EFFECTS OF RHIZOBIUM STRAINS ON SEED QUALITY, YIELD AND YIELD RELATED TRAITS OF (*Cicer arietinum* L.) VARIETIES AT AMBO, ETHIOPIA

MSc THESIS

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Effects of Rhizobium Strains on Seed Quality, Yield and Yield Related Traits of Chickpea (*Cicer arietinum* L.) **Varieties at Ambo, Ethiopia**

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Master of Science in Seed Science and Technology

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HARAMAYA UNIVERSITY POSTGRADUATE PROGRAM DIRECTORATE

We hereby certify that we have read and evaluated this Thesis entitled **"Effects of *Rhizobium* Strains on Seed Quality, Yield and Yield Related Traits of Chickpea** (*Cicer arietinum* L.) **Varieties at Ambo, Ethiopia",** which has been prepared under our Guidance by **Berhanu Soboka**. We recommend that the Thesis be accepted as fulfills the thesis requirements.

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As members of the Board of Examiners of the M.Sc. thesis open defense examination, we certify that we have read and evaluated the thesis prepared by **Berhanu Soboka** and examined him. We recommend that the thesis be accepted as it fulfills the requirements for the Degree of Master of Science in Agriculture **(Seed Science and Technology).**

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Final approval and acceptance of the thesis is contingent upon submission of the final copy of the thesis to the council of graduate studies (CGS) through the graduate committee (SGC) of the candidate's School.

DEDICATION

I dedicated this thesis to God and my family.

STATEMENT OF THE AUTHOR

First, I declare that this thesis is a result of my genuine work and that I have duly acknowledged all sources of materials used for writing it. I submit this thesis to Haramaya University in partial fulfillment of the requirements for the Degree of Master of Science. The thesis is deposited at the library of the University to be made available to borrowers for reference. I solemnly declare that I have not so far submitted this thesis to any other institution anywhere for the award of any academic degree, diploma, or certificate.

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The author was born on August 1989 at Jibat, West Shoa Zone of Oromiya Regional States, and Ethiopia from his father Soboka Bayisa and his mother Desi Dabal. He completed elementary (1994-2001) and secondary education (2005-2007) at Icha Adadi Elementary and shenen Preparatory Schools, respectively. He joined chiro Agricultural Technical Vocational Education and Training College (ATVET) graduated with Diploma in 2010. Thereafter, he was recruited by Oromia Agricultural and Natural Resource Development Bureau and served as Development Agent until 2011. He joined Arsi University in summer program and graduated with B.Sc. degree in Plant Sciences in 2019.Thereafter, he was recruited by Oromia Agricultural and Natural Resource Development Bureau and served as Development Agent until 2014 of West Shoa zone, jibat as coffee Agronomy expert until 2015. In 2021 he joined Oromia Agricultural input and production regulatory authority seed inspector. He joined the School of Graduate Studies of Haramaya University in 2022 to pursue a study leading to the degree Master of Sciences in agriculture (Seed Science and Technology).

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ACRONYMS AND ABBREVIATIONS

Am ARC	Ambo Agricultural Research Center
AOSA	Association of Official Seed Analyst
BNF	Biological Nitrogen Fixation
CSA	Central Statistical Agency.
CV	Coefficient of Variation
EIAR	Ethiopian Institute of Agricultural Research
EU	European Union
FAO	Food and Agriculture Organization
FAOSTAT	Food and Agriculture Organization Statistics
ICARDA	International Center of Agricultural Research in the Dry Areas
ICRISAT	International Crop Research Institute for the Semi-Arid Tropic
ISTA	International Seed Testing Association
LSD	Least Significant Difference
M.A.S.L	Mass above sea level
PH	Potential of Hydrogen
SSA	Sub-Sahara Africa
USDA	United States Department of Agriculture
WHO	World Health Organization

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ABSTRACT

Chickpea (Cicer chairietinum L.) is an important legume food crop in West Showa used as food and income generation for farmers, however, unavailability of quality seeds and low soil fertility are among the production constraints. Therefore, this study was conducted to assess effect of Rhizobium inoculation on growth, yield and its components and seed quality of chickpea varieties. The four varieties (Eshete, Dimtu, Teketay and Local) x four Rhizobium strains (Cp11, Cp17, Cp41 and control) arranged in factorial combinations were evaluated in randomized complete block design with three replications at research field of Ambo Agricultural Research Centre. The seed quality test was conducted in acompletely randomized design with four replications in lab. Variety and Rhizobium strain had significant influence on all phenology and growth traits, yield components, seed yield and productivity indices except crop phenology and hundred seed weight were not significant influenced by Rhizobium strain. The interaction of variety and R strain had significant effect on plant height, number of pods per plant,percentage of normal seedlings germination, seedlings shoot length, seedlings root length and seedlings vigour index one. The main factors, variety and R. strain significantly influenced seedlings dry weight and seedlings vigour index two, and speed of germination was significantly influenced by variety. The seed yield obtained from four varieties ranged from 2013.89 to 2777.78 kg ha^{-1}, and the seed yield from inoculation treatments ranged from 2152.78 to 2690.97 kg ha^{-1}. The highest seed yield was obtained from Teketay variety seeds innoculated with Cp17 Rhizobium strain while the seed yield was ombtained from farmers' cultivar and seeds not innoculated. Teketay variety and seeds innoculated with Cp17 Rhizobium strain also had higher seed production efficiency, grain yield index per day, Rhizobium sensitive or infection index and yield index. Percentage of normal seedlings germination ranged from 97.25 to 98.75% that the farmers' cultivar innoculated with Cp17 Rhizobium strain and uninnoculated seed and Teketay variety innoculated with Cp11 Rhizobium strain had highest germination. The seedling vigour index one ranged from 693.81 to 1138.01 in which the lowest and highest seedling vigour index one was registered for Teketay and Eshete varieties innoculated with Cp17 Rhizobium strain, respectively. Farmers' cultivar and Dimtu variety had the lowest (56.26) and highest (62.85) seeding vigour index two, respectively, whereas uninnoculated seeds and seeds innoculated with Cp17 Rhizobium strain had the lowest (57.93) and highest (60.73), respectively. Rhizobium strain have significantly increases the seed yield and quality of variety which is possibly due to the effectiveness of the train in solubilization and availability of nutrients.

Keywords: Growth, Productivity index, Seed, Seedlings germination and Vigour index.

1. INTRODUCTION

Chickpea (*Cicer arietinum* L.) is the world's third most significant food legume after beans and peas on the basis of production level. It is grown in over 50 countries throughout the world and consumed in over 120 countries. Ethiopia is one of the top five chickpea producing countries in the world only exceeded by India, Australia, Myanmar and Turkey (FAO, 2019). Ethiopia is considered as second center of origin of chickpea (Anbessa and Bejiga *et al*., 2002; ICARDA, 2009) and the country is the largest chickpea grower that ranks first in Africa. Chickpea is widely grown in the central, northern and eastern highland areas of the country at an altitude range of 1400-2300 m.a.s.l., where annual rainfall ranges between 500 and 2000 mm (Anbessa and Bejiga *et al*., 2002; Bejiga and Van der Maesen *et al*., 2006). The crop has numerous roles in traditional farming system. It is a significant source of human food and animal feed, and it is a source of cash income for farmers in developing countries (Erman *et al.,* 2011). Chickpea seeds contain 20.6% protein, 61.2% carbohydrate and 2.2% fat (Togay *et al*., 2008) and hence, it is a good source of relatively cheap energy, protein and potentially health-beneficial phytochemicals (Wood and Grusakb *et al.*, 2007). The crop further plays a vital function in the sustaining of soil fertility (Funga *et al.*, 2016), primarily in dry and rain-fed areas through N_2-fixation, and, as a rotation crop allowing the diversification of agricultural production systems (Shurigin *et al.*, 2015) and solubilisation of unavailable soil phosphorus (Selvakumar 2012) that may be useful to other crops in the system.

In Ethiopia during 2019/20 *Meher* Season, 779,033 farmers produced chickpea on 208,838 hectares of land. A total of 435193.21 tons of red and white chickpeas produced with the average yield of 2 t ha^{-1}. Oromia Regional State was the major producer of chickpea which accounts 40.52 % of land cultivated and 35.71% of total production of the country. However, the average yield of chickpea in the Regional State was 1.834 t ha^{-1} and it was lower by 8.85% than the national average yield. The average yield of red chick-peas (*Desi* type) was much lower by 13.1% than the national average yield (CSA, 2020). The productivity is, thus, far below the potential yield of 4.5 t ha^{-1} (Fikre,*et al*., 2016). The limited access to high quality seeds of improved varieties and the low level implementation of improved technologies and agronomic practices by farmers are considered major limiting factors for low chickpea production (Sheleme *et al*., 2016).

Many research reports showed that chick pea fixes atmospheric nitrogen in the range of 60-140 kg N ha^{-1} in one cropping season (Burton 1984; Crouch *et al*., 2004 and Shiferaw *et a l*.,2004).The chickpea grain and Stover yield increased by 8 to 40% and 8 to 10%, respectively, if seeds are inoculated by *Rhizobium* strains, it further reaches about 60% grain and Stover yields increase if inoculation is combined with the application of phosphorus fertilizer (Ali *et al*., 2004 as cited in Ronner and Giller *et al*.,2011; Giller *et al*., 2013). Inoculation of seed with its own specific and suitable *Rhizobium* strain before planting is crucial to fully benefited from grain legume crop in terms of maximum yield and soil improvement (Ayaz, *et al*., 2010). In Ethiopia, there are many *Rhizobia* strains that have been identified and proven to be effective and competitive (Wolde-Meskel 2012) and 17 and 14 *Desi* and *Kabuli* type varieties, respectively, released up to 2019 (2011). However, the use of high yielding varieties alone may not result in the required level of productivity without inclusion of compatible and effective rhizobia strains (Mulongoy 1995; Shantharam *et al.,* 1997). Keneni *et al.,* 2013) suggested the need to conduct specific tests for specific breeding materials, strain and environments to improve the selection process to gain host genotype compatible to specific Rhizobia strain that eventually enhance BNF and subsequently ensure productivity and sustain soil fertility.

West Showa Administrative Zone of Oromia Regional State had large share (15.27%) of cultivated land for red chickpeas in the Regional State (CSA, 2020). However, the yield of the crop is low as compared to the potential of the crop to produce up to 5.5 t ha^{-1} (Belay *et al.,* 2006). The use of local cultivars and poor-quality seeds, seed borne diseases, low population density of plants are among many factors are responsible for the low yield of chickpea in the country (Melese *et al.*, 2005). It was reported that chemical fertilizers that are capable to increase yields by 10 to 80% the production and productivity of pulse crops including chickpea but the fertilizers usage is low in Ethiopia (Alemneh *et al*., 2017). The production constraints of chickpea in the country are also true to West Showa. Therefore, the production and availability of quality seed of improved varieties, identifying specific and compatible *Rhizobia* strains is vital to improve the chickpea productivity in West Showa, reduce the need for costly fertilizer applications and sustained soil fertility. Seed is the single most important input in crop production. The lack of quality seed limits smallholder farmers' production and productivity of developing countries in sub-Saharan Africa (SSA). The seed alone as input is estimated to contribute up to 20% of total crop productivity. Despite this, only an average of about 20% of farmers in sub-Saharan Africa use seeds of improved varieties (Akpo *et al*., 2021). Ethiopia as one of sub-Saharan Africa countries also face the unavailability of seeds of improved chickpea varieties and the use of local cultivars seeds by smallholder farmers passed on from generation to generation as the production constraints. Several authors reported the positive effect of *Rhizobium* inoculation and NP fertilizer application on chickpeas growth and yields in Ethiopia (Amanuel *et al*.,2000; Ayneabeba *et al*., 2001; Angaw and Asfa *et al*., 2006; Mulugeta *et al.,* 2018 and Beza *et al*., 2019), but the authors didn't indicate the effect of these practices on seed quality of chickpea.

