JUATIKA
JURNAL AGRONOMI TANAMAN TROPIKA
VOL. 8 No. 3 September 2026

DOI :https://doi.org/10.36378/juatika.v8i3. 6169
eissn 2656-1727
pissn 2684-785X
pages : 894 – 899

RESEARCH ARTICLE Open Access

Effectiveness of Arabica Coffee Seed Germination (*Coffea arabica* L.) with Various Combinations of Soaking Length and PGPR Concentration

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Abstract

Arabica coffee (*Coffea arabica* L.) is a significant plantation commodity in Indonesia, ranking after oil palm, rubber, and cocoa. Despite Indonesia's status as one of the world's leading coffee producers, national coffee productivity remains relatively low compared to several other prominent coffee-producing countries. One promising approach to enhance seedling performance is biopriming with Plant Growth-Promoting Rhizobacteria (PGPR). This study aims to identify the optimal soaking duration and PGPR concentration for Arabica coffee seed germination. The research was conducted from September to December 2025, with the experimental trials carried out at the Land of the Surabaya City Food and Agriculture Security Agency (DKPP) and at the Biotechnology Laboratory of the Veteran National Development University of East Java. The experimental design employed was a two-factorial Completely Randomized Design (CRD), comprising soaking time (12, 24, and 36 hours) and PGPR concentration (0, 15, 30, and 45 ml/L). We assessed seedling vigor index, root length, fresh seedling weight, and dry seedling weight. We analyzed the data using analysis of variance (ANOVA) at a 5% significance level. The findings revealed significant interactions between soaking time and PGPR concentration affecting root length, seedling wet weight, and dry weight, indicating the importance of optimizing these factors to improve seedling growth performance.

Keywords: Biopriming, Biostimulant, Imbibition, Rhizobacteria, Seed Viability

1. Introduction

Arabica coffee (*Coffea arabica* L.) is a vital plantation commodity in Indonesia. Although Indonesia is one of the world's largest coffee producers, national coffee productivity remains lower than that of several other coffee-producing countries (Torok et al., 2018). One of the primary factors contributing to this issue is the use of low-quality seeds. These seeds are characterized by slow germination, uneven growth, abnormal physical traits, and low vigor, which ultimately reduce both yield and quality of the coffee harvest. Consequently, this diminishes the international competitiveness of Indonesian coffee. To address these challenges, sustainable and environmentally friendly nursery techniques are needed to improve seed quality. One promising approach is the application of pre-germination treatments utilizing biological agents, such as Plant Growth-Promoting Rhizobacteria (PGPR), through biopriming.

Plant Growth-Promoting Rhizobacteria (PGPR) are bacteria that reside in the root zone of plants and play a crucial role in enhancing plant growth. They do so through several mechanisms, including the production of phytohormones, increasing nutrient availability, and protecting plants from pathogens (Kasifah et al., 2022). PGPR promote plant growth through three main functions. First, as biostimulants, they produce and regulate concentrations of plant growth regulators (phytohormones) such as indole-3-acetic acid (IAA), gibberellins, cytokinins, and ethylene around the roots, stimulating plant growth. Second, as biofertilizers, they facilitate nutrient acquisition by fixing atmospheric nitrogen (N_2) and dissolving phosphorus (P) bound in the soil. Third, as biological control agents (bioprotectants), PGPR produce various antimicrobial compounds—including siderophores, β-1,3-

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glucanase enzymes, chitinases, antibiotics, and cyanide—that inhibit the growth of pathogenic microorganisms (Irawan et al., 2022).

Biopriming coffee seeds can be done by soaking them in PGPR before germination. Some PGPR bacteria frequently used in biopriming include *Pseudomonas* sp., *Bacillus* sp., *Azospirillum* sp., *Azotobacter* sp., and *Rhizobium* sp. These bacteria have been extensively studied and proven effective in increasing seed vigor and quality during germination. For coffee seeds, soaking in PGPR can aid imbibition, accelerate germination, and increase seed vigor (Muawwana *et al*., 2022).

