Effect of rhizobial inoculants and micronutrients on yield and yield components of faba bean (Vicia faba L.) on vertisol of Wereillu district, South Wollo, Ethiopia

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Abstract: Nitrogen fixation by legumes like faba bean is a cheap way of fixing atmospheric nitrogen to plant available form. However, the inoculation of grain legumes with rhizobium bacteria along with the addition of micronutrients is poorly researched in Amhara Region of Ethiopia. Thus a study to examine the effects of rhizobium leguminosarum (var vicae) strains and micronutrients on nodulation, growth, and yield of faba bean was conducted in Wereillu district of Amhara Region, Ethiopia during the rainy season of 2018. The treatments comprised three faba bean strains (un-inoculated, EAL-1018, EAL-1035 and EAL-17) and two micro nutrients (without, zinc, boron) arranged in a randomized complete block design with three replications. The collected data on yield and yield-related parameters were analyzed using Statistical Analysis System (2003), version 9.1 and subjected to Duncan’s Multiple Range Test for mean separation when the analysis of variance was significant. The mean separation revealed that the combined effect of EAL-1018 and boron brought significantly (P ≤ 0.05) higher difference on nodule number, nodulation volume, nodule dry weight, biomass yield and grain yield compared to the control. The combined effect of faba bean strain, EAL-1018 and boron gave
65.9% grain yield advantage over the control and 13.9% compared to the second promising treatment (EAL-1018 alone). Hence, EAL-1018 combined with boron is recommended for the study area and similar agro-ecologies.

**Keywords:** Symbiosis; faba bean; strain; nitrogen fixation; legume

1. Introduction

1.1. Background and justification

Biological nitrogen fixation, which is the conversion of atmospheric nitrogen to ammonia by microorganisms in root nodules of legume plants, plays a significant role in the global nitrogen cycle and world agriculture. The ability to fix nitrogen is widely but sporadically distributed among Archaea and Bacteria which includes these families: Proteobacteria, Firmicutes, Cyanobacteria, Actinobacteria and Chlorobi (Santos et al., 2012).

Symbiosis refers to a close and prolonged interaction between organisms of different species. Previously, the term is restricted to a mutualistic relationship wherein both organisms benefit from the interaction. This cooperative interaction promotes the fitness of both species, and thereby reinforces their symbiotic relationship. The legume secretes a cocktail of phenolic molecules, predominantly flavonoids and isoflavonoids, into the rhizosphere. These signals are taken up by rhizobia, bind the transcriptional regulator Nod, and activate a suite of bacterial nodulation genes (Long, 1996). These nodulation genes are responsible for the production of lipochito-oligosaccharides (LCOs) called Nod factors. Nod factors are key symbiotic signals and are indispensable in the specific Host–Rhizobium interaction and at later stages in the infection process and nodule organogenesis (Oldroyd & Downie, 2008). Nod factors trigger plant cell division and meristem formation, and the rhizobia infect legume roots through crack entry, intercellular colonization of epidermal cells, or the well-studied formation of infection threads (Sprent & James, 2007).

One of the well-known symbioses occurs between legume plants and rhizobia (nitrogen-fixing soil bacteria). Rhizobia set up harmonious organs named root nodules on the underlying foundations of their host, and multiply by separating supplements from the host plant. Thus, they supply their host plants with nitrogen assets delivered by nitrogen gas obsession (Fujita et al., 2014).

Faba bean (Vicia faba L.) is an important grain-legume crop and a good source of dietary protein (Hanelt & Mettin, 1989). It is a major food and feed legume because of the high nutritional value of its seeds, which are rich in protein and starch (Duc et al., 2010). China has been the main producing country of faba bean, followed by Ethiopia, Egypt, Italy and Morocco. Ethiopia is considered as the secondary center of diversity and also one of the nine major agro-geographical production regions of faba bean (Hailu et al., 2014).

Faba bean takes the first place among pulse crops’ production cultivated in Ethiopia which is leading the protein source for the rural people and used to make various traditional dishes. According to Central Statistical Agency CSA (2018), faba bean takes over 3.45% of cultivated land with an average national productivity of 2.1 tons ha⁻¹. It is also the first among pulse crops cultivated in Amhara region and South Wollo zone with average productivity of 1.88 and 1.4 tons ha⁻¹, respectively (CSA, 2018). However, the optimal yield production in faba bean is dependent on symbiosis with Rhizobium leguminosarum to produce nitrogen-fixing root nodules as well as on the pollination services of wild bee populations to ensure both optimal seed set and out crossing rates (Sullivan & Angra, 2016).

Rhizobium symbiosis with legumes produces 50% of 175 million tons of total biological N2 fixation annually worldwide (Yadav & Verma, 2014). As Herridge et al. (2008) reported, the symbiosis of rhizobia form nodules on the roots or stems of the host plant and legume–rhizobia symbiosis accounts 60% of the total biological nitrogen fixation. Therefore, inoculation of legumes with efficient
rhizobia is believed to increase the yield and yield components of legumes while maintaining soil health. It is also supposed to be eco-friendly practices used for improvement of N fixation resulted in increased shoot growth, number of pods and grain yield of faba bean (Siczek & Lipiec, 2016).

In Ethiopia, there is a gradual increase in chemical fertilizer usage without letting any crop and animal manure residues in farm land. This action leads to the distraction of microorganisms in the soil, poor soil structure, low nutrient and water holding capacity of soil and reduce soil fauna–flora distribution. To minimize these environmental and soil damages caused by chemical fertilizers, the contribution of biological nitrogen fixation while increasing crop production plays indisputable role especially in maintaining soil health. In Ethiopia, the use of bacterial strain is not well advanced due to the lack of considerable effective strain selection, production and lack of appropriate inoculation of strains on the field.

From the 18 elements known to be essential for plant and microorganism growth, 8 are required in small quantities that they are called micronutrients. Among such micronutrients zinc and boron are known for their advantage on promoting of co-factors and involve in numbers of enzymes (Brady & Weil, 2008). These micronutrients are also involved in different functions of biological nitrogen fixation like, involved in nitrogen fixation (Bolanos et al., 1996), nodulation (Bonilla et al., 1980; Carpena et al., 2000), nodule protein synthesis (ENOD2) in nodule parenchyma cells and malfunction of oxygen diffusion barrier (Bonilla et al., 1997). Number of nodules and leghemoglobin content of nodules increased with increasing zinc application up to 7.5 μg/g soil. Faba bean plant dry-matter yield and N fixation increased with zinc up to 10 μg/g soil. Both nodulation and N fixation decreased at higher levels (Yadav & Shukla, 1983). The previous studies were not able also to fully evaluate the performance of different strains under balanced application of micro and macro plant nutrients. Whether the applications of micro-nutrients like boron and zinc is needed or not is also not well studied in the study district. The objective of this study is therefore to evaluate the effect of micronutrients and rhizobial inoculants on yield and yield components of faba bean.

2. Materials and methods

2.1. Description of the study area

2.1.1. Location

The study was carried out in Wereillu District, of South Wollo zone of the Amhara Region, Ethiopia. The experimental site is located at geographical coordinates of 10° 40’ 6" latitude and 39° 26’ 19" longitude at an altitude of 2,662 m above sea level (m.a.s.l) (Figure 1).

