

**IN VIVO TESTING OF ANTIPYRETIC LEAVES OF FENCE
(JATROPHA CURCAS L) ORIGIN GORONTALO**

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Abstract

Jatropha (*Jatropha Curcas L*) is a plant that has been used empirically as a fever reducer. Fever is one of the symptoms of the disease which is characterized by a significant increase in body temperature. The novelty of this study was due to the effectiveness of *Jatropha Curca L*. leaf extract as an antipyretic. The purpose of this study was to determine the dose of antipyretic effectiveness of *Jatropha Curcas* (*Jatropha Curcas L*) Leaf Extract in Male Mice (*Mus Musculus*). Extraction by maceration using 96% ethanol solvent. 15 male mice (*Mus Musculus*) were divided into 5 groups, namely negative control (Na-cmc 1%), positive control (Paracetamol), and the treatment group given *Jatropha* leaf extract with doses of 150, 200, 250 mg/kgBW. Peptone 10% 1 ml orally as a fever inducer. And observed at 30, 60, 90 and 120 minutes after treatment. The data obtained were analyzed using one-way ANNOVA followed by the LSD test to determine differences between groups. The results showed that the leaf extract of *Jatropha Curcas* (*Jatropha Curcas L*) had an antipyretic effect where the most effective dose was at a dose of 250 mg/kgBW because it has a very stable temperature reducing power and effectiveness is comparable to Paracetamol. The conclusion of this study is that *Jatropha Curcas L* extract at a dose of 250 mg/kg can be used as an antipyretic.

Keywords: Antipyretic; *Jatropha* (*Jatropha Curcas L*) Leaf Extract; Flavonoids.

INTRODUCTION

Antipyretics are drugs used to lower body temperature in a feverish state (1). Indonesia has a variety of plant species totaling 30,000 species and 7,000 species which are already considered nutritious plants and have been scientifically tested. Traditional medicine in Indonesia uses ingredients found in the natural surroundings and is part of the nation's culture that has been passed down from generation to generation (2). In 1904, the use of medicinal plants was gradually and systematically abandoned along with the development of modern medicine in Indonesia. Since then, the use of medicinal plants began to be considered ancient, dangerous, and backward. But in the last few decades, there has been a global tendency to return to nature or "back to nature". This trend is starting to be very strong in developed countries and has a big influence in developing countries such as Indonesia (3). One of the plants that can be used as traditional medicine is *Jatropha* (*Jatropha curcas* L.).

Jatropha is commonly used as a medicine, namely the leaves are used for medicine for canker sores, new wounds and reducing fever (4). The results obtained from this study are that the ethanol extract of *Jatropha* leaves has antipyretic activity that can reduce body temperature in male white rats at doses of 200 mg, 400 mg and 800 mg (5). *Jatropha* (*Jatropha curcas* L.) is believed to be a pain reliever (analgesic) plant. The results

obtained from the study showed that the overall decrease in the average response value was the lowest in group 4 with a dose of *jatropha* leaves 600 mg/kg BW, it can be concluded that *jatropha* leaf extract has an analgesic effect on male white rats (6). *Jatropha* leaves have analgesic effect because they contain flavonoid compounds. The results obtained in *jatropha* leaf extract as much as 125 mg/kg BW, 250 mg/kg BW and 375 mg/kg BW decreased more than the negative control group (7). A single dose of ethanol extract of *Jatropha* leaves in mice starting at a dose of 1400 mg/kg BW to 5734 mg/kg BW did not cause clinically toxic symptoms (8). A concentration of 50% there was a visible clear zone (dark to slightly green) which was not overgrown by *Candida albicans* (9).

