D-Ribose-L-Cysteine Mitigates Hepato-Renal Dysfunction in a STZ + HFD Rat Model of Diabetes.

[1]Isibor H, [1]Eduviere AT, [1,2]Akpovwre CO, [1]Ben-Azu B.

Abstract

Introduction: Diabetes mellitus is a major global health concern often associated with oxidative stress–induced organ complications, particularly in the liver and kidneys. D-ribose-L-cysteine (DRLC), a cysteine prodrug and glutathione precursor, has been suggested as a potential antioxidant therapy. This study investigated the ameliorative effects of DRLC on hepato-renal dysfunction in HFD-fed, and Streptozotocin (STZ)-induced diabetic rats.

Materials and Methods: Male Wistar rats were randomly assigned to five groups. For 28 days, the experimental groups were treated orally with either HFD+STZ, metformin (100 mg/kg), or DRLC at doses of 150 mg/kg and 300 mg/kg. Biochemical parameters, including oxidative stress biomarkers, liver function indices like Alanine Aminotransferase (ALT), Alkaline Phosphatase (ALP), Aspartate Aminotransferase (AST), and Gamma-Glutamyl Transferase (GGT), renal markers, proteins (albumin, total protein, globulin), and electrolytes were evaluated.

Results: HFD+STZ significantly increased MDA, MPO, NO, liver enzymes, creatinine, urea, and bilirubin, while markedly reducing SOD, CAT, GSH, GST, GPx, albumin, globulin, total protein, and electrolytes ($p < 0.05$), indicating hepato-renal dysfunction. These alterations were significantly mitigated ($p<0.05$) following DRLC treatment, with effects comparable to metformin. DRLC decreased pro-oxidant biomarkers, enhanced antioxidant defense, normalized serum proteins, and restored electrolyte balance.

Conclusion: DRLC effectively attenuated oxidative stress and improved liver and kidney function in HFD+STZ-induced diabetic rats, suggesting its therapeutic potential as an antioxidant-based intervention against diabetes-associated organ complications.

Keywords: D-Ribose-L-Cysteine, Diabetes Mellitus, High-Fat Diet, Streptozotocin

1.Department of Pharmacology, Faculty of Allied Health Sciences, Delta State University, Abraka.
2.Department of Pharmacology, School of Basic Medical Sciences, Igbinedion University, Okada

Corresponding author: Isibor Happy, Department of Pharmacology, Faculty of Allied Health Sciences, Delta State University, Abraka. Email: happy-isibor@delsu.edu.ng.

INTRODUCTION

Metabolic diseases, such as diabetes, represent a major public health challenge. The combination of a high-fat diet (HFD) and low-dose streptozotocin (STZ) in rodents serves as a validated model for type 2 diabetes, effectively mimicking key clinical features including hyperglycemia, insulin resistance, dyslipidemia, and oxidative stress.[1,2]. This model posits oxidative stress as a key mediator of hepato-renal impairment in diabetes. An overproduction of reactive oxygen species (ROS) depletes endogenous antioxidant capacity, triggering lipid peroxidation and resulting in organ dysfunction[3,4].

The liver, central to metabolic regulation and the kidneys which both manifest and exacerbate metabolic disturbances via glycosuria, oxidative stress, and advanced glycation end-product (AGE) formation are critically impacted[5,6]. In diabetes, both the liver and kidneys undergo profound metabolic disturbances that contribute to disease progression and complications[7-12].

The sustained metabolic and hydraulic stress of diabetes inflicts severe structural damage to the glomerulus. This includes thickening of the glomerular basement membrane (GBM), podocyte injury and loss, and mesangial matrix expansion, which collectively lead to glomerulosclerosis and capillary loss. This structural decline manifests clinically as progressive diabetic nephropathy, beginning with microalbuminuria and advancing through proteinuria to a falling glomerular filtration rate (GFR), culminating in end-stage renal disease (ESRD)[13-19].

D-Ribose-L-cysteine (DRLC), a cysteine prodrug and glutathione precursor, has recently garnered attention as a potent antioxidant intervention. Unlike N-acetylcysteine, DRLC provides cysteine in a stable, bioavailable form while simultaneously supplying ribose to support cellular metabolism[20]. Preclinical studies demonstrate that DRLC elevates intracellular GSH levels, enhances antioxidant enzyme activities, and reduces oxidative stress in models of heavy metal toxicity, high-fat feeding, and neurotoxicity[21,22,23]. Moreover, DRLC admini--stration has been shown to improve liver histology, mitigate renal oxidative injury, and normalize serum markers of hepatic and renal function[22,24]. These findings underscore its therapeutic potential in metabolic disease–related organ dysfunction.

Despite this evidence, limited data exists on the integrated effect of DRLC on hepato-renal parameters in a combined STZ and HFD model of type 2 diabetes, particularly encompassing liver oxidative indices, renal oxidative markers, liver function tests, and electrolyte profiles. Addressing this gap is crucial for understanding whether glutathione restoration translates into measurable biochemical and functional protection. Therefore, this study investigates the effect of D-ribose-L-cysteine on a comprehensive panel of hepato-renal biomarkers including oxidative parameters, functional indices, and electrolyte balance in male Wistar rats exposed to STZ and HFD.

MATERIALS AND METHODS

Drugs and Chemicals

DRLC and STZ (Sigma-Aldrich, St. Louis, MO, USA), Antioxidant enzyme assays were conducted using the following kits: Superoxide Dismutase (SOD) Activity Assay Kit from Sigma-Aldrich (catalog no. 19160; St. Louis, MO, USA), Catalase (CAT) Activity Assay Kit from Abcam (catalog no. ab83464; Cambridge, UK), Glutathione (GSH) Assay Kit from Sigma-Aldrich (catalog no. CS0260; St. Louis, MO, USA), Glutathione S-Transferase (GST) Activity Assay Kit from Abcam (catalog no. ab65326; Cambridge, UK), and Glutathione Peroxidase (GPx) Activity Assay Kit from Sigma-Aldrich (catalog no. CGP1; St. Louis, MO, USA). Malondialdehyde (MDA) assay reagents, including thiobarbituric acid (TBA) and trichloroacetic acid (TCA), were sourced from Sigma-Aldrich (St. Louis, MO, USA; catalog no. MAK085). Myeloperoxidase (MPO) activity was measured using a Myeloperoxidase Activity Assay Kit from Abcam (Cambridge, UK; catalog no. ab105136).

Animal care

Experimental animals were obtained from the Laboratory Animal Centre, College of Health Science, Delta State University, Abraka, Nigeria. The animals were maintained under standard laboratory conditions and acclimatized for a period of ten days before experimentation. Throughout the study, they were provided with unrestricted access to standard feed and clean water. Ethical approval (RBC/FBMC/ DELSU /25/655) for the study was granted by the Animal Care and Use Research Ethics Committee of the Faculty of Basic Medical Sciences, Delta State University, in accordance with the National Institutes of Health (NIH) Guide for the Care and Use of Laboratory Animals.

Experimental Diet

Purchase of Animal Feeds

Groundnut seeds were obtained from a local market in Abraka Latitude: 5.7917° N Longitude: 6.1043° E Delta State, while commercial pelleted feed was sourced from Rainbow Top Feed Company, Amukpe, Latitude: 5.5099° N Longitude: 5.7508° E Sapele, Nigeria.