2.1.2. Climate

Wereillu is one of the highland districts of South Wollo. But it encompasses all kolo, woina dega, dega and Wurch climatic zones sharing 2%, 31%, 64% and 3% of the district, respectively. The rainfall pattern is predominantly bimodal, the short (belg) rains falling from mid-January to the end of May and the main or kiremt rains falling from June to mid-October. Based on the last 10 years (2008–2017) meteorological data obtained from Ethiopian Meteorological Service Agency, Kombolcha station, the district receives a mean annual rainfall of 873.0 mm with mean minimum and maximum temperatures of 10.3 and 21.6°C, respectively.

2.2. Geology of the study area

The geology of Eastern Amhara including the study area is covered by Cenozoic volcanic rocks with some sedimentary rocks (Kogal et al., 2012). The major formations are Ashang, Tarmaber-Megezez, Alojae, Aiba basalts and Ambo-Aradom formations covering 49%, 18%, 14%, 12% and 3%, respectively (Tefera et al., 1996). The geology of the study area belongs to Tarmaber-Megezez formations. Tarmaber-Megezez formations are transitional and alkaline basalts (Tefera et al.,
According to Food and Agriculture Organization FAO (1984), the soils of Wollo area have been developed almost exclusively on Trap Series volcano.

2.3. Farming system of Wereillu district

According to Wereillu district agriculture office (2011), Wereillu encompasses an area of 110,001 ha of land, from which, 57,383 ha, 11,354 ha, 8767 ha of land are allocated for crop production, grazing, natural forest and bush, respectively. The remaining unutilized and water bodies cover 8767 ha and 115.5 ha of land, respectively. Agriculture takes a greater part as a source of income for the farmers; however, it is unable to fully meet one of its most basic important functions i.e. the provision of food for the large and expanding population. Cereals as wheat (*Triticum aestivum* L.), teff (*Eragrostis teff* L.), pulse (*Fabciea* L.) and other oil and spices are the source of food and cash income. There are two-production seasons (*meher* and *belg*). *Meher* is the main season and *belg* producing kebeles are small in number. Out of the total land allocated for crop production, 50,116 ha is used for *meher* and 7267 ha for *belg* production season. The remaining 770 ha of land is cultivated by using a small-scale irrigation system in very few kebeles.
2.4. Experimental materials and methodology

2.4.1. Materials
The testing crop was Faba bean (Wolki variety), which is already adapted and still perform best in the study area.

2.4.2. Treatments
The experiment was done by using three selected Faba bean rhizobial strains based on their performance as recommended by Holeta micro biology laboratory.

(1) Control (without both micro nutrient and bacterial strain)
(2) Strain 1 (faba bean strain 17) (EAL 17)
(3) Strain 2 (faba bean strain 1018) (EAL 1018)
(4) Strain 3 (faba bean strain 1035) (EAL 1035)
(5) Zn (2 kg ha\(^{-1}\))
(6) B (1.5 kg ha\(^{-1}\))
(7) Strain 1 + Zn (faba bean strain 17 and Zn)
(8) Strain 2 + Zn (faba bean strain 1018 and Zn)
(9) Strain 3 + Zn (faba bean strain 1035 and Zn)
(10) Strain 1 + B (faba bean strain 17 and B)
(11) Strain 2 + B (faba bean strain 1018 and B)
(12) Strain 3 + B (faba bean strain 1035 and B)

Eighteen kilogram per hectare nitrogen from urea as starter fertilizer and 20 kg ha\(^{-1}\) phosphorus from triple super phosphate which is recommended for faba bean in the study district were applied at planting to all plots. Before the experimentation, the viability of inoculates was tested to know whether they are viable or not. Thus they were found that their number was above \(10^7\) per one carrier (125 g) and was taken as they are viable.

2.4.3. Experimental setup
Carrier peat based strains of faba bean EAL 17, EAL 1018 and EAL 1035 were obtained from Soil Microbiology Laboratory of Holeta Agricultural Research Center (HARC). First the faba bean seeds were immersed in to lukewarm water (36°C) so as to create anchoring environment for the stains. Then under umbrella shade, the bacterial strains were opened and mixed with the faba bean seeds in mixing container. Finally, seeds were allowed to air dry for a few minutes and were planted immediately after air drying. Three faba bean rhizobial strains were combined with zinc and boron in factorial randomized complete block design (RCBD) with three replications and hence there were 12 treatments in each replication. Nitrogen and phosphorus fertilizer were applied at planting by drilling with faba bean seeds in row. The plot size was 3.6-m length by 3-m width and spacing between plots and blocks was 1 m and 1 m, respectively. The spacing between plants and rows was also 0.1 m and 0.4 m, respectively. Before sowing each plot was prepared with the dimension of 0.8 m and 0.4-m length of beds and furrows, respectively. This dimension was selected based on the principle of broad bed and furrow in order to drain excessive water from the plot. Ridges were also made between each plot to reduce the movement of bacteria and to reduce the leaching/addition of nutrients from plot to plot or from external environment.

2.4.4. Agronomic and cultural practices
Zinc and boron were dissolved in water separately and were applied in foliar application by using knapsack sprayer during the vegetative stage of the faba bean (i.e., after the faba bean possess enough leaves to capture the sprayed Zn and B, and before flowering stage). Regarding the rates and the amount of water used to dissolve, the recommendation from Tisdal et al. (2003) and Troeh
and Thompson (1993) were taken. Thus boron was applied foliarly with the rate of 1.5 kg ha$^{-1}$ in 4 m$^3$ ha$^{-1}$ of water and zinc was also applied foliarly with the rate of 2 kg ha$^{-1}$ with 7 m$^3$ ha$^{-1}$ of water. The source of boron was borax (Na$_2$B$_4$O$_7$·10H$_2$O) which is the major source of B fertilizer (Troeh & Thompson, 1993). Similarly, the source of zinc was zinc sulfate (ZnSO$_4$) containing about 35% of zinc and is most commonly used zinc fertilizer (Tisdal et al., 2003).

2.5. Data collection procedures

2.5.1. Nodulation
Sampling for nodulation was performed by excavating the roots of five randomly selected plants in each plot at mid flowering stage of faba bean by destructive sampling from the border rows. A hoe was used to dig out the root surrounding soil and the spade was used to excavate the dug soil around 20 cm depth which is approximately the rooting depth of faba bean, and the radius of approximately 12 cm extending out from the central stem containing entire root system of the faba bean. The excavated soil was washed from the roots using washing bottle. Nodules from crown region and lateral roots subsequently were removed from the roots and collected in plastic bag for counting. The total number of nodules were counted and the effective and non-effective nodules were counted separately (visual observation) in those five sample plants by taking the intensity of the pink color into consideration (i.e. nodules which have pink color are considered as effective nodules) and the effective nodules undergo further analysis like for nodule number, nodule volume and nodule dry weight. The mean values of effective nodules from the five plants were recorded as number of nodules per plant.

The collected effective nodules were immersed in previously measured volume of water in a measuring cylinder. The volume of water displaced by the nodules was considered as nodule volume (cm$^3$). After the determination of the nodule volume, the collected nodules were dried in an oven for 65 h at 75°C to a constant weight to determine nodule dry weight per plant. The average from five plants was taken as nodule dry weight per plant.

