

**RESEARCH ARTICLE****Antidiabetic potential of aqueous and alcoholic leaf extracts of *Pithecellobium dulce***M Sugumaran<sup>1\*</sup>, T Vetrichelvan<sup>1</sup> and S Darlin Quine<sup>2</sup><sup>1</sup>Adhiparasakthi College of Pharmacy, Melmaruvathur-603319.<sup>2</sup>School of Chemical and Biotechnology, SASTRA University, Thanjavur-613402.\* Corresponding Author E-mail: [murugesansugumaran@yahoo.com](mailto:murugesansugumaran@yahoo.com)**ABSTRACT**

Ethanollic and aqueous leaf extract of *Pithecellobium dulce* was studied for its antidiabetic activity using streptozotocin-induced diabetic model in rats. The aqueous extract showed significant activity ( $P < 0.01$ ) than the alcoholic extract at the tested dose level which was comparable to glibenclamide, a standard antidiabetic drug. HPTLC fingerprinting profile of the aqueous extract was also developed which would serve as reference standard for quality control of this extract.

**KEY WORDS**Alloxan , Anti-diabetic, Leaf extracts, *Pithecellobium dulce***INTRODUCTION:**

*Pithecellobium dulce* Benth. (Leguminosae) is a small to medium sized, evergreen, spiny tree up to 18 m height, native of tropical America and cultivated throughout the plains of India and in the Andamans<sup>1</sup>. It is known as 'Vilayati Babul' in Hindi and 'Kodukkapuli' in Tamil. The bark of the plant is reported to be used as astringent in dysentery, febrifuge and it is also useful in dermatitis and eye inflammation. The leaves have been reported to possess astringent, emollient, abortifacient and antidiabetic properties. The presence of steroids, saponins, lipids, phospholipids, glycosides, glycolipids and polysaccharides have been reported in the seeds<sup>2-5</sup>. The bark contains 37% of tannins of catechol type. Quericitin, kaempferol, dulcitol and afezililn have been reported from the leaves<sup>6&7</sup>. Roots have been reported to possess estrogenic activity<sup>8</sup>. Studies on alkylated resins from seed oil have been reported recently<sup>9</sup>.

According to ethanobotanical data, *P. dulce* possesses an antidiabetic effect, which has never been experimentally demonstrated<sup>10</sup>. Therefore, we considered it worth while to undertake a study to evaluate the antidiabetic effect of the aqueous and alcoholic leaf extracts in rats using streptozotocin - induced diabetic model and compared with glibenclamide, a standard antidiabetic drug.

**MATERIALS AND METHODS:****Collection of Medicinal plant:**

Fresh leaves of *Pithecellobium dulce* were collected from Sembulam Village at Kancheepuram District, T.N. in the month of January 2005. The plant was identified by local people of that village and authenticated by Dr. P. Jayaraman, Director, Plant Anatomy Research Centre (PARC), Chennai. A herbarium specimen of the plant (APCP-3/205) was preserved in the Department of Pharmacognosy of our institute for further reference.

**Procurement of Animals:**

Adult wistar albino rats of either sex maintained under standard condition (temperature:  $23 \pm 2^\circ\text{C}$ , relative humidity:  $55 \pm 10\%$  and 12 h light and dark place) were used for pharmacological study. The animals were allowed standard laboratory feed and water *ad libitum* (Amrut lab animals feed, sanli-416436). Ethical clearance for performing the experiments on animals was obtained (Reg. No. - 409/ 2001/ CPCSEA) from the Institutional Animal Ethics Committee. Streptozotocin was obtained from SD Fine Chemicals Pvt Ltd., Mumbai. All other chemicals and solvents used for this study were of analytical grade.

