Physiology and Pharmacology 30 (2026) 70-80
Experimental Research Article

Comparison of the effects of platelet-rich plasma and erythropoietin on nephropathy and hepatopathy in streptozotocin-induced diabetic rats: a biochemical, oxidative stress, and histopathological study

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ABSTRACT

Introduction: This research evaluates the treatment benefits of platelet-rich plasma (PRP) and erythropoietin (EPO) on diabetic nephropathy and hepatopathy in rats.

Methods: Thirty male rats were separated into five groups: control, diabetic control, PRP-treated diabetic (1 mL PRP subcutaneously twice weekly), EPO-treated diabetic (300 units/kg EPO subcutaneously three times weekly), and a combination of PRP and EPO treatment (1 mL PRP twice weekly and 300 units/kg EPO three times weekly). Diabetes was induced using streptozotocin (65 mg/kg), and treatments were administered over four weeks. Serum markers for renal and hepatic function, oxidative stress indices, and histopathological changes in liver and kidney tissues were assessed.

Results: In diabetic rats, serum urea, creatinine, and liver enzymes (ALT, AST, ALP, LDH, GGT) increased significantly ($p<0.05$), while treatment with PRP, EPO, or both significantly reduced ALT, AST, ALP, LDH, and GGT (except in the combined group for GGT; $p>0.05$). Renal and hepatic SOD decreased significantly in diabetes ($p<0.05$) but improved with treatment, especially in the combined group ($p<0.05$). Renal TAC increased in diabetes and decreased significantly after treatment ($p<0.05$). No significant changes were observed in GPX or MDA levels ($p>0.05$). Damage in kidney and liver tissues were obtained in diabetic group. Histopathological improvements were evident, in hepatocyte integrity and glomerular structure, with EPO and PRP treatments.

Conclusion: The results emphasize the potential of EPO and PRP as complementary therapeutic strategies to reduce diabetes-induced oxidative damage and structural deterioration in liver and kidney tissues. However, further adjunctive approaches are required to achieve comprehensive organ protection.

Keywords:
Platelet-rich plasma
Erythropoietin
Diabetes
Kidney
Liver

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Received 23 January 2025; Revised from 1 June 2024; Accepted 13 May 2025

Citation: Ahooghalandari SB, Jalali SM, Jamshidian J, Rezaie A. Comparison of the effects of platelet-rich plasma and erythropoietin on nephropathy and hepatopathy in streptozotocin-induced diabetic rats: a biochemical, oxidative stress, and histopathological study. Physiology and Pharmacology 2026; 30: 70-80.

Introduction

Diabetes mellitus is a chronic metabolic disorder characterized by persistent hyperglycemia due to impaired insulin secretion or action (Unnikrishnan et al., 2016). Both type 1 and type 2 diabetes are associated with long-term complications, including damage to the kidneys and liver (Kottaisamy et al., 2021). Diabetic nephropathy and liver dysfunction in diabetes involve complex mechanisms such as oxidative stress, chronic inflammation, mitochondrial dysfunction, and activation of pro-apoptotic pathways (Yu & Bonventre, 2018; Targher et al., 2018).

Oxidative stress plays a central role in diabetic tissue injury, leading to increased production of reactive oxygen species (ROS) and lipid peroxidation, marked by elevated levels of malondialdehyde (MDA), and reduced antioxidant enzyme activities, including superoxide dismutase (SOD) and glutathione peroxidase (GPX). These processes are regulated by molecular pathways such as the Nrf2/ARE antioxidant response pathway, the NF-κB inflammatory pathway, and PI3K/Akt signaling pathway, which is crucial for cell survival and tissue repair.

Erythropoietin (EPO), originally known for its role in erythropoiesis, also exhibits cytoprotective, anti-apoptotic, and anti-inflammatory properties via modulation of the aforementioned pathways, particularly PI3K/Akt and NF-κB (Nekoui & Blaise, 2016; Kavas & Yazihan, 2010). It has shown protective effects in renal and hepatic ischemia-reperfusion injuries and other models of organ damage.

