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Supplementation of a Novel Microbial Biopolymer, PGB1, from New *Enterobacter* sp. BL-2 Delays the Deterioration of Type 2 Diabetic Mice

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Abstract Antidiabetic effects of a novel microbial biopolymer (PGB)1 excreted from new *Enterobacter* sp. BL-2 were tested in the *db/db* mice. The animals were divided into normal control, rosiglitazone (0.005%, wt/wt), low PGB1 (0.1%, wt/wt), and high PGB1 (0.25%, wt/wt) groups. After 5 weeks, the blood glucose levels of high PGB1 and rosiglitazone supplemented groups were significantly lower than those of the control group. In hepatic glucose metabolic enzyme activities, the glucokinase activities of PGB1 supplemented groups were significantly higher than the control group, whereas the PEPCK activities were significantly lower. The plasma insulin and hepatic glycogen levels of the low and high PGB1 supplemented groups were significantly higher compared with the control group. Specifically, the insulin and glycogen increases were dose-responsive to PGB1 supplement. PGB1 supplement did not affect the IPGTT and IPITT compared with the control group; however, rosiglitazone significantly improved IPITT. High PGB1 and rosiglitazone supplementation preserved the appearance of islets and insulin-positive cells in immunohistochemical photographs of the pancreas compared with the control group. These results demonstrated that high PGB1 (0.25% in the diet) supplementation seemingly contributes to preventing the onset and progression of type 2 diabetes by stimulating insulin secretion and enhancing the hepatic glucose metabolic enzyme activities.

Keywords: Microbial biopolymer, *Enterobacter* sp. BL-2, PGB1, type 2 diabetes, blood glucose lowering, C57BL/KsJ-*db/db* mice

Diabetes mellitus (DM) is a common disorder of glycemic control, the prevalence of which is increasing rapidly [24]. DM has been classified into two forms. Type 1 diabetes is caused by the autoimmune destruction of pancreatic β-cells, producing insulin deficiency that requires hormone replacement therapy. This type 1 form accounts for about 10% of all cases of diabetes. The more prevalent form (90%) is type 2 (non-insulin-dependent) diabetes, which results from the combination of insulin resistance plus a β-cell secretory defect. An explosive increase in the prevalence of type 2 diabetes worldwide is predicted for the future [8]. Type 2 diabetes is characterized by pancreatic β-cell dysfunction accompanied by insulin resistance.

Recently, microbial biopolymers have received considerable attention as flocculating agents; *e.g.*, those from *Bacillus* [13, 53], *Rhodovulum* [48], *Rhodococcus* [27], *Klebsiella* [11], *Paenibacillus* [38], *Alcaligenes* [46], *Enterobacter* [52], and *Citrobacter* [15, 21]. The polysaccharide-type bioflocculants are more effective and stable than protein and glycoprotein-type bioflocculants. Nevertheless, a microbial cationic bioflocculant, except for the neutral biopolymer chitosan, has not been developed.

Son *et al.* [42] reported in 2005 that a new microbial strain, BL-2, excreting a cationic biopolymer was screened from spoiled leaves of Chinese cabbage, based on its hexosamine content and flocculating efficiency against a kaolin suspension. The newly identified *Enterobacter* sp. BL-2 was cultivated in pH-stat fed-batch culture for excretion of the biopolymer, and the excreted biopolymer was purified by the procedure [55], which they had named as a microbial biopolymer, PGB1. The purified PGB1 included glucosamine, rhamnose, and galactose, respectively, at the molar ratio of 86.4:1.6:1.0. It implied that PGB1 was almost a homobiopolymer, mainly consisting of glucosamine, even a small amount of the others.

