ISSN 1996-0700

Asian Journal of Biotechnology

http://knowledgiascientific.com

Knowledgia SCIENTIFIC
A Place to Publish Outstanding Research

OPEN ACCESS Asian Journal of Biotechnology

ISSN 1996-0700
DOI: 10.3923/ajbkr.2020.1.8

Research Article
Antioxidant and Antidiabetic Properties of *Mimosa pudica* Seeds in Streptozotocin-induced Diabetic Wistar Rats

[1]Rita Maneju Sunday, [2]Efere Martins Obuotor and [1]Anil Kumar

[1]School of Biotechnology, Devi Ahilya University, Khandwa Road, 452001 Indore, India
[2]Department of Biochemistry and Molecular Biology, Faculty of Biological Sciences, Obafemi Awolowo University, Ile-Ife, Nigeria

Abstract

Background and Objectives: In India and many other Asian countries, traditional medicinal plants are used for treatment of various diseases. Under the category, *Mimosa pudica* is used as a traditional medicine for the treatment of numerous diseases including diabetes. In recent time, diabetes has become much common disease perhaps due to changed life style. Therefore, in the present study, antidiabetic effect of *Mimosa pudica* seeds ethanolic extract and extract fractions (n-butanol, ethyl acetate and n-hexane) was investigated *in vivo* in Wistar rats. Antioxidant study was carried out using *in vitro* and *in vivo* models. **Materials and Methods:** The free radical (2,2-Diphenyl-1-picrylhydrazyl and nitric oxide) scavenging activity and total antioxidant (flavonoid and phenol) contents of the extracts were determined *in vitro* by spectrophotometric method. In the *in vivo* studies, 100, 200 and 400 mg kg^{-1} *Mimosa pudica* seeds ethanolic extract and 200 mg kg^{-1} of each extract fractions were administered to streptozotocin (60 mg kg^{-1}) induced diabetic Wistar rats. **Results:** *Mimosa pudica* seed extracts possess moderate amount of antioxidants and scavenged 2,2-Diphenyl-1-picrylhydrazyl and nitric oxide free radicals. *Mimosa pudica* seed extracts also decreased fasting blood glucose level and lipid peroxidation (thiobarbituric acid reactive substances) and increased serum insulin, serum α-amylase and hepatic antioxidants (catalase and reduced glutathione) levels in 21 days treated diabetic rats. The ethyl acetate extract fraction scavenged 2,2-Diphenyl-1-picrylhydrazyl (65.41%) and nitric oxide (90.16%) radicals than the other extracts. The ethyl acetate extract fraction also exerted more antidiabetic activity and increased hepatic antioxidant levels than the other extracts and the untreated group. **Conclusion:** *Mimosa pudica* seeds exerted its antidiabetic effect by enhancing secretion of pancreatic insulin and α-amylase. *Mimosa pudica* seeds also possess antioxidant activity and therefore, can be employed in preventing hepatic damage associated with diabetes.

Key words: *Mimosa pudica*, seeds, diabetes, antioxidants, Wistar rats, streptozotocin, serum insulin, metabolic disorders

Citation: Rita Maneju Sunday, Efere Martins Obuotor and Anil Kumar, 2020. Antioxidant and antidiabetic properties of *Mimosa pudica* seeds in streptozotocin-induced diabetic Wistar rats. Asian J. Biotechnol., 12: 1-8.

Corresponding Author: Anil Kumar, School of Biotechnology, Devi Ahilya University, Khandwa Road, 452001 Indore, India Tel: +91 9425058373

Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Abnormally high levels of free radicals produced during normal biological processes have significant effect in the causation of diabetes as well as diabetes related complications in humans[1]. Diabetes mellitus is a metabolic disorder that has to do with the inability of the pancreas to produce insulin (type 1 diabetes mellitus) and also due to the resistance of body cells to insulin (type 2 diabetes)[2]. In diabetes, oxidative stress co-exists with a reduction in the anti-oxidant status due to the presence of high levels of free radicals (reactive oxygen species and reactive nitrogen species)[3-5]. High concentrations of these radicals may lead to diseases including diabetes[1], cardiovascular disease[6], Alzheimer's disease[7] and Parkinson's disease[8]. The harmful effect of free radicals is neutralized by endogenous antioxidants produced in the body and exogenous antioxidants gotten from the diet. Endogenous antioxidants include catalase, glutathione reductase, superoxide dismutase[9] and exogenous antioxidants received from the diet include vitamin C, vitamin E, β-carotene, lycopene and polyphenols (flavonoids, flavones and flavonols)[10]. Medicinal plants are major sources of antioxidants and some medicinal plants also possess antidiabetic properties[11-13]. *Mimosa pudica* L., belonging to the family Fabaceae is a plant with numerous common names viz. Humble Plant, Sensitive Plant, Touch-Me-Not, Sleepy Plant, Laajvanti and Chui-Mui[14]. The plant is found Europe, Africa and Asia[14]. All parts of *Mimosa pudica* are used as herbs for the treatment of various diseases. The roots are used for the treatment of dysentery, leprosy, fever and insomnia[15]. The leaves are used for the treatment of haemorrhoids and urinary tract infections[15]. Research works carried out on the roots of *M. pudica* documented that it has anti-fertility[16] and wound healing effect[17]. Studies carried out on the leaves of *Mimosa pudica* showed that the leaves possess antidepressant[18] and anticonvulsant activity[19]. Previous antidiabetic studies carried out on *M. pudica* using animal model showed that the leaves ethanolic extract reduced blood glucose level in alloxan-induced diabetic Wistar rats[20], whereas the whole plant ethanolic extract reduced blood glucose level in streptozotocin-induced diabetic Wistar rats[21,22].