Platelet-rich plasma (PRP) is an autologous concentrate of platelets that releases growth factors such as VEGF, PDGF, FGF, and EGF upon activation. These factors contribute to tissue regeneration by enhancing angiogenesis, cell proliferation, and repair processes (Alves & Grimalt, 2018). Emerging evidence suggests that PRP also modulates oxidative stress and inflammation by influencing key molecular pathways like NF-κB and Nrf2 (Arif et al., 2022; Samy et al., 2020).

Considering the overlapping protective mechanisms of EPO and PRP, including their impact on oxidative stress, inflammatory signaling, and cellular repair, this study aimed to investigate their individual and combined effects on renal and hepatic injury in a streptozotocin-induced diabetic rat model, with a focus on biochemical and histopathological outcomes.

Materials and Methods

Animals

A total of 30 male Wistar rats, weighing approximately the same, were selected and randomly divided into five equal groups, with six rats in each group. The rats had unrestricted access to food and water and were housed in a controlled environment at a temperature of 23 ± 2 °C and humidity of 24 ± 6%. The light and dark cycles were maintained at 12 hours each.

The groups received the following treatments:

A. Control Group: Rats received a subcutaneous injection of 0.5 ml of normal saline.

B. Diabetic Control Group: Diabetes was induced in these rats through an intraperitoneal injection of streptozotocin at a dose of 65 mg/kg.

C. Diabetic PRP Group: Exogenous PRP was administered subcutaneously to diabetic rats two times a week for a duration of four weeks (Zarin et al., 2019).

D. Diabetic EPO Group: EPO was administered subcutaneously to diabetic rats at a dose of 300 units/kg three times a week for four weeks (Eren et al., 2016).

E. Diabetic PRP and EPO Group: In these diabetic rats, EPO and PRP were administered subcutaneously three times a week and twice a week, respectively, for four weeks (Zarin et al., 2019).

It is important to note that a glucose level higher than 250 mg/dL was used as an indicator to confirm diabetes (Wang-Fischer and Garyantes 2018). In addition, specified treatments (PRP, EPO, or combination) started at 72 hours post-diabetes induction.

PRP Preparation

Twenty male Wistar rats were used for the study. The rats were sedated using a combination of ketamine and xylazine (10.5 mg/kg), and whole blood was collected via cardiac puncture using sodium citrate as an anticoagulant. The blood was subsequently centrifuged at 1,000 rpm for 15 minutes, resulting in three distinct layers: plasma (upper layer), buffy coat (middle thin layer), and red blood cells (lower layer). The plasma and the buffy coat were transferred to a sterile tube and centrifuged again at 3,000 rpm for 5 minutes at room temperature. The upper two-thirds of the resulting supernatant, which comprised platelet-poor plasma (PPP), was discarded. The residual lower one-third was designated as PRP. To determine the platelet count, the PRP was mixed with phosphate-buffered saline (PBS) and counted per unit

TABLE 1: Mean ± SE of serum biochemical analytes in different groups.

	Control A	Diabetic Control B	Diabetic PRP C	Diabetic EPO D	Diabetic PRP and EPO E
Urea (mg/dl)	31.60±1.50	53.00±1.00 [a]	49.33±1.35 [a]	45.33±2.53 [a]	52.60±2.22 [a]
Cr (mg/dl)	0.38±0.02	0.58±0.02 [a]	0.50±0.01 [a]	0.54±0.01 [a]	0.51±0.01 [a]
ALT (U/L)	37.05±3.56	178.50±1.50 [a]	93.16±26.42 [a,b]	115.52±12.90 [a,b]	129.11±16.74 [a,b]
AST (U/L)	12.25±3.56	281.50±11.50 [a]	166.27±40.70 [a,b]	161.16±59.48 [a,b]	179.25±52.66 [a,b]
ALP (U/L)	254.40±53.73	6572.58±127.54 [a]	4135.16±840.96 [a,b]	4156.72±763.51 [a,b]	5317.43±517.22 [a,b]
LDH (U/L)	865.40±157.73	3265.29±135.32 [a]	1470.36±271.85 [b]	1535.51±360.68 [b]	1497.82±277.10 [b]
GGT (U/L)	11.25±3.27	14.50±0.50 [a]	9.22±2.55 [b]	8.66±1.28 [b]	16.39±6.01

