International Journal of PharmTech Research
CODEN (USA): IJPRIF ISSN : 0974-4304
Vol.6, No.6, pp 1870-1874, Oct-Nov 2014

Effect of Methanolic Extract of *Smilax Wightii* A.Dc. on Serum Protein Profile in Streptozotocin Induced Diabetic Rats

Uma Maheswari P[1]*, Shalimol A[1], Arumugasamy K[1], Udhayasankar MR[1], and Punitha D[2].

[1]PG and Research Department of Botany, Kongunadu arts and science college (Autonomous), Coimbatore-641029, Tamilnadu, India.
[2]Department of Botany, Providence College for Women, Coonor, The Nilgiris, Tamilnadu, India.

***Corres.author: umascience@gmail.com**

Abstract: The present study was carried out to investigate the effects of methanolic extract of *Smilax wightii* (MESW) on the biochemical parameters (serum- protein, albumin, globulin, SGPT, SGOT and SALP levels) in diabetic induced rats. . Diabetes was induced in Wistar albino rats by intraperitoneal (i.p) injection of streptozotocin (STZ) at a dose of 55 mg/kgb.wt . Forty eight hours after streptozotocin administration, blood samples were drawn from tail and glucose levels were determined to confirm diabetes. The rats were divided into 6 groups as follows; first group served as normal control which received normal saline, the second group served as diabetic control, received normal saline, the third, fourth, and fifth groups received graded dosage (100, 200 and 400mg/kg b. wt) of the plant extract and Glibenclamide (600m/kg b.wt) was used as the standard drug for the sixth group. The decrease in the levels of serum protein, albumin, and globulin in the diabetic rats were elevated to near normal levels by the extract treatment. The altered levels of serum SGPT, SGOT and ALP were normalized upon treatment with the plant extract. The results of the present study suggest that the plant extract can improve the protein metabolism and is beneficial in preventing diabetic complications.
Keywords: Streptozotocin; *Smilax wightii*; Glibenclamide.

Introduction

Nature has been a source of medicinal agents for thousands of years. Of the 265,000 species of flowering plants that have been identified on this planet, only 0.5% of them have been studied in detail for chemical composition and medicinal value. An impressive number of drugs have been isolated from natural sources, many based on their use in traditional medicine. The plant-based traditional medicine systems continue to play an essential role in health care. Herbal medicine is a major component in all traditional medicine systems and a common element in Ayurveda, Homeopathy, Naturopathy, Traditional Chinese medicine, and Native American medicine. Indigenous people use a wide range of plants therapeutically to maintain their health[1].

Smilax is a large genus of climbing shrubs distributed in temperate and tropical regions. The plant *Smilax wightii* is a member of the family Smilacaceae. Species of *Smilax* have been reported to contain phytoconstituents dioscin, plant steroids such as smilagenin and sarsapogenin[2]. Several species of genus *Smilax* are well- known Chinese traditional medicines, and are used as anti-inflammatory, anticancer and analgesic agents [3].

Materials and Methods

Plant Material

The plant materials were collected from Kodanadu, the Nilgiri Hills, Western Ghats, Southern India, Tamil Nadu. The plant was identified and authenticated by a plant taxonomist.

Animals

Male Wistar Albino rats weighing 180-250 g were obtained from Agricultural University, Animal house lab, Trissur, Kerala. The animals were fed on a standard pellet diet (Hindustan Lever, Kolkata, India) and water *ad libitum*. They were maintained in a controlled environment (12 h/12 h light/dark) and temperature (25 ± 2^0C). The animals were acclimatized to the laboratory conditions for one week before starting the experiment. All the procedures performed on animals were approved and conducted in accordance with the Institution of Animal Ethics committee and by the Regulatory body of the government (659/02/a/CPCSEA).

Preperation of Extract

The whole plant materials were dried in shade after washing with cold water and then powdered using pulveriser and passed through sieve. About100 g of dried plant powder was extracted with petroleum ether using soxhlet apparatus for 18 hours. The petroleum ether was evaporated from the extract and then the residue was re-extracted with methanol. This extract after evaporation of methanol, the filtered residue was stored at 4^0C in refrigerator for further use.

Toxicity Studies

The acute toxicity studies were carried out in adult male albino rats weighing180-250g.The animals were fasted overnight and 100- 1000 mg/kgb.wt of the testextract was given to various groups containing six animals in eachgroup. The treated animals were monitored for 14 days, for mortality and general behaviour.

