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Research Article
Effect of Barley (*Hordeum vulgare*) on Renal Function in a Diabetic Animal Model: A Study in Experimental Rats

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Abstract

Background and Objective: Barley has been traditionally used in traditional Persian medicine (TPM) for treating diabetes mellitus (DM). Previous research suggests its protective effects on various tissues in diabetic rats, including the liver, pancreas, kidneys and heart. This study aimed to investigate the impact of barley on renal function in a diabetic animal model using experimental rats. **Materials and Methods:** Male Wistar rats were divided into 4 groups control, barley, diabetic and barley administrated diabetic groups, with diabetes induced by streptozotocin injection. Barley was administered through dietary supplementation. Biochemical investigations, oral glucose tolerance tests, insulin sensitivity tests and histological examinations were conducted to evaluate renal function. **Results:** Firstly, it mitigated significant weight loss observed in diabetic rats, leading to higher body weight compared to untreated rats by the study's end. Secondly, barley improved glucose tolerance, reduced fasting blood glucose levels and enhanced insulin sensitivity, as evidenced by the Oral Glucose Tolerance Test (OGTT) and insulin sensitivity test (IST) results. Thirdly, barley supplementation demonstrated potential renal protective effects by reducing blood urea nitrogen, albumin and creatinine levels, indicative of mitigating diabetes-induced renal damage. Additionally, barley elevated levels of antioxidant enzymes, counteracting reduced levels seen in untreated diabetic rats and partially reversed pathological changes associated with diabetic nephropathy in histological analysis. **Conclusion:** This study provides insights into the potential of barley as a dietary intervention for improving renal function in diabetic conditions, offering valuable implications for managing diabetes-related complications.

Key words: Barley, diabetic, kidney, hyperglycemia, phytotherapy, renal function

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Competing Interest: The author has declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Barley has long been recognized in traditional Persian medicine (TPM) as a valuable treatment for diabetes mellitus (DM) due to its numerous health benefits[1]. Research conducted on both normal and streptozotocin-induced diabetic rats has shown that barley can have a protective effect on various tissues such as the liver, pancreas, kidneys and heart[2]. This protective effect was evident through histopathological analysis, which revealed decreased abnormal histological signs in diabetic rats treated with barley compared to those left untreated[3]. Furthermore, non-diabetic rats treated with barley also exhibited protective effects when compared to the control group. These results underscore the potential of barley as a therapeutic intervention for addressing diabetes-related tissue damage[4].

The antioxidant properties of phenolic compounds present in barley contribute to its nutritional value and may aid in its protective effects[5]. Antioxidants are crucial in combating chronic diseases by neutralizing reactive oxygen species (ROS), often elevated in individuals with diabetes. Studies have demonstrated that consumption of barley can lead to reduced serum glucose levels in diabetic rats, highlighting its potential as a dietary intervention for managing diabetes-related complications[1,6].

In conclusion, the research on the impact of barley on renal function in diabetic rats emphasizes the importance of exploring natural remedies for managing diabetes mellitus and its associated complications. By studying the effects of barley on various tissues affected by diabetes, valuable insights can be gained into its therapeutic potential for enhancing renal function and overall health outcomes in individuals with diabetes. Previous studies have pointed out the ability of barley to alleviate histopathological alterations in the liver, pancreas, kidneys and cardiac tissues of streptozotocin-induced diabetic rats[4,7-9]. Furthermore, barley-glucans have demonstrated a capacity to decrease systemic inflammation, renal damage and aortic calcification by inhibiting ADAM17 and neutral-sphingomyelinase[2,10,11]. Barley grass powder contains beneficial compounds like GABA, flavonoids, SOD, vitamins and polyphenols that provide a wide array of health advantages such as enhancing sleep quality, regulating blood pressure, boosting immunity and exerting anti-inflammatory effects[12].

The preventive and therapeutic potentials of barley grass for chronic ailments have been extensively researched, showcasing its effectiveness as a functional food with multiple health benefits[13]. Barley grains are recognized for their high glucan content which can enhance gut health and lower blood glucose levels[14]. Analyses of nutrient composition have revealed that barley is rich in essential minerals like calcium, zinc, iron, potassium and phosphorus which support diverse bodily functions from maintaining bone health to aiding in wound healing[15]. Furthermore, consuming whole grain barley has been associated with reduced risks of chronic conditions like diabetes and cardiovascular diseases due to its phytochemical content including glucan and phenolic acids[10,16].

Diabetes is a widespread metabolic disorder that has a significant impact on renal function, resulting in conditions like Diabetic Kidney Disease (DKD)[17]. There have been various animal models developed to examine the effects of diabetes on kidney health, with the db/db mouse model being one of the most extensively researched. This model, distinguished by a mutation in the leptin receptor gene, displays consistent and strong characteristics of diabetic nephropathy, such as increased albuminuria and expansion of the mesangial matrix[18]. Research has indicated that diabetic animals, like db/db mice, experience glomerular pathology over time, showcasing changes like enlarged glomeruli, expanded mesangial matrix and thickened basement membranes[19].

Additionally, studies have brought to light the link between high blood sugar levels and oxidative stress in diabetic rats, resulting in impaired renal function[20]. The generation of reactive oxygen species (ROS) due to elevated blood sugar levels can lead to cellular damage and dysfunction of endothelial cells[21]. In diabetic rats, there have been observations of increased levels of malondialdehyde and decreased catalase activities[22]. Furthermore, examinations at a histopathological level have unveiled structural modifications in the pancreas, liver and kidneys of diabetic rats.