Research (10) found that alcoholic extracts from the roots and stems gave effects like Pentazocine. The aqueous extract of *Jatropha* leaves at a dose of 150 mg/kg BW showed significant anti-inflammatory activity in carrageenan-induced rat paw edema models (11). The administration of ethanol leaf extract of *Jatropha curcas* caused morphological changes, histopathological changes and behavioral changes in test animals (12). Effect of *jatropha* extract against pathogenic bacterial strains can introduce the plant as a potential candidate for drug development in the treatment of diseases caused by human pathogens (13). Flavonoids have various kinds of bioactivity including antipyretic, analgesic and anti-

inflammatory effects. Flavonoids are found in almost all parts of plants such as flowers, fruits, seeds, and leaves (14). The results of the qualitative test of the class of metabolites present in the *Jatropha* leaf extract were positive for antibacterial compounds, namely flavonoids, tannins, and saponins (15). The active components of the plant, namely flavonoids, can inhibit fever-inducing prostaglandins, protein kinases, monoaminoxidase, DNA polymerase and cyclooxygenase (16).

Jatropha contains chemical compounds or secondary metabolites in all parts of its body from roots to leaves (17). The roots of these plants contain methyltrans-2-dekene-4,6,8-trienoate and 1-tridekene-3,5,7,9,11-pentene-beta-sitosterol. *Jatropha* leaves also contain compounds such as kaempferol, kaempferol-3-rutinoside, nicotiflorin, quercetin, isoquercetin and rutin. In addition, *jatropha* leaves also contain astragalol, reiniutrin and vitamin C. *Jatropha* stems contain saponins, flavonoids, tannins and polyphenolic compounds. *Jatropha* seeds contain 40-50% castor oil which contains various triglycerides, palmitic acid, ricinoleic acid, isorisinoleic acid, oleic acid, linoleic acid, linolenic acid, stearic acid, and dihydroxystearic acid. Besides that, *Jatropha* seeds also contain ricinin, several kinds of toxalbumin called ricin (acid ricin, and basic ricin) and several kinds of enzymes including lipase.

Objective: to determine the effective

dose of antipyretic from leaf extract of *Jatropha curcas* (*Jatropha curcas* L) in male mice (*Mus musculus*).

METHODS

Manufacture of *Jatropha* (*Jatropha curcas* L) Leaf Extract

Jatropha Leaves which has become simplicia made in powder form weighed as much as 500 grams and then extracted using the maceration method (immersion), the weighed sample is put into a maceration container and then moistened with 96% ethanol solvent until all samples are completely submerged and macerated for 3 days. Then every 1x24 hours stirred for 15 minutes using a digital stirrer at a room temperature that meets the requirements in room air ranging from 18-30°C (18). After 3 days the sample was filtered with the aim of separating the sample extract from the filtrate that was no longer needed. After that, the sample which is the result of maceration, namely the filtrate, is then evaporated using a rotavapor type evaporator until a thick extract is formed.

Peptone Production 10%

A 10% peptone solution was prepared by weighing 10 grams of peptone then dissolved in 100 ml of distilled water.

Preparation of Paracetamol Suspension in 1% Na-CMC

20 tablets of paracetamol were weighed and the average weight was calculated. After that, all the paracetamol tablets were put into a mortar and ground until they became powder.

Weighed 0.234 g of paracetamol powder and then suspended in 1% Na-CMC little by little while stirring, the volume was made up to 100 ml.

Preparation of Ethanol Extract Suspension of *Jatropha* Leaves (*Jatropha curcas* L)

Extracts from *Jatropha* Leaves that will be used in this study are 150 mg/kg BW, 200 mg/kg BW, and 250 mg/kg BW. Each of which was put into a mortar and then ground and added a colloidal solution of Na-CMC 1% w/v little by little until homogeneous. The homogeneous solution was then made up to volume with colloidal NaCMC 1% w/v solution to 50 ml in a calibrated bottle.

Grouping of test animals

The test animals that had been acclimatized for 7 days were weighed and given normal feed, before being given treatment the test animals were fasted for 8 hours. then divided into 5 groups. Animals in 1 group are placed together in 1 cage. In group 1 as a negative control and group 2 as a positive control while groups 3 to 5 were given ethanol extract of *Jatropha* leaves (*Jatropha curcas* L) orally according to the dose level, where each group consisted of 3 mice. Each test animal in the group will be induced by 10% peptone solution orally as a mediator or fever inducer.

Treatment of test animals

Each test animal was weighed and grouped into 5. First, the initial rectal temperature of the mice was measured, then the fever was induced using 10% peptone solution.