Identification

Groundnut seeds were identified and authenticated in the Department of Botany, Faculty of Science, Delta State University, Abraka, Nigeria, by Mr. Michael O.E

Preparation of High-Fat Diet

The groundnut diet was prepared according to the method of Ossai et al. (2024)[25]. Seeds were cleaned, air-dried, and milled using a manual grinder. The feed was stored under hygienic conditions to prevent pest and mold contamination. To preserve nutrient integrity, fresh diets were prepared weekly.

Composition of High-Fat Diet

Eighty grams (80 g) of the milled Groundnut were added to 20 g of the normal commercial animal pellet to make a High Fat Diet.

Nutritional composition

Eighty grams (80 g) of the milled Groundnut were added to 20 g of the normal commercial animal pellet to make a high-fat diet of nutrient composition below

Nutrients	Composition of groundnut in 80 g	Composition of groundnut in 20 g
Fat	39.1	15.9
Carbohydrate	19.0	2.1
Protein	21.9	2.0

Experimental Design

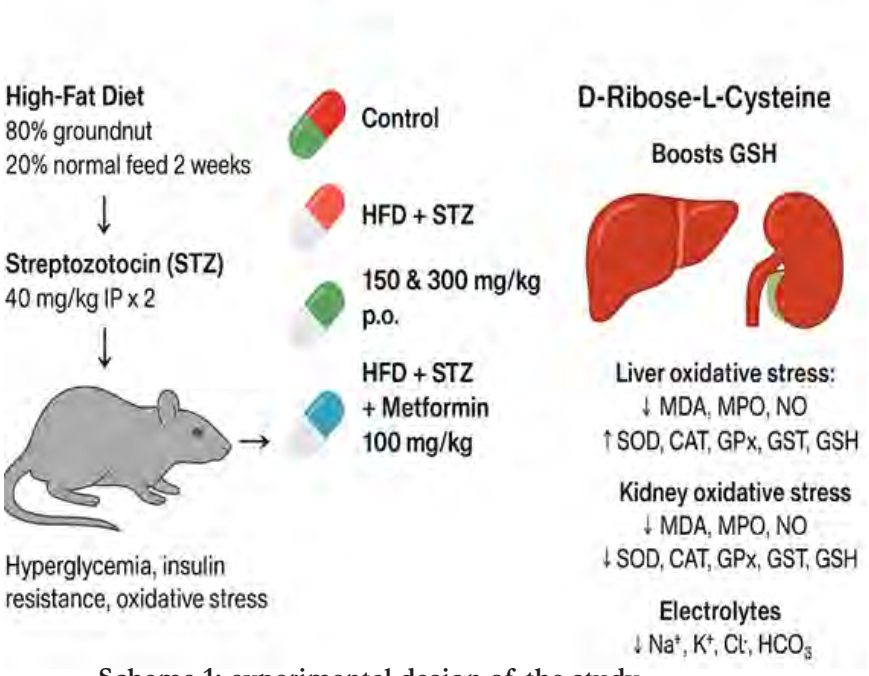
Animals were maintained on a high-fat diet (HFD) for four weeks. On the 14th day of the protocol, they received an intraperitoneal injection (i.p) of streptozotocin (STZ) at 40 mg/kg, followed by a second dose on day 28. The onset of DM was confirmed by measuring fasting blood glucose, with rats classified as diabetic when postprandial glucose levels were ≥288 mg/dL. Male Wistar rats (200–250 g) were randomly assigned to five groups (n = 6). The control group received a standard rodent diet, while the second group was maintained on HFD-STZ only. Groups 3 and 4 were given HFD-STZ, along with oral doses of D-ribose-L-cysteine (DRLC) at 150 mg/kg and 300 mg/kg, respectively. Group five was treated with HFD-STZ and the standard drug, metformin (100

mg/kg body weight)[26]. DRLC was prepared by dissolving 125 mg in 12.5 mL of distilled water to yield a stock solution of 10 mg/mL. The doses of STZ and HFD composition were adapted from an earlier study[25]. Treatments were continued for four weeks, and DRLC was administered orally.

Sample Collection

At the end of the experimental protocol, animals were euthanasia under ketamine anesthesia. Blood and serum were collected and stored. Liver and kidney were also isolated on an ice trail, homogenized, centrifuged (10000 rpm for 10 min at 4°C in a phosphate buffer, PBS), and supernatants were stored at -20 °C for analysis of biochemical levels as previously described by Akpovwre et al. (2024)[27].

Liver oxidative parameters (GSH, SOD, CAT, GPx, GST, MDA, MPO, NO), liver function test parameters (ALT, ALP, AST, and GGT), renal oxidative stress parameters (GSH, SOD, CAT, GPx, GST, MDA, MPO, NO) renal function markers (creatinine, urea, bilirubin, albumin, and globulin) and serum electrolyte (Na^+, K^+, Cl^-, HCO^{3-}) were all assayed for.



Scheme 1: experimental design of the study

Determination of reduced glutathione (GSH), glutathione-s-transferase (GST), glutathione peroxidase (GPx) concentration

Aliquots of liver and kidney supernatants of rat in the respective treatment groups were taken, and reduced glutathione (GSH), glutathione-s-transferase (GST), and glutathione peroxidase (GPx) concentration was determined using the Moron' method[28].

Determination of superoxide dismutase (SOD) activity

The level of SOD activity was determined liver

and kidney supernatants of rat in the respective treatment groups by the method of Misra and Fridovich[29].

Estimation of catalase activity

Catalase activity in liver and kidney tissues was determined according to the method previously described by Sinha[30].

Estimation of brain level of malondialdehyde (MDA)

The liver and kidney levels of MDA, a biomarker of lipid peroxidation, was estimated according to the method of Adam-Vizi and Seregi[31].

Determination of brain myeloperoxidase (MPO) activity

The myeloperoxidase activity in liver and kidney tissues of rat in the respective treatment groups was determined according to the method previously described by Bradley[32].

Estimation of brain nitrite level

Liver and kidney nitrite concentration was estimated using Greiss reagent, which serves as an indicator of nitric oxide production.

Determination of Alanine Aminotransferase (ALT) and Aspartate Aminotransferase (AST)

ALT and AST activities are commonly determined using the Reitman and Frankel colorimetric method[34].

Determination of Alkaline Phosphatase (ALP)

ALP was assayed using a colorimetric method based on its ability to hydrolyze p-nitrophenyl phosphate into p-nitrophenol and phosphate in an alkaline medium. The liberated p-nitrophenol produces a yellow color, measured spectrophotometrically at 405 nm. Enzyme activity is proportional to the rate of color formation[35].

Determination of Gamma-Glutamyl Transferase (GGT)

GGT activity is determined by its transfer of the γ-glutamyl group from the substrate γ-glutamyl-p-nitroanilide to glycylglycine, releasing p-nitroaniline. The formation of p-nitroaniline is monitored spectrophotometrically at 405 nm, with activity expressed in IU/L. This assay is highly sensitive for hepatobiliary dysfunction[36].

Determination of Creatinine, Urea, Bilirubin, Albumin, Total protein, And Globulin

The biochemical parameters were determined using standard spectrophotometric and enzymatic methods. Serum creatinine was assayed based on the Jaffe reaction[37]. Urea concentration was determined by the urease–Berthelot method[38]. Serum bilirubin was estimated by the diazo method[39]. Albumin concentration was measured by the bromocresol green (BCG) dye-binding method, and globulin concentration was calculated as the difference between total serum protein and albumin levels[40].

