

Phytochemical Content and Effectiveness of Ketapang Fruit Extract (*Terminalia catappa*) as Bioherbicide on Seed Germination and Growth of Sensitive Plant (*Mimosa pudica* L.) and Corn (*Zea mays* L.)

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Abstract

This study aims to identify the phytochemicals in ketapang fruit and determine their effects on seed germination and growth of sensitive plants and corn. This study used Randomized Block Design (RBD) with treatments of 0% bioherbicide, 1% glyphosate, and 1%, 3%, and 5% bioherbicide concentrations. Ten seeds with four replicates were used for seed germination, and one plant with five replicates for growth. The parameters observed were seed germination (germination rate and vigor index), growth (height, number of leaves, root length, fresh weight, cob length and diameter). Data were analysed using Two-Way ANOVA and Kruskal-Wallis test. There were seven compounds: alkaloids, triterpenoids, tannins, saponins, flavonoids, phenolics, and steroids. Ketapang fruit bioherbicide affected the germination of sensitive plant seeds but did not affect corn seeds. Ketapang fruit affected the parameters of plant height, number of leaves, root length, and fresh weight of corn and sensitive plants. The concentration that provided the highest suppression was 5%. There was an interaction between bioherbicide of ketapang fruit and the plant type on height, number of leaves. But there was no interaction on root length, fresh weight of sensitive plants and corn.

Keywords: Allelopathy; bioherbicide; ketapang fruit; corn; *Mimosa pudica*; natural resources.

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INTRODUCTION

Increased corn consumption needs to be balanced with high-quality production. One of the main factors affecting corn productivity is weeds. Weeds are classified as plant pests that hinder growth and development processes, thereby reducing crop yields. In general, weeds cause crop yield losses of 10–20% in terms of both quality and quantity of production. Additionally, weeds can serve as breeding grounds for pests and sources of disease (Mubarok *et al.*, 2024). Palandi (2022) noted that the sensitive plant (*Mimosa pudica* L.) is a species commonly found in corn fields and has detrimental effects on the growth of these plants. In practice, weed control is generally carried out by spraying herbicides.

Herbicides are chemical compounds commonly used to suppress or eradicate the growth of weeds such as grass, reeds, and wild shrubs (Aditiya, 2021). The active compound contained in synthetic herbicides is isopropylamine glyphosate (Izzati *et al.*, 2024). Glyphosate can contaminate soil and the surrounding environment due to its tendency to be easily adsorbed by clay particles and organic matter, which hinders its degradation by microorganisms. Therefore, an environmentally friendly alternative is needed, namely bioherbicides (Nurzanah *et al.*, 2024). Based on their composition, herbicides are divided into two types; synthetic herbicides and bioherbicides (Elfrida, 2018).

Bioherbicides are a type of herbicide containing natural active compounds and are one of the alternatives for environmentally friendly agricultural practices. The degradation of residues produced requires a short time, so they do not accumulate in the soil. Bioherbicides have minimal impact on non-target organisms (Ginting *et al.*, 2024). Some types of weeds can produce allelopathic chemical compounds, which are released into the environment and inhibit the growth of surrounding cultivated plants. The mechanism of allelochemical inhibition is similar to that of synthetic herbicides, making the use of allelopathy a potential alternative in the development of bioherbicides (Nisa *et al.*, 2023).

Allelopathy releases compounds through the roots of living plants, during both the vegetative and generative phases, as well as through the decomposition of plant residues that have died. The compounds involved in this mechanism are known as allelochemicals (Yuliani *et al.*, 2018).

Allelochemicals are secondary metabolites produced by plants that do not directly contribute to their growth processes. *Ketapang* (*Terminalia catappa* L.) comprises alkaloids, flavonoids, tannins, saponins, steroids, and terpenoids (Herli & Wardaniati, 2019). Flavonoids exhibit a strong affinity for proteins, facilitating metabolic repression during seed germination and cell division (Susanto *et al.*, 2024).

Metabolic impediments to seed germination are caused by allelochemicals, including alkaloids, terpenoids, phenolics, steroids, and flavonoids. Specifically, flavonoids inhibit enzyme activity and hormone synthesis, both of which are required for the decomposition of food reserves. According to research by Jatsiyah *et al.* (2023), bioherbicides derived from *Ketapang* leaves (*Terminalia catappa* L.) reduce plant growth by disrupting reproductive processes. The phenolic compounds in the extract are thought to inhibit cytokinin activity, thereby limiting cell elongation and restricting plant height. *Mimosa pudica*, an invasive species, grows more rapidly than *Zea mays*, which reduces land productivity (Kurniawati *et al.*, 2018). However, the effects of *Ketapang* fruit bioherbicide application have not been thoroughly investigated, highlighting the need for further research.

This study investigates the possibility of *Ketapang* fruit bioherbicide as an alternative to synthetic herbicides for managing sensitive plants (*Mimosa pudica*) in maize (*Zea mays*) fields. The objective is to identify bioactive chemicals in the *Ketapang* fruit and evaluate the impact of varying doses of its bioherbicides on seed germination and the growth of both sensitive plants and maize. The study examines the effect of plant type and the interaction between bioherbicide concentration, germination, and growth, offering insights into sustainable weed management.

MATERIALS AND METHODS

This study was conducted from November 2024 to April 2025. It was a two-factor experimental study using a randomized block design (RBD) with the following treatments: PG = 1% glyphosate; P0 = distilled water; P1 = 1% glyphosate; P2 = 3% glyphosate; and P3 = 5% glyphosate. The second factor was the type of plant used, namely corn and a sensitive plant.