In general, these discoveries emphasize the intricate relationship between diabetes and renal function in animal models. Grasping the mechanisms that drive these interactions is essential for formulating targeted measures to lessen the impact of diabetes on kidney well-being.

Barley is gaining recognition for its potential as a dietary intervention in various health conditions, including diabetes[23,24]. The health benefits of barley are linked to its nutrient-rich profile, containing proteins, vitamins, minerals and fiber[25,26]. A key component in barley is beta-glucan, a soluble fiber known for reducing cholesterol levels and lowering the risk of heart disease[27]. Research has shown that beta-glucan can help regulate blood sugar levels, making it a favorable choice for individuals with diabetes[28]. Additionally, the high fiber content in barley can assist in managing blood glucose levels and promoting gut health[14].

Moreover, barley lings contain compounds like-glucan, tocols and resistant starch that offer various health benefits such as anticancer properties, immune system enhancement, blood sugar regulation and anti-inflammatory effects[1,23]. Products made from barley like flour, flatbread, muffins, pasta and whole-grain options like noodles have been found to provide additional advantages like boosting immunity and preventing conditions like tumors or bone disorders[27,29].

Studies suggest that including barley in meals can enhance glucose utilization due to factors like -glucan content and the amylose/amylopectin ratio present in barley grains. Individuals who consume whole barley meals have shown improvements in glycemic tolerance and fasting glucose levels[30]. Furthermore, incorporating resistant starches from barley-based bread may also help lower blood sugar levels[8].

In summary, incorporating barley into the diet has shown promising results for improving renal function and blood sugar control in diabetic rats[31,32]. Its nutrient-dense profile makes it a valuable dietary choice with potential benefits for individuals managing diabetes[33].

Prior research has showcased the potential of barley to positively influence kidney function in diabetic rats. An example is a study conducted by Kalra and Jood[9], which highlighted the beneficial impact of barley sprouts juice on diabetic rats. This study revealed significant reductions in fasting blood glucose levels and improvements in lipid profiles. Furthermore, supplementation with barley sprouts juice[34] resulted in a decrease in malondialdehyde values and an increase in catalase activities, indicating its antioxidant properties. The examination of kidney tissue through histopathological analysis also displayed enhancements following treatment with barley sprouts juice, suggesting a protective effect on renal function.

Additionally, another study demonstrated that barley sprouts juice, when combined with other dietary interventions like cell-free probiotic extract and whey protein hydrolysate[35,36], alleviated histopathological changes in the pancreas, liver and kidneys of diabetic rats. These interventions contributed to reducing oxidative stress and enhancing kidney function biomarkers, underscoring the potential of barley as a dietary intervention for diabetic complications[37].

Moreover, research by PubMed Disclaimer underscored the significance of modifying one's diet to manage diabetes-related complications[38]. The inclusion of barley microgreen in the diet was proven effective in controlling type 2 diabetes mellitus and improving oxidative stress biomarkers. This indicates that dietary strategies involving barley could play a substantial role in mitigating the impact of diabetes on kidney function[1]. In conclusion, past studies have presented compelling evidence supporting the utilization of barley as a dietary intervention to enhance kidney function in diabetic rats[39]. The antioxidant properties and potential protective effects of barley on kidney tissue make it a promising avenue for further exploration concerning diabetic nephropathy[40].

The importance of this study lies in the potential of barley to have a positive impact on renal function in diabetic rats. Prior research has emphasized the antioxidant properties of barley, attributed to its abundance of phenolic compounds, making it a promising dietary intervention for conditions like diabetes[41]. Barley has been proven to offer various health advantages, including lowering cholesterol levels, regulating blood sugar levels[40] and possessing anti-cancer properties[23,27]. Furthermore, products derived from barley have been associated with anti-inflammatory, antioxidant, anti-arthritic effects and detoxifying properties[12,42,43]. This highlights the significance of investigating how barley affects renal function in diabetic rats[44,45].

Studies have shown that barley can decrease blood sugar levels by influencing glucose utilization and glycemic responses through components like-glucan and amylose/amylopectin ratio[46]. Additionally, the barley's antioxidant and detoxifying abilities alleviate oxidative stress and eliminate toxins from the body[28]. The phytochemicals present in barley leaves have also demonstrated positive effects on human health, including anti-inflammatory, immune-boosting and antioxidant properties.

The preventive and therapeutic potential of functional ingredients in barley grass further strengthens its role as a functional food for chronic diseases. Barley grass is abundant in beneficial components such as Gamma-Aminobutyric Acid (GABA), flavonoids, SOD, vitamins (A, B1, C, E), chlorophyll, dietary fiber, polysaccharide, alkaloid and polyphenols among others[11,15,47,48]. These discoveries indicate that studying the impact of barley on renal function in diabetic rats could offer valuable insights into its therapeutic effects beyond its well-known health benefits.

The main objective of this research endeavor is to delve into the effects of barley on renal function in diabetic rats. Barley, a recommended remedy in traditional Persian medicine for diabetes mellitus, has been shown to have protective properties on different tissues in diabetic rats. In summary, this study aims to expand upon current research by focusing specifically on how barley influences renal function in diabetic rats. By investigating the protective effects of barley and its potential to alleviate histopathological changes in various tissues affected by diabetes mellitus, this research strives to provide valuable insights into the role of barley as a dietary intervention for individuals with diabetes.

