

Potential of Rhizobacteria from the Rhizosphere of Coffee Plants to Increase the Viability of Rice Plant Seeds

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Abstract: Rhizobacteria is a type of biological fertilizer developed to increase plant production. The research aims to explore bacteria in the rhizosphere area of coffee plants and test their potential to increase the viability of rice seeds. The research was structured using a completely randomized design with treatment of 21 bacterial isolates and 1 control (without rhizobacteria) which was repeated 3 times so that there were 66 experimental units. The variables observed were vigor index, germination capacity, maximum growth potential, dry weight of sprouts, relative growth speed, uniformity of growth and 50% seed germination time (T-50). Observational data were analyzed using analysis of variance, and data that showed a real effect was followed by Duncan's Multiple Range Test at a confidence level of 95%. The results showed that rhizobacteria isolated from the roots of coffee plants could increase rice viability. The best treatment was obtained on isolate A8 both on the variables maximum growth potential, germination capacity, vigor index, uniformity of growth and dry weight of sprouts with an increase in each variable respectively by 21.63%, 23.61%, 27.03%, 25.17% and 82.05% compared to control.

Keywords: coffee, rhizobacteria, potential, viability.

1. Introduction

Food crop production is currently being encouraged in various countries to meet the consumption needs of their population. The development of food crops is currently still very intensive using chemical fertilizers to produce high production. Excessive application of chemical fertilizers can cause soil damage, including decreasing organic matter levels, soil compaction, and water pollution (Abebe et al., 2022). The concept of sustainable agriculture must prioritize cultivation techniques that are environmentally friendly and effective in increasing crop production both now and in the future.

One approach to developing sustainable agriculture is to utilize bacteria that are explored in plant root areas or what are known as rhizobacteria (Nehra and Choudhary, 2015). Rhizobacteria can be used as biofertilizer to increase crop production and streamline the use of chemical fertilizers at the farmer level.

Mahmud et al. (2021) revealed that the application of various microbes isolated from plant roots was able to increase crop yields and maintain environmental conditions. Rhizobacteria can aggressively colonize root areas by producing various chemical compounds that act as growth promoters and also increase plant resistance to disease attacks (Etesami and Maheshwari, 2018; Chittora et al., 2020; Mhatre et al., 2019; Djayaet al., 2019). Rhizobacteria are able to stimulate plant growth through the mechanism of increasing the IAA hormone, phosphate solubilization, nitrogen fixation (Agustiyani et al., 2021; Gianfreda, 2015; Jha and Saraf, 2015; Saharan and Nehra, 2011) and can indirectly increase plant resistance to stress, drought (Ahluwalia et al., 2021) and disease attacks (Syamsuddin et al., 2021; Goswami et al., 2020; Shobha et al., 2021; Wang et al., 2021). These bacteria are able to dissolve insoluble phosphate in the soil, mineralize organic matter to produce dissolved phosphate ions (Pi, HPO₄²⁻, H₂PO₄⁻), and produce the hormone IAA. The bacteria that have been isolated and are able to increase plant growth are Azotobacter, Pseudomonas, Bacillus, Burkholderia,

Rhizococcus, Arthrobacter, Xanthomonas, Enterobacter, Klebsiella, Rhizobium, Bradyrhizobium, and Thiobacillus (Nehra and Choudhary, 2015).

In this research, bacteria isolated from the roots of coffee plants that grow naturally in Buton Regency were used. This coffee grows naturally without fertilization, which is thought to contain bacteria associated with plant roots. Bacteria can utilize root exudates produced by plants as a source of energy and nutrients, whereas plants can utilize metabolites produced by bacteria so that they grow well and are resistant to disease attacks. Souza et al. (2012), also revealed that 4 bacterial isolates explored from plants were able to increase rice production. Sutariati et al. (2020), reported that bacteria isolated from the roots of areca nut plants have the ability to increase the viability of rice plants. Based on the description above, each plant has a different diversity of bacteria and potential to influence plant growth and production. Therefore, this research aims to explore bacteria in coffee plants and test their potential to increase the viability of rice seeds.

2. Methods

1) Place and time of research

The research was carried out at the Agrotechnology Laboratory, Faculty of Agriculture, Halu Oleo University, from July to November 2022.

2) Research design

The treatments in this study consisted of 21 bacterial isolates and 1 control (without bacteria) which were arranged using a completely randomized design which was repeated 3 times.