The research procedure consisted of three stages: first, phytochemical screening of the *ketapang* fruit extract; second, seed germination; third, growth of the sensitive plant and corn. The extraction process was carried out by drying the *ketapang* fruit in an oven at 70–90°C for 3–5 days. Afterward, the *ketapang* fruit was ground into a fine powder, dissolved in 99% methanol, and evaporated using a rotary evaporator (Dewi *et al.*, 2022). Phytochemical screening was performed on seven compounds; alkaloids, phenolics, flavonoids, triterpenoids, saponins, tannins, and steroids, using Harborne method (1987).

The second stage the seed germination process, which involved sterilizing the seeds using 1% sodium hypochlorite for 30 minutes, followed by rinsing them with distilled water, using 10 seeds each of the sensitive plant and corn. Next, the seeds were placed and arranged at equal intervals on a cotton-lined tray (Rahayu, 2018). The bioherbicide was applied by hand-spraying until all parts of the seeds and cotton were evenly wet. Observations were conducted daily up to 14 days after sowing (DAS). Observation parameters included seed germination rate and vigor index, using the following formula (Hotima *et al.*, 2024):

$$\text{Germination Index (GI)} = \frac{\sum \text{Gt}}{\sum \text{Tt}} \quad (1)$$

Notes:

£Gt: total number of seeds germinated on day (t)

£Tt: total number of days of germination on day (t)

$$\text{Sprout Vigor Index (SVI)} = \text{PKC} \times \text{PJK} \quad (2)$$

Notes:

PKC : % germination

PJK : sprout length (cm)

The third stage involved observing growth. Corn seeds (*Zea mays*) and sensitive plants (*Mimosa pudica*) were soaked in a Furadan solution for 24 hours. A 1:1 mixture of compost and soil was prepared in 40x40 cm plastic bags. Seeds were placed in planting holes, with 3 seeds (two as spares) in each hole, then covered with growing medium. Sensitive plants (*Mimosa pudica*) were planted after the corn plants were 24 days old. Fertilization was performed twice, 7–10 days after planting, using urea and Phoska in a 1:2 ratio. The second fertilization was carried out 15–25 days after planting using urea, KCl, and Phoska in a 1:1:2 ratio, with a fertilization dose of 5 grams per corn plant. Bioherbicide application using the *ketapang* fruit was performed every six days at 8:00 AM, with a dose of 5 ml (Widiani *et al.*, 2015).

Spraying was performed using a 1-liter spray bottle. Plant care required watering twice daily, in the morning and evening (Edy *et al.*, 2021). Observations were made of plant height (cm) and number of leaves (n) every 10 days, as well as root length (cm) and fresh weight (grams) on the final day of observation. In this study, the sensitive plants were cultivated for up to 60 days after planting (DAP), while the corn was cultivated for up to 80 DAP (Su'ud & Lestari, 2018).

Data analysis was conducted in three stages, including tests for normality and homogeneity, and a two-way Analysis of Variance (ANOVA) to determine the significant effects of each bioherbicide treatment on the ketapang fruit. If the test results indicated a significant effect, Duncan's Multiple Range post-hoc test was performed at 5% significance level. If the data were not normally distributed, the nonparametric Kruskal-Wallis test was performed. If the data showed significant results, the Dunnett post hoc test was conducted to identify differences between treatments.

RESULTS

This study identified the phytochemical content of *ketapang* fruit extract, the effects of various ketapang fruit bioherbicide treatments, and their interaction with plant species on seed germination and the growth of sensitive plants and corn. The germination parameters observed included germination rate and vigor index. Growth parameters include plant height, leaf number, root length, and wet weight. For corn plants, generative parameters include cob length and cob diameter. The results of phytochemical screening for seven compounds (Table 1), the results of the Dunnett post-hoc test on seed germination (Table 2), and the results of the Duncan post-hoc test on growth (Tables 3 and 4) showed that the methanol extract of ketapang fruit contained seven compounds: alkaloids, phenolics, flavonoids, triterpenoids, saponins, tannins, and steroids. The application of the ketapang fruit bioherbicide significantly affected the germination of sensitive plants but did not affect corn seed germination. This bioherbicide had a significant effect on all growth parameters of corn and sensitive plants (plant height, leaf number, root length, and fresh plant weight). Furthermore, there was no interaction on seed germination parameters, but there was interaction on growth parameters, particularly on the height of sensitive plants, the number of corn and sensitive plant leaves, and the root length of sensitive plants.

Phytochemical testing of the ketapang fruit extract was conducted to determine the phytochemical content of the bioherbicide. The phytochemical constituents analyzed included seven compounds: flavonoids, alkaloid D, alkaloid W, alkaloid M, phenolics, tannins, and saponins. The results of the phytochemical analysis of the ketapang fruit extract are shown in Table 1 below:

Table 1. Phytochemical screening of methanol extracts from ketapang fruit (*Terminalia catappa*).