Chemically, chitosan is a polymeric D-glucosamine, a basic polysaccharide, which is produced by deacetylating chitin, a polymer *N*-acetyl-β-D-glucosamine, with 40%–45%

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NaOH at 120°C [35]. Chitosan is a bioadhesive polysaccharide derived from deacetylation of chitin, which is, after cellulose, the most abundant polymer found in nature [51]. Therefore, chitosan has also received substantial attention in novel bioadhesive drug delivery systems with the aim to improve the bioavailability of drugs by prolonging the residence time at the site of absorption [17, 22, 32]. Moreover, chitosan is widely used for anticancer [29, 37], hypocholesterolemic [31, 56], and hemagglutinating [49] purposes, as it is a polymer of glucosamine. In 1995, Miura *et al.* [36] reported that chitosan given as a 5% food mixture produces consistent blood glucose and lipid-lowering effects in normal and neonatal streptozotocin (STZ)-induced diabetic mice, but the compound is ineffective in improving glucose and lipid biomarkers in KK-A^y mice, one of the animal models of genetically obese type NIDDM with hyperinsulinemia. Koji and Mikio [25] reported that when the low molecular weight chitosan was given as 0.05%, 0.2%, and 0.8% levels in drinking water to genetically obese diabetic KK-A^y mice, it was dose-dependently effective in improving serum biochemical parameters.

Accordingly, the current study examined the role of PGB1 in regulating blood glucose, plasma insulin, and hepatic glycogen levels in *db/db* mice, type 2 diabetic mice, along with the activities of hepatic glucose-regulating enzymes in glycolysis and gluconeogenesis.

MATERIALS AND METHODS

Preparation of Microbial Biopolymer (PGB)1 Powder

A unique cationic microbial polyglucosamine biopolymer, PGB1, excreted from *Enterobacter* sp. BL-2 was composed of D-glucosamine content of more than 95%, showing similar FT-IR and NMR spectra with those of chitosan from crab shell. PGB1 was received from ENZbio Corp. as a lyophilized fine powder, followed by authentication in the Department of Genetic Engineering of Kyungpook National University, Korea.

Animals and Diets

Thirty-six male 5-wk-old *db/db* mice were purchased from Jackson Laboratory (Maine, U.S.A.). All mice were individually housed in stainless-steel cages in a room with controlled temperature (24°C) and lighting (alternating 12 h periods of light and dark). Following arrival, all mice were fed a pelleted commercial diet (Samyang) for 1 wk; then, they were randomly divided into 4 groups (n=9). Thereafter, the control group of *db/db* mice were fed a standard semisynthetic diet (AIN-76) [3, 4], whereas the other three groups of *db/db* mice were fed standard semisynthetic diet with rosiglitazone (0.005%, wt/wt), a low dose of PGB1 (low PGB1, 0.1%, wt/wt), a high dose of PGB1 (high PGB1,0.25%, wt/wt) for 5 wks. Mice had free access to food and water. We measured food consumption and body weight everyday and every week, respectively. At the end of the experimental period, the mice were anesthetized with ketamine after withholding food for 12 h, and blood samples were drawn from the inferior vena cava for the determination of HbA_{1c}, plasma insulin.

The livers were removed after collecting blood, rinsed with physiological saline solution, weighed, and immediately stored at −70°C. Mice were treated in strict accordance with the Kyungpook National University Guide for Care and Use of Laboratory Animals.

Blood Biomarkers

The blood glucose concentration was measured at 6, 7, 8, 9, 10, and 11 wk of age, which is equivalent to 0, 1, 2, 3, 4, and 5 wk of rosiglitazone, low PGB1, and high PGB1 supplementation. After withholding food for 6 h, the blood glucose concentration was measured, with whole blood obtained from the tail veins, by using a glucose analyzer based on the glucose oxidase method (GlucoDr Supersensor, Allmedicus, Korea). The blood glycated hemoglobin (HbA_{1c}) concentration was measured after hemolysis of the anticoagulated whole-blood specimen. HbA_{1c} was determined immunoturbidimetrically. Plasma insulin (RIA kit, Diagnostic Systems Laboratories) levels were measured based on a radioimmunometric assay.

