ANTIOXIDANT AND ANTIDIABETIC PROPERTIES OF *TAMARINDUS INDICA* LEAF ETHANOLIC EXTRACT FROM MALAYSIA

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Abstract. *Tamarindus indica* (*T. indica*; Family Leguminosae) is widely used in various traditional medicine and food preparations. Antioxidant and antidiabetic activities of *T. indica* leaf extracts from Malaysian macerated (TIME) and Soxhlet (TISE) were investigated. In TIME and TISE, total phenolic (TP) content was 1.80 mg gallic acid equivalent (GAE)/g and 1.01 mg GAE/g respectively, and total flavonoid (TF) content 1.44 mg rutin equivalent (RUE)/g and 1.04 mg RUE/g respectively. TIME was selected for further studies due to its higher TP and TF contents. Using 2,2-diphenyl-1-picrylhydrazyl and 2,2′-azino-bis-3-ethylbenzothiazoline-6-sulphonic acid radical scavenging assays, TIME radical scavenging capacity was 1.42±0.3 µg/ml and 1.62±0.66 µg/ml, respectively; and employing α-amylase and α-glucosidase inhibition assays, TIME *in vitro* antidiabetic ability was 2.24±0.07 µg/ml and 2.26±0.07 µg/ml. Acute oral toxicity study in rat revealed TIME was safe up to 2,000 mg/kg body weight (BW), and treatment with 200 mg/kg BW TIME significantly lowered elevated blood glucose levels to those of glucose-loaded normoglycemic and streptozotocin-induced diabetic rats. The results suggest TIME from Malaysia has therapeutic potential as a natural product antioxidant and antidiabetic.

Keywords: *Tamarindus indica*, antidiabetic, antioxidant, polyphenol, streptozotocin

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INTRODUCTION

Medicinal plants are utilized in alternative medicine to treat various illnesses, such as metabolic diseases such as diabetes or diabetes-associated oxidative stress with fewer side effects

compared with synthetic drugs (Wang *et al*, 2013). Such medicinal plants contain phytochemical constituents or their secondary metabolites, which can be beneficial for therapeutic purposes, such as antidiabetics and antioxidants, and in certain circumstances have similar properties as conventional pharmaceutical drugs (Vikram *et al*, 2014).

Recently there has been an increase in interest and research in medicinal plants for cure and management of diabetes mellitus in developing and developed countries (Rahimi-Madiseh *et al*, 2016). Hypoglycemic properties of plant extracts are normally determined by their ability to inhibit α-amylase and α-glucosidase, enzymes responsible for breakdown in the body of large sugar molecules to monosaccharides (Kazeem *et al*, 2013). Inhibition of α-amylase and α-glucosidase enable prolonged carbohydrate digestion time and lead to a marked decrease in rate of glucose absorption resulting in an antidiabetic effect (Mohamed *et al*, 2015).

Tamarindus indica, commonly known as tamarind, is a well-known medicinal plant globally and different parts of the plant have been well explored for their biological activities (Azman *et al*, 2012; Agnihotri and Singh 2013; Sole *et al*, 2013; Chigurupati *et al*, 2018a) including antioxidant and antidiabetic properties (Maiti *et al*, 2004; Rehana *et al*, 2017). Although many studies have been performed on *T. indica* extracts from various geographical origins (Rao *et al*, 1999), there are a limited number of reports on antioxidant and antidiabetic properties of the plant of Malaysian origin (Rao *et al*, 1999).

T. indica leaves are readily available and they also used in foods in certain parts of Malaysia. Since the antioxidant and antidiabetic potentials of *T. indica* obtained from Malaysian geographical zone is not yet been explored, our research focused on its *in vitro* pharmacological activities so that it could be beneficial for future *in vivo* studies. Here, in the present study, we targeted *T. indica* leaves for finding the flavonoid and phenolic contents and then investigating for antidiabetic and antioxidant potentials.

