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Anti Diabetic Activity of Hydroethanolic Extracts of *Mirabilis jalapa* Leaves in Streptozotocin Induced Diabetic Rats



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ABSTRACT

Mirabilis jalapa Linn (Nyctaginaceae) is a perennial herb which is widely used in Himalayan region as a traditional treatment for diabetes mellitus. The present study was designed to evaluate the Anti-hyperglycemic effect of hydroethanolic leaf extract of *Mirabilis jalapa* in streptozotocin induced diabetic rats. The hydroethanolic leaf extract at the concentration of 200 and 400 mg/kg of body weight showed significant decrease in the levels of glucose, Urea, Creatinine, serum marker enzymes such as Aspartate transaminase (AST), Alanine transaminase (ALT) and Alkaline Phosphatase (ALP) in streptozotocin induced diabetic rats. Thus, the study clearly shows that the hydroethanolic leaf extract of *Mirabilis jalapa* possess potent anti-diabetic activity.



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INTRODUCTION

Diabetes mellitus is characterized by an increased concentration of blood glucose due to disarrangement in carbohydrate metabolism and defective secretion of insulin. These metabolic disturbances result in acute and long term complications^[1]. It is characterized by chronic hyperglycemia with altered metabolism to damaged beta cells of pancreas^[2]. Oxidative stress plays an important role in Diabetes mellitus right from its genesis to the development of microvascular complications and the generation of free radicals by hyperglycemia is related to glucose autooxidation^[3]. As the treatment of diabetes mellitus using oral hypoglycaemic drugs produces undesirable side effects, people moved on to plant based therapies. So the medicinal plants are used to treat many ailments^[4].

Administration of several medicinal plants have restored the activities of certain serum enzymes when compared to normal control groups^[5]. The active principles of many plant species are isolated for direct use as drugs, lead compounds or pharmacological agents^[6]. Traditional plant medicines or herbal formulations might offer a natural key to unlock diabetic complications^[7].

Mirabilis jalapa is a perennial herb, belonging to the family Nyctaginaceae, used widely in Himalayan region as a traditional medicine. In herbal medicine, root of the plant are used as a diuretic, purgative and for vulnerary wound healing purposes. Leaves of whole plant are used as anti-inflammatory, antiviral, anti-bacterial, anti-fungal, antispasmodic and anti-noceptive agents^[8]. *Mirabilis jalapa* is characterised as a quick growing much-branched herb with erect, angular, distinctly joined stem swollen at the nodes^[9].

The current study was designed to evaluate the anti-hyperglycemic activity of *Mirabilis jalapa* on streptozotocin induced diabetic rats.

MATERIALS AND METHODS

a) Plant Collection and extraction

Mirabilis jalapa was collected from local areas of Coimbatore, Tamilnadu. The plant was identified and certified (BSI/SRC/5/23/2014-15/Tech/1461) by the Taxonomist, Botanical Survey of India (BSI), Southern Regional Centre, Coimbatore, Tamilnadu, India. The leaves

were dried and ground to a coarse powder. The coarse powder was extracted using hydroethanol. The extracts were condensed to dryness using rotatory evaporator and crystals were obtained. These crystals were weighed, dissolved in distilled water and administrated orally to the experimental animal for the treatment of diabetes.

b) Procurement of animals

Male Spraque-Dawley rats of 120-150 g weight procured from scientific suppliers were used for the study. The ethical clearance for handling experimental animals was obtained from Institutional Animal Ethics Committee (IAEC) (260/2014/IAEC). The animals were maintained under standard and laboratory conditions with controlled humidity and temperature where they are allowed to get acclimatized to standard laboratory diet and filtered water. The animal house was well ventilated and maintained in large spacious hygienic cages during the course of the experimental period.

c) Induction of diabetes

Severe diabetes was induced in overnight fasting rats by a single intraperitoneal injection of streptozotocin (52 mg/kg of body weight). After 3 days, glucose levels were measured and the animals showing high blood glucose levels (> 250 mg/dl) were used for the experiments.

d) Experimental design

The experimental rats were divided into 5 groups of 6 animals in each group. The animals were fasted overnight before the experimental schedule began but allowed to free access of water.