In this study, the antioxidant and antidiabetic effects of *Mimosa pudica* seeds ethanolic extract and extract fractions (n-butanol, ethyl acetate and n-hexane) were investigated due to the traditional use of the plant for the treatment of various diseases including diabetes. The antidiabetic mechanism of *Mimosa pudica* seeds extracts was also investigated in streptozotocin-induced diabetic Wistar rats.

MATERIALS AND METHODS

This research work was carried out in 5 months (September, 2018 to February, 2019) in the School of Biotechnology, Devi Ahilya University, Indore, India.

Plant material: *Mimosa pudica* pulverized seeds was a kind gift from AMSAR Private Limited, Indore, India.

Extraction procedure: The dried pulverized seeds of *Mimosa pudica* were macerated in 70% ethanol for 72 h and the suspension was filtered using Whatman No.1 filter paper[6]. The filtrate (ethanolic extract) was concentrated to a solid form at 45°C and stored in a refrigerator at 4-8°C prior to use.

Fractionation of ethanolic extract: A mass of 5 g *Mimosa pudica* seeds ethanolic extract was prepared in 20 mL distilled water and thereafter was partitioned using various solvents of different polarities in the following order: n-hexane, ethyl acetate and n-butanol (one solvent at a time) to obtain the respective fraction[23]. (All the 4 reagents viz. ethanol, n-hexane, ethyl acetate and n-butanol were purchased from Merck Specialities Pvt. Ltd., Mumbai, India). A volume of 50 mL n-hexane was added to the liquid paste of *Mimosa pudica* seeds ethanolic extract (5 g *M. pudica* seeds ethanolic extract in 20 mL distilled water) and swirled gently for 10 min. The mixture was allowed to stand for 30 min in a separating funnel and the n-hexane fraction was then collected in a beaker. The same method was used for the preparation of ethyl acetate and n-butanol extract fractions. Each of the partitioned fractions was air dried and stored in a refrigerator at 4-8°C prior to use.

In vitro antioxidant assay

Qualitative antioxidant assays: The ethanolic extract at 62.5, 125, 250, 500 and 1000 μg extract mL^{-1}, each extract fractions at 62.5 μg mL^{-1} and standard (acarbose) at 100 μg mL^{-1} distilled water was used for the free radical scavenging assays.

2,2-Diphenyl-1-picrylhydrazyl (DPPH) radical scavenging assay: A volume of 1.0 mL of 0.3 mM DPPH in methanol was added to 1.0 mL of samples in different test tubes. The contents were mixed and incubated in the dark for 30 min and thereafter absorbance was read at 517 nm against a reagent blank[24]. The percentage radical scavenging activity (RSA) of the samples was calculated using the equation below Eq. 1:

$$\text{RSA (\%)} = \frac{\text{Absorbance of reagent blank} - (\text{Absorbance of test} - \text{Absorbance of test blank})}{\text{Absorbance of reagent blank}} \times 100 \quad (1)$$

Nitric oxide (NO) radical scavenging assay: A 3 mL of 10 mM sodium nitroprusside in 0.2 M phosphate buffered saline (pH 7.4) was added to 1.0 mL of samples in different test tubes and then incubated for 60 min at 25°C. After incubation, 5.0 mL of Griess reagent (1% sulfanilamide, 0.1% naphthylethylene-diamine-dihydrochloride in 2% H_3PO_4) was added and absorbance was measured at 546 nm[25,26]. The percentage radical scavenging activity of the samples was calculated using Eq. 1 written above.

Quantitative antioxidant assay: The concentration of the ethanolic extract and each of the extract fractions used for the quantitative antioxidant assay was 1.0 mg mL^{-1}.

Total antioxidant capacity (TAC) assay: A 1.0 mL of reagent (0.6 M sulfuric acid, 28 mM sodium phosphate and 4 mM ammonium molybdate) was added to 1.0 mL of the samples in different test tubes and then incubated in a water bath at 95°C for 30 min. Thereafter, the mixture was cooled to room temperature and the absorbance was measured at 695 nm against a reagent blank[27]. The total antioxidant capacity of the extracts expressed as mg standard (ascorbic acid) equivalent/g of the plant extract was calculated using the following Eq. 2:

$$C = c.\frac{v}{m} \quad (2)$$

Where:

C = Total antioxidant (mg) standard
c = Concentration of standard established from the calibration curve (mg mL^{-1})
v = Volume of the extract (mL)
m = Weight of the extract (g)

Total flavonoid content assay: To 0.5 mL of the samples in different test tubes, 0.3 mL of 5% sodium nitrite was added. After 5 min, 0.3 mL of 10 % aluminum chloride was added and subsequently after 6 min, 2.0 mL of 1 M sodium hydroxide was added and the volume was made up to 5.0 mL with distilled water. The absorbance was measured at 510 nm against a reagent blank[27]. The total flavonoid contents expressed as mg standard (quercetin) equivalents/g of the plant extract was calculated using Eq. 2 written above.

