RESPONSE OF CHICKPEA (*Cicer arietinum* L.) TO VARIETIES, INOCULATION WITH *MESORHIZOBIUM* STRAINS, AND APPLICATION OF BLENDED NPSB FERTILIZER AT CHEHA DISTRICT, GURAGE ZONE, SOUTHERN ETHIOPIA

MSc. THESIS

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A THESIS SUBMITTED TO WOLKITE UNIVERSITY,

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DEDICATION

This MSc thesis manuscript is dedicated to my father Nahusenay G/Egziabher who was not lucky to see my success. I also dedicate this piece of work to my beloved mother Fentaye Baysase who had always been supporting me towards and sought my success.

STATEMENT OF THE AUTHOR

First, by my signature below, I declare and affirm that this Thesis is the result of my genuine work. I have followed all ethical and technical principles of scholarship in the preparation, data collection, data analysis and compilation of this Thesis. Any scholarly matter that is included in the Thesis has been given recognition through citation. This Thesis is submitted in partial fulfillment of the requirement for an MSc. degree at Wolkite University. The Thesis is deposited in the Wolkite University Library and is made available to borrowers under the rules of the Library. I solemnly declare that this Thesis has not been submitted to any other institution anywhere for the award of any academic degree, diploma, or certificate.

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LIST OF ACRONYMS AND ABBREVIATIONS

ANOVA	Analysis of Variance
ATA	Agricultural Transformation Agency
ATP	Adenosine triphosphate
BNF	Biological Nitrogen Fixation
CEC	Cation Exchange Capacity
CIMMYT	International Maize and Wheat Improvement Center
CSA	Central Statistics Agency
CWANRO	Cheha Woreda Agriculture and Natural Resource Office
DAP	Di-ammonium phosphate
DZARC	Debre Zeyit Agricultural Research Center
EIAR	Ethiopian Institute of Agricultural Research
ETB	Ethiopian Birr
Ethio-SIS	Ethiopian Soil Information System
FAOSTAT	Food and Agricultural Organization Statics
GLM	General Liner Model
ILRI	International Livestock Research Institute
LSD	Least Significance Difference
MRR	Marginal Rate of Return
Mbp	Mega Base Pairs
masl	Meter Above Sea Level
MBI PLC	Menagesha Biotech Industry Private Limited Company
MoA	Ministry of Agriculture
MoARD	Ministry of Agriculture and Rural Development
NSTC	National Soil Testing Centre
OC	Organic carbon
OM	Organic Matter
PLC	Private Limited Company
SAS	Statistical Analysis Software
SNNPR	Southern Nation Nationalities and People Region
TN	Total Nitrogen

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Response of Chickpea (*Cicer arietinum* L.) **to Varieties, Inoculation with *Mesorhizobium* Strains and Application of Blended NPSB Fertilizer at Cheha District, Gurage Zone, Southern Ethiopia**

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ABSTRACT

Chickpea (Cicer arietinum L.) is a major legume crop in Ethiopia and provide multiple benefits, due to high nutritive value as well as the ability of the crop to enrich nitrogen poor soils due to biological nitrogen fixation with different strains of endosymbiotic Mesorhizobium spp. However, the effectiveness of the strains varies due to inherent physiological characteristics of the endo-symbionts, the host varieties, and nutrient availability in the soil. Its' cultivation in the study area is constrained mainly by low soil fertility (low N, low soil OM, low available P, S and B) causing ineffective nodulation, and lack of improved agronomic practices. Therefore, the field experiment was conducted during the main cropping season of 2021/22 at Cheha district in Buchach Kebele with the objective of evaluating the effect of inoculation with Mesorhizobium strains and NPSB fertilizer application on nodulation, growth, and yield performance of different chickpea (Cicer arietinum L.) varieties. Factorial combinations of three Mesorhizobium strains CP-M41, CP-EAL 029, CP-M20b, and un-inoculated control, two NPSB levels (0 and 121 kg NPSB ha^{-1}) and two chickpea varieties (Local and Arerti) were laid out in randomized complete block design (RCBD) with three replications. Analysis of variance showed that crop phenology, nodulation parameters, growth parameters, yield and yield components were significantly affected by the main effects. The effect was more pronounced in the interaction effect of different Mesorhizobium strains with NPSB application and varieties. The maximum values of most studied parameters were found from Arerti variety at the combined application of CP-M41 Mesorhizobium strain with NPSB fertilizer (121 kg ha^{-1}) as compared with the single application and the control. The highest grain yield (3177.16 kg ha^{-1}) was recorded from the use of Arerti variety at the combined application of NPSB fertilizer with CP-M41 Mesorhizobium strain, which resulted in 15.96%, 24.06% and 37.93% increment over the use of Arerti x CP-M41 strain, Arerti x NPSB, and the control, respectively. From the economic point of view, the partial budget analysis of the study treatments revealed that the highest net return (102,092.6 ETB ha^{-1}) with an acceptable marginal rate of return (618%) was gained from Arerti variety with the combined application of 121 kg NPSB ha^{-1} and Mesorhizobium strain CP-M41 inoculation. Hence, it can be concluded that the use of Arerti variety with combined application of 121 kg NPSB ha^{-1} with CP-M41 strain is found to be an appropriate combination to get higher yield and profit of chickpea crop at the study area. However, verification of the result on farmers' fields across season and areas could be required before wide use of this study to put the recommendation in firm ground.

Key words: Chickpea varieties, *Mesorhizobium* strains, Blended NPSB Fertilizer, Nodulation, Effective Nodules.

1. INTRODUCTION

Chickpea (*Cicer arietinum* L.) is an ancient legume crop of great economic importance; ranked third in terms of production and second in terms of area harvested among the grain legumes in the world (Vishnyakova *et al*. 2017; Zehara, 2021). It is the third most important legume crop both in terms of area and production in Ethiopia (CSA, 2021). The crop is produced almost in all the regions of the country as food crop (CSA, 2021). In addition to its contribution for food security in Ethiopia, chickpea plays an important role in nutritional security of the nation by providing protein, fat, carbohydrates, fiber, essential amino acids and a considerable amount of vitamins (Gowda *et al*., 2016). The crop is also an important component of crop rotation to enhance its yield and succeeding crops as it improves the nitrogen (N) content of soil through symbiotic nitrogen (N_2) fixation (Zehara *et al*., 2020).

In Ethiopia, area coverage and total production of both Desi and Kabuli chickpea types during 2020/21 cropping season were about 220, 719 ha and 4,573,193 quintals (CSA, 2021). Despite its wide spread cultivation and uses, the national average yield of the crop in the country under farmer's condition is 2 t ha^{-1} (CSA, 2021), which is far below the average potential yield of the crop under improved management conditions (5 t ha^{-1}) (Gemechu *et al*., 2011). Poor soil fertility (Korbu *et al*., 2020; Sultan *et al*., 2018), lack of nutrient supply (Endalkachew *et al*., 2018), lack or insufficient availability of effective indigenous *Rhizobia* populations in soils frequently cultivated with cereals (Endalkachew *et al*., 2018; Wondwosen *et al*., 2016a and 2017) which in turn resulted in lower biological N_2 fixation (BNF) by the crop (Dar *et al*., 2016), and shortage of improved varieties (Sheleme *et al*., 2015) are causes for the low yield of the crop among others.

Most agricultural soils in Ethiopia are reported to be deficient in Sulphur (S) and boron (B) in addition to nitrogen (N) and phosphorous (P) deficiencies (EthioSIS, 2016). Soil erosion, intensive cultivation over long period of time, and minimal or unbalanced fertilizer application such as NP fertilizer (among others) are reported to be the major causes of poor soil fertility in the country (Beza, 2017; Endalkachew *et al*., 2018). Yield reduction on chickpea due to deficiency of these nutrients has been reported in Ethiopia (Beza, 2017; Endalkachew *et al*., 2018; Korbu *et al*., 2020). Several authors such as Kiros and Atsede (2020); Tesfahun *et al*. (2018); and Wang *et al*. (2008) reported a significant yield response of chickpea to the application of inorganic fertilizers containing N, P, S and B.

In terms of N supply, leguminous crops can make use of *Rhizobia*, which are bacteria living symbiotically with legume crops, by fixing atmospheric N_2 (Wondwosen *et al*., 2016b; Lindström and Musavi, 2020). Chickpea requires inoculation with effective *Rhizobia* to obtain 60-80% of its N requirement through symbiotic N_2 fixation (Giller, 2001). The rest of N gained from soil inorganic N, mineralized organic matter, fertilizer application (Endalkachew *et al*., 2018). The crop is selective in its symbiotic requirement, nodulating with only in association with effective and compatible *Mesorhizobium* strain (Wondwosen *et al*., 2016a). However, efficiency of nodulation and nitrogen fixation depends on the compatibility between the chickpea genotype and the *Rhizobia* strain, and environmental conditions (mostly soil factors such as nutrient availability) (Ashenafi *et al*., 2020; Endalkachew *et al*., 2018). Different research reports showed a significant yield increment in chickpea as a result of seed inoculation with effective *Rhizobia* inoculants (bio-fertilizers) as compared to the un-inoculated plants (Mulugeta *et al*., 2018; Wondwosen *et al*., 2017). This indicated the need of *Rhizobia* inoculation to the seed.

Although chickpea can fix up 60–80% of its required nitrogen (Giller 2001), actual symbiotic effectiveness is likely to differ between *Rhizobia* strain genotypes (G_R), legume genotypes (G_L) as well as their combination (G_L x G_R) (Giller *et al*., 2013; Ashenafi *et al*., 2020). In line with this, many authors also showed that symbiotic and agronomic responses of chickpea crops vary with varieties (Wondwosen *et al*., 2016a; Gemechu *et al*., 2018; Tamiru and Girma, 2019a and b), *Rhizobium* strains (Abere and Getahun, 2019), variety by strain combination (Molla, 2016), and also the environmental conditions in which the crop grows (Endalkachew *et al*., 2018; Genet *et al*., 2020). Moreover, different researchers have recommended the combined application of inorganic fertilizer with *Rhizobia* inoculation of seeds as way of correcting nitrogen deficiency for the enhancement of productivity of pulse crops including chickpea (Daur *et al*., 2008; Sheleme *et al*., 2015; Wondwosen *et al*., 2016a; Kiros and Atsede, 2020). Other authors also reported that application of mineral nutrients such as P, S and B along with *Rhizobia* inoculants in nutrient deficient soils are important for the symbiotic relationship that enables bacteria to fix more nitrogen from the air and thereby improving growth and yield of chickpea through increasing nutrient availability (Admasu, 2019; Mulugeta *et al*., 2018; Pal *et al*., 2021). Likewise, Kiros and Atsede (2020) also recommended the application of blended fertilizer along with *Rhizobia* inoculation of seeds to enhance chickpea grain yield and profitability in Laelay maichew district of Tigray region, Ethiopia.

Above background indicated that seed inoculation with proper *Rhizobia* strains has positive benefits for improving chickpea yield in nutrient depleted soils of Ethiopia including soil in the study area which is deficient in N, P, S and B (ATA, 2016). Furthermore, several authors have also recommended the combined application of inorganic fertilizers with *Rhizobia* inoculants along with superior varieties as way to enhance chickpea productivity. Based on these facts, therefore, there is a need to evaluate the symbiotic and agronomic performance of different chickpea varieties, *Mesorhizobium* strains as well as their combination in order to identify better yield improvement technology in chickpea production environments in Ethiopia.

Cheha district (the study area) is one of the areas found in Gurage Zone of SNNPRS having a favorable environment for chickpea cultivation. Vertisols are the dominant soil type in the area. Farmers produce the crop as a source of food, cash and also rotate it with cereal crops. As per soil fertility map of SNNRS developed by ATA (2016), N and S in soils of the study area is rated as low while P and B are very low and this contradict the crop requirement. In Ethiopia in general and in the study area in particular, in order to reduce the negative impact of poor soil fertility on the yield of the crop, blended NPSB fertilizer is recommended at a rate of 121 kg ha^{-1} for food legumes (MoA, 2014; ATA, 2014). However, most farmers in the study area do not apply any external inputs for chickpea production with the common perception among farmers that it does not need any fertilizer application (Cheha District Office of Agriculture, October 2021; Endalkachew *et al*., 2018) and fertilizer is costly. As a result, productivity of chickpea in the study area is lower than the national average (CSA 2021). Hence, symbiotic N_2-fixation can be used as a cheaper alternative for improving soil N-content. To our knowledge, chickpea varieties which give high yield as well as respond favorably to the application of inorganic fertilizers and *Mesorhizobium* inoculants were not identified for Cheha district though different chickpea varieties are easily available in the area.

Moreover, there is limited, if any, documented information regarding the response of chickpea to the combined application of *Rhizobia* inoculation and inorganic fertilizers under Cheha condition in Southern Ethiopia. Thus, identifying suitable *Rhizobia* strains in combination with the use of blended chemical fertilizer would be useful in developing effective nutrient management for chickpea production in the study area.

In addition, there is also a need to consider the relative cost and profitability of these technologies with respect to their adoption by small-holder farmers in the study area. And it is hypothesized that application of the recommended rate of blended NPSB fertilizer combined with suitable and effective *Rhizobia* strain would increase growth and yield of chickpea varieties in the study area. Therefore, the general objective of this study was to evaluate the effect of seed inoculation with *Mesorhizobium* strains and NPSB fertilizer application on nodulation, growth, and yield performance of different chickpea (*Cicer arietinum* L.) varieties in Cheha District, Gurage zone, Southern Ethiopia. The specific objective**s** were:

- To evaluate the effects of *Mesorhizobium* strains, NPSB fertilizer application and varieties on nodulation, growth, yield and yield components of chickpea (*Cicer arietinum* L.).
- To identify the suitable combination of *Mesorhizobium* inoculum, NPSB fertilizer and variety that could improve productivity in the study area.
- To assess the economic feasibility of *Mesorhizobium* inoculation, NPSB fertilization and variety for chickpea production at Cheha district.

2. LITERATURE REVIEW

2.1. Origin, Distribution and Botany of Chickpea

Chickpea (*Cicer arietinum* L.) is one of the most important cool season legume crop. It originated in around 10,000 years ago from Southeast Turkey and Syria, having its primary center of diversity there with secondary centers of diversity in India and Ethiopia (Plekhanova *et al*. 2017). Currently, it is produced across 50 countries worldwide (FAOSTAT, 2021), and the top ten chickpea producing countries are; India, Turkey, Pakistan, Australia, Russia, Myanmar, Ethiopia, Iran, Mexico, and Canada (FAOSTAT, 2021).

Chickpea (*Cicer arietinum* L.) is an annual diploid with 16 chromosomes and genome of 740 Mbp (Jain *et al*., 2013). It is a member of genus *Cicer*, tribe *Cicereae*, subfamily *Papilionoidea*e, family *faba ceae* (Van der Maesen *et al*., 2007). Based on distinct botanical or morphological features and molecular diversity analysis, chickpea was primarily classified into two major groups: desi and kabuli type (Sheleme *et al*., 2015). Desi type of chickpea is characterized by the small size seeds (microsperma), pods, leaflets, and plantlets. However, large variations in flower and seed coat color, and seed shape commonly observed. The flower color ranges from white to red, purple and blue, and seed coat with different shades of brown. The seed shape is angular, and seed coat varies from smooth to wrinkled, which reflects the genetic diversity (Sheleme *et al*., 2015).

The kabuli type, also known as the “macrosperma” chickpea, is known for the “ram’s head” shaped seeds. In contrast to the desi type, the kabuli type is characterized by large seeds, pods, leaflets and plantlets. The flower and seed coat colors are usually white or cream colored. The seed coat of kabuli type is thinner and less winkled than that of the desi type (Sheleme *et al*., 2015). The desi type of chickpea accounts for up to 85% of production; whereas the cream to yellow colored seeded kabuli type contributes to 15% of the global production (Imran *et al*., 2015; Thudi *et al*., 2017; Zafar *et al*., 2017). Chickpea has the ability to convert atmospheric nitrogen into ammonia, by a process known as biological N_2 fixation through a symbiotic relationship with *Mesorhizobium* (Nour *et al*., 1994).

2.2. Agro-ecological Requirements of Chickpea

Chickpea is traditionally grown in the northern hemisphere, mostly at relatively high elevations in India and Ethiopia. It is grown between 20°N and 40°N in the northern hemisphere and 27°S and 38°S in the southern hemisphere (Imtiaz *et al*., 2011). These environmental conditions give significance difference in photoperiod, temperature and precipitation, all of which have a profound effect on growth and development (Geletu and van der Maesen, 2006). Currently chickpea is mainly grow in the central, northern and eastern highland areas of Ethiopia at an altitude of 1400-2300 meter above sea level (m.a.s.l.), where annual rainfall ranges between 500 and 2000 mm (Geletu and van der Maesen, 2006).

The crop is a quantitative long-day plant that needs a moderately high temperature for its normal growth. It also requires a daily temperature fluctuation with narrow amplitude. Chickpea grows best on heavy clay soils and in a rough seedbed and it is moderately tolerant to drought conditions (Yasin, 2014). Cool nights, moderate relative humidity, evenly distributed rainfall and drained seedbeds are conducive for the crop and considered as the ecological optimum of the crop for its normal growth and development (Menale *et al*., 2009). Chickpea can be widely grown on different soil types provided there is good moisture and drainage. Well drained black soils (usually Vertisols), which have good water holding capacity, are suitable soil types for optimum growth of chickpea (Yasin, 2014).

2.3. Significance of Chickpea Production

Chickpea plays an important role in human nutrition by providing protein, fat, carbohydrates, fiber, essential amino acids (mainly, lysine, methionine, threonine, valine, isolucine and leucine), and a considerable amount of vitamins such as B1 and B2, ascorbic acid (vitamin C), and niacin (Gowda *et al*., 2016). It is sown with low water inputs, largely reliant on stored residual moisture following wet seasons and can grow well even in the marginal soil and soils of varying textures (Imran *et al*., 2015). Consequently, it is a relatively drought tolerant grain legume grown in semi-arid regions may act as an insurance crop in poor seasons when the main crop fails due to drought (Ogola, 2015). Agronomically, it is an important part of crop rotations along with major cereal crops, especially in the Vertisols where other food legumes poorly adapted (Lijalem *et al*., 2016).

Grain husks after threshing the pods, dry haulms, stems, and leaves are also used as protein feed in smallholder crop-livestock system, particularly in the central highland farming systems where small-scale animal fatting farms have flourished. More importantly, domestic demand for its grain has been substantially increasing recently, and the crop commanded higher prices in local markets providing favorable returns on investment to the smallholder farmers (Setotaw *et al*., 2018). Nitrogen fixation ability of chickpea increases the protein content its seeds, enhance soil fertility and benefits both chickpea and its following crops there by reducing the use of chemical N fertilizers (Khaitov and Abdiev, 2018). Greenlon *et al*. (2019) estimated that up to 181 kg N ha^{-1} is obtained when chickpea pods are harvested and crop residues are tilled into the soil.

