Antidiabetic Activities Analysis by Oral Glucose Tolerance Test in Rats

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ABSTRACT

In this study, we conducted an oral glucose tolerance test (OGTT) so as to compare antidiabetic activities of general potatoes, purple-flesh potatoes, and potato pigments in rats at various concentration levels. After allowing the rats to abstain from food for 12 hours, 10%/20% general potato, purple-flesh potato, and potato extract was orally administered to rats at 100 and 500 mg/kg concentrations. The blood glucose level was measured after an hour. Then, immediately, 1.5 g/kg of sucrose was administered through the abdominal cavity and the blood glucose measured after 30, 60, 120, and 180 minutes. 20% purple-flesh potato group and 10% general potato group, both 100 and 500 mg/kg, showed a significant concentration-dependent decrease in blood glucose levels after 30 minutes. The 100 mg/kg potato pigment group also showed a statistically significant decrease after 30 minutes. In conclusion, administration of 10% general potato, 20% purple-flesh potato, and potato pigment can reduce blood glucose level in an OGTT using rats.

(Key words : OGTT, Potato, Antidiabetic Activities, Rats)

INTRODUCTION

A potato (*Solanum tuberosum* L.) has a short period of growth, high production per unit area, and relatively good adaptability to environment, which makes it widely cultivated around the world (Park *et al.*, 2009). Although the colors of potato tubers are mainly white or light yellow, there is a variety of colored potatoes, containing anthocyanin pigments such as red and purple depending on genes involved in deciding tuber colors.

Colored potatoes with red or purple tubers are known to contain a large amount of anthocyanin pigments and phenolic acid (Brown, 2005). Anthocyanin pigments, contained in various parts of higher plants such as a flower, a fruit, a stem, leaves, a root, and so on, are water-soluble flavonoid pigments that produce red, purple, or blue color. The pigment is expressed in different colors according to parts of a plant. Recently, more research efforts are being put into development of food products using anthocyanin and its evaluation as various physiological active effects of the pigment, including anti-oxidation related with chronical diseases, anti-inflammation, anti-carcinogenic effects, prevention of artery hardening, inhibition of lipid peroxidation, protection of DNA cleavage, and so on, have been reported (Acquaviva *et al.*, 2003; Lazze *et al.*, 2003; Lefevre *et al.*, 2004).

The prevalence rate of diabetes in Korea sharply rose compared to that of developed countries in the same period (Ahn, 2010). Diabetes is fatal not only in that it causes disorders of carbohydrate metabolism, preventing the body from using glucose, but in that it causes various complications. Though diabetes-related complications are linked with pathophysiology in a complex manner, it has been known that oxidative stress caused by hyperglycemia plays a major role, which has motivated research on the influence of anti-carcinogenic substances on diabetes (Wolff, 1993; Baynes and Thorpe, 1996). Moreover, as the level of income has greatly improved and public concern over prevention/treatment of diabetes has increased, it has become more important to discover natural substances that can help to alleviate diabetes.

In this study, therefore, we examined antidiabetic activities by conducting an oral glucose tolerance test (OGTT) at various levels of general potatoes, purple-flesh potatoes, and potato pigments in rats so as to provide the basis for developing anti-diabetic food products and promoting use of colored potatoes.

MATERIALS AND METHODS

Materials

The potatoes used in this experiment were cultivated in Potato Valley (Chuncheon, Gangwon-do). We used both white potatoes (Daeseo) and purple-flesh potatoes (Bora Valley). The potatoes were washed clean, sliced, freeze-dried, and crushed to 200 mesh. The potato pigment extract was obtained by shaking the freeze-dried colored

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potatoes in 80% EtOH for 12 hours at room temperature, repeating to filter (Whatman No. 2) the contents 3 times and, finally, vacuum evaporating (Rotary evaporator N-1000, EYELA) at 40℃. All the reagents were extra pure reagents.

Animals

We purchased 78 male rats (CrjBgi:CD(SD)IGS), 7 weeks old, from Orient Co., Ltd. (Gapyeong-gun, Gyeonggi-do) and nurtured/monitored them for 7 days under the lab conditions provided by the Safety Evaluation Department of the Korea Testing & Research Institute. This experiment complied with the protocol of experimental animal use and management established by the Korea Testing & Research Institute.

Oral Glucose Tolerance Test

Table 1 shows the oral glucose tolerance test plan. After allowing the rats to abstain from food for 12 hours, each experimental substance was orally administered. The blood glucose level was measured after an hour. Immediately, 1.5 g/kg of sucrose was administered through the abdominal cavity and the blood glucose level measured after 30, 60, 120, and 180 minutes. Of the test substances, general diet was formulated based on AIN-93 (Reeves *et al.*, 1993) while purple-flesh and general potatoes by mixing freeze-dried potato powder with 10% and 20% of the rat food. As to the potato pigment, concentrated potato extract was dissolved in sterile normal saline so as to achieve 250 mg/kg body weight. Test substances were dissolved in sterile normal saline according to the dosage as described in Table 1. Blood was taken from caudal veins using a lancet. The collected blood was exposed to blood glucose test strip (Taidoc Technology Corp., Taiwan) according to the manufacturer's directions and the blood glucose level measured by a glucometer (COSMO sensor 2, Taidoc Technology Corp., Taiwan). All measurement was done 3 times.

Statistical Analysis

All the test results were analyzed by SAS program (version 9.1, SAS Institute Inc., Cary, NC) to obtain the average and standard deviation. We used Student's *t*-test to compare the control group and the test group. When compared with the control group, the result was deemed significant if $p<5\%$.