Total phenol content assay: To 0.2 mL of the samples in different test tubes, 8.0 mL of 1 M Folin-Ciocalteu's phenol reagent was added. The mixture was vortexed and after 5 min, 1 mL of saturated sodium carbonate solution (8% w/v) was added and the volume was made up to 3 mL with distilled water. The reaction was kept in the dark for 30 min and the absorbance was measured at 765 nm against a reagent[25]. The total phenolics contents of the extracts expressed as mg standard (gallic acid) equivalent/g of extract were calculated using Eq. 2 written above.

***In vivo* studies in streptozotocin-induced diabetic Wistar rats:** The experiments using Wistar rats were carried out after approval (approval No. 779/CPCSEA/IAEC/2018/010) from the Institutional Animal Ethics Committee, Devi Ahilya University, Indore, India. The approved guidelines were strictly followed.

Animals: Wistar rats of both sexes (200-220 g) were procured from the Institutional Animal house of the University. These rats were kept in well ventilated polypropylene rat cages, fed with broilers mash and were given water *ad libitum*. The rats were allowed to acclimatize with the environment, under natural day light and night conditions for 2 weeks before starting the experiments.

Acute toxicity studies: Three Wistar rats/group (6 groups) were orally administered 10, 100, 1000, 1600, 2900 and 5000 mg extract/kg body weight (group 1-6, respectively) and were monitored for signs of toxicity/mortality every hour for the first 4 h and daily for 3 days after acute administration of the ethanolic extract[28].

Antidiabetic and *in vivo* antioxidant studies in streptozotocin-induced diabetic Wistar rats: Wistar rats were fasted overnight (for 12 h) and diabetes was induced by a single intra-peritoneal injection of 60 mg streptozotocin/kg body weight (b.wt.). After seventy 2 h, blood was drawn from the vein of the tail and the fasting blood glucose level (FBGL) was measured using glucometer and glucose strips (Accu-Check Active Glucometer, model: GC0088, Mannheim Germany). Animals with FBGL above 11.1 mmol L^{-1} were selected for the experiment. The streptozotocin-induced diabetic rats were randomly divided into 8 groups with 6 rats each. Group 1, normoglycemic Wistar rats (5 mL distilled water only), group 2-4 (100, 200 and 400 mg ethanolic extract/kg b.wt., respectively), group 5 (200 mg kg^{-1} n-butanol extract fraction), group 6 (200 mg kg^{-1} ethyl acetate extract fraction), group 7 (200 mg kg^{-1} ethyl acetate extract), group 8 (5 mg glibenclamide/kg b.wt.) and group 9, control (10 mL distilled water/kg). The extracts and glibenclamide were administered (p.o.) daily for 21 days and

the FBGL was measured[29] on day 0, 4, 7, 14 and 21. On the 21st day, animals were sacrificed and blood samples were collected by cardiac puncture into plain bottles. The serum was collected for insulin[30] and α-amylase assay[31]. The rats liver were also immediately harvested, washed in ice cold normal saline, then in 0.15 M Tris-HCl (pH 7.4), blotted dry and weighed. Liver homogenate (10% w/v) prepared in 0.15 M Tris-HCl, pH 7.4 buffer was used for the estimation of lipid peroxidation (by measuring thiobarbituric acid reactive substance)[32], reduced glutathione[33] and catalase[34].

Statistical analysis: The results were analyzed using one-way analysis of variance (ANOVA) followed by Dunnett *post hoc* multiple comparisons tests at 95% ($p<0.05$) level of significance using Primer (version 3.01). All results were expressed as mean±standard error of mean (SEM). The 50% inhibition (IC_{50}) of the plant extract was calculated from graph plotted using excel.

RESULTS

Radical scavenging activity of *Mimosa pudica* seeds extracts: *Mimosa pudica* seeds ethanolic extract significantly ($p<0.05$) scavenged 2,2-Diphenyl-1-picrylhydrazyl (DPPH) and nitric oxide (NO) radicals in a concentration dependent manner (Table 1). The ethyl acetate extract fraction at 62.5 μg mL^{-1} distilled water caused a more significant ($p<0.05$) increase in scavenging DPPH and NO radicals than the ethanolic extract and other extract fractions (Table 2). Ascorbic acid (standard) at 100 μg mL^{-1} distilled water exerted a higher significant ($p<0.05$) radical scavenging activity when compared with the ethanolic extract and other extract fractions (Table 1 and 2).

Flavonoids and phenolic contents of *Mimosa pudica* seeds extracts: *Mimosa pudica* seeds ethanolic extract and extract fractions have moderate amount of antioxidants, flavonoids and phenols (Table 3). The ethyl acetate extract fraction possess a significant ($p<0.05$) higher total antioxidant, flavonoids and phenolic contents when compared with the ethanolic extract and other extract fractions (Table 3).