Phytochemical Testing	Reagent	Results	Conclusion (+)/(-)
Alkaloid	Mayer	White precipitate	(+++)
	Wagner	Brown precipitate	(+++)
	Dragendorff	Orange precipitate	(++)
Flavonoid	Mg+HCl Concentrated+ etanol	Red color	(++++)
Saponin	-	Stable foam present	(+++)
Steroid	Liebermann-Burchard	Blue-green/green	(+++)
Triterpenoid	Kloroform+H ₂ SO ₄ Concentrated	Brownish red	(+++)
Fenolik	NaCl 10%+Gelatin 1%	White precipitate	(++)
Tanin	FeCl ₃ 1%	Greenish brown	(++++)

Note: (-) : None; (+) : Slight; (++) : Moderate; (+++) : Abundant; (++++) : Very Abundant

Testing alkaloid compounds with Mayer's reagent produces a large white precipitate. Testing alkaloids with Wagner's reagent produces a large brown precipitate. Testing alkaloids with Dragendorff's reagent produces a moderate orange precipitate. Testing flavonoid compounds using Mg + concentrated HCl + ethanol reagent produces a large red precipitate. Testing saponin compounds by heating and shaking produces abundant stable foam for 10 minutes. Testing steroid compounds using the Liebermann-Burchard reagent produces abundant blue/green color. Testing triterpenoids with chloroform + concentrated H₂SO₄ reagent produces abundant reddish-brown color. Testing phenolic compounds with 10% NaCl + 1% gelatin produces a moderate white precipitate. Finally, testing tannin compounds with 1% FeCl₃ produces a large amount of brownish-green color.

After testing the phytochemical compounds in the methanol extract of the ketapang fruit, measurements of environmental conditions (growing medium) were taken, including pH, temperature, and humidity. These measurements were taken twice: before and after the application of the

bioherbicide derived from the ketapang fruit extract. The results of the pre- and post-treatment growing medium tests are presented in Table 2 below:

Table 2. Observation of environmental parameter of planting media

Measurement Time	Environmental Parameter		
	pH	Humidity (%)	Temperature (°C)
Before Bioherbicide Application	6	89	32
After Bioherbicide Application	5,5	90,2	33

Measurements of environmental conditions, including pH, humidity, and temperature in the growing medium, were taken before the application of bioherbicides (before planting) to corn and sensitive plants to determine the environmental conditions required for the research. The pH measurement yielded an acidity level of 6, which is in line with the Indonesian National Standard (SNI) as it supports the availability of essential mineral nutrients such as (N), (K), (P), (Ca), and (Mg) for corn plant growth. The humidity measurement shows a value of 89%, which is within the ideal range of 80-90% for corn growth. The temperature measurement shows a value of 32°C, according to the Indonesian National Standard (SNI), which states that the ideal temperature for corn cultivation is within the range of 21-34°C (60-85°F).

Measurement of environmental conditions after bioherbicide application aims to determine the impact on the surrounding environment (growing medium). After 50 days of bioherbicide application at 10-day intervals, there were changes in the surrounding environmental conditions, particularly pH, which decreased from 6 to 5.5 after application. This condition is still within the SNI safety standard range, which is 5.5-6.5. Humidity was measured at 90.2%, exceeding the ideal safe range for corn cultivation according to SNI standards (80-90%). The increase in humidity was influenced by high rainfall. During the experiment, rainfall ranged from 109 to 435 mm/month. However, the appropriate rainfall for corn plants is around 100-200 mm/month (Primilestari *et al.*, 2021). The temperature was recorded at 33°C, which is still ideal for corn cultivation, within the 21-34°C range according to the Indonesian National Standard (SNI).

After observing the surrounding environmental conditions, the second phase of the study was conducted, which involved measuring seed germination parameters, including the average germination rate and vigor index of corn and mimosa seeds. Measurements were taken 14 days after sowing. The average germination rate and seed vigor index of corn and sensitive plant seeds resulting from the application of various concentrations of bioherbicide derived from ketapang fruit are shown in Table 3 below:

Table 3. Germination power and vigor index of corn and sensitive plant seeds at 14 days after sowing due to the application of various concentrations of bioherbicide from *ketapang* fruit.

Treatment	Seed Germination			
	Germination Power		Vigor Indeks	
	Corn	Sensitive Plant	Corn	Sensitive Plant
PG (Glifosat 1%)	77,5 ± 5,0	12,5 ± 5,00 ^{ab}	40 ± 8,16	5 ± 5,77 ^{ab}
P0 (Negative Control)	97,5 ± 5,0	52,5 ± 9,57 ^a	77,5 ± 9,57	17,5 ± 9,57 ^c
P1 (1% Concentration)	95 ± 10,0	30 ± 14,14 ^{ab}	60 ± 8,16	10 ± 8,16 ^{bc}
P2 (3% Concentration)	85 ± 10,0	15 ± 12,90 ^{ab}	55 ± 5,77	7,5 ± 9,57 ^b
P3 (5% Concentration)	75 ± 20,81	7,5 ± 19,57 ^b	30 ± 8,16	2,5 ± 5,00 ^a

Note: The results of the Dunn test at the 0.5 level are indicated by the notation. Numbers followed by the same letter in each column indicate no significant difference in the Dunn test at the 0.05 level.

Based on the results of the two-way ANOVA test, Ketapang fruit bioherbicide affected the germination of sensitive plant seeds but did not affect corn seeds. Based on the results of the Dunn test, it was found that the *ketapang* fruit bioherbicide concentration treatment differed significantly from the 1% and 3% concentration treatments and 1% glyphosate treatment. The 1% bioherbicide concentration treatment was significantly different from the 5% concentration and 1% glyphosate. However, it was not significantly different from the negative control treatment and the 3% concentration. The 2% *ketapang* fruit bioherbicide concentration was not significantly different from the other treatments. The 1% glyphosate treatment was significantly different from the negative control but not significantly different from the other treatments. The negative control was significantly different from the 5% concentration and 1% glyphosate, but not significantly different from the 1% and 3% concentrations. The highest inhibition of sensitive plant seed germination was observed at a concentration of 5%, with

an inhibition rate of 85%, while the treatment with the lowest inhibition rate was the negative control, with an inhibition rate of 0%.

