



## UTILIZATION OF LIQUID ORGANIC FERTILIZER TO BREAK SEED DORMANCY AND GROWTH OF SUGAR APPLE (*Annona squamosa* L.) SAPPLINGS

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### ABSTRACT

The sugar apple (*Annona squamosa* L.) is generative propagation had obstacles caused by the hard seed coat that inhibited the imbibition process, and the seeds became dormant. Liquid organic fertilizer (LOF) can be utilized to break seed dormancy and the growth of sugar apple seedlings. This study was conducted to decide the correct concentration and splashing time of LOF for breaking seed dormancy and growth of sugar apple seedlings. The research was conducted using factorial design with two factors, LOF concentration (0%, 25%, 50%, 75%, 100%) and soaking time (6, 12, 18, 24 hours). Data used ANOVA and DMRT. The results showed that the LOF concentration, soaking time and the interaction between the combination of LOF concentration and soaking time were not yet obtained to break seed dormancy and the growth of sugar apple seedlings. The use of 25% LOF concentration showed significant differences with other LOF concentration treatments (50, 75, 100%) on growth parameters (plant height, number of leaves and fresh biomass) but not significantly different compared to the control on all germination and growth parameters. Soaking times of 6 hours and 12 hours were not significantly different on all germination and growth parameters but substantially different from soaking 18 hours and 24 hours on germination parameters (germination percentage and vigour index). The interaction of LOF concentration and soaking time was not significantly different on all germination and growth parameters.

## INTRODUCTION

Sugar apple (*Annona squamosa* L.) may be a tropical fruit plant native to Latin America. It belongs to the genus *Annona* and has a relatively high nutritional content of

vitamin C (Sari et al., 2024). Sugar apples can be cultivated generatively (seeds) or vegetatively (grafting and budding) (Suryani, 2011). Generative cultivation of sugar apples is preferred because the root system is stronger, the plant has a longer lifespan, and it is more economical (Roslinda *et al.*, 2022).

Generative cultivation of sugar apples is hampered by the fact that the seed coat is hard and rigid, requiring a relatively long time for seed germination (Wahyuni *et al.*, 2023). Germination of sugar apple seeds without treatment or naturally takes 38 days (Sunder *et al.*, 2024). The structure of the apple seed consists of the integument and endosperm, with a layer of macrosclerotic tissue forming the outermost layer of the integument (Muwarni, 2012). The cell walls of macrosclerotic tissue contain lignin, which makes the cell walls thick and hard (Ramdhini *et al.*, 2022). This can hinder the imbibition process, leading to sugar apple seed dormancy, requiring precise and effective efforts to break seed dormancy.

Seed dormancy can be lifted by scarification, either mechanically or chemically. (Amalia 2016). Scarification using chemicals is quite widely used, but continuous use and high doses will damage the environment and affect plant physiology (Wijayanti, 2023). The high cost of chemicals is also an obstacle to their use. An alternative is therefore needed, such as the use of liquid organic fertilizer (LOF) derived from organic waste, which is easy to obtain, economical and environmentally friendly. Liquid organic fertilizer quickly dissolves in the soil, offering numerous advantages. It is simple to produce and requires absorption by plants. Additionally, it enhances soil structure and is user-friendly (Tuapattinaya *et al.*, 2024).

Rissa & Bangun (2023) proved that the application of LOF from a combination of banana stalks, coconut water and rice wash water at a concentration of 45 ml/L can increase the height and stalk diameter of forage maize seedlings compared with the control. Nutrients such as phosphorus and potassium from banana stems and carbohydrates from rice wash water can support the growth of soursop seedlings. Coconut water contains growth hormones that can activate the enzymes that destroy food reserves in the seed germination process. This is supported by research by Pranata *et al.* (2018), who found that soaking soursop seeds in coconut water at a concentration of 50% resulted in the highest germination rate of 70.14%. Another study by Okoli (2022) on the effects of soaking time and soursop seed weight revealed that seeds weighing 0.6 to 0.8 g had a

higher germination percentage (92.90%) compared with lighter seeds weighing 0.3 to 0.5 g (78.60%). In addition, soaking the seeds in coconut water for 60 hours resulted in a higher germination percentage (83.30%) than seeds not soaked in coconut water, which had a germination percentage of 75%.

