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Interaction of Rhizobium Inoculation, Phosphorus Application and Planting Density Affects Green Gram Yield and Economic Benefit

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

There exists limited green gram production in Kirinyaga County. Increasing green gram production and yield in this area can make it a substitute of beans, cowpeas and maize. Farmers in this area have limited information on use of rhizobia and phosphorus and using the right spacing hence resulting to low yields. This study aimed at evaluating the effect of rhizobia inoculation, phosphorus application and planting density on yield and net economic analysis of green gram production. The study was conducted at Kenya Agricultural and Livestock Research Organization (KALRO), Industrial Crops Research Centre – Mwea where the experiment was laid out as a 3 x 3 x 3 factorial arrangement using Randomised Complete Block Design and replicated three times and in two consecutive cultivations. Treatments included three levels of phosphorus (0 kg/ha, 60 kg/ha and 90 kg/ha), three planting densities (40 x 10 cm, 45 x 15 cm, and 50 x 20 cm) and three levels

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of rhizobium inoculation (0 g, 20 g and 40 g per kg of seeds) in K26 variety of green gram. Data was collected on number of pods and grains per pod, grain yield and net economic benefit. Data collected was subjected to analysis of variance using Statistical Analysis Software (SAS) and significantly different means separated using the Turkeys test at α = 0.05. The findings indicated that rhizobia and phosphorus application and plant density had a significant effect on the grain yield and net economic benefit. There was significant ($p < 0.05$) interaction effect for rhizobia inoculation, phosphorus application and planting density on the grain yield per green gram plant. The yield increased from 98.81 kg and 212.29 kg; 64.15 kg and 118.52 kg in trial I and II, respectively. The analysis of integrated treatment effect of rhizobia rates, phosphorus application and densities showed that the net economic benefit ranged from KSh -21205 to KSh 24512 .5 and KSh -600 to Ksh -24600 for trial I and II, respectively. Treatment with the highest net economic benefit was rhizobia 40 g, phosphorus 20 kg/Ha and 45 x 15 cm. Application of rhizobia rate 40g/Ha, phosphorus 60 kg/Ha and at planting density of 45 × 15 cm produced the highest grain yield. The study recommends application of rhizobia at 40 g/Ha, phosphorus at 60 kg/Ha and planting density at 45 x 15 cm to produce the highest grain yield and rhizobia 40 g, phosphorus 20 kg/Ha and 45 x 15 cm for highest net economic benefit.

Keywords: Green gram; net economic benefit; planting density; phosphorus; rhizobia; yield.

1. INTRODUCTION

The availability of sufficient and nutritionally balanced food is of highest priority while planning to provide the basic needs or improving living standards of small-scale farmers (Manono, 2025). In developed countries pulses account for about an eighth of global pulse area but for more than a fifth of global production, because yields in developed countries are almost twice those in developing countries (Bhat et al., 2022). The bulk of the production is for local consumption, although, in the last two to three decades, large quantities have been grown for export to developing countries (Kinnunen et al., 2020). Recently in most parts of the world, pulses are being used as a major food component due to their nutritional value and health benefits especially to vegetarians. Some of pulses grown include soybean, chick pea, common beans, pigeon peas and green grams (Singh et al*.,* 2025).

Green gram (*Vigna radiata L*.) is important crop in the cropping pattern of low-income countries where the bulk of the production comes from small farms (Wambua, 2021). Green gram generate income for farmers and contribute to soil fertility owing to their nitrogen-fixing ability, thus reducing usage of chemical fertilizers for subsequent crops (Xing et al*.,* 2025). Green grams are an important accompaniment in the diet along with staples like rice and wheat and they are of particular importance for nutrition security, particularly for low-income consumers whose major sources of protein are vegetable crops (Von Bargen, 2025). They can potentially help improve health and nutrition, reduce poverty and hunger and enhance ecosystem resilience (Joshi and Rao, 2016). Climate change, rising cost of inputs, growing demand for food and scarcity of land and water form some challenges facing efforts to ensure food security (El Bilali et al., 2020). Green gram is a short duration crop with low input requirements, a high global demand and an ideal rotation crop for smallholder farmers (Stagnari et al., 2017). Green grams form a cheaper source of protein, which can be used to overcome malnutrition as they have nearly three times as high protein content as compared to cereals (Saxena, 2018). In Kenya, green grams are not only the principal source of protein but also a major source of smallholder cash income (Wambua et al., 2019).

There are large prospects of green gram production in Kenya. However, these prospects have not been realized due to some limiting factors in production such as agronomic practices, unreliable rainfall, and lack of high yielding disease tolerant varieties, price fluctuations and lack of institutional support (Begna, 2020). The leading areas in green gram production in Kenya are Kitui, Makueni, Tharaka Nithi, Machakos and Mbeere. These regions have erratic rainfall patterns and continuous cropping has been draining the critical soil nutrients and hence climate smart agricultural interventions are needed to improve farm level productivity. Green grams require large amounts of phosphorus, potassium, magnesium, calcium and sulphur for seed development (Pati and Pattanayak, 2016). Phosphorus is an essential nutrient for animals and plants and its quantities in soil are generally small which limits plant growth and can result in a decreased crop yield. Phosphorus application significantly improves dry matter yield, grain production, and number of branches (Feng et al*.,* 2021) as it is crucial in formation of proteins, DNA, and RNA, which are vital for producing crops with higher nutritional value, including protein content and mineral density (Khan et al*.,* 2023). Among the essential plant nutrients phosphorous is the most important for seed production, due to its role in cell division and flowering. Many plant crops need more phosphorus than is dissolved in the soil to grow optimally. In addition, crops are usually harvested and removed, leaving no decaying vegetation to replace phosphorus. Therefore, farmers replenish the phosphorus 'pool' by adding phosphate fertilizers.