Determination of serum electrolyte (Na^+, K^+, Cl^-, HCO^{3-})

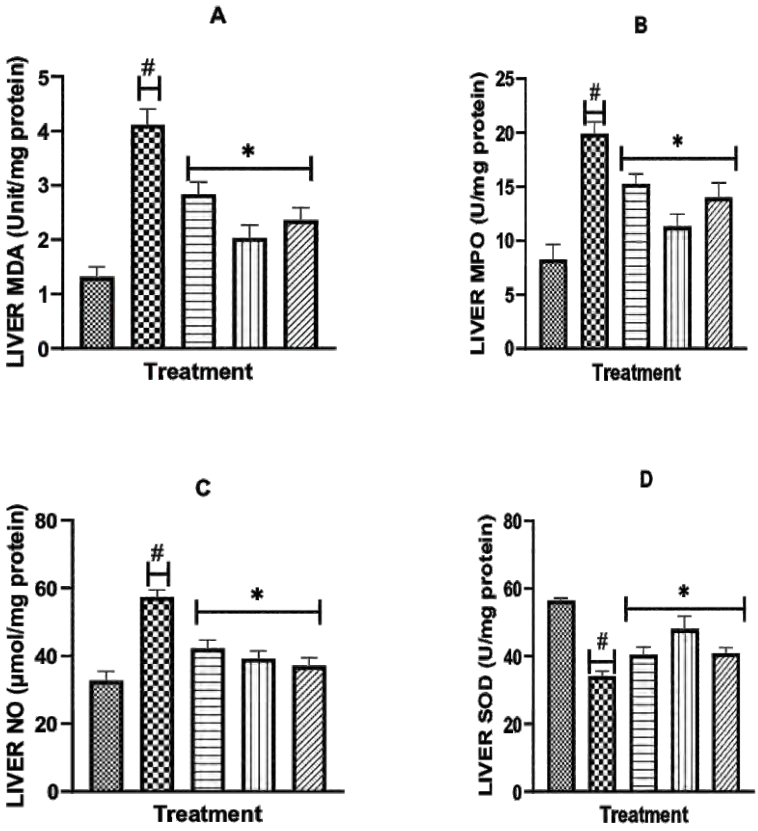
Electrolyte estimation was performed using an ion-selective electrode method. Sodium, potassium, and chloride ions were measured directly by their respective electrodes and reported in mmol/L. Serum bicarbonate (HCO^{3-}) was estimated as total CO^2 by enzymatic methods, with values reflecting the principal buffer base in plasma[41].

RESULTS

Effect of D-ribose-L-cysteine on Liver oxidative biomarkers in rats exposed to STZ and HFD

The effect of D-Ribose-L-Cysteine given daily for 28 days on liver oxidative biomarkers is shown in **Figures 1(A-H), respectively**. One-way ANOVA from figures 1A-C showed that there was significant difference between treatment groups. Post hoc analysis using Tukey's post hoc test revealed that the high fat diet and streptozotocin (HFD + STZ) group showed a significant ($p<0.05$) increase in prooxidant biomarkers (MDA, MPO and NO) when compared to the control group, while DRLC (150, 300 mg/kg), metformin 100mg/kg, given daily for 28 days produced a significant ($p<0.05$) decrease in prooxidant biomarkers (MDA, MPO and NO) when compared with the (HFD+ STZ) group only.

Post hoc analysis using Tukey's post hoc test from figure 1D-H revealed that the high fat diet and streptozotocin (HFD + STZ) group showed a significant ($p<0.05$) decrease in antioxidant biomarkers (SOD, CAT, GSH, GST, GPx) when compared to the control group, while DRLC (150, 300 mg/kg), metformin 100mg/kg, given daily for 28 days produced a significant ($p<0.05$) increase in prooxidant biomarkers (SOD, CAT, GSH, GST, GPx) when compared with the (HFD+ STZ) group only.



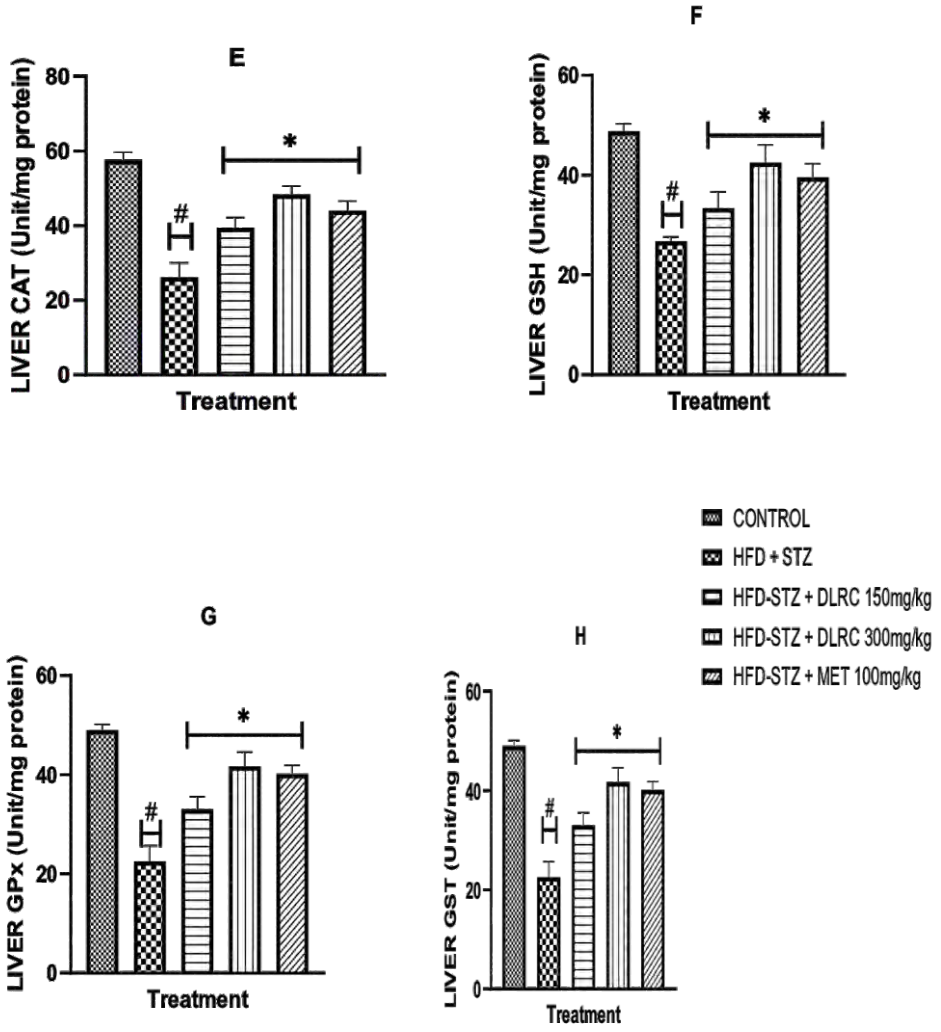


Figure 1: Effect of D-ribose-L-cysteine on liver (A) malondialdehyde (MDA), (B) myeloperoxidase (MPO), (C) nitric oxide (NO), (D) superoxide dismutase (SOD), (E) catalase (CAT), (F) reduced glutathione (GSH), (G) glutathione peroxidase (GPx), (H) glutathione-S-transferase (GST) in rats exposed to STZ and HFD

