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# Effect of Sulfur and Bradyrhizobium Inoculation on Nodulation and Yield of Soybean (*Glycine max* L.) on Nitisols of Southwestern Ethiopia

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**Abstract:** A field experiment was conducted at Jimma Agricultural Research Center (JARC) during 2017/18 growing season to evaluate the effect of integrated use of Bradyrhizobium strain and sulfur fertilization on nodulation and yield of soybean. The experiment consisted of four levels of S (0, 20, 30 and 40 kg ha$^{-1}$) and three Bradyrhizobium strains (MAR-1495, SB-12 and Murdock) arranged factorially in completely randomized block design (RCBD) with three replications. Grain and straw yield increased due to inoculation of Bradyrhizobium strain whether used alone or in combination with S. Nodulation, parameters (nodule number per plant and nodule dry weight) and number of pods per plant were highly significantly influenced due to combined use of Bradyrhizobium strains with sulfur fertilization. The yield and yield components namely (grain yield, hundred seed weight, biomass yield and harvest index) were highly significantly ($P < 0.01$) affected by individual application of sulfur and inoculation of Bradyrhizobium strains alone. Accordingly, the highest grain yield (1496.35 kg ha$^{-1}$) was obtained from application of S at a rate of 30 kg ha$^{-1}$ corresponding 30.96% yield advantages compared with control and 1548.55 kg ha$^{-1}$ grain yield was recorded due to inoculation with Murdock strain corresponding to 30.32% increase over MAR-1495 strain. The results clearly suggested that proper application of S along with Bradyrhizobium strain affect nodulation, grain and straw yield of soybean.

**Keywords:** Sulfur, Strain, Nodulation, Yield

## 1. Introduction

Soybean (*Glycine max* (L.) *Merrill*) is one of the most important legume crop cultivated worldwide and domesticated first in China and it is the world's leading source of oil and protein. Depending on the variety, it can grow best from 0-2200m altitude and under rainfall ranging from 300 to 1200mm. Thus, altitude influences temperature that in turn affects the physiological (esp. flowering and maturity) cycle of the crop. It has the highest protein content (40%) of all food crops and is second only to groundnut in terms of oil content (20%) among food legumes [11]. Soybean requires large amounts of nitrogen (N), phosphorus (P) and potassium (K) as well as a smaller amount of sulfur (S) and some micronutrients. Although soybean requires considerably less P and S than N or K, all are important for plant growth and development. Nitrogen nutrition is not a serious problem as the plant has the inherent ability to obtain most of its N requirement from the atmosphere through biological fixation by forming a symbiotic relationship with Rhizobium bacteria in the soil. Soybean requires 378 kg ha$^{-1}$ N to complete its growth cycle; however, it has the potential to obtain 60 to 70% of its requirement from biological N fixation. Nonetheless, this relationship could not occur without inoculating the seed with appropriate strain of Bradyrhizobium bacteria.

Ethiopia has suitable natural conditions and vast land for investing soybean production. Southwestern part of Ethiopia, particularly Jimma area is favorable location for soybean cultivation and has an economic importance for smallholders [30]. However, the actual yield productivity of the crop has ranged from 920 -1146 kg ha$^{-1}$, if properly managed; it is possible to achieve the yield up to 2000-3500 kg ha$^{-1}$.

Biological nitrogen fixation (BNF) offers an economically attractive and ecologically sound means of improving soybean yield, reducing external N inputs and enhancing the quality of soil, which consequently reduce the dependence on mineral fertilizers that could be costly and unavailable to smallholder farmers. This is achieved when effective and compatible strain is inoculated to the soil/or seed. Legume crops including soybean obtain between 50 - 80% of their N requirements through biological nitrogen fixation [24]. However, [21] reported that the current promiscuous soybean genotypes could not meet all their demand for growth and seed development only by $N_2$ fixation rather it needs addition of mineral nutrient sources and Bradyrhizobium strain which may hardly exist in highly weathered soils because soybean is an exotic crop. Moreover, population of Bradyrhizobia is seldom available in depleted; therefore, nodulation of soybean may require specific species of Bradyrhizobium for effective $N_2$ fixation [1]. However, the success of inoculation depends not only on high quality inoculants but also on the establishment of effective and efficient BNF through optimization of the factors that affect its performance such as legume genotype, climatic, edaphic and management factors [10].