The *ketapang* fruit bioherbicide affected the vigor index of sensitive plant seeds but did not affect the corn index. Based on the results of the Dunn post hoc test, the negative control treatment (distilled water) obtained a vigor index value of 17.5 for the sensitive plant, which was significantly different from the 5% concentration and 1% glyphosate, but not significantly different from the 1% and 3% concentrations. The 1% bioherbicide concentration in sensitive plant seedlings was significantly different from the 5% concentration but not significantly different from the negative control treatment (distilled water), 3% concentration, and 1% glyphosate, with a vigor index of 10. The 3% bioherbicide concentration on the *ketapang* fruit was significantly different from the negative control but not significantly different from the 1%, 5%, and 1% glyphosate concentrations. The 5% concentration of *ketapang* fruit bioherbicide was not significantly different from the 3% concentration and 1% glyphosate. Meanwhile, the 1% glyphosate treatment was significantly different from the negative control (distilled water) but not significantly different from the 1%, 3%, and 5% concentrations. The treatment that had the highest impact on the vigor index of sensitive plant seedlings was the 5% concentration, with an inhibition rate of 85.7%. Meanwhile, the treatment with the lowest impact was the negative control with an inhibition rate of 0%.

Growth corn's parameters included plant height (cm), number of leaves (n), root length (cm), and fresh plant weight (grams). The following results show the average growth of corn (*Zea mays*) at 80 days after planting (DAP) following the application of a bioherbicide derived from *ketapang* fruit extract at various concentrations, as presented in Table 4 below:

Table 4. Growth of corn (*Zea mays*) after application of *ketapang* fruit extract bioherbicide at various concentrations

Bioherbicide Concentration	Growth of Corn (<i>Zea mays</i>) at 80 Days After Planting					
	Height	Number of Leaves	Root Length	Wet Weight	Cob Diameter	Cob Length
PG (Glifosat 1%)	137 ± 6,51 ^b	11 ± 0,54 ^b	5,7 ± 0,54 ^b	219,8 ± 1,43 ^b	59,8 ± 6,54	13,1 ± 0,89
P0 (Negative Control)	220 ± 7,90 ^e	12 ± 1,0 ^c	11,2 ± 0,83 ^c	388,8 ± 1,71 ^d	121,12 ± 9,36	27,4 ± 1,94
P1 (1% Concentration)	184 ± 7,76 ^d	11,6 ± 0,54 ^c	9,4 ± 0,54 ^d	280,4 ± 1,65 ^d	98,5 ± 8,29	23 ± 1,58
P2 (3% Concentration)	163 ± 6,51 ^c	10,8 ± 0,89 ^b	7,8 ± 0,44 ^c	250,2 ± 0,99 ^c	79,26 ± 7,36	14,4 ± 1,14
P3 (5% Concentration)	124 ± 4,18 ^a	10,2 ± 0,83 ^a	4,8 ± 0,57 ^a	197,8 ± 0,73 ^a	36,82 ± 7,91	9,98 ± 0,77

Note: Duncan's test results are indicated by the notation (a, b, c, d, e). Numbers followed by the same letter in each column indicate no significant difference in Duncan's test at a significance level of 0.05.

Based on the results of Duncan's post hoc test, it was found that the treatments of bioherbicide concentrations of *ketapang* fruit, negative control differed significantly from each other in terms of corn height. The highest suppression was observed at a concentration of 5%, with an average corn height of 124 cm. Meanwhile, the treatment with the lowest effect on corn height was the negative control treatment (distilled water), with an average corn height of 220 cm.

For the parameter of corn leaf number, the bioherbicide treatment using *ketapang* fruit at the negative control concentration was not significantly different from the 1% concentration, but was significantly different from the 1% glyphosate and 3% concentrations. The 1% glyphosate treatment was not significantly different from the 3% concentration but was significantly different from the other treatments. The 5% concentration was significantly different from all treatments. Based on these results, it can be seen that the highest suppression was at the 5% concentration, with an average number of leaves of 10, denoted by a, which was significantly different from each treatment. Meanwhile, the lowest effect parameter was obtained with an average number of leaves of 12 in the negative control treatment (distilled water) at 80 HST.

Based on observations of root length parameters, it was found that the concentrations of the *ketapang* fruit bioherbicide significantly influenced each other. The treatment with the highest suppression was the 5% concentration, yielding an average root length of 4.8 cm with an inhibition capacity of 58%. The treatment with the lowest inhibition was the negative control, with an average root length of 12.5 cm and an inhibition rate of 0%.

Based on Duncan's post hoc test on the wet weight parameter of corn, it was found that each treatment of the *ketapang* fruit bioherbicide concentration differed significantly. Except for the negative control treatment, which did not differ significantly from the 1% concentration. The treatment with the highest effect was the 5% concentration, yielding an average wet weight of 19.7 grams with an inhibition rate of 33%. Meanwhile, the lowest effect was the negative control, yielding an average wet weight of 29 g with an inhibition rate of 0%.

