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In vivo hypoglycemic investigation, antihyperglycemic and antihyperlipidemic potentials of *Pereskia bleo* Kunth. in normal and streptozotocin-induced diabetic rats

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ABSTRACT

Objective: To elucidate the *in vivo* hypoglycemic capability, antihyperglycemic and antihyperlipidemic activities of *Pereskia bleo* (Kunth) leaves extracts and bioactive fraction. **Methods:** The various solvent extracts of *Pereskia bleo* were investigated for the hypoglycemic and antihyperglycemic activities using a relevant *in vivo* normal rat model and streptozotocin-induced diabetic rat model with glibenclamide and metformin utilized as positive controls. The effects of the most potent extract and its bioactive fraction on the insulin level, lipid profile and body weight of the diabetic rats were also analyzed. **Results:** All the extracts showed no hypoglycemic effect while petroleum ether, chloroform and aqueous extracts demonstrated significant (P<0.05) reduction in blood sugar level in the intraperitoneal glucose tolerance test. Aqueous extract and aqueous fraction significantly (P<0.05) reduced the blood glucose level in streptozotocin-induced diabetic rats as early as day 6 compared to the diabetic control as well as significantly restored the serum insulin of diabetic rats. Moreover, the aqueous extract and aqueous fraction disclosed a significant (P<0.05) reduction in total cholesterol, triglycerides, and low-density lipoprotein levels. An elevation in high-density lipoprotein as well as improved body weight loss of the diabetic rats were also observed. **Conclusions:** In summary, *Pereskia bleo* appears effective in the management of diabetes and correlated impairments arising from high blood sugar level. Further studies will possibly bring about the discovery of effective and secure plant derived antidiabetic drugs.

1. Introduction

To date, the occurrence of diabetes mellitus is greatly augmenting. Diabetes, which is well known as the high blood sugar level, is a metabolic ailment. Lack of insulin production or the cells failure to utilize insulin competently in the body contributes to the development of diabetes. By the year 2030, WHO foresees that diabetes will be one of the top ten major reasons of all deaths globally[1]. This is due to the fact that diabetes engages in the advancement of other health complications such as heart attacks, stroke and kidney failure[2]. There are two main elements ascertained to be associated with the accelerated occurrence of diabetes, including lifestyle factors and genetic makeup. Unhealthy lifestyle that provokes the lessening of physical activity, poor diet

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and obesity are the leading contributors to this chronic condition[3].

At present, pharmaceutical market caters a variety of diabetes drugs and insulin therapy. Nevertheless, these diabetic drugs are often encountered with various side effects which include hypoglycemia, weight gain, lactic acidosis, nausea and peripheral edema[4]. Besides, the clinically established diabetes drugs were also reported not economical as well as confronted prominent treatment failures[5]. Therefore, a better therapeutic agent is critically needed for the prevention and management of diabetes.

Over the centuries, plants have documented countless medicinal significances for many diseases. There are a number of data reported on the usage of medicinal plants for the treatment of diabetes[6]. Metformin is one of the substantial examples of diabetic drug isolated from medicinal plant. Hence, the need for novel antidiabetic drug from herbal medicines is indispensable. *Pereskia bleo* (*P. bleo*) (Kunth) is a member of Cactaceae family with thorny leaves native to West and South America and is widely distributed in tropical and subtropical parts of Asia such as India, Indonesia, Malaysia and Singapore[7,8]. Leaves extract of *P. bleo* had established prominent pharmacological activities such as antidiabetic, antimicrobial, antioxidant, antihypertension, antinociceptive and antiproliferative properties[9,10]. The leaves decoction of the plant is also reportedly used by pregnant ladies in the villages of Kelantan, Malaysia as antidiabetic remedy[11].

Previous studies had reported that aqueous leaf extract of *P. bleo* significantly reduced the blood sugar concentration of alloxan induced diabetic Sprague-Dawley rats[12]. However, there have been no reports on the antidiabetic activities from the fractions of *P. bleo* extract. It follows that the related antidiabetic activity of the plant crude extract might possibly be enhanced by a subsequent bioactivity guided fractionation step. The resulting bioactive fractions may also exhibit better antidiabetic potency in the *in vivo* study. Inspired by this, the current study was conducted in order to explore the hypoglycemic, antihyperglycemic and antihyperlipidemic potentials of crude leaf extracts of *P. bleo* as well as its bioactive fraction.