A total of 1 mL/30 grams of body weight of the mice was taken orally. The rectal temperature was measured using a digital thermometer. The data are initial temperature (T₀), temperature 30 minutes after administration of 10% peptone solution, and interval temperature every 30 minutes after treatment. and measured again using a digital thermometer, then each group was treated as follows:

1) Group 1 (negative control)

In the first group, male mice that had been acclimatized for 1 week were then fasted for approximately 12 hours and only given water, then 3 mice were treated with Na-CMC with a concentration of 1% w/v as a negative control of 1 mL orally induced and then seen the effect of decreasing body temperature in the mice. The rectal temperature of the mice was then measured again at 0, 30, 60, 90 and 120 minutes, respectively.

2) Group 2 (positive control)

In the second group of male mice that had been acclimatized for 1 week then fasted for approximately 12 hours and only given drinking then 3 mice were treated with antipyretic drugs as positive controls, in this study paracetamol 0.234% w/v then induced orally and then seen the effect of decreasing body temperature on the mice, the rectal temperature of the mice was then measured again at 0, 30, 60, 90 and 120 minutes, respectively.

3) Group 3 (Test I)

In the third group, after the mice were

induced using 10% peptone orally to increase body temperature, they were then treated with ethanol extract of *Jatropha* leaves (*Jatropha curcas* L) orally at a dose of 150 mg/kg BW mice, the extract was administered to see what dose of The extract was effective in lowering body temperature in mice with each dose given to 3 different mice that had been fasted beforehand. After being treated, the rectal temperature of the mice was then measured again at 0, 30, 60, 90 and 120 minutes, respectively.

4) Group 4 (Test II)

In the fourth group, after the mice were induced using 10% peptone orally to increase body temperature, they were then treated with ethanol extract of *Jatropha* leaves (*Jatropha curcas* L) orally at a dose of 200 mg/kg BW mice, the extract was administered to see what dose of The extract was effective in lowering body temperature in mice with each dose given

to 3 different mice that had been fasted beforehand. After being treated, the rectal temperature of the mice was then measured again at 0, 30, 60, 90 and 120 minutes, respectively.

5) Group 5 (Test III)

In the fifth group, after the mice were induced using 10% peptone orally to increase body temperature, they were then treated with the ethanol extract of *Jatropha* (*Jatropha curcas* L) leaves orally at a dose of 250 mg/kg BW mice, the extract was administered to see what dose of The extract was effective in lowering body temperature in mice with each dose given to 3 different mice that had been fasted beforehand. After being treated, the rectal temperature of the mice was then measured again at 0, 30, 60, 90 and 120 minutes, respectively.

RESULTS AND DISCUSSION

Table 1. Measurement Of The Decrease In Temperature Of Mice

Group	Mice	Temperature After Treatment (oC)					
		T	T0	T1	T2	T3	T4
Control (-) (Na-CMC 1%)	1	34.3	35.1	35.0	35.0	34.9	34.9
	2	34.2	35.4	35.4	35.4	35.1	34.8
	3	34.7	35.4	35.4	35.5	35.5	35.4
	Average	34.4	35.3	35.2	35.3	35.1	35.0
Control (+) (Paracetamol)	1	36.6	37.2	36.9	36.8	36.6	36.4
	2	36.6	37.3	36.6	36.3	36.1	36.0
	3	36.2	36.9	36.0	35.8	36.0	35.5
	Average	36.4	37.1	36.5	36.3	36.2	35.9
Dose I (<i>Jatropha</i> Leaf Extract 150 mg/kg BW)	1	35.1	36.5	36.3	36.1	36.0	35.9
	2	34.1	35.1	35	35.2	35.2	35.1
	3	35.3	36.0	35.9	35.6	35.3	35
	Average	34.8	35.8	35.7	35.6	35.5	35.3
Dose II (<i>Jatropha</i> Leaf Extract 200 mg/kg BW)	1	35.2	36.5	36.3	36.0	36	36.0
	2	35.0	36.2	36.0	35.8	35.5	35.3
	3	34.8	35.9	35.7	35.5	35.3	35.0
	Average	35.0	36.2	36.0	35.7	35.6	35.4

