The effectiveness of germination pre-treatment on mung beans, peanuts, and tomatoes

M Muttaqin1*, R I Putri1, D A Putri1 and D D Matra2

1 Department of Biology, IPB University, Bogor 16680, Indonesia
2 Department of Agronomy and Horticulture, IPB University, Bogor 16680, Indonesia

*E-mail: mafrikhul.bio@apps.ipb.ac.id

Abstract. Improvement in the germination process will improve the productivity of mung beans (Vigna radiata), peanuts (Arachis hypogaea), and tomatoes (Solanum lycopersicum), the three Indonesian important crops. Seed immersion in the water is one of the germination pre-treatments. The aim of the study was to evaluate the effects of seed immersion in pre-determined water on mung beans, peanuts, and tomatoes germination. The study used a factorial completely randomized design with three factors consisted of water pH (5, 7 and 9), water temperature (27 ºC and 50 ºC), and immersion time (15 and 30 minutes) on 3 replications. Seed germination process was carried out in a rolled paper method. The study showed that different water pH, water temperature, and immersion time gave a significant effect on the germinability, germination speed, seed vigor index, and the percentage of normal growth seeds. Moreover, the seed immersed in the water (pH 7, 50ºC) for 15 minutes showed the best effect on germination quality.

1. Introduction
Mung beans (Vigna radiata), peanuts (Arachis hypogaea), and tomatoes (Solanum lycopersicum) are known as the three important crops utilized in Indonesia mainly as dishes or raw materials. Mung beans classified as the third important crop in the category of nuts [1] while peanut was one of Indonesian most focus commodity based on the Indonesian Ministry of Agriculture [2]. Moreover, based on Indonesian export projection in 2014-2019 developed by Indonesian Ministry of Agriculture, tomato production should be improved due to the future local and ASEAN market high demand [3].

In a period of 2014-2015, the production (ton) of mung beans in Indonesia was drop by 10% while an increase in production (ton) was experienced by peanuts (6%) and tomatoes (4%) [4]. However, the increasing number in peanut and tomato production was relatively low. Moreover, not only by using a new variety development of these crop [5, 6], but the improvement in production also can be achieved by observing the seed quality and testing its germination process [7].

Seed germination is the process of plant seed growing into a plant. The process affected by some environmental factors, such as water, temperature, light, as well as plant hormones [8, 9]. Germination pre-treatment, pre-germination in short, described as the treatments for seeds to induce germination before planting. Seed priming, for instance, is able to increase the rate and uniformity of emergence in many vegetable and flower species [10] or to prepare the plant to respond to imminent abiotic stress aggressively [11]. One of seed priming treatment is seed immersion in water which has different pre-condition.
To improve the seed germination process, the water used for immersion can be pre-determined by its acidity level, temperature, and time. Water availability is the main factor that breaks seed dormancy and will lead to several physiological processes towards seed germination. Different range of water pH affected the seed germination rate differently depend on its species \[12,13,14\]. Meanwhile, Roberts \[8\] said that high temperatures generally reinforce dormancy or may even induce it and low temperatures may also induce dormancy in some circumstances. Those effect on seed germination will be varied depending on other environmental factor or seed genetics or physiology.

In order to improve the germination quality of mung bean, peanut, and tomato seeds, this study observed the effect of pre-determined water, varied by pH, temperature, and time, used for seed immersion. So, the aim of the study was to evaluate the effects of seed immersion in pre-determined water on mung beans, peanuts, and tomatoes germination.

2. Materials and Methods

The materials used in this study were seeds of mung bean (Vima-2), peanut (Bogor local variety), and tomato (Karina) obtained from local farmer shop. Other materials such as sterilized water, fungicide, cameras, pH meter, rulers, opaque paper, clear plastic, Petri dishes, computers, thermometers, autoclaves, and germinator (GTM-01 Stainless steel) were used in this study.

The mung beans, peanuts, and tomatoes seeds were sorted, water cleaned, then soaked in fungicide (DithaneTM M-45 80 WP, 0.5 g/L) in two minutes. The seeds then rinsed by sterilized water three times, dried, and immersed in water contains varying degrees of water acidity and temperature in different immerse times by a completely randomized design. The design has factors: pH (5, 7 and 9), temperature (27ºC and 50ºC), and time (15 and 30 minutes) and conducted in 3 replications which each replication consisted of 10 seeds.

The soaked seed then germinated using the rolled paper packed in plastic method and stored in a GTM-01 Germinator for 7 days. In these days, the observation \[15\] on the seed germinability (1), germination speed (2), seed vigor index (3), and the percentage of normal growth seed (4) based on following equations were conducted. Then, an analysis of variance was performed by using IBM SPSS ver. 16.

