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Protective effect of *Mollugo cerviana* extract on liver markers in alloxan induced diabetic rats

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ABSTRACT

Diabetes mellitus is a metabolic disorder of multiple aetiology characterized by chronic hyperglycaemia with disturbances of carbohydrate, fat and protein metabolism resulting from defects in insulin secretion, insulin action, or both. Liver is the central metabolic organ in body responsible for glucose homeostasis, diabetes leads to hepatic dysfunction. In the present study to investigate the protective effect of *Mollugo cerviana* extract on liver markers in alloxan induced diabetic rats. Hepatospecific enzymes (ALP, ALT and AST), protein and albumin content were activated when hepatocellular damage gave rise to abnormalities of liver function and these enzymes are remarkably increased in diabetic rats. Further confirmed in the histopathological studies of liver. *Mollugo cerviana* helps in parenchymal cell regeneration in liver, thus protecting membrane integrity and thereby minimizing enzyme leakage. Histopathological studies also supported the biochemical parameters. This result suggested that *Mollugo cerviana* possess potential hepato regenerating activity.

Keywords: Alloxan, Diabetes mellitus, Hepatospecific enzymes, Mollugo cerviana.

INTRODUCTION

Diabetes mellitus is a metabolic disorder of multiple aetiology characterized by chronic hyperglycaemia with disturbances of carbohydrate, fat and protein metabolism resulting from defects in insulin secretion, insulin action, or both. Diabetes mellitus is classified into insulin dependent diabetes mellitus (IDDM) or Type 1, and Non-Insulin Dependent Diabetes Mellitus (NIDDM) or Type 2. The aetiological type named Type 1 encompasses cases due to pancreatic islet beta–cell destruction and are prone to ketoacidosis. Type 1 includes those cases attributable to an autoimmune process, as well as those with beta-cell destruction and who are prone to ketoacidosis for which neither an aetiology nor a pathogenesis is known (idiopathic). The type named Type 2 includes the common major form of diabetes which results from defect(s) in insulin secretion with a major contribution from insulin resistance [1, 2].

The liver plays an important role in maintaining normal glucose concentrations during both, fast and postprandial phase. It is also a major site for insulin clearance. The loss of a direct effect of insulin to suppress hepatic glucose production and glycogenolysis in the liver causes an increase in hepatic glucose production.7 Hepatic dysfunction resulting from the insulin resistance syndrome may contribute to the development of T2DM. Few studies in Mexico have examined the relationship between liver enzymes and diabetes and have suggested that elevated liver enzymes such as aspartate aminotransferase (AST), alanine aminotransferase (ALT) and alkaline phosphatase (ALP) are associated with the development of diabetes and cardiovascular disease [3, 4, 5]. In the present study to investicate the protective effect of *Mollugo cerviana* extract on liver markers in alloxan induced diabetic rats.

MATERIALS AND METHODS

Animals

Male albino rats of Wistar strain approximately weighing 180-220g were used in this study. They were healthy animals procured from Sri Venkateswara enterprises, Bangalore, India. The animals were housed in spacious polypropylene cages bedded with rice husk. The animal room was well ventilated and maintained under standard experimental conditions (Temperature 27±2ºC and 12 hours light / dark cycle) throughout the experimental period. All the animals were fed with standard pellet diet (Gold Mohur, Mumbai, India) and water *ad libitum*. They were acclimatization to the environment for 1 week prior to experimental use. The experiment was carried out according to the guidelines of the Committee (Ethical No: MC/1416/a/11/CPCSEA) for the Purpose of Control and Supervision of Experiments on

Animals (CPCSEA), New Delhi, India.

Plant material

The whole plant of *Mollugo cerviana* was collected in January 2015 from Tamil University, Thanjavur District, Tamil Nadu, India from a single herb. The whole plant was identified and authenticated by Dr. S. John Britto, The Director, the Rabiant Herbarium and centre for molecular systematics, St. Joseph's college Trichy-Tamil Nadu. India. A Voucher specimen has been deposited at the Rabinat Herbarium, St. Joseph's College, Thiruchirappalli, Tamil nadu, India.