The biological nitrogen fixation (BNF) is a natural process of rhizobial inoculants production. The rhizobial inoculant is a cheap and non-polluting source of bacteria that can provide high rates of nitrogen to the legume, and the use of improved inoculants is a strategy to increase BNF rates in agro-ecosystems (Deaker *et al.,* 2004; Araujo *et al.,* 2011).The atmosphere contains about 1015 tons of N2 gas, and the nitrogen cycle involves the transformation of 3x109 tones of N2 per year on global bases (Posgate, 1982). However, N-fixation is not exclusively biological, lightning probably accounts for about 10% of the world's supply of fixed nitrogen (Sprent.J.I. and Sprent.p, 1990). FAO, (1990) reported that, world production of fixed nitrogen in the form of chemical fertilizers accounts for about 25% of the earth's newly fixed nitrogen, and biological processes accounts for about 60%.

The use quality seed of chickpea varieties with inoculation of *Rhizobium* strains to improve chickpea yield in West Showa has been studied (Abere Mnalku 2015; Assefa *et al.,* 2016). The researchers were reported the importance of using improved varieties, *Rhizobium* inoculation along with NP fertilizer application to increase the grain yield. However, none of the authors reported about the effect of *Rhizobium* inoculation and NP fertilizer application on the production of quality seed of improved chickpea varieties though it is well known that the use of high quality seed is the first step in establishing a rapidly emerging, vigorous stand, and producing a high quality and profitable crop (*Eshete, Dimtu,Teketay* and Farmers' cultivar). Thus, it is vital to identify of improved varieties that would have specific Rhizobial strain(s) association to produce quality and high seed yield important in West Showa Zone of Oromia Region state to enhance the productivity of the crop. This research was therefore initiated to achieve the following objectives:

- To evaluate effects of seeds inoculation with *Rhizobium* strains on nodulation, phenology, growth and seed yield of chickpea varieties, and
- To determinate the effects of seed inoculation with *Rhizobium* strains on seed quality of chickpea varieties.

2. LITERATURE REVIEW

2.1. The Chickpea Crop

Chickpea (*Cicer arietinum* L.) belongs to the genus Cicer, tribe Ciccereae, family Fabaceae and subfamily Papilionaceae. It is one of the most important cool season crops, is believed to have originated in present-day south eastern Turkey and adjoining Syria where several of its natural species are found (Van der Maesen 1987). The crop later spread to India, Europe and subsequently reached Africa, Latin and central American countries. Ethiopia being secondary center of origin is one of the largest chickpeas producing countries of the world and ranks first in Africa. It is widely grown in the central, northern and eastern highland areas of the country (Anbessa and Bejiga 2002). Chickpea is self-pollinated annual crops that can complete its life cycle in 90 to 180 days depending on the prevailing meteorological conditions. There are two types' of chickpea, *Desi* and *Kabuli*. *Desi* types are generally small seeded with thick and dark coloured seed coat, while *Kabuli* types are usually large seeded with thin and lighter coloured seed coat. Phenolic compound s (tannin) impart seed coat colour, which are considered to also have antifungal, antimicrobial properties and reported to protect the seed from insect pests and precocious germination (Caldes and Blair *et al*., 2009). Currently, about 75% of the area all over the world is covered by the *Desi* and the remaining 25% by the *Kabuli* types (Kassie *et al.,* 2009).

Chickpea is herbaceous and the height generally ranges from 30-70cm depending on the suitability of the growing environment (Melka, 2014). Stems are branched, flexuous or straight, erect to prostrate, and more or less ribbed. The leaves are serrated leaflets and arise alternatively from the third node with 2-3 bisexual flowers in a node. Flowers are 4 to 30 mm long which may be white, pink, purple or blue in color. The crop may set 1-3 seeds in a pod and the seed is a good source of protein (23%), carbohydrates (64%), fat (5%) and crude fiber (6%) (Bejiga and van der Maesen *et al*., 2006). It has tap root system; which is usually deep and strong. The lateral roots develop nodules with the symbiotic *Rhizobium* bacteria, capable of fixing atmospheric nitrogen in plant usable form. The nodules (slightly flattened, fan-like lobes) are visible about one month after plant emergence and generally to the top 15 cm of the surface (ICRISAT, 2010).

2.2. Agronomic and Economic Importance of chickpea

Chickpea serves as a multi-purpose legume. First, it fixed atmospheric nitrogen in soil and thus improve soil fertility and saves fertilizer costs in subsequent crops rotation. Ibsa (2013) observed improvement in Chickpea yield (2 tone ha^{-2}) after inoculation, in response to soil fertility improvement, through enhanced biological nitrogen as compared to no inoculation (1.6 tone ha^{-1}). Chickpea is an excellent source of protein, fire, complex carbohydrates, Vitamins, and minerals, as a cash crop for smallholder producers and a good source of energy also contains potentially health-beneficial phytochemicals (Wood and Grusak *et al.,* 2007). Chickpea is rich in proteins and serves as an economical source of nutritious food for many poor households. Its seed contains 28.5% protein, 59% carbohydrate, 3% fibber, 5% oil and 4% others. The protein is rich in l*ysine* and *arginine* but most deficient in sulphur-containing amino acids such as methionin*e* and cysteine (Irbil *et al.,* 2006).

Chickpea is a good source of absorbable Ca, P, Mg, Fe, and K (chuvin *et al.,* 1986*;* Christodoulou *et al.,* 2005). It is mainly used as human food in Ethiopia and seed are consumed green, cooked, roasted, or bolt/germinated. Sometimes the dry seed are mixed with wheat and /or barley and ground nut to powder to make *''kyat Injera''* (a type of local bread). Split seeds (kick) and powdered seed (*shiro*) are also used in making *wet* (type of sauce) or soup which is usually eaten with *injera* (Gulite *et al.,* 1972). Because of its ability to withstand drought stress, smallholder farmers in Ethiopia grow Chickpea at the end of the main rainy season using residual soil moisture. This permits farmers to grow a second crop and secure an additional source of income and protein through efficient use of the residual moisture in black soil at the end of the rains. This improves food security for the house hold while the nitrogen fixed by the crop enriches soil nutrient for the subsequent cereal crops (*teff* and *wheat*) that follow in the rotation. Legumes are multipurpose crops and are consumed either directly as food or in various processed forms or as feed in many farming systems (Kumara Charyulu and Deb *et al.,* 2014).

Chickpea is playing a leading role in food safety in the world by covering the deficit in proteins of daily food ration of Indian and African Sub Sahara populations. The designed chickpea-based infant follow-on formula meets the WHO/FAO requirements on complementary foods and also the EU regulations to follow on formula with minimal addition of oils, minerals, and vitamins. It uses chickpea as a common source of carbohydrate and protein hence making it more economical and affordable for the developing countries without compromising the nutrition quality (Malunga *et al*., 2014).

2.3. Production of Chickpea in Ethiopia

Ethiopia is the top producer of chickpea in Africa. In Ethiopia, chickpea is the third most important grain legume after faba bean (*Vicia faba*) and common bean (*Phaseolus vulgaris* L.) by volume for small-scale farm production. It is mainly grown in the central, northern and eastern highland areas of the country at an altitude of 1400-2300 m.a.s.l., where annual rainfall ranges between 500 and 2000 mm (Anbessa and Bejiga *et al.,* 2002 and Bejiga and van der Maesen *et al*., 2006). During the (2014/15) cropping year, Ethiopia produced 458,682.26 tons of chickpea on 239,747.51 hak. of land (CSA, 2015). The average productivity of Ethiopian chickpea in the same year was about 1.9 tons ha^{-1} which is below half of the 5.5 tons ha^{-1} that can be produced potentially (Belay *et al*.,2006). Chickpea is one of the major crops grown in the area next to *teff* and *wheat*. The overall chickpea production contributed for the betterment of producer farmers in food availability, farm income, household expenditure and productivity compared with the counter parts. Producer farmers generated more gross-margin and field benefits from chickpea compared to non-grower farmers and placed long time food availability. Ethiopia has the highest yield gain of chickpea among 10 producing countries in the world (Foyer *et al*., 2016).

According to CSA (2021) agricultural sample survey report in the belg cropping season of 2020/2021 about 4.6 million smallholder farmers cultivated 0.40 million hectares of land with pulse crops, from which about 2.58 million quintals of pulses was harvested. The amount produced is second next to cereal crops. Horse beans, field pea and chickpea take the first three largest proportions, which is about 0.055, 0.0204 and 0.015 million quintals, respectively. SNNPR, Oromia, Amhara and Sidama regions are the first four leading regions in producing pulses crops in the country. The total cultivated area under pulses in SNNPR, Oromia, Amhara and Sidama regions is 0.18, 0.103, 0.102, and 0.011 million hectares of land, respectively. Considering the volume of production these four regions, SNNPR, Oromia, Amhara and Sidama regions take the largest percentage proportions which are 1.19; 0.83; 0.42 and 0.09, respectively.

The crop can be grown on different soil types as long as good drainage is ensured. However, to achieve optimum growth well drained black soils (usually Vertisols) are identified as the most suitable soil type (Kassie *et al.,* 2009). In Ethiopia, chickpea is mostly grown on vertisols which have good water holding capacity. Ethiopian farmers essentially plant chickpea on residual moisture after the end of the main rainy season, which is usually in late August-October. Of the two types of chickpea, traditionally the *Desi* types are more widely cultivated in Ethiopia (Kassie *et al*., 2009). Even though there exist many improved chickpea cultivars released from Ethiopian Institute of Agricultural Research (EIAR) focused mainly on breeding and selection of improved cultivars with better yield and disease resistance, the mean average productivity is as low as 1.9 tons ha-^{1} (FAOSTAT, 2015) which is much below its potential productivity of 5.5 tons ha^{-1} (Belay, *et al.,*2006). Therefore, one way of improving yield of leguminous crops is inoculation of the seeds with *Rhizobium* bacteria proven to be effective in other African countries (Woomer *et al*., 2012).

2.4. Mechanism and Factors Affecting Symbiotic Nitrogen Fixation

2.4.1. Mechanism of Symbiotic Nitrogen Fixation

Binding of Rhizobia to seed surface is essential for establishing a symbiotic relationship between the host plant and bacterium. In early stages of symbiosis, a complex "molecular dialogue" takes place, which involves production of Nod factors by the bacterium and flavonoids by the roots of legume plants. This helps in the host recognition and initiation of the nodulation process. When bacteria colonize the root surface, it induces curling of root hair tips. Rhizobia caught in the root hairs, locally degrade the plant cell walls and form an invagination called infection thread through which bacterium reaches the cortical cells and within a week after infection, small sac like structures called nodules are visible on the roots of legume plants which enclose the bacteriod and are actual site for nitrogen fixation (Garg and Geetanjali, *2007* and Laranjo *et al*,. 2014). Rhizobia are able to metabolize atmospheric nitrogen and convert it into available form of nitrogen that plant can take up. In exchange, Rhizobia take advantage of carbon substrates derived from the plant photosynthesis.

