Check for updates

Original Research Article

Gardenin A Alleviates Diabetic Nephropathy by Modulating Oxidative Stress and Inflammation in Experimental Rats

Pharmacognosy Magazine
22(4) 1651–1660, 2026
© The Author(s) 2026
Article reuse guidelines:
in.sagepub.com/journals-permissions-india
DOI: 10.1177/09731296261464762
journals.sagepub.com/home/phm
Sage

Juan Ji[1,2], Longhai Song[3], Chong Yang[3], Yan Gao[3], Song Li[3] and Qiaoying Wu[1,2]

Abstract

Background: Diabetic nephropathy is a formidable microvascular complication of diabetes mellitus, considerably contributing to global morbidity and mortality due to its role as the primary factor of end-stage renal disease.
Objectives: This work was focused on evaluating the therapeutic effects of gardenin A on the streptozotocin (STZ)-induced diabetic nephropathy in rats.
Materials and Methods: Diabetic nephropathy was initiated in the experimental rats by administering 65 mg/kg of STZ. The rats with diabetic nephropathy were subsequently treated with gardenin A for a period of 10 weeks. Following the completion of the treatments, the body weight, glucose, and glycated hemoglobin (HbA_{1C}) levels were evaluated. The renal dysfunction markers creatinine, urea, and uric acid levels were evaluated in the experimental rats. The concentrations of oxidative stress markers and pro-inflammatory markers were evaluated in the kidney tissues of the rats. The kidneys of the rats were subjected to histopathological analysis.
Results: Gardenin treatment markedly decreased body weight, glucose, and HbA_{1C} levels in rats with diabetic nephropathy. Moreover, gardenin A treatment decreased the levels of renal dysfunction markers in the diabetic rats. The gardenin A treatment also markedly reduced inflammatory cytokine levels and oxidative stress response by elevating antioxidant concentrations in STZ-induced rats. Additionally, the results of histological analysis confirmed the therapeutic efficacy of gardenin A against diabetic nephropathy.
Conclusion: The present study suggests that gardenin A treatment may ameliorate diabetic nephropathy in STZ-induced rats. Thus, it was evident that gardenin A may have a therapeutic effect against diabetic nephropathy.

Keywords

Gardenin A, diabetic nephropathy, creatinine, malondialdehyde
Received 31 October 2025; **revised** 27 January 2026; **accepted** 21 May 2026

Introduction

Diabetic nephropathy, a serious microvascular complication of diabetes mellitus, is defined by advanced structural and functional injury to the kidneys, resulting in chronic renal dysfunction and eventually end-stage renal disease (ESRD). This debilitating condition significantly contributes to high morbidity and mortality worldwide, often necessitating renal replacement therapies like dialysis or transplantation (Shan et al., 2024). The global prevalence of diabetes mellitus, a primary driver of diabetic nephropathy, has escalated dramatically, with predictions suggesting an increase to 642 million affected individuals by 2040. Consequently, diabetic nephropathy has appeared as the major cause of chronic kidney disease globally, affecting roughly 40% of all diabetic patients

Juan Ji and Longhai Song contributed equally to the work and can be considered co-first authors.
[1]Department of Nephrology, Affiliated Hospital of Hebei University, Baoding, Hebei, China
[2]Department of Key Laboratory of Bone Metabolism and Physiology in Chronic Kidney Disease of Hebei Province, Baoding, Hebei, China
[3]Urology Department, Affiliated Hospital of Hebei University, Baoding, Hebei, China

Corresponding authors:
Qiaoying Wu, Department of Nephrology, Affiliated Hospital of Hebei University, Baoding, Hebei 071000, China; Department of Key Laboratory of Bone Metabolism and Physiology in Chronic Kidney Disease of Hebei Province, Baoding, Hebei 071000, China.
E-mail: 15512285236@163.com
Song Li, Urology Department, Affiliated Hospital of Hebei University, Baoding, Hebei 071000, China.
E-mail: 15830201117@163.com

Creative Commons Non Commercial CC BY-NC: This article is distributed under the terms of the Creative Commons Attribution-NonCommercial 4.0 License (http://www.creativecommons.org/licenses/by-nc/4.0/) which permits non-Commercial use, reproduction and distribution of the work without further permission provided the original work is attributed as specified on the SAGE and Open Access pages (https://us.sagepub.com/en-us/nam/open-access-at-sage).

(Hu et al., 2025). Type 2 diabetes is responsible for most of the diabetic individuals, and a pronounced increase in diabetic nephropathy is observed in this population. This complex disorder is characterized by intricate pathophysiological mechanisms involving inflammatory processes, genetic predispositions, and epigenetic modifications, which are frequently exacerbated by comorbidities such as hypertension and dyslipidemia (Zhang et al., 2025).

Diabetic nephropathy is also characterized by the overaccumulation of extracellular matrix within the tubulointerstitial and glomerular compartments, along with thickening of the intrarenal vasculature. These structural alterations are primarily driven by chronic hyperglycemia, which instigates a series of cellular events and signaling pathways that culminate in renal injury. The early stages of nephropathy are characterized by glomerular hyperfiltration, which progresses to albuminuria, a decrease in glomerular filtration rate, and, subsequently, renal fibrosis (Raval et al., 2020). The onset of diabetic nephropathy is multifactorial, primarily driven by hyperglycemia-induced inflammatory processes that affect both hemodynamic and metabolic pathways. This involves chronic low-grade inflammation and activation of the immune system, resulting in elevated serum levels of inflammatory cytokines (Moreno et al., 2018). These inflammatory mediators contribute to glomerular and tubulointerstitial damage, exacerbating renal dysfunction and development to ESRD. Moreover, persistent hyperglycemia induces oxidative stress, which further contributes to renal injury by generating reactive oxygen species that damage cellular components and activate pro-inflammatory signaling pathways, thereby accelerating the decline in renal function (Jin et al., 2023).

Despite advancements in therapeutic approaches, a considerable number of individuals with diabetic nephropathy inevitably develop ESRD, requiring either dialysis or kidney transplantation. This progression highlights the current limitations in preventing irreparable renal damage, especially given the complex interplay of oxidative stress, inflammation, and fibrotic mechanisms underlying diabetic nephropathy pathology (Jung & Yoo, 2022). Existing clinical interventions, including glucose-lowering medications and renin-angiotensin system inhibitors, frequently fail to prevent the inexorable development of diabetic nephropathy, leading to a persistent demand for improved and innovative therapeutic modalities. Consequently, there is an urgent clinical need for new therapies that target the multifaceted mechanisms underlying diabetic nephropathy, moving beyond symptomatic management to achieve disease modification (Yamazaki et al., 2021). Gardenin A is a major bioactive flavonoid compound widely present in various medicinal plants, including *Gardenia resinifera* and *Gardenia gummifera* plants. Several previous studies have reported that gardenin A has many biological activities, including anti-tumor (Cabrera et al., 2016), neuroprotective (Alonso-Castro et al., 2020), hepatoprotective and anti-hyperlipidemic (Toppo et al., 2017), and anti-parkinsonian (Hack et al., 2024). However, there is no such evidence for its therapeutic effects against diabetes, and diabetic nephropathy has been reported. Hence, this work focused on evaluating the therapeutic effects of gardenin A against streptozotocin (STZ)-induced diabetic nephropathy in rats.