Due to low soil fertility status in the country legume crops are generally grown in sever soil conditions, which are inherently low in nutrient content including S and low soil pH value especially in Southwestern part of Ethiopia [2]. The Ethiopian Soil Information Service is currently involved in mapping the entire country for all nutrients, and has found extensive areas of S, Zn, and B deficiency [29]. Among secondary nutrients S is an essential nutrient for plant growth accounting about 10% of the total N and legume crops require it in a similar or more than that of P for high yield and quality [3, 15]. On the other hand, in areas like Ethiopia where subsistence farming is common, addition of S through organic decomposition is mostly insufficient to meet crop requirement. Thus, continuous application of N containing fertilizers in the form of DAP excluding sulfur further makes its deficiency worse because of widen N:S ratio. Despite the important roles of S in agriculture, research pertaining to its status in soil and its response in crops are almost nonexistent in Ethiopia [12]. Therefore, supplying adequate S to the soil has significant potential to increase the capacity of N fixation by legumes and their grain yield, thereby to improve soil fertility [7]. A research finding revealed that lower N accumulation and a yield reduction of legume crops was recorded when S was limiting in the soil and also recognized not only decreased yield but also quality of products when S is deficient [15, 22]. Therefore, the most important constraints to soybean crop growth may be due to shortage of plant nutrients (S and N). Therefore, a field experiment was conducted to evaluate the effect of S fertilization and inoculation of Bradyrhizobium strain on nodulation, yield and yield components of soybean.

# 2. Materials and Methods

### *2.1. Description of the Study Area*

A field experiment was conducted at Jimma Agricultural Research Center (JARC) Southwestern Ethiopia under rain fed conditions during 2017/2018 cropping season to evaluate the effect of combined application of S and Bradyrhizobium strain on nodulation and grain yield of soybean. Jimma Agricultural Research Center is located at about 358 km far from Addis Ababa city and 12 km from Jimma town to Southwest direction. Geographically it is located at 07°40′.212″ N latitude and 036°47′.055″ E longitude and an altitude of 1763 masl. The average annual maximum and minimum air temperature were 27.2°C and 11.8°C, respectively and the area receives adequate amount of rainfall, 1198 mm per annum. The major reference soil group in the area is dominated by Nitisols, Acrisols, Ferralsols, Vertisols and Planosols.

### *2.2. Treatment Set up and Experimental Procedure*

The experiment consist four levels of sulfur (0, 20, 30 and 40 kg ha$^{-1}$) and three strains of Bradyrhizobium (MAR-1495, SB -12 and Murdock). It was laid out in a completely randomized block design (RCBD) in factorial arrangement with three replications which accommodating seven rows and net plot area was 3m x 3.9m = 11.7m$^2$. Planting was done early June, 2017 based on farmer's local planting calendar where two seeds per hill were drilled on ridges made with 0.05m and 0.60m spacing between plants and each row respectively. After planting the seed was covered with soil and thinned to one seed per hill two weeks later, which accommodates a total of plant population levels 333333 ha$^{-1}$. Harvesting was done at physiological maturity by leaving the outer most rows on both sides of each plot to avoid border effects.

### *2.3. Crop Management*

The field was prepared by removing all unwanted materials and before sowing, the field plowed with oxen, four times to make a fine seedbed. Sulfur was applied as potassium sulfate ($K_2SO_4$) in solid form. In addition to sulfur, other nutrients, such as starter dose of N fertilizer at 18 kg ha$^{-1}$ as Urea [25] and P fertilizer as TSP at recommended dose of 23 kg ha$^{-1}$ was applied at planting for each treatment. Since S fertilizer was applied in the form of $K_2SO_4$, the disproportionate addition of potassium in different treatments was counter balanced by the addition of proportionate amount of potassium chloride (KCl).