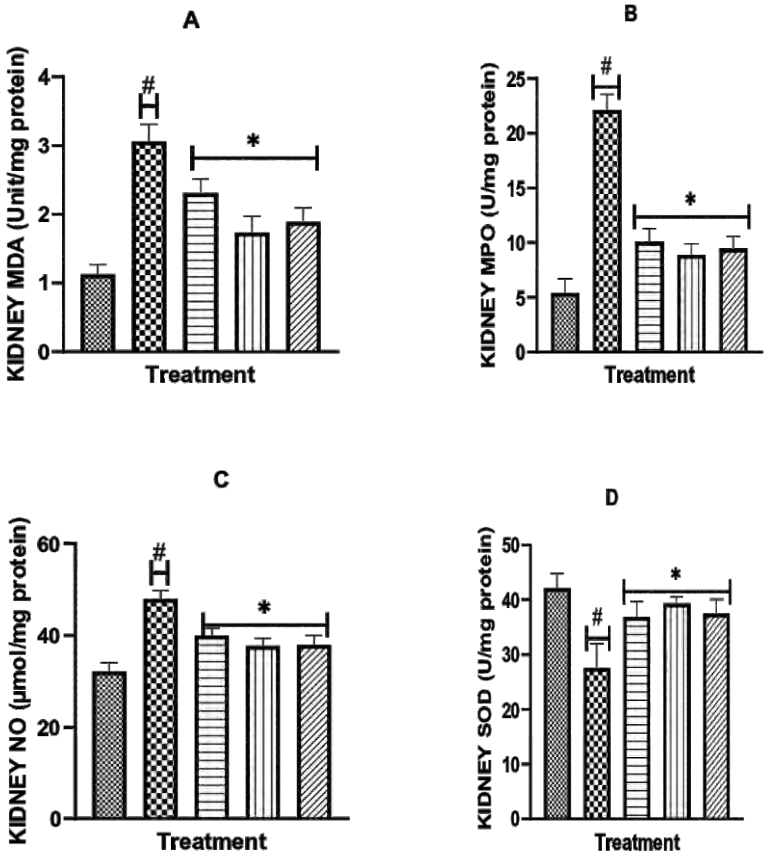
Values represent the mean ± S.E.M for 5 animals per group
# $p<0.05$ compared to the control group (ANOVA followed by Tukey's *post hoc* test).
* $p<0.05$ compared to the pathologic group (ANOVA followed by Tukey's *post hoc* test).

Effect of D-ribose-L-cysteine on Renal oxidative biomarkers in rats exposed to STZ and HFD

The effect of D-Ribose-L-Cysteine given daily for 28 days on renal oxidative biomarkers is shown in **Figures 2A-H, respectively**. One-way ANOVA from figures 2A-C showed that there was significant difference between treatment groups. Post hoc analysis using Tukey's post hoc test revealed that the high fat diet and

streptozotocin (HFD + STZ) group showed a significant ($p<0.05$) increase in prooxidant biomarkers (MDA, MPO and NO) when compared to the control group, while DRLC (150, 300 mg/kg), metformin 100mg/kg, given daily for 28 days produced a significant ($p<0.05$) decrease in prooxidant biomarkers (MDA, MPO and NO) when compared with the (HFD+ STZ) group only.

Post hoc analysis using Tukey's post hoc test from figure 2D-H revealed that the high fat diet and streptozotocin (HFD + STZ) group showed a significant ($p<0.05$) decrease in antioxidant biomarkers (SOD, CAT, GSH, GST, GPx) when compared to the control group, while DRLC (150, 300 mg/kg), metformin 100mg/kg, given daily for 28 days produced a significant ($p<0.05$) increase in prooxidant biomarkers (SOD, CAT, GSH, GST, GPx) when compared with the (HFD+ STZ) group only.



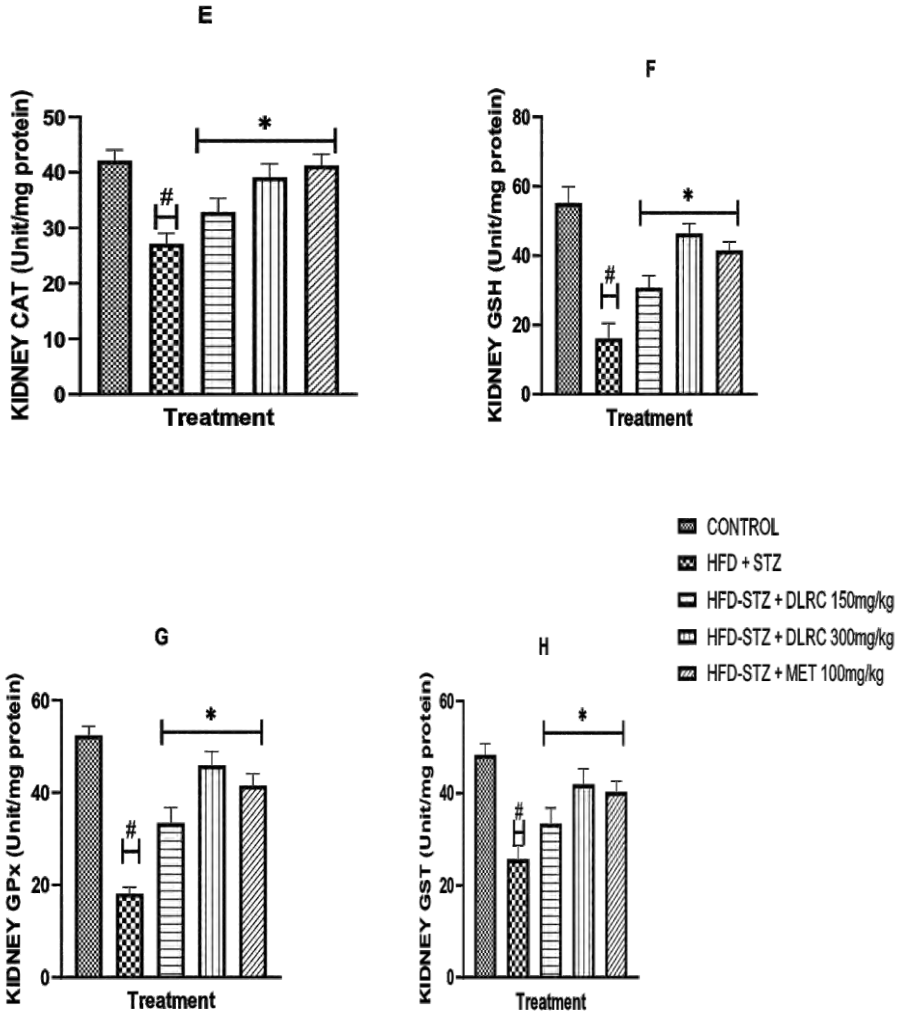


Figure 2: Effect of D-ribose-L-cysteine on kidney (A) malondialdehyde (MDA), (B) myeloperoxidase (MPO), (C) nitric oxide (NO), (D) superoxide dismutase (SOD), (E) catalase (CAT), (F) reduced glutathione (GSH), (G) glutathione peroxidase (GPx), (H) glutathione-S-transferase (GST) in rats exposed to STZ and HFD

Values represent the mean ± S.E.M for 5 animals per group

# $p<0.05$ compared to the control group (ANOVA followed by Tukey's *post hoc* test).

* $p<0.05$ compared to the pathologic group (ANOVA followed by Tukey's *post hoc* test).

