# Response of soybean to inoculation with *bradyhrizobia spp.*: effect on root nodulation, yield and residual soil nitrogen

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## Abstract

*Legume cro*

*partially or fully meet their N requirement. Soybean is an exotic crop to Ethiopia and thus inoculation with effective rhizobia strains is important to improve its productivity. This study was done with the objective of evaluating the effect of different indigenous and commercial rhizobia strains on nodulation and yield of soybean. The study was conducted in Jabi tehahn district of West Gojam Zone of Amhara Region, Ethiopia in 2015 and 2016. The study was comprised of rhizobia strains MAR-1495, SB-12 and TAL-379 factorial combined with 0 and 23 kg* $P_2O_5$ $ha^{-1}$ *and there was a control (non-inoculated and non-fertilized) treatment. The treatments were laid in randomized complete block design with three replications. The agronomic data analysis result showed that there was a statistically significant* ($P<0.05$) *effect of the use of the rhizobia strains and P fertilizer on nodulation and yield of soybean. The highest grain yield of 2.7 t* $ha^{-1}$ *was obtained from the combined use of 23 kg* $P_2O_5$ $ha^{-1}$ *with the strain MAR-1495 statistically at par with the grain yield recorded from MAR-1495 alone. The strain SB-12 alone also increased the yield significantly as compared to the control treatment. Grain yield advantages of 30.8 and 21.8% over the grain yield obtained from control treatment were found from the strain MAR-1495 and SB-12 alone, respectively. The maximum number of effective nodules per plant was recorded from MAR-1495+23 kg* $P_2O_5$ $ha^{-1}$ *statistically at par with the number of effective nodules counted from MAR-1495 alone. The residual soil nitrogen analysis indicated that the residual soil N differences of 0.029 and 0.011% (0.29 and 0.11 g total N per 1 kg soil, respectively) from MAR-1495 and SB-12 inoculated treatments against the non-inoculated once respectively, while the lowest residual soil N was recorded from the control treatment. Therefore, inoculation of soybean with MAR-1495 primarily and SB-12 as an alternative strain can be recommended for maximum soybean production in Jabi-Tehnan district and similar agro-ecologies.*

**Keywords:** Inoculation, MAR-1495, Nodulation, SB-12, Soybean

## Introduction

Nitrogen and phosphorus are the two major nutrients that limit plant growth in smallholder farms in Africa. Leguminous plants require large amount of nitrogen (N) for grain yield (Hungria and Kaschuk, 2014) but it is difficult for smallholder farmers with limited resources to supply the required high N quantities. Most low income farmers tend to plant legumes without any major external inputs; thus obtaining low grain yields. Under such conditions, legumes depend on biological nitrogen fixation through symbiosis with rhizobia to partially or fully meet their N requirement (Hungria and Kaschuk, 2014).

Rhizobia are bacteria that fix atmospheric N through symbiosis with leguminous plants in a process referred as biological N fixation (BNF). It is estimated that about 50 to 300 kg N $ha^{-1}$ can be fixed by rhizobia bacteria (Bokhara and Sakurai, 2005). Thus, their contribution to the N economy of the soil is quite substantial. Moreover, BNF is believed to consume less energy than nitrogen fixation through the mineral process (Dubey, 2006). For these reasons, inoculation with strains of rhizobia has become an important agronomic practice to augment N supply to legumes and reduce the amount of inorganic N fertilizers required. In addition, legumes are presumed to rely less on external inorganic N sources than non-leguminous crops.

Soybean is a grain legume cultivated in many areas in the world from tropics to temperate regions with a seed yield of 1.4-2 t $ha^{-1}$. The seed (bean) contains about 18% oil and 38% protein and the extraction residue represents more than 40% of the utilization value of the plant (Asiedu, 1989). Soybean fixes up to 200 kg N $ha^{-1}$ $year^{-1}$ when in symbiotic association with *Bradyrhizobium japonicum* (Zhang et al., 2002) reducing the need for potentially environmental damaging N fertilizer (Aseidu, 1989).

