



# Response of Arabica Coffee Seed Germination to *Moringa oleifera* Leaf Extract Concentration and Soaking Duration

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## Abstract

**Background:** Hard Arabica coffee seeds germinate slowly, but this can be accelerated by increasing their viability through seed priming techniques. The objective of this study was to determine the effects of concentration and immersion duration in an organic priming solution extracted from moringa leaves on germination and to identify the optimal concentration and immersion duration.

**Methods:** This study was conducted from October to December 2024 at the Greenhouse in Sidoarjo Village, Krian District, Sidoarjo, and the Production Laboratory of the Faculty of Agriculture, Universitas Pembangunan Nasional "Veteran" Jawa Timur. The study was conducted by applying two treatment factors: the concentration of the organic priming solution extracted from moringa leaves and the soaking duration. Data analysis was performed using ANOVA and regression analysis.

**Results:** The combination of treatment factors—concentration of moringa leaf extract solution and soaking duration—showed a significantly higher number of normal seedlings compared to abnormal seedlings, dead seeds, hard seeds, and fresh seeds. **Conclusions:** Based on the study's results, a 15% concentration with a soaking time of 42 hours yielded the highest results. Moreover, the regression analysis showed that the optimal predicted concentration of the moringa leaf extract solution was 12%, and the optimal predicted soaking time was 38 hours.

**Keywords:** Concentration; Germination Evaluation; Invigoration; Soaking Time

## Introduction

Coffee is highly valued in Indonesia, especially Arabica, which has a distinctive taste and aroma that coffee lovers highly value. Over time, the number of Arabica coffee enthusiasts continues to grow, driving up consumer demand. However, to meet this demand, coffee seed production must be increased through both generative and vegetative propagation. Generative propagation of coffee plants is considered quite effective, but coffee seed germination takes a significant amount of time.

Coffee seeds require approximately 3 to 4 weeks to germinate when planted in lowland areas with temperatures ranging from 30°C to 35°C, while in highland areas, the process takes approximately 6 to 8 weeks (Putra et al., 2012). Two types of seed dormancy affect germination: primary and secondary. Primary dormancy is physiological and generally caused by internal seed characteristics, such as the inability of the seed coat to absorb air. Meanwhile, secondary dormancy results from environmental factors and can occur when seeds are exposed to specific temperature and humidity levels. (Souza et al., 2025). Coffee seeds are classified as primary dormancy because Arabica coffee seeds are hard seeds, consisting of three parts in the coffee pulp: the exocarp (outer skin layer), mesocarp (flesh layer), and the hard-shelled seed coat (Anggari, 2018). The presence of a hard seed coat reduces water permeability, slowing the entry of water and oxygen into the seed (Marano & Tandirerung, 2016).



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The quality of Arabica coffee seeds is one of the efforts to improve seed viability and vigor, thereby increasing production yields. One method to enhance seed viability and vigor is through priming. Seed priming involves treating seeds before germination to improve seed performance even under unfavorable environmental conditions (Anwar et al., 2020). Seed priming can use chemical or organic materials, including moringa leaf extract (*Moringa oleifera*). Moringa leaves contain the cytokinin zeatin, which is classified as an active form that triggers growth responses, translocation, and storage (Strnad et al., 2005). Cytokinins are highly effective in increasing protein synthesis, stimulating cell division, regulating the cell cycle, and accelerating bud initiation in plants (Amriyanti & Ajiningrum, 2019). Additionally, research by Suryani et al. (2019) For Arabica coffee seeds soaked in coconut water, the best results were observed in germination rate, growth rate, and plant height. Therefore, this study was conducted to determine the effects of the concentration and soaking time of an organic priming solution from moringa leaf extract on germination, and to identify the optimal concentration and soaking time to increase the germination percentage of Arabica coffee seeds.

## Method

The research was conducted from October to December 2024 at the Green House in Sidorejo Village, Krian District, Sidoarjo Regency, and at the Production Laboratory of Universitas Pembangunan Nasional "Veteran" Jawa Timur. The equipment used included germination trays, a blender, an analytical balance, a cloth sieve, a soaking tank, label paper, a hand sprayer, a timer, a room thermometer, and a digital scale. The materials used included Arabica coffee seeds, distilled water, moringa leaves, sand, quartz paper, and water. This research employed a factorial experiment with a Completely Randomized Design (CRD) consisting of two factors: the concentration of the organic priming solution of moringa leaf extract (K), with four levels, and the duration of soaking in the organic priming solution of moringa leaf extract (L), with four levels. Each treatment was tested for K x L interaction, yielding 16 treatment combinations with three replicates per treatment. The data were then analyzed using ANOVA (Analysis of Variance) and regression analysis.

