Porang flour (Amorphophallus oncophyllus) with and without soaking of keji beling extract increases the value of ureum on toxicity test in wistar rat (Rattus norvegicus)

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ABSTRACT

Background: The porang tuber (Amorphophallus oncophyllus) is a functional food containing glucomannan that has many advantages in health. However, porang flour can not be consumed, because the high content of calcium oxalate that have the risk on kidney disease. It can be reduced by physical or chemical treatment. Keji beling (Strobilanthes crispa L. Blume) has been proved for its function in dissolving the calcium oxalate, but its uses in decreasing of calcium oxalate has not been studied yet.

Objectives: To evaluate the effect of porang flour on ureum levels of wistar rat blood in acute toxicity test. **Methods**: The research was experimental with pre and post without control group design. The samples were 20 female Wistar rats, aged 8-10 weeks with body weight of 100-180 grams. Rats were divided into 4 groups of treatment those were native porang with the dose of 2000, 5000 mg/kg of body weight, porang flour with soaking of extract at the dose 2000 and 5000 mg/kg of body weight. Porang was incorporated orally into the mouth of rats after 18 hours of adaptation. At the 24th and 72nd hours after treatment, the bloods were collected and analyzed for their ureum levels.

Results: The statistical test showed that there was an effect of porang flour with and without soaking of keji beling extract before and after treatment on ureum level at the dose of 2000 and 5000 mg/kg body weight, however there was no significant difference ureum level of the same dose at 24^{th} or 72^{nd} hours, except on the dose of 2000 mg / kg weight at the 72^{nd} hour. Results of observation between the 24^{th} hour compared to the 72^{nd} hour showed that there was no significant difference of urea value (p> 0.05). Increased levels of ureum was influenced by the calcium oxalate content contained in porang flour. In TPM, ureum level was higher than that in TPK.

Conclusions : The increase in urea levels was still in normal range, therefore porang flour is still safe for consumption.

KEYWORDS: acute toxicity, porang flour, urea, keji beling

INTRODUCTION

Porang tuber (*Amorphophallus oncophyllus*) is local plant belonging to the Araceae family and can grow in almost all forest in Indonesia. Porang contains glucomannan that is 15-64% (dry base) (1). It also contains other carbohydrate, such as starch, polyose, and crude fiber that are approximately 2%, 14%, and 8.0%, respectively (2). The high content of glucomannan or other polysaccharide in porang is potential to be developed in food industry and health science (3).

Glucomannan had many advantages in health, such as improve the digestive function, immune system, and also lowered the cholesterol, blood sugar, and body weight (4). High fiber content could also reduce cholesterol levels in the blood because its ability to bind the fat (5–6).

Porang could not be consumed directly, because of calcium oxalate presence. It may cause the itching when consumed and trigger the occurrence of kidney stones (7). Kidney damage was also may be occurred. It was characterized by the high level of protein in the urine (proteinuria or albuminuria), blood in urine (hematuria) and elevated levels of urea or creatinine (residual production of protein metabolism) in the blood (8).

Ureum is the last product of nitrogen metabolism synthesized from ammonia, carbon dioxide, and aspamarated amit nitrogen. The nitrogen balance in urea excretion is approximately 25 mg/day. Renal disease was usually accompanied with the decrease of glomerular filtration rate leading to high plasma urea. High plasma urea was an abnormal feature of kidney disease (9,10).

There are many procedures that have been developed to decrease the levels of calcium oxalate in porang. However, it seems not effective, because the oxalate residue is still high. In this research, the alternative way to reduce the calcium oxlate in porang was studied by using keji beling (Strobilanthes crispa L. Blume). Keji beling contains alkaloids, saponins, flovonoids, potassium and polyphenols. Potassium in hepatic diuretics is strong and can dissolve stones from calcium salts. Previous study proved that keji beling could dissolved calcium and oxalate in the urine (12). However, there is still limited study about the effect of purified porang consumption on the ureum level in the blood as one of kidney's damage in the body. The objective of this study was to evaluate the effect of porang flour on ureum levels of wistar rat blood in acute toxicity test.

MATERIALS AND METHODS

Porang was obtained from Madiun, East Java. It was dried, grinded, and decreased the content of calcium oxalate by mechanic treatment. Porang was then purified with the soaking with kejibeling ethanol extract for a day with 2 times of rinsing.

This was an experimental study with pre- and post- without control group design. The subjects were 20 female Wistar (*Rattus novergicus*) rat, that was divided into 4 groups. They were TPM (native porang flour) dose of 2000 (TPM 2000) and 5000 mg/kg body weight (TPM 5000) and TPK (purified porang flour) dose of 2000 (TPK 2000) and 5000 mg/kg of body weight (TPK 5000). Rats were chosen with the mean of weight 100-180 gram and age of 8-10 weeks.

Rats were feed with laboratory standard diet. In the beginning of study, rats were adapted for 18 hours. TPM or TPK was given orally to all of rats after adaptation. Body weight was measured on the first and the end of study. The ureum was analyzed from blood serum and analysed a while after the treatment, and 24 also 72th hours after the treatment.

