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# Increasing Papaya Seed Viability By Osmoconditioning Treatment With Polyethylene Glycol (PEG) 6000

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**ABSTRACT**
Papaya (Carica papaya L.) is a plant that has high economic value, although its distribution is limited. One of the obstacles in increasing papaya production is the limited availability of seeds caused by a decrease in seed quality due to less than optimal storage. Seed viability is thought to be able to be increased through osmoconditioning techniques using Polyethylene Glycol (PEG) 6000. This study aims to evaluate the effect of osmoconditioning with PEG 6000 on the viability of papaya seeds (Carica papaya L.). The experimental design used was a Randomized Block Design with two treatment factors repeated three times. The first factor is the concentration of PEG 6000 consisting of 0%, 2%, 4%, and 6%. The second factor is the soaking time, which is 4 hours, 8 hours, and 12 hours. The results showed that osmoconditioning treatment with PEG 6000 had a significant effect on the viability of Papaya seeds (Carica papaya L.). The most effective concentration of PEG 6000 is 4%, while the optimal soaking time is 6 hours.



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## INTRODUCTION

Papaya is a very profitable herbaceous fruit plant from the Caracecae family. Increasing public awareness of the importance of this fruit can increase its demand, which means that the supply and quantity of papaya must be increased. To overcome this problem, papaya cultivation must be developed and increase its productivity by increasing production efficiency and expanding the scale of the business. Technological improvements are very important to increase papaya production [1].

Papaya is one of the fruits that has very good nutritional content. This fruit can be consumed as fresh fruit and can also be in the form of processed products. Papaya fruit contains 1.0% to 1.5% protein, 1% to 1.5% vitamin A, and also contains 69 mg to 71 mg of vitamin C. Meanwhile, for the mineral content of papaya fruit, it contains 11 mg to 31 mg of calcium and 39 mg to 337 mg of potassium. Other contents in low-fat papaya fruit are 0.1%, 7% to 13% carbohydrates, 35 kcal to 59 kcal, 200 kJ of energy and 85% to 90% water. Other parts of the papaya plant such as roots, leaves, fruit, and seeds are known to contain phytochemicals in the form of polysaccharides, minerals, vitamins, enzymes, alkaloids, proteins, saponins, glycosides, and flavonoids, all of which can be used as a source of nutrition

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and medicinal ingredients [2].

Increasing papaya production must begin with ensuring the availability of high-quality and affordable seeds in sufficient quantities. Papaya is a monocotyledonous plant that can only be grown from seeds. Therefore, quality seeds are needed to achieve high production results. Seed quality includes genetic, physiological and physical quality. Meanwhile, papaya seeds have a dormancy period of 12 to 15 days. This fairly long dormancy period is caused by the presence of aryl and phenolic compounds in the seed aryl [3].

Naturally, when papaya seeds are stored, the seeds will experience decreased viability, which means that their germination and vigor are reduced. Long storage, high room temperature, and increased respiration can cause decreased food reserves. Storing seeds at cold temperatures can make it difficult for seeds to imbibe water, so viability will decrease. According to [4], invigoration is a way to accelerate germination, which can be done by soaking seeds in an osmotic solution (which has the ability to control the amount and speed of water absorption). Faster germination begins with rapid imbibition, which triggers subsequent earlier processes, such as seed coat rupture, hormone and enzyme activation, food reserve breakdown, radical release, and nutrient translocation.

Invigoration techniques are widely used to improve seed quality so that seed viability will be more optimal and will result in seeds being able to grow quickly and uniformly in diverse environments. The invigoration technique used in this study is the osmoconditioning technique. Osmoconditioning is the process of improving the biochemical and physiological quality of seeds that occurs during the dormancy period [5]. The purpose of osmoconditioning is to accelerate and also synchronize germination, in addition to being able to improve the potential for seed germination. [6] stated that the basic principle of the osmoconditioning technique begins when the seeds absorb water until they reach a water potential equilibrium between the seeds and the imbibition medium. One of the methods used in osmoconditioning is the use of polyethylene glycol (PEG).

Based on the results of research that has been conducted on several seeds, it can be seen that the osmoconditioning technique using PEG is considered effective in increasing the germination rate of seeds that have low viability. The use of PEG can accelerate the germination process. PEG is a compound that can reduce the osmotic potential of a solution by binding water. Osmoconditioning using PEG has been successfully applied to rice, carrot, soybean, and cashew seeds [4].