Materials and Methods

Experimental Rats

The 6-week-old male Sprague–Dawley rats weighing about 230–250 g were employed in this work, which were housed in sterilized polypropylene cages under laboratory settings, with a temperature range of 22 °C–26 °C and humidity of 40%–60%. A 12-h cycle of alternating light and dark was established. During the study period, all rats had free access to a standard diet (Bio-Serv, USA) and purified drinking water. Prior to starting the experiments, all rats were assigned a 7-day acclimation period in a laboratory environment.

Experimental Groups

Following a 1-week acclimatization period, rats were divided into four groups, each consisting of six rats ($n = 6$). Group I was the control, which was administered with normal saline solution. Group II rats received a single administration of STZ at a 65 mg/kg dosage to initiate diabetic nephropathy. The rats were given glucose (0.5%) to prevent death from acute hypoglycemia. After a 3-day administration of STZ, blood glucose was evaluated, and rats with a glucose level exceeding 250 mg/dL were marked as diabetic and designated for additional evaluations. The Group III rats were undergone to STZ induction as specified in Group II and were treated with 50 mg/kg of gardenin A for 10 weeks. In Group IV, rats underwent STZ induction as specified in Group II and were treated with 5 mg/kg of gliclazide, a standard drug, for a duration of 10 weeks. The body weight of each rat was measured using a sensitive electronic balance. The glucose level was assessed utilizing a commercial glucometer (Roche, Germany). The animals were fasted overnight for 12 h before sacrification and blood sample collection.

Analysis of Glycated Hemoglobin and Kidney Function Marker Levels

The concentrations of HbA_{1C} in the experimental rats were evaluated using a commercial kit sourced from Elabscience, USA. The HbA_{1C} levels were assessed by measuring the absorbance at 450 nm. The levels of renal dysfunction markers, including urea, creatinine, and uric acid in the serum of experimental rats, were assessed using an assay kit obtained from Abcam, USA. Each test was performed with three replicates using the manufacturer's guidelines.

Assessment of Inflammatory Cytokines and Oxidative Stress Marker Levels

The removed kidney tissues from the experimental rats were homogenized with saline, and the suspension was centrifuged at 5,000 rpm for 20 min. The resulting supernatant was used to assess oxidative stress biomarkers using kits. The glutathione peroxidase (GPx), superoxide dismutase (SOD), catalase (CAT), and malondialdehyde (MDA) were assessed using kits (Abcam, USA). The concentrations of inflammatory cytokines interleukin (IL)-6 and tumor necrosis factor (TNF)-α in the renal tissue homogenates were evaluated using commercial kits (MyBioSource, USA). The experiments were conducted in triplicate using the manufacturer's procedures.

Histopathological Analysis

The kidney tissue samples were obtained and subjected to a 10% neutral formalin solution to assess the histopathological alterations in kidney architecture. Thereafter, the tissues were paraffinized, sectioned to 5 µm thickness, and stained with eosin-hematoxylin. Finally, the kidney tissues were examined microscopically to evaluate the histological changes.

Statistical Analysis

The data were assessed using GraphPad Prism (version 9.5.0), presented as a mean ± standard deviation (SD) of three replicates. The results are evaluated using one-way analysis of variance and subsequently Tukey's *post hoc* test, with significance established at $p < .05$.

Results

Effect of Gardenin A on Bodyweight, Glucose, and Glycated Hemoglobin Levels in Experimental Rats

The current results indicated that STZ-induced rats with diabetic nephropathy showed a substantial increase in body weight, glucose, and HbA_{1C} concentrations in comparison to the control. However, treatment with gardenin A at a dosage of 50 mg/kg markedly diminished the body weight, glucose, and HbA_{1C} concentrations in the STZ-induced rats with diabetic nephropathy (Figure 1A). Similarly, the standard drug gliclazide treatment also decreased these parameters, supporting the activity of gardenin A.