### Preparation of Growing Medium

The growing medium used was sand, which was then placed in germination trays measuring 26 cm x 21.5 cm with a sand depth of 3 cm. Each tray was labeled with the treatment details and then placed on the germination table in the greenhouse.

### Preparation of Moringa Leaf Extract Solution

The moringa leaf extract solution was prepared from young moringa leaves, which were first washed and separated from their stems. The leaves were then ground in a blender with 1 kg of moringa leaves added to 1,000 ml of water. After grinding, the leaves were filtered through a cloth sieve to obtain the moringa leaf extract.

### Coffee Seed Preparation

This research used 3-month-old Arabica coffee seeds of the Komasti variety, obtained from the Indonesian Coffee and Cocoa Research Center (Puslitkoka). The number of Arabica coffee seeds required was 1,200, with 25 seeds per replicate across 16 treatment combinations obtained from the interaction of two treatments. The seeds were previously selected based on uniform size, shape, and color.

### Preparation of Solution Concentrations

The solution concentrations used were 0% (distilled water/control), 10%, 15% and 20%. The moringa leaf crude extract was diluted using a solution volume of 1,000 ml. The solution concentrations were obtained using the formula (Chairunnisak et al., 2015):

$$M1 \times V1 = M2 \times V2$$

Notes:

- M1 = Original solution concentration  
 M2 = Concentration to be prepared  
 V1 = Volume of original solution  
 V2 = Volume of solution to be prepared

### *Soaking of Arabica Coffee Seeds*

Selected Arabica coffee seeds are soaked in an organic priming solution made from moringa leaf extract in a soaking tank labeled with the treatment concentration. The seeds are soaked for 4 treatment periods: 24, 30, 36, and 42 hours. After soaking, the seeds are rinsed with distilled water and are ready for planting.

### *Planting Coffee Seeds*

Arabica coffee seeds are planted in prepared germination trays. Each tray contains 25 Arabica coffee seeds spaced 3 cm apart, with the bud eyes facing upward.

### *Maintenance*

Maintenance involves watering twice daily, in the morning and evening, using a hand sprayer, and weeding by pulling weeds directly from the growing medium. If the growing medium is still moist, watering is not performed.

### *Data Analysis*

The data obtained will be analyzed using Anova (Analysis of Variance) with the formula used, which is (Montgomery, 2012):

$$Y_{ijk} = \mu + \alpha_i + \beta_j + \gamma_k + (\alpha\beta)_{ij} + \epsilon_{ijk}$$

Noted:

- $Y_{ijk}$  = Observation or response values for factor A obtained at level i with treatment of organic priming solution concentration of moringa leaf extract and level j of treatment duration of organic priming solution of moringa leaf extract and repetition-k.
- $\mu$  = General average.
- $\alpha_i$  = The additive effect of the concentration factor of the organic priming solution of moringa leaf extract at level I.
- $\beta_j$  = The additive effect of the immersion time of the organic priming solution extracted from moringa leaves at level-j.
- $(\alpha\beta)_{ij}$  = Interaction between the i-th level of treatment with organic priming solution concentration from moringa leaf extract and the j-th level of treatment with organic priming solution immersion duration from moringa leaf extract.
- $\epsilon_{ijk}$  = The effect of experimental errors (errors) in the concentration of organic priming solution from moringa leaf extract level i and the duration of immersion in organic priming solution from moringa leaf extract level j on observation-k

Next, a regression analysis was conducted to predict the effect of the regression analysis values on the relationship between the independent and dependent variables. The regression analysis formula used is (Suyono, 2015):

$$Y = a + bX^1 + cX^2$$

Note:

- Y = Dependent variable  
 X = Independent variable  
 a,b,c = Regression constants