Dose III	1	36.0	37.2	36.8	36.5	36.3	36.1
(Jatropha Leaf	2	36.2	37.0	36.7	36.3	36.0	36.0
Extract 250 mg/kg	3	35.8	36.7	36.0	36.0	35.9	35.7
BW)	Average	36.0	36.9	36.5	36.2	36.0	35.9

Information :

T = Initial temperature of mice

T0 = Temperature after administration of Peptone 10% (fever inducer)

T1 = Temperature at 30 minutes after drug administration

T2 = Temperature at 60 minutes after drug administration

T3 = Temperature at 90 minutes after drug administration

T4 = Temperature at 120 minutes after drug administration

Table 2. Results of Post Hoc Analysis with LSD test

Results of Post Hoc Analysis with LSD Test					
Treatment	Control Negative	Positive Control	1 dose	2 dose	Dose 3
Negative Control	-	0.000	0.064	0.010	0.000
Positive Control	0.000	-	0.001	0.005	0.538
1 dose	0.064	0.001	-	0.413	0.003
2 dose	0.010	0.005	0.413	-	0.020
Dose 3	0.000	0.538	0.003	0.020	-

The results of the study listed in table 1, it can be seen that there are different or varied temperature changes in each treatment group. Based on the table, it was found that group 1 (negative control) induced with Na-CMC did not show a significant change in body temperature, so it can be seen that Na-CMC did not affect the decrease in body temperature of mice. In contrast to group 2 (positive control) who were given paracetamol

suspension, a better decrease in body temperature was seen, because the body temperature seemed to be able to reach a normal state (normal body temperature).

The results of temperature measurements for each treatment group are shown by the graph in Figure 1 about the average decrease in body temperature of mice, as follows.

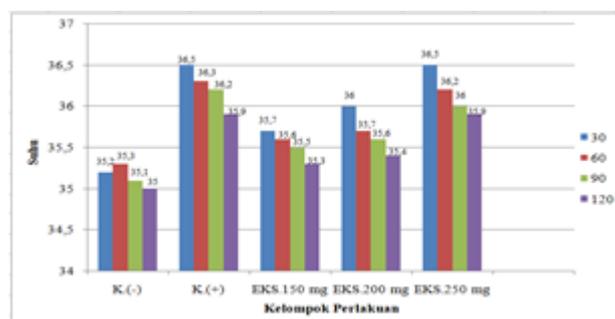


Figure 1. Average Decrease In Body Temperature Of Mice

Based on the data obtained from the series of measurements, it was found that in each extract there was a significant decrease but after averaging, it was found that the 250 mg dose had good effectiveness in reducing body temperature / fever that occurred in mice.

If it can be observed again, should each increase in the dose of the drug in a control group by the extract give an effect that is proportional to the increased dose, but with this increase the dose increase in the final response only decreases. Although there is a slight decrease. This often occurs in drugs, especially natural ingredients, because the components of the compounds contained are not single but are composed of several kinds of chemical compounds, where these components work together to cause an effect, but with an increase in the dose of the number of chemical compounds required. automatically contains more and more, so that adverse interactions can occur, this can lead to a decrease in the effect, besides that the limited number of receptors also affects so that even though the dose variation is increased,

The 250 mg extract had a gradual decrease in temperature which indicated that the antipyretic activity of the dose was running well. Based on the parameters from the bar chart that have been presented, showing the stages of decreasing body temperature well at that dose up to 120 minutes compared to 150 mg and 200 mg, the temperature drop did

occur but experienced a fluctuating decrease, so it can be assumed that the best dose variant which is effective against decreasing body temperature is at a dose of 250 mg. Fever is a condition where there is an increase in temperature above normal. When measured rectally the temperature reaches $> 38^{\circ}\text{C}$, if measured orally, the temperature is above 37.8°C and if measured through the axilla the temperature is above 37.2°C (99°F) (19,20). Meanwhile, according to NAPN (National Association of Pediatrics Nurses) it is stated that fever occurs when infants less than 3 months old have a rectal temperature exceeding 38°C , in children older than 3 months the axillary and oral temperature is more than 38.3°C . The theory becomes a benchmark that the increase in body temperature that occurs in mice after being induced by Peptone is in accordance with the existing theory, where the increase in temperature experienced ranges from $36.9-37.8^{\circ}\text{C}$.