MATERIALS AND METHODS

Study area: This study was conducted over a period of six months from January, 2023 to June, 2023. The research was carried out at the Department of Animal Physiology, Al-Jamoum University College, located in Umm Al-Qura University, Kingdom of Saudi Arabia.

Experimental animals: Eighty fertile male albino rats (*Rattus norvegicus*), weighing approximately 200 g[49], procured from the Umm Al-Qura University Breeding Farm, KSA, will be divided into four groups (n = 20 per group): Control, barely supplementation, diabetes and diabetes with barely supplementation (250 mg/kg)[50]. The experimental period will extend from diabetes induction initiation to 8 weeks thereafter, with *ad libitum* access to water[49].

A control group will consist of rats receiving no treatment, serving as a baseline for assessing the effects of barley treatment on kidney health.

Type 2 diabetes mellitus will be induced in all rats via a single intraperitoneal injection of streptozotocin (60 mg/kg) in citrate buffer (0.05 M, pH 4.5), accompanied by 100 mg/kg nicotinamide[49,50]. Hyperglycemia is induction will be confirmed by measuring blood glucose levels, with rats exhibiting levels of 250 mg/dl selected for the study by Aly *et al*.[51].

Formula used for barley bread (80%) preparation consists of refined flour 20 g, whole barley flour 80 g, yeast 0.5 g, salt 1.5 g and water 75-80 mL tap water[52].

Ethical consideration: In terms of ethical considerations, it is crucial that all experimental procedures receive approval from Local Animal Care Committees and adhere to International Guidelines for the Ethical Treatment and usage of laboratory animals. This ensures that the study is conducted ethically and prioritizes the well-being of the animals involved, following meticulous procedures in compliance with the guidelines and regulations set by the Animal Ethics Committee of the Umm Al-Qura University.

Blood sample collection: Blood samples will be obtained at 7-day intervals by extracting blood from the distal end of the tail of experimental animals[41]. After an overnight fasting period, rats in the study groups will be euthanized humanely with an appropriate anesthetic dose. Subsequently, blood collection will be performed via cardiac puncture using a sterile syringe, focusing on the artery. The collected blood will undergo centrifugation to remove impurities, with the resulting supernatant preserved at -20°C for subsequent biochemical analyses[53-56].

Biochemical investigations: During the barely treatment period, regular measurements of fasting blood glucose (FBG) in rats will be conducted using a glucometer from Roche in Germany. Urine samples will be collected weekly, specifically one day after each treatment. The total urinary protein quantity will be assessed using a commercially available kit.

Blood insulin and glycation levels (AGE) will be evaluated using Enzyme-Linked Immunosorbent Assay (ELISA) kits from commercial sources[38,57]. Measurements will be conducted following the manufacturers' instructions to ensure precise and standardized assessment of insulin and glycation levels in the blood. The Homeostatic Model Assessment of β-cell function (HOMA-β) will be created using a specific equation. Various biochemical parameters, such as High-Density Lipoprotein (HDL), blood urea nitrogen (BUN), serum triglyceride (TG), total cholesterol (TC), catalase (CAT), Low-Density Lipoprotein (LDL), superoxide dismutase (SOD), glutathione peroxidase (GPx), glutathione S transferase and creatinine levels, will be measured using kits from Rawabi Marketing International (RMI), Riyadh, Kingdom of Saudi Arabia[58]. The measurement of free fatty acids (FFA) will be conducted using a kit provided by RMI. Furthermore, total urine protein concentrations will be determined and all calculations for these indicators will be performed following the manufacturer's specifications[49].

Oral Glucose Tolerance Test (OGTT) and insulin sensitivity test (IST): All animals in the study will undergo an Oral Glucose Tolerance Test (OGTT) and insulin sensitivity test (IST) following a 10 hrs fasting period, according to established protocols. During the OGTT, glucose levels will be measured at specified intervals after the administration of a glucose solution. Simultaneously, the insulin sensitivity test (IST) will evaluate the animals' response to insulin following hormone administration. These tests are pivotal for evaluating glucose metabolism and insulin sensitivity, providing valuable insights into the animals' metabolic state[59-61]. Blood samples will be taken from the tails of the mice at 0, 30, 60, 90 and 120 min after either glucose supplementation or intraperitoneal insulin injection. The area under the curve (AUC) will be calculated for both the IST and OGTT using the trapezoidal rule to comprehensively evaluate glucose response over specified time intervals, providing valuable information for the study of researchers[62,63].

Histological examination: Kidney samples tissue from the different study groups will be isolated and promptly immersed in a 10% phosphate-buffered formalin solution with a pH of 7.4 for fixation. After fixation, the specimens will undergo dehydration using progressively increasing concentrations of ethyl alcohol, followed by clearing in xylene and finally embedding in molten paraplast at a temperature range of 58-62°C. Histological sections, each approximately 4 µm thick, will be prepared and stained with Hematoxylin and Eosin (H&E). The assessment of cells and Nissl bodies in the hippocampus will be performed using Strata Quest software (version 7.0.1, Tissue Gnostics GmbH, Vienna, Austria). Specifically, the CA1 region of the right hippocampus will be manually outlined and the software algorithm will automatically determine the number of cells with intact nuclear membranes and visible nucleoli. The resulting quantification will be normalized to the cell count per CA1 area[64,65].