The seed germination process begins with the seed absorbing water (imbibition), which triggers the activation of various enzymes and initiates germination. Germination in coffee seeds is often hampered by the presence of a hard, *impermeable, horny layer*, which impedes the diffusion of water and oxygen. Similarly, seed germination is relatively difficult because of the seeds' hard shells and dormancy. One factor that can inhibit seed germination is the presence of a hard seed coat, which hinders the Water absorption and gas exchange necessary for germination (Siregar *et al*., 2023).

Soaking Arabica coffee seeds in a PGPR suspension not only aids the imbibition process but also increases seed vigor, one of the effects of which is the presence of *Indole Acetic Acid* (IAA), a growth phytohormone commonly produced by PGPR (Faiz & Sulistyono, 2019). Soaking with a PGPR suspension not only stimulates seed germination but also improves seed vigor and seedling quality for subsequent growth.

Two factors influence the success of biopriming on Arabica coffee seeds: soaking time and PGPR suspension concentration. Soaking time that is too short causes water absorption, or the imbibition process, to be hampered, and the PGPR effect is less than optimal, while soaking too long can inhibit seed respiration and can even result in seed death. In addition, PGPR concentration that is too low can cause the number of bacteria to be insufficient to provide an optimal stimulation effect on coffee seeds, resulting in suboptimal performance. At the same time, concentrations that are too high can cause toxicity, reduce germination power, disrupt microbial balance, and trigger excessive colonization that reduces seed germination (Marom *et al*., 2017). Therefore, the purpose of this study was to determine the interaction in each treatment between the soaking time and the right PGPR concentration in order to obtain the right and optimal results for Arabica coffee seed germination.

2. Material and Methods

2.1. Time and Place of Research

This research was conducted in September - December 2025. The research was conducted of the Surabaya City Food Security and Agriculture Service (DKPP), located on Jl. Jambangan I, Jambangan District, Surabaya City, East Java Province, at coordinates -7.320788, 112.715389, and in the Biotechnology Laboratory of the Veteran National Development University of East Java at coordinates -7.333997,112.791188. The research land was located at an altitude of 3-6 meters above sea level, with a temperature of 33-34 ^{0}C and a relative humidity of 68-75%.

2.2. Tools and materials

The tools used in this study consisted of a germination tank, seed soaking container, ruler, spray bottle or hand sprayer, label paper, label board, analytical balance, measuring cup, petri dish, Bunsen burner, loop needle, *Erlenmeyer flask, rotary shaker, autoclave, spectrophotometer*, and stationery.

The materials used in this study were Arabica coffee seeds of the Komasti variety, a PGPR isolate of Pseudomonas fluo*rescens* ATCC 13525, *nutrient broth* (NB), distilled water, sand, and soil growing media.

2.3. Research methods

This research used a two-factorial Completely Randomized Design (CRD). The first factor had 3 treatment levels, namely soaking time (12, 24, 36 hours), and the second factor had 4 treatment levels, namely PGPR concentration (0, 15, 30, 45 ml/L). The combination of these two factors resulted in 12 treatment combinations, each repeated three times, resulting in 36 experimental units. In each experimental unit, 50 seeds were germinated, requiring a total of 1,800 Arabica coffee seeds. The parameters observed in this study were the vigor index (%), seedling root length (cm), seedling wet weight (g), and seedling dry weight (g).

