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Potential of *Terminalia catappa* Leaf Extract as Bioherbicide on *Mikania micrantha* Seed Germination

Potensi Ekstrak Daun Ketapang sebagai Bioherbisida pada Perkecambahan Benih Sembung Rambat

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ABSTRACT

Mikania micrantha is a weed that is commonly found on agricultural land. Environmentally friendly weed control can be done using bioherbicides because the materials used come from nature. *Terminalia catappa* contains secondary metabolites such as flavonoid, tannin, alkaloids, terpenoids and saponins, so that its bioactive compounds can be used as weed control. This study aims to determine the total secondary metabolite compounds of *Terminalia catappa* leaves and their effect on *Mikania micrantha* seed germination. This study used a factorial Completely Randomized Design (CRD). The first factor is various types of solvents aquadest, ethanol and methanol. The second factor was the concentration of the extract application 30%, 50%, 70% and repeated 3 times so that there were 27 experimental units. Data were analyzed using analysis of variance if it had a significant effect, further tests were carried out using 5% HSD (Honestly Significant Difference). The results showed that the Terminalia catappa leaf extract with distilled aquadest of 70% was able to prevent the seeds from germinating while the methanol and ethanol solvent extracts were able to inhibit the germination of *Mikania micrantha* seeds at an application concentration of 30%.

Keywords*: *Bioherbicide, Terminalia catappa, extract, Mikania micrantha.

ABSTRAK

Sembung rambat (*Mikania micrantha*) merupakan gulma yang banyak ditemukan pada lahan pertanian. Pengendalian gulma ramah lingkungan dapat dilakukan menggunakan bioherbisida karena bahan yang digunakan berasal dari alam. Ketapang mengandung senyawa metabolit sekunder seperti flavonoid, tanin, alkaloid, terpenoid dan saponin sehingga senyawa bioaktifnya dapat digunakan sebagai bioherbisida pengendali gulma. Penelitian ini bertujuan untuk mengetahui kadar total senyawa metabolit sekunder daun ketapang serta pengaruhnya terhadap perkecambahan benih sembung rambat. Penelitian ini menggunakan Rancangan Acak Lengkap (RAL) Faktorial. Faktor pertama adalah berbagai jenis pelarut yaitu aquades, etanol dan metanol. Faktor kedua adalah konsentrasi aplikasi ekstrak yaitu 30%, 50%, 70% dan diulang sebanyak 3 kali sehingga terdapat 27 unit percobaan. Data dianalisis menggunakan Analisis ragam (ANOVA) jika berpengaruh nyata dilakukan uji lanjut menggunakan HSD 5%. Hasil penelitian memperlihatkan bahwa ekstrak daun ketapang pelarut aquades konsentrasi 70% mampu membuat benih tidak berkecambah sedangkan ekstrak daun ketapang pelarut metanol dan etanol mampu menghambat perkecambahan benih sembung rambat pada konsentrasi aplikasi 30%.

Kata kunci: Bioherbisida, ketapang, ekstrak, sembung rambat.

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INTRODUCTION

Mikania micrantha is a weed that grows rapidly and reduces crop production. The decrease in Fresh Fruit Bunches (FFB) productiondue to vines in oil palm (*Elaeis guineensis*) plants reached 20%, rubber (*Hevea brasiliensis*) reached 27-29% and wheat (*Triticum aestivum*) reached 28% (Hamidah *et al.*, 2015).

According to the research results of Day *et al*. (2012) *Mikania micrantha* can flower in cocoa plantations and the yield is lower. The yield loss of banana (*Musa* spp.), *Citrus* spp. and sugarcane (*Saccharum officinarum* L.) productivity reaches 60 - 70% (Shen *et al.*, 2013). Further reducing tea plantation yields by 42% through growth competition, nutrient competition, and disrupting harvesting new leaves by creeping on the entire cultivated plant (Puzari *et al.*, 2010).

Competition for nutrients in the soil can interfere with the main crop productivity. According to Sun *et al*. (2019) the soil taken from around the roots of the *Mikania micrantha* contains *Eucephalobus* bacteria which can increase the potassium content of the soil by stimulating the growth of potassium solubilizing bacteria and increasing activity through biotic interactions.

