e-ISSN: 2320-4230, p-ISSN: 2961-6085

Journal of Drug Discovery and Therapeutics

Available Online at www.jddt.in

CODEN: - JDDTBP (Source: - American Chemical Society)

Volume 11, Issue 03, May-June: 2023, 42-47

# Evaluation of *in-vitro* and *in-vivo* Anti-diabetic Activity of Herbal Tablet

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**Received: 14-04-2023 / Revised: 16-05-2023 / Accepted: 14-06-2023**

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**Conflict of interest: No conflict of interest.**

**Abstract:**

The herbal tablet significantly reduced fasting blood glucose levels compared to the placebo group. Improvements in insulin sensitivity and reductions in oxidative stress markers were observed. Histopathological analysis indicated a protective effect on pancreatic β-cells. The efficacy of the herbal tablet was comparable to that of the standard anti-diabetic medication.

**Keyword:** Anti-diabetic, Herbal Tablet, Glycemic Control, Streptozotocin-Induced Diabetes, Insulin Sensitivity, Oxidative Stress.

## INTRODUCTION

Diabetes mellitus represents a significant global health challenge, with conventional treatments often associated with various side effects. Herbal medicine offers a potential alternative or adjunct to conventional therapies. This study investigates the anti-diabetic activity of a proprietary herbal tablet formulation composed of traditional medicinal plants reputed for their glucose-lowering effects. This study was aimed to evaluate the efficacy and safety of the herbal tablet in reducing hyperglycemia and improving glycemic control in a preclinical model of diabetes[1].

**Materials and method:**

**Formulation of tablet**

The formulation of 650 mg of herbal anti-diabetic tablets was made using the direct compression technique. The active substances used were dry powdered standardized extracts of several plants, including G. sylvestre (150 mg), T. foenum-graecum (150 mg), C. longa (150 mg), and P. nigrum (50 mg). Anhydrous lactose served as a filler, croscarmellose sodium as a disintegrant, and directly compressible microcrystalline cellulose as a binder. Anhydrous lactose was completely combined with 500 mg of the standardized extracts to regulate their moisture-absorbing characteristics. In addition, MCC and CCS were added to the mixture and stirred vigorously for 10 minutes. Formulation batches were adjusted according to the DoE by varying the amounts of binder and disintegrant. For the purposes of lubrication and glidant, talc and magnesium stearate were thoroughly combined with the

aforementioned combination. A Rimek single Rotary tableting machine with 12mm biconvex facing punches was used to crush the mix until it reached a desired weight of 650 mg/Tablet.

**Table 1:**

| Ingredients (mg) | Formulation |
|---|---|
| *G. sylvestre* extract | 150 |
| *T. foenum- graecum* extract | 150 |
| *C. longa* extract | 150 |
| P. nigrum extract | 50 |
| Microcrystalline cellulose | 50 |
| Crosscarmellose sodium | 30 |
| Anhydrous lactose | 65 |
| Talc | 3 |
| Magnesium stearate | 2 |

***In-vivo* anti-diabetic activity**

To assess the formulation's efficacy in relieving diabetic state, the optimized tablet formulation was tested for in-vivo anti-diabetic activity using the streptozocin induced diabetic rat model. Ethical approval from the Institutional Animal Ethics Committee at KLE College of Pharmacy Belagavi was obtained before any animal research was conducted (KLECOP/CPCSEA-Reg,No. 221/Po/Re/S/2000/CPCSEA, Res. 30-13/03/2021). The animals were sourced from a vendor that is registered with the CPCSEA. Prior to the trial, they were allowed to acclimate in a pathogen-free environment for one week. The anti-diabetic investigation used 36 mature albino Wistar rats (210 ± 10 g).

**Induction of diabetes**

The intra-peritoneal method was used to administer streptozocin at a dosage of 35 mg/kg in order to induce diabetes. Fresh streptozocin solution was made by dissolving in citrate buffer that was cooled to 4.5 pH. We included rats in the trial if their fasting blood glucose level was more than 200 mg/dL[2].

**Dose preparation**

To evaluate its anti-diabetic efficacy, the optimized tablet formulation was suspended in RO water, which served as the research vehicle. Previous research indicates that the LD50 of all herbal extracts is more than 2000 mg/kg. Therefore, the dose for anti-diabetic action was assessed as 1/2, 1/10, and 1/20 of 2000 mg/kg. Hence, the per oral (p.o.) doses for the treatment groups were chosen as 100, 200, and 400 mg/kg.