Although synthetic fertilizers are an alternative for improving green gram production they are expensive and most farmers cannot afford them. Therefore, a cheaper alternative such as the use of climate smart innovations like efficient biological nitrogen fixation, cropping density, and appropriate nutrient sources are necessary. A common agronomic practice to increase pulse production is use of rhizobium inoculation. Use of rhizobia inoculants has been recommended as a sustainable practice of improving adaptability to environmental stress (Sindhu et al., 2020). Improvement in grain yield due to inoculation with rhizobia has been observed in crops such as soya bean (Ntambo et al., 2017). Also use of Optimum planting density minimize competition of the nutrients between crops. Green gram is an important legume crop in Kenya due to its short growth cycle, adaptability to marginal environments, nutritional value, and role in improving household income and soil fertility. In semi-arid regions such as Kirinyaga County, with average on-farm yields reported to be less than 1 tonne per hectare, green gram has the potential (2-3 tonnes per hectare) to serve as a viable alternative to other staples such as beans, cowpeas, and maize, particularly under conditions of limited rainfall. Despite this potential, its productivity in the county remains low compared to the attainable yields under optimal management. Several factors contribute to this yield gap. Soils in Kirinyaga County are characterized by nutrient deficiencies, particularly phosphorus, which is critical for root development and biological nitrogen fixation. In addition, farmers often apply phosphorus fertilizers either inadequately or excessively, leading to suboptimal yields or ecological damage. Similarly, rhizobia inoculation, which enhances nitrogen fixation and crop productivity, is rarely adopted by farmers despite its proven benefits. The economic benefit of Rhizobia inoculation can be enhanced by providing adequate phosphorus, which is essential for the nitrogen fixation process. The use of Rhizobia promotes sustainable farming practices by reducing the environmental impact of synthetic fertilizers and improving soil fertility for future crops.

An important aspect of crop arrangement is spacing and its relationship to yield as the plant density greatly determines competition and utilization of growth resources such as light, nutrients and water. Adequate spacing between plants will ensure that each plant receives and efficiently utilizes the growth resources yet inappropriate spacing remains a common practice. The net economic benefit of planting density is achieved by finding an optimal balance between increasing crop yield and controlling production costs, as too high a density can lead to reduced individual plant yields and increased input requirements (seeds, water, labor), which lowers profitability (Hu et al*.,* 2025). These challenges point to a critical need for integrated soil and crop management practices tailored to local conditions. It is important to determine the planting density that maximizes profit, not just yield, for a specific crop and location. Also, there is need to coordinate planting density with other management practices, such as phosphorus application rates, to enhance green gram yield and alleviate negative impacts of high density. However, there is limited research on the combined effects of rhizobia inoculation, phosphorus application, and planting density on the growth, yield, and economic viability of green gram production in Kirinyaga County. Addressing this gap is essential for improving productivity, promoting sustainable land use, and enhancing food and nutrition security. This study sought to determine the effect of different levels of phosphorus, plant population and rhizobium inoculums on yield and economic benefit of green gram production.

2. MATERIALS AND METHODS

2.1 Study Site

The study was carried out in two consecutive cultivations (Trial I and II) at Kenya Agricultural and Livestock Research Organization (KALRO), Industrial Crops Research Centre – Mwea in

Mwea East Sub- County, Kirinyaga County between January and May 2022. The Centre is 21 km South West of Embu town and about 112 km North East of Nairobi (0°39′ S latitude, 37°22′ E longitude), at an elevation of 1162 m above sea level. The average rainfall is about 850 mm with a range of 500 - 1250 mm divided into long rains from March – June with an average of 450 mm and short rains from mid-October to December with an average of 350 mm. The temperature ranges from 15.6 ºC to 28.6 ºC with a mean of about 22 ºC (Kavu et al., 2019). The centre has 16.8 hectares under research. The soils characteristics are; well drained dusky red to dark reddish brown, nitosols and friable clay. This soil has low fertility (Bii & Bii, 2018). Along Mwea irrigation scheme (72%) are rice farmers, 15% grow horticultural crops, 7% engaged in trading activities while 2% do livestock production, are wage earners or do boda boda business (Mwai et al., 2023).

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2.2 Experimental Design, Treatments and Field Layout

The study used a Randomized Complete Block Design with three replications, which include three levels of phosphorus (0 kg/Ha, 60 kg/Ha and 90 kg/Ha), three planting densities (40 x 10 cm [250000 plants per hectare], 45 x 15 cm [148148 plants per hectare], 50 x 20 cm [100000 plants per hectare] and three levels of rhizobium inoculation (0 g, 20 g and 40 g per kg of seeds). The recommended rate of phosphorus application for K26 is 60 kg / Ha which translates to 26.4 kg of P / Ha. The three rates of phosphorus were provided as triple super phosphate (TSP) applied at planting translated into 0, 26.4 and 52.8 kg of P/Ha for 0 kg/Ha, 60 kg/Ha and 90 kg/Ha, respectively. The TSP was uniformly distributed on planting holes and thoroughly mixed with soil to avoid direct contact with the green gram seeds. Each block of the three replicates measured 50 m × 2.5 m. Paths between plots were 0.5 m. Each plot had 5 rows each with 5 green gram plants, giving a total of 25 plants per plot. The first and the last rows including the first and last green gram plant per row formed the guard rows. Data was taken on the 3 middle plants.

2.3 Land Preparation, Planting Materials, Planting and Crop Maintenance in the Field

A piece of land was cleared using a panga and primary land preparation was done by digging using a forked jembe to a depth of 30 cm. Secondary cultivation was done to obtain a fine tilth. K26 seeds was prepared by coating with inoculants using 4% gum Arabica supplied with the inoculums at rates of 0, 20 and 40 g/kg of seeds. Green gram seeds were inoculated with rhizobium inoculums three hours before sowing. Rhizobium inoculated seeds were planted when there was sufficient moisture in the soil. TSP at 0, 60 and 90 kg / Ha was applied at planting. Two seeds were planted per hole and at a planting density of 40 x 10 cm, 45 x 15 cm, and 50 x 20 cm. Certified seeds of K26 was procured from Kenya Seed Company. The rhizobium inoculum was procured from MEA Company Limited, Nakuru. Thinning was done 14 days after planting leaving one seedling per hole. Weeding was done as weeds emerged and pests and diseases controlled as they emerged. Harvesting of green gram was done when most of the pods have turned black. Dry individual pods or the whole plant were picked and dried for 2 days before threshing and winnowing was done to remove chaff and dirt.