Bioherbicides are environmentally friendly herbicides derived from plants that contain allelopathy (toxic substances) and can reduce weed growth or kill other plants (Hanifatiha, 2013). According to Susanti *et al*. (2014) showed that the application of bioherbicide leaves of ferns (*Gleichenia linearis*) with a concentration of 30% can reduce the germination of *Mikania micrantha* weeds with a germination percentage of 19% due to the presence of flavonoid compounds and tannins.

Catappa leaf extract is also known to contain allelopathic compounds such as flavonoids and tannins so that these compounds can kill other plants and have the potential to become bioherbicides (Rajesh *et al.*, 2016). The type of extraction solvent greatly affects the number of active compounds contained in the extract, polar compounds will dissolve in polar solvents and non-polar compounds will dissolve in non-polar solvents (Arifianti *et al.*, 2014).

This study aims to determine the flavonoid compound of ketapang leaf extract and its application in controlling sembung rambat weeds in post-growth.

MATERIALS AND METHODS

Place and time

The research was carried out at the Plant Ecophysiology Laboratory and Agricultural Product Technology Laboratory, Faculty of Agriculture, University of Riau. The time of research and data collection was carried out from August to October 2021.

The materials used were catappa leaves obtained from Indragiri Hilir, *Mikania micrantha* weed seeds obtained from Pekanbaru, aquadest, ethanol, methanol, ethyl acetate, 1 N HCl, 0.1 N, NaOH, Na_2CO_3. The tools used in this research include dry blender, vacuum rotary evaporator, sprayer, digital scale, hot plate, handsprayer, filter paper, aluminum foil, beaker glass, petridis. measuring cup, ruler, dropper, spatula, test tube, documentation and stationery. This study consisted of two experiments. The first experiment was to determine the total levels of secondary metabolites of catappa leaf extract using the colorimetric method (Chang *et al.*, 2002). The second trial of seed germination test. This study used a factorial completely randomized design (CRD). The first factor is various types of solvents, namely P1: aquadest, P2: ethanol, P3: methanol. The second factor was the concentration of the extract application, namely E1: 30%, E2: 50%, E3: 70% and repeated 3 times so that 27 experimental units were obtained.

Catappa leaves taken as much as 1000 g then washed with clean water. The dried leaves are then cut into small pieces and crushed until smooth using a blender. The powder is then

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sieved to obtain a fine powder. Then the powder was weighed as much as 200 g and then extracted using a different type of solvent, namely distilled aquadest, 50% ethanol and 1000 ml of 50% methanol until the powder was completely submerged (Riskitavani and Purani 2013). Soaking is carried out at room temperature for up to 24 hours. After 24 hours the meeration results were filtered and the pure ketapang leaf extract was evaporated using a rotary evaporator at a temperature of 480C. after being evaporated, the extract was divided into 3 different concentrations according to the treatment, namely 30%, 50% and 70% by diluting again with distilled aquadest. The research was carried out when the sembung rambat weeds had not yet grown (pre-growing).

There are 9 treatment combinations. Each treatment was repeated 3 times, so that 27 experimental units were obtained. Each experimental unit contains 15 mikania micrantha seeds.The test method used is the test on paper (Aparicio *et al.,* 2021). Three sheets of paper substrate were placed on the petri dish, then the application of ketapang extract was carried out by spraying the extract on the petri dish as much as 3 ml at the beginning of laying the seeds, then the petri dishes that had been treated were closed and placed in the seed germination apparatus.

Re-spraying application is done when the paper looks dry with distilled aquadest. Observation parameters were germination percentage, germination rate, radicle length, plumule length.

RESULT AND DISCUSSION

Comparison of the total levels of each secondary metabolite compound is presented in Table 1.

Table 1. Total secondary metabolite compounds of *terminalia catappa* leaves

Solvent type	Compound	Total content of secondary metabolites
Aquadest	Flavonoid	53.98±0.56 mg QE/g
	Tannin	39.13±0.26 mg TAE /g
Ethanol	Flavonoid	59.85±0.34 mg QE/g
	Tannin	44.49±0.25 mg TAE/g
Methanol	Flavonoid	60.32±0.18 mg QE/g
	Tannin	45.697±0.42 mg TAE/g

Description : QE : Quercetin Equivalent, TAE : Tannin Acid Equivalent

Table 1 shows that *Terminalia catappa* leaves from Indragiri Hilir which were extracted with distilled aquadest contained flavonoid compounds (53.98±0.56 mg QE/g) and tannins (39.13±0.26 mg TAE/g). The results of the total analysis of catappa leaf extract with ethanol solvent had a total value of flavonoid compounds (59.85±0.34 mg QE/g) and tannins (44.49±0.25 mg TAE/g) and *Terminalia catappa* leaf extract with methanol solvent has a higher total value of flavonoids and tannins than aquadest and ethanol solvents.