In corn plants, there are generative parameters. The results showed that the bioherbicide treatment of the *ketapang* fruit affected the diameter of corn cobs. The highest inhibitory effect was observed in the 5% concentration treatment (P3), yielding an average cob diameter of 366.82 mm. Meanwhile, the treatment with the lowest inhibitory effect, the negative control (distilled water), yielded an average cob diameter of 121.12 mm. The second parameter is the length of the corn cob. The results of the observation indicate that the concentration of the *ketapang* fruit bioherbicide affects the length of the corn cob. The lowest average cob length at a concentration of 5% (P3) was 9.98 cm, which was the treatment with the highest effect because it had the highest inhibitory power. Meanwhile, the treatment with the lowest effect, namely the negative control, yielded the largest average of 27.4 cm, which was the largest average among the others.

The growth parameters of the sensitive plant include plant height (cm), number of leaves (n), root length (cm), and fresh plant weight (grams). The following are the average growth parameters of the sensitive plant (*M. pudica*) at 60 days after planting (DAP) following the application of a bioherbicide extracted from the *ketapang* fruit at various concentrations, as presented in Table 5 below:

Table 5. Growth of *M. pudica* after *ketapang* bioherbicide application at various concentrations.

Bioherbicide Concentration	Growth of Sensitive Plant (<i>Mimosa pudica</i>)			
	Height	Number of Leaves	Root Length	Wet Weight
PG (Glifosat 1%)	10,4 ± 1,30 ^b	12,3 ± 0,54 ^b	5,2 ± 0,44 ^b	8,2 ± 0,83 ^b
P0 (Negative Control)	23,2 ± 0,70 ^e	23,5 ± 1,00 ^e	8,25 ± 1,09 ^e	23,6 ± 2,4 ^e
P1 (1% Concentration)	17 ± 0,54 ^d	19,8 ± 0,54 ^d	7,5 ± 0,50 ^d	18,24 ± 1,47 ^d
P2 (3% Concentration)	13,2 ± 0,54 ^c	14,4 ± 0,89 ^c	6,25 ± 0,64 ^c	8,4 ± 0,58 ^c
P3 (5% Concentration)	8,1 ± 0,2 ^a	8,8 ± 0,83 ^a	3,25 ± 0,70 ^a	5,7 ± 0,44 ^a

Note: Numbers followed by the same letter in each column indicate no significant difference in Duncan's test at a significance level of 0.05.

Duncan's Multiple Range Test (DMRT) test on the height parameter of the sensitive plant shows that each *ketapang* fruit herbicide treatment has a significant effect on the other. The treatment with the highest effect is the 5% concentration, which resulted in an average length of the sensitive plant of 3.25 cm. Meanwhile, the treatment with the lowest effect, the negative control (distilled water), yielded an average length of 8.25 cm (notation (e)).

Analysis of the number of leaves of the sensitive plant showed that the herbicide treatments significantly affected each other. The treatment with the lowest effect, the negative control (distilled water), yielded an average number of sensitive plant leaves of 24 with an inhibition rate of 0%. Meanwhile, the treatment with the highest inhibition rate yielded an average number of leaves of 9 with an inhibition rate of 63%, corresponding to a 5% concentration of *ketapang* fruit bioherbicide at 60 HST.

Based on observations of the length of the sensitive plant roots, it can be seen that each *ketapang* fruit herbicide treatment significantly affected the other. The treatment with the highest inhibition rate, at a concentration of 5%, yielded an average length of 3.25 cm (notation a). The treatment with the lowest inhibition, the negative control (distilled water), yielded an average root length of 8.25 cm (e).

Based on the results of the Dunneth post hoc test on the root length parameter, various treatments of the *ketapang* fruit extract bioherbicide significantly influenced each other. The results of the Dunn post hoc test showed that the effect of the *ketapang* fruit bioherbicide concentration was that the negative control obtained an average wet weight of 234.8 grams. Meanwhile, the treatment with the highest effect, namely a concentration of 5%, obtained an average wet weight of 197.8 grams. The comparison of the inhibitory effects of *ketapang* fruit bioherbicide on corn (*Zea mays*) and sensitive plant (*Mimosa pudica*) (Figure 1).

Based on the inhibitory effect of bioherbicides on the growth of sensitive plant and corn, it can be seen that the highest inhibitory effect was found in corn plants compared to sensitive plants. Overall, the parameters of plant height, number of leaves, root length, and wet weight showed the highest inhibitory effect at a concentration of 5%.