Groups	Rats
Group I	Normal control rats
Group II	Streptozotocin induced diabetic rats (60 mg/kg body weight)
Group III	Diabetic rats treated with Glibenclamide drug (120 mg/kg body weight)
Group IV	Diabetic rats treated with 50% ethanolic extract of <i>Mirabilis jalapa</i> leaves (200 mg/kg body weight)
Group V	Diabetic rats treated with 50% ethanolic extract of <i>Mirabilis jalapa</i> leaves (400 mg/kg body weight)

After the end of the experimental procedure, the animals were fasted overnight and sacrificed by cervical dislocation under mild chloroform anaesthesia. Blood was collected by cardiac puncture and the serum was separated by centrifugation at 2500 rpm for 15 minutes.

Biochemical estimation

Biochemical analysis in serum such as blood glucose was estimated according to the method of Trinder 1969^[10]. Urea was measured according to the method of Wybenga 1971^[11]. Creatinine was measured according to the method of Slot and Scand, 1965^[12]. Alanine Transaminase and Aspartate Transaminase was estimated according to the method of Reitman and Frankel 1957^[13]. The activity of serum alkaline phosphatase was measured according to the method of King and Armstrong 1934^[14].

Statistical Analysis

Data obtained was expressed as mean \pm SD. Statistical analysis was performed by using the method of distribution statistics (Standard descriptive analysis) and analysis of means (Student 't' test) using R – Statistical Computing and Graphical Tools (formerly AT & T, Lucent technology). A probability of $p < 0.05$ was considered significant.

RESULTS AND DISCUSSION

Serum glucose

Administration of streptozotocin led to significant elevation in the levels of serum glucose in diabetic rats when compared to the normal rats. After the treatment with 50% ethanolic extract of *Mirabilis jalapa* (200 and 400 mg/kg of b.w), there was a significant ($p < 0.05$) decrease in the level of serum glucose in diabetic treated rats. Decrease in blood glucose might be due to the regeneration of beta cells of pancreas and potentiation of insulin secretion^[15].

TABLE 1: Estimation of Serum glucose, Urea and Creatinine

Groups	Serum Glucose	Urea	Creatinine
I	88.14 ± 2.51	26.63 ± 0.49	0.76 ± 0.05
II	284.77 ± 15.62 ^{a*}	37.26 ± 0.39 ^{a*}	3.28 ± 0.16 ^{a*}
III	116.25 ± 11.60 ^{b*}	30.94 ± 0.68 ^{b*}	1.65 ± 0.31 ^{b*}
IV	178.16 ± 5.55 ^{c*}	28.22 ± 1.02 ^{c*}	2.44 ± 0.36 ^{c*}
V	148.31 ± 8.77 ^{d*}	26.97 ± 0.78 ^{d*}	1.60 ± 0.34 ^{d*}

Urea and Creatinine

Many studies have shown significant increase in the rate of kidney damage in diabetes. It is evident from Table 1, that there was a significant increase in Urea and Creatinine level in Group II rats and after the treatment with the plant extract, the level was found to be decreased as that of the normal group.

Serum marker enzymes

It was observed (Table 2) that there was a significant increase in the serum enzyme levels in diabetic control group when compared to normal group. On oral administration of 50% ethanolic extract of *Mirabilis jalapa* (200 (Group IV) and 400 mg/kg (Group V)), there was a significant decrease in ALT, AST and ALP when compared to Group II.