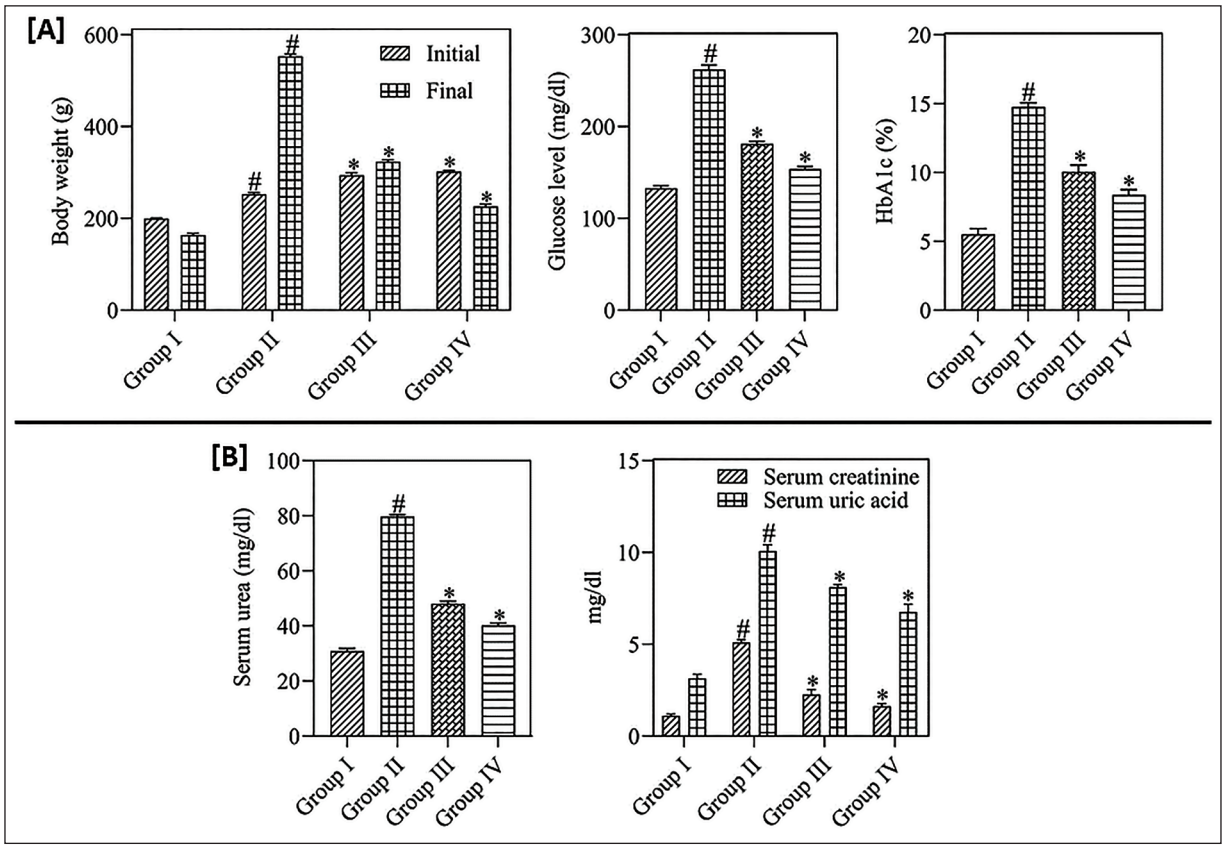


Figure 1. Effect of Gardenin A on the Bodyweight, Glucose, Glycated Hemoglobin (HbA_{1C}), and Kidney Function Marker Levels in the Experimental Rats. The Data Were Evaluated Using GraphPad Prism Software, and Values are Presented as Mean ± Standard Deviation (SD) of Triplicates. The Level of Statistical Significance for Treatment Groups Were Established as "#" $p < .01$ for Comparisons Between the Control (Group I) and Streptozotocin (STZ)-induced (Group II) Groups; "*" $p < .05$ for Comparisons Between the STZ-induced (Group II) and the Gardenin A (Group III) and/or Standard Drug Gliclazide-treated (Group IV) Groups.

Effect of Gardenin A on the Kidney Function Markers in the Experimental Rats

The current findings demonstrated a significant elevation in the renal dysfunction markers, including creatinine, urea, and uric acid levels in the serum of STZ-induced rats with diabetic nephropathy compared to controls (Figure 1B). Captivatingly, the treatment with a 50 mg/kg dosage of gardenin A to the STZ-induced rats resulted in a marked reduction in urea, creatinine, and uric acid in their serum. Supportively, the standard drug gliclazide treatment also decreased these renal dysfunction markers in the serum of STZ-induced rats.

Effect of Gardenin A on the Oxidative Stress Markers in Experimental Rats

The oxidative stress biomarker levels in the kidney tissues of rats were evaluated to determine the antioxidant effect of gardenin A (Figure 2). The experimental rats with diabetic nephropathy had a significant elevation in MDA levels, accompanied by a decrease in antioxidant GPx, SOD, and CAT concentrations related to the control. Whereas gardenin A treatment at a dosage of 50 mg/kg markedly decreased the MDA levels and subsequently increased the GPx, SOD, and CAT concentrations in the kidneys of diabetic rats. Furthermore, the treatment with gliclazide also decreased the MDA and increased the antioxidants in the kidneys of diabetic rats, which suggests the antioxidant activity of gardenin A.

Effect of Gardenin A on the Inflammatory Cytokine Levels in the Experimental Rats

Figure 3 illustrates the concentrations of pro-inflammatory cytokine levels in the kidney tissues of experimental rats. The experimental rats with diabetic nephropathy demonstrated a

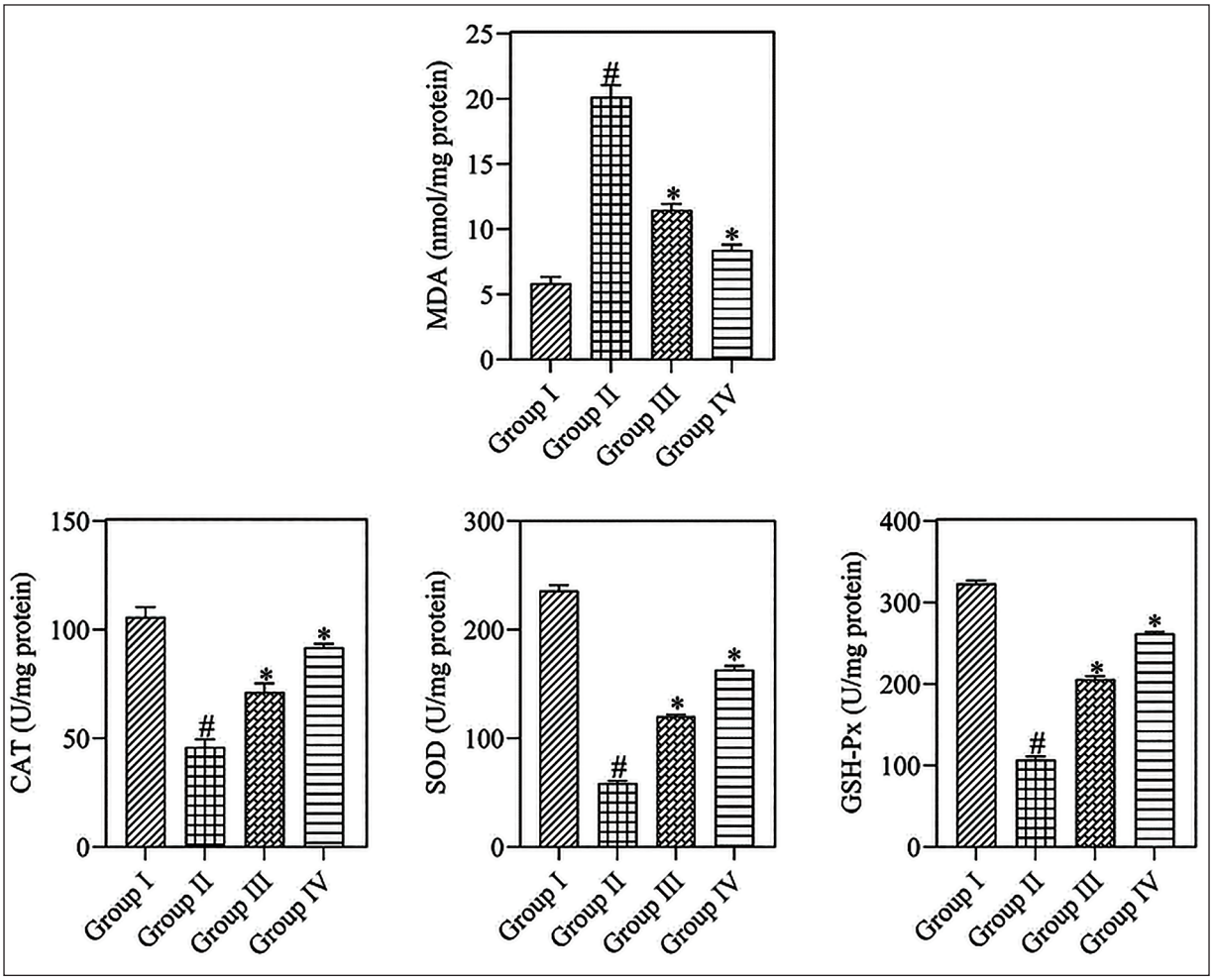


Figure 2. Effect of Gardenin A on the Oxidative Stress Marker Levels in the Experimental Rats. The Data Were Evaluated Using GraphPad Prism Software, and Values are Presented as Mean ± Standard Deviation (SD) of Triplicates. The Level of Statistical Significance for Treatment Groups Were Established as "#" $p < .01$ for Comparisons Between the Control (Group I) and Streptozotocin (STZ)-induced (Group II) Groups; "*" $p < .05$ for Comparisons Between the STZ-induced (Group II) and the Gardenin A (Group III) and/or Standard Drug Gliclazide-treated (Group IV) Groups.