2.5.2. Plant dry matter
Plant dry matter was determined at mid flowering stage of the crop from plants sampled for nodulation. After the nodules had been collected from roots, the plant samples were placed in a labeled perforated paper bags and oven-dried to a constant weight for 65 h at 75°C to determine the plant dry matter. The average dry weight of five plants was measured to determine dry weight per plant.

2.5.3. Plant height
Five plants from central rows were randomly selected for measuring their height at physiological maturity using measuring tape. The average height of five plants was taken from each plot and was considered as plant height.

2.5.4. Number of pods per plant
Five plants were randomly selected from harvestable rows of each plot. The pods were collected and counted separately from each plant and their average was taken and reported as number of pods per plant.

2.5.5. Number of grains per pod
After pods had been counted from each of the five randomly selected non-border plants, grains were separated from pods to get the number of grains per plant. For each plant the number of grains per pod was calculated by dividing the total number of grains per plant with the number of pods per plant. Finally, the mean value of five plants was taken as number of grains per pod.
2.5.6. Biomass yield, grain yield and hundred-grain weight

At physiological maturity, plants from four rows of net plot size 1.6-m × 3 m (4.8 m²) were manually harvested close to the ground surface. The harvested plant was sun-dried in an open air, and was weighed to determine above ground plant biomass yield. Grain yield of each plot was also determined after threshing. Finally, yield per plot was converted to grain yield per hectare basis. The grain yield was adjusted to 10% moisture content. The weight of random sample of 100 grains in gram was reported as 100 grain-weight.

2.5.7. Symbiotic effectiveness

The symbiotic effectiveness of faba bean strains was calculated based on the formula set by Mulongoy (2004) as:

\[
SE = \frac{\text{Inoculated Plant DM}}{\text{Uninoculated Plant DM}} \times 100\%
\]

Where, DM = dry matter in gram, SE = Symbiotic effectiveness (%)

The rate of nitrogen-fixing effectiveness is evaluated as highly effective if the value is greater than 80%, effective if it was between 50% and 80%, less effective if the values are between 35% and 49% and infective when the values were below 35%.

2.5.8. Soil sample collection and processing

Disturbed composite surface soil samples (0–30 cm depth) were collected from experimental area by zigzag sampling method before planting for determination of physico-chemical properties of the soil. Similarly, after harvesting one soil sample from each plot was taken for determination of physico-chemical properties of the soil. Then after, the collected samples were air dried at room temperature and were ground to pass through a 2 mm sieve for most parameters and through 0.5 mm sieve to determine total nitrogen and organic carbon. Using core sampler, one soil sample was taken from the representative soil of the study site to determine bulk density of the soil.

2.5.9. Soil physico-chemical analysis

Soil samples were analyzed for determination of bulk density, particle size distribution, pH, organic carbon, cation exchange capacity, exchangeable bases (K, Ca and Mg), base saturation and total nitrogen, from the representative bulk soil sample at planting. Available P was taken two weeks after incubation clearly to know the amount of P which was not fixed with the soil exchange sites like with aluminum. Total N was analyzed for each treatment in all replications following the standard procedure after harvest.

Soil bulk density was estimated by core method up to 30 cm depth and was calculated as \( \rho_b = \frac{M_s}{V_t} \)

Where \( \rho_b \) is the bulk density of the soil (g/cm³), \( M_s = \) Mass of the dry soil (g) and \( V_t = \) total volume of the soil sample (cm³) (Blake, 1965). Soil particle size distribution was determined by hydrometer method (Bouyoucous, 1951). Soil pH was determined by potentiometric method at soil:water ratio of 1:2.5 (Van Reeuwijk, 1992). Cation exchange capacity was determined by 1 M ammonium acetate method at pH 7 (Chapman, 1965), whereas organic carbon was determined by the dichromate oxidation method (Walkley & Black, 1934) and total nitrogen by the micro Kjeldhal method (Jackson, 1985), available P was analyzed by Olsen method (Olsen et al., 1954). Ca⁺⁺ and Mg⁺⁺ values were analyzed from Atomic Absorption Spectrophotometer reading while K⁺ was determined using flame photometer.

2.5.10. Partial budget analysis

The farm gate prices of 12.00 and 1.00 Ethiopian Birr (ETB) were used for faba bean grain and straw price kg⁻¹. One hundred fifty and 160 (ETB) ha⁻¹ for EAL 17 and EAL 1018 price respectively were used. Eighteen and 16 ETB were used for Zn and B price kg⁻¹. Having such prices, the partial
budget analysis was done following the CIMMYT procedure (CIMMYT (International Maize and Wheat Improvement Center), 1988). The mean grain yields used in the partial budget analysis were adjusted to 90 % of the actual yield.

### 2.6. Statistical analysis

The data measured were subjected to a statistical analysis using SAS software package 2003, version 9.1 for analysis of variance. The DMRT mean comparison method at 5% level of significance was used to separate the significant treatment means.

### 3. Results and discussion

#### 3.1. Soil analysis results

According to Jones (2003), Murphy (1968) and Tadese (1991), the pH is slightly acidic range (Table 1) and such a pH range is conducive for biological nitrogen fixation (Jordan, 1984). The soil analysis results (Table 1) revealed that the organic matter and total nitrogen were in a range of low and medium respectively (Debele, 1980; Murphy, 1968; Tadese, 1991). Thus in the study area, the application of nitrogen-containing fertilizer or inoculation with effective strains are mandatory to refill the remaining nitrogen that the soil couldn’t supply to the crop. The available phosphorus was found low (Olsen et al., 1954) and moderate (Cottenie, 1980) and to fill the nutrient P requirement of the crop, the recommended phosphorus fertilizer was added. According to FAO (2006) CEC, exchangeable Ca, Mg and K were in very high range and no need to apply any one of them in the study site. The bulk density of the soil laid in a range of moderate according to (Hazelton & Murphy, 2007) and nitrogen fixation is not difficult to take place under such bulk density. In Table 1 it is also seen that boron and zinc are deficient according to Jonse (2003) and implies that the area needs additional boron and zinc to boost crop production.

| Soil Properties                  | Values |
|----------------------------------|--------|
| pH (H₂O)                         | 6.4    |
| Organic matter (OM) (%)          | 1.57   |
| Total Nitrogen (TN) (%)          | 0.21   |
| Available P (mg kg⁻¹ soil)       | 8.7    |
| Boron (ppm)                      | 0.7    |
| Zinc (ppm)                       | 0.4    |
| Exchangeable Ca (cmolc kg⁻¹)     | 38     |
| Exchangeable Mg (cmolc kg⁻¹)     | 10.2   |
| Exchangeable K (cmolc kg⁻¹)      | 0.8    |
| Cation exchange capacity (CEC)   | 58     |
| Bulk density (gm cm⁻³)           | 1.14   |
| Sand%                            | 21.5   |
| Silt%                            | 23.5   |
| Clay%                            | 55     |
| Textural class                   | Clay   |

As it is indicated in Table 2, after harvesting total nitrogen across all treatments fall under the same nitrogen ratting range (medium) (Debele, 1980; Tadese, 1991). But numerically, the nitrogen content of the soil after harvesting is better than the nitrogen content of the soil before planting. This may be due to the addition of nitrogen into the soil as nitrogen passes into the soil itself,
either by excretion or more probably by sloughing off of the roots and especially of the nodules (Brady & Weil, 2008).