Note: a, significant difference compared to control group ($p<0.05$); b, significant difference compared to diabetic control group ($p<0.05$). Abbreviations: Cr, creatinine; ALT, alanine aminotransferase; AST, aspartate aminotransferase; ALP, alkaline phosphatase; LDH, lactate dehydrogenase; GGT, gamma-glutamyl transferase.

volume. The platelet count of the prepared PRP was 1.5–2.2 × 10^6/μL, representing a 4- to 5-fold increase compared to baseline peripheral blood levels. Ultimately, the prepared PRP was frozen at -70°C until subsequent use (Zarin et al., 2019).

Sampling

Once the treatment period ended, after anesthesia with ketamine and xylazine, blood samples were collected from the rats' hearts into standard collection tubes. Serum samples were then separated through centrifugation and kept at -70°C until the experiments were conducted.

After euthanasia, the rats were necropsied, and their liver and kidney organs were extracted and also stored in a freezer at -70°C. Additionally, portions of the liver and kidney were fixed in 10% buffered formalin for histopathological examination.

Laboratory analysis

Serum samples were analyzed for indicators of kidney function, including urea and creatinine levels, in addition to liver enzyme activity comprising AST, ALT, ALP, and LDH. This analysis utilized kits from Biorex Fars Company in Iran and was conducted using a Biotecnica biochemical autoanalyzer (BT-1500) from Italy.

To evaluate oxidative stress, the activities of SOD and GPX enzymes were quantified with Randox kits (England). Additionally, TAC and MDA concentration were assessed using Zellbio kits (Germany) in renal and hepatic tissue samples and were reported relative to tissue protein content.

Histopathologic assessment

Tissue samples from kidney and liver were fixed in 10% neutral buffered formalin for 48 hours, dehydrated through a graded ethanol series, cleared in xylene, and embedded in paraffin. Sections of 5 μm thickness were cut using a microtome, mounted on glass slides, and stained with hematoxylin and eosin (H&E) for histopathological evaluation under a light microscope, following standard protocols (Bancroft & Gamble, 2023).

Liver and kidney tissue specimens were examined for histopathological changes following sectioning and staining with hematoxylin and eosin (H&E).

Statistical analysis methods

Test data from different groups are presented as mean ± standard error. The normality of the data distribution was assessed using the Shapiro-Wilk test. A one-way ANOVA with a subsequent Tukey post-test was utilized to analyze the differences between group means. A P value of less than 0.05 was regarded as statistically significant.

Results

Serum biochemical analysis

According to Table 1, serum urea and creatinine levels were significantly increased in all diabetic groups (B, C,

TABLE 2: Mean ± SE of renal tissue oxidant/antioxidant markers in different groups.

	Control **A**	Diabetic Control B	Diabetic PRP C	Diabetic EPO D	**Diabetic PRP and EPO E**
SOD (U/gpr)	1559.04±84.54	493.74±105.90 [a]	982.12±191.99 [b]	1158.01±165.23 [b]	1359.20±181.67 [b]
GPX (U/mgpr)	53.16±12.91	55.54±0.37	59.34±8.09	53.29±1.62	53.80±3.82
TAC (mM/gpr)	1.16±0.15	1.38±0.10 [a]	1.16±0.09 [b]	1.13±0.11 [b]	1.16±0.08 [b]
MDA (nmol/gpr)	191.53±10.72	212.33±8.81	221.42±12.58	217.54±7.7	209.47±11.24

Abbreviations: SOD, superoxide dismutase; GPX, glutathione peroxidase; TAC, total antioxidant capacity; MDA, malondialdehyde; gpr, gram of protein in the tissue.
Note: a, significant difference compared to control group ($p<0.05$); b, significant difference compared to diabetic control group ($p<0.05$).