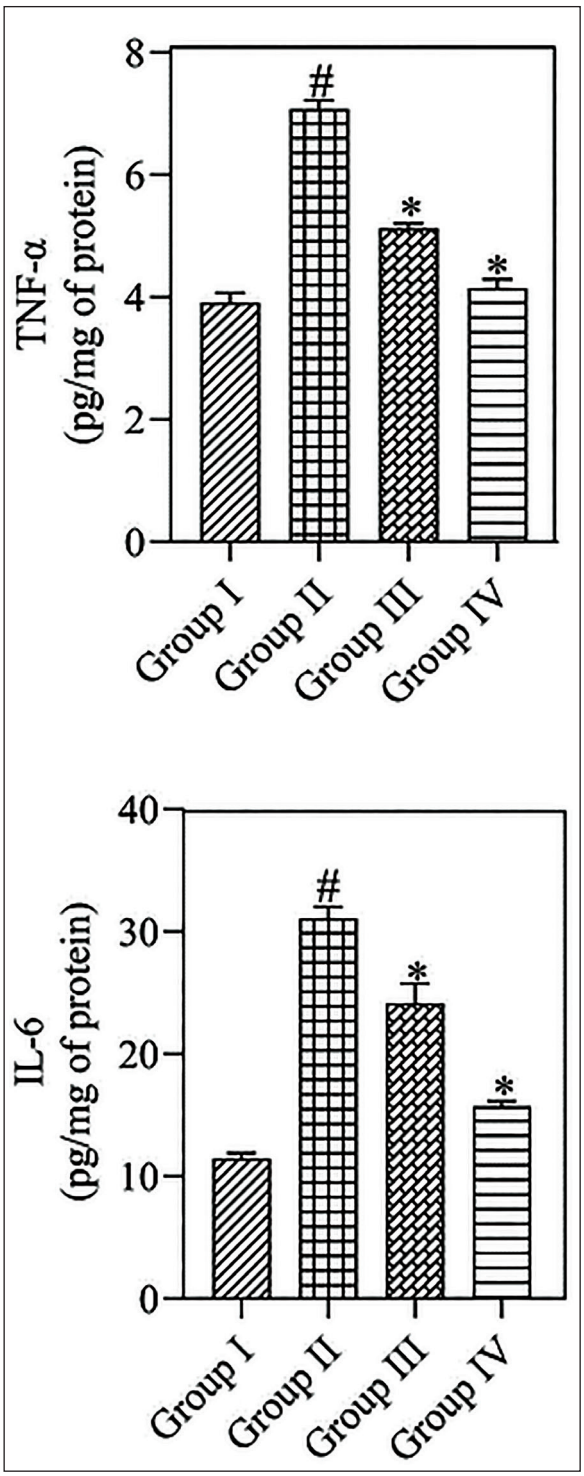


Figure 3. Effect of Gardenin A on the Inflammatory Cytokine Levels in the Experimental Rats. The Data Were Evaluated Using GraphPad Prism Software, and Values are Presented as Mean ± Standard Deviation (SD) of Triplicates. The Level of Statistical Significance for Treatment Groups Were Established as "#" $p < .01$ for Comparisons Between the Control (Group I) and Streptozotocin (STZ)-induced (Group II) Groups; "*" $p < .05$ for Comparisons Between the STZ-induced (Group II) and the Gardenin A (Group III) and/or Standard Drug Gliclazide-treated (Group IV) Groups.

significant increase in IL-6 and TNF-α levels in their kidneys compared to controls. Interestingly, the gardenin A treatment at a 50 mg/kg dosage showed a marked diminution in the IL-6 and TNF-α levels in the STZ-induced rats. Moreover, gliclazide treatment also considerably reduced these cytokines in the kidneys of diabetic rats, further validating the anti-inflammatory activity of gardenin A.

Effect of Gardenin A on the Kidney Tissue Histopathology of Experimental Rats

The histological examination was performed on the renal tissues of the experimental rats, with the results illustrated in Figure 4. The kidneys of control rats exhibited normal morphology with clear renal histoarchitectures. Conversely, the kidney tissues of the rats with diabetic nephropathy demonstrated glomerular shrinkage, damage to renal tubular epithelial cells, and increased infiltration of inflammatory cells when compared with the control. Fascinatingly, the gardenin A treatment at a 50 mg/kg dosage effectively mitigated these histological abnormalities in the renal tissues of diabetic rats. Furthermore, gliclazide treatment also reduced the histological abnormalities in the renal tissues of diabetic rats.

Discussion

Diabetic nephropathy represents a formidable microvascular complication of diabetes mellitus, significantly contributing to global morbidity and mortality due to its role as the foremost cause of ESRD, requiring dialysis or kidney transplantation in advanced stages (van Raalte et al., 2024). Despite advances in understanding its complex pathogenesis, conventional therapeutic approaches often fall short, leading to progressive renal dysfunction and a significant public health challenge. The current treatment methods offer some renoprotective benefits but are often insufficient to halt disease progression in many patients. Moreover, while rigorous glycemic and blood pressure control remains foundational, a substantial number of patients still advance to ESRD, underscoring the limitations of current strategies (Bell & Jerkins, 2024). The multifaceted etiology of diabetic kidney disease, encompassing inflammatory and fibrotic pathways, necessitates the development of novel therapeutic interventions that can effectively target these underlying mechanisms. The inherent difficulties in fully elucidating the precise pathogenic mechanisms of diabetic nephropathy further complicate effective intervention, contributing to the high mortality associated with this condition (Kushwaha et al., 2022).

