ORIGINAL ARTICLE

# Formulation and Assessment of Antidiabetic Polyherbal Preparations Containing *Azadirachta indica, Tinospora cordifolia,* and *Ocimum sanctum* in Streptozotocin-Nicotinamide-Triggered Diabetic Rats: Comparative Evaluation with Glibenclamide

**Vipinchandra B. Pande[1], Sharad R. Manapure[2], Saket Singh Chandel[3], Krishnakant Bhojrajji Bhelkar[4]**

*[1]Department of Pharmacology, Gurunanak College of Pharmacy, Nagpur, Maharashtra, India, [2]Department of Pharmaceutical Chemistry, Gurunanak College of Pharmacy, Nagpur, Maharashtra, India, [3]Faculty of Pharmacy, Dr. C. V. Raman Institute of Pharmacy, Dr. C. V. Raman University, Kota, Bilaspur, Chhattisgarh, India, [4]Department of Pharmaceutics, Gurunanak College of Pharmacy, Nagpur, Maharashtra, India*

## Abstract

**Background:** A chronic metabolic abnormality marked by elevated plasma glucose concentration that results from defects in insulin production or function is termed diabetes mellitus. This study was conducted to formulate and investigate polyherbal combinations with *Azadirachta indica*, *Tinospora cordifolia*, and *Ocimum sanctum* for their antioxidant and antidiabetic potential. **Methods:** Standardization of plant materials was carried out by pharmacognostic and physicochemical evaluations, followed by preparation of ethanol and aqueous extracts. The analysis of phytochemical constituents confirmed the occurrence of glycosides, flavonoids, polyphenols, and alkaloids. The antioxidant potential of sample to scavenge free radicals was analyzed employing the 2,2-diphenyl-1-picrylhydrazyl (DPPH) method, hydrogen peroxide scavenging, total phenolic, and flavonoid content assays. Eight polyherbal formulations (F1–F8) were prepared and evaluated to investigate the antidiabetic potential *in vivo* activity in Wistar rats through oral glucose tolerance test and diabetes triggered by streptozotocin-nicotinamide in experimental animals. **Results:** Among individual extracts, the hydroalcoholic-free phytochemical extract of *Ocimum sanctum* exhibited the highest antioxidant activity inhibitory concentration 50 ($IC_{50}$) of 28.29 µg/mL, DPPH assay. The extract contained phenolics at 69.5 mg (gallic acid equivalents per gram [GAE/g]) and flavonoids at 65.7 mg (GAE/g). *In-vivo*, polyherbal formulations significantly reduced blood glucose compared with diabetic controls, with F7 and F8 showing the greatest improvement. Formulations also improved lipid profile, enhanced insulin levels, and preserved pancreatic β-cell structure. **Conclusion:** The synergistic effects of *A. indica*, *T. cordifolia*, and *O. sanctum* in polyherbal formulations demonstrated potent antioxidant and antidiabetic activities, supporting their traditional use and therapeutic potential for diabetes management.

**Key words:** Blood glucose, diabetes mellitus, glibenclamide, histopathology, pancreas, streptozotocin-nicotinamide-induced

## INTRODUCTION

Diabetes mellitus is a prolonged metabolic dysfunction in which persistent hyperglycemia promotes oxidative stress, β-cell dysfunction, and progressive complications. Experimental models such as streptozotocin-nicotinamide-induced hyperglycemic rats remain essential for exploring new therapeutic approaches

**Address for correspondence:**
Dr. Saket Singh Chandel, Professor, Faculty of Pharmacy, Dr. C.V. Raman Institute of Pharmacy, Dr. C.V. Raman University, Kota, Bilaspur, Chhattisgarh - 495 113, India.
Mobile: +919827181552. E-mail: singhpharma@gmail.com