Insulin Tolerance Test (IPITT) and Intraperitoneal Glucose Tolerance Test (IPGTT)

An insulin tolerance test was performed 4 weeks after feeding with the experimental diet. After 12 h fasting, an insulin solution of 2 unit/kg body weight was injected intraperitoneally into the mice, and blood samples were obtained for glucose determination 0 (prior to insulin administration), 30, 60, and 90 min later. An intraperitoneal glucose tolerance test was performed at the end of the repeated administration period. Twelve-hour-fasted mice received an intraperitoneal injection of glucose (0.5 mg glucose/g body weight) and blood samples were collected for glucose level determination 0, 30, 60, and 120 min following glucose injection. Blood glucose was determined with a GlucoDr Supersensor (Allmedicus, Korea) on 10 μl of blood collected from the tip of the tail vein after the insulin or glucose administration.

Hepatic Glucose Metabolic Enzyme Preparation

The hepatic enzyme source was prepared according to the method developed by Hulcher and Oleson [19] with a slight modification. A 20% (w/v) homogenate was prepared in a buffer containing 100 mmol/l of triethanolamine, 20 mmol/l of EDTA, and 2 mmol/l of dithiothreitol, pH 7.0. The homogenates were centrifuged at 600 ×*g* for 10 min to discard any cell debris, and then the supernatant was centrifuged at 10,000 ×*g* followed by 12,000 ×*g* at 4°C for

20 min to remove the mitochondria pellet. The supernatant was finally ultracentrifuged twice at 100,000 ×*g* for 60 min at 4°C to obtain the cytosol supernatant. The resulting microsomal pellets were then redissolved in 1 ml of a homogenization buffer and the protein content determined by the Bradford method [7] using bovine serum albumin as the standard.

Hepatic Glucose Metabolic Enzyme Activities

Glucokinase activity was determined from liver samples homogenized in a buffer containing 50 mmol/l Tris-HCL, pH 7.4, 100 mmol/l KCl, 10 mmol/l mercaptoethanol, and 1 mmol/l EDTA. Homogenates were centrifuged at 100,000 ×*g* for 1 h; the post microsomal supernatant (cytosol) was used for the spectrophotometric continuous assay as described by Davidson and Arion [9], with a slight modification, whereby the formation of glucose-6-phosphate from glucose at 27°C was coupled to its oxidation by glucose-6- phosphate dehydrogenase and nicotinamide adenine dinucleotide (NAD).

Glucose-6-phosphatase activity was determined in the hepatic microsome using a spectrophotometric assay according to the method of Alegre *et al.* [1], with a slight modification; the reaction mixture contained 100 mmol/l sodium Hepes (pH 6.5); 26.5 mmol/l glucose-6-phosphate and 1.8 mmol/l EDTA, both previously adjusted to pH 6.5; 2 mmol/l NADP; 0.6 kIU/l mutarotase; and 6 kIU/l glucose dehydrogenase.

Phosphoenolpyruvate carboxykinase (PEPCK) activity was measured using the spectrophotometric assay developed by Bentle and Lardy [6], with a slight modification. A 1 ml final volume of the hepatic cytosolic enzyme pipetted with the reaction mixture, pH 7.0, contained 50 mmol/l sodium Hepes, 1 mmol/l IDP, 1 mmol/l $MnCl_2$, 1 mmol/l dithiothreitol, 0.25 mmol/l nicotinamide adenine dinucleotide reduced form (NADH), 2 mmol/l phosphoenolpyruvate, 50 mmol/l $NaHCO_3$, and 7.2 U of malic dehydrogenase. The enzyme activity was measured at 25°C, based on a decrease in the absorbance at 340 nm.