Median lethal dose (LD_{50}) of *Mimosa pudica* seeds extracts: The ethanolic extract and each of the extract fractions (n-butanol, ethyl acetate and n-hexane extract fraction) of *Mimosa pudica* seeds caused no mortality in Wistar rats after acute oral administration of the samples. The median lethal dose (LD_{50}) of the ethanolic extract and each of the extract fractions was $\geq$5000 mg kg^{-1} body weight.

Table 1: Radical scavenging activity of *Mimosa pudica* seeds ethanolic extract

Ethanolic extract (μg mL^{-1})	2,2-Diphenyl-1-picrylhydrazyl (%)	Nitric oxide (%)
62.5	36.33±0.06*	34.18±0.49*
125	48.52±1.81*	56.00±1.65*
250	69.01±0.57*	73.04±0.66*
500	80.74±0.31*	81.27±0.43*
1000	92.88±0.35*	95.40±1.22
Ascorbic acid (100)	97.73±0.37	97.44±0.62
IC_{50} of ethanolic extract	107.41μg mL^{-1}	78.91μg mL^{-1}

Values are given as Mean±SEM, n = 3, *$p<0.05$ compared to ascorbic acid

Table 2: Radical scavenging activity of *Mimosa pudica* seeds ethanolic extract and fractions at 62.5 (μg mL^{-1})

Samples (μg mL^{-1})	2,2-Diphenyl-1-picrylhydrazyl (%)	Nitric oxide (%)
Ethanolic extract	36.33±0.06*#	34.18±0.49*#
n-butanol fraction	46.91±0.41*#	81.17±0.07*#
Ethyl acetate fraction	65.41±0.33*	90.16±0.11*
n-hexane fraction	34.84±0.44*#	33.78±0.38*#
Ascorbic acid	97.73±0.37	97.44±0.62

Values are given as Mean±SEM, n = 3, *$p<0.05$ compared to ascorbic acid, #$p<0.05$ compared to ethyl acetate fraction

Table 3: Total antioxidant, flavonoids and phenolics contents of *Mimosa pudica* seeds ethanolic extract and fractions

Samples (μg mL^{-1})	Total antioxidant capacity (mg ascorbic acid equivalent/g)	Total flavonoids (mg quercetin equivalent/g)	Total phenol (mg gallic acid equivalent/g)
Ethanolic extract	55.73±0.50*	49.99±0.14*	32.21±0.15*
n-butanol fraction	47.62±0.07*	44.20±0.62*	31.29±0.06*
Ethyl acetate fraction	61.96±0.27	61.11±0.20	46.37±0.17
n-hexane fraction	35.90±0.37*	35.03±0.53*	20.55±0.26*

Values are given as Mean±SEM, n = 3, *$p<0.05$ compared to ethyl acetate fraction

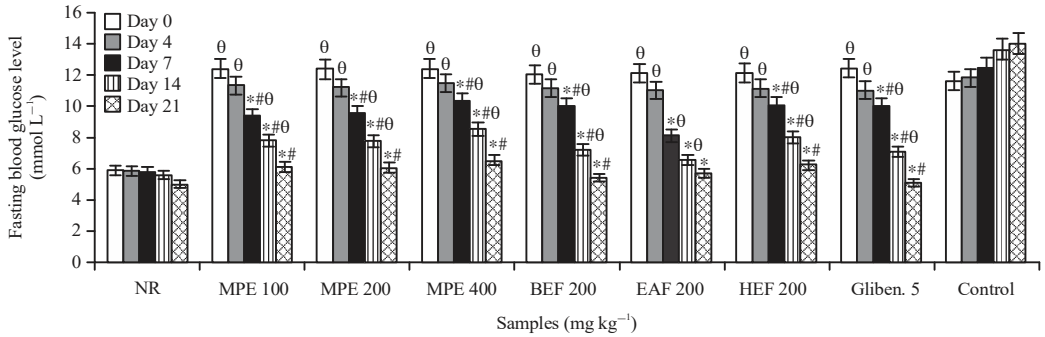

Fig. 1: Antidiabetic effect of *Mimosa pudica* seeds ethanolic extract and extract fractions on fasting blood glucose levels (FBGL) of streptozotocin-induced diabetic Wistar rats

Values are given as Mean±SEM, n = 5, NR: Normoglycemic rats, MPE: *Mimosa pudica* seeds ethanolic extract, BEF: n-butanol extract fraction, EAF: Ethyl acetae extract fraction, HEF: n-Hexane extract fraction, Gliben.: Glibenclamide,* p<0.05: Significant decrease in FBGL of treated streptozotocin-induced diabetic Wistar rats compared to the untreated diabetic rats (control), #p<0.05: Compared to ethyl acetate extract fraction, θp<0.05compared to day 21

Table 4: Effect of *Mimosa pudica* seeds ethanolic extract and extract fractions on serum insulin and serum α-amylase levels