Soybean is a recently introduced crop to Ethiopia and its average yield per hectare is by far below the world’s average. To acquire a high yield and use its potential, the crop needs association with *Bradyrhizobium japonicum* to fix atmospheric nitrogen. However, as the crop is exotic indigenous rhizobia bacteria in the district where this study was conducted were not found effective and competitive compared to a commercial rhizobia strain TAL 379 in producing effective nodulation and increasing yield (Tesfaye et.al. 2010). This indicates a need for further screening and selection of alternative and effective commercial rhizobia strains through research. This study was therefore proposed to evaluate the effect of different indigenous and commercial rhizobia strains on nodulation and yield of soybean.

## Materials and methods

### Study site description

This study was conducted in Jabi-Tehnan District of West Gojam Zone of Amhara region in Ethiopia in 2015 and 2016 main cropping seasons. The study district is located between the coordinates of 37$^o$5’59’’ - 37$^o$29’59’’ E and 10$^o$21’26’’ - 10$^o$57’20’’ N with altitudinal ranges of 1500-2500 meters above sea level. The dominant soil type in the study site is Rhodic Nitisols. The district receives a mean annual rainfall of 1250 mm with mean minimum and maximum temperatures of 14 and 32 $^oC$, respectively. The physico-chemical characteristics of surface soil (0-20 cm) for the study area were as given below (Table 1).

**Table 1.** Some physico-chemical characteristics of surface soils of the study area

| Soil parameter | Value |
|---|---|
| Organic carbon (%) | 0.55-3.10 |
| Total nitrogen (%) | 0.044-0.136 |
| Available phosphorus (mg kg$^{-1}$ soil) | 2.6-6.16 |
| Cation exchange capacity CEC (meq 100 g$^{-1}$soil) | 14.8-22.6 |
| Base saturation (%) | 36-55 |
| Texture class | Clay |

### Experimental procedure and treatments

The study was conducted on three farmers’ fields in 2015 and two farmers’ fields in 2016. The farmers’ field was plowed and prepared with oxen-drawn traditional *Maresha* and was divided into experimental plots which had an area of 4 m * 3 m. The space between each plot and block was 1 m. An improved variety of soybean *Gishama 335* was planted in a row with 40 cm and 10 cm spacings between rows and plants, respectively. The P fertilizer was applied in diammonium phosphate form at a rate of 23 kg P2O5 ha$^{-1}$ at planting. Soybean seeds were soaked in water andsugar (90:10 ratio) solution, inoculated with respective strains after gently decanting the excess water under shade just before planting. Three rhizobia strains, MAR-1495, SB-

**Soil sample collection and analysis**

Composite surface (0-20 cm) soil samples were collected at planting for the determination of pH, texture, organic carbon, total nitrogen and available phosphorus analysis. Similarly, surface soil samples at a depth of 0-20 cm were taken plot-wise at harvesting for the determination of residual nitrogen to evaluate the carryover effect of rhizobia bacteria inoculation on soil available nitrogen for the subsequent cropping season. Each soil sample collected was thoroughly mixed and air-dried at room temperature. Air-dried soil samples were ground and passed through a 2 mm sieve. The physico-chemical characteristics of the soil samples were analyzed at Adet Agricultural Research Center Soil testing laboratory in Bahir dar Ethiopia following the standard soil testing procedure.

**Data collection and analysis**

Nodule count data was collected at 50% flowering stage randomly from five plants in the border rows. The inner rows excluding the border were harvested at maturity and yield and yield-related parameters were measured. The grain yield measured was adjusted to 14% moisture content. The agronomic and soil data collected were analyzed statistically using SAS statistical software version 9.0 (SAS, 2009). Mean separation was made by using the Least Significance Difference (LSD) at 5% level of significance.

**Results and discussion**

**Soybean dry matter yield and yield components**

**First year (2015)**

The first year result indicates that there was a significant effect of treatments on the yield of soybean in the two sites (Table 2 and 3). At the first testing site, the highest grain yield (3.37 t $ha^{-1}$) and dry matter yield (8.43 t $ha^{-1}$) was measured from the combined use of the strain MAR-1495 with 23 kg $P_2O_5$ $ha^{-1}$. The maximum 100 seed weight (18.3 g) was also measured from the combined use of the strain MAR-1495 with 23 kg $P_2O_5$ $ha^{-1}$. Statistically similar and comparable yield was recorded from the use of the strains MAR-1495 and SB-12 alone.