3) Seed Treatment Using Rhizobacteria

Bacteria isolated from the rhizosphere of coffee plants from Buton Regency were grown on TSA media and suspended at a density of 109 CFU mL⁻¹. The rice seeds used were washed using 2% sodium hypochlorite and washed 3 times using sterile water, then dried in a laminar. The dried rice seeds were put into an Erlenmeyer containing 50 ml of bacterial suspension and shaker for 24 hours.

4) Rice Plant Seed Viability Test

Rice seeds treated with rhizobacteria were germinated in plastic trays containing husk charcoal media. The viability of germinated rice seeds is calculated using the following variables:

a. **Germination capacity (DB)**, calculated based on the percentage of normal sprouts (KN) first count (I) at 4 days after planting (DAT) and second count (II) at 7 days after planting (DAT), with the formula:

$$DB = \frac{\sum KNHitunganI + \sum KNHitunganII}{\sum Beni hyangditanam} \times 100\%$$

b. **Relative Growth Speed (KCT-R)**, calculated based on the accumulated daily growth speed using the formula:

$$KCT-R = \frac{KCT}{KCT_{max}}$$

$$KCT = \sum_{t=0}^{tn} \frac{N}{t}$$

$$KCT \text{ maks} = \frac{100}{\sum Hari \text{ hitunganI}}$$

Information:

tn = End time of observation

N = Percentage (%) of normal sprouts at each observation time

t = Observation time

c. **Maximum Growth Potential (PTM)**, calculated based on the percentage of seeds showing symptoms of growth from the first day (3rd day) to the last day (30th day), using the formula:

$$PTM = \frac{\sum Tumbu h}{\sum Beni hyangdiTanam} \times 100\%$$

d. **Emergence of sprouts (T50)**, observed by counting the number of seeds that germinate every day which is calculated using a formula:

$$T_{50} = ti + \frac{(n50\% - ni)}{nj - ni} (tj - ti)$$

Information:

ti = The time between, at or before the seeds germinate 50%

tj = The time between, after the seeds germinate 50%

n50% = Number of germinated seeds (50% of the total germinated seeds).

nj = Number of germinated seeds at time tj.

ni = number of germinated seeds at time ti.

e. **Vigor Index (IV)**, calculated using the following:

$$IV = \frac{\sum KNHitunganI}{\sum Beni\ hyangditanam} \times 100\%$$

f. **Dry Weight of Sprouts**, calculated by weighing the dry weight of rice sprouts that were oven-dried at 600C for 2x24 hours using an analytical balance.

Data analysis. The observational data was analyzed for variance and the data showed a real effect followed by Duncan's Multiple Range Test at the confidence level 95%.

3. Results

a. Maximum Growth Potential

Treatment with rhizobacteria isolates can increase the maximum growth potential of rice seeds (Figure 1). The highest maximum growth potential was obtained in isolate A8 at 98.67% which was not significantly different from other treatments but was significantly different from isolates A18, A19 and control.

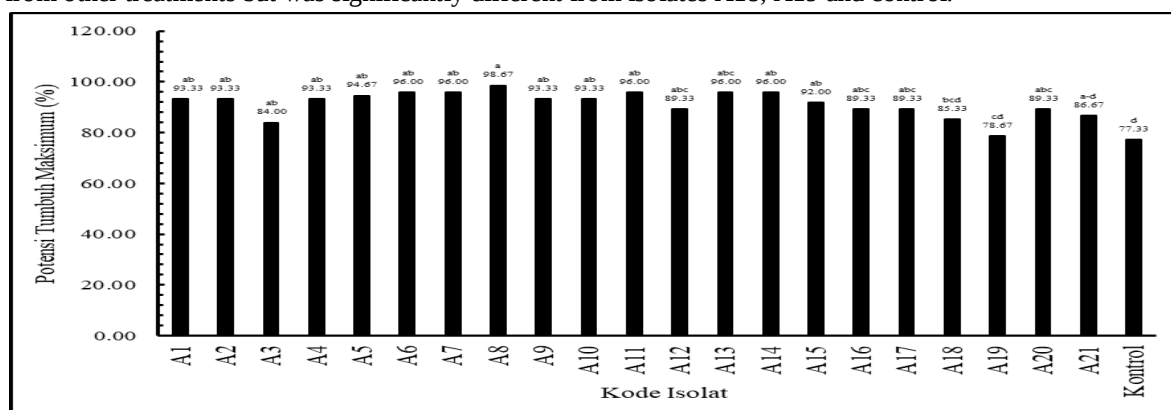


Figure 1. Maximum growth potential of rice seeds treated with rhizobacteria isolates from the rhizosphere of coffee plants.

b. Germination Power

Treatment with rhizobacteria isolates can increase the germination of rice seeds (Figure 2). The highest germination capacity was obtained in isolates A8, A13, and A14 at 96.00% which was not significantly different from other treatments but was significantly different from isolates A19 and the control.