**Received:** 03-11-2025
**Revised:** 16-12-2025
**Accepted:** 26-12-2025

because they reliably reproduce pancreatic β-cell injury and oxidative imbalance.[1] Among oral hypoglycemic drugs, glibenclamide is often chosen as the benchmark, thanks to its insulin secretagogue action and well-documented antioxidant benefits in diabetic animals.[2] In the past decade, considerable attention shifted toward bioactive plant products with comparable efficacy. For example, *Pleurotus cornucopiae* extract reduced blood glucose, improved Nrf2 signaling, and protected renal tissue in streptozotocin-nicotinamide-treated rats, showing effects close to those of glibenclamide.[3] Likewise, *Berberis calliobotrys* demonstrated significant hypoglycemic, lipid-lowering, and nephroprotective properties in the same model.[4] Parallel developments in formulation science such as solid lipid nanoparticles, SNEDDS, and hybrid carriers have further improved the stability and bioavailability of phytoconstituents, enhancing their antidiabetic activity.[5] Current treatments involve insulin and oral hypoglycemic agents, which often present side effects and long-term limitations. Herbal medicines, rich in bioactive phytoconstituents, are considered safer and more cost-effective alternatives. Among such botanicals, *Azadirachta indica* (Neem), *Tinospora cordifolia* (Guduchi), and *Ocimum sanctum* (Tulsi) have been extensively used in traditional Ayurvedic medicine for their antidiabetic, antioxidant, immunomodulatory, and protective properties against diabetic complications.[6,7] The current investigation was therefore designed for the assessment and standardization of antioxidant capacity, *in vivo* antidiabetic ability to manage diabetes of polyherbal formulations containing these plants.

## EXPERIMENTAL DESIGN AND PROCEDURES

### Collection of Parts of plants and authentication

Leaves collected in fresh condition of *A. indica* (Neem), stems of *T. cordifolia* (Guduchi), and leaves of *Ocimum sanctum* (Tulsi) were sourced from the residential area of Gurunanak College of Pharmacy, Nagpur. Plant parts were cleaned, shade-dried, coarsely powdered, and stored in airtight containers for further use. The plant specimens were authenticated by Rashtrasant Tukadoji Maharaj Nagpur University, Nagpur. The authenticated specimens have been assigned the following specimen numbers: *T. cordifolia* (10318), *O. sanctum* (10316), and *A. indica* (10321). These specimen numbers serve as official references for the verified plant materials used in research and related studies [Figure 1].

### Standardization of plant materials

To ensure the quality and authenticity of raw materials, standard pharmacognostic and physicochemical evaluations were performed.

### Standardization of plant materials

Pharmacognostic and physicochemical evaluation was carried out to ensure quality and purity. The measurements of total ash value with acid-insoluble ash value and loss on drying (LOD) were performed as well as determined.

### Extraction and phytochemical screening

Powdered drugs were extracted separately using Soxhlet extraction with ethanol and distilled water. The preliminary assessment of the extract confirmed the occurrence of carbohydrates, glycosides, alkaloids, flavonoids, and polyphenols including saponins in diverse amounts was abundant in aqueous extracts, suggesting potential antioxidant activity. Ethanol and aqueous extracts were prepared using coarsely powdered plant materials in a Soxhlet extractor. Ethanol (90%) and distilled water were used as solvents in a ratio of 5 g plant powder to 100 mL solvent. After extraction, the hot extracts were filtered and the solvents were removed by distillation under reduced pressure. Extractive values were determined by macerating 5 g powder in 100 mL solvent for 24 h (6 h shaking, 18 h standing) and evaporating 25 mL filtrate at 105°C. The alcohol-soluble extractive yields were 22.9%, 21.4%, and 24.6%, whereas water-soluble extractive yields were 26.1%, 23.9%, and 27.1% for *A. indica*, *T. cordifolia,* and *Ocimum sanctum*, respectively.

### Evaluation of antioxidant activity

Antioxidant potential of extracts was assessed by 2,2-diphenyl-1-picrylhydrazyl (DPPH) and hydrogen peroxide radical scavenging assays.

### Formulation of polyherbal combinations

Eight formulations (F1-F8) were prepared by combining powdered plant materials in different ratios. Decoctions were prepared by macerating and boiling the powders, and the final volumes were standardized. The ratios in the polyherbal formulations were selected based on the individual antioxidant strength, extractive values, and phytochemical richness of each plant extract. Different proportions were tested to identify the most synergistic combination, ensuring balanced phytoconstituent contribution and maximizing overall therapeutic potential. This systematic variation allowed the selection of the most consistent and effective formulation (F7).

### *In-vivo* antidiabetic activity

#### *Experimental animals*

Male Wistar rats (6–8 weeks, 200–270 g) were housed under standard conditions (21 ± 3°C, 12-h light and 12-h dark cycle). Animals were allowed to acclimatize for 7 days

and subsequently allocated randomly according to their blood glucose levels. The study protocol was approved by the Institutional Ethics Committee (IAEC No: GNCP/IAEC/2019-20/Pharmacology-I).

