

## EFFECT OF NELUMBO NUCIFERAAQUEOUS SEED EXTRACT ON GLUCOSE AND LIPID PROFILE ON ALLOXAN INDUCED DIABETIC ALBINO RATS

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### ABSTRACT

*Nelumbo nucifera* (Lotus) have been used in the traditional system of medicine for various ailments including diabetes. This study was conducted to elucidate the effect of its aqueous seed extract on blood glucose and lipid profile of alloxan induced diabetic rats. Therefore, in the design, eighteen rats were divided into three groups of six each and maintained under ideal laboratory conditions. Group 1 & 2 was taken as normal & diabetic control. Group 3 was treated orally with 200mg/kg body weight aqueous seed extract of *N. nucifera* for 45 days. The diet and water were given *ad libitum*. Blood was collected by retro-orbital puncture for glucose and lipid profile; serum triglyceride, total cholesterol, HDL-cholesterol, LDL-cholesterol and VLDL-Cholesterol estimation. The results showed that administration dose of *N. nucifera* seed extract had significant reduction ( $P<0.05$ ) in both fasting and post-prandial blood glucose level as well as serum lipid profile while significant elevation ( $P<0.05$ ) was found in HDL-cholesterol when compared with diabetic control. This result clearly indicated that *nucifera* seed extract can be key contributor in treatment of diabetes and also could play a cardio protective role.

**Key words:** *Nelumbo nucifera*, Fasting, Post-Prandial, Lipid profile, Alloxan, Diabetes Mellitus

### INTRODUCTION

In recent past numerous workers all over the globe have tried to explore the possibilities of using medicinal plants for prevention, treatment and management of various diseases<sup>1</sup>. One of such diseases, Diabetes Mellitus is a major degenerative disorder that characterized by hyperglycemia and hyperlipidemia which has been rated as a major contributing factors underlying the development of several metabolic diseases<sup>2</sup>. Recently, some medicinal plants have been reported to be useful in diabetes worldwide and have been used pragmatically as antihyperglycemic and antihyperlipidemic reme-

dies. Antidiabetic effects of these plants are attributed to their ability to restore the function of pancreatic tissues by causing an increase in insulin output or inhibit the intestinal absorption of glucose or to the facilitation of metabolites in insulin dependent processes. More than 400 plant species having hypoglycemic activity have been available in literature, however, searching for new antidiabetic drugs from natural plants is still attractive because they contain substances which take alternative and safe effect on diabetes mellitus<sup>3</sup>.

*Nelumbo nucifera* is aquatic plant grown in Asian countries for its edible rhizomes and seeds<sup>4</sup>. The seed of *N.nucifera* Gaertn is known as 'kamalgatta' and widely sold in Indian markets as a vegetable. They are sweet, slightly astringent, flavory, and edible and medicinally versatile<sup>5</sup>. Several pharmacologically active constituents have been used for its medicinal properties that include improving learning and memory<sup>6</sup>, hepatoprotective<sup>7</sup>, anti-obesity, anti-HIV activity, anti-tumor effect<sup>8</sup>, diuretic activity, antipyretic activity, antidepressant, anti-inflammatory<sup>9</sup>, and antihypertensive effect<sup>10</sup>.

With increasing incidence of diabetes mellitus in population throughout the world and due to adverse effects of synthetic medicine, there is a clear need for development of indigenous, inexpensive botanical sources for anti-diabetic crude or purified drugs. The confinements of available oral antidiabetic agents either in terms of efficacy/safety coupled with the emergence of the disease into a global epidemic have encouraged a concerted effort to discover herbal drugs that can manage diabetes more efficiently.

## MATERIALS AND METHODS

**Plant Material and Preparation of Aqueous extract:** Fresh seeds of lotus used in the present study were collected from village Ichoi, Milkipur and authenticated by horticulture department of NDUAT, Kumarganj, Faizabad (UP). The seeds of plant *N.nucifera* were dried and then pounded in a mortar; it was then further ground to powder and stored in an air tight container until required. Two hundred grams of the powder was mixed with 1000ml of distilled water and shaken thoroughly at intervals to ensure ad-

equate extraction. It was then soaked for 48h and then filtered using muslin cloth after which a filter paper was used to obtain a pure filtrate. The filtrate was collected and then evaporated to dryness in a steam bath to give a brownish black residue which was stored in a small plastic container at 4°C<sup>11</sup>.