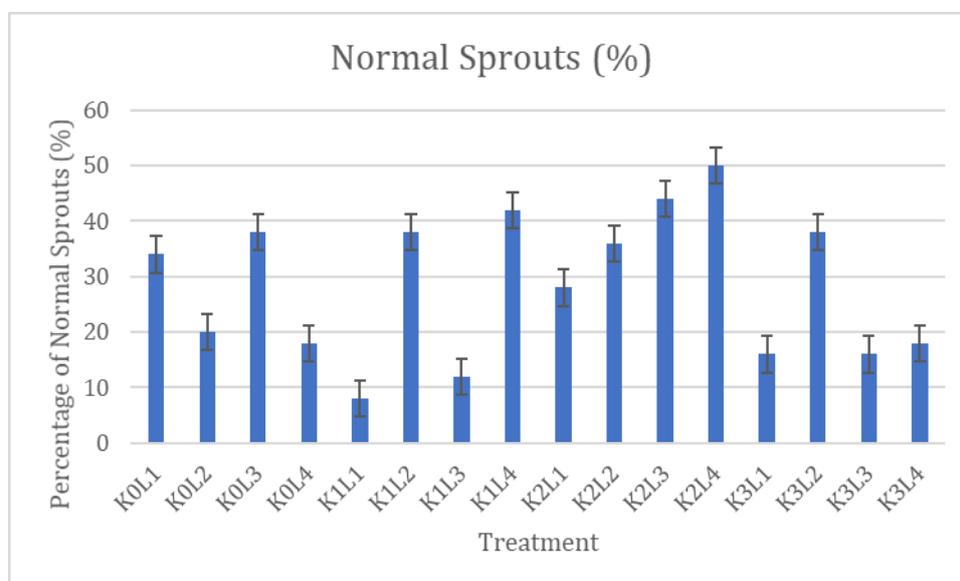
## Result

### Criteria for Arabica Coffee Seed Germination

**Table 1.** Percentage of Arabica Coffee Sprouts Based on Criteria

Treatment	Normal Sprout (%)	Abnormal Sprout (%)	Dead Seed (%)	Hard Seed (%)	Fresh Seed (%)
K <sub>0</sub> L <sub>1</sub>	34±2,83	20±5,66	20±11,31	14±8,49	12±5,66
K <sub>0</sub> L <sub>2</sub>	20±11,31	8±11,31	14±2,83	34±2,83	24±22,63
K <sub>0</sub> L <sub>3</sub>	38±8,49	12±11,31	30±8,49	18±2,83	2±2,83
K <sub>0</sub> L <sub>4</sub>	18±2,83	6±8,49	36±16,97	34±2,83	6±8,49
K <sub>1</sub> L <sub>1</sub>	8±0,00	2±2,83	30±14,14	24±5,66	36±5,66
K <sub>1</sub> L <sub>2</sub>	38±14,14	12±5,66	18±2,83	24±5,66	8±0,00
K <sub>1</sub> L <sub>3</sub>	12±0,00	8±0,00	22±8,49	18±2,83	40±5,66
K <sub>1</sub> L <sub>4</sub>	42±2,83	4±5,66	26±2,83	18±2,83	10±2,83
K <sub>2</sub> L <sub>1</sub>	28±0,00	14±8,49	28±5,66	12±0,00	18±14,14
K <sub>2</sub> L <sub>2</sub>	36±5,66	12±11,31	28±5,66	2±2,83	22±8,49
K <sub>2</sub> L <sub>3</sub>	44±5,66	6±2,83	28±11,31	8±11,31	14±2,83
K <sub>2</sub> L <sub>4</sub>	50±2,83	4±0,00	22±8,49	12±5,66	12±5,66
K <sub>3</sub> L <sub>1</sub>	16±0,00	10±2,83	32±28,28	18±14,14	24±16,97
K <sub>3</sub> L <sub>2</sub>	38±2,83	12±5,66	32±5,66	14±2,83	4±5,66
K <sub>3</sub> L <sub>3</sub>	16±5,66	8±0,00	40±5,66	22±2,83	14±2,83
K <sub>3</sub> L <sub>4</sub>	18±2,83	12±0,00	30±8,49	32±0,00	8±5,66

**Notes:** K<sub>0</sub>: Concentration 0%; K<sub>1</sub>: Concentration 10%; K<sub>2</sub>: Concentration 15%; K<sub>3</sub>: Concentration 20%; L<sub>1</sub>: Soaking time 24 hours; L<sub>2</sub>: Soaking time 30 hours; L<sub>3</sub>: Soaking time 36 hours; L<sub>4</sub>: Soaking time 42 hours.