**Table 2.** Effect of phosphorus and inoculation on yield and yield components of soybean at testing site 1 in 2015

| Treatment* | Plant height (cm) | Pod no. per plant | 100 seed weight (g) | Grain yield (kg ha$^{-1}$) | Dry biomass (kg ha$^{-1}$) |
|---|---|---|---|---|---|
| 1. Control | 83.0 | 32.9 | 15.7bc | 2238.8c | 6423.6cd |
| 2. MAR-1495 | 90.0 | 33.3 | 16.9abc | 3034.6ab | 7472.4b |
| 3. SB-12 | 82.7 | 33.0 | 15.4bc | 3141.1ab | 7484.4b |
| 4. TAL 379 | 81.9 | 29.3 | 15.1c | 1984.8c | 5618.4d |
| 5. MAR-1495 + 23 kg $P_2O_5$ ha$^{-1}$ | 95.9 | 30.7 | 18.3a | 3374.9a | 8430.6a |
| 6. SB-12 + 23 kg $P_2O_5$ ha$^{-1}$ | 84.1 | 25.8 | 17.5ab | 2574.8bc | 6894.2bc |
| 7. TAL-379 + 23 kg $P_2O_5$ ha$^{-1}$ | 92.7 | 27.2 | 17.1abc | 2554.9bc | 7326.4bc |
| Grand Mean | 87.2 | 30.3 | 16.64 | 2678.5 | 7073.3 |
| CV (%) | 9.2 | 24.1 | 7.36 | 14.9 | 7.1 |
| LSD (5%) | NS | NS | 2.28 | 740.6 | 935.6 |

** Means within a column followed by the same letter are not significantly different at p = 0.05. NS: Non-significant at P=0.05.*

However, at site 2, the dry biomass and grain yield obtained from inoculation alone with the strain MAR-1495 outweighed the other treatments (Table 3). Morover, this did not show statistically significant grain and biomass yield with that of SB-12, MAR-1495 plus P, and SB-12 plus P treatments. The plant height and average pod number per plant showed no significant effect among treatments at both testing sites. The lowest grain and biomass yields were recorded from the control treatment in both testing sites.

**Table 3.** Effect of P and inoculation on yield and yield components of soybean at testing site 2 in 2015

| Treatment* | Plant height (cm) | Pod no. per plant | 100 seed weight (g) | Grain yield (kg ha$^{-1}$) | Dry biomass (kg ha$^{-1}$) |
|---|---|---|---|---|---|
| 1. Control | 64.9 | 34.3 | 14.6 | 1547.1c | 4357.7d |
| 2. MAR-1495 | 75.9 | 31.0 | 15.8 | 2642.2a | 6628.5a |
| 3. SB-12 | 73.1 | 38.7 | 16.8 | 2375.7ab | 6083.3ab |
| 4. TAL-379 | 71.6 | 29.3 | 15.5 | 1837.8bc | 5138.9cd |
| 5. MAR-1495 + 23 kg $P_2O_5$ ha$^{-1}$ | 76.5 | 31.0 | 16.6 | 2610.0a | 6524.3a |
| 6. SB-12 + 23 kg $P_2O_5$ ha$^{-1}$ | 75.9 | 30.6 | 17.0 | 2474.9a | 6371.5ab |
| 7. TAL-379 + 23 kg $P_2O_5$ ha$^{-1}$ | 72.3 | 38.3 | 15.7 | 1682.9c | 5343.7bcd |
| Grand Mean | 72.9 | 33.3 | 16 | 2167.2 | 5778.3 |
| CV (%) | 6.9 | 15.3 | 6.64 | 15.3 | 10.3 |
| LSD (5%) | NS | NS | NS | 588.7 | 1062.6 |

** Means within a column followed by the same letter are not significantly different at p = 0.05. NS: Non-significant at P=0.05.*

At the third testing site (on a research station), however, the yield difference obtained from the control treatment versus both the use of the strains alone and their combined use with P fertilizer (Table 4) was insignificant. This might be most likely due to the availability of residual soil N and P in the surface soil of the research site as a result of long years of N and P fertilization which might have led to nodulation failure and insignificant response to the addition of P fertilizer.