Preparation of plant extract

The whole plant of *Mollugo cerviana* was first washed well and dust was removed from the plant. Whole plant was washed several times with distilled water to remove the traces of impurities from the plant. The whole plant was dried at room temperature and coarsely powdered. The powder was extracted with 70% methanol for 24 hours. A semi solid extract was obtained after complete elimination of alcohol under reduced pressure. The *Mollugo cerviana* extract (MCE) was stored in refrigerator until used.

Experimental design

Body weights of the animals were recorded and they were divided into 4 groups of 6 animals each as follows

Group 1: Control rats (untreated) were fed with standard diet and water *adlibitum.*
Group 2: Diabetic rats.
Group 3: Diabetic rats treated with *Mollugo cerviana* by oral gavage daily at a dose of 500 mg/kg body weight for a period of 30 days.
Group 4: Diabetic treated with glibenclamide (0.5mg/kg body weight) for a period of 30 days

Collection of samples

On completion of the experimental period, animals were anaesthetized with thiopentone sodium (50mg/kg). The blood was collected with or without EDTA as anticoagulant. Serum were separated for the estimation of various biochemical parameters.

Biochemical estimation

The serum GOT was estimated by the method of [6]. The serum GPT was estimated by the method of [6]. The serum alkaline phosphatase activity was estimated by the method of [7]. Protein was estimated by the method of [8].

RESULTS

Hepatospecific enzymes (ALP, ALT and AST), protein and albumin content were activated when hepatocellular damage gave rise to abnormalities of liver function and these enzymes are remarkably increased in diabetic rats (Table 1). Further confirmed in the histopathological studies of liver (Figure 1). *Mollugo cerviana* helps in parenchymal cell regeneration in liver, thus protecting membrane integrity and thereby minimizing enzyme leakage. This result suggested that *Mollugo cerviana* possess potential hepato regenerating activity.

Table 1: Effect of *Mollugo cerviana* extract on liver markers in experimental animals.

S.No.	Parameters	Group – I	Group - II	Group – III	Group – IV
1	SGOT(IU/L)	49.17 ± 3.70 [a]	116.44±0.97[b]	46.23± 2.49 [a]	48.00 ± 2.46 [a]
2	ALP (IU/L)	34.72 ± 1.02 [a]	60.66± 2.16 [b]	36.10 ±2.65 [a]	34.99 ± 1.24 [a]
3	SGPT (IU/L)	29.63 ± 0.44 [a]	42.72±0.17 [b]	29.38 ±0.18 [a]	31.01 ± 0.07 [a]
4	Protein (gm/dl)	7.32 ± 0.12 [a]	4.93 ± 0.03 [b]	7.49 ±0.14 [a]	7.31 ± 0.13 [a]
5	Albumin (gm/dl)	4.62 ± 0.15 [a]	2.99 ± 0.14 [b]	4.90 ±0.24 [a]	4.84 ± 0.18 [a]

Values are expressed as Mean ± SD for six rats. Mean values within a row followed by different letters are significantly different from each other at P <0.05 level comparison by Duncan's multiple range test (DMRT).