Experimental Design

The rats were divided into six groups comprising of six animals in each group as follows:

Group I. : - Rats given normal saline daily for 14 days, orally (by using an intragastric catheter tube. (IGC)**.** (Normal control).
Group II: - Diabetic rats given normal saline daily for 14 days, orally by using IGC.(Diabetic control).
Group III : -Diabetic rats given Methanolic extract of *Smilax wightii* (MESW) at the dose of 100 mg/ Kg body weight daily for 14 days , orally by IGC.
Group IV : -Diabetic rats given Methanolic extract of *Smilax wightii* (MESW) at the dose of 200 mg/ Kg body weight daily for 14 days, orally by IGC.
Group V : - Diabetic rats given Methanolic extract of *Smilax wightii* (MESW) at the dose of 400 mg/Kg body weight daily for 14 days , orally by IGC.
Group VI: -Diabetic rats given Glibenclamide at the dose of 600 µg/kg/ body weight daily for 14 days, orally by IGC.

Diabetes was induced in Wistar albino rats by intraperitoneal (i.p injection of streptozotocin (STZ) at a dose of 55 mg/kgb.wt . Forty eight hours after streptozotocin administration, blood samples were drawn from tail and glucose levels were determined to confirm diabetes. The rats with blood glucose level above 200mg/kgb.wt. were considered diabetic and used for the study.

Estimation of Biochemical Parameters

The protein was estimated by the method of Lowry *et al* [4]., albumin and globulin by the method of Wolfson *et al* [5]., SGPT (Alanine Transaminase) and SGOT (Aspartate Transaminase) by Reitman and Frankel[6] and SALP (Alkaline phosphotase by King and Armstrong[7].

Statistical Analysis

All Biochemical data were expressed as Mean ± SEM. Statistical analysis was performed using one-way ANOVA using SPSS statistical analysis programme. In all cases, a, p-value of less than 0.05 was considered to be significant.

Results

Acute toxicity studies revealed that the methanolic extract of *Smilax wightii* (MESW) was found safe to up to a dose of 1000 mg/kg body weight. No sign of toxicity was noticed on the general health of the animals, and no death was observed in the animals when exposed to the extract.

The activity of the plant extract on the protein profile in STZ induced diabetic rats are indicated in Table -1. The protein, albumin and globulin levels were found to be significantly decreased in diabetic control group as compared with normal control group ($P<0.05$). The administration of the plant extract increased the protein content in diabetic animals significantly ($P<0.05$) as compared with the diabetic control group, whereas a slight increase was observed in the albumin and globulin contents.

The activities of SGPT, SGOT and SALP, in the serum of control and experimental groups are presented in Table-2.The activities of SGPT, SGOT and SALP in diabetic controlgroup were significantly ($P<0.01$) elevated as compared to the normal control group. Treatment with *Smilax wightii* methanolic extract at a dose of 400 mg/kgb.wt significantly ($P<0.05$) brought the levels of SGPT, SGOT and SALP towards normal in a dose dependent manner.

Discussion

The aim of the present study was to demonstrate the efficacy of methanolic extract of *Smilax wightii* in recovering the alteredbiochemical parameters to normal levels in STZ induced diabetic rats.

Diabetes mellitus is a group of metabolic disease characterized by chronic hyperglycemia resulting from defects in insulin secretion. The abnormalities in carbohydrate, fat and protein metabolism that are found in diabetes are due to deficient action of insulin on target tissues[8]. Diabetes mellitus is also associated with long-term complications, including retinopathy, nephropathy, neuropathy and angiopathy and several others [9]. Medicinal plants are frequently considered to be less toxic and free from side effects than the synthetic ones. The world health organization has also recommended that this should be encouraged, especially in countries where conventional treatment of daibetes seems inufficient [10].

Currently, the two most widely used chemicals for inducing experimental diabetes are alloxan (2, 4, 5, 6 tetra oxohexahy dropyrimidine, CAS Nº 50 -71 - 5) and streptozotocin (2 – deoxy –2 - (3- methyl -3-nitrosoureido) -D-gluco pyranose, CAS Nº 18883- 66-4). Alloxan and streptozotocin are toxic glucose analogues that preferentially accumulate in pancreatic ß cells via the GLUT2 glucose transporter[11]. Their diabetogenic action is due to the ability to destroy pancreatic ß cells[12]. There is an evidence that the generation of hydrogen peroxide, superoxide anion radicals,and hydroxyl radicals play a critical role in the cytotoxicity of streptozotocin[13].

Transaminases (SGOT,SGPT), SALP, creatinine and urea are good indices of liver and kidney damage respectively[14]. SGOT and SGPT are determined predominantly for hepatocellular damage. High levels of SGOT indicates that the liver is damaged which might be due to toxicants present and it is also seen increased during cardiac infection and muscle injury. Whereas SGPT is more specific to the liver for detecting hepatocellular damage.Increase in the serum level of SALP is due to increased synthesis in presence of increasing biliary pressure[15].

Presently, decrease in SGPT and SGOT and SALP levels in rats were observed after plant extract treatment when compared with controls. Similar results were reported in streptozoin induced diabetic rats with the administration of polyphenolic extracts of *Ichnocarpus frutescens*[16].

Conclusion

The results of the present study reveal that the plant extract showed beneficial effects on the serum biochemical parameters in streptozotocin induced diabetic rats.