Furthermore, the combined analysis pooled over the three testing sites revealed that there was a statistically significant effect of the use of the strains MAR-1495 and SB-12 with 23 kg P2O5 ha$^{-1}$ and use of MAR-1495 and SB-12 alone on the yield of soybean as compared to the control treatment (Table 5).

**Table 4.** Effect of P and inoculation on yield and yield components of soybean at testing site 3 in 2015

| Treatment* | Plant height (cm) | Pod no. per plant | 100 Seed weight (g) | Grain yield (kg ha$^{-1}$) | Dry biomass (kg ha$^{-1}$) |
|---|---|---|---|---|---|
| 1. Control | 84.7 | 26.9 | 16.7 | 3024.4 | 7320.2 |
| 2. MAR-1495 | 80.5 | 21.4 | 17.9 | 3100.3 | 6937.5 |
| 3. SB-12 | 81.9 | 26.9 | 17.3 | 3012.8 | 6843.8 |
| 4. TAL-379 | 78.7 | 24.7 | 16.8 | 2938.8 | 6812.9 |
| 5. MAR-1495 + 23 kg $P_2O_5$ ha$^{-1}$ | 87.3 | 26.3 | 17.3 | 3124.4 | 7621.5 |
| 6. SB-12 + 23 kg $P_2O_5$ ha$^{-1}$ | 86.9 | 25.2 | 17.4 | 3105.2 | 7350.7 |
| 7. TAL-379 + 23 kg $P_2O_5$ ha$^{-1}$ | 87.2 | 23.3 | 16.5 | 3229.8 | 7201.4 |
| Grand Mean | 83.9 | 24.9 | 17.1 | 3076.5 | 7155.4 |
| CV (%) | 5.3 | 17.5 | 4.96 | 7.6 | 6.6 |
| LSD (5%) | NS | NS | NS | NS | NS |

** NS: Non-significant at P=0.05.*

The pooled analysis of overall testing sites, as shown in Table 5 below, revealed that there was a statistically significant difference among plant height, 100 seed weight, grain and dry matter yield due to the effect of treatments. The maximum plant height (86.6 cm), 100 seed weight (17.4 g), grain yield (3.03 t ha$^{-1}$) and dry matter yield (7.5 t ha$^{-1}$) was measured from the combined use of the strain MAR-1495 with 23 kg $P_2O_5$ ha$^{-1}$ followed with insignificant difference by the grain yield (2.9 t ha$^{-1}$) and dry matter yield (7.0 t ha$^{-1}$) obtained from the use of the strain MAR-1495 alone. The use of strain SB-12 had also statistically similar and comparable yield advantages as compared to the yield measured from strain MAR-1495 with 23 kg $P_2O_5$ ha$^{-1}$. The lowest yield (2.3 t ha$^{-1}$) was recorded from the control treatment.

**Table 5.** Pooled analysis of the effect of P and inoculation on yield and yield components of soybean over the three testing sites in 2015

| Treatment* | Plant height (cm) | Pod no. per plant | 100 Seed weight (g) | Grain yield (kg $ha^{-1}$) | Dry biomass (kg $ha^{-1}$) |
|---|---|---|---|---|---|
| 1. Control | 77.6c | 31.4 | 15.6c | 2270.1c | 6033.8c |
| 2. MAR-1495 | 82.1abc | 28.6 | 16.8ab | 2925.7a | 7012.8ab |
| 3. SB-12 | 79.2bc | 32.9 | 16.5abc | 2806.0ab | 6718.8b |
| 4. TAL-379 | 77.4c | 27.8 | 15.9bc | 2253.8c | 5856.7c |
| 5. MAR-1495 + 23 kg $P_2O_5$ $ha^{-1}$ | 86.6a | 29.3 | 17.4a | 3036.4a | 7525.5a |
| 6. SB-12 + 23 kg $P_2O_5$ $ha^{-1}$ | 82.3abc | 27.2 | 17.3a | 2718.3ab | 6872.1b |
| 7. TAL-379 + 23 kg $P_2O_5$ $ha^{-1}$ | 84.1ab | 29.6 | 16.4abc | 2489.2bc | 6623.8b |
| Grand Mean | 81.3 | 29.5 | 16.6 | 2640.1 | 6662.5 |
| CV (%) | 7.4 | 18.6 | 6.6 | 13.8 | 8.6 |
| LSD (5%) | 5.8 | NS | 1.05 | 350.1 | 554.9 |