**Experimental design**

The research used a total of 36 rats, broken down into 6 groups of 6 animals each: 6 normal rats and 30 STZ-diabetic rats. In the first group, the rats were given a vehicle intraperitoneally. In the second group, the rats were given glibenclamide at a dose of 5 mg/kg intraperitoneally. In the third group, the rats were given a positive control, STZ+GLIB, and in the fourth group, the rats were given a formulation at a lower dose of

100 mg/kg intraperitoneally. In the fifth group, the rats were given a formulation at a median dose of 200 mg/kg intraperitoneally, and in the sixth group, the rats were given a formulation at a higher dose of 400 mg/kg intraperitoneally. All medications were taken orally for a total of 28 days beginning on the fourth day following STZ delivery[3].

**Study Parameters**

On days 0, 7, 14, 21, and 28, participants reported their weight, as well as their food and drink consumption. We also tracked the percentage change in body weight, as well as food and water consumption. The glucometer (Janaushadi, India) was used to test the fasting blood glucose level. The animals were fasted overnight after the therapy was successfully completed in order to estimate their fasting blood glucose level. In order to evaluate glucose clearance, a glucometer was used to record blood glucose levels from 0 to 120 minutes apart at 30-minute intervals during an oral glucose tolerance test (OGTT). This test included delivering 4 g/kg of exogenous glucose.

Following the administration of a mild anesthetic, the animals were sacrificed. Blood was drawn from the heart through a cardiac puncture, and serum was separated. This allowed for the estimation of biochemical parameters using kits that were available on the market, including HDL, TG, total cholesterol, LDL, and VLDL. Furthermore, the pancreas was removed for the purpose of the histological analysis.

**Histopathology**

Hematoxylin and eosin (H&E) staining, sectioning, and fixing the pancreas with 10% formalin were all steps in the histological evaluation[4].

**Results and discussion**

***In-vivo* anti-diabetic activity**

**Effect on body weight, food intake, and water intake**

There was a change in body weight, food consumption, and water intake across all groups when diabetes was induced. Following the treatment period, the effects on body weight, food consumption, and water intake.

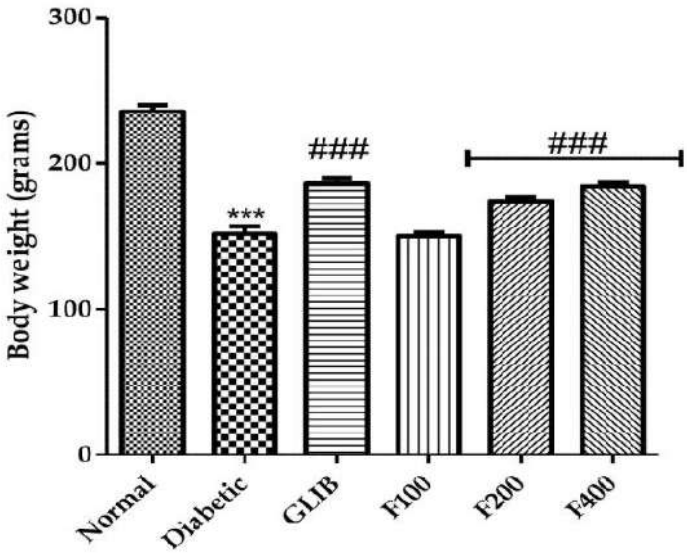


**Figure 2: Effect of formulation on body weight,** ***p<0.001 compared to normal,###p<0.001 compared to diabetic group

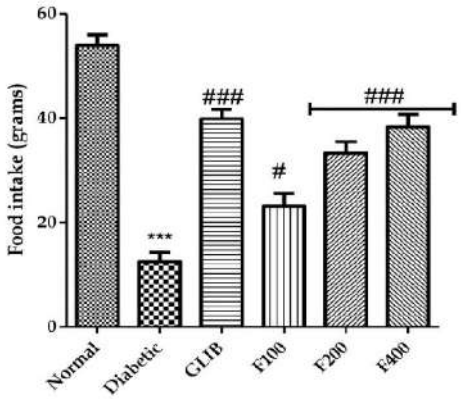


**Figure 3: Effect of formulation on food intake.** ***p<0.001 compared to normal,#p<0.05, ##p<0.01, ###p<0.001 compared to % change in food intake

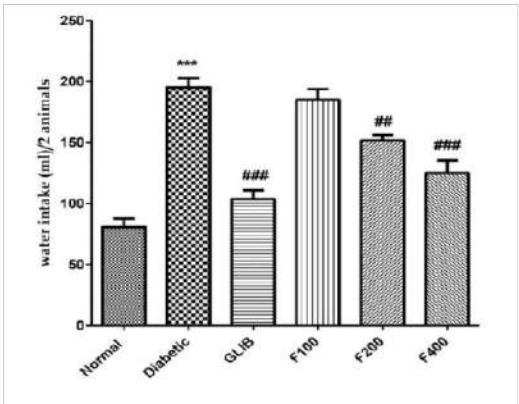


**Figure 4: Effect of formulation on water intake** ***p<0.001 compared to normal,##p<0.01, ###p<0.001 compared to Diabetic

**4.7.2.Effect on Fasting blood glucose level and OGTT glucose level**

After the treatment period, the researchers used oral glucose tolerance testing (OGTT) to see how the formulation affected fasting blood glucose levels and exogenous glucose clearance. The effects on fasting blood glucose level, AUC during OGTT, and Effect are shown in Figures 4.33, 4.34, and 4.35.

