

**ANTIDIABETIC AND ANTIHYPERLIPIDEMIC EFFECT OF *MORINDA CITROFOLIA* AND *COCCINIA INDICA* IN ALLOXAN INDUCED DIABETIC RATS**

**<sup>1</sup>Manish Kumar and <sup>2</sup>Deepmala Verma**

<sup>1</sup>Department of Zoology, Govt. P.G. College, Jalesar, Etah, U.P

<sup>2</sup>Department of Zoology, Government P.G. College, Etah, U.P

<sup>1</sup>Corresponding Author: manishzoology06@gmail.com

**Summary**

Antidiabetic effects of combined aqueous fruit extract of *Morinda citrifolia* and *Coccinia indica* were evaluated in alloxan induced diabetic rats. The combined aqueous fruit extract of *M. citrifolia* and *C. indica* at a concentration of 300 mg/kg body weight/rat/day was orally administered to Alloxan induced diabetic rats for a period of 7,15 and 30 days. The elevated levels of blood glucose in the diabetic rats reverted back to near normal after treatment with the combined aqueous fruit extract of *Morinda citrifolia* and *Coccinia indica*. Similarly significant decrease in the levels of plasma insulin elevated to near normal after treatment with fruit extract. The extract also showed significant antihyperlipidemic activity with significantly reduction serum cholesterol, triglycerides (TG) and LDL-cholesterol. HDL-cholesterol level was found to be improved ( $p < 0.01$ ) as compared to diabetic control group. The result of this experimental study indicates that combined fruit extract possess a significant antidiabetic and antihyperlipidemic effect.

**Key Words:** Diabetes mellitus, Alloxan, Fruit extract

**Address for Correspondence**

**Dr. Manish Kumar**

Director, Academy of Environment and Life Sciences

27, B.N. Puram, Near Banshi Vihar Colony

Paschim Puri Road, Sikandra-Bodla

Agra-282007, U.P. India

Contact: +91-9457202960

**Introduction**

Diabetes mellitus describes 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. Approximately 140 million people worldwide suffer from diabetes. The disease becomes a real problem of public health in developing countries, where its prevalence is increasing steadily and adequate treatment is often expensive or unavailable. Alternative strategies to the current modern pharmacotherapy of diabetes mellitus are urgently needed, because of the inability of existing modern therapies to control all the pathological aspects of the disorder, as well as the enormous cost and poor availability of the modern therapies for many rural populations in developing countries[1,2].

Plants used in traditional medicine to treat diabetes mellitus represent a valuable alternative for the control of this disease. *Morinda citrifolia* L (Noni) is one of the traditional folk medicinal plants that has been used for over 2000 years in southeast asia and used in india. It has been reported to have a broad range of therapeutic and nutritional value, including antibacterial, antiviral, antifungal, antitumor, antihelmin, analgesic, hypotensive, anti-inflammatory, and immune enhancing effects [3,4, 5].

*Coccinia indica* (Bimba in Sanskrit) known as Ivy Gourd has a long history in ancient Indian medicinal system for its use in diabetes, bronchitis and skin diseases. It is a climbing perennial herb, growing wild throughout India [6].

In the present study, the antidiabetic effect of the fruit extract of *Morinda citrifolia* L and *Coccinia indica* in Alloxan induced diabetic rats was evaluated.

## **Materials and Methods**

### **Experimental Animals**

Albino rats of the Wister strain were used. They weighed between 180 and 200g and were fed on Goldmohar brand feed (manufactured by Lipton India Ltd., New Delhi) and water *ad libitum*. The albino rats maintained under the Good Laboratory Practices (GLP) and follow the guidelines of Committee for the Purpose of Control and Supervision on Experiments on Animals (CPCSEA)[7].

### **Medicinal plants**

The fruits of *Morinda citrifolia* and *Coccinia indica* were used. The plants were obtained from medicinal plant farm house [Govt Regd.], Lucknow. They were identified and authenticated by plant experts in the Department of Botany, Govt P.G.College, Jalesar, Etah.

### **Preparation and administration of plant extracts**

For the preparation of aqueous extract, a decoction was prepared by bringing 500 gm/L of material to boil in the water. The suspension was filtered (Whatmann No.1 filter paper) and the filtered solution was brought to dryness under vacuum. The dried fraction was stored at -20°C until required. The dried fraction was re-extracted with distilled water for experimentations. The rats were given free access to the extracts (300 mg/kg b.wt.) for a period of 30 days.

### **Induction of diabetes in rats**

The rats were injected intraperitoneal with alloxan monohydrate (Span Chemical Co. Mumbai) dissolved in sterile normal saline at a dose of 120 mg kg<sup>-1</sup> b.wt. The rats were kept for 15 days to stabilize the diabetic condition. Only rats with a fasting blood glucose level of at least 200 mg/dl [8].