** Means within a column followed by the same letter are not significantly different at p = 0.05. NS: Non-significant at P=0.05.*

### Second year (2016)

The second year data analysis results indicated that there was a significant effect of treatments on the grain and dry biomass yield of soybean at two testing sites (Table 6 and 7). At the first testing site, the highest yield was recorded from the use of strains SB-12 with 23 kg $P_2O_5$ $ha^{-1}$and MAR-1495 with 23 kg $P_2O_5$ $ha^{-1}$ (Table 6). However, a statistically similar yield advantage was obtained from the use of the strain MAR-1495 alone at both testing sites. The combined analysis over the two testing sites also revealed that the maximum grain yield of 2.2 t $ha^{-1}$ was obtained from the use of the strain MAR-1495 with 23 kg $P_2O_5$ $ha^{-1}$ followed by the grain yield of 2.1 t $ha^{-1}$ from the strain SB-12 with 23 kg $P_2O_5$ $ha^{-1}$ and the grain yield of 2.0 t $ha^{-1}$ from MAR-1495 alone (Table 8). There were no significant yield differences among the above treatments. Similarly, there was no significant yield differences between separate application of MAR-1495 and SB-12.

**Table 6.** Effect of P and inoculation on the yield and yield components of soybean at testing site 1 in 2016

| Treatment* | Plant height (cm) | Pod no. per plant | Grain yield (kg $ha^{-1}$) | Dry biomass (kg $ha^{-1}$) |
|---|---|---|---|---|
| 1. Control | 72.1 | 30.2 | 1998.8c | 3871.5d |
| 2. MAR-1495 | 65.5 | 30.7 | 2386.0ab | 4517.4bcd |
| 3. SB-12 | 72.0 | 29.1 | 2088.1bc | 5541.2a |
| 4. TAL-379 | 67.5 | 32.3 | 1928.9c | 4128.5d |
| 5. MAR-1495 + 23 kg $P_2O_5$ $ha^{-1}$ | 76.6 | 29.8 | 2580.8a | 5024.3abc |
| 6. SB-12 + 23 kg $P_2O_5$ $ha^{-1}$ | 79.3 | 36.3 | 2628.1a | 5138.9ab |
| 7. TAL-379 + 23 kg $P_2O_5$ $ha^{-1}$ | 69.5 | 35.9 | 2185.7bc | 4274.3cd |
| Grand Mean | 71.8 | 32.0 | 2256.6 | 4642.3 |
| CV (%) | 8.4 | 12.9 | 9.6 | 10.0 |
| LSD (5%) | NS | NS | 385.6 | 826.5 |

** Means within a column followed by the same letter are not significantly different at p = 0.05. NS: Non-significant at P=0.05.*

**Table 7.** Effect of P and inoculation on yield and yield components of soybean at testing site 2 in 2016

| Treatment* | Plant height (cm) | Pod no. per plant | Grain yield (kg $ha^{-1}$) | Dry biomass (kg $ha^{-1}$) |
|---|---|---|---|---|
| 1. Control | 58.5 | 22.5 | 901.7b | 2227.1b |
| 2. MAR-1495 | 63.7 | 26.3 | 1541.9a | 3349.0a |
| 3. SB-12 | 62.7 | 26.1 | 1471.8a | 3180.6a |
| 4. TAL-379 | 59.9 | 22.6 | 1618.0a | 3520.8a |
| 5. MAR-1495 + 23 kg $P_2O_5$ $ha^{-1}$ | 69.4 | 27.8 | 1576.1a | 3312.5a |
| 6. SB-12 + 23 kg $P_2O_5$ $ha^{-1}$ | 62.1 | 27.7 | 1310.1ab | 3020.8ab |
| 7. TAL-379 + 23 kg $P_2O_5$ $ha^{-1}$ | 65.5 | 29.8 | 1327.0a | 3066.0ab |
| Grand Mean | 63.1 | 26.1 | 1374.2 | 3065.3 |
| CV (%) | 11.5 | 10.3 | 14.9 | 13.9 |
| LSD (5%) | NS | NS | 416.3 | 873.3 |

