

Analysis of Allelopathic Content in *Acacia auriculiformis* and *Melia azadirachta* on Mung Bean Germination

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Abstract. This study aims to analyze the effect of allelopathy from leaf extracts of *Acacia auriculiformis* and *Melia azadirachta* on the germination of mung beans (*Vigna radiata*). Allelopathy is a biological phenomenon in which chemical compounds released by certain plants affect the growth and development of other plants. This study used an experimental method with various concentrations of leaf extracts of the two tree species (0%, 10%, 20%, and 40%). The parameters observed included germination percentage, growth rate, root length, and mung bean bud length. The results showed that the increase in the concentration of *Acacia auriculiformis* and *Melia azadirachta* leaf extracts significantly inhibited the germination process and early growth of mung beans. Allelopathic compounds such as flavonoids, tannins, and total phenols are thought to be the main cause of the barrier. Growth and development in acacia leaf extraction (*Acacia auriculiformis*) showed that the growth and development of green bean seeds experienced rapid growth, even exceeding growth in the control treatment. The difference in concentration in the analysis of further tests of neem leaf extraction and acacia gave a quite significant effect, in acacia leaf extraction there was no major effect on germination, but in neem leaf extraction on green beans which was given a negative effect on germination which at a concentration of 40% was much slower compared to other concentrations.

Keywords: allelopathy, *Acacia auriculiformis*, *Melia azadirachta*, mung beans, germination

INTRODUCTION

Competition in plant populations is something that is common in their growing habitats, the occurrence of this competition causes a natural selection process that makes plants that are able to survive can grow and develop well until an ecosystem becomes a climax (*steady state*) (Aarssen, 1983; Werner, 1976). One form of competition/competition response that often occurs in the habitat of this population is that plants can produce allelopathy (Foy, 2024; Poljuha et al., 2024).

Allelopathy is a chemical compound produced by plants in secondary metabolite mechanisms in the form of phenols, flavonoids, and terpenoids (Bachheti et al., 2020; Rahaman et al., 2022). Allelopathy can function as a defense system against disruptive biotic factors and can also be a factor that inhibits the growth and development of surrounding plants due to nutrient competition (Choudhary et al., 2023; Shan et al., 2023). The amount and chemical compounds of allelopathy in each plant vary, allelopathy in plants can be found in leaves, stems, roots, seeds, rhizomas, flowers and fruits. Allopathic compounds can basically have a negative effect on other species, namely:

affecting abnormal seed germination, inhibited root growth, changes in root cell arrangement, inhibiting cell division, inhibiting photosynthesis activity, affecting respiration, affecting protein synthesis, affecting membrane tension, inhibiting enzyme activity, affecting plant succession, inhibiting nitrogen fixation, and nitrification, inhibiting dispersal patterns plants, inhibiting seed decay and germination (Smith-Ramesh, 2020; Hickman et al., 2021; Kong et al., 2024).

Leaves and roots are important sources of allelochemical producers which are compounds that cause allelopathy in plants which are not always abundant but are very important for the survival of plant life (Hierro & Callaway, 2021; Tlak Gajger & Dar, 2021). Root exudate plays an active role in regulating symbiosis and plant protection against microorganisms. In allelopathic interactions, donor plants use secondary metabolites that the roots expel into the rhizosphere to interfere with the growth and development of other plants in the vicinity (Thakre, 2020; Sharma, 2022; Fite et al., 2023).

To be able to observe and study the influence of allelopathic content on other plants, it is very necessary to conduct research using *Acacia auriculiformis* leaf samples and *Melia azadirachta* leaves as a comparison and aquades water as a control for the germination of mung bean seeds, the selection of mung beans as a sample germination due to the growth rate of green beans is relatively very fast and easy. The information from this study can later be an initial reference for environmental conservation management to take policies in conservation wisely without affecting and negatively impacting the surrounding plants of *Acacia auriculiformis* and *Melia azadirachta* in forest reforestation which can affect the distribution of other plants at the bottom of the canopy.

The purpose of this study was to determine the effect of leaf extraction of *Acacia auriculiformis* and *Melia azadirachta* at concentrations of 10%, 20% and 40% on the germination of mung bean seeds for 10 days with 5 mL watering at each replicate, so that it can be observed that the influence of the leaf type has a significant effect and what concentration has the greatest influence on the germination of the mung bean seeds.