3.2. Effect of strains and micronutrients on nodulation parameters

3.2.1. Nodule number

As it indicated in Table 3, both the main effect and the combined effects of strains and micro nutrients significantly affected nodule number plant\(^{-1}\). Thus the combined effect of EAL 1018 and boron gave higher (81.2) nodule number plant\(^{-1}\) compared to all other treatments. The nodule number obtained from the combined effect of (0, uninoculated), (Zn, uninoculated) and (Zn, EAL 17) shows statistically ($P \leq 0.05$) lower nodule number compared to other treatments. The result revealed that boron alone and in combination with strains have positive effect on nodule number. The combined effect of zinc with EAL 1018 and EAL 1035 was also found promising nodule number in the study area. The result is in line with the findings of Bolanos et al. (1996), who stated that the application of boron could increase nodulation by developing infection threads associated with covalently bounded hydroxyproline/proline rich proteins. Likewise, Ahmad et al. (2009) reported that the absence of boron in culture medium resulted in lower number of nodules and alterations in nodule development. In addition, Alemayehu Workalemahu (2009) and Nagwa et al. (2012) reported that inoculation of strain to faba bean significantly increased nodule number. Yohannes

| Table 2. Soil analysis result of total nitrogen after harvesting |
|---------------------------------------------------------------|
| **Treatments** | **Total nitrogen (%)** |
| Control | 0.23 |
| EAL 17 | 0.25 |
| EAL 1018 | 0.25 |
| EAL 1035 | 0.25 |
| Zn | 0.22 |
| B | 0.23 |
| EAL 17 + ZN | 0.23 |
| EAL 1018 + ZN | 0.25 |
| EAL 1035 + ZN | 0.24 |
| EAL 17 + B | 0.24 |
| EAL 1018 + B | 0.25 |
| EAL 1035 + B | 0.26 |

EAL = Ethiopian agricultural legumes.

| Table 3. Combined effect of rhizobial inoculation and micronutrients on effective nodule (nodule plant\(^{-1}\)) |
|---------------------------------------------------------------|
| **Faba bean Strains** | **Micro nutrient (Zn and B)** | **Zn** | **B** |
| Un-inoculated | 49.1\(^{a}\) | 57.6\(^{cde}\) | 71.9\(^{abc}\) |
| EAL 17 | 63.8\(^{bcd}\) | 54.3\(^{bc}\) | 64.8\(^{bcd}\) |
| EAL 1018 | 69.4\(^{abc}\) | 73.3\(^{ab}\) | 81.2\(^{a}\) |
| EAL 1035 | 64.8\(^{bcd}\) | 69.0\(^{bc}\) | 69.7\(^{bcd}\) |

| LSD(0.05) | CV = 10.15 | SE (\(\pm\)) = 6.93 |

Where EAL = Ethiopian Agricultural Legumes, CV = Coefficient of variation, LSD = Least significant difference, SE = standard error. Means followed by the same letter with in a column are non-significantly different at $P = 0.05$ (Duncan's Multiple Range Test).
Desta et al. (2015) also reported that both inoculation alone and/or the combined effect of strains with zinc significantly increase nodule number plant$^{-1}$. But in contrary this work, Endalkachew Fekadu et al. (2018) reported that faba bean plants that were not inoculated with rhizobium but treated with FYM, lime and P, formed nodules with a higher number as compared with plants inoculated and received the same treatment.

### 3.2.2. Nodule volume

In Table 4, nodule volume was significantly ($P \leq 0.05$) affected by strain and micro nutrient and their interactions. The highest nodule volume (1.83 cm$^3$ plant$^{-1}$) was recorded from the combination of EAL 1018 and boron. The main effect in terms of nodule volume was also significant ($P \leq 0.05$). As Rodelas et al. (1999) described, while using strains nodule dry weight accumulation increased, this work also showed that inoculation of strains increased nodule volume. Thus the individual effects of EAL 1018, boron and EAL 1035 gave better nodule volume as compared to the uninoculated treatment and zinc alone.

### 3.3. Nodule dry weight

Nodule dry weight is affected by the strains, micronutrient and their interactions as indicated in Table 5. The highest nodule dry weight (0.75 g plant$^{-1}$) was recorded from the combined effect of EAL 1018 and boron by the combined effect of EAL 1018 with zinc. But, EAL 1018 with Zn is statistically at par with EAL 1018 alone, EAL 1015 alone, EAL 1015 with boron and EAL 1035 with zinc (Table 5). The control treatment is not also statistically different with all treatments except the effect of EAL 1018 alone and in combination with micronutrients, and the combined effect of EAL1035 and boron. This result is in agreement with the result reported by EL-Wakeil and EL-Sebai (2007) who stated that application of rhizobium strains brought significantly higher nodule dry

### Table 4. Combined effect of rhizobial inoculation and micronutrients on faba bean nodule volume per plant (cm$^3$ plant$^{-1}$)

| Faba bean strains | Micro nutrient (Zn and B) | 0 | Zn | B |
|-------------------|---------------------------|---|----|---|
| Un-inoculated     |                           | 0.63$^d$ | 0.87$^cd$ | 1.13$^b$ |
| EAL 17            |                           | 1.0$^{bc}$ | 1.07$^{bc}$ | 1.20$^a$ |
| EAL 1018          |                           | 1.13$^b$ | 1.17$^b$ | 1.83$^b$ |
| EAL 1035          |                           | 1.07$^{bc}$ | 1.13$^b$ | 1.17$^b$ |
| LSD(0.05) = 0.26  | CV = 10.01                | SE (±) = 0.13 |

Where EAL = Ethiopian Agricultural Legumes, CV = Coefficient of variation, LSD = Least significant difference, SE = standard error. Means followed by the same letter with in a column are non-significantly different at $P = 0.05$ (Duncan’s Multiple Range Test).

### Table 5. Combined effect of rhizobium inoculation and micronutrients on nodule dry weight (g plant$^{-1}$)

| Faba bean strains | Micro nutrient (Zn and B) | 0 | Zn | B |
|-------------------|---------------------------|---|----|---|
| Un-inoculated     |                           | 0.24$^a$ | 0.28$^{cd}$ | 0.31$^{cd}$ |
| EAL 17            |                           | 0.31$^{cd}$ | 0.30$^{cd}$ | 0.33$^{bcd}$ |
| EAL 1018          |                           | 0.39$^{bc}$ | 0.42$^b$ | 0.75$^a$ |
| EAL 1035          |                           | 0.33$^{bcd}$ | 0.32$^{bc}$ | 0.35$^{bc}$ |
| LSD(0.05) = 0.104 | CV = 15.67                | SE (±) = 0.05 |

Where EAL = Ethiopian Agricultural Legumes, CV = Coefficient of variation, LSD = Least significant difference, SE = standard error. Means followed by the same letter with in a column are non-significantly different at $P = 0.05$ (Duncan’s Multiple Range Test).
weight and nodule number. Rodelas et al. (1999) also reported that nodule dry weight was significantly increased (38–53%) by co-inoculation with strains.