MATERIALS AND METHODS

Chemicals

Ascorbic acid and 2,2-diphenyl-1-picrylhydrazyl (DPPH) were obtained from Merck Millipore Co (Temecula, CA); and acarbose, α-amylase, 2,2′-azino-bis-3-ethylbenzothiazoline-6-sulphonic acid (ABTS), dinitrosalicylic acid (DNSA), glibenclamide, α-glucosidase, and streptozotocin (STZ) from Sigma Chemicals Co (St Louis, MI). All other chemicals were of analytical grade.

Experimental

Healthy Sprague Dawley rats (6-8 weeks of age, 150-180 g), obtained from the central animal house, Asian Institute of Medicine, Science and Technology (AIMST) University, Malaysia were housed at 25±2°C, 45-55% relative humidity, with a 12-hour dark and light cycle and fed commercial rat pellet and water *ad libitum*. Test solutions were administered by gavage. Experimental protocols were approved by AIMST University Animal Ethics Committee (permit no. AUHAEC8/FOP/2016).

Plant material and extract preparations

T. indica leaves were obtained from AIMST University, Kedah, Malaysia, authenticated by (Dr. Mukesh Singh, Department of Pharmaceutical Chemistry and Pharmacognosy, AIMST University where a reference sample was deposited (ref. no. AIMST/FOP/12). Mature and

healthy *T. indica* leaves were hand-picked, stems removed, washed in tap water to remove soil and dirt, dried in shade, coarsely ground using a blender, and stored out of direct sunlight at ambient temperature until used.

The extraction of *T. indica* leaves were subject to two methods of solvent extraction, namely, maceration and Soxhlet extractions. In the former technique, 200 g of coarsely powdered *T. indica* leaves were added to 500 ml of absolute alcohol, shaken using a mechanical shaker (Orbital Shaker, Thermo Fisher Scientific, Waltham, MA) at 100 rpm for seven days. Maceration was filtered and filtrate concentrated under reduced pressure in a rotary flash evaporator (Rotavapour R-220 Pro, Buchi, Flawil, Switzerland) and freeze-dried (labelled TIME) (Chigurupati *et al*, 2018a). In Soxhlet extraction technique, 20 g of coarsely powdered leaves were continuously extracted with 250 ml of absolute ethanol using a Soxhlet apparatus (Fischer Scientific, Marsiling Industrial Estate Road, Singapore) until the solvent was colorless (or nearly so) and supernatant concentrated and freeze-dried (labelled TISE) (Parimala *et al*, 2009).

Qualitative phytochemical tests

Stock solutions of TIME and TISE were prepared at 1 mg/ml and used in qualitative phytochemical tests as previously describe (Jaradat *et al*, 2015; Chigurupati *et al*, 2018b). Test for alkaloids was conducted by incubating 5 ml aliquot of each extract with 2% (v/v) H_2SO_4 at 30°C for two minutes, filtered and few drops of the following reagents were added separately: Mayer's reagent (formation of opalescence or yellowish precipitate indicating occurrence of alkaloids) and Dragendorff's reagent (formation of turbid solution or precipitate indicating occurrence of alkaloids). Test for flavonoids was performed by adding to 2 ml of each plant extract a few drops of 10% (w/v) lead acetate solution (formation of a yellow precipitate indicating occurrence of flavonoids). Test for phenols was carried out by adding to 2 ml aliquot of each extract a few drops of 5% (w/v) ferric chloride solution (formation of a dark green color indicating occurrence of phenols). Test for tannins was conducted by adding to 2 ml aliquot of each extract 3-4 drops of 5% (w/v) ferric chloride solution (appearance of bluish-green color indicating occurrence of tannins). Test for glycosides was carried out by adding 1 ml of 0.1 M NaOH solution to 1 ml aliquot of each extract and making up to 10 ml with distilled water and distilled water (appearance of a yellow color indicating occurrence of glycosides). Test for saponins was performed by adding 2 ml aliquot of each extract to 2 ml of 5% (w/v) $NaHCO_3$ solution, followed by vigorous shaking and allowing to stand for three minutes (formation of a honeycomb-like foam indicating occurrence of saponins).

