

Original Research Article

Evaluation of Elites Isolates of Bradyrhizobia Nodulating on Groundnut Varieties (*Arachis hypogaea* L.) at Assosa District of Western Ethiopia.

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Abstract

A field experiment was conducted on Nitisols of Asossa Agricultural Research Centre during 2017/18 2018/19 cropping season to investigate the response of yield, partial budget analysis of soybean varieties to bio-fertilizer and inorganic nitrogen. Two locally suitable varieties 4 groundnut rhizobial strains with control and 18 kg N ha⁻¹, totally 12 treatments had combined in factorial RCBD. TSP fertilizer was applied uniformly to all plots at 46 kg P₂O₅ ha⁻¹. The treatments consists of: two Varieties of groundnut (V1= Muniputer V2= Babile 2) and 4-strains with 18 kg N ha⁻¹ and uninoculated (control) factorial treatment combination were tested. Analysis of variance for two factors randomized complete block design (Table 1) revealed highly significant difference ($P < 0.001$) due to the main effects of rhizobia and varieties for the means of seed yield. On the other hand the interaction effects of rhizobia and varieties had non-significant ($p > 0.05$) for the means of seed yield, nodule dry weight and straw dry weight. The highest (186.5) nodules per plants were recorded from the interaction effect of Dibate moderate and Maniputer variety, while less nodules per plant (76.25) were produced by Dibate moderate with Babile variety. The Dibate 2 rhizobia had the highest net-benefit of 67,655 Ethiopian birr. The lowest net benefit was obtained by the application of the Dibate moderate and none inoculant. The profitability of the study showed that application of Moderate 2 rhizobia which provided the relatively high net benefit (67,655 ETB), was recommended to apply bio fertilizers. The % MRR between any pair of undominated treatments denotes the return per unit of investment in fertilizer expressed as a percentage. Economic analysis revealed that maximum marginal rate of return was recorded with application of Dibate 2 (9257.2%). So we could be recommended Dibate 2 strain for ground nut production in Asossa district. The best recommendation for treatments is not necessarily based on the highest marginal rate of return, rather based on the minimum acceptable marginal rate of return and the treatment with the high net benefit, relatively low variable cost together with an acceptable MRR becomes the tentative recommendation.

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Key words, ground nut, strain, varieties, yield partial budget analysis

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1. Background

Groundnut (*Arachis hypogaea* L.) is an important monoecious annual legume used for oilseed, food and animal feed all over the world (Upadhyaya *et al.*, 2006). Ground nut was grown on nearly 25.45 million ha worldwide with the total production of 45.23 million tons and an average yield of 1.777 tons/ha in 2009; (FAOSTAT (2010). Groundnut has several uses. For people in many developing countries, groundnut is the principal source of digestible protein (25 to 34%), cooking oil (44 to 56%), and vitamins like thiamine, riboflavin, and niacin (Savage and GP;

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Keenan, ~~J~~ (1994). The estimated production area and yield of groundnut in Ethiopia in 2015/2016 cropping season were 67,062.4 hectares and 1,039,395.34 quintals, respectively, and the largest groundnut production areas are found in Oromiya (39,469.11 ha), Benshangul-Gumuz (18,192.29 ha), Amhara (6585.51 ha) and SNNPR (543.84 ha) in regional states (CSA, 2016).

FAOSTAT (2010) reveals that, groundnut yield in Africa is lower (980 kg ha^{-1}) than the average world groundnut yields. Researchers associate these lower yields to abiotic, biotic and socio-economic factors (Caliskan *et al.*, 2008). The most known contaminants of groundnuts are aflatoxins, which are metabolic by-products of the moulds of different *Aspergillus* species. The moulds are common saprophytic fungi found in seeds and soils throughout the major groundnut producing areas of the world (Griffin and Garren, 1974). Contamination of groundnut with fungi and aflatoxin endangers health of humans and animals and lowers market value (Abdalla *et al.*, 2005).