Samples (mg kg^{-1})	Serum insulin (μIU mL^{-1})	Serum α-amylase (U L^{-1})
Normoglycemic rats	3.99±0.21*	52.90±0.25*#
100 Ethanolic extract	2.22±0.26*#	25.27±0.20*#
200 Ethanolic extract	2.30±0.24*#	29.01±0.22*#
400 Ethanolic extract	2.59±0.20*	36.00±0.29*#
200 n-Butanol fraction	2.61±0.19*	38.11±0.19*#
200 Ethyl acetate fraction	3.32±0.27*	45.01±0.23*
200 n-Hexane fraction	2.31±0.19*#	32.09±0.27*#
5 Glibenclamide	3.61±0.24*	56.07±0.18*#
Control (untreated diabetic rats)	1.00±0.16	19.21±0.31

Values are given as Mean±SEM, n = 5, *p<0.05 compared to untreated diabetic rats (control), #p<0.05 compared to ethyl acetate fraction

Effect of *Mimosa pudica* seeds extracts on fasting blood glucose levels of streptozotocin-induced diabetic Wistar rats: The ethanolic extract, extract fractions and glibenclamide caused a significant (p<0.05) reduction in fasting blood glucose level (FBGL) in streptozotocin-induced diabetic Wistar rats on day 7, 14 and 21 (Fig. 1) when compared to the control (untreated diabetic rats). The ethyl acetate extract fraction caused a more significant (p<0.05) decrease in FBGL from day 7 onwards when compared with the ethanolic extract and other extract fractions (Fig. 1). There was also a significant decrease in FBGL on day 0, 4, 7 and 14 when compared to day 21 (Fig. 1).

Effect of *Mimosa pudica* seeds extracts on serum insulin and α-amylase level of streptozotocin-induced diabetic Wistar rats: *Mimosa pudica* seeds ethanolic extract and extract fractions significantly (p<0.05) increased the level of serum insulin and α-amylase when compared to the control (Table 4). The ethyl acetate extract fraction caused a more significant increase in serum insulin and α-amylase level when compared to the ethanolic extract and other extract fractions (Table 4).

Antioxidant effect of *Mimosa pudica* seeds in streptozotocin-induced diabetic Wistar rats: The ethanolic extract and extract fractions of *Mimosa pudica* seeds significantly (p<0.05) decreased the level of thiobarbituric acid reactive substance (TBARS) and increased hepatic glutathione GSH) and catalase levels when compared to the control (Fig. 2). The ethyl acetate extract fraction caused a more significant decrease in the level of TBARS and increase in hepatic GSH and catalase levels when compared to the ethanolic extract and other extract fractions (Fig. 2).

DISCUSSION

The results of this study showed that the ethanolic extract and extract fractions of *Mimosa pudica* seeds possess moderate amount of antioxidants (flavonoids and phenols) and scavenged 2,2-Diphenyl-1-picrylhydrazyl and nitric oxide free radicals. The results also showed that *M. pudica* seeds increased endogenous antioxidants (catalase and reduced glutathione levels) and decreased lipid peroxidation in streptozotocin-induced diabetic Wistar rats. *M. pudica* seeds also reduced fasting blood glucose level and increased insulin

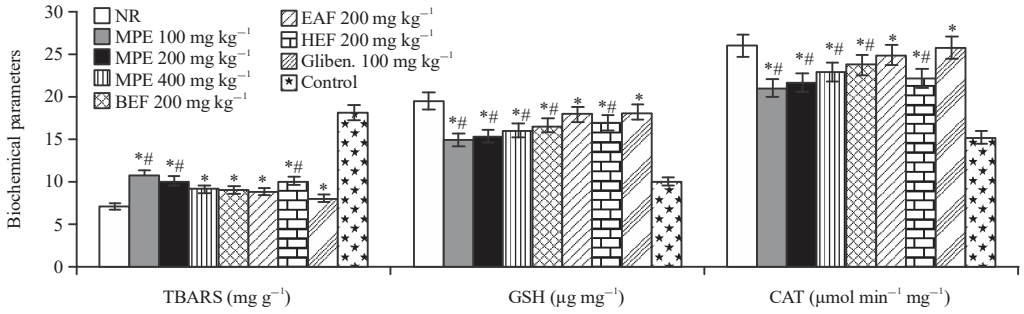

Fig. 2: Effect of *Mimosa pudica* seeds ethanolic extract and extract fractions on thiobarbituric acid reactive substances, glutathione and catalase of streptozotocin-induced diabetic Wistar rats

Values are given as Mean±SEM, n = 5, NR: Normoglycemic rats, MPE: *Mimosa pudica* seeds ethanolic extract, BEF: n-butanol extract fraction, EAF: Ethyl acetate extract fraction, HEF: n-Hexane extract fraction, Gliben.: Glibenclamide, *p<0.05: Significant decrease in FBGL of treated streptozotocin-induced diabetic Wistar rats compared to the untreated diabetic rats (control), #p<0.05: Compared to ethyl acetate extract fraction

and α-amylase levels in streptozotocin-induced diabetic Wistar rats. The ethyl acetate extract fraction exerted a more antioxidant and antidiabetic activity than other extract fractions.