D, and E) in comparison to the control group ($p<0.05$). In the groups treated with PRP, EPO, or a combination of both, there was a minor insignificant decrease in these indices ($p>0.05$).

Regarding serum enzyme activity, all enzymes studied, including ALT, AST, ALP, LDH, and GGT, showed a significant increase in activity in the diabetic group compared to the control group ($p<0.05$) (Table 1). The activity levels of ALT, AST, ALP, and LDH in the treated diabetic groups were significantly lower than those in the diabetic control group (group B) ($p<0.05$). Additionally, a significant decrease in serum GGT activity was observed in the groups treated with PRP or EPO (Groups C and D) compared to the untreated diabetic group; however, there was no significant difference in the combined treatment group (Group E) compared to the diabetic control group ($p>0.05$).

It is important to note that among the treated diabetic groups, the maximum levels of ALT, AST, ALP, and GGT enzyme activity were found in the combined treatment group. Nevertheless, no significant differences were noted when compared to the other treatment groups ($p>0.05$).

Renal tissue oxidant/antioxidant analysis

According to Table 2, which outlines the status of oxidant and antioxidant indices in renal tissue, the level of SOD enzyme activity in the untreated diabetic group was significantly lower than that in the control group, reaching only one-third of the control group's level ($p<0.05$). In all treated groups, there was a significant increase in SOD activity compared to the diabetic control group (group B) ($p<0.05$). Among the treated groups, the highest SOD activity was observed in the group receiving the combined treatment of PRP and EPO (group E), while the lowest activity was found in the group treated only with PRP. The difference in SOD levels between these two groups was statistically significant ($p<0.05$).

Additionally, the TAC level in the diabetic group (group B) was significantly higher than in the control group ($p<0.05$). However, in the treated diabetic groups, there was a significant decrease in TAC compared to the untreated group ($p<0.05$). There was no significant difference in TAC levels among the various treatment groups ($p>0.05$) (Table 2).

Regarding GPX and MDA levels, no significant differences were found among the groups ($p>0.05$) (Table 2).

Hepatic tissue oxidant/antioxidant analysis

Upon examining the oxidant/antioxidant indices in hepatic tissue (Table 3), a significant decrease was found in SOD activity in the diabetic control group (Group B) compared to the control group ($p < 0.05$). Among the treated groups, only the combined treatment group (Group E) demonstrated a significant increase in SOD levels compared to the untreated diabetic group ($p < 0.05$).

Additionally, GPX activity was significantly reduced in all diabetic groups when compared to the control group ($p < 0.05$). However, no significant differences were observed between the untreated and treated diabetic groups ($p > 0.05$) (Table 3).

Furthermore, there were no significant differences in serum MDA and TAC values among the study groups ($p > 0.05$).

TABLE 3: Mean ± SE of hepatic tissue oxidant/antioxidant markers in different groups.

	Control A	Diabetic Control B	Diabetic PRP C	Diabetic EPO D	**Diabetic PRP and EPO** E
SOD (U/gpr)	2329.1±302.47	1476.41±743.85 [a]	1153.14±245.16 [a]	1436.78±545.85 [a]	3296.48±1113.24 [b]
GPX (U/mgpr)	49.44±15.1	32.4±0.4 [a]	32.41±2.1 [a]	44.91±2.9 [a]	33.88±2.1 [a]
TAC (mM/gpr)	1.72±0.06	1.55±0.0	1.37±0.05	1.61±0.15	1.49±0.09
MDA (nmol/gpr)	131.48±23.59	114.57±5.29	110.4±5.91	147.15±6.54	120.78±4.81

Note: a, significant difference compared to control group ($p<0.05$); b, significant difference compared to diabetic control group ($p<0.05$). Abbreviations: SOD, superoxide dismutase; GPX, glutathione peroxidase; TAC, total antioxidant capacity; MDA, malondialdehyde; gpr, gram of protein in the tissue.