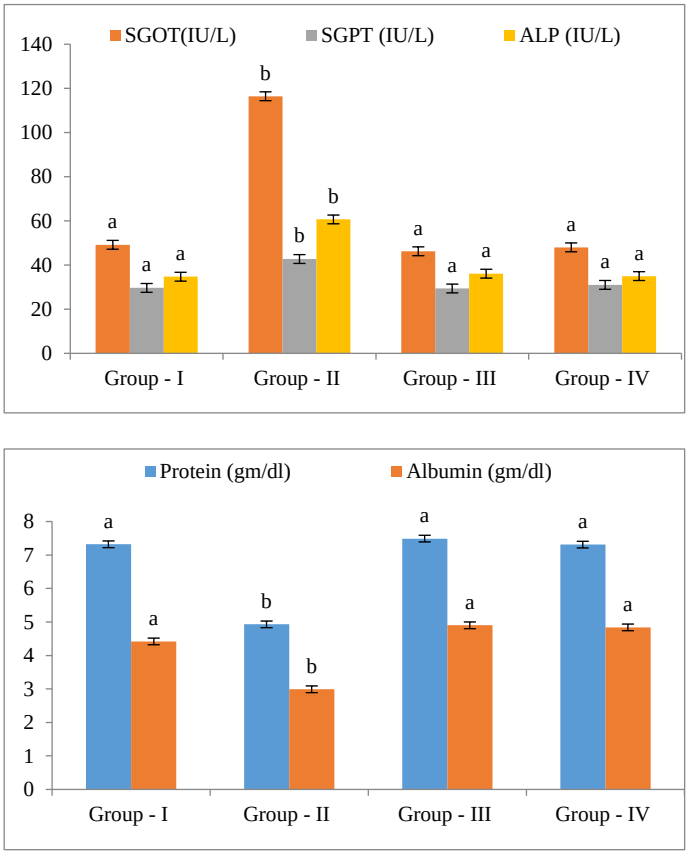

Figure 1: Effect of *Mollugo cerviana* extract on liver markers in experimental animals.

Histology of liver

Liver sections of normal control rats showing normal hepatic cells with well-preserved cytoplasm, well brought out central vein, prominent nucleus and nucleolus (Figure 2a). Liver section of alloxan treated rats showing irregular hepatocytes, ballooning degeneration and kupffer cells around the central vein and the loss of cellular boundaries (Figure 2b). Liver section of rats treated with alloxan and *Mollugo cerviana* extract (500 mg/kg, p.o.) showing well brought out central vein, hepatic cell with well-preserved cytoplasm, prominent nucleus and nucleolus (Figure 2c). The hepatic cells of the glibenclamide treated group seem to be normal architectures comparable to normal group (Figure 2d).