2.4.2. Factors Affecting Symbiotic Nitrogen Fixation

The ability of symbiotic nitrogen fixing agents to fix N_2 is strongly influenced by the prevailing environmental conditions that can mainly be categorized as physical factors, chemical factors, and nutrient deficiencies (Giller *et al.*, 2001). The survival of *Rhizobium* sp. in the soil is influenced by a combination of factors such as nitrogen and phosphorus availability, acidity fertility status of the soil (Giller *et al*., 2001 and Slattery *et al.,* 2001). Soil Mineral Content Depending on the accessible quantity present, the mineral nitrogen content of the soil can have both positive and negative effects on yield and growth response of chickpea to inoculation. Usually, a higher mineral nitrogen content in the Rhizosphere leads to poor N2 fixation through inhibition of nodulation of chickpea (Namvar *et al*., 2011). On the other hand, small amounts of soil or fertilizer N often have a stimulatory effect on nodulation and N2 fixation which is principally due to the positive effect of N on growth and plant establishment during the period between root emergence and the onset of active N2 fixation (Giller and Cadisch *et al*., 1995).

The process of symbiotic nitrogen fixation is significantly influenced by the application of phosphorus (P) to the legumes. In presence of adequate phosphorus (P), the bacterial cells became motile and flagellate which is the pre-requisite for bacterial migration, but in the absence of phosphorus or with inadequate supply the infection remains latent leading to the poor development of nodules (Dutta and Bandyopadhyay *et al.*, 2009). High soil temperature is one of the most critical factors which can prevent the development of a symbiotic association between the host plant and microsymbiont especially in arid and semi-arid regions. For most of rhizobia, optimum temperature range for growth is 28 to 31^{o}C and majority of them are unable to grow at 38^{o}C (Graham *et al*,.1992). Temperature influences the survival of free rhizobia as well as the molecular dialogue between host and rhizobia. Elevated temperature can have inhibiting effect on micro symbiont adherence to root nodules, on nodule structure and on legume root nodule functioning (Zahran *et al*., 1999). Similarly, the soil pH greatly influences rhizobia content of soils and their ability to nodulate pulse crops (Slattery *et al*., 2004). Soil acidity reduces nitrogen fixation in legumes, particularly affecting *Rhizobium* survival in soil and reducing nodulation. Nodulation of soya bean and haricot bean was drastically reduced at pH of 4.5, whereas a pH of 5.2 resulted in good nodulation as well as satisfactory N2 fixation (Hungria and Varga *et al*., 2000).

Moreover, production of grain legumes is severely reduced in salt affected soils mainly due to the impairing effect of both salinity and sodicity on the plant ability to form and maintain nitrogen fixing nodules (Rao *et al*., 2002). For chickpea, very small nodule dry mass was recorded for all the genotypes tested under highly saline soil (Rao *et al*., 2002). With increasing salinity, a sharp decrease in both nodule number and nodule biomass was observed for all chickpea genotypes tested. In water logged conditions, there is inhibition of root hair development as well as nodulation sites. Moreover, normal diffusion of oxygen (O_2) in the root system of plants is also negatively affected. This lack of Oxygen (O_2) is a major problem for root respiration and results in inhibition of nitrogenase activity and nodule oxygen permeability (Serraj and Sinclair *et al*., 1996).

2.5. Effect of *Rhizobium* Inoculation on Growth and Seed Yield of Chickpea

Nitrogen fixers benefit the plant by providing them atmospheric nitrogen, which contributes to the development of plant growth and biomass production. Rudresh *et al*., 2005) studied the effect of inoculation with *Rhizobium* on growth attributes and observed that chickpea gave higher plant height (3.3%), number of branches per plant (23.3%) and biomass per plant (144%) as compared to uninoculated control. In similar findings (Elkoca *et al*., (2008) revealed that *Rhizobium* inoculation increased plant height; shoot dry weight and chlorophyll content in chickpea. The increment in the root length was also observed in the inoculated treatments (Solaiman, *et al.,* 2010) which in return resulted in increased root surface area. The increase in root surface area enhances the nutrient acquisition by plant from the soil (Yadav and Verma, *et al.,* 2014). Branches are subsidiary parameter of plant biometry which is important to realize higher seed yields. Singh *et al*., (2011) found that number of primary and secondary branches were higher when chickpea was components (Reddy *et al.,* 2012). Many researchers around the globe have reported positive effect of *Rhizobium* inoculation on various seed yield components viz. number of pods per plant, seeds per pod and 1000-seed weight in chickpea. Number of pods per plant and number of seeds per plant were reported to be 21.8% and 10.5% higher, respectively in chickpea inoculated with *Rhizobium* over uninoculated control (Sharar *et al.,* 2000).

Further Ali *et al*., (2004) revealed that 1000-seed weight was significantly better with inoculation. Khan *(*2003) in a similar study observed significantly higher number of pods per plant, number of seeds per plant and 1000-seed weight in chickpea. These findings are in agreement with that of Akhtar and Siddiqui *et al., (*2009) and Meena *et al.,* (2013). El Hadi and Elsheikh (1999) studied the effect of *Rhizobium* inoculation on the seed yield of chickpea and reported that inoculation with *Rhizobium* increased seed yield by 72 and 70% in the first and second year, respectively, in comparison to uninoculated control Similarly Maleki *et al.,* (2009) conducted a pot experiment in Iran and reported higher seed yield where rhizobial inoculation was done as compared to uninoculated control.

Ogola and Furthur, (2015) from South Africa reported increase in the seed yield to the tune of 7.9% with *Rhizobium* inoculation in a clayey and slightly acidic soil. The increase in the seed yield components through seed inoculation might be due to higher nodulation and more nutrient availability, resulting in vigorous plant growth and dry matter accumulation, which in turn resulted in higher seed yield (Namwar *et al.,* 2013 and Uddin *et al.,* 2014)**.** Residual soil nutrient status: Use of *Rhizobium* inoculants helps in improving available N and P in the soil after crop harvest which can be utilized by the next crop (Abdalla *et al.,* 2013). Similarly Chandra and Pareek (2015) recorded 0.6%, 6.5% and 4.3% higher organic carbon(C), available Nitrogen (N) and available Phosphorus (p), respectively, in a *Rhizobium* inoculated chickpea. These findings are in line with those of Zaidi *et al.,* (2003) and Tagore *et al., (*2013). Moreover, higher soil microbial biomass carbon was found in *Meso Rhizobium* inoculated chickpea (Bhattacharjya and Chandra *et al.,* 2013). These results may be attributed to production of more crop biomass due to inoculation and consequently higher return of organic residues and exudates into the soil enhancing microbial biomass and activities (Babu *et al.,* 2015). The pulse crops residues have lower C: N ratio, as a result of which immobilization after incorporation in the soil is low, which consequently enhances N availability to subsequent crops.

2.6. Effect of *Rhizobium* Inoculation on Seed Quality

Inoculation with *Rhizobium* strains not only have many nodules formed on chickpea genotypes, but also increased chickpea plant growth and yield, and improved seed protein and oil content. Inoculation with *Rhizobium* strains not only have many nodules formed on chickpea genotypes, but also increased chickpea plant growth and seed yield, and improved seed protein and oil content of seed’s quality. Inoculation *Rhizobium* showed the highest efficiency in increasing the seed yield. Rhizobial strains could significantly increase seed protein and oil contents of all chickpea genotypes. There were significant genotypic differences in terms of chickpea response to inoculation with Rhizobial strains. The results of many experiments are in agreement with previous studies that inoculation of legume seed with rhizobia inoculants increased settings of pods, the number of seeds per pod and per plant, and finally the quality of seeds (Rokhzadi *et al.,* 2008). Many researchers declared that inoculation with appropriate *Rhizobium* strains is an effective measure to increase N2 fixation, promote N nutrition and enhance yield in legumes (Mirza *et al.,* 2007). Meanwhile experiment, confirmed that inoculation with Rhizobial inoculants showe d its advantages in saline soil with significantly increasing chickpea shoot and root biomass and improve seed yield. It was revealed that different chickpea genotypes and Rhizobial strains differed in their tolerance to environmental stresses.

2.7. Seed Quality Parameters

2.7.1. Physical Quality

Quality seed is defined as varietal pure seed with a high germination, free from diseases and disease causing organisms and proper moisture content and weight (ISTA, 1985). Quality seeds imply good germination, rapid emergence and vigorous growth. Seed vigour is defined by the International Seed Testing Association (1987) as the sum total of those properties of the seeds that determine the level of activity and performance of the seed during germination and seedling emergence. Tests have been developed to evaluate the quality or vigour of seed lot. Seed vigour asses the ability to germinate under a wide range of field conditions (Woodstock 1969). According to Cromwell and Zambezi (1990) genetic quality of seeds refers to the true to typeness of the seed. Zewdie (2004) clearly elaborated the genetic quality of seed as: the inherent genetic information in the seed which provides the potential for higher yield, better grain quality, greater tolerance to biotic or abiotic stresses and varietal identity, i.e., the transfer of seed desired variety from the breeder to the farmer through successive generations of seed multiplications. If the seed possesses all the genetic qualities that the breeder has placed in the variety, it is said to be genetically pure (Agrawal *et al.,* 20015). The genetic quality has direct effect on ultimate yields. If there is any deterioration in the genetic makeup of the variety during seed multiplication and distribution cycle, there would definitely be proportionate decrease in its performance and disease resistance (Agrawal *et al.,* 2015).

Physical purity is the first seed quality attribute recognized in seed trade to protect farmers against the use of impure seed quality which is contaminated with other species, or inert matter. Analytical purity indicates the proportion of a seed lot that is pure seed of the species concerned. According to ISTA (1999) quality test determines the weight and the nature of the contaminant pre sent in the seed sample and by inference that of a seed lot it represents. Analytical purity analysis determines the percentage purity of the seed sample. Farmers require seed that is uncontaminated with seed quality of different crop species, or inert matter that may reduce the quality of their product (ISTA*,* 1996). Pure seed refers to the species stated by the sender, whereas other seeds include seeds and seed like structures of any species other than that of pure seed, and inert matter include soil, sand, stone, chaff, leaves, cone scales, and pieces of bark, flower, nematode galls, and fungus bodies such as ergot, schelerotia and bunt balls (ISTA,2001). Other seeds cover seed units of any plant species than that of pure seed while inert matter include seed units and all other matter and structures not defined as pure seed or other seed.

2.7.2. Physiological Seed Quality

The **Physiological quality** of the seed includes germination capacity and vigour. Germination is a major component of any seed quality assessment. After analyzing the physical purity, germination test is conducted from the pure seed. According to ISTA rules seed germination, is defined as normal seedlings produced during standard germination test. Germination refers to the ability of the seed to emerge and develop into a normal plant under favorable conditions (Deaker *et al.,* 2004). Seed germination, to the seed physiologi st, is the emergence of the radicle through the seed coat; to the seed analysis it is the emergence and development from the seed embryo of those essential structures (especially plumule and radicle), which, for the kind of seed in question, are indicative of the ability to produce a normal plant under favourable conditions (AOSA, 2000). The aim of standard germination test is to estimate the germination potential of a seed lot, which can then in turn be used to compare the quality of different lots and also estimate the field planting value of all crops (ISTA, 2001.)