RESEARCH METHODS

Time and Place

This research was conducted on March 13, 2024 at the Bio-2 Laboratory of the Department of Biology, Faculty of Mathematics and Natural Sciences, Bogor Agricultural University, Dramaga, Bogor Regency, West Java.

Tools and Materials

The tools used in this study were petri dishes, measuring cups, filter paper, erlenmayers, blenders, filter funnels, buckets, droppers, scissors, refrigerators, analytical scales, filter tools and cellphone cameras. Meanwhile, the ingredients used are mindi leaves (*Melia azadirachta*), acacia leaves (*Acacia auriculiformis*), quades, and mung bean seeds.

Procedures

This research began by cutting mindi leaves (*Melia azadirachta*) and acacia leaves (*Acacia auriculiformis*). To make it smaller to make it easier to blend, each leaf is then weighed as much as 150 grams, after which it is poured into a blender and mixed with 1000 mL of aquades water. After that, each leaf that has been blended with a strainer and the final filtration is done with filter paper to separate the extraction liquid from the sediment. After that, each attraction is stored in the refrigerator for 24 hours. The next stage is the dilution of the extraction solution with a concentration of 10%, 20%, 40, and aquades water as a plant control of 21 petri dishes were prepared, each

concentration was done 3 times by placing filter paper on the base of the petri dish as a substitute for cotton. After that, 10 good quality green beans are placed in each petri dish with watering for 10 days at each different concentration and with the same treatment, namely watering every day as much as 5 mL in each petri dish on 3 repeats. Each watering is documented and observes and counts live and dead peanut seeds every day. Each day the results of the effect of allelopathy on each concentration of mindi leaves were observed and compared (*Melia azadirachta*), and acasia (*Acacia auriculiformis*).

RESULTS AND DISCUSSION

Allelopathy research is one of the studies that has been widely conducted to see the influence of allelopathy on the influence of the germination of other plants, in this study mung bean sprouts were used with 3 replicates at each concentration of mindi leaf extract (*Melia azadirachta*), and acacia leaf (*Acacia auriculiformis*). The first stage is leaf extraction with concentrations of 10%, 20% and 40% using aquades, the results of the calculation of the formula to form 100 dilution solutions, namely in 10% extract with 10 mL of extract added 90 mL of aquades, in 20% of extract with a ratio of 20 mL of extract added 80 mL of aquades, while for 40% with a ratio of 40 mL of extract added 60 mL of aquades.

Observations were made every day from day zero to day seven by measuring the growth of epicot length (area above the cotyledon), hypocotile (the part of the sprout below the cotyledon) and observing the number of dead sprouts each day at each treatment and repeat. Based on the observation of the increase in epicot and hypocotyl length in the sprouts in the two treatments, it appears that the seeds treated with aquades drip as a control experience faster growth than the seeds dripped with mindi leaf extract (*Melia azadirachta*). The effect of watering treatment with mindi leaf extract clearly shows that sprout growth will be inflamed when exposed to allelopathic compounds from other plants.

