Ethanol Mangosteen Rind Extract
(Garcinia x-mangostana L.) Activity on Wistar White Male Rats Ureum Creatinine Levels

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Abstract—Objectives: Kidney is organ that plays an important role in maintaining the homeostasis of the body. Kidney damage can be caused by several things, such as hypovolemia, sepsis, acute glomerulonephritis, rhabdomyolysis and drugs (gentamicin and NSAIDs). Consumption of drugs like gentamicin can cause damage and decreased kidney function through the formation of free radicals. Mangosteen rind could be expected to provide nephroprotective antioxidant effect. These research was to determine the activity and effective doses of mangosteen rind (Garcinia x-mangostana L.) ethanol extract on gentamicin induced male rat creatinine and Blood Urea Nitrogen (BUN) level. Rats were divided into 5 groups. Group I as normal control (Aquadest po), the second group of negative control (Gentamicin 60 mg / kgBW ip), group III, IV, and V (extract of mangosteen rind dose of 50 mg / kgBW; 100 mg / kgBW; and 200 mg / kgBW orally for 7 days). After dosage administered, followed by induction of gentamicin for 7 days. According to ANOVA with 95% level of confidence, the results showed that mangosteen rind ethanol extract gives a significant differences, which Dose II and Dose III can reduce creatinine levels, as well as protects from necrosis with average levels of creatinine 1,086 mg/dl and 1,158 mg/dl. However, mangosteen rind (Garcinia x-mangostana L.) ethanol extract had no activity on BUN levels.

Keywords: Garcinia x-mangostana L., gentamicin, creatinine, BUN

I. INTRODUCTION

Indonesia from time immemorial has been familiar with the treatment by utilizing the existing plants in the forest and plants that exist around the yard of the house to treat various diseases both external and internal medicine diseases [8]. Kidneys are vital organs that play an important role in maintaining the stability of the environment in the body. Kidney damage can be caused by many things, such as hypovolemia, sepsis, acute glomerulonephritis, rhabdomyolysis and drugs (gentamicin and NSAIDS) [7]. Gentamicin is an aminoglycoside class of antibiotics which are widely used primarily to treat infections caused by gram-negative bacteria [5].

Aminoglycoside antibiotic gentamicin classified as a known toxic to the kidneys. A result that can be caused by nephrotoxic agents is one of them is renal tubular damage, by activating protease which causes oxidative injury tubular epithelial and capillary endothelium caused by the formation of reactive oxygen species (ROS) [7].

In these conditions the necessary antioxidants that are useful to bind free radicals is excessive. One of the plants that allegedly capable of functioning as an antioxidant is the rind of the mangosteen (Garcinia x-mangostana L.) contains compounds that have pharmacological activity and antioxidant. The compounds are tannins and xanton [3].

The skin of the mangosteen fruit contains flavonoids, tannins and polyphenols that have antioxidant activity [1]. This study aims to determine the activity nephroprotectif ethanol extract of mangosteen rind on levels of urea, creatinine renal function as a parameter.

II. MATERIAL AND METHOD

A. Procedure

Tools and material
Analytical balance, beaker glass (Pyrex), cup vaporizer, a pipette, a funnel, a water bath, set of tools maserator, rotary evaporator, syringe, sonde oral, stopwatch, tube eppendorf, centrifugator, centrifuge tubes, micropipette, photometer (Intherma-168), Mangosteen rind, 96% ethanol, distilled water, gentamicin, ammonia 10%, hydrogen chloride (HCl) 1N, hydrogen chloride (HCl) 2N, sodium hydroxide (NaOH), metal Mg, reagent Mayer, reagent Dragendroff, reagent Liberman-Buchard, anisaldehid, chloroform (CHCl3), vanillin, reagent kit of creatinine (DiaSys), reagent kits urea (DiaSys) and test animal feed.