Germination test when conducted accurately according to internationally standardized rules indicates the percentage of seed, fall under normal seedlings and /or abnormal seedlings. This failed to produce seedling (because they are dead or fresh germinated). Germination capacity indicate the percentage of pure seed fraction that produces normal seedling under optimal condition in the laboratory test and there by inference the field planning value under favorable environment in the soil (ISTA,1999). Seed vigor indicates that seed properties, which determine the potential for rapid, uniform emergence and development into normal seedlings under a wide range of field condition (Tekrony and *et, al.,* 1991). Vigor is not a single measurable character but a concept describing different attributes, which are associated with the various aspects of performance of germinating seed or the subsequently growing seedling (Agrawal *et al.,* 1987). According to Agrawal (1980) vigor can also be measured by dry weight of seedling and germination percentages. Measuring the length of seedling and their weight is often used to determine physiological seed vigor.

The results obtained from previous studies showed that vigor test could provide better results for ranking the quality and for indicating planting value of seed lots than the standard germination test. Sometimes standardized laboratory germination procedures are critized as not predicting field performance very well. Seed vigor tests can detect differences in potential seed lot performance. The critical requirements of a vigor test includes that (1) vigor test must provide a more sensitive index of seed quality than seed germination (Donald *et al.,*1980) and (ii) it must better predict planting value of high germination seed lot than seed germination (Hampton and Tekrony *et al.,*1995). Difference in vigor is also reflected in seed longevity. A high vigor seed lot has good storage potential and retains high germination potential during stage, whereas low vigor seed lots show poor storage potential and may show a rapid decline in germination (Hampon *et al.,* 1990)

3. MATERIALS AND METHODS

3.1. Description of the Experimental Site

The study was conducted at Ambo Agricultural Research Centre in Oromia Regional State, Ethiopia. The Centre is located at 08°57'N latitude 38°07' E longitude, and at an altitude of 2195 meters above sea level. The area has an annual average rainfall of 1036 mm and means annual maximum and minimum temperatures of 25.4 and 11.7 ^{0}C, respectively (Am ARC, 2018). Soil Type Vertisol (Heavy black clay soil) (Am ARC, 2018)

3.2. Description of Experimental Materials

The seeds of three chickpea varieties (*Eshete*, *Dimtu* and *Teketay*) produced in 2021 cropping season was collected from Debre Zeit and Ambo Agriculture Research Centers. The three varieties were released by Debre zeit Agriculture Research. In addition, the seeds of one chickpea cultivar largely produced by the farmers of the study area were collected from farmers who produced the cultivar seeds in 2021 cropping season. The description of chickpea varieties and farmers' cultivar is presented in Table 1.

Table 1 Description of improved varieties and farmers cultivar

	Variety			
Variable	*Eshete*	*Dimtu*	*Teketay*	Farmers' cultivar
Year of released	2020	2016	2013	----
Altitude (m.a.s.l.)	700-1900	1800-2800	1800-2700	----
Days to maturity	119-138	122	90-145	----
Yield on farmers field qt/ha	10-14	23-36	16-22	----
Yield in research site qt/ha	10-18	25-47	20-27	----
Seed rate kg ha^{-1}	60-90	120-140	130-135	----
Breeder center	DZARC/EIARRR	DZARC/EIARR	DZARC/EIAR	----

Source: Ministry of Agriculture Plant Variety Release, Protection and Seed Quality Control Director. (In .2020, 2016 & 2013)

3.3. Treatments and Experimental Design.

The treatments were consisting of three *Rhizobium* strains namely, CP41 (commercial), CP11 (local) and CP17 (local) inoculation and without inoculation of four chickpea varieties (three improved and one farmer's cultivar). Therefore, there was a total of 16 treatments as

4 (inoculations) x 4 (varieties) in factorial combination. The treatments were laid out in randomized complete block design (RCBD) with three replications at field and complete randomized design (CRD) with four replications at laboratory. A treatment was randomly assigned to each plot at field. The size of individual plots was 1.2 m and 4m width and length, respectively. The seed was placed at 0.1 m and 0.3 m intra-row and inter-row spacing, respectively. The gap between blocks and plot was 1m and 0.5m, respectively.

3.4. Experimental Procedures and Management

3.4.1. *Rhizobium* Inoculant and Inoculation

The strains CP 11 and CP 17 was obtained from the soil microbiology of Holetta Agriculture Research Centre. The inoculant of the two strains were isolated and purified from the nodules of local chickpea genotype Ararti grown in green house and innoculated by the two *Rhizobium* strains. Strain CP 41 was obtained from Damot Gale district, Southern Nations, Nationalities and Peoples Regional State by Menagesha biotech. (Collee and Mackie *et al.,* 1989). The inoculant CP 41 *Rhizobium* strain was isolated and purified by Menagasha biotech institute from local strain identified in Damot Gale district, Southern Nations, Nationalities and Peoples Regional State.

The seeds inoculation was performed before sowing using the procedure developed by (Fatima *et al*., 2007). The inoculation with peat-based inoculums seeds was moistened in sugar solution (48%) before application of inoculums to get a thin uniform coating of inoculums on seeds immediately before sowing. 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 10g per kg of seed. Inoculation was done just under shade to maintain the viability of cells and allow to air dry for a few minutes and then the inoculated seeds was sown at recommended rate and spacing to the respective plots. To avoid contamination, plots with un-inoculated seeds were planted first followed by the inoculated ones.

3.4.2. Field Management

Land preparation was done using tractor at Ambo Agricultural Research. At planting the experimental plots was prepared well and according to the design, field layouts was made and each treatment was assigned randomly to the experimental units within a block. Planting was done on September 29, 2022. Hand weeding was practiced three times during the growing period. Application of nitrogen, phosphate and sulfur (NPS) 121kg /hek as per of recommendation was done at the depth of 5cm below and 5cm around the seed at panting. Harvesting was done when the leaves of the chick-pea plant senescent and showed yellowing of leaves and threshing was done manually.

3.4.3. Seed Quality Tests at Laboratory

A seed sample of 1 kg was obtained from each experimental (Plots) treatment for laboratory analysis. The 1kg of seeds from each treatment was divided into 4 each 250 g which was used for the laboratory tests that include purity, moisture content determination, germination and vigour. Each sample was sorted into three components that include pure seed, inert matter and other crop as suggested by the International Seed Testing Association (ISTA, 2014). Sand was used as a substrate for germination. The germination test was done from the pure seed fractions obtained from the purity test. Four hundred seed (400) pure seeds were taking from each treatment, and divided into four replicates of 100 seeds. Each replicate was sown on a sterilized sand medium on plastic box. Finally, at the 10^{th} day of the planting seedling was categorized in to normal, abnormal, ungerminated seeds, dead seeds and their percentages was calculated.

3.5. Data Collection

3.5.1. Crop Phenology and Growth

Days to emergency (DE**):** Number of days recorded from sowing to the stage when 50% of the plants in the plot emerged.

Days to flowering (DF): Number of days recorded from sowing to the stage when 50% of the plants in the plot showed the first flower.

Days to maturity (DM): The number of days from sowing to the stage when 90% of the plants in a plot have reached physiological maturity.

Plant height (cm): plant height from ground level to the tip of main stem at 90% physiological maturity, it was measured using a ruler on ten plants randomly taken from the middle rows.

3.5.2. Seed Yield and Yield Components

Number of pods per plant: This refers to total number of pods per plant at full maturity, and was obtained from ten randomly sample plants within the entire net plot area

Number of seeds per pod: This refers to the total number of seed per pod, and was obtained from 20 randomly sample pod.

Hundred seed weight (g): Was determined by weighting the weights of 100 seeds sample from plot using an electric sensitive balance.

3.5.3. Production Efficiency.

Seed production efficiency (SPE): Seed filling duration divided by duration of vegetative period and then multiplied by grain yield.

Grain yield per day (GYPD): Calculate as the ratio of gain yield (kg ha^{-1}) to days to physiological maturity and expressed as kg ha^{-1} day^{-1}.
The chickpea varieties seed yield response to the three *Rhizobium* strains namely, CP41, CP11 and CP17 inoculation was estimated as inoculation sensitivity index and yield index with modification of stress sensitivity index and yield index proposed by Gavuzzi *et al*.,1997 and Farshadfar and Javadinia *et al*.,2011), respectively, as follow:
Rhizobium strains sensitivity index (RSSI): it was estimate for each variety with each *Rhizobium* strain inoculation in comparison to the seed yield obtained from plot without inoculation as follow:
Rhizobium strains sensitivity index (RSSI): $\frac{YR-YUI}{YUI}$, Where; YR = seed yield obtained from plot inoculated with each *Rhizobium* strain and YUI = seed yield obtained from plot without inoculation of four chickpea varieties in each replication.

Yield index: it was estimate for each variety as ratio of mean seed yield of variety obtained from plot inoculated with all *Rhizobium* strains to seed yield obtained from plot without inoculation in each replication as follow:

Yield index: µYs/µYUI. Where; µYR = mean seed yield of each variety obtained from plot inoculated with all *Rhizobium* strains and µYUI = mean seed yield of each variety obtained from plot without inoculation in each replication.

3.5.4. Data Collected from Seed Quality Test

3.5.4.1. Physical purity

A seed sample of 1000 g was obtained from each experimental treatment for laboratory analysis.1000 g of seeds from each treatment was used for the laboratory tests that include purity, moisture content determination, germination, vigour and seed health tests. Each sample was sort into three components that include pure seed, inert matter and other crop as suggested by the International Seed Testing Association (ISTA, 2014). The percentage of each fraction will be recorded on a weight-by-weight basis.

$$\text{Purity (\%)} = \frac{\text{Weight of pure seed (g)}}{\text{Total weight of sample (g)}} \text{ x } 100$$

Seed moisture content (%): The amount of water within the seed will determined with low constant temperature oven method and the temperature was maintained at 103±2C^0 and the seed was dried for 17±1 hours. The difference between the weight of fresh seed and the dry one will be established by the formula:

$$\text{MC} = \frac{\text{M2}-\text{M3}}{\text{M2}-\text{M1}} \times 100$$

Where; MC= Seed moisture content.

M1=Weight of the empty container with its cover

M2=Weight of the container with its cover and seeds before drying.

M3=Weight of the container with its cover and the seed after drying.

3.5.4.2. Seed physiological test

Standard germination (StG) test: The germination test for chickpea was done at the temperature of 25^{o}C (ISTA, 1996). Thus, the first and second count was done at 8 days after planting, respectively. Sand was use as a substrate for germination. The germination test was done from the pure seed fractions obtain from the pure test. Four hundred seed (400) pure seeds was taken from each treatment, and divide into four replicates of 100 seeds. Each replicate was sown on a sterilized sand medium on plastic box. Finally, at the 10 day of the planting seedling was categorized in to normal, abnormal, un germinated seeds, dead seeds and their percentages was calculated.

$$\text{StG (\%)} = \frac{\text{Total number of normal seedling}}{\text{Total number of seeds planted}} \times 100$$

Seedling Vigour Test: Seed vigour test was determined by measuring shoot and root length of seedlings. The seedling shoot and root length was measured after the final count of the germination test. Ten normal seedlings were randomly selected from each replicate and shoot length was measured from the point of attachment to the cotyledon. Similarly, the root length was measured. The averages of shoot and root a length was computed by dividing the total shoot or root lengths by the total number of normal seedling measured (Fiala *et al.*, 1987).

