# Antihyperglycemic, Antiapoptotic efficacy of Psidium guajava L. in alleviating type-2 diabetic complications in streptozotocin-induced male albino rats

**Sowmya BH**

bio.zsowmya@gmail.com

Bangalore University

**Usha Anandhi D**

Bangalore University

**Research Article**

**Keywords:** Psidium guajava L., Diabetes, β-cells, Streptozotocin, Apoptosis

**Posted Date:** July 16th, 2026

**DOI:** https://doi.org/10.21203/rs.3.rs-10246692/v1



**Additional Declarations:** No competing interests reported.

# Antihyperglycemic, Antiapoptotic efficacy of *Psidium guajava* L. in alleviating type-2 diabetic complications in streptozotocin-induced male albino rats

**Sowmya BH*** **and Usha Anandhi D**
Reproductive Physiology Laboratory, Department of Zoology, Bangalore University, Bengaluru, Karnataka, India.

**Corresponding author:**

**Sowmya BH:** Reproductive Physiology Laboratory, Department of Zoology, Bangalore University, Bengaluru, Karnataka, India. ORCID- https://orcid.org/0009-0005-9230-7978

**Email: bio.zsowmya@gmail.com**

## ABSTRACT

**Introduction:** Diabetes mellitus is a metabolic disease characterized by elevated blood glucose levels resulting from insufficient insulin or insulin resistance. In view of its high morbidity and mortality rate, it presents a significant global health challenge. Traditional medicine is the oldest medical practice that existed in human communities before contemporary science was applied to health. *Psidium guajava* L.,(PG), belonging to the family Myrtaceae, is a well-known indigenous plant known for its wide range of nutritive properties. It is rich in vitamin C and antioxidants, capable of inhibiting reactive oxygen species by modulating antioxidant defense enzymes. This study aims to explore the efficacy PG (Lalit variety-LA.L) leaf extracts in preventing type-2 diabetes, and its mechanisms of action on pancreatic β-cells.

**Methods:** Adult male *Wistar* rats (n=6) weighing 180-200grams were divided into four groups of six each. The first group served as the control. The second group was the diabetic group, wherein diabetes was induced by intraperitoneal injection of 45mg/kg/bw of streptozotocin (STZ). The third group was the STZ-induced diabetic group treated with the standard drug Glibenclamide (GLB) of 5mg/kg bw. The fourth group was STZ induced diabetic group treated with the leaf extract of LA.L (150 mg/kg/b.w) for 8 weeks.

**Results:** The outcomes of this study revealed that LA.L extract had a potential effect on fasting blood glucose levels, improved body weight, normalized feed and water intake, and showed a regenerative effect on pancreatic Islets. Further, the treatment of LA.L improved the altered blood lipid profiles. In addition, the treatment restored histo-morphometric parameters, increased organ weight, total protein levels, antioxidant activities, and decreased lipid peroxidation levels in the pancreas. Furthermore, LA.L extract effectively modulated

immunohistochemical findings, with downregulation of Caspase-3 expression and upregulation of Bcl-2 expression. The liquid chromatography-mass spectrometry (LC-MS) analysis indicated the presence of compounds that might exert an anti-hyperglycemic effect. Whereas the anti-diabetic drug GLB showed a moderate recovery in all the parameters.

**Conclusion:** Our findings suggest that LA.L extract has a beneficial effect in protecting pancreatic β-cells by improving their functions by activating antioxidant mechanisms and exerting anti-apoptotic effects.

**Keywords:** *Psidium guajava* L., Diabetes, β-cells, Streptozotocin, Apoptosis.

## 1. **Introduction**

Diabetes mellitus (DM) is turning into the third 'Quiet enemy' in humans, after cardiovascular disorders and cancer, due to its high prevalence of mortality and morbidness (Siam et al., 2024). The International Diabetes Federation (IDF), during the year 2024, estimated that around 589 million adults live with diabetes, and it is predicted to rise upto 853 million by 2050 (Genitsaridi et al., 2026). Type 2 Diabetes mellitus (T2-DM) is the most commonly occurring form of diabetes and appears in roughly 90% of cases (Wagh et al., 2025). T2-DM is caused by decreased cellular sensitivity to insulin's metabolic actions, known as insulin resistance. Insulin regulates the proper utilization of glucose by cells, the transport of glucose within cells, and the maintenance of homeostasis in the body (Nellaiappan et al., 2022). Insulin resistance is closely associated with complications such as hypertension, obesity, dyslipidemia, a cluster of cardiovascular and metabolic abnormalities, which ultimately defines diabetes as a metabolic syndrome (Galicia-Garcia et al., 2020). Immoderate hyperglycemia leads to oxidative stress, thereby causing detrimental effects on various organs through the overproduction of free radicals, especially reactive oxygen species

(ROS). Hyperglycemia-induced excess production of ROS causes cell injury. When the defence mechanisms in the body system fail to curb the increased production of ROS, this leads to oxidative stress. Thus, it acts as an intermediary in insulin sensitivity and the progression of glucose intolerance, consequently leading to various disorders (Chen et al., 2025). Even though the human system has check-in mechanisms to deal with oxidative injury and free radical formation via endogenous and exogenous antioxidants, overproduction of ROS leads to structural deterioration and instability of macromolecules, which in turn affects cell signalling pathways and gene regulation (Afzal et al., 2023). One of the root causes of β-cell damage in T2-DM is the activation of intrinsic apoptotic pathways and mitochondrial dysfunction. Apoptosis can be triggered by a variety of factors, both within and outside the cell, including ligation of cell surface receptors, huge DNA damage, faulty DNA repair mechanisms, irradiation, cytotoxic drugs, loss of survival signals, and contradictory signalling during the cell cycle. Diabetic-related apoptosis is characterized by an imbalance between pro-apoptotic and anti-apoptotic regulators and affects insulin production and pancreatic function (Chandirasegaran et al., 2017).

The use of pharmaceutical interventions like GLB, a sulfonylurea class antidiabetic agent, reduces insulin resistance during DM (Mohan et al., 2022). It is well established that GLB promotes insulin secretion by targeting β cells. GLB promotes insulin release and further activats autophagy through the adenosine 5′-monophosphate (AMP) activated protein kinase (AMPK) pathway in MIN-6 cells. GLB-induced autophagy plays an inhibitory role in promoting insulin secretion by activating the AMPK pathway instead of altering the mammalian target of rapamycin (Zhou et al.,2019). Furthermore, GLB's capacity to reduce oxidative stress and lipid peroxidation in diabetes has been established in multiple

experimental trials in which GLB was used as a control anti-diabetic drug (Madhuri and Naik, 2017). This drug is well known for its ability to prevent hyperglycemia during the course of treatment. Aside from their pharmacological effects, these medications might produce drug-related side effects such as hypoglycemia and cardiovascular problems (Simpson et al., 2006).

In the current era, there is no exceptionally effective treatment for DM, but when insulin treatment is given for a longer period, it can lead to insulin resistance, anorexia nervosa, brain shrinkage, and fatty liver disorders (Nolan and Prentki, 2019).

Traditional medicine is the oldest medical practice that existed in human communities before contemporary science was applied to health. Indian Ayurveda is one of the oldest folk medicines still in practice today (Patwardhan et al., 2005). A vast array of medicinal plants flourishes in India, which is known for its traditional ethnomedicine practices based primarily on plants and plant products (Farnsworth et al., 1985). It has been recognized by the World Health Organization that herbal medicines have a significant role to play in the treatment, prevention, and management of chronic diseases (Pulivarthi et al.,2021). Traditional medicines rely heavily on phytocompounds, which can be identified and standardized through cumulative pharmacognostic studies to develop better treatments (Vignesh et al., 2021). Natural antioxidants, whether in the form of raw extracts or synthesised components, are effective in avoiding oxidative stress-related damage. Most of the medicinal plants lack a complete toxicity description; it is widely believed that remedies derived from plants and fruits are more dependable than their synthetic counterparts (Muscolo et al., 2024).

PG also known as poor man's apple, is an evergreen fruit-bearing tree or shrub with wide-spreading branches and downy twigs that grows 6 to 25 feet tall, belonging to the Myrtaceae family, used as therapeutic medicine worldwide (Joseph and Priya, 2011). Fruits are regarded

as highly nutritious due to their abundant ascorbic acid content (50–300mg/100g fresh weight), which is three to six times greater than that of oranges. It is an exceptional source of fiber, potassium, and vitamin C, and is devoid of fat and cholesterol (Thaipong et al., 2006). PG leaves possess diverse phytoconstituents mainly phenolic compounds like gallic acid, protocatechuic acid, caffeic acid, ferulic acid, ellagic acid, gauvin B, and flavonoids like quercetin, leucocyanidin, and kaempferol, which shows a promising anti-hyperglycemic activity. The leaves of PG were extensively employed for various ailments such as gastrointestinal tract, dysentery, sore throats, bleeding gums, rheumatoid arthritis, vertigo, regulate menstrual cycles, and as a douche for vaginal discharge. A decoction of bark and leaves is given after delivery to expel the placenta. PG tincture was given for the pubescents suffering from seizures and a jelly of the plant is used as a tonic for constipational issues. An infusion of the new shoots can be taken as a febrifuge, and leaves are chewed to relieve toothaches (Conway, 2002). The flavonols present in PG leaf tea prevented viral entry into host cells, counteracting influenza virus growth, including oseltamivir-resistant strains (Sriwilaijaroen et al., 2012). The first electrophysiological investigation based on UV-induced melanogenesis in PG leaves was conducted by Lee et al. (2016). The authors recommended, the usage of methanolic leaf extract inhibit UV-induced skin melanogenesis. The extract has the capability to block both the ORAI1 channel, which has been linked to UV-induced melanogenesis, as well as tyrosinase, an enzyme essential to melanin production. In addition, another studies demonstrated that aqueous extract of PG leaves enhanced insulin secretion from the β-cells of the pancreas and promoted glycogen synthesis in the liver. The extract also inhibited glucose absorption from the upper intestine and partially improved dyslipidemia (Rahman et al., 2023).

The current research focused on evaluating anti-hyperglycemic and anti-apoptotic effects in the pancreas of streptozotocin-induced diabetic rats treated with ethanolic leaf extract of PG.

## 2. Materials and Methods

### 2.1. Collection and preparation of plant materials

The fresh leaves of PG *cultivar Lalit* (LA.L) variety were collected in the post-monsoon season from the Regional Horticultural Research and Extension Centre, University of Horticulture Science, Bengaluru{longitude of 77°56′ E and latitude of 13° 9′ N and 942.37} and were authenticated by a botanist (Acc. No: RRCBI-18272). Fresh leaves of LA.L were cleansed in tap water and then rinsed with distilled water, followed by a shade drying (approx. 45/50days), then the leaves were pulverized using a mechanical blender. 40grams of powdered extract was subjected to soxhlet apparatus with increasing-polarity solvents in the extraction process, including acetone, methanol, ethanol, chloroform, and petroleum ether. For the present study, the ethanolic filtrate was used; later, the extract (ratio 4:1) was then dried in a desiccator.

### 2.2. LCMS characterization

The determination of bioactive compounds in *in vivo* leaf extracts was carried out using LC-MS. Ethanolic extract of LA.L was centrifuged at 12,000 rpm for 10 minutes before analysis. HPLC was performed using two pumps and an automated injector, with a C-18 column and two mobile phases: A-0.01% formic acid and B-90% acetonitrile, at a flow rate of 500 μL min− 1. There was an LC of 5% at 0 and 3 minutes in B, increasing linearly to 20%

between 3 and 25 minutes, to 40% between 25 and 40 minutes, and to 50% between 40 and 55 minutes, and finally to 95% at 55 to 63 minutes (Pai et al., 2015).

### 2.3. Procurement of Animals

Three-month-old, twenty-four male albino rats (*Wistar* strain) weighing about 180-200grams (n=6) were acclimated for a week at 25°C, 40%-60% humidity, 12-hour light/12-hour dark cycles at room temperature (Silambujanaki et al., 2009). During the 8-week experimental period, all animals were fed with a standard rodent diet (Amruth Feeds, India) and water *ad libitum*.

### Induction of Diabetes

After overnight fasting, rats were deprived of food for 16hrs but allowed free access to water. Rats were rendered diabetes intraperitoneally by freshly prepared STZ at a single dose of 45mg/kg/b.w in 0.1M citrate buffer (pH- 4.5) in a volume of 0.5ml (Siddiqui *et al*., 1987). After 72hrs of STZ injection, blood glucose levels were examined for the confirmation of induction of diabetes. STZ induced rats showing fasting blood glucose levels above 200mg/dl were regarded as diabetic and used for the experimental studies (Chauhan *et al*., 2012).

### 2.4. Experimental design

Rats were randomly assigned to four groups of six each. The first group is the control and received distilled water as a vehicle. The second group was the STZ-induced group. The third group was STZ induced and administered with GLB (5mg/kg/b.w/day), and the fourth group

was STZ induced and administered with the LA.L extract (150mg/kg/b.w/day) daily for a period of 8 weeks.

Prior to the setting up of the following groups, a pilot study (50, 100, 150, 200, 250, 500mg/kg/b.w) was conducted in the leaf extract. For the standard anti-diabetic drug GLB, a dose of 5mg/kg/b.w was selected according to Ebokaiwe et al. (2018).

**2.5. Assessment of physical parameters: Body weight, Feed intake, Fluid intake, and Blood glucose level.**

During the experimental period, the initial and final body weights of the rats were recorded using an electronic weighing balance. Food intake was determined by measuring the difference between the pre-weighed food and the weight of the food remaining in the hopper or spilled every 24 hrs. The water intake was measured by recording the quantity of water remaining in the feeding bottle. Blood was acquired by snipping off the tail vein (Flutteret et al., 2000) from control and experimental rats to examine blood glucose levels (Accu-chek active glucometer, Roche Diagnostics, Indianapolis, IN)

**2.6. Biochemical analysis**

After the treatment period, the rats were euthanized by using 1% pentobarbital sodium (0.4mL/100g/b.w). Blood was collected via cardiac puncture, allowed to clot, and centrifuged to separate serum. The pancreas was dissected and weighed after trimming excess fat, the relative organ weight is calculated by dividing the absolute weight of the pancreas by the rat's total body weight. Commercially available enzymatic kit manufactured by Sigma Diagnostics (Pvt.) Ltd., Baroda, India, was used for the following serum lipid profiles analysis (Total cholesterol, Triglycerides, High density lipoprotein, low density lipoprotein, very low-density lipoprotein). Estimation of total proteins (Lowry et al., 1951), Lipid peroxidation (LPO)

{Niehaus and Samuelsson,(1968)}, Superoxide dismutase (SOD) {Misra and Fridovich,(1972)}, Catalase(CAT) {Aebi,1984)}, reduced Glutathione(GSH) {Ellman,(1959)}, glutathione peroxidase(GPx) {Lawrence and Burk, (1976)}, glutathione S transferase(GST) {Habig et al.,(1974)} were carried out in pancreas tissue.