Statistical analysis: Data analysis will be conducted using Graph Prism 9.0 (GraphPad Software Inc., USA) for three separate multiple trials. The findings will be presented as mean values accompanied by the standard deviation. Statistical assessments will be performed using One-way Analysis of Variance (ANOVA), followed by Tukey's *post hoc* test for multiple comparisons. The predetermined level of significance will be established at $p<0.05$, indicating statistical significance in observed differences[58].

RESULTS

Influence of barley on body weight gain in type 2 diabetic rats: Throughout the experimental period, meticulous documentation and tracking of the rats' body weight were conducted, as depicted in Fig. 1. Upon the induction of diabetes in the D rats using streptozotocin (STZ), a rapid decline in body weight was observed, contrasting sharply with the steady increase observed in the control (C) group. Notably, administration of barley attenuated the loss of body weight. Specifically, at the conclusion of the study, the body weight of the rats treated with barley was significantly higher compared to that of the D group ($p<0.05$), indicating a substantial impact of barley in ameliorating body weight loss in D rats.

Investigation the impact of barley on glucose tolerance and insulin sensitivity in diabetic rats: Barley has proven to have a considerable impact on blood glucose levels in diabetic rats. The results were shown that following the onset of diabetes in rats, fasting blood glucose levels tend to rise gradually. However, when diabetic rats were administered barley, there was a noticeable reduction in fasting blood glucose levels compared to control groups. This indicates that barley may effectively lower blood glucose levels in diabetic rats. Subfigure (A) shows how intervention with barley (B) mitigates fasting blood glucose (FBG) levels in rats with type 2 Diabetes (T2D). A thorough examination of the beneficial effects of barley on diabetic rats involved a comprehensive analysis of Homeostasis Model Assessment for Insulin Resistance (HOMA-IR) and Homeostasis Model Assessment of β-cell function (HOMA-β) concentrations. Both Oral Glucose Tolerance Test (OGTT) and insulin sensitivity test (IST) assays were conducted. Compared to the diabetic (D) group, the group administered with barley (B) showed a significant decrease in HOMA-IR levels ($p<0.0001$, Fig. 2b). Furthermore, rats in the barley group exhibited significantly higher HOMA-β values than those in the diabetic group ($p<0.0001$, Fig. 2c). These findings suggest that barley effectively attenuates

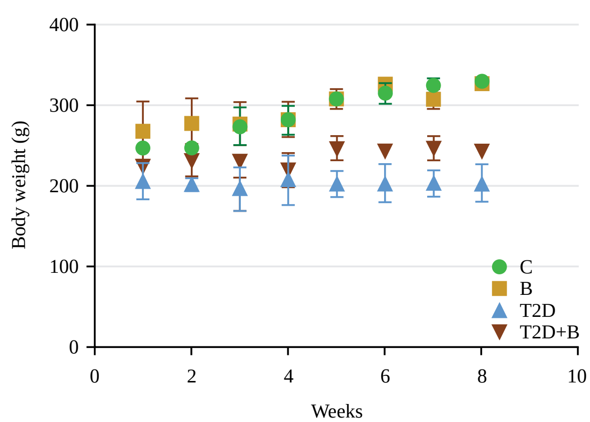

Fig. 1: Effects of barley (B) on body weight gain in different groups of rats

Subfigure illustrates the variations in body weight observed in rats over the course of the experiment. Data are presented as Means±Standard Deviation (n = 20), with statistical significance indicated by $p<0.05$, $p<0.01$ and $p<0.0001$