### **Blood Collection**

The blood samples were collected directly from ventricle of heart. The blood samples which were collected in heparinized tubes were then centrifuged at 3000g for 15 minutes. The clear serum obtained was used for the analysis of glucose and insulin.

### **Experimental Design**

Animals were divided into three groups and for each group six animals were taken. Group I (Normal control) (0.9% NaCl; 5ml/kg.b.w.), Group II served as Alloxan induced diabetic control, Group III received combined fruit extract of *M.citrifolia* and *C.indica* (300mg/kg.b.w.) once a day for 30 days.

**Oral Glucose Tolerance Test (OGTT)**

The oral glucose tolerance test (OGTT) was performed for dose of aqueous fruit extract of *M.citrofolia* and *C.indica* (300mg/kg.b.w.) and blood glucose level was measured by one touch glucometer (accu-check). The glucose level was measured at the interval of 0, 30, 60, and 120 min after the administration of extract.

**Biochemical assays**

Fasting blood glucose level, lipid profiles and serum insulin were evaluated in normal and diabetic rats. The blood glucose level was estimated by one touch glucometer (Accu check). The serum insulin was estimated by Radioimmunoassay (RIA) method [9]. The serum total cholesterol was estimated by the method of Wybenga *et al.* [10]; while triglyceride estimated by the method of McGowan [11]. The serum low-density lipoprotein (LDL-C) was estimated by Fried Wald [12] and HDL-C was estimated by the method of Warnick *et al.* [13].

**Statistical analysis**

All the values in the test are presented as mean  $\pm$  SEM. Statistical differences between the means of the various groups were evaluated by one-way analysis of variance (ANOVA) using the SPSS program followed by Students' *t*-test. P values of 0.05 or less were considered to be significant.

**Results and Discussion**

The mean blood glucose concentration of controlled and aqueous fruit extract of *M.citrofolia* and *C.indica* treated animals on 0, 30, 60 and 120 min are in table-1. The significant reduction ( $p < 0.01$ ) of blood glucose was observed at 60 and 120min of the experiment. Table-2 showed serum glucose decrease and insulin was increased significantly  $p < 0.001$  after oral administration of combined aqueous fruit extract of *M.citrofolia* and *C.indica* in alloxan induced diabetic rats during 7,15 and 30 days respectively as compared to diabetic controls. In the present study, oral administration of *M.citrofolia* and *C.indica* aqueous fruit extract decreased serum glucose and enhanced insulin level in diabetic rats. This hypoglycemic effect may be due to depression of key gluconeogenic or the increase in the levels of glucose transporters and stimulation of uptake in peripheral tissues [8]. Another effect of these plants extract may be that it preserve the cells of islets of langerhans of  $\beta$ -cells functions, which results in a significant increase in insulin activity [14,15].

The most commonly observed lipid abnormalities in diabetes are hypertriglyceridemia and hypercholesterolemia and contribute to coronary artery disease [1]. The repeated administration the extract for a period of 7,15 and 30 days resulted in a significant reduction in lipid profile levels when compared to the diabetic control. In the present study, the total cholesterol, triglycerides and LDL levels was increased in diabetic control groups and it was reduced in 7, 15 and 30 days treatment with combined fruit extract of *M. citrofolia* and *Coccinia indica*. After the treatment with combined fruit extract the HDL cholesterol levels was significantly increased due to mechanism by which, HDL acts as a scavengers by removing cholesterol from peripheral cells such as smooth muscle. In this process free cholesterol in plasma membranes of peripheral tissues comes in contact with the surface of HDL, where it is esterified by lecithin cholesterol acyltransferase (LCAT) and store in core of the HDL [3,6]. This suggests that the combined fruit extract may also inhibit the pathway of cholesterol synthesis.

**Table-1: The Effect of plant extracts on blood glucose levels in alloxan-induced diabetic rats (mg /dl)**

Treatment	0min	30min	60min	120min
Normal	93±3.2	133 ± 6.7	129± 5.8	125 ± 6.0
Diabetic control	298±10.8	301 ± 7.9	300±12.2	302.6 ±14.2
<i>M.citroflia</i> and <i>C.indica</i> (300mg/kg b.wt.)	275 ±19.8	234 ± 4.6*	210±9.5*	177 ± 3.4*

Values are mean concentration of blood glucose ± S.E. (n=6)

Significantly increased or decreased values compared with 0 min data (p<0.05)\*.