2. Materials and methods

2.1. Chemicals and standards

Streptozotocin (STZ) was obtained from the Sigma-Aldrich company. Glibenclamide and metformin (standard drugs) used were from Hovid Bhd. (Malaysia). All other commercial reagents and solvents used were of analytical grade.

2.2. Plant collection and authentication

Leaves of *P. bleo* were collected from Kubang Semang, Bukit Mertajam, Penang, Malaysia. The leaves were authenticated by the institution botanist and the voucher specimen (USM/ Herbarium/11609) was kept at Herbarium Unit.

2.3. Extraction of P. bleo

The dried powders were extracted for 24 h by maceration using petroleum ether. The residues were then re-extracted by chloroform, methanol and water serially using the same technique. Each extract was then filtered and was concentrated by using rotary evaporator (Rotavapor R-200, Buchi, Switzerland) under reduced pressure at 40 ℃. The water extract was subjected to freeze-drying (FreeZone, MO, USA). All the four different solvent extracts were stored at 4 ℃ until use.

2.4. Animals

Experiments were performed in healthy male adult Sprague Dawley rats (200 to 250 g) which were acquired from the Animal Research and Services Centre (ARASC), Universiti Sains Malaysia (USM) and acclimatized for one week in the Animal Transit Room, USM prior to experiments. The animals were housed in sanitized polypropylene cages (3 per cage) under the temperature of (23 ± 2) ℃ and (50 ± 5)% humidity. The rats were fed on standard laboratory diet with free access to water. The experimental protocol has been approved by the institutional animal ethics committee (Approval no. USM/ Animal Ethics Approval / 2013/ (87) (463)).

2.5. Hypoglycemic activity in normal rats

In this experiment, a total of 36 male Sprague Dawley rats were used. All the rats were kept fasting overnight providing water *ad libitum*. All the rats were casually allocated into six groups (n=6). Group 1, the negative control group received 10% tween 20; group 2, the positive control group received glibenclamide (10 mg/kg b.w.); groups 3 to 6, the treatment groups received 1 000 mg/kg b.w. of petroleum ether extract, chloroform extract, methanol extract and aqueous extract of *P. bleo*, respectively. The treatments were administered orally to the rats. The effects of the vehicle, crude extracts treatment or glibenclamide on blood sugar levels were measured in fasted rats at zero, one, two, three, five and seven hours respectively after a single oral administration. Blood samples were withdrawn from the caudal tail vein in anesthetized rats. Blood sugar levels were ascertained using a glucometer (ACCU-CHEK® Performa).

2.6. Intraperitoneal glucose tolerance test

A total of 36 male Sprague Dawley rats were randomly assigned into six different groups (n=6 in each group) and fasted overnight. Group 1, the negative control group received 10% tween 20; group 2, the positive control group received metformin (500 mg/kg b.w.); groups 3 to 6, the treatment groups received 1 000 mg/kg b.w. of petroleum ether extract, chloroform extract, methanol extract and aqueous extract of *P. bleo* respectively. Glucose (1 g/kg b.w.) was administered intraperitoneally within 1 h after the extracts were fed. Blood samples were withdrawn from rats at 0, 15, 30, 45, 60, 90 and 120 min. Blood sugar levels were verified using a glucometer (ACCU-CHEK® Performa).

2.7. Antihyperglycemic activity of P. bleo crude extracts

2.7.1. Induction of diabetes

In short, a high fat diet was given to the rats for 4 weeks. At the end of 4 weeks, STZ (50 mg/kg b.w.) was injected intraperitoneally to the rats overnight fasted. After the STZ injection, the rats were given 5% glucose solution overnight as drinking water. After 72 h, rats with fasting blood sugar levels of more than 12 mmol/L were selected to include in the study. Rats in the normal control group (n=6) were fed with normal pellet diet and were administered buffer alone.