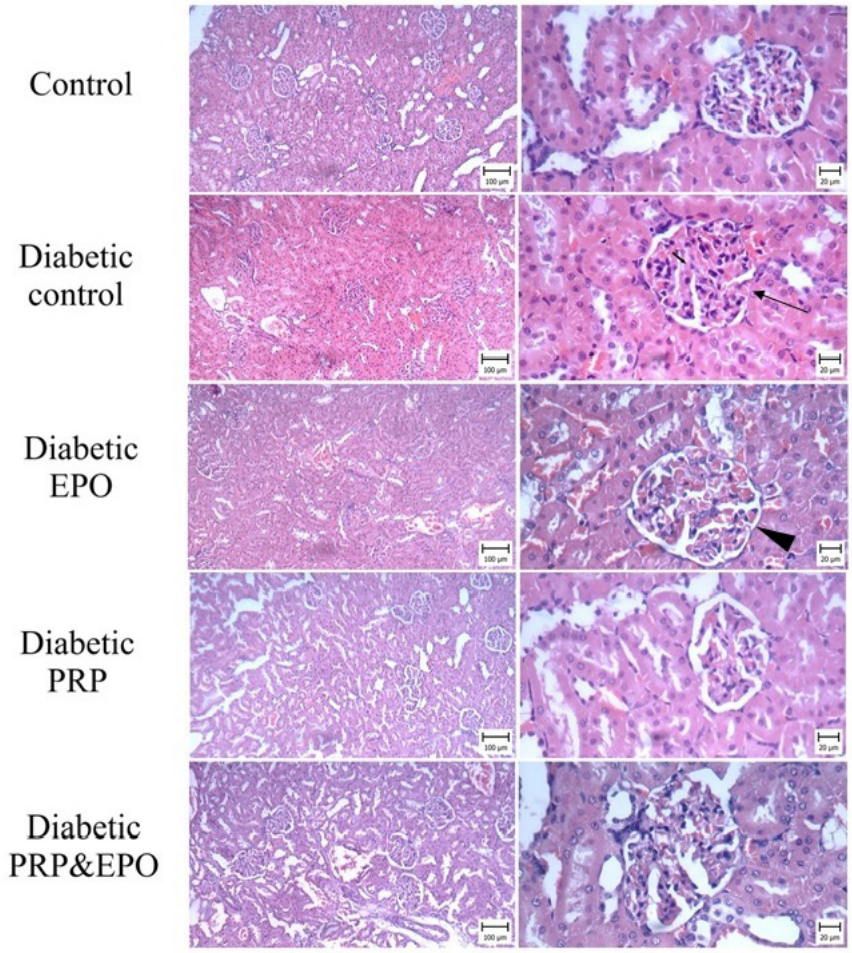

FIGURE 1. H&E-stained sections of kidney tissue in different groups (six rats in each group): Diabetic control, diabetic EPO, diabetic PRP, and diabetic PRP & EPO group. Glomerular damage is characterized by glomerulonephritis and proteinuria (arrow). Diabetic EPO group exhibited less glomerular damage (arrow head). A scale bar is provided alongside each histopathological image.

Histopathology

Histopathological studies indicate that glomerular damage, characterized by glomerulonephritis and proteinuria, was observed in the diabetic control group (Group B) as well as in the treatment groups (C, D, and E) when compared to the control group. Notably, the erythropoietin treatment group exhibited less glomerular damage (Figure 1).