Diabetic nephropathy is a multifactorial condition, comprising glucose metabolism disorders, inflammatory processes, oxidative stress, and renal hemodynamic alterations. However, the precise mechanisms are still under extensive investigation. Central to its development is chronic hyperglycemia, which instigates numerous cellular and molecular

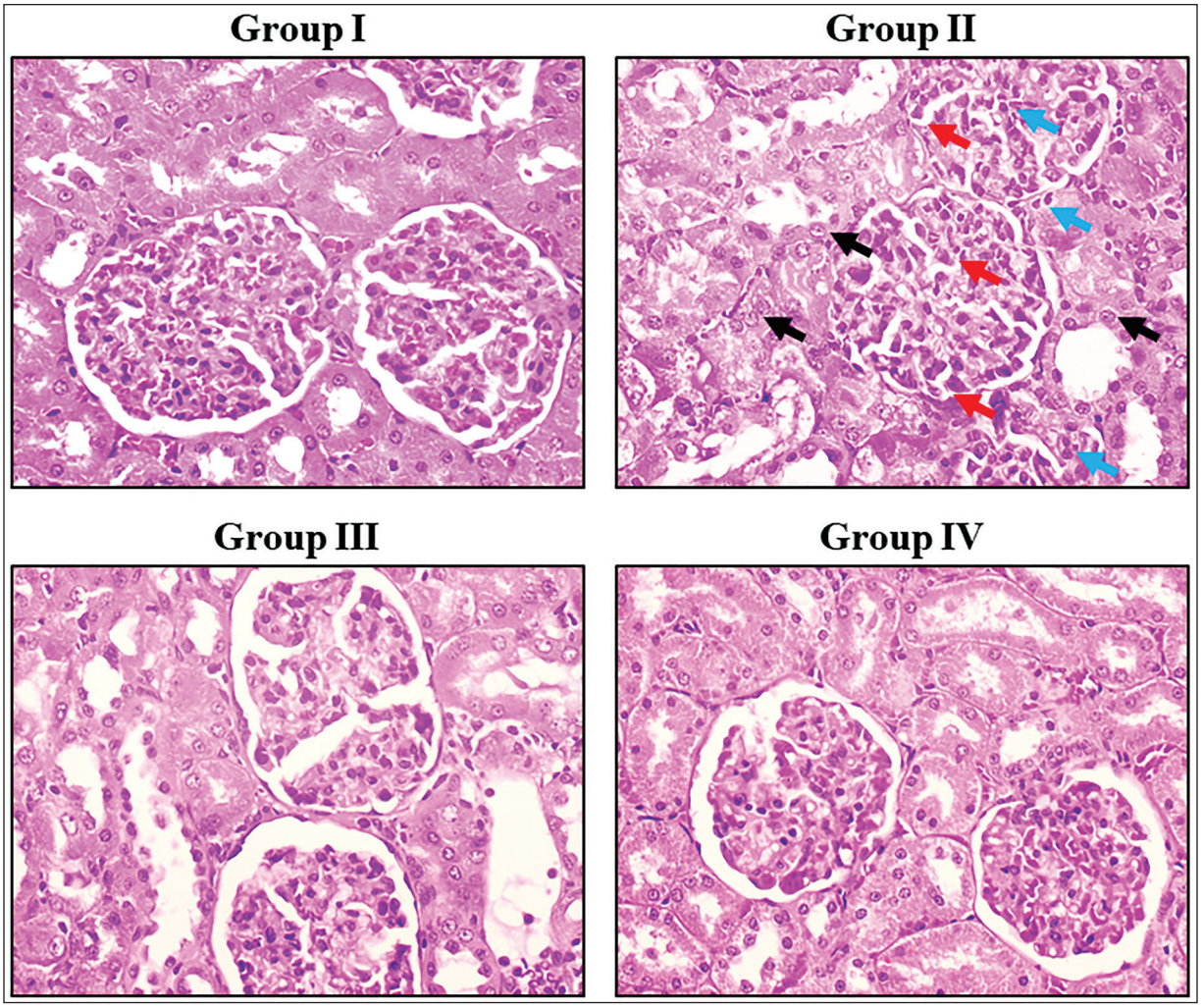


Figure 4. Effect of Gardenin A on the Kidney Tissue Histopathology of Experimental Rats. Group I: Normal Control Rats. Group II: Streptozotocin (STZ)-induced Diabetic Nephropathy Rats. Group III: STZ-induced Diabetic Nephropathy + 50 mg/kg of Gardenin A-treated Rats. Group IV: STZ-induced Diabetic Nephropathy + 5 mg/kg of Standard Drug Gliclazide-treated Rats. Black Arrows: Glomerular Shrinkage. Red Arrows: Damage to Renal Tubular Epithelial Cells. Blue Arrows: Infiltration of Inflammatory Cells.

changes within the renal milieu, eventually resulting in glomerular and tubular damage (Wu et al., 2017). Persistent elevated blood glucose levels directly impair the filtering capacity of the kidneys by damaging the small blood vessels. This persistent hyperglycemia leads to morphological and functional alterations, including mesangial expansion, glomerulosclerosis, and thickening of the glomerular basement membrane. Furthermore, long-term glycemic exposure, reflected by elevated HbA_{1C} levels over time, serves as a primary risk factor for the onset of diabetic complications, including diabetic nephropathy. Poor glycemic control, evidenced by persistently high HbA_{1C} levels, exacerbates these pathological features and drives structural changes within the kidney, such as telangiectasia and increased extracellular matrix expansion (Tian et al., 2020). This chronic hyperglycemia also triggers the overproduction of extracellular matrix components, which are key instigators of renal damage in diabetic conditions. The excessive channeling of glucose intermediaries into alternative metabolic cascades contributes to the accumulation of advanced glycation end-products, further exacerbating kidney damage. Moreover, hyperglycemia triggers renal endothelial dysfunction and injury through sustained metabolic anomalies, thereby compromising renal hemodynamics (Coca et al., 2012).

The chronic hyperglycemia characteristic of diabetes also leads to an overproduction of ROS, causing oxidative damage to critical cellular macromolecules and ultimately resulting in organelle dysfunction and energy failure, particularly within mitochondria. These metabolic derangements collectively initiate a pathological feed-forward cycle that drives structural and functional kidney abnormalities characteristic of diabetic nephropathy. This intricate interplay of metabolic imbalances, hemodynamic alterations, and chronic inflammation establishes a feedback loop that promotes progressive

kidney damage (Guo et al., 2017). Therefore, targeting the management of blood glucose and HbA_{1C} levels in diabetic conditions is crucial to treat or prevent diabetic nephropathy. In this work, the findings indicated the increased glucose and HbA_{1C} in the serum of STZ-induced rats. Interestingly, the gardenin A treatment remarkably decreased these markers in the diabetic rats, which indicates that gardenin A can alleviate diabetic complications in the STZ-induced rats.