PEG is a compound that is soluble in water and can enter cells, so it can be used for the invigoration process. PEG has the ability to bind water, which can support seeds in the water imbibition process. Invigoration treatment is carried out physiologically to increase the germination rate by water imbibition, which is the basis for seed invigoration techniques. According to [7], the invigoration technique is the treatment of seeds using osmotic solutions that aim to increase speed and uniformity during the germination process.

Based on previous studies, osmoconditioning using PEG 6000 has shown potential in increasing seed viability. The results of the study by [8] stated that the PEG 6000 concentration treatment had a very significant effect on germination power and a significant effect on the simultaneity of soybean seed growth. Osmoconditioning using a 15% PEG-

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6000 solution produced the highest germination power value in both varieties, which was 79.33%. However, this value was not significantly different compared to other concentrations. The 15% concentration also produced the highest growth simultaneity value, which was 57.62%. Thus, it is known that the osmoconditioning treatment using a 15% PEG-6000 concentration is the best treatment because it can increase the germination power and simultaneity of soybean seed growth, so it can be used as one solution to increase the viability and vigor of seeds that have experienced a decline in quality.

Meanwhile, the research results of [9] also concluded that the interaction of soybean seed treatment soaked for 12 hours using a PEG 6000 solution with a concentration of 15%, effectively produced the most optimal growth simultaneity and hypocotyl length values. In the single factor of osmoconditioning invigoration, the use of a PEG-6000 solution with a concentration of 15%, produced the highest germination power, growth rate and normal dry weight of seedlings. In the single factor of soaking time, soaking seeds for 12 hours, provided optimal germination power, vigor index and root length values.

[10] also conducted research on soybean seeds and concluded the same thing, that osmoconditioning invigoration with PEG 6000 solution gave the best results compared to soaking treatments with other solutions to increase soybean seed viability. Where the best concentration is 15% and soaking for 3 hours. The positive effect of invigoration can even increase production results, as shown in the results of research on Detam-1 and Detam-2 soybean seeds reported by [11]. The effect of invigoration increased production by 13% compared to the control.

In contrast to soybean seed osmoconditioning, [12] in their research conducted osmoconditioning with PEG 6000 on rice seeds. The results obtained were that PEG 6000 -2.0 Bar had a very good effect on seed viability and vigor of expired rice seeds. This is in line with the results of [13] research that PEG 6000 -2.0 Bar solution was effective in increasing PTM values in expired Ciherang rice. Based on Marleni's (2009) research, osmoconditioning using PEG with an osmotic potential of -12.5 Bar provided the best vigor and viability in local red field rice seeds.

Based on several things that have been described above, the author has conducted a study entitled "Increasing Papaya Seed Viability with Osmoconditioning Treatment with Polyethylene Glycol (PEG) 6000". Here the author wants to know the best concentration and soaking time of PEG 6000 in increasing papaya seed invigoration.

## METHOD

This research was conducted in the laboratory of Al-Azhar University, Medan Johor District, Medan City, Indonesia. This research was conducted from April to August 2024, using a Randomized Block Design with 2 factors. The first factor is the concentration of PEG 6000 (P) consisting of P0 (0%), P1 (2%), P2 (4%), P3 (6%). The second factor is the soaking time (L), namely L1 (3 hours), L2 (6 hours), and L3 (9 hours), and with 3 replications. The variables observed in this study were germination rate and germination ability.

### Seed selection

The selected seeds are put into water and the seeds that sink to the bottom of the

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container are classified as full seeds and will be used in the study, while the floating seeds are discarded. After soaking, the seeds are aired on a tissue so that the outer skin is not damp.

**Dilution or dissolution of PEG 6000**

The preparation of PEG 6000 solutions with concentrations of 2%, 4%, and 6% was carried out by dissolving 2 grams, 4 grams, and 6 grams of PEG 6000 respectively in distilled water until the volume of each solution reached 100 ml.

**Osmoconditioning treatment with PEG 6000**

Papaya seeds that have been selected as research materials are separated into 4 parts (according to the level of osmoconditioning treatment). Seeds that will be given osmoconditioning treatment are soaked in PEG 6000 solution with concentrations of 2%, 4%, and 6%.

**Seed nursery**

Prior to the nursery, the seeds were soaked with a fungicide dose of 2 g/l. After that, the seeds were planted into poly bags that already contained planting media, where one poly bag for one seed and one treatment plot consisted of 10 poly bags. Seeds are germinated with a depth of ± 1 cm. Seed germination is expected up to 6 weeks after germination.