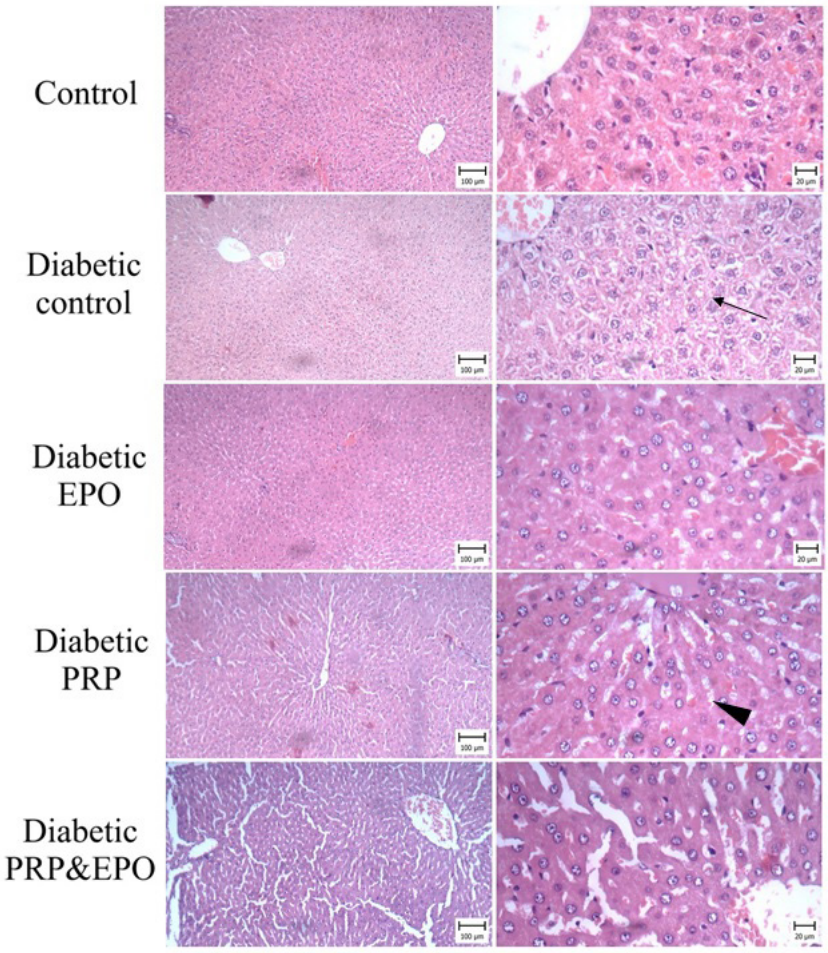

FIGURE 2. H&E-stained sections of liver tissue in different groups (six rats in each group). Diabetic control group: revealed cellular degeneration (arrow). Diabetic EPO, diabetic PRP and diabetic PRP & EPO group: demonstrated improvements in hepatocyte condition, with no signs of degeneration (arrow head). A scale bar is provided alongside each histopathological image.

In the assessment of liver tissue in the patient group, the histopathological evaluation revealed cellular degeneration in the untreated diabetic group (Group B). In contrast, the treatment groups demonstrated improvements in hepatocyte condition, with no signs of degeneration (Figure 2).

Discussion

Diabetes mellitus (DM) is a long-term metabolic condition marked by elevated blood sugar levels, which triggers oxidative stress and inflammation, leading to widespread tissue damage, including in the kidneys and liver (Cole and Florez 2020). The interaction between oxidative stress and impaired antioxidant defenses is a crucial factor in the development of diabetic nephropathy and hepatopathy (Wang and Zhang 2024; Mobasheri et al., 2023). This study evaluates the effects of PRP and EPO, both separately and in combination, on biochemical markers, oxidative-antioxidant balance, and histopathological changes in diabetic rats. By addressing the multifaceted nature of diabetic complications, this discussion explores how these treatments influence organ function and structural integrity, providing insights into their therapeutic potential and mechanisms of action.

The combined analysis of kidney biochemical, oxidative-antioxidant, and histopathological results in this study, highlights the complex impact of diabetes-induced oxidative stress on renal function and structure. The significant elevation in serum urea and creatinine levels in all diabetic groups, including untreated and treated rats, indicates renal dysfunction due to impaired filtration capacity. Histopathological assessments revealed glomerular damage (glomerulonephritis) and proteinuria in the diabetic control group, corroborating these biochemical

abnormalities. Similar findings of glomerular structural damage in diabetic nephropathy have been reported in previous studies, where hyperglycemia-induced oxidative stress was identified as the primary cause of glomerular endothelial dysfunction and increased permeability (Darenskaya et al., 2023).