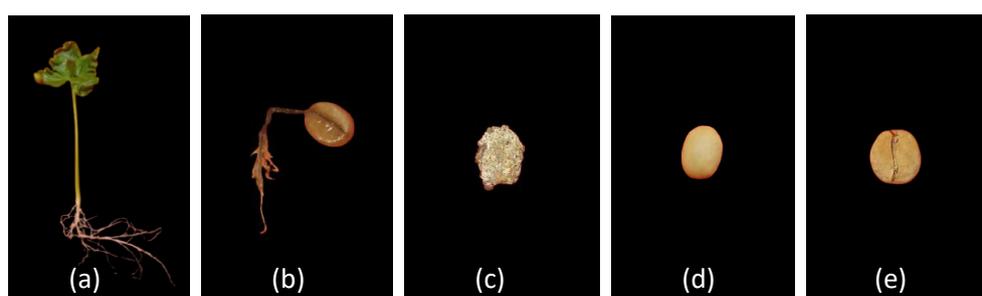


**Figure 1.** Error bar from the effect of moringa leaf extract concentration and soaking duration on the percentage of normal sprouts of Arabica coffee.

The results in Table 1 show that the combination of a 15% moringa leaf extract solution concentration and a 42-hour soaking period (K<sub>2</sub>L<sub>4</sub>) produced the highest average number of normal seedlings among the treatments. Normal seedlings are those whose structures develop properly, including the cotyledon, hypocotyl, radicle, and plumule (Figure 1). A high number of normal seedlings indicates high seed viability (Suhendra et al., 2016). Seed viability refers to the ability of seeds to germinate, usually under optimal environmental conditions or under stress (Agustiansyah et al., 2020). High seed viability indicates high quality. The response observed in K<sub>2</sub>L<sub>4</sub> may be related to the combined effects of moringa leaf extract concentration and immersion duration. Moringa leaf extract is a natural growth regulator containing zeatin, a natural component of cytokinin, phenolics, and several mineral elements. Cytokinin helps induce cell division, delay cell aging, and stimulate plant cell

growth (Mare et al., 2023). However, physiological parameters were not directly measured in this study, so the underlying mechanism can only be inferred from previous findings. Adequate soaking time can facilitate the absorption of these compounds, thereby supporting seed germination.

The combination of a 0% concentration of moringa leaf extract solution (distilled water/control) and a soaking time of 24 hours (K<sub>0</sub>L<sub>1</sub>) showed a relatively higher average abnormal germination rate than other treatments. Abnormal seedlings include rotten seedlings with organ decay due to infection, damaged seedlings with severe organ damage, and disproportionate seedlings with structural abnormalities that prevent normal plant development (Figure 1). The cause of abnormal germination is suspected to be seeds that did not receive sufficient growth stimulants to develop normally, and a soaking period that was too short compared to other treatments. According to Indriaty et al. (2022) Abnormal germination occurs because water has not been fully absorbed, resulting in unstable enzyme activation and insufficient materials to be converted into energy. However, the embryo tissue in the seeds has already ruptured.



**Figure 2.** Criteria for Arabica coffee germination: (a) Normal sprout, (b) Abnormal sprout, (c) Dead seed, (d) Hard seed, (e) Fresh seed

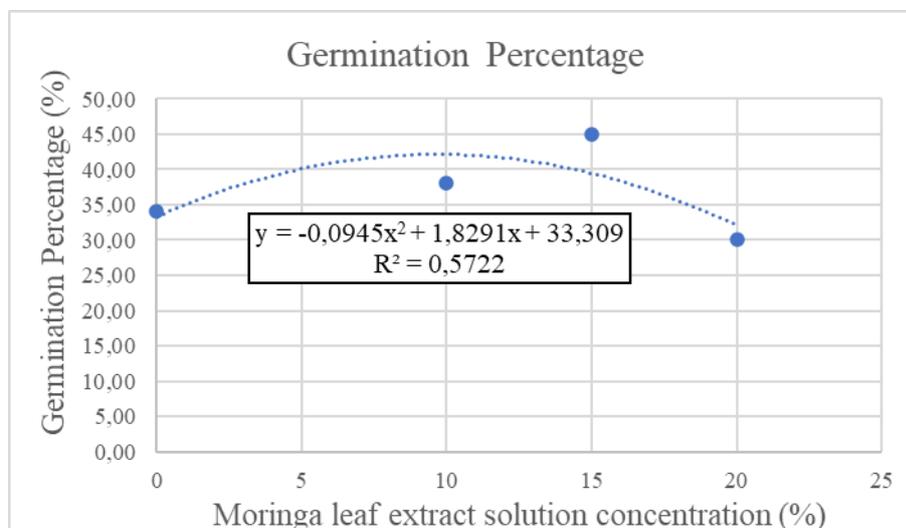
In addition to regular and abnormal sprouts, there are dead Arabica coffee seeds. Dead Arabica coffee seeds are characterized by seeds that have imbibed but have become rotten or moldy, releasing a white liquid containing bacteria, and are therefore unable to sprout (Figure 1). Table 1 shows that the combination of 20% moringa leaf extract and a 36-hour soaking time (K<sub>3</sub>L<sub>3</sub>) resulted in the highest percentage of dead seeds, accounting for 40% of the tested seeds. This was due to the high dose of the moringa leaf extract solution administered. According to Nur et al. (2024) Plants become stressed when the administered concentration is too high because the compounds in the moringa leaf extract become toxic, thereby inhibiting seed germination.