### *2.4. Soil Fertilization and Planting*

Soybean seed was selected based on size and healthiness by physical observation. Then the seeds were weighed and surface sterilized by soaking them first with 70% (v/v) ethanol for 10 seconds and 4% (v/v) sodium hypochlorite (NaOCl) solution for five minutes and late washed five times with sterilized water as indicated in [26]. Each strain was applied at the rate of 5gm powder inocula per 1kg of seed. In order to ensure that all the applied inoculum stick to the seed, the required quantity of inoculants was suspended in 1:1 10% sugar solution. The sugar slurry was gently mixed with dry seed and then with carrier-based inoculant so that all the seeds received a thin coating of the inoculant. Then the strain was mixed thoroughly with seeds. For each inoculation, separate plastic bag was used and care was taken to avoid contact among each strain during sowing. The seed was allowed to shaded air dry for a few minutes and then sown at 60 kg $ha^{-1}$ seed rate.

### *2.5. Soil Sampling and Analysis*

A composite soil sample was collected using auger in a diagonal pattern at 0-20cm depth before treatment application. The sample was air-dried, ground using a pestle and a mortar and allowed to pass through a 2 mm sieve. Working samples were prepared and analyzed for soil pH, organic carbon (OC), total nitrogen (TN), available phosphorus (Av.P) and cation exchange capacity (CEC). Total N was determined by Kjeldahl digestion, distillation and titration method [6]. Soil pH was determined (1:2.5 soil: water ratio) according to [20]. Available P was determined using Bray II extraction method as described by [5]. Organic carbon content of the soil was determined by reduction of potassium dichromate by OC compound and determined by reduction of potassium dichromate by oxidation-reduction titration with ferrous ammonium sulfate and OC was determined using the method of [19]. Extracted Phosphorus was determined with atomic absorption spectrophotometer calorimetrically according to molybdenum blue color method described by [18].

### *2.6. Data Collection*

#### *2.6.1. Effective Number of Nodules*

It was determined by carefully uprooting and washing the roots of five plants randomly selected from each plot at 50% flowering stage. The effective nodules were identified by their color where a cross section of an effective nodule made with pocket knife a pink to dark red color whereas the green color indicate non- effective nodules.

#### *2.6.2. Nodule Dry Weight*

It was determined by carefully uprooting and detached the nodules from the root and dried in oven dry until constant weight is attained then the dried nodule was weighed with sensitive balance.

#### *2.6.3. Yield and Yield Components*

Number of pods per plant: It was counted from five randomly selected plants of the middle rows at the time of harvesting from each plot by visual observation and their averages were recorded.

Number of seeds per pod: Was determined as the total number of seeds counted per pod in each plot during harvesting from five randomly selected plants.

Grain yield (kg $ha^{-1}$): It was measured from each plot and converted into hectare base.

Biomass yield (kg $ha^{-1}$): Plants from the net plot area was harvested at physiological maturity, after gained constant weight the dried straw was weighed and converted into hectare base.

Hundred seed weight (gm): Was determined by counting seeds manually from ten randomly selected plants and its weight was measured by sensitive balance and adjusted to 10% standard moisture content of legumes. Finally, the average value was taken as hundred seed weight.

Harvest Index (%): It was expressed as the ratio of economic yield per plant to the total above ground biomass calculated as follow;

$$\text{HI}(\%) = \frac{\text{GY kg/ ha}}{\text{TBY kg/ ha}} \times 100$$

Where, HI = harvest index, GY = Grain yield (at 10% moisture level), TBY = Total biomass yield.

### *2.7. Statistical Analysis*

The collected data were summarized and statistically analyzed using the analysis of variance (ANOVA) procedure for RCBD using SAS 9.3 version software [8]. Treatment means that differed significantly was separated using LSD procedure at 5% level of significance. Correlation coefficient was determined for parameters using the same software to determine relations between yield and yield contributing characters.

## 3. Results and Discussion

### *3.1. Number of Effective Nodules*

The ANOVA table showed that there was highly significant interaction effect on number of effective nodules per plant ($P < 0.01$) due to combined application of sulfur and bio-inoculant strain. Accordingly, the highest effective nodule number per plant (15.33) having pink in color was recorded from application of S at rate of 20 kg $ha^{-1}$ in combination with MAR-1495 strain which produced 31.70% nodule number advantages compared with control (no S fertilization) combined with SB-12 strain. The increase in nodule number is an indication that native strains of Bradyrhizobium bacteria could form symbiotic relation with soybean are absent in the soils of study site. Effective strains are larger in size and few in number which are congregated on main roots and are pink and/ red in colour due to the presence of iron containing substance called leghemoglobin while the nodules produced by ineffective strains have neither leghemoglobin nor pink or red pigmentation. This result is supported by [31] who reported varietal differences in terms of nodule numbers was observed due to inoculation of different rhizobial strains and observations in Chinese milk vetch, respectively.