Seedling dry weight: The seedling dry weight was measured after the final count of the StG test. Ten randomly selected seedlings from each replicate was selected and placed in envelopes and dried in an oven at 80°C for 24 hrs. The dried seedling was weighed by using sensitive balance and the average seedling dry weight was calculated.

Vigour index one and Vigour index two: for each treatment, two vigor indexes were calculated. Seedling vigor index one was calculated by multiplying the number of normal seedlings with the average sum of shoot and root length and vigor index two was calculated by multiplying the standard germination percentage with mean seedling dry weight.

Mathematically; Vigor Index One (VIG-I) = Average Seedling Length × Normal seedling % Vigor Index Two (VIG-II) =Normal seedling % × Seedling mean Dry Weight

Speed of germination: Speed of germination was calculated from the daily germination records. In case chickpea speed of germination shall be obtained was 10 days of germination period; the first count was done on the 5^{th} day. Finally, speed of germination was calculated using the following formula which was given by (Fiala *et al.*, 1987).

$$Speed\ of\ Germination = \frac{N1}{C1} + \frac{N2}{C2} + \cdots + \frac{NF}{CF}$$

Where: N1= number of normal seedlings at first count, N2= number of normal seedlings at second count, NF= number of normal seedlings at final count, C1= days to the first count, C2= days to the second count and CF= days to the final count.

3.6. Data Analysis

The data that was collected from field experiment was subjected to analysis of variance (ANOVA) for RCBD factorial while the data that was collected from seed quality test was subjected to ANOVA for using CRD factorial using Gen Stat 15th edition statistical software package. The mean values comparison was performed following the significance test results from ANOVA using least significant difference (LSD) at 5% level of probability.

4. RESULTS AND DISCUSSION

4.1. Seed Yield and Yield Related Traits of Chickpea Varieties

4.1.1. Phenology and Growth of Chickpea Varieties

4.1.1.1. Phenology of chickpea varieties.

Analysis of variance revealed that the chickpea varieties had significant differences for days to emergency, days to flowering and days to maturity. The main factor inoculation of *Rhizobium* strains and the interaction of variety and *Rhizobium* strains had nonsignificant effect on phenology of chickpea (Appendix Table 1). Longest duration (13 days) to days to emergence took for *Dimtu* variety while shortest days to emergence registered for *Eshete* (11.66 days) variety with statistical parity with *Teketay* and farmers cultivar of chickpea. *Eshete* variety showed delayed days to flowering (64.33 days) and *Teketay* variety was late for maturity (136.33 days) but had non-significant difference for days to maturity of *Eshete* variety. The farmers' cultivar took shortest duration to days to flowering (53.25) and maturity (130 days) (Table 2). The presence of variations among varieties was also indicated in variety register that *Teketay* variety had up to 145 days to maturity (MoANR, 2013) while *Eshete* and *Dimtu* varieties had up to 138 and 122 days to maturity, respectively (MoANR, 2020 and 2016).

Other authors also reported the presence of significant variations among Desi type chickpea genotypes in Ethiopia. Awol and Bulti (2018) reported significant variation among 202 Desi type chickpea landraces and two released varieties (*Fetenech* and *Minjar*) evaluated at Sirinka and Jari, North Wollo. Zerihun and Shiferaw (2019) also observed significant differences among 19 elite varieties for days to flowering and maturity. Sintayehu,(2021) reported significant variations among 79 chickpea genotypes and two improved varieties for days to flowering and maturity evaluated at Debre Zeit for two consecutive years.

Table.2**.** Variation of chickpea varieties for days to emergence, flowering and maturity at Ambo in 2022/2023 cropping season

Variety	Days to emergence	Days to flowering	Days to maturity
Eshete	11.66^{b}	64.33^{a}	135.42^{a}
Dimtu	13.00^{a}	55.25^{b}	132.92^{b}
Teketay	12.75^{b}	55.50^{b}	136.33^{a}
Farmers cultivar	12.00^{b}	53.25^{c}	130.0^{c}
LSD (0.05)	0.47	1.39	1.94
CV (%)	4.62	2.93	1.74

Mean values within column designated with similar letters are not significant each other at $p<0.05$, LSD (5%) = Least significant difference at $p\leq 0.05$ and CV (%) = percentage of coefficient of variation.

4.1.1.2. Interaction effect of variety and *Rhizobium* strain on plant height of chickpea.

The plant height of chickpea was significantly influenced by the main factors variety and inoculation of *Rhizobium* strains as well as the two factors interaction (Appendix Table 1). The seeds of *Eshete* variety innoculated with CP17 *Rhizobium* strain had tallest plants (52.26 cm); however, it had nonsignificant difference with the height of plants grown from seeds of *Eshete* and *Teketay* varieties innoculated with CP11 and CP41 *Rhizobium* strains. The seeds of farmers' cultivar sown without inoculation and innoculated with CP11 and CP41 *Rhizobium* strains had shorter plants with non-significant difference for height of plants. The seeds of *Dimtu* variety without inoculation and innoculated with CP41 *Rhizobium* strain also had shorter plants and taller plants of this variety were observed at plots where the plants grown from seeds innoculated with CP17 *Rhizobium* strain but had nonsignificant difference with plants grown from seeds of *Teketay* variety innoculated with CP11 *Rhizobium* strain (Table 3).

The results showed that varieties were responded to different *Rhizobium* strains in terms of plant height and the compatibility of varieties to *Rhizobium* strains also varied. The seeds of *Teketay* and *Eshete* varieties inoculated with the three *Rhizobium* strains showed increased the height of plants ranged from 29.84 to 34.83% and 15.46 to 20.06%, respectively, as compared to plants grown from seeds of varieties without inoculation. On the other hand, seeds of farmers' cultivar and *Dimtu* variety innoculated only with CP17 *Rhizobium* strain had significantly taller plants by 14.09 and 13.91%, respectively, than plants grown seeds without inoculation (Table 3). The inoculations of chickpea seeds with *Rhizobium* strains help the plants to fix atmospheric nitrogen which contributes to the development of plant growth that increase height of plants (Rudresh, *et al.,* 2005; Elkoca, *et al.,* 2008). The presence of compatibility and differences of effective rhizobia strains among chickpea varieties was also reported by other authors (Mulongoy 1995; Shantharam and Mattoo *et al.,* 1997). Gemechu, *et al*., (2013) suggested the need to conduct specific tests for specific breeding materials, strain and environments to identify host genotype compatible to specific *Rhizobia* strain.

Table 3. Interaction effects of variety and *Rhizobium* strain on plant height of chickpea varieties at Ambo in 2022/2023 cropping season

Treatment	*Rhizobium* strain			
Variety	CP11	CP17	CP41	Control

Eshete	50.26^{ab}	52.26^{a}	51.66^{ab}	43.53^{def}
Dimtu	44.00^{cde}	47.73^{bcd}	43.03^{ef}	41.90^{ef}
Teketay	47.60^{bcd}	49.43^{ab}	48.06^{abc}	36.66^{gh}
Farmers cultivar	36.26^{gh}	39.36^{fg}	35.36^{gh}	34.50^{h}
LSD (0.05)		4.36		
CV (%)		5.98		

Mean values in columns and rows designated with similar letters are not significant each other at $p<0.05$. LSD (5%) = Least significant difference at $p\leq0.05$ and CV (%) = percentage of coefficient of variation.

4.1.2. Yield Components and Seed Yield of Chickpea.

4.1.2.1 Influence of varieties and *Rhizobium* strains on yield components and biomass yield.

Rhizobium strains have significaly increase the seed yield and quality of variety with possibility due to the extremeness of strain of solubilization and availability of nutrients. Analysis of variance revealed that chickpea varieties and inoculation of seeds with *Rhizobium* strains significantly influenced number of seeds per pod and biomass yield, while hundred seeds weight was influenced by varieties. Chickpea varieties and *Rhizobium* strains did not interact to influence number of seeds per pod; hundred seeds weight and biomass yield (Appendix Table1). *Teketay* (2.49kg) and *Eshete* (2.42 kg) varieties produced higher biomass yield per plot while farmers cultivar produced lower biomass yield of 1.68 kg per plot. *Teketey* variety produced higher biomass yield than *Eshete, Dimtu* and farmers' cultivar by about 2.89, 14.75 and 48.21%, respectively (Table 4). Amare *et al*., (2020) reported significant difference among 100 advanced lines of *Desi*-type chickpea germplasms evaluated at two location of North Gondar.

Farmers' cultivar, *Eshete* and *Teketey* varieties produced 1.64, 1.61 and 1.56 average number of seeds per pod, respectively, without significant difference each other, but significantly higher number of seeds per pod of *Dimtu* variety. *Dimtu* variety had significantly highest hundred seeds weight (34g) and *Teketey* variety had also heavier hundred seed weight (28.75g) next to the Farmers' cultivar whereas farmers cultivar followed by *Eshete* variety had lower hundred seed weight (Table 4). Amare *et al*., (2020) reported 1 to 1.7 with 1.15 mean numbers of seeds per pod and 10.1 to 35.5g with 22.7g average hundred seeds weight of 100 advanced lines of *Desi*-type chickpea germplasms evaluated at two location of North Gondar. Yasin and Alamir (2023) reported 1.03 to 1.53 numbers of seeds per pod and 14.83 to 32.17g hundred seeds weight of eight chickpea varieties evaluated in two districts of South Gonder, northwestern Ethiopia. Ahmad *et al*., (2003) and Jakhar (2014) reported significant variability in chickpea genotypes for seeds per pod and hundred seeds weight.

The seeds of chickpea varieties inoculated with CP17 *Rhizobium* strain produced highest biomass (2.48 kg) but had nonsignificant difference with biomass yield of chickpea varieties produced from seeds inoculated with CP 11 *Rhizobium* strain. The uninnoculated seeds and seeds of chickpea varieties inoculated with CP 41 *Rhizobium* strain produced lowed biomass yield than the former varieties but without significant difference each other. The seeds of chickpea varieties inoculated with CP17 *Rhizobium* strain produced highest number of seeds per pod (1.8) and seeds inoculated with CP 11 and CP 41 *Rhizobium* strains produced significantly lower number of seeds per pod than seeds innoculated with CP17 strain but nonsignificant difference with each other. The seeds of chickpea varieties sown without inoculation produced lowest number seeds per pod (Table 4)

The *Natoli* chickpea variety produced 1.6 numbers of seeds per pod from plants grown from inoculated seeds by CP41 *Mesorhizobium ciceri* strain but plants grown from uninnoculated seeds produced 1.4 seeds per pod. Inoculation of seeds had also a positive and highly significant effect on total biomass of chickpea variety (Ibsa *et al.,* 2013).

This is because the increased supply of nitrogen through biological nitrogen fixation played important roles in enhanced growth and assimilate accumulation, thereby improving the reproductive performance of the plants and total biomass of chickpea. This is in agreement with many authors reported that inoculation of chickpea seeds highly significantly improved the supply of nitrogen that had positive effects on reproductive performance, vegetative growth and assimilate accumulation of plants which in turn increase seeds per pod and biomass yield (Ali *et al*., 2004; Elkoca *et al*., 2007; Togay *et al*., 2008; Dutta and Bandyopadhyay *et al.,* 2009; Ahmed *et al.,* 2010 and Jakhar *et al.,* 2014).