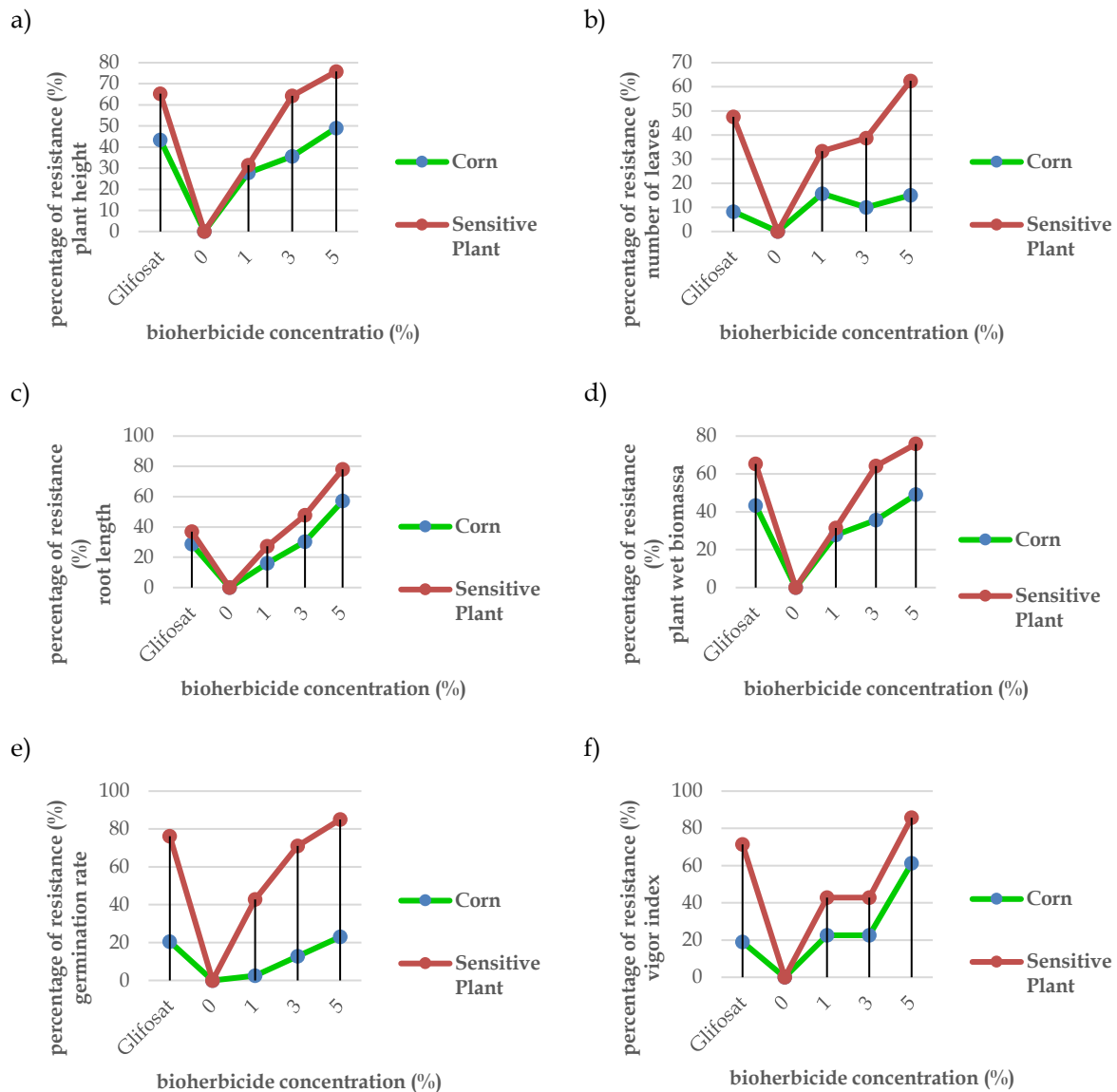


Figure 1. Inhibitory effect of bioherbicide from *ketapang* fruit (*Terminalia catappa*) on sensitive plant (*Mimosa pudica*) and corn (*Zea mays*) a) plant height, b) number of leaves, c) root length, d) plant wet biomass, e) germination rate, and d) vigor index

DISCUSSION

Phytochemical testing of *ketapang* fruit was conducted on seven compounds, namely: alkaloids, flavonoids, terpenoids, saponins, tannins, phenolics, and steroids. Alkaloid testing was conducted using the following three reagents: Mayer, Wagner, and Dragendorff. A positive result for alkaloids with Mayer reagent is indicated by the presence of a white precipitate. This white precipitate is a potassium-alkaloid complex. The addition of iodide in excess causes the formation of potassium tetraiodomercurate (II). A positive Wagner test result is indicated by a reddish-brown precipitate.

According to Jannah (2022), a positive result for alkaloids in the Wagner test indicates the formation of a light brown to yellow precipitate, a potassium alkaloid complex. Positive alkaloid results in the Dragendorff test indicate the presence of light brown to yellowish-green deposits. These sediments are potassium alkaloid complexes (Tandi *et al.*, 2020). The test results yielded orange residues, indicating that the methanol extract of the *ketapang* fruit was positive for alkaloids.

The Flavonoid testing on the *ketapang* fruit methanol extract showed positive results for the content of these compounds. The reagents used were magnesium (Mg) and hydrochloric acid (HCl), which reduced the benzopyron nucleus in the flavonoid structure, thereby forming a red-orange flavilium salt (Rismayuti & Supriningrum, 2024). The tannin test on the *ketapang* fruit methanol extract yielded positive results, indicated by the appearance of a purple-green/blue precipitate. In this tannin

test, a black precipitate was produced. The use of the FeCl_3 reagent in the tanin test determines the presence of phenolic groups in the sample, which is indicated by the appearance of a dark green color after the addition of FeCl_3 (Listiana *et al.*, 2022).

The steroid compound test of the ketapang fruit methanol extract using the Liebermann-Burchard reagent showed positive results. The test results indicated a color change to purple-blue/greenish. This is supported by Ayuchecaria *et al.* (2024), who stated that the formation of a green-blue color is a positive indicator of steroids. The addition of anhydrous acetate aims to form acetyl derivatives. Meanwhile, the addition of H_2SO_4 aims to hydrolyze the solvent, which then reacts with acetyl derivatives to form a red-purple to green-to-blue ring. Methanol extracts of the ketapang fruit are positive for triterpenoid compounds, which are characterized by a brownish-red color. This is supported by research conducted by Sholehah *et al.* (2023), which found that ketapang contains terpenoid compounds present in lipids. Terpenoid compounds are a type of hardened fat.