The phytohormone content of coconut water is suspected of promoting germination, and certain nutrients in banana stems and rice wash water are likely to support the growth of sugar apple seedlings after germination. Therefore, this investigation was conducted to decide the appropriate LOF concentration and splashing time, as well as to understand the interaction between LOF concentration and splashing time on seed dormancy breaking and sugar apple seedling growth. The results of this study should provide information on the use of LOF from banana stems, coconut water and rice washing water with the appropriate concentration and soaking time as a cost-effective alternative for seed dormancy breaking and sugar apple seedling growth.

## **MATERIALS AND METHODS**

### ***Place and Time of The Research***

The research was conducted from July to September 2024. The LOF was made in the Botany Laboratory of the Biology Department, Faculty of Mathematics and Natural Sciences, University of Riau. The planting and maintenance of the sugar apple were carried out in the Biological Experimental Garden, University of Riau, Pekanbaru.

### ***Material and Tools***

The materials needed were banana stems (40-day-old bananas), coconut water, rice wash water, brown sugar solution, EM4 (Songgolangit Persada), sugar apple seeds (1000 seeds), aquades, and mineral soil. The tools used were buckets, 25×30 polybags, plastic containers, hoes, shovels, mobile phone cameras, knives, measuring flasks, measuring cups, hand sprayers, rulers, digital scales (Digipounds I-2000) and stationery.

### ***Research Plan***

The study utilized a total factorial randomized plan (RAL Factorial) with two components. This plan was chosen for its ability to efficiently test multiple factors and their interactions. The first factor was LOF concentration (0, 25, 50, 75, 100%), and the

second factor was soaking time (6, 12, 18, 24 hours). Twenty combinations with five replications resulted in 100 experimental units.

### ***Research Procedure***

#### **Making LOF and Preparation of Planting Media**

The preparation of LOF was carried out based on Rissa & Bangun (2023) using 5 kg of banana stems (40-day-old bananas), 10 L of rice wash water, 10 L of coconut water, 3 L of brown sugar solution, and 250 ml of EM4. The inner part of the banana stems was selected, amounting to 5 kg, then coarsely chopped and cleaned with water. All the ingredients were mixed in a container and stirred evenly. The container was tightly closed and placed in a location protected from sunlight. The ingredients were fermented for 2 weeks, and every 2 days, the container was opened to prevent gas buildup.

The planting medium used was mineral soil that had been cleaned of impurities. The soil was then placed into polybags and positioned in the Biological Experimental Garden.

#### **Selection, Treatment and Planting of Seeds, Maintenance**

The seeds, carefully selected from ripe sugar apples, underwent a rigorous treatment process. They were soaked in water, and only the seeds that sank were used in the study. These selected seeds were then immersed in a LOF solution according to the treatment concentration and soaking duration. The seeds that had been soaked according to the treatment were then planted in polybags at a depth of 2 cm from the soil surface and maintained until 70 days after planting (DAP). Maintenance includes daily watering and weeding, ensuring the health and accuracy of the study.

#### **Observation**

Observations include germination parameters (germination percentage (%), germination time (days), vigour index) conducted until 30 days after planting (DAP) and continued with growth parameter observations performed at the end of the study (70 DAP), including plant height (cm), number of leaves (leaves), fresh biomass (g), and root length (cm).

### ***Data Analysis***

The quantitative data obtained were meticulously analyzed using ANOVA and the DMRT post hoc test at the 5% level. This thorough analysis ensures the reliability of the study's conclusions.

RESULTS AND DISCUSSION

Germination Parameter

The results in Table 1 show that the use of 25% LOF is significantly different from the other LOF concentrations but not substantially different from the control in terms of germination rate. Seeds without LOF (control) were able to germinate more than the other treatments. Seeds with 50% LOF, however, exhibited a unique behavior, with faster germination times than the other LOF concentrations and Even the control. As the LOF concentration increased, the germination rate and vigour index decreased, and the seeds took longer to germinate. In terms of germination capacity, the control (no treatment) showed higher germination rates than the use of LOF. However, the overall germination rates were less than 50% for all treatments, indicating that the use of LOF or the absence of LOF is not effective in breaking the dormancy of sugar apple seeds.