Animal Testing
Experimental animals used were male rats wistar strain 2-3 months old and weighs 200-250 grams as much as 25 animals. All animals were acclimatized for 7 days.

Sample Collection and Determination
The sample used in this study is the mangosteen rind obtained from plantations in the district Urug, Tasimalaya district. The plant material used in this study is the skin of the
fruit. Determination plants is conducted in Jatinangor Herbarium, Plant Taxonomy Laboratory, Department of Biological Science.

Preparation of Extract
Preparation of ethanol extract of mangosteen rind (Garcinia x-mangostana L.) of 400 grams was extracted by maceration, by immersing the mangosteen rind that has been blended in 96% ethanol for 24 hours with occasional stirring. Then filtered using filter paper. In residues do remasensi with the type and amount of the same solvent until the filtrate becomes colorless. After that evaporated with a rotary evaporator to obtain a thick extract of mangosteen rind.

Phytochemical screening
Phytochemical screening is done by testing the class of alkaloids, flavonoids, tannins and polyphenols, saponins, monoterpenes and sesquiterpenes, steroids and triterpenoids and quinones.

Testing Activities Nefroprotectif Ethanol Extract Mangosteen Fruit Leather
This research was conducted by using The Posttest Only Control Group Design, with five treatment groups. Animal test levels measured creatinine and Blood Urea Nitrogen (BUN) after being given treatment at a dose of 50 mg / kgBW Rat, Dose II = 100 mg / kgBW Rat, Dose III = 200 mg / kgBW Rat. Categorization of animal experiments as follows:

The first group as normal control mice that were only given food and drink ad libitum.

b. The second group as a negative control which rats were given CMC 1% po, 2 hours and then induced gentamicin 60 mg / kg intraperitoneally for 7 days.

c. The third group as the first test dose group were given ethanol extract of mangosteen rind 50 mg / Kg BW rats in CMC 1% po, 2 hours and then induced gentamicin 60 mg / kg in the IP (conducted for 7 days).

d. The fourth group as group II were given a test dose of ethanol extract of mangosteen rind 100mg / kg body weight of mice in CMC 1% po, 2 hours and then induced gentamicin 60mg / kg in the IP (conducted for 7 days).

e. The fifth group as a group III were given a test dose of ethanol extract of mangosteen rind 200mg / Kg BW rats in CMC 1% po, 2 hours and then induced gentamicin 60mg / kg in the IP (conducted for 7 days).

On day 8 after 24 hours measured creatinine levels and levels of Blood Urea Nitrogen (BUN).

Serum Sample Preparation
Creatinine was carried out by the method of Jaffe reaction and BUN examination carried out by the method of Urease. Blood sampling is done by cutting the rats and the blood is collected in tubes setrifugasi, then centrifuged for 20 min at 2000 rpm with the aim to separate the serum to be used for screening [4].

Examination of creatinine and BUN levels
The level of creatinine was conducted by Jaffe Reaction Time Fixed BUN levels while the investigation was conducted by Urease Fixed Time.

B. Data Analysis
Analysis of the data used in this study using SPSS 16 statistical method ANOVA between normality test, homogeneity test, ANOVA and LSD.

III. RESULTS AND DISCUSSION

Extraction
Extraction is done by using a 3x24-hour maceration method, with 96% ethanol as a solvent. 96% ethanol is used as solvent has the ability to attract secondary metabolites contained in the sample well. Ethanol 96% are able to produce the highest yield allegedly because it can dissolve phytochemical compounds as a whole and able to attract some of phytochemical compounds that are dissolved in a polar solvent [6]. Mesenrit is then concentrated by rotary evaporator. The results of the extraction process 400 grams of crude drugs obtained mangosteen rind extract as much as 119.22 grams viscous and yield of 29.80%.

Phytochemical screening
Based on the results of phytochemical screening on samples obtained crude drugs and extracts the positive results of flavonoids and polyphenols are believed to have antioxidant activity in this study. In addition, the positive results obtained also against alkaloids, saponins, tannins, triterpenoids, monoterpenes and sesquiterpenes.

