

**EFFECT OF GLIMEPIRIDE ON SERUM GLUCOSE, HbA1c AND
LIPID PROFILE IN NORMAL AND TYPE 2 DIABETIC RATS**

Jagdish Kakadiya*, Dr. N. J. Shah

Pharmacology Department, Dharmaj Degree Pharmacy College, Petlad-Khambhat Road,
Dharmaj, Anand-388430, Gujarat, INDIA.

SUMMARY

Present study was designed to evaluate effect of Glimepiride on serum glucose, HbA1c and lipid profile in normal and Streptozotocin-Nicotinamide induced diabetic in rats. Glimepiride (0.5 mg/kg, p.o) was administered for 28 days in rats injected with single dose of Streptozotocin (65 mg/kg, i.p, STZ) and Nicotinamide (110 mg/kg, i.p, NIC). Administration of STZ–NIC in rats showed a significant ($p<0.001$) increased in the levels of serum glucose, glycosylated hemoglobin (HbA1c), Total Cholesterol (TC), Triglycerides (TG) and High density lipoprotein (HDL) whereas the levels of Low density lipoprotein (LDL) were found to be non significant. Treatment with Glimepiride significantly ($P<0.001$) decreased HbA1c, glucose level, LDL and significantly ($P<0.05$) decreased TC, TG level but no significantly change HDL in compared to diabetic control group. We concluded that GLI (0.5 mg/kg) is effective in controlling blood glucose levels and improves lipid profile in diabetic rats.

KEYWORDS: Glimepiride, Streptozotocin, Nicotinamide, lipid profile

ADDRESS FOR CORRESPONDENCE

Mr. Jagdish L. Kakadiya

Dharmaj Degree Pharmacy College,

Petlad-Khambhat Road, Dharmaj,

Anand-388430, Gujarat, INDIA.

jagdishkakadiya@yahoo.co.in

INTRODUCTION

Three major metabolic abnormalities contribute to the development of hyperglycemia in Type 2 diabetes mellitus such as impaired insulin secretion in response to glucose, increased hepatic glucose production and decreased insulin-stimulated glucose uptake in peripheral tissues. The latter 2 abnormalities are primarily due to insulin resistance (1, 2). Non insulin dependent diabetic mellitus has also been associated with an increased risk for premature arteriosclerosis due to increase in triglycerides and low density lipoprotein levels. About 70-80 % of deaths in diabetic patients are due to vascular diseases. An ideal treatment for diabetes would be a drug that not only controls the glycemic level but also prevent the development of arteriosclerosis and other complication of diabetes.

Glimepiride (GLI) an oral blood glucose lowering drug of the sulfonylurea class is reported to have pancreatic and extra pancreatic effects as well. The blockages of K_{ATP} channels of pancreatic cells by sulphonylureas are critical in the regulation of glucose regulated insulin secretion. It has been demonstrated to be effective in the treatment of non-insulin dependent diabetes mellitus with insulin resistance. GLI is pharmacologically distinct from Glibenclamide because of differences in receptor-binding properties which could result in a reduced binding to cardiomyocyte K_{ATP} channels. Some have reported the antioxidative properties of GLI. Moreover, GLI is pharmacologically different than other sulfonylurea drugs for its cardiovascular effects.

Literature survey showed that, there was no report regarding the effect of GLI on glucose, HbA1c and lipid parameter in diabetic rats. Hence, the purpose of the present study was to instigate the effect Glimepiride on serum glucose, HbA1c and lipid profile in normal and Streptozotocin-Nicotinamide induced diabetic in rats.

MATERIALS AND METHOD

Drugs and Chemicals

Glimepiride was obtained as a gift sample from Alembic Pharmaceuticals Pvt. Ltd., Baroda, India. STZ and NIC were obtained from SIGMA, St. Louis, MO, USA. All other chemicals and reagents used in the study were of analytical grade.

Experimental Animals

All experiments and protocols described in present study were approved by the Institutional Animal Ethics Committee (IAEC) of Dharmaj Degree Pharmacy College, Anand. Sprague Dawley rats (205 ± 15 g) were housed in-group of 3 animals per cage and maintained under standardized laboratory conditions (12- h light/dark cycle, 24°C) and provided free access to palletted CHAKKAN diet (Nav Maharashtra Oil Mills Pvt., Pune) and purified drinking water *ad libitum*.