RESULTS

Table 2 shows the result of OGTT conducted in order to compare antidiabetic activities of general potatoes, purple-flesh potatoes, and potato pigments. As to the blood test, the value of each rat was measured and the value before the administration of sucrose is deemed 100%. In all groups, changes over time in blood glucose level reached the highest point 30 minutes after the administration of sucrose and then the level kept decreasing. Compared to the control group, the 20% purple-flesh potato group and 10% general potato group, both 100 and 500 mg/kg, showed a significant concentration-dependent decrease in blood glucose level after 30 minutes. The 100 mg/kg potato pigment group also showed a statistically significant decrease after 30 minutes. The blood glucose level in the 10% purple-flesh potato and general potato groups significantly dropped 180 minutes after the administration of sucrose. Particularly, the result in the 10% general potato group was concentration dependent.

Table 1. Experimental design

Group	Treatment	Dose (mg/kg, P.O.)	No. of animal	Animal No.
G1	Saline	-	7	1101~1107
G2	General diet	100	7	1108~1114
G3	General diet	500	7	1115~1121
G4	10% Purple-flesh potato	100	7	1122~1128
G5	10% Purple-flesh potato	500	7	1129~1135
G6	20% Purple-flesh potato	100	7	1136~1142
G7	20% Purple-flesh potato	500	7	1143~1149
G8	10% White-flesh potato	100	6	1150~1155
G9	10% White-flesh potato	500	5	1156~1160
G10	20% White-flesh potato	100	6	1161~1166
G11	20% White-flesh potato	500	6	1167~1172
G12	Purple pigment extract	100	6	1173~1178

DISCUSSION

In this study, we carried out an OGTT in order to compare antidiabetic activities of general potatoes, purple-flesh potatoes, and potato pigments in rats. Diabetes is a symptom that a surplus of glucose exists in blood because peripheral tissue cells cannot make full use of glucose supplied due to the body's declined ability to use glucose. And it has been known that oxidative stress caused by hyperglycemia, which damages tissues, plays a major role in causing complications (Baynes, 1991). This has motivated research on use of anti-carcinogenic

Table 2. Blood glucose levels in oral glucose tolerance test of rats

Group	Treatment	Dose (mg/kg, P.O.)	Glucose level (%)				
			0 min	30 min	60 min	120 min	180 min
G1	Saline	-	100.0±0.0	247.0±13.9	174.7±11.2	121.7± 8.0	117.3±5.9
G2	General diet	100	100.0±0.0	231.1±17.3	194.6±15.2	111.8± 7.9	112.3±6.3
G3	General diet	500	100.0±0.0	210.5±13.4	147.4± 7.2	104.2± 6.9	103.9±3.0
G4	10% PP	100	100.0±0.0	163.0± 7.0**	149.5± 8.8	112.6± 4.6	94.8±5.3*
G5	10% PP	500	100.0±0.0	175.3±15.1**	138.9±11.8	106.2± 5.7	95.6±5.1*
G6	20% PP	100	100.0±0.0	203.5±12.9*	169.1±12.3	117.5± 7.1	101.9±5.0
G7	20% PP	500	100.0±0.0	191.3± 9.5**	153.7±12.0	107.0± 2.0	104.9±5.7
G8	10% WP	100	100.0±0.0	219.4±17.5	160.5± 9.6	111.7± 7.1	121.7±6.4
G9	10% WP	500	100.0±0.0	195.0±10.2*	145.3± 7.5	100.6± 4.1*	95.0±3.8**
G10	20% WP	100	100.0±0.0	185.3±12.0**	132.4± 6.1**	102.3± 5.6	99.6±5.0*
G11	20% WP	500	100.0±0.0	186.9± 8.7*	151.6± 3.8	104.7± 3.0	102.3±4.2
G12	PPE	100	100.0±0.0	182.1±16.9*	164.8±10.8	114.2±11.5	102.0±6.0

*, $p>0.05$, **, $p<0.01$: compared to G1 group.
PP, purple-flesh potato; WP, white-flesh potato; PPE, purple pigment extract.

substances in food to improve blood sugar levels and delay diabetes-related complications.

It has been reported that polyphenol in green tea helps to improve blood sugar levels and control diabetes by increasing the life of islet cells in the pancreas (Hyon and Kim, 2001). The blood glucose level of the experimental animals rose because nitric oxide, produced by streptozotocin administration, reacts to superoxide anion and, as a result, peroxynitrite is produced. Peroxynitrite destroys β-cells, causing lack of insulin, and thus lowers sensitivity of β-cells to glucose. Then, the use of glucose in cells decreases, promoting production of glucose, which leads to hyperglycemia. This, in turn, produces free radicals by causing a disorder of vascular oxidation metabolism (Kahn, 1985).

Prior to this study, there has been a precedent study using purple potatoes rich in anthocyanin, which has anti-oxidation activities. It has been reported that extract of purple sweet potato orally administered to 8-week-old SD rats could reduce blood glucose and production of insulin (Matsui *et al.*, 2002). In this study, involving a GTT, the 20% purple-flesh potato group and 10% general potato group, was showed a significant concentration-dependent decrease in blood sugar after 30 minutes. The 100 mg/kg potato pigment group also showed a statistically significant decrease after 30 minutes. Therefore, administration of 10% general potato, 20% purple-flesh potato, and, 100 mg/kg potato pigment can reduce blood glucose in a GTT using rats. However, this research has only examined if various types of potatoes have anti-diabetic effects. Hence, in order to explain the anti-diabetic effects of general potatoes, purple-flesh potatoes, and potato pigments in detail and more directly, there should be follow-up research on blood glucose and insulin level changes after long-term administration of the test substances showing significant results in the study.

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