ANALGESIC POWER TEST OF MAHOGANY LEAF EXTRACT (*Swietenia mahagoni*) ON MALE MICE (*Mus musculus*. L)

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ABSTRACT

Mahogany leaves (Swietenia mahagoni) have flavonoids in them that might help with pain by stopping the cyclooxygenase enzyme from working. The purpose of this study is to see if compounds from mahogany leaves (Swietenia mahagoni) can help relieve pain in mice that have been given acetic acid. This study used the chemical stimulation method (writhing). Twenty-five mice were divided into 5 treatment groups, namely group 1 was given Na-CMC as a negative control, group 2 was given paracetamol at a dose of 1.3 mg/20 gBW as a positive control, and groups 3, 4, and 5 were given mahogany leaf extract (Swietenia mahagoni) at a dose of 2.8 mg/20 gBW, 5.6 mg/20 gBW, and 11.2 mg/20 gBW. Each group was given treatment orally; 30 minutes later, the mice were injected with 1% acetic acid intraperitoneally. Additionally, researchers observed and counted the number of wriggles every 15 minutes for a duration of 90 minutes. This study shows that the ethanol extract of mahogany leaves (Swietenia mahagoni) at a dose of 11.2 mg/20 g BB with a 100% success rate can relieve pain.

Keywords: *Analgesic, mahogany leaves, Swietenia mahagoni, ethanol extract*

INTRODUCTION

Indonesia, blessed with endless natural wealth, boasts the second largest biodiversity in the world after Brazil. There are approximately 30,000 types of plants, of which 2,500 are medicinal. Many experts from various countries, such as Germany, India, China, and Australia, conduct research and testing with various plants that are traditionally used by the community to cure various diseases (Ramadhiani et al., 2022).

Mahogany plants (*Swietenia macrophylla*) are widely planted as shade trees because of their heat-resistant nature and excellent adaptability to various soil conditions, so they continue to decorate roadsides in several areas. The Java region initially developed this plant during the Dutch colonial era. Mahogany is a type of wood that is easy to cultivate because it can grow in various places and in different types of soil (Alfayed et al., 2022).

According to previous research, hibiscus leaves (*Hibiscus rosasinensis*) are effective in providing analgesic effects on male mice (*Mus musculus* L.). Based on the phytochemical approach, hibiscus leaves are thought to contain flavonoid compounds that can ease pain by blocking the activity of the cyclooxygenase enzyme. This means that the leaves are thought to have an analgesic effect (Yustisi & Rahmawati, 2019).

Based on previous research, research has been conducted on mahogany seeds as antidiabetic, mahogany bark as antihyperlipidemic (Isdadiyanto et al., 2024), and mahogany leaves as biolarvacide (Rizkika Nur Amelia et al., 2017). However, there has been no research on mahogany leaves as analgesics. Therefore, researchers are interested in conducting research on the analgesic power test of mahogany leaf extract (*Swietenia mahagoni*) in male mice (*Mus musculus* L.).

METHODE

Some of the things that were used in this research were mahogany leaves, 1% acetic acid, 0.5% CMC-Na, 70% ethanol, distilled water, 500 mg of paracetamol, aluminum foil, parchment paper, $FeCl_3$, concentrated HCl, 2 N HCl, the Mayer reagent, the Dragendrof reagent, and H_2SO_4.

Mahogany leaf extraction: We use the maceration method to extract mahogany leaves. The maceration method involves mixing fine extraction materials with other extraction materials. The advantages of the maceration method are that it is straightforward to do and the tools used are simpler. We use maceration for simplicia extracts, which contain active substances easily dissolved in the solvent liquid. You can use solvent liquids such as water, ethanol, water-ethanol, or other combinations. We carry out the maceration extraction method three times in a 24-hour period to attract the compounds present in the herbs.

The analgesic testing method: The chemical stimulation method is used as a method for testing analgesic effects because this method is simple, straightforward to do, and sensitive enough for testing compounds that have weak analgesic power. Giving analgesics will reduce pain so that the amount of writhing in a certain period of time will decrease. Mice express their pain through a response. The writhing movement is both pairs of mouse legs moving forward and backward and the stomach pressing the floor, which appears within five minutes after induction.

