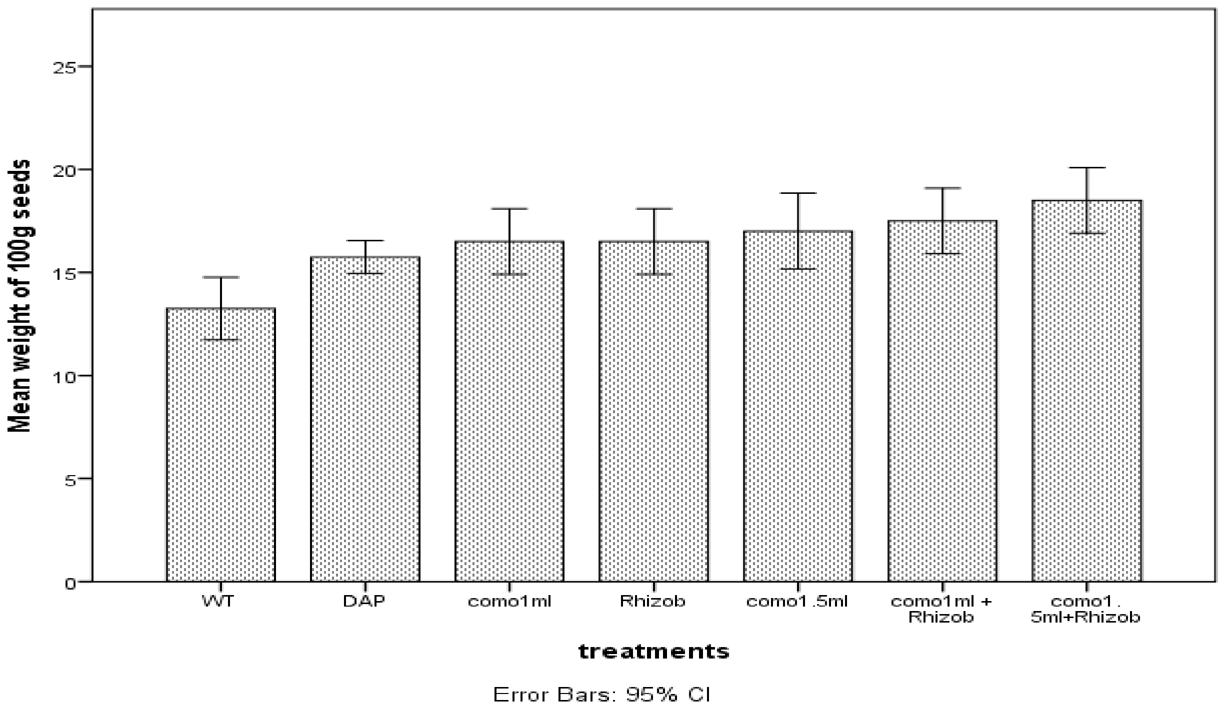

Figure 2: Mean weight of 100g seed weight

Table 1*:* Univariate ANOVA of treatments on 100 seed weight in grams.

Dependent Variable: weight of 100g seeds

Source	Type III Sum of Squares	Df	Mean Square	F	Sig.
Block	5.143	3	1.714	2.149	.130
Treat	65.357	6	10.893	13.657	.000
Error	14.357	18	.798		
Total	84.857	27			

a. R Squared = .831 (Adjusted R Squared = .746)

c) Effect of treatments on yield in kilograms per hectare

At maturity all plants per treatment (per plot) were harvested, sundried, threshed and winnowed, the grains were then sundried at moisture content of 13%, weighed in kilograms per hectare. The highest yielding treatment was COMO1ml+Rhizob. ANOVA showed that there was significant difference among the treatment's yield per ha, (p= 0.006). Post hoc analysis depicted COMO1.5ml +Rhizobium to have the highest yields.

Table 2: Univariate ANOVA of treatments on yield in kg ha

Dependent Variable: yield kg ha

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Block	11515.236	1	11515.236	1.323	.264
Treat	226632.867	6	37772.144	4.339	.006
Error	174088.265	20	8704.413		
Total	412236.367	27			

d) Effect of treatments on weight of seeds whole plot.

The weight of seeds from the whole plot were harvested for each treatment per plot, dried and winnowed. The average weight for each treatment was taken and the highest effect was observed from COMO1ml+Rhizob, it gave the heaviest weight. There was also a significant difference, (p=0.005). The post hoc indicated COMO1ml+Rhizob to be the highest compared to other treatments.