Tests for other biological constituents were carried as follows: (i) presence of protein by adding 2 ml aliquot of each extract to an equal volume of 0.1 M NaOH solution, followed by gentle mixing and addition of 1% (w/v) copper sulfate solution (a violet color indicating presence of protein) (Narasimhan, 2014); (ii) presence of monosaccharides by adding 2 ml aliquot of each extract to 1 m of Barfoed's reagent (Central Drug House (P) Ltd, New Delhi, India), followed by heating at 100°C for two minutes (red precipitate indicating presence of monosaccharides) (Weiner and Harrison 2009); (iii) presence of sterols by mixing 2 ml aliquot of each extract with 2 ml

of chloroform and 2 ml of 0.1 M H_2SO_4 solution (red chloroform layer and greenish-yellow fluorescent aqueous layer indicating presence of sterols) (Auwal *et al*, 2014); (iv) presence of amino acids by adding to 2 ml aliquot of each extract a few drops of 5% (w/v) ninhydrin solution, followed by heating at 100°C for 10 minutes (purple color indicating presence of amino acids) (Weiner and Harrison 2009); (v) presence of carbohydrates by adding to 2 ml aliquot of each extract a few drops of Molisch's solution (Fischer Scientific, Selangor, Malaysia) and then allowing few drops of 0.1 M H_2SO_4 solution to run down the side reaction tube without shaking (purple color indicating presence of carbohydrates (Weiner and Harrison 2009); (vi) presence of reducing sugars by adding 2 ml aliquot of each extract to equal volumes of Fehling's solutions I and II (Fischer Scientific, Selangor, Malaysia) followed by heating in boiling water for two minutes (brick-red precipitate indicating presence of reducing sugars) (Weiner and Harrison 2009); (vii) presence of non-reducing sugars by adding 2 ml aliquot of each extract to 2 ml of Benedict's reagent II (Fischer Scientific, Selangor, Malaysia) and heating in boiling water for 3 minutes (reddish-brown color indicating presence of non-reducing sugars) (Weiner and Harrison 2009); and (viii) presence of mucilage and gums by drop-wise adding of 10 ml aliquot of each extract to 25 ml of absolute alcohol with gentle agitation (precipitation indicating presence of gums and mucilage) (Weiner and Harrison 2009).

Quantitative phytochemical determination

Total phenolic content

Total phenolic (TP) contents of TIME and TISE were determined as previously described (Chigurupati *et al*, 2017b). In brief, a solution of 100 µl aliquot of each extract (1 mg/ml 70% ethanol), 2 ml of 2% (w/v) Na_2CO_3 solution and 100 µl of Folin-Ciocalteu reagent (Fischer Scientific, Selangor, Malaysia) was incubated at ambient temperature for 30 minutes, then $A_{750\ nm}$ measured. A standard curve using gallic acid (GA) (0.1-1.0 mg/ml 70% ethanol) was constructed ($r^2 = 0.81$) and TP content expressed as mg of GA equivalent per gram of plant extract (mg GAE/g).

Total flavonoid content

Total flavonoid (TF) contents of TIME and TISE were measured as previously described (Chigurupati *et al*, 2017b). In short, a solution of 1 ml aliquot of each extract (1 mg/ml 70% ethanol), 10 ml of 30% ethanol and 0.7 ml of 5% (w/v) $NaNO_2$ and 10% (w/v) $AlCl_3$ was incubated at ambient temperature for 6 minutes, then 10 ml aliquot of 1 M NaOH solution was added, solution adjusted to 25 ml with 30% ethanol and incubated at ambient temperature for 10 minutes before $A_{450\ nm}$ measurement. A standard curve using rutin (RU) was constructed as described above ($r^2 = 0.97$) and TF content expressed as mg RUE/g.