The procedure of toxicity test was based on Peraturan Kepala Badan Pengawas Obat dan Makanan Republik Indonesia with the No 7, year 2014 about the *in vivo* non-clinical toxicity test. The toxic symptoms were qualitative observed from the rat behavior,

RESULTS AND DISCUSSION

The research was firstly done by doing preliminary study to know the dose of porang that should be given. By using a starting dose of 300 mg/ kg body weight of rats, there was no toxic symptoms and mortality in all samples. Therefore, the dose was increased to a maximum dose, i.e. 2000 and 5000 mg/kg of body weight.

Qualitative observation

After giving the porang orally, observation on the behavior and toxic symptoms were conducted. There was no dead rats in each treatment group and no toxic symptoms that emerged after intervention (11).

Body weight of rat during the study

The body weight of rat was increased after 72 hours observations. The increase did not cause by porang consumption, but due to rat growth and feed consumption during the study (**Figure 1**).

Normality test of the urea content

The overall data of urea content after intervention was tested for its distribution using SPSS using Shapiro-wilk test. The results of normality test data of blood ureum content of rats was presented in **Table 1**.

Table 1 showed that normality test resulted in p-value<0.05. It meant that both of data had normal distributions.

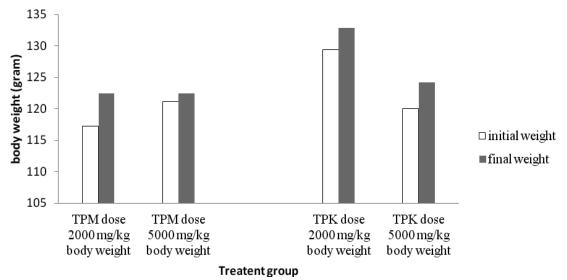


Figure 1. Body weight of rats at the begin and the end of study (72 hours)

Table 1	Normality	test of	ureal	content
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Treatment	S	Shapiro-w	vilk
group	Statistic	Df	p-value
TPM	0.930	10	0.449
TPK	0.942	10	0.576

Note: TPM, native porang flour TPK, keji beling purified porang flour

The effect of porang consumption on the urea levels after 24th hour of treatment

Table 2 showed that all of the rats had normal urea levels (11.00 - 19.90 mg/dL). There were no significant different between the urea level of all groups at the initial study. However, after 24th hour of treatment there were found the effect of porang

Table 2. Effect of porang consumption on ure	a
levels of rat blood after 24th hours of treatmen	٦t

Group	Urea levels of rat blood Group (mg/dL) Ρ Δκ			
	Before	After		
TPM 2000	10.71 ± 0.90ª	11.52 ± 0.26ª	0.08	0.81
TPM 5000	11.11 ± 0.51ª	13.64 ± 0.48 ^b	0.00	2.53
TPK 2000	10.85 ± 0.42^{a}	11.65 ± 0.39ª	0.08	0.80
TPK 5000	10.84 ± 0.48^{a}	13.64 ± 0.41 ^b	0.00	2.80

Note: Superscript within the same coloumn with the same letters are not significantly different (p>0.05). ΔK , different of urea level. TPM 2000&5000, native porang flour with the dosage 2000 & 5000 mg/kg body weight. TPK 2000 & 5000, keji beling purified porang flour with the dosage 2000 & 5000 mg/kg body weight.

consumption (p<0.05). Among the same type of porang (TPM or TPK) group, the increase of dose gave the improvement of urea levels. Meanwhile, the result of comparation between urea levels of the different type of porang when the same dose used showed no different value. At higher doses, blood urea levels become higher. This is because in large doses the amount of calcium oxalate in porang flour is more numerous than the smaller dose. In addition to calcium oxalate, there is also a protein content in porang flour that affects elevated levels of urea.

Calcium oxalate will be settle and accumulated in the body and may affect the kidneys work. In acute conditions, calcium oxalate causes anatomic pathology changes in the form of gastric inflammation in TPM dose 5000 mg/dL and congestion, hemorrhage in the kidney resulting in decreased renal function (11,12).

Decrease in kidney function can be seen from the increase of blood ureum level. In the long term, calcium oxalate will form crystals resulting in calcium oxalate stones formation. Calcium oxalate will affect kidney function. If there is impaired renal function due to the presence of calcium oxalate, it will increase the level of urea. Ureum can be an indication of kidney disorders.

The effect of porang consumption on the urea levels after 72th hour of treatment

Table 3 shows the effect of porang consumption on urea levels of rat blood after 72th hour of treatment. At the begin of study, all of urea levels were the same (p>0.05). It was also the same urea levels when the samples were treated with different type of porang although the dosages (TPM and TPK 2000) were the same. However, at the higher dosage (5000 mg/kg body weight), the urea levels were different (p<0.05). This is caused by the more content of calcium oxalate in native porang flour than that in purified porang flour.