Table 4 . Effects of Variety and *Rhizobium* strain on biomass yield, number of seed per pod and hundred seed weight chickpea at Ambo in 2022/2023 cropping season

Variety	Biomass yield (kg)	Number seed per pod	Hundred seed weight (g)
Eshete	2.42a	1.61a	20.41c

Dimtu	2.17b	1.45b	34a
Teketey	2.49a	1.56a	28.75b
Farmers cultivar	1.68c	1.64a	13.33d
LSD (0.05)	0.22	0.09	2.41
Rhizobium strains			
CP 11	2.33a	1.59b	25.25
CP 17	2.48a	1.80a	25.25
CP 41	2.02b	1.57b	24.41
Control	1.93b	1.30c	22.02
LSD (0.05)	0.22	0.09	NS
CV (%)	12.46	7.55	11.99

Mean values within column designated with similar letters are not significant each other at p<0.05., NS = Nonsignificant, LSD (5%) = Least significant difference at p≤0.05 and CV (%) = percentage of coefficient of variation.

4.1.2.2 Interaction effect of variety and *Rhizobium* strain on number pods per plants of chickpea.

Number of pods per plant was significantly influenced by varieties, *Rhizobium* strains and the interaction of the two factors (Appendix Table 1). Highest number of pods per plant (114.66) was obtained from *Eshete* variety inoculated by CP17 *Rhizobium* strain which had statistical parity with *Dimtu, Teketay* and farmers' cultivar inoculated with the same CP17 strains. *Eshete* variety without inoculation had the lowest number of pods per plant (69) which had no statistical difference with seeds of same *Eshete* variety inoculated by CP11 and CP41, *Dimtu, Teketay* and farmers' cultivar without inoculation and farmers cultivar inoculated by CP11 and CP41 *Rhizobium* strains. *Eshete* variety was more responsive to the three *Rhizobium* strains to increased number of pods per plant. All other varieties increased number of pods per plant when inoculated with CP17 *Rhizobium* strain (Table 5). The reports Karadavut and Ozdemir *et al.,* (2001) are in agreement with this finding that inoculation of chickpea variety by *Rhizobium* strain increase number of pod per plant as compared to control. Number of pods per were reported to be higher, in chickpea inoculated with *Rhizobium* over uninoculated control (Sharar *et al.*, 2000; Khan *et al.*, 2003). Tena *et al.*, (2016) also reported a 48% increment of number of pods per plant through inoculation of indigenous CP-41 Rhizobial strain over the uninoculated seeds.

Table 5. Interaction effects of variety and *Rhizobium* strain on number of pod per plant of chickpea varieties at Ambo in 2022/2023 cropping season.

Treatment	*Rhizobium* strain			
Variety	CP11	CP17	CP41	Control
Eshete	80.66efg	114.66a	76.66fg	69.00g
Dimtu	96.33bcd	104.66abc	83.66def	81.33efg
Teketay	91.66cde	106.00ab	99.00bc	81.66efg
Farmers' cultivar	70.33fg	102.66abc	77.66fg	75.66fg
LSD (0.05)		13.82		
CV (%)		9.42		

Mean values in column and rows designated with similar letters are not significant each other at p<0.05, LSD (5%) = Least significant difference at p≤0.05 and CV (%) = percentage of coefficient of variation.

4.1.2.3. Seed yield

The yield of chickpea was significantly influenced by varieties and inoculation of *Rhizobium* strains, but the interaction of the two factors had nonsignificant effect on it (Appendix Table 1). *Teketay* variety produced highest yield (2777.78 kg/ha) but had nonsignificant difference with yield (2552.08 kg/ha) of *Dimtu* variety. *Eshete* variety produced statistically nonsignificant different grain yield of *Dimtu* variety.

Farmers' cultivar produced significantly lowest yield. *Teketay* variety had 8.84, 17.65 and 37.93% yield advantages over *Dimtu, Eshete* and Farmers cultivar, respectively (Table 6).

The difference of *Teketay, Dimtu* and *Eshete* varieties for yield potential at farmers field and research site were reported at the time of varieties release (MoA, 2020, 2016 and 2013). According to the variety register description, *Dimtu* variety released in 2016 had higher yield potential at farmers field (2300 to 3600 kg /ha) and research site (2500 to 4700 kg/ha) than the two varieties (Table 1). However, the yield of varieties might not be the same all environments due to effect of environment and genotype x environment interactio n*Dimtu* and *Teketay* varieties produced 3218.9 and 2296.3 kg/ha yield, respectively, at Kobo district, North Wollo under irrigation during 2018 and 2019 (Awol *et al*., 2020). *Dimtu* and *Teketay* varieties produced 1740 and 1970 kg/ha mean yield, respectively at five locations of Western Ethiopia, during the 2016/2017 main cropping season (Biru and Dagnachew, *et al*., 2018). The highest yield (2690.97 kg/ha) and significantly different from other treatments was produced from seeds inoculated with CP17 *Rhizobium* strain**.** The seeds inoculated with CP41 *Rhizobium* strain produced the second higher yield but had nonsignificant with seeds innoculated with CP11 *Rhizobium* strain while the lowest yield was obtained from uninnoculated seeds. The inoculation of chickpea seeds with CP17 *Rhizobium* strain had 25, 11.51 and 9.93,% yield advantages over uninnoculated seeds, innoculated seeds with CP11 and CP41 *Rhizobium* strains, respectively (Table 6).

Various studies on *Rhizobium* inoculation have proved its beneficial effects in increasing the performance of yield components of chickpea. Seed inoculation dramatically increases the nodulation which in turn causes produces plants of high vigor. This results in more accumulation of dry matter and ultimately higher grain yield is obtained (Namvar *et al*, 2013 and Uddin *et al*, 2014). One of the major limiting factors in increasing the production of crops is Nitrogen deficiency. This drawback of Nitrogen deficiency can be addressed to a great extent by the use of bio fertilizers. As a legume, chickpea can obtain a significant portion of its N requirement through symbiotic N_2 fixation when grown in association with effective and compatible *Rhizobium* strains, but there is variability in amount of N_2 fixation due to biotic and abiotic factors that can influence the success of the legume-*Rhizobium* association (Fran *et al.,* 2004). The finding of this research revealed that the inoculation of chickpea seeds with CP17 *Rhizobium* strain was better than the other two *Rhizobium* strains (CP11 and CP41). The inoculation of seeds with CP17, CP41 and CP11 *Rhizobium* strains produced 25, 13.7 and 12.1% yield advantages over uninnoculated seeds, respectively (Table 6). The legume yields and nitrogen fixation depends on the genotype of the legume, the *Rhizobium* strain and the interactions of these with the bio-physical environment and management practices (Giller *et al*., 2013), thus, it is important to identify effective *Rhizobium* strain(s) compatible to the cultivar(s) and growing area(s).

Table 6. Effects of variety and *Rhizobium* strain on seed yield of chickpea at Ambo in 2022/2023 cropping season

Variety	Seed yield kg/ha.
Eshete	2361.11b
Dimtu	2552.08ab
Teketay	2777.78a
Farmers cultivar	2013.89c
LSD (0.05)	290.65
Rhizobium strains	

CP 11	2413.19bc
CP 17	2690.97a
CP 41	2447.92b
Control	2152.78c
LSD (0.05)	290.65
CV (%)	14.38

Mean values within column designated with similar letters are not significant each other at $p<0.05$, LSD (5%) = Least significant difference at $p\leq0.05$ and CV (%) = percentage of coefficient of variation.

4.1.3. Production Efficiency

Analysis of variance revealed that chickpea varieties and *Rhizobium* strain had significant effect on seed production efficiency, grain yield per day, *Rhizobium* strains sensitivity index for varieties but effectiveness and compatibility for strains and yield index. However, the interaction of variety and *Rhizobium* strains had non-significant effect on production efficiency of chickpea (Appendix Table 1).

Teketay and *Dimtu* varieties were better for seed production efficiency, grain yield index p er day, *Rhizobium* sensitive index and yield index with nonsignificant difference between the two but significantly higher than *Eshete* variety and farmers' cultivar though *Eshete* variety showed statistically nonsignificant difference with the farmer two for yield index and with *Dimtu* variety for *Rhizobium* sensitive index. *Eshete* variety and farmers cultivar had nonsignificant difference for seed production efficiency and grain yield index per day but this variety was better than farmers' cultivar for *Rhizobium* sensitive index and yield index (Table 7). In this as other authors suggested the high yielding varieties alone may not result in the required level of productivity without inclusion of compatible and effective *Rhizobia* strains. A given legume cultivar nodulated by different strains of the same species of *Rhizobium* would fix different amounts of nitrogen and thereby yield (Mulongoy, *et al*., 1995 and Shantharam *et al*., 1997). Thus, this research results showed that *Teketay* followed by *Dimtu* varieties were better compatible to the *Rhizobium* strains used for seed inoculation.

The CP17 *Rhizobium* strain was best for seed production efficiency, grain yield index per day, *Rhizobium* sensitive index (effectiveness and compatibility) and yield index though it had statisticaly nonsignificant difference with the other two strains for seed production efficiency and grain yield index per day and with CP41 *Rhizobium* strain for yield index. The control plot showed significantly lowest values for all estimates except it had nonsignificant difference with CP11 *Rhizobium* strain for yield index (Table 7). It is generally agreed that the symbiotic association operates at highest efficiency in Nitrogen limiting systems, particularly if other potentially limiting factors are supplied in adequate quantities (Abaidoo *et al*., 1990 and Abdiev *et al.,* 2019).

A given strain of *Rhizobium* wills nodulate and fix different amounts of Nitrogen in symbiosis with a range of cultivars of the same plant species (Mulongoy *et al*., 1995). Thus, CP17 could be considered as best *Rhizobium* strain in the study area to increase the productivity of chickpea. But chickpea may be inferior in terms of N_2 fixation (Smith *et al*., 1987), particularly where native Rhizobial populations are low or inefficient (Beck 1992). Thus the lowest estimates of seed production efficiency, grain yield index per day, *Rhizobium* sensitive index.