**2.7. Histology and Histomorphometric analysis**

Tissues were washed in buffered saline, fixed in fixative (10% buffered formalin) for 24hrs and dehydrated through grades of alcohol, embedded in paraffin wax (58-60°C) and tissue blocks were made. Sections were cut at 5-6µm thickness, stained with haematoxylin and counterstained with eosin then slides were mounted in DPX and examined under the microscope (Vijayavel and Aswath, 2013).

For, histo-morphometric analysis, stained slides of the pancreas were examined to determine, i) the number of Islets, ii) the diameter of the Islets, iii) the number of β-cells in the pancreatic Islets, and iv) the diameter of β-cells in the pancreatic Islets. Measurement of cells were done using image analysis system (version 1 e).

**2.8. Determination of Immunohistochemical analysis**

Immunohistochemistry was carried out to evaluate the protein expression of Caspase-3 and Bcl-2 in pancreatic tissue. On poly-L-lysine-coated glass slides, 4–5mm tissue sections were mounted. The slides were deparaffinized in 60°C oven for 10minutes before being rinsed twice in xylene for 10minutes each time. Slides were hydrated for 10minutes in a succession of graded ethanol and then in double-distilled water. When exposed to 0.5% hydrogen peroxide, endogenous peroxidases were blocked. The slides were also rinsed in Phosphate

buffer saline (PBS) before being treated with primary antibodies overnight (Caspase-3 and Bcl-2). After 30 minutes of incubation with biotinylated secondary antibody, the sections were again rinsed in PBS and incubated for 20minutes. Then, stained with 3,30-diaminobenzidine for 20 minutes and counterstained with haematoxylin and the intensities of Caspase-3 and Bcl-2 immunoreaction in the pancreas were measured using an image analysis system and recorded on a scale of 0 to 10 (Chandirasegaran et al., 2017).

### 2.9. Determination of gene expression levels of apoptotic (Caspase-3) and anti-apoptotic (Bcl-2) proteins in pancreas by quantitative reverse transcription-polymerase chain reaction (qRT-PCR) method

By using, Takara Prime Script RT kit, gene expression levels were determined. Total RNA was extracted from various concentrations of pancreas of control and experimental groups and purified using RNAiso Plus. Reverse transcription was carried out with an RNA PCR kit and the PCR reaction was performed by One Real Time PCR using the SYBR Green Chemistry (Sensifast SYBR HiRoxkit, Bioline,USA). Reverse transcription reaction was carried out at 37°C for 30mins.

The qRT PCR conditions are mentioned below.

| | | | |
|---|---|---|---|
| Initial denaturation | 95ºC | 20 sec | |
| Denaturation | 95ºC | 15 sec | 40 cycles |
| Annealing & Extension | 60ºC | 30 sec | |

Besides, the specific primers (final concentration 10µmol) were used as mentioned below. For, raw fluorescence data (Ct values) generated by the real-time PCR instrument

(Applied Biosystems) will be exported to qBase plus software 13, to analyze the relative mRNA expression of the genes.

### 2.10. Statistical analysis

Results were expressed as Mean ± S.E.M for six rats in each experimental group. Statistical analysis was performed using IBM SPSS Statistics for Windows, Version 20.0 (IBM Corp., Armonk, N.Y., USA). The data were analysed using one-way analysis of variance (ANOVA), and experimental groups were compared with Tukey's *post hoc* test compared to control group. Significant difference among control and experimental groups when compared to the STZ group ($P<0.05$).

## 3. RESULTS

### 3.1. LC-MS studies;

The obtained chromatogram contained several intense peaks, indicating the presence of various bioactive compounds (Table 1). The most significant ones were identified based on their mass-to-charge (m/z) ratios as gallic acid (m/z 124.87), and caffeic acid (m/z 168.99), Quinic acid (190.94 m/z) quercetin (m/z 300.83), myricetin derivatives (m/z 274.87, 473.06), kaempferol (m/z 316.91), Glycosylated flavonoids were also present, including quercetin-glucoside (m/z 471.04), quercetin-rhamnoside (m/z 491.06). It is well known that these phytochemicals have strong antioxidant, anti-inflammatory, and anti-diabetic effects, which may explain the pharmacological effects observed in the treated groups. The identification of these compounds supports the ethno-medicinal use of LA.L in reducing hyperglycemia and oxidative damage (Figure 1).

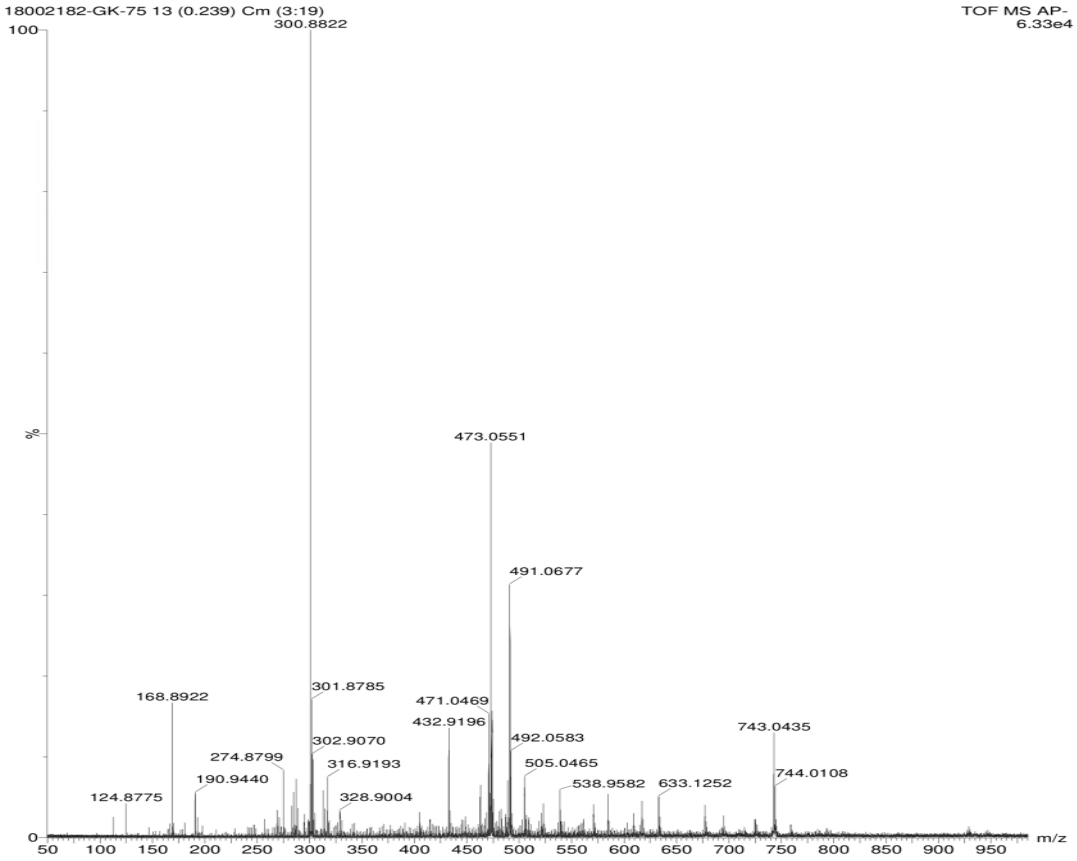
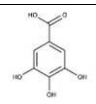
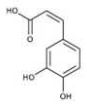
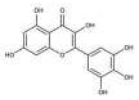
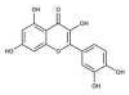
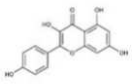
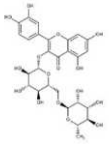
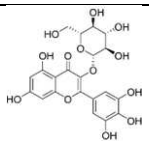
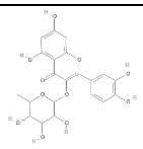
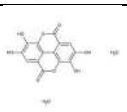



**Figure 1: LC-MS of ethanolic leaf extract of PG**

**Table 1: Identification of phytoconstituents in the ethanolic extract of PG leaves using LCMS analysis.**

| m/z | Compound Identified | Phytochemical Classification | Chemical Structure |
|---|---|---|---|
| 124.87 | Gallic acid (C7H6O5) | Phenolic acid |  |
| 168.99 | Caffeic acid (C9H8O4) | Hydroxycinnamic acid |  |
| 190.94 | Quinic acid (C7H12O6) | Polyol derivative |  |
| 274.87 | Myricetin (C15H10O8) | Flavonoid |  |
| 300.83 | Quercetin (C15H10O7) | Flavonoid |  |
| 316.91 | Kaempferol (C15H10O6) | Flavonol |  |
| 471.05 | Quercetin-glucoside (e.g., Rutin) | Flavonoid glycoside |  |
| 473.06 | Myricetin-hexoside (putative myricetin 3-O-glucoside) | Flavonoid glycoside |  |
| 491.06 | Quercetin-rhamnoside (C21H20O11) | Flavonoid glycoside |  |
| 505.04 | Ellagic acid derivative | Polyphenol |  |

**3.2a. Effect of PG extract on Bodyweight, Feed intake and Water intake level in experimental rats.**

The body weight fluctuations of the experimental and control rats are depicted in Figure 2. From week 1 to week 8, the STZ group's body weight significantly decreased in comparison to the control and other experimental groups ($p < 0.05$). Over the course of the experiment, the body weight of the control group increased steadily. At week 1, the GLB group lost weight, later moderate weight gain was observed. Whereas in the LA.L treated group initial weight reduction at week 1 was noted, followed by gradual weight gain in all the weeks.

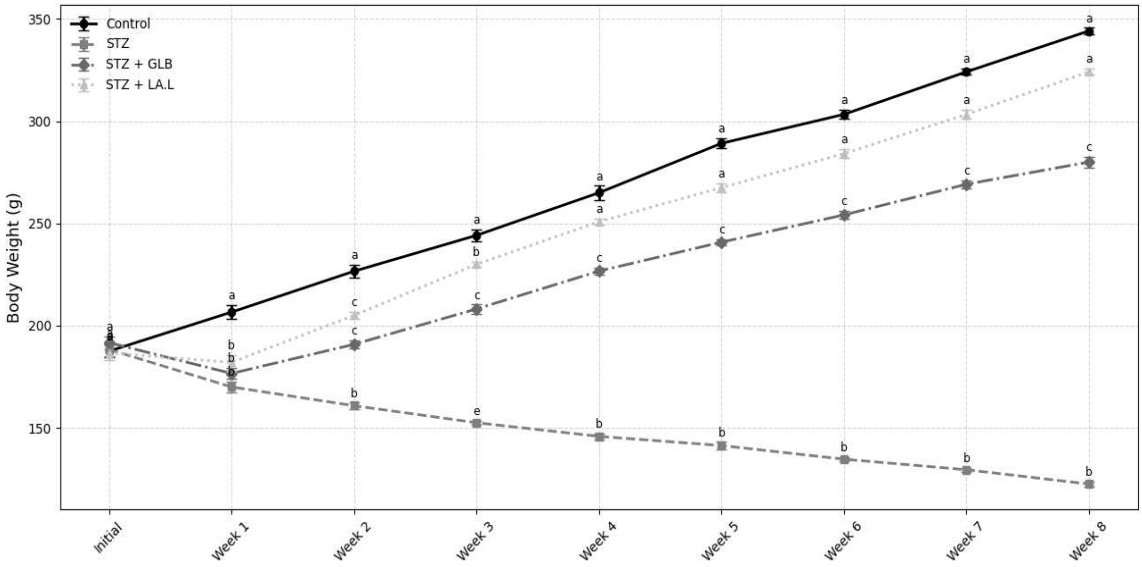



**Figure 2: Effect of PG extract on body weight (gm)in experimental rats.**
(Based on Tukey's *post hoc* test, letters in the same column with the same letters are not significantly different, but those with different letters are significantly different (P<0.05)

In Figure 3, The STZ group showed a substantial increase in food consumption from week 1 to week 8 when compared to the control group ($p < 0.05$), suggesting hyperphagia, which is frequently linked to DM. Also, the GLB and LA.L-treated rats consumed more food during this period, but at a moderate rate compared to STZ group.

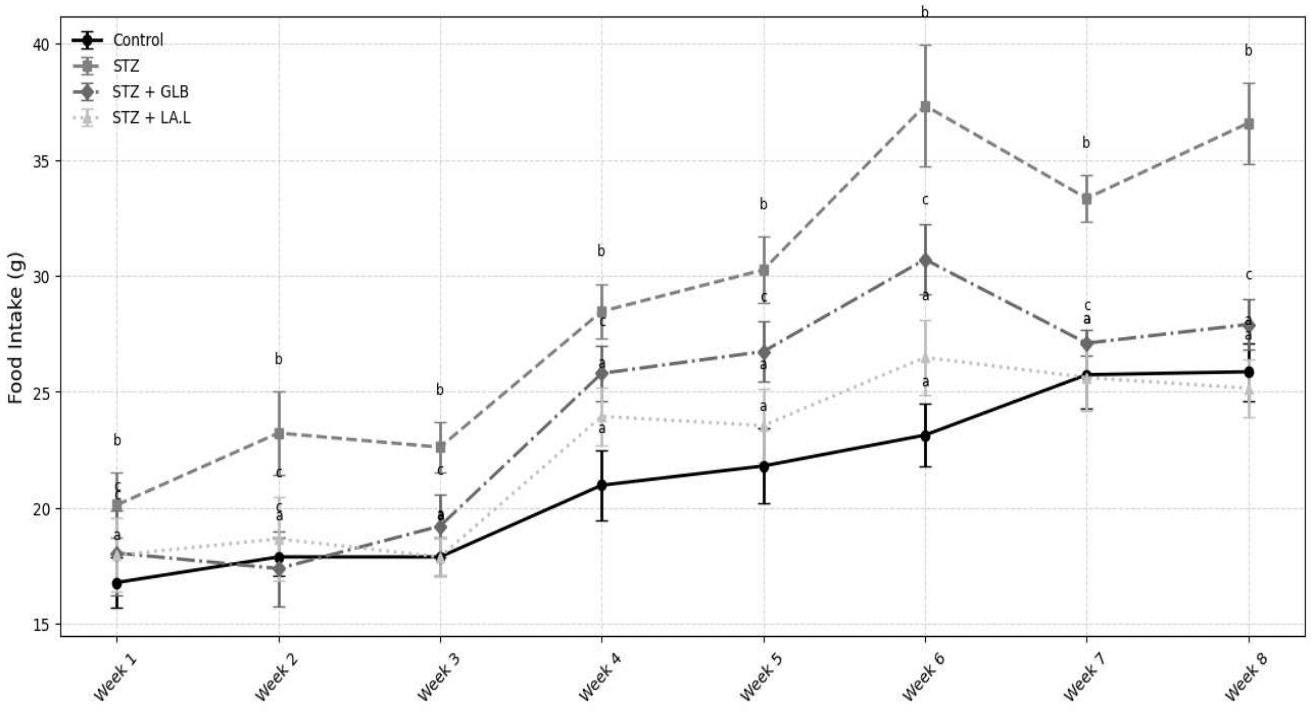



**Figure 3: Effect of PG extract on food intake (gm) in experimental rats.** (Based on Tukey's *post hoc* test, alphabets in the same column with the same letters are not significantly different, but those with different letters are significantly different (P<0.05)

Figure 4 depicts a significant increase in water consumption from week 1 to 5 in STZ group reflecting the typical polydipsia. It is likely that a sudden decline in thirst regulation in week 6 is due to renal adaptation or impaired thirst regulation, while a rise in week 7 is likely due to disease progression with renewed osmotic diuresis. A dynamic physiological response to diabetes over time can be seen in these fluctuations. Among the GLB group, water intake increased moderately, showing partial improvement. Whereas, the LA.L treated group did not demonstrate a significant difference in water consumption in comparison with the control group, indicating that LA.L extract effectively normalized water intake in diabetic rats.