The oxidative stress profile further supports these observations. The reduction in SOD activity in untreated diabetic rats reflects impaired antioxidant defenses, consistent with findings from prior research showing decreased SOD and GPX activity in diabetic kidneys due to excessive reactive oxygen species (ROS) production (Vodošek et al., 2020; Sakashita et al., 2021). Treatment with PRP, EPO, or their combination significantly restored SOD activity, particularly in the combined treatment group, indicating an improvement in antioxidant defenses. The renoprotective effects of EPO align with studies demonstrating its ability to activate redox-sensitive signaling pathways, reduce oxidative stress, and enhance endothelial stability, which may explain its effectiveness in reducing glomerular damage observed histologically (Wu and Yang 2021; Altun et al., 2020).

The histopathological results in this study further highlight EPO's superior efficacy in mitigating structural kidney damage, showing less glomerular injury compared to other groups. These findings align with studies by Zeng et al. (2023) and Wang et al. (2023), which reported that EPO reduces glomerular sclerosis and preserves podocyte integrity in diabetic nephropathy models (Zeng et al., 2023; Wang et al., 2023). PRP, known for its growth factors that promote cellular repair and angiogenesis, also contributed to renal protection but was less effective in reducing glomerular damage than EPO.

However, despite these improvements, the persistent reduction in GPX activity and unchanged MDA levels across groups suggest that lipid peroxidation and oxidative damage remain inadequately addressed. This limitation aligns with findings from Mallik et al., (2024), who reported that hyperglycemia-induced oxidative damage in diabetes is often profound and difficult to reverse, even with antioxidant therapies (Mallik et al., 2024).

Taken together, the biochemical, oxidative-antioxidant, and histopathological results demonstrate that while PRP, EPO, and their combination can partially alleviate oxidative stress and reduce renal injury, structural and functional damage in diabetic kidneys remains incompletely reversed. These findings emphasize the need for prolonged treatment durations or adjunctive therapies targeting additional antioxidative pathways to fully address renal damage in diabetes (Młynarska et al., 2024; Zoja et al., 2020).

The combined biochemical, oxidative-antioxidant, and histopathological findings highlight the profound impact of diabetes-induced oxidative stress on liver function and structure. Serum levels of liver enzymes, including ALT, AST, ALP, LDH, and GGT, were significantly elevated in the diabetic control group, reflecting hepatic damage and dysfunction. This increase corresponds to previous studies showing hyperglycemia-induced oxidative stress as a driver of hepatocellular injury, leading to the leakage of enzymes into the bloodstream (Bhatti et al., 2022; Dey and Lakshmanan 2013).

Histopathological evaluation of liver tissues further supported these biochemical findings, with cellular degeneration observed in the diabetic control group. The untreated diabetic group exhibited hepatocyte degeneration consistent with diabetic hepatopathy, a condition characterized by steatosis, fibrosis, and apoptosis due to oxidative damage and inflammatory responses (Palma et al., 2022; Singh et al., 2024).

Treatment with PRP, EPO, or their combination demonstrated a protective effect, as indicated by reduced serum enzyme levels and improved histopathological findings. The treated groups showed no signs of cellular degeneration, suggesting that these therapies supported hepatocyte recovery and reduced oxidative injury. This aligns with previous research indicating that PRP, rich in growth factors such as VEGF and PDGF, promotes tissue regeneration and hepatocyte repair (Shivaramu et al., 2024). Similarly, EPO has been demonstrated to exert antioxidative, anti-inflammatory, and anti-apoptotic effects on the liver, mitigating oxidative damage and improving cellular function (Liu et al., 2015; Peng et al., 2020).

In terms of oxidative stress markers, the significant reduction in SOD and GPX activity in the diabetic control group underscores the disruption of the antioxidant defense system in diabetes, in line with prior studies (Mobasheri et al., 2023; Ghasemi et al., 2020). Treatment, particularly with the combined PRP and EPO therapy, significantly improved SOD activity, reflecting enhanced antioxidative capacity. Notably, the combined therapy was the only group that significantly restored

SOD levels compared to the untreated diabetic group, implying a synergistic effect. However, the persistent reduction in GPX activity and unchanged MDA and TAC levels across all groups indicate that the therapies were not fully effective in reversing lipid peroxidation or restoring total antioxidant capacity, a limitation commonly reported in studies on diabetic oxidative stress management (Singh et al., 2024).