*Observation*

1. Germination rate (Day)
   According to [14], germination rate can be measured by calculating the number of days required for the emergence of the radicle or plumule.
   $$\text{Germination Rate} = \frac{\text{N1T1} + \text{N2T2} + \cdots + \text{NxTx}}{\text{total number of seeds that germinated}}$$
   N = number of seeds that germinate in a certain time unit, T = shows the amount of time between thestart of the test to the end of a certain interval of an observation.
2. Germination Ability Test (%)
   Germination ability is determined by calculating the number of seeds that germinate normally for 14 days. Using the formula:
   $$\text{Germination Ability} = \frac{\text{Total Number of Normal Sprouts}}{\text{Number of seeds tested}}\ x\ 100\%$$

## RESULTS AND DISCUSSION

**Evaluation**

Based on the results of the analysis of variance, it is known that the concentration of PEG 6000, the duration of soaking, and the interaction between the two gave very significant different results on the germination rate and germination ability of papaya seeds.

**Germination Rate (Days)**

The results of observations of the germination rate were carried out from the first day after sowing until the end of observation or 42 days. From the analysis of variance, it can be seen that the interaction between the concentration of PEG 6000 and the duration of soaking showed a very significant effect on the germination rate. The results of the mean difference test with the DMRT (Duncan Multiple Range Test) on the effect of interaction between the application of plant growth regulators and osmoconditioning treatment on the germination

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rate are presented in Table 1.

**Table 1**. Average Germination Rate (Days)

| Teatment | Germination Rate (Days) | |
|---|---|---|
| P0L1 (PEG 0% + Soaking time 3 hours) | 8.07 | i |
| P0L2 (PEG 0% + Soaking time 6 hours) | 8.20 | ij |
| P0L3 (PEG 0% + Soaking time 9 hours) | 8.80 | k |
| P1L1 (PEG 2% + Soaking time 3 hours) | 5.87 | h |
| P1L2 (PEG 2% + Soaking time 6 hours) | 5.47 | efg |
| P1L3 (PEG 2% + Soaking time 9 hours) | 5.67 | gh |
| P2L1 (PEG 4% + Soaking time 3 hours) | 4.80 | bc |
| P2L2 (PEG 4% + Soaking time 6 hours) | 4.40 | a |
| P2L3 (PEG 4% + Soaking time 9 hours) | 4.60 | ab |
| P3L1 (PEG 6% + Soaking time 3 hours) | 5.27 | def |
| P3L2 (PEG 6% + Soaking time 6 hours) | 5.00 | cd |
| P3L3 (PEG 6% + Soaking time 9 hours) | 5.20 | de |

Description: The numbers in the columns followed by the same letter are not significantly different at the 5% level according to the DMRT (Duncan Multiple Range Test) mean difference test.

From table 1, it can be seen regarding the effect of interaction between concentration and soaking time of PEG 6000 on the germination rate, that the interaction that produces the fastest germination rate is in the interaction between 4% concentration and 6 hours soaking time (P2L2) which is 4.40 HSS. Meanwhile, the longest germination rate value is produced by P0L3 (Concentration 0% and soaking time 9 hours) which is 8.80 HSS which is significantly different from all existing treatment combinations. From the results of the analysis of variance, it can be seen that the concentration of PEG 6000 and the soaking time of papaya seeds have a very significant effect on the germination rate. The results of the average difference test with DMRT (Duncan Multiple Range Test) on the effect of concentration and soaking time of PEG 6000 are presented in Figures 1 and 2.

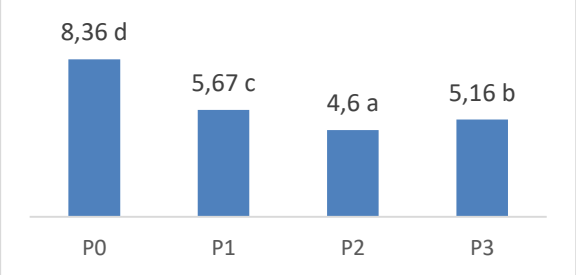


**Figure 1.** Effect of PEG 6000 Concentration on Germination Rate (Days)

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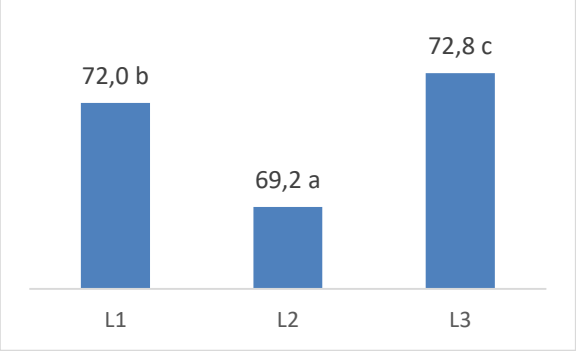