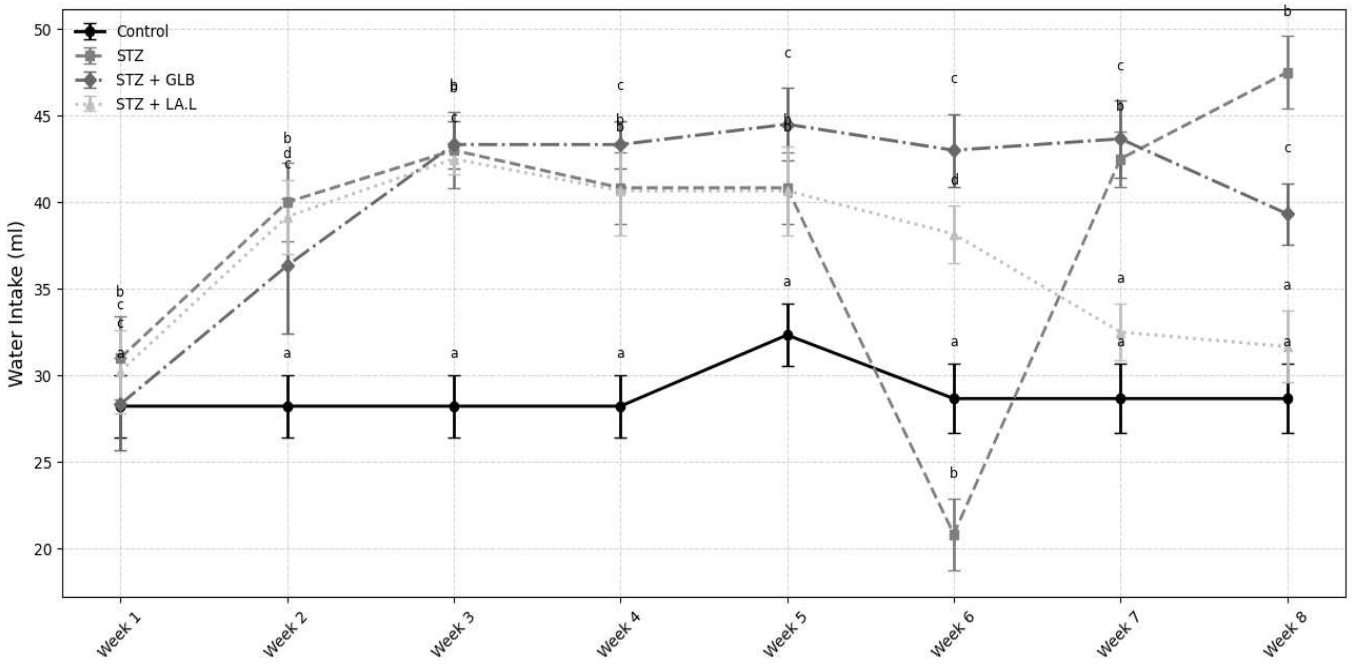



**Figure 4: Effect of PG extract on water intake (ml) in experimental rats.**
(Based on Tukey's *post hoc* test, alphabets in the same column with the same letters are not significantly different, but those with different letters are significantly different (P<0.05)

### 3.2b. Effect of PG extract on Blood glucose level in experimental rats.

Figure 5, illustrates a marked elevation in the blood glucose level (week 1-8) in STZ group in contrast to other groups ($p < 0.05$) confirming hyperglycemia. At week 1, LA.L administered group showed a transient increase in blood glucose levels; however, over time, glucose levels decreased substantially. This indicates the potential antihyperglycemic effect of LA.L extract. Similarly, the GLB-treated group experienced an initial rise in blood glucose level, and gradual reduction was noted by week 8. As, GLB treatment effectively reduced blood glucose to nearly-normal levels possessing its anti-diabetic efficacy. Nonetheless, control group rats maintained normal blood glucose levels throughout the experiment, suggesting a stable glucose regulation.

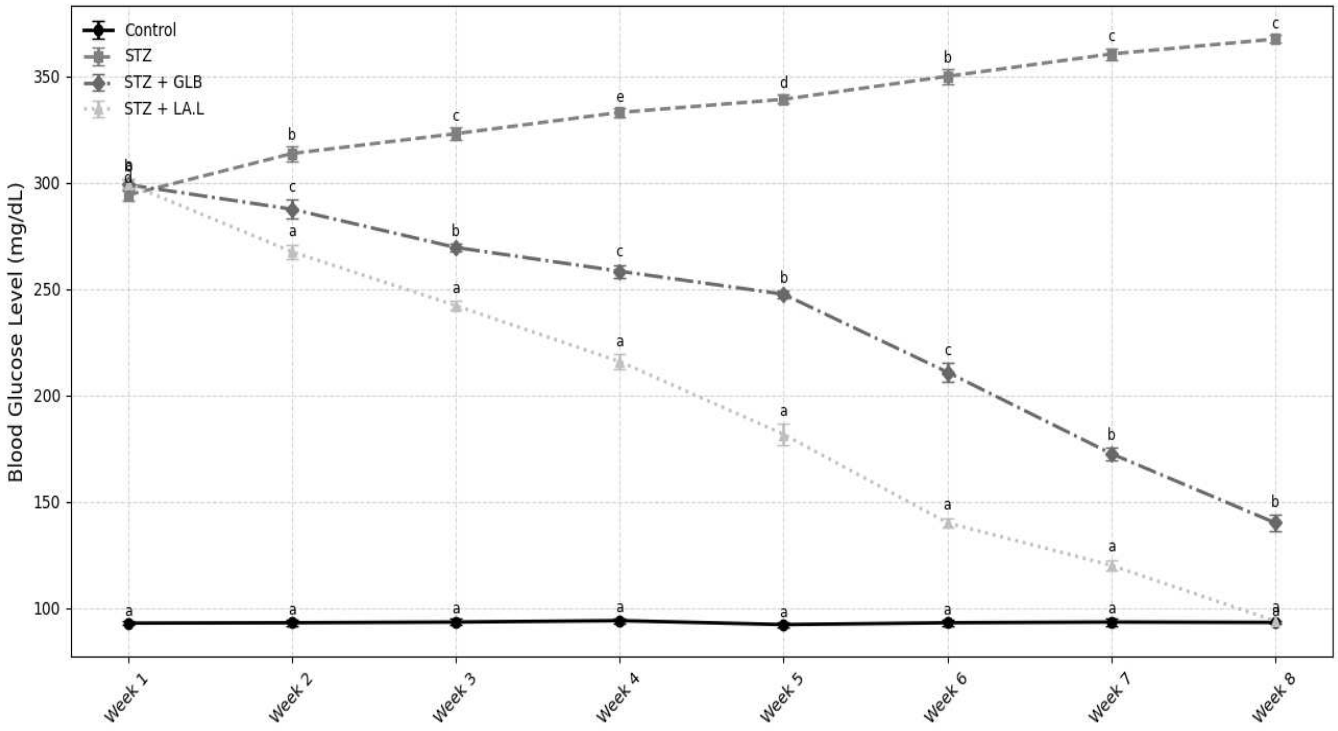



**Figure 5: Effect of PG extract on blood glucose levels (mg/dl) in experimental rats.**
(Based on Tukey's *post hoc* test, alphabets in the same column with the same letters are not significantly different, but those with different letters are significantly different (P<0.05)

### 3.3. Effect of PG extracts on Serum lipid profiles in experimental rats

As diabetes progresses, hyperglycemia and hyperlipidemia become more prevalent. As a biomarker of hyperlipidemia, total cholesterol (TC), triglycerides(TG), low-density lipoprotein(LDL) and very-low-density lipoprotein (VLDL) were significantly increased, and the high density lipo protein (HDL) levels were significantly decreased in STZ group rats in comparison with the control and other groups. The GLB-treated group showed a mild improvement, Whereas, the rats administered with the LA.L extract displayed almost similar values to that of control group. TC, TG, LDL and VLDL were markedly decreased, with

increased HDL level (P<0.05) by statistical comparisons (Figure 6).

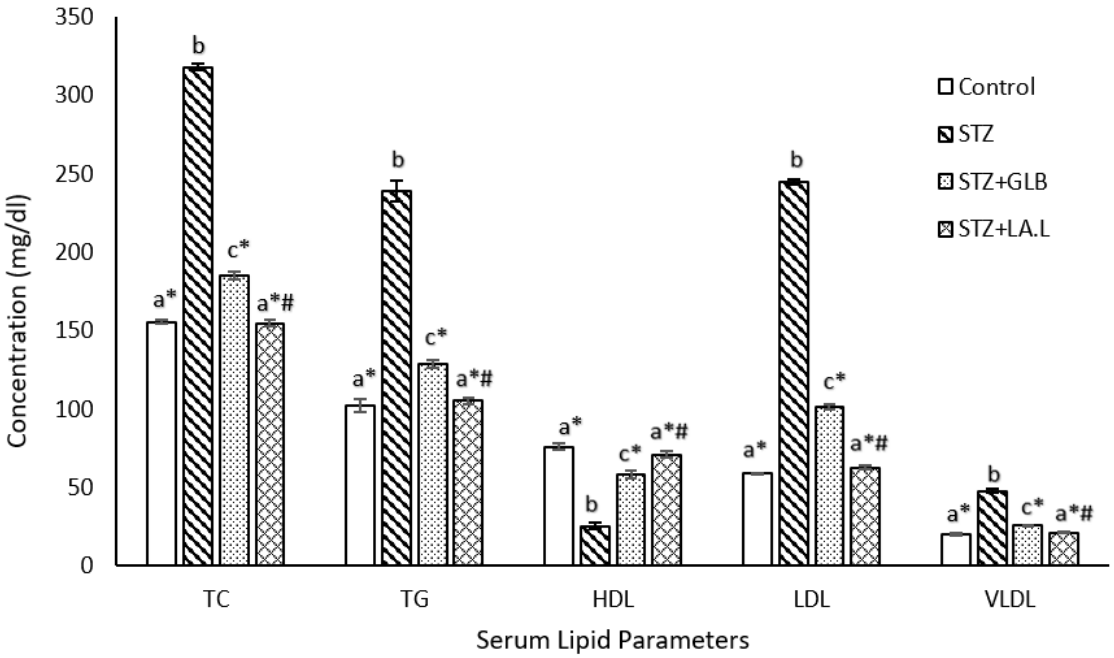



**Figure 6: Effect of PG extract on Serum lipid profiles in diabetes induced male albino rats.** (Alphabets 'a', 'b', and 'c' represent significant differences among experimental groups based on Tukey's *post hoc* test. '*' represents significantly different (P< 0.05) between the experimental groups compared to diabetic induced group. Whereas '#' represents a significant difference between the LA.L group and the GLB-treated group (P<0.05)

### 3.4. Effect of PG extract on Relative organ weight, Total protein, LPO, and antioxidant enzymes in the pancreas of experimental rats.

The relative pancreas weight was significantly reduced in STZ-treated rats, which indicates the adverse effects of chronic hyperglycemia. LA.L extract treatment remarkably increased pancreas weight to nearly the control value compared to the GLB-treated group.

These results suggest that LA.L extract can preserve the integrity of the pancreas (Figure 7 A).

Total protein content and the activity of antioxidant enzymes, SOD, GSH, Gpx and GST were significantly decreased and levels LPO found to be increased, which indicates a high level of oxidative damage in the pancreas tissue of STZ group ($P<0.05$). Moderate restoration was noticed in the GLB-treated group. It is worth noting that administration with LA.L displayed almost normalization of the total protein levels (Figure 7B) and antioxidant enzyme activities (Figure 7D- 7H) with significant reduction in LPO levels (Figure 7C) in experimental rats, which implies that LA.L extract was more effective than GLB in all the parameters, indicating its strong antioxidative property.

