



## IMPACT OF SELAGINELLA BRYOPTERIS ON BIOCHEMICAL ANALYSIS OF DIABETIC SWISS ALBINO MICE CAUSED INDUCED BY ALLOXAN

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### ABSTRACT:

The present study was carried out to evaluate the antidiabetic effect of *Selaginella bryopteris* extract and to study the activities of Blood Glucose, Total cholesterol, Triglyceride, Amylase, Creatinine, Urea, Uric acid in different group of normal, alloxan induced diabetic, insulin alloxan induced diabetic and *Selaginella bryopteris* alloxan induced diabetic. Alloxan (120mg/dl) was administered intra-peritoneal to swiss albino mice and after establishment of diabetes, aqueous extract of *Selaginella bryopteris* (150mg/kg b.w) was orally administered for 26 day. It was noticed that *Selaginella bryopteris* reduced the level of glucose, total cholesterol, triglyceride, amylase, creatinine, urea and uric acid in mice.

**KEYWORD:** *Selaginella bryopteris*, Antidiabetic, Alloxan and International Diabetes Federation (IDF)

### INTRODUCTION:

Diabetes mellitus is endocrine metabolic disorder. All parts of the world mankind health is affected through this disease. The International Diabetes Federation (IDF) estimated that around 15.8 million people currently in India have diabetes and will rise up to 87 million by 2030 (Singh et al., 2014). This disease is a chronic disorder in metabolism of proteins, cholesterol and fat due to absolute or relative deficiency of insulin secretion with/without varying degree of insulin resistance (Ranjana, 2002). It may produce little insulin/ceases to produce insulin, or becomes progressively resistant to its action. It

becomes an epidemic with a worldwide incidence of 5% in the general population. The countries with the largest number of diabetic people in the year 2025 will be India, China and United States (Torban, et al., 2002, Ramachandran, et al., 2002). Patients suffering from diabetes have significant morbidity and mortality from macrovascular complications (stroke, peripheral vascular disease and heart attack) and microvascular (nephropathy, retinopathy, and neuropathy). There is large number of chemical agents available to control the diabetic but complete recovery from diabetes has not been reported till date. Alternative of these synthetic agents, plants extract may provide a potential source of hypoglycemic drugs and are widely used in several traditional systems of medicine to prevent diabetes. Several medicinal

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plants have been investigated for their beneficial use in different types of diabetes. The effects of these plants may delay the development of diabetic complications and improve the metabolic abnormalities using variety of mechanisms. A large number of plants were subjected to clinical trials and were found effective against diabetes. Science during the past few years many phytoconstituents is responsible for antidiabetic effects have been isolated from different plants. During the screening *Selaginella bryopteris* is seems very important. Several *Selaginella* species are used as a traditional medicine in many countries to treat a variety of diseases such as cancer, cardiovascular problems (Lin, 1994), diabetes (Darias, 1989), gastritis (Han, 1984), hepatitis (Lin, 1990), skin diseases (MacFoy, 1983) and urinary tract infections (Winkelman, 1986). Extracts from some *Selaginella* species have shown activity as anti-inflammatory (Han, 1972), antimutagenic (Meng, 1990), antispasmodic (Itokawa, 1983), cytotoxic (Rhan, 1992) and immunestimulant and RNA reverse transcriptase inhibitory agents (Ono, 1810). However, only a few studies on the bioactive components of species in this genus have been performed. A previous investigation on *S. doederleinii* demonstrated that its cytotoxic activity against L929 murine carcinoma cells was correlated to its lignan constituents (Lin, 1994), although several biflavones also isolated from this species were found to be inactive in this same assay. Thus, the folkloric use of many of

these species to treat cancer may not be entirely explained by the presence of lignans, since such compounds have been isolated thus far from only the above-mentioned *Selaginella* species.

#### **MATERIALS METHODS:**

##### **Preparation of plant extracts:**

Aqueous extract of *Selaginella bryopteris* was prepared by shocking 40gms of air dry clean *S. bryopteris* in 200ml distilled water for 12hr at room temperature. Aqueous substance was filtered and effect of *Selaginella bryopteris* finally dried in a vacuum desiccator. Different concentration stock solution was prepared and LD50 was calculated.

##### **Animals:**

6 weeks aged in bred Swiss albino mice with average body weight  $22 \pm 2$ gm were obtained from animal house of University Department of Zoology, T. M. B. U., Bhagalpur (Bihar) India and approval of the ethical committee was sought prior the commencement of the experiment, L. N. M. U., Darbhanga (Bihar). Food and water to mice were provided Ad libitum (prepared mixed formulated feed by the laboratory itself). Mice were housed in colony rooms with 12 hrs light/dark cycle at  $22 \pm 2^{\circ}$  C and were grouped in four different groups.