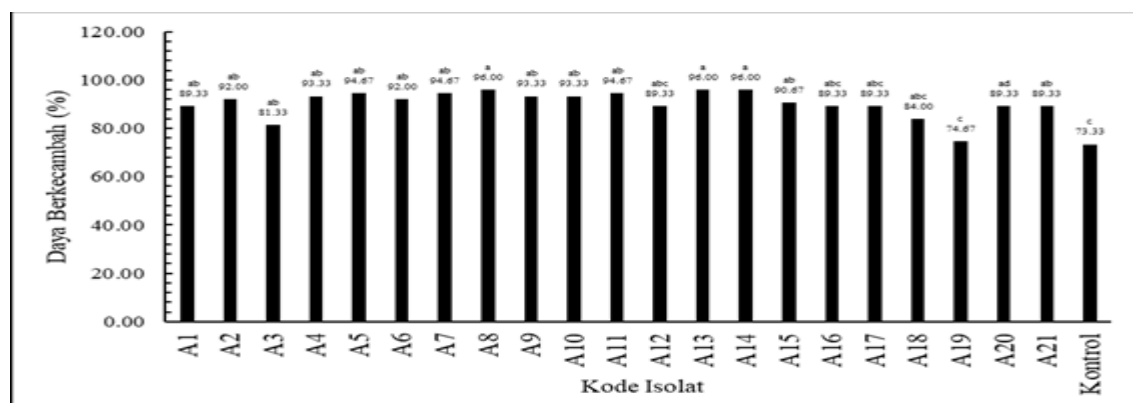


Figure 2. Germination capacity of rice seeds treated with rhizobacteria isolates from the rhizosphere of coffee plants.

c. Vigor Index

Treatment of rhizobacteria isolates can increase the vigor index of rice seeds (Figure 3). The highest vigor index obtained by isolate A8 was 98.67% which was not significantly different from other treatments but was significantly different from isolates A3, A18, A19, A21 and control.

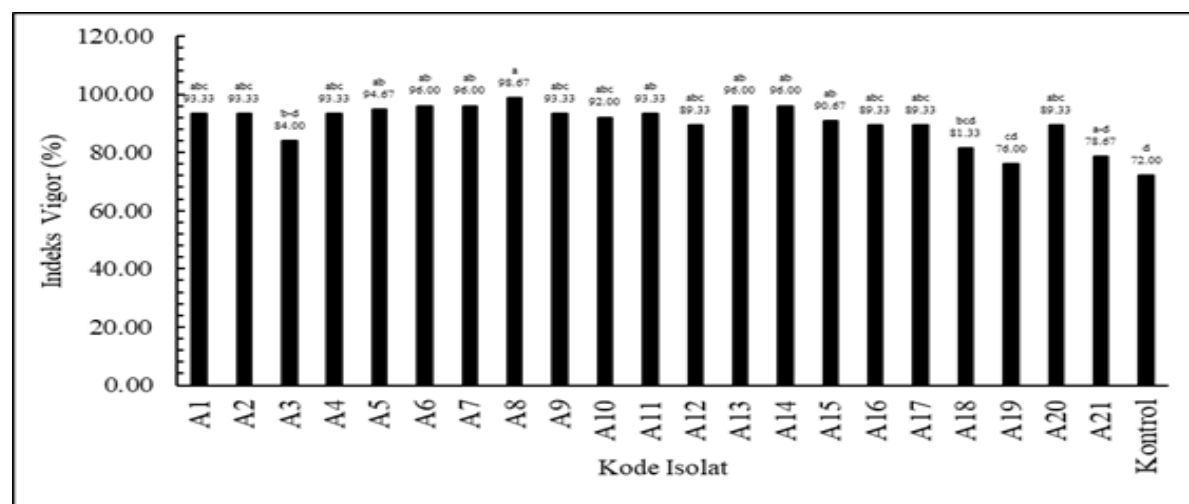


Figure 3. Vigor index of rice seeds treated with rhizobacteria isolates from the rhizosphere of coffee plants.

d. Simplicity Grows

Treatment of rhizobacteria isolates can increase the uniformity of rice seeds (Figure 4). The highest uniformity of growth was obtained for isolate A8 at 98.00% which was not significantly different from other treatments but was significantly different from isolates A3, A18, A19, and the control.