Hepatic Glycogen Assay

The glycogen concentration was determined as previously described by Seifter *et al.* [41], with slight modifications. Briefly, the liver tissue was homogenized in 5 volumes of an iced-cold 300 g/l KOH solution and was dissolved in a boiling water bath (100°C) for 30 min. The glycogen was precipitated with ethanol, and then was pelleted, washed, and resolublized in distilled water. The glycogen concentration was determined by treatment with an anthrone reagent [2 g anthrone per 1 l of 95% (v:v) H_2SO_4] and by measuring the absorbance at 620 nm.

Pancreas Immunohistochemistry

Briefly, the pancreas was rapidly removed after sacrificing, fixed in 10% (v:v) neutral-buffered formalin, processed routinely, and embedded in paraffin. Paraffin sections were cut at 4-μm thickness, deparaffinized in xylene twice for 5 min, and rehydrated with graded ethanol. The rehydrated sections were treated with 3% (v:v) H_2O_2 in methanol for 30 min to block endogenous peroxidase, washed in 0.01 mol/l phosphate buffer for 10 min, and then immunostained with the primary antibody, monoclonal mouse anti-insulin. The antigen-antibody complex was visualized by an avidin-biotin peroxidase complex solution by using an ABC kit (Vector Laboratories) with 3,3-diamino benzidine (Zymed Laboratories). For the negative control, the primary antibody was replaced by phosphate-buffered saline solution.

Statistical Analysis

All data are presented as the means±SEM. The data were evaluated by a one-way ANOVA with SPSS for Windows and the differences between the means were assessed using Duncan's multiple-range test. Statistical significance was considered at $p<0.05$.

RESULTS

Body Weight and Food Intake

As shown in Fig. 1, the body weights of the low PGB1, high PGB1, and rosiglitazone supplemented groups increased throughtout the experimental period. On the other hand, those of the control group were slightly decreased after 3 weeks. The body weight of the rosiglitazone supplemented group was significantly higher, but those of the control, low PGB1, and high PGB1 supplemented groups did not differ during the whole period. The food intake of the rosiglitazone supplemented group was significantly lower than the other groups, 4.50±0.20 g vs. 5.90±0.15 g.

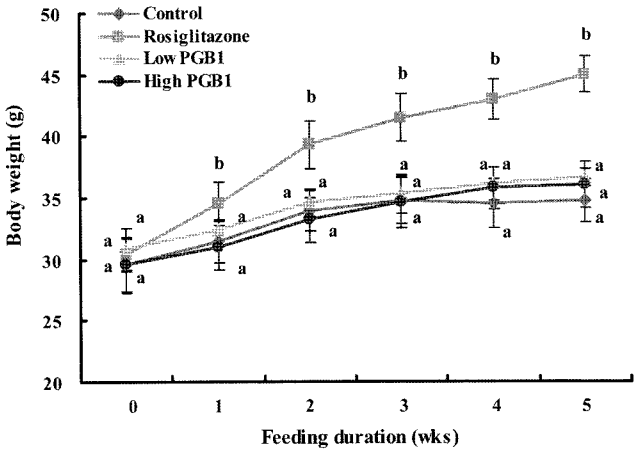

Fig. 1. Weekly changes in body weight of PGB1 and rosiglitazone supplemented C57BL/KsJ-*db/db* mice.
Values are means±SEM, n=9. [ab]The means not sharing a common letter are significantly different between groups ($p<0.05$).

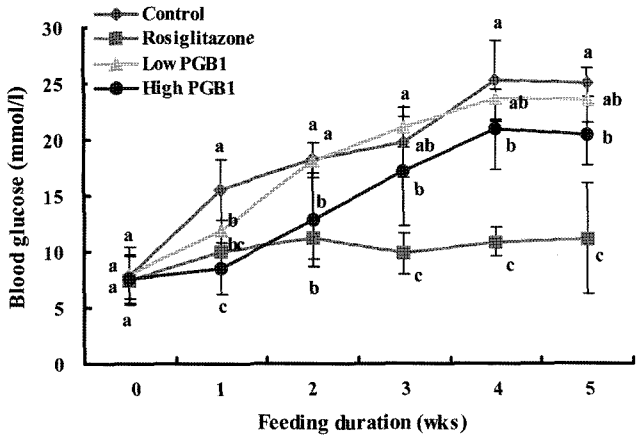

Fig. 2. Weekly changes in blood glucose concentration of PGB1 and rosiglitazone supplemented in C57BL/KsJ-*db/db* mice.
Values are means±SEM, n=9. [abc]The means not sharing a common letter are significantly different between groups ($P<0.05$).