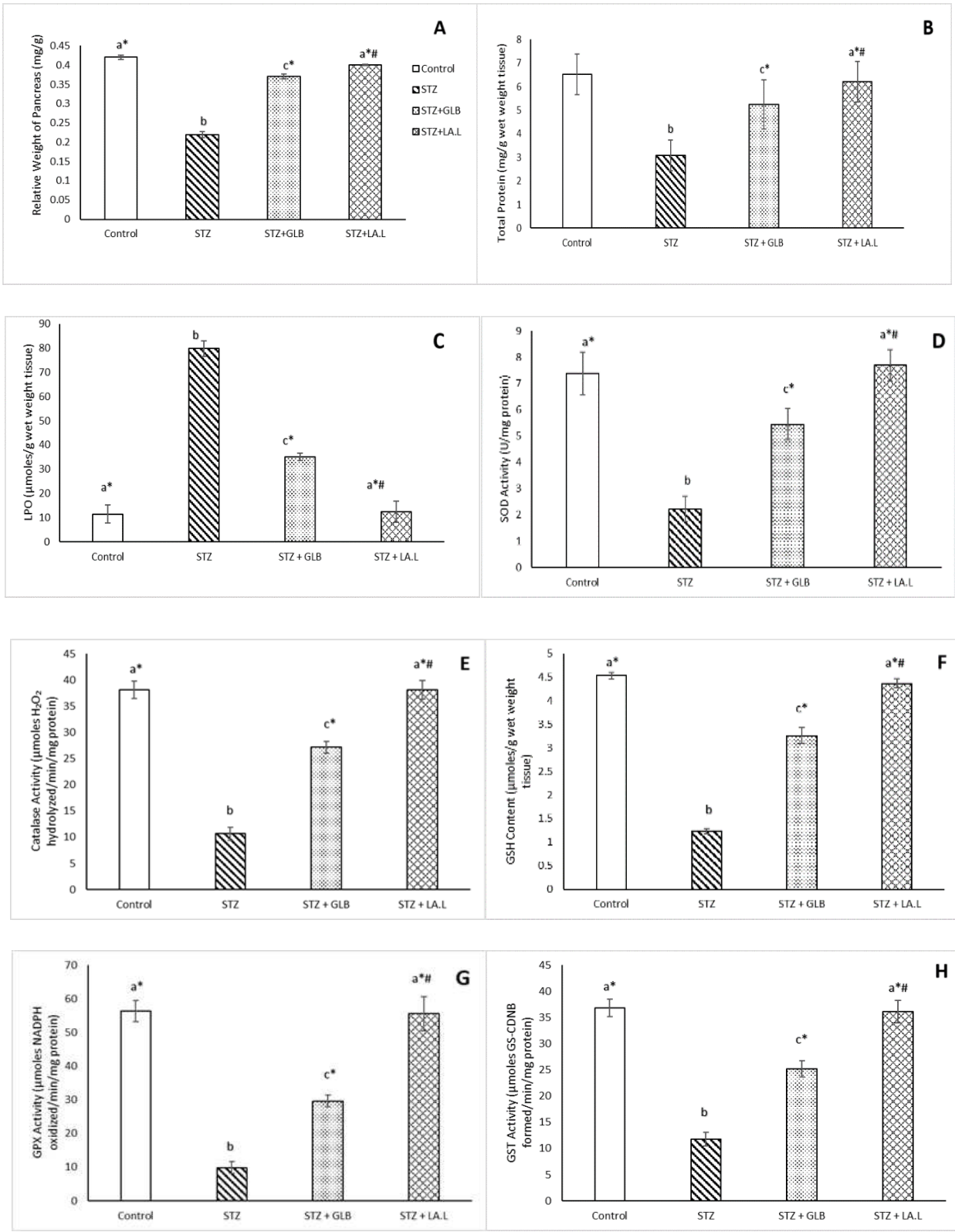



**Figure 7 (A-H): Effect of PG extract on Relative organ weight, Total protein, LPO, and antioxidant status in the pancreas of experimental rats** (Alphabets 'a', 'b', and 'c' represent significant differences among experimental groups based on Tukey's *post hoc* test. '*' represents

significantly different (P< 0.05) between the experimental groups compared to diabetic induced group. Whereas '#' represents a significant difference between the LA.L group and the GLB-treated group (P<0.05)

### 3.5. Effect of PG extract on Histology and Histomorphometric analysis in pancreas of experimental rats.

In Plate 1A (Control), the pancreas tissue is well-organized, and the Islets of Langerhans (IL) and the surrounding acinar cells (AC) are intact, which shows normal histoarchitecture. Plate-1B (STZ) shows extreme degeneration of pancreatic Islet cells (Dg.IL) and distorted acinar cells (D.AC) which is a manifestation of the cytotoxic effect of diabetes. In Plate 1C, GLB group- the acinar cells are loosely arranged and the Islet cells are moderately disorganized, which indicates that GLB has limited protective effects. In Plate-1D LA.L group-, the Islets and acinar cells are fully recovered, which shows that the tissue repair and regeneration are highly accomplished after the administration of LA.L extract.

Figure 8(A,B) illustrates the histo-morphometric analysis of the pancreas. A mild restoration of cell number and diameter were observed in GLB group. No significant difference were noticed in number and diameter of Islets and β-cells between the LA.L-treated and control group. Concurrently, as a result of degenerated Islets of Langerhans, the number and diameter of Islets and β-cells were significantly decreased in STZ group (P<0.05).

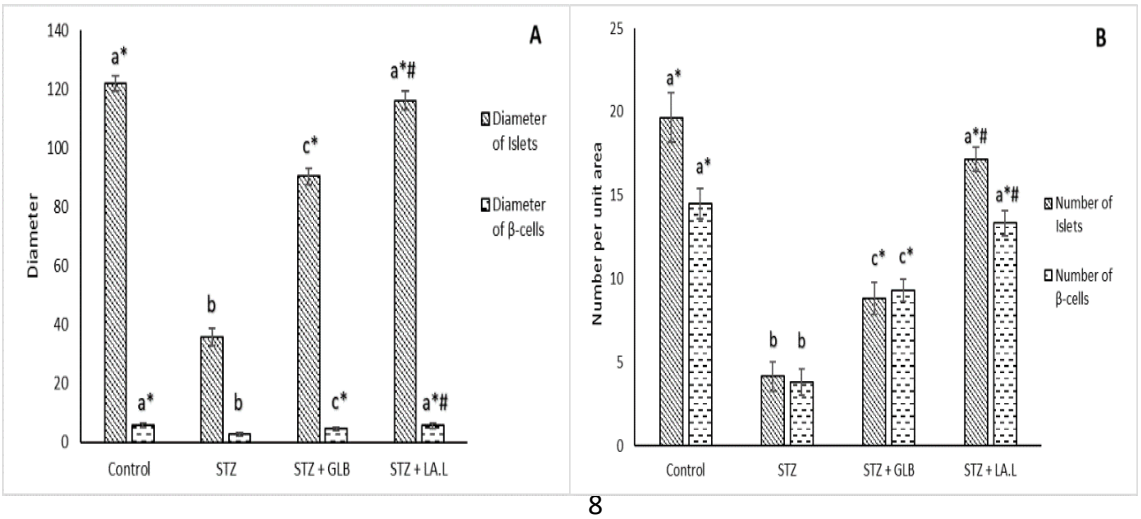


**Figure 8 (A, B). Effect of PG extract on Histomorphometric analysis of pancreas of experimental rats** (Alphabets 'a', 'b', and 'c' represent significant differences among experimental groups based on Tukey's *post hoc* test. '*' represents significantly different (P< 0.05) between the experimental groups compared to diabetic induced group. Whereas '#' represents a significant difference between the LA.L group and the GLB-treated group (P<0.05)

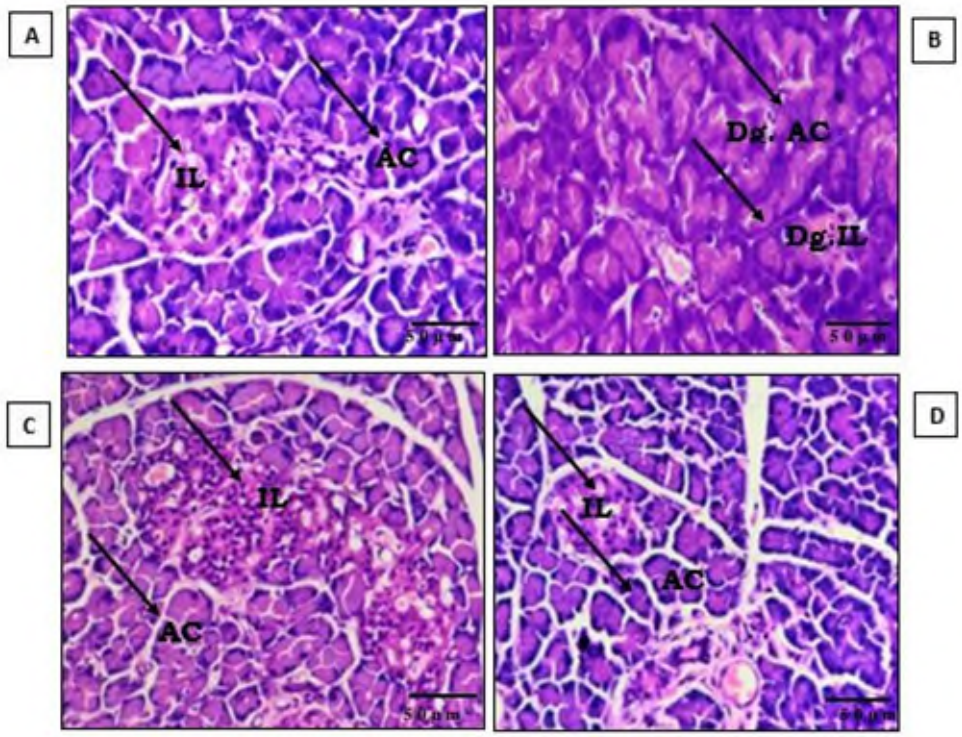



**Plate 1: (A-D) Photomicrographs of T.S of pancreas in experimental rats.**

Note: IL-Islets of Langerhans, AC-Acinar cells, D.AC-Disrupted acinar cells, Dg.IL- Degenerated Islets of Langerhans (Hematoxylin and Eosin staining (400X, scale bar 50µm)

### 3.6. Effect of PG extract on Immunohistochemistry of Caspase-3 and Bcl-2 in the pancreas of experimental rats

Caspase-3(Plate-2 A-D) and Bcl-2 (Plate-3 A-D) in pancreas of control and experimental groups. Immuno-reactivity of Caspase-3 level was significantly elevated in STZ group, which means that there is more apoptosis and there is a significant decrease in the expression of Bcl-2, which implies that, there is impaired cell survival. Partially, these markers were ameliorated by GLB treatment, which reduced Caspase-3 and increased Bcl-2 in comparison with the STZ induced diabetic group. It is important to note that LA.L extract caused a significant decreasein Caspase-3 immunoreaction, almost to the level of control and a significant increase in the expression of Bcl-2.

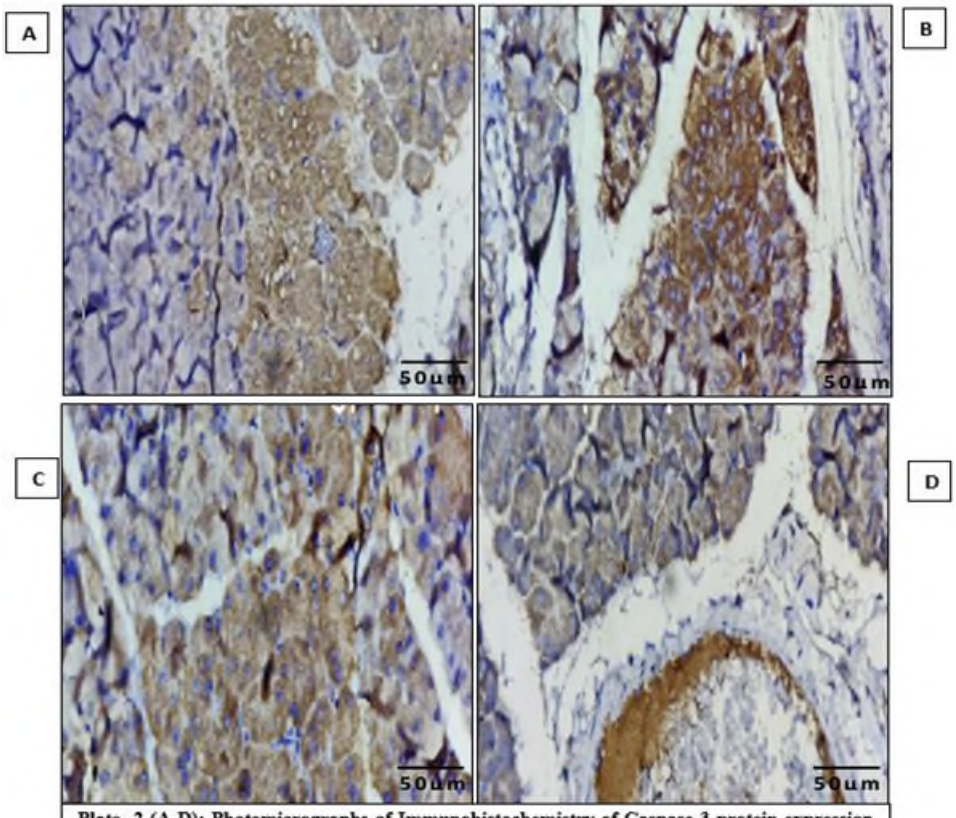



**Plate -2 (A-D): Photomicrographs of Immunohistochemistry of Caspase-3 protein expression in pancreas of control and experimental groups. (Intensity of immunoreaction are measured 0 to10) Scale:** (400X, scale bar 50µm).**A-Control, B- STZ, C STZ+GLB and D-STZ+LA.L**

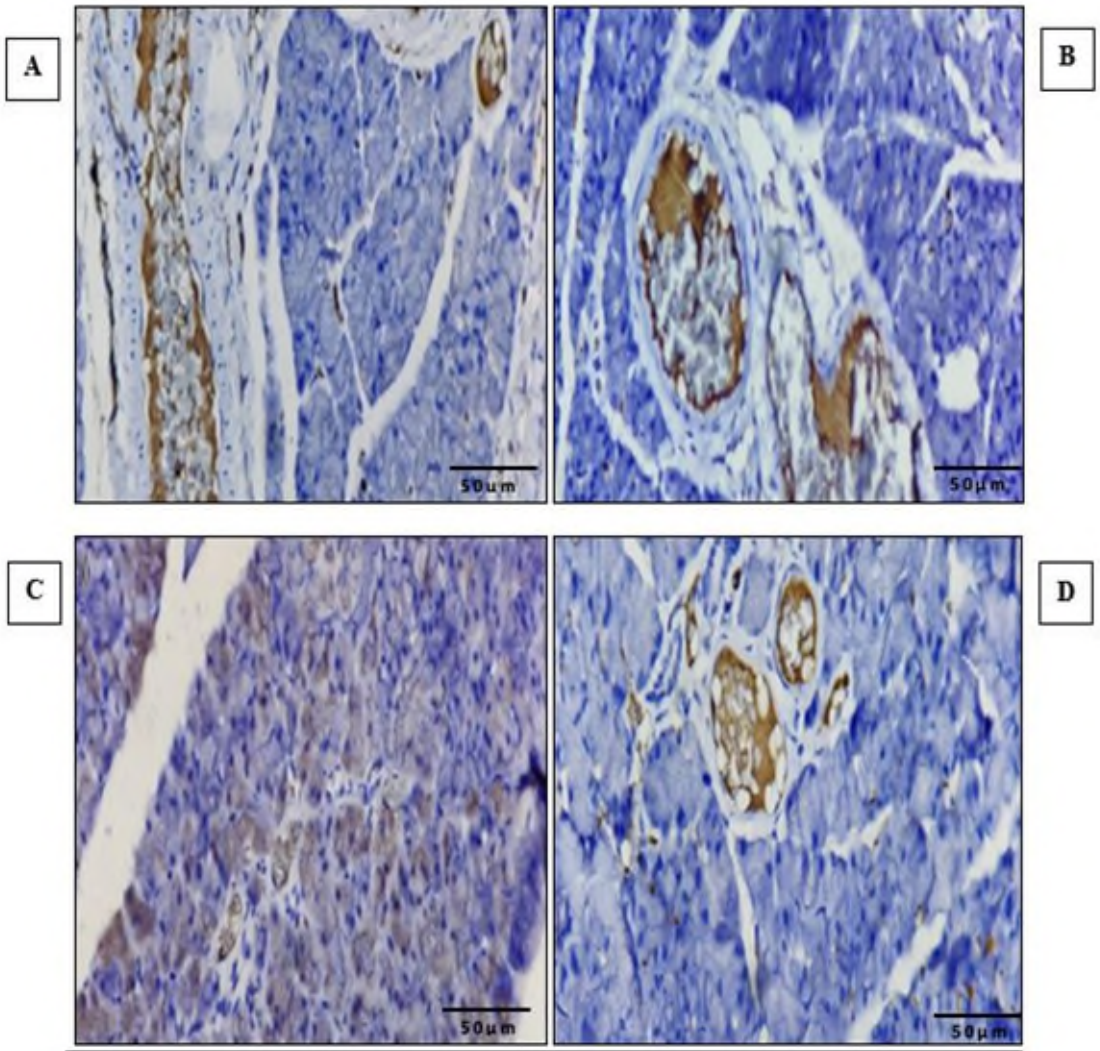



**Plate –3(A-D): Photomicrographs of Immunohistochemistry of Bcl-2 protein expression in pancreas of control and experimental groups. (Intensity of immunoreaction are measured 0 to10)** (400X, scale bar 50µm).**A-Control, B-STZ, C -STZ+GLB and D -STZ+LA.L**