##### **Experimental Protocol:**

Mice were grouped into four different groups. Group-I Normal Control, Group-II Alloxan Control, Group- III Alloxan + Insulin, Group-IV Alloxan + *Selaginella*. Alloxan (120mg/dl) was prepared in

distilled water and administered intra-peritoneal at the interval of 48 hrs, diabetes was confirmed by blood sugar test, with the help of glucometer (Lever check Pvt. Ltd.) and bio-chemical method. Animals have more than 200mg/dl blood sugar level was selected for the further study and maintained for next 4 days in diabetic condition for well establishment of diabetes. Insulin (0.05U/Mouse) (Singh, *et al.*, 2014) at the interval of 72 hrs was administered intra-peritoneal and *S. bryopteris* (150mg/kg bw) for next 22 days.

**BIOCHEMICAL ASSAY:**

Blood was collected from retro orbital venous puncture (Madway, *et al.*, 1969) into sodium fluoride treated tubes and for other biochemical test; blood was collected in plane tubes. Plasma / Serum were separated by centrifugation. Different biochemical test like Serum glucose, Total cholesterol, Triacylglycerol, amylase, protein, Total lipid, Creatinine Urea Uric acid and LPO were measured by standard kits.

**STATISTICAL DATA:**

Comparison between control and drug treated groups were analysed by sigma plot software with one way ANOVA. The results were expressed as mean ± Standard Error of Mean (S. E. M), N = 6. P-Values < 0.001 were considered to be statistically significant.

**RESULTS AND DISCUSSION:**

The induction of diabetes mellitus in mice by

intraperitoneal administration of alloxan 120 mg/kg.b.w. was established after 7th day of the experiment. Table 1 and Table 2 shows the changes in different biochemical test in experiment mice. Further, Alloxan treated mice exhibited frequent urination, decreased physical activities and sluggishness in comparison to Normal control group mice. Blood sugar, Total cholesterol Triglyceride Amylase Protein Total Lipid and LPO have normal concentration but in alloxan treated group have increased concentration. After 16 days treatment with insulin all test concentration was decreased and after 28th day of *Selaginella bryopteris* treatment concentration was also decreased but it takes more time as compared to insulin treatment (Table 1 and Table 2). It is also reported that alloxan increased hydroxyl radical's in vitro (Grankvist, 1981, Munday, 1988) and in vivo (Kurahashi, *et al.*, 1993). From this experiment it is analysed that *Selaginella bryopteris* is help in reduction of LPO concentration that means it have potential to reduced the generation of hydroxyl free radicals.

**Table: 1 different biochemical analysis in different group of mice (Result represents the mean value of six replicates).**

Group	Blood Glucose (90-120mg/dl)	Total cholesterol (mg/dl)	Triglyceride (mg/dl)	Amylase (U/L)	Protein (NR: 4.0-6.0 mg/dl)	Total Lipid (NR: 51-145 mg/dl)	LPO (NR: 0-1.3 nmol/ml)
Normal Control	112.5±2	47.2±2	90.5±3.2	80±3	5.4± 0.5	95 ± 4	1.05±0.03
Diabetic (after 7 days)	179±4	76.4±2	210±5.5	170±5	3.2±0.5	48± 3	2.25±0.04
Insulin (after 16 days)	116±3	51.5±1.7	97±3.5	90±3	4.0±0.4	70± 5	1.36±0.02
<i>Selaginella bryopteris</i> (after 28 days)	117±3	53.1±1.8	110±4	95±3	4.9±0.3	75± 4	1.15±0.01

**Table: - 2 Showing different biochemical parameters of kidney functions tests in different groups of experimental mice (Result represents the mean value of six).**

Different Groups	Creatinine (NR: 0.6 – 1.2mg/dl)	Urea (NR: 14-40mg/dl)	Uric acid (NR: 3.5-7.2mg/dl)
Normal Control	0.6±0.05	16.0±1.0	4.4±0.6
Diabetic (after 7 days)	2.7±0.06	51.0±1.5	9.7±0.5
Insulin (after 16 days)	1.4±0.05	36.5±1.0	5.0±0.5
<i>Sellaginella bryopteris</i> (after 28 days)	1.3±0.04	30.00±2.2	6.0±0.5

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