2.4 Data Collection

2.4.1 Green gram yield and yield components

Green gram variety K26 took between 60 - 65 days to mature. Pods from three selected plants per treatment that had started turning brown were harvested and dried in Khaki sampling bags to prevent shattering of pods that would result in decline in the average yield of the green grams. Different Khaki bags were used to store the pods from plants with different treatments.

2.4.1.1 Pod number per plant

This was done at physiological maturity. Three plants from two inner rows in each experimental plot were selected and number of pods per plant was counted and recorded.

2.4.1.2 Length of the pod

Three pods from the shoot tip were selected from 3 plants per treatment and length record between the two tips of the pod.

2.4.1.3 Number of seeds per pod

Number of seed in each pod was recorded from 3 plants selected per treatment and converted to kg/Hectare.

Table 1. Cost of green gram production per hectare

Variables	No. of units	Unit Cost (Kshs)	Total Cost (Kshs)
Ploughing	1 ha	7500	7500
Planting (1 ha)	8 man-days	400	3200
1st Weeding (1 ha x2)	8 man-days	400	3200
Harvesting (1 Ha)	6 man-days	400	2400
Threshing/packaging	3 man-days	400	1200
Inputs			
TSP	2 bags	4500	9,000
Duduthrin pesticide	1 Litre	1500	1.500
Seeds	8 pcs 2kg	500	4,000
Rhizobia	40g	200	200
Total			32,200

2.4.1.4 Grain yield

All the grains from pods in selected plants per treatment were counted per pod and then combined for each treatment. The pods were further air dried then threshed manually and winnowed to separate the seeds from the debris. The grains were air dried under the sun to for three days on a mat to reduce moisture content and dry weights determined using electronic balance. Then grains were put in a khaki bag per treatment. This was converted to yield in kg per hectare. Grain yield in kg per ha= 10000 m^2 x yield (kg)/Harvest area (m^2).

2.4.2 Net economic benefit/ economic analysis

Economic analysis (net economic benefit) of the green gram production enterprise was performed after harvest. It was calculated by deducting the (variable cost) gross production cost from (revenue) gross green grams output. The gross green gram output was determined by multiplying the weight of harvested green grams by the prevailing market price. The gross benefit is the gross income derived from sale of the grain. The gross production cost included cost of; phosphate fertilizer, rhizobia, seeds and labour cost in man days which included cultivation, planting, weeding, crop protection, harvesting. The net benefit per plot was translated to net economic benefit per hectare (Bhatia et al, 2018) [Table 1].

2.5 Data Analysis

Data collected was subjected to analysis of variance (ANOVA) to determine the effects of the treatments applied. Data was analysed using Statistical Analysis Software (SAS) version 9.3. Least Significant Difference (LSD) test was used for means comparison at probability level of 5% (Mertler & Reinhart, 2016).

3. RESULTS AND DISCUSSION

3.1 Soil Analysis

Soil sampling was done at the experimental site before planting and the sample was analysed for soil chemical properties at Crop Nuts Laboratory, Nairobi to determine the initial soil nutrient status. The parameters assessed included soil reaction (pH), total nitrogen, organic carbon, available phosphorus, and selected exchangeable cations (K, Mg, Ca) [Table 2]. The soil analysis revealed that the pH was 5.64 which was within the optimum range for green gram production, suggesting no major limitation due to soil acidity. However, the total nitrogen content (0.14%) was below the critical threshold, indicating a deficiency that could constrain crop growth if not supplemented. Similarly, the organic carbon level (0.14%) was also lower than the recommended range, reflecting poor organic matter content and reduced soil fertility status. Exchangeable potassium (389 cmol/kg) was above the critical level and therefore adequate to support crop growth, while magnesium (371 cmol/kg) exceeded the recommended range, raising the possibility of nutrient imbalance that could hinder calcium uptake. In contrast, exchangeable calcium (1030 cmol/kg) was below the critical range, suggesting a deficiency that may negatively affect cell wall development and nodulation. Available phosphorus (8.6 ppm) was markedly lower than the optimum range of 40–100 ppm, signifying severe phosphorus deficiency likely to limit root development and symbiotic nitrogen fixation. Overall, the soils at the trial site were characterized by low levels of nitrogen, organic carbon, calcium, and phosphorus, all of which are essential for green gram growth and biological nitrogen fixation. These deficiencies justified the need to evaluate the effects of rhizobia inoculation, phosphorus application, and planting density on the performance of green gram in the study area.

The soil analysis revealed a pH of 5.64, indicating slightly acidic conditions. Green gram thrives best in soils with a pH range of 6.0–7.5, where nutrient availability and microbial activity are optimized (Basha & Prasada Rao, 2017). Although a pH of 5.64 is not severely acidic, it falls below the optimum range and may impose constraints on crop performance. Soil acidity influences nutrient dynamics by increasing the solubility of toxic elements such as aluminum and manganese while simultaneously reducing the availability of essential nutrients such as phosphorus, calcium, and magnesium (Rahman et al., 2018). Thus, even moderate acidity can compromise crop growth by limiting root development and nutrient uptake efficiency. One of the major consequences of sub-optimal pH is its effect on phosphorus availability. At pH levels below 6.0, phosphorus tends to be fixed by iron and aluminum oxides, rendering it unavailable to plants (Muindi, 2019). This aligns with the observed low phosphorus content (8.6 ppm) in the trial soils, which is well below the critical range of 40–100 ppm. Since phosphorus is essential for energy transfer, root development, and nodulation in legumes, its deficiency under slightly acidic conditions is likely to constrain both early crop establishment and biological nitrogen fixation. Previous studies have shown that phosphorus deficiency not only reduces vegetative growth but also limits the symbiotic efficiency of rhizobia in legumes such as cowpea and green gram (Dikr & Abayechaw, 2022).