<table>
<thead>
<tr>
<th>TABLE 1: PHYTOCHEMICAL SCREENING RESULTS</th>
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<tbody>
<tr>
<td>Secondary metabolites</td>
</tr>
<tr>
<td>------------------------</td>
</tr>
<tr>
<td>alkaloids</td>
</tr>
<tr>
<td>flavonoids</td>
</tr>
<tr>
<td>saponin</td>
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<tr>
<td>polyphenols</td>
</tr>
<tr>
<td>tanin</td>
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<tr>
<td>monoterpenes and sesquiterpen</td>
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<tr>
<td>steroids and triterpenoids</td>
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<tr>
<td>quinone</td>
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</tbody>
</table>

Information :  
+ = Unidentified chemical compounds  
- = Not identified chemical compounds
Creatinine Levels examination and Blood Urea Nitrogen (BUN)

TABLE 2: THE AVERAGE LEVELS OF CREATININE AND BUN (mg/dl)

<table>
<thead>
<tr>
<th>Group</th>
<th>On average Creatinine (mg/dl)</th>
<th>Average Blood Urea Nitrogen (BUN) (mg/dl)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal</td>
<td>1.258 ± 0.233</td>
<td>15.467 ± 3.133</td>
</tr>
<tr>
<td>Negative</td>
<td>2.196 ± 0.818</td>
<td>26.992 ± 12.354</td>
</tr>
<tr>
<td>dose I</td>
<td>1.732 ± 0.272</td>
<td>51.555 ± 5.634</td>
</tr>
<tr>
<td>dosage II</td>
<td>1.086 ± 0.202</td>
<td>21.001 ± 4.920</td>
</tr>
<tr>
<td>dose III</td>
<td>1.158 ± 0.333</td>
<td>23.667 ± 5.287</td>
</tr>
</tbody>
</table>

Based on the ANOVA test results obtained that the significance value of 0.003 <0.05, which means that Ho is rejected, meaning that the data vary considerably. This means that the mangosteen rind extract provides significant differences against impairment of creatinine in mice. So it continued with LSD test to determine which groups are giving a significant difference. The negative control group was significantly different to the normal group (p = 0.003), the test dose group II (p = 0.001) and with the third test dose group (p = 0.001). Test dose groups II and III test dose of mangosteen peel extract has the ability to reduce levels of creatinine with creatinine average of 1,086 and 1,158. Dosage Test II is the most effective dose in lowering levels of creatinine.

A decrease in creatinine levels at the test dose groups II and III trials doses due to the presence of secondary metabolites, which act as antioxidants to restore the function of damaged cells caused by free radicals produced by gentamicin.

Based on the results obtained that the ANOVA significance of 0.000 <0.05, which means that Ho is rejected it means the data vary considerably. This means that the ethanol extract of mangosteen rind of the test dose groups II and III were given for 7 days did not cause a decrease in BUN levels in rats. It is also in accordance with the average yield BUN levels have increased compared to normal controls, which means mangosteen rind does not have the ability to reduce levels of BUN (Blood Urea Nitrogen) when compared to normal controls. While the first dose had an increased compared to the negative control. Levels of urea is one sign that is commonly used to estimate the glomerulus Filtration Rate (GFR), but examination of urea only as a check supporter, this can be caused by several internal and external factors among which the intake of animal protein test can affect test parameters [9].

IV. CONCLUSION

According to ANOVA with 95% level of confidence, the results showed that mangosteen rind ethanol extract gives a significant differences, which Dose II and Dose III can reduce creatinine levels, as well as protects from necrosis with average levels of creatinine 1,086 mg/dl and 1,158 mg/dl. However, mangosteen rind (Garcinia x-mangostana L.) ethanol extract had no activity on BUN levels.

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