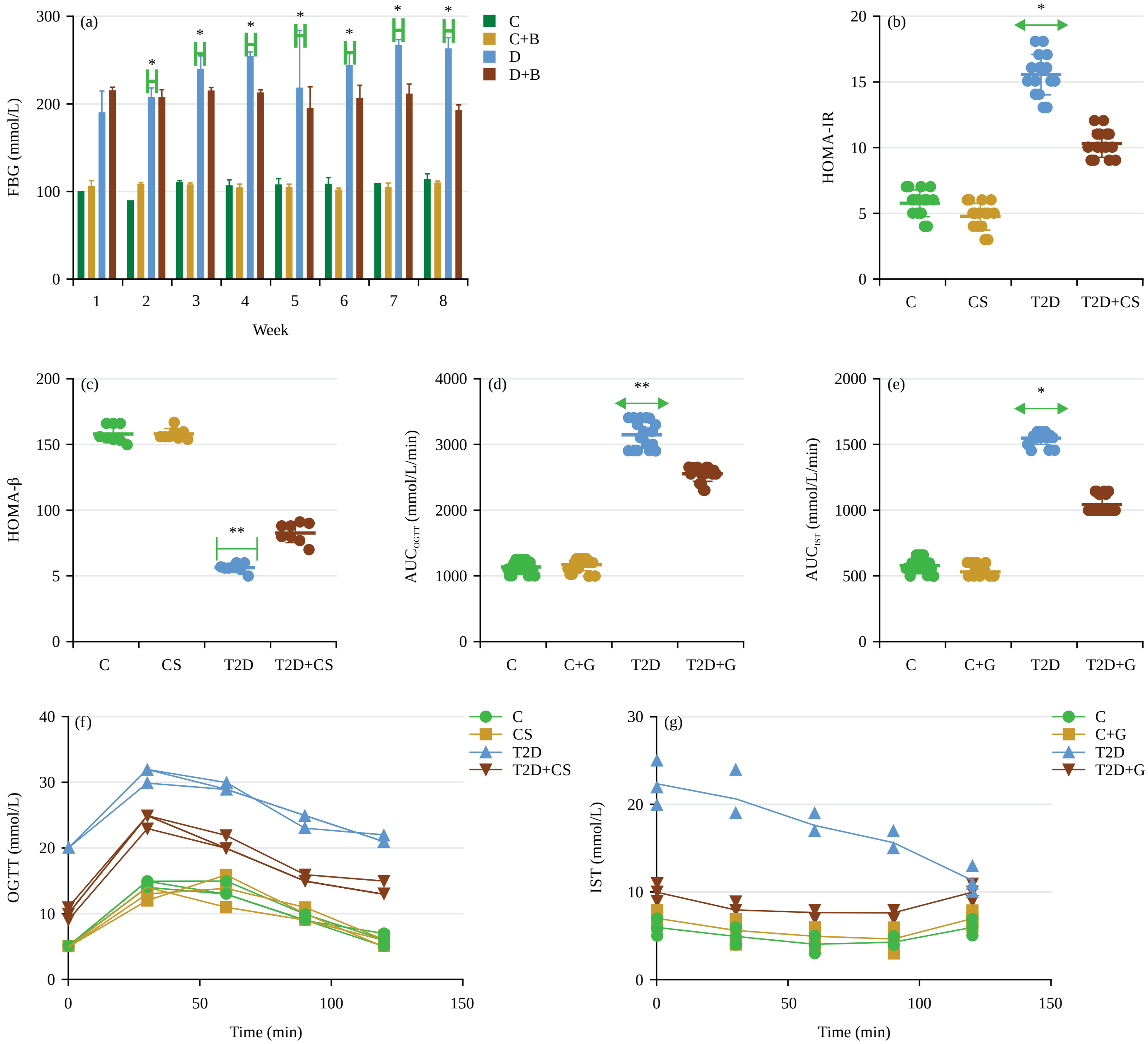

Fig. 2(a-g):Portrays the influence of barley on glucose tolerance and insulin sensitivity in rats with type 2 diabetes (D); (a) How intervention with barley (B) mitigates fasting blood glucose (FBG) levels in rats with type 2 Diabetes (T2D), (b) Computations for Homeostatic Model Assessment for Insulin Resistance (HOMA-IR), (c) Homeostatic Model Assessment for Beta Cell Function (HOMA-β), (d) Additionally, the area under the curve (AUC) calculations for Oral Glucose Tolerance Test (OGTT), (e) Insulin sensitivity test (IST) are illustrated, (f) Along with the results from the OGTT and (g) IST at week 8

Data are expressed as Means±Standard Deviation (n = 20). Statistical significance is denoted as p<0.05, p<0.001, p<0.0001 versus D; $^{\#}$p<0.05 and $^{\#\#}$p<0.01 versus B

insulin resistance to varying extents in type 2 diabetic rats. The results of the Oral Glucose Tolerance Test (OGTT) and insulin sensitivity test (IST) were depicted in Fig. 2d-e. Rats in the diabetic (D) group displayed a notably heightened glycemic response to glucose administration compared to the control (C) rats. However, relative to the diabetic (D) group, rats treated with barley (B) exhibited significantly lower glycemic responses throughout the entire 120 min observation period (p<0.01, Fig. 2f-g). These results indicate that barley has the potential to ameliorate glucose metabolic disorders in diabetic rats, consequently mitigating the risk factors associated with kidney damage.

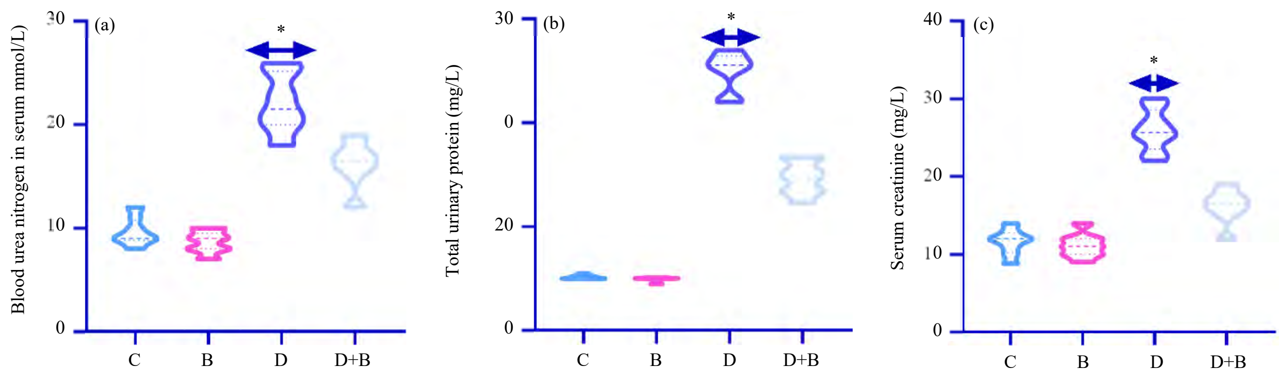

Fig. 3(a-c): Impact of the of barley (B) on various serum biochemistries parameters related to serum kidney function in rats with type 2 diabetes (D). The subplots delineate various parameters; (a) Blood urea nitrogen in serum (mmol/L), (b) Total urinary protein (mg/L) and (c) Creatinine (mg/L)

Data are presented as Means±Standard Deviation (n = 8-10). Statistical significance is denoted as p<0.05, p<0.01 versus D; #p<0.05 and ##p<0.01 versus B

Effects of barley on renal function biomarkers: The data analysis and presentation in this study involved comparing the control group to the barley-treated group, assessing improvements in kidney function. These results suggest potential health benefits of barley in reducing diabetes-induced renal damage.