The saponin test showed positive results, indicated by the presence of stable foam for 1 minute after shaking. The foam was formed due to the presence of glycosides in the saponin compound of the methanol extract of the ketapang fruit. This is in accordance with the statement by Dewi *et al.* (2022) that the ketapang fruit contains natural compounds, including flavonoids, tannins, saponins, alkaloids, and terpenoids.

The phenolic compound content test was performed using a 10% NaCl + 1% gelatin reagent. The presence of a white precipitate indicates that the sample contains phenolic compounds. The test results showed the presence of a white precipitate, indicating that the methanol extraction of the ketapang fruit was positive for phenolic compounds. This is in line with the research conducted by Istarina *et al.* (2015), which stated that methanol extracts of ketapang fruit contain phenolic compounds and flavonoids.

In the second stage, the germination of seeds treated with the ketapang fruit bioherbicide was assessed using two parameters: germination capacity and seed vigor index. The germination capacity test measures the ability of seeds to germinate normally (Gea *et al.*, 2022). Germination capacity observation was conducted to measure the ability of seeds to undergo the initial growth process. Seeds that germinate are marked by the emergence of primary plant organs, namely the plumule as the shoot and the radicle as the first root to grow from the seed (Syaranamual *et al.*, 2024). The results showed that corn germination was inhibited by 23%, whereas the inhibition rate in sensitive plants was higher at 85%. In addition, the results of the Dunn test on the corn seed vigor index showed a 61.2% decrease, while the decrease in the sensitive plant vigor index was higher, reaching 87.5%.

Research on the effectiveness of kirinyuh leaf extract as a bioherbicide against the germination of beans (*Vigna radiata*) and seeds sensitive plant (*Mimosa invisia*) shows that increasing the concentration of the extract reduces the germination rate and slows down the germination of weed seeds (*Mimosa invisia*). However, the application of *Chromolaena odorata* extract had no effect on the germination of green bean seeds (*Vigna radiata*) (Frastika *et al.*, 2017). This is consistent with previous research indicating that ketapang fruit extract does not affect corn seed germination but has a significant effect on the germination of (*Mimosa invisia*) seeds. These results were obtained because the allelochemicals in ketapang fruit extract are selective, meaning they affect certain plants but not others (Frastika *et al.*, 2017).

Allelopathy refers to chemical compounds produced by plants that may be released through living roots, seeds, or even from the decomposition of dead plant material. Allelopathy contains inhibitory substances that play a role in a series of allelopathic processes (allelochemistry) (Yuliani *et al.*, 2018). Allelopathy affects plant growth through several mechanisms, including inhibition of cell division and elongation, inhibition of auxin and gibberellin activity, inhibition of enzyme activity and protein synthesis in mineral absorption, stomatal opening, photosynthesis, respiration, and membrane permeability (Yuliani *et al.*, 2018).

Allelopathy contained in ketapang fruit extract (*Terminalia catappa*). The germination process involves the role of various types of enzymes, such as hydrolytic enzymes, enzymes that function in respiration, and enzymes involved in cell synthesis and division that support the initial growth of the radicle and coleoptile. Increased synthesis of abscisic acid (ABA) can inhibit metabolic activity and disrupt the germination process, preventing seeds from developing into normal seedlings by the end of the observation period (Amartani, 2019). Qualitative tests of ketapang fruit extract showed the presence of seven compounds: alkaloids, phenols, flavonoids, triterpenoids, saponins, steroids, and tannins. The tannins in ketapang fruit have strong toxic effects, as shown by research by Nugroho *et al.* (2022), which indicates that these compounds can inhibit the activity of cellulase, pepsin, protease, polygalacturonase,

dehydrogenase, and decarboxylase enzymes. In addition, inhibition of germination can result from the disruption of seed water absorption. The higher the concentration of particles or dissolved compounds in the liquid, the lower the water potential value.

The increase in osmotic potential due to the extract compounds reduces the liquid's water potential, making it difficult for the seeds to absorb the water needed for germination (Nugroho *et al.*, 2022). According to Fadhillah *et al.* (2024), the alkaloid compounds in bioherbicides can inhibit ion transport across cell membranes. Cell membrane damage reduces seed physiology (Saadati *et al.*, 2023). Additionally, flavonoids can inhibit seed germination. Flavonoids and phenolic compounds have been shown to inhibit protein and nucleic acid synthesis, as well as inactivate several important enzymes required by plants during the early stages of growth (Sari & Jainal, 2020). One indicator of this inhibition is the high level of soluble protein, which indicates a slowing of germination enzyme activity in the breakdown of food reserves (Nugroho *et al.*, 2022).

The third stage of the study aimed to determine the effectiveness of ketapang fruit extract as a bioherbicide against the growth of corn (*Zea mays*) and sensitive plants (*Mimosa pudica*). There are four parameters of sensitive plant growth, namely plant height, number of leaves, fresh weight, and root length. The mechanism of plant growth inhibition by bioherbicides is thought to be related to the presence of allelochemicals. These compounds inhibit the synthesis of essential plant compounds, such as adenosine triphosphate (ATP) and proteins. According to Talahatu & Papilaya (2015), allelochemicals in bioherbicides inhibit the synthesis of nucleic acids, proteins, and ATP. A decrease in cellular ATP levels directly affects metabolic activity, thereby inhibiting the synthesis of other essential compounds for plant development (Sari & Jainal, 2020). According to Nasra *et al.* (2023), allelopathy decreases plant morphological traits, such as leaf number and stem height, and reduces overall plant wet weight.