The pathogenesis of diabetic nephropathy is complex, involving various risk factors. The early identification of diabetic nephropathy is crucial for timely intervention and improved patient outcomes, necessitating a deeper understanding of its biochemical markers. Traditional biomarkers such as serum urea, creatinine, and uric acid are well-established indicators of renal function, reflecting the severity of kidney damage in uncontrolled diabetic patients. Among these, creatinine, an uncharged small molecule produced from creatine and phosphocreatine, is filtered by the glomeruli and excreted, making it a critical indicator of glomerular function. Elevated creatinine levels directly correlate with a diminished glomerular filtration rate, indicating a decline in renal excretory capacity. Moreover, it has been mentioned that increased serum creatinine levels in diabetic patients are related to an increased likelihood of developing nephropathy (Kawamoto et al., 2019). Similarly, urea, a waste product of protein metabolism, accumulates in the blood when kidney function is compromised, further reflecting impaired renal clearance. Additionally, uric acid, a product of purine metabolism, has garnered attention for its crucial role in the onset of diabetic nephropathy, with elevated levels contributing to inflammation and oxidative stress within the renal microenvironment (Han et al., 2023). Higher serum uric acid levels are also independently related to microalbuminuria, serving as an early indicator of renal damage in diabetes mellitus. This intricate interplay of these renal dysfunction markers highlights their critical role in both diagnosing and monitoring the progression of nephropathy. Additionally, the increase in urea, creatinine, and uric acid often aligns with a linear progression in diabetic nephropathy severity, correlating with declines in estimated glomerular filtration rate and increased inflammatory markers (Pandya et al., 2016). Therefore, targeting these renal dysfunction markers is crucial to developing novel therapeutic candidates against diabetic nephropathy. This study showed increased serum concentrations of urea, creatinine, and uric acid in STZ-induced rats. Though treatment with gardenin A has effectively decreased the concentrations of these renal dysfunction markers in the serum of diabetic rats, which suggests its nephroprotective properties.

Oxidative stress, a condition of imbalanced ROS accumulation and the body's antioxidant mechanisms, plays a crucial role in the initiation of nephropathy. This imbalance, often exacerbated by chronic hyperglycemia, leads to increased ROS generation and a concurrent reduction in antioxidant mechanisms, which subsequently cause oxidative damage to DNA and proteins (Hernandez et al., 2022). The primary sources of these ROS in the kidney are mitochondria and the nicotinamide adenine dinucleotide phosphate oxidase family, with additional contributions from endoplasmic reticulum and peroxisomes. In diabetic kidneys, NADPH oxidase and the mitochondrial electron transport chain are key contributors to ROS production, which is believed to initiate albuminuria and subsequent renal damage via podocyte depletion (Darenskaya et al., 2021). This intricate interplay of heightened oxidative stress and diminished antioxidant capacity ultimately contributes to structural alterations in the kidney, including glomerular hypertrophy, thickening of basement membranes, and renal interstitial fibrosis, which are hallmarks of diabetic nephropathy (Qiu et al., 2022).

Chronic hyperglycemia further triggers oxidative stress by increasing ROS production, diminishing antioxidant mechanisms, and triggering oxidative damage to cells. This elevated oxidative state activates various signaling pathways, including those involving transcription factors, inflammatory cytokines, chemokines, and vasoactive substances, eventually contributing to the initiation of renal injury (Rousseau et al., 2022). MDA serves as a key biomarker of lipid peroxidation, which is significantly elevated in diabetic nephropathy due to increased ROS production, contributing to renal damage and fibrosis. Elevated MDA levels are a consistent marker of oxidative stress that impacts renal metabolic pathways, resulting in inflammation, fibrosis, and endothelial dysfunction in diabetic nephropathy (Wang et al., 2020). Conversely, the antioxidant enzymes, including SOD, CAT, and GPx, are typically diminished in diabetic nephropathy, highlighting a critical imbalance in the prooxidant/antioxidant homeostasis that further exacerbates renal pathology. This imbalance compromises the kidney's ability to neutralize damaging free radicals, leading to sustained oxidative injury to renal cells and tissues. Specifically, a decrease in the activity and expression of the antioxidants has been observed in diabetic microvascular disease, indicating a compromised defense against oxidative damage (Zhang et al., 2021). This reduction in endogenous antioxidant capacity, coupled with increased ROS, results in the initiation of nephropathy by fostering renal interstitial fibrosis, glomerular hypertrophy, and podocyte injury. This intricate interplay between heightened oxidative stress, marked by elevated MDA, and diminished antioxidant defenses, such as reduced SOD, CAT, and GPx, drives the pathological progression of diabetic nephropathy (Ren et al., 2020). Consequently, targeting the oxidative stress and its biomarker levels is crucial to developing novel therapies to mitigate the progression of nephropathy. In this study, the rats with diabetic nephropathy showed an elevated MDA level and subsequently diminished SOD, CAT, and GPx levels in their renal tissues, which suggests the onset of oxidative stress in the kidneys of diabetic rats. Captivatingly, the treatment with gardenin A remarkably decreased the MDA levels and subsequently increased the antioxidant levels in the kidneys of diabetic rats. These findings suggest the antioxidant effects of gardenin A in mitigating the oxidative stress response in the kidneys of diabetic rats.

Inflammation, a complex biological response to harmful stimuli, is defined by the activation of immune cells and the subsequent release of various mediators, including cytokines. In the context of diabetic nephropathy, sustained low-grade inflammation significantly contributes to disease initiation and progression, exacerbating renal damage (Rayego-Mateos et al., 2020). This inflammatory stress, stemming from both metabolic and hemodynamic derangements inherent to diabetic conditions, results in an elevated accumulation of cytokines. Specifically, elevated TNF-α and IL-6 levels contribute to renal podocyte injury and glomerular endothelial dysfunction, thereby accelerating the progression of nephropathy. These cytokines induce structural and functional alterations within the glomeruli by promoting oxidative stress and dysregulating vasoactive pathways, eventually resulting in albuminuria and a decrease in renal function (Hou et al., 2025). Furthermore, the sustained presence of these cytokines can lead to a chronic inflammatory state within the kidney, promoting fibrosis and scarring that irrevocably impairs renal function. TNF-α is a crucial cytokine that directly contributes to microvascular problems in diabetes, including nephropathy, through its cytotoxicity on glomerular, mesangial, and epithelial cells. This cytokine not only orchestrates cellular apoptosis but also enhances the expression of adhesion molecules and chemokines, thereby augmenting inflammatory cell infiltration into renal tissues (Lim & Tesch, 2012). In parallel, IL-6 contributes to renal dysfunction by promoting mesangial cell proliferation, extracellular matrix accumulation, and the induction of profibrotic growth factors, exacerbating glomerular injury in diabetic nephropathy.

The heightened systemic and local inflammatory responses observed in diabetes, characterized by increased TNF-α and IL-6, are closely correlated with the severity of albuminuria, indicating their early involvement in the pathogenic cascade (Wada & Makino, 2013). Specifically, IL-6 promotes glomerular basement membrane thickening and extracellular matrix synthesis, which are early morphological changes in nephropathy. Indeed, it has been shown that patients with progressive nephropathy exhibit higher TNF-α and IL-6 levels, correlating with the degree of albuminuria. The intricate interplay between these cytokines further potentiates renal damage, thereby reducing renal function and exacerbating the pathogenesis of nephropathy (Zhao et al., 2024). In line with this statement, we observed the increased TNF-α and IL-6 levels in the kidney tissues of the STZ-induced rats with diabetic nephropathy, which shows its crucial role in the initiation and development of nephropathy. Interestingly, the gardenin A treatment to the STZ-induced rats resulted in a reduction in the TNF-α and IL-6 in their renal tissues. These results propose the anti-inflammatory activity of gardenin A in ameliorating inflammatory response in the kidneys of diabetic rats, thereby hindering the initiation and development of nephropathy.