Furthermore, it was noted that feeding rats with barley significantly lowered blood urea nitrogen in serum level (Fig. 3a), albumin (Fig. 3b) and creatinine (Fig. 3c) in the urine of a metabolic syndrome model, effectively countering kidney failure.

Evaluation of antioxidant parameters: This section elucidates the impact of the of barley (B) on antioxidant enzymes in rats with type 2 diabetes (D). The D group exhibited significantly reduced levels of antioxidant enzymes, namely catalase (CAT), superoxide dismutase (SOD) and glutathione peroxidases (GPx) levels were significantly reduced (p<0.01, Fig. 4a-d) in comparison to the control group (C). However, administration of barely led to an elevation in SOD, catalase (CAT) and GPx levels. It is noteworthy that glutathione-S-transferase activity exhibited a marked increase in the diabetic group compared to the control or B-treated groups.

Histological analysis of kidney tissue: Overall, the diabetic kidney architecture appears disrupted Fig. 5a-c with less distinct boundaries between structures. Increased mesangial matrix material is noticeable too. These changes develop as a result of prolonged hyperglycemia and are associated with declining renal function in diabetic kidney disease.

Careful analysis of slides like this one helps physicians monitor disease progression and potential treatment responses. It also aids research into new therapeutic strategies and prevention of diabetes complications.

The data analysis and presentation in this study Fig. 5d-f involved comparing the control group to the barley-treated group, assessing improvements in kidney structure and evaluating enhancements in kidney function. Previous research on the effects of barley on kidney health showed that adding to the diet prevented dyslipidemia and insulin resistance, normalized glucose metabolism, reduced adipose tissue, improved glucose and insulin tolerance and restored liver function. These results suggest potential health benefits of barley's in reducing diabetes-induced renal damage.

Furthermore, it was noted that feeding rats with barley's significantly lowered albumin and glucose levels in the urine of a metabolic syndrome model, effectively countering kidney failure. Also reversed fibrosis, morphological abnormalities and altered markers of renal glucose metabolism. In addition to these structural improvements, barley intake resulted in a decrease in triglyceride buildup and oxidative stress indicators in the kidneys.

Regarding functional improvements, treatment successfully countered oxidative stress-induced damage caused by diabetes. This was evidenced by a decrease in liver triglycerides and an improvement in insulin resistance and dyslipidemia. Additionally, barley was found to regulate body weight and increase glucose levels in diabetic rats without significant deterioration.

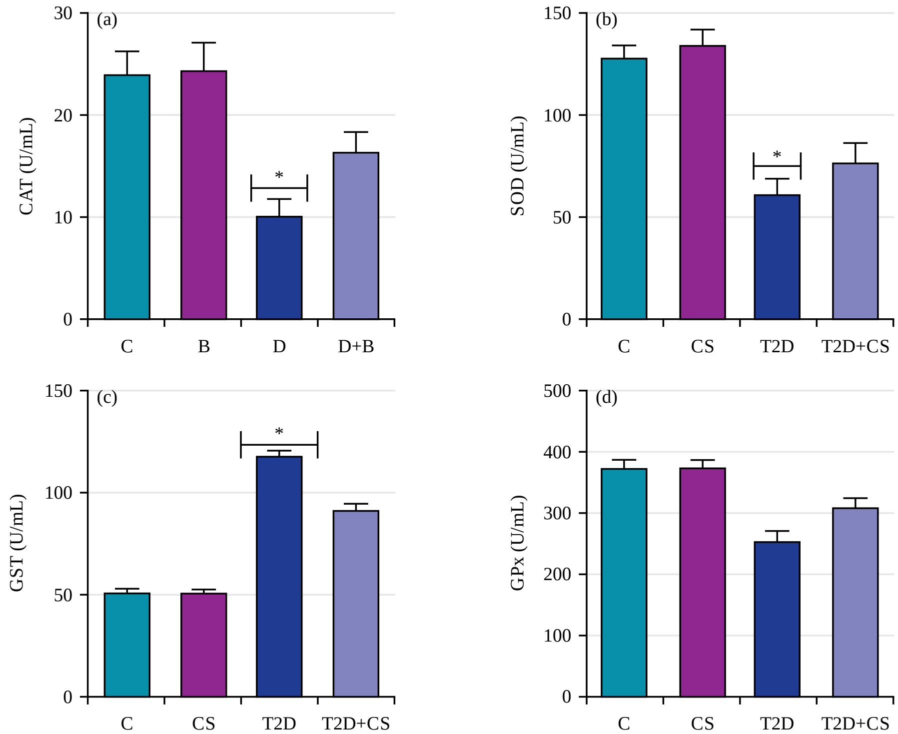

Fig. 4(a-d): Impact of the of barley (B) on various antioxidant parameters related to serum kidney function in rats with type 2 diabetes (D). The subplots delineate various parameters; (a) Catalase (CAT), (b) Superoxide dismutase (SOD), (c) Glutathione peroxidases (GPx) and (d) Glutathione-S-transferase (GST) activity (expressed as U/g tissue)