**Table 1.** The mean germination percentage (%), germination time (days), and vigor index (seeds/day) at the end of the observation (70 DAP)

Parameter	Soaking Time (Hours)	LOF Concentration (%)					Mean
		0	25	50	75	100	
Germination Percentage (%)	6	20	16	18	20	16	18 <sup>b</sup>
	12	20	14	12	16	16	15.6 <sup>b</sup>
	18	8	20	12	6	2	9.6 <sup>a</sup>
	24	14	8	6	4	-	6.4 <sup>a</sup>
Mean		15.5	14.5	12	11.5	8.5	
Germination Time (days)	6	16	17	17	18	18	17.5
	12	17	17	16	18	21	17.7
	18	21	17	17	17	15	17.9
	24	17	21	21	18	-	19
Mean		18	18	17	18	19	
Vigor Index (seed/day)	6	0.12	0.10	0.11	0.11	0.09	0.10 <sup>b</sup>
	12	0.12	0.08	0.08	0.09	0.08	0.09 <sup>b</sup>
	18	0.05	0.12	0.07	0.03	0.01	0.06 <sup>a</sup>
	24	0.09	0.04	0.02	0.02	-	0.03 <sup>a</sup>
Mean		0.09	0.09	0.07	0.06	0.05	

Note:

- The symbol (-) indicates seeds that did not germinate
- Numbers in the same column followed by the same letter are not significantly different according to the DMRT test at the 5% level

Some things that inhibit the imbibition process so that the breaking of dormancy of sugar apple seeds have not been effectively carried out, among others, was thought to be because the seeds used come from the same level of fruit maturity (fruit is not physiologically mature). In addition, in the treatment, there were obstacles in seed

germination, presumably because LOF had hormone content from coconut water and rice washing water, so the concentration may be an obstacle in the process of seed germination. It is evident that as the concentration increases, LOF is used, the lower the percentage of germination, as well as the length of soaking time; the longer soaking can also affect the germination time and reduce the vigour index. Kadir *et al.* (2020), in their research, showed that exogenous application of high concentrations of hormones (GA3) inhibited Arabica coffee beans from germinating.

Seeds that germinate may be due to the concentration of hormones given exogenously, not disturbing the balance of endogenous hormone concentrations in the seeds so that the seeds are able to germinate. Seeds that did not germinate were thought to be because the concentration of hormones in the LOF was too high or insufficient to trigger seed germination. Imansari & Sri (2017) stated that another factor that affects the ability of seed germination is the availability of nutrients in each different seed. In addition, according to Murrinie *et al.* (2021), exogenously administered hormones require a longer time to help the process of  $\alpha$ -amylase formation during the germination process.

The study found that a soaking time of 6 hours resulted in a significantly different mean of 18 and 24 hours, but not substantially different from 12 hours of soaking on germination percentage, germination time, and vigour index. This was because seeds that were soaked for longer periods in the treatment could be deprived of the oxygen needed for respiration. Respiration, a stage that occurs after the imbibition process, produces ATP as an energy source for seeds to process food reserves. If the respiration process is inhibited, so is the germination process. The study also found that the food reserves in seeds were used as an energy source for germination, and their quantity and quality affected the germination of seeds. Research by Polhaupessy & Sinay (2014) showed that soaking soursop seeds in 20 ppm GA for 24 hours resulted in a lower germination percentage than soaking for 6 and 12 hours. Srilaba *et al.* (2018) stated that while soaking seeds can accelerate germination, excessive soaking can actually damage the seeds.

The interaction of concentration treatment and soaking time showed that no seeds germinated at a concentration of 100% LOF for 24 hours. This was thought to be because the concentration of hormones given exogenously exceeds the optimal hormone concentration limit in seeds, making it an obstacle for seeds to germinate. According to Alghofar *et al.* (2017), seeds that have reached optimum imbibition when still soaked will

cause a faster energy breakdown process and will lack oxygen in the breakdown process, causing the seeds to be damaged or die.

***Growth Parameter***

The mean plant height (cm), number of leaves (blade), fresh biomass (g) and root length (cm) at the end of observation (70 days) are presented in Table 2. There was a significant difference between 25% LOF concentration and other LOF concentrations, but not significantly different from the control on plant height, number of leaves and fresh biomass. Plant height, number of leaves and fresh biomass in the control tended to be higher but not significantly different compared to the 25% LOF concentration.