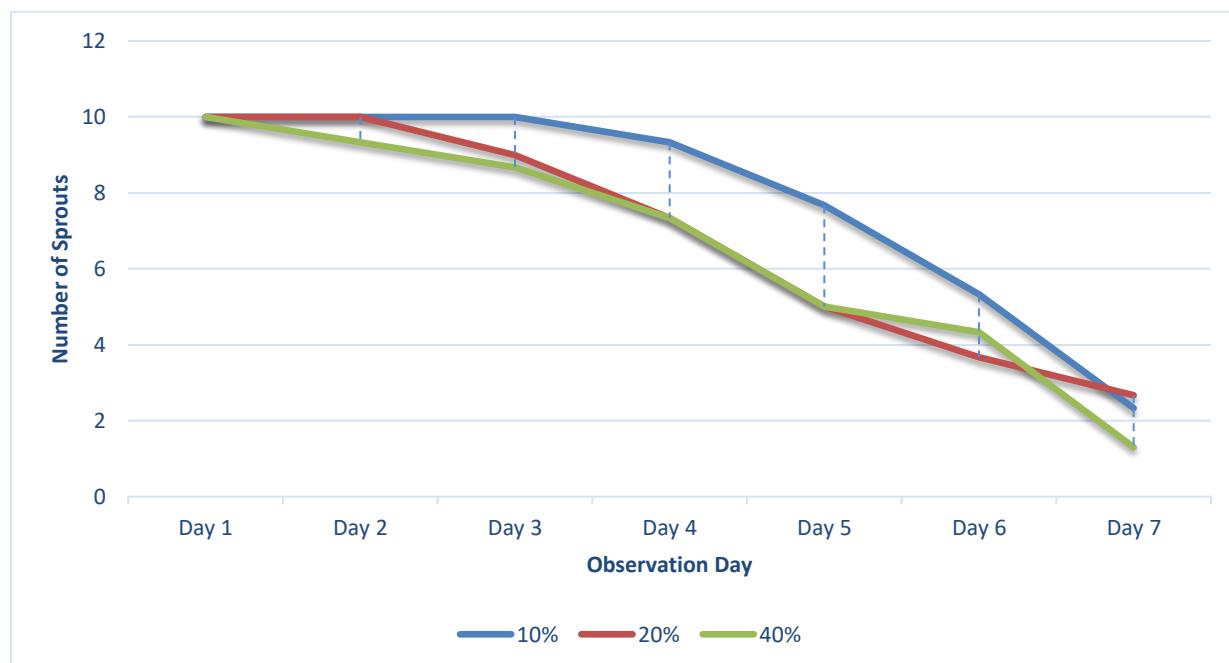


Figure 1. Graph of mung bean death on *Melia azedarac* leaf extraction.

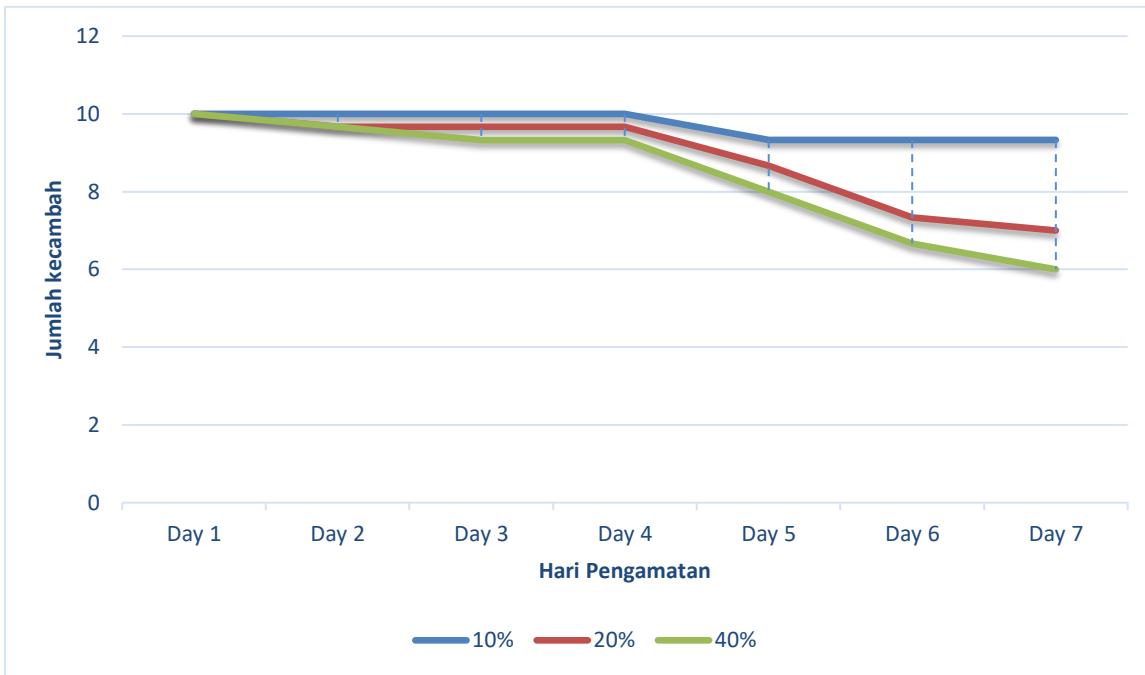


Figure 2. Graph of mung bean death on *Acacia auriculiformis* leaf extraction.