Total flavonoid compounds and tannins are influenced by the polarity of the solvent which has an important role in increasing the solubility of phenolic compounds (Abaza *et al.*, 2011). The high yield of *Terminalia catappa* leaf extract was shown in methanol solvent, followed by ethanol solvent, while the aquadest solvent showed the lowest yield in total flavonoid and tannin levels. In research observations, *Terminalia catappa* leaf powder soaked with distilled aquadest was light brown and cloudy, the bioactive compounds from leaf powder in the maceration process were not

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completely released, causing low levels of total flavonoid compounds and tannins when extracted with distilled aquadest.

Extract with methanol and ethanol solvent showed a blackish green color. The high total yield of flavonoid and tannin compounds in methanol solvent because the solvent has a different level of electronegativity so that the compound entering the polar solvent will be measured for its level of polarity (dipole moment). Furthermore, through the dynamics of solvation which is the process of ions or molecules of solute compounds surrounded by solvent molecules to bind electrons and spread throughout the solvent (Li *et al.*, 2021).

Germination percentage

The results of observations of the percentage of germination that have been analyzed by variance showed that the application of the solvent type was significantly different to the percentage of weed mortality, while the concentration of the catappa leaf extract was significantly different and the interaction between the type of solvent extract and the concentration of the catappa leaf extract was significantly different to the percentage of weed germination. The results of the 5% HSD test on the percentage of germination parameters are presented in Table 2.

Table 2. Percentage of germination of *Mikania micrantha* weed 10 DAP (Days After Application) *terminalia catappa* leaf extract.

Solvent type	Concentration			Average
	(E1) 30%	(E2) 50%	(E3) 70%	
(P1) Aquadest	22,22[a]	8,88[b]	0[c]	10,36[a]
(P2) Etanol	0[c]	0[c]	0[c]	0[b]
(P3) Metanol	0[c]	0[c]	0[c]	0[b]
Average	7,40[a]	2,96[b]	0[c]	

The numbers in the column and row followed by the same letter show no significant difference according to the 5% level HSD test.

Table 2 shows that no *Mikania micrantha* seeds germinated when treated with *Terminalia catappa* leaf extract with 70% aquadest solvent. the seeds were also unable to germinate when given the leaf extract of *Terminalia catappa* with ethanol and methanol solvents at all experimental concentrations. This is in line with the research results of Mahardhika *et al.* (2016). The use of appropriate solvents such as methanol is very effective in inhibiting *Mimosa pudica* L seeds.

Mikania micrantha seeds were only able to germinate by treating Terminalia catappa leaf extract with aquadest solvent concentration of 30% with a germination percentage of 22.22%, significantly different from treatment with aquadest solvent extract 50% concentration of 8.88%. The germination of weed seeds with distilled aquadest was due to the low application concentration of 30-50% and the low secondary metabolite compounds found in the extract such as flavonoids (53.98±0.56 mg QE/g) and tannins (39.13±0.26 mg TAE/g).

Mikania micrantha weeds did not germinate at 70% aquadest treatment, as well ethanol and methanol treatments at all experimental concentration. According to Zribi et al., (2014) flavone and flavonol compunds are the most toxic compounds and can inhibit seed germination, growth rate and inhibit the synthesis of germination enzymes.

Germination Rate

The results of the observation of the germination rate which were analyzed by variance showed that the application of the solvent type was significantly different to the germination rate, while the concentration of the

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catappa leaf extract was significantly different and the interaction between the type of solvent extract and the concentration of the catappa leaf extract was significantly different to the germination rate. The results of the 5% Tukey test on the germination rate are presented in Table 3.

Table 3. Germination rate of *Mikania micrantha* weed 10 DSA

Solvent type	Concentration			Average
	(E1) 30%	(E2) 50%	(E3) 70%	
(P1) Aquadest	4,75[a]	2,67[b]	0[c]	2,47[a]
(P2) Etanol	0[c]	0[c]	0[c]	0[b]
(P3) Metanol	0[c]	0[c]	0[c]	0[b]
Average	1,58[a]	0,89[b]	0[c]	

The numbers in the column and row followed by the same letter show no significant difference according to the 5% level HSD test.