In addition to phosphorus limitation, slightly acidic soils can influence rhizobia survival and effectiveness. Rhizobia are highly sensitive to soil pH, with sub-optimal conditions reducing their proliferation, root colonization, and nodule formation (Dash, 2016). This, in turn, lowers nitrogen fixation efficiency, which is a key driver of legume productivity. Therefore, the combination of low soil pH, low phosphorus, and imbalanced nutrient availability at the trial site highlights the importance of integrated soil fertility management. Corrective measures such as phosphorus supplementation, rhizobia inoculation, and, where feasible, liming to raise soil pH could improve the soil environment for green gram production. This justifies the present study's focus on evaluating the combined effects of rhizobia inoculation, phosphorus application, and planting density on the growth and yield of green gram in Kirinyaga County. The soil pH of 5.64 indicated slightly acidic conditions. Green gram grows optimally in soils with a pH of 6.0–7.5 (Basha & Prasada Rao, 2017). Although the pH observed was not extremely low, it is below the optimum, and acidity at this level can reduce nutrient availability, especially phosphorus, and may also impair rhizobia activity (Dash, 2016). Consequently, sub-optimal pH is likely to contribute to poor nodulation and nitrogen fixation, both of which are vital for legume productivity.

The total nitrogen content of the soil (0.14%) was below the critical range of 0.20–0.50%, indicating nitrogen deficiency. Nitrogen is essential for protein synthesis, chlorophyll formation, and vegetative growth in legumes (Leghari et al., 2016). In legumes such as green gram, nitrogen deficiency limits photosynthetic capacity and reduces biomass accumulation. Although legumes can fix atmospheric nitrogen through symbiosis with rhizobia, low soil nitrogen often reduces initial seedling growth before nodulation is established (Schwember et al*.,* 2019). Therefore, the low N content in the soil underscores the importance of effective rhizobia inoculation and phosphorus application to enhance biological nitrogen fixation and crop performance. The organic carbon content (0.14%) was also below the critical range of 0.20–0.50%, reflecting poor soil organic matter. Organic matter is crucial for soil fertility as it improves soil structure, water retention, and nutrient supply through mineralization (Gerke, 2022). Low organic carbon is associated with reduced microbial activity and limited nutrient cycling, which further constrains the establishment of rhizobia and nodulation efficiency. Studies in African soils have shown that low organic matter often correlates with poor crop yields and low fertilizer-use efficiency (Zingore et al*.,* 2021). Thus, the deficiency in organic carbon at the study site highlights the need for integrated fertility management, such as organic amendments combined with rhizobia inoculation and phosphorus fertilization.

Table 2. Soil analysis from the trial site before planting

Properties	Value	Range	Class
Ph	5.64	5.4-7	Optimum
Nitrogen (N) %	0.14	0.2-0.5	Low
Organic Carbon (OC) %	0.14	0.2-0.5	Optimum
Potassium (K) (cmol/ kg)	389	>140	Optimum
Magnesium (Mg) (cmol/kg)	371	164-263	High
Calcium (Ca) (cmol/kg)	1030	1640-2060	Low
Phosphorus (P) (ppm)	8.6	40-100	Low

Exchangeable potassium (389 cmol/kg) was well above the critical level (>140 cmol/kg), indicating that K was sufficient for crop growth. Potassium plays a vital role in osmoregulation, enzyme activation, and translocation of assimilates, and adequate levels are particularly beneficial under water-limited conditions (Bhattacharya, 2021). Since the study site already had adequate potassium, K deficiency was unlikely to limit green gram production. However, excess potassium can sometimes induce nutrient imbalances, particularly affecting magnesium and calcium uptake (Sardans & Peñuelas, 2021). The high Mg and low Ca values observed suggest that nutrient interactions may need to be considered. Magnesium concentration (371 cmol/kg) was higher than the recommended range (164–263 cmol/kg), indicating excess Mg in the soil. While Mg is important for chlorophyll formation and enzyme activation, high levels may cause nutrient antagonism, especially with calcium and potassium (Ahmed et al*.,* 2023). This imbalance could interfere with root function and nutrient uptake, thereby affecting plant growth. In legumes, adequate Mg is essential for photosynthesis, but excessive levels may not directly enhance yield and could instead reduce calcium availability, as observed at the study site.

Exchangeable calcium (1030 cmol/kg) was below the critical range of 1640–2060 cmol/kg, indicating a deficiency. Calcium is vital for cell wall structure, root elongation, and nodule formation in legumes (Zartdinova and Nikitin, 2023). Inadequate Ca can reduce nodulation efficiency and root development, thereby limiting the crop’s ability to exploit soil resources. The deficiency in Ca, coupled with excess Mg, suggests a cation imbalance that may affect nutrient uptake and overall plant vigor. This makes calcium limitation an important factor influencing green gram performance in the study site. The available phosphorus content (8.6 ppm) was critically low compared to the optimum range of 40–100 ppm. Phosphorus is essential for root growth, energy transfer (ATP), and symbiotic nitrogen fixation in legumes (Ma & Chen, 2021). Deficiency results in stunted growth, delayed maturity, and reduced nodulation efficiency (Singh et al., 2011). The low P levels in the soil are consistent with the slightly acidic pH, which promotes phosphorus fixation by iron and aluminum oxides, reducing its availability to plants. Therefore, phosphorus deficiency was likely a key limiting factor for green gram production at the trial site, justifying the evaluation of phosphorus application in this study.

The soil at the experimental site was characterized by slightly acidic pH, low nitrogen, low organic carbon, low calcium, and critically low phosphorus, while potassium was adequate and magnesium was in excess. This nutrient profile suggests that green gram production was limited mainly by deficiencies in N, P, Ca, and organic matter, coupled with potential nutrient imbalances due to high Mg. Since green gram relies heavily on symbiotic nitrogen fixation, the low soil fertility and sub-optimal pH highlight the importance of integrated interventions such as rhizobia inoculation, phosphorus supplementation, and proper planting density to enhance growth, yield, and economic returns.

