Effect of Breadfruit Leaves Infusion on Acute Renal Failure Rat Model

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Abstract

Background: Empirically, breadfruit (Artocarpusaltilis) leaves have been used as traditional medicine for several diseases, such as acute renal failure. This study aimed to find the effects of breadfruit leaves infusion on blood creatinine levels in rats to model acute renal failure.

Methods: This laboratory experimental study was conducted by using 30 male wistar rats, weighing150–250 g. The rats were divided into 5 groups. The first group is a negative control and the second group is a positive control. Groups 2–5 were inducedby Gentamicine and peroxicam for 7 days. Groups 3–5 were given infusion of breadfruit leaves in different dosage for 7 days then the blood creatinine levels were tested at the 15th day. This data was analyzed using a multiple comparison Dunnett T3 test because this data was not homogenous. This study was done in Pharmacological Laboratory Universitas Padjadjaran from October toNovember 2012.

Results: The average of creatinine levels was as follows 0.62, 0.87, 0.98, and 0.87 mg/dL observed from the different groups. It was statistically shown that the significantly different creatinine levels, differing by 0.038 ($p<0.05$) were observed for the control group and the group which was given an infusion of breadfruit leaves. The significant difference on the average of creatinine levels were among the groups with (p–value <0.05).Comparison of group 1 and 3 ($p < 0.05$) gave a significant increase among the groups.

Conclusions: Infusions of breadfruit leaves have some effects on blood creatinine levels in acute renal failure according to the dosage and duration of administration. [AMJ.2015;2(4):511–5]

Keywords: Acute renal failure, breadfruit leaves, creatinine

Introduction

Renal disease is a decrease of renal function which is sudden within a couple hours to several days marked by urea, creatinine, potassium, and drugs retentions which are normally excreted by the kidneys.[1,2] Decrease of renal function can be due to the decrease of blood flow to the kidneys caused by infection, ischemia, or toxins such as drugs or nephrotoxic chemicals.[3] Drugs that are nephrotoxic are Non-steroidal anti-inflamatory drugs (NSAIDs), Aminoglycoside antibiotics, Amphotericin B, cisplatin, radiocontrast media, and other nephrotoxic drugs.[4,5]

Drugs used in this study which are gentamycin and piroxicam are proven to be able to damage the kidneys for 7 days to cause renal failure.[6] Breadfruit leaves have anti–oxidant effects and are believed to be able to lessen renal damage because of its flavonoid contents that can decrease the creatinine levels in blood by inhibiting the activity of free radicals and lessening the damages.[7]

According to assumption that breadfruit leaves contain flavonoid compounds which are anti–oxidants. It was needed to study the effects of breadfruit leaves on creatinine levels using experimental animals induced by gentamycin and piroxicam.

Methods

This study was conducted in the animal laboratory of the Department of Pharmacology and Therapy of the Faculty of Medicine, Universitas Padjadjaran, Bandung in October 2012. Breadfruit leaves (Artocarpusaltilis) were obtained from around Bandung, and identified in the herbarium of Jatinangor, Universitas Padjadjaran. The study used 30 male wistar rats as subjects (2–3 months old)

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Table 1 Creatinine Levels Average

Group	Creatinine Levels Average (Mg/dl)	Standard Deviation
1	0.62	0.05
2	0.87	0.27
3	0.83	0.10
4	0.96	0.18
5	0.89	0.22

Note: Group 1: Negative Control, Group 2: Positive Control, Group 3: 1 time dose treatment, Group 4: ½ time dose treatment, Group 5: 2 times dose treatment

which were healthy and weighing 150–250 g and divided into 5 groups. All rats were adapted for 7 days and given food and drink homogeneously.

Infusion water was made out of dried breadfruit leaves which were weighed according to the dose and 100 mL of water for treatment on each group. All base materials were mixed and interred to infusion pot for 15 minutes until the temperature reached 900 C and stirred for every 5 minutes. Seventy milliliters of infusion water was obtained and used for treatment for 4 mL on each rats.