2.4. Production and Productivity of Chickpea in the World and Ethiopia

In 2020/21 cropping season, chickpea was grown on about 15 million hectares of land across the world with an average productivity of 1.01 t ha^{-1} globally (FAOSTAT, 2021). In Africa, Ethiopia stands first in area coverage (220,719 ha) and production (457,319 tons) during 2020/21 (CSA, 2021; FAOSTAT, 2021). About 65 and 3 % of chickpea produced in Africa and the world, respectively is from Ethiopia (FAOSTAT, 2021). Ethiopia has suitable agro-climatic conditions for production of chickpea. The crop is highly integrated into the farming system and ecologically friendly for growing in many areas. Chickpea is grown in almost all regions of Ethiopia with residual moisture on black Vertisol soils (Menale *et al*., 2009). Amhara and Oromia regions together produce 93% of total chickpea production in Ethiopia while SNNPR and Tigray produce 3.5% and 3%, respectively (Sheleme *et al*., 2015).

2.5. Biological Nitrogen Fixation by *Rhizobia* Bio-fertilizers

Though molecular nitrogen represents nearly 80% of the earth's atmosphere, it is chemically inert and cannot be directly assimilated by plants. N_2 needs to be converted in to NH_4^+ and NO_3^- to be used by plants (Hoffman *et al*., 2014; Geurts *et al*., 2016). Naturally, only limited numbers of prokaryotes are able to convert the N_2 into a usable form of N through a process known as biological N_2 fixation (BNF) (Bayou *et al*., 2015). *Rhizobium*-legume symbiosis is a host specific association and the need to identify specific strains and the diversity of rhizobia associated with legumes is vital for better exploitation of the benefits of the rhizobia as bio-fertilizers (Koskey *et al*., 2018). The efficiency of nitrogen fixation varied between strain either due to genomic background of the rhizobia and/ or the combinations between strains, plant varieties and soil factors (Wang *et al*., 2019).

It is, therefore, of paramount importance to understand the major phases of the general symbiotic process involved such as plant infection, nodulation and nodule maturation, senescence, release of rhizobia and persistence of rhizobial populations in soil (Musarrat *et al*., 2010).

The developmental stages start with the attachment of the bacteria within root hairs followed by root hair curling, epidermal invasion and crack entry for the formation of root nodules that provide an environment suitable for nitrogen fixation by rhizobia (Oldroyd *et al*., 2011). The success of the symbiosis depends on the recognition of rhizobia by the legume host to activate the expression of a group of bacterial nodulation(nod) genes leading to initiation of bacterial infection (Gibson *et al*., 2008). Chickpea establish symbiosis with the type of root nodule bacteria belonging to the specific genus *Mesorhizobium* with an intermediate growth rate between the genera *Rhizobium* and *Bradyrhizobium* (Nour *et al*., 1994).

Rhizobium inoculants or rhizobial bio-fertilizers are selected strains of beneficial soil microorganisms cultured in a laboratory and packed in with or without a carrier. They are host-specific, low cost and an environmentally friendly source of nitrogen (EIAR, 2014; Getahun *et al*., 2018). Carrier-based rhizobial bio-fertilizers are coated on legume seeds before planting (process called as inoculation) to enhance growth and yield of legume crops and provide nitrogen and organic carbon for subsequent or associated crops. Rhizobial dressed seeds must be planted in moist soil as soon as possible. Rhizobial bio-fertilizers can improve and sustain soil fertility and soil health when used as part of a long-term rotation system (Getahun *et al*., 2018). Daniel and Endalkachew (2017) reported that *Rhizobium* inoculation with strain GN100 increased soil N by 51.6% compared to soil N content before planting. Rhizobial biofertilizers help enhance production and productivity of grain legume crops. It improves grain or biomass yield up to 10% (particularly with 100 kg DAP ha^{-1}) in any cropping system through boosting plant growth promoting enzymes, hormones and auxins and increasing yields leads to higher income that leads to greater margins when favorable markets exist for the farm produces (Getahun *et al*., 2018).

So far, in Ethiopia, only few native strains of chickpea rhizobia were identified, among which the two *Mesorhizobium* strains “CP-41” and “CP-029” are reported as effective commercial inoculants widely used by farmers (Wondwosen *et al*., 2016a; Endalkachew *et al*., 2018).

Rhizobia biofertilizer is cheaper and usually more effective agronomic practice for ensuring adequate N nutrition of legumes than chemical N fertilizer (Chianu *et al*., 2011). Inoculating legumes with adaptable and effective species of specific rhizobia increase the success of their establishment, nodulation, biomass and N yields or uptake (Adamu *et al*., 2001; Habtegebrail and Singh, 2006). Therefore, there is a need to apply rhizobia inoculation technology for enhancing chickpea production and productivity in chickpea growing areas of Ethiopia.

2.6. Response of Chickpea to *Mesorhizobium* Inoculation

Nitrogen fixation in agriculture has been improved by inoculation of legume crops with suitable rhizobia. Inoculation with compatible and appropriate rhizobia is critical for sustained yield in farmlands where N supply limits production of grain legume (Abd-Alla *et al*., 2014). Inoculation of legume with rhizobia is known to increase nodulation, N uptake, growth, yield and yield attributes of legume crops (Erman *et al*., 2011). If specific and effective rhizobia are absent in a soil or if they are present in low numbers, it is necessary to introduce the rhizobia in that soil to ensure proper nodulation and N_2 fixation. Mulugeta *et al*. (2018) indicated that knowledge of the biodiversity of rhizobia and of local populations is important for the design of successful inoculation strategies. According to Endalkachew *et al*. (2018), two situations identified which are the absence of compatible strains and the small rhizobia populations in the soil are important limitations for nodule formation in chickpea.

Chickpea require effective rhizobia to fix atmospheric nitrogen. It is selective in its symbiotic requirement for nodulating with only a specific group of rhizobia species (Wondwosen *et al*., 2017). *Mesorhizobium* strains are commonly used in chickpea. The beneficial effects of *Mesorhizobium* inoculation in chickpea crops has been primarily related to increase in the N uptake, nodulation, growth and yield on chickpea (Wondwosen *et al*., 2017). Mulugeta *et al*. (2018) from their experiment reported that chickpea inoculation with *Mesorhizobium* significantly increased number of nodules per plant, nodule volume, nodule dry weight, number of seeds per plant, biomass yield, and grain yield by 41.43, 28, 53.33, 31.86, 15.79, and 27.91% respectively as compared to un-inoculated control. Similarly, Singh *et al*., 2018 reported higher nodule number and dry weight per plant from rhizobia inoculated chickpea plot as compared to un-inoculated plots, which is 33.57 and 0.68g, respectively.

An increase in nodule number, nodule dry weight, shoot dry weight, plant height, number of branches, number of pods, grain and straw yields of chickpea with *Mesorhizobium* inoculation has been reported (Sharma *et al*. 2015; Wondwosen *et al*. 2016a). Recently, Endalkachew *et al*. (2018) also found that chickpea genotypes inoculated with rhizobia strains showed higher N uptake and substantially increased grain yield by many folds compared with un-inoculated control.

The enhanced yield of chickpea through inoculation could be due to the fact that inoculation improved nodulation and nutrient availability that improved plant growth, dry matter accumulation and consequently the yield (Namvar *et al*., 2013). Similarly, Togay *et al*. (2008) attributed the growth and yield response of chickpea to rhizobia inoculation due to its ability to fix N and increase N uptake of the crop. Moreover, improved growth and yield response of chickpea to the *Rhizobium* bacteria might be attributed to the ability of the bacteria to produce growth-promoting substances (phytohormones). *Rhizobia* bacteria synthesized phytohormones including auxin as secondary metabolites in inoculated plants, which promote seed germination, root elongation and stimulation of leaf expansion. In addition, improved root development and proliferation of plants in response to rhizobia activities enhance water, nutrient uptake and consequently the yield (Erman *et al*., 2011).

Researchers indicated that an increased nodulation, growth yield and yield attrinutes of chickpea due to *Rhizobia* inoculation as compared to un-inoculated treatment might be because of the reason that inoculated bacteria strain had good nodule inducing capacity and N fixing ability over the native soil *Rhizobia* population (Singh *et al*., 2011). The higher nodulation due to inoculation resulted in higher nitrogen fixation and eventually produced higher number of pods per plant which bring about higher grain yields as a whole (Singh *et al*., 2011).

However, it has been observed from several studies that the nodulation, N_2 fixation, growth and yield responses of chickpea plants to *Mesorhizobium* inoculation are influenced by the use of different rhizobia strains (Assefa, 2016; Wondwosen *et al*., 2017; Tamiru and Girma, 2019a and b; and Ashenafi *et al*., 2020). This indicated that rhizobia strains varies in their symbiotic effectiveness and N fixing capacity (Albareda *et al*., 2008).

Similarly, Wondwosen *et al*. (2016a) from their field experiment observed that chickpea plants inoculated with CP 41 strain significantly increased number of nodules, nodule dry weight, shoot dry weight per plant, number of branches, number of pod per plant, straw and grain yield by 85.96, 84.62, 43.75, 29.89, 20.58, 24.06, and 26.65 %, respectively as compared with CpNSTC strain in Southern Ethiopia.

In addition, the study by Tamiru and Girma (2019b) showed a significant variation between two *Mesorhizobium* strains (EAL 018 and EAL 029) in nodulation, N_2 fixation, growth and yield characteristics of chickpea. They reported that among the strains, the higher nodule number per plant (37.83 $plant^{-1}$), nodule volume (0.67 ml), nodule dry weight (139.80 mg $plant^{-1}$), amount of N fixed (43.537 kg ha^{-1}), and percent N derived from the atmosphere (0.341 %) was recorded from inoculation with EAL 029 strain than EAL 018 strain that scored 35.68, 0.64 ml, 132.90, 36.123 kg ha^{-1}, and 0.309 %, in that order.

In general, the use of bio-fertilizers such as *Rhizobium* inoculants has enormous benefits in chickpea production in nutrient depleted soils of Ethiopia. Furthermore, previous research findings showed varations among *Mesorhizobium* inoculants efficiency on plant growth promoting, symbiotic and agronomic performance of chickpea crop. Therefore, such like field trails or investigations in different chickpea growing agro-ecologies of Ethiopia including the current study area are highly needed to identify the potential of *Rhizobium* inoculants in improving the growth and yield performances of chickpea varieties.

2.7. Response of Chickpea to N, P, S and B Fertilizers Application

2.7.1. Nitrogen

Nitrogen is the fourth most abundant element in plant and is an essential component of amino acid, enzyme, hormones, phenolics, phytoalexins and protein. It is available as the oxidized anion NO_3^- or the reduced cations $NH_4^+$and the form can affect the uptake of other element (Brady and Weil, 2016). For legumes including chickpea, it is more useful because it is the main component of amino acids as well as proteins. Adequate supply of N is essential for normal growth and yield. Without N it is not possible to synthesize the necessary proteins, enzymes, DNA and RNA required in virtually all plant cells for their initial development, sustained growth and functioning to support other tissues of the plant (Mozumder *et al*., 2003).

There are increasing evidences demonstrating the positive benefits of N fertilizer application on chickpea. Addisu *et al*. (2016) also reported that N_2 application markedly improved overall agronomic performances of chickpea cultivars studied on Vertisols at Debre Zeit site. Similarly, an on-farm fertilizer trail conducted in the southern region revealed that NP fertilizer application has significantly increased the biomass and grain yield of chickpea cultivar compared with unfertilized control (Lemma *et al*. 2013). According to this study, 11.5 kg N and 10–20 kg P rates were found economically viable and hence recommended for optimum chickpea production in the region. Endalkachew *et al*. (2018) also reported that nutrient (specifically N) application has significantly enhanced symbiotic efficiency in chickpea. Other authors such as Girma *et al*. (2018) revealed that the application of half the recommended rate of NP fertilizers (in the form of DAP) gave double grain yield and biomass of chickpea as compared to the unfertilized control.

2.7.2. Phosphorus

Phosphorus is one of the essential nutrients for legume growth and BNF (Mulugeta *et al*., 2018). Phosphorus influenced initiation of nodules development as well as efficiency of the symbiosis relationship between rhizobia and legumes (Nyoki and Ndakidemi, 2014). Because it influences nodule development through its basic functions in plants as an energy source and plays a vital function in increasing plant tip and root growth, decreasing the time needed for developing nodules to become active and benefit to the host legume. Legumes such as chickpea need P for adequate growth and N_2 fixation. Sufficient P levels are also required to enhance different plant organs growth and promote nodulation and early maturity (Kamara *et al*., 2010).

Phosphorus application at a rate of 30 kg ha^{-1} significantly increased nodule number per plant, nodule dry weight per plant, plant height, number of branches per plant, number of pods per plant, number of seed per pod, thousand seed weight, biomass yield and grain yields of chickpea as compared with the control (no P application) (Lemma *et al*., 2013). Application of P influenced plant height, number of nodules per plant, number of branches per plant, number of pods per plant, number of grains per pod, grain yield and thousand-grain weight significantly in chickpea (Sharma *et al*., 2015). A field trail conducted in the northeastern highland Vertisols of Ethiopia also revealed that P has significantly improved key agronomic traits (Seid *et al*. 2013), and hence, 60 kg P ha^{-1} was recommended for the study areas and other chickpea production zones having similar agro-ecology.

A more comprehensive on-farm field study revealed that the application of P, either alone or along with rhizobial inoculation, increased N uptake of chickpea genotypes by 14–25 kg ha^{-1} comparing with the control (Endalkachew *et al*. 2018). This study further indicated that P application substantially enhanced N-fixation and increased grain yield by many folds.

2.7.3. Sulphur

Sulfur fertilizers are known to enhance crop yield and uptake of macronutrients especially nitrogen (Islam *et al*. 2012). Sulphur has been found to be an indispensable element for higher pulse production and it is an integral part of proteins, sulpholipids, enzymes etc. (Das and Misra, 1991). It is involved in various metabolic and enzymatic processes including photosynthesis, respiration and Legume-*Rhizobium* Symbiotic N fixation (Rao *et al*., 2001). Sulfur improves nodulation activity (Scherer *et al*., 2008) and affects growth of leguminous plants through its effects on N_2-fixation (Varin *et al*., 2009). It increases nitrogenase activity due to higher ferredoxin and ATP concentration in bacteroids of root nodules of legumes (Scherer *et al*., 2008).

Apart from the commonly used NP fertilizers, sulfur (S) is also one of the essential plant nutrients often neglected in many crop production systems. Chickpea is responsive to S application as well (Das *et al*. 2016). Recently, Beza *et al*. (2019) reported that the application of 30 kg S ha^{-1} substantially increased grain yield of chickpea genotypes by about 22.18% over the non-applied plots under rain-fed conditions in the Ethiopian highlands. Islam *et al*. (2012) described that application of 30 kg S ha^{-1} increased the seed yield of chickpea up to 12% over the control (no sulfur application).

Moreover, Das *et al*. (2016) reported that application of S at a rate of 20 kg ha^{-1} responded significantly higher seed yield mainly attributed to significantly higher number of primary branches per plant, number of pods per plant, and 1000 seed weight of chickpea compared with the less rate and it improved the seed yield by 16% over its non-application or control. Increase in yield due to S application may be due to the fact that sulfur application increased rate of photosynthesis due to an increment in protein synthesis and maintenance of high chlorophyll content (Patra *et al*., 2012).

2.7.4. Boron

Boron ranks third among the micronutrient and has a chief role in plant cell wall and membrane constancy (Bassil *et al*., 2004). Boron (B) is an essential micronutrient for plants, and plant requirements for this nutrient are lower than the requirements for all other nutrients except molybdenum and copper. It is required for normal development of reproductive tissues and its deficiency results in low grain set and poor seed quality. Even the cereals (like wheat and rice) and pulse like chickpea with small B requirement can suffer from impaired seed set due to B shortage at a critical growth stage (Brady and Weil, 2016).

According to Renukadevi *et al*., (2002), the application of boron maximizes the light interception ratio, biomass production, leaf area index, net assimilation rate, crop growth rate and seed yield in pulses. Boron increases nodulation activity, which may have increased the nitrogen content. Moreover, application of boron in boron deficient soil might have resulted in increased availability of boron that in turn has influenced DNA and protein synthesis leading to increased nitrogen content, uptake and consequently seed yield (Debnath *et al*., 2011). Kumar *et al*. (2018) showed that the grain yield of chickpea crop was increased by 38% due to the application of 2.5 kg B ha^{-1} compared with the non-applied plots. Similarly, Hoque *et al*. (2021) observed that application of boron at a rate of 2 kg ha^{-1} significantly increased the plant height, number of nodules, branche number, total pod number, seed yield and straw yield of chickpea genotypes by 3.51, 4.15, 7.95, 3.39, 12.5 and 7.25 % over the control. They specified that boron works in reproductive tissues and control flower drop which in result increases the amount of pods $plant^{-1}$ and consequently the seed yield of chickpea crop. Deficiency of boron showed a noticeable decline in the amount of flowers and fail to fruit, causing decreases in grain and pod yield. Boron plays an important role on seed yield of chickpea as it regulates plant hormone level, photosynthetic activity and generative growth in plants which increase yield of chickpea (Hoque *et al*., 2021).

From the above background, it can be concluded that the common perception among farmers that chickpea does not need nutrient application has contributed to a greater extent for lower national yield in Ethiopia. Moreover, the results of the above fertilizer studies indicated that Nitrogen, Phosphorus, Sulphur and Boron are important elements in chickpea nutrition. Recently, there is an increasing evidence to suggest that the application of inorganic fertilizers significantly enhanced plant growth, grain and biomass yield, and symbiotic efficiency of chickpea.

Therefore, considering the increasing nutrient depletion in most of the agricultural soils including the current study area, the use of major yield-enhancing chemical fertilizers as blended form such as NPSB for chickpea production is becoming indispensable to alleviate soil nutrient deficiencies and narrow down the huge yield gaps existing between the actual and attainable level towards realizing food security in such like chickpea growing areas of the country. Hence, such like field trails are needed to study the synergistic effects of N, P, S and B fertilizers on growth, yield component and yield performances of chickpea varieties in chickpea growing areas of Ethiopia.

2.8. Response of Chickpea to Different Varieties

Phenological and growth traits of chickpea varies with varieties (Assefa, 2016; Wondwosen *et al*., 2016a). Tamiru and Girma (2019a) reported delayed flowering and days to physiological maturity in Arerti than Habru variety, due to distinction in genetic makeup of the two cultivars, and on the other hand Habru variety produced the tallest plant height than Arerti. Similarly, Tamiru *et al*. (2021) reported significant differences between two chickpea varieties (Acos Dubie and Habru) for number of days to reach 50% flowering and maturity, plant height, number of branch, number of pods per plant, biological yield, grain yield and thousand grain weight. This could be attributed to genetic differences among the varities with respect to absorption of nutrients, nitrogen fixation and accumulation of other relevant nutrients (Chabot *et al*. 1996). Moreover, nodulation and nitrogen fixing capacity of chickpea also varied with varieties. In line with this, Wondwosen *et al*. (2016a) observed a higher nodule number and dry weight in Natoli than Shasho varities of chickpea, however, the later varity accumulated higher N in its biomass. Variation among chickpra varities in nodulation and N fixing capacity has been well documented (Zehara *et al*. (2020). The use of different varieties also influences the yield and yield component traits (number of pods, number of grains, grain weights, total aboveground biomass yield, straw yield, grain yield, and grain harvest index) of chickpea plants (Molla, 2016; Tamiru and Girma, 2019a; Tamiru *et al*., 2021).