3.4. Symbiotic effectiveness

The result of the symbiotic effectiveness (Table 6) revealed that application of all the three faba bean strains in the study area were highly effective based on the formula that was developed by Mulongoy (2004). This implies that inoculation of S1, S2 and S3 creates high symbiotic association with the host plant, faba bean than the existing strains in the study district. This result supported by previous findings of Gebremariam and Assefa (2018) in such a way that the symbiotic effectiveness of isolates of faba bean strains was highly effective.

3.5. Effect strains and micronutrients on agronomic parameters

3.5.1. Plant height

Micronutrients and the combined effect of strains and micro nutrients didn’t affect plant height. While inoculation significantly affected \( P \leq 0.05 \) plant height, as a result, EAL 1018 alone gave significantly higher (63.46 cm) plant height compared to the other treatments (Table 7). This could be attributed to inoculation of strains to faba bean supplying additional nitrogen through symbiotic nitrogen fixation and lead to increased plant height. This result agreed with the result of Farfour

Table 6. The effect inoculation of faba bean on symbiosis effectiveness of strains at study site

| Treatment       | Plant dry weight | Percentage (%) | SE | Effectiveness |
|-----------------|------------------|----------------|----|---------------|
| Un-inoculated   | 8.66 \(^{c}\)    | -              | -  | -             |
| EAL 17          | 9.54 \(^{bc}\)   | 110.2          | HE |               |
| EAL 1018        | 10.61 \(^{a}\)   | 122.5          | HE |               |
| EAL 1035        | 9.68 \(^{b}\)    | 111.8          | HE |               |

Where EAL = Ethiopian Agricultural Legumes, SE = Symbiosis effectiveness and HE = Highly effective.

Table 7. Main effects of rhizobium inoculation and micronutrients on plant height (cm), plant dry weight (g plant\(^{-1}\)), number of pod plant\(^{-1}\), number of seed pod\(^{-1}\) and 100 seed weight (g) of faba bean

| Factors          | Plant height | Number of Pod plant\(^{-1}\) | Plant dry weight | Number of Seed pod\(^{-1}\) | 100 seed Weight |
|------------------|--------------|------------------------------|------------------|-----------------------------|-----------------|
| Strain           |              |                              |                  |                             |                 |
| Uninoculated     | 57.00 \(^{a}\) | 20.52 \(^{ab}\)              | 8.66 \(^{c}\)    | 2.24 \(^{a}\)              | 54.62 \(^{c}\)  |
| EAL 17           | 58.80 \(^{a}\) | 23.13 \(^{ab}\)              | 9.54 \(^{bc}\)   | 2.24 \(^{a}\)              | 55.80 \(^{bc}\) |
| EAL 1018         | 63.46 \(^{a}\) | 26.88 \(^{a}\)               | 10.61 \(^{a}\)   | 2.46 \(^{a}\)              | 58.80 \(^{a}\)  |
| EAL 1035         | 58.75 \(^{b}\) | 23.44 \(^{ab}\)              | 9.68 \(^{b}\)    | 3.11 \(^{a}\)              | 56.96 \(^{b}\)  |
| LSD(0.05)        | 4.24         | 3.04                         | 0.79             | Ns                          | 2.06            |
| SE(\(\pm\))     | 2.04         | 1.46                         | 0.38             | 0.08                        | 0.99            |
| MN               |              |                              |                  |                             |                 |
| O                | 57.26 \(^{a}\) | 23.26 \(^{a}\)              | 9.36 \(^{c}\)    | 2.26 \(^{a}\)              | 54.72 \(^{c}\)  |
| Zn               | 59.76 \(^{a}\) | 22.13 \(^{bc}\)              | 9.54 \(^{a}\)    | 2.31 \(^{a}\)              | 58.08 \(^{a}\)  |
| B                | 61.48 \(^{a}\) | 25.03 \(^{bc}\)              | 9.96 \(^{a}\)    | 2.36 \(^{a}\)              | 56.83 \(^{b}\)  |
| LSD(0.05)        | Ns           | Ns                           | Ns               | Ns                          | 1.79            |
| CV               | 8.92         | 16.16                        | 9.27             | 8.93                        | 2.18            |
| SE(\(\pm\))     | 1.77         | 1.26                         | 0.33             | 0.07                        | 0.86            |

Where EAL = Ethiopian Agricultural Legumes, MN = Micro nutrient, CV = Coefficient of variation, Ns = None—significant, * = Significant \( P \leq 0.05 \) but > 0.01 and ** = Highly significant \( P \leq 0.01 \). LSD = Least significant difference SE = Standard error. Means followed by the same letter within a column are not significantly different at \( P = 0.05 \) (Duncan’s Multiple Range Test).
(2013) which affirmed that application of rhizobium stains could increase plant height of faba bean. Similarly, EL-Azeem et al. (2007) reported that application of bacterial strain significantly increase plant height compared to the uninoculated treatments.

3.6. Plant dry weight
The data on plant dry weight (Table 7) showed that the interaction effect (strain × micronutrient) was not significant ($P \leq 0.05$). But, inoculation of faba bean with EAL 1018 alone gave statistically significant plant dry weight (10.61 g) followed by EAL 1035 and EAL 17 with the lowest mean dry weight from the un-inoculated treatment. Our result is supported by Farfour (2013) who reported that, inoculation of faba bean with rhizobial strain caused noticeable increase in plant length, fresh weight and dry weight. Sameh et al. (2017) also reported that inoculation of strains increases plants dry weight. In this work, micro nutrient didn’t impose significant difference in plant dry weight compared with the control treatment. Our result disagreed with that of Salem et al. (2014) who stated that application of micronutrients could increase the plant dry weight of faba bean. Having in mind all this arguments, this work showed that only inoculation of strains rather than application of micronutrients could cause an increased plant dry weight. This could be attributed to meanwhile micronutrients were applied at around vegetative stage, and this may not have contributed for the increased plant dry weight in short time frame (i.e., plant dry weight was taken at flowering stage) rather application of micronutrients could influence seed weight and other yield and yield-related parameters.

3.6.1. Number of pod plant$^{-1}$
The number of pods plant$^{-1}$ was affected only by the inoculation. Application of micro nutrients didn’t impose significant number of pods/plant on faba bean. Accordingly, inoculation of faba bean with EAL 1018 brought considerably higher number of pods plant$^{-1}$ compared to all other strains and statistically significant ($P \leq 0.05$) difference compared to the un-inoculated treatment (Table 7). In view of strains, the inoculation of all the three faba bean strains (EAL 17, EAL 1018 and EAL 1035) on the study area impose statistically significant ($P \leq 0.05$) number of pods plant$^{-1}$ compared to the un-inoculated treatment. The research conducted by Yohannes Desta et al. (2015) reported that rhizobial strain alone could significantly increase number of pods plant$^{-1}$ as in support with this work but in contrary it disagreed with this work in the way that the combined effect of zinc and strain increase pod plant$^{-1}$.

3.6.2. Number of seed pod$^{-1}$
The main effects of both strains and micronutrients and their interaction didn’t impose significant ($P > 0.05$) number of seed difference on faba bean. But, still EAL 1018 gave relatively numerically higher (2.64) number of seeds pod$^{-1}$ as shown in Table 4.7 compared to other treatments. In consent with this result the finding by Zerihun and Abera (2014) indicated that inoculation of rhizobial strain did not impose significant difference over the un-inoculated one. But it disagreed with the finding of Salem et al. (2014), who stated that application of micro nutrients significantly increased number of seeds pod$^{-1}$.