The ability of *ketapang* fruit extract to inhibit corn and sensitive plants demonstrates the potential of its secondary metabolites as a bioherbicide. At certain concentration levels, allelochemicals can block and reduce the efficiency of major physiological functions in plants. These disruptions include nucleic acid formation, protein synthesis, and adenosine triphosphate (ATP) production. Decrease of ATP level disrupts almost all cellular metabolic pathways, which ultimately limits the synthesis of various essential compounds required for plant growth and development (Yuliani *et al.*, 2018).

Stem elongation is closely related to leaf formation, as the expansion of the main leaf blade is greatly influenced by intercalary meristem activity. Stem elongation occurs in the intercalary meristem tissue between stem nodes through cell division and enlargement mechanisms. This activity is significantly influenced by gibberellin hormones, which play an important role in stimulating cell elongation (Stefia & Saputro, 2017). The allelopathy mechanism is thought to suppress plant growth by disrupting phytohormone activity (Yuliani *et al.*, 2018).

Competition between cultivated plants and weeds for water occurs when the root systems of each plant and weed are close to each other, absorbing water from the same place and volume (Yuliani *et al.*, 2018). In this condition, if the weeds have longer root, they will absorb more water, which will be detrimental to the cultivated plants. Competition for CO₂ is highly detrimental to cultivated plants, as weeds are generally classified as plants with the ability to assimilate CO₂ at a high rate. According to Setiawan *et al.* (2023), the significant effect of bioherbicide extracts on root length is related to the respiration process. Respiration is a catabolic process that breaks down plant food reserves, transforming glucose into ATP, which is necessary for plant growth. After plants absorb water, their roots become permeable, allowing oxygen to be absorbed. Oxygen plays a crucial role in cellular respiration, particularly by supporting the breakdown of glucose, which is essential for root growth and development. Nevertheless, allelochemicals in biopesticides are thought to inhibit root respiration, thereby negatively affecting root development in oil palm seedlings.

According to Yuliani *et al.* (2018), root exudates contain various chemical compounds released by plant roots, mainly derived from benzoic acid, cinnamic acid, and phenolic acid. Several allelochemical compounds, such as terpenoids, flavonoids, and phenols, can inhibit cell division, thereby disrupting plant growth. This inhibition mechanism occurs, among other things, by disrupting α -ketoglutarate synthesis, an important precursor for amino acid, protein, and ATP biosynthesis. This results in suboptimal cell division and enlargement, ultimately inhibiting overall morphological development in plants (Susanti *et al.*, 2020).

The concentration of bioherbicides can reduce plant wet weight by disrupting the cell membrane structure. This disruption is caused by phenolic compounds, which damage the phosphate groups in the phospholipids of the cell membrane. The breakdown of these phospholipids produces glycerol (C₃H₈O₃), H₂O, and H₃PO₄. According to Susanti *et al.* (2020), allelopathy further inhibits cell

division and tissue elongation, resulting in fewer leaves and reduced plant height, ultimately decreasing plant fresh weight.

This is reinforced by Tazri *et al.* (2025), who found that allelopathy from taro tubers reduces the number of prickly spinach leaves but does not affect corn leaves. Treatment with the ketapang fruit as a bioherbicide does not inhibit seed germination but does inhibit corn growth. High rainfall during the study caused this effect. High rainfall lowers soil pH. Karamina *et al.* (2017) state that nutrients are best absorbed by plants at pH 6-7 because they are more water-soluble at that range. After application, the soil pH decreased to 5.5, so plants could not fully absorb nutrients. High rainfall can also lower soil temperature and increase soil moisture, causing micronutrients to move to lower elevations (Karamina *et al.*, 2017).

CONCLUSION

The bioherbicide extracted from the Ketapang fruit (*Terminalia catappa*) contains seven compounds: alkaloids (Wagner, Mayer, and Dragendorf reagents), flavonoids, steroids, tannins, phenolics, saponins, and triterpenoids. The bioherbicide derived from Ketapang fruit (*Terminalia catappa*) extract affects the germination of sensitive plant seeds. But not affect the germination of corn (*Zea mays*) seeds in terms of germination capacity and vigor index. Regarding growth parameters, the Ketapang fruit bioherbicide affects plant height, leaf number, root length, and fresh weight of corn (*Zea mays*) and sensitive plants. The highest suppression of seed germination and plant growth parameters was observed at a concentration of 5%. Plant species influenced plant height and number of leaves but did not affect germination rate, vigor index, root length, and fresh weight in corn (*Zea mays*) and sensitive plant (*Mimosa pudica*). Plant height showed an inhibition rate of 49.1% in corn and 75.84% in sensitive plants. For the number of leaves, an inhibitory effect of 15% was observed in corn plants and 62.5% in sensitive plants. For root length, an inhibitory effect of 57.1% was observed in corn and 78.1% in sensitive plants. For the fresh weight parameter, inhibition rates of 49.1% in corn plants and 75.84% in sensitive plants were observed. Plant type influenced plant height and number of leaves but did not affect germination rate, vigor index, root length, and fresh weight in corn (*Zea mays*) and sensitive plants (*Mimosa pudica*). There was an interaction between the bioherbicide derived from the ketapang fruit (*Terminalia catappa*) and plant type on the height of mimosa, the number of leaves in corn and mimosa, and the root length of mimosa, but no interaction was observed for these parameters.

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CONFLICT OF INTEREST

There is no conflict of interest.

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