***In vitro* antioxidant assays**

DPPH assay

Capacity of TIME to scavenge DPPH radicals was determined as previously described with minor modifications (Chigurupati *et al*, 2017a; Salar *et al*, 2017). A solution containing 0.5 ml of extract (1 mg/ml 70% ethanol) and 0.5 ml of 2 µM DPPH were mixed in test tubes and incubated at ambient temperature for 20 minutes, then $A_{517\ nm}$ measured. Ascorbic acid (10-1,000 mg/70% ethanol) was used as a standard and 1 ml of 2 µM DPPH was used as control. Percent scavenging capacity was calculated using Eq 1 and the concentration of a drug required for 50% inhibition *in vitro* (IC_{50}) values

for both sample and ascorbic acid were obtained from the graph of percentage free radical inhibition (y-axis) *vs* concentration (x-axis).

$$[(A_{517\ nm\ control} - A_{517\ nm\ sample})/ A_{517\ nm\ control}] \times 100 \quad \text{(Eq 1)}$$

ABTS assay

ABTS free radical cation scavenging capacity of TIME as previously described (Chigurupati *et al*, 2017a). A solution of 0.5 ml of extract (prepared as described above) and 0.5 ml of 7 mM ABTS solution were mixed in test tube and incubated at ambient temperature for 30 minutes, then $A_{734\ nm}$ measured. Percent scavenging capacity was calculated using Eq 1 and the IC_{50} values for both sample and ascorbic acid were obtained from the graph of percentage free radical inhibition (y-axis) *vs* concentration (x-axis).

***In vitro* antidiabetic assays**

α-Amylase inhibition assay

In vitro antidiabetic activity of TIME using an α-amylase inhibition assay was performed as previously described (Salar *et al*, 2019). A solution of 0.5 ml of extract (1 mg/ml 70% ethanol) and 0.5 ml of α-amylase (2 U/ml 0.2 mM phosphate buffer pH 6.9) was incubated at 25°C for 10 minutes, then 0.5 ml aliquot of 1% (w/v) starch solution in above mentioned buffer was added and the mixture further incubated at 25°C for 10 minutes, followed by addition of 1 ml of DNSA reagent (12 g of sodium potassium tartrate tetrahydrate in 8.0 mL of 2 M NaOH and 20 ml of 96 mM of 3,5-dinitrosalicylic acid solution) in same buffer solution. The reaction mixture was heated in boiling water for five minutes, cooled to room temperature, then 10 ml of distilled water added and $A_{540\ nm}$ measured. A standard curve using acarbose (0.1-1.0 mg/mL) was constructed as previously described (Chigurupati *et al*, 2019) and percent α-amylase inhibition calculated using the following Eq 2. Control was also made similarly except that 0.5 ml of extract was replaced with 0.5 ml of distilled water and acarbose (0.1-1.0 mg/mL) was used as standard.

$$[(A_{540\ nm\ control} - A_{540\ nm\ sample})/ A_{540\ nm\ control}] \times 100 \quad \text{(Eq 2)}$$

α-Glucosidase inhibition assay

In vitro antidiabetic activity of TIME using an α-glucosidase inhibition assay was performed as previously described (Kajaria *et al*, 2013). A mixture of 1 ml of extract (prepared as described above) and 1 ml of α-glucosidase (1 U/ml) was incubated at 37°C for five minutes, then 1 ml of 2% (w/v) sucrose in Tris buffer pH 8 was added and solution further incubated at 37°C for 20 minutes before the reaction was terminated by heating in boiling water for two minutes. The solution was cooled to ambient temperature and $A_{505\ nm}$ of released glucose measured according to GOD-POD method (Yeye *et al*, 2020). Acarbose standard curve (0.1-1.0 mg/ml) was constructed and percent of α-glucosidase inhibition calculated using Eq 2.

***In vivo* assays**

Acute oral toxicity assay

Acute oral toxicity assay of TIME was carried out according to OECD guidelines (OECD, 2001; Kesavanarayanan *et al*, 2013). Overnight fasted female rats ($n = 3$) were fed 2 g/kg body weight (BW) extract in 0.5% (w/v) carboxymethyl cellulose (CMC) solution and monitored for clinical signs and mortality for 24 hours, then observed at least once a day for another 14 days.