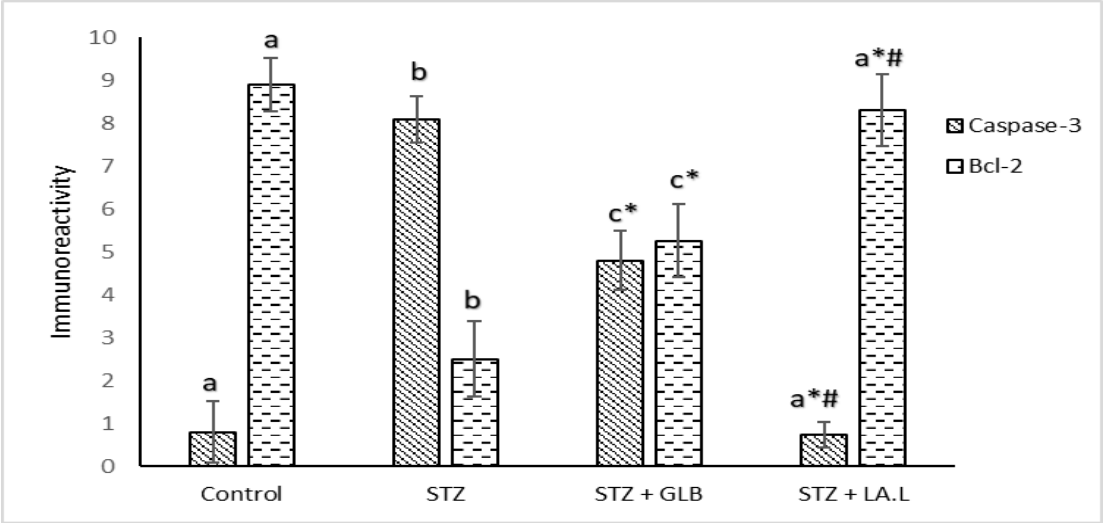



**Figure 9: Effect of PG extract on Immunoreaction of Caspase-3 and Bcl-2 in pancreas of experimental rats** (Alphabets 'a', 'b', and 'c' represent significant differences among experimental groups based on Tukey's *post hoc* test. '*' represents significantly different (P< 0.05) between the experimental groups compared to diabetic induced group. Whereas '#' represents a significant difference between the LA.L group and the GLB-treated group (P<0.05)

### 3.7. Effect of PG extract on gene expression levels of apoptotic (caspase-3) and anti- apoptotic (Bcl-2) proteins in the pancreas of experimental rats by qRT-PCR.

**CASPASE-3**, **Forward primer** :- GGGGAGCTTGGAACGCTAAG

**Reverse primer** :- CCGTACCAGAGCGAGATGAC

**Product length** :- 232 bp

**Bcl-2** , **Forward primer :-** TCTTTGAGTTCGGTGGGGTC

**Reverse primer** :- TAGTTCCACAAAGGCATCCCAG

**Product length :-** 155bp

Figure 10 llustrates, the protein expression of caspase-3 and Bcl-2, in the pancreas of control and experimental rats. STZ induced diabetic group showed elevated expression levels of

Caspase-3. Whereas Bcl-2 protein levels was found to be reduced when compared to control group. The levels of Caspase -3 proteins were significantly down-regulated as well as Bcl-2 protein levels were significantly up-regulated in LA.L (150 mg/kg/ b.w) extract treated group compared to STZ group.

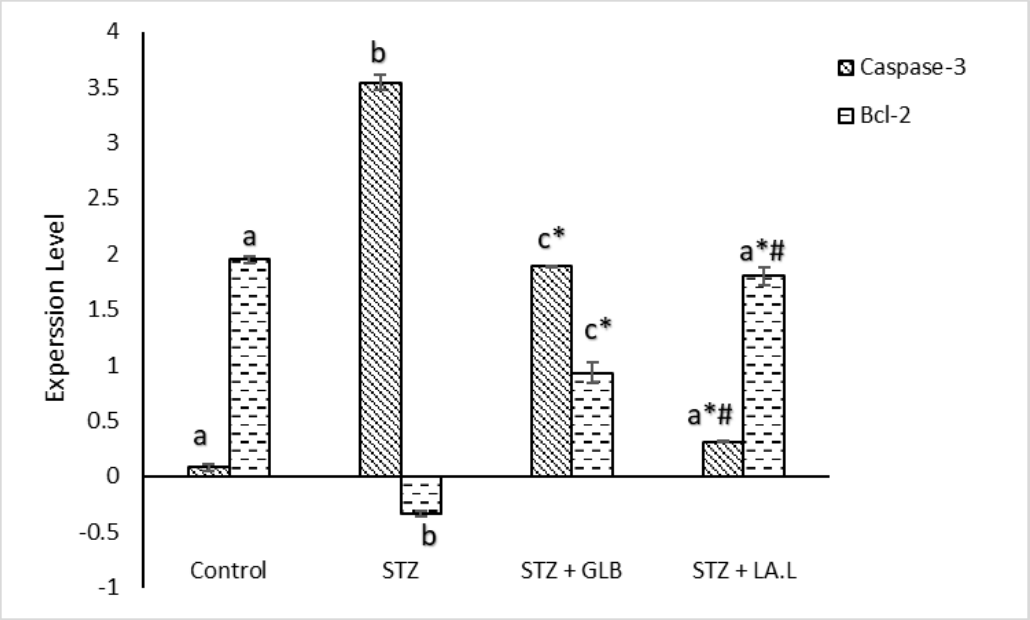



**Figure 10: Effect of PG extract on apoptotic (Caspase-3) and anti-apoptotic (Bcl-2) expression in the pancreas of experimental rats** (Alphabets 'a', 'b', and 'c' represent significant differences among experimental groups based on Tukey's *post hoc* test. '*' represents significantly different (P< 0.05) between the experimental groups compared to diabetic induced group. Whereas '#' represents a significant difference between the LA.L group and the GLB-treated group (P<0.05)

## 4. DISCUSSION

Approximately 70 to 80% of the world's population relies on non-conventional medicine, mostly herbs, as their primary health care (Chan, 2003; El-Sayed and Youssef, 2019). As a promising therapy for the treatment of DM in the foreseeable future, herbal medicine emerged as an effective treatment with negligible side effects (Okur et al., 2017). PG has been extensively studied for its therapeutic effects, and the results showed potent anti-diarrheal,

antihypertensive, antigenotoxic, antiplasmodial, hepatoprotective, antimicrobial, anti-inflammatory, and antimutagenic properties (Panche et al., 2016). The leaves of P. guajava contain enriched mineral elements, such as calcium, potassium, sodium, iron, boron, magnesium, and manganese, which make them an ideal source for human nutrition and as feed for animals in order to prevent micronutrient deficiencies (Adrian et al.,2015). Numerous active phytoconstituents are present in this plant, which include flavonoids, phenolic compounds, tannins, saponins, carotenoids, terpenoids, and triterpenes (Pietta, 2000). Phenols and flavonoids are vital secondary metabolites that play a crucial role in physiological function. In order to scavenge free radicals, it is linked to other hydroxyl group compounds like proteins, lignins, cellulose, and esters (Panche et al., 2016). Antioxidants from this plant can also function at different levels, such as blocking the generation of ROS or regulating the antioxidant defence enzymes. Some enzymatic antioxidant structures, such as copper, zinc, manganese, superoxide dismutase, glutathione peroxidase, glutathione reductase, and catalase, also protect from ROS accumulation and adverse health effects (Oroian et al., 2015). Our previous research demonstrated high phenolic and flavonoid content with a strong antioxidant scavenging capacity in the ethanolic leaf extract of PG (Sowmya and Usha, 2020a). In the present study, LC-MS results revealed various phytocompounds such as Quercetin, Myricetin, flavonoids, kaempferol, Quercetin-glucoside, Gallic acid, Ellagic acid, Quercetin rhamnoside, which are the derivatives of phenols and flavonoids. These compounds are known for their potent bioactive properties, and their combined presence underpins the extract's multifaceted effects on diabetes management.

One of the detrimental factors associated with the onset of DM is dysfunction of β-cell of the pancreas. As a consequence, we examined the anti-diabetic, anti-hyperlipidemic,

and anti-apoptotic effects of LA.L extract using albino rats as an experimental model. In the current study, STZ-induced diabetic rats showed increased food and water consumption with elevated blood glucose levels and significant weight loss. Such changes may be due to insufficient insulin secretion and reduced ATP levels, and imbalances in the synthesis of protein. As soon as blood glucose levels surpass the renal threshold levels, glucose will be excreted in urine, water escort glucose owing to the osmotic effect, and to recompense this water loss in the body, the thirst centre gets activated, and excess water will be consumed (Murray et al., 2003). On physiological premises, the decline in blood glucose fixation causes hunger, which has prompted the glucostatic hypothesis of cravings and feeding regulations. The satiety community is sensitive to the arterio-venous angle of blood glucose level, so elevated arteriovenous blood glucose inclination invigorates the satiety center and hinders the feeding centre and focus instigating anorexia. In diabetes, the blood glucose level will be elevated and polyphagia will be expanded and the fact that the arterio venous slope is low as the cells cannot utilize the glucose because of the absence of insulin secretion (Hall, 2011). In this study, administration with LA.L extract (150mg/kg/b.w) normalized the excess food and water consumption and decreased the blood glucose level in diabetic rats and these changes might be due to the nutrients present in the PG leaf extract, which could have improved the insulin secreting capacity of β-cells and also due to the synergistic action of polyphenolic compounds present in it. Our findings are consistent with the earlier findings of *C. americanum* leaves lipid fraction shows significant reduction of blood glucose levels in diabetes-induced rats (Araujo et al., 2024). Thus, PG can be a defensive part in forestalling the imbalances caused in the diabetic model by subsequently working on the glycemic control.

Aggravations in glucose digestion, modified lipid levels, and oxidative stress are significant danger variables of diabetes. Hyperglycemia with dyslipidemia, a condition which is characterized by an elevation in TG, TC, LDL, VLDL, and a fall in HDL, is a hazardous factor in the body system. It has been suggested that a deficiency in insulin leads to a variety of de-arrangements in regulatory metabolic processes, thus leading to amassment of lipids (Goldberg,1981). In the current study, the STZ group showed an enormous imbalance in serum lipid profiles, elevated TC, TG, LDL, VLDL, and reduced HDL levels in contrast to control and other experimental groups. Increased fatty substances and diminished HDL cholesterol levels are the vital qualities of dyslipidemia in T2-DM. (Strikic et al., 2023). Supplementation with LA.L extract reversed the trend by a marked decrease in the levels of TC, TG, LDL, and VLDL with an increase in HDL level. LA.L extract might have prevented the metabolic abnormality and subsequently re-established the typical digestion by shifting the equilibrium from high lipids to high carb turnover. Present results are in agreement with reports of (Srivastava et al., 2020).

Destruction of β-cells and production of free radicals can be identified by determining LPO levels (Gerber and Rutter, 2017). The present investigation showed increased LPO in the pancreas with concomitant reduced levels of total protein content and organ weight in the STZ group, and this might be due to glucose autoxidation, elevated LPO associated with insulin inadequacy leading to elevated protein catabolism with reduced protein synthesis (Latha and Daisy, 2010). Sustained hyperglycemia elevates ROS levels in the pancreas, in turn causing oxidative stress and leading to a reduction in organ weight, protein content, and an increase in LPO levels (Sowmya and Usha, 2020b). However, the LA.L supplemented group played an effective role in normalizing the pancreas weight, increasing total protein, and also reducing

the elevated LPO levels compared to GLB and STZ treated groups. This could be due to the free radical quenching capacity of LA.L extract. Further, the production of antioxidant defense system, namely, SOD, CAT, GSH, GPx, and GST levels, showed impairment in the pancreas of the STZ group compared to the control group. Superoxide radicals are catalyzed by SOD into hydrogen peroxide and atomic oxygen, which are harmful to tissues by causing free radical damage. The noticed reduction in SOD action in STZ group might be due to the inactivation of $H_2O_2$ or by enzyme glycations. Hemeprotein, CAT is a catalyst for hydrogen peroxide degradation and protects cells from reactive hydroxyl radicals. It may be inhibited by glycation, causing its action to decrease. By suppressing hydrogen peroxide generated by dismutation, CAT also protects peroxisomes from oxidative damage caused by hydrogen peroxide. The decreased levels of CAT in STZ group could resulted from amassing of $H_2O_2$, which causes injurious outcomes (Jomova et al., 2024). Typically, GSH maintains cell integrity and functions by balancing redox homeostasis, eliminating free radicals, and detoxifying cells. In addition to scavenging free radicals, it also serves as a substrate for glutathione peroxidase to detoxify peroxides. In this study, declined levels of GSH in pancreas was noted in the STZ group and this could be owing to diminished combination or expanded debasement of GSH by oxidative pressure in diabetes. By oxidizing reduced GSH, GPx forms $H_2O_2$ through the detoxification of $H_2O_2$. Diminished movement of GPx may be attributed to inactivation and glycation induced by extremists. GST, together with GSH, in disintegrating hydrogen peroxide and other natural hydroperoxides to nontoxic items, leads to a detrimental decrease in GSH content (Jena et al., 2023). The STZ group showed a reduction in GST levels, and this may be due to the inactivation of the catalyst by ROS. A moderate increase in SOD, CAT, GSH, GpX, and GST levels were observed in GLB treated group. While the LA.L supplemented group showed a marked elevation in all the antioxidant parameters. This might be owing to the presence of

more phenols and flavonoid contents present in the LA.L extract, as these compounds are associated with the mending system of free extremist intervened sicknesses, including diabetes. The active compounds present in the LA.L leaves might have helped in anti-hyperglycemia with restoration of enzymatic alterations. The present results correlate with the previous reports of Banerjee et al. (2020) and Jayachandran et al. (2018)

Approximately β-cells, which constitute 65-80% of Islet cells, which secrete insulin, and these cells are sensitive to glucose levels in the blood (Iida and Watada, 2025). As a result of diabetes, the Islets of Langerhans can undergo quantitative changes, such as reduction of size and number, and qualitative changes, such as necrosis and degeneration. The present result revealed degeneration of islet cells, deterioration of acinar cells, disruption of interlobular connective tissue in STZ treated group, and this may be owing to the detrimental effect on β-cells and also toxicity induced by STZ, wherein cells might have experienced degenerative changes (Szkudelski, 2001). Supplementation with GLB showed loosely arranged degenerative acinar cells and disorganized islet cells. Administration with LA.L extract showed regeneration and restoration of Islets of Langerhans and acinar cells similar to the control group, and thus LA.L has proven to be beneficial in promoting regeneration of pancreatic Islets. The presence of various phytoconstituents in the LA.L extract might have contributed to the regeneration of islet cells. Our results are consistent with the earlier reports that *D. excelsa* extract caused a reduced blood glucose concentration with an increase in the number of Islets in the pancreas of alloxan-induced diabetic animals (Matusin et al., 2021). The reduction in β-cell count can be as low as 50% during diabetes. STZ group resulted in a significant decrease in the number of Islet cells (N/10mm$^2$), the number of β-cells (N/1000μm$^2$), and also a decrease in the diameter (μm) of Islet cells and β-cells. While

moderate elevation in diameter, number of Islets and β-cells were noticed in GLB groups. LA.L extract-treated group showed a marked elevation in the number and diameter of β-cells and Islet cells. Thus the normalcy of the Islets and β-cells might be owing to the improvement in the secretion of insulin by recovered β-cells. Current result reveals, LA.L extract might have acted upon Islet cell function, by elevating the number of Islets upon stimulating release of insulin, thereby potentiating the existing β-cells of the Islets of Langerhans in diabetic rats.