While our study demonstrates gardenin A's promising therapeutic effects on diabetic nephropathy, it has several limitations. The present study's single *in vivo* model may not fully replicate human disease complexity. Additionally, the study's sample size was relatively small, and the longer-term effects of gardenin A remain unclear. Further research is needed to elucidate the molecular mechanisms underlying gardenin A's renoprotective effects. Future studies should investigate gardenin A's efficacy in combination with existing treatments and explore its potential in other kidney diseases. Larger, randomized controlled trials are necessary to confirm our findings. Moreover, investigating gardenin A's pharmacokinetics and toxicity profile will be essential for its clinical translation. Research on gardenin A's effects on other organs and pathways is also warranted, providing a more comprehensive understanding of its therapeutic potential. Addressing these limitations will help establish gardenin A as a viable treatment option for diabetic nephropathy.

Conclusion

The present study suggests that gardenin A treatment may ameliorate diabetic nephropathy in experimental rats. The gardenin A treatment significantly diminished the blood glucose, HbA_{1C}, and renal dysfunction marker levels in the STZ-induced rats. Moreover, gardenin A treatment significantly diminished the TNF-α and IL-6 levels and mitigated oxidative stress via increasing antioxidant levels in the kidney tissues of diabetic rats. The histological findings of the kidney tissues also suggested the therapeutic effects of gardenin A against diabetic nephropathy. Thus, it was evident that gardenin A may have a therapeutic effect against diabetic nephropathy. However, future in-depth studies are strongly advised to thoroughly understand the underlying mechanisms by which gardenin A shows its therapeutic effects on diabetic nephropathy.

Abbreviations

CAT: Catalase; ESRD: End-stage renal disease; GPx: Glutathione peroxidase; IL: Interleukin; MDA: Malondialdehyde; SOD: Superoxide dismutase; STZ: Streptozotocin.

Declaration of Conflicting Interests

The authors declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Ethical Approval

The animal experiments were approved by the Ethics Committee for Animal Care and Use of Affiliated Hospital of Hebei University, China.

Funding

The authors disclosed receipt of the following financial support for the research, authorship, and/or publication of this article: The project was supported by the Baoding Science and Technology Plan Project: Tyrosine phosphatase SHP-1 delays the progression of

diabetic nephropathy by inhibiting apoptosis and fibrosis of HK2 cells induced by the SP1/HIF1α/HDAC3/FOXO1/ROS signaling pathway (Grant No. 2541ZF104).

Informed Consent

Not applicable.

ORCID iD

Qiaoying Wu https://orcid.org/0009-0003-6926-8806

References

Alonso-Castro, A. J., Gasca-Martínez, D., Cortez-Mendoza, L. V., Alba-Betancourt, C., Ruiz-Padilla, A. J., & Zapata-Morales, J. R. (2020). Evaluation of the neuropharmacological effects of Gardenin A in mice. *Drug Development Research*, *81*(5), 600–608.

Bell, D. S. H., & Jerkins, T. (2024). The potential for improved outcomes in the prevention and therapy of diabetic kidney disease through "stacking" of drugs from different classes. *Diabetes, Obesity and Metabolism*, *26*, 2046–2053.

Cabrera, J., Saavedra, E., Del Rosario, H., Perdomo, J., Loro, J. F., Cifuente, D. A., Tonn, C. E., García, C., Quintana, J., & Estévez, F. (2016). Gardenin B-induced cell death in human leukemia cells involves multiple caspases but is independent of the generation of reactive oxygen species. *Chemico-Biological Interactions*, *256*, 220–227.

Coca, S. G., Ismail-Beigi, F., Haq, N., Krumholz, H. M., & Parikh, C. R. (2012). Role of intensive glucose control in development of renal end points in type 2 diabetes mellitus: Systematic review and meta-analysis intensive glucose control in type 2 diabetes. *Archives of Internal Medicine*, *172*, 761–769.

Darenskaya, M. A., Kolesnikova, L. I., & Kolesnikov, S. I. (2021). Oxidative stress: Pathogenetic role in diabetes mellitus and its complications and therapeutic approaches to correction. *Bulletin of Experimental Biology and Medicine*, *171*(2), 179–189.

Guo, J., Lu, C., Zhang, F., Yu, H., Zhou, M., He, M., Wang, C., Zhao, Z., & Liu, Z. (2017). VDR activation reduces proteinuria and high-glucose-induced injury of kidneys and podocytes by regulating Wnt signaling pathway. *Cellular Physiology and Biochemistry*, *43*(1), 39–51.

Hack, W., Gladen-Kolarsky, N., Chatterjee, S., Liang, Q., Maitra, U., Ciesla, L., & Gray, N. E. (2024). Gardenin A treatment attenuates inflammatory markers, synuclein pathology and deficits in tyrosine hydroxylase expression and improves cognitive and motor function in A53T-α-syn mice. *Biomedicine & Pharmacotherapy*, *173*, 116370.

Han, R., Duan, L., Zhang, Y., & Jiang, X. (2023). Serum uric acid is a better indicator of kidney impairment than serum uric acid-to-creatinine ratio and serum uric acid-to-high-density lipoprotein ratio: A cross-sectional study of type 2 diabetes mellitus patients. *Diabetes, Metabolic Syndrome and Obesity*, *16*, 2695–2703.

Hernandez, L. F., Eguchi, N., Whaley, D., Alexander, M., Tantisattamo, E., & Ichii, H. (2022). Anti-oxidative therapy in diabetic nephropathy. *Frontiers in Bioscience*, *14*(2), 14.

Hou, G., Dong, Y., Jiang, Y., Zhao, W., Zhou, L., Cao, S., & Li, W. (2025). Immune inflammation and metabolic interactions in the pathogenesis of diabetic nephropathy. *Frontiers in Endocrinology*, *16*, 1602594.

Hu, R., Zhao, Z., Xie, L., Ma, Z., Wu, W., & Li, S. (2025). Global, regional, and national burden of chronic kidney disease due to diabetes mellitus type 2 from 1990 to 2021, with projections to 2036: A systematic analysis for the Global Burden of Disease Study 2021. *Frontiers in Medicine*, *12*, 1531811.

Jin, Q., Liu, T., Qiao, Y., Liu, D., Yang, L., Mao, H., Ma, F., Wang, Y., Peng, L., & Zhan, Y. (2023). Oxidative stress and inflammation in diabetic nephropathy: Role of polyphenols. *Frontiers in Immunology*, *14*, 1185317.