Effect of TIME in normoglycemic rats

Changes in blood glucose levels at different times following a single oral dose of TIME were evaluated in normoglycemic rats. Overnight fasted male rats were randomly divided into 4 groups (5 rats per group): group 1 (normal control) received 10 ml/kg BW 0.5% (w/v) CMC solution; group 2 received 5 mg/kg BW glibenclamide; group 3 received 100 mg/kg BW TIME; and group 4 received 200 mg/kg BW TIME. Except for group 1, the other group received 2 g/kg BW aqueous glucose solution one hour prior to treatment. All administered solutions were freshly prepared using 0.5% CMC one hour before dosing. Basal blood glucose level was measured using a glucometer (Accu-Chek Aviva Blood Glucose Meter, Roche Diagnostics, Selangor, Malaysia) of a blood sample collected from tail vein under light ether anesthesia. Rats with basal blood glucose level ≥7 mM were not included in the study. Blood glucose level was measured at hour 0, 2, 4, 8 and 12 h post-dosing.

Effect of TIME in STZ-induced diabetic rats

Diabetes was induced in overnight-fasted male rats by a single intraperitoneal injection of 55 mg/kg BW freshly prepared STZ in ice-cold 10 mM citrate buffer pH 4.5. At hour-6 post-injection, rats were orally dosed with 2 ml/kg BW 5% (w/v) aqueous glucose for every 6 hours for the next 24 hours (to prevent hypoglycemic shock). Fasting blood glucose was determined at hour-48 post STZ treatment to confirm induction of diabetes induction (blood glucose ≥12 mM). Diabetic rats were randomly divided into three groups, with one control group (5 rats per group): group 1 (untreated normal control) received 10 ml/kg BW 0.5% (w/v) CMC solution; group 2 received 10 ml/kg BW 0.5% (w/v) CMC solution; group 3 received 200 mg/kg BW TIME; and group 4 received 5 mg/kg BW glibenclamide. All administered solutions were prepared similar to that of the normoglycemic experiment. Blood glucose levels were measured as described above at hours 0, 2, 4, 8 and 12 post-administration.

Statistical Analysis

Results from *in vitro* experiments are expressed as mean ± SEM of three independent measurements. IC_{50} value was determined from a non-linear regression analysis. Results from *in vivo* studies are expressed as mean ± SD of five experiments. Statistical analysis between groups was conducted using a one-way analysis of variance followed by Tukey's multiple comparison *post-hoc* test, with p-value <0.050 considered significant. All calculations were carried out using a Graph Pad Prism Software version 5 (GraphPad Software Inc, San Diego, CA).

RESULTS

Constituents of TIME and TISE

Percent yield of ethanolic TIME and TISE was 39.7% (w/w) (after three macerations) and 25.4% (w/w) respectively. Qualitative analysis of seven phytochemicals, amino acids, protein, sugars, and carbohydrate demonstrated all were present, except reducing and non-reducing sugars, in TIME and TISE (Table 1). TP content in TIME and TISE was 1.80 and 1.01 mg GAE/g respectively and TF content 1.44 and 1.04 mg RUE/g respectively.

Free radical scavenging and *in vitro* antidiabetic capacities

Based on yield and TP and TF contents, TIME was selected for further studies. Using DPPH and ABTS scavenging assays, IC_{50} values for TIME were found to be 1.42±0.3

Table 1
Qualitative screening of phytochemical and other biological constituents of ethanolic *Tamarindus indica* leaf macerated (TIME) and Soxhlet (TISE) extracts.

Constituent	TIME	TISE
Alkaloids	+	+
Amino acids	-	-
Carbohydrates	+	+
Glycosides	+	+
Monosaccharides	+	+
Mucilage and gums	+	+
Non-reducing sugars	-	-
Proteins	+	+
Reducing sugars	-	-
Saponins	+	+
Steroids	+	+
Tannins	+	+

+: present; -: absent.