2.7.2. Diabetic study

A total of 42 STZ induced rats and 6 normal rats were assigned into eight groups. Group 1 and 2 which consisted of normal and diabetic control rats were delivered 10% tween 20. Groups 3 and 4 received glibenclamide (10 mg/kg b.w.) and metformin (500 mg/kg b.w.), respectively. Groups 5 to 8 were delivered 1 000 mg/kg b.w. of petroleum ether extract, chloroform extract, methanol extract and aqueous extract, respectively. Treatments were delivered orally twice a day for 12 d. Blood sugar levels were ascertained using glucose meter (ACCU-CHEK® Performa).

2.8. Fractionation of aqueous extract of P. bleo

P. bleo aqueous extracts were further partitioned by solvent-solvent extraction method. An amount of 40 g of aqueous extract were suspended in 250 mL distilled water and were poured in 1 L separatory funnel. This extract was then partitioned by adding 750 mL of ethyl acetate. The fraction of ethyl acetate separated was collected and concentrated using rotary evaporator. Similar process was repeated to obtain the n-butanol fraction by replacing ethyl acetate with 750 mL of n-butanol. The residual water layer was collected and denoted as aqueous fraction.

2.9. Diabetic study of P. bleo fractions

The diabetic induction was done in Sprague Dawley rats according to the method described in section 2.7.1. Only those rats with fasting glucose level exceeding 12 mmol/L were used. Thirty-six diabetic rats and 6 normal rats were allocated into seven groups (n=6). Groups 1 to 4 were given the same treatments as described previously in section 2.7.2. Group 5 to 7 received 500 mg/kg b.w. of ethyl acetate fraction, n-butanol fraction and aqueous fraction of *P. bleo*, respectively. Treatments were delivered orally twice a day for 12 d and the blood sugar levels were ascertained.

2.10. Dose optimization study of aqueous fraction of P. bleo

The diabetic induction was done in Sprague Dawley rats according to the method described in section 2.7.1. The aqueous fraction of *P. bleo* aqueous extract was observed to be the most active fraction, hence it was used in this study to determine the minimum effective dose. The number of rats used was as same as depicted in section 2.9. Groups 1 to 4 were given the same treatments as described previously in section 2.7.2. Groups 5 to 7 received different doses of aqueous fractions that include 500 mg/kg b.w., 250 mg/kg b.w. and 125 mg/kg b.w., respectively. Treatments were delivered orally twice a day for 12 d and the blood sugar levels were ascertained.

2.11. Antidiabetic effects of P. bleo aqueous extract and aqueous fractions

The diabetic induction was done in Sprague Dawley rats according to the method described in section 2.7.1. The number of rats used was as same as depicted in section 2.9. Groups 1 to 4 were given the same treatments as described previously in section 2.7.2. Group 5 received crude aqueous extract of 1 000 mg/kg b.w. Group 6 and 7 received aqueous fractions of 500 mg/kg b.w and 250 mg/kg b.w., respectively. Treatments were delivered orally twice a day for 12 d. After 12 days of treatment, the rats were anaesthetised with pentobarbitone sodium at 50 mg/kg b.w. intraperitoneally before the procedure was done. About 5 mL of blood samples were collected from the heart using 25 G needle and 5 mL syringe which then were transferred to 10 mL heparinised tubes for further studies.

2.11.1. Effect on plasma insulin level

The heparinized tubes were immediately centrifuged at 2 000 ×g for 20 min at 4 ℃ to obtain plasma. Plasma insulin levels were measured by ELISA method using Ultra Sensitive Rat Insulin ELISA Kit (Crystal Chem, USA).

2.11.2. Effect on serum lipid profile

The blood samples were collected in a tube. Lipid profile analysis was done by analysis of blood samples at a clinical laboratory [Gribbles Pathology (M) Sdn Bhd] to measure total cholesterol (TC), triglycerides (TG), low-density lipoprotein (LDL) and high-density lipoprotein (HDL).

2.11.3. Effect on body weight

The weights of all the rats were measured and recorded on the third day of the diabetes induction (first day of treatment) and the last day of the treatment (day 12). The rats were fasted overnight before measurements were taken.