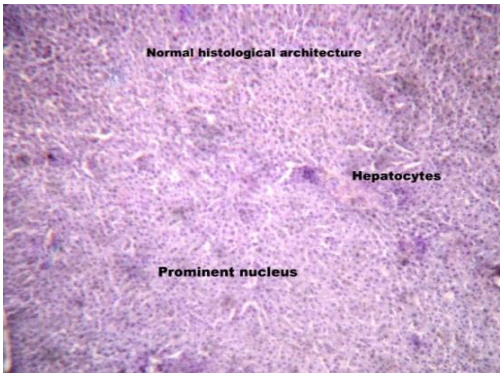

Figure 2a: Normal group

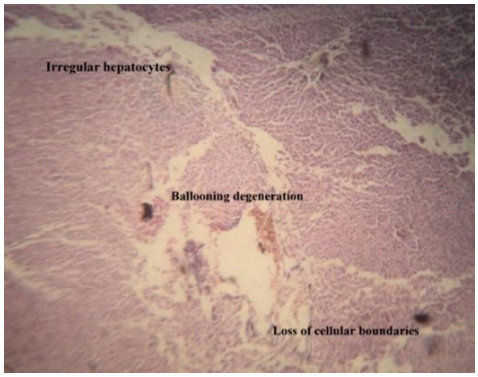

Figure 2b: Diabetic group

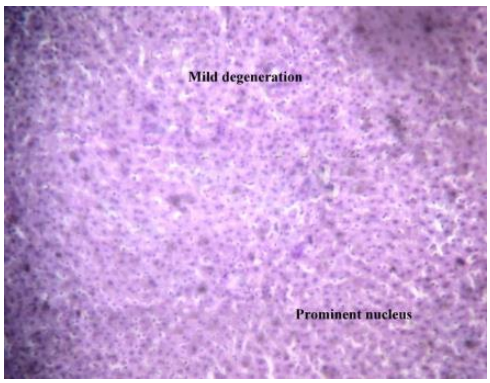

Figure 2c: Diabetic group + Mollugo cerviana treated

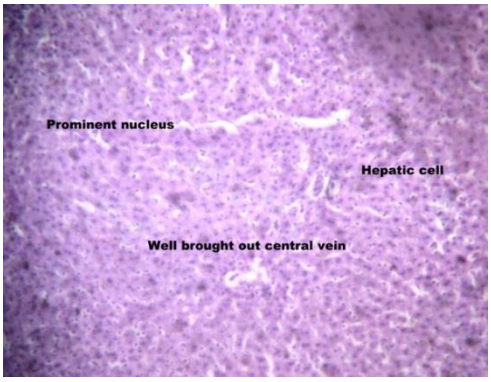

Figure 2d: Diabetic group + standard treated

Figure 2: Histology of pancreas in normal and experimental rats (40x Magnification)

DISCUSSION

Effect of *Mollugo cerviana* extract on liver and kidney markers in experimental animals.

Liver is the central metabolic organ in body responsible for glucose and lipid homeostasis, diabetes leads to hepatic dysfunction. The elevation of plasma concentrations of hepatic tissues enzymes AST, ALP and ALT is an indicator of hepatic damage [9]. Serum transaminases (AST and ALT) and alkaline phosphatase have long been considered as sensitive indicator of hepatic injury [10]. Injury to the hepatocytes alters their transport functions and membrane permeability, leading to the leakage of enzymes from their cells [11]. This leakage causes an increase in levels of serum ALT, AST and ALP. Streptozotocin induced diabetes in rats causes the elevated activities of liver marker enzymes (SGPT, SGOT, and ALP) due to the reported destruction of hepatocytes [12]. Oxidative stress may also be one of the factors which may alter liver enzymes (ALT, AST, and ALP). ALP is also used for the assessment of the liver function. It reaches extremely high levels in biliary obstruction. The altered ALP activity may reflect an increased hepatic insulin resistance or oxidative stress [13].

The present work indicated that the STZ treated rats showed a significant elevating tendency in the serum ALT, AST, ALP. These similar results reported other plants by [14]. The measurement of enzymatic activities of phosphates such as alkaline phosphatase (ALP) is of clinical and toxicological importance as changed in their activities are indicative of tissue damage by toxicants [15]. ALP is present in all tissues of the body, especially in cell membrane and the levels are high in the liver, kidney, bone and placenta. Administration of methanolic extract of *Mollugo cerviana* significantly restored in the activities of AST, ALT and ALP activity was observed. Both Glibenclamide and the extract of *Mollugo cerviana* decreased the alloxan-induced elevated enzyme levels in tested groups, indicating the protection of structural integrity of hepatocytic cell membrane or regeneration of damaged liver cells. These results are in agreement with those reported by [16] who observed a significant reduction of liver markers in hydroethanolic and methanolic extracts of *Artemisia amygdalina* to diabetic rats.

In diabetes mellitus, a variety of proteins are subjected to non-enzymatic glycation and this is thought to contribute to the long term complications of the disease [17]. The level of serum total proteins were found to be decreased in diabetic rats may be ascribed to i) decreased amino acid uptake ii) greatly decreased concentration of variety of essential amino acids. iii) Increased conversion rate of glycogenic amino acids to carbon dioxide and water iv) reduction in protein synthesis secondary to a decreased amount and availability of mRNA [18]. There is a reduced level in total protein content plasma in alloxan induced diabetic rats in the present study. *Mollugo cerviana* significantly increased proteins. Histopathological observation also supported the biochemical parameters. These results are in agreement with those reported by [19] who observed that the *Passiflora ligularis* extract showed significant restoration of proteins content. This results of the present study confirmed that the hepatoprotecive activity of *Mollugo cerviana*.

CONCLUSSION

Administration of methanolic extract of *Mollugo cerviana* significantly restored in the activities of AST, ALT and ALP activity

was observed. Histopathological studies also supported the biochemical parameters. *Mollugo cerviana* helps in parenchymal cell regeneration in liver, thus protecting membrane integrity and thereby minimizing enzyme leakage. The result of the study concluded that *Mollugo cerviana* possess potential hepato regenerating activity.

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