3.6.3. 100 seed weight
The result clearly indicated a marked difference in terms of 100 seed weight. It was observed that both strains and micronutrients independently significantly affected ($P \leq 0.05$) 100 seeds weight (Table 7). EAL 1018 gave statistically significant ($P \leq 0.05$) 100 seed weight (58.8 gram) over the control as well as the other strains. This result was also in line with the work of EL-Wakeil and EL-Sebai (2007) who reported that inoculation of faba bean with rhizobial strain alone could increase 100 seed weight. Inoculation of faba bean with EAL 17 and EAL 1035 are also made 100 seed weight difference over the control treatment though they gave statistically lower seed weight compared to EAL 1018. This result was in contrary with the finding of Bolland et al. (2000) which stated that inoculation of faba bean strains didn’t brought significant seed weight difference on faba bean. Alike the inoculants, application of micro nutrients significantly ($P \leq 0.05$) affected 100 seed weight. Thus, application of zinc alone gave the maximum (58.08 g) 100 seed weight.
contest with this study, other findings revealed that foliar spraying of zinc enhanced soybean yield, and number and weight of seeds plant\(^{-1}\) (Kobraee et al., 2011). Similarly, Mohamed et al. (2017) stated that application of micro nutrients like zinc significantly affected 100 seed weight of faba bean. Gizawy and Mehasen (2009) also confirmed that application of zinc significantly increased 100 seed weight of faba bean compared to the control treatment. The weight difference gained from this work could be attributed the effect of micronutrients on the quality of seed.

### 3.6.4. Grain yield

As the analysis of variance indicated in Table 8, the main effect of strains and micronutrients as well as their interaction significantly \((P \leq 0.05)\) affected faba bean grain yield. As it is shown in Table 9, the maximum grain yield \((4,820 \text{ kg ha}^{-1})\) was obtained from the combined application of EAL 1018 and boron, whereas the lowest grain yield was obtained from the control. Application of zinc and EAL 17 alone and their combination as well as the combined effect of faba bean EAL 1035 and zinc did not considerably increase faba bean grain yield. The combination of EAL 1018 with boron as well as their main effect highly affected faba bean grain yield as compared to all other combinations and the main effects. The result revealed that application of the combination of faba bean strain, EAL 1018 and boron gave a grain yield advantage from 13.9% to 65.9% over the second promising treatment EAL 1018 alone and the control treatment, respectively. This work is in agreement with the work of Rugheim and Abdelgani (2012) who indicated that co inoculation of rhizobial strains significantly increased faba bean grain yield. Different authors testified that application of micro nutrients increased faba bean grain yield. In this respect, Mady (2009) reported that foliar application of zinc significantly increased grain yield of faba bean as compared

| Table 8. Analysis of variance for grain yield analysis |
|------------------------------------------------------|
| Source of variation | DF | SS         | MS         | F value | Pr > F |
|----------------------|----|------------|------------|---------|--------|
| MN                   | 2  | 2190219.396| 1095109.698| 26.29   | 0001** |
| Rep                  | 2  | 477831.733 | 238915.866 | 5.74    | 0.0179*|
| Strain               | 3  | 3515437.944| 1171812.648| 28.13   | 0.0001**|
| Rep*MN               | 4  | 42708.099  | 10677.025  | 0.26    | 0.902**|
| Strain*MN            | 6  | 925426.022 | 154237.670 | 3.70    | 0.0256*|
| Rep*strain           | 6  | 80147.255  | 13357.876  | 0.32    | 0.9137**|
| R-Square             |    | 0.93348    |
| Root MSE             |    | 204.0971   |
| Mean                 |    | 3405.410   |

Where MN = Micro nutrient, Rep = Replication, DF = Degree of freedom, SS = Sum squares, MS = Mean squares, ns = None—significant, * = Significant \((P \leq 0.05\) but > 0.01), ** = highly significant \((P \leq 0.01)\) and MSE = Mean square of error.

| Table 9. Interaction effect of rhizobium inoculation and micronutrients on grain yield (kg ha\(^{-1}\)) |
|---------------------------------------------------------|
| Faba bean strains | Micro nutrient (Zn and B) |
|-------------------|--------------------------|
|                   | 0   | Zn  | B    |
| Un-inoculated     | 2614.5\(^{7}\) | 3230.8\(^{dc}\) | 3619.1\(^{bc}\) |
| EAL 17            | 2995.1\(^{a}\) | 2985.4\(^{a}\) | 3522.8\(^{bc}\) |
| EAL 1018          | 3806.5\(^{d}\) | 3631.3\(^{bc}\) | 4338.2\(^{a}\) |
| EAL 1035          | 3411.7\(^{cd}\) | 3177.4\(^{de}\) | 3532.2\(^{bc}\) |
| LSD(0.05) = 284.87| CV  = 5.99| SE (±) = 137.36 |

Where EAL = Ethiopian Agricultural Legumes, CV = Coefficient of variation, LSD = Least significant difference, SE = standard error. Means followed by the same letter with in a column are non-significantly different at \(P = 0.05\) (Duncan’s Multiple Range Test).
to the control. Yazied and Mady (2012) also stated that foliar application of boron significantly increased faba bean grain yield. According to Salem et al. (2014), zinc and boron significantly increased faba bean yield compared with control. The finding of Yohannes Desta et al. (2015) was also in line with this finding in a way that application of effective strain alone and/or in combination with zinc significantly increased grain yield of faba bean. According to Sameh et al. (2017), application of effective strains increases grain yield of faba bean up to 44–47%. Unlike the above mentioned findings, Zerihun and Abera (2014) showed that rhizobium inoculation didn’t increase faba bean grain yield over the control. The increments in grain yield due to inoculation of strains indicated that the soils nitrogen is a limiting factor and the existing rhizobium bacteria may not be

| Table 10. Pearson’s correlation coefficient among yield and yield related parameters under strain and micronutrient treatments |
| GY | BIWt | NDWt | NN | NP | NS | NV | PDWt. | PH | Sedwt. |
|---|---|---|---|---|---|---|---|---|---|
| GY | p-value | *** | | | | | | | |
| BIWt. | 0.9569 | | | | | | | | |
| p | *** | *** | | | | | | | |
| NDWt. | 0.7138 | 0.74 | | | | | | | |
| p | *** | *** | *** | | | | | | |
| NN | 0.694 | 0.689 | 0.5893 | | | | | | |
| p | *** | *** | *** | *** | | | | | |
| NP | 0.6113 | 0.6671 | 0.5741 | 0.5708 | | | | | |
| p | *** | *** | *** | *** | *** | | | | |
| NS | 0.595 | 0.577 | 0.4756 | 0.4891 | 0.4533 | | | | |
| p | *** | *** | *** | *** | *** | *** | | | |
| NV | 0.7144 | 0.7505 | 0.8518 | 0.6806 | 0.5869 | 0.3517 | | | |
| p | *** | *** | *** | *** | *** | *** | *** | | |
| PDWt. | 0.6042 | 0.6417 | 0.6066 | 0.4283 | 0.453 | 0.5718 | 0.6022 | | |
| p | *** | *** | *** | *** | *** | *** | *** | *** | |
| PH | 0.5482 | 0.5463 | 0.5847 | 0.4676 | 0.6111 | 0.5267 | 0.5945 | 0.47 | |
| p | *** | *** | *** | *** | *** | *** | *** | *** | |
| Sedwt. | 0.5449 | 0.6034 | 0.401 | 0.2797 | 0.299 | 0.212 | 0.3419 | 0.4228 | 0.4276 |
| p | *** | *** | *** | *** | *** | *** | *** | *** | *** |