Apoptosis is massively associated with differentiation, proliferation, and the capability of the immune system in the removal of defects and harmful cells (Gewies, 2003). Numerous apoptotic signaling pathways, including caspase-3 activation pathways, have been demonstrated to be activated by oxidative stress and hyperlipidemia (Korkmaz-Icoz et al.,2016). Caspases are cysteine-aspartyl-specific proteases that play a crucial role in apoptosis (Creagh et al., 2003). The most characteristic anti-apoptotic protein is Bcl-2, which inhibits the secretion of cytochrome c from mitochondria, thereby preventing apoptosis (Jafri et al., 2019). In the present study, immunohistochemistry showed lower Bcl-2 protein expression and higher Caspase-3 expression in the STZ group, which could be due to STZ-induced toxicity. Further, the LA.L extract-treated group significantly increased Bcl-2 immunoreactivity and decreased caspase-3 in the pancreas of diabetic rats. One of the studies reported that treatment with Genistein for two months showed an up-regulation in Bcl-2 and down-regulation of caspase-3 in diabetic rats, which was similar to the results of our investigation (Yousefi et al., 2017). The outcome of our results might be due to the enriched phytoconstituents present in the LA.L extract, which would have counteracted the oxidative stress, in turn protecting the pancreatic β-cells from apoptosis.

Corresponding results of qRT-PCR revealed a drastic down-regulation of anti-apoptotic Bcl-2 and up-regulation of pro-apoptotic caspase-3 in the STZ group. It has been shown that expression of anti-apoptotic proteins might have prevented the β-cell damage caused by STZ (Lee et al., 2017). In the current study, supplementation with GLB showed a moderate up-regulation of Bcl-2 and down-regulation of caspase-3. Further, administration with LA.L extract showed up-regulation in anti-apoptotic Bcl-2 protein expression and inhibited the caspase-3 protein expression in pancreatic tissue of STZ-treated diabetic rats. Present results are in line with the previous study of Khamchan et al. (2018). The anti-apoptotic effects of PG extract may be attributed to its therapeutic efficacy, which has been observed in this study, and this might be owing to the bioactive phytocompounds of flavonoids and phenols present in it.

## 5. CONCLUSION:

This study demonstrates the efficacy of the ethanolic leaf extract of PG on the pancreas of diabetic albino rats. The LA.L extract showed significant anti-hyperglycemic and anti-hyperlipidemic activities. The results also indicated that LA.L mitigated the oxidative stress by enhancing the activity of antioxidant enzymes. Histological examination of the pancreas showed restoration of the Islets of Langerhans and acinar cells. The LA.L extract appears to improve islet cell function by increasing the number of islets and stimulating insulin secretion, thereby enhancing β-cell activity in diabetic rats. Further, the Immuno-histochemical analysis revealed a marked increase in Bcl-2 expression, an anti-apoptotic protein, and a decrease in caspase-3 activity, which is associated with apoptosis. Our investigation also emphasized the gene expression analysis by RT-qPCR, which showed that the LA.L extract effectively modulated apoptosis-related genes, with significant upregulation

of anti-apoptotic gene Bcl-2 and downregulation of the pro-apoptotic gene caspase-3. The ameliorative efficacy of LA.L extract might have arisen from synergistic interactions among the bioactive constituents present in it, which offer a broader and holistic therapeutic potential in managing type-2 diabetes.

**LIST OF ABBREVATIONS**

Psidium gujava L. –PG
Diabetes mellitus - DM
Type-2 Diabetes mellitus - T2-DM
Lalit leaf - LA.L
Glibenclamide - GLB
Liquid chromatography-mass spectrometry - LC-MS
Streptozotocin - STZ
Quantitative reverse transcription- polymerase chain reaction - qRT-PCR
Reactive oxygen species – ROS
Total cholesterol - TC
Triglycerides- TG
Low density lipo protein – LDL
High-density lipoprotein - HDL
Lipid peroxidation - LPO
Superoxide dismutase - SOD
Catalase - CAT
Gluatathione - GSH
Glutathione transferase - GST
Glutathione peroxidase – GPx
Islets of Langerhans - IL
Acinar cells - AC
Disrupted acinar cells – D.AC
Degenerated Islets of Langerhans – Dg.IL

## Author's contribution

**Sowmya BH**- Conceptualization, Methodology, Investigation, Formal analysis, Writing-review & editing original draft.

**Usha Anandhi D**- Conceptualization, Supervision, Review & editing original draft.

## Ethics approval and Consent to participate

The Institutional Animal Ethics Committee of Bangalore University, Bengaluru, approved the proposed animal experiments (NO: 402/GO/Re/S/01/CPCSEA). The committee's ethical standards were followed for the purpose of controlling and supervising animal experiments.

## Competing interests

The authors declare no competing interests.


## Funding source

No funding has been received for this experiment.


## Consent for publication

Not applicable.


## Acknowledgements

Not applicable


## Data availability statement

Data can be available on reasonable request from the corresponding author.

## 6. REFERRRENCES


[1]. Siam, N. H., Snigdha, N. N., Tabasumma, N., & Parvin, I. (2024). Diabetes mellitus and cardiovascular disease: exploring epidemiology, pathophysiology, and treatment strategies. *Reviews in Cardiovascular Medicine*, *25*(12), 436. https://doi.org/10.31083/j.rcm2512436

[2]. Genitsaridi, I., Salpea, P., Salim, A., Sajjadi, S. F., Tomic, D., James, S., ... & Magliano, D. J. (2026). 11th edition of IDF Diabetes Atlas: global, regional, and national diabetes prevalence estimates for 2024 and projections for 2050. *The Lancet Diabetes & Endocrinology*, *14*(2), 149-156.

[3]. Wagh, K., Kirpich, A., & Chowell, G. (2025). The Future Diabetes Mortality: Challenges in Meeting the 2030 Sustainable Development Goal of Reducing Premature Mortality from Diabetes. *Journal of Clinical Medicine*, *14*(10), 3364. https://doi.org/10.3390/jcm14103364

[4]. Nellaiappan, K., Preeti, K., Khatri, D. K., & Singh, S. B. (2022). Diabetic complications: an update on pathobiology and therapeutic strategies. *Current diabetes reviews*, *18*(1), 31-44. https://doi.org/10.2174/1573399817666210309104203

[5]. Galicia-Garcia, U., Benito-Vicente, A., Jebari, S., Larrea-Sebal, A., Siddiqi, H., Uribe, K. B., ... & Martín, C. (2020). Pathophysiology of type 2 diabetes mellitus. *International journal of molecular sciences*, *21*(17), 6275. https://doi.org/10.3390/ijms21176275

[6]. Chen, X., Xie, N., Feng, L., Huang, Y., Wu, Y., Zhu, H., ... & Zhang, Y. (2025). Oxidative stress in diabetes mellitus and its complications: From pathophysiology to therapeutic strategies. *Chinese Medical Journal*, *138*(1), 15-27. https://doi.org/10.1097/CM9.0000000000003230

[7]. Afzal, S., Abdul Manap, A. S., Attiq, A., Albokhadaim, I., Kandeel, M., & Alhojaily, S. M. (2023). From imbalance to impairment: the central role of reactive oxygen species in oxidative stress-induced disorders and therapeutic exploration. *Frontiers in pharmacology*, *14*, 1269581. https://doi.org/10.3389/fphar.2023.1269581

[8]. Chandirasegaran, G., Elanchezhiyan, C., Ghosh, K., & Sethupathy, S. (2017). Berberine chloride ameliorates oxidative stress, inflammation and apoptosis in the

pancreas of Streptozotocin induced diabetic rats. *Biomedicine & Pharmacotherapy*, *95*, 175-185. https://doi.org/10.1016/j.biopha.2017.08.040

[9]. Mohan, V., Wangnoo, S., Das, S., Dhediya, R., & Gaurav, K. (2022). Comparison of gliclazide vs linagliptin on hypoglycemia and cardiovascular events in type 2 diabetes mellitus: A systematic review. *World Journal of Diabetes*, *13*(12), 1168. https://doi.org/10.4239/wjd.v13.i12.1168

[10]. Zhou, J., Kang, X., Luo, Y., Yuan, Y., Wu, Y., Wang, M., & Liu, D. (2019). Glibenclamide-induced autophagy inhibits its insulin secretion-improving function in β cells. *International journal of endocrinology*, *2019*(1), 1265175. https://doi.org/10.1155/2019/1265175

[11]. Madhuri, K., & Naik, P. R. (2017). Ameliorative effect of borneol, a natural bicyclic monoterpene against hyperglycemia, hyperlipidemia and oxidative stress in streptozotocin-induced diabetic Wistar rats. *Biomedicine & Pharmacotherapy*, *96*, 336-347. https://doi.org/10.1016/j.biopha.2017.09.122

[12]. Simpson, S. H., Majumdar, S. R., Tsuyuki, R. T., Eurich, D. T., & Johnson, J. A. (2006). Dose–response relation between sulfonylurea drugs and mortality in type 2 diabetes mellitus: a population-based cohort study. *Cmaj*, *174*(2), 169-174. https://doi.org/10.1503/cmaj.050748

[13]. Nolan CJ, Prentki M. Insulin resistance and insulin hypersecretion in the metabolic syndrome and type 2 diabetes: Time for a conceptual framework shift. Diab Vasc Dis Res. 2019 Mar;16(2):118-127. https://doi.org/10.1177/1479164119827611

[14]. Patwardhan, B., Warude, D., Pushpangadan, P., & Bhatt, N. (2005). Ayurveda and traditional Chinese medicine: a comparative overview. *Evidence-Based Complementary and Alternative Medicine*, *2*(4), 465-473. https://doi.org/10.1093/ecam/neh140

[15]. Farnsworth, N. R., Akerele, O., Bingel, A. S., Soejarto, D. D., & Guo, Z. (1985). Medicinal plants in therapy. *Bulletin of the world health organization*, *63*(6), 965. https://pubmed.ncbi.nlm.nih.gov/3879679/

[16]. Pulivarthi, V., Josthna, P., & Naidu, C. V. (2021). Ameliorative effect of Annona reticulata L. leaf extract on antihyperglycemic activity and its hepato-renal protective potential in streptozotocin induced diabetic rats. *Journal of Ayurveda and Integrative Medicine*, *12*(3), 415-426. https://doi.org/10.1016/j.jaim.2021.01.010

[17]. Vignesh, A., Sivalingam, R., Selvakumar, S., & Vasanth, K. (2022). A review on ethnomedicinal and phytopharmacological potential of traditionally wild and endemic plant Berberis tinctoria Lesch. *The Thai Journal of Pharmaceutical Sciences*, *46*(2), 137-148. https://doi.org/10.56808/3027-7922.2554

[18]. Muscolo, A., Mariateresa, O., Giulio, T., & Mariateresa, R. (2024). Oxidative stress: the role of antioxidant phytochemicals in the prevention and treatment of diseases. *International journal of molecular sciences*, *25*(6), 3264. https://doi.org/10.3390/ijms25063264

[19]. Joseph, B., & Priya, M. (2011). Review on nutritional, medicinal and pharmacological properties of guava (Psidium guajava Linn.). *International Journal of pharma and bio sciences*, *2*(1), 53-69.

[20]. Thaipong, K., Boonprakob, U., Crosby, K., Cisneros-Zevallos, L., & Byrne, D. H. (2006). Comparison of ABTS, DPPH, FRAP, and ORAC assays for estimating antioxidant activity from guava fruit extracts. *Journal of food composition and analysis*, *19*(6-7), 669-675.

https://doi.org/10.1016/j.jfca.2006.01.003

[21]. Conway, P. (2002). Tree medicine a comprehensive guide to the healing power of over 170 trees.

[22]. Sriwilaijaroen, N., Fukumoto, S., Kumagai, K., Hiramatsu, H., Odagiri, T., Tashiro, M., & Suzuki, Y. (2012). Antiviral effects of Psidium guajava Linn.(guava) tea on the growth of clinical isolated H1N1 viruses: Its role in viral hemagglutination and neuraminidase inhibition. *Antiviral research*, *94*(2), 139-146. https://doi.org/10.1016/j.antiviral.2012.02.013

[23]. Lee, D. U., Weon, K. Y., Nam, D. Y., Nam, J. H., & Kim, W. K. (2016). Skin protective effect of guava leaves against UV-induced melanogenesis via inhibition of ORAI1 channel and tyrosinase activity. *Experimental Dermatology*, *25*(12), 977-982. https://doi.org/10.1111/exd.13151

[24]. Rahman, M. H., Asrafuzzaman, M., Tusher, M. M. H., Mosihuzzaman, M., Khan, M. S. H., Shoeb, M., & Rokeya, B. (2023). Elucidation of anti-hyperglycemic activity of Psidium guajava L. leaves extract on streptozotocin induced neonatal diabetic Long-Evans rats. *Journal of Ayurveda and Integrative Medicine*, *14*(5), 100776. https://doi.org/10.1016/j.jaim.2023.100776

[25]. Pai, T.V., Sawant, S.Y., Ghatak, A.A. *et al.* Characterization of Indian beers: chemical composition and antioxidant potential. *J Food Sci Technol* **52**, 1414–1423 (2015). https://doi.org/10.1007/s13197-013-1152-2

[26]. Silambujanaki, P., Chitra, V., Soni, D., Raju, D., & Sankari, M. (2009). Hypoglycemic activity of Stachytarpheta indica on streptozotocin induced wistar strain rats. *International Journal of PharmTech Research*, *1*(4), 1564-1567.