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Biological nitrogen fixation (BNF) is very useful for small holder farmers as it is cost effective and environmentally friendly that improves the N requirement of legumes and other succeeding crops. Inoculation with compatible and effective rhizobia may be necessary to optimize nitrogen fixation and legumes grain yields, where a low population of native rhizobial strains predominates (Chianu *et al.*, 2011). Therefore, evaluation and identification of appropriate and effective rhizobial strains are crucial to enhance nitrogen fixation and yield of ground nut. In the present investigation, therefore, the influences of rhizobia types and having no rhizobial association with soybean were thoroughly examined under field conditions rain season. Therefore this experiment was designed for the purpose of evaluated the *rhizobial* strain types and inorganic N for ground-nut under field condition of Asossa District.

2. Material and methodology

2.1. Treatments and Experimental Design

The experiment was conducted in Benshal-gul Gumuz Regional State, at Asossa Agricultural Research Center (AsARC) from 2017 to 2019 main cropping season under rain fed field condition. Two locally suitable varieties (V1= Muniputer V2= Babile 2) und 4 groundnut *rhizobial* strains (Dibate moderate, Dibate2, A7 und A10) with control and 18 kg N ha⁻¹, totally 12 treatments had combined in factorial RCBD. TSP fertilizer was applied uniformly to all plots at 46 kg P₂O₅ ha⁻¹. Two Varieties of groundnut (V1= Muniputer V2= Babile 2) and 4-strains with 18 kg N ha⁻¹ and uninoculated (control) factorial treatment combination were tested

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2.2 Statistical Data Analysis

Analyses of variances for the data were recorded and conducted using the SAS GLM procedure (SAS 1998). Least significant difference (LSD) test at 5% probability used for mean separation when the analyses of variance indicate the presence of significant differences.

2.3. Economic Analysis

Mean grain yield of the selected treatment was used in partial budget analysis (CIMMTY, 1988). Economic analysis was performed to investigate the economic feasibility of the treatments (fertilizer rates). A partial budget, dominance and marginal analysis were used. The average open market price (Birr kg⁻¹) for ground nut and the official prices of Urea and bio fertilizers were used for economic analysis. The dominance analysis procedure as detailed in CIMMYT (1988) was used to select potentially profitable treatments from the range that was tested. The selected and discarded treatments using this technique are referred to as undominated and Dominated' treatments, respectively. The undominated treatments were ranked from the lowest (the farmers' practice) to the highest cost treatment. For each pair of ranked treatments, a % marginal rate of return (MRR) was calculated. The % MRR between any pair of undominated treatments denotes the return per unit of investment in fertiliser expressed as a percentage.

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3. Result and discussion

Analysis of variance for two factors randomized complete block design (Table 1) revealed highly significant difference ($P < 0.001$) due to the main effects of rhizobia and varieties for the means of seed yield. Similarly the analyzed data indicated that the varieties had significant ($p < 0.05$) and highly ($p < 0.001$) difference for the means of nodule dry weight and straw dry weight (Table 1). However there were no significant difference between the rhizobia, inorganic N and negative N control for the means of nodule dry weight and straw dry weight. On the other hand the interaction effects of rhizobia and varieties had non-significant ($p > 0.05$) for the means of seed yield, nodule dry weight and straw dry weight. The variety Maniputer has been found to be the most promising for increasing nodule dry weight, straw dry weight and yield than that of Babile. Rhizobial strains (Dibate 2, A7 and A10) performed well to produce more yield. The reason might be better nodulation because of perfect Rhizobium supply in the rhizosphere and thus nitrogen fixation giving healthy, environment for plant growth and ultimately better yield production. Relatively smallest mean of nodule dry weight, straw dry weight and yields were observed with non-inoculated treatment. This might be due to poor nutrition in the soils and inefficiency of natural Rhizobium present in the soil.