The histopathological evidence of hepatocyte improvement in treated groups further corroborates the biochemical and antioxidant findings. EPO's known role in modulating oxidative stress and inflammation likely contributed to its effectiveness, while PRP's regenerative properties facilitated cellular repair. These findings are consistent with studies by Liu et al., (2015) and Elbaset et al., (2023), which demonstrated the hepatoprotective effects of EPO and PRP in liver damage caused by diabetes (Liu et al., 2015; Elbaset et al., 2023).

In comparing the mechanisms by which PRP and EPO exert their therapeutic effects, it becomes evident that they operate through distinct yet potentially complementary biological pathways. PRP primarily facilitates tissue regeneration through the localized release of a wide array of growth factors, such as platelet-derived growth factor (PDGF), transforming growth factor-beta (TGF-β), and vascular endothelial growth factor (VEGF), which stimulate cell proliferation, migration, angiogenesis, and extracellular matrix remodeling (Pineda-Cortel et al., 2024; Zhu et al., 2024). These effects are particularly prominent in fibroblasts, mesenchymal stem cells, and endothelial cells, thereby promoting repair and regeneration at the site of injury. In contrast, EPO functions mainly as an anti-inflammatory and cytoprotective agent by binding to erythropoietin receptors (EPORs) expressed on various non-hematopoietic cells, including immune and endothelial cells. Through activation of downstream signaling pathways such as JAK2/STAT5, PI3K/Akt, and MAPK, EPO downregulates pro-inflammatory cytokines like tumor necrosis factor-alpha (TNF-α) and interleukin-6 (IL-6), while upregulating anti-inflammatory mediators such as interleukin-10 (IL-10) (Mateus et al., 2023; Arik et al., 2022). Additionally, EPO reduces apoptosis and oxidative stress, and enhances phagocytosis, contributing to tissue preservation rather than direct regeneration (Bin et al., 2023; Arik et al., 2022). Thus, while PRP promotes cellular repair through growth factor-mediated proliferation and matrix remodeling, EPO exerts its effects primarily by modulating inflammation and enhancing cell survival, highlighting their distinct but potentially synergistic roles in therapeutic applications.

In summary, the combination of biochemical, oxidative-antioxidant, and histopathological assessments demonstrates that PRP and EPO therapies mitigate liver damage by enhancing antioxidant defenses and promoting hepatocyte recovery. While these treatments show promise, the incomplete restoration of certain oxidative markers highlights the need for optimized or adjunctive strategies to fully address diabetes-related hepatic injury.

Conclusion

The findings demonstrate that diabetes-induced oxidative stress significantly impairs kidney and liver function, as evidenced by elevated serum biochemical markers, reduced antioxidant enzyme activity, and histopathological damage. While treatment with PRP, EPO, or their combination partially mitigated oxidative stress, improved antioxidant defenses (notably SOD activity), and facilitated structural recovery, the restoration of overall renal and hepatic function was incomplete. Histopathological improvements, particularly in hepatocytes and glomerular structure, were more pronounced in the EPO-treated groups, highlighting its potent antioxidative and cytoprotective properties. These results underscore the potential of EPO and PRP therapies in addressing diabetic complications, though adjunctive strategies targeting persistent oxidative damage are needed for comprehensive organ protection.

Acknowledgements

The authors would like to thank the Vice Chancellor of Research and Technology of Shahid Chamran University of Ahvaz for the financial support of this project.

Conflicts of interest

The authors declare that there is no conflict of interest.

Ethics approval

The experimental procedures were performed with the approval of Shahid Chamran university of Ahvaz laboratory Animal Care and Use Committee (Ethical code: EE/1401.2.24.226815/scu.ac.ir).

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