2.12. Statistical analysis

All the data reported are expressed as mean ± SEM for 6 animals in each group. The hypothesis testing method used was one-way ANOVA followed by Dunnette's comparison tests. The values were considered significantly different at P<0.05.

3. Results

3.1. Hypoglycemic activity

Figure 1 represents the variation of blood sugar levels after delivering a single oral dose of various solvent extracts and aqueous extract of *P. bleo* to the normal rats. The rats which received treatments of methanol, chloroform and aqueous extracts displayed no hypoglycemic effect throughout test compared to the control

group. Petroleum ether extract-treated rats showed a reduction on blood glucose level at the third hour (4.1 mmol/L) but increased at the final hour of study. At this dose, entire rats were found conscious, and the behaviour and activity were normal compared to the control. Glibenclamide-treated rats demonstrated significant (P<0.05) hypoglycemic effect consistently from the first time point.

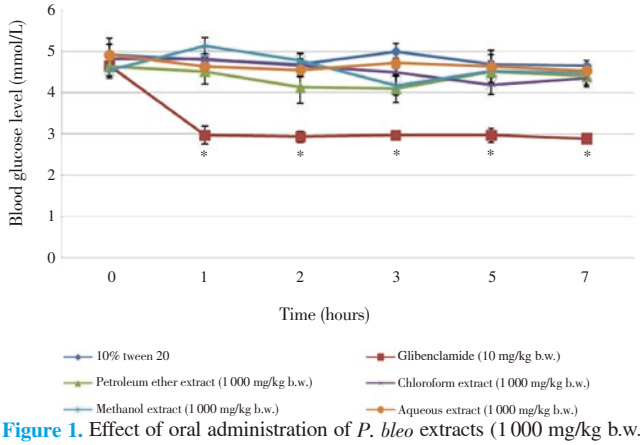

Figure 1. Effect of oral administration of *P. bleo* extracts (1 000 mg/kg b.w.) on fasting blood glucose level in normal rats.

Data are presented as mean ± SEM (n = 6). *Significant difference from control (P<0.05).

3.2. Intraperitoneal glucose tolerance test

The glucose tolerance effect of *P. bleo* crude extracts is illustrated in Figure 2. Rats in all the tested groups exhibited a sudden increase in their blood glucose level after glucose administration until the 15 min (*i.p.*). However, during the period of 30 min to 90 min, all the tested groups presented potential glucose lowering activity. At 120 min, rats treated with 1 000 mg/kg b.w. of petroleum ether, chloroform and aqueous extracts of *P. bleo* demonstrated significant (P<0.05) reduction on the blood glucose level compared to the control group. Notably, the efficiency of *P. bleo* extracts and metformin (500 mg/kg b.w.) in reducing the blood sugar level was almost comparable. Metformin has showed significant glucose lowering affect from 30 to 120 min.

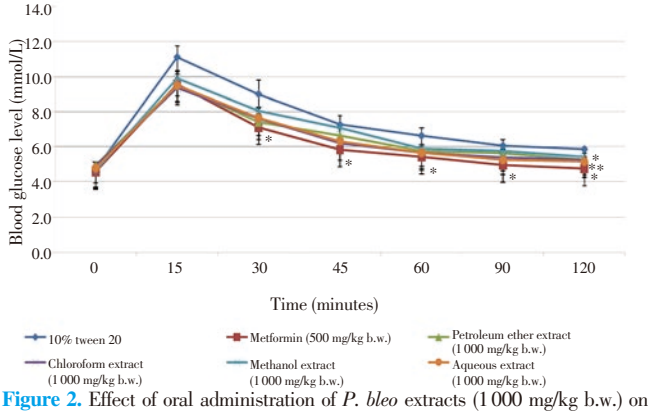

Figure 2. Effect of oral administration of *P. bleo* extracts (1 000 mg/kg b.w.) on fasting blood glucose level after intraperitoneal loading of 1 g/kg b.w. of glucose in normal rats.

Data are presented as mean ± SEM (n = 6). *Significant difference from control (P<0.05).