Table 7.Variation of chickpea varieties and *Rhizobium* strains for seed production efficiency, grain yield per day, *Rhizobium* strains sensitivity index and yield index

Variety	SPE	GYPD	RSSI	YI
Eshete	2412.67b	20.00b	1874.25b	1.17a
Dimtu	3235.00a	23.52a	2030.50ab	1.18a
Teketay	3535.46a	25.27a	2117.30a	1.12a
Farmers cultivar	2593.83b	18.89b	1527.10c	0.85b
LSD (0.05)	387.08	2.73	196.36	0.24
Rhizobium strains				
Cp 11	2932.32ab	21.75ab	2412.19b	0.96c
CP 17	3286.03a	24.24a	2689.97a	1.29a
Cp 41	2950.91ab	22.16ab	2446.92b	1.15ab

Control	2607.70c	19.53c	0.07c	0.91c
LSD (0.05)	387.08	2.73	192.36	0.24
CV (%)	15.76	14.98	12.22	16.89

Mean values within column designated with similar letters are not significant each other at $p<0.05$, LSD (5%) = Least significant difference at $p\leq0.05$ and CV (%) = percentage of coefficient of variation, SPE=Seed production efficiency; GYPD= Grain yield index per da y; RSSI =*Rhizobium* sensitive index; YI=yield index

4.2. Seed Quality Test

4.2.1. Seeds physiological quality test

4.2.1.1. Effect of variety and *Rhizobium* strain on seed germination and speed of germination.

Percentage of normal seedling and speed of germination was significantly influenced by chickpea varieties. Percentage of normal seedling was also significantly influenced by interaction of chickpea variety with *Rhizobium* strain. However, both percentage of normal seedling and speed of germination was not significantly influenced by main effect of *Rhizobium* strain. Similarly, proportion of dead seeds and abnormal seedlings was not significantly influenced by main effect of variety, *Rhizobium* strain and their interaction (Appendix Table 2). Highest speed of germination (17.8) was registered for Farmers cultivar which had statistical parity with speed of germination from both *Dimtu* and *Teketay* variety but lowest speed of germination (16.13) was registered from *Eshete* variety (Table 8). The significant difference observed among chickpea varieties for speed of germination of the crop indicated the varieties had inherent variations for the studied seed physiology parameters. More seedlings per a day were emerged for Farmers cultivar. Also, both *Dimtu* and *Teketay* varieties was fastest emerged per a day but *Eshete* variety was delayed to emerged per a day. Toumey and Korstian (1942) emphasized rapid germination as an important component of the seed vigor, and it usually corresponds to more rapid seedling emergence in the field. Seeds that have high germination speed were found vigorous in the field and could be escaped harsh conditions.

Table 8. Main effect of variety on speed of germination of chickpea

Variety	SG (No/days)
Eshete	16.13b
Dimtu	17.13a

Teketey	16.95a
Farmers cultivar	17.8a
LSD (0.05)	0.60
CV (%)	4.99

Mean values within column designated with similar letters are not significant each other at p<0.05, LSD (5%) = Least significant difference at p≤0.05 and CV (%) = percentage of coefficient of variation.

4.2.1.2. Interaction effect of variety and *Rhizobium* strain on normal seedling of chickpea.

The normal seedling percentage of chickpea was significantly influenced by the main factor variety and the two factors interaction (Appendix Table 2). Highest normal seedling percentage (98.75%) was registered from *Teketay* variety inoculated by Cp11 strain which had statistical parity with normal seedling percentage from *Eshete, Dimtu* and Farmers cultivar inoculated by Cp17 strains and no inoculation and it also had non-significance difference with local variety inoculated by Cp11 and Cp41 and *Teketay* variety inoculated by Cp17 strain. *Dimtu* variety inoculated by Cp11 and Cp41 had least normal seedling (97.25%) which had no statistical difference with *Teketay* variety without inoculation (Table 9). The study showed that significant difference observed among chickpea varieties and inoculation of *Rhizobium* strains for normal seedling percentage of chickpea. This indicated that the chickpea varieties had significant difference for inherent characteristics of normal seedling percentage which might be the function of genetic factor and *Rhizobium* strain compatibility. *Teketay* variety was more responsive to Cp11 and Cp17 strains and increased the normal seedling also Farmers cultivar increased normal seedling when innoculated with all *Rhizobium* strains. *Dimtu* and *Eshete* varieties increased normal seedling percentage when inoculated with Cp17 strain and no inoculation. Adebisi (2011) who reported that seed size is one of the components of seed quality which affect the performance of the whole plant and their germination.

Table **9**. Interaction effects of variety and *Rhizobium* strain on percentage of normal seedlings germination of chickpea Varieties

Variety	*Rhizobium* strain	Normal seedling (%)
	Cp11	97.75bcd
	Cp17	98.00abcd
Eshete	Cp41	97.50cd
	Control	98.25abc

Dimtu	Cp11	97.25d
	Cp17	98.00abcd
	Cp41	97.25d
	Control	98.50ab
Teketay	Cp11	98.75a
	Cp17	98.00abcd
	Cp41	97.75bcd
	Control	97.25d
Farmers cultivar	Cp11	98.50ab
	Cp17	98.75a
	Cp41	98.00abcd
	Control	98.75a
LSD (0.05)		0.95
CV (%)		0.68

Mean values within column designated with similar letters are not significant each other at p<0.05, LSD (5%) = Least significant difference at p≤0.05 and CV (%) = percentage of coefficient of variation.

4.2.1.3. Effect of variety on seedlings shoot length, root length and dry weight

Analysis of variance revealed that both main effect of variety and interaction of variety with *Rhizobium* strain had significant influence on shoot and root length. In addition, root length and seedling dry weight was significantly influenced by main effect of *Rhizobium* strains. Seedling dry weight also had significantly influenced by chickpea variety but not significantly influenced by the interaction of variety with *Rhizobium* strain. Main effect of *Rhizobium* strain had not significantly influenced on shoot length (Appendix 2). The highest seedling dry weight (0.64gm) was registered for seeds of *Dimtu* variety while least seedling dry weight (0.57gm) was measured for Farmers cultivar of chickpea. The highest seedling dry weight (0.61gm) was registered for both seed of chickpea inoculated by both Cp17 and Cp41 strains which had non-significant effect with seeds of chickpea inoculated by Cp11 strain. Least seedling dry weight (0.59gm) was observed from seed of chickpea without inoculation (Table 10).

The study indicated that chickpea verities and *Rhizobium* strains significantly influences seedling dry weight. This indicated that seedling dry weight was influenced by varietal variation and *Rhizobium* inoculation compatibility. *Dimtu* variety was evaluated more than other varieties and inoculating of chickpea variety by Cp17 and Cp41 was increased weight of the chickpea. Gharineh and Moshatati (2012) reported that more seedling weight of the heavy seeds might be attributed to large food reserves of the seeds.

Table 10.Variatin of chickpea Varieties and *Rhizobium* strains for seedling dry weight

Variety	SDW(gm)
Eshete	0.60b
Dimtu	0.64a
Teketay	0.61b
Farmers cultivar	0.57c
LSD (0.05)	0.018
R Strain	
Cp 11	0.60ab
CP 17	0.61a
Cp 41	0.61a
Control	0.59b
LSD (0.05)	0.018
CV (%)	4.29

Mean values within column designated with similar letters are not significant each other at $p<0.05$, LSD (5%) = Least significant difference at $p\leq0.05$ and CV (%) = percentage of coefficient of variation.

4.2.1.4. Interaction effect of variety and *Rhizobium* strain on seedling shoot and root length

Shoot and Root length was significantly influenced by main effect of variety and interaction of variety with *Rhizobium* strain. Root length was significantly influenced by main effect of *Rhizobium* strain. Highest shoot length (11.61cm) was registered from *Eshete* variety inoculated by Cp17 strain which had statistical parity with shoot length from Farmers cultivar inoculated by Cp11, Cp 17, Cp41 strains and no inoculation. And it also had non-significance difference with *Eshete* inoculated by Cp41 and no inoculation. *Teketay* variety inoculated by Cp17 had least shoot length (7.08cm) which had no statistical difference with Shoot Length from *Dimtu* variety inoculated by Cp11 and without inoculation. Highest root length (23.27cm) was registered from Farmers cultivar inoculated by Cp11 strain which had statistical parity with root length from *Teketay* and farmers cultivar inoculated by Cp17, Cp41 strains and no inoculation. And it also had non-significance difference with *Teketay* vareity inoculated by Cp11. *Dinmtu* variety without inoculaation had least root length

(15.22cm) (Table 11). Highest seedling length (34cm) was registered from Farmers cultivar inoculated by Cp11 strain which had statistical parity with seedling length from Farmers cultivar inoculated by Cp17 and *Teketay* variety inoculated by Cp11 while least (22.25cm) seedling length was registered for *Dimtu* variety without inoculation (Table 11).

The results showed significant effects of inoculation of *Rhizobium* strains and varieties on shoot and root length of chickpea (Appendix Table 4). *Eshete* variety was more sensitive to the Cp17, Cp41 *Rhizobium* strains and no inoculation and increased the shoot of plants also Farmers cultivar variety increased shoot length when inoculated with CP11, 41 *Rhizobium* strains and no inoculation. *Teketay* variety was most responsive to three *Rhizobium* strains and no inoculation and increased the root of plants and also Farmers cultivar was more responsive to CP11, 17 *Rhizobium* strains and no inoculation and increased the root of plants. Gharineh and Moshatati, (2012) reported that more seedling length might be attributed to large food reserves of the seeds.

Table 11.Interaction effects of variety and *Rhizobium* strain on shoot, root length and seedling length of chickpea Varieties.

Variety	R strain	SL(cm)	RL(c	Seedling length
Eshete	Cp11	10.09bc	18.92de	29.25ef
	Cp17	11.61a	19.80cde	31.5bcd
	Cp41	11.26a	19.44de	30.5cdef
	Control	10.72ab	18.46e	29.5ef
Dimtu	Cp11	7.16f	19.25de	26.5h
	Cp17	7.60ef	19.59de	27.25gh
	Cp41	8.56de	20.50bcde	29fg
	Control	7.13f	15.22f	22.25i
	Cp11	9.59cd	22.82a	32.5ab
	Cp17	7.08f	21.95abc	29fg
	Cp41	8.15ef	21.99abc	30.25cdef

Teketay	Control	8.48e	21.12abcd	29.75def
	Cp11	10.74ab	23.17a	34a
	Cp17	9.85bc	22.74ab	32.7ab
Farmers cultivar	Cp41	10.93ab	20.04cde	31bcde
	Control	10.88ab	21.04abcd	32bc
LSD (0.05)		1.08	2.25	1.99
CV (%)		8.17	7.77	4.7

Mean values within column designated with similar letters are not significant each other at p<0.05, LSD (5%) = Least significant difference at p≤0.05 and CV (%) = percentage of coefficient of variation.

4.2.1.5. Effect of variety and *Rhizobium* strain on seedling vigour index

Analysis of variance showed that vigor index one and vigor index two was significantly influenced by chickpea variety and *Rhizobium* strain. Vigor index one also influenced by interaction of variety with *Rhizobium* strain but vigor index two not significantly influenced by interaction of variety with *Rhizobium* strain (Appendix Table 2). *Dimtu* variety had higher vigor index two (62.85) while least vigor index two (56.26) was registered for Farmers cultivar. Highest vigor index two (60.73) was Cp17 strain which was statically non-significant with both Cp11 and Cp41 strain while least vigor index two (57.93) was estimated for non-inoculation (Table 12).

The significant difference observed among chickpea varieties for seedling vigor index of the crop indicated the varieties and *Rhizobium* strain had inherent variations for the studied seed quality parameters. Samson (2013) reported that seed vigour index was affected by varietal variation in two varieties of *teff*. The *Rhizobium* legume symbiotic relationship is highly specific and most legume plants form association with only a limited number of the *Rhizobium* strain (Subba Rao *et al.,* 1999)

Table .12. Main effects of variety and *Rhizobium* strain on vigor index two of chickpea variety

Variety	VI2
Eshete	59.08b
Dimtu	62.85a
Teketay	59.98b
Farmers cultivar	56.26c
LSD (0.05)	1.79

R Strains	
Cp 11	59.62ab
CP 17	60.73a
Cp 41	59.9a
Control	57.93b
LSD (0.05)	1.79
CV (%)	4.23

Mean values within column designated with similar letters are not significant each other at $p<0.05$, LSD (5%) = Least significant difference at $p \leq 0.05$ and CV (%) = percentage of coefficient of variation.