The presence of allelopathic compounds in the extract Mindi leaves and acacia leaf extraction causing the growth of the plant to be inhibited so that the graph decreases. Allelopathy is a compound produced by plants which is usually in the form of phenol and patented. The administration of allelopathic compounds can inhibit germination growth (in leaf treatment). This can be seen in the leaf treatment which shows very slow growth and is increasingly inhibited as the concentration of compounds increases. Allelopathy. The mechanism of allelochemical influence (especially the inhibiting one) on the growth and development of the target organism (especially the plant) is through a series of fairly complex processes, the process begins in the plasma membrane with the occurrence of structural chaos, modification of membrane channels, or loss of function of the ATP-ase enzyme. This will affect the absorption and concentration of ions and water which then affects the opening of the stomata and the process of photosynthesis (Driesen et al., 2020; Kimura et al., 2020; Guo et al., 2023). At certain concentrations of allelopathic compounds can inhibit and reduce the plant's main processes. Obstacles are for example in the formation of nucleic acids, proteins, and ATP. The reduced amount of ATP can suppress almost all cell metabolic processes, so that the synthesis of other substances needed by plants will be reduced (Marchiosi et al., 2020; Efenberger-Szmechtyk et al., 2021). Subsequent inhibitions occur in the synthesis of proteins, pigments and other carbon compounds, as well as the activity of several phytohormones (Carlos et al., 2021; EL Sabagh et al., 2022). Some or all of these obstacles then lead to disruption of cell division and enlargement which ultimately hinders plant growth and development (Lundgren & Fleming, 2020; Pedroza-Garcia et al., 2022).

The organ-forming organs and types of allelochemicals are specific to each species. In general, allelochemicals are secondary metabolites that are grouped into 14 groups, namely water-soluble organic acids, lactones, long-chain fatty acids, quinones, terpenoids, flavonoids, tannins, cinnamic acid and its derivatives, benzoic acid and its derivatives, coumarins, phenols and phenolic acids, nonprotein amino acids, sulfides, and nucleosides (Singh et al., 2021; Khamare et al., 2022). The release of allelochemicals generally occurs at a certain stage of development and the level is influenced by biotic and abiotic stress (Stout, 2014; Harvey, 2015).

Contents of Acacia (*Acacia auriculiformis*)

Allelopathy is certainly beneficial for the species that produces it, but detrimental to the target plant. Therefore, plants that produce allelochemicals generally dominate certain areas, so that the residential population is generally a population of allelochemical-producing plant species. With this interaction process, nutrient and water absorption can be concentrated in allelochemical-producing plants and certain plants that are tolerant of these compounds. Allelopathic compounds exert effects that prevent species from sprouting and newly grown plants, possibly linked to the availability of *Acacia auriculiformis* leaf allelopathic compounds containing tanus, wax, flavonoide and phenolic acids. Furthermore, toxicity has a synergistic effect and phenolic acid has shown toxic effects on the process of budding and plant growth (Chattopadhyay et al., 2021; Oliveira et al., 2021; Misra et al., 2023).

Contains Allelopathy Mindi (*Melia azadirachta*)

The content of active substances contained in the mindi plant is azadirachtin, salannin, and nimbin, which are mainly found in the leaves and seeds of the mindi plant. Azadirachtin is believed to have killing power against pests. Mindy leaves and seeds contain various chemical compounds such as phenols, quinones, alkaloids, and other nitrogen substances, acidic acids and terpenes. Compounds believed to be bioactive ingredients of plant-based pesticides are nimbin, thioneemon, meliantriol, azadirachtin, and silanenine, which are chemical compounds of the terpene group. In addition to containing these compounds, mindi also contains high protein up to 15% and low fiber. Mindi plant waste contains nitrogen, phosphorus, and potassium.

The condition of plant cells due to allelopathy makes the cells inelastic so that they inhibit the flow of water and dissolved nutrients through the cell membrane. Another disturbance of high concentrations is the degradation of enzymes from the cell wall, so that the activity of enzymes becomes inhibited or may not function. Inhibition of enzyme function in seeds causes the growth energy produced during the germination process to be very little and slow, so that the germination process decreases or even the seeds cannot germinate (Kermode, 1990; Jimenez-Lopez, 2017).

In addition, the bark and bark of mindi root contain toosendanin ($C_3H_{38}O_{11}$) and soluble components ($C_{30}H_{40}O_{12}$). In addition, there are also azaridine (margosine) alkaloids, kaempferol, resin, tannins, n-triacontane, β -sitosterol, and triterpene culinone. The bark of the root is toxic compared to the bark while the seeds contain a highly toxic resin, 60% fatty oil is composed of stearic acid, palmitate, oleic, linoleic, laurate, valerianate, butyrate, and a small amount of sulfur essential oil. The fruit contains sterols, catechols, vanillic acid, and bacyanic acid. The leaves contain paraisine alkaloids, routine flavonoids, bitter substances, saponins, flavonoids, tannins, stenoids, and kaemphenols.