** Means within a column followed by the same letter are not significantly different at p = 0.05. NS: Non-significant at P=0.05.*

**Table 8.** Effect of P fertilizer and inoculation on yield and yield components of soybean pooled over the two testing sites in 2016

| Treatment* | Plant height (cm) | Pod no. per plant | Grain yield (kg ha$^{-1}$) | Dry biomass (kg ha$^{-1}$) |
|---|---|---|---|---|
| 1. Control | 65.3 | 26.3c | 1450.3c | 3049.3c |
| 2. MAR-1495 | 64.6 | 28.5abc | 1964.0ab | 3933.2ab |
| 3. SB-12 | 67.4 | 27.6bc | 1779.9b | 4360.9a |
| 4. TAL-379 | 63.7 | 27.5c | 1804.5b | 3885.4ab |
| 5. MAR-1495 + 23 kg $P_2O_5$ ha$^{-1}$ | 73.0 | 28.8abc | 2179.0a | 4339.6a |
| 6. SB-12 + 23 kg $P_2O_5$ ha$^{-1}$ | 70.7 | 31.9ab | 2100.9a | 4291.7a |
| 7. TAL-379 + 23 kg $P_2O_5$ ha$^{-1}$ | 67.5 | 32.9a | 1756.3b | 3670.1b |
| Grand Mean | 67.4 | 29.1 | 1849.3 | 3914.5 |
| CV (%) | 10.1 | 12.7 | 12.4 | 11.6 |
| LSD (5%) | NS | 4.4 | 286.1 | 567.4 |

** Means within a column followed by the same letter are not significantly different at p = 0.05. NS: Non-significant at P=0.05.*

Similarly, the combined analysis result over the two experimental years revealed that there was a highly significant effect of treatments on the yield of soybean (Table 9). The highest grain and dry matter yields of 2.7 t ha$^{-1}$ and 6.4 t ha$^{-1}$, respectively were obtained from the use of the strain MAR-1495 with 23 kg $P_2O_5$ ha$^{-1}$. The use of the strain MAR-1495 alone had a statistically similar yield advantage as compared to its combined use with 23 kg $P_2O_5$ ha$^{-1}$ and increased the yield by 30.8% when compared with the control treatment. Inoculation of soybean with the strain SB-12 had a statistically similar yield advantage as that of the strain MAR-1495 and had a 21.8% yield advantage over the yield obtained from the control treatment. A similar result was reported by Fitsum et.al. (2016) at Pawe. In line with this, the results of Mahmood et al. (2009) revealed that the application of *rhizobial* inoculant alone significantly increased nodulation of soybean. The reason might be due to inoculation with *Rhizobia*, which increased the number of bacteria and hence more nodules per plant were produced.

**Table 9.** Effect of use of different rhizobia strains and P fertilizer on the yield (kg ha$^{-1}$) of soybean combined over years

| Treatment* | Plant height (cm) | Pod no. per plant | Grain yield (kg ha$^{-1}$) | Biomass yield (kg ha$^{-1}$) |
|---|---|---|---|---|
| 1. Control | 72.7c | 29.3 | 1942.2e | 4840.0e |
| 2. MAR-1495 | 75.1bc | 28.5 | 2541.0ab | 5780.9bc |
| 3. SB-12 | 74.5bc | 30.8 | 2366.2bc | 5708.2bc |
| 4. TAL-379 | 71.9c | 27.6 | 2093.3de | 5152.7de |
| 5. MAR-1495+ 23 kg $P_2O_5$ ha$^{-1}$ | 81.1a | 29.1 | 2730.2a | 6387.6a |
| 6. SB-12+ 23 kg $P_2O_5$ ha$^{-1}$ | 77.7ab | 29.1 | 2497.8ab | 5950.5b |
| 7. TAL-379+ 23 kg $P_2O_5$ ha$^{-1}$ | 77.4ab | 30.9 | 2196.1cd | 5442.4cd |
| Grand Mean | 75.7 | 29.3 | 2334.8 | 5601.4 |
| CV (%) | 8.6 | 16.7 | 13.6 | 9.6 |
| LSD (5%) | 4.8 | NS | 236.2 | 401.4 |

** Means within a column followed by the same letter are not significantly different at p = 0.05. NS: Non-significant at P=0.05.*