In agreement with this, Wondwosen *et al*. (2016a) from their field experiments observed higher pod number and hundred-grain weight in Shasho than Natoli varieties. They indicated that Natoli variety recorded significantly higher straw yield during the 2011/2012 study season field experiment, but during the 2012/2013 study season field experiment Shasho yielded significantly higher straw yield as compared to Natoli.

This result was because of the presence of good rainfall distribution throughout the growing season during the 2012/2013 study season. In terms of grain yield, Natoli variety recorded significantly higher grain yield during the 2011/2012 study season field experiment, but during the 2012/2013 study season field experiment Shasho yielded significantly higher grain yield as compared to Natoli. In addition, Tesfahun *et al*. (2018) reported significant differences between two chickpea varieties (Mariye-Desi type and Arerti-Kabuli type) for number of seeds per pod, straw yield, grain yield, hundred grain weight and harvest index. Thus, due to the fact that agronomic and symbiotic responses of chickpea varies between varieties, their potential need to be investigated using multiple released varieties in a given agro-ecological zones.

2.9. Response of Chickpea Varieties to Combined Application of Inorganic Fertilizers with *Mesorhizobium* Inoculants

Variations in nodulation and N_2-fixation efficiency frequently occur in a bacteria strain-legume cultivar specific manner (Sanginga *et al*., 2000), and hence genotype of both the host and the competing rhizobia strains have been shown to influence the outcome. Genetic variation for N_2-fixation ability has been reported involving both the legume and *Rhizobium* components of the symbiotic association (Sanginga *et al*., 2000). Understanding the rhizobia-legume interaction is important for the development of rhizobial strains and legume cultivars with high N_2-fixation potential. Simultaneous selection for the optimal combination of the *Rhizobium* and the host usually results in more effective symbiosis and better growth of the host plant (Bayou *et al*., 2015).

Compatibility of *Mesorhizobium* with chickpea is strain and cultivar dependent, and efficiency of nodulation and nitrogen fixation depends on the compatibility between the chickpea genotype and the *Rhizobia* strain and the environmental (mostly soil factors) conditions (Ashenafi *et al*., 2020; Assefa, 2016). Tamiru and Girma (2019a and b) reported significant interactions between two chickpea varieties (Arerti and Habru) and *Rhizobium* strains (EAL 029 and EAL 018) for nodule dry weight and nodulation rating, where Arerti variety inoculated with EAL 029 strain produced maximum values over the other treatments. Intercation between chickpea varities and *Mesorhizobium* strains for nodulation, N-fixation and yield has also been documanetd by Wondwosen *et al*. (2016a).

The substantial effects (enhanced nodulation, growth, yield, and its attributes) due to the interaction of variety and strain might be attributed to varying symbiotic effectiveness of strains to fix atmospheric N_2 with different chickpea genotype or varied response of cultivars to inoculums (Molla, 2016).

Moreover, differences between chickpea varieties regarding their agronomic and symbiotic responses due to inorganic fertilizer application have also been reported (Tesfahun *et al*., 2018; China, 2018). The study by China (2018) revealed that blended NPSZnB fertilizer rates had a significant interaction effect with chickpea varieties on most of the agronomic parameters. In line with this findings, Tesfahun *et al*. (2018) indicated that the combined use of two chickpea varieties (Arerti and Mariye) and fertilizers significantly increased growth, yield components, and yield of chickpea plants as compared to their single effect.

Several authors also reported a significant effect of combining *Rhizobium* inoculant with N, P, S, B, N+P, P+S, P+B, NPS, and NPSB on symbiotic, growth and yield performances of chickpea crop (Ibsa, 2013; Singh *et al*., 2018; Mulugeta *et al*., 2018). Similarly, Endalkachew *et al*. (2018) reported that *Rhizobium* inoculation and P application increased chickpea grain yield by 21% and 25% respectively, while the combined application of inoculant and P resulted in a 38% increase. Furthermore, nodulation and yield of chickpea can be influenced by the interaction effect of varieties, *Rhizobium* inoculants and inorganic fertilizer (Nuru, 2020). In agreement with this, Tamiru and Girma (2019b) from their three factor experiment reported highest nodulation rate per plant form Arerti varity of chickpea when it was inoculated with EAL029 strain under 45 kg P_2O_5 application. This could be due to improved nodulation and healthy root system resulting from an adequate P supply and effective *Rhizobiu*m colonization in the root zone for successful N_2-fixation.

Similarly, Gan *et al*. (2010) observed that the application of N fertilization with *Rhizobium* inoculants significantly increased the seed yield and N uptake of different chickpea cultivars in southern Saskatchewan. The authors concluded that optimum productivity of chickpea can be achieved by applying effective *Rhizobium* inoculants and/or using sufficient amounts of N fertilizer regardless of cropping systems. Nuru (2020) also reported that the combined application of *Rhizobium* inoculation with NPS rates and varieties had significant effect on number of total nodules per plant and seeds per pod in common bean plants. Where, the highest number of total nodules per plant (68.53) and seeds per pod (7.5) were recorded from *Rhizobium* inoculation with NPS rate 100 kg ha^{-1} and for variety Nasir.

The author concluded that *Rhizobium* inoculation with application of NPS rate of 100 kg ha^{-1} found to be appropriate for common bean variety Nasir.

Generally, in many studies, it has been evidenced that nitrogen fixation capacity in legumes varies with environmental conditions (Kurdali *et al*., 2002), Cultivars (Walley *et al*., 2007), *Rhizobium* inoculants (Beck, 1992), and their interactions. Furthermore, several reports indicated that different chickpea varieties respond differently to the combinations of supplied N, P, S and B fertilizers and *Rhizobium* inoculant treatments in terms of their nodulation, N_2 fixation, growth and yield performances. Agronomic and symbiotic performances of different chickpea varieties significantly varied as a result of combining *Rhizobium* inoculation with inorganic fertilizers. Hence, field investigations are needed to identify and establish the interactive effects of different inorganic fertilizers and *Rhizobium* inoculants on growth, N_2 fixation, yield performance of different chickpea varieties, and the economic benefits in each specific agro-ecological zones found in Ethiopia.

3. MATERIALS AND METHODS

3.1. Description of the Study area

The field experiment was conducted at Buchach kebele of Cheha district. Cheha is one of the district in the Gurage Zone of Southern Nations, Nationalities, and Peoples' Region (SNNPR), Ethiopia. The site is located at 170 km away in the South West from the country’s capital Addis Ababa and 15 km away in the South East from the Zonal capital Wolkite city. The geographical position of the experimental site is 8^0 12’54’’ N longitude and 37^0 48’30’’ E latitude with an altitude of 1929 m.a.s.l. (Figure 1). The district has divers Agro-ecology i.e. 9 % dry-land (kola), 71 % mid-land (Woina dega), and 20 % highland (Dega) (Alemayehu, 2015). The major economic activity in the district is predominantly depended on agriculture and agribusiness related trading. Vertisols are the dominant soil type in the district (Alemayehu, 2015). The agricultural production is primarily based on Enset together with maize, wheat, barley, chickpea, yams, kale, and some other crops. The major cash crops include teff, niger seed, coffee, and Khat (CWANRO, 2020; CWAFEDO, 2020).

According to the ten years (2012-2022) metrological data found from Hawassa Metrological center, the area characterized by a unimodal rainfall pattern that receives mean annual rainfall of 986.24 mm. The mean monthly maximum and minimum temperatures are 25.44 and 11.35 ºC, respectively (Figure 2).

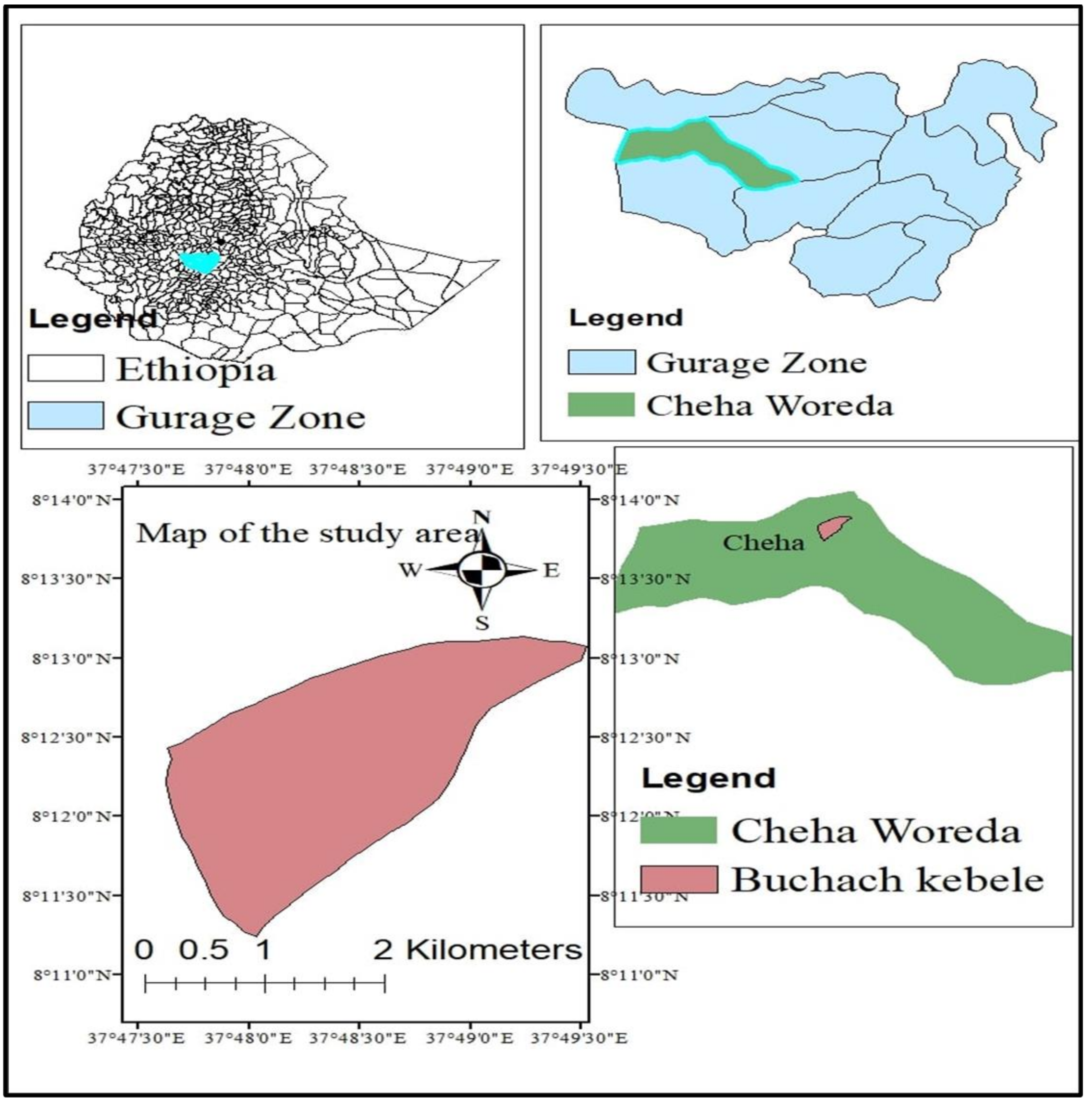


Figure 1: Location Map of the study area

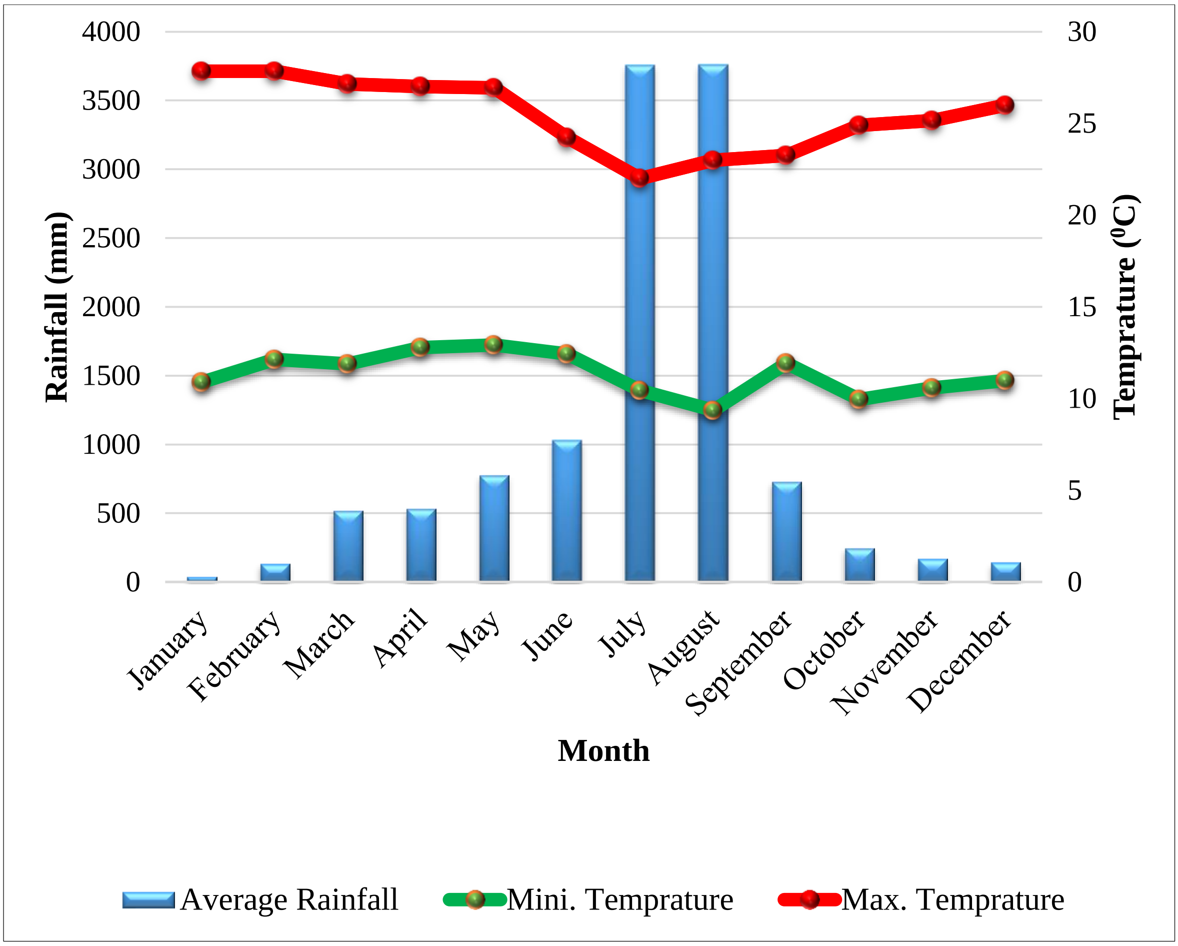


Figure 2: The mean monthly rainfalls (mm), maximum and minimum temperatures (^{0}C) of Cheha district for ten years (2012-2021)

3.2. Soil Sampling, Preparation and Analysis

Before planting, a composite soil sample was prepared by taking sub-samples from 20 sampling spot of the entire experimental site to a depth of 0-30 cm using an auger following zigzag sampling technique for determination of the physicochemical properties of the soil. The composite sample was air dried, ground using mortal and mixed thoroughly and then passed through a 2 mm sieve for most parameters except for organic carbon (OC) and total nitrogen (TN) which was passed through 0.5 mm sieve. Particle size distribution, pH, available P, OC, CEC, were analysed at soil laboratories of Wolkite University and Wolkite city, Ethiopia. kjeldhal N, available S and extractable B were analyzed at soil laboratory of Areka Agricultural Research Center.

Soil bulk density was determined on the undisturbed core sampling method after drying the soil samples in an oven at 105^{o}C to constant weights. Soil particle size distribution was determined by hydrometer method (Day, 1965).

Soil pH was measured with digital pH meter potentiometerically in supernatant suspension of 1:2.5 soils to distilled water ratio (Van Reeuwijk, 1992). The cation exchange capacity (CEC) was determined by 1M ammonium acetate method at pH 7 (Chapman, 1965). Organic carbon(OC) content was determined as described by Walkley and Black (1934). Kjeldhal nitrogen in the soil was analyzed by the micro kjeldhal method (Jackson, 1958). Available P was analyzed by Olsen method (Olsen *et al*., 1954). Available S was measured in the soil by mono-calcium phosphate extraction method (Hariram and Dwivedi, 1994), and extractable B was determined using hot water method (Berger and Truog, 1939).

3.3. Description of Experimental Materials

3.3.1. Source of *Mesorhizobium* strains

Mesorhizobium strains CP-M41, CP-EAL 029, and CP M20b (on lignite based carrier) used as inoculant bio-fertilizer were obtained from Menagesha Biotechnology Industry Private Limited Company (MBI PLC), Addis Ababa (Table 1). These *Mesorhizobium* strains were selected based on their symbiotic effectiveness in chickpea and ability to enhance chickpea yield under wide ecological condition in Ethiopia (Wondwosen *et al*., 2016a; Tadele, 2017; Zehara *et al*., 2020).

Table 1: Profile of *Rhizobium* bio-fertilizer recommended for chickpea crop in Ethiopia from the research system

Name of Strain	Year of Recommendation	Maintainer*
CP M41	2016	Hawassa University/MBI
CP EAL 029	2016	NSTC
CP M20b	-	MBI/ILRI

*NSTC = National Soil Testing Centre; ILRI = International Livestock Research Institute; MBI = Menagesha Biotech Industry PLC, CP = Chickpea, EAL= Ethiopian Agricultural Legumes, M= Menagesha (Source: Gemechu *et al*., 2021)

3.3.2. Source of chickpea varieties

The improved chickpea variety Arerti (Kabuli type), and the local variety (Desi type) were used in this experiment. The seeds of the improved variety were received from Debre Zeyit Agricultural Research Center (DZARC), Ethiopia, and seeds of the local variety were obtained from the farmers.

The agronomic and phenologic characteristics of the varieties are shown in (Table 2). The improved Arerti variety, representing Kabuli type, was chosen based on higher yield, well adapted and widely grown by smallholder farmers and market preferable.

