The Effectiveness Extract of *Curcuma Xanthorriza* Roxb to Decrease Histopathology Brain Pressure in Male Mice (*Mus Musculus*)

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**Abstract.** This study was to determine the effect of *Curcuma* extract on brain histopathology in hypertensive-induced male mice compared with captopril administration. *Curcuma* was extracted using remaseration method using 96% ethanol solvent. Induction of hypertension using L-name for 4 weeks where the first 2 weeks are given before therapy then the next 2 weeks are given with therapy. The therapy used is *curcuma* extract and captopril as control therapy. This study used four groups of mice that group distilled water, L-Name, captopril and *curcuma* with the average yield is 42 ± 11.81, 57.51 ± 166, 119 ± 8.40, and 111.5 ± 14.20. Results of analysis using Tukey's Post Hoc test showed that there was no significant difference between groups aquades, captopril, and extract *Curcuma xanthorriza* but there are significant differences between the groups of L-Name with the other three groups. It can be concluded that captopril can reduce the amount of damage to brain cells in male mice but not as good as *Curcuma xanthorriza* extract.

1. **Introduction**

Hypertension can cause complications in the form of damage to organs in the heart, brain, kidneys and blood vessels. Damage to the heart causes left ventricular hypertrophy to heart failure, damage to the brain causes strokes, damage to the kidneys causes chronic kidney disease to kidney failure and the aorta can cause aneurysms and tearing of the intima layer [1]. Treatment given to patients is to conduct therapy in patients with the use of antihypertensive drugs. One of the anti-hypertensive drugs used is captopril, which is a class of Angiotensin Converting Enzyme (ACE) inhibitors. This drug is used because it has no effect on heart rate and cardiac output and does not reduce blood flow to the brain, coronary arteries, or kidneys [2].

Captopril is quickly absorbed but has a short duration of action, so it is useful to determine whether a patient will respond well to ACE-inhibitors. The first dose of ACE-inhibitor must be given at night because a sudden drop in blood pressure may occur, this effect will increase if the patient has low sodium levels. Captopril is quickly absorbed but has a short duration of action, so it is useful to determine whether a patient will respond well to ACE-inhibitors. The first dose of ACE-inhibitor must be given at night because a sudden drop in blood pressure may occur, this effect will increase if the patient has low sodium levels. Hypertension is caused due to the activity of Angiotensin II in the lungs which plays an important physiological role in regulating blood pressure. Angiotensin I is changed by ACE (Angiotensin Converting Enzyme) to Angiotensin II. Angiotensin II plays an important role in raising blood pressure. Therefore, research is needed regarding the bioactive compound inhibitors Angiotensin Converting Enzyme.

In general, controlling the activity of Angiotensin II uses captopril drugs. However, the high price of synthetic drugs and the emergence of human awareness of the negative side effects of the use of synthetic drugs have encouraged people to use natural medicines derived from plants [3]. Even recently there has...
been an increase in research on herbs and natural ingredients to treat various diseases [4]. The pharmaceutical industry is also trying to find opportunities for the use of natural materials and their derivatives as ingredients for medicine.

Traditional medicine has advantages, namely: it has relatively small side effects, in one herb with different components there is a mutual support effect, one plant has more than one pharmacological effect and is more suitable for metabolic and degenerative diseases. Traditional plants have properties that work as an anti-oxidant, analgesic, and anti-inflammatory, therefore leading to healing of a disease. Research also conducted by [5] with curcumin and tetrahydro curcumin doses of 50 mg and 100 mg/kg given to hypertensive-induced mice using L-Name showed the average MAP (mmHg) at a dose of 50 mg with a MAP value of 140.4 ± 2.2, a dose of 100 mg 136.1 ± 0.9 and a placebo group 157.7 ± 2.2. So far the testing of *Curcuma xanthorriza Roxb* extract. As an antihypertensive has not been done much, therefore this study aims to determine the benefits of *Curcuma xanthorriza Roxb* ethanol extract in reducing hypertension in male mice (*Mus musculus*) by giving L-Name as an induction of hypertension in male mice (*Mus musculus*).