3.2 Effect of Inoculation with Rhizobia, Rate of Phosphorus Application and Planting Densities on Green Gram Yield

3.2.1 Effects on number of pods, length of pods and number of seeds

There was significant ($p < 0.05$) difference in number of pods per plant trial I and II (Table 3). Number of pods was significantly affected by rhizobia application, density, phosphorus application, rhizobia and density, density and phosphorus, rhizobia and phosphorus and combined application of rhizobia, density and phosphorus ($p < 0.05$) for trial I. For trial II, rhizobia application and density significantly affected number of pods ($p < 0.05$). Number of pods per plant was recorded as 16.56 to 22.89 and 17.11 to 21.11 in trial I and II, Rhizobia 40 g/Ha, 60 kg/Ha phosphorus, 50 x 20 cm and Rhizobia 20 g/Ha, 0 kg/Ha phosphorus and 50 x 20 cm produced highest number of pods (Table 3). Length of pods was significantly affected by rhizobia application, phosphorus application, density, rhizobia and density, density and phosphorus, rhizobia and phosphorus and combined application of rhizobia, density and phosphorus ($p < 0.05$) for trial I. For trial II, rhizobia application, phosphorus application, rhizobia and density, density and phosphorus and combined application of rhizobia, density and phosphorus significantly affected length of pods ($p < 0.05$). Length of pods per plant was recorded as 8.36 to 11.53 and 8.64 to 10.63 in trial I and II, rhizobia 20 g/Ha, 60 kg/Ha phosphorus, 50 x 20 cm and Rhizobia 20 g/Ha, 0 kg/Ha phosphorus and 40 x 10 cm produced highest length of pods (Table 3). Number of seeds was significantly affected by rhizobia application, phosphorus application, density, rhizobia and density, density and phosphorus, rhizobia and phosphorus and combined application of rhizobia, density and phosphorus ($p < 0.05$) for trial I. For trial II rhizobia application and density significantly, number of seeds ($p < 0.05$). Number of seeds per pod was recorded as 10.22 to 13.33 and 10.56 to 12.67 in trial I and II, Rhizobia 0 g/Ha, 60 kg/Ha phosphorus, 45 x 15 cm and Rhizobia 0 g/Ha, 60 kg/Ha phosphorus and 40 x 10 cm produced highest number of seeds per pod (Table 3).

This study finding showed that high phosphorus had a significant positive effect on increasing the number of pods per plant, the length of pods and the number of seeds per pod in many crops leading to increased overall yield. Moderate spacing generally increased the number of pods and seeds per plant. The observation that 0 g rhizobia, 60 kg/Ha phosphorus and a planting density of 45 x 15 cm in trial I and II which resulted in the longest pods can be explained by a combination of physiological and agronomic factors; in aabsence of rhizobia (0 g), the plant relied more on soil nutrients, especially phosphorus, for growth. Dikr and Abayechaw (2022) findings were that it led to fewer but larger pods, as the plant allocated resources to fewer reproductive structures. Phosphorus at 60 kg/Ha enhanced flowering, pod formation, and grain development. Liu (2021) found that in the absence of rhizobia, phosphorus becomes even more critical for energy transfer and root development. Meena et al*.* (2021) findings state that increasing phosphorus caused pod elongation.

Planting density at a spacing of 45 × 15 cm allowed for moderate plant population. Fajriani et al*.* (2025) reported that plants with enough space to grow without excessive competition, more light and air circulation led to better pod development and longer pods. Without rhizobia, nitrogen was limited. Kubure et al*.* (2016) findings were the plant produced fewer pods but each pod received more nutrients, resulting in longer pods. Also, the observation that 0 g rhizobia, 0 kg/ha phosphorus and a planting density of 50 x 20 cm in trial II resulted in the longest pods. Several biological and environmental factors could explain this outcome such as low competition at 50 × 20 cm spacing which provided ample room for each plant. With fewer plants competing for light, water and nutrients, each plant allocated more resources to pod development. According to Manjesh et al*.* (2019) this could have led to fewer but longer pods as the plant invested more in each reproductive structure.

There was stress-induced elongation, in the absence of rhizobia and phosphorus, where the plant experienced nutrient stress. Green gram plants responded to stress by producing fewer but longer pods as a survival mechanism. Júnior et al*.* (2018) findings were that this was a form of compensatory growth whereby plants had fewer pods but each one was longer to maximize grain output. The observation that 0 g rhizobia, 60 kg/ha phosphorus and a planting density of 45 cm × 15 cm in resulted in the highest number of grains per plant was explained by a combination of nutrient availability, plant spacing and physiological responses. High phosphorus (60 kg/ha) played a key role in flower and grain formation, energy transfer (ATP) during reproductive stages and root development. Xie et al*.* (2022) reported root growth enhanced nutrient uptake. Even without rhizobia, the high phosphorus level likely compensated for nitrogen deficiency to some extent. Moderate planting density (45 × 15 cm) allowed higher plant population than wider spacing (50×20 cm), efficient use of space without severe overcrowding and moderate competition, which stimulated reproductive effort. This balance according to findings of Pant and Sah

(2020) had encouraged plants to produce more pods and grains to compete for survival and allowed enough light and air for healthy development. Without rhizobia, nitrogen fixation is limited. However, according to findings of Wendlandt et al*.* (2022), if the soil had residual nitrogen or native rhizobia, the plants performed well. Bhattacharya (2021) findings were that the plant had prioritized grain production over vegetative growth, leading to more grain per pod or more pods per plant.

The observation that 20 g rhizobia, 0 kg/Ha phosphorus and a planting density of 40 x 10 cm resulted in the highest number of pods per plant could be explained by the following agronomic and physiological factors, rhizobia bacteria formed symbiotic relationships with legume roots fixing atmospheric nitrogen. In this study, with 20 g of rhizobia, nitrogen availability was enhanced, According to Mejri et al*.* (2018), even without added phosphorus the nitrogen boosted from rhizobia significantly improved reproductive output.