Table 2: Description of chickpea varieties to be used for the study

Variety	Chickpea type	Year of release	Adaptation /Elevation (m.a.s.l)	Maturity days	Seed color	Grain yield (t ha^{-1})	
						On farm	Potential
Arerti	Kabuli	1999	1900-2600	90-115	White	2.0-3.2	2.6-4.6
Local	Desi	-	-	-			

Sources: MoARD (2009)

3.3.3. Source of fertilizer materials

The blended fertilizer NPSB (18.9 N – 37.7 P_2O_5 – 0 K_2O - 6.95 S - 0.1 B) was used (ATA, 2016), and obtained from Plant Science department, Wolkite University, Wolkite.

3.4. Experimental Treatments, Design, and Layout

A three factor experiment was conducted in this investigation. Factorial combination of two chickpea varieties (local and Arerti), four inoculation levels (Un-inoculated, CP-M41, CP EAL 029, and CP M20b), and two levels of NPSB fertilizer (without, 0 kg ha^{-1}, and with 121 kg ha^{-1}) were laid out in factorial randomized complete block design (RCBD) with three replications. The experiment had sixteen treatment combinations (Appendex Table 1). Each replication had sixteen plots corresponds to the sixteen treatment combinations. A total of 48 plots having a dimension of 2 m × 2.4 m (4.8 m^2) were prepared. The plots were kept 1 m apart and the spacing between blocks was 1.5 m to minimize contamination (Abere *et al*., 2019). In accordance with specification of the design, treatments with in the block were assigned randomly to experimental units using lottery method. Seeds were sown with a spacing of 30 cm and 10 cm between row and plants (ATA, 2007), respectively having 8 rows per plot. The total plant population in each plot was 160 plants.

3.5. Experimental Procedures

3.5.1. Farm selection and land preparation

The experimental site with no inoculation history, cereal crops in the preceding season, flat slope (4%) and free from waterlogged condition was selected and cleaned. Before planting, the experimental field was thoroughly ploughed and levelled. Ploughing was done based on the recommendation given to the crop and following the conventional practice to make the field suitable for planting. The experimental land was leveled and divided into blocks and individual plots based on the experimental design. According to the design, a field layout was made.

3.5.2. Seed inoculation and sowing

Seed inoculation was performed using the procedure developed by Fatima *et al*. (2007) as described in Abere *et al*. (2019). Seeds were inoculated with the respective rhizobial strains just before planting. The recommended lignite based carrier- rhizobial biofertilizers at 500 g ha^{-1} was used for chickpea inoculation (Abere *et al*., 2019). To ensure the sticking of the applied inoculant to the seeds, the required quantity of seed was suspended in 1:1 ratio in 10% sugar solution. The inoculant was gently mixed with dry seeds at the rate of 10 g kg^{-1} of seed. Inoculation was done just before sowing under shade to maintain the viability of cells and allow to air dry for a few minutes and then the inoculated seeds were sown at recommended rate and spacing to the respective plots. Sowing was done by hand at about 5 cm depth on randomly allocated plots within each replication at the recommended row spacing (ATA, 2007). To avoid contamination, un-inoculated plots were sown first. Ridges were made between each plot and block to reduce the movement of bacteria and fertilizer from one plot to the other by rain.

3.6. Data Collected

3.6.1. Nodulation and symbiotic related parameters

Sampling for nodulation was performed by excavating the roots of ten randomly selected plants from one row next to the boarder row of each plot at the mid flowering stage of the crop (Abere *et al*., 2019). Uprooting was done by spade to expose the whole-root system and avoid loss of nodules and then the soil was removed from the root system by hand (Molla, 2016). The adhering soil was removed by washing the roots with intact nodules gently with tap water over a metal sieve. Nodules remaining in the soil were picked up by hand.

Nodule number per plant (NNPP): Number of nodules from ten uprooted plants per plot were counted and average was taken as number of nodules per plant (Tekalign and Asgelil, 1994).

Nodule volume per plant (**NV, ml plant^{-1}**): Nodules collected from the ten uprooted plants were immersed in to 50 ml measuring cylinder filled with water up to 30 ml. Volume of water displaced by the nodules was obtained, and the average was taken as nodule volume (ml) per plant.

Number of Effective nodules per plant: Ten representative nodules were taken from each of the ten up rooted plants and dissected with blade to observe their color at the center. The color score was made in 1-4 scale as: 1 = white, 2 = pink, 3 = slightly dark red and 4 = deep dark red (Tekalign and Asgelel, 1994).

Nodule dry weight per plant (NDWPP, g plant^{-1}**)**: Nodules collected from ten plants were oven dried at 65^0C to a constant weight, and average dried weight was recorded as nodule dry weight per plant was attained (Abere *et al*., 2019).

3.6.2. Phenological parameters

Days to 50% flowering (DTF, Days**):** Days from sowing to 50% of the chickpea in the plot produce flower was recorded as days to 50% flowering (Dejene *et al*., 2020).

Days to physiological maturity (DTM, Days**)**: It was recorded as the number of days from planting to when foliage and pod color of 90% of the plants in a plot turned yellowish (Dejene *et al*., 2020).

3.6.3. Growth parameters

Plant height (PH, cm**)**: Height, at physiological maturity, of ten randomly selected plants from central rows of each plot from the ground to the top of the main plant were measured using meter and averaged (Dejene *et al*., 2020).

Number of primary branches per plant (**NPBPP**): Average of the total number of primary branches recorded from 10 sample plants at maturity (Nuru and Kemila, 2021).

Number of secondary branches per plant (**NSBPP**): It was recorded as the averages of the number of secondary branches from 10 sample plants at maturity (Nuru and Kemila, 2021).

Shoot dry weight per plant (SHDWPP, g plant^{-1}): The ten plants used for nodule scoring at mid-flowering stage were oven dried at 65°C for 72 hours to determine this parameter by using sensitive balance (Abere *et al*., 2019).

Root dry weight per plant (RDWPP, g plant^{-1}): Roots from the ten uprooted plants used for nodule scoring were removed, oven dried at 65°C for 72 hours and weighted by using sensitive balance, then average was used as RDWPP (Abere *et al*., 2019).

3.6.4. Yield component traits

During the harvesting time, data were recorded for yield components including number of pods per plant, number of seeds per pod, and hundred seed weight for each plot.

Number of pods per plant (NPPP, number plant^{-1}**)**: It was determined as the average number of pods from ten randomly taken plants from the net plot area at physiological maturity (Dejene *et al*., 2020).

Number of seeds per pod (NSP): Total number of pods from ten randomly selected plants was threshed and numbers of seeds was counted and total numbers of seeds was divided by total number of pods to compute average number of seeds pod^{-1} (Desta and Ermias, 2019).

Hundred-seed weight (HSW, g**):** It was determined, after harvesting, by counting 100 randomly selected seeds (adjusted to 10% grain moisture content) from the net plot harvest area and weighing them with sensitive balance (Dejene *et al*., 2020).

3.6.5. Yield Parameters

Aboveground dry biomass yield (BY, kg ha^{-1}): It was measured from plants harvested from the net plot area (1.92 m^2) after sun drying to a constant weight and converted to kg ha^{-1} (Dejene *et al*., 2020).

Grain yield (GY, kg ha^{-1}**)**: It was determined by weighting grain received from the harvestable plot (1.92 m^2) area and adjusting it to 10% moisture level to give adjusted yield and converted to kg ha^{-1} (Dejene *et al*., 2020).

Yield (at 10% grain moisture) = Grains yield x (100-actual grain moisture %)/90

Straw yield (SY, kg ha^{-1}): It was calculated by subtracting grain yield from the corresponding total above ground biomass yield obtained from the net plot area.

Harvest index (HI, ratio): It was calculated as the ratio of grain yield to above ground dry biomass multiplied by 100 at harvest from the respective treatments (Dejene *et al*., 2020).

3.7. Statistical Data Analysis

The collected data were analyzed through a three-way analysis of variance (ANOVA) using general liner model (GLM procedure) in statistical analysis software (SAS) version 9.3 (SAS Institute, 2012). Wherever significant differences detected in the F- test, means were compared by using the least significance difference (LSD) test at 5% level of probability. Simple correlation analysis was conducted between yield, yield components and other relevant parameters.

3.8. Partial Budget Analysis

Economic analysis was performed to investigate the economic feasibility of the treatments. The average yield from experimental plots was adjusted downward by 15%, i.e. 10% for management difference and 5% for plot size differences, to reflect the difference between the experimental yield and the yield that farmers could expect from the same treatment (CIMMYT, 1998). Accordingly, the mean grain yields for *Mesorhizobium* strains, NPSB fertilizer, and variety treatment combinations were subjected to a discrete partial budget analysis using the procedures outlined by CIMMYT (1988). To estimate economic parameters, the variable cost of *Mesorhizobium* strains (45 ETB bag^{-1} (125g)), NPSB fertilizer (18.60 ETB kg^{-1}) and chickpea seeds (25 and 32 ETB ka^{-1} for local and Arerti, respectively) were taken by considering the price at the time of planting (September, 2021). Field price of current chickpea grain at the time of harvesting (30 and 35 ETB kg^{-1} for local and Arerti varieties, respectively) was taken from Office of Trade and Transportation marketing team of Cheha district (January to February, 2022). The price of NPSB fertilizer and *Mesorhizobium* strains were taken from Agricultural Inputs Supply Enterprise and Menagesha Biotechnology Industry PLC, respectively. All cost and benefits were calculated on hectare basis in Ethiopian Birr (ETB ha^{-1}). For a treatment to be considered as a worthwhile option to farmers, the minimum acceptable rate of return (MRR) needs to be at least between 50 and 100% (CIMMYT, 1998), So to draw farmers’ recommendations from marginal analysis in this study, 100% return to the investment was reasonable minimum acceptable rate of return.

4. RESULTS AND DISCUSSION

4.1. Physico-chemical Characteristics of Soil before Planting

Results of composite soil sample analysis of the study area indicated that the particle size distribution in the soil of the experimental site was 11% sand, 21% silt, and 68% clay. Hence, based on the soil analysis made, the soil texture of the experimental site was clay. the bulk density of the soil was 1.323 gm/cm^3, which laid in a range of moderate according to (Hunt and Gilkes, 1992). The pH of the soil was 5.7, which is moderately acidic in reaction (Tekalign *et al*, 1991). This pH value is in the optimum range for most legume crops including chickpea (Horneck *et al*. 2011), and such a pH range is conducive for biological nitrogen fixation (Jordan, 1984). The analysis for other soil chemical properties before sowing showed that the experimental soil had values of 2.1%, 44.8 cmol (+) ka^{-1}, 0.16%, 2.69 mg kg^{-1}, 6.34 mg kg^{-1}, and 0.443 mg kg^{-1} (ppm), organic carbon (OC) content, cation exchange capacity (CEC), kjeldhal nitrogen, available phosphorus, available sulfur and extractable boron, respectively (Table 3).

Thus, the organic carbon (OC) content, cation exchange capacity (CEC), total nitrogen, available phosphrous, available sulphur, and extractable boron of the experimental soil was rated as low, very high, low, very low, low and low, in accordance with Landon (1991), Hazelton and Murphy (2007), Landon (1991), Olsen *et al*. (1954), Lewis (1999), and Reisenaur et al. (1973), respectively. The low OC and low N content in the study area indicate low fertility status of the soil. This indicates that nitrogen was a limiting factor for crop growth at the study area, possibly due to continuous cultivation and lack of incorporation of organic materials, and therefore the application of nitrogen-containing fertilizer and/or inoculation with effective strains are mandatory to reduce this limiting factor of growth. Existence of low contents of available P is a common characteristic of most soils in Ethiopia (Tekalign *et al*., 1991; Yihenew, 2002; Wakene and Heluf, 2003). The low S was expected because the experimental soil had low organic matter content (source of about 95% of S) indicating that its potential to supply S to plant growth through mineralization is low. The deficiency of S in the study site could also be due to the fact that the Ethiopian agriculture mainly emphasizes on the use of high analysis NP fertilizers that contain little available S and continuous mono-cropping (Habtegebrial and Singh, 2006). Low micronutrients similar to this result (low B content) were reported by EthioSIS (2016).

Hence phosphrous, sulphur and boron containing fertilizer such as NPSB should be added to fill the crop requirement.

Table 3: Physico-chemical characteristics of surface soil before planting

Soil characters	**Unit**	**Value**	**Rating**	**References**
Bulk density	gm/cm^3	1.323	Moderate	Hunt and Gilkes, 1992
Sand	%	11	_	_
Silt	%	21	_	_
Clay	%	68	_	_
Textural class	_	Clay	_	_
PH (H_20)	_	5.7	Moderately acid	Tekalign *et al*. (1991)
Organic carbon	%	2.1	Low	Landon (1991)
CEC	cmol (+) kg^{-1}	44.8	Very high	Hazelton and Murphy (2007)
Kjeldhal nitrogen	%	0.16	Low	Landon (1991)
Available P	mg kg^{-1}	2.69	Very low	Olsen *et al*. (1954)
Available S	mg kg^{-1}	6.34	Low	Lewis (1996)
Extractable B	mg kg^{-1}	0.443	Low	Reisenauer *et al*. (1973)

4.2. Effects of *Mesorhizobium* Strains and NPSB Fertilizer on Nodulation Parameters of Chickpea Varieties

4.2.1. Number of nodules and nodule volume per plant

The analysis of variance showed that the total number of nodules and nodule volume per plant of chickpea were significantly influenced by the main effects of varieties, strains, and NPSB fertilizer, and their two-way interaction effects except for the interaction effect of variety with NPSB fertilizer. Moreover, the three-way interaction effects of varieties, strains, and NPSB fertilizer had significant effect on these parameters (Appendix Table 3). The highest nodule number (19.2) and volume (9.9 ml $plant^{-1}$) per plant of chickpea was recorded in Arerti variety with CP-M41 strain and NPSB fertilizer application followed by the use of Local variety with CP-EAL 029 strain and NPSB fertilizer application. In contrast, the lowest nodule number (10.79) and volume (4.13 ml $plant^{-1}$) was recorded when Local variety is grown without inoculation and fertilizer application (control treatment) (Table 4).

Compared to the control treatment (un-inoculated and unfertilized), nodule number and volume per plant of Ararti variety was increased by 36.61 and 51.21%, respectively due to the combined application of CP-M41 strain and NPSB fertilizer.

In this study, noticeable numbers of nodules were also observed from the control treatments, which reflected the presence of indigenous *Rhizobia* species capable of forming small sized nodules on lateral roots and most of them were ineffective (white in colour) (Table 4). However, higher number and volume of nodules due to inoculation with strains suggested that there was better synergism between introduced *Rhizobia* and chickpea plant. On the other hand, different nodulation response of the two chickpea varieties to the combined application of strains and NPSB fertilizer might be due to the presence of Cultivar-*Rhizobia* strain specificity between tested varieties and strains used (Ashenafi *et al*., 2020). This result might also be due to the fact that symbiotic effectiveness of strain to fix atmospheric N_2 varying with different genotype or may be due to varied response of cultivars to inoculums (Bayou *et al*., 2021).

The highest value of nodule number and volume due to interaction of CP-M41 strain and NPSB fertilizer application might be due to the vital role of P, S and B combined with CP-M41 strain for improving nodule formation as well as for the rhizobia bacteria to infect the roots to form nodules (Bolanos *et al*., 1996; Dotaniya *et al*., 2014; Dhage *et al*., 2014). But in CP-M20b * NPSB and CP-EAL 029 * NPSB interactions, the lower performance of both CP-M20b and CP-EAL 029 strains than CP-M41 strain might resulted significantly lower values of nodule number and volume than CP-M41 * NPSB strain interaction. The result was in agreement with (Singh *et al*., 2018; Tamiru and Girma, 2019b; Mulugeta *et al*., 2018). In addition, Merkebu (2019) reported that the diameter of nodules increased due to the combined application of NPSZn blended fertilizer with MB003 *Rhizobium* strain in Mung-bean crop. Other authors such as Kamara *et al*. (2010), Jakasaniya *et al*. (2012), Beza (2017), and Endalkachew *et al*. (2018) have also reported that combined application of inorganic fertilizers with rhizobia inoculant increased the number of nodule in chickpea.

Table 4: Three-way interaction effects of variety, *Mesorhizobium* strains, and NPSB fertilizer on number of nodules per plant (NNPP), number of effective nodules per plant (NENPP), nodule volume (NV) and nodule dry weight (NDW) of chickpea in Cheha district.

Treatments			NNPP	NV (ml plant^{-1})	NENPP	NDW (g plant^{-1})
Variety	**NPSB**	**Strains**				
Arerti	Control	Control	12.17^{g}	4.83^{j}	1.90^{g}	2.83^{i}
		CP M41	16.43^{c}	7.10^{ef}	2.57^{cd}	3.40^{ef}
		CP EAL-029	14.97^{de}	6.50^{g}	2.33^{e}	3.11^{gh}
		CP M20b	15.60^{cd}	7.00^{ef}	2.50^{de}	3.28^{fg}
	121 kg ha^{-1}	Control	15.73^{cd}	6.50^{g}	1.94^{g}	3.29^{fg}
		CP M41	19.2^{a}	9.90^{a}	3.37^{a}	4.35^{a}
		CP EAL-029	15.90^{cd}	7.33^{de}	2.53^{cd}	3.58^{de}
		CP M20b	17.53^{b}	8.37^{c}	2.87^{b}	3.87^{c}
Local	Control	Control	10.97^{h}	4.13^{k}	1.80^{gh}	2.49^{j}
		CP M41	14.00^{ef}	5.80^{h}	2.13^{f}	2.93^{hi}
		CP EAL-029	14.90^{de}	6.83^{fg}	2.46^{de}	3.21^{fg}
		CP M20b	14.40^{ef}	6.50^{g}	2.33^{e}	3.16^{g}
	121 kg ha^{-1}	Control	13.40^{f}	5.33^{i}	1.87^{g}	2.82^{i}
		CP M41	15.03^{de}	6.93^{efg}	2.46^{de}	3.22^{fg}
		CP EAL-029	17.9^{b}	9.10^{b}	3.23^{a}	4.09^{b}
		CP M20b	15.93^{cd}	7.73^{d}	2.70^{bc}	3.70^{cd}
P value			***	***	***	***
CV (%)			4.14	4.01	4.8	3.47

Interaction means within a column followed by the same letter (s) are not significantly different from each other at 5% level of significance based on Fishers LSD test; *** = significant at $P \leq 0.001$; CV= Coefficient of variation.

4.2.2. Number of effective nodules per plant

The main effect of Rhizobial strains, NPSB application, varieties and their two way interactions except for the interaction effect of variety with NPSB signigicantlly affected the number of effective nodules per plant of chickpea.

This parameter was also significantly ($P \leq 0.001$) influenced by the three-way interaction effects of varieties, *Mesorhizobium* strains, and NPSB fertilizer application (Appendix Table 3).