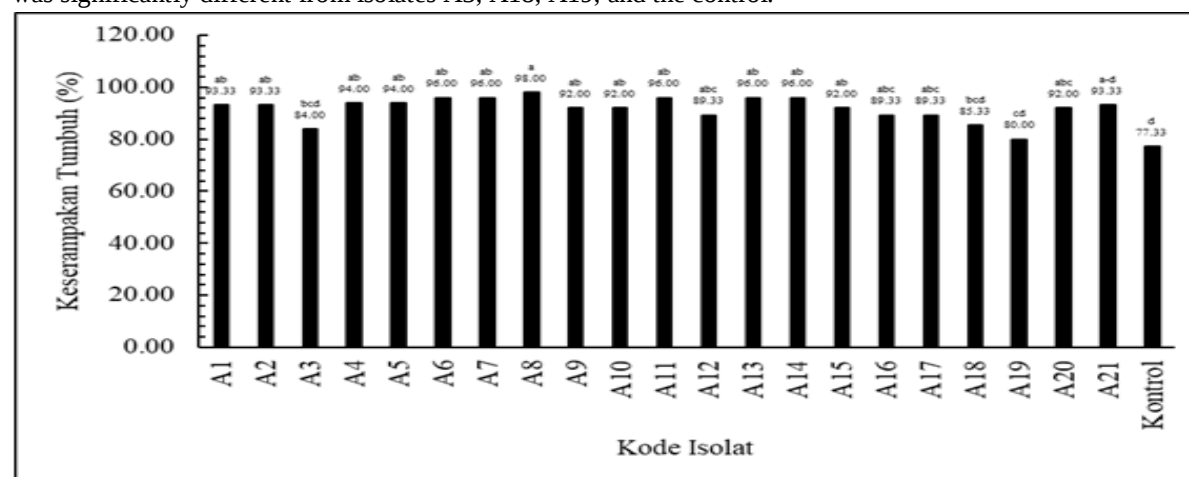


Figure 4. Uniformity of growth of rice seeds treated with rhizobacteria isolates from the rhizosphere of coffee plants.

e. KCT-R

Treatment with rhizobacteria isolates can increase the relative growth rate of rice seeds (Figure 5). The highest relative seed growth speed was obtained in isolate A13 at 94.14% which was not significantly different from other treatments but was significantly different from isolate A19 and the control.

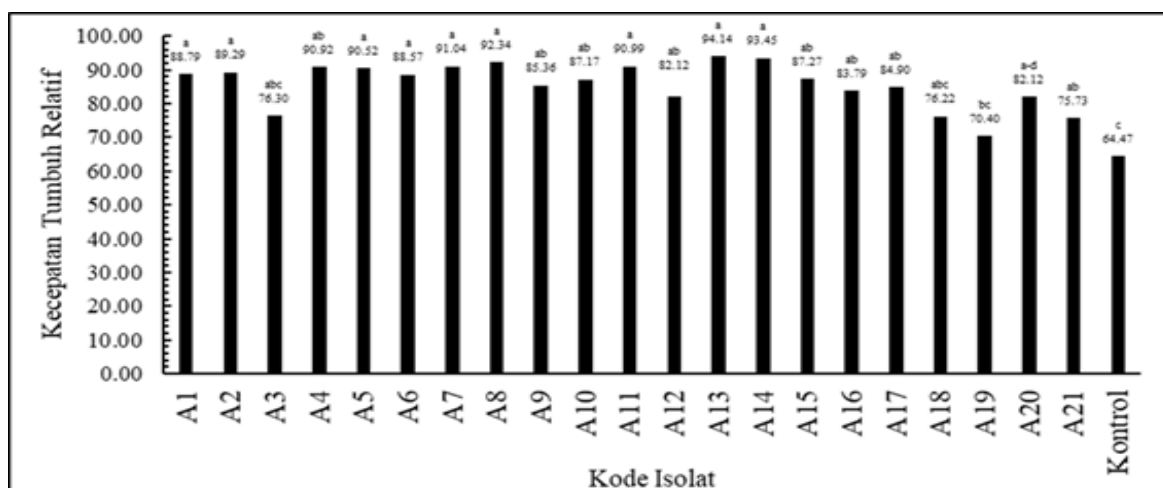


Figure 5. Relative growth speed of rice seeds treated with rhizobacteria isolates from the rhizosphere of coffee plants.

f. T-50

Treatment with rhizobacteria isolates can speed up the germination time of rice seeds by 50% (Figure 6). The fastest 50% germination time was obtained for isolate A8 for 2.83 days, which was not significantly different from other treatments but was significantly different from isolates A10, A19, A21, and the control was the longest germination treatment.