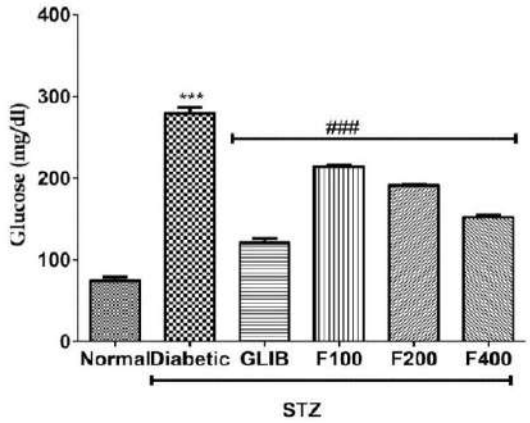


**Figure 5: Effect on fasting blood glucose level.** All the data are presented inmean±SEM (n=6) ***p<0.001 compared to normal, ###p<0.001 compared to Diabetic

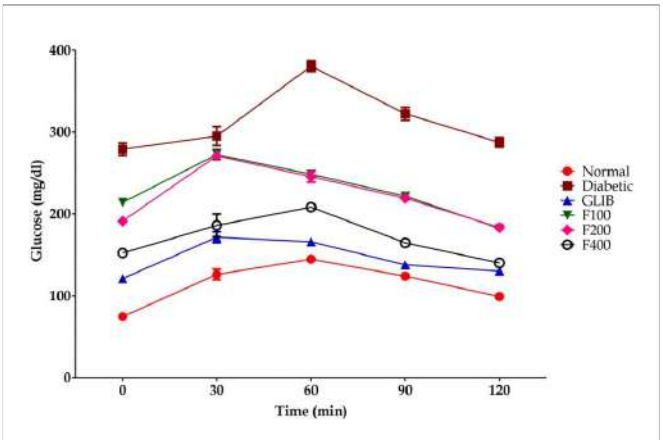


**Figure 6: Total area under the curve of glucose during OGTT**

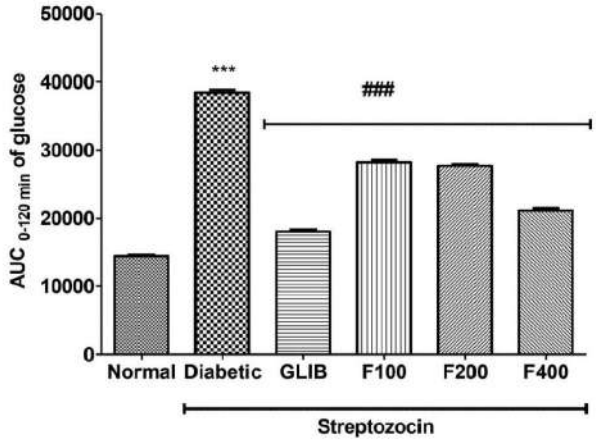


**Figure 7: Effect on total AUC $_{0\text{-}120\ min}$ of glucose on OGTT**

**Histopathology of liver and pancreas**

The histopathological examinations of pancreas are represented in figure.

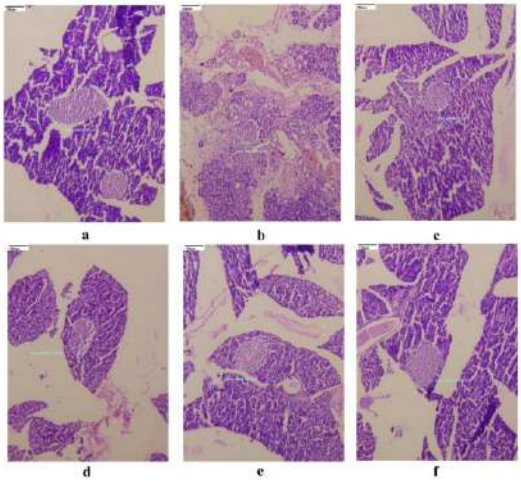


**Figure 8: Effect on the pancreatic histology (a) normal, (b) diabetic, (c) GLIB, (d) F100, (e) F200, and (f) F400**

**Conclusion:**

The herbal tablet demonstrated substantial anti-diabetic activity, evidenced by improvements in glycemic control and pancreatic health. These findings support the potential of the herbal formulation as a complementary approach in managing diabetes. Further clinical studies are warranted to confirm these results and establish long-term safety and efficacy in human subjects.

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