The highest number of effective nodule (3.37) was scored when Arerti variety inoculated with CP M41 strain with the application of NPSB fertilizer, which however was statistically at par with that obtained from the local variety inoculated with CP EAL-029 under NPSB application. The lowest value of this parameter was registered when the local variety planted without inoculation and fertilizer application (Table 4). The nodule color due to the combined application of *Mesorhizobium* strains with NPSB fertilizer on both varieties was ranged between pink and deep dark red, while white and/or green nodule color was observed on the varieties planted without inoculation and fertilizer application, which indicated the ineffectiveness of the existing native *Rhizobia* in the soil. The effective nodules of chickpea at the combined application of *Mesorhizobium* strains with NPSB fertilizer might be due to the positive influence of optimum amount of mineral NPSB fertilizer with inoculation on nodule formation and production of leghemoglobin (Nuru, 2020). This is also indicated that the added fertilizer is not fulfilling all the requirements of the crops N demand. This result is in line with the findings of Beza (2017), who reported that *Rhzobium* inoculation combined with application of inorganic fertilizers improved effective nodules on chickpea.

4.2.4. Nodule dry weight per plant

Mesorhizobium strains, varieties and NPSB fertilizer were signifcantlly affected the dry weight of nodules per plant of chickpea. Both the two- and three-way interaction effects were significantly ($P \leq 0.001$) affected this parameter (Appendix Table 3). Among the three-way interactions, variety Arerti recorded the maximum nodule dry weight of 4.35 g $plant^{-1}$ when inoculated with CP M41 strain and applied with 121 kg NPSB ha^{-1} followed by the local variety (4.09 g $plant^{-1}$) inoculated with CP EAL 029 strain and applied with 121 kg NPSB ha^{-1}. On the other hand, the minimum nodule dry weight was obtained from local variety planted without inoculation and fertilizer application (Table 4).

Inoculation of CP-M41 strain with application of NPSB fertilizer increased this parameter of Arerti chickpea variety by 8.07 and 14% as compared to interactions of CP-M20b strain * NPSB and CP-EAL 029 strain * NPSB fertilizer, respectively.

The significant variation between NPSB fertilized chickpea varieties on their dry weight of nodules due to inoculation with different *Mesorhizobium* strains might be due to the presence of Cultivar-*Rhizobia* strain specificity between tested variety and strain used (Ashenafit *et al*., 2020). Furthermore, the increased dry weight of nodules per plant on Arerti for the interaction effects of NPSB * CP-M41 strain might be due to the fact that P and S combined with rhizobia bacteria have the role to improve the leghaemoglobin contents of nodular tissue and increase in the nitrogenase activity which might in turn increase nodules size and ultimately increased the dry weight of nodules (Dutta and Bandhyopadhyay, 2009). Comparable result was also reported by (Mulugeta *et al*. 2018; Admasu, 2019).

4.3. Effects of *Mesorhizobium* Strains, Varieties and NPSB Fertilizer on Chickpea Phenological Parameters

4.3.1. Days to 50% flowering and 90% physiological maturity

Mesorhizobium strains, varieties and NPSB fertilizer application significantly affected both days to 50 % flowering and 90 % physiological maturity. However, interaction effects were not significant for both parameters (Appendix Table 4).

The result revealed that flowering and physiological maturity of variety Arerti delayed by 2.13 and 5.34 days, respectively compaired to the local variety (Table 5). This could be due to their genetic difference in response to flowering as chickpea has very high diversity in such phenological characters (Gemechu *et al*., 2012). Similarly, Tamiru and Girma (2019a) reported the longest days to reach flowering and maturity for Arerti variety of chickpea. In line with this result, Molla (2016) showed a highly significant effects of chickpea cultivars on the number of days to reach 50% flowering and 90% physiological matuirity in southern Ethiopia. Inoculation with *Mesorhizobium* strains significantly ($P \leq 0.001$) delayed both days to 50 % flowring and 90 % physiological maturity of chickpea plants. The longest day to flowering and matuirity were recorded from CP EAL 029 strain and CP M41 strain respectively, which however, was statistically at par with that scored from CP M20b strain. The lowest values of both parameters were recorded from un-inoculated plants. The delay in flowering and maturity with the *Rhizobial* inoculation might be due to the fact that inoculation enhanced N fixation and thereby increasing N uptake by plants contributed to improve vegetative growth of chickpea and delayed flowering and maturity (Admasu, 2019). This result is in agreement with the finding of Verma *et al*. (2013) who reported that seeds inoculated with rhizobia increased the days to flowering and maturity of chickpea.

Similarly, Assefa (2016) found significant effect of *Rhizobial* inoculation on days to 50 % flowering and days to 90 % maturity in chickpea varieties at Debre Zeit.

NPSB fertilizer was also significantly ($P \leq 0.01$) affected flowering and matuirity time of chickpea. Chickpea plant grown without NPSB fertilization delayed days to 50 % flowering and 90 % physiological maturity compared to NPSB applied plots (Table 5). This might be due to the fact that the importance of P found in NPSB fertilizer type for flowering and seed formation and fastening crop maturity (Kamara *et al*., 2010; Admasu, 2019). In agreement with this result, Wazir *et al*. (2018) and Admasu (2019) reported that increasing P rate decreased days to flowering and matuirity. Similarly, China (2018) observed reduced days to flowering and physiological maturity in chickpea varieties at NPSZnB applied treatments compared to the control treatment.

Table 5: Mean effects of *Mesorhizobium* inoculants, NPSB fertilizer and varieties on days to 50% flowering (DF) and 90% physiological maturity (DM) of chickpea at Cheha district.

Treatments	**DF**	**DM**
Variety		
Local	56.70^{b}	98.00^{b}
Arerti	58.83^{a}	103.37^{a}
P value	***	***
LSD (5%)	0.63	1.65
***Mesorhizobium* Strains**		
Un-inoculated	56.50^{b}	98.41^{b}
CP M41	58.17^{a}	102.16^{a}
CP EAL029	58.41^{a}	101.16^{a}
CP M20b	58.00^{a}	101.00^{a}
P value	***	**
LSD (5%)	0.90	2.34
NPSB Fertilization		
Without NPSB (0 kg ha^{-1})	58.20^{a}	102.29^{a}
With NPSB (121 kg ha^{-1})	57.33^{b}	99.08^{b}
P value	**	***
LSD (5%)	0.63	1.65
CV (%)	1.87	2.79

Main effect means within a columns followed by the same letter(s) are not significantly different from each other at P < 0.05 according to Fisheres LSD test; NS = non-significant; ** = significant at P ≤ 0.01; *** = significant at P ≤ 0.001; LSD = Least Significance Difference; CV= Coefficient of variation.

4.4. Effects of *Mesorhizobium* Strains, Varieties and NPSB Fertilizer on Growth Parameters of Chickpea

4.4.1. Plant Height

Analysis of variance revealed that the main effects of varieties, *Mesorhizobium* strains, and NPSB fertilizer application had very highly significant (P≤0.001) effect on plant height of chickpea, and also their two-way interaction effects except interaction effect of variety with NPSB had also a significant effect.

Similarly, the three-way interaction effects of *Mesorhizobium* strains, varieties and NPSB fertilizer had a highly significant ($P \leq 0.01$) effect on this parameter (Appendix Table 4).

Inoculation of *Mesorhizobium* strains with the application of NPSB fertilizer increased plant height for both chickpea varieties as compared to un-inoculated and un-fertilized plot. The longest plant height (56.4 cm) of chickpea was recorded in Arerti due to the combined application of blended NPSB fertilizer with CP-M41 strain followed by the use of Local variety with CP-EAL 029 strain and NPSB fertilizer application (49.73 cm), while the shortest value (38.05 cm) was recorded in Local variety planted without inoculation and fertilizer application (Table 6).

The longest plant height result due to interaction of NPSB * CP-M41 strain compared to un-inoculated and unfertilized treatment might be due to the enhancement of nitrogen fixation by the strain and thereby increased N uptake of the plants which might contribute to improve vegetative growth of chickpea (Admasu, 2019). Phosphorus also enhanced meristematic cell activities and early root growth leading to more absorption of other nutrients from deeper layers of soil that ultimately might increased plant height (Makoi *et al*., 2013). It might also be due to the reason that S increased meristematic tissue activity, activity of the rhizobia and nutrient (N, P and B) availability to the crop (Muhammad *et al*., 2013; Singh *et al*., 2018).

In line with the present finding, Tarekegn (2011) reported that an increased in plant height due to the interaction effect of *Rhizobium* inoculation and inorganic fertilizer application which they attributed to the increased supply of N through BNF. An increased in plant height with the interaction effect of *Rhizobium* inoculation and inorganic fertilizer on chickpea has been well documented (Kumar, 2009; Verma *et al*., 2013; Tripathi *et al*., 2013; Singh *et al*., 2018; and Admasu, 2019).

Table 6: Three-way interaction effects of varieties, *Mesorhizobium* strains, and NPSB fertilizer on plant height (PH), number of primary branches per plant (NPBPP), number of secondary branches per plant (NSBPP) and shoot dry weight (SHDW) per plant of chickpea in Cheha district.

	Treatments		**PH (cm)**	**NPBPP**	**NSBPP**	**SHDW (g plant^{-1})**
Variety	**NPSB (kg ha^{-1})**	**Strains**				
Arerti	Control	Control	42.73^{e}	2.36^{h}	7.93^{f}	34.87^{hi}
		CP M41	46.75^{bcd}	3.03^{cde}	10.43^{d}	39.94^{de}
		CP EAL-029	44.93^{cde}	2.83^{ef}	9.80^{de}	38.92^{ef}
		CP M20b	45.13^{cde}	2.90^{def}	9.93^{de}	39.68^{de}
	121	Control	44.41^{cde}	2.67^{fg}	8.90^{ef}	39.58^{de}
		CP M41	56.40^{a}	3.70^{a}	14.23^{a}	48.32^{a}
		CP EAL-029	47.60^{bc}	3.13^{cd}	11.73^{bc}	41.02^{d}
		CP M20b	49.70^{b}	3.26^{bc}	12.60^{b}	43.72^{c}
Local	Control	Control	38.05^{f}	2.06^{i}	6.63^{g}	29.89^{k}
		CP M41	42.70^{ef}	2.83^{ef}	8.83^{ef}	34.58^{ij}
		CP EAL-029	44.96^{cde}	3.10^{cd}	9.93^{de}	37.58^{fg}
		CP M20b	43.53^{def}	2.96^{de}	9.30^{e}	36.42^{gh}
	121	Control	42.96^{def}	2.56^{gh}	8.10^{f}	32.94^{j}
		CP M41	45.26^{cde}	3.10^{cd}	10.70^{cd}	38.88^{ef}
		CP EAL-029	49.73^{b}	3.50^{ab}	12.70^{b}	45.56^{b}
		CP M20b	46.06^{bcde}	3.13^{cd}	11.77^{bc}	43.08^{c}
P value			**	***	**	***
CV (%)			5.25	5.19	6.53	2.58

Means within a column followed by the same letter (s) are not significantly different from each other at 5% level of significance based on Fishers LSD test; ** = significant at $P \leq 0.01$; *** = significant at $P \leq 0.001$; CV= Coefficient of variation.

4.4.2. Number of primary and secondary branches per plant

Primary and secondary branches of chickpea were significantly affected by the main effects of *Mesorhizobium* strains, varieties and NPSB application.

The two and three-way interactions of the three factors were also significantly influenced these papamaters except the interaction between varity and NPSB fertilizer effect on secondary branches (Appendix Table 4). Both primary and secondary branches per plant were significantly ($P \leq 0.001$) influenced by the three-way interaction effects of varieties, *Mesorhizobium* strains, and blended NPSB fertilizer application. Arerti variety of chickpea inoculated with CP-M41 strain and supplied with 121 kg NPSB ha^{-1} scored the highest number of primary branches per plant (3.7) which, however, was statistically at par with that obtained from local variety inoculated with CP-EAL 029 strain under NPSB fertilizer application (3.5 branches). Similarly, the maximum number of secondary branches per plant (14.23) was registerd from Arerti variety inoculated with CP-M41 strain and supplied with 121 kg NPSB ha^{-1}. In contrast, both un-inoculated Arerti and Local chickpea varieties without NPSB application scored the lowest secondary branch per plant of 2.06 and 6.63, respectively (Table 6).

The increased number of branches due to inoculant with NPSB fertilizer application might be due to the reason that rhizobia inoculant increased the ratio of N and increased uptake of P due to the presence of sulfur (Raj *et al*., 2017). This could be due to the synergistic effect of P and S for the utilization of high quantities of nutrients through their well-developed root system and nodules formation, which might have resulted in better vegetative growth (Kumar *et al*., 2017). Moreover, this might also be due to the role of P and S to facilitate plant roots development and enhanced atmospheric nitrogen fixation and consequently branch number (Hayat *et al*., 2010). Furthermore, the application of inoculant and P might improve the vegetative growth of chickpea that may enhance N availability through biological nitrogen fixation and might be due to this fact that the crop produces most of its secondary branches during the early vegetative growth period when there was high soil nitrogen or effective nodules.

In line with this result, Togay *et al*. (2008) and Uddin *et al*. (2014) observed that the application of different doses of sulfur and phosphorus with inoculant significantly increased the number of branches in chickpea. Similarly, Kiros and Atsede (2020) described the significant effect of combined application of NPSB fertilizer with *Rhizobium* inoculation on the growth of chickpea crop in Tigray Region, Ethiopia.

4.4.3. Shoot dry weight per plant

The analysis of variance showed that shoot dry weight per plant of chickpea was significantly influenced by the main effects of *Mesorhizobium* strains, varieties and NPSB fertilizer and their two-way interaction effects except for the interaction effect of variety with NPSB. Moreover, the three-way interaction effects of *Mesorhizobium* strains, varieties and NPSB fertilizer had also a significant ($P \leq 0.001$) effect on this parameter (Appendix Table 4). The result revealed that the maximum shoot dry weight per plant of 48.32 g was obtained from Arerti variety when combined with CP-M41 strain inoculation and NPSB blend fertilizer application, whereas the lowest value of 29.89 g was recorded from un-inoculated and unfertilized Local variety (Table 6). *Mesorhizobium* inoculation with CP-M41, CP-M20b, CP-EAL 029 strains with NPSB fertilizer application increased the shoot dry weight of Arerti chickpea variety by 27.83%, 20.24% and 14.99%, respectively over un-inoculated and unfertilized Arerti, while inoculation of CP-M41, CP-M20b and CP-EAL 029 strains with the application of NPSB fertilizer on local varity resulted 23.08, 34.39 and 30.61% increase in dry biomass per plant, respectively.

This significant variation between varieties on their shoot dry matter production due to inoculation with different *Mesorhizobium* inoculants and NPSB application might be due to the existence of genetic difference between the varieties for response to both inputs. This might also be due to the fact that symbiotic effectiveness of strain to fix atmospheric N_2 varying with different genotype (Molla, 2016). Phosphorus and rhizobia inoculants have increased uptake of nutrients such as N through the BNF thereby improving N availability to plants (Ndakidemi *et al*., 2011). This was mainly due to the fact that P found in the blended NPSB fertilizer caused well-developed root system having higher nitrogen-fixing capacity and inoculant enhanced the number of rhizobia resulting the better availability of N, better growth and development of plants that ultimately increase all the growth attributes including shoot dry weight of plants (Singh *et al*., 2018). Also, the increased shoot dry weight of chickpea due to combined application of NPSB fertilizer with *Mesorhizobium* inoculant might be because S nutrient improved nitrogenase activity and N fixation that enhance vegetative growth and increased shoot dry weight (Zerihun *et al*., 2017). In agreement with the present result, Singh *et al*. (2018) reported that the combined application of nutrients with rhizobia increased shoot dry weight of chickpea over un-inoculated and unfertilized control. The result was also in agreement with the finding of Namvar and Sharifi (2011) and Sharma *et al*. (2015).

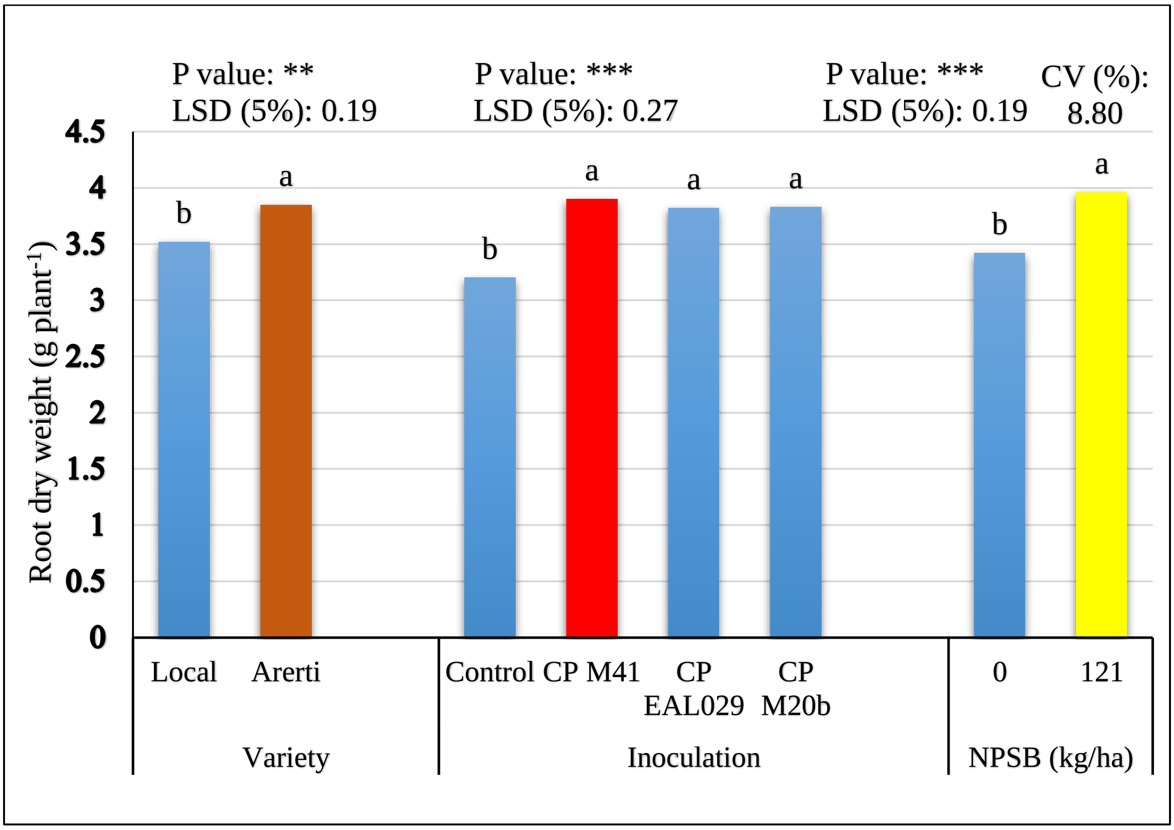
4.4.4. Root dry weight per plant

The main effects of varieties, *Mesorhizobium* strains, and NPSB application significantly influenced root dry weight per plant of chickpea. However, their two-way and three-way interaction effects were not significant for this parameter (Appendix Table 4).

