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Porang (*Amorphophallus oncophyllus*) flour with *Strobilanthes crispus* maceration reduced blood glucose level in streptozotocin-induced diabetic rats

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Porang (*Amorphophallus oncophyllus*) flour with *Strobilanthes crispus* maceration reduced blood glucose level in streptozotocin-induced diabetic rats

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Abstract

BACKGROUND: Diabetes mellitus (DM) is a metabolic disease indicated by hyperglycemia. Dietary regulation is an alternative way to control blood glucose levels. *Amorphophallus oncophyllus* or porang is a local tuber that has low glycemic index due to glucomannan content. Its combination with *Strobilanthes crispus* (SC) that is rich in antioxidant is potential for diabetes treatment.

OBJECTIVES: This study aimed to know the effect of porang flour macerated with SC on blood glucose levels in diabetic rats.

METHODS: Thirty-five Wistar (*Rattus norvegicus*) rats were divided into 5 groups, based on the types of diets. They were standard AIN-93 diet for NC group (normal/negative control, non-diabetic), PC group (positive control, streptozotocin (STZ)-induced diabetic), GB group (STZ-induced diabetic, n^gdicated with glibenclamide 100 mg/200 g body weight (BW)). The treatment groups were fed with a modified AIN-93 diet substituted its fiber with 11% (equal with 1.1 g/200 g BW) of porang (NP) group and SC-macerated porang (SP) group. Food intake, stools, and BW were recorded during the study, while blood glucose levels were measured before, 6 d after the induction of STZ, and at the end of the study (14 d of treatments periods). The data were statistically analyzed by one-way analysis of variance (ANOVA) continued with Duncan's multiple range test.

RESULTS: Feed intake of rats during 14 d of treatment were almost the same and influence the bodyweight. After the STZ induction, the body weight of rats seemed down, but statistically not different. This weight loss may be better controlled in treatment groups because of the role of glucomannan content in the improvement of glucose metabolism, especially in NP and SP. Stools looked normal in consistency and moisture and confirmed that there was no diarrhea incident. The role of glucomannan also proved to decrease blood glucose levels in NP and SP groups. SP groups showed the best performance in decreasing glucose levels due to the addition of SC as the source of antioxidant.

CONCLUSION: Porang had antidiabetic activity that was comparable with glibenclamide as a commercial drug. It was forced by combination with SC that contains many antioxidants. It should be studied further to optimize its antidiabetic potency and its uses as functional food or nutraceuticals.

Keywords: *Diabetes mellitus*, *blood glucose*, *Amorphophallus oncophyllus*, *Strobilanthes crispus*, *porang*

Introduction

Diabetes mellitus (DM) is a metabolic disease indicated by hyperglycemia that is caused by insulin function or secretion disorder (1,2). The bad impact of DM is very influential on other comorbidities, like multiple secondary complications micro- and macro-vascular, also neuropathic disorder (3). If it is not taken seriously, it may decrease the quality of human resources and increase health costs.

Dietary regulation is a way that patients can do to control blood glucose levels, in addition to consuming hypoglycemic drugs. A diet with foods that have a low glycemic index had a positive effect on lowering blood glucose levels (4). *Amorphophallus oncophyllus* or known as porang is a kind of konjac tuber that is widely cultivated in Indonesia. It has low glycemic index (5) because of its glucomannan content. The study about the health effect (6,7) and its application has been carried out (8–14), but its uses as raw porang flour are still very rare due to the limited availability of calcium oxalate-free flour.

Porang flour macerated with *Strobilanthes crispus* (SC) has been studied for its safety (15) and low content of calcium oxalate (16). The SC content that is rich in flavonoid and phenolic acid (17) in porang potentially to add benefits in diabetes treatment. However, the study to prove that potency has not been studied. The aim of this study was to know the effect of porang flour macerated with SC on blood glucose levels in diabetic rats.

Methods

Plant material

Porang tuber was obtained from the farmer in Madiun, East Java. The tuber was cleaned from the sand, sliced, and dried. It was then ground and sifted 40 mesh to make powder. The flour was macerated with SC as described in Patent Application No. S00202006668 (16).