Table 3. Effect of Rhizobia, phosphorus application and planting density on green gram number of pods, length of pods and number of seeds per pod during trial i and ii

Number of Pods, Length of Pods and Number of Seeds per Pod						
	Trial I			**Trial I**		
Treatment combinations	**Nnumber of Pods**	**Length of Pod**	**Number of Seeds**	**Number of Pods**	**Length of Pods**	**Number of Seeds**
R0P0D1	20.22^{bcdef*}	9.43^{fghi}	11.11^{fghij}	17.11^{f}	10.63^{a}	10.78^{ef}
R0P0D2	19.56^{cdefgh}	10.63^{abcd}	11.78^{cdefgh}	19.78^{abcde}	10.54^{ab}	12^{abcd}
R0P0D3	18.56^{fghij}	10.43^{bcde}	11.56^{cdefgh}	20.89^{ab}	10.52^{ab}	12.67^{a}
R0P1D1	18.89^{fghij}	8.9^{hij}	10.56^{hij}	18.89^{abcdef}	10.31^{ab}	11.44^{abcdef}
R0P1D2	19.56^{cdefgh}	9.11^{ghij}	13.33^{a}	17.89^{def}	10.18^{abc}	11.11^{cdef}
R0P1D3	19^{efghij}	9.81^{defgh}	10.33^{ij}	19.44^{abcdef}	10.13^{abcd}	11.78^{abcde}
R0P2D1	21.44^{abc}	9.33^{ghij}	12.11^{bcdef}	18^{def}	10.09^{abcde}	10.89^{def}
R0P2D2	18.67^{fghij}	8.91^{hij}	11.33^{efghij}	18.56^{dcdef}	10.08^{abcde}	11.33^{bcdef}
R0P2D3	20.33^{bcdef}	9.61^{efghi}	11.33^{efghij}	18.78^{abcdef}	10.03^{abcde}	11.33^{bcdef}
R1P0D1	19.22^{defghij}	8.36^{j}	12.33^{abcde}	17.44^{f}	9.98^{abcde}	10.56^{f}
R1P0D2	19.89^{bcdefg}	10.27^{bcdef}	11.46^{defgh}	20^{abcd}	9.93^{abcde}	12.33^{ab}
R1P0D3	21^{abcd}	9.51^{efghi}	10.78^{ghij}	21.11^{a}	9.92^{abcde}	12.44^{ab}
R1P1D1	18^{fghij}	11.53^{a}	12^{bcdef}	18^{def}	9.89^{abcde}	10.78^{ef}
R1P1D2	19.78^{cdefg}	9.66^{efghi}	10.22^{j}	18.78^{abcdef}	9.88^{abcde}	11.33^{bcdef}
R1P1D3	16.56^{k}	10.23^{bcdef}	12.67^{abc}	19.67^{abcde}	9.86^{abcde}	11.67^{abcdef}
R1P2D1	19.33^{defghij}	9.93^{defgh}	12^{bcdef}	19.78^{abcde}	9.57^{abcde}	11.56^{abcdef}
R1P2D2	17.56^{hijk}	8.71^{ij}	13^{ab}	18.44^{cdef}	9.44^{bcdef}	11.11^{cdef}
R1P2D3	18.78^{fghij}	9.43^{fghi}	11.11^{fghij}	18.67^{bcdef}	9.43^{bcdef}	11.33^{bcdef}
R2P0D1	17.56^{ijk}	10.09^{cdefg}	11.56^{cdefgh}	19.67^{abcde}	9.42^{bcdef}	11.89^{abcde}
R2P0D2	17.67^{ghijk}	9.43^{fghi}	11^{fghij}	19.67^{abcde}	9.4^{bcdef}	11.67^{abcdef}
R2P0D3	20.89^{bcde}	9.81^{defgh}	12.56^{abcd}	19.89^{abcd}	9.32^{cdef}	12^{abcd}
R2P1D1	22.89^{a}	10.22^{bcdef}	11.33^{defghi}	19.44^{abcdef}	9.11^{def}	11.78^{abcdef}
R2P1D2	18.78^{fghij}	8.88^{ij}	11.89^{cdefg}	20.11^{abcd}	9.11^{def}	12.33^{abc}
R2P1D3	17.11^{jk}	10.97^{ab}	12^{bcdef}	20.44^{abc}	8.99^{ef}	12.11^{abc}
R2P2D1	19.78^{cdefg}	9.98^{cdefg}	11.22^{efghij}	17.78^{ef}	8.97^{ef}	10.67^{ef}
R2P2D2	21.78^{ab}	9.8^{defgh}	11.67^{cdefgh}	20.78^{abc}	8.78^{f}	12.56^{ab}
R2P2D3	20.33^{bcdef}	10.79^{abc}	11.22^{efghij}	20.11^{abcd}	8.64^{f}	12.22^{abc}
LSD	1.9	0.96	1.15	2.35	1.17	1.28
CV	10.67	10.72	10.76	13.29	13.14	12
R2	0.54	0.54	0.4	0.34	0.34	0.3

**Means followed by same letter along the column for each treatment are not significantly different from each other at 5% probability level. Where: R= Rhizobium levels 0, 20, 40 g/ha; P = Phosphorus level at 0, 60, 90 kg/ha and D= Planting density at 50 x 20, 45 x 15, and 40 x 10 cm; LSD=Least Significant Different; CV=Coefficient of variation;* R^2 *=R-Square*

Table 4. Effect of Rhizobia, phosphorus, and planting density on green gram grain yield during trial i and ii

	Grain Yield	
Treatment combinations	**Trial I**	**Trial II**
R0P0D1	1.3^{bc}	0.77^{ab}
R0P0D2	1.2^{cd}	0.8^{ab}
R0P0D3	1.27^{bcd}	0.57^{cd}
R0P1D1	1.27^{bcd}	0.77^{ab}
R0P1D2	1.3^{bc}	0.77^{ab}
R0P1D3	1.3^{bc}	0.5^{ef}
R0P2D1	1.3^{bc}	0.57^{cd}
R0P2D2	1.17^{d}	0.8^{ab}
R0P2D3	1.27^{bcd}	0.77^{ab}
R1P0D1	0.77^{efg}	0.62^{cd}
R1P0D2	0.77^{efg}	0.53^{def}
R1P0D3	1.27^{bcd}	0.6^{cd}
R1P1D1	1.23^{cd}	0.57^{cd}
R1P1D2	0.83^{ef}	0.63^{bc}
R1P1D3	0.67^{g}	0.63^{bc}
R1P2D1	1.3^{bc}	0.5^{ef}
R1P2D2	1.27^{bcd}	0.43^{f}
R1P2D3	1.23^{cd}	0.8^{ab}
R2P0D1	0.87^{e}	0.63^{bc}
R2P0D2	1.17^{d}	0.8^{ab}
R2P0D3	0.67^{g}	0.53^{def}
R2P1D1	1.43^{a}	0.83^{a}
R2P1D2	1.3^{bc}	0.57^{cd}
R2P1D3	0.8^{ef}	0.63^{bc}
R2P2D1	0.73^{fg}	0.6^{cd}
R2P2D2	1.37^{ab}	0.53^{def}
R2P2D3	0.7^{fg}	0.67^{bc}
LSD	0.11	0.11
CV	10.59	18.5
R^2	0.85	0.65