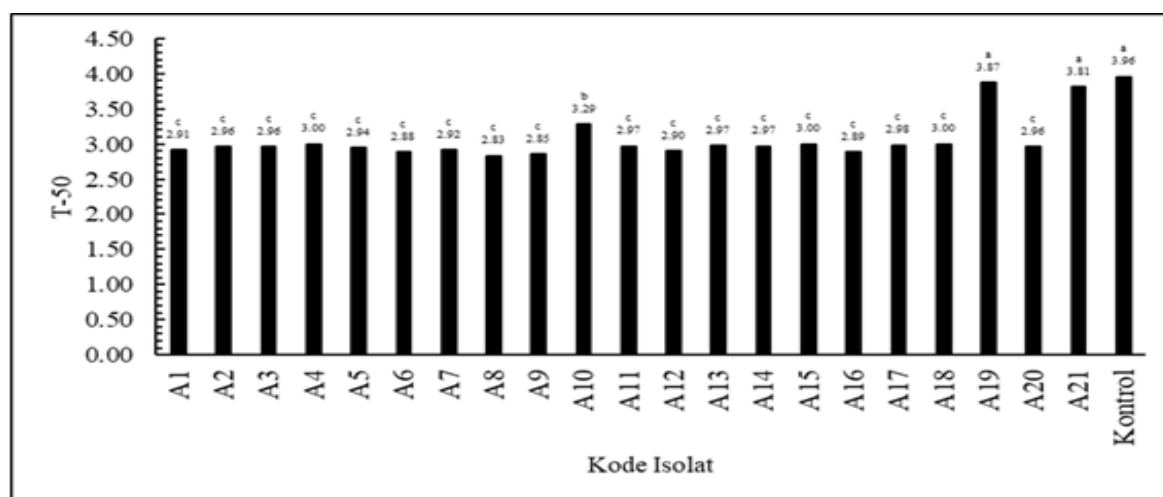


Figure 6. Time to 50% germination (T-50) of rice seeds treated with rhizobacteria isolates from the rhizosphere of coffee plants.

g. Dry Weight of Sprouts

Treatment with rhizobacteria isolates can increase the dry weight of rice seed sprouts (Figure 7). The highest dry weight of sprouts was obtained by isolate A8 at 0.39 g which was not significantly different from other treatments but was significantly different from isolates A2, A5, A10, A12, A15, A15, A16, A18, A19, and control.

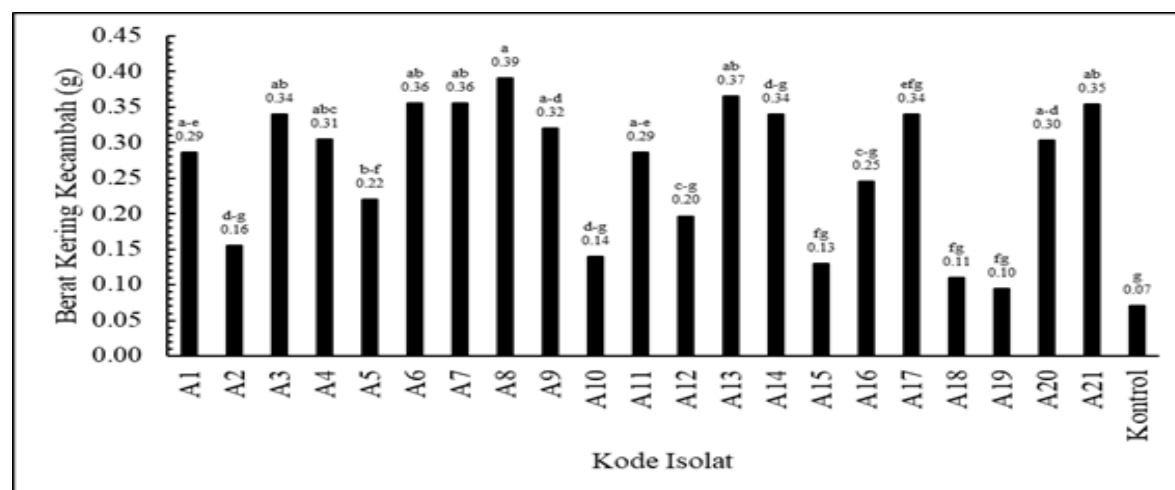


Figure 7. Dry weight of rice seed sprouts treated with rhizobacteria isolates from the rhizosphere of coffee plants.