Statistical Analysis of Experiments

To be able to scientifically prove the observation of the effect of allelopathy on the extraction of mindi leaves and acacia leaves, statistical testing must be carried out so that the significance of the influence of allelopathy on germination in mung beans can be known with several treatments, namely differences in concentration. In this study, a complete two-factor random design was used. These factors consist of leaf type and leaf dilution concentration. The first factor consists of AI (*Melia azadirachta*) and AC (*Acacia auculiformis*). While the second factor is the level of concentration which consists of 4, namely K0, K10, K20, K40. The experiment used 3 repetitions with a total of 24 experimental units (Table 1).

Table 1 can be seen there was a decrease in the percentage of mung bean germination in different leaf extractions with different concentrations as treatments. The pattern formed from the observation results is that there is a decrease in the percentage of mung bean survival rate, this

decrease is greater in the extraction of mindi leaves than acacia leaves, this is because the allelopathy rate of mindi leaves is stronger than that of acacia leaves, the difference is when we compare the growth rate of acacia leaves with the control (water aquades), there is a growth rate of mung bean germination that is at least similar. This is suspected because the extraction from acacia leaves using a blender and the filtration is not effective so it is suspected that the allelopathic content in acacia leaves is not optimal when extracted.

Table 1. Percentage Of Germination Of Green Beans

Species	Deuteronomy	Concentration			
		K0%	K10%	K20%	K40%
<i>Melia azadirachta (MA)</i>	1	93.333	77.143	72.857	70.000
	2	83.333	81.429	65.714	65.714
	3	100.000	75.714	65.714	61.429
<i>Acasia aculeiformis (AC)</i>	1	93.333	100.000	80.000	77.143
	2	83.333	91.429	98.571	90.000
	3	100.000	100.000	87.143	85.714

Data analysis using SPSS Software version 21, with analysis using the Anova test, several observation factors were seen to be able to determine the influence of allelopathy on objects from observations on mung bean germination. The results of statistical analysis are presented in the Table 2.

Table 2. Anova Test Results

Tests of Between-Subjects Effects					
Dependent Variable: Growth percentage	Source	Type III Sum of Squares	Df	Mean Square	F
	Corrected Model	2841.383a	7	405.912	9.549
	Intercept	166507.974	1	166507.974	3917.13
	Leaf Type	1265.646	1	1265.646	29.775
	Concentration	1150.907	3	383.636	9.025
	Leaf Type * Concentration	424.830	3	141.610	3.331
	Error	680.121	16	42.508	.000
	Total	170029.478	24		
a. R Squared = .807 (Adjusted R Squared = .722)	Corrected Total	3521.504	23		

From the results of this study, it is also seen that the difference in concentration has a fairly obvious effect, mung beans that are given a concentration on the extraction of mindi leaves and acacia, this can be seen in the observation of watering mindi leaf extraction with a concentration of 40% germination much slower than others. The allelopathic effect of the plant *A. indica* It has a negative influence in general like other allelopathic contained in plants, this can be seen from the results of watering which proves that there is no significant effect of disturbing changes from the extraction of acacia leaves, it is possible that this occurs because the percentage of acacia leaf extraction is carried out in a blender so that the allelopathic factor is not eroded optimally, this is

different from the extraction of mindi leaves which is entirely from the results of watering the green bean sprouts. It has a significant influence that can be evidenced by the large amount of decay and death of mung bean sprouts. Inhibition can occur in cell division, seed germination, nutrient uptake, photosynthesis and other enzymatic processes so that it can interfere with the growth of mung bean germination which on the 7th day causes sprout rot and death.

CONCLUSION

From the results of allelopathic research that uses the extrusion of mindi leaves (*Melia azadirachta*) and acacia leaves (*Acacia auriculiformis*) in each spraying, it can be concluded that This study shows that there are different allelopathic effects of mindi leaf extract (*Melia azadirachta*) and acacia leaves (*Acacia auriculiformis*) on mung bean seed germination. Mindi leaf extract has a significant negative effect on germination, especially at high concentrations (40%), where almost all sprouts die in the first 1-3 days. In contrast, acacia leaf extract showed surprising results by increasing sprout growth even exceeding the control, although this finding still requires further research to validate the results. This different response pattern is clearly seen in tests with various concentrations (10%, 20%, and 40%), where the higher the concentration of mindi leaf extract, the stronger the inhibitory effect on germination. This finding confirms that allelopathic compounds can have varying effects on mung bean seed samples.

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