Data are presented as Means±Standard Deviation (n = 20). Statistical significance is denoted as p<0.05, p<0.01 versus D; #p<0.05 and ##p<0.01 versus B

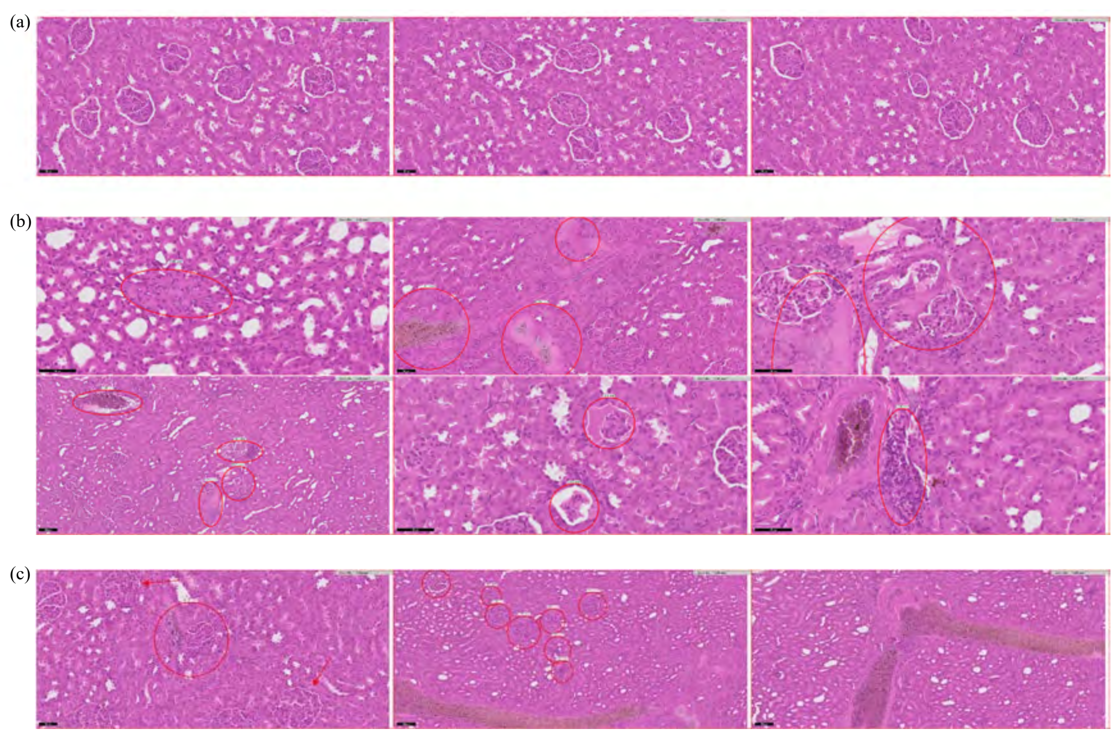

Fig. 5(a-g): Continue

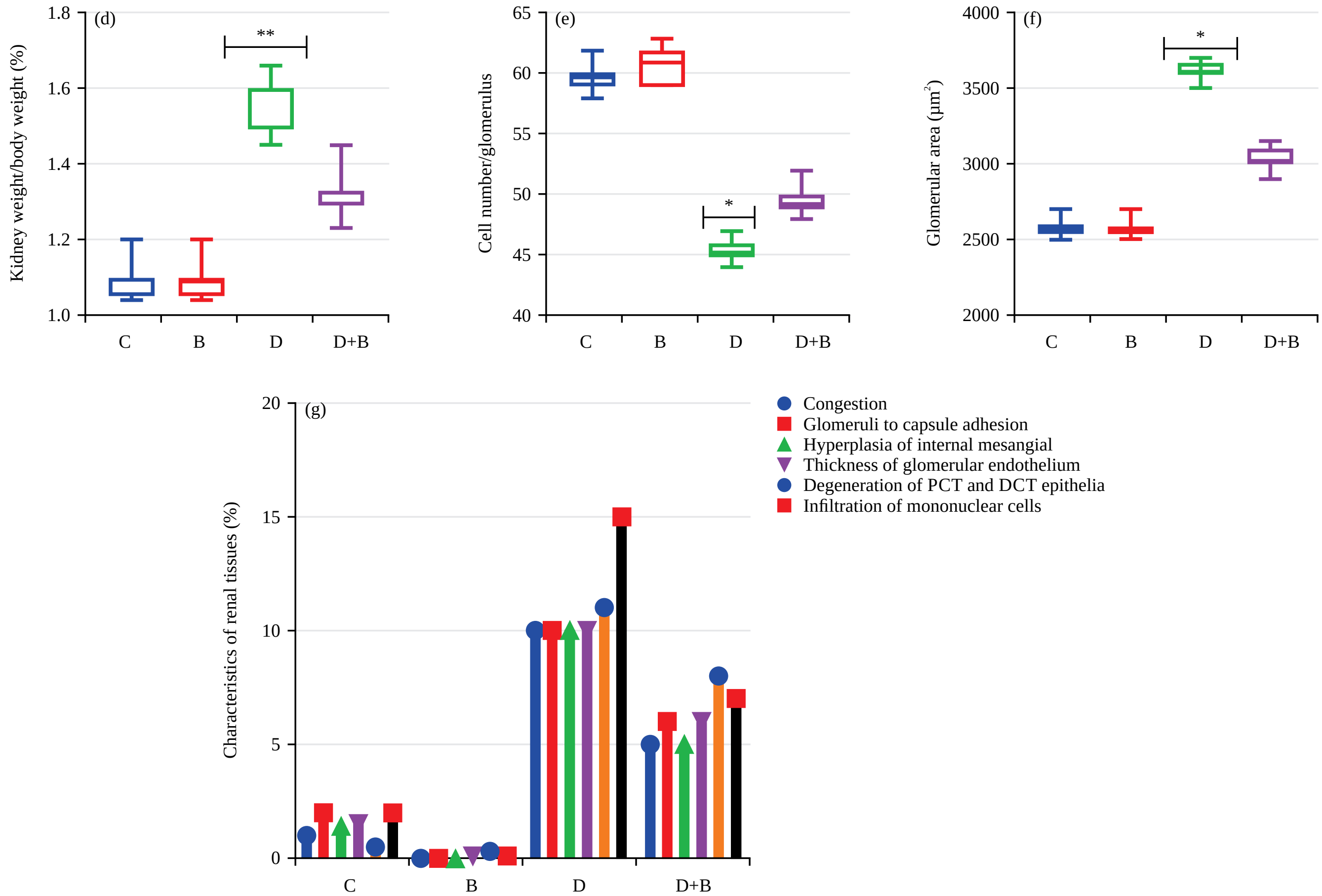

Fig. 5(a-g): (a) Comparison between control and barley-fed rat groups, (b) Histological photomicrograph of diabetic rat kidney tissue displays characteristic pathological changes of diabetic nephropathy, (c) Histology slide of diabetic rat kidney tissue treated with barley and (d-g) Kidney protective effects of barley against kidney tissue damage in D rats

(a) Reveals preserved normal renal histology, showcasing distinct podocytes around glomeruli and well-defined tubular boundaries, suggesting minimal impact of barley supplementation on renal tissue morphology; (b) Glomerular hypertrophy due to increased filtration pressure from high blood glucose levels, focal sclerosis indicating advanced kidney damage and tubular epithelial damage with vacuolization and tubular casts formation from factors released during diabetes (H&E staining); (c) Reveals improvements compared to untreated diabetic samples, with less glomerular hypertrophy and sclerosis, reduced tubular injury signs, fewer tubular casts and clearer kidney architecture. Barley's anti-hyperglycemic and antioxidant properties likely contributed to these protective changes, indicating partial reversal of diabetic kidney damage. While not fully preventive, barley treatment demonstrates Reno protective effects against hyperglycemia-induced alterations, urging further investigation into its therapeutic potential for diabetic nephropathy (H&E staining) and (d-g) Statistical significance is denoted as $p<0.05$, $p<0.01$ and $p<0.001$ compared to the normal control group. Additionally, $^{\#}p<0.05$ and $p<0.01$ are indicated versus the diabetic control group. The data are presented as median Interquartile Range (IQR), with n = 20 in each group

These findings collectively demonstrate the potential of barley as a natural therapeutic option for alleviating diabetes-induced renal damage through both structural and functional improvements.