Phytochemical Screening Test:

Flavonoid Test

We put 1 gram of sample extract into a test tube, added concentrated HCl, and heated it on a water bath for 15 minutes. If a red or yellow color is formed, it means it is positive for flavonoids.

Terpenoid Test: 2 grams of sample extract were put into a test tube, then added with 2 ml of ethyl acetate and shaken; the ethyl acetate layer was taken and then dripped on a dropper plate and left to dry. Following the drying process, we added 2 drops of anhydrous acetic acid and 1 drop of concentrated sulfuric acid. It indicates a positive result for terpenoids if a red or yellow color forms.

Alkaloid Test: We evaporated 2 ml of the test solution over a porcelain cup. The resulting residue was then dissolved with 5 ml of 2 N HCl; the solution obtained was divided into 3 test tubes. We added 3 drops of 2 N HCl to the first tube, which served as a blank. The second tube was added with 3 drops of Dragendorff's reagent, and the third tube was added with 3 drops of Mayer's reagent. The

formation of orange sediment in the second tube and yellow sediment in the third tube indicates the presence of alkaloids.

Saponin Test: Add 100 mg of plant powder and 10 ml of distilled water to a test tube. Cover and shake vigorously for 30 minutes. If foam appears approximately 3 cm from the surface, it indicates the presence of saponins.

Tannin Test: The test is carried out by taking 2 mL of each mahogany leaf sample that has been extracted with water and ethanol solvents, then heated for approximately 5 minutes. After heating, several drops of 1% $FeCl_3$ are added to each. Each solution is confirmed to contain tannins if it turns greenish-brown or blackish-blue.

Analgesic Test: We divided the test animals into 5 groups, each consisting of 5 test animals. Each group was given oral treatment with a specified dose.

Group I: 0.5% Na-CMC solution (as a negative control)

Group II: Given paracetamol (as a positive control) with a dose of 1.3 mg/20 gBW suspended in 0.5% Na-CMC.

Group III: Ethanol extract of mahogany leaves (2.8 mg/20 g BW)

Group IV: Ethanol extract of mahogany leaves (5.6 mg/20 gBW)

Group V: Ethanol extract of mahogany leaves (11.2 mg/20 g BW)

Following a single oral treatment dose, we administered 1% acetic acid as a pain stimulant to the mice 15 minutes later. We then observed and counted the characteristic writhing every 15 minutes for a total of 90 minutes.

The calculation of analgesic power in each group can be determined after obtaining the number of mice writhing for 90 minutes by calculating the percentage of analgesic power of each group with the formula:

$$\%DA = 100\% - (P/K \times 100\%)$$

Description:

DA : Analgesic Power

P :Cumulative number of wriggles in mice given analgesic drugs.

K :Cumulative number of wriggles in mice given 1% acetic acid.

RESULT AND DISCUSSION

We conducted this study to test the analgesic activity of mahogany leaf compounds (*Swietenia mahagoni*) on mice. Mahogany leaf extract was made by macerating the leaves with

70% ethanol. The goal is to get the chemical compounds that make up mahogany leaves. Ethanol can dissolve in both nonpolar and polar solvents; it is also semipolar and has the lowest toxicity (Pudjiastuti & Nugroho, 2009).

Alkaloids, flavonoids, saponins, and tannins are chemical parts of mahogany leaves that have been tested phytochemically before. These results are similar to those from earlier studies that showed that mahogany leaf extract has active compounds like alkaloids, flavonoids, saponins, and tannins.

The analgesic effect is caused by the compound components contained in mahogany leaves, namely alkaloids, flavonoids, saponins, and tannins. Pain is lessened by flavonoids because they stop the cyclooxygenase enzyme from working by stopping arachidonic acid from making prostaglandins. Also, flavonoids stop neutrophils from releasing their outer layer, which stops the release of cytokines, free radicals, and enzymes that cause inflammation (Sasongko et al., 2016). Alkaloids stop important steps in prostaglandin biosynthesis, specifically the cyclooxygenase pathway. Tannins work by encouraging the release of the lipomodulin enzyme, which stops the phospholipase enzyme. This stops the cyclooxygenase and lipoxygenase pathways and their byproducts from being made.