μg/ml and 1.62±0.66 μg/ml, respectively and for standard drug, ascorbic acid, it was 1.09±0.02 μg/ml and 1.02±0.03 μg/ml, respectively (data not shown).

Similarly, for TIME using *in vitro* α-amylase and α-glucosidase inhibition assays, IC_{50} values were found to be was 2.24±0.07 μg/ml and 2.26±0.07 μg/ml, respectively and for standard drug, acarbose, it was found to be and 2.02±0.02 and 2.11±0.02 μg/ml, respectively (data not shown)

Acute oral toxicity and effect of TIME in normoglycemic and STZ-induced diabetic rats

Acute toxicity test showed no lethality over a 14-day period after dosing with TIME at 2,000 mg/kg BW and normal behavior of animals was observed throughout the clinical observation period. Thus, 1/20th and 1/10th dose of

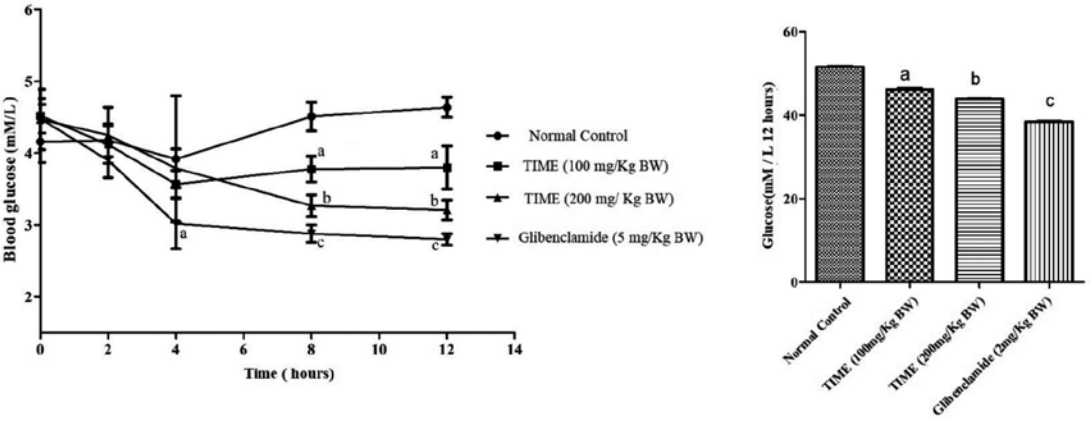

Fig 1-Blood glucose levels in glucose-loaded normoglycemic rats following a single oral dose of *Tamarindus indica* macerated leaf extract (TIME) or glibenclamide.
Overnight fasted male Sprague Dawley rats (5 rats per group) were fed by gavage 10 ml/kg body weight (BW) 0.5% (w/v) carboxymethyl cellulose (CMC) solution (normal control), 100 or 200 mg/kg BW TIME, and 5 mg/kg BW glibenclamide. Except for normal control, all groups received one hour prior to treatment 2 g/kg BW glucose. Blood was drawn from tail vein and blood glucose level was measured with a glucometer. Values are presented as mean± SD.

[a]*p*-value <0.050; [b]*p*-value <0.010; [c]*p*-value <0.001 compared to normal control.

2,000 mg/kg (*ie* 100 and 200 mg/kg BW) were selected for the efficacy study.

Normoglycemic rats treated with TIME at 100 and 200 mg/kg BW exhibited a significant dose-dependent reduction in blood glucose levels at hour 8 and 12 post-dosing compared to untreated controls, but not as effective as the antidiabetic drug glibenclamide (5 mg/kg BW), which manifested a hypoglycemic effect as soon as hour-4 post-dosing (Fig 1).

Diabetic rats (blood glucose level ≥12 mM) induced by a single intraperitoneal injection of STZ (55 mg/kg BW) treated with TIME (200 mg/kg BW) exhibited a significant reduction in blood glucose level at 8 and 12 hours post-dosing compared to untreated diabetic rats (Table 2). As expected, treatment of diabetic rats with glibenclamide (5 mg/kg BW) resulted in significant lowering of blood glucose levels as early as at hour 4 post-dosing compared to untreated diabetic controls (Table 2).