**Table-2: Effect of aqueous fruit extract of *M. citroflia* and *C.indica* on blood glucose and insulin activity in Alloxan induced diabetic rats after 7,15 and 30 days.**

Parameters	Normal	Diabetic control	Diabetic control + fruit extract of <i>M.citroflia</i> and <i>C.indica</i> (300 mg/kg b.wt.)		
			7day	15 day	30 day
Serum glucose (mg/dl)	105.5±2.11	298±10.8	227 + 12.9*	195±17.8**	124±4.6**
Serum Insulin (µu/ml)	2.12±0.11	0.34±0.05	0.89±0.045*	1.32±0.06**	1.89±0.07**

Values are given as mean S.Em. for 6 rats per groups

Significant at P<0.01\*; P<0.001\*\*

**Table-3: Effect of aqueous fruit extract of *M. citroflia* and *C.indica* on serum lipid profile in Alloxan induced diabetic rats.**

Parameters	Normal	Diabetic control	Diabetic control+ fruit extract of <i>M.citroflia</i> and <i>C.indica</i> (300 mg/kg b.wt.)		
Total Cholesterol (mg/dl)	136.50±0.75	247.52±1.35	212.30±2.2*1	164.70±2.0*1	148.40±1.4**0
Triglyceride (mg/dl)	81.35±1.32	175.30±2.24	155.18±2.65ns	128.33±2.01*	98.35±1.82**
HDL-C (mg/dl)	29.75±1.35	18.23±1.24	22.40±1.50ns	24.75±0.86*	28.80±1.41**
LDL-C (mg/dl)	40.20±0.70	80.35±0.69	60.40±1.57*	54.56±1.23**	41.85±0.93**

Values are given as mean S.Em. for 6 rats per groups

Significant at P<0.01\*; P<0.001\*\*

### Conclusions

Our study has shown that the combined fruit extract of *M. cirofolia* and *C.indica* is most effective in glucose and lipid lowering effect in diabetic rats.

### References

- [1] Scheen, J.A., Drug treatment of non-insulin dependent diabetes mellitus in the 1990s. Achievement and future development. Drug, 1997.54:355-368.
- [2] Lyra, R., M. Oliveira, D. Lins and N. Cavalcanti. Prevention of type 2 diabetes mellitus. Arq. Bras. Endocrinol. Metabo., 2006. 50: 239-249.
- [3] Y Y Soon and B K H Tan. Evaluation of the Hypoglycemic and Anti- Oxidant Activities of *Morinda officinalis* in Streptozotocin-induced Diabetic Rats. Singapore Med J. 2002. Vol 43(2) : 077-085
- [4] Yoshikawa M, Yamaguchi S, Nishisaka H, Yamahara J, Murakami N. Chemical constituents of Chinese natural medicine, Morindae Radix, the dried roots of *Morinda officinalis* How: structures of morindolide and morofficaloside. Chem Pharm Bull. 1995. 43:1462-5.
- [5] Kamiya K, Hamabe W, Harada S, Murakami R, Tokuyama S, and Satake T. Chemical constituents of *Morinda citrifolia* roots exhibit hypoglycemic effects in streptozotocin-induced diabetic mice. Biol Pharm Bull. 2008. 31(5):935-8
- [6] Hossain MZ, Shibib BA, Rahman R. Hypoglycemic effects of *Coccinia indica*: inhibition of key gluconeogenic enzyme, glucose-6-phosphatase. Indian J Exp Biol.;1992. 30(5):418-20.
- [7] CPCSEA (Committee for the Purpose of Control and Supervision on Experiments on Animals). CPCSEA guidelines for laboratory animal facility. Indian J. Pharmacol., 2003. 35: 257-274.
- [8] Ji Su Kim, Jung Bong Ju, Chang Won Choi and Sei Chang Kim. Hypoglycemic and Antihyperlipidemic Effect of Four Korean Medicinal Plants in Alloxan Induced Diabetic Rats. Am. J. Biochem. Biotechnol. 2006. 2 (4): 154-160, 2006
- [9] Scott, M.D., H.M. Heick and N.B. Heick. An improved double antibody radioimmunoassay for the determination in serum, plasma and incubation media. Can. J. Biochem., 1982. 60(10):962-966.
- [10] Wybenga, D.R. Estimation of total cholesterol in the serum. Clin. Chem., 1970. 16: 980
- [11] McGowan, M.W. Estimation of the concentration of triglyceride in plasma. Clin. Chem., 1983. 29: 538.
- [12] Friedwald, W.T., R.I. Lovy and D.S. Fredrickson. Estimation of the concentration of low density lipoprotein cholesterol in plasma without use of the preparative ultracentrifuge. Clin. Chem. 1972. 18: 499.
- [13] Warnick, G., T. Nguyen and A.A. Albert. Comparison of improved precipitation method for quantification of high density lipoprotein cholesterol. Clin. Chem. 1985. 39:39-46.
- [14] Horsfal, A.U., O.A. Olabiyi, A.A. Osinubi, C.C. Noronha and A.O. Okanlawon. Anti diabetic effect of fruit juice of *Morinda citrifolia* (Tahitian noni juice) on experimentally induced diabetic rats. Nigerian Journal of Health and Biomedical Sciences 2008. 7: (2) 34-39.
- [15] Palayan Muralidharan and Dhanasekaran Sivaraman. Antihyperglycemic and antidiabetic effects of *Morinda tinctoria Roxb* using streptozotocin-induced diabetic rats. Asian Biomedicine. 2009. Vol. 3 No. 4 ; 433-437.