Table 1: Effect of MESW on Serum protein, albumin and globulin levels in STZ induced diabetic rats

Groups	Protein (g/dl)	Albumin (g/dl)	Globulin (g/dl)
Normal control	7.96±0.39	4.26±0.14	3.70±0.45
Diabetic control	6.34±0.21*	3.41±0.22	2.93±0.67*
MESW (100 mg/kg)	6.97±0.13	3.54±0.17	3.43±0.89
MESW (200mg/kg)	7.28±0.54	4.05±0.21	3.23±0.12
MESW (400mg/kg)	8.23±0.41[a]	4.23±0.12	4.00±0.23
Glibenclamide (600mg/kg)	8.07±0.53a	4.17±0.36	3.90±0.18

Each Value is SEM of 6 animals * P <0.05 ; ** P< 0.01 comparison with Normal control vs diabetic and drug treated. a ,Comparison made between diabetic control to drug treated groups Level of significance a: P<0.05 , aa: P<0.01

Table 2: Effect of MESW on Serum SGPT, SGOT and SALP levels in STZ induced diabetic rats

Groups	SGPT (u/l)	SGOT (u/l)	SALP (u/l)
Normal control	13.56±0.93	12.42±0.74	183.91±6.54
Diabetic control	69.88±0.84**	57.84±0.81**	286.27±10.93**
MESW (100 mg/kg)	47.28±0.54*	34.96±0.76	207.18±9.84*
MESW (200mg/kg)	31.16±0.86[aa]	18.39±0.27[a]	198.27±7.16[a]
MESW (400mg/kg)	17.81±0.16[a]	11.54±0.27[a]	178.36±5.16[a]
Glibenclamide (600mg/kg)	15.86±0.21[a]	12.67±0.33[a]	157.91±3.96[aa]

Each Value is SEM of 6 animals * P <0.05 ; ** P< 0.01 comparison with Normal control vs diabetic and drug treated. a ,Comparison made between diabetic control to drug treated groups Level of significance a: P<0.05 , aa: P<0.01

References

1. Sayed Nudrat Z., Herbal medicine: Evolution of medicine and Human society., New India Publishing Agency, Book: Medicinal plants-holistic approaches, 2011, p. 1-34.
2. Coimbatore: Botanical survey of India;1989.
3. Shu XS., Gao ZH., and Yang XL., Anti-inflammatory and anti-nociceptive activities of *Smilax china* L.Aqueous extract J.Ethno pharmacol,2006,103(3):327-332.
4. Lowry, OH., Rosenbrough NJ., Farr AL. and Randall R.J., Protein measurement with Folin's phenol reagent. Journal of Biological Chemistry, 1951,193: 265-275.
5. Wolfson, W.Q., Cohn C.,Calvary E. and Ichiba F.,A rapid procedure for the estimation of total protein, true albumin, and globulin in 1ml of serum. American Journal of Clinical Pathology, 1948, 18: 723-730.
6. Reitman, S., and Prankel S. A., Colorimetric method for the determination serum Glutamic oxalo acetic acid and glutamic pyruvic transaminges. Amaerican Journal of Clinical Pathology,1957, 28: 56-63.
7. King, E.J., and Armstrong A.R., Convenient method for determining serum and bile phosphatase activity. Canadian MedicalAssociation Journal, 1934, 31: 376.
8. Craig ME., Hattersley A., Donaghue KC., Definition, epidemiology and classification of diabetes in children and adolescents. Pedia Diab 2009., 10:3-12.
9. Kristova, V., Liskoya Sotnikova S., Vojtko R. and Kurtansky A.,Physiol. Res., 2008,5: 491-494.
10. Santhakumari, P., Prakasam A. and Pugalendi K.V.,I . J. Med. Food., 2006, 9: 108-112.

11. Lenze S., The mechanisms of alloxan- and streptozotocin-induced diabetes. Diabetologia 2008, *51*,216-226.
12. Szkudelski T., The mechanism of alloxan and streptozotocin action in B Cells of the rat pancreas.Physiol Res., 2001, 50, 536-546.
13. Matsumoto S., Koshiishi I.,Inoguchi T., Nawata H , and Utsumi H., *Free Radical Res.*, 2003, **37**, 767.
14. Martin DW., Mayes PA., Rodwell YW., Harper's Review of Biochemistry, 18th Ed, Lange Medical, California, 1981, p. 61.
15. Moss D W, Butterworth PJ., Enzymology and Medicine, 1st Ed, Pitman Medical, London, 1974, p. 139.
16. KumarappanC.T.,Thilagam E.,Vijaya kumar M., and Subhash C. Mandal.,Modulatory effect of polyphenolic extracts of *Ichnocarpus frutescens* on oxidative stress in rats with experimentally induced diabetes, Indian J Med Res 136, November 2012, pp 815-821.

*****