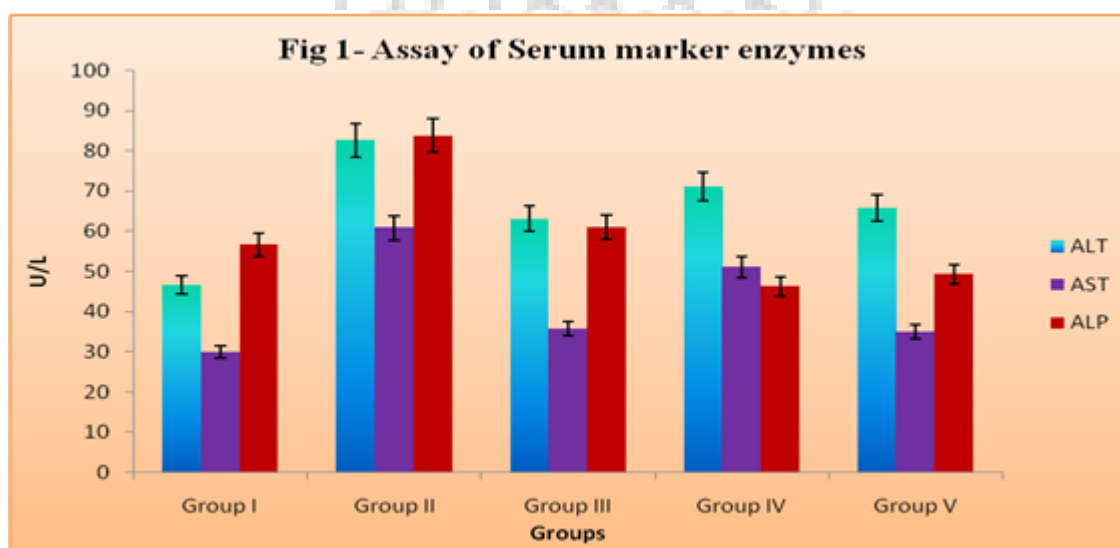


TABLE 2: Assay of serum marker enzymes

Groups	ALT (U/L)	AST (U/L)	ALP(U/L)
I	46.59 ± 0.27	29.88 ± 0.67	56.63 ± 0.32
II	82.57 ± 0.29 ^{a*}	60.85 ± 0.78 ^{a*}	83.73 ± 0.22 ^{a*}
III	63.07 ± 0.14 ^{b*}	35.82 ± 0.86 ^{b*}	60.99 ± 0.02 ^{b*}
IV	71.08 ± 0.79 ^{c*}	51.02 ± 0.81 ^{c*}	46.26 ± 0.35 ^{c*}
V	65.83 ± 0.71 ^{d*}	35.09 ± 0.58 ^{d*}	49.26 ± 0.55 ^{d*}

The increased levels of ALP, AST and ALT in the circulation indicates hepatic damage. In the present study, treatment with *Mirabilis jalapa* extracts had effectively reduced serum AST, ALT and ALP in diabetic rats suggesting that the extracts of experimental plant prevent hepatic injury associated with diabetes. This may be due to the fact that *Mirabilis jalapa* is rich in Flavonoids, Phenols, Phytosterols, Carotenoids and Glycosides^[16]. Stigmasterol is a Phytosterol compound reported to have antidiabetic activity^[17].

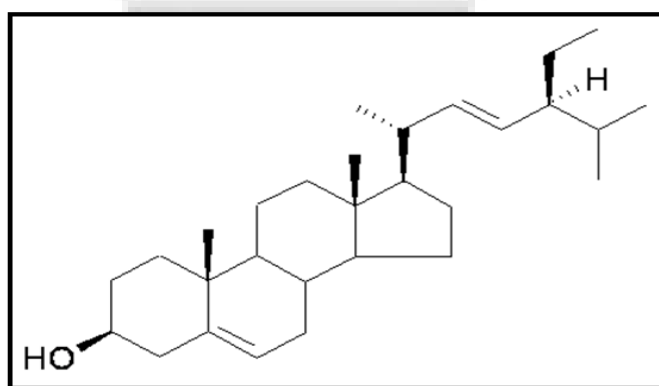


Fig. 2. Structure of Stigmasterol

Mirabilis jalapa contains Stigmasterol a Phytosterol^[18], which had been reported to have cholesterol lowering^[19] and antidiabetic activities^[20]. Thus, this study predicts that Stigmasterol of *Mirabilis jalapa* could be the major phytoconstituent that is responsible for the hypoglycemic effect. Hence, this could lead to future prospects of isolating this compound for evaluating it as an effective hypoglycemic agent.

CONCLUSION

The present work implies that the hydroethanolic extract of *Mirabilis jalapa* leaves showed a significant decrease in the Glucose, Urea, Creatinine, Serum Markers Enzymes which may be due to the phytochemicals present in the plant. Further studies may be taken up for the isolation and characterization of Stigmasterol and also other bioactive compounds which may exert potent antioxidant and anti-diabetic effects.

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