Root length in all treatments showed no significant difference, but the highest root length was found in the control. These results indicate that soaking seeds in the given LOF concentration did not affect increasing the growth of sugar apple seedlings until the 70th DAP. Similarly, the treatment of seed soaking time showed that there was no effect on all growth parameters. However, there was a tendency that without treatment or with a low concentration of LOF (25% LOF) used, it can trigger better growth at the beginning of tiller growth than other treatments.

The occurrence of obstacles in the germination period will also affect all further tiller growth parameters. This means that soaking seeds in LOF is not effective in increasing tiller growth throughout the vegetative phase, which may also affect the generative phase. Research by Budiawan *et al.* (2023) used rice washing water for soaking coffee seeds (*Coffea* sp.); the results also did not affect seed growth.

**Table 2.** The mean plant height (cm), number of leaves (blade), fresh biomass (g) and root length (cm) at the end of observation (70 DAP)

Parameter	Soaking Time (Jam)	LOF Concentration (%)					Mean
		0	25	50	75	100	
Plant Height (cm)	6	7	5.8	3.2	9.1	5.6	6.1
	12	12.5	3.9	1.3	5.2	6.1	5.8
	18	5.3	8	4	1.8	1.3	4.1
	24	5.8	6.9	3.2	*	-	3.2
	Mean		7.7 <sup>b</sup>	6.2 <sup>ab</sup>	2.9 <sup>a</sup>	4 <sup>a</sup>	3.3 <sup>a</sup>
Number of Leaves (blade)	6	6	3.6	2	5.8	3.4	4.2
	12	6.6	3.2	1.2	3.4	3.4	3.6
	18	3.4	6.4	2.2	1.4	1	2.9
	24	4.8	4.4	2	*	-	2.2
	Mean		5.2 <sup>b</sup>	4.4 <sup>ab</sup>	1.9 <sup>a</sup>	2.7 <sup>a</sup>	2 <sup>a</sup>
Fresh Biomass (g)	6	0.8	0.6	0.3	0.7	0.5	0.6
	12	1.1	0.4	0.1	0.4	0.6	0.5
	18	0.5	1.2	0.4	0.5	0.1	0.5

Parameter	Soaking Time (Jam)	LOF Concentration (%)					Mean
		0	25	50	75	100	
	24	2.3	0.8	0.3	*	-	0.7
Mean		1.2 <sup>b</sup>	0.8 <sup>ab</sup>	0.3 <sup>a</sup>	0.4 <sup>a</sup>	0.3 <sup>a</sup>	
Root Length (cm)	6	15.7	13.9	12.6	20.6	13.6	15.3
	12	21.9	9.8	6.4	12	15.9	13.2
	18	15.9	16.5	13.6	3.4	3	10.5
	24	11.8	7.8	11.8	*	-	6.3
Mean		16.3	12	11.1	9	8.1	

Note:

- Signs (-) indicate seeds that did not grow
- Sign (\*) plants died at 50 day after planting (DAP)
- The numbers in the same row followed by the same lowercase letters are not significantly different according to the DMRT test at the 5% level.

The study's main findings reveal that the interaction of LOF concentration treatment and soaking time did not significantly affect the mean of all growth parameters. The control group, interestingly, showed a tendency for higher mean growth parameters compared to the LOF concentration and soaking time treatment. These results suggest that the nutrients and growth hormones in LOF may not be sufficient to significantly enhance seedling growth up to 70 DAP. This conclusion is in line with Kurnia *et al.*'s (2016) statement that prolonged seed soaking can lead to cell damage, hindering germination and growth.

## CONCLUSION

LOF concentration, soaking time, and the interaction between the combination of LOF concentration and soaking time have not yet been obtained to break seed dormancy and the growth of sugar apple seedlings. The use of 25% LOF concentration showed significant differences with other LOF concentration treatments (50, 75, 100%) on growth parameters (plant height, number of leaves and fresh biomass) but not significantly different compared to the control on all germination and growth parameters. Soaking times of 6 hours and 12 hours were not significantly different on all germination and growth parameters but substantially different from soaking 18 hours and 24 hours on germination parameters, namely germination percentage and vigour index. Importantly, the interaction between LOF concentration and soaking time did not reveal any significant difference in all the measured germination and growth parameters, providing a clear conclusion.



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