Variety Arerti scored significantly ($P \leq 0.01$) highest mean dry weight of roots per plant (3.86 g) than the Local variety (3.52 g) (Figure 3). This might be due to genetic differences among the cultivars. Significant difference between chickpea varieties in terms of root dry weight per plant has been reported (Molla, 2016; Khaitov *et al*., 2020). Similarly, *Mesorhizobium* inoculation treatments significantly ($P \leq 0.001$) affected the roots dry weight per plant of chickpea. The highest root dry weight per plant (3.90 g) was obtained from CP M41 inoculated chickpea, followed by CP M20b (3.83g). On the other hand, the lowest dry weight of roots per plant (3.20 g) was recorded from un-inoculated plants (Figure 3). Chickpea inoculation with CP M41 strain increased dry weight of roots by 1.8%, 2.05%, and 17.95% over CP M20b, CP EAL 029, and un-inoculated chickpea, respectively. This study revealed that inoculation significantly improved root dry weight per plant as compared to un-inoculated treatment. This might be due to an increase in root nodulation due to *Rhizobial* inoculation that resulted in N availability in soil and enhanced growth and development of plants hence increased root dry weight (Admasu, 2019). Moreover, the highest root dry weight from CP M41 might be due to the its higher N fixing capacity (Wondwosen *et al*., 2016a). Similarly, Tamiru and Girma. (2019b)) had also observed that inoculation with rhizobial inoculants increased root dry weight of chickpea.

Regarding the NPSB fertilizer treatments, the result in Figure 3 indicate that NPSB fertilizer treatments significantly ($P \leq 0.001$) increased chickpea root dry weight per plant by 13.64% over unfertilized treatment. The increased root dry weight with NPSB fertilizer application could be because of P that required for root development and growth (Admasu, 2019). Phosphorus is known to initiate nodule formation as well as influence the efficiency of the rhizobium-legume symbiosis thereby enhancing nitrogen fixation and consequently root biomass (Haruna and Aliyu, 2011). The highest value of root dry weight of chickpea with application of blended NPSB fertilizer might also be due to that application of S found in NPSB enhanced N and P uptake, stimulate photosynthetic activity and synthesis of chloroplast protein resulting in higher root dry matter production (Merkebu, 2019).

In line with this result, Merkebu (2019) reported that application of NPSB fertilizer increased the root dry weight of Mung bean crop by 41.46% over the control treatment.



Main effect means of each factor within the bar graph followed by the same letter (s) are not significantly different from each other at 5% level of significance based on Fishers LSD test; ** = significant at $P \leq 0.01$; *** = significant at $P \leq 0.001$; LSD= least significance difference, CV= Coefficient of variation.

Figure 3: Root dry weight of chickpea as influenced by Varieties, *Mesorhizobium* strains, and NPSB fertilizer application

4.5. Effects of *Mesorhizobium* Strains, Varieties and NPSB Fertilizer on Yield Components of Chickpea

4.5.1. Total number of pods per plant

Pod number per plant of chickpea was significantly affected by the main effects of *Mesorhizobium* strains, varieties and NPSB application, their two and three-way ($p \leq 0.01$) interaction (Appendix Table 5).

As indicated in Table 7, the maximum pod number per plant (84.6) was obtained from CP-M41 inoculated Arerti variety under NPSB fertilizer application. In contrast, the lowest pod number per plant (54.41) was obtained from the Local variety planted without inoculation and NPSB fertilizer application. Arerti variety inoculated with CP-M41 with NPSB application scored almost 8% higher pod number than the local varity recived the same inoculant and fertilizer. This might be attributed to relatively higher nutrient uptake and/or utilization efficiency of Arerti variety than local variety. Moreover, the highest number of pods per plant from combined effect of NPSB * CP-M41 on Arerti might be due to the important role of S in the growth and development of plants including chlorophyll and nitrogenase formation, promotes nodule formation and enzyme activation (Fageria, 2009), and due to higher N fixing performance of CP-M41 strain than CP-EAL 029 and CP-M20b strains.

It might also be due to adequate availability of N through BNF and P that might have improved the production of primary branches, secondary branches and plant height that could in turn have contributed for the production of the higher number of total pods. This might be supported by positive correlation of primary branches, secondary branches and plant height with pod number (Table 9). The positive effect of N and P on the production of chickpea pods might be attributed to the merit of the nutrient in promoting of both vegetative and reproductive, thereby improving the photosynthetic efficiency and partitioning of carbohydrate that in turn had increased number of pods per plant (Kumar *et al*., 2017; Admasu, 2019). On the other hand, S might have played a pivotal role in the regulation of the metabolic and enzymatic processes resulting in increased yield attributes of chickpea (Favorite *et al*., 2012). The increased number of pods per plant from NPSB fertilizer with *Rhizobial* application might also be because of B that increased the pollen producing capacity and pollen tube growth in legumes (Verma *et al*., 2004). The increment of pods per plant with NPSB fertilizer application might be due to adequate supply of nutrients in NPSB fertilizer for nodule formation, protein synthesis, fruiting, and seed formation (Nuru, 2020).

The current result is in line with the findings of Mulugeta *et al*. (2018); Sharma *et al*. (2015); Kumar *et al*. (2017; and Meena *et al*. (2013). Similarly, Kiros and Atsede (2020) also indicated that the combined application of NPSB fertilizer with *Rhizobium* inoculation increased the number of pods per plant as compared to the control.

Table 7: Three-way interaction effects of varieties, *Mesorhizobium* strains, and NPSB fertilizer on number of pods per plant (NPPP), number of seeds per pod (NSP) and hundred seed weight (HSW) per plant of chickpea in Cheha district.

Treatments			NPPP	NSP	HSW (g plant^{-1})
Variety	**NPSB (kg ha^{-1})**	**Strains**			
Arerti	Control	Control	58.73^{ij}	1.06^{hi}	21.28^{h}
		CP M41	66.86^{ef}	1.103^{def}	26.61^{c}
		CP EAL-029	63.26^{fgh}	1.08^{gh}	25.12^{def}
		CP M20b	64.80^{efgh}	1.086^{fg}	25.50^{cde}
	121	Control	61.80^{hi}	1.086^{fg}	24.21^{fg}
		CP M41	84.60^{a}	1.213^{a}	31.14^{a}
		CP EAL-029	71.50^{cd}	1.117^{cd}	28.22^{b}
		CP M20b	74.21^{bc}	1.133^{bc}	29.12^{b}
Local	Control	Control	54.41^{k}	1.03^{j}	19.16^{i}
		CP M41	60.81^{hij}	1.063^{hi}	23.02^{g}
		CP EAL-029	66.46^{efg}	1.09^{efg}	24.62^{ef}
		CP M20b	62.48^{ghi}	1.07^{ghi}	23.35^{g}
	121	Control	57.16^{jk}	1.056^{i}	21.32^{h}
		CP M41	65.96^{efg}	1.09^{efg}	25.79^{cde}
		CP EAL-029	77.36^{b}	1.153^{b}	28.02^{b}
		CP M20b	68.33^{de}	1.11^{de}	26.30^{cd}
P value			**	***	*
CV (%)			3.62	1.12	2.88

Interaction means within a column followed by the same letter (s) are not significantly different from each other at 5% level of significance based on Fishers LSD test; * = significant at P ≤ 0.05; ** = significant at P ≤ 0.01; *** = significant at P ≤ 0.001; CV= Coefficient of variation.

4.5.3. Number of seeds per pod

Analysis of variance showed that seed number per pod of chickpea was significantly affected by the main effects of *Mesorhizobium* strains, varieties and NPSB fertilizer application, their two and three-way ($p \leq 0.001$) interaction effects (Appendix Table 5).

As shown in Table 7, significantly highest number of seeds per pod (93.26) was observed from the interaction effect of NPSB fertilization (121 kg ha^{-1}) with CP-M41 strain for variety Arerti while the lowest number of seeds per pod (57.08) was from Local variety planted without inoculation and NPSB fertilizer application. This might be due to the fact that adequate supply of nutrients from NPSB fertilizer combined with inoculant might be the reason for improved nodule formation, protein synthesis, fruit and seed formation (Nuru, 2020).

The number of seeds per pod of Arerti chickpea variety was increased by 12.61% due to the interaction of NPSB blended fertilizers with CP-M41 inoculant as compared to the control. The increment of seeds per pods at the combined application of NPSB with CP-M41 inoculant on variety Arerti could be because of inoculation provides an adequate supply of N and P for plant, P caused well-developed root system having higher nitrogen-fixing capacity resulting in better growth, development of plants and better diversion of photosynthates towards sink (Singh *et al*., 2018), and B improved biomass production and seed formation in pulses (Renukadvi *et al*., 2002). In addition, the possible reason for increased seeds per pod might be due to interaction effect of P and *Rhizobial* inoculants that resulted in more availability of nutrients that contributed to root improvement, stem vigor, flower and seed formation thereby increasing the number of the seeds (Endalkachew *et al*., 2018). Moreover, this might be due to the fact that P and N playing important role translocation of photosynthetic products from vegetative growth to reproductive growth ultimately increase grain per pod (Admasu, 2019).

The result of the present study was in conformity with Endalkachew *et al*. (2018) and Admasu (2019) who reported that the combined application of P containing fertilizer with inoculant significantly increased the number of grains per pod in chickpea. Similarly, Malhur *et al*. (2003), Basir *et al*. (2008), Togay *et al*. (2008) and Reddy (2012) also reported that interaction of rhizobia inoculant with inorganic fertilizer increased the number of grains per pod compare with individual application in chickpea.

4.5.4. Hundred-seed weight (HSW)

Mesorhizobium strains, varieties, NPSB fertilizer application, their two and three-way interaction effects significantly influenced hundred-seed weight (HSW) of chickpea (Appendix Table 5).

Analysis of data indicated that the three-way interaction effects of *Mesorhizobium* strains, NPSB fertilizer, and varieties significantly ($P \leq 0.05$) influenced the HSW of chickpea (Table 7). The highest (31.14 g) and the lowest (19.16 g) hundred-grain weight were obtained from variety Arerti at the combined application of NPSB (121 kg ha^{-1}) with CP-M41 strain and variety Local at the control treatment, respectively. M*esorhizobium* inoculation with CP-M41 strain with NPSB fertilizer application significantly increased the HSW in Arerti variety by 14.54%, 22.25%, and 31.66% as compared with the individual effects of CP-M41 strain without fertilizer, NPSB fertilized plants without inoculation, and the control treatment, respectively. The maximum hundred-grain weight recorded in the combined application of NPSB and inoculant might be due to the supply of nitrogen through BNF and the cumulative effect of phosphorus and sulphur on the processes of cell division and help as balanced nutrition. Then it plays an important role in improving growth and assimilates accumulations, thereby increase the reproductive performance of the plants, which led to larger seed production (Tarekegn, 2011).

The resulting increased N availability through inoculant might have promoted the supply of assimilates to seed thereby enabling them to gain more weight (Wazier *et al*., 2018). The result showed that the highest hundred-grain weight might have obtained from the synergistic effect of P and S on the growth processes of the plant (Singh *et al*., 2011). The synergetic effect of P and S could allow the utilization of high quantities of nutrients through their well-developed root system and nodules, which might have resulted in increases vegetative and reproductive growth as a result the supply of dry matter to grain that ultimately increased the weight of grain. This result could also be due to the vital role of P and S for plant metabolism and thier positive effect on the nodulation of plants. The better growth and development of crop plants due to P and S supply and N uptake might have increased the supply of assimilates to seed, which ultimately gained more weight (Patil *et al*., 2011). Similarly, Kiros and Atsede (2020) reported that the combined application of rhizobia inoculant with NPSB fertilizer increased hundred-seed weight on chickpea.

Other authors, Uddin *et al*. (2018), Mulugeta *et al*. (2018) and Wazier *et al*. (2018) have observed that the combined application of inoculant and inorganic fertilizer increased grain weight in chickpea. The finding of this study is also in confirmation with the findings of Habtamu (2015), Nazmun *et al*. (2009), Sadeghipour *et al*. (2010) and Yin *et al*. (2018).

Hossain *et al*. (2011) and Merkebu (2019) reported that combination of *Rhizobia* inoculum and inorganic nutrients (N, P, S, Zn and K) showed significant effects on seed weight of mung bean. Moreover, Islam *et al*., (2017) studied that interaction of S and B level had significant influence in respect of 100-grains weight of mung bean. Significance difference was found by Habtamu (2015) on 100- seed weight of legumes due to combined application of NP fertilizer with inoculants. On another study, Beza (2017) described that the combined application of *Rhizobium* inoculation with inorganic fertilizers is significant on 100-seed weight of chickpea.

4.6. Effects of *Mesorhizobium* Strains, Varieties and NPSB Fertilizer on Yield Traits of Chickpea

4.6.1. Aboveground dry biomass yield

The productivity of a crop is largely determined by the biological yield. Production of large amount of biomass is among the attributes of seed yield. Increase in dry matter accumulation is one of the criterions of crop growth (Tesfahun *et al*., 2018). The analysis of variance showed that the total above ground dry biomass yield of chickpea was significantly affected by the main effects of *Mesorhizobium* strains, varieties and NPSB fertilizer application, and their two-way interaction effects (except interaction effect of *Mesorhizobium* strains and NPSB fertilizer application) as well as their three-way interaction effects (Appendix Table 6).

As indicated in Table 8, the total above ground dry biomass yield of chickpea was significantly ($P \leq 0.01$) influenced by the interaction effect of chickpea varieties and combined application of *Mesorhizobium* strains with NPSB fertilizer. The highest total above ground dry biomass (5924.33 kg ha^{-1}) was obtained from variety Arerti at the combination of NPSB fertilization with CP-M41 *Mesorhizobium* strain. In contrast, the lowest total above ground dry biomass yield of 3726.83 kg ha^{-1} was obtained from the Local variety planted without inoculation and fertilizer application. Arerti variety grown under the combined application of CP M41 strain and blended NPSB fertilizer increased the above ground biomass yield of chickpea by 37.09% as compared to local variety grown without inoculation and NPSB fertilization.

The result also indicated that *Mesorhizobium* inoculation with CP-M41 strain, CP-M20b strain, CP-EAL 029 strain, and NPSB fertilizer application increased the total above ground dry biomass yield of Arerti chickpea variety by 15.42%, 13.53%, 12.69% and 12.46% over the control check, while the interactions of CP-M41 strain with NPSB fertilizer, CP-M20b * NPSB, and CP-EAL 029 * NPSB resulted in a 27.58%, 21.17%, and 18.01% increase, respectively. Moreover, CP-M41 strain inoculation integrated with NPSB fertilizer application increased the total above ground dry biomass yield of Arerti chickpea variety by 8.12 and 11.66% as compared to interactions of CP-M20b strain * NPSB and CP-EAL 029 strain * NPSB fertilizer, respectively.The observed variation between varieties on their dry matter production due to inoculation with different strains might be due to presence of variety-strain specificity between tested variety and strain used (Molla, 2016).

The increment in dry matter yield with application of NPSB fertilizer might be due to the adequate supply of nutrients that could have increased the number of branches per plant, and leaf area which in turn increased photosynthetic area and number of pods per plant thereby dry matter accumulation (Nuru, 2020). The increased total dry biomass yield due to interaction of CP-M41 strain with NPSB fertilizer application could be because of the fact that the combined application NPSB and CP-M41 rhizobia inoculant significantly increased nodulation and improves vegetative growth and development of plants, which leads to increased dry matter yield comparatively to the single use of NPSB or inoculant (Akpalu *et al*., 2014). The observed total dry biomass yield improvements could also be due to the increased N, P, S and B availability and are in line with the improvements observed for the growth related traits such as plant height, number of branches and shoot dry weight. But, the lower performance of CP-M20b and CP-EAL 029 strains than CP-M41 strain might resulted lower total biomass yield. Comparable results were reported by Chandra and Khaldelwal (2009), Shiri-Janagard *et al*. (2012) and Admasu (2019).

Table 8: Three-way interaction effects of varieties, *Mesorhizobium* strains, and NPSB fertilizer on grain yield (GY), aboveground dry biomass yield (BY), straw yield (SY), and harvest index (HI) of chickpea in Cheha district.

Treatments			**BY (kg ha^{-1})**	**GY (kg ha^{-1})**	**SY (kg ha^{-1})**	**HI (ratio)**
Variety	**NPSB (kg ha^{-1})**	**Strains**				
Arerti	Control	Control	4290.41^{f}	1971.87^{h}	2318.54^{efghi}	0.46^{g}
		CP M41	5073.10^{cd}	2669.88^{d}	2403.22^{def}	0.52^{cd}
		CP EAL-029	4914.05^{d}	2523.69^{e}	2390.36^{defg}	0.51^{e}
		CP M20b	4961.84^{d}	2580.92^{e}	2380.92^{defgh}	0.52^{cd}
	121	Control	4901.44^{d}	2412.72^{f}	2488.72^{bcd}	0.49^{f}
		CP M41	5924.33^{a}	3177.16^{a}	2747.17^{a}	0.55^{a}
		CP EAL-029	5233.24^{c}	2816.62^{c}	2416.12^{cde}	0.53^{b}
		CP M20b	5442.69^{b}	2890.68^{bc}	2552.01^{b}	0.53^{b}
Local	Control	Control	3726.82^{g}	1693.91^{i}	2032.91^{k}	0.45^{h}
		CP M41	4615.09^{e}	2405.88^{f}	2209.21^{ij}	0.52^{cd}
		CP EAL-029	4905.84^{d}	2620.42^{d}	2285.42^{ghij}	0.53^{b}
		CP M20b	4655.7^{e}	2427.85^{f}	2227.85^{ij}	0.52^{cd}
	121	Control	4264.66^{f}	2115.66^{g}	2149.00^{jk}	0.49^{f}
		CP M41	5130.43^{c}	2720.05^{d}	2410.38^{hij}	0.53^{b}
		CP EAL-029	5486.71^{b}	2952.19^{b}	2534.52^{bc}	0.53^{b}
		CP M20b	5150.51^{c}	2727.42^{d}	2423.09^{fghi}	0.53^{b}
P value			**	**	**	**
CV (%)			2.25	1.93	3.22	1.36

Interaction means within a column followed by the same letter (s) are not significantly different from each other at 5% level of significance based on Fishers LSD test; ** = significant at $P \leq 0.01$; CV= Coefficient of variation.

4.6.2. Grain yield

Dry matter production and its transformation into economic yield is the ultimate outcome of various physiological, biochemical, phonological and morphological events occurring in the plant system.

Seed yield of a variety is the result of interplay of its genetic makeup and environmental factors in which plant grow (Tamiru *et al*., 2021). In other words, yield depends, directly or indirectly, on the performances of other related traits. Therefore, reductions in the performances of other traits result in decrease in seed yield (Tesfahun *et al*., 2018). The analysis of variance showed that the grain yield of chickpea was significantly affected by the main effects of *Mesorhizobium* strains, varieties and NPSB fertilizer application, and their two-way and three-way interaction effects (Appendix Table 6).