Experimental animals

This study was carried out according to Health Research Ethics Committee Universitas Alma Ata with the Ref No: KE/AA/VI/273/EC/2017. Thirty-five Wistar (*Rattus norvegicus*) rats with the category of eight weeks of ages and the body weight of 121-159 g were used in this study. The rats were divided into 5 groups, based on the types of diets. Each group was treated with standard AIN-93 diet (18) for NC group (normal/negative control, non-diabetic), PC group (positive control, streptozotocin (STZ)-induced diabetic), and GB group (STZ-induced diabetic, medicated with glibenclamide 100 mg/200 g bodyweight (BW)). The treatment groups were fed with a modified AIN-93 diet substituted its fiber with 11% (equal with 1.1 g/200 g BW) of porang (NP) group and SC-macerated porang (SP) group. They were individually housed in wire cages under an ambient temperature with 12 h light-dark cycle. Food intake, stools, and bodyweight (BW) were recorded during the study, while blood glucose levels were measured before, 3 d after the induction of STZ, and at the end of the study (14 d of treatments periods).

Induction of diabetes, blood preparation, and biochemical analysis

The intraperitoneal injection of nicotinamide 110 mg/kg BW was done before diabetic inducement with 8 mg/200 g BW of STZ. The rats were categorized as diabetic when glucose level was more than 126 mg/dL (1). The blood glucose levels were determined by GOD-PAP (glucose oxidase-peroxidase aminoantypirin) enzymatic methods from the rat plasma samples. This plasma was prepared from centrifugation of blood samples (at 400 rpm for 15 minutes) that were drawn from the vena retroorbital part by microcapillary technique.

Statistical analysis

Data were presented as mean±standard deviation (SD). They were analyzed by one-way analysis of variance (ANOVA) continued with Duncan's Multiple Range Test (DMRT) at $p < 0.05$ to compare

the data between groups. Paired T-test analysis was also used to compare the data between before and after the treatment. All of the analysis used statistical package for the social sciences (SPSS) software (version 16.0 SPSS Inc., Chicago, USA).

Results and Discussions

Feed intake, body weight of rat, and observation of stools

Feed intake of rats during 14 d of treatment periods could be seen in Table 1. The amount of intake was almost the same every day with the mean value recorded in the range of 6.18-9.59 g/dL. Among the groups, NC showed the lowest amount of feed intake and was statistically different from others ($p < 0.05$). It may be due to NC rats' behavior that looked more active than other groups. It caused some feeds were fallen or be mixed in the drinking water, so that could not be weighed. The feed intake of NC was also statistically different with PC ($p < 0.05$). It indicated that the inducement of diabetes influenced the feed intake or rat's appetite.

Feed intake relates to the body weight of rats. The body weight of rats during the study could be seen in Table 1. During 14 d of treatment, the bodyweight of rats seemed down, but not statistically different ($p > 0.05$). This weight loss value increased after STZ induction that may be caused by insufficient insulin. It leads to an inability of glucose to be used as energy, therefore the availability in the body was provided by fat catabolism. If it was happened continuously can lose body weight (19,20,21). However, the body weight in treatment groups (GB, NP, and SP) could be controlled by increasing the insulin sensitivity resulting in the improvement of glucose metabolism (22) for GB groups or by the role of glucomannan as fiber in NP and SP groups that fulfilled the intestine and decrease the feed intake (6,23). It may also decrease postprandial glucose and improve insulin sensitivity (24,25). The previous study has also been studied for the potency of porang glucomannan as a prebiotic that increased short-chain fatty acid (SCFA) in the colon (6) leading to the improvement of glucose and lipid metabolism (23,26) and resulting in controlling of body weight (24).

The inability of body weight to increase in this study was also confirmed by the observation of stools that could be seen in Table 1, especially to know whether there was diarrhea or other disorder in gastrointestinal. The data showed that stools in all groups were in normal moisture and consistency. It meant that there was no diarrhea, instead there were also no difficulties in defecating process. The previous study proved that glucomannan absorbed much water and influenced the dry and wet stool weight, the defecation frequency, and colonic flora in stool (27). The porang consumption and its combination with SC did not affect the gastrointestinal response.

Table 1: Feed intake, body weight, and stool character of rats during 14 d of treatment periods

| Groups | Feed intake (g) | Stools character | | Body weight (g) | | $\Delta.K$ |
|--------|-----------------------|--------------------------|---------------|---------------------|--------------------|--------------------|
| | | Water content (%) | Consistency | Before intervention | After intervention | |
| NC | 6.18±0.8 ^a | 39.40 ±2.2 ^a | Slightly hard | 133.2±7.3 | 130.4±13.0 | -2.8 ^a |
| PC | 9.56±1.1 ^b | 76.20 ± 5.3 ^a | Soft | 134.4±11.8 | 123.4±13.8 | -11.0 ^a |
| GB | 9.59±1.9 ^b | 71.40 ± 3.0 ^a | Soft | 141.2±9.9 | 133.0±10.3 | -8.2 ^a |
| NP | 9.64±0.6 ^b | 78.80 ± 5.1 ^a | Soft | 150.0±6.0 | 142.6±27.6 | -7.4 ^a |
| SP | 8.84±0.8 ^b | 70.20 ± 2.8 ^a | Soft | 138.8±13.4 | 132.80±4.1 | -6.0 ^a |