Reduced glutathione and catalase are endogenous antioxidants found in liver that functions in the scavenging of free radicals and prevention of lipid peroxidation[35,36]. In this study, the increase in thiobarbituric acid reactive substances level in untreated streptozotocin-induced diabetic Wistar rats suggested enhanced lipid peroxidation[32]. *Mimosa pudica* seeds ethanolic extract and extract fractions reduced lipid peroxidation and increased the levels of catalase and reduced glutathione in streptozotocin-induced diabetic Wistar rats. Previous studies on free radical scavenging activities of plants due to the presence of antioxidants in different parts of plants reported that plants with antioxidants (mostly phenolics and flavonoids) have a correlation with therapeutic agent for the treatment of diabetes[24,37]. In this study, *Mimosa pudica* seeds possess moderate amount of antioxidants (flavonoids and phenols) and this might be correlated with its antidiabetic activity in streptozotocin-induced diabetic Wistar rats.

Under normal physiological conditions, the islet of Langerhans beta cells found in the pancreas produce the hormone, insulin which functions in the uptake of glucose by body cells[38]. The pancreas also secretes the enzyme α-amylase which catalyzes digestion of starchy carbohydrate to glucose[39]. Streptozotocin exerts its diabetogenic action in experimental animals by selectively destroying the insulin secreting beta cells of the pancreas thereby leading to a decrease in endogenous insulin resulting to insulin dependent diabetes (type 1 diabetes) or non-insulin dependent diabetes (type 2 diabetes) depending on the degree of damage to beta cells in Wistar rats[40-41]. In this study, the fasting blood glucose level of streptozocin-induced diabetic rats used range from 11-12 mmol L^{-1} which is similar to type 2 diabetes mellitus with partially functional pancreas[42]. The defect in the secretion of insulin by the pancreas in type 2 diabetes mellitus also affects the synthesis of α-amylase[43]. Madole *et al.*[44] also reported that low serum α-amylase was observed when there is high blood glucose level in type 2 diabetes mellitus and this signifies a defect in the cells of the pancreas. Previous studies carried out on leaves ethanolic extract and petroleum ether extract of *Mimosa pudica* reported that the extracts decreased blood glucose level in alloxan-induced diabetic Wistar rats[20]. Antidiabetic studies carried out on the whole plant ethanolic extract of *Mimosa pudica* showed that the plant extract exerted hypoglycemic effect in normal and streptozotocin-induced diabetic rats[21,22]. In this study, the seeds of *Mimosa pudica* reduced the fasting blood glucose level from the 4th day onwards in treated streptozotocin-induced diabetic Wistar rats. The serum insulin and α-amylase levels were also increased in treated streptozotocin-induced diabetic Wistar rats at the end of the study. The ethyl acetate extract fractions exerted more antidiabetic activity compared to other extract fractions.

Numerous studies have reported that the antidiabetic mechanism of conventional pharmacological drugs includes inhibition of α-glucosidase (acarbose), stimulation of the pancreas to produce more insulin (sulfonylureas) and increase in the sensitivity of the body cells to insulin (thiazolidinediones)[45]. The results of this study suggested that the antidiabetic mechanism of *Mimosa pudica* seeds is by stimulation of pancreatic cells to produce more insulin.

CONCLUSION

Mimosa pudica seeds possess moderate amount of flavonoids and phenolic compounds and scavenged

2,2-Diphenyl-1-picrylhydrazyl and nitric oxide free radicals. *Mimosa pudica* seeds also reduced lipid peroxidation and increased endogenous antioxidant (catalase and reduced glutathione) levels in streptozotocin-induced diabetic Wistar rats. The results of this study also showed that *Mimosa pudica* seeds exerted antidiabetic effect by enhancing secretion of pancreatic insulin.

SIGNIFICANCE STATEMENT

This study discovered that *Mimosa pudica* seeds exerted antidiabetic effect by enhancing secretion of pancreatic insulin and α-amylase and also due to the presence of flavonoids and phenols. *M. pudica* seeds also reduced lipid peroxidation and increased the level of endogenous antioxidants (reduced glutathione and catalase). This study will help researchers to uncover the critical areas in the use of the plants for the treatment of free radical induced diseases including diabetes that many researchers were not able to explore. Thus a new theory in the treatment of diabetes will be uncovered.

ACKNOWLEDGMENTS

Rita M Sunday acknowledges the Department of Biotechnology, Ministry of Science and Technology, Government of India, New Delhi (DBT) and The World Academy of Sciences (TWAS), Italy for funding this research work. Authors acknowledge the AMSAR Private Limited, Indore, India for providing the pulverized seeds of *Mimosa pudica*. Facilities of DBT available in the Department under M.Sc. Biotechnology program and also under Distributed Bioinformatics Sub-Centre are also gratefully acknowledged. Authors also acknowledge the help rendered by Dr. H.S. Parmar in getting *Asparagus* sample from AMSAR Private Limited, Indore.