Table 3. Effect of porang consumption on urea levels of rat blood after 72nd hours of treatment

Group	Urea levels of rat blood Group (mg/dL)			ΔK
	Before	After	-	
TPM 2000	10.71 ± 0.90ª	11.47 ± 0.26 ^a	0.00	0.76
TPM 5000	11.11 ± 0.51ª	14.61 ± 0.48⁵	0.00	3.50
TPK 2000	10.85 ± 0.42^{a}	11.54 ± 0.39ª	0.01	0.69
TPK 5000	10.84 ± 0.48^{a}	13.84 ± 0.41°	0.00	3.00

Note: Superscript within the same coloumn with the same letters are not significantly different (p>0.05). ΔK , different of urea level. TPM 2000&5000, native porang flour with the dosage 2000 & 5000 mg/kg body weight. TPK 2000 & 5000, keji beling purified porang flour with the dosage 2000 & 5000 mg/kg body weight.

The difference of urea level measured at $24^{\mbox{th}}$ and $72^{\mbox{nd}}$ hour

Differences in ureal levels were compared to determine changes in ureal content between at the 24th hour and the 72nd hour. The results of the ratio of urea can be seen in **Table 4**.

Table 4. Levels of urea after treatment between the24th hour and 72nd hour

Treatment	Urea leve	P-value		
group	24 th hour	72 nd hour	P-value	
TPM 2000	11.52	11.47	0.38	
TPM 5000	13.64	14.61		
TPK 2000	11.65	11.54		
TPK 5000	13.64	13.84		

In all samples, there were no different urea levels measured at 24 and 72 hour after treatment

(p>0.05). It meant that there was no increase of urea levels at the prolonged period of analysis.

CONCLUSIONS AND SUGGESTIONS

The consumption of porang was still safe for the body that was found from the normality levels of urea in the blood. The consumption of keji beling that followed in porang after the soaking process may also have the advantage effect in the body. Therefore, the subsequent researcher was suggested to study it.

ACKNOWLEDGEMENT

The authors gratefully thanked to Alma Ata Centre for Healthy Life and Food (ACHEAF) for the supporting facilities and accompaniment.

REFERENCES

- Supriati Yati. Keanekaragaman Iles-Iles (Amorphophallus spp.) dan Potensinya Untuk Industri Pangan fungsuonal, Kosmetik, Dan Boietanol. 2016;
- Ohtsuki T. Studies on Reverse Carbohydrates of Flour Amorphophallus Species, with Special Reference of Mannan. Bot Mag Tokyo. 1968;81:119–26.
- Zhang, Y., Xie, B., dan Gan X. Advance in Application of Konjac Glucomannan and its Derivatives. Carbohydr Polim. 2005;60:27–31.
- Faridah, Anni et al. Optimasi Produksi Tepung Porang Dari Chip Porang Secara Mekanis Dengan Metode Permukaan Respon. 2009.
- Katsuraya, K., Okuyama, K., Hatanaka, K. OK, Sato, T., dan Matsuzaki K. Contitution of Konjac Glucomannan: Chemical Analysis and 13C NMR Spectroscopy. Carbohydr Polym. 2003;53:183–189.
- Yang, X. H., Zhu, W.L., dan Yan, J.F. A Time-Temperature Rheological Study of Konjac Glucomannan Hydrocolloid. J Biomater Sci. 2006;17((1–2),):53–59.
- 7. Natalia ED, Widjanarko SB, Ningtyas DW. Acute Toxicity Test Of Glucomannan Flour (A.

Porang flour (Amorphophallus oncophyllus) with and without soaking of keji beling extract increases the value of ureum 97

muelleri Blume) Toward Potassium Of Wistar Rats. 2014;2(1):132–6.

- Reksodiputro, H dan Prayoga N. Eritropoesis dalam Ilmu Penyakit Dalam. III. Jakarta: FKUI; 2001. 494 p.
- 9. Murray. Biokimia Harper. 27152nd–94th ed. Jakarta: .EGC; 2009. 152-94 p.
- Widmann FK. Tinjauan klinis atas hasil pemeriksaan laboratorium. kresno Boedina S, Soebrata A, editors. Jakarta: EGC; 1995.
- Hasanah U. Uji Toksisitas Akut Tepung Porang (Amorphophallus oncophyllus) Dengan dan Tanpa Maserasi Ekstrak Keji Beling Serta

Histopatologi Ginjal Pada Tikus Wistar (Rattus norvegicus). Universitas Alma Ata, Yogyakarta; 2017.

- Ernawati. Pengaruh Pemberian Tepung Porang (A.onchophyllus) Tanpa Dan Dengan Maserasi Ekstrak Etanol Daun Keji Beling Terhadap Kadar Kreatinin Darah Dan Histopatologi Lambung Tikus Wistar (Rattus norvegicus) Pada Uji Toksisitas Akut. Universitas Alma Ata Yogyakarta;
- Sudarsono, D. Gunawan SW, I.A. Donatus dan P. Tumbuhan Obat II. Yogyakarta: UGM Pusat Studi Obat Tradisional UGM; 2002.