**Figure 2.** Effect of Soaking Time in PEG 6000 on Germination Rate (Days)

### Germination Ability Test (%)

The results of observations of germination ability conducted at 14 Days. From the analysis of variance, it can be seen that the interaction between the concentration and soaking time of PEG 6000 showed a very significant effect on germination ability. The results of the mean difference test with the DMRT (Duncan Multiple Range Test) on the effect of interaction between concentration and soaking time of PEG 6000 can be seen in table 2.

**Table 2.** Average Germination Ability Value (%)

| Treatment | Germination Ability (%) | |
|---|---|---|
| P0L1 (PEG 0% + Soaking time 3 hours) | 53,00 | a |
| P0L2 (PEG 0% + Soaking time 6 hours) | 62,67 | c |
| P0L3 (PEG 0% + Soaking time 9 hours) | 54.00 | ab |
| P1L1 (PEG 2% + Soaking time 3 hours) | 87,00 | d |
| P1L2 (PEG 2% + Soaking time 6 hours) | 88,00 | def |
| P1L3 (PEG 2% + Soaking time 9 hours) | 87,67 | de |
| P2L1 (PEG 4% + Soaking time 3 hours) | 97,67 | j |
| P2L2 (PEG 4% + Soaking time 6 hours) | 99,00 | j |
| P2L3 (PEG 4% + Soaking time 9 hours) | 98,00 | j |
| P3L1 (PEG 6% + Soaking time 3 hours) | 88,33 | defg |
| P3L2 (PEG 6% + Soaking time 6 hours) | 89,33 | defghi |
| P3L3 (PEG 6% + Soaking time 9 hours) | 89,00 | defgh |

Description: The numbers in the columns followed by the same letter are not significantly different at the 5% level according to the DMRT (Duncan Multiple Range Test) mean difference test.

From table 2, it can be seen regarding the effect of interaction between concentration and soaking time of PEG 6000 on germination rate, that the interaction that produces the highest germination ability is in the interaction between concentration of 4% and soaking time for 6 hours (P2L2) which is 99%. Meanwhile, the longest germination rate value is produced by P0L1 (Concentration 0% and soaking time 3 hours) which is 53% which is significantly different from all existing treatment combinations. From the results of the analysis of variance, it can be seen that the concentration of PEG 6000 and the soaking time of papaya seeds have a very significant effect on the germination ability of papaya seeds. The

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results of the average difference test with DMRT (Duncan Multiple Range Test) on the effect of concentration and soaking time of PEG 6000 are presented in Figures 3 and 4

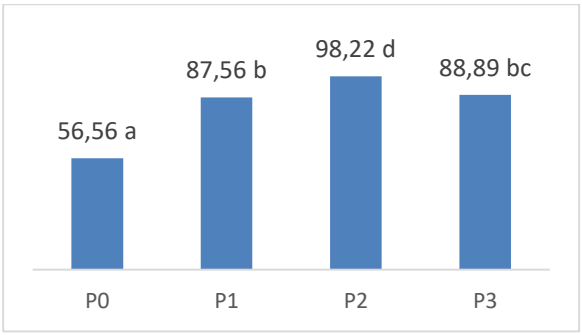


**Figure 3.** Effect of PEG 6000 Concentration on Germination Ability (%)

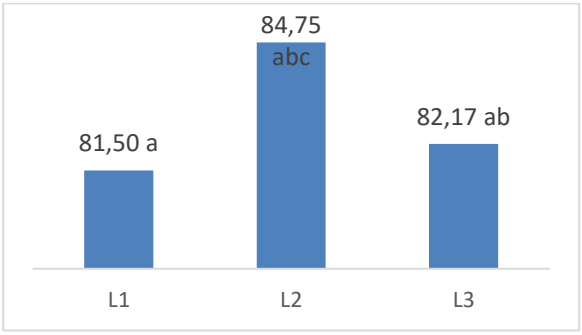


**Figure 4.** Effect of Soaking Time in PEG 6000 on Germination Power (%)

## Discussion

In general, seeds given different concentration treatments from seeds given control treatments because seeds given treatments with different PEG 6000 concentrations experience controlled water imbibition, which allows water to enter the seeds gradually until they reach equilibrium.