DISCUSSION

Tamarind leaves are considered as a potential source of numerous phytochemicals including phenolic and flavonoid compounds (Escalona-Arranz *et al*, 2014). Ethanolic TIME generated higher TP and TS yields than TISE, in agreement with a previous study (Sankeshwari *et al*, 2018). Food grade solvents, *eg* ethanol, are safe for consumption and plants extracted with such solvents have high TP and TF contents as well as *in vitro* antioxidant activity (Do *et al*, 2014). Highly polar solvents can readily extract hydrophilic phytocompounds such as polyphenols (Tsao, 2010).

DPPH and ABTS tests are widely used to assess *in vitro* antioxidant capacity of compounds of interest as they are easy to carry out and the results are readily measured by spectrophotometric methods (Lü *et al,* 2009). The likely antioxidant mechanism of TIME might be due to the presence of polyphenols (Gautam *et al*, 2012) and their ability to transfer hydrogen atoms to the stable DPPH and ABTS free radicals (Nahar *et al*, 2014).

Polyphenols have been shown to inhibit intestinal absorption of carbohydrates by inhibiting the enzymes α-amylase and α-glucosidase and

Table 2
Blood glucose levels of streptozotocin (STZ)-induced diabetic rats treated with *Tamarindus indica* macerated leaf extract (TIME) or glibenclamide.

Animals (5 animals in each group)	Blood glucose level (mM) post-dosing (Mean±SD)				
	0 hour	2 hours	4 hours	8 hours	12 hours
Normoglycemic control	4.3 ± 0.3	4.5 ± 0.4	4.0 ± 0.1	4.1 ± 0.1	4.1 ± 0.2
STZ-induced diabetic control	24 ± 2	21 ± 3	20 ± 2	19 ± 1	18 ± 1
TIME (200 mg/kg BW) treated	26 ± 1	23 ± 2	20 ± 1	13 ± 1*	12.0 ± 0.5**
Glibenclamide (5 mg/kg BW) treated	24 ± 1	16 ± 0.9	14 ± 1*	11 ± 1**	8 ± 1**

*p-value <0.010; **p-value <0.001 compared to STZ-induced diabetic control; BW: body weight; mM: milli moles; SD: standard deviation

helping to provide better management of postprandial blood glucose level (Noreen *et al*, 2017). The *in vitro* antidiabetic property of TIME could be attributed to the presence of polyphenols (Gautam *et al*, 2012; Nahar *et al*, 2014).

In acute oral toxicity test of rat, TIME up to a dose 2 g/kg BW was considered as orally safe (Nahar *et al*, 2014). At a presumed therapeutic dose (10% of maximum safe oral dosing), TIME significantly reduced elevated blood glucose levels in glucose-loaded normoglycemic and STZ-induced diabetic rats. Treatment with methanol extract of tamarind seeds at the same dosage produced a significant decline in blood glucose levels in normoglycemic fasted mice (Nahar *et al*, 2014). Administration of aqueous extract of tamarind seeds has been reported to promote glycogenesis and attenuate altered activity of carbohydrate metabolizing key enzymes such as glucose-6-phosphate dehydrogenase and glucose 6-phosphatase in STZ-induced diabetic rats (Maiti *et al*, 2004). The antihyperglycemic effect of TIME might be attributed to its free radical scavenging property and rich polyphenolic content.

In conclusion, the study contributes to the knowledge on use of *Tamarindus indica* leaf extract as an antioxidant and antidiabetic agent, and its safety profile (in a rat model) indicates its potential to be developed as a safe treatment of type 2 diabetes in humans. However, further studies into its pharmacological profile and appropriate formulation will be needed before this natural product comes into clinical trial as an alternative to synthetic drugs in the treatment of diabetic patients.

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