***Table 1.*** *Soil Physico - Chemical properties of experimental site before sowing in 2017/18 season.*

| Soil Properties | Value | Rating | References |
|---|---|---|---|
| pH (1:2.5 $H_2O$) | 5.52 | Moderate acidic | Tekalign Mamo (1991) |
| Soil BD (g $cm^{-3}$) | 1.25 | Optimum | Hunt and Gilkes (1992) |
| CEC (cmol (+) $Kg^{-1}$) | 16.06 | Medium | Hazelton and Murphy (2016) |
| Total N (%) | 0.10 | Low | Tekalign Mamo (1991) |
| Avail. P (mg $kg^{-1}$) | 8.18 | Low | Landon (1991) |
| OC (%) | 1.84 | Medium | Charman and Roper (2007) |
| OM (%) | 3.17 | Medium | Nelson and Sommers (1982) |
| Soil Textural Class | Clay loam | Ideal | Onwueme and Sinha (1991) |

Moreover, legumes have a high internal P requirement for their symbiotic $N_2$ fixation. [23] Reported that, in addition to the nodule formation, deficiency of P in legumes also markedly affects the development of effective nodules and the nodule leghemoglobin content. It is therefore, suggested that the presence of high amount of available phosphorous in soils as in the soils of the experimental field may be beneficial to nodule nitrogen fixation through the prevention of the decrease of the phosphorous concentration in the plants at the later growth stage.

### *3.2. Nodule Dry Weight*

Nodule dry weight was significantly ($P<0.05$) affected due to the interaction of sulfur fertilization in combination with rhizobial strain (Table 2). The maximum nodule dry weight (4.57 gm $plant^{-1}$) was obtained from application of sulfur at 20 kg $ha^{-1}$ in combination with MAR-1495 strain, which is statistically at par with inoculation of Murdock strain in combination with application of S at 30 kg $ha^{-1}$ (4.50g mg $plant^{-1}$), while the minimum nodule dry weight (2.63 gm $plant^{-1}$) was recorded from application of 40 kg $ha^{-1}$ S in combination with SB-12 strain. The extent of establishment of inoculated rhizobia in soil through bio-fertilizer is dependent on the type of legumes, effectiveness of strains, inorganic content of N, level of available P, K and presence of compatible forms of secondary nutrients (esp. S). At field condition, soybean has shown positive correlation in yield with nodule weight not with nodule number. Plant nodulated by ineffective strains showed lower dry matter yield, grain yield and N content, irrespective of the number of nodules than those nodulated by effective rhizobium strains. The yield increase obtained from the use of bio-fertilizer relates not only to the N enrichment but also to the antagonistic effect on harmful organisms in the rhizosphere. This is in agreement with the report of [17] who observed that nodule dry weight was significantly dependent on Rhizobium strain and soybean variety interaction.

### *3.3. Number of Pods Per Plant*

The ANOVA table showed that there was highly significantly interaction effects ($P < 0.01$) on number of pods per plant due to application of S in combination with Bradyrhizobium strain. Sulfur fertilization at a rate of 30 kg $ha^{-1}$ in combination with Murdock strain recorded significantly the highest number of pods per plant (52.60) which produced 32.07% more number of pods per plant as compared to inoculation of MAR-1495 strain alone. This revealed the presence of positive interactive effect of S and Bradyrhizobium strain on number of pods per plant on Nitisols of Jimma area. Application of S in combination with Bradyrhizobium strains plays an important role in physiological and developmental processes in plant life and the favorable effect of these important nutrients combination (N and S) might accelerate the growth processes, which ultimately resulted in increased yield and quality. The number of pods per plant formed is influenced by the treatment given by Bradyrhizobium strain. This proves that bacteria that work with root nodules affect plants in forming pods. This result agreed with [14] who reported that number of pods produced by soybean is largely determined by vegetative growth such as photosynthesis rate and assimilation.