[27]. Siddiqui, O., Sun, Y., Liu, J. C., & Chien, Y. W. (1987). Facilitated transdermal transport of insulin. *Journal of Pharmaceutical Sciences*, *76*(4), 341-345.

https://doi.org/10.1002/jps.2600760416

[28]. Chauhan, K., Sharma, S., Rohatgai, K., & Chauhan, B. (2012). Antihyperlipidemic and antioxidative efficacy of Catharanthus roseus Linn [Sadabahar] in

streptozotocin induced diabetic rats. *Asian Journal of Pharmaceutical and Health Sciences*, *2*(1).

[29]. Ebokaiwe, A. P., Ijomone, O. M., Osawe, S. O., Chukwu, C. J., Ejike, C. E., Zhang, G., & Wang, F. (2018). Alteration in sperm characteristics, endocrine balance and redox status in rats rendered diabetic by streptozotocin treatment: attenuating role of Loranthus micranthus. *Redox Report*, *23*(1), 194-205. https://doi.org/10.1080/13510002.2018.1540675

[30]. Fluttert, M., Dalm, S., & Oitzl, M. S. (2000). A refined method for sequential blood sampling by tail incision in rats. *Laboratory animals*, *34*(4), 372-378. https://doi.org/10.1258/002367700780387714

[31]. Lowry, O. H., Rosebrough, N. J., Farr, A. L., & Randall, R. J. (1951). Protein measurement with the Folin phenol reagent. Journal of Biological Chemistry, 193(1), 265-275.

[32]. Niehaus, W. G., & Samuelsson, B. (1968). Formation of malonaldehyde from phospholipid arachidonate during microsomal lipid peroxidation. The FEBS Journal, 6(1), 126-130 https://doi.org/10.1111/j.1432-1033.1968.tb00428.x

[33]. Misra, H. P., & Fridovich, I. (1972). The role of superoxide anion in the autoxidation of epinephrine and a simple assay for superoxide dismutase. Journal of Biological Chemistry, 247(10), 3170-3175. https://doi.org/10.1016/S0021-9258(19)45228-9

[34]. Aebi, H. (1984). [13] Catalase in vitro. In *Methods in enzymology* (Vol. 105, pp. 121-126). Academic press. https://doi.org/10.1016/S0076-6879(84)05016-3

[35]. Lawrence, R. A., & Burk, R. F. (1976). Glutathione peroxidase activity in selenium deficient rat liver. Biochemical and Biophysical Research Communications, 71(4), 952-958. https://doi.org/10.1016/0006-291X(76)90747-6

[36]. Habig, W. H., Pabst, M. J., & Jakoby, W. B. (1974). Glutathione S-transferases the first enzymatic step in mercapturic acid formation. Journal of Biological Chemistry, 249(22), 7130-7139. https://doi.org/10.1016/S0021-9258(19)42083-8

[37]. Ellman, G. L. (1959). Tissue sulfahydryl groups. Archives of Biochemistry and Biophysics, 82, 70-77

[38]. Vijayavel, T., & Aswath, N. (2013). Correlation between histological grading and ploidy status in potentially malignant disorders of the oral mucosa: A flow cytometric analysis. *Journal of oral and maxillofacial pathology: JOMFP*, *17*(2), 169.

https://doi.org/10.4103/0973-029X.119747

[39]. Chan, K. (2003). Some aspects of toxic contaminants in herbal medicines. Chemosphere 52, 1361–1371 https://doi.org/10.1016/S0045-6535(03)00471-5

[40]. El-Sayed, S. M., & Youssef, A. M. (2019). Potential application of herbs and spices and their effects in functional dairy products. *Heliyon*, *5*(6). https://doi.org/10.1016/j.heliyon.2019.e01989

[41]. Okur, M. E., Karantas, I. D., & Siafaka, P. I. (2017). Diabetes Mellitus: a review on pathophysiology, current status of oral pathophysiology, current status of oral medications and future perspectives. *Acta Pharmaceutica Sciencia*, *55*(1). https://doi.org/10.23893/1307-2080.APS.0555

[42]. Panche, A. N., Diwan, A. D., & Chandra, S. R. (2016). Flavonoids: an overview. *Journal of Nutritional Science*, *5*, e47. https://doi.org/10.1017/jns.2016.41

[43]. Adrian, J. A. L., Arancon, N. Q., Mathews, B. W., & Carpenter, J. R. (2015). Mineral composition and soil-plant relationships for common guava (Psidium guajava L.) and yellow strawberry guava (Psidium cattleianum var. Lucidum) tree parts and fruits. *Communications in Soil Science and Plant Analysis*, *46*(15), 1960-1979. https://doi.org/10.1080/00103624.2015.1069310

[44]. Pietta, P. G. (2000). Flavonoids as antioxidants. *Journal of natural products*, *63*(7), 1035-1042. https://doi.org/10.1021/np9904509

[45]. Oroian, M., & Escriche, I. (2015). Antioxidants: Characterization, natural sources, extraction and analysis. *Food Research International*, *74*, 10-36. https://doi.org/10.1016/j.foodres.2015.04.018

[46]. Sowmya, B. H., & Usha A.D. (2020 a). Quantification of Total Phenolics, Flavonoids and Evaluation of in vitro Free Radical Scavenging Activities in Psidium guajava L. *Indian Journal of Pharmaceutical Sciences*, *82*(4). https://doi.org/10.36468/pharmaceutical-sciences.683

[47]. Murray, H. D., Schneider, D. A., & Gourse, R. L. (2003). Control of rRNA expression by small molecules is dynamic and nonredundant. *Molecular cell*, *12*(1), 125-134. https://doi.org/10.1016/S1097-2765(03)00266-1

[48]. Hall, J. E. (2011). Dietary Balances; Regulation of Feeding; Obesity and Starvation; Vitamins and Minerals. U: Hall JE, ur. Guyton and Hall Textbook of Medical Physiology, 12. izdanje.

[49]. Araujo, R. L., de Pinho, C. L. C., Farias, F. O., da Silva Zanzarini, I., Moure, V. R., Valdameri, G., ... & Mafra, M. R. (2024). Anti-diabetic, antiglycant,

antioxidant, and anticancer activities of Crinum americanum L. leaves lipid fraction. *Industrial Crops and Products*, *209*, 118012. https://doi.org/10.1016/j.indcrop.2023.1180

[50]. Goldberg, R. B. (1981). Lipid disorders in diabetes. *Diabetes care*, *4*(5), 561-572. https://doi.org/10.2337/diacare.4.5.561

[51]. Strikic, D., Vujevic, A., Perica, D., Leskovar, D., Paponja, K., Pećin, I., & Merćep, I. (2023). Importance of Dyslipidaemia Treatment in Individuals with Type 2 Diabetes Mellitus—A Narrative Review. *Diabetology*, *4*(4), 538-552. https://doi.org/10.3390/diabetology4040048

[52]. Srivastava, A. K., Mukerjee, A., & Tripathi, A. (2020). Antidiabetic and antihyperlipidemic activities of Cucumis melo var. momordica fruit extract on experimental animals. *Future Journal of Pharmaceutical Sciences*, *6*(1), 1-9 https://doi.org/10.1186/s43094-020-00116-z

[53]. Gerber, P. A., & Rutter, G. A. (2017). The role of oxidative stress and hypoxia in pancreatic beta-cell dysfunction in diabetes mellitus. *Antioxidants & redox signaling*, *26*(10), 501-518.

[54]. Latha, R. C., & Daisy, P. (2010). Parameters in Streptozotocin diabetic rats. *International Journal of Pharmacology*, *6*(2), 89-96.

[55]. Sowmya, B. H., & Usha, A. D.(2020 b). Antidiabetic and ameliorative efficacy of two varieties of Psidium guajava L. fruit extracts in type 2 diabetes and indices of complications in diabetic albino rats. *International Journal of Medical Science and Public Health*, 9 (10), 599-603. doi:10.5455/ijmsph.2020.12180202008012021

[56]. Jomova, K., Alomar, S. Y., Alwasel, S. H., Nepovimova, E., Kuca, K., & Valko, M. (2024). Several lines of antioxidant defense against oxidative stress: antioxidant enzymes, nanomaterials with multiple enzyme-mimicking activities, and low-molecular-weight

antioxidants. *Archives of toxicology*, *98*(5), 1323-1367. https://doi.org/10.1007/s00204-024-03696-4

[57]. Jena, A. B., Samal, R. R., Bhol, N. K., & Duttaroy, A. K. (2023). Cellular Red-Ox system in health and disease: The latest update. *Biomedicine & Pharmacotherapy*, *162*, 114606. https://doi.org/10.1016/j.biopha.2023.114606

[58]. Banerjee, A., Singh, S., Prasad, S. K., Kumar, S., Banerjee, O., Seal, T., & Maji, B. K. (2020). Protective efficacy of Tinospora sinensis against streptozotocin induced pancreatic islet cell injuries of diabetic rats and its correlation to its phytochemical profiles. *Journal of ethnopharmacology*, *248*, 112356. https://doi.org/10.1016/j.jep.2019.112356

[59]. Jayachandran, M., Vinayagam, R., Ambati, R. R., Xu, B., & Chung, S. S. M. (2018). Guava leaf extract diminishes hyperglycemia and oxidative stress, prevents β-cell death, inhibits inflammation, and regulates NF-kB signaling pathway in STZ induced diabetic rats. *BioMed research international*, *2018*. https://doi.org/10.1155/2018/4601649

[60]. Iida, H., & Watada, H. (2025). Pancreatic β-cell Dysfunction and Diabetes. *Juntendo Medical Journal*, *71*(3), 158-165. doi: 10.14789/ejmj.JMJ25-0001-R

[61]. Szkudelski, T. (2001). The mechanism of alloxan and streptozotocin action in B cells of the rat pancreas. *Physiological research*, *50*(6), 537-546.

[62]. Matusin, A. H. A., Abd Ghani, N. I., & Ahmad, N. (2021). Pancreatic islet regenerative capability of Dillenia excelsa in alloxan-induced diabetic rats. *Journal of Applied Pharmaceutical Science*, *11*(3), 121-129. https://dx.doi.org/10.7324/JAPS.2021.110315

[63]. Gewies, A. (2003). Introduction to apoptosis. *Apo Review*, *3*(1), 1-26.

[64]. Kim, W. H., Lee, J. W., Suh, Y. H., Hong, S. H., Choi, J. S., Lim, J. H., & Jung, M. H. (2005). Exposure to chronic high glucose induces β-cell apoptosis through decreased interaction of glucokinase with mitochondria: downregulation of glucokinase in pancreatic β-cells. *Diabetes*, *54*(9), 2602-2611. https://doi.org/10.2337/diabetes.54.9.2602

[65]. Korkmaz-Icöz, S., Al Said, S., Radovits, T., Li, S., Brune, M., Hegedűs, P., ... & Szabó, G. (2016). Oral treatment with a zinc complex of acetylsalicylic acid prevents diabetic cardiomyopathy in a rat model of type-2 diabetes: activation of the Akt pathway. *Cardiovascular diabetology*, *15*(1), https://doi.org/10.1186/s12933-016-0383-8

[66]. Creagh, E. M., Conroy, H., & Martin, S. J. (2003). Caspase-activation pathways in apoptosis and immunity. *Immunological reviews*, *193*(1), 10-21. https://doi.org/10.1034/j.1600-065X.2003.00048.x

[67]. Jafri, A., Siddiqui, S., Rais, J., Ahmad, M. S., Kumar, S., Jafar, T., ... & Arshad, M. (2019). Induction of apoptosis by piperine in human cervical adenocarcinoma via ROS mediated mitochondrial pathway and caspase-3 activation. *Excli Journal*, *18*, 154. https://doi.org/10.17179/excli2018-1928

[68]. Yousefi, H., Karimi, P., Alihemmati, A., Alipour, M. R., Habibi, P., & Ahmadiasl, N. (2017). Therapeutic potential of genistein in ovariectomy-induced pancreatic injury in diabetic rats: The regulation of MAPK pathway and apoptosis. *Iranian Journal of Basic Medical Sciences*, *20*(9), 1009. https://doi.org/10.22038/IJBMS.2017.9269

[69]. Lee, D., Kim, K. H., Lee, J., Hwang, G. S., Lee, H. L., Hahm, D. H., & Kang, K. S. (2017). Protective effect of cirsimaritin against streptozotocin-induced apoptosis in pancreatic beta cells. *Journal of Pharmacy and Pharmacology*, *69*(7), 875-883. https://doi.org/10.1111/jphp.12719

[70]. Khamchan, A., Paseephol, T., & Hanchang, W. (2018). Protective effect of wax apple (Syzygium samarangense (Blume) Merr. & LM Perry) against streptozotocin-induced pancreatic ß-cell damage in diabetic rats. *Biomedicine & Pharmacotherapy*, *108*, 634-645. https://doi.org/10.1016/j.biopha.2018.09.072.

**Table 2: Effect of PG extracts on body weight (gm) in experimental rats.**

| Groups | Initial | Week 1 | Week 2 | Week 3 | Week 4 | Week 5 | Week 6 | Week 7 | Week 8 |
|---|---|---|---|---|---|---|---|---|---|
| Control | 187.5 ± 3.10$^{a}$ | 206.6 ± 3.34$^{a}$ | 226.6 ± 3.34$^{a}$ | 244.16 ± 2.72$^{a}$ | 265 ± 3.42$^{a}$ | 289.16 ± 2.39$^{a}$ | 303.33 ± 2.11$^{a}$ | 324.16 ± 1.54$^{a}$ | 344.16 ± 1.54$^{a}$ |
| STZ | 188.33 ± 3.08$^{a}$ | 170 ± 2.59$^{b}$ | 160.83 ± 1.54$^{b}$ | 152.5 ± 1.12$^{e}$ | 145.83 ± 1.54$^{b}$ | 141.33 ± 1.86$^{b}$ | 134.66 ± 1.05$^{b}$ | 129.5 ± 0.96$^{b}$ | 122.5 ± 1.12$^{b}$ |
| STZ + GLB | 191.66 ± 3.08$^{a}$ | 176.6 ± 2.74$^{b}$ | 190.83 ± 2.01$^{c}$ | 208.16 ± 2.25$^{c}$ | 226.66 ± 1.67$^{c}$ | 240.83 ± 1.54$^{c}$ | 254.16 ± 2.01$^{c}$ | 269.16 ± 2.01$^{c}$ | 280 ± 2.59$^{c}$ |
| STZ + LA.L | 186.66± 3.34$^{a}$ | 182± 3.61$^{b}$ | 205 ± 1.83$^{c}$ | 230 ± 1.29$^{b}$ | 250.83 ± 1.54$^{a}$ | 267.5 ± 2.14$^{a}$ | 284.16 ± 2.01$^{a}$ | 303.33 ± 2.11$^{a}$ | 324.16 ± 1.54$^{a}$ |

.