Jung, C. Y., & Yoo, T. H. (2022). Pathophysiologic mechanisms and potential biomarkers in diabetic kidney disease. *Diabetes & Metabolism Journal*, *46*(2), 181–197.

Kawamoto, R., Ninomiya, D., Kikuchi, A., Akase, T., Kasai, Y., Ohtsuka, N., & Kumagi, T. (2019). Serum uric acid to creatinine ratio is a useful predictor of renal dysfunction among diabetic persons. *Diabetology & Metabolic Syndrome*, *13*(3), 1851–1856.

Kushwaha, K., Kabra, U., Dubey, R., & Gupta, J. (2022). Diabetic nephropathy: Pathogenesis to cure. *Current Drug Targets*, *23*, 1418–1429.

Lim, A. K. H., & Tesch, G. H. (2012). Inflammation in diabetic nephropathy. *Mediators of inflammation*, *2012*, 146154.

Moreno, J. A., Gomez-Guerrero, C., Mas, S., Sanz, A. B., Lorenzo, O., Ruiz-Ortega, M., Opazo, L., Mezzano, S., & Egido, J. (2018). Targeting inflammation in diabetic nephropathy: A tale of hope. *Expert Opinion on Investigational Drugs*, *27*(11), 917–930.

Pandya, D., Nagrajappa, A. K., & Ravi, K. S. (2016). Assessment and correlation of urea and creatinine levels in saliva and serum of patients with chronic kidney disease, diabetes and hypertension—A research study. *Journal of Clinical and Diagnostic Research*, *10*(10), ZC58-ZC62.

Qiu, D., Song, S., Wang, Y., Bian, Y., Wu, M., Wu, H., Shi, Y., & Duan, H. (2022). NAD(P)H: Quinone oxidoreductase 1 attenuates oxidative stress and apoptosis by regulating Sirt1 in diabetic nephropathy. *Journal of Translational Medicine*, *20*(1), 44.

Raval, N., Kumawat, A., Kalyane, D., Kalia, K., & Tekade, R. K. (2020). Understanding molecular upsets in diabetic nephropathy to identify novel targets and treatment opportunities. *Drug Discovery Today*, *25*(5), 862–878.

Rayego-Mateos, S., Morgado-Pascual, J. L., Opazo-Ríos, L., Guerrero-Hue, M., García-Caballero, C., Vázquez-Carballo, C., Mas, S., Sanz, A. B., Herencia, C., Mezzano, S., Gómez-Guerrero, C., Moreno, J. A., & Egido, J. (2020). Pathogenic pathways and therapeutic approaches targeting inflammation in diabetic nephropathy. *International Journal of Molecular Sciences*, *21*(11), 3798.

Ren, H., Shao, Y., Wu, C., Ma, X., Lv, C., & Wang, Q. (2020). Metformin alleviates oxidative stress and enhances autophagy in diabetic kidney disease via AMPK/SIRT1-FoxO1 pathway. *Molecular and Cellular Endocrinology*, *500*, 110628.

Rousseau, M., Denhez, B., Spino, C., Lizotte, F., Guay, A., Côté, A. M., Burger, D., & Geraldes, P. (2022). Reduction of DUSP4 contributes to podocytes oxidative stress, insulin resistance and diabetic nephropathy. *Biochemical and Biophysical Research Communications*, *624*, 127–133.

Shan, S., Luo, Z., Yao, L., Zhou, J., Wu, J., Jiang, D., Ying, J., Cao, J., Zhou, L., Li, S., & Song, P. (2024). Cross-country inequalities in disease burden and care quality of chronic kidney disease due to type 2 diabetes mellitus, 1990-2021: Findings from

the global burden of disease study 2021. *Diabetes, Obesity and Metabolism*, *26*(12), 5950–5959.

Tian, J., Ohkuma, T., Cooper, M., Harrap, S., Mancia, G., Poulter, N., Wang, J. G., Zoungas, S., Woodward, M., & Chalmers, J. (2020). Effects of intensive glycemic control on clinical outcomes among patients with type 2 diabetes with different levels of cardiovascular risk and hemoglobin A_{1c} in the ADVANCE trial. *Diabetes Care*, *43*(6), 1293–1299.

Toppo, E., Darvin, S. S., Esakkimuthu, S., Stalin, A., Balakrishna, K., Sivasankaran, K., Pandikumar, P., Ignacimuthu, S., & Al-Dhabi, N. A. (2017). Antihyperlipidemic and hepatoprotective effects of Gardenin A in cellular and high fat diet fed rodent models. *Chemico-Biological Interactions*, *269*, 9–17.

van Raalte, D. H., Bjornstad, P., Cherney, D. Z. I., de Boer, I. H., Fioretto, P., Gordin, D., Persson, F., Rosas, S. E., Rossing, P., Schaub, J. A., Tuttle, K., Waikar, S. S., & Heerspink, H. J. L. (2024). Combination therapy for kidney disease in people with diabetes mellitus. *Nature Reviews Nephrology*, *20*(7), 433–446.

Wada, J., & Makino, H. (2013). Inflammation and the pathogenesis of diabetic nephropathy. *Clinical Science*, *124*, 139–152.

Wang, F., Li, R., Zhao, L., Ma, S., & Qin, G. (2020). Resveratrol ameliorates renal damage by inhibiting oxidative stress-mediated apoptosis of podocytes in diabetic nephropathy. *European Journal of Pharmacology*, *885*, 173387.

Wu, X., Gao, Y., Xu, L., Dang, W., Yan, H., Zou, D., Zhu, Z., Luo, L., Tian, N., Wang, X., Tong, Y., & Han, Z. (2017). Exosomes from high glucose-treated glomerular endothelial cells trigger the epithelial-mesenchymal transition and dysfunction of podocytes. *Scientific Reports*, *7*(1), 9371.

Yamazaki, T., Mimura, I., Tanaka, T., & Nangaku, M. (2021). Treatment of diabetic kidney disease: Current and future. *Diabetes & Metabolism Journal*, *45*, 11–26.

Zhang, M., He, L., Liu, J., & Zhou, L. (2021). Luteolin attenuates diabetic nephropathy through suppressing inflammatory response and oxidative stress by inhibiting STAT3 pathway. *Experimental and Clinical Endocrinology & Diabetes*, *129*(10), 729–739.

Zhang, L., Jiang, L., Xu, R., Zhang, X., Zhang, B., & Yue, R. (2025). Epidemiological research on diabetic nephropathy at global, regional, and national levels from 1990 to 2021: An analysis derived from the global burden of disease 2021 study. *Frontiers in Endocrinology*, *16*, 1647064.

Zhao, L., Hu, H., Zhang, L., Liu, Z., Huang, Y., Liu, Q., Jin, L., Zhu, M., & Zhang, L. (2024). Inflammation in diabetes complications: Molecular mechanisms and therapeutic interventions. *MedComm (2020)*, *5*(4), e516.