Blood Glucose Levels

The blood glucose levels of *db/db* mice treated with low PGB1, high PGB1, and rosiglitazone supplementation are shown in Fig. 2. The baseline (0 wk) fasting glucose levels did not differ between the groups (≥7.5 mmol/l). The blood glucose level of the high-PGB1 group was significantly lower compared to the control group during the whole experimental period except for the 3rd wk, whereas that of the control group gradually increased with age. However, this increase was completely prevented in the rosiglitazone supplementation group. The low PGB1 group showed a similar blood glucose change to the control *db/db* group.

Intraperitoneal Glucose Tolerance Test (IPGTT) and Insulin Tolerance Test (IPITT)

Insulin and glucose tolerance were evaluated by the IPGTT and IPITT method on the 4th and 5th week of the experimental period. Neither doses of PGB1 affect the glucose tolerance or insulin tolerance when compared with the control group and the rosiglitazone-supplemented group affected in overall glucose response for the insulin tolerance test (data not shown).

Blood HbA_{1c}, Plasma Insulin, and Plasma Glucagons Levels

The blood HbA_{1c} concentration of the rosiglitazone group was significantly lower compared with the control and PGB1 groups. The plasma insulin levels of the two PGB1 groups were significantly higher than the control or rosiglitazone groups, whereas the plasma glucagon level was not different from the control group. The glucagon level of the rosiglitazone group showed the highest level among the groups. However, the ratio of insulin/glucagon of both PGB1 groups was significantly higher than those of the control and rosiglitazone groups. The hepatic glycogen content of the high-PGB1 group was significantly higher than the rosiglitazone, low-PGB1, or control groups, although the low-PGB1 group showed a higher glycogen content than the rosiglitazone group (Table 1).

Hepatic Glucose Metabolic Enzyme Activities

Hepatic glucokinase activity was significantly higher in the PGB1 and rosiglitazone groups when compared with the control group. In contrast, phosphoenolpyruvate carboxykinase (PEPCK) activity was remarkably lower in the PGB1 groups compared with the control and rosiglitazone groups. Glucose-6-phosphatase activity was significantly lower in the rosiglitazone group than in the other groups (Table 2).

Pancreas Immunohistochemistry

The islets of Langerhan in the *db/db* mice appeared to have abnormal architecture. Intact and specific insulin-positive cells in the low PGB1, high PGB1, and rosiglitazone supplemented groups were confirmed in the pancreatic islet β-cells. There were more insulin-positive β-cells in these 3 groups compared with the control group (Fig. 3).

DISCUSSION

In the current study, the antidiabetic potential of a novel microbial biopolymer, PGB1, was compared with that of rosiglitazone in a type 2 diabetic animal model, *db/db* mice. We demonstrated that PGB1 exhibited blood glucose

Table 1. Concentration of blood HbA_{1c}, plasma insulin, glucagon, and hepatic glycogen in C57BL/KsJ-*db/db* mice supplemented with PGB1 or rosiglitazone.