REFERENCES

1. Asmat, U., K. Abad and K. Ismail, 2016. Diabetes mellitus and oxidative stress: A concise review. Saudi Pharm. J., 24: 547-553.
2. American Diabetes Association, 2011. Diagnosis and classification of diabetes mellitus. Diabetes Care, 34: S62-S69.
3. Pham-Huy, L.A., H. He and C. Pham-Huy, 2008. Free radicals, antioxidants in disease and health. Int. J. Biomed. Sci., 4: 89-96.
4. Sen, S., R. Chakraborty, C. Sridhar, Y.S.R. Reddy and B. De, 2010. Free radicals, antioxidants, diseases and phytomedicines: Current status and future prospect. Int. J. Pharmaceut. Sci. Rev. Res., 3: 91-100.
5. Phaniendra, A., D.B. Jestadi and L. Periyasamy, 2015. Free radicals: Properties, sources, targets and their implication in various diseases. Indian J. Clin. Biochem., 30: 11-26.
6. Singh, U. and I. Jialal, 2006. Oxidative stress and atherosclerosis. Pathophysiology, 13: 129-142.
7. Smith, M.A., C.A. Rottkamp, A. Nunomura, A.K. Raina and G. Perry, 2000. Oxidative stress in Alzheimer's disease. Biochimica Biophysica Acta (BBA)-Mol. Basis Dis., 1502: 139-144.
8. Bolton, J.L., M.A. Trush, T.M. Penning, G. Dryhurst and T.J. Monks, 2000. The role of quinones in toxicology. Chem. Res. Toxicol., 13: 135-160.
9. Jeeva, J.S., J. Sunitha, R. Ananthalakshmi, S. Rajkumari, M. Ramesh and R. Krishnan, 2015. Enzymatic antioxidants and its role in oral diseases. J. Pharm. BioAllied Sci., 7: 331-333.
10. Rahal, A., A. Kumar, V. Singh, B. Yadav, R. Tiwari, S. Chakraborty and K. Dhama, 2014. Oxidative stress, prooxidants and antioxidants: The interplay. BioMed Res. Int., Vol. 2014. 10.1155/2014/761264.
11. Ayoola, G.A., H.A. Coker, S.A. Adesegun, A.A. Adepoju-Bello, K. Obaweya, E.C. Ezennia and T.O. Atangbayila, 2008. Phytochemical screening and antioxidant activities of some selected medicinal plants used for malaria therapy in Southwestern Nigeria. Trop. J. Pharm. Res., 7: 1019-1024.
12. Jackie, T., N. Haleagrahara and S. Chakravarthi, 2011. Antioxidant effects of *Etlingera elatior* flower extract against lead acetate induced perturbations in free radical scavenging enzymes and lipid peroxidation in rats. BMC Res. Notes, Vol., 4. 10.1186/1756-0500-4-67.
13. Balde, N., A. Youla, M.D. Balde, A. Kake, M.M. Diallo, M.A. Balde and D. Maugendre, 2006. Herbal medicine and treatment of diabetes in Africa: An example from Guinea. Diabetes Metab., 32: 171-175.
14. Ahmad, H., S. Sehgal, A. Mishra and R. Gupta, 2012. *Mimosa pudica* L. (Laajvanti): An overview. Pharmacogn. Rev., 6: 115-124.
15. Prajapati, N.D., S.S. Purohit, A.K. Sharma and T.A. Kumar, 2003. Handbook of Medicinal Plants: A Complete Source Book. Agrobios Publication, ISBN-13: 9788177541342, India, Pages: 134.
16. Ganguly, M., N. Devi, R. Mahanta and M.K. Borthakur, 2007. Effect of *Mimosa pudica* root extract on vaginal estrous and serum hormones for screening of antifertility activity in albino mice. Contraception, 76: 482-485.
17. Kokane, D.D., R.Y. More, M.B. Kale, M.N. Nehete, P.C. Mehendale and C.H. Gadgoli, 2009. Evaluation of wound healing activity of root of *Mimosa pudica*. J. Ethnopharmacol., 124: 311-315.
18. Molina, M., C.M. Contreras and P. Tellez-Alcantara, 1999. *Mimosa pudica* may possess antidepressant actions in the rat. Phytomedicine, 6: 319-323.
19. Bum, E.N., D.L. Dawack, M. Schmutz, A. Rakotonirina and S.V. Rakotonirina *et al*., 2004. Anticonvulsant activity of *Mimosa pudica* decoction. Fitoterapia, 75: 309-314.