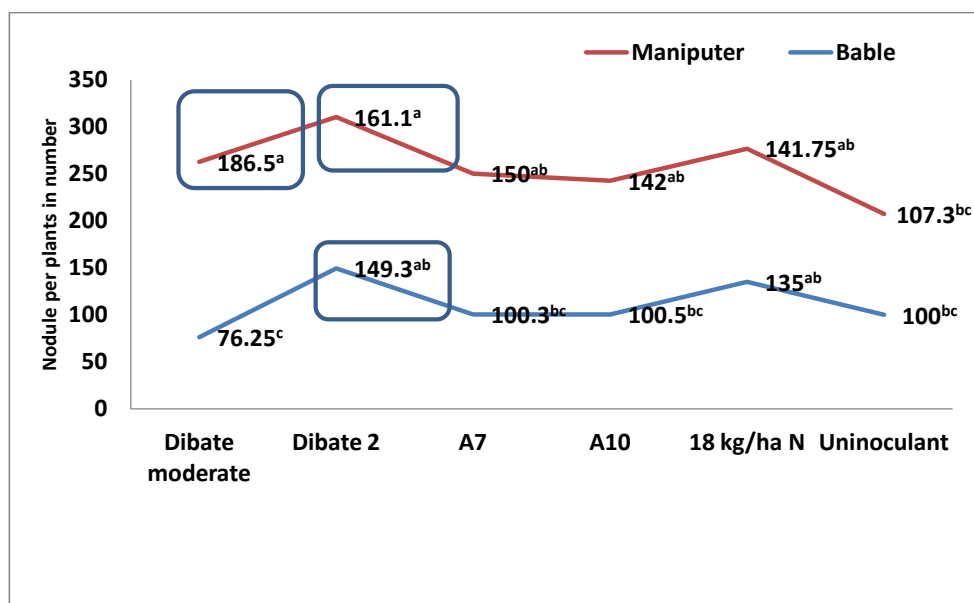
Table1. The effect of rhizobium and variety main effects on soybean yield and nodule dry weight.

Source of variation	Crop yield kg/ha	Nodule dry weight/ plants	Straw dry weight kg/plants
Rhizobium			
Dibate moderate	1158.1b	0.72	3.74
Dibate 2	1507.0a	0.72	4.56
A7	1380.1ab	0.67	3.86
A10	1345.1ab	0.66	3.34
18 kg N /ha	1275.4ab	0.67	3.97

Un inoculants	1174.3b	0.64	3.61
LSD	245	-	-
Varieties			
Maniputer	1734.0a	0.92a	4.45a
Babile	879.0b	0.44b	3.24b
LSD	145.1***	0.2*	0.57***
CV%	18.13	33.59	24.87

Means followed by the same letter within a column or row are not significantly different at 5% level of significance; * = 0.05, ** = 0.01 and *** = indicates significant difference at 0.1% level of significance.

The interaction effect due to rhizobia and varieties were highly ($p < 0.01$) significant difference for the number of number nodule per plants. The highest (186.5) nodules per plants were recorded from the interaction effect of Dibate moderate and Maniputer variety, while less nodules per plant (76.25) were produced by Dibate moderate with Babile variety. Maximum nodule mass per plant were observed where Rhizobium inoculation was applied as Dibate moderate followed by Dibate 2 while inoculated to Maniputer variety. There was no significant difference among the individual Rhizobium strains and inorganic N but performed better than that of control treatment, while treated to Maniputer variety. The reason might be due to the synthetic inoculation of Rhizobia, which increased the number of bacteria and hence more nodules per plants were produced. Maniputer proved to be the most efficient in nodulation and hence nitrogen fixation ultimately in improving crop yield as compared to the variety Babile might be the genetic potential of the crop. The results are in agreement with the findings of Thongchai-Mala (1988), who tested four Rhizobium strains with five groundnut cultivars. After 40-60 days, the results indicated that, for five cultivars, Rhizobium strain NC-92 had the highest tendency to increase nodule number.



Means followed by the same letter are not significantly different at 1% level of significance; LSD (1%) = 53.6 to compare rhizobia variety interaction; **= indicates significant difference at 1% level of significance; and CV (%) = 28.4