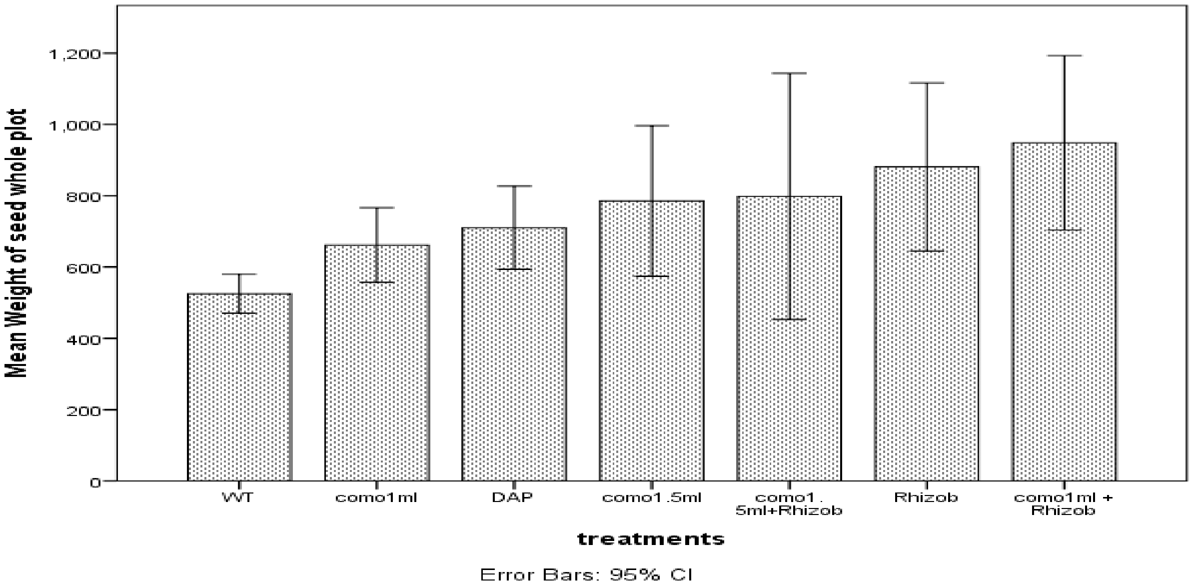

Figure 3: Mean weight of seed whole plot.

Table 3. Univariate ANOVA of treatment on Weight of seed whole plot

Dependent Variable: Weight of seed whole plot

Source	Type III Sum of Squares	Df	Mean Square	F	Sig.
Block	10841.600	1	10841.600	.615	.442
Treat	477272.929	6	79545.488	4.515	.005
Error	352323.900	20	17616.195		
Total	840438.429	27			

a. R Squared = .581 (Adjusted R Squared = .434)

Discussion

Preliminary soil analysis showed that soil in Kisii is moderately acidic and low in nitrogen and phosphorus that is necessary for nitrogen fixation. From the study results, the effect of the treatments used showed significant increase in yield of soya beans. The results are similar with from other research studies done worldwide. For instance, a study conducted in Ethiopia, showed the use of co inoculation with rhizobium to exhibit significant increase from 7-19% in seeds per pod (Abera et al., 2019). Increase in 100 seed weight was shown in an experiment using co inoculants with *A. brasilense* (Ferdous et al., 2019). Signifying that use of inoculant is a viable option to increasing yield in soya beans production in Kenya. Use *Bradyrhizobium* sp. and A. *brasilense* together with cobalt and molybdenum is valuable for sustenance and soybean yields. Increase in yield was also seen in a study done by Filho et al., 2017, it showed significant increase in mean yield of 109 sacks of 60 kg, the yield was approximately 17 sacks of grains more than the control.

Another study done to evaluate the efficacy of nitrogen-fixing bacteria inoculation and nitrogen fertilization, soybean yield was noted to increase. They results depicted 10% (237 kg ha^{-1}) and 15% (336 kg ha^{-1}) higher grain yields , compared to other inoculant without treatment (Carlos Felipe Cordeiro & Echer, 2019). Increase in yield was also observed in a large-scale study in Brazil, inoculation using Bradyrhizobium *japonicum* and diazoefficiens. However, the yields were not significantly different from other treatments (Araujo et al., 2017). In Ghana, use of inoculants showed similar results with increased yield (1262 kg/ha) compared output from uninoculated treatments (1044 kg/ha) (Lamptey et al., 2014). In a study conducted in Zimbabwe, rhizobium inoculant showed increase in pods dry weight. The plants coated with the bacterial *B. japonicum*, and applied alongside 0 and 50 kg N ha-1, exhibited higher yield (Ntambo et al., 2019). Further, an experiment carried out at Nyankpala, manga and Kpongu study site showed a significant effect on yield, treatment used were Teprosyn Mo+Legumefix, Legumefix and Teprosyn Mo, it increased yield by 205.62%, 135.54% and 110.24%in Nyankpala, respectively. It was also found that co inoculation of Teprosyn Mo+Legumefix was the most economically viable option among the treatments where (VCR = 2.65)(Rechiatu, 2015).

The observed positive effect of treatment on yield parameters in this study indicate use of improved quality varieties and inoculation could result to increased yield in Kenya. It can then be concluded that, soya bean grain yield is highly dependent on the effect inoculants on yield parameters such as; six plants pod weight,100 seed weight, yield per hectare among others. Commitment from the government in the achievement of sustainable agricultural systems and realization of the sustainable development goals is important. Access to high quality soya beans products and utilization of bacterial inoculants by local farmers is anticipated to potentially increase in future. (Carlos Felipe Cordeiro & Echer, 2019).

Conclusion and Recommendation. .

Low soil fertility in SSA is characterized by low available phosphorous (P), nitrogen (N), organic matter and soil acidity among other abiotic and social economic factors (Mbebe et al., 2019). The findings from this study indicate that soil in Kisii is moderately acidic and has low nitrogen and phosphorus levels, important in nitrogen fixation in plants. All yield parameters; six plant pod weight, 100 seed weight, weight of seeds whole plot and yield in Kg per ha was noted to increase with application of co-inoculation. COMO and Rhizobium inoculants at either 1ml COMO or 1.5mls COMO was shown have the highest effect compared to other treatments.

Inoculation is effective in enhancement of rhizobia, which subsequently aids in crop growth and soil fertility. Interaction of cobalt, molybdenum and rhizobium has shown significant effect on several yield parameters of soya beans. Combination of inoculants prior to planting of soya beans in Kenya is an effective method to increase yield of soya beans. Further research is recommended to evaluate the effect of soil pH on soya been production in Kisii County.

Additionally, research to investigate effects of Nitrogen and phosphorus levels on soya bean production in Kisii is highly recommended

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