20. Sutar, N.G., U.N. Sutar and B.C. Behera, 2009. Antidiabetic activity of the leaves of *Mimosa pudica* Linn in albino rats. J. Herbal Med. Toxicol., 3: 123-126.
21. Konsue, A., C. Picheansoonthon and C. Talubmook, 2017. Fasting blood glucose levels and hematological values in normal and streptozotocin-induced diabetic rats of *Mimosa pudica* L. extracts. Pharmacogn. J., 9: 315-322.
22. Yupparach, P. and A. Konsue, 2017. Hypoglycemic and hypolipidemic activities of ethanolic extract from *Mimosa pudica* L. in normal and streptozotocin-induced diabetic rats. Pharmacogn. J., 9: 834-837.
23. Sas, K., H. Robotka, J. Toldi and L. Vecsei, 2007. Mitochondria, metabolic disturbances, oxidative stress and the kynurenine system, with focus on neurodegenerative disorders. J. Neurol. Sci., 257: 221-239.
24. Boora, F., E. Chirisa and S. Mukanganyama, 2014. Evaluation of nitrite radical scavenging properties of selected Zimbabwean plant extracts and their phytoconstituents. J. Food Process., Vol. 2014. 10.1155/2014/918018.
25. Hyun, D.H., J.O. Hernandez, M.P. Mattson and R. de Cabo, 2006. The plasma membrane redox system in aging. Ageing Res. Rev., 5: 209-220.
26. Kinnula, V.L. and J.D. Crapo, 2004. Superoxide dismutases in malignant cells and human tumors. Free Radical Biol. Med., 36: 718-744.
27. Ruch, R.J., S.J. Cheng and J.E. Klaunig, 1989. Prevention of cytotoxicity and inhibition of intercellular communication by antioxidant catechins isolated from Chinese green tea. Carcinogenesis, 10: 1003-1008.
28. Lorke, D., 1983. A new approach to practical acute toxicity testing. Arch. Toxicol., 54: 275-287.
29. Sharma, S.B., S. Gupta, R. Ac, U.R. Singh, R. Rajpoot and S.K. Shukla, 2010. Antidiabetogenic action of *Morus rubra* L. leaf extract in streptozotocin-induced diabetic rats. J. Pharm. Pharmacol., 62: 247-255.
30. Temple, R., P.M.S. Clark and C.N. Hales, 1992. Measurement of insulin secretion in type 2 diabetes: Problems and pitfalls. Diabetic Med., 9: 503-512.
31. Burtis, C.A. and E.R. Ashwood, 1994. Tietz Textbook of Clinical Chemistry. 2nd Edn., W.B. Saunders Company, Philadelphia, USA., ISBN-13: 9780721644721, pp: 852-863.
32. Ohkawa, H., N. Ohishi and K. Yagi, 1979. Assay for lipid peroxides in animal tissues by thiobarbituric acid reaction. Anal. Biochem., 95: 351-358.
33. Ellman, G.L., 1959. Tissue sulfhydryl groups. Arch. Biochem. Biophys., 82: 70-77.
34. Aebi, H., 1974. Catalase. In: Methods of Enzymatic Analysis, Bergmeyer, H.U. (Ed.). Vol. 2, Academic Press Inc., New York, USA., ISBN: 352725370X, pp: 673-684.
35. Ghalia, A.H.A. and I.M. Fouad, 2000. Glutathione and its metabolizing enzymes in patients with different benign and malignant diseases. Clin. Biochem., 33: 657-662.
36. Collins, A.R. and E. Horvathova, 2001. Oxidative DNA damage, antioxidants and DNA repair: Applications of the comet assay. Biochem. Soc. Trans., 29: 337-340.
37. Sales, P.M., P.M. Souza, L.A. Simeoni, P.O. Magalhaes and D. Silveira, 2012. α-amylase inhibitors: A review of raw material and isolated compounds from plant source. J. Pharmacy Pharm. Sci., 15: 141-183.
38. Masicampo, E.J. and R.F. Baumeister, 2008. Toward a physiology of dual-process reasoning and judgment: Lemonade, willpower and expensive rule-based analysis. Psychol. Sci., 19: 255-260.
39. Gupta, R., P. Gigras, H. Mohapatra, V.K. Goswami and B. Chauhan, 2003. Microbial α-amylases: A biotechnological perspective. Process Biochem., 38: 1599-1616.
40. Lenzen, S. and R. Munday, 1991. Thiol-group reactivity, hydrophilicity and stability of alloxan, its reduction products and its n-methyl derivatives and a comparison with ninhydrin. Biochem. Pharmacol., 42: 1385-1391.
41. Eddouks, M., D. Chattopadhyay and N.A. Zeggwagh, 2012. Animal models as tools to investigate antidiabetic and anti-inflammatory plants. Evidence-Based Complement. Altern. Med., Vol. 12. 10.1155/2012/142087.
42. Gupta, K.R., N.A. Kesari, G. Watal, P.S. Murthy, R. Chandra, K. Maithal and V. Tandon, 2005. Hypoglycaemic and antidiabetic effect of aqueous extract of leaves of *Annona squamosa* (L.) in experimental animal. Curr. Sci., 88: 1244-1254.
43. American Diabetes Association, 2010. Diagnosis and classification of diabetes mellitus. Diabetes Care, 33: S62-S69.
44. Madole, M.B., C.M. Iyer, M.T. Madivalar, S.K. Wadde and D.S. Howale, 2016. Evaluation of biochemical markers serum amylase and serum lipase for the assessment of pancreatic exocrine function in diabetes mellitus. J. Clin. Diagn. Res., 10: BC01-BC04.
45. Meneses, M.J., B.M. Silva, M. Sousa, R. Sá, P.F. Oliveira and M.G. Alves, 2015. Antidiabetic drugs: Mechanisms of action and potential outcomes on cellular metabolism. Curr. Pharmaceut. Des., 21: 3606-3620.