Effect of D-ribose-L-cysteine on liver function test in rats exposed to STZ and HFD

The effect of D-Ribose-L-Cysteine given daily for 28 days liver function test parameters was shown in **Figure 3A-D respectively**. One-way ANOVA from figure 3A-D showed that there was significant difference between treatment groups. Post hoc analysis using Tukey's post hoc test revealed that the high fat diet and streptozotocin

(HFD + STZ) group showed a significant ($p<0.05$) increase in liver function parameters (ALT, ALP, GGT, and AST) when compared to the control group, while DRLC (150, 300 mg/kg), metformin 100mg/kg, given daily for 28 days produced a significant ($p<0.05$) decrease in liver function parameters ALT, ALP, GGT, and AST) when compared with the (HFD+ STZ) group only.

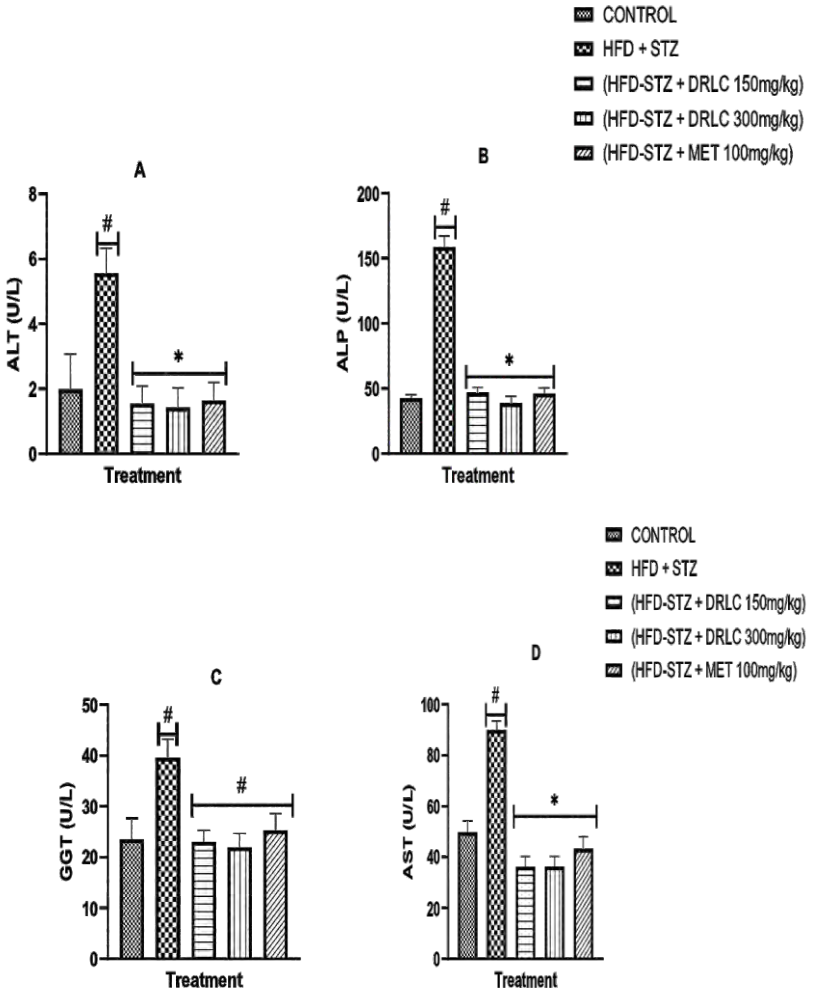


Figure 3: Effect of D-ribose-L-cysteine on (A) Alanine Aminotransferase, (B) Alkaline Phosphatase, (C) Gamma-Glutamyl Transferase, (D) Aspartate Aminotransferase in rats exposed to STZ and HFD

Values represent the mean ± S.E.M for 5 animals per group

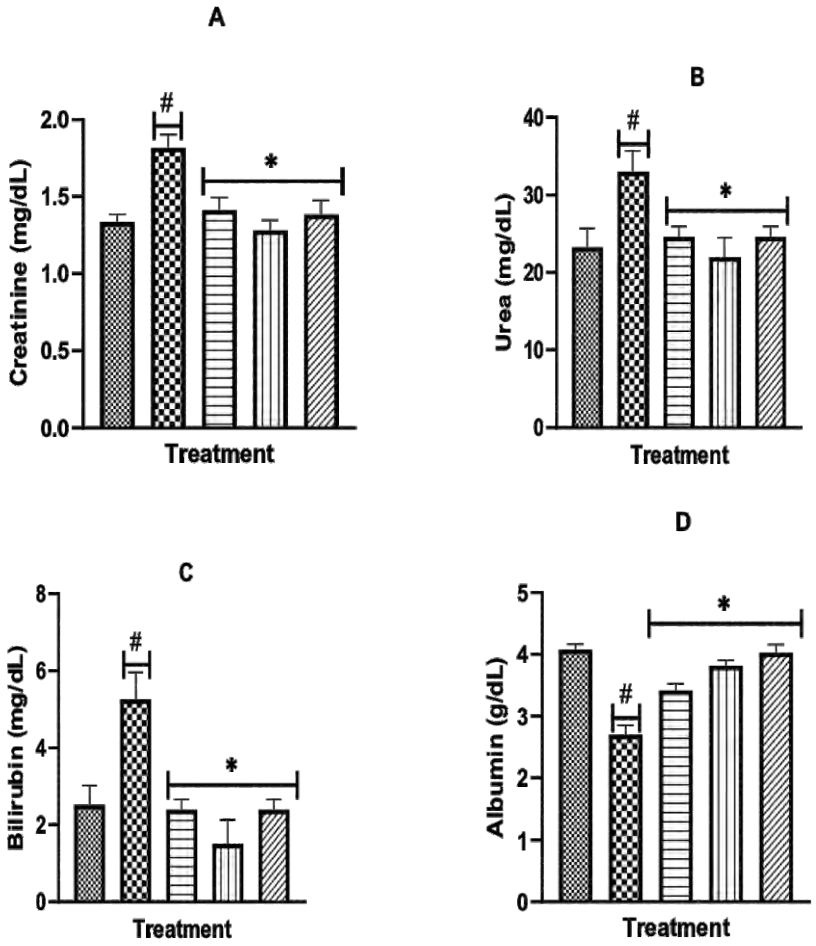
# $p<0.05$ compared to the control group (ANOVA followed by Tukey's *post hoc* test).

* $p<0.05$ compared to the pathologic group (ANOVA followed by Tukey's *post hoc* test).

Effect of D-ribose-L-cysteine on Creatinine, Urea, Bilirubin, Albumin, Total protein And Globulin in rats exposed to STZ and HFD

The effect of D-Ribose-L-Cysteine given daily for 28 days on liver function test parameters was shown in **Figures 4A-F, respectively**. One-way ANOVA from figures 4A-C showed that there was significant difference between treatment groups. Post hoc analysis using Tukey's post hoc test revealed that the high fat diet and streptozotocin (HFD + STZ) group showed a significant ($p<0.05$) increase in Creatinine, Urea, Bilirubin when compared to the control group, while DRLC (150, 300 mg/kg), metformin 100mg/kg, given daily for 28 days produced a significant ($p<0.05$) decrease in Creatinine, Urea, Bilirubin when compared with the (HFD+ STZ) group only.