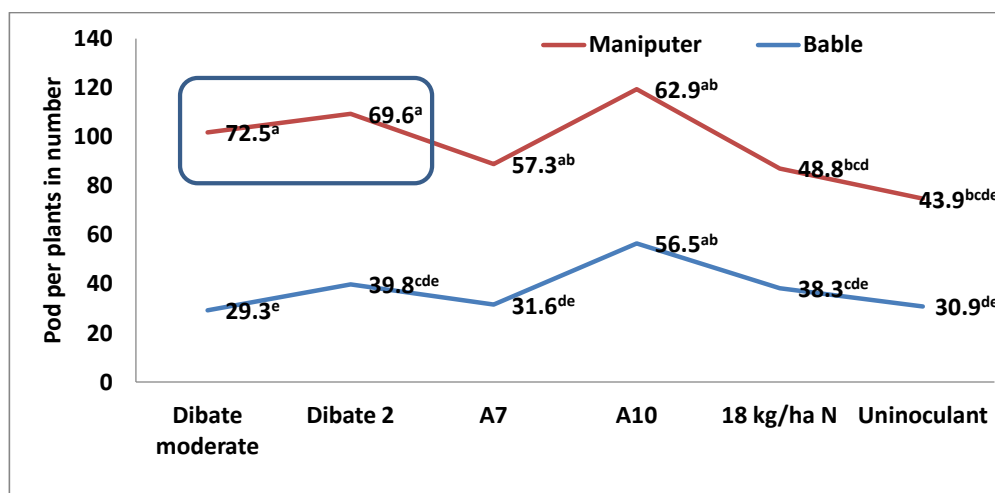
Figure 1. Interaction effect of rhizobia strain and varieties on number of nodule per plant of soybean at Asossa district.

Similar to the previous the interaction effect of rhizobia and varieties were highly ($p < 0.01$) significant difference for the number pod per plants. Maximum pod per plant were observed where Rhizobium inoculation was applied as Dibate moderate followed by Dibate 2 while inoculated to Maniputer variety. Similar to nodule per plant the variety Maniputer has been found to be the most promising for increasing number of pods per plant than that of Babile. Minimum number of pods per plant was observed with Dibate moderate and non-inoculated treatments, while treated to Babile variety. This might be due to poor nutrition in the soils and inefficiency of natural Rhizobium present in the soil, and the genetic potential of Babile variety

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Means followed by the same letter are not significantly different at 1% level of significance; LSD (1%) = 19.3 to compare rhizobia variety interaction; **= indicates significant difference at 1% level of significance; and CV (%) = 27.22

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Figure 2. Interaction effect of rhizobia strain and varieties on number of pod per plant of soybean at Asossa district.

3.1. Partial budget analysis

The Dibate 2 rhizobia had the highest net-benefit of 67,655 Ethiopian birr, followed by A7 rhizobia, and A10 rhizobia which also had a total of 61,944.5 and 60,369.5 Ethiopian birr net benefit respectively. The lowest net benefit was obtained by the application of the Dibate moderate and none inoculant with net benefit of 51,954.5 and 52,843.5 ETB the respectively. The increased production of the crop due to the application of inputs might or might not be beneficiary to farmers (CIMMYT, 1988). Therefore, partial budget analysis (CIMMYT, 1988) was employed to estimate the net benefit, dominance analysis and marginal rate of return that could be obtained from various alternative treatments (CIMMYT, 1988). The profitability of the study showed that application of Moderate 2 rhizobia which provided the relatively high net benefit (67,655 ETB), was recommended to apply bio fertilizers. The highest net benefits from the application of inputs for the production of the crop might not be sufficient for the farmers to

accept as good practices. In most cases, farmers prefer the highest profit (with low cost and high income). For this purpose it is necessary to conduct dominated treatment analysis (CIMMITY, 1988).

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Table 2. Partial Budget Analysis of rhizobia application types on soybean at Asossa Zone

Rhizobia	Crop yield kg/ha	GR (ETB ha ⁻¹)		VC (ETBha ⁻¹)	NB(ETBha ⁻¹)	MRR%
		from yield				
Un inoculants	1174.3	52,843.5		0	52,843.5	0
Dibate 2	1507.0	67,815.0	160	67,655.0	9257.2%	
Dibate Moderate	1158.1	52,114.5	160	51,954.5	D	
A7	1380.1	62,104.5	160.0	61,944.5	D	
A10	1345.1	60,529.5	160.0	60,369.5	D	
18 kg N /ha	1275.4	57,393	366.42	57026.658	D	

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N. B: Prices - Urea= 8.24 birr/kg, Price of ground nut = 45 birr/kg, & Labor cost =30 birr/ person/day for 8 hours, TC=Total cost, GR = Gross return (Return from Grain) =price /kg* yield in kg and NB = Net benefit = gross return – Variable cost, VC = variable cost.