3.3. Antihyperglycemic activities of P. bleo crude extracts

Figure 3 displays the influence of the *P. bleo* extracts on diabetic rats during 12 days of daily oral administration. Diabetic rats treated with petroleum ether and chloroform extracts showed no significant effect on the blood glucose levels throughout the study. Methanol extracts, on the contrary, had significantly (P<0.05) lowered the blood glucose level of the diabetic rats on the day 12. Aqueous extract-treated diabetic rats have initiated reduction in blood glucose levels as early as day 3 and this reduction became more significantly pronounced on day 6 (11 mmol/L) till day 12 (8.1 mmol/L). Aqueous extract of *P. bleo* revealed potent blood glucose lowering effect compared to the clinically established diabetic drugs, glibenclamide and metformin (Figure 3). Therefore, the crude aqueous extract was subjected to the fractionation study to further improve and investigate the antidiabetic activity.

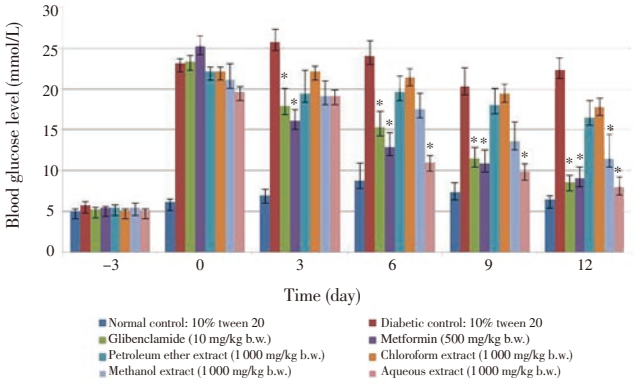

Figure 3. Antihyperglycemic effect of oral administration of *P. bleo* extracts (1 000 mg/kg b.w.) on fasting blood glucose level of STZ-induced diabetic rats.

Data are presented as mean ± SEM (n = 6). *Significant difference from diabetic control (P<0.05).

3.4. Antihyperglycemic activities of fractions of crude aqueous extract

A total of three fractions namely ethyl acetate, n-butanol and aqueous fraction were obtained after the partitioning of the potent crude aqueous extract of *P. bleo* by solvent-solvent fractionation method. The effect of the *P. bleo* fractions on the blood glucose level in STZ induced diabetic rats is shown in Figure 4. A significant (P<0.05) reduction in blood glucose levels was observed after 12 days of oral treatment with ethyl acetate (12.3 mmol/L) and n-butanol (9.7 mmol/L) fractions in contrast to diabetic control (22.3 mmol/L). On the other side, the aqueous fraction treated rats had lowered the blood glucose significantly (P<0.05) from day 6 (14 mmol/L) to day 12 (6.2 mmol/L) compared to diabetic control. Interestingly, the aqueous fraction demonstrated comparable potency to those of glibenclamide and metformin which had lowered the blood sugar levels as soon as day 3 with 17.9 mmol/L and 16 mmol/L respectively. Therefore, aqueous fraction was chosen to dose response relationship study.

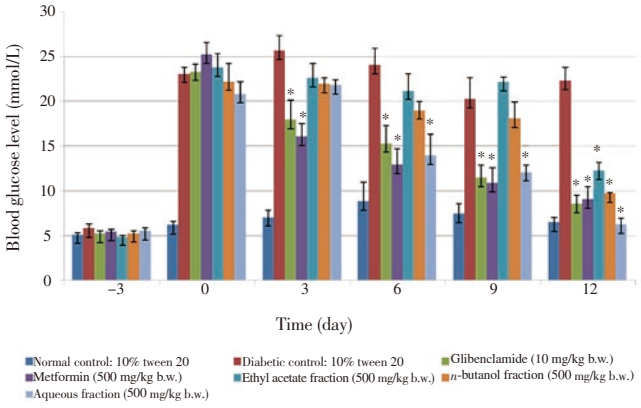

Figure 4. Antihyperglycemic effect of oral administration of *P. bleo* aqueous extract fractions (500 mg/kg b.w.) on blood glucose of STZ-induced diabetic rats.

Data are presented as mean ± SEM (n = 6). *Significant difference from diabetic control (P<0.05).