**Preparation of aqueous and alcoholic extract:**

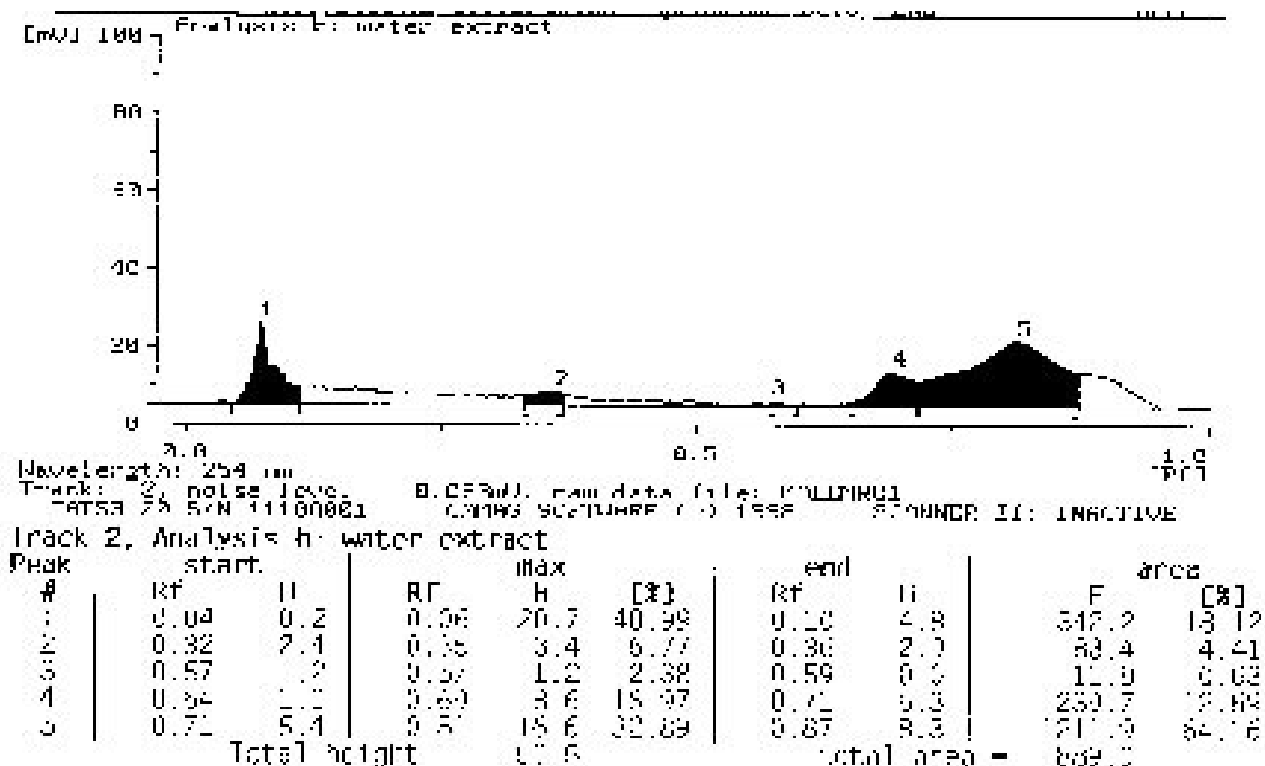
The fresh leaves of *P. dulce* were washed with water, air-dried at room temperature and then reduced to coarse powder. The powdered mass of leaf was successively extracted with petroleum ether (60-80°), benzene, chloroform, acetone, alcohol (95% v/v) and distilled water by using soxhlet extraction apparatus for about 18 h<sup>11</sup>. The extracts were filtered and the filtrates were concentrated under reduced pressure to obtain the extracts as solid residues. Extractive value (%w/w) of alcohol and aqueous extracts were 17.93 and 18.58 respectively. The

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**Table 1. Antidiabetic activity of *P. dulce* on streptozotocin – induced diabetic model in rats**

Group	Blood glucose levels in mg/dl					
	0 h	1 h	2 h	5 h	8 h	7 <sup>th</sup> day
Diabetic Control	191.16±1.70	194.83±0.30	200.16±0.40	205.83±0.40	210.00±0.85	224.83±0.47
Glibenclamide (10 mg/kg)	192.50±1.11	176.00±0.89*	156.66±0.76**	121.83±0.74**	110.16±0.60**	99.16±1.16**
Aqueous extract (250 mg/kg)	192.66±0.71	181.50±0.42*	163.00±0.73**	130.33±0.76**	119.00±0.36**	107.66±0.71**
Ethanol extract (250 mg/kg)	196.00±1.00	187.66±0.55	174.66±0.80	166.83±0.65	153.83±0.60	149.33±0.33

n=6; values expressed as mean±SEM \*P<0.05 & \*\*P<0.01 vs control (ANOVA)