*Different superscript letter in the same columns indicated significantly different result ($p < 0.05$). NC group (normal/negative control, non-diabetic, AIN-93 diets), PC group (positive control, streptozotocin (STZ)-induced diabetic, AIN-93 diets), GB group (STZ-induced diabetic, AIN-93 diets, medicated with glibenclamide), NP group (STZ-induced diabetic, AIN-93 modified diets with 11% porang flour), SP group (STZ-induced diabetic, AIN-93 modified diets with 11% SC-macerated porang flour).

Blood glucose levels

Table 2 showed that the initial blood glucose levels were in the range of 50-135 mg/dL. It meant that all intervention groups were normal. The measurement of initial blood glucose levels of rats aimed to ensure that the rats were in normal condition. After being induced with STZ, blood glucose levels in diabetic groups increased in the range value between 217-244 mg/dL. The rats with blood glucose levels more than 126 mg/dL were included in diabetic groups (1). STZ induced diabetes by damaging β -cells through the production of radicals NO that may block the Fe-containing enzymes and the breakdown of secondary radicals caused peroxide of lipids, reduced antioxidant, and led to DNA damage (28, 29).

The intervention of commercial drug (GB group) and porang flour (NP and SP) significantly decreased blood glucose levels ($p<0.05$), although they were still above 126 mg/dL, especially for NP and SP (Table 2). GB worked by stimulating the insulin secretion in the pancreas gland through sensitizing of β -cells allowing glucose-induced changes in the synthesis and release of insulin (22). NP and SP worked as the fiber that has high water absorption and is very viscous (6) that may decrease food absorption in the small intestine and decrease postprandial glucose and insulin secretion leads to the improvement of insulin sensitivity (24,25). This was also confirmed by the histopathological study of the pancreas that showed the maintaining of pancreatic structure in diabetic rats treated by konjac glucomannan (24).

This study also presented that SP groups had a similar antihyperglycemic ability with GB. The role of fiber in porang was strengthened by the existence of SC. SC is the source of flavonoids and phenolic acids that possessed antidiabetic activity in diabetic rats (17). SC was also useful in decreasing calcium oxalate in porang and had no acute toxicity result (15) which allows its use as a functional food.

Table 2: Blood glucose levels of rats in various treatment during 14 days of treatment

| Groups | Blood glucose levels (mg/dL) | | | ΔK |
|--------|------------------------------|---------------------|--------------------|--------------------|
| | Before STZ induction | After STZ induction | After intervention | |
| NC | 69.6 \pm 0.9 ^b | 67.17 \pm 1.4 | 67.4 \pm 2.0 | 0.67 ^a |
| PC | 66.7 \pm 1.1 ^a | 244.4 \pm 7.4 | 215.7 \pm 6.1 | 1.26 ^a |
| GB | 70.4 \pm 2.2 ^b | 217.40 \pm 7.8 | 121.8 \pm 5.3 | -96.0 ^b |
| NP | 68.3 \pm 1.1 ^{ab} | 217.4 \pm 9.2 | 156.8 \pm 4.3 | -62.9 ^c |
| SP | 68.7 \pm 1.9 ^{ab} | 224.5 \pm 7.3 | 133.2 \pm 1.9 | -91.3 ^b |

* Different superscript letter in the same column indicated significantly different result ($p<0.05$). ΔK showed the different value between 0 d and 14 d using T-test. NC group (normal/negative control, non-diabetic, AIN-93 diets), PC group (positive control, streptozotocin (STZ)-induced diabetic, AIN-93 diets), GB group (STZ-induced diabetic, AIN-93 diets, medicated with glibenclamide), NP group (STZ-induced diabetic, AIN-93 modified diets with 11% porang flour), SP group (STZ-induced diabetic, AIN-93 modified diets with 11% SC-macerated porang flour).

Conclusions

This study concluded that porang had antidiabetic capacity. This capacity was forced by the presence of SC and proved the comparable result with the glibenclamide as a commercial drug. Further study is needed in the optimization of doses to improve the antidiabetic capacity and probability uses as functional food or nutraceuticals.

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