**Note: Mean values with same superscript letters in the given column are not significantly different, whereas those with different superscript letters are significantly (P<0.05) different as judged by Tukey's *post hoc* test**

**Table 3: Effect of PG extract on food intake (gm) in experimental rats.**

| Groups | Week 1 | Week 2 | Week 3 | Week 4 | Week 5 | Week 6 | Week 7 | Week 8 |
|---|---|---|---|---|---|---|---|---|
| Control | 16.78 ± 1.09$^{a}$ | 17.88 ± 0.81$^{a}$ | 17.88 ± 0.81$^{a}$ | 20.97 ± 1.49$^{a}$ | 21.80 ± 1.61$^{a}$ | 23.13 ± 1.34$^{a}$ | 25.73 ± 1.46$^{a}$ | 25.86 ± 1.24$^{a}$ |
| STZ | 20.12 ± 1.42$^{b}$ | 23.22 ± 1.80$^{b}$ | 22.61 ± 1.07$^{b}$ | 28.46 ± 1.19$^{b}$ | 30.25 ± 1.42$^{b}$ | 37.33 ± 2.62$^{b}$ | 33.33 ± 1.02$^{b}$ | 36.57 ± 1.73$^{b}$ |
| STZ + GLB | 18.05 ± 1.81$^{c}$ | 17.38 ± 1.61$^{c}$ | 19.21 ± 1.34$^{c}$ | 25.79 ± 1.17$^{c}$ | 26.73 ± 1.31$^{c}$ | 30.70 ± 1.52$^{c}$ | 27.09 ± 0.56$^{c}$ | 27.90 ± 1.08$^{c}$ |
| STZ + LA.L | 17.96 ± 1.58$^{c}$ | 18.66 ± 1.81$^{c}$ | 17.88 ± 0.81$^{a}$ | 23.94 ± 1.23$^{a}$ | 23.54 ± 1.57$^{a}$ | 26.48 ± 1.63$^{a}$ | 25.61 ± 1.46$^{a}$ | 25.15 ± 1.24$^{a}$ |

**Note: Mean values with same superscript letters in the given column are not significantly different, whereas those with different superscript letters are significantly (P<0.05) different as judged by Tukey's *post hoc* test**

**Table 4: Effect of PG extract on water intake (ml) in experimental rats.**

| Groups | Week 1 | Week 2 | Week 3 | Week 4 | Week 5 | Week 6 | Week 7 | Week 8 |
|---|---|---|---|---|---|---|---|---|
| **Control** | 28.22 ± 1.79$^{a}$ | 28.22 ± 1.80$^{a}$ | 28.22 ± 1.80$^{a}$ | 28.22 ± 1.80$^{a}$ | 32.33± 1.80$^{a}$ | 28.66 ± 2.00$^{a}$ | 28.66 ± 2$^{a}$ | 28.66 ± 2$^{a}$ |
| **STZ** | 31 ± 2.40$^{b}$ | 40 ± 2.25$^{b}$ | 43 ± 2.20$^{b}$ | 40.83 ± 2.08$^{b}$ | 40.83 ± 2.08$^{b}$ | 20.83 ± 2.08$^{b}$ | 42.5 ± 1.61$^{b}$ | 47.5 ± 2.10$^{b}$ |
| **STZ + GLB** | 28.33 ± 2.66$^{c}$ | 36.33 ± 3.90$^{c}$ | 43.33 ± 1.38$^{b}$ | 43.33 ± 1.38$^{c}$ | 44.5 ± 2.11$^{c}$ | 43 ± 2.11$^{c}$ | 43.66 ± 2.23$^{c}$ | 39.33 ± 1.77$^{c}$ |
| **STZ + LA.L** | 30.22 ± 2.40$^{c}$ | 39.16 ± 2.15$^{d}$ | 42.5 ± 0.92$^{c}$ | 40.66 ± 2.58$^{b}$ | 40.66 ± 2.58$^{b}$ | 38.16 ± 1.66$^{d}$ | 32.5 ± 1.65$^{a}$ | 31.66 ± 2.06$^{a}$ |

**Note: Mean values with same superscript letters in the given column are not significantly different, whereas those with different superscript letters are significantly (P<0.05) different as judged by Tukey's *post hoc* test**

**Table 5: Effect of PG extract on blood glucose levels (mg/dl) in experimental rats.**

| Groups | Week 1 | Week 2 | Week 3 | Week 4 | Week 5 | Week 6 | Week 7 | Week 8 |
|---|---|---|---|---|---|---|---|---|
| Control | 92.83 ± 1.05$^{a}$ | 93 ± 1.26$^{a}$ | 93.33 ± 1.45$^{a}$ | 94 ± 1.24$^{a}$ | 92.16 ± 1.11$^{a}$ | 93 ± 1.51$^{a}$ | 93.33 ± 1.78$^{a}$ | 93.16 ± 0.54$^{a}$ |
| STZ | 294.16 ± 2.49$^{d}$ | 313.66 ± 3.44$^{b}$ | 323 ± 2.89$^{c}$ | 333 ± 1.99$^{e}$ | 339.16 ± 2.08$^{d}$ | 350 ± 3.42$^{b}$ | 360.5 ± 2.64$^{c}$ | 367.5 ± 1.79$^{c}$ |
| STZ + GLB | 299 ± 2.71$^{b}$ | 287.5 ± 4.62$^{c}$ | 269.5 ± 1.88$^{b}$ | 258.33 ± 3.08$^{c}$ | 247.5 ± 1.71$^{b}$ | 210.83 ± 4.18$^{c}$ | 172.5 ± 3.04$^{b}$ | 140 ± 3.79$^{b}$ |
| STZ + LA.L | 299.5 ± 2.32$^{a}$ | 267.5± 3.36$^{a}$ | 242.16 ± 2.09$^{a}$ | 215.83 ± 3.52$^{b}$ | 181.66 ± 4.79$^{a}$ | 140 ± 2.24$^{a}$ | 120 ± 2.59$^{a}$ | 93.83 ± 0.60$^{a}$ |

**Note:** Different alphabets (a, b, c) within each column indicates significant ($P< 0.05$) differences among experimental groups compared to control group using Tukey's *post hoc* test, whereas same alphabets indicates no significant difference.

**Table 6: Effect of PG extract on Serum lipid profiles in experimental rats.**

| Groups | Total Cholesterol (TC) mg/dl | Triglycerides (TG) mg/dl | High density lipoprotein (HDL) mg/dl | Low density lipoprotein (LDL) mg/dl | Very low density lipoprotein (VLDL) mg/dl |
|---|---|---|---|---|---|
| **Control** | 155.5 ± 1.48$^{a*}$ | 102.16± 3.80 $^{a*}$ | 75.83 ± 2.08 $^{a*}$ | 59.23 ± 0.19 $^{a*}$ | 20.43 ± 0.76 $^{a*}$ |
| **STZ** | 318.03 ± 2.14$^{b}$ | 238.83 ± 6.66 $^{b}$ | 25.33 ± 2.22 $^{b}$ | 244.93 ± 1.67 $^{b}$ | 47.76 ± 1.33 $^{b}$ |
| **STZ + GLB** | 185 ± 2.59$^{c*}$ | 128.5 ± 2.31 $^{c*}$ | 58.16 ± 2.47 $^{c*}$ | 101.13 ± 1.67 $^{c*}$ | 25.7 ± 0.46 $^{c*}$ |
| **STZ + LA.L** | 154.7 ± 2.18 $^{a*\#}$ | 105.16 ± 1.84 $^{a*\#}$ | 71 ± 2.08 $^{a*\#}$ | 62.66 ± 1.14 $^{a*\#}$ | 21.03 ± 0.36 $^{a*\#}$ |

**Note: Mean values with same superscript letters in the given column are not significantly different, whereas those with different superscript letters are significantly (P<0.05) different as judged by Tukey's *post hoc* test. *indicates significantly different (*P*< 0.05) among control and experimental groups compared to STZ group whereas # indicates significantly different (*P*<0.05) among extract treated groups compared to GLB group using *post hoc* Bonneferoni's test**.

**Table 7: Effect of PG extract on relative organ weight of pancreas in experimental rats**

| Groups | Relative weight of pancreas (mg/g) |
|---|---|
| **Control** | $0.42 \pm 0.005^{a*}$ |
| **STZ** | $0.22 \pm 0.008^{b}$ |
| **STZ+GLB** | $0.37 \pm 0.006^{c*}$ |
| **STZ+LA.L** | $0.40 \pm 0.002^{a*\#}$ |

**Note: Mean values with same superscript letters in the given column are not significantly different, whereas those with different superscript letters are significantly (P<0.05) different as judged by Tukey's *post hoc* test. *indicates significantly different (*P*< 0.05) among control and experimental groups compared to STZ group whereas #indicates significantly different (*P*<0.05) among extract treated group compared to GLB group using *post hoc* Bonneferoni's test**.

**Table 8: Effect of PG extract on total protein, LPO and antioxidant status of pancreas in experimental rats**

| Groups | Total protein (mg/g/wet.wt. tissue) | LPO (µmoles/g wet.wt tissue) | SOD (U/mg protein) | CAT (µ moles of $H_2O_2$ hydrolyzed/min/mg/ protein) | GSH (µ moles/g wet.wt tissue) | GPX (µ moles of NADPH oxidised/min/mg protein) | GST (µ moles of GS-CDNB formed/min/mg protein) |
|---|---|---|---|---|---|---|---|
| **Control** | $6.52 \pm 0.85^{a*}$ | $11.45 \pm 3.69^{a*}$ | $7.39 \pm 0.82^{a*}$ | $38.17 \pm 1.66^{a*}$ | $4.53 \pm 0.07^{a*}$ | $56.38 \pm 3.06^{a*}$ | $36.87 \pm 1.68^{a*}$ |
| **STZ** | $3.09 \pm 0.65^{b}$ | $79.81 \pm 3.3^{b}$ | $2.22 \pm 0.48^{b}$ | $10.65 \pm 1.28^{b}$ | $1.23 \pm 0.06^{b}$ | $9.83 \pm 1.76^{b}$ | $11.77 \pm 1.31^{b}$ |
| **STZ + GLB** | $5.24 \pm 1.04^{c*}$ | $34.98 \pm 1.5^{c*}$ | $5.45 \pm 0.6^{c*}$ | $27.2 \pm 1.12^{c*}$ | $3.26 \pm 0.17^{c*}$ | $29.64 \pm 1.72^{c*}$ | $25.18 \pm 1.53^{c*}$ |
| **STZ + LA.L** | $6.21 \pm 0.86^{a*\#}$ | $12.45 \pm 4.4^{a*\#}$ | $7.69 \pm 0.6^{a*\#}$ | $38.15 \pm 1.8^{a*\#}$ | $4.37 \pm 0.1^{c}a*\#$ | $55.60 \pm 5.05^{a*\#}$ | $36.08 \pm 2.14^{a*\#}$ |

**Note: Mean values with same superscript letters in the given column are not significantly different, whereas those with different superscript letters are significantly (P<0.05) different as judged by Tukey's *post hoc* test. *indicates significantly different (*P*< 0.05) among control and experimental groups compared to STZ group whereas #indicates significantly different (*P*<0.05) among extract treated groups compared to GLB group**

**Table 9: Effect of PG extract on morphometric analysis of pancreas in experimental rats**

| Groups | Number of Islets ($N/10mm^2$) | Number of β-cells ($N/1000\mu m^2$) | Diameter of Islets (µm) | Diameter of β-cells (µm) |
|---|---|---|---|---|
| **Control** | $19.66 \pm 1.48^{a*}$ | $14.5 \pm 0.92^{a*}$ | $122 \pm 2.64^{a*}$ | $5.77 \pm 0.63^{a*\#}$ |
| **STZ** | $4.16 \pm 0.87^{b}$ | $3.83 \pm 0.79^{b}$ | $35.83 \pm 3.09^{b}$ | $2.7 \pm 0.54^{b}$ |
| **STZ + GLB** | $8.83 \pm 0.94^{c*}$ | $9.33 \pm 0.66^{c*}$ | $90.33 \pm 2.76^{c*}$ | $4.66 \pm 0.61^{c*}$ |
| **STZ + LA.L** | $17.16 \pm 0.75^{a*\#}$ | $13.33 \pm 0.76^{a*\#}$ | $116.16 \pm 3.2^{a*\#}$ | $5.66 \pm 0.84^{a*\#}$ |

**Note: Mean values with same superscript letters in the given column are not significantly different, whereas those with different superscript letters are significantly (P<0.05) different as judged by Tukey's *post hoc* test. *indicates significantly different (*P*< 0.05) among control and experimental groups compared to STZ group whereas #indicates significantly different (*P*<0.05) among extract treated groups compared to GLB group**.

**Table 10: Effect of PG extract on Immunoreaction of Caspase-3 and Bcl-2 pancreas in experimental rats**

| Groups | Caspase-3 | Bcl-2 |
|---|---|---|
| **Control** | 0.8 ± 0.71 $^{a*}$ | 8.9 ± 0.61 $^{a*}$ |
| **STZ** | 8.1 ± 0.54 $^{b}$ | 2.5 ± 0.88 $^{b}$ |
| **STZ + GLB** | 4.8 ± 0.7 $^{c*}$ | 5.26 ± 0.86 $^{c*}$ |
| **STZ + LA.L** | 0.73 ± 0.3 $^{a*\#}$ | 8.3 ± 0.84 $^{a*\#}$ |

**Table 11: Effect of PG extract on apoptotic (Caspase-3) and anti apoptotic (Bcl-2) expression of pancreas in experimental rats**

| Groups | Caspase-3 | Bcl-2 |
|---|---|---|
| **Control** | 0.083 ± 0.03 $^{a*}$ | 1.95 ± 0.03 $^{a*}$ |
| **STZ** | 3.54 ± 0.07 $^{b}$ | -0.33 ± 0.025 $^{b}$ |
| **STZ + GLB** | 1.89 ± 0.006 $^{c*}$ | 0.93 ± 0.09 $^{c*}$ |
| **STZ + LA.L** | 0.31 ± 0.007 $^{a*\#}$ | 1.8 ± 0.08 $^{a*\#}$ |

**Note: Mean values with same superscript letters in the given column are not significantly different, whereas those with different superscript letters are significantly (P<0.05) different as judged by Tukey's *post hoc* test. *indicates significantly different (*P*< 0.05) among control and experimental groups compared to STZ group whereas #indicates significantly different (*P*<0.05) among extract treated groups compared to GLB group**