**Means followed by same letter along the column for each treatment are not significantly different from each other at 5% probability level. Where: R= Rhizobium levels 0, 20, 40 g/ha; P = Phosphorus level at 0, 60, 90 kg/ha and D= Planting density at 50 x 20, 45 x 15, and 40 x 10 cm; LSD=Least Significant Different; CV=Coefficient of variation;* R^2 *=R-Square*

High planting density at 40 x 10 cm resulted in a high plant population. It is possible that even if this increased competition, it stimulated reproductive effort as plants hence they competed for light, space and lead to more pods per plant were produced as a survival strategy. However, according to findings of Williams II (2016) this effect depended on the crop's ability to tolerate crowding. No phosphorus (0 kg/Ha), according to findings of Concha and Doerner (2020) the crop had prioritized pod formation over pod size or grain number and relied on efficient root systems supported by rhizobia-enhanced nitrogen.

3.2.2 Effects on grain yield per plant

Grain yield was significantly affected by rhizobia application, phosphorus application, density, rhizobia and density, density and phosphorus, rhizobia and phosphorus and combined application of rhizobia, density and phosphorus ($p < 0.05$) for trial I. For trail II, grain yield was significantly affected by rhizobia application, density, rhizobia and density, density and phosphorus, rhizobia and phosphorus and combined application of rhizobia, density and phosphorus, the grain weight ranged from 0.67 g and 1.43 g; 0.43 g and 0.8 g in trial I and II, respectively. Rhizobia rate 40 g/Ha, phosphorus 60 kg/Ha and 45 x 15 cm produced the highest grain yield in trial I and trial II (Table 4).

It is possible that during this study the high rates of rhizobia promoted high nitrogen fixation and boosted crop grain yield by promoting growth and photosynthesis. Moderate phosphorus effect on grain yield was positive and significant, as it was essential for energy conversion, photosynthesis, and the development of strong roots and stems, which directly contributed to higher grain yields. Further, reasonable spacing affected grain yield by influencing crop to crop competition for resources like light, water, and nutrients, with optimal spacing leading to higher yields by minimizing competition and maximizing resource use.

The treatment with 40 g rhizobia, 60 kg/Ha phosphorus and 45 x 15 cm spacing resulted in the highest grain yield which was attributed to a combination of biological, chemical and spatial factors that optimized plant growth and productivity. During the study it is possible that high rhizobia inoculation (40 g) fixed atmospheric nitrogen, provided a natural and sustained nitrogen source. Gebremariam and Tesfay (2021) finding indicate that increasing rate of rhizobia inoculum during inoculation was likely cause of optimal nodulation, enhancing vegetative growth, flowering, grain filling and protein synthesis in grains. Malhotra et al*.* (2018) found that adequate phosphorus (60 kg/ha) was essential for: root development, energy transfer (ATP) and grain formation and grain development. 60 kg/Ha was often considered optimal for legumes and cereals, especially when paired with Rhizobia.

There is a possibility that moderate spacing (45 x 15 cm) provided enough room for each plant to access sunlight and nutrients due to just enough plant population and reduced competition, which allowed full expression of genetic yield potential. Plant spacing at 45 x 15 cm produced the highest grain yield with less rhizobia 20 g/Ha and higher phosphorus rate of 90 kg/Ha. Phosphorus played a key role in flower and grain formation, energy transfer (ATP) during reproductive stages and root development, which enhanced nutrient uptake. Even with less rhizobia, the high phosphorus level likely compensated for nitrogen deficiency to some extent and promoted more flowers and higher grain weight in trial II. Zhang et al*.* (2024) found that the appropriate application of fertilizers, such as nitrogen and phosphorus, is vital for optimizing grain production. Kanonge-Mafaune et al*.* (2018) findings were that inoculating legume seeds with rhizobia can lead to significantly increased grain yields per plant. Grain yield is a crucial performance indicator in agriculture, reflecting the success of crop production and influencing economic development by improving farm competitiveness, ensuring food security, and reducing production costs for raw materials (Koptaeva et al*.,* 2019).

3.3 Effect of Rhizobia Inoculation, Phosphorus Application and Planting Density on Net Benefit

There was no significant ($p < 0.05$) interaction between trial I and trial II on the net economic benefit green gram plant (Table 5). With a market price of KSh 150/ kg and cost of production ranged from KSh 23000 to 32000, the analysis of integrated treatment effect of rhizobia rates, phosphorus application and densities showed that the net economic benefit from the ranged from KSh -21205 to KSh 24512 .5 and KSh -600 to Ksh -24600 for trial I and II, respectively. The results showed that the treatment with the highest net economic benefit was rhizobia 40 g, phosphorus 20 kg/Ha and 45 x 15 cm and the lowest net economic benefit was rhizobia 40 g, phosphorus 60 kg/Ha and 50 x 20 cm for trial I and II, respectively (Table 5).

Trial I was done under irrigation but trail II had a mixture of irrigation and rains in the last month of growth. This contributed to abscission of flowers which affected yield for trial II. During the study, there was significant interaction of combined treatments effect on the net economic benefit green gram plant. The findings showed that the treatment with the highest net economic benefit was rhizobia 40 g, phosphorus 60 kg/Ha and 45 x 15 cm and the lowest net economic benefit was rhizobia 40g, phosphorus 60 kg/Ha and 50 x 20 cm for trial I and II, respectively.