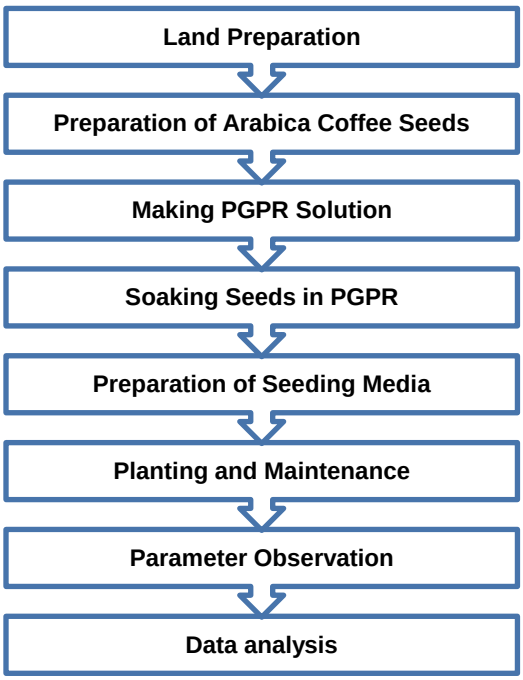


Figure 1 Research Implementation Flowchart

2.4. Data analysis

The observation data were analyzed using Analysis *of Variance* (ANOVA) through the F-test at a significance level of 5% to determine the effect of the treatment on each observed parameter. All data processing and analysis were carried out using the Minitab application version 20.4. If the results of the analysis of variance indicated a significant effect, the analysis continued with the Honestly Significant Difference (HSD) test at a significance level of 5% to determine differences between treatments.

3. Results and Discussion

3.1. Vigor Index

The results of the analysis of the average vigor index data showed that the combination of soaking time and PGPR concentration treatments did not significantly affect the seed vigor index. The single factor of PGPR concentration treatment significantly affected the vigor index. In contrast, the single factor of soaking time treatment did not significantly affect the vigor index of Arabica coffee seeds (Table 1).

The PGPR concentration treatment gave a better average value of the Arabica coffee seed vigor index in the PGPR concentration treatment of 15 ml/L, namely 13.84%, and was not significantly different in the PGPR concentration treatments of 0 ml/L and 45 ml/L, but was significantly different from the PGPR concentration treatment of 30 ml/L, namely 11.57%. Meanwhile, the single factor of soaking time on the Arabica coffee seed vigor index did not differ significantly in the soaking time treatments of 12 hours, 24 hours, and 36 hours.

The vigor index reflects the physiological quality of seeds and their potential to produce strong seedlings. Seed vigor relates to germination strength, the ability of seeds to produce strong roots, and resistance to attack by organisms (Handayani et al., 2024). The effectiveness of PGPR in increasing seed vigor is closely related to its ability to produce phytohormones and can influence metabolism during early germination. The significant effect of PGPR concentration treatment on the seed vigor index tends to indicate a good response to PGPR treatment because it has a more effective physiological system in utilizing bioactive compounds produced by rhizobacteria (Fadila *et al* ., 2026)**.** Differences in response at each concentration indicate that PGPR effectiveness depends on the concentration used.

Table 1 Average Seed Vigor Index for Soaking Length and PGPR Concentration Treatments on Arabica Coffee Seed Germination

Treatment	**Vigor Index (%)**
Soaking Time (hours)	
12	12.67
24	12.27
36	12.29
BNJ 5%	Mr.
PGPR concentration (ml/L)	**Vigor Index**
0	12.35 ± 1.42 ab
15	13.84 ± 1.75 b
30	11.57 ± 1.91 a
45	11.89 ± ab
BNJ 5%	2.12

Description: Numbers followed by the same letter indicate no significant difference in the 5% BNJ test; tn = not significantly different.

3.2. Length of Sprout Roots

The results of the analysis of the average length of the sprout roots showed that the interaction of the soaking time and PGPR concentration treatments on the germination of Arabica coffee seeds significantly affected the length of the sprout roots. The analysis results proved that the highest sprout root length was obtained in the combination of a soaking time of 36 hours + a PGPR concentration of 15 ml/L with a value of 4.84 cm, significantly different from the combination of a soaking time of 12 hours + a PGPR concentration of 45 ml/L with a value of 3.72 cm (Table 2).