The results of this study are in line with [15], imbibition assisted by osmoconditioning treatment allows seeds to optimize their internal components to initiate germination, such as restoring membrane integrity, because deteriorating seeds can produce better seeds. From the results of the study, it can be seen that giving a PEG 6000 concentration of 6% actually reduces the germination rate and germination ability. This is because the higher the concentration of PEG 6000 given, the more water will be bound to enter the seeds. This has been explained by [16] who stated that the higher the concentration of PEG, the faster the seed's chance of imbibing water becomes, so that the amount of water entering the seed also increases. If water seeps into the seed excessively, it can reduce the oxygen space needed by the seed to breathe. When respiration is blocked, seedling growth will also be inhibited, so that its ability to germinate will also decrease.

According to [17], the concentration of osmotic solution that is too high causes the seeds to stop absorbing water in phase II. According to [18], the process of water absorption by seeds follows a triphasic pattern (3 phases). Phase I begins with rapid water absorption,

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this is due to the potential difference between water and seeds. Water has a potential value of 0 Mpa, while the potential value in seeds (especially orthodox seeds) is between -50 and -350 Mpa. Furthermore, in phase II, water absorption is slow, because the water potential of the seeds with their environment is in balance, but seed metabolism is actively taking place. In phase III, water absorption increases again, which is the germination process has been completed, marked by the emergence of radicles [17].

In general, the pattern of water absorption in seeds treated with osmoconditioning is no different compared to seeds without treatment, only the rate of water absorption is slowed down and controlled [19]. Osmoconditioning causes the potential of the seed environment to be lower, so that the rate of water absorption at the beginning of imbibition (phase I) can be slowed down.

The germination rate is slow at low concentrations, because when soaking seeds with low concentrations of PEG 6000 only a little water can be bound so that very little water can enter the seed. Lack of water in the seed increases the time required for seed germination by inhibiting the process of seed metabolism and the breakdown of food needed for germination.

According to [20], water is the most important basic requirement for germination. It is used to soften the seed coat, causing the embryo and endosperm to swell and break the seed coat. In addition, water also helps move stored food to the growth points that need it. The effective soaking time in increasing papaya seed viability in this study was 6 hours and was very different from other treatments. Where soaking for 3 hours was less effective and soaking for 9 hours also slowed down the germination rate and could not increase germination power compared to soaking for 6 hours. Soaking seeds in PEG 6000 solution aims to insert PEG material into the seeds. PEG has water-binding properties so that when absorbed by the seeds it can help the absorption process and start the germination process.

Soaking PEG 6000 for 9 hours did not seem to be able to accelerate the germination rate and increase germination power, this happened because the germination process requires an optimal amount of water. Water absorption occurs through the seed coat through diffusion and osmosis. The longer the seeds are in the PEG solution, the more PEG is absorbed into the seeds and the greater the possibility that the seeds will absorb water quickly and excessively [21].

According to [9], water is absolutely necessary for germination. However, soaking for too long can cause anoxia (loss of oxygen) and inhibit the respiration process. Respiration is a stage of the germination process that occurs after the water imbibition process. When the respiration process is limited, the germination process also slows down. According to the results of research conducted by [22] on the effect of PEG 6000 on cotton seed viability, a 9-hour soaking period effectively increased the germination rate, and the average germination rate increased by 6.04 days.

## CONCLUSION

From the results of the research that has been done, it can be concluded that there is a very significant effect of the concentration of Polyethylene Glycol (PEG) 6000 on the viability of papaya seeds (Carica Papaya L.), a concentration of 4% gives the highest results in increasing

the germination and germination power of papaya seeds. There is also a very significant effect of the duration of soaking Polyethylene Glycol (PEG) 6000 on the viability of papaya seeds (Carica Papaya L.). The highest germination and germination power are produced at a soaking time of 6 hours. It can also be concluded that there is a very significant interaction effect of the concentration and duration of soaking on both parameters, the highest results are seen given by the combination of a concentration of PEG 6000 4% with a soaking time of 6 hours. The results of this study can be used as a reference in efforts to increase the viability of papaya seeds that are declining, it is recommended to soak PEG 6000 with a concentration of 4% for 6 hours.

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