**Experimental Animals:** Adult male albino wistar rats weighing around 120- 150 g were purchased from Disease free animal house, CCSHAU, Hissar, India. The animals were kept in polypropylene cages (six in each) at an ambient temperature of 25±2°C and 55-65% relative humidity 12±1 h light and dark schedule was maintained in the animal house till the animals were acclimatized to the laboratory conditions, and were fed with commercially available rat chow (Asshiraad Industries, 1544, Sector 38-B, Chandigarh). The water was given *ad libitum*. The experiments were designed into three groups; first group received only normal diet (control group), second group was diabetic control group (without treatment) and last group was experimental group which was treated with 200mg/kg body weight aqueous seed extract of *Nelumbo nucifera* orally for 45 days and conducted according to the guidelines approved by the institutional animal ethics committee CPCSEA Registration No. 574/02/ab/CPCSEA. .

**Induction of diabetes:** Diabetes was induced by injection of a single intraperitoneal (i.p.) dose of alloxan monohydrate freshly prepared in sterile normal saline (0.9%)<sup>12</sup>. The diabetic state was determined after 7 days of alloxination by moderate blood glucose level and gain of body weight. At every 15 days interval, blood glucose level was estimated by glucometer

sticks and at the end of 45 days treatment, serum lipid profile were examined.

### Collection of serum

Animals from each group were deprived of food overnight but with free access of water before taking the sample of blood. On every 15 days of the experiment, the blood samples were collected in test tubes containing EDTA from the orbital plexus by pricking a needle and centrifuged at 2000 rpm at 4°C for 10 minutes to separate serum for the estimation of the various biochemical parameters.

### Biochemical analysis

Biochemical parameters blood glucose estimated by GOD-POD method<sup>13</sup>, Serum total cholesterol (TC) estimated by CHOD/PAP

method<sup>14</sup>, serum Triglyceride (TG) carried out by enzymatic method<sup>15</sup> and serum high density lipoprotein cholesterol (HDL)<sup>16</sup> and serum low density lipoprotein-cholesterol (LDL-C) and very low density lipoprotein-cholesterol (VLDL-C)<sup>17</sup> as per equation: Serum VLDL-C = Serum Triglyceride/5 ; Serum LDL-C = Total serum cholesterol - (Serum HDL-C + Serum VLDL-C)

### Statistical analysis

All the values of blood glucose level and other biochemical estimations were expressed as Mean ± SD of six determinations. Statistical analysis was done by Analysis of Variance (ANOVA) between the groups were considered significance at p 0.05 level<sup>18</sup>.

## RESULTS AND DISCUSSION

**Table 1: Effect of Aqueous Seed Extract of *N nucifera* on Fasting Blood Glucose of Control and Experimental Animals**

Groups	Fasting Blood Glucose Levels (mg/100ml)			
	BT	After Treatment		
		AT <sub>1</sub>	AT <sub>2</sub>	AT <sub>3</sub>
NC	88.09±7.37 <sup>a</sup>	88.70±5.22 <sup>a</sup> (0.69% )	88.68±4.17 <sup>a</sup> (0.66% )	91.12±0.88 <sup>a</sup> (3.43% )
DC	152.13±3.82 <sup>b</sup>	157.47±0.8 <sup>c</sup> (3.51% )	176.71±12.6 <sup>d</sup> (16.15% )	182.88±6.65 <sup>e</sup> (20.21% )
DSE	147.01±1.91 <sup>f</sup>	134.36±4.21 <sup>g</sup> (8.60% )	121.95±3.89 <sup>h</sup> (17.04 )	112.39±4.22 <sup>i</sup> (23.54 )

**Table 2: Effect of Aqueous Extract of *N nucifera* seeds on Post-Prandial Blood Glucose Level of Control and Experimental Animals**

Group	Post-Prandial Blood Glucose Levels (mg/100ml)			
	BT	After Treatment		
		AT <sub>1</sub>	AT <sub>2</sub>	AT <sub>3</sub>
NC	92.85±7.49 <sup>a</sup>	94.63±8.37 <sup>a</sup> (1.91%)	94.63±3.74 <sup>a</sup> (1.91%)	98.38±1.52 <sup>a</sup> (5.95%)
DC	211.17±0.56 <sup>b</sup>	220.01±11.81 <sup>c</sup> (4.18% )	230.71±5.15 <sup>d</sup> (9.25% )	241.23±4.15 <sup>e</sup> (14.22% )
DSE	201.19±12.85 <sup>f</sup>	171.01±14.68 <sup>g</sup>	157.06±11.54 <sup>h</sup>	135.77±3.89 <sup>i</sup>

		(15.0% )	(21.93% )	(32.51% )
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The results are expressed as Mean±SD for six animals in each group. Values not sharing common alphabets (a-i) are differ significantly at p<0.05. NC- Normal Control, DC- Diabetic Control and DSE- Diabetic+200mg/kg body weight seeds aqueous extract treatment. BT= before treatment 0 day, AT1, AT2, AT3= Treatment after 15, 30, 45 days

The effect of aqueous seed extracts of *N. nucifera* on the fasting and post-prandial blood glucose level of experimental animals were determined for 45 days The results were summarized in Table1&2 and Figure1.