### *Diabetes mellitus induction in experimental animals*

A diabetic model was generated through intraperitoneal injection of prepared streptozotocin-nicotinamide monohydrate freshly (200 mg/kg). After 48 h, animals with blood glucose higher than 200 mg/dL and symptoms of frequent urination and thirst were classified as diabetic.

### *Study design*

Animals were considered diabetic if their fasting glucose exceeded 200 mg/dL, 48 h post-streptozotocin-nicotinamide injection. Subsequently, they were randomly divided into four groups, each comprising six animals.

- Group 1 – Normal control: Healthy rats administered deionized water (DI water) orally.
- Group 2 – Diabetic control: Diabetic Rats receiving oral administration of vehicle (0.5% sodium carboxymethylcellulose, 10 mL/kg.
- Group 3 – Standard group: The experimental animals received an oral dose of glibenclamide at 10 mg/kg, body weight.
- Group 4 – Test group: The experimental animals received formulation 7 orally, with the dose decided according to preliminary findings.

The treatments were delivered to the animals through oral administration once daily for 21 successive days. Animals were examined every day for clinical signs such as polyuria, polydipsia, and changes in activity. Feed and water intake as well as body weight were recorded daily throughout the experimental period. Fasting blood samples were collected at baseline (day 0) and on days 7, 14, and 21 to assess biochemical parameters.[8-10]

### *Biochemical and histological analysis*

Samples of blood are collected through the retro-orbital plexus using mild anesthesia. The collected blood is centrifuged to obtain serum, which is then used for biochemical analyses. Animals were kept fasting overnight before each collection (on days 0, 7, 14, and 21).

### Fasting blood glucose (FBG)

Blood glucose levels following an overnight fast were assessed instantly following blood collection with an Accu-Chek® glucometer (Roche Diagnostics, Germany).

### Lipid profile

Serum levels of total cholesterol, high-density lipoprotein cholesterol, low-density lipoprotein cholesterol, and triglycerides were measured enzymatically through colorimetric methods, following the protocols provided by the kit manufacturer.

### Glycated hemoglobin (HbA1c)

HbA1c percentage was estimated on day 21 of the study using a commercially available diagnostic kit to assess long-term glycemic control.

### Renal and hepatic function markers

Levels of serum urea, creatinine, serum glutamic-oxaloacetic transaminase (aspartate aminotransferase), and serum glutamic pyruvic transaminase (alanine aminotransferase) were determined through colorimetric assays with automated diagnostic kits procured locally.

### Serum insulin

Following the experimental period, supplementary blood samples were drawn, and serum insulin concentration was analyzed using an enzyme-linked immunosorbent assay kit specific for rat insulin (Elabscience Biotechnology Inc., USA) in accordance with the supplier's guidelines.

### Histopathological examination

After 21 days, animals were humanely euthanized, and the pancreas was removed, washed with PBS, and fixed in 10% neutral-buffered formalin. Tissues were dehydrated through graded alcohols, cleared with xylene, and embedded in paraffin. Sections of 3–5 µm were cut using a rotary microtome, placed on slides, and stained with H&E. Microscopic analysis was conducted at 400× magnification, and images were captured with an Olympus Magnus digital microscope.

### Statistical analysis

Results are reported as mean ± standard error of the mean for six animals per group. Statistical significance was determined using one-way analysis of variance (ANOVA) followed by Tukey's *post hoc* test in GraphPad Prism, with $P < 0.05$ considered significant.

## RESULTS

### Standardization of plant materials

Pharmacognostic and physicochemical evaluation was carried out to ensure quality and purity. The measurements of total ash value with acid-insoluble ash value and LOD


**Figure 1:** Morphology and microscopy of (a) *Azadirachta indica*, (b) *Tinospora cordifolia,* and (c) *Ocimum sanctum*

**Table 1:** Physicochemical parameters of crude drug samples

| Sample of plants | Percentage of total ash content | Percentage of acid-insoluble ash content | Percentage of loss on drying |
|---|---|---|---|
| *Azadirachta indica* (Neem) | 8.1 | 3.5 | 14.8 |
| *Tinospora cordifolia* (Guduchi) | 6.8 | 2.1 | 14.6 |
| *Ocimum sanctum* (Tulsi) | 7.3 | 1.9 | 9.6 |

were performed as well as determined. Neem leaves showed the highest total ash value (8.1%), while Guduchi exhibited the lowest acid-insoluble ash (2.1%). Tulsi leaves had comparatively lower LOD (9.6%), indicating less moisture content and better stability [Table 1].