Post hoc analysis using Tukey's post hoc test from 4D-F revealed that the high fat diet and streptozotocin (HFD + STZ) group showed a significant ($p<0.05$) decrease in Albumin, Total protein And Globulin when compared to the control group, while DRLC (150, 300 mg/kg), metformin 100mg/kg, given daily for 28 days produced a significant ($p<0.05$) increase in Albumin, Total protein And Globulin when compared with the (HFD+ STZ) group only.



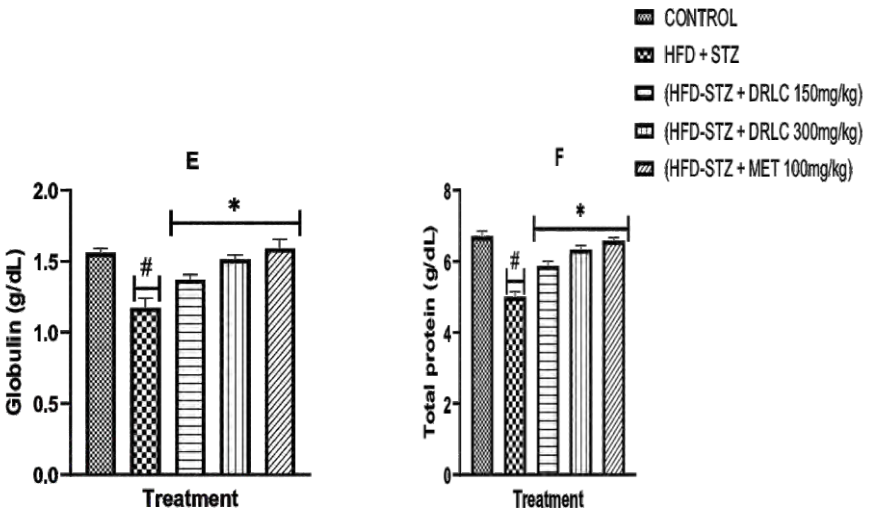
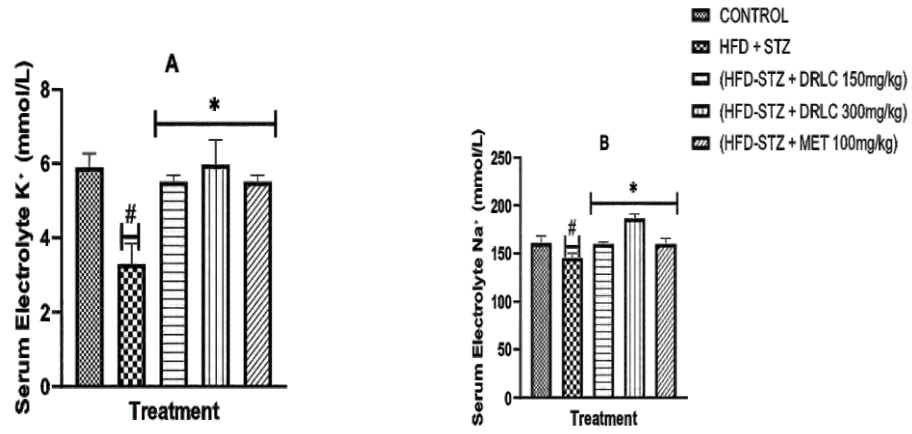


Figure 4: Effect of D-ribose-L-cysteine on (A) creatinine, (B) urea, (C) bilirubin, (D) Albumin, (E) globulin, (F) total protein in rats exposed to STZ and HFD
Values represent the mean ± S.E.M for 5 animals per group
# p<0.05 compared to the control group (ANOVA followed by Tukey's *post hoc* test).
* p<0.05 compared to the pathologic group (ANOVA followed by Tukey's *post hoc* test).

Effect of D-ribose-L-cysteine on Serum Electrolytes in rats exposed to STZ and HFD

The effect of D-Ribose-L-Cysteine given daily for 28 days on liver function test parameters is shown in **Figures 5A-D, respectively**. One-way ANOVA from figures 5 A-D showed that there was significant difference between treatment groups. Post hoc analysis using Tukey's post hoc test revealed that the high fat diet and streptozotocin (HFD + STZ) group showed a significant (p<0.05) decrease in **serum electrolyte** when compared to the control group, while DRLC (150, 300 mg/kg), metformin 100mg/kg, given daily for 28 days produced a significant (p<0.05) increase in **serum electrolytes** when compared with the (HFD+ STZ) group only.



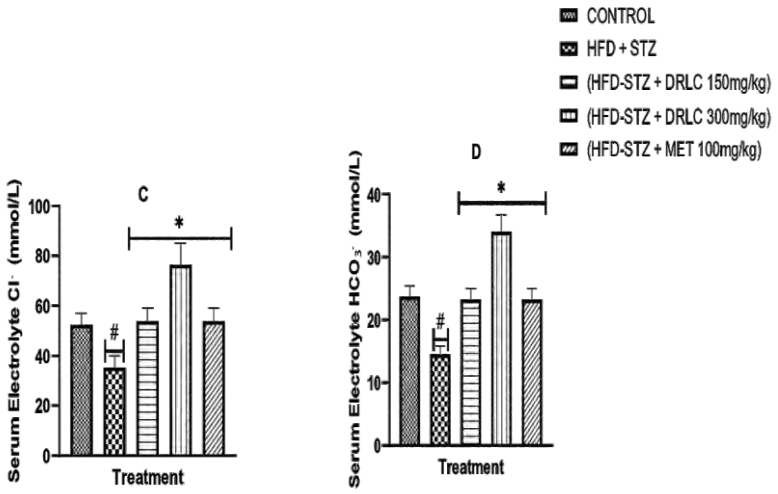


Figure 5: Effect of D-ribose-L-cysteine on (A) potassium ion, (B) sodium ion, (C) chloride ion, (D) biocarbonate ion in rats exposed to STZ and HFD
Values represent the mean ± S.E.M for 5 animals per group
# $p<0.05$ compared to the control group (ANOVA followed by Tukey's *post hoc* test).
* $p<0.05$ compared to the pathologic group (ANOVA followed by Tukey's *post hoc* test).

DISCUSSIONS

The present findings show that combined High-Fat Diet (HFD) and Streptozotocin (STZ) produced a clear hepato–renal oxidative and functional derangement characterized by increased lipid peroxidation and pro-oxidant activity (MDA, MPO, NO), suppression of enzymatic and non-enzymatic antioxidants (SOD, CAT, GSH, GST, GPx), and concurrent impairment of liver and kidney function (elevated ALT, AST, ALP, GGT, creatinine, urea, bilirubin; reduced albumin, total protein, and globulin, and disturbed electrolytes). These changes are consistent with an imbalance between reactive oxygen/nitrogen species generation and endogenous antioxidant defenses induced by metabolic stress and β-cell toxin exposure, and they mirror what has been reported in a diet- and toxin-induced model of metabolic and oxidative injury[24]. Administration of D-ribose-L-cysteine (DRLC) at both 150 and 300 mg/kg for 28 days, and metformin (100 mg/kg), substantially reversed these pathological alterations: DRLC lowered pro-oxidant markers, restored antioxidant enzyme activities in liver and kidney, reduced markers of hepatocellular and renal injury, and improved serum protein and electrolyte profiles. The effect of DRLC on pro-oxidant and antioxidant markers is similar to the effect seen in Ejiro et al. (2025)[21], where coadministration of lead with DRLC led to significantly ($p<0.05$) reduced MDA levels when compared to the negative control group, and significantly ($p<0.05$) improved antioxidant defense in the kidneys of the mice exposed to lead for a period of 21 days. Taken together, these data indicate that DRLC exerts a multifaceted protective effect on hepato-renal systems in the HFD+STZ model.