The three-way interaction among varieties, *Mesorhizobium* strains and NPSB fertilizer application had significant ($P \leq 0.01$) effect on the grain yield of chickpea (Table 8). Among the interaction, the highest grain yield (3177.16 kg ha^{-1}) was recorded from Arerti variety in combined application of CP-M41 strain with NPSB fertilizer at the recommended rate of 121 kg ha^{-1} followed by the use of Local variety with combination of CP-EAL 029 strain with NPSB fertilizer (2952.19 kg ha^{-1}), while the lowest grain yield (1693.91 kg ha^{-1}) was recorded from variety Local when grown without inculation and fertilizer application. The result showed planting of Arerti variety with CP M41 strain and NPSB application increased the grain yield of chickpea by 46.68% as compared to growing of local variety without inoculation and fertilization. The result also showed that *Mesorhizobium* inoculation with CP-M41 strain and NPSB fertilizer application increased the grain yield of Arerti variety by 26.14%, and 18.27% over the control check respectively, while the interactions of CP-M41 strain with NPSB fertilizer resulted in a 37.93% increase. Moreover, CP-M41 strain inoculation integrated with NPSB fertilizer application increased the grain yield of Arerti chickpea variety by 9.01 and 11.34% as compared to interactions of CP-M20b strain * NPSB and CP-EAL 029 strain * NPSB fertilizer, respectively.

The highest grain yield result as a result of interaction effects of CP-M41 strain and NPSB blend fertilizer application might be due to the vital role of N, P, S, B nutrients combined with CP-M41 strain for improving plant growth, biomass production, nodule formation and development, N_2 fixation and yield contributing characters (Renukadevi *et al*., 2002; Aslam *et al*., 2010; Sadeghipour *et al*., 2010). This might have resulted from the positive effects of P, S and B on the process of nitrogen fixation where the increased supply of N through inoculation resulted in enhanced plant growth that eventually leads to higher yield (Singh *et al*., 2018; Beza, 2017 and Kiros and Atsed, 2020).

But, in CP-M20b *NPSB and CP-EAL 029 * NPSB interactions, the lower performance of both CP-M20b and CP-EAL 029 strains than CP-M41 strain might resulted significantly lower grain yield values than CP-M41 * NPSB strain interaction.

This result is in line with Mulugeta *et al*. (2018) who reported that the combined application of *Mesorhizobium* inoculant with P significantly increased grain yield on chickpea. Endalkachew *et al*. (2018) also reported that rhizobia inoculant with P in chickpea plants significantly increased grain yield comparable to the individual use of P or rhizobia inoculant. Similarly, Kiros and Atsede (2020) indicated that application of NPSB fertilizer and *Rhizobium* inoculants at the same time had synergistic effect on the yield of chickpea that increased the yield by 34% over the untreated check. Other authors, Meena *et al*. (2013), Sharma *et al*. (2015) and Das *et al*. (2016) have observed that the combined application of P and S increased grain yield of chickpea.

4.6.3. Straw yield

The straw yield of chickpea was significantly ($P \leq 0.001$) influenced by the main effect of *Mesorhizobium* strains, NPSB application, and varieties. Their two-way interaction effect of variety with strain and three-way interaction were also significant for this parameter. However, their two-way interaction effects of variety with NPSB and strain with NPSB did not significantly affect this parameter (Appendix Table 6). The three-way interaction among varieties, *Mesorhizobium* strains and NPSB application had significant ($P \leq 0.01$) effect on the straw yield of chickpea (Table 8). The highest straw yield (2747.16 kg ha^{-1}) was recorded when Arerti variety inoculated with CP M41 strain with the application of NPSB fertilizer, while the lowest straw yield (2032.91 kg ha^{-1}) was obtained from the local variety when planted without inoculation and fertilizer application (Table 9). *Mesorhizobium* inoculation with CP-M41 strain and NPSB application increased the straw yield of Arerti variety by 3.52 % and 6.83 % over that of the control respectively, while the combined use of CP-M41 strain with NPSB fertilizer resulted in a 15.6 % increase.

The result indicated that the combined application NPSB and rhizobia inoculant in legume plants significantly increased nodulation and improves vegetative growth and development of plants, which leads to increased straw yield comparatively to the single use of NPSB or inoculant (Akpalu *et al*., 2014; Beza, 2017, Endalkachew *et al*., 2018).

The observed straw yield improvements are due to the increased N, P, S and B availability and are in line with the improvements observed for the growth related traits such as plant height, number of branches and shoot dry weight. The combined application of P, S and B with inoculant increased plant height, number of branches per plant, nodules per plant, nodule dry weight and straw yield in chickpea (Parkash *et al*., 2017, Admasu, 2019). This might be due to P is important for chlorophyll synthesis, enhances nodule formation and nitrogen availability to plant that improves plant growth like the number of branches and plant height in turn resulted in higher straw yield. An increase in the N supply not only stimulates growth but also changes the morphology of the plant, which ultimately increases the straw yield. The application of S increased the availability of N, which has important function to improve the vegetative growth that resulted in straw yield increment (Admasu, 2019).

Similarly, Wazier *et al*. (2018) also reported that the straw yield increased at the combined application of P fertilizer with inoculant in chickpea plant. In addition, Fatima *et al*. (2007) reported that the combined application of inoculation and P increased cowpea straw yield by 63% over that of the control. Other authors such as Ibsa (2013) and Sheleme *et al*. (2014) described that the application of specific rhizobium inoculums (Bio-fertilizer) along with application of chemical fertilizer improved straw yield of chickpea as compared to the control and the single use of inoculant and chemical fertilizer.

4.6.4. Harvest index

Harvest index (HI) is a measure of physiological productivity potential of a crop or variety. It is the ability of a crop to convert the dry matter into economic yield. It is very useful in measuring nutrient partitioning in crop plants, which provides an indication of how efficiently the plant acquired nutrients are partitioned for grain production. The higher the harvest index value, the more the production efficiency and vice versa. So the highest harvest index also implies higher partitioning of dry matter into grain. Thus, selection for varieties having high seed to biomass ration (HI) could enhance chickpea yield (Assefa, 2016; Tesfahun *et al*., 2018).

The analysis of variance showed that the HI of chickpea was significantly affected by the main effects of *Mesorhizobium* strains, varieties and NPSB fertilizer application, and their two-way interaction effects of variety with strains and NPSB application as well as their three-way interaction effects.

However, the two-way interaction effect of inoculation with *Mesorhizobium* strains and NPSB fertilizer application did not significantly affect this parameter (Appendix Table 6).

As shown in Table 8, the HI of chickpea was significantly ($P \leq 0.01$) influenced by the three-way interaction effect of varieties, *Mesorhizobium* strains and NPSB application. Significantly the highest harvest index (0.55) was observed from the interaction effect of NPSB fertilization with CP-M41 strain and for variety Arerti while the lowest HI (0.45) was for variety Local when grown without inoculant and fertilizer application. The growing of Arerti chickpea variety with combined application of CP M41 strain and blended NPSB fertilizer increased the HI of chickpea by 18.18% over that of local variety grown without inoculation and fertilization. Moreover, *Mesorhizobium* inoculation with CP M41 strain and NPSB application increased the HI value of Arerti variety by 11.32 % and 6 % over that of the control respectively, while the combined use of CP-M41 strain with NPSB fertilizer resulted in a 20.34 % increase.

The highest harvest index due to interaction of NPSB with CP-M41 inoculant might be due to the fact that P application resulted the highest biological yield coupled with the highest uptake of P as a result that P translocated higher photosynthetic product to the seed than the straw ultimately increases grain yield (Admasu, 2019). The increment in harvest index by NPSB fertilizer combined with *Rhizobial* inoculant might be ascribed to greater photo-assimilates production and its ultimate partitioning to the grains compared to the partition to the straw (Zelalem, 2018). Moreover, inoculation increased the number of effective nodules per plant and N availability that in turn enhanced dry matter partitioning in favor of grain showing a greater harvest index, and application of balanced NPSB nutrients increased hundred grains weight of chickpea crop (Endalkachew *et al*., 2018; Nuru, 2020). Previous research findings also indicated the positive role of *Mesorhizobium inoculation*, N, P, S and B nutrient application in increasing harvest index of chickpea crop (Valenciano *et al*., 2011; Sheleme *et al*., 2014; Beza, 2017). Similarly, Zelalem (2018) reported that lentil crop supplied with NPSZnB with *Rhizobium* inoculants produced significantly highest harvest index compared to the control.

4.7. Correlation Analysis

The correlation analysis of nodulation, growth, yield components, and yields of chickpea indicated strongly and very highly significant positive correlation among nodulation, grain yield and yield traits of chickpea (Table 9). Nodulation parameters such as total and effective nodule number, nodule volume and nodule dry weight were strongly correlated with all of the grain yield components. Growth parameters like plant height, number of branches, shoot dry weight and root dry weight were also strongly correlated with all of the yield components. This strong correlation indicated that nodulation contributes better to attained more nutrient availability, resulting in vigorous plant growth and dry matter accumulation, which in turn resulted in higher grain yield. Similarly, above ground dry biomass yield revealed very highly positive association with grain yield (r=0.97), implies the higher above ground biomass predicted superior yield behind positive response of inoculation. Number of pods per plant was much correlated with grain yield (r = 0.89), and grain yield had strong and positive very highly significant association with number of seeds per pod. The correlation results also indicate that straw yield, hundred seed weight and harvest index were more closely related to the different nodulation and yield parameters including seed yield. This indicated that the grain yield of crop is a dependent variable that is significantly and positively affected by all other growth and yield contributing components (Gomez and Gomez, 1984; Zelalem, 2018).

The positive and significant correlation result among all the nodule characteristics, growth and yield component parameters indicated that the development of promising nodules of the crop, and the improvement of growth traits due to supply of NPSB fertilizers and *Mesorhizobium* inoculation could promote nutrient uptake through the process of BNF that ultimately enhance the final grain yield and yield attributes of the crop (Zelalem, 2018). In this regard, the strong association between nodulation and grain yield are useful in selecting *Rhizobia* as inoculants to promote nodulation, plant growth and seed yields in enhancing grain yield of chickpea. Similar result was reported that nodulation traits, growth parameters, yield components and yield traits of chickpea, faba-bean, mung-bean, lentil and other pulse crop strongly and positively associated with grain yield, and hence, contributed to the maximum yield of crops (Delic *et al*., 2011; Yirga *et al*., 2013; Wondwosen *et al*., 2017; Zelalem, 2018).

Table 9: Relationship among nodulation, growth, grain yield and yield traits of chickpea crop

	TNN	ENN	NV	NDW	SHDW	PHPL	NPB	NSB	NPPL	NSP	DBY	SY	HSW	HI	GY
TNN	1														
ENN	0.93***	1													
NV	0.91***	0.94***	1												
NDW	0.92***	0.95***	0.94***	1											
SHDW	0.91***	0.9***	0.95***	0.93***	1										
PHPL	0.76***	0.76***	0.83***	0.76***	0.8***	1									
NPB	0.8***	0.82***	0.92***	0.82***	0.85***	0.79***	1								
NSB	0.88***	0.92***	0.91***	0.94***	0.9***	0.74***	0.81***	1							
NPPL	0.85***	0.9***	0.93***	0.91***	0.91***	0.84***	0.87***	0.9***	1						
NSP	0.89***	0.93***	0.91***	0.94***	0.88***	0.81***	0.8***	0.89***	0.91***	1					
DBY	0.94***	0.9***	0.94***	0.91***	0.93***	0.8***	0.87***	0.89***	0.9***	0.9***	1				
SY	0.83***	0.78***	0.78***	0.79***	0.82***	0.49***	0.72***	0.71***	0.63***	0.77***	0.88***	1			
HSW	0.9***	0.89***	0.93***	0.92***	0.92***	0.74***	0.86***	0.92***	0.9***	0.89***	0.94***	0.79***	1		
HI	0.89***	0.9***	0.93***	0.9***	0.85***	0.75***	0.91***	0.91***	0.87***	0.84***	0.9***	0.64***	0.91***	1	
GY	0.91***	0.89***	0.94***	0.89***	0.91***	0.77***	0.92***	0.89***	0.89***	0.86***	0.97***	0.75***	0.93***	0.95***	1

TNN = Total nodule number; ENN = Effective nodules number; NV = Nodule volume; NDW = Nodule dry weight; SHDW = Shoot dry weight; PHPL = Plant height per plant; NPB = Number of primary branch; NSB = Number of secondary branch; NPPL = Number of pods per plant; NSP = Number of seeds per pod; DBY = Dry biomass yield; SY = Straw yield; HSW = Hundred seed weight; HI = Harvest index; GY = Grain yield. *** = Very highly significant correlation.

4.8. Partial Budget Analysis of Treatment Effects

Data presented in Table 10 indicated the economic analysis of chickpea as affected by different varieties, *Mesorhizobium* strains, and NPSB application. From the use of various interactions of different varieties with *Mesorhizobium* strains and NPSB fertilizer, the total variable cost and net benefits were estimated. The total variable cost (TVC) was estimated during field experimental period that was included cost of seeds, fertilizers and *Mesorhizobium* strains as well as cost of labor for fertilizers and *Mesorhizobium* strains applications (Table 10). Daily labor cost during experimental period was 150 ETB per person per day and the field price of chickpea yield during harvesting period was 30 ETB kg^{-1} (for Local variety) and 35 ETB kg^{-1} (for Arerti variety). The total variable cost (TVC) was subtracted from gross benefit (GB) to obtain net benefits (NB). In most cases, farmers prefer the highest profit (low cost with high income). For this purpose, it is necessary to conduct dominance analysis. A dominated treatment is any treatment that has net benefits that are less than or equal to those of a treatment with lower costs that vary (CIMMYT, 1988). The dominance analysis procedure as detailed in CIMMYT (1998) was used to select potentially profitable treatments from the range that was tested and serve to eliminate some of the treatments from further consideration and thereby simplify the analysis. Therefore, after estimating net benefits, the treatments were arranged in ascending or increasing order of total variable costs to identify dominated and non-dominated treatments (Table 11).

According to the dominance analysis, from sixteen (16) treatments, eleven were dominated and five treatments were non-dominated (Table 11). Untreated control treatment showed the least variable cost (2,500.00 birr) and the treatment with Arerti variety at NPSB and inoculants application showed the maximum variable cost (5930.6 birr), and all the remaining treatments were confined between these treatments (Table 11). The dominated treatments have high total cost of variable but lower net benefit, and hence eliminated for further economic analysis. Though, the rest of treatment had both higher variable cost and net benefit, hence not dominated and was considered for marginal rate of return. The Un-dominated treatments were ranked from the lowest (the farmers' practice) to the highest costs that vary. For each pair of ranked treatments, a % marginal rate of return (MRR) was calculated to obtain an estimate of the returns per unit of investment in fertilizer and inoculants since the % MRR between any pair of un-dominated treatments denotes the return per unit of investment in fertilizer and inoculants expressed as a percentage.

The marginal rate of return indicates what farmers can expect to gain, on the average, in return for their investment when they decide to change from one practice (or set of practices) to another (CIMMYT, 1988).

According to the economic analysis data, all marginal rates were above 100% that is in a range of acceptance (CIMMYT, 1988). According to CIMMYT (1988), treatments with high net benefit, relatively low variable cost together with an acceptable and maximum MRR becomes the tentative economically preferable treatments. Thus, although Local variety with CP-M41 *Mesorhizobium* inoculation ranked first (followed by Arerti varity with inoculated with the same strain) among the treatments with the highest marginal rate of return in percent, the benefit obtained was lower than the treatment in the combined use of Arerti variety with CP-M41 *Mesorhizobium* inoculant and NPSB application.

Therefore, from the budget summary of economic analysis, the highest net return (102092.6 ETB ha^{-1}) with acceptable marginal rate of return (618%) was obtained from Arerti variety grown with CP-M41 *Mesorhizobium* strain and 121 kg NPSB ha^{-1} application. This implies that the grower in the study area can gate additional benefit of 6.18 ETB/ha for every 1 ETB expense by changing the current practice and adopting the new treatment, followed by Arerti variety with *Mesorhizobium* CP-M41 inoculation and no supply of NPSB blend fertilizer having calculated net return of (87245.6 ETB ha^{-1}), while the lowest net economic return (40694.6) was recorded from Local variety planted without inoculation and NPSB fertilizer application (Table 12). Therefore, the use of Arerti variety with *Mesorhizobium* inoculation with strain CP-M41 and 121 kg NPSB ha^{-1} application found to be economically feasible at Cheha area, Gurage zone, SNNPRS of Ethiopia. In line with this finding, Kiros and Atsede (2020) reported that planting of the cultivar Arerti with blended NPSB application and *Rhizobia* inoculation produced the highest net benefit (67132.2025 ETB ha^{-1}) with acceptable marginal rate of return (4106.48 %) compared to other treatments at Hatsebo research site in Laelay maichew district, Tigray regional state, Northern Ethiopia.

Table 10: Partial budget analysis of interaction effect of different varieties and *Mesorhizobium* strains with NPSB fertilizer on yield of chickpea

Treatments	**AVGY (kg ha^{-1})**	**AJGY (kg ha^{-1})**	**TGB (ETB ha^{-1})**	**Seed cost (ETB ha^{-1})**	**NPSB Cost (ETB ha^{-1})**	**STC cost (ETB ha^{-1})**	**LC 1 (ETB ha^{-1})**	**LC 2 (ETB ha^{-1})**	**TVC (ETB ha^{-1})**	**NB (ETB ha^{-1})**
L	1693.91	1439.82	43194.6	2500	0	0	0	0	2500	40694.6
L x S1	2405.88	2044.99	61349.7	2500	0	180	0	150	2830	58519.7
L x S2	2673.42	2272.4	68172	2500	0	180	0	150	2830	65342
L x S3	2427.85	2063.67	61910.1	2500	0	180	0	150	2830	59080.1
L x NPSB	2115.66	1798.31	53949.3	2500	2250.6	0	150	0	4900.6	49048.7
L x S1 x NPSB	2675.05	2273.79	68213.7	2500	2250.6	180	150	150	5230.6	62983.1
L x S2 x NPSB	2952.19	2509.36	75280.8	2500	2250.6	180	150	150	5230.6	70050.2
L x S3 x NPSB	2697.42	2292.8	68784	2500	2250.6	180	150	150	5230.6	63553.6
A	1971.87	1676.08	67043.2	3200	0	0	0	0	3200	63843.2
A x S1	2669.88	2269.39	90775.6	3200	0	180	0	150	3530	87245.6
A x S2	2523.69	2145.13	85805.2	3200	0	180	0	150	3530	82275.2
A x S3	2580.92	2193.78	87751.2	3200	0	180	0	150	3530	84221.2
A x NPSB	2412.72	2050.81	82032.4	3200	2250.6	0	150	0	5600.6	76431.8
A x S1 x NPSB	3177.16	2700.58	108023.2	3200	2250.6	180	150	150	5930.6	102092.6
A x S2 x NPSB	2816.62	2394.12	95764.8	3200	2250.6	180	150	150	5930.6	89834.2
A x S3 x NPSB	2890.68	2457.07	98282.8	3200	2250.6	180	150	150	5930.6	92352.2

L = Local; A = Arerti; S1 = CP-M41 strain; S2 = CP-EAL-029 strain; S3 = CP-M20b strain; AVGY = Average grain yield; AJGY = adjusted grain yield (-15% of the gross average grain yield); TGB = total gross benefit (AJGY * 30 ETB kg^{-1} (field price of Local grain) and 35 ETB kg^{-1} (field price of Arerti grain); Seed cost (25 and 32 ETB kg^{-1} for Local and Arerti varieties, respectively); NPSB fertilizers cost (NPSB = 18.60 ETB kg^{-1}), inoculants cost (CP M41, EAL-029 & M20b = 45 ETB bag^{-1}), STC = strain cost; LC1 = fertilizer application labor cost (Labor cost = 150 ETB day^{-1}), LC2 = inoculants application labor cost (Labor cost = 150 ETB day^{-1}), TVC = total variable cost, NB = net benefit (GB-TVC).