2. RESEARCH METHODS

2.1. Tools and ingredients.

This study used mice (*Mus musculus*) with inclusion and exclusion criteria as follows: Male sex, 2-3 months old, weight 20-30 grams, have never experienced any treatment or have never received a chemical intake, do not suffer from disease or in a healthy condition (active move, hair does not fall out, not deformed, not bleed, hypertension with BP > 120 mm/Hg.

2.2. Method

Extraction is done by remaseration method. The curcuma rhizome is dried at room temperature and protected from direct sunlight for one week. Dry rhizomes are then crushed using a grinding tool. This method is a cold extraction method so it does not use heat in the process. No use of heating in this method is expected to minimize the possibility of damage to curcuminoids contained in ginger. Furthermore, the extraction process in this study was carried out through the use of room temperature with a pressure of 1 atm and stirring 200 rpm. Induced hypertension in male mice (*Mus musculus*) by giving L-name orally through a drinking bottle with a dose of 1.75 mg/25gBB every day. Within 2 weeks it is expected that there will be an increase in systolic and diastolic blood pressure in male mice from physiological pressure> (120/71mmHg). In weeks 2 and 4, blood pressure measurements were performed in male mice (*Mus musculus*) in each group, blood pressure measurements using a Blood Pressure Recorder. After that, brain organ surgery and histopathological observation of the brain using parameters, the first treatment of mice was turned off with anathesis, then the brain organs were taken. Brain organs were treated following a standard histological method by HE staining. Standardization of histopathological examination preparations of brain organs, a picture of mouse brain damage seen by observing histopathological preparations using a microscope with a magnification of 40x. The target being read is a change in the histology of the brain organ in astrocyte cells contained a nucleus into picnotics [6]. Then the examination is recorded in the form for analysis. The analysis in this study used a test with the Normality and Post Hoc methods.

3. Results and Discussion

The results of making *Curcuma Xanthorriza Roxb* ethanol extract, A total of 1000 grams, the results obtained in the form of thick dark brown extract weighing 159 grams with curcumin content of 7.23%.
3.1 Results of Brain Histopathology

Histopathological observations of the brain were carried out using a light microscope at 40x magnification using laboratory optical software and were counted in 5 visual fields. In Figure 1, Histopathological observations of male mice brain cells (Mus Musculus) showed that the induction of L-Name in animal models of male mice (Mus Musculus) with a dose of 1.75mg/kg 25gBB resulted in damage to the brain marked by the presence of cerebral edema that can be seen from changes in astrocyte cells, astrocyte cell damage is characterized by picnotic nuclei and vacuolated cytoplasm.

![Image of histopathological observations](image.png)

Figure 1. Microscopic vacuolated cells with a magnification of 40 times. (A) normal glia cells, (B) glia cells with picnotic nuclei, vacuolated cytoplasm, (C) = cerebral cortex 10 times ob.

The mean number of astroit cells edema per field in histopathological preparations of the brain in male mice obtained from microscopic observations through five different fields of view of the whole group namely the LN group as a negative control (L-NAME), the PC positive control group (L-NAME) + 0.04875 mg/30gBB captopril), and group T is the treatment of temulawak extract at a dose of 31.25 mg/30gBB).

3.2 Analysis

Histopathological observation data analysis of male mice (Mus Musculus) brain cells is managed using the SPSS (Statistical Program for Social Science) application 18.0 for windows. Curcuma xanthorrhiza Roxb ethanol extract was tested against histopathology of mouse brain cells. The sample normality test results obtained all groups have a Sig. or P> 0.05 indicates that the data used are normally distributed or taken from the normal population. Because the normal data distribution was then analyzed using the One Way ANOVA test, and it was found that the data variant was normal (p> 0.05) so the ANOVA test results would be valid. One Way ANOVA test on the value of the number of brain nerve cells in each field of view found a significant difference (p <0.05). This shows that there are at least significant differences in the number of brain nerve cells in each field of view in the two groups.
Figure 2. From the post hoc test found significant differences between the T group with PC and LN groups. The difference in the number of brain nerve cells in each field of view is significant in the two groups, namely the T group with PC and LN. During the research, there was one mouse that died in the LN group, the dead mouse was not replaced with a backup sample because it still met the specified requirements.