***Table 2.*** *Interaction effect of S and Bradyrhizobium strains on soybean nodule number, nodule dry weight and number of pods plant$^{-1}$.*

| Treatments | Sulfur rates (kg $ha^{-1}$) | | | |
|---|---|---|---|---|
| Bradyrhizobium Inoculation | 0 | 20 | 30 | 40 |
| | Number of nodule $plant^{-1}$ | | | |
| MAR-1495 strain | 11.27e | 15.33a | 13.47bc | 14.33abc |
| SB -12 strain | 10.47e | 13.07cd | 13.20bcd | 13.33bcd |
| Murdock strain | 13.60bc | 14.80ab | 13.53bc | 11.73de |
| LSD (0.05) | 1.64 | | | |
| CV (%) | 6.89 | | | |
| Bradyrhizobium Inoculation | Nodule dry weight (gm) | | | |
| MAR-1495 strain | 3.53a-d | 4.56a | 4.32ab | 3.17cd |
| SB -12 strain | 2.68d | 4.03abc | 4.03abc | 2.63d |
| Murdock strain | 3.35bcd | 3.27cd | 4.50a | 3.93abc |
| LSD (0.05) | 1.05 | | | |
| CV (%) | 15.01 | | | |

| **Treatments** | **Sulfur rates (kg ha$^{-1}$)** | | | |
|---|---|---|---|---|
| **Bradyrhizobium Inoculation** | **0** | **20** | **30** | **40** |
| Bradyrhizobium Inoculation | Number of pods plant$^{-1}$ | | | |
| MAR-1495 strain | 35.73e | 38.80de | 43.40cd | 43.20cd |
| SB -12 strain | 43.53cd | 43.87cd | 42.13cde | 38.73de |
| Murdock strain | 44.47bcd | 51.20ab | 52.60a | 46.87abc |
| LSD (0.05) | 7.15 | | | |
| CV (%) | 7.41 | | | |

Different small letters denote significant difference between treatments.

### *3.4. Grain Yield*

The ANOVA table showed that there was non-significant interaction effect due to combined application of S and bio-inoculant but main effect was observed highly significantly ($P < 0.01$) affected due to application of S and Bradyrhizobium strain separately. Accordingly, the maximum yield (1496.35 kg ha$^{-1}$) was obtained from application of S at a rate of 30 kg ha$^{-1}$, which produced 30.95% yield advantages compared with control. In case of Bradyrhizobium inoculation 1548.55 kg ha$^{-1}$ was recorded from inoculation of Murdock strain that accounts 30.32% and 22.64% yield advantages compared with MAR-1495 and SB-12 strains, respectively. From this, it has demonstrated that integration of sulfur with Bradyrhizobium inoculation did not increase grain yield compared to the individual application. The result also revealed that grain yield of inoculated plants showed the increasing trends as the rates of S. This implies that S application increases the effectiveness of residual indigenous rhizobia rather than the inoculated isolate. It is not clear how high rates of S application reduce the effectiveness of inoculated isolate. However, there was a main effect on grain yield due to S fertilization. This result agreed with the finding of [16] who reported that maximum seed yield has been recorded by inoculation of soil with biological strains.

Application of S plays an important role in physiological and developmental processes in plant life and the favorable effect of these important nutrient combinations (N and S) might accelerate the growth processes, which ultimately resulted in increased grain yield and quality of the crop. The synergistic effect of N and S is common thus, might be due to utilization of high quantities of nutrients through their well-developed roots and nodules, which might have resulted in better growth and yield. Sulfur and N relationship was established in many studies in terms of dry matter and yield in several crops [4] while working on tobacco plant concluded that there was apparent accumulation of one nutrient when the other nutrient was limited and the accumulated nutrient was used in protein synthesis when the treatment were reversed. A shortage in S supply to the crops lowers the utilization of available soil N, thereby increasing nitrate leaching thus N status in soil become unsatisfactory. This result agreed with the findings of [13].