3.5. Dose response relationship of aqueous fractions

Figure 5 reveals the effect of different doses of *P. bleo* aqueous fraction on fasting blood glucose level in STZ induced diabetic rats. The treatment of 500 mg/kg b.w. and 250 mg/kg b.w. of *P. bleo* aqueous fractions showed reduction in the blood glucose level since day 6 (14 mmol/L and 16 mmol/L, respectively) compared to diabetic control (24.1 mmol/L). Nevertheless, rats treated with 125 mg/kg b.w. of *P. bleo* aqueous fraction did not show any significant blood glucose reduction.

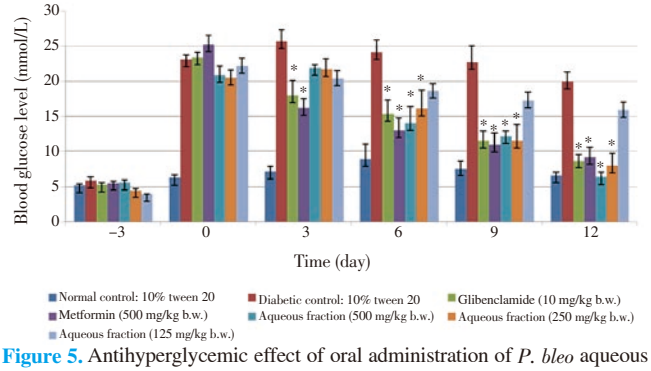

Figure 5. Antihyperglycemic effect of oral administration of *P. bleo* aqueous fraction at different doses on blood glucose of STZ-induced diabetic rats.

Data are presented as mean ± SEM (n = 6). *Significant difference from diabetic control (P<0.05).

3.6. Antidiabetic effects of P. bleo aqueous extract and aqueous fractions

3.6.1. Effect on insulin level

Figure 6 depicts the insulin level of the diabetic rats treated with crude aqueous extract (1 000 mg/kg b.w.) and aqueous fractions (500 and 250 mg/kg b.w.) of *P. bleo* for 12 d. Both the aqueous extract and fractions presented a significant (P<0.05) increase of the insulin level compared to diabetic control. The diabetic rats treated with 500 mg/kg b.w. of aqueous fraction, displayed the highest insulin concentration of 0.42 ng/mL, which was slightly higher than the normal control (0.41 ng/mL). Moreover, the insulin concentration of *P. bleo* aqueous fraction outshone the clinically established drug metformin at the same dosage (500 mg/kg b.w.) used. On the other hand, glibenclamide displayed an insulin rise of 0.25 ng/mL compared to diabetic control (0.10 ng/mL).

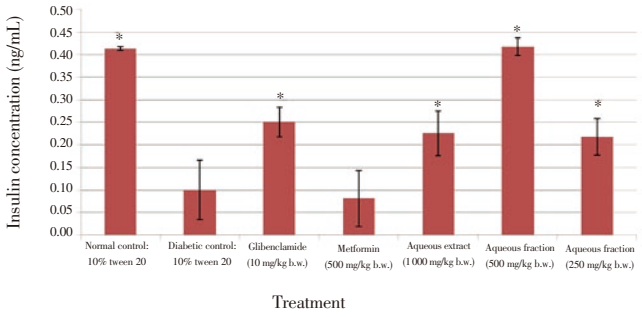

Figure 6. Antihyperglycemic effect of oral administration of *P. bleo* aqueous extract and aqueous fractions on plasma insulin of STZ-induced diabetic rats.

Data are presented as mean ± SEM (n = 6). *Significant difference from diabetic control (P<0.05).

3.6.2. Effect on lipid profile

The lipid profile of *P. bleo* aqueous extract (1 000 mg/kg b.w.) and aqueous fractions (500 mg/kg b.w. and 250 mg/kg b.w.) is shown in Figure 7. The serum TC, TG and LDL values were observed to be decreased for all treatments. HDL level was found to be increased significantly (P<0.05), except for aqueous extract treated rats which showed an increase in HDL level but not significant when compared to diabetic control.