Mechanistically, the protective profile observed here is biologically plausible because DRLC is a

cysteine-donor pro-drug that supports intracellular glutathione (GSH) synthesis[22], thereby replenishing the major cellular thiol buffer and enabling recovery of downstream antioxidant enzyme activities and redox-sensitive pathways. Several recent preclinical reports show comparable patterns[24,42] DRLC or riboceine supplementation increases tissue GSH, reduces MDA and nitrosative stress, and attenuates pro-inflammatory cytokines in models of heavy-metal[22], xenobiotic or diet-induced oxidative injury[43], supporting the fact that enhanced GSH biosynthesis underpins the observed normalization of antioxidant indices and reduced lipid peroxidation. The parallel benefit of metformin in the current experiment provides a useful comparator: metformin's protective effects in metabolic disease models have been attributed in part to reductions in mitochondrial ROS generation and indirect enhancement of cellular antioxidant responses[44], so the similar direction of effect seen with DRLC suggests complementary, perhaps synergistic, mechanisms — metformin improving metabolic flux and mitochondrial redox tone, and DRLC directly increasing the glutathione pool and thiol-dependent detoxification. This idea is consistent with broader recent hypotheses arguing that bolstering glutathione and its precursors is a rational strategy to prevent or reduce tissue damage in conditions of metabolic stress and early diabetes[45,46]. Also, restoration of serum proteins and reduction in transaminases after DRLC indicate not only reduced oxidative injury but also partial recovery of synthetic and metabolic liver function — an important translational signal when considering hepato-renal protection[47]. Thirdly, the renal improvements (electrolyte normalization, lower creatinine/urea) suggest DRLC protects not just by systemic antioxidant replenishment but also by preserving tubular and glomerular integrity in the context of metabolic-toxic stress.

In summary, the data supports that DRLC ameliorates HFD+STZ-induced oxidative stress and associated hepato-renal dysfunction, most likely through augmentation of glutathione-dependent antioxidant defenses and suppression of pro-oxidant and pro-inflammatory cascades. These findings align with recent preclinical work showing DRLC/riboceine raises tissue GSH, lowers lipid peroxidation and inflammatory mediators, and improves organ-specific functional endpoints, and they fit within a growing conceptual framework that glutathione precursors can be protective in early metabolic disease. Taken together, DRLC merits further mechanistic and translational study as a candidate adjunctive strategy to reduce oxidative injury and preserve liver and kidney function in metabolic and toxin-related disease models.

REFERENCE

1. Phillips S, Feola A, Solomon J, Cardelle L, Douglass A, Bales KL, et al. Retinal and metabolic changes in a high-fat diet (HFD)+STZ model of Type II diabetes. Mol Vis. 2024;30:e239.
2. Brito AK, Mendes AV, Timah-Acha B, Santos-Oliveira AS, Lopes-Macedo J, Suzuki-Cruzio A, et al. Experimental Models of Type 2 Diabetes Mellitus Induced by Hyperlipidemic Diet (HFD) and Streptozotocin Administration in Rats: An Integrative Review. Biomedicines. 2025; 13(5):1158.
3. Abozaid OA, Moawed FS, Gabr HF, Esmat MA. Combined Effects of Metformin, Quercetin, and Fractionated Gamma Irradiation on MiR-107-Mediated Brain Injury in HFD/STZ-Induced Diabetic Rats. Dose-Response. 2025;23(3):e1559 3258251367627.
4. Ha E, Alhakami A, Alqahtani S, Alqahtani Y, Shati A, Almohaimeed H. Mesenchymal Stem Cells Attenuate Renal Microscopic Alterations in Induced Diabetic

Nephropathy in Rats through Suppression of Oxidative Stress, Inflammation, Apoptosis and Upregulation of Nrf2/PPAR-γ Inflammatory Signaling Pathway. Int J Morphol. 2025;43(1):226-236.
5. Maiorana A, Tagliaferri F, Dionisi-Vici C. Current understanding on pathogenesis and effective treatment of glycogen storage disease type Ib with empagliflozin: new insights coming from diabetes for its potential implications in other metabolic disorders. Front Endocrinol. 2023;14: e1145111.
6. Kumar M, Dev S, Khalid MU, Siddenthi SM, Noman M, John C, et al. The bidirectional link between diabetes and kidney disease: mechanisms and management. Cureus. 2023;15(9):e45615.
7. Tomic D, Shaw JE, Magliano DJ. The burden and risks of emerging complications of diabetes mellitus. Nat Rev Endocrinol. 2022;18(9):525-539.
8. Allameh A, Niayesh-Mehr R, Aliarab A, Sebastiani G, Pantopoulos K. Oxidative stress in liver pathophysiology and disease. Antioxidants. 2023;12(9):e1653.
9. Hanke J, Romejko K, Niemczyk S. Sodium-Glucose Cotransporter-2 Inhibitors in Diabetes and Beyond: Mechanisms, Pleiotropic Benefits, and Clinical Use—Reviewing Protective Effects Exceeding Glycemic Control. Molecules. 2025;30(20):e4125.
10. Ravindran S, Munusamy S. Renoprotective mechanisms of sodium glucose co transporter 2 (SGLT2) inhibitors against the progression of diabetic kidney disease. J Cell Physiol. 2022;237(2):1182-1205.
11. Pardawala N, Al Hassan M, Bahig J, Szaszi K, Doan H, Shoker A, et al. Exploring Glomerular Filtration Mechanisms and Molecular Pathways: Insights for Advancing Hemodialysis Membrane Design. ACS Biomater Sci Eng. 2025. [Epub ahead of print]
12. Yang Y, Xu G. Update on pathogenesis of glomerular hyperfiltration in early diabetic kidney disease. Front Endocrinol. 2022; 13:e872918.
13. Wu T, Ding L, Andoh V, Zhang J, Chen L. The mechanism of hyperglycemia-induced renal cell injury in diabetic nephropathy disease: an update. Life. 2023;13(2):e539.
14. Ebefors K, Lassén E, Anandakrishnan N, Azeloglu EU, Daehn IS. Modeling the glomerular filtration barrier and intercellular crosstalk. Front Physiol. 2021;12:e689083.
15. Thomas HY, Ford Versypt AN. Pathophysiology of mesangial expansion in diabetic nephropathy: mesangial structure, glomerular biomechanics, and biochemical signaling and regulation. J Biol Eng. 2022;16(1):e19.
16. Thomas HY. Computational Modeling of Mesangial Expansion and Glomerular Fibrosis in Diabetes [dissertation]. State University of New York at Buffalo; 2022.
17. Wang N, Zhang C. Recent advances in the management of diabetic kidney disease: slowing progression. Int J Mol Sci. 2024;25(6):e3086.
18. Ahmed K, Dubey MK, Dubey S, Pandey DK. Chronic kidney disease: Causes, treatment, management, and future scope. In: Computational Intelligence for Genomics Data. Academic Press; 2025:99-111.
19. Pottanat N, Friederich A, LaMotte J. End-Stage Kidney Disease, Dialysis, and Transplant Basics. In: Psychosocial Considerations in Pediatric Kidney Conditions: Guidance for Collaborative Practice. Cham: Springer Nature Switzerland; 2024: 67-94.
20. Obukohwo OM, Faith FY, Ejiro OP, Onyebuchi MM, Ajayi AF. Medicinal applications of d-ribose l-cysteine in neuro-endocrinopathy, reproductive dysfunction and cardio-metabolic syndrome: A review of evidence in animal investigations. Eur J Med Chem Rep. 2024;11:e100153.
21. Ejiro A, Ikechukwu AA. Nephroprotective