	Control	Rosiglitazone	Low PGB1	High PGB1
HbA_{1c} (%)	10.5±0.28[b]	4.4±0.09[a]	12.1±0.12[c]	10.1±0.17[b]
Insulin (pmol/l)	723.2±47.4[a]	981.2±52.3[b]	2,047.6±83.8[e]	2,293.6±56.4[d]
Glucagon (pg/ml)	315.7±15.41[ab]	388.1±15.14[c]	298.9±11.52[a]	347.8±9.49[bc]
Insulin/Glucagon	2.29±0.15[a]	2.53±0.13[a]	6.85±0.28[b]	6.59±0.16[b]
Glycogen (mg/g liver)	169.0±12.8[ab]	135.9±6.3[a]	176.4±5.0[b]	211.4±16.9[c]

Values are means±SEM, n=9.
[abcd]The means in the row not sharing a common letter are significantly different between groups ($p<0.05$).

Table 2. The activities of hepatic glucokinase, glucose-6-phosphatase, and phosphoenolpyruvate carboxykinase in C57BL/KsJ-*db/db* mice supplemented with PGB1 or rosiglitazone.

	Control	Rosiglitazone	Low PGB1	High PGB1
	(nmol/min/mg protein)			
Glucokinase	8.47±0.37[a]	16.68±1.26[c]	12.03±0.97[b]	12.70±0.71[b]
Glucose-6-phosphatase	169.6±7.68[a]	114.0±9.83[b]	153.6±13.16[a]	158.8±8.76[a]
Glucokinase/G-6-P	55.70±3.50[a]	108.2±4.27[c]	77.35±2.32[b]	75.91±2.99[b]
Phosphoenolpyruvate carboxykinase	57.64±3.32[a]	64.35±1.91[a]	48.30±1.64[b]	45.83±1.61[b]

Values are means±SEM, n=9.
[abc]The means in the row not sharing a common letter are significantly different between groups ($p<0.05$). G-6-P, glucose-6-phosphatase.

lowering property in *db/db* mice; specifically, its high dose (0.25%, wt/wt) was significantly effective. High dose of PGB1 significantly lowered the blood glucose level compared with the control group throughout the experimental period, except at the 3rd week.

Rosiglitazone, low PGB1, and high PGB1 significantly elevated the plasma insulin level compared with the control group by 1.4 times, 2.8 times, and 3.2 times, respectively. Thus, the plasma insulin level, an index of insulin secretion [14], of the PGB1-supplemented group was higher than the rosiglitazone group at the end of the study. In the *db/db* mice, the plasma insulin level increased rapidly during the first few weeks of life, and then declined successively after 8–10 wk of age [20]. Thus, the plasma insulin levels in the *db/db* mice might be decreased after reaching a peak, whereas PGB1 supplementation maintained its insulin level, although the plasma insulin level was not measured throughout this study. In our previous study, we confirmed that the plasma insulin concentration of *db/db* mice begins to decrease from 11 wk of age [44], which is equivalent to the 5th wk of the experimental period in this study. Increased insulin level may lead to a significant reduction of the blood glucose level. In addition, we observed that the relative number of insulin-positive β-cells of PGB1 groups was larger than in the control group. The histology of the pancreatic islets from both the rosiglitazone and PGB1 groups exhibited a similar pattern. Insulin production observed in the high PGB1-supplemented group seemed to be well-preserved, as shown in the pancreatic islet morphology and compared with the islet minification, structure disorganization, and sparsity of insulin production displayed in the control group. The appearance of islets from the PGB1-supplemented *db/db* mice with their metabolic changes strongly suggests a protective role for PGB1, in particular a high dose of PGB1, against type 2 diabetes. In the present study, the ratio of insulin/glucagon of PGB1-supplemented groups was significantly higher than that of the control group, yet the plasma glucagon level of PGB1 groups was similar to the control group. This result corresponded with Park *et al.* [45]. In their study, the plasma glucagon level was not different between the *db/+* mice and the *db/db* mice. However, the ratio of insulin/glucagon was significantly higher in the *db/+* mice than in the control *db/db* mice. Thus, they suggested that the insulin/glucagon ratio rather than the absolute concentration of insulin or glucagon may be more important for altering blood glucose concentration, although the exact mechanism was not elucidated.