The dominance analysis showed that the net benefits of all treatments were dominated except application of Moderate 2. The % MRR between any pair of undominated treatments denotes the return per unit of investment in fertilizer expressed as a percentage. Economic analysis revealed that maximum marginal rate of return was recorded with application of Dibate 2 (9257.2%). The marginal rate of this treatment was well above the 100% minimum (CIMMYT, 1988). Accordingly, the study revealed that application of Dibate 2 was considered as the best for

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recommendation. The best recommendation for treatments subjected to marginal rate of return is not necessarily based on the highest marginal rate of return, rather based on the minimum acceptable marginal rate of return and the treatment with the highest net benefit, relatively low variable cost together with an acceptable MRR becomes the tentative recommendation (CIMMYT, 1988).

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4. Summary and recommendation

In recent years, crop productivity in Ethiopia in general and in Benshal-gul Gumuz region in particular has shown a declining trend, in spite of the best use of improved varieties. The most possible causes of this decline soil fertility depletion and the continuous use of the traditional fertilizer, which have limited the yield and crop quality. The present study was conducted in Benishangul Gumuz Regional State, at Asossa Agricultural Research Center station in the 2017/2018 to 2018/2019 main cropping season under rain fed field conditions. Therefore this experiment was designed for the purpose of evaluated the *rhizobial* strain types and inorganic N for ground nut under field condition of Asossa District. The rhizobia strain on grain yield highly significant difference ($P \leq 0.001$), however there was no significant differences between the rhizobia strain and inorganic fertilizer (**18 kg N /ha**). Accordingly, the study revealed that application of DIBATE 2, A10 and A7 as the best strain recommended agronomical for soybean production at Assosa area.

The profitability of the study showed that DIBATE 2 strain which provided the relatively highest net benefit (67,655 ETB), was the best strain for Asossa district. The best recommendation for treatments subjected to marginal rate of return is not necessarily based on the highest marginal rate of return, rather based on the minimum acceptable marginal rate of return and the treatment with the high net benefit, relatively low variable cost together with an acceptable MRR becomes the tentative recommendation. Therefore economically we recommend the treatments DIBATE2 that have acceptable marginal rate of return, relatively high net benefit and relatively small total cost of production for soybean production at Begi and Asossa district respectively.

5. Reference

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Chianu, J.N., Nkonya, E.M., Mairura, F.S., Chianu, J.N. & Akinnifesi, F. K. (2011) Biological nitrogen fixation and socioeconomic factors for legume production in sub-Saharan Africa: a review. *Agronomy for Sustainable Development*, 31 (1):139-154.

Hungria M, Vargas MAT. Environmental factors affecting N₂ fixation in grain legumes in the tropics, with an emphasis on Brazil. *Field Crops Res.* 2000;65:151–164.

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Thongchai, M. 1988. Response of groundnut to different Rhizobium strains. *Thai. J. Soils & Fert.* 10: 261-271.

FAOSTAT. Groundnut world production; 2015. Available: <http://www.faostat.fao.org>.

Upadhyaya, H.D., Reddy, L.J., Gowda, C.L. and Singh, S. 2006. Identification of diverse groundnut germplasm: Sources of early maturity in a core collection. *Field Crops Research* 97: 261-271.

FAOSTAT. 2010. Groundnut world production. <http://www.faostat.fao.org>.

Central Statistical Agency (CSA). 2016. Agricultural sample survey in 2009/2010. Addis Ababa, Ethiopia

Caliskan, S., Arslan, M. and Arioglu, H. 2008. Effects of sowing date and growth duration on growth and yield of groundnut in a Mediterranean-type environment in Turkey. *Field Crops Research* 105: 131-140

Abdalla, A.T., Stigter, C.J., Mohamed, H.A. and Gough, M.C. 2005. Identification of microorganisms and mycotoxin contamination in stored groundnut in central Sudan. 10 p.

Griffin, G. J. and Garren, K.H. 1974. Population levels of *A. flavus* and the *A. niger* group in Virginia peanut field soils. *Phytopathology* 64: 322-325.

Savage GP, Keenan JI. The composition and nutritive value of groundnut kernels. Pages 173–213 in the groundnut crop: A scientific basis for improvement (Smartt J, ed.). London, UK: Chapman and Hall; 1994.

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