Where GY = Grain yield, BIWt. = Biomass weight, PH = Plant height, NN = Nodule number, NV = Nodule volume, NDWt. = nodule dry weight, PDWt. = Plant dry weight, NP = Number of pod, NS = Number of seed and SDWt. = 100 g seed weight, ns = No significant correlation, * = Significant correlation (P ≤ 0.05 but > 0.01) and ** = highly significant correlation (P ≤ 0.01).

| Table 11. Interaction effect of rhizobium inoculation and micro nutrients on biomass weight (kg ha⁻¹) |
| Faba bean strains | Micro nutrient (Zn and B) |
|---|---|---|
| Uninoculated | 7326.6 | 8839.9 | 9862.4 |
| EAL 17 | 8328.7 | 8898.9 | 9524.5 |
| EAL 1018 | 10,277.3 | 10,144.5 | 12,360.0 |
| EAL 1035 | 9360.7 | 9195.1 | 9655.0 |
| LSD(0.05) = 1034 | CV = 7.59 | SE (±) = 498.57 |

Where EAL = Ethiopian Agricultural Legumes, CV = Coefficient of variation, LSD = Least significant difference, SE = standard error. Means followed by the same letter with in a column are non-significantly different at P = 0.05 (Duncan’s Multiple Range Test).
capable to supply nitrogen through biological nitrogen fixation. Thus the grain yield could be strongly improved by means of inoculation or fertilization.

The correlation analysis (Table 10) indicated that there was highly positively significant ($P \leq 0.01$) correlation grain yield with yield related parameters. The correlation coefficient value of all parameters in relation to the grain yield of faba bean was almost more than 0.5 especially there was a strong correlation ($r = 0.96$) between the grain yield and the biomass yield of faba bean.

### 3.6.5. Biomass yield

Both the main effect and the interaction of strains and micronutrients significantly ($P \leq 0.05$) affected biomass yield. From Table 11, it is clearly indicated that the combined effect of EAL 1018 and boron gave maximum (12,360 kg ha$^{-1}$) biomass yield while the lowest biomass yield was obtained from the control followed by the main effect of EAL 17 alone or in combination with zinc and boron. This result is in agreement with the work of EL-Azeem et al. (2007) in a way that, inoculation of bacterial strain cause a significant biomass yield on faba bean. Gizawy and Mehasen (2009) who stated that application of zinc significantly increases biomass yield and Salem et al. (2014) also testified that Zinc and Boron significantly affects biomass yield of faba bean. The biomass yield difference obtained from the inoculation of faba bean strains could be from the additional supply of nitrogen through the remarkable biological nitrogen fixation by the inoculated strains.

### 3.7. Partial budget analysis

The partial budget analysis for marginal rate of return in Table 12 showed that the strains and the combination of EAL 1018 and B gave acceptable marginal rate of return (i.e., MRR greater than 100%). According to CIMMYT (International Maize and Wheat Improvement Center) (1988), when there are two and more treatments with MRR greater than 100%, the treatment with greater net benefit should be selected for recommendation. Therefore, application of the combination of EAL 1018 and boron brought the maximum net benefit (53,690.4 Ethiopian Birr) per hectare while possessing MRR of greater than 100% and thus it is economically feasible for Wereillu district.

### Table 12. Partial budget analysis of the variable costs on mean grain and straw yields (biomass yield—grain yield) of faba bean in the study district for strains and micronutrients

| Treatments    | AGY  | GYP  | ASY  | SYP  | TR   | SC   | FC   | LC   | TVC  | NB   | DA   | MRR  |
|---------------|------|------|------|------|------|------|------|------|------|------|------|------|
| Control       | 2353.1 | 12   | 4712.5 | 1    | 32949.1 | 0    | 0    | 0    | 32949.1 |      |
| EAL 17        | 2695.6 | 12   | 5333.9 | 1    | 37680.9 | 150   | 0    | 400   | 550   | 37131 | 760.3 |
| EAL 1018      | 3425.9 | 12   | 6470.5 | 1    | 47580.7 | 160   | 0    | 400   | 560   | 47020.7 | 98897 |
| EAL 1035      | 3070.5 | 12   | 5949.3 | 1    | 42795.6 | 160   | 0    | 400   | 560   | 42325.7 |      |
| B             | 3257.2 | 12   | 6242.9 | 1    | 45329.2 | 0    | 24   | 600   | 624   | 44705.2 |      |
| Zn            | 2907.7 | 12   | 5609.2 | 1    | 40501.8 | 0    | 36   | 600   | 636   | 3965.8 |      |
| EAL 17 + B    | 3170.5 | 12   | 6001.2 | 1    | 44047.4 | 160   | 24   | 1000  | 1174  | 42873.4 |      |
| EAL 1018 + B  | 3904.4 | 12   | 8021.8 | 1    | 54874.4 | 160   | 24   | 1000  | 1184  | 53690.4 | 1068.9 |
| EAL 1035 + B  | 3179  | 12   | 6122.8 | 1    | 44270.6 | 160   | 24   | 1000  | 1184  | 43086.6 |      |
| EAL 17 + ZN   | 2686.9 | 12   | 5913.6 | 1    | 38155.9 | 150   | 36   | 1000  | 1186  | 36969.9 |      |
| EAL 1018 + ZN | 3268.2 | 12   | 6153.7 | 1    | 45731.5 | 160   | 36   | 1000  | 1196  | 44535.7 |      |
| EAL 1035 + ZN | 2859.7 | 12   | 6967.6 | 1    | 41283.5 | 160   | 36   | 1000  | 1196  | 40087.5 |      |

All costs are expressed in Ethiopian birr (ETB). AGY = Adjusted grain yield (kg ha$^{-1}$), GYC = Grain yield price kg$^{-1}$ (ETB), ASY = Adjusted straw yield (kg ha$^{-1}$), SYP = Straw yield price kg$^{-1}$ (ETB), TR = Total revenue (ETB), SC = Strain cost kg$^{-1}$ (ETB), FC = fertilizer cost kg$^{-1}$ (ETB), LC = Labor cost (ETB), TVC = Total variable cost, NB = Net benefit (ETB), DA = Dominance analysis, MRR = Marginal rate of return.
4. Conclusions and recommendations

4.1. Conclusions

Biological nitrogen fixation through microbial processes ensures the greatest quantitative impact on the nitrogen cycle and has tremendous potential for the contribution of fixed N in the soil. Accordingly, this research work was conducted to examine this remarkable activity of rhizobium strains under field conditions. As indicated from the result, micronutrients and rhizobial inoculants significantly \((P \leq 0.05)\) affected yield and yield components of faba bean except for a number of seeds pod\(^{-1}\). The combined effect of micronutrients and rhizobium strains brought statistically significant difference on nodulation of faba bean like nodule number, nodule volume, and nodule dry weight of faba bean. Similarly, the interaction effect also imposes considerably significant biomass and grain yield difference over the control treatment. The main effect of micronutrients significantly affects only the weight of 100 seeds of faba beans. On the other hand, inoculation of faba bean with strains alone imposes statistically significant difference in plant height, plant dry weight and number of pods plant\(^{-1}\) as compared to uninoculated treatments.