4. Discussion

Rhizobacteria are bacteria in the root area that are explored to increase the productivity of food crops, horticulture, plantations and forestry (Nehra and Choudhary, 2015). Bacteria in the plant rhizosphere have been widely reported and are effective in increasing plant production. Rhizobacteria can increase plant growth and production through their ability to produce the hormone IAA, dissolve phosphate, fix nitrogen and the ability to produce other chemical compounds (Gupta et al., 2015; Moustaine et al. Mehmood et al., 2018; 2020; Lebrazi et al., 2020; Leontido et al., 2020).

The results of the research showed that in general rhizobacteria isolates could increase the viability of rice seeds in terms of vigor index, germination capacity, maximum growth potential, dry weight of sprouts, uniformity of growth and seed germination time of 50% (T-50) with different percentages for each bacterial isolate. The influence of rhizobacteria on the variables maximum growth potential, germination capacity, vigor index and uniformity of growth was highest in the A8 isolate treatment at 98.67%, 96.00%, 98.67% and 98.00% respectively, which were not significantly different. with other treatments but significantly different from isolates A18, A19 and control treatments. Meanwhile, the highest KCT-R variable was obtained in isolate A13 at 94.14%, which was not significantly different from other treatments but was significantly different from the control treatment. The fastest 50% germination time variable (T-50) was obtained in isolate A8 which was not significantly different from other treatments but was significantly different from isolate A2 and the control.

Increasing seed viability is thought to be influenced by the presence of bacteria that can stimulate seed germination. Pérez-García et al. (2023), revealed that the KBEndo3P1 isolate had the highest germination percentage compared to the control. Rhizobacteria isolates are able to stimulate the growth of meristem tissue in both roots and shoots through the synthesis of the hormone auxin (IAA) which ultimately increases seed germination and supports cell division. Paul & Lade, (2014); Shultana et al., (2020); Ha-Tran et al., (2021); Purwanto et al. (2022), revealed that the application of rhizobacteria from saline soil can increase the vigor of rice seedlings in saline conditions. Rhizobacteria are able to increase plant growth through their ability to produce the hormone auxin, especially Indole Acetic Acid. Apart from that, rhizobacteria are able to facilitate seeds to speed up the imbibition process with their ability to degrade plant seed coats so that plants germinate easily. The hard skin of coffee beans and areca nut can inhibit the plant imbibition process. Rhizobacteria are thought to be able to soften the seed coat and make it easier for water to enter the seed so that it can speed up the seed germination (Guo et al., 2020; Sutariati et al., 2021; Javed et al., 2022; Zaim et al., 2023). Rhizobacteria can produce cellulase enzymes which play a role in degrading plant seed cell walls. Rathod et al. (2021), revealed that bacteria can increase seed germination and seedling growth after germination of lemon plants.

Increasing the dry weight of rice sprouts in treatment A8 is closely related to increasing germination capacity, vigor index and uniformity of growth. It is thought that bacteria can dissolve phosphate bound in the soil (Fatimah et al., 2023; Arfarita et al., 2017). Arfarita et al. (2017) revealed that strain SPP2 showed the highest phosphate solubilization activity compared to other isolates. Atekan et al. (2014), revealed that eight bacterial isolates dissolved phosphate with isolates T-K5 and T-K6 being superior in dissolving P. The element phosphorus plays a role in the transfer of energy, coenzymes, nucleic acids and other metabolic compounds so that it can accelerate the process of forming good plant optic organs. roots, stems and leaves (Alamgir, 2018; Landi et al., 2020; Silva et al., 2023). Mukhtar et al., (2017); Pande et al., (2017); Manzoor et al., (2017); Jiang et al., (2019); Tariq et al., (2020); Tariq et al. (2022), revealed that phosphate solubilizing bacteria applied to plants significantly increased fresh weight, dry weight, plant height, root and shoot length compared to controls. Based on the description of the research results, rhizobacteria isolated on the roots of coffee plants have great potential to be developed to increase plant growth and production.

5. Conclusion

Rhizobacteria isolated from the roots of coffee plants from Buton Regency can increase the viability of rice seeds. The best treatment was obtained for isolate A8 in terms of maximum growth potential, germination, vigor index, uniformity of growth and dry weight of sprouts with an increase in each variable respectively by 21.63%, 23.61%, 27.03%, 25.17% and 82.05% compared to control.

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