Effect of D-Ribose-L-Cysteine (DRLC) On Renal Oxidative Stress and Proinflammatory Markers in Lead Acetate-Treated Mice. Trop J Chem. 2025;1(1):26-30.
22. Isibor H, Ajayi AM, Ben-Azu B, Omeiza NA, Ademola AP, Umukoro S. D-ribose-L-cysteine reduces oxidative stress and inflammatory cytokines to mitigate liver damage, and memory decline induced by copper sulfate in mice. J Trace Elem Med Biol. 2022;73:127001.
23. Ramos V, Reis M, Ferreira L, Silva AM, Ferraz R, Vieira M, et al. Stalling the course of neurodegenerative diseases: could cyanobacteria constitute a new approach toward therapy? Biomolecules. 2023;13 (10):e1444.
24. Ojetola AA, Adedeji TG, Fasanmade AA. Changes in antioxidants status, atherogenic index and cardiovascular variables after prolonged doses of D-ribose-L-cysteine in male Wistar rats. Heliyon. 2021;7(2):e06113.
25. Ossai NR, Ojieh AE, Nwangwa EK, Nwogueze BC. Attenuating Potential of Modified Diets on Testicular Inflammatory Biomarkers in Streptozotocin-Induced Diabetic Wistar Rats. Pak Heart J. 2024; 57(01):297-318.
26. Hacioglu C, Kar F, Kara Y, Yucel E, Donmez DB, Sentürk H, et al. Comparative effects of metformin and Cistus laurifolius L. extract in streptozotocin-induced diabetic rat model: oxidative, inflammatory, apoptotic, and histopathological analyzes. Environ Sci Pollut Res. 2021;28(41):57888-57901.
27. Akpovwre CO, Eduviere AT, Isibor H, Edje KE, Ewhre LO, Akinsola AO, et al. Theaflavin Alleviates Memory Deficit and Anxiety-Like Phenotypes in Valproic Acid Murine Model of Autism: Impact on Cholinergic and Oxido-Nitrigic Stress Mechanisms. Trop J Nat Prod Res. 2024; 8(12):9345-9355.
28. Moron MS, Depierre JW, Mannervik B. Levels of glutathione, glutathione reductase and glutathione S-transferase activities in rat lung and liver. Biochim Biophys Acta Gen Subj. 1979;582(1):67-78.
29. Misra HP, Fridovich I. The role of superoxide anion in the autoxidation of epinephrine and a simple assay for superoxide dismutase. J Biol Chem. 1972;247(10):3170-3175.
30. Sinha AK. Colorimetric assay of catalase. Anal Biochem. 1972;47(2):389-394.
31. Ádám-Vizi V, Seregi A. Receptor independent stimulatory effect of noradrenaline on Na, K-ATPase in rat brain homogenate: Role of lipid peroxidation. Biochem Pharmacol. 1982;31(13):2231-2236.
32. Bradley PP, Christensen RD, Rothstein G. Cellular and extracellular myeloperoxidase in pyogenic inflammation. Blood. 1982; 60(3):618-622.
33. Green CL. Analysis of nitrate, nitrite and [15N] nitrate in biological fluids. Infect Immun. 1994;62:e1171.
34. Reitman S, Frankel S. A colorimetric method for the determination of serum glutamic oxalacetic and glutamic pyruvic transaminases. Am J Clin Pathol. 1957; 28(1):56-63.
35. Tang Z, Chen H, He H, Ma C. Assays for alkaline phosphatase activity: Progress and prospects. TrAC Trends Anal Chem. 2019;113:32-43.
36. Goldberg DM, Martin JV. Role of γ-glutamyl transpeptidase activity in the diagnosis of hepatobiliary disease. Digestion. 1975;12(4-6):232-246.
37. Chromý V, Rozkošná K, Sedlak P. Determination of serum creatinine by Jaffe method and how to calibrate to eliminate matrix interference problems. Clin Chem Lab Med. 2008;46(8):1127-1133.
38. Taylor AJ, Vadgama P. Analytical reviews in clinical biochemistry: the estimation of urea. Ann Clin Biochem. 1992;29(3):245-264.
39. Thaler M, Luppa PB, Schlebusch H. Bilirubin measurement-an updated survey.

Laboratoriums Medizin. 2008;32(1):1-9.
40. Adair GS, Robinson ME. The specific refraction increments of serum-albumin and serum-globulin. Biochem J. 1930; 24(4):e993.
41. Burnett RW, Covington AK, Fogh-Andersen N, Külpmann WR, Lewenstam A, Maas AH, et al. Use of ion-selective electrodes for blood-electrolyte analysis. Recommendations for nomenclature, definitions and conventions. J Int Fed Clin Chem. 1994;6(4):154-159.
42. Chidebe EO, Moke EG, Asiwe JN, Ben-Azu B, Demaki WE, Oritsemuelebi B, et al. Therapeutic effect of coenzyme-Q10 pretreatment on isoprenaline-induced cardiogenic hepatorenal complications in rats. Int J Physiol Pathophysiol Pharmacol. 2024;16(6):126-138.
43. Jayanti S, Vitek L, Verde CD, Llido JP, Sukowati C, Tiribelli C, et al. Role of natural compounds modulating Heme catabolic pathway in gut, liver, cardiovascular, and brain diseases. Biomolecules. 2024;14(1):63.
44. Buczyńska A, Sidorkiewicz I, Krętowski AJ, Adamska A. Examining the clinical relevance of metformin as an antioxidant intervention. Front Pharmacol. 2024;15:31330797.
45. Dawi J, Misakyan Y, Affa S, Kades S, Narasimhan A, Hajjar F, et al. Oxidative stress, glutathione insufficiency, and inflammatory pathways in type 2 diabetes mellitus: Implications for therapeutic interventions. Biomedicines. 2024;13(1):e18.
46. Tuell D, Ford G, Los E, Stone W. The Role of glutathione and its precursors in type 2 diabetes. Antioxidants. 2024;13(2):e184.
47. Li Z, Lin D, Miao J, Fan J, Lu F, Zhang X, et al. Effects of transcatheter tricuspid valve replacement on hepatic and renal function in severe TR. IJC Heart Vasc. 2025;59:e101714.

Isibor H, Eduviere AT, Akpovwre CO, Ben-Azu B.. D-Ribose-L-Cysteine Mitigates Hepato-Renal Dysfunction in a STZ + HFD Rat Model of Diabetes. Afr. J. Trop. Med. & Biomed. Res. 2026; 9(1) 114-130
https://dx.doi.org/10.4314/ajtmbr.v9i1.7