The use of paracetamol as a positive control was aimed at determining the antipyretic effectiveness of the extract of *Jatropha Curcas L* (*Jatropha Curcas L*) leaves from several extract dosage variants which were made as a comparison. This drug has activity by inhibiting the cyclooxygenase enzyme so that the formation of prostaglandins is inhibited. The flavonoids contained in the leaf extract of *Jatropha* (*Jatropha Curcas L*)

have the same mechanism of action as Paracetamol where this compound also inhibits the increase in prostaglandin synthesis through inhibition of the cyclooxygenase enzyme (14).

So in this study it can be assumed that the pharmacological effect in the form of antipyretics in the extract of *Jatropha Curcas* (*Jatropha Curcas* L) leaves in male mice (*Mus musculus*) has proven to be efficacious, and this is also evidence from empirical data obtained in the community that *Jatropha* leaves have been used as a fever-reducing drug long before this study began.

Data Analysis

From the data resulting from the decrease in body temperature that has been obtained, then proceed with statistical analysis to see whether the effect of treatment on the animals that have been tested. Before further statistical tests are carried out, the normality test and homogeneity test are carried out, this test needs to be carried out because the results will determine further statistical tests (Fathoni, n.d.). If the data obtained are normally distributed and the variation is homogeneous, then hypothesis testing can be done using a parametric statistical model. If the data obtained are homogeneous and normally distributed, then proceed with the One Way Anova analysis (Murti, 1996). The normality test was carried out to determine whether the data obtained came from a normally distributed population or not. The

homogeneity test is used to determine whether there are several population variants that are the same or not. Based on the results obtained, the data is normally distributed and homogeneous so that it can be continued with the ANOVA test.

The statistical test used next was the One Way Anova test with a 95% confidence level or a significant level (α) = 0.05 to find out the significant differences between all treatment groups. One Way Anova statistical test is used to test the difference in the average treatment in an experiment for more than two groups by comparing the variances. In this case, it was used to see whether there was a significant (significant) difference in the antipyretic effect between each treatment group. The results of statistical tests on the decrease in the temperature of the mice were obtained significantly less than 0.05 ($p < 0.05$), which means that there is a significant difference or significant difference in the decrease in temperature between the treatment groups.

The Post Hoc test was carried out after the results of the One Way Anova test were obtained. The post hoc test used is the LSD (Least Significant Different) test. This test was conducted to compare the presence or absence of significant differences in each group. The results of the LSD test showed a significant or significant difference if the significance value of each treatment group was less than 0.05 (≤ 0.05). Based on the results of the analysis

that has been done, it can be seen that the negative control group had a significantly different effect with the positive control group, doses 2 and 3 because they had a p value <0.05 . This is because Na CMC does not contain compounds that are efficacious as antipyretics so that the effect is different from the other four groups. Meanwhile, the positive control group with doses 2 and 3 did not show any difference in antipyretic effectiveness against male mice because a significant value was >0.05 so it can be said that both doses of the extract had antipyretic effectiveness. Between the two extract doses, it was found that the highest antipyretic effect was found at a dose of 250 mg/kg BW of *Jatropha curcas* L. extract because it had a greater significance value against the positive control, so that the antipyretic effectiveness of paracetamol could be said to be comparable to that of the *Jatropha* Leaf extract (*Jatropha curcas* L.) 250 mg/kg BW. Besides being comparable to the positive control dose, this dose can also be said to be stable because the resulting decrease in body temperature experienced a significant decrease or did not change.

CONCLUSION

Based on the results of the research that has been done, several conclusions can be drawn including the following: *Jatropha* leaves (*Jatropha curcas* L.) have been shown to have good effectiveness in reducing body temperature of mice (*Mus musculus*) based on the chemical content contained in the plant

after phytochemical screening, which contains flavonoids where flavonoids have an active role as antipyretics. The dose that had good effectiveness as an antipyretic in male mice after being averaged was at a dose of 250 mg/kg BW because there was a decrease that was comparable to the positive and stable control.

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