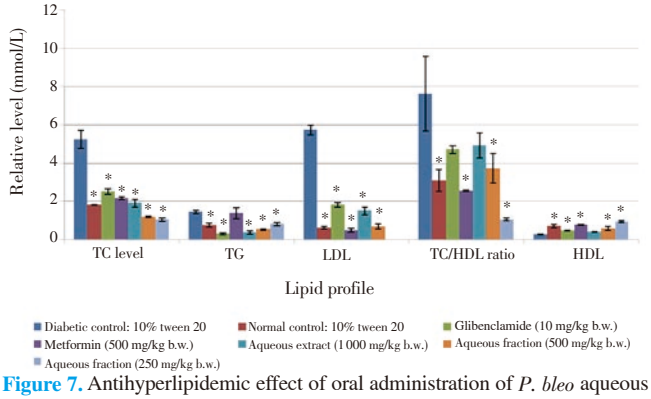

Figure 7. Antihyperlipidemic effect of oral administration of *P. bleo* aqueous extract and aqueous fractions on lipid profile of STZ-induced diabetic rats.

Data are presented as mean ± SEM (n = 6). *Significant difference from diabetic control (P<0.05).

3.6.3. Effect on body weight

All the diabetic rats reduced some body weight increment after diabetic induction (Figure 8). However, the administration of aqueous extract (1 000 mg/kg b.w.) and aqueous fraction (250 mg/kg b.w.) exhibited significant (P<0.05) effect in ameliorating the decreasing percentage of the body weight in contrast to diabetic control rats.

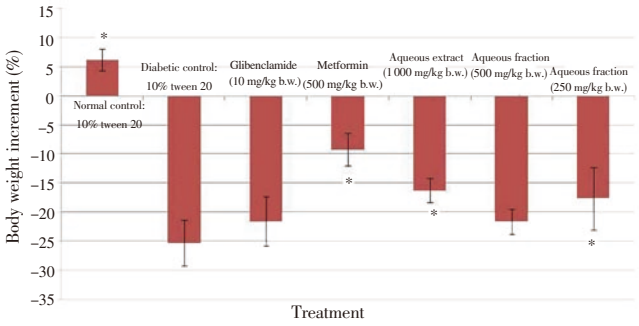

Figure 8. Effect of oral administration of *P. bleo* aqueous extract and aqueous fractions on body weight increment of STZ-induced diabetic rats.
Data are presented as mean ± SEM (n = 6). *Significant difference from diabetic control (P<0.05).

4. Discussion

In the current study, *P. bleo* extracts were investigated for hypoglycemic properties in normal rats following an acute dose of 1 000 mg/kg b.w. The findings distinctly showed that *P. bleo* extracts do not provoke hypoglycemic action on acute dose and are capable to maintain the glycemic condition in non-diabetic rats. Meanwhile, glibenclamide lacks the credibility to regulate normal blood sugar level, which had exhibited functional hypoglycemic action in non-diabetic rats. This is because glibenclamide operates by provoking the β-cells in producing additional insulin from pancreas regardless of the glycemic status of the body[13]. Apparently, the results indicate that *P. bleo* extracts may not manage the blood glucose levels through insulin secretion at an acute dose. This condition is very favourable as *P. bleo* extracts can be potential candidates for diabetes therapy replacing many clinical drugs that yield adverse reaction such as hypoglycemia. Our findings are the first one to demonstrate that acute dose of *P. bleo* extracts prevents hypoglycemia in normal rats.

P. bleo extracts had compelling glucose resilience effects in normal rats. All the extracts demonstrated remarkable abilities to abstain the sudden arise of blood sugar level upon glucose injection. Though the reductions in glucose by petroleum ether, chloroform and aqueous extract were only significant at the end of 120th minute, the glucose lowering pattern by these extracts was comparable to that of standard drug used. In contrast, metformin showed significant blood glucose reduction as soon as 30th minute of the test. This is expected as metformin performs by extra-pancreatically obstructing hepatic gluconeogenesis without raising the insulin production[14-16]. The similarity in the glucose lowering pattern as metformin may suggest that *P. bleo* extracts may be slow acting and require more time for the bioactivity potential.