3.3 Discussion

In the research that has been done, using extracts from the ginger rhizome (*Curcuma xanthorriza* Roxb.) Obtained from Materia Medica, Batu city. The extract was made by remaseration method which is a modification of the literature, where to do the remaseration method used a fixed ratio of 1:10, both in the first maceration and other maceration. A total of 1000 grams, the results obtained in the form of thick dark brown extract weighing 159 grams with curcumin content of 7.23%.

The experimental animal used was male mice (*Mus musculus*) weighing 20-30 grams aged 2-3 months and then acclimatized for 1 week at the study site for adjustment to the environment. After that the mice were randomly divided into 5 groups, P1 by giving placebo in the form of aquadest as negative control, P2 by giving L-Name as negative control, P3 by giving Captopril as positive control, P4 by giving *Curcuma xanthorriza* roxb extract at a dose of 31.2 mg/30 g BB. The dose of Captopril used is the usual dose that is often used at 12.5 mg/day. Based on research [5] L-NAME administration as an induction of hypertension used a dose of 50 mg/kg BW and using a [7] received a conversion dose of 70 mg/kg BW in mice. Based on research of curcumin dose to mice using three doses, namely 50 mg/kg body weight, 75 mg/kg body weight, 100 mg/kg body weight, and effectively reducing blood pressure at a dose of 75 mg/kg body weight.

The treatment of mice was carried out for 4 weeks, the mice were adapted for 1 week so that the mice could adjust to the new place and environment. Mice are placed in a clean, well-ventilated cage with a length of 50 cm, width 50 cm, height 40 cm. During the 4-week trial period the mice were given a standard food consumption and drinking water. Induced hypertension in male mice (*Mus Musculus*) by giving L-name orally through a drinking bottle with a dose of 1.75 mg/25gBB every day. Within 2 weeks it is expected that there will be an increase in systolic and diastolic blood pressure in male mice from physiological pressure>=(120/71 mmHg). *Curcuma xanthorriza* Roxb extract is given every day for the past two weeks by oral route through food carriers. The extract is done once a day at 09.00 am. In weeks 2 and 4, blood mice (*Mus musculus*) were measured in each group. Mice were measured their blood pressure using a Blood Pressure Recorder by putting mice in a mice holder, then blood pressure was measured through the tail and the results recorded.
Histopathology of male mice brain cells (*Mus Musculus*) shows that the induction of L-Name in animal models of male mice (*Mus Musculus*) at a dose of 1.75 mg/25gBB results in damage to the brain characterized by the presence of edema which can be seen from changes in astrocyte cells. Astrocyte cell damage is characterized by picnotic nuclei and vacuolated cytoplasm.

In the histopathological figure of the brain nerve cells as seen in Figure 1 shows the glia cells with picnotic nuclei. In the LN group (L-name) showed that the average brain nerve cells in each field of view were the highest 166. This is because the LN group was given L-NAME as induction of hypertension for 4 weeks. And the mean brain nerve cells per field was lowest in the T group (curcuma) with a smaller number 111.5 there was a decrease in the average number of brain nerve cells per field of the LN group (L-name) as a negative control compared to the T group (curcuma) with curcuma extract treatment.

This shows that the administration of curcuma extract as a therapy for hypertension at a dose of 31.25 mg/30gBB in male mice induced by L-NAME has an effect on the number of brain nerve cells in each field of view. There is a difference in the number of brain nerve cells in each field of view in male mice (*Mus musculus*).

4. Conclusion
The results showed that administration of *curcuma* extract at a dose of 75 mg/kg BW could reduce the number of brain nerve cells in male mice (*Mus musculus*) induced by hypertension. The decrease in the number of brain nerve cells in the Captopril group was different from the decrease in the number of brain nerve cells in the curcumin group (curcuma extract 75 mg/kg BW), which decreased better and each field of view found a significant difference (p <0.05).

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