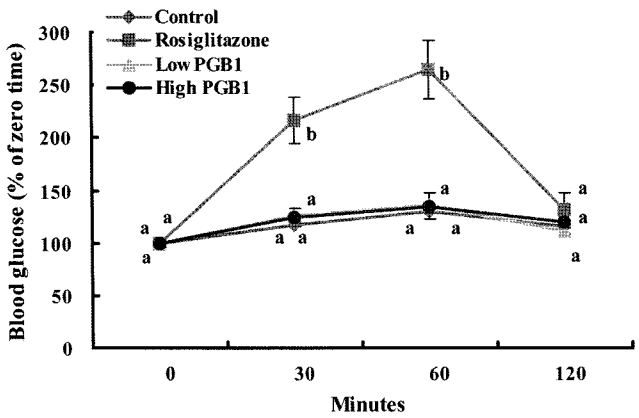

Fig. 3. Effects of PGB1 and rosiglitazone supplementation on intraperitoneal glucose tolerance test in C57BL/KsJ-*db/db* mice. Values are means±SEM, n=9. [abc]The means not sharing a common letter are significantly different between groups ($p<0.05$).

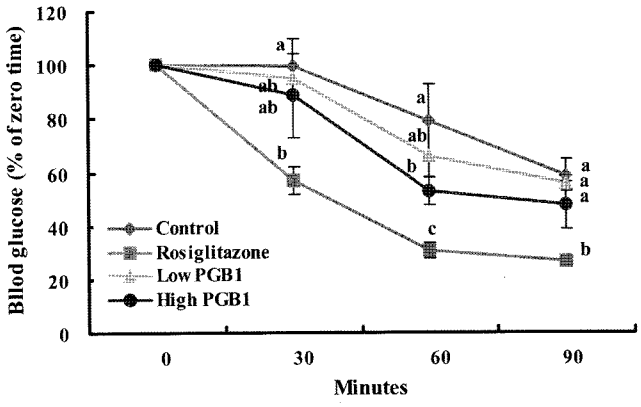

Fig. 4. Effects of PGB1 and rosiglitazone supplementation on intraperitoneal insulin tolerance test in C57BL/KsJ-*db/db* mice. Values are means±SEM, n=9. [abc]The means not sharing a common letter are significantly different between groups ($p<0.05$).

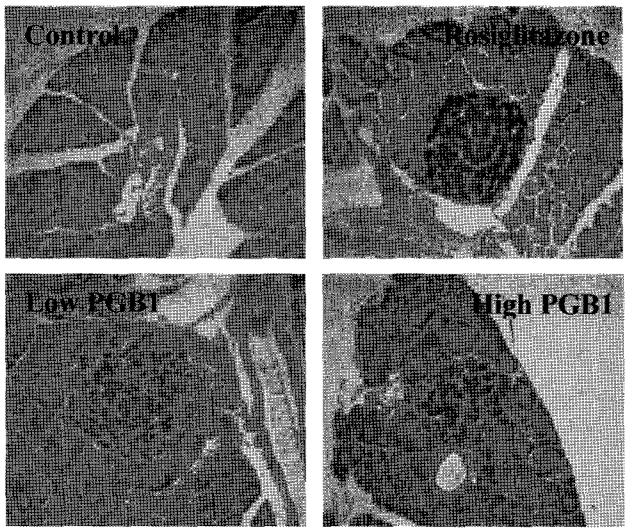

Fig. 5. Immunohistochemical photographs of pancreas in C57BL/KsJ-*db/db* mice supplemented with PGB1 and rosiglitazone. Original magnification ×200.