There is a combination of treatment interactions that significantly affect the length of the sprout roots, presumably because the long soaking treatment of the seeds provides sufficient time for the water absorption process (imbibition) so that the metabolic process takes place properly. Treatment at a PGPR concentration of 15 ml / L is also able to provide an optimal bacterial population so that it can colonize the root surface without causing competition between microbes, causing root growth to increase due to an increase in the process of mineral nutrient absorption, increased phytohormone production, especially IAA, which has a role in stimulating root cell division and elongation and root formation. As a result, the root system can develop so that the plant's ability to absorb water and nutrients also increases (Walida, 2018).

The increase in the length of the sprouted roots is suspected to be due to the administration of PGPR, one of which contains the bacterium *Pseudomonas fluorescens*, which is capable of acting as a biostimulant, synthesizing growth hormones that play a role in the division and elongation of plant root cells. Salicylic acid and phytoalexin are obtained from *Pseudomonas bacteria* by colonizing the roots so that they can induce plant resistance

(Mursiana *et al*., 2021). According to Handayani et al. (2024), the use of rhizobacteria from the Bacillus sp., Pseudomonas sp., and Serratia sp. groups is effective. It can improve the physiological quality of seeds, especially in seed viability and vigor. Similar results were reported in the study conducted by Mudi *et al.* (2018), which showed that the combination of treatments using biopriming rhizobacteria, *Pseudomonas fluorescens* PG01 + *Bacillus* sp CKD061, was proven to provide the best treatment results, namely being able to improve the physiological quality of seeds, as well as providing the best results on seed viability and vigor, and also proven to increase root length, plumule length, sprout weight, and seed vigor index. It is also proven that the results of observations conducted by Kasifah *et al.* (2022) found that PGPR treatment with a concentration of 20 ml/L produced longer root length than 10 and 30 ml/L.

Plants develop through several stages and have a long life cycle. One example is the increase in sprouts, which is then accompanied by growth of roots, stems, and leaves. This is supported by research conducted by Baihaqi et al. (2018) that found that increasing leaf area allows for light reception and optimizes photosynthesis, thus influencing plant biomass formation, as indicated by plant wet and dry weight.

3.3. Fresh Weight of Sprouts

The results of data analysis (Table 3) on the average fresh weight of the sprouts indicate that there is an interaction of soaking time and PGPR concentration on the germination of Arabica coffee seeds that significantly affects the fresh weight of the sprouts. The fresh weight of the sprouts is one of the accumulations of the coffee sprout parts that can increase according to plant growth. The increase in sprout fresh weight is also influenced by the presence of growing plant organs, such as leaf length, number of leaves, root growth, and plant height.

Table 2 Average Root Length of Seedlings in Combinations of Soaking Length and PGPR Concentration Treatments on Arabica Coffee Seed Germination

Soaking Time (hours)	Root Length (cm)			
	PGPR concentration (ml/L)			
	0	15	30	45
12	4.27 ± 0.09 ab	4.33 ± 0.50 ab	4.73 ± 0.31 ab	3.72 ± 0.69 a
24	4.60 ± 0.33 ab	4.40 ± 0.30 ab	4.33 ± 0.27 ab	4.40 ± 0.35 ab
36	4.52 ± 0.23 ab	4.84 ± 0.08 b	4.03 ± 0.28 ab	4.23 ± 0.24 ab
BNJ 5%	1.01			

Description: Numbers followed by the same letter indicate no significant difference in the 5% BNJ test.

Table 3 Average Fresh Weight of Sprouts in Combinations of Soaking Length and PGPR Concentration Treatments on Arabica Coffee Seed Germination

Soaking Time (hours)	Fresh Weight of Sprouts (g)			
	PGPR concentration (ml/L)			
	0	15	30	45
12	0.53 ± 0.05 a	0.61 ± 0.03 ab	0.63 ± 0.09 ab	0.60 ± 0.09 ab
24	0.57 ± 0.09 ab	0.58 ± 0.07 ab	0.69 ± 0.00 ab	0.61 ± 0.03 ab
36	0.68 ± 0.03 ab	0.73 ± 0.06 b	0.59 ± 0.07 ab	0.60 ± 0.07 ab
BNJ 5%	0.186			

Description: Numbers followed by the same letter indicate no significant difference in the 5% BNJ test.