Experimental Induction of Type 2 Diabetes in Rats

Type 2 Diabetes was induced in rats by a single intraperitoneal (i.p) injection of Streptozotocin (65 mg/kg, STZ) in overnight fasting rats or mice followed by the i.p administration of Nicotinamide (10.5 mg/kg, NIC) after 15 minutes. STZ was dissolved in citrate buffer (pH 4.5) and NIC was dissolved in normal saline. After 7 days following STZ and NIC administration, blood was collected from retro-orbital puncture and serum samples were analyzed for blood glucose (3). Animals showing fasting blood glucose higher than 300 mg/dL were considered as diabetic and used for the further study.

Experimental Protocol

Animals were divided into following groups, each group containing 6 animals and the treatment period for whole study was 4 weeks.

Group 1: Non-diabetic control [0.5 % Sodium CMC (1 ml/kg/day, p.o) as vehicle for 4 weeks (ND-CON)].

Group 2: Non-diabetic control treated with GLI (0.5 mg/kg/day, p.o) as a suspension [0.5 % Sodium CMC for 4 weeks (ND-GLI)].

Group 3: STZ-NIC diabetic control [0.5 % Sodium CMC (1 ml/kg/day, p.o) as vehicle for 4 weeks (D-CON)].

Group 4: STZ-NIC diabetic rats treated with GLI (0.5 mg/kg/day, p.o) as a suspension [0.5 % Sodium CMC for 4 weeks (ND-GLI)]

BIOCHEMICAL ESTIMATIONS

Characterization of Type 2 Diabetes Model

Type 2 diabetes was confirmed by measuring fasting serum glucose using standard diagnostic kit (SPAN diagnostics Pvt., India) and the degree of uncontrolled diabetic state was confirmed by measuring HbA1c (Ion Exchange Resin method). After 4 weeks, diabetes was confirmed by measuring glucose and HbA1c as mentioned above.

Estimation of Serum Markers

On 4th week blood samples were collected from retro-orbital plexus under light ether anesthesia and centrifuged at 2500 rpm for 20 minutes to separate serum. Glucose, HbA1c, TC, TG, HDL and LDL were estimated using diagnostic kits (SPAN Diagnostics Pvt. India).

Statistical Analysis

All of the data are expressed as mean \pm SEM. Statistical significance between more than two groups was tested using one-way ANOVA followed by the Bonferroni multiple comparisons test or unpaired two-tailed student's t-test as appropriate using a computer-based fitting program (Prism, Graphpad 5). Differences were considered to be statistically significant when $p < 0.05$.

RESULTS

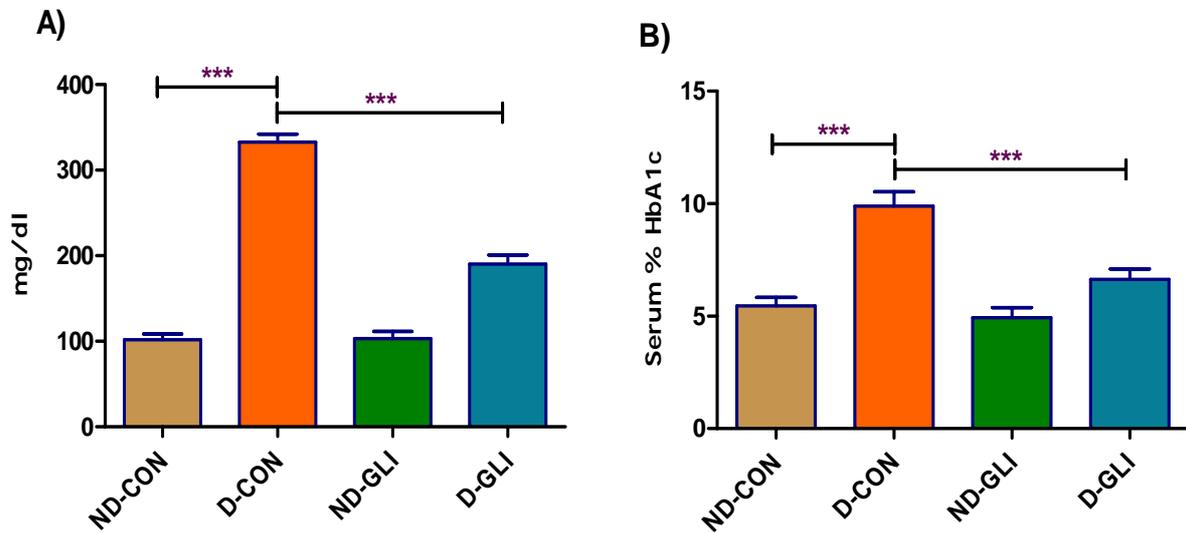
CHARACTERIZATION OF TYPE 2 DIABETES

Single intraperitoneal (i.p) injection of Streptozotocin (65mg/kg) followed by i.p administration of Nicotinamide (10.5 mg/kg) to rats produced severe hyperglycemia and increased HBA1C in 70 to 80 % the animals.