Table 1. Phytochemical Screening

Compounds	Result
Flavonoids	+
Terpenoids	-
Alkaloids	+
	+
Tannins	+
Saponins	+

In this study, male white mice were used as experimental animals because they are easy to handle and have physiological similarities to humans. Male mice were selected because female mice can experience fluctuation cycles from time to time due to hormone cycles that can affect research results (Suwarni et al., 2016).

The method used is the chemical stimulation method; this method is used as a method for testing analgesic effects because this method is simple, straightforward to do, and sensitive enough to test compounds that have

weak analgesic power. Giving analgesics will reduce pain so that the amount of writhing in a certain period of time will decrease; this pain in mice is observed in the form of a writhing movement response, namely, the abdomen touches the base of the footrest, and both pairs of legs are pulled back, and the stomach presses the floor.

In the chemical stimulation method, a substance is said to have an analgesic effect if it can stop writhing by at least 50% of the time. For minimal analgesic effects, the amount of writhing that is stopped by less than 70% is considered minimal. Tables 2 and 3 below display the cumulative number of mice wriggling every 15 minutes for 90 minutes.

Showing that paracetamol treatment and doses I, II, and III of mahogany leaf extract can reduce the wriggling of mice by more than 50% compared to negative controls, this means that paracetamol treatment and ethanol extract at doses of 2.8 mg/20 gBB, 5.6 mg/20 gBB, and 11.2 mg/20 gBB have an analgesic effect. These doses have an analgesic effect. A drug is said to have analgesic activity if it is able to reduce the number of mice wriggling by 50% of the number of wriggling in the negative control group. We chose acetic acid as a pain inducer because it triggers a local acute inflammatory reaction that produces pain. This reaction releases arachidonic acid from phospholipid tissue through the cyclooxygenase pathway. It also makes prostaglandins in the intraperitoneal fluid, which makes mice move around.

The wriggling response is characterized by abdominal muscle contractions, the stomach touching the floor, pulling the hind legs, bending the head, and twisting the back (Febri et al., 2007). Furthermore, we observed the number of wriggles produced every 15 minutes for 90 minutes. We averaged each group's wriggles and compared them between the treatment group and the control group. The average number of wriggles that was less than the control group indicated analgesic activity in the mice. Table 2 displays the average number of wriggles observed in each group.

Table 2. Number of wrigglings every 15 minutes for 90 minutes

Group	Minute						Average
	15	30	45	60	75	90	± SD
Negative control	29,2 ±13,14	39,8 ±13,7	41,8 ±7,7	27,6 ±8,41	20,6 ±8,3	21,6 ±10,5	30,1 ± 10,29
Positive control	13,6 ±,.5	10,6 ±2,3	7 ±3,08	6,6 ±1,14	5,2 ±1,3	4,6 ±2,3	7,93 ± 2,11
Dose 1	18 ±7,31	15,2 ±4,3	12,6 ±4,4	1,4 ±5,18	8,4 ±3,8	6,4 ±2,7	12,33 ± 4,62
Dose 2	10,6 ±1,4	10,4 ±2,8	10,2 ±2,8	8,8 ±1,92	6,2 ±3,5	3,8 ±1,6	8,33 ± 2,74
Dose 3	14,8 ±1,3	8,6 ±1,14	7,8 ±1,8	6,6 ±2,61	5,8 ±1,9	2,8 ±0,8	7,73 ± 1,55