This shows that good root development can increase water absorption so that plant tissue has a higher water content and reflects the plant's ability to form new tissue through the process of optimal water and nutrient absorption that affects the wet weight of the sprouts. The provision of PGPR to Arabica coffee seeds can support plant growth, especially in the wet weight of the sprouts, because PGPR has a nutrient content that increases the process of sprout growth in Arabica coffee seeds. This is in line with the statement of Khanna et al. (2019) that the application of PGPR can improve the process of photosynthesis performance and consequently the process of plant biomass formation takes place better towards increasing the wet weight of the sprouts.

3.4. Dry Weight of Sprouts

The results of the data analysis of the average value of the interaction of the combination of soaking time and PGPR concentration treatments significantly affected the dry weight parameters of Arabica coffee seed sprouts. According to the results of the analysis of variance, it was found that there was a significant effect of the combination of treatments; the largest dry weight of sprouts was in the combination of 36 hours soaking time + 0 ml/L PGPR concentration with a value of 0.22 g, which was significantly different from the combination of 12 hours soaking time + 0 ml/L PGPR concentration, which showed the smallest average dry weight of sprouts with a value of 0.17 g (Table 4).



Figure 2 Visualization of Coffee Sprouts Resulting from Treatment Combinations. L1: 12 Hours; L2: 24 Hours; L3: 36 Hours; K0: Aquades; K1: 15 ml/L; K2: 30 ml/L; K3: 45 ml/L

Table 4 Average Dry Weight of Sprouts in Combinations of Soaking Length and PGPR Concentration Treatments on Arabica Coffee Seed Germination

Soaking Time (hours)	Dry Weight of Sprouts (g)			
	PGPR concentration (ml/L)			
	0	15	30	45
12	0.17 ± 0.01 a	0.19 ± 0.01 ab	0.18 ± 0.01 ab	0.19 ± 0.02 ab
24	0.19 ± 0.01 ab	0.18 ± 0.01 ab	0.20 ± 0.01 ab	0.18 ± 0.01 ab
36	0.22 ± 0.01 b	0.20 ± 0.01 ab	0.18 ± 0.01 ab	0.19 ± 0.01 ab
BNJ 5%	0.046			

Description: Numbers followed by the same letter indicate no significant difference in the 5% BNJ test.

Dry weight describes the amount of biomass formed as a result of the accumulation of organic matter during the plant growth process. According to Saputri (2020), sprout dry weight indicates biomass accumulation from the results of organic compounds formed during growth from inorganic compounds, water, and carbon dioxide, thus contributing to the plant's dry weight. A well-developed root system allows optimal water and nutrient absorption, thereby increasing photosynthesis. The results of photosynthesis are then converted into carbohydrates, proteins, and other organic compounds, which are then stored in plant tissue, thereby increasing the dry weight of the sprouts and roots. Dry weight reflects the result of plant physiological activity during the early growth phase of the plant. This is also supported by Ferraro et al. (2023), who stated that inoculation of growth-promoting bacteria in coffee seedlings can improve seedling quality through root development and nutrient utilization efficiency, which ultimately leads to increased accumulation of plant biomass.

4. Conclusion

Based on the research findings, the 36-hour soaking duration and 15 ml/L concentration interact to yield optimal results for parameters such as seedling root length, seedling wet weight, and seedling dry weight. The application of PGPR in Arabica coffee seed germination can support technological innovations in the production of high-quality coffee seedlings, thereby contributing to efforts to increase coffee productivity in Indonesia.

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