After being adapted, each group was given different treatments. Negative control group (I) were only fed and given water for 14 days, positive (II) and treatment groups (III, IV, and V) were induced by gentamycin and piroxicam for 7 days to promote inflammatory response so that the rats on each group experienced renal failure.[6] After induction, the treatment groups were given breadfruit leaves infusion by per oral and with different doses of 4.25%, 9.5%, and 19% for 7 days. After the treatments were done, the rats were fasted for 24 hours and each group were entered to a heater to dilate their veins for 5 minutes, after their tails were cut off and the blood taken into a tube and labeled for plasma creatinine measurement.

The data after measurement were analyzed statistically using multiple comparison tests with dunnett T3 because the data distribution
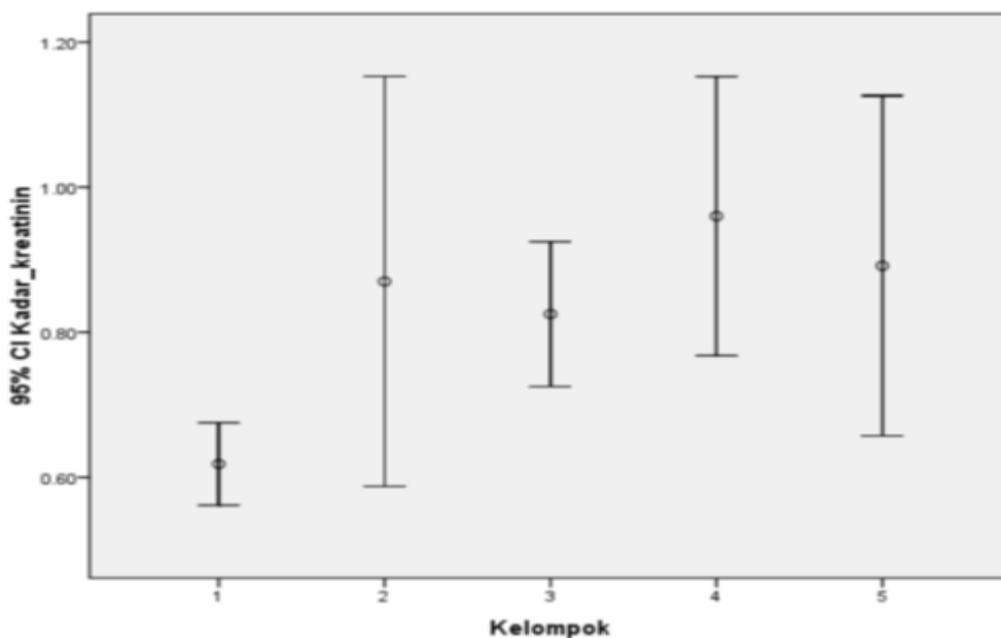

Figure 1 Creatinine Levels Average

Table 2 The Creatinine Levels with Normality Test

	Kolmogorov–Smirnov*			Shapiro–Wilk		
	Statistic	Df	Sig.	Statistic	Df	Sig.
Group1	.260	6	.200**	.847	6	.148
Group2	.211	6	.200**	.887	6	.304
Group3	.270	6	.195	.915	6	.472
Group4	.188	6	.200**	.942	6	.678
Group5	.159	6	.200**	.986	6	.976

Note: *: Lilliefors Significance Correction, **: This is a lower bound of the true significance, Information:$p \geq 0.05$ --> data distributed normally, $p < 0.05$ --> data distributed abnormally

Table 3 ANOVA Test

Creatinine levels	Sum of Squares	Df	Mean Square	F	Sig.
Between Groups	.403	4	.101	2.992	.038
Within Groups	.841	25	.034		
Total	1.243	29			

was not homogeneous.

Results

After the creatinine levels measurement on control group and treatments for 14 days, the results were obtained with the lowest creatinine levels of group I (negative) at 0.62 mg/dL, while the largest creatinine levels were found in group IV (treatment 19%).

The positive control group had wider ranges compared to other groups which means that the larger the range, then the more deviating from the truth (Figure 1). The creatinine

levels were normally distributed with p–value ($p > 0.05$)[8]

Homogenity test showed p–value 0.041 ($p < 0.05$) (Table 2). The data of creatinine levels was not homogenous. There was a significant difference on the average of creatinine levels among the groups with (p–value <0.05)[8](Table 3).