Kouam et al*.* (2020) found out that low plant density increased the net returns and benefit cost ratios of common beans. Muchomba et al*.* (2023) found that in Kenya, green gram production is done mainly by smallholder farmers for food and sale. Kihoro et al*.* (2016) find concluded that farmers prefer marketing channels where they incur low production and transport cost and that offer higher prices to maximize profits. Wambua (2021) findings were that various policy concerns were to be adopted for transforming subsistence-oriented production into market-oriented production focusing on green gram and pigeon pea food crops. Mugure et al*.* (2023) findings recommend that increasing green gram yield requires a supportive price.

Table 5. Effect of Rhizobia, phosphorus application and planting density on green gram net economic benefit

	Net Economic Benefit	
Treatment combination	**Trial I**	**Trial II**
R0P0D1	-3500	-11495
R0P0D2	3666.55	-5222.3
R0P0D3	24512.5	-1737.5
R0P1D1	-8495	-15995
R0P1D2	1388.8	-10455.6
R0P1D3	21250	-8750
R0P2D1	-12500	-23495
R0P2D2	-6066.8	-14222.3
R0P2D3	15512.5	-3237.5
R1P0D1	-11595	-13849.5
R1P0D2	-6055.65	-11255.7
R1P0D3	24412.5	-600
R1P1D1	-9105	-19095
R1P1D2	-9088.95	-13533.45
R1P1D3	-2587.5	-3862.5
R1P2D1	-12600	-24600
R1P2D2	-3944.55	-13588.95
R1P2D3	14137.5	-2100
R2P0D1	-10195	--13705
R2P0D2	2733.2	-5422.3
R2P0D3	1812.5	-3212.5
R2P1D1	-5605	-20605
R2P1D2	1788.8	-14500.15
R2P1D3	2900	-3362.5
R2P2D1	-21205	-23200
R2P2D2	-1822.3	-20355.7
R2P2D3	-5950	-7187.5
LSD	3339.5	3137.7
CV	-30.92	-501.51
R-squared	0.84	0.93

**Means followed by same letter along the column for each treatment are not significantly different from each other at 5% probability level. Where: R= Rhizobium levels 0, 20, 40 g/ha; P = Phosphorus level at 0, 60, 90 kg/ha and D= Planting density at 50 x 20, 45 x 15, and 40 x 10 cm; LSD=Least Significant Different; CV=Coefficient of variation;* R^2 *=R-Square*

Based on this study findings net economic benefit being negative in some treatments, production cost needs to be minimized. 54% of cost of total production cost was KSh 7500 ploughing cost, KSh 3200 planting cost, KSh 3200 weeding cost, KSh 2400 harvesting and KSh 1200 threshing and packaging. Cost of rhizobia of KSh 200 lowered production cost that could have been used to buy nitrogen fertilizer. According to Kebede (2021), most grain legumes including can obtain between 50 and 80% of their nitrogen concentration requirements through biological fixation. Some, like fababean will fix up to 90% (Argaw & Mnalku, 2017). The amount of nitrogen fertilizer required for green grams varies but a basal dose of 10-15 kg/Ha of nitrogen may be sufficient for highly eroded soils. Further measures to reduce cost of production are use of herbicide instead of manual weed. Herbicide cost KSh 1500 saving KSh 1700 from weeding cost. To lower cost of cultivation, seeds can be placed directly into the soil with minimal to no prior cultivation, improving efficiency and soil health. Adopting conservation tillage practices such as zero tillage can enhance soil quality, compared with conventional tillage practices that disrupt soil structure (Torabian et al., 2019).

A net economic analysis refers to the overall economic impact, often calculated by comparing the total costs against the total benefits to determine a net positive or negative outcome. This approach evaluates both the direct and indirect costs and benefits, allowing for a comprehensive understanding of the economic efficiency of various options by focusing on net human welfare (Schmid, 2019). The primary goal is to assess economic efficiency, determining whether the benefits outweigh the costs and thus creating a net positive economic impact (Ekins et al., 2016). Net economic analysis of seed inoculation quantifies the financial benefits versus costs of using beneficial microorganisms (inoculants) to enhance crop growth and yield (Messias et al*.,* 2024). It involves comparing the costs of the inoculant and its application with the increased revenue from higher yields, which is assessed using metrics like net revenue, benefit-cost ratio, and profitability indices (Kannan and Moorthy, 2022). Research shows seed inoculation can improve profitability by reducing production costs and increasing gross revenue for crops like common beans improved nutrient uptake (Karavidas et al*.,* 2022). Inoculants, such as rhizobia, fix atmospheric nitrogen, reducing the need for synthetic nitrogen fertilizers and lowering production costs (Abd-Alla et al*.,* 2023).

Net economic analysis of phosphorus fertilizer involves evaluating the economic benefits of increased crop yields against the costs of purchasing and applying P fertilizer and the associated environmental costs of overuse (Jiao et al*.,* 2016). Net economic analysis of varying plant density assesses how crop yield, input costs, and market value interact to determine the most profitable planting rate, aiming to maximize net profit per hectare. Studies show that increasing plant density can improve yield up to a certain point, but excessively high densities can lead to diminishing returns, reduced individual plant yield, and negative economic impacts due to increased competition for resources, higher costs (like for seed and fertilizer), and potential environmental damage (Willy et al*.*, 2019). The optimal plant density is a complex interaction between the specific crop, variety, environmental conditions, and management practices.

4. CONCLUSION AND RECOMMENDATION

Application of rhizobia at 40 g/Ha, phosphorus at 60 kg/Ha and a planting density of 45 x 15 cm produced the highest grain yield while aapplication of rhizobia at 40 g/Ha, phosphorus at 20 kg/Ha and at planting density of 45 x 15 cm produced the highest net economic benefit Therefore, application of rhizobia, phosphorus and selecting an appropriate plant density is essential for maximizing yield. Based on the findings of the study, to optimize green gram production, farmers in Kirinyaga should adopt an integrated application of rhizobia at 40 g/Ha, phosphorus at 60 kg/Ha and using a 45 x 15 cm planting density.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of this manuscript.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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