The standardization of crude drugs revealed acceptable physicochemical parameters [Table 1]. Neem leaves showed the significant total ash value (8.1%), while *Guduchi* stems had the smallest amount of acid-insoluble ash content (2.1%). Tulsi leaves demonstrated the least moisture content with a LOD of 9.6%, indicating better stability compared to the other two plants. Preliminary phytochemical evaluation confirmed the occurrence of carbohydrates, glycosides, flavonoids, polyphenols, and alkaloids in extracts. Saponins were detected in aqueous extracts only, while steroids and triterpenoids were absent. These findings suggest that the three plants are rich in multiple classes of bioactive compounds from secondary metabolites that may contribute to antioxidant and antidiabetic activity.

## Evaluation of antioxidant activity

Tulsi aqueous extract demonstrated the strongest free radical scavenging activity ($IC_{50}$ = 28.29 µg/mL, DPPH), which was comparable to ascorbic acid ($IC_{50}$ = 22.49 µg/mL). Neem showed the least activity among the three plants. All extracts demonstrated significant free radical scavenging potential in antioxidant assays [Table 2]. Among them, tulsi aqueous extract exhibited the strongest activity in both DPPH and $H_2O_2$ assays, with $IC_{50}$ values of 28.29 µg/mL and 47.45 µg/mL, respectively. Statistical analysis has now been incorporated into the antioxidant results. All experimental values are expressed as mean ± SD ($n$ = 3), and group comparisons were performed using one-way ANOVA followed by Tukey's *post hoc* test. Statistical significance has been indicated as $P < 0.05$, $P < 0.01$, and $P < 0.001$ wherever applicable. These significance markers have been added to the tables/graphs in the antioxidant section to clearly distinguish differences between formulations [Table 2].

Estimation of total phenolic and flavonoid content further supported these results [Table 3]. Tulsi aqueous extract contained the greatest concentration of phenolic compounds (69.5 mg gallic acid equivalents per gram), while neem aqueous extract showed the highest flavonoid concentration (74.6 mg QE/g). A significant correlation was observed among phenolic/flavonoid content and antioxidant efficacy. Eight polyherbal formulations were prepared, out of which F7 (neem 50%, guduchi 40%, tulsi 10%) and F8 (neem 50%,

guduchi 10%, tulsi 40%) demonstrated superior antioxidant properties [Table 3].

The selected formulations included F7 and F8, which showed the most promising results in subsequent antidiabetic evaluation.

### Blood glucose level

The polyherbal formulations significantly decreased blood glucose in streptozotocin-nicotinamide diabetic rats relative to diabetic controls. Formulations F7 and F8 showed the greatest hypoglycemic effect. Furthermore, the intervention improved lipid metabolism, raised serum insulin levels, and protected pancreatic β-cells, as demonstrated by histological analysis [Table 4].

**Table 2:** Antioxidant activity of extracts ($IC_{50}$ values)

| Plant/Standard | DPPH $IC_{50}$ (µg/mL) | $H_2O_2$ $IC_{50}$ (µg/mL) |
|---|---|---|
| Neem (Aq. extract) | 73.16 | 97.21 |
| Guduchi (Aq. extract) | 62.99 | 76.02 |
| Tulsi (Aq. extract) | 28.29 | 47.45 |
| Ascorbic acid (Std.) | 22.49 | 43.02 |

DPPH: 2,2-diphenyl-1-picrylhydrazyl

**Table 3:** Selected polyherbal formulations (F1, F4, F7, F8)

| Formulation code | Neem (%) | Guduchi (%) | Tulsi (%) |
|---|---|---|---|
| F1 | 40 | 30 | 30 |
| F4 | 50 | 25 | 25 |
| F7 | 50 | 40 | 10 |
| F8 | 50 | 10 | 40 |

### Lipid profile

The effect of Formulation 7 on lipid profile is shown in Table 5. Diabetic control animals showed elevated total cholesterol, LDL, and triglycerides with reduced HDL levels. Treatment with Formulation 7 significantly reduced total cholesterol, LDL, and triglycerides levels and improved HDL level compared to diabetic control, suggesting improvement in diabetic dyslipidemia [Table 5].