EFFECT OF GLI ON SERUM ENZYMES

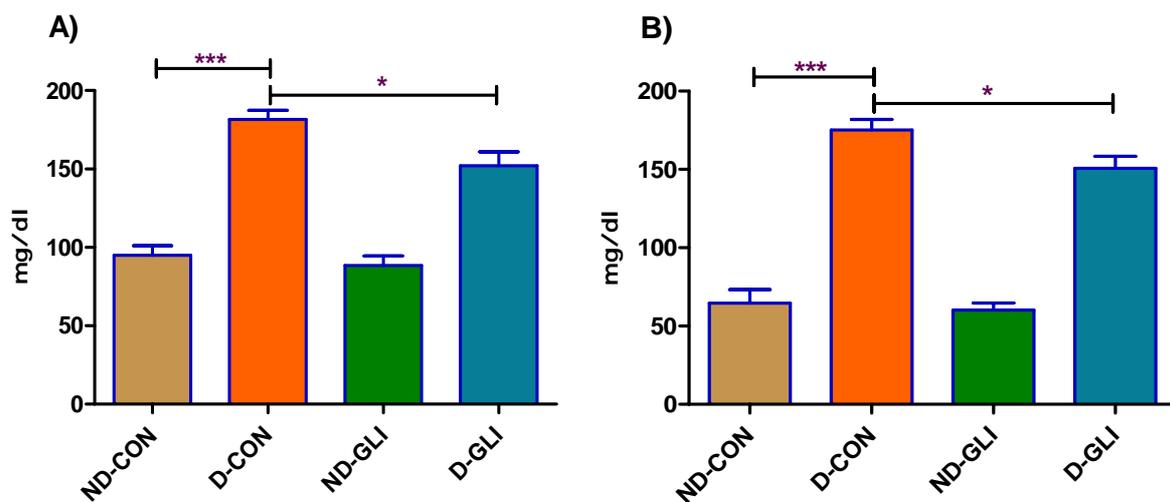
There was a significant ($p < 0.001$) increase in blood glucose and HbA1c level of STZ-NIC injected animals compared to ND-CON group (Fig. 1). Serum total cholesterol, LDL and triglyceride were significant ($p < 0.001$) increase in STZ-NIC diabetic rats as compared to non-diabetic rats. Administration of GLI in STZ-NIC diabetic rats (D-GLI) significant ($p < 0.001$) decrease on blood glucose and HbA1c, while it significantly significant ($p < 0.05$) reduced serum total cholesterol, triglyceride and LDL ($p < 0.01$) in D-GLI group as compared to D-CON group but there was no significant changes in the levels of HDL (Fig. 2).

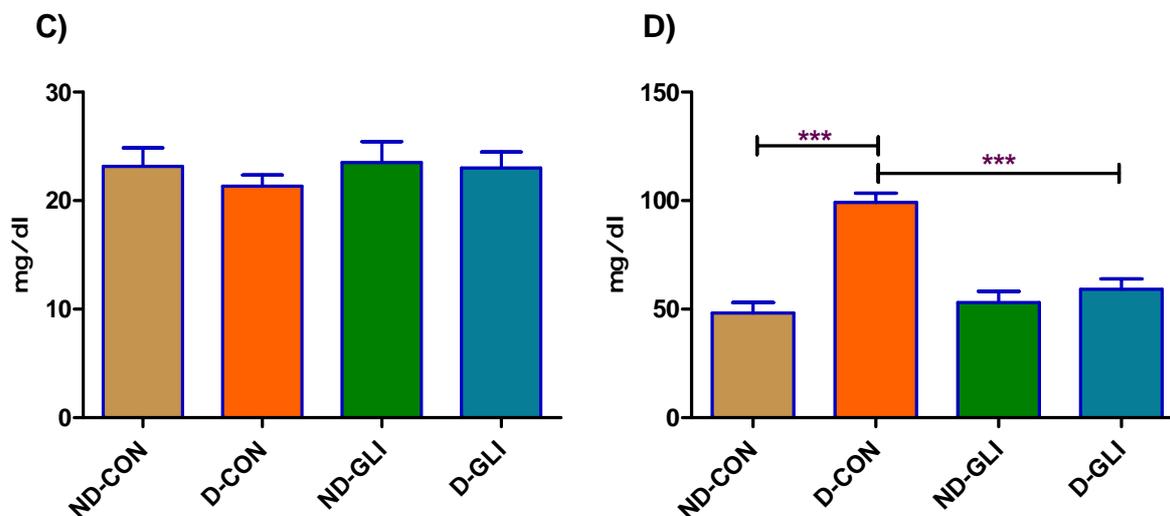
Figure 1. Effect of Glimpiride (0.5 mg/kg/day, p.o) on changes in serum glucose and HbA1c level in normal and STZ-NIC induced diabetic rats.