The criteria for hard seeds are those that do not undergo imbibition and are unable to germinate by the end of the testing period (Figure 1). The research results show that the combination treatment of 0% moringa leaf extract solution concentration (distilled water/control) with a soaking time of 30 hours (K<sub>0</sub>L<sub>2</sub>) and the combination treatment of 0% moringa leaf extract solution concentration (distilled water/control) with a soaking time of 42 hours (K<sub>0</sub>L<sub>4</sub>) had the same number of hard seeds, namely 34% of the total seeds tested. Arabica coffee seeds remain hard because of their water-impermeable seed coat, which prevents them from absorbing water and undergoing imbibition to stimulate germination. (Nurrachmamila & Saputro, 2017).

The results also showed that fresh Arabica coffee seeds were relatively abundant, accounting for 40% of the total seeds tested. This percentage was observed in the treatment combination of a 10% moringa leaf extract solution with a soaking time of 36 hours (K<sub>1</sub>L<sub>3</sub>). Arabica coffee seeds classified as fresh seeds are those that have undergone imbibition but did not germinate by the end of the testing period; however, if the research period were extended, they could potentially germinate (Nurrachmamila & Saputro, 2017).

### Regression Analysis of Concentration and Soaking Time

The results of the regression analysis on the single treatment of the concentration of the organic priming solution extracted from moringa leaves showed that it significantly affected the germination rate of Arabica coffee seeds. Based on the regression analysis equation  $Y = -0.0945x^2 + 1.8291x + 33.309$ , the  $R^2$  value was 0.5722, indicating that the concentration of moringa leaf extract solution influences the germination percentage of Arabica coffee seeds by 57.22% (Figure 2). The regression analysis indicates that a solution concentration of 12% is predicted to be optimal for influencing the germination rate of Arabica coffee seeds.

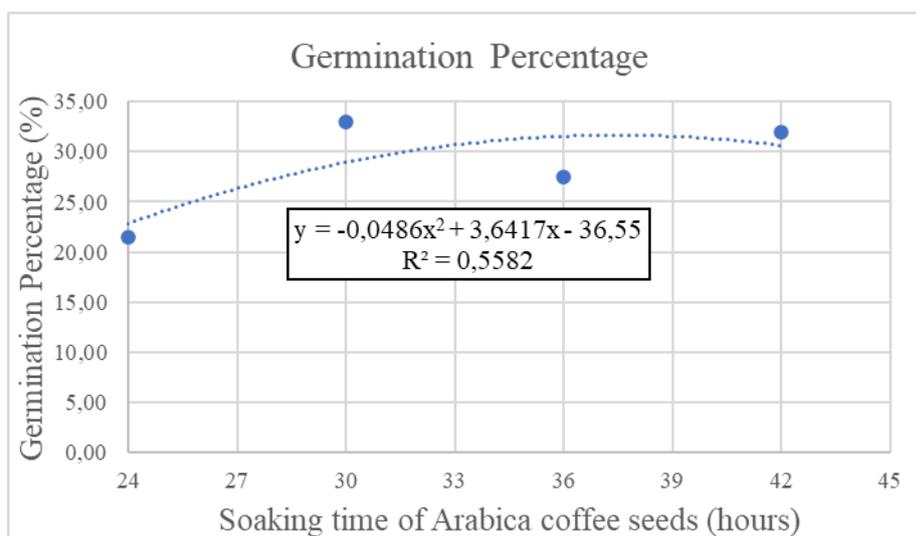


**Figure 3.** Quadratic Regression Graph of the Relationship Between the Concentration of Organic Priming Solution from Moringa Leaf Extract and the Germination Rate