### Glycated hemoglobin

Glycated hemoglobin levels were significantly increased in diabetic control animals compared to normal control. Treatment with Formulation 7 significantly reduced HbA1c levels compared to diabetic control, indicating improved long-term glycemic control shown in table 6.

### Urine profile

Diabetic control animals showed elevated urea, creatinine, SGPT, and SGOT levels indicating renal and hepatic dysfunction. Treatment with Formulation 7 significantly reduced these parameters compared to diabetic control, suggesting protective effects on kidney and liver function [Table 7].

**Table 4:** Effect of test compound - Formulation 7 on the level of blood glucose over the experimental period

| Animal groups | Treatment | Blood glucose concentrations (mg/dL) at different days (mg/dL) | | | |
|---|---|---|---|---|---|
| | | D-0 | D-7 | D-14 | D-21 |
| G1 | Normal control | 86.8±3.5 | 91.5±3.0 | 86.3±3.3 | 88.2±1.6 |
| G2 | Diabetes control (Na-CMC) | 310.8±6.7* | 391.8±16.6* | 411.2±13.2* | 420.3±16.5* |
| G3 | Reference drug-Glibenclamide | 318.2±6.8 | 297.0±5.5*** | 250.3±14.2*** | 161.8±4.8*** |
| G4 | Formulation 7 | 320.2±6.2 | 357.7±19.3 | 307.7±23.8*** | 224.5±7.0*** |

*p < 0.05 vs Diabetic Control group. ** p < 0.01 vs Diabetic Control group. *** p < 0.001 vs Diabetic Control group. Statistical analysis was performed using one-way ANOVA followed by Tukey's post-hoc test.

**Table 5:** Effect of test compound - Formulation 7 on blood lipid level

| Groups | Treatment | Lipid level | | | |
|---|---|---|---|---|---|
| | | Total cholesterol (mg/dL) | HDL (mg/dL) | LDL (mg/dL) | Triglycerides (mg/dL) |
| G1 | Normal control | 72.2±2.8 | 17.0±1.9 | 38.7±2.5 | 74.1±3.4 |
| G2 | Diabetes control (Na-CMC) | 135.6±16.6* | 7.4±0.6* | 80.3±2.4* | 120.3±5.2* |
| G3 | Reference drug-glibenclamide | 91.7±4.4** | 12.5±0.6** | 60.7±3.5** | 80.3±4.2** |
| G4 | Formulation 7 | 103.8±2.3* | 10.0±0.3 | 69.6±1.7* | 102.3±3.8* |

HDL: High-density lipoprotein, LDL: Low-density lipoprotein. *p < 0.05 vs Diabetic Control group. ** p < 0.01 vs Diabetic Control group. *** p < 0.001 vs Diabetic Control group. Statistical analysis was performed using one-way ANOVA followed by Tukey's post-hoc test.

### Blood insulin level

Blood insulin levels were altered in diabetic animals. Treatment with Formulation 7 significantly improved insulin levels compared to diabetic control, suggesting possible improvement in pancreatic function or insulin sensitivity [Table 8].

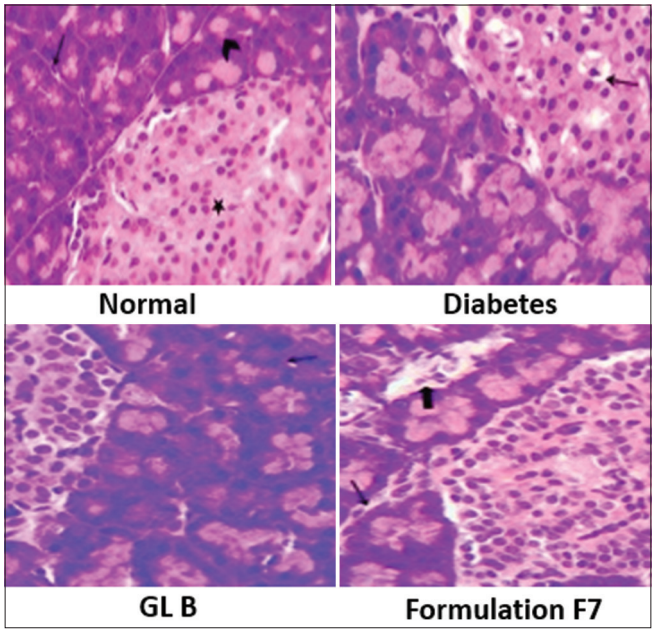


**Figure 2:** Histology of pancreas in experimental rats treated with streptozotocin-nicotinamide and Na-CMC, glibenclamide (GLB, 10 mg/kg) and formulation F7 for 21 days (H and E, ×400)