4.2.1.6. Interaction effect of variety and *Rhizobium* strain on vigour index one

The vigor index one of chickpea was significantly influenced by the main factors variety and inoculation of *Rhizobium* strains as well as the two factors interaction (Appendix Table 2). Highest vigor index one (3340) was registered for Farmers cultivar inoculated by Cp11 strain which had statistical parity with vigor index one for Farmers cultivar inoculated by Cp17strains and no inoculation as well as *Teketay* variety inoculated by Cp11. *Dimtu* variety without inoculation (2204) had least vigor index one (Table 13). The study indicated that significant effects of inoculation of *Rhizobium* strains and varieties on vigor index one of chickpea (Appendix Table 3). *Eshete* variety was more responsive to the two *Rhizobium* strains (Cp17 and Cp41) and increased vigor index one of the crop. Vigor index one was increased when Farmers cultivar was inoculated with Cp11, Cp41 strains and non-inoculation. Vanangamudi *et al*., (2006) reported that the vigor index of any seed is the sum of those properties of seed which determine the potential level of activity which help to withstand under a wide range of field condition. Zewdie (2004) indicated that seedlings with well-developed shoot and root system would withstand adverse conditions and provide better seedling emergence and seedling establishment in the field and thus, seedlings with higher index are expected to show rapid germination and emergence that eventually leads to escape adverse field conditions.

Table 13. Interaction effects of Variety and *Rhizobium* strain on vigor index one of chickpea varieties

Variety	R strain	Vigor index one
	Cp11	2835.8fg
	Cp17	3078.3bcde

	Cp41	2994.5cdef
	Control	2867efg
Dimtu	Cp11	2568.8h
	Cp17	2666.3gh
	Cp41	2827fg
	Control	2204i
Teketay	Cp11	3201abc
	Cp17	2846.3fg
	Cp41	2946def
	Control	2880.3ef
Farmers cultivar	Cp11	3340a
	Cp17	3219.8ab
	Cp41	3035.3bcdef
	Control	3152.5abcd
LSD (0.05)		211.7
CV (%)		5.09

Mean values within column designated with similar letters are not significant each other at $p<0.05$, LSD (5%) = Least significant difference at $p \leq 0.05$ and CV (%) = percentage of coefficient of variation.

5. SUMMARY AND CONCLUSION

Chickpea is among the most significant food legumes that are used as a source of food and feed, cash income for farmers, and in sustaining soil fertility. However, its productivity has been declined probably due to P-deficiency and the usual native soil Rhizobial populations are insufficient/ ineffective in N2-fixation. On the other hand, sustainability of agriculture has become a major issue of global concern as the intensive use of chemical inputs has an adverse impact on the environment. Use of bio fertilizers such as *Rhizobium* can reduce the need for chemical fertilizers and decrease adverse environmental effects. To supply adequate rhizobial population in the rhizosphere, seed inoculation with an effective and importunate *Rhizobial* strain is essential for chickpea in soils having no/feeble bacterial existence. In this regard, *Rhizobium* inoculation of the seed may substitute costly N-fertilizers and is a useful way of achieving sustainable production. Moreover, chickpea has high demand for P for effective N2-fixation, growth, and yield. Therefore, this study was conducted to assess the effect of different *Rhizobium* strains on seed quality of four chickpea varieties and its subsequent effect on growth, yield and its components and seed quality of chickpea.

The effect of chickpea seeds as influenced by two main factors (variety and *Rhizobium* strain) and all possible two factors' interactions on phenology, growth, yield components and seed yield were evaluated at field condition. The results of field experiment indicated that all phenology, growth, seed yield and its components were significantly influenced by varietal difference. Plant height, biomass, pod per plant, seed per pod, seed yield, seed production efficiency, grain yield per day, *Rhizobium* strains sensitivity index and yield index were significantly influenced by main effect of *Rhizobium* strains. Besides, the interactions of variety x *Rhizobium* strain had significant effect on plant height and pod per plant. The growing of *Teketay* and *Dimtu* varieties higher seed yield of 2777.78 and 2552.08kg ha^{-1}, respectively, while growing of farmer cultivar produced significantly lower seed yield (2013.89kg ha^{-1}). Inoculation of chickpea varieties by Cp17 strains increases seed yields by 538.19 kg ha^{-1} over control. The chickpea seeds quality produced from seeds influenced by two main factors (variety and *Rhizobium* strain) and all possible two factors' interactions were evaluated.

The results of seed quality test after harvest showed that the varieties had inherent significant difference for moisture content, normal seedling, speed of germination, seedlings shoot and root length, seedling dry weight, seedlings vigor index one and two. *Rhizobium* strains had significant effect on seedlings root length; seedling weight and vigor index two. The variety and *Rhizobium* strain interacted to influence significantly seeds moisture content, normal seedling, shoot length, root length and vigor index one.

In general, results from the two experiments showed that after growing the chickpea seeds of different quality influenced by variety, *Rhizobium* strain and interaction of the two factors had significant effect on seed yield, yield related traits and seed quality parameters. In most cases, the seeds of improved varieties inoculated by Cp17 strain produced high yield and quality seeds. Thus, it is suggested to consider varieties and *Rhizobium* strain to produce high yield and quality seeds to improve the productivity of chickpea. However, to come to a conclusive recommendation, additional experiments should be conducted at different locations using more number of varieties and *Rhizobium* strain.

6. REFERENCES

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APPENDIX

Appendix Table 1. Mean squares from analysis of variance for phenology, growth, yield components and seed yield of chickpea variety at Ambo in 2022/2023 cropping season.

Parameter	Replication (2)	Variety (3)	R strain (3)	Variety * R.strain	Error (32)	CV (%)
Days to50% emergence	2.77	4.68**	0.90 ^{ns}	0.31^{ns}	0.57	4.62
Day to 50% flowering	0.33	292.50**	0.72 ^{ns}	1.92^{ns}	1.67	2.93
Days to maturity	0.145	96.72**	2.72 ^{ns}	5.59^{ns}	2.33	1.74
Plant height (cm)	38.27	358.73**	136.98**	16.22*	2.19	4.99
Plant biomass (kg)	0.22	1.61**	0.79 **	0.06^{ns}	0.27	12.46
Hundred seed weight (g)	29.31	996.47**	36.30 ^{ns}	6.32^{ns}	2.89	11.99
Number of pods per plant	62.27	416.46*	2033.02**	166.37*	8.33	9.22
Number of seed per pod	0.002	0.08*	0.48**	0.00 ^{ns}	0.11	7.55
Seed yield (g)	14467.82	1254757.07**	582020.39*	83691.75^{ns}	348.6	14.36
Seed production efficiency	3712.57	3357710.25**	921074.17*	139514.64^{ns}	464.26	15.76
Grain yield per day	1.43	106.51**	44.60*	6.42 ^{ns}	3.28	14.98

Rhizobium strains sensitivity index	170931.71	813294.40**	19178416.17**	121534.47^{ns}	230.7	12.22
Yield index	0.54	0.28*	0.37*	0.12^{ns}	0.29	11.87

Ns, * and **, nonsignificant, significant, significant at p>0.05 and p<0.001 probability level, respectively, R strain= *Rhizobium* strain, CV (%) = percentage of coefficient of variation. Number in parenthesis represents degree of freedom for the respective source of variation.

Appendix Table 2. Mean squares from analysis of variance for physiological seed quality parameters of chickpea varieties as influenced by *Rhizobium* strain at Ambo.

Parameter	Variety (3)	R strain (3)	Variety * R	Error	CV (%)
Percentage of pure seed	0.01^{ns}	0.01 ^{ns}	0.01 ^{ns}	0.12	0.12
Percentage of inert matter	0.01 ^{ns}	0.01 ^{ns}	0.01 ^{ns}	0.12	12.3
Proportion of normal seedlings (%)	1.76*	1.14ns	0.94*	0.66	0.68
Proportion of abnormal seedlings (%)	0.51ns	0.43ns	0.40ns	0.55	8.5
Proportion of dead seeds (%)	0.54ns	0.29ns	0.36ns	0.55	7.7
Speed of germination (No/days)	4.10*	1.39ns	0.84ns	0.84	4.9
Seedlings shoot length (cm)	43.18**	1.29ns	2.50*	0.78	8.34
Seedlings root length (cm)	47.59**	15.32**	6.16*	1.29	6.37
Seedling length (cm)	106.8**	15.29**	12.41**	1.4	4.7
Seedlings dry weight (g)	0.013**	0.002*	0.0004^{ns}	0.02	4.29
Seedlings vigour index one	1061562**	144608*	117769*	148	5.09
Seedlings vigour index two	117.84**	22.23*	4.06^{ns}	2.52	4.23

Ns, *and **, nonsignificant, significant, significant at $p>0.05$ and $p<0.001$ probability level, respectively, R strain= *Rhizobium* strain, CV (%) = percentage of coefficient of variation. Number in parenthesis represents degree of freedom for the respective source of variation.

Appendix Table 3. Effects of Variety and *Rhizobium* strain on plant height and number of pod per plant of chickpea at Ambo during 2022/2023 cropping season

Variety	Plant height (cm)	Number of pods per plant
Eshete	49.43^{a}	$85.25b^{c}$
Dimtu	44.16^{b}	91.50^{ab}
Teketey	45.44^{b}	94.58^{a}
Farmers cultivar	36.37^{c}	81.58^{c}
LSD (0.05)	1.82	6.95
CV (%)	4.99	9.22
Rhizobium strain		
Cp 11	44.53^{b}	84.75^{b}
CP 17	47.20^{a}	107.00^{a}
Cp 41	44.53^{b}	84.25^{b}
Control	39.15^{c}	76.91^{c}
LSD (0.05)	1.82	6.95
CV (%)	4.99	9.45

Mean values within column of each parameter and treatment effect designated with similar letters are not significant each other at $p<0.05$. LSD (5%) = Least significant difference at $p\leq 0.05$ and CV (%) = percentage of coefficient of variation

Appendix Table 4. Effect of variety and *Rhizobium* strain on Normal Seedling, Shoot Length Root length and Vigor Index one of chickpea at Ambo during 2022/2023 Cropping Season

Variety	NS%	SL(cm)	RL(cm)	VI1
Eshete	97.87b	10.922a	19.15b	1069.03a
Dimtu	97.75b	7.61c	18.64b	744.33c
Teketey	97.93b	8.32b	21.97a	815.65b
Farmers cultivar	98.50a	10.60a	21.75a	1044.33a
LSD (0.05)	0.475	0.556	0.9247	55.25
Rhizobium strain				
Cp 11	98.06ab	9.39ab	21.04a	922.14ab
CP 17	98.18a	9.03b	21.02a	887.51b
Cp 41	97.62b	9.72a	20.49a	949.58a
Control	98.18a	9.30ab	18.96b	914.11ab
LSD (0.05)	0.47	0.55	0.92	55.25
CV (%)	0.68	8.34	6.37	8.45

Mean values within column of each parameter and treatment effect designated with similar letters are not significant each other at $p<0.05$. LSD (5%) = Least significant difference at $p\leq 0.05$ and CV (%) = percentage of coefficient of variation