Application of S might have increased the availability of nutrient to soybean plant due to improved nutritional environment, which in turn, favorably influenced the energy transformation activation of enzymes, chlorophyll synthesis as well as increased carbohydrate metabolism [9]. It also constitutes the main element of amino acids including cysteine and methionine, which are of essential nutrient value and can increase grain yield. Therefore, soybean production and protein quality could significantly improve by increasing the concentration of the S containing amino acids through S fertilization. Thus, yield and quality of legume seeds including soybean are limited by the amount of S partitioned to the seeds as reported by [28].

### *3.5. Biomass Yield*

The ANOVA table showed that there was non- significant interaction effect on biomass yield due to combined application of S and bio-inoculation as shown (Table 3). However, the main effect of S fertilization and inoculation of strain separately was highly significant ($P<0.01$) effects on biomass yield, which might be the presence of indigenous strain in the soil and less competent capacity of the inoculated strain. Accordingly, the highest biomass yield (4924.10 kg ha$^{-1}$) was recorded from S fertilization at a rate of 30 kg ha$^{-1}$, which is statically at par with plots treated with 40 kg ha$^{-1}$ S (4825 kg ha$^{-1}$). Application of 30 kg ha$^{-1}$ S produced 8.24% biomass yield advantages over control. This showed that S is an essential nutrient for plant growth accounting 10% of the total N content [3] and legume crops including soybean generally require S in a similar quantity or more than that of P for high yield and quality there by maximize above ground biomass. The result is in line with the finding of [15].

With regard to strain, the maximum biomass yield (1548.55 kg ha$^{-1}$) was obtained from inoculation of Murdock strain while the minimum biomass yield (1079.02 kg ha$^{-1}$) was recorded from inoculation of SB-12 strain. From the current result, inoculation of Murdock strain produced 22.64% and 30.32% biomass yield advantages compared with SB-12 and MAR- 1495 strain, respectively. Inoculation of Bradyrhizobium strain to soybean seed increased growth and yield contributing parameters including seed weight. Each strain has its own synergetic effect on production of seed weight as seed weight also increased when Rhizobium strains (significantly greater for Murdock) were applied individually but not in combination with S. An increase in biomass was observed because rhizobial inoculation is known to increase yield of several legume crops by increasing nodulation and biomass of root and shoot, which is supported by [27].

### *3.6. Hundred Seed Weight*

The ANVA table showed that hundred seed weight was non- significant interaction effects due to combined application of S and Bradyrhizobium strain but significantly ($P < 0.05$) affected by main effects due to S application alone. The highest hundred seed weight (11.32gm) was recorded from application of S at a rate of 20 kg ha$^{-1}$, which produced 10.60% advantages compared with control plots. This result showed soybean seed quality can significantly improve by increasing the concentration of S containing nutrient sources because S application is an important nutritional element to get full and better soybean seed. The role of S in seed production of soybean has also reported by [9, 14] who found that soybean seed yield and straw yield increased significantly due to application of 60 kg ha$^{-1}$ S followed by 40 kg ha$^{-1}$ S over control.

### *3.7. Harvest Index*

The ANOVA table showed that harvest index was observed to be significantly ($P < 0.05$) affected by inoculation of strain alone and the interaction effect of fertilizer and strain was highly affected at ($P < 0.01$). Sulfur is an important nutrient for plant growth and development. Sulfur interaction with N are directly related to the alteration of physiological and biochemical responses of crops, and thus required to be studied in depth. This would help to understand nutritional behavior of S in relation to N and provide guidelines for inventing balanced fertilizer recommendations in order to optimize yield and quality of crops. A strong interaction of S and N for seed yield was found in Soybean [14].

***Table 3.*** *Effects of sulfur and Bradyrhizobium on soybean yield, biomass, hundred seed weight and harvest index at Jimma in 2017/18 cropping season.*