**Table 6:** Effect of test compound - Formulation 7 on glycated hemoglobin

| Groups | Treatment | Glycated hemoglobin (%) |
|---|---|---|
| G1 | Normal control | 5.9±0.3 |
| G2 | Diabetes control (Na-CMC) | 9.1±0.2* |
| G3 | Reference drug - Glibenclamide | 6.9±0.3*** |
| G4 | Formulation 7 | 7.3±0.1*** |

*p < 0.05 vs Diabetic Control group. ** p < 0.01 vs Diabetic Control group. *** p < 0.001 vs Diabetic Control group. Statistical analysis was performed using one-way ANOVA followed by Tukey's post-hoc test.

### Histopathology of pancreas

Histopathological studies revealed normal pancreatic architecture in normal control rats, whereas diabetic control rats showed severe islet degeneration and β-cell destruction. Treatment with glibenclamide and Formulation F7 showed improvement in pancreatic histology with restoration of islet structure and reduced cellular damage [Figure 2].

## DISCUSSION

The present investigation confirmed that *A. indica* (Neem), *T. cordifolia* (Guduchi), and *Ocimum sanctum* (Tulsi) contain important phytoconstituents such as flavonoids, polyphenols, glycosides, and alkaloids which are known to play a crucial role in diabetes management. Physicochemical constants [Table 1] were within pharmacopoeial limits, ensuring purity and quality of crude materials. Antioxidant assays demonstrated strong free radical scavenging activity in aqueous extracts, especially Tulsi, which showed the lowest $IC_{50}$ in DPPH and $H_2O_2$ assays [Table 2]. These findings correlated with the higher phenolic and flavonoid content of Tulsi and Neem [Table 3]. Since oxidative stress is a major contributor to β-cell damage in diabetes, the antioxidant potential of these plants supports their therapeutic role. The polyherbal formulations [Table 4] showed synergistic benefits. F7 and F8, with higher proportions of Neem and variable amounts of Guduchi or Tulsi, produced the most pronounced hypoglycemic effects in streptozotocin-nicotinamide induced diabetic rats. Among the formulations showing higher *in vitro* potential (F7 and F8), preliminary *in vivo* observations were conducted. F7 showed more consistent hypoglycemic activity, whereas F8 displayed higher variability; therefore, only F7 was selected for full *in vivo* testing. These formulations significantly lowered blood glucose levels, improved lipid profile, increased serum insulin concentration, and preserved pancreatic islet morphology compared with diabetic control. The histopathological analysis supported functional restoration of β-cells, indicating that the herbal extracts not only reduced hyperglycemia but also protected pancreatic tissue. The observed effects can be attributed to multiple mechanisms: Neem and Tulsi are rich in flavonoids and terpenoids known for insulinotropic and antioxidant properties, while Guduchi contains alkaloids

**Table 7:** Effect of test compound - Formulation 7 on urine profile

| Groups | Treatment | Urine profile (mg/dL) | | | |
|---|---|---|---|---|---|
| | | Urea (mg/dL) | Creatinine (mg/dL) | SGPT (IU/L) | SGOT (IU/L) |
| G1 | Normal control | 29.3±1.8 | 0.7±0.0 | 33.9±1.4 | 40.4±1.4 |
| G2 | Diabetes control (Na-CMC) | 144.8±2.7* | 1.5±0.1* | 98.5±2.7* | 119.0±1.8* |
| G3 | Reference drug-glibenclamide | 39.6±2.5*** | 0.9±0.0*** | 39.0±1.1 | 52.2±2.0*** |
| G4 | Formulation 7 | 61.8±5.1*** | 1.1±0.0*** | 44.4±2.2 | 62.1±2.0*** |

SGOT: Serum glutamic-oxaloacetic transaminase, SGPT: Serum glutamic pyruvic transaminase. *p < 0.05 vs Diabetic Control group. ** p < 0.01 vs Diabetic Control group. *** p < 0.001 vs Diabetic Control group. Statistical analysis was performed using one-way ANOVA followed by Tukey's post-hoc test.