4.2. Recommendations

From the result, inoculation of faba bean strain, EAL-1018 with boron brought biologically higher significant yield and economically maximum net benefit compared to other treatments. Thus the combination of EAL-1018 could be recommended in the study district and similar agro ecological zones.

The concentration of zinc above 300 ppm (Prasad et al., 1999) and concentration of boron more than 250–300 ppm could be toxic for most crops. So seasonal soil analysis on boron and zinc concentration should be conducted to avoid toxicity of these nutrients through gradual accumulation in the soil.

Bolland, M. D. A., Siddique, K. H. M., & Brennan, R. F. (2000). Grain yield responses of faba bean (Vicia faba L.) to applications of fertiliser phosphorus and zinc. *Australian Journal of Experimental Agriculture*, 40(6), 849. 10.1071/EA99164

Bonilla, I., Cadahía, O., Hernando, V., & Cadahía, C. (1980). Effects of boron on nitrogen metabolism and sugar levels of sugar beet. *Plant and Soil*, 57(1), 3–9. https://doi.org/10.1007/BF02139636

Bollia, I., Mergold-Villasen, C., Campos, M. E., Sanchez, N., Perez, H., Lopez, L., Castrejon, L., Sanchez, F., & Cassab, G. I. (1997). The aberrant cell walls of boron-deficient bean root nodules have no covalently bound hydroxyproline-/proline-rich proteins. *Plant Physiology*, 115(4), 1329–1340. https://doi.org/10.1104/pp.115.4.1329

Bouyoucos, C. J. (1951). A reclamation of the hydrometer method for making chemical analysis of soils. *Agronomy Journal*, 43(9), 434–438. 10.2134/ agronj1951.0002196200430009005x

Brady, N. C., & Weil, R. R. (2008). *The nature and properties of soils* (14th ed.). Pearson Prentice hall.

Carpena, R. O., Esteban, E., Sarro, M. J., Peñalosa, J., Gárate, A., Lucena, J. J., & Zornoza, P. (2000). Boron and calcium distribution in nitrogen-fixing pea plants. *Plant Science*, 151(2), 163–170. https://doi.org/10.1016/S0168-9452(99)00210-1

Chapman, H. D. (1965). Cation exchange capacity. In C. A. Black (Ed.), *Ensminger* (pp. 891–901).

CIMMYT (International Maize and Wheat Improvement Center). (1988). *From agronomic data to farmers recommendations: An economic training manual* (Revised ed.).

Cottenie, A. (1980). Boron physiology, tissue analysis and boron in soil. In *Boron in crop plants* (pp. 849–901). American Society of Agronomy.

Cottenie, A. (1980). Boron, calcium and nitrogen interactions in field legumes. In *Boron in crop plants* (pp. 473–503). American Society of Agronomy.

Cottenie, A. (1980). Boron in soil and plants. In *Boron in crop plants* (pp. 65–84). American Society of Agronomy.

Cottenie, A. (1980). Boron, calcium and nitrogen interactions in field legumes. In *Boron in crop plants* (pp. 473–503). American Society of Agronomy.

Cottenie, A. (1980). Boron, calcium and nitrogen interactions in field legumes. In *Boron in crop plants* (pp. 473–503). American Society of Agronomy.
Sameh, H., Youssef, F. H., El-Megeed, A., & Saleh, A. S. (2017). Improvement of faba bean yield using rhizobium/agrobacterium inoculant in low-fertility sandy soil. Agronomy, MDP.

Santos, P. C., Fang, Z., Mason, S. W., Setubal, J. C., & Dixon, R. (2012). Distribution of nitrogen fixation and nitrogenase-like sequences amongst microbial genomes. BMC Genomics, 13(1), 162. 10.1186/1471-2164-13-162

SAS (Statistical Analysis System). (2003). Statistical analysis system, version 9.1. SAS Institute Inc.

Siczek, A., & Lipiec, J. (2016). Impact of aba bean-seed rhizobial inoculation on microbial activity in the rhizosphere soil during growing season. International Journal of Molecular Science, 17(5), 784. 10.3390/ijms17050784

Sprent, J. I., & James, E. K. (2007). Legume evolution: Where do nodules and mycorrhizas fit in? Plant Physiology, 144(2), 575–581. 10.1104/pp.107.096156

Sullivan, D. M. O., & Angra, D. (2016). Advances in faba bean genetics and genomics. Genetics in Frontier, 7, Article 150.

Tedese, T. (1991). Soil, plant, water, fertilizer, animal manure and compost analysis. (Working Document No. 13). International Livestock Research Center for Africa.

Tefera, M., Chernet, T., & Haro, W. (1996). Geological map of Ethiopia. Ministry of Mines Geological Survey of Ethiopia, GSE.

Tisdal, S. L., Nelson, W., Beaton, J. D., & Havlin, J. L. (2003). Soil fertility and fertilizers. Asoke k. Ghosh. Troeh, F. R., & Thompson, L. M. (1993). Soil and soil fertility. McGrawHill, Inc.

Van Reewijk, L. P. (1992). Procedures for soil analysis (3rd ed.). International Soil Reference Center (ISRIC). L.E. and Clark, F.E. (eds.). Methods of soil analysis. Agronomy No. 9.

Walkley, A., & Black, A. L. (1934). An example of the digestion method for determining soil organic matter and a proposed modification of the chromic acid titration method. Soil Science, 34(1), 29–38. 10.1097/00010694-193401000-00003

Werkalemahu, A. (2009). The effect of indigenous root-nodulating bacteria on nodulation and growth of faba bean (vicia faba L.) in the low-input agricultural systems of Tigray highlands, northern Ethiopia. CNCS, 1, 30–43.

Yadav, J., & Verma, J. P. (2014). Effect of seed inoculation with indigenous Rhizobium and plant growth promoting rhizobacteria on nutrients uptake and yields of chickpea (Cicer arietinum L.). European Journal of Soil Biology, 63, 70–77. https://doi.org/10.1016/j.ejsobi.2014.05.001

Yadav, O., & Shukla, U. (1983). Effect of zinc on nodulation and nitrogen fixation in chickpea (Cicer arietinum L.). The Journal of Agricultural Science, 101(3), 559–563. https:// doi.org/10.1017/S0021859600038582

Yazied, A., & Mady, M. A. (2012). Effect of boron and yeast extract foliar application on growth, pod setting and both green pod and seed yield of broad bean (Vicia faba L.). Journal of Applied Sciences Eseach, 2, 1240–1251.

Zerihun, A., & Abera, T. (2014). Yield response of faba bean to fertilizer rate, rhizobium inoculation and lime rate at Gedo highland, Western Ethiopia. Global Science Research Journals, 2, 135–139.