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(1) \text{Seed Germinability} (\%) = \frac{\sum (NSd3+NSd7) \times 100}{\sum \text{planted seeds}}
\]
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(2) \text{Germination Speed} (\% NS/etmal) = \frac{\% NSd1 + \% NSd7}{etmal} + \ldots + \frac{\% NSd1 + \% NSd7}{etmal}
\]
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(3) \text{Seed Vigor Index} (\%) = \frac{\sum NSd1 \times 100}{\sum \text{planted seeds}}
\]
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(4) \text{Normal Growth} (\%) = \frac{\sum NSd7 \times 100}{\sum \text{planted seeds}}
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Note: NSd3: number of normal sprouts in day-3; NSd7: number of normal sprouts in day-7; NSd1: number of normal sprouts in day-1.
3. Results and Discussion

3.1. Seed germinability and vigor index

Table 1 gives information about the mung bean, peanut, and tomato seed germinability and vigor index. Based on Table 1, a combination of seed immersion treatment in water pH 7 and 50 °C for 15 minutes showed the best effect on these seed germinability and vigor index.

| Treatment/Seed | Mung Bean | Peanut | Tomato | Mung Bean | Peanut | Tomato |
|---------------|-----------|--------|--------|-----------|--------|--------|
| A1B1C1        | 90,00     | 30,00  | 57,33  | 86,67     | 30,00  | 50,00  |
| A1B1C2        | 80,00     | 46,67  | 57,67  | 76,67     | 50,00  | 50,00  |
| A1B2C1        | 93,33     | 56,67  | 71,33  | 93,33     | 56,67  | 63,33  |
| A1B2C2        | 93,33     | 60,00  | 59,00  | 86,67     | 60,00  | 50,00  |
| A2B1C1        | 100,00    | 70,00  | 53,33  | 96,67     | 70,00  | 46,67  |
| A2B1C2        | 96,67     | 66,67  | 53,00  | 96,67     | 63,33  | 46,67  |
| A2B2C1        | 100,00    | 73,33  | 72,00  | 100,00    | 76,67  | 63,33  |
| A2B2C2        | 100,00    | 76,67  | 49,67  | 100,00    | 80,00  | 43,33  |
| A3B1C1        | 90,00     | 53,33  | 36,67  | 96,67     | 56,67  | 30,00  |
| A3B1C2        | 100,00    | 63,33  | 32,33  | 90,00     | 30,00  | 26,67  |
| A3B2C1        | 100,00    | 66,67  | 55,00  | 96,67     | 66,67  | 46,67  |
| A3B2C2        | 100,00    | 66,67  | 54,33  | 93,33     | 70,00  | 46,67  |

- Average data followed by similar letter in the same column means not significantly different based on 5% of DMRT (Duncan’s Multiple Range Test)
- A1 = pH 5, A2 = pH 7, A3 = pH 9; B1 = 27°C, B2 = 50°C; C1 = 15 minutes, C2 = 30 minutes

Overall, the mung bean seed germinability was higher than e-CFR (Electronic Code of Federal Regulations, LII, 1994) and SeedNet (seednet.gov.in) bean standard which is 70% whereas the other seeds tend to lower than standard, 65-75%. The seed germinability, expressed in percent units, illustrates the ability of plant seeds to germinate and grow normally and reflects seed physical, physiological, and genetic conditions.

Water acidity can influence seed germinability. Lower acidity has a detrimental effect on the seed germination because of inhibition of enzyme synthesis needed for germination [16]. In addition, a lower or higher pH in water also affects seed germination negatively [17].

The high-temperature water used for seed immersion yielded the highest germinability in mung bean and peanut seeds. Hot water often is used as a breaking dormancy agent [18] by stresses and ruptures the seed micro sclereids layer. This will lead to higher cell surface tension increase allowed the cell membrane cracking, moreover, water is able to enter the seed.

A consistency with [19] research, the seed immersed in water for 30 minutes shows the best seed germinability. In 30 minutes the water is able to lower seed coat hardness level so water can enter the seed to trigger the germination process.

The seed vigor index stated as uniformity and speed of seeds germination at certain times. The seed vigor test has an aim to obtain information on seed growth in diverse and wide environmental conditions and it provides information on seed quality during storage [15]. It also reflects a combination of seed age, seed resistance, seed strength, and seed health that can then be tested by seed stress test or biochemical analysis [20]. Higher vigor index will allow the seed to survive better in unfavorable conditions when compared to low vigor index seeds. Seeds with high vigor values will also grow faster than seeds with low vigor [21].

Seed vigor index is affected by acidity which is, in this research, the water acidity. Water pH 5, for instance, affect negatively in seed protein synthesis, thereby reducing the value of seed vigor index [22].
In this research water pH 7 combined by hot temperature resulted in the highest vigor index value on mung bean, peanut and tomato seeds. This is in accordance with [23] which states that the treatment of soaking the seeds in distilled water with normal pH for 24 hours also produces a good effect on seed vigor index on lamtoro seeds. Low vigor index in seeds can be caused by genetic, physiological, morphological, mechanical and microbial factors [24] as well as external factors such as harvesting and storing process [25]. The low seed vigor index causes low leaf chlorophyll content leads to a significant decrease in crop production [26].