**Table 8:** Effect of test compound Formulation 7 on blood insulin level

| Groups | Treatment | Insulin level (µIU/mL) |
|---|---|---|
| G1 | Normal control | 5.9±0.3 |
| G2 | Diabetes control (Na-CMC) | 9.1±0.2* |
| G3 | Reference drug- Glibenclamide | 6.9±0.3*** |
| G4 | Formulation 7 | 7.3±0.1*** |

*p < 0.05 vs Diabetic Control group. ** p < 0.01 vs Diabetic Control group. *** p < 0.001 vs Diabetic Control group. Statistical analysis was performed using one-way ANOVA followed by Tukey's post-hoc test.

and diterpenoid lactones that enhance glucose utilization and modulate immune responses. The synergistic combination of these phytoconstituents appears more effective than individual extracts, aligning with Ayurvedic principles of polyherbalism. Thus, the results validate the traditional use of Neem, Guduchi, and Tulsi in diabetes and demonstrate that polyherbal formulations could provide safe and effective alternatives or adjuncts to conventional therapies.

## CONCLUSION

The study established that aqueous and ethanolic extracts of *A. indica*, *T. cordifolia*, and *Ocimum sanctum* possess potent antioxidant and antidiabetic activities. Among the prepared formulations, F7 and F8 exhibited the highest efficacy, significantly reducing blood glucose, improving lipid profile, and enhancing insulin secretion in diabetic rats. Histopathological evidence further confirmed the protective effect on pancreatic β-cells. In conclusion, these findings highlight the therapeutic potential of Neem, Guduchi, and Tulsi in combination as a polyherbal formulation for diabetes management. The observed synergistic action suggests that such formulations could serve as effective, affordable, and safer alternatives to synthetic drugs. Further investigations and well-designed clinical trials are required to validate the safety and efficacy of these findings before clinical application.

## REFERENCES

1. Bhaskarrao PV, Singh CS, Soni V. Novel antidiabetic polyherbal formulation for synergistic therapeutic effects in streptozotocin (STZ)induced diabetic rats. Int J Drug Deliv Technol 2022;12:1612-7.
2. Chougule S, Shirolkar A, Gacche R, Gaidhani S, Pawar S. Proteomics-based Evaluation of the Antidiabetic Activity of a Polyherbal Formulation in StreptozotocinInduced Hyperglycemia in Rats. Research Square [Preprint]; 2024.
3. Chougule S, Shirolkar A, Gacche R, Gaidhani S, Pawar SD. Proteomics-based assessment of the antidiabetic activity by polyherbal formulation in streptozotocininduced hyperglycemic rat model. J Proteins Proteomics 2025;16:253-67.
4. Anushree VB, Jha DK, Bhattacharjee S. Polyherbal formulation as a potential treatment for type 2 diabetes: An experimental study in streptozotocin - nicotinamide induced diabetic in rats. Int J Pharm Sci Drug Res 2025;17:444-51.
5. Kambale EK, Quetin-Leclercq J, Memvanga PB, Beloqui A. An overview of herbal-based antidiabetic drug delivery systems: Focus on lipid- and inorganic-based nanoformulations. Pharmaceutics 2022;14:2135.
6. Sathyanarayan S, Pillai KS. Antidiabetic and antidyslipidemic properties of GOA-111, a mixture of gymnema sylvestrae, *Ocimum sanctum* and *Azadirachta indica* extract in the ratio of 1:1:1 studied in high fat diet fed- low dose streptozotocin induced experimental type 2 diabetes in rat. J Drug Deliv Ther 2019;9:115-21.
7. Arya R, Faruquee HM, Shakya H, Rahman SA, Begum MM, Biswas SK, *et al*. Harnessing the antibacterial, anti-diabetic and anti-carcinogenic properties of *Ocimum sanctum* Linn (Tulsi). Plants (Basel) 2024;13:3516.
8. Mutia MS, Sipayung YR, Elmiam D, Sihotang WY, Lina J. Granule nanoparticle *Citrus sinensis* (L.) Osbeck peel lowers blood glucose levels and HbA1c in alloxan-induced diabetic rats. Indones Biomed J 2025;17:60-9.
9. Verma S, Pande MS. *Invitro*, *invivo* evaluation and formulation development of polyherbal extract in streptozotocininduced diabetic rat. South East Eur J Public Health 2025;116:1176-83.
10. Pande VB, Chandel SS, Soni V. Synergistic and safe antidiabetic effect of polyherbal formulation: Comprehensive overview. Int J Life Sci Pharma Res 2021;11:P51-7.

**Source of Support:** Nil. **Conflicts of Interest:** None declared.