## Nodule formation

The maximum effective average number of nodules (14.9) per plant was recorded from the combined use of MAR-1495 with 23 kg $P_2O_5$ ha$^{-1}$ fertilizer followed with insignificant difference by the average effective nodule number (11.6) per plant recorded from the use of the strain MAR-1495 alone (Table 10). The number of effective nodules per plant recorded in this study are supported by Pedersen (2003; 2004), who reported that a successful nodulation by the V3 to V4 growth stage should produce 8 to 10 healthy nodules per plant. Tolera et. al. (2015) also reported that higher nodule dry biomass and dry plant biomass were produced from soybean inoculated with rhizobia strain MAR-1495.

**Table 10.** Effect of inoculation trains and P on nodulation of soybean in 2015

| | Effective nodule number per plant | | |
|---|---|---|---|
| Treatment* | Site 1 | Site 2 | Combined |
| 1. Control | 0.4c | 1.6d | 1.0d |
| 2. MAR-1495 | 5.0a | 18.1ab | 11.6ab |
| 3. SB-12 | 4.2ab | 12.9abc | 9.4bc |
| 4. TAL 379 | 1.3bc | 8.3cd | 5.9cd |
| 5. MAR-1495 + 23 kg $P_2O_5$ ha$^{-1}$ | 4.6ab | 20.2a | 14.9a |
| 6. SB-12 + 23 kg $P_2O_5$ ha$^{-1}$ | 6.2a | 15.9abc | 10.1abc |
| 7. TAL-379 + 23 kg $P_2O_5$ ha$^{-1}$ | 6.6a | 11.3bc | 8.9bc |
| Grand Mean | 4.1 | 12.6 | 8.8 |
| CV (%) | 40.7 | 34.2 | 48.9 |
| LSD (5%) | 3.4 | 7.7 | 5.12 |

** Means within a column followed by the same letter are not significantly different at p = 0.05. NS: Non-significant at P=0.05.*

## Residual soil nitrogen

The statistical analysis result of the surface soil total nitrogen content in percent indicated that although statistically insignificant, at site I, the highest residual total nitrogen (0.239%) was recorded from plots where MAR-1495 was inoculated followed by the residual soil total nitrogen (0.200%) where SB-12+P was applied. While, at the other testing site, the highest residual soil nitrogen was recorded from plots where TAL-379 and SB-12 were inoculated followed by MAR -1495 (Table 11).

**Table 11.** Effect of inoculation on residual soil total nitrogen (%)

| Treatment* | Site 1 | Site 2 | Combined |
|---|---|---|---|
| 1. Control | 0.1877 | 0.1731 | 0.1804 |
| 2. MAR-1495 | 0.2392 | 0.1796 | 0.2094 |
| 3. SB-12 | 0.1758 | 0.2145 | 0.1913 |
| 4. TAL-379 | 0.1849 | 0.2244 | 0.2007 |
| 5. MAR-1495+ 23 kg $P_2O_5$ $ha^{-1}$ | 0.1878 | 0.1944 | 0.1911 |
| 6. SB-12+ 23 kg $P_2O_5$ $ha^{-1}$ | 0.2007 | 0.1743 | 0.1901 |
| 7. TAL-379+ 23 kg $P_2O_5$ $ha^{-1}$ | 0.1872 | 0.1661 | 0.1787 |
| Grand Mean | 0.194 | 0.188 | 0.1918 |
| CV (%) | 17.5 | 17.6 | 17.9 |
| LSD (0.05) | NS | NS | NS |

**NS: Non-significant at P=0.05*

## Conclusion and recommendation

The use of the rhizobia strains MAR-1495 and SB-12 combined with P fertilizer (23 kg $ha^{-1}$ $P_2O_5$) gave the highest yield. However, the use of the rhizobia strains MAR-1495 and SB-12 alone gave significantly higher yields than the control treatment with sizeable yield advantages. Moreover, significant residual soil nitrogen was recorded from plots where MAR-1495 and SB-12 were inoculated which is beneficial for subsequent crop production. Thus, sole use of the strains MAR-1495 primarily and alternatively the strain SB-12 can be recommended for soybean production in Jabi tehnan district and similar agro-ecologies.

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