Values are expressed as mean \pm SEM for six animals in the group. ***P<0.001 considered statistically significant as compared to Control group.

Figure 2. Effect of Glimpiride (0.5 mg/kg/day, p.o) on changes in serum Total Cholesterol (A), Triglycerides (B), High density lipoprotein (C) and Low density lipoprotein (D) level in normal and STZ-NIC induced diabetic rats.





Values are expressed as mean \pm SEM for six animals in the group. * $P < 0.05$, ** $P < 0.001$, *** $P < 0.001$ considered statistically significant as compared to Control group.

DISCUSSION

The present study was undertaken with the objective of exploring the hypoglycemic and hypolipidemic of GLI in STZ-NIC induced diabetic rats. Recent studies have suggested that prevalence of type 2 diabetes is rapidly increasing.

Diabetes mellitus is a chronic disorder caused by partial or complete insulin deficiency, which produces inadequate glucose control and leads to chronic complication. Premature and extensive arteriosclerosis involving renal, peripheral, and cardiovascular vessels remain the major complication of diabetes mellitus. Alteration in the serum lipid profile is known to occur in diabetes and this is likely to increase the risk for coronary heart disease. A reduction in serum lipids, particularly of the LDL and VLDL fraction and TG, should be considered as being beneficial for the long term prognosis of these patients. Lower of blood glucose and plasma lipid levels through dietary modification and drug therapy seem to be associated with a decrease in the risk of vascular disease.

In the present study, an increase in the levels of serum glucose and HbA1c in STZ-NIC treated rats confirmed the induction of diabetes mellitus. Significant decrease was observed in the glucose and HbA1c level in diabetic rats after treatment with GLI (0.5 mg/kg) when compared with D-CON rats at the end of experimental period. STZ causes diabetes by the rapid

depletion of β -cells and thereby brings about a reduction in insulin release. HbA1c level has been reported to be increased in patients with diabetes mellitus (4). It was reported that during diabetes mellitus, the excess of glucose present in the blood reacts with hemoglobin to form HbA1c (5). The level of HbA1c is always monitored as a reliable index of glycemic control in diabetes (6). Elevated levels of HbA1c observed in our study reveal that diabetes animals had prior high blood glucose level.

The chronic diabetic state was also associated with dyslipidemia. Administration of STZ caused increase in serum TC, TG, LDL and Glimepiride (0.5 mg/kg, p.o) could reduce them. This study concluded that GLI at 0.5 mg/kg may show some protection on glucose, HbA1c and lipid parameter in STZ-NIC induced diabetic rats.

REFERENCES

1. Kahn SE, Porte DJ. The pathophysiology of type II (noninsulin-dependent) diabetes mellitus: Implications for treatment. In: Rifkin H, Porte DJ, eds. Ellenberg and Rifkin's Diabetes Mellitus: Theory and Practice. New York: Elsevier Science 1990:436-456.
2. Leibowitz HE. Oral hypoglycemic agents. In: Rifkin H, Porte DJ, eds. Ellenberg and Rifkin's Diabetes Mellitus: Theory and Practice. New York: Elsevier Science 1990:554-574.
3. Masiello, P., Broca, C., Gross, R., Roye, M., Manteghetti, M., Hillaire-Buys, D., Novelli, M., Ribes, G., 1998. Experimental NIDDM: development of a new model in adult rats administered Streptozotocin and Nicotinamide. *Diabetes* 47, 224–229.
4. Paulsen, E.P. Hemoglobin A1C in childhood of diabetes. *Metabolism* 1973; 22: 269- 271.
5. Koenig, R.L., Peterson, C.M. Jones, R.L. Saudek, C. Lehrman, M. and Cerami, A. Correlation of glucose regulation and hemoglobin A1C in diabetes mellitus. *New England Journal of Medicine* 1976; 295: 417-420.
6. Gabbay, K.H. Glycosylated hemoglobin and diabetic control. *New England Journal of Medicine* 1976; 95: 443-454.