Multiple comparison dunnett T3 was done to recognize the difference among each group because the data was not homogenous.

According to the result, between control groups and treatments had significant difference. This meant that the infusion of

Table 4 Dunnett T3 test

Comparison Groups		Significance
1	2	0.379
1	3	0.014
1	4	0.035
1	5	0.181
2	3	1.000
2	4	0.997
2	5	1.000
3	4	0.690
3	5	0.997
4	5	0.999

Note: p–value> 0.05: No significant difference, p–value< 0.05: Significant difference, Group 1: Negative Control, Group 2: Positive Control, Group 3: 1 time dose treatment, Group 4: ½ time dose treatment, Group 5: 2 times dose treatment

breadfruit caused an increase of creatinine levels compared to the control groups, with the comparison of group 1 and 2 gave a significance of 0.379 ($p > 0.05$)There was no significant difference between both groups. Comparison of group 1 and 3 ($p < 0.05$) gave a significant increase among the groups. There were no significant differences on average creatinine levels in Group 2 and 5 ($p > 0.05$).

Comparison between group 2 and groups 3 and 4 ($p > 0.05$), group 3 and groups 4 and 5 ($p>0.05$) and comparison of group 4 and 5 ($p > 0.05$) showed that there was no significant difference on the average creatinine levels between the positive group and treatments.

Discussions

From the result of this study, the breadfruit leaves test did not have effects to reduce the creatinin of acute renal failure model rats which were induced by Gentamycin and Piroxicam per intraperitoneal.

According to a research done by Walker and Shah[9], Gentamycin and piroxicam can induce renal failure after induced for 7 days. Gentamycin and piroxicam also cause an increase in reactive oxygen metabolites in the renal cortex mitochondria which causes lipid hidroxyperoxydase. This peroxydase causes the damage on renal tissue, and inflammation as such that causes renal dysfunction. Renal dysfunction can be seen from creatinin examination which is one of the indicators to assess renal failure and confirms a person as having renal failure. In this study, there is an evidence on positive control group after induced by gentamycin and piroxicam compared to negative control group.

Cells or tissues experience damage and cause renal dysfunction because of these oxidative stress inducing molecules can be inhibited by a compound called anti–oxidants.

Anti–oxidants can inhibit or prevent oxidation in forming free radicals. One of the anti–oxidants contained in breadfruit leaves is flavonoid indonesianin. Flavonoid can prevent covalent bonding on lipid. Lipid peroxidation is inhibited which can lessen or prevent renal damage.

According to the statistical tests between control and treatment groups, there were no significant difference with a p-value ($p > 0.05$) between group 2 and 3, 2 and 4, and group 2 and 5, because the average creatinine levels on treatment groups were higher than the control. From this study, it was known that the

anti–oxidant effects on breadfruit leaves were considered as an agent to reduce renal damage but it had not been scientifically supported.

This study is not in accordance with the research by Saija et al.[10] because it did not prove that the flavonoid content in breadfruit leaves had anti–oxidant effects that can lessen renal failure cells induced by nephrotoxic agents and the anti–oxidant effects of breadfruit leaves are dubious in reducing the renal damage.

From this study, it was also possible that there were toxic effects from the breadfruit leaves that can increase renal damage and the creatinine levels on the experimental animals experiencing acute renal failure, It can be concluded that breadfruit leaves infusion did not effect to reduce blood creatinine levels on acute renal failure model rats after induced by gentamycin and piroxicam.

However, breadfruit leaves infusion administration for 7 days had an effect on creatinin levels of rats. The creatinine levels on groups which were given breadfruit leaves infusion at dose 4.25%, 9.5%, and 19% were higher and significantly different compared to the negative control group. The highest creatinine levels were obtained from the group which were given an infusion dose of 4.25%

As a conclusion, the breadfruit leaves infusion did not effect to reduce blood creatinine levels on acute renal failure model rats after induced by gentamycin and piroxicam. The breadfruit leaves infusion administration for 7 days had an effects on creatinin levels of rats according to the dosage and the duration of administration.

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