The regression analysis results in Figure 1 indicate that the moringa leaf extract solution has a 57.22% effect, meaning that its concentration has a moderate effect on the germination percentage of Arabica coffee seeds. Moringa leaf extract contains cytokinin, a plant hormone that enhances plant performance by optimizing tissue water status, strengthening plant growth, increasing plant antioxidant content, and improving membrane stability. (Rehman et al., 2017). According to Krisnadi (2015) Moringa leaves contain approximately 0.00002  $\mu\text{g}$  to 0.02  $\mu\text{g}/\text{g}$  of zeatin concentration. This amount is sufficient to stimulate the germination of hard Arabica coffee seeds.

Arabica coffee seeds are hard, so an appropriate concentration is needed to increase germination. Based on regression analysis, the optimal concentration of moringa leaf extract solution is 12%. The concentration applied to Arabica coffee seeds must be at the appropriate dose to enhance their viability. According to Adelia & Sunarti (2024) If the concentration applied to the seeds is too low, it will not significantly affect the plants or seeds being studied or even be detected. Meanwhile, according to Sari et al. (2014) If the applied concentration is too high, it can disrupt plant growth and development, or even cause the plant to die, as high doses damage the plant's hormonal functions.

Similarly, the single treatment of prolonged soaking in moringa leaf extract solution significantly affects the germination percentage of Arabica coffee seeds. The regression analysis equation obtained is  $Y = -0.0486x^2 + 3.6417x + 36.55$ , with an R-squared ( $R^2$ ) value of 0.5582 (Figure 3). Based on the quadratic model, a soaking time of 38 hours for Arabica coffee seeds is predicted to be optimal for influencing germination rate.



**Figure 4.** Quadratic Regression Graph of the Relationship Between the Soaking Time of Organic Priming Solution from Moringa Leaf Extract and the Germination Rate

The results of the regression analysis of the relationship between the soaking time of the organic priming solution of Moringa leaf extract and the germination percentage of Arabica coffee seeds indicate that soaking time has a 55.82% influence on germination percentage. This figure falls into the moderate influence category. The duration of seed soaking can influence seed viability and vigor. Seed invigoration techniques, such as priming, can regulate seed hydration and dehydration to facilitate metabolic processes before seed germination. (Triyadi et al., 2023). According to Puspitaningtyas et al. (2018) Through seed-soaking techniques, the seed coat of Arabica coffee seeds can be softened, allowing water to enter and the seeds to imbibe. The seeds will activate the embryo once they reach the optimal water content. Imbibition occurs due to osmotic differences between the seeds and their surrounding medium, softening the seed coat and increasing seed growth rate.

Based on regression analysis results, the predicted optimum soaking time for Arabica coffee seeds is 38 hours. Seeds subjected to soaking treatment must be soaked for the appropriate duration to achieve good seed viability. According to Wahdah et al. (2021) Seeds that are soaked for too short a time may not be able to induce germination. Similarly, if seeds are soaked for too long, seed viability may be affected.

The difference between the empirical optimum (15% concentration with 42 hours of immersion) and the regression-based optimum (12% concentration with 38 hours of immersion) may be due to several factors. First, the regression model is constructed from discrete experimental data points, whereas the predicted optimum represents a continuous estimate between treatment levels. Second, biological responses such as seed germination are inherently variable and may not follow a perfectly symmetric quadratic pattern. In addition, environmental and physiological variability between seeds may contribute to deviations between observed and predicted values. Therefore, regression analysis should be interpreted as a predictive tool rather than an absolute determinant of optimal conditions. Its application is limited by the number of treatment levels, the discrete nature of the experimental design, and the biological variability inherent in seeds.

## Conclusion

Based on the study's results, a 15% concentration of moringa leaf extract and 42 hours of soaking produced the highest percentage of normal Arabica coffee germination among the treatments tested. Meanwhile, the regression analysis predicted an optimal soaking time of 38 hours at a 12% concentration of moringa leaf extract. This optimal prediction indicates a model estimate and does not entirely replace the research observation results. Further studies with more precise treatment intervals are needed to validate and align the research and predictive results.

## Declaration statement

The authors report no potential conflict of interest.

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