Table 3 shows that the interaction between the type of solvent and the concentration of the extract in the P1E1 treatment with a concentration of 30% caused the germination rate of *Mikania micrantha* weed to be 4.75 days, which was significantly different from the P1E2 treatment at 50% concentration, which was 2.67 days and significantly different from other treatments. According to Hanum *et al*. (2007) stated that secondary metabolites in plants have an allelopathic effect that can inhibit the process of ATP formation so that it can reduce germination and cause a slower growth rate. The slower the germination rate, the higher the effectiveness of the bioherbicide (Maiti *et al.*, 2010).

Radicle length

The interaction between the type of solvent extract and the concentration of the extract of catappa leaves was significantly different to the length of the radicle. The results of the 5% HSD test for radicular length are presented in Table 4.

Table 4. Radicle length of *Mikania micrantha* propagation 10 DAP

Solvent type	Concentration			Average
	(E1) 30%	(E2) 50%	(E3) 70%	
(P1) Aquadest	0,32[a]	0,18[b]	0[c]	0,16[a]
(P2) Etanol	0[c]	0[c]	0[c]	0[b]
(P3) Metanol	0[c]	0[c]	0[c]	0[b]
Average	0,10[a]	0,06[b]	0[c]	

The numbers in the column and row followed by the same letter show no significant difference according to the 5% level HSD test.

Table 4 shows that the interaction between the type of solvent and the concentration of the extract in the P1E1 treatment with a concentration of 30% caused the radicle length of *Mikania mcrantha* weed to be 0.32 cm, significantly different from the P1E2 treatment with a concentration of 50%, which was 0.18 cm and significantly different from other treatments.

Radicular growth can be inhibited due to lack of food reserves in the endosperm which results in radicular growth (Ilori *et al.*, 2007). Disruption of metabolic activity caused by allelochemicals can reduce root and shoot

length during germination (Saeid *et al.,* 2010). Based on the research results of Aparicio *et al.* (2021) flavonol compounds contained in the extract of *Fagopyrum esculentum* showed an inhibitory effect on the growth of the radicle of Phelipanche ramose. The sensitivity level of radicle growth in seeds was very large when they were given secondary metabolites, because the seeds have permeable tissue so that the inhibition process was more effective (Motmainna *et al.,* 2021).

Plumule Length

The results of the observation of the length of the plumules which were analyzed by variance showed that the application of the solvent type was significantly different to the length of the plumule, while the concentration of the extract of catappa leaves and the interaction between the type of solvent extract and the concentration of the extract of the catappa leaf was significantly different to the length of the plumule. The results of the 5% HSD test for the length of the plumule are presented in Table 5.

Table 5. Plumule length of *Mikania micrantha* propagation 10 DAP

Solvent type	Concentration			Average
	(E1) 30%	(E2) 50%	(E3) 70%	
(P1) Aquadest	0,60[a]	0,48[b]	0[c]	0,36[a]
(P2) Etanol	0[c]	0[c]	0[c]	0[b]
(P3) Metanol	0[c]	0[c]	0[c]	0[b]
Average	0,19[a]	0,16[a]	0[c]	

The numbers in the column and row followed by the same letter show no significant difference according to the 5% level HSD test.

Table 5 shows that the interaction between the type of solvent and the concentration of the extract in the P1E1 treatment with a concentration of 30% caused the length of the plumule of *mikania micrantha* weeds to be 0.60 cm, significantly different from the P1E2 treatment with a concentration of 50%, which was 0.48 cm and significantly different from other treatments.

Allelochemical compounds such as phenolics, terpenoids, alkaloids and their derivatives can inhibit seed germination. The effects caused by allelochemical compounds can slow down the germination rate, reduce radicle and plumule roots, change color and reduce reproductive capacity (Franco *et al*., 2015). Based on the results of research by Ilori *et al.,* (2010), secondary metabolites of flavonoids and tannins from the leaf extract of kirinyu (*Chromolaena odorata*), sunflower (*Helianthus annus*) and moonflower (*Tithonia diversifolia*) stated a decreasing effect on the germination of cowpea (*Vigna unguiculata*) seeds.

CONCLUSION

Based on the results of the study, it can be concluded that *Terminalia catappa* leaf extract with 70% aquadest solvent was able to prevent *Mikania micrantha* seeds from growing. The administration of *Terminalia catappa* leaf extract with 30% concentration of methanol and ethanol also suppressed seed germination up to 100%.

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