**Fig 1: HPTLC finger printing of water extract of *P. dulce***

freshly prepared extracts were chemically tested for the presence of different constituents using standard methods<sup>12</sup>.

### Screening of anti-diabetic activity<sup>13</sup>

Streptozotocin 65 mg/kg body weight prepared in ice cold citrate buffer of (0.01 M, pH 4.5) was injected intraperitoneally (i.p) to overnight fasted wistar albino rats to induce diabetes. After 72 h of injection, blood glucose level was measured by using glucose oxidase method<sup>14</sup>. Rats having blood glucose level above 175 mg/dl were selected and grouped into four groups consisting of 6 animals each. The rats in the first group (diabetic control) were administered CMC orally. Second group was treated orally with 10 mg/kg of glibenclamide, while CMC suspension of alcoholic and aqueous extract of *P.dulce* (250 mg/kg body

weight, p.o) was given to third and fourth group respectively as a single dose for seven days. Blood samples (0.5 ml) were collected from rat tail vein under mild anaesthesia condition just prior to and 1<sup>st</sup>, 2<sup>nd</sup>, 5<sup>th</sup>, 8<sup>th</sup> h and 7<sup>th</sup> day after drug administration for acute and prolong study respectively.

### STATISTICAL ANALYSIS :

The results were expressed as Mean ± SEM. The data was analysed by one-way ANOVA followed by Dunnet's test to find out the level of significance<sup>15</sup>.

**RESULTS AND DISCUSSION:**

The fresh alcoholic and aqueous extract of *P. dulce* gave positive phytochemical tests for glycosides, saponins, and flavonoids. These compounds have been earlier elucidated for their structures in this plant. The results of present study indicates that the streptozotocin can act as an effective diabetogenic agent<sup>16</sup>. It is cytotoxic to beta-cells of islets of langerhans and is capable of inducing chemical diabetes in a wide variety of animal species through damage of the insulin secreting cells<sup>17</sup>. Both the extracts showed activity (Table1) at the tested dose level by decreasing glucose level when compared to control group animals in acute and prolong treatment. The aqueous extract significantly decreased the glucose level at 2<sup>nd</sup>, 5<sup>th</sup> and 8<sup>th</sup> h in hyperglycemic animals (P<0.01) than alcohol extract. Glibenclamide reduces the elevated blood glucose level from 192.5 mg% to 110 mg%. The treatment with aqueous and alcoholic extract reduced the blood glucose level to 119 and 153.8 mg % respectively from the basal value. The Flavonoid compounds present in this extract (indicated by preliminary phytochemical studies) may be involved for this action as flavonoids have been reported to possess significant anti-diabetic activity in various experimental models<sup>18</sup>. HPTLC fingerprinting profile of the aqueous extract was also developed (Figure 1) which would serve as reference standard for quality control of this extract.

From the study it can be concluded that *P.dulce* leaves freely available throughout India, the people in rural area, where they are more aware of decoction procedures of plants than the conventional anti-diabetic drugs, can very well use it. These extracts can be further used to formulate poly herbal preparations using other plant extracts with anti-diabetic activity. The common adverse effects and drug interactions by using conventional antidiabetic drugs on prolonged use can be minimized by using the leaf extracts of *P.dulce* benth. As it reduces the blood glucose level significantly in oral route, it may be used as an alternative medicine to insulin in diabetic treatment. A commonly herbal drug produces their antidiabetic effect by protecting the cells in pancreas from destruction, by restricting glucose load as well as by promoting unrestricted endogenous insulin action. However further studies are required to establish its exact mode of action and the active principles involved in this effect.

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