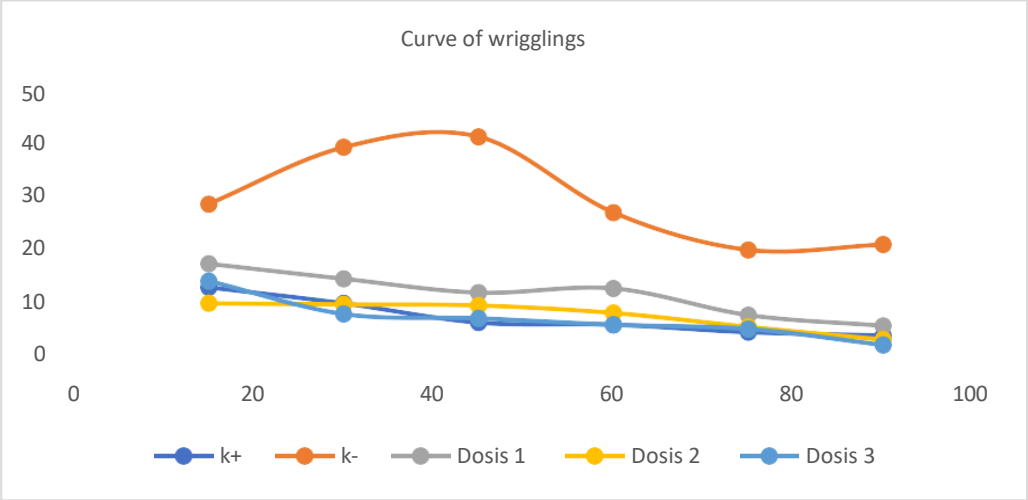

Figure 1. Average Curve of Number of Wriggles Every 15 Minutes for 90 Minutes

Each of the three groups that received different amounts of mahogany leaf compound and the positive control group (paracetamol 1.3 mg/20 gBW) had a different average number of wriggles. The negative group had 30.1, while the positive control group had 7.93. This evidence indicates an analgesic effect of mahogany leaf compounds and paracetamol.

The group that produced the least average number of wriggles was the positive control group, while of the three doses of mahogany leaf extract, the best average number of wriggles was dose 3 (dose 11.2 mg/20 gBW). The lower the average number of wriggles produced, the better the analgesic effect is. Once the percentage of protection was calculated, the results can be seen in Table 3. They show what percentage of protection each group had against acetic acid pain induction.

Table 3. Results of Calculation of % Protection of Writhing Against the Amount of Acetic Acid Pain

Group	% Protection
Negative control	0 %
Positifve control	99,73 %
Dose 1	99,59 %
Dose 2	99,72 %
Dose 3	99,74 %

In the table, the negative control group's protection percentage is zero because it's a comparison. In the test group, the higher the dose, the greater the percentage of protection. We conduct statistical analysis to determine if there is a significant difference between the treatment groups. An ANOVA test with a 95% confidence level is used for statistical analysis because the sample population is spread out and has the same amount of variation across all individuals. Table 6 displays the results of the ANOVA analysis. The results of the calculation of the largest % protection are paracetamol; the positive control is greater than the dose of 3.

In this study, three doses were used to see if there was a dose-effect relationship in the results. If a test material has a dose-effect relationship, which means that the effect gets stronger as the dose goes up, then the test material does have the effect that was expected. The ability of mahogany leaf extract provides an analgesic effect at doses of 2.8 mg/20 g BB, 5.6 mg/20 g BB, and 11.2 mg/20 g BB.

The results prove that the higher the concentration of mahogany leaf extract, the more it can provide an analgesic effect. The results of the data

obtained were completely unique. At a dose of 11.2 mg/20 g BB, it turned out to be able to provide a 100% analgesic effect. A study showed that an ethanol extract of eucalyptus leaves (*Melaleuca leucadendron* L.) can help male white mice feel less pain. Ethanol extract of eucalyptus leaves at a dose of 5.12 g/kg BW has a percentage of protection that is not significantly different from paracetamol at a dose of 65 mg/kg BW, or in other words, has the same efficacy as paracetamol at a dose of 65 mg/kg BW, while mahogany leaf extract at a dose of 5.6 mg/20 g BW and 11.2 mg/20 g BW has a percentage of protection that is not significantly distinct from paracetamol at a dose of 1.3 mg/20 g BW, with the same content, namely flavonoids, alkaloids, saponins, and tannins.

The results of the statistical analysis using the ANOVA test (0.001 < 0.05) showed that there was a difference in analgesic effectiveness in each treatment. Continued with the LSD test, a significant difference was obtained between the dose of 2.8 mg/20 gBW with the dose of 5.6 mg/20 gBW and 11.2 mg/20 gBW. There was no significant difference in analgesic effectiveness between the dose of 5.6 mg/20 gBW and the dose of 11.2 mg/20 gBW.

CONCLUSION

As a result of the study, we can say that the analgesic effect of mahogany leaf extract on male mice using a chemical stimulation method at a dose of 11.2 mg/20 g BB is greater than that of paracetamol at 1.3 mg/20 g BB.

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