| Treatments | Grain yield (Kg ha$^{-1}$) | Biomass yield (Kg ha$^{-1}$) | 100 - Seed weight (gm) | Harvest Index (%) |
|---|---|---|---|---|
| Sulfur rate (kg ha$^{-1}$) | | | | |
| 0 | 1033.15c | 4518.50b | 10.12b | 21.93 |
| 20 | 1243.93b | 4549.10b | 11.32a | 21.96 |
| 30 | 14963.35a | 4924.10a | 11.03a | 22.21 |
| 40 | 1327.37b | 4825.90a | 11.18a | 21.30 |
| LSD (0.05) | 154.69 | 256.76 | 0.79 | ns |
| P – value | ** | ** | * | - |
| Bradyrhizobium inoculation | | | | |
| MAR-1495 | 1079.02b | 4809.00a | 11.20 | 20.64c |
| SB -12 | 1198.02b | 4441.70b | 10.75 | 21.94b |
| Murdock | 1548.55a | 4862.50a | 10.80 | 22.96a |
| LSD (0.05) | 133.97 | 222.36 | ns | 0.96 |
| P - value | ** | ** | - | ** |

Different small letters denote significant difference between treatments.

***Table 4.*** *Pearson Correlation Coefficients between growth and yield component of soybean.*

| Variables | GY | BY | NPP | NN | NDW | HSW | HI |
|---|---|---|---|---|---|---|---|
| GY | 1.00 | | | | | | |
| BY | 0.38* | 1.00 | | | | | |
| NPP | 0.52** | 0.44** | 1.00 | | | | |
| NN | 0.12$^{ns}$ | 0.25$^{ns}$ | 0.14$^{ns}$ | 1.00 | | | |
| NDW | 0.6$^{ns}$ | 0.35* | 0.26$^{ns}$ | 0.27$^{ns}$ | 1.00 | | |
| HSW | 0.05$^{ns}$ | 0.12$^{ns}$ | -0.05$^{ns}$ | 0.32* | 0.37* | 1.00 | |
| HI | 0.53** | -0.17$^{ns}$ | 0.35* | -0.14$^{ns}$ | 0.04$^{ns}$ | -0.04$^{ns}$ | 1.00 |

Where, GY = Grain yield, BY = Biomass yield, NPP = Number pods per plant, NN = Number of nodules per plant, NDW= Nodule dry weight, HSW= Hundred seed weight, HI = Harvest index, ** *= significant at P < 0.01,* * *= significant at P < 0.05, ns = non-significant.*

# 4. Correlation Analysis

Correlation analysis between growth and yield related parameters is presented in (Table 4). Crop yield is a cumulative interaction effect of all the dependent and independent characters in an experiment. This is because it has a positive relationship with those parameters and contributes a great growth performances and yield parameter for the plants. According to present result, grain yield was highly significantly ($P < 0.01$) and positively correlated with number of pods per plant ($r = 0.52$), and harvest index ($r = 0.53$) and significantly correlated with biomass yield ($r = 0.38$) indicating they have direct relationship on yield. Number of pods per plant was highly significantly ($P < 0.01$) and positively correlated with biomass yield ($r = 0.44$) and significantly ($P < 0.05$) correlated with nodule dry weight ($r = 0.35$). Number of pods per plant was significantly ($P < 0.05$) and positively correlated with harvest index ($r = 0.35$). Number of nodule and nodule dry weight was significantly ($P < 0.05$) and positively correlated with hundred seed weight $r = 0.32$ and $r = 0.37$, respectively.

# 5. Conclusion

Currently increasing soybean production is a mandatory practice through integrated soil fertility management because it has greater prospects in improving soil fertility and achieving high crop yield via combination of mineral fertilizers and bio-inoculants; which could synergistically improves the overall properties of the soil. Based on the result it is possible to conclude that application of sulfur in combination with Bradyrhizobium strain is an imperative to enhance not only soybean production but also sustainable soil health. Thus, efforts should be made, wherever possible, to introduce inoculation technology to the farming community in Jimma area. Sulfur application at the rate of 20kg $ha^{-1}$ and 30kg $ha^{-1}$ in combination with Murdock Bradyrhizobium strain increased nodulation and yield of soybean. More research based on standard methods needs to be undertaken to assess the contribution of nitrogen-fixing plants to the overall nitrogen budget. Rhizobial inoculants are not locally available, and farmers are not aware about this new technology. Therefore, more efforts need to be done to popularize this cheap and eco-friendly technology among resource poor farming community of the nation. For an alternative use, Murdock and MAR-1495 strains can recommended for soybean inoculum in Jimma area.

# Data Availability Statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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