Unlike sulfonylureas (*e.g.*, glipizide, glyburide), which activate insulin secretion from β-cell of the pancreas, and metformin, which reduces hepatic glucose output, thiazolidinediones improve insulin sensitivity and thus counteract insulin resistance [40]. Rosiglitazone is an insulin sensitizing agent that improves the response to insulin in target tissues without stimulating insulin secretion [5, 43, 54]. The exact mechanism of increased insulin sensitivity has yet to be defined, but several pharmacological activities have been identified. Rosiglitazone is an agonist of the peroxisome proliferator-activated receptor gamma (PPAR-γ), which is found in insulin-dependent glucose requiring tissues such as adipose tissue, skeletal muscle, and liver tissue [2, 30]. Once PPAR-γ is stimulated, glucose transporter-4 (GLUT-4) production and translocation occur. Under normal circumstances, insulin facilitates the migration of GLUT-4 to the cell surface, which stimulates glucose uptake. Therefore, increasing transcription of GLUT-4 assists in overcoming this resistance [26, 40]. The end point of PPAR-γ activation is a reduction in hepatic glucose production and increased in insulin-dependent glucose uptake in fat and skeletal tissues [2, 23, 30]. As previously stated, rosiglitazone did not stimulate insulin secretion as compared with PGB1-supplemented group in this study, although the insulin level of rosiglitazone group was higher than that of the control group. We observed that rosiglitazone increased insulin sensitivity, which is suggested by decreased blood glucose levels of rosiglitazone group compared to the control or PGB1 groups in IPITT. We measured the insulin resistance of *db/db* mice using the IPGTT, commonly employed in the diagnosis of diabetes mellitus, as well as the IPITT. As the result of IPGTT, glucose tolerance was not improved by PGB1 supplement. Although PGB1 supplementation dropped the blood glucose level in *db/db* mice, it did not affect the glucose and insulin tolerance tests. This needs to be elucidated in a further study.

Generally, increased hepatic glucose production, plus decreased hepatic glycogen synthesis and glycolysis, are the major symptoms in type 2 diabetes that results in hyperglycemia; these would seem to be the consequence of the low glucokinase activity and high glucose-6-phosphatase and PEPCK activities in diabetic mice [12, 16, 28, 39]. In the current study, both low and high PGB1 supplements enhanced the glucokinase activity, regardless of its dose, in *db/db* mice. Valera and Bosch [47] reported that a short term increase in hepatic GK expression increases glycolytic flux and glycogen storage. Hepatic glucokinase is the most sensitive indicator of the glycolytic pathway in diabetes, and its increase can in turn increase the utilization of blood glucose for glycogen storage in the liver [33]. In addition, in the current study, high PGB1 significantly elevated the hepatic glycogen concentration compared with the control group. Hepatic glycogen reserves are important for the whole body glucose homeostasis and are markedly low in the diabetic state [18, 34, 50]. In the present study, changes of hepatic glucokinase activity and glycogen concentration by the high-PGB1 dose were more effective than by the low-PGB1 dose. The gluconeogenic enzyme hepatic G6Pase activities of the PGB1 groups were not different from the control group, although rosiglitazone lowered this enzyme activity. However, PGB1, regardless of its dose, significantly improved the glucokinase/G6Pase ratio compared with the control group.

Among the glucose regulating genes, an enhanced expression of hepatic PEPCK gene has been identified in most forms of diabetes, and contributes to increased hepatic glucose output [10]. PGB1 supplement led to significantly lower hepatic PEPCK activity compared with the control group, whereas rosiglitazone did not affect the hepatic PEPCK activity. Interestingly, there was no dose-dependent response of PGB1 supplementation for hepatic PEPCK activity. Thus, PGB1 supplementation, overall, may have the ability to alter the hepatic gluconeogenesis that influences glucose homeostasis.

At this time, the exact mechanism by which PGB1 supplement influences plasma glucose needs to be elucidated. Nevertheless, the data obtained in this study suggest that high PGB1 is an effective antihyperglycemic agent, which seems to be mediated through the increased plasma insulin level, activation of GK, and inhibition of PEPCK in the liver in the type 2 diabetic animal model.

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