Table 11: Dominance analysis of treatments and marginal analysis of un-dominated treatments

Treatments	**TVC (ETB ha^{-1})**	**NB (ETB ha^{-1})**	**Dominance**	**MC (ETB ha^{-1})**	**MB (ETB ha^{-1})**	**MRR (%)**
L	2500	40694.6	UD	-	-	-
L x S1	2830	58519.7	UD	330	17825.1	5401
L x S2	2830	65342	UD	0	6822.3	-
L x S3	2830	59080.1	D			
A	3200	63843.2	D			
A x S1	3530	87245.6	UD	700	21903.6	3129
A x S2	3530	82275.2	D			
A x S3	3530	84221.2	D			
L x NPSB	4900.6	49048.7	D			
L x S1 x NPSB	5230.6	62983.1	D			
L x S2 x NPSB	5230.6	70050.2	D			
L x S3 x NPSB	5230.6	63553.6	D			
A x NPSB	5600.6	76431.8	D			
A x S1 x NPSB	5930.6	102092.6	UD	2400.6	14847	618
A x S2 x NPSB	5930.6	89834.2	D			
A x S3 x NPSB	5930.6	92352.2	D			

UD = Un-dominated; D = Dominated treatments; MC = Marginal cost (Change in total variable cost between treatments); MB = Marginal benefit (Change in net benefits between treatments); MRR = Marginal rate of return (MB/MC * 100)

5. SUMMARY, CONCLUSION AND RECOMMENDATIONS

5.1. Summary and Conclusion

Chickpea (*Cicer arietinum* L.) is a major legume crop in Ethiopia and provide multiple benefits, due to high nutritive value as well as the ability of the crop to enrich nitrogen poor soils due to biological nitrogen fixation with different strains of endosymbiotic *Mesorhizobium spp*. However, the effectiveness of the *Mesorhizobium* strains varies due to inherent physiological characteristics of the endo-symbionts, the host varieties and availability of nutrients in the soil. In Ethiopia, the current national average productivity of chickpea is 2 t ha^{-1} which is much lower than its' potential (5 t ha^{-1}), obtained under good management conditions, due to low soil fertility and ineffective nodulation or low BNF by the crop. Declining soil fertility due to unbalanced fertilization was the major factor for lower productivity of chickpea in Ethiopia and to combat these problems, balanced commercial fertilizers containing N, P, S and B in blend form are recommended in the country.

Despite of the role of nitrogen, phosphorus, sulfur, boron, and *Rhizobia* bio-fertilizers (inoculants) in influencing nitrogen fixation, growth and productivity of chickpea, most farmers in the study area did not apply any fertilizer materials for improving productivity of the crop and thereby facing productivity problem. Moreover, low soil fertility and lack of improved agronomic practices are the major chickpea production constraints at Cheha district (the study area) in Gurage Zone, Southern Ethiopia. The major soil related constraints in the study area are low total N, low soil OM, low available P, S and B. Currently, there is no use of blended commercial fertilizers, *Rhizobia* inoculants and improved varieties for chickpea cultivation in the study area. Hence, ensuring a well-balanced supply of blended NPSB fertilizer and *Rhizobia* inoculation to the crop may result in higher seed yield. Similarly, using effective *Rhizobia* inoculants as Nitrogen sources for improving chickpea growth, symbiotic and yield performance is very important for sustainable crop production under Cheha conditions.

Limited research has been done on the effect of seed inoculation with *Mesorhizobium* strains and blended NPSB fertilizer application on growth, symbiotic, and yield and yield components of chickpea varieties. Therefore, the field experiment was conducted to evaluate the effect of different *Mesorhizobium* strains and blended NPSB fertilizer on the growth, nodulation, yield and yield components of chickpea varieties and to identify economically appropriate combination of *Mesorhizobium* strains and blended NPSB that provided maximum yield for the chickpea varieties. The experiment consisted of four levels of *Mesorhizobium* inoculation (un-inoculated, inoculated with strains CP-M41, CP EAL-029, and CP-M20b), two levels of NPSB rates (0 and 121 kg ha^{-1}) and two chickpea varieties (Local and Arerti) that were tested in randomized complete block design with factorial arrangement using three replications.

A composite surface soil sample (0-30 cm depth) was collected before planting from the experimental site where responsiveness to *Mesorhizobium* inoculation and NPSB fertilizer application for chickpea has not been studied. The soil analysis indicated that the soil is clayey in texture. The pH (H2O) of the soil ranged as moderately acid (pH 5.7). The site was very high in CEC, low in OM content (3.62%), low in total N content (0.18%), very low in available (Olsen) P (2.69 mg kg^{-1}). The tested site was also deficient in available S and B. Based on the soil testing result, S and Zn had been considered as the nutrients to be corrected by applying chemical fertilizers along with *Rhizobia* inoculation to improve nutrient availability and productivity of legume crops including chickpea.

Analysis of variance showed that the main effect due to *Mesorhizobium* inoculants, NPSB fertilizer application, and varieties significantly influenced all of the studied parameters. Moreover, the effect on nodulation, growth, yield and yield component parameters was more pronounced in the combined application of *Mesorhizobium* inoculants with NPSB application and varieties. Significantly maximum number of total nodules (19.2), volume of nodules (9.9 ml plant^{-1}) and dry weight of nodules (4.35 g plant^{-1}) were obtained from combination of NPSB fertilizer with CP-M41 *Mesorhizobium* strain and variety Arerti, which resulted in 36.61%, 51.21% and 34.48% increment over the control check, in that order. The nodule color due to the combined application of *Mesorhizobium* inoculants with NPSB blend fertilizer on the tested varieties was found ranging between pink and slightly dark red, while white nodule color was observed at the control check plots.

The combined application of CP-M41 strain with NPSB fertilizer and Arerti variety significantly gave the maximum values on most of the growth parameters including plant height, number of primary and secondary branches and shoot dry weight when compared to the single use and the control treatments. The results also demonstrate that, the interaction of the three factors significantly affect all the yield components studied (number of pods and seeds per plant, number of seeds per pod, and hundred seed weight). The combined application of CP-M41 *Mesorhizobium* strain with NPSB blend fertilizer and Arerti variety gave the maximum number of pods, number of seeds per pod, and hundred seed weight. Whereas, the minimum values of these parameters were observed from the combined application of Local variety at the control. Significantly the highest number of total pods (84.6 pods plant^{-1}), number of seeds per pod (1.213) and hundred seeds weight (31.14 g plant^{-1}) were obtained from combination of NPSB fertilizer with CP-M41 *Mesorhizobium* strain for variety Arerti, which resulted in 30.57%, 12.61% and 31.66% increment over the control check, in that order.

Yield traits of chickpea like above ground dry biomass yield, grain yield and straw yield were also significantly affected by the three-way interaction of *Mesorhizobium* strains, varieties and NPSB application. The highest grain yield (3177.16 kg ha^{-1}) and straw yield (2747.91 kg $^{ha-1}$) were obtained from Arerti variety at the combined application of NPSB blend fertilizer with CP-M41 *Mesorhizobium* strain, while the lowest grain yield (1693.91 kg ha^{-1}) and straw yield (2032.91 kg ha^{-1}) were obtained from Local variety grown without inoculation and NPSB fertilizer application. Furthermore, the highest (5924.33 kg ha^{-1}) and lowest (3726.83 kg ha^{-1}) dry biomass yield was obtained from variety Arerti when grown at the combined application of NPSB blend fertilizer with CP-M41 *Mesorhizobium* strain, and from un-inoculated and unfertilized Local variety, respectively. Generally, the combined application of NPSB blend fertilizer with CP-M41 *Mesorhizobium* strain and variety Arerti also gave maximum grain yield, total dry biomass yield and harvest index of chickpea, which resulted in 37.93%, 27.58% and 20.34% increment over the control.

Therefore, interaction effects of *Mesorhizobium* strains with NPSB blend fertilizer application showed a synergistic effect justifying that the combined application of nutrients gave highest grain yield than the individual application. Correlation analysis of this study indicated that grain yield was positively very highly significantly correlated with measured nodulation, growth and yield related parameters.

The economic analysis data of the present study also showed that the highest net return (102, 092.6 ETB ha^{-1}) with acceptable marginal rate of return (618%) was obtained from Arerti variety with CP-M41 *Mesorhizobium* inoculation and 121 kg NPSB ha^{-1} application, followed by the combined use of Arerti variety with CP-M41 strain alone, which gave the second highest net benefit of 87, 245.6 ETB ha^{-1} with an acceptable marginal rate of return of 3129% (Table 9). Based on the results of this study, it can be concluded that combined application of Arerti variety with CP-M41 *Mesorhizobium* inoculation and 121 kg NPSB ha^{-1} application gave the highest agronomic yield as well as economic benefit. The result also indicated that combined application of *Mesorhizobium* inoculants and NPSB fertilizer has more yield benefits than single application in both chickpea varieties.

5.2. Recommendations

In this study, therefore, the use of Arerti variety with *Mesorhizobium* inoculation with CP-M41 inoculant and 121 kg NPSB ha^{-1} application found to be highest yield and economically feasible and could be recommended for chickpea production in cheha district. However, the results presented here is based on one season and one location experiment and need to be confirmed through further on farm research under various soil and agro climatic conditions. Therefore, attention shall be given to the following issue for future research:

- ✓ Conducting similar research over locations and seasons would be relevant to verify the current result and to get conclusive result for the best recommendation, and hence to put it in a firm ground.
- ✓ Evaluating and reconfirming the present research result with different levels of NPSB rates less than and greater than 121 kg ha^{-1} along with the *Mesorhizobium* inoculants is needed under different agro-ecologies in order to reach to a conclusive recommendation.
- ✓ The effectiveness of these commercial inoculants of chickpea should be evaluated over location and their relationship with respect to native rhizobia population, soil fertility status and cropping system need further investigation.

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7. APPENDICES

Appendix Table 1: Treatment Combinations and Descriptions

Treatment number	Factors		
	Variety (V)	*Mesorhizobium* Strains	NPSB Fertilizer (kg ha^{-1})
T1	Local	Un-inoculated	0
T2	Local	CP M41	0
T3	Local	CP EAL 029	0
T4	Local	CP M20b	0
T5	Local	Un-inoculated	121
T6	Local	CP M41	121
T7	Local	CP EAL 029	121
T8	Local	CP M20b	121
T9	Arerti	Un-inoculated	0
T10	Arerti	CP M41	0
T11	Arerti	CP EAL 029	0
T12	Arerti	CP M20b	0
T13	Arerti	Un-inoculated	121
T14	Arerti	CP M41	121
T15	Arerti	CP EAL 029	121
T16	Arerti	CP M20b	121

Appendix Table 2: Mean monthly rainfall (mm) and mean temperatures (ºC) from 2012 to 2021 at Cheha district

Month	Meteorological data		
	Rainfall (mm)	Maximum temperature (^{0}C)	Minimum temperature (^{0}C)
January	37.85	27.85	10.89
February	132.29	27.84	12.13
March	519.32	27.15	11.89
April	532.45	27	12.78
May	776.57	26.94	12.92
June	1030.03	24.23	12.43
July	3758.73	22	10.45
August	3760.4	23	9.36
September	730.2	23.25	11.93
October	246.31	24.9	9.95
November	170.02	25.15	10.58
December	140.73	26	10.99
Mean	986.24	25.44	11.35

Source: Hawassa Metrological Station, Ethiopia

Appendix Table 3: Mean squares ANOVA for nodulation parameters of chickpea to seed inoculated by *Mesorhizobium* strains, Varieties, and NPSB fertilization.

Sources of variation	**DF**	**TNNPL**	**ENNPL**	**NVPL (ml plant^{-1})**	**NDWPL (g plant^{-1})**
Rep	2	2.00	0.11	0.31	0.11
V	1	22.68***	0.3***	5.005***	0.82***
RI	3	25.72***	1.02***	14.85***	1.21***
NPSB	1	55.47***	2.43***	29.29***	3.83***
V*RI	3	9.34***	0.59***	5.17***	0.64***
V*NPSB	1	0.27^{NS}	0.003^{NS}	0.13^{NS}	0.037*
RI*NPSB	3	0.99*	0.026*	0.24**	0.043**
V*RI*NPSB	3	2.09***	0.14***	1.22***	0.14***
Error	30	0.29	0.0077	0.06	0.006
Total	47				

* Significant at $p \leq 0.05$; ** significant at $p \leq 0.01$; *** significant at $p \leq 0.001$ based on LSD at $p \leq 0.05$; NS= non- significant at $p > 0.05$. DF= Degree of freedom, N=nitrogen, P= phosphorus, S= Sulfur, B=boron; V = Variety; RI=*Mesorhizobium* inoculants, TNNPL= Total number of nodules per plant, ENNPL= Effective nodule number per plant, NVPL = Nodule volume per plant; NDWPL= Nodule dry weight per plant.

Appendix Table 4: Mean squares ANOVA for phenological and growth parameters of chickpea to seed inoculated by *Mesorhizobium* strains, Varieties, and NPSB fertilization.

SV	DF	DTF	DTM	PHPL	NPBPL	NSBPL	SHDWPL (g plant^{-1})	RDWPL (g plant^{-1})
Rep	2	0.39	24.93	63.39	0.28	3.88	2.81	3.04
V	1	54.18***	346.68***	93.94***	0.05***	9.99***	138.07***	1.29**
RI	3	8.96***	30.68**	62.58***	1.42***	27.60***	120.17***	1.28***
NPSB	1	9.18**	123.52***	184.27***	1.54***	58.30***	318.42***	3.47***
V*RI	3	1.63^{NS}	13.68^{NS}	38.61***	0.26***	4.90***	48.87***	0.2^{NS}
V*NPSB	1	0.02^{NS}	6.02^{NS}	6.27^{NS}	0.03*	0.22^{NS}	1.42^{NS}	0.009^{NS}
RI*NPSB	3	0.85^{NS}	10.41^{NS}	7.59*	0.02*	1.84***	3.08*	0.21^{NS}
V*RI*NPSB	3	0.24^{NS}	8.35^{NS}	12.96**	0.04***	1.05**	14.73***	0.07^{NS}
Error	30	1.17	7.89	1.96	0.006	0.21	0.89	0.105
Total	47							

SV = Sources of variation; * Significant at $p \leq 0.05$; ** significant at $p \leq 0.01$; *** significant at $p \leq 0.001$ by LSD; NS= non- significant at $p > 0.05$. DF= Degree of freedom; N=nitrogen, P= phosphorus, S= Sulfur, B=boron; V= Variety; RI=*Mesorhizobium* inoculants; DTF= Days to 50% flowering; DTM= Days to 90% Physiological maturity; PHPL= Plant height per plant; NPBPL= number of primary branches per plant; NSBPL= number of secondary branches per plant; SHDWPL=Shoot dry weight per plant; RDWPL=Root dry weight per plant.

Appendix Table 5: Mean squares ANOVA for yield components of chickpea to seed inoculation with *Mesorhizobium* strains, Varieties, and NPSB fertilization.

SV	DF	NPPL (no)	NSP (no)	HSW (g plant^{-1})
Rep	2	2.33	0.0005	5.808
V	1	201.31***	0.0085***	72.201***
RI	3	366.20***	0.0081***	72.977***
NPSB	1	746.55***	0.026***	121.444***
V*RI	3	142.60***	0.0055***	8.494***
V*NPSB	1	35.70*	0.00083*	1.573**
RI*NPSB	3	40.30**	0.00096***	0.641*
V*RI*NPSB	3	32.66**	0.00165***	0.524*
Error	30	5.97	0.00012	0.173
Total	47			

SV= Sources of variation; * Significant at $p \leq 0.05$; ** significant at $p \leq 0.01$; *** significant at $p \leq 0.001$ by LSD; NS= non- significant at $p > 0.05$; DF= Degree of freedom; N=nitrogen; P= phosphorus; S= Sulfur; B=boron; V= Variety; RI=*Mesorhizobium* inoculants; NPPL= number pods per plant, NSP= number of seeds per pod; HSW=Hundred seed weight; GY=Grain yield; BY=Biomass yield; HI=Grain harvest index

Appendix Table 6: Mean squares ANOVA for yield traits of chickpea to seed inoculation with *Mesorhizobium* strains, Varieties, and NPSB fertilization.

SV	DF	GY (kg ha^{-1})	BY (kg ha^{-1})	SY (kg ha^{-1})	HI (%)
Rep	2	2851.833	1576.304	3721.66	0.000152
V	1	368638.380***	1845536.333***	564525.63***	0.000675***
RI	3	1323539.286***	1971354.516***	65756.8***	0.010905***
NPSB	1	1459588.501***	2965798.041***	264211.36***	0.012675***
V*RI	3	156850.604***	440252.817***	71934.62***	0.001347***
V*NPSB	1	18198.999**	56189.242*	10432.38^{NS}	0.000300*
RI*NPSB	3	15820.094**	27695.382^{NS}	3831.19^{NS}	0.000113^{NS}
V*RI*NPSB	3	8652.700**	72339.991**	36476.69**	0.000150**
Error	30	2403.912	12897.06	5914.39	0.000045
Total	47				

SV= Sources of variation; * Significant at $p \leq 0.05$; ** significant at $p \leq 0.01$; *** significant at $p \leq 0.001$ by LSD; NS= non- significant at $p > 0.05$; DF= Degree of freedom; N=nitrogen; P= phosphorus; S= Sulfur; B=boron; V= Variety; RI=*Mesorhizobium* inoculants; NPPL= number pods per plant, NSP= number of seeds per pod, NSPL=number of seeds per plant; HSW=Hundred seed weight; GY=Grain yield; BY=Biomass yield; HI=Grain harvest index

BIOGRAPHICAL SKETCH

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