Seed immersion in water for 30 minutes resulted in the highest vigor index value. It is said that 30 minutes were the most effective treatment to improve seed vigor [27]. However, the longer immersion time also tends to increase the seed vigor index, for example, 24-hour peanut seeds immersion [28] and 48-hour Setaria italica seeds immersion [29].

3.2. Seed normal growth and germination speed

| Treatment/Seed | Normal growth seeds (% NS) | Germination speed (%NS/etmal) |
|---------------|---------------------------|------------------------------|
|               | Mung Bean  | Peanut  | Tomato | Mung Bean  | Peanut  | Tomato |
| A1B1C1        | 86.67 b    | 16.67 a | 73.33 ab | 2.08 b    | 1.04 ab | 16.97 ab |
| A1B1C2        | 76.67 a    | 20.00 a | 76.67 ab | 1.85 a    | 0.99 ab | 16.70 abc |
| A1B2C1        | 86.67 b    | 33.33 cd | 80.00 ab | 2.17 c    | 1.09 c | 19.07 bc |
| A1B2C2        | 86.67 b    | 33.33 cd | 90.00 b  | 2.12 c    | 1.14 c | 18.43 bc |
| A2B1C1        | 96.67 bc   | 36.67 de | 73.33 ab | 2.25 c    | 1.33 c | 15.57 abc |
| A2B1C2        | 93.33 bc   | 30.00 cd | 70.00 ab | 2.28 c    | 1.19 c | 15.10 abc |
| A2B2C1        | 100.00 c   | 46.67 e  | 86.67 b  | 2.41 c    | 1.42 c | 20.63 c  |
| A2B2C2        | 100.00 c   | 23.33 abc | 66.67 a  | 2.41 c    | 1.29 c | 14.60 ab |
| A3B1C1        | 96.67 bc   | 20.00 abc | 66.67 a  | 2.28 c    | 0.98 b | 13.67 a  |
| A3B1C2        | 90.00 bc   | 23.33 abc | 56.67 a  | 2.17 c    | 0.59 a | 12.37 a  |
| A3B2C1        | 96.67 bc   | 23.33 abc | 83.33 ab | 2.34 c    | 1.15 c | 18.10 bc |
| A3B2C2        | 96.67 bc   | 36.67 de | 76.67 ab | 2.24 c    | 1.27 c | 17.00 abc |

*a Average data followed by similar letter in the same column means not significantly different based on 5% of DMRT (Duncan’s Multiple Range Test)

*b A1 = pH 5, A2 = pH 7, A3 = pH 9; B1 = 27°C, B2 = 50°C; C1 = 15 minutes, C2 = 30 minutes

Table 2 shows that combination of seed immersion treatment in water pH 7 and 50 °C for 15 minutes showed the best effect on the mung bean, peanut, and tomato seeds normal growth as well as seed germination speed. Figure 1 shows the normal growth of mung bean, peanut, and tomato seeds that characterized by well developed important sprout structures (root systems, axial shoots, cotyledons, and terminal buds). Abnormal sprouts described as sprouts that do not show the potential to develop into normal sprouts characterized by damage on, deformed structure, rotten, hypocotyl or epicotyl strongly twisted, curved, forming a spiral, the seeds do not grow and lost it important structure.
Figure 1. Normal growth of mung bean (left), peanut (middle), and tomato (right) seeds. The bar scale is 1 cm in length.

Lower or higher water pH will lead to a lower value of normal growth seed. Lower and higher acidic environment affect negatively on seed germination because of inhibition of enzyme activity such as amylase synthesis that will reduce starch hydrolysis in the endosperm lead to seed abnormal growth [14, 22]. Meanwhile, the mung bean and peanut seeds immersed in pH 7 water combined with a hot temperature resulted in the highest normal seed growth [30]. Seeds that grow normally indicate that the germination process in the seeds runs well.

Seed growth speed indicates the seed ability to grow in suboptimum conditions. Research [31] stated that the immersion treatment in water for 24 hours was the best treatment in accelerating the appearance of sprouts, increasing the average germination, and the dry weight of the canopy. Seeds that grow faster, are better able to deal with suboptimum field conditions.

Acid water will inhibit germination speed by increasing cell electrolyte leakage, inhibit protein synthesis, and respiration rate [22]. Concurrently, normal pH water mixed with hot-cold water treatment can increase water permeability into the seeds and eliminate metabolic barriers so that the seed germination speed increases [32].

Overall, based on this research, the seeds immersed in the water (pH 7, 50°C) for 15 minutes showed the best effect on germination quality: germinability, germination speed, seed vigor index, and the percentage of normal growth seeds. The farmer, then, may adopt this research finding in their planting practices especially in seed germination to improve the plant productivity.

4. Conclusion
Overall, based on this research, the Mung bean, peanut, and tomato seeds that immersed in the water (pH 7, 50°C) for 15 minutes showed the best effect on germination quality: germinability, germination speed, seed vigor index, and the percentage of normal growth seeds. The farmer, then, may adopt this research finding in their planting practices especially in seed germination to improve the plant productivity.

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