The blood glucose level in control normal group showed no significant difference at p>0.05 level. There was a significant (p<0.05) elevation in the fasting and post prandial blood glucose levels in diabetic group by 20 and 14% respectively when compared to normal group whereas the administration of seeds extract caused significant reduction in fasting and post-prandial blood glucose level after 45 day by 23.5% and 32.5 % respectively at p<0.05 level when compared to diabetic control group.

**Table 3: Effect of Aqueous Extract of *N. nucifera* Seeds on Serum Lipid Profile of Control and Experimental Animals**

Groups	Triglyceride (mg/100ml)	Total Cholesterol (mg/100ml)	HDL-C (mg/100ml)	VLDL-C (mg/100ml)	LDL-C (mg/100ml)
NC	82.11±14.26 <sup>a</sup>	77.26±9.53 <sup>a</sup>	26.87±2.46 <sup>a</sup>	16.42±2.85 <sup>a</sup>	38.97±7.57 <sup>a</sup>
DC	177.17±17.59 <sup>b</sup>	113.63±12.52 <sup>b</sup>	22.05±1.58 <sup>b</sup>	35.43±3.52 <sup>b</sup>	56.15±14.94 <sup>b</sup>
DSE	138.83±9.85 <sup>c</sup>	89.41±4.66 <sup>c</sup>	28.49±4.33 <sup>c</sup>	27.76±1.96 <sup>c</sup>	33.16±12.28 <sup>c</sup>

The values are expressed as mean±SD for six animals in each group. Values not sharing common alphabets (a,b,c) are differ significantly at p<0.05. NC- Normal Control, DC- Diabetic Control and DSE- Diabetic+200mg/kg body weight seeds aqueous extract treatment

Table 3 represent the effects of daily administration of aqueous extract of *N. nucifera* seeds at 200mg/kg body weight on various parameters of serum lipid profile on normal and diabetic rats. The levels of triglyceride(177.17±17.59), total cholesterol (113.63±12.52), VLDL-C(35.43±3.52)and LDL-C (57.98±14.94)were significantly elevated and the level of serum HDL-C(22.32±3.06) was decreased in the DC

group as compared to NC group at p<0.05level. After supplementation with aqueous extract of *N. nucifera* seed, the alteration in lipid metabolism was significantly at attenuated as evidenced by decreased serum triglyceride (TG) 138.83±9.85, total cholesterol (TC) 89.45±11.00, VLDL-cholesterol 27.76±1.96and LDL-cholesterol 33.16±12.28levels and increased HDL-cholesterol28.16±4.18concentration in diabetic rats.

### DISCUSSION

In India a number of plants are mentioned in Ayurveda for the treatment of diabetes and some of them have been treated orally in folk medicine with variety of plant extracts<sup>19</sup>.In fact, phytotherapy has been wide-

ly used because of the low cost and the easy availability of medicinal plants. Results of the present investigation are in agreement with the study of Sivasankari<sup>20</sup> that streptozotocin-induced diabetic rats with *nelumbo nucifera* seeds aqueous extract had significantly decreased the blood glucose level. Saini and Sharma<sup>22</sup> also found that *helianthus annuus* seeds ethanolic extract had anti-diabetic effect on diabetic rats and also analyzed that the *nucifera* seed contains appreciable concentrations of these essential elements (Ca, Cu, Na, Zn, V, Mg, Cr, Mn, K and Fe). Diabetic dyslipidaemia is marked by alloxan in diabetic rats by elevated triglycerides, total cholesterol, LDL-C and decreased HDL-C, constitutes important cardiovascular risk factors<sup>22</sup>. These effects may be due to low activity of cholesterol biosynthesis enzymes and or low level of lipolysis which are under the control of insulin<sup>23</sup>. The seed of *nucifera* supplementation also results to the significant attenuation in the level of serum HDL-C toward the control level which again strengthens the hypolipidemic effect of this extract. The findings are in agreement with Kaleem and others revealed that ethanolic extract of *Nigella sativa* seeds produced significant decreased in serum lipid profile (total cholesterol, LDL-C and triglyceride) and increased HDL-cholesterol level<sup>24</sup>. Similar data was reported by Mbakafor *Raphiahooker* seed extract on diabetic rats<sup>25</sup>.

## CONCLUSION

The study had shown that the aqueous extract of *N. Nucifera* seeds possesses blood glucose, cholesterol and triglycerides lowering effect in alloxan induced diabetic rats. Therefore, it may be regarded as a useful

therapy for hyperglycemia and hyperlipidemia which can help to prevent coronary atherosclerosis and may be used for the control and management of diabetes. Further chemical and pharmacological investigations are required to elucidate the exact mechanism of action of this extract and to isolate the active principles responsible for such effects.

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