The mechanism underlying the antidiabetic property of *P. bleo* extracts was further investigated. In this study, human insulin-dependent diabetes mellitus was portrayed in animal model using STZ-induced diabetic rats[17]. STZ forms reactive oxygen species that destructs the pancreatic β-cells thus leading to a condition where insulin is reliant on[18]. Therefore, to overcome the drastic reduction of blood insulin concentration, the induced rats were immediately given 5% glucose solution *ad libitum* which had aided in regulating the insulin production and glucose restoration[14,19]. In current study, blood glucose readings were recorded on day 0, 3, 6, 9 and 12. Aqueous extract of *P. bleo* had significantly decreased blood glucose levels from day 6 till day 12 compared to other extracts. The results disclosed that the 12 days of routine treatment of *P. bleo* aqueous extract had stopped the blood sugar increase in STZ-diabetic rats. According to Arunachalam, the pancreatic release of insulin from the remaining β-cells may contribute to the antihyperglycemic response by the aqueous extract[20]. Another possibility is that, after the eradication of β-cells by STZ, the aqueous extract administration had stimulated the regeneration of β-cells which eventually secretes insulin to reduce the blood sugar level in rats[21]. Notably, the aqueous extract presented a superior blood sugar lowering activity in the 12 days of investigation compared to intraperitoneal glucose tolerance test. This could be due to the aqueous extract that required initial period of bioaccumulation to concert the antidiabetic effect[22].

The aqueous extract being the most potent in antidiabetic activity compared to other solvent extracts of *P. bleo*, was further fractionated serially using different solvents of varying polarity. Of all the fractions studied, aqueous fraction demonstrated a strong antihyperglycemic property by significantly lowering the blood glucose level as early as day 6 of the experiment. Hence, a subsequent dose response relationship test was conducted on the aqueous fraction and the results showed that two different doses of 500 and 250 mg/kg b.w. of the aqueous fraction had significantly lowered the blood sugar level of the diabetic rats. These two doses of aqueous fraction along with aqueous extract were chosen to be tested on for insulin level, body weight and antihyperlipidemic properties.

Insulin is one of the major hormones that regulates the blood glucose concentration. As stated by Ali *et al*, the most appropriate treatment for diabetes can be optimized by determining the insulin level in blood[22]. Previously, various studies have proved that antihyperglycemic effect of plant extracts can be accredited to their insulin-trophic influence[23,24]. This is consistent with our results which had revealed the administration of 500 mg/kg b.w. of *P. bleo* aqueous fraction had escalated the plasma insulin significantly in contrast to diabetic control. Thereby, the aqueous fraction displayed direct insulinotropic action by stimulating the plasma insulin to improve the blood sugar level. Diversely, metformin exhibited the lowest insulin level which verifies its blood glucose reducing mechanism is through extrapancreatic cells.

Besides managing the blood glucose level, the aqueous fractions of *P. bleo* showed potent antihyperlipidemic activity by controlling lipid contents of the diabetic rats. The aqueous fractions have lowered the TC, TG and LDL levels in STZ-induced diabetic rats followed by a significant increase of HDL contrast to the diabetic rats. Earlier, Mat Darus reported the similar lipid profile by the aqueous extract of *P. bleo*[12]. Probably, the aqueous fractions of *P. bleo* also regulate the fatty acids flow. The serum lipid profile by the *P. bleo* aqueous fractions directly denotes its protective effects in diabetes associated complication such as cardiovascular disease. In diabetic condition, the imbalance of the metabolic pathways often contributes to a body

weight decrease[25]. However, aqueous extract (1 000 mg/kg b.w.) and aqueous fraction (250 mg/kg b.w.) of *P. bleo* had significantly reduced the body weight loss. According to Adiga *et al*, successful antidiabetic treatments are able to avoid body weight loss by monitoring muscle wasting[26].

In conclusion, the overall study proved *P. bleo* as potential ethnomedicinal plant having remarkable antidiabetic and antihyperlipidemic activities. Aqueous fraction of *P. bleo* which possesses promising clinical value awaits further research and development as a new chemotherapeutic agent.

Conflict of interest statement

We declare no conflict of interest.

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