DISCUSSION

Current research has demonstrated that diabetic rats display histological changes in their kidneys, including glomerular atrophy, widening of urinary spaces, cell swelling, hydropic degeneration of tubules, capillary congestion and tubular necrosis. Additionally, diabetic rats were found to have glomeruli adhering to the capsule, an increased number of glomerular cells, thickening of the glomerular endothelium and destruction of epithelial cells in both the distal and proximal convoluted tubules. These alterations signify renal dysfunction linked with diabetes, consistent with findings from Aly *et al.*[51]. Histopathological assessment indicated that supplementation with barley alleviated these damages in diabetic rats, suggesting potential protective properties of barley on kidney tissue under diabetic conditions, possibly due to its antioxidant characteristics.

Histopathological assessment indicated that supplementation with barley alleviated these damages in diabetic rats. This indicates that barley could potentially have protective properties on kidney tissue under diabetic conditions. The antioxidant characteristics of barley may contribute to reducing oxidative stress and safeguarding renal function.

Examining the influence of barley on blood glucose levels in diabetic rats is a fundamental aspect when considering its potential impact on renal function. Aly *et al.*[51] investigating the hypoglycemic, hypolipidemic and antioxidant effects of various dietary interventions in diabetic rats revealed that administering barley resulted in a notable decrease in fasting blood glucose levels. This reduction in blood glucose levels coincided with enhancements in the lipid profile of the diabetic rats.

Furthermore, another research focused on the consequences of incorporating whole barley flour into bread on serum glucose levels in obese rats[51]. The outcomes displayed that rat consuming barley-enriched bread experienced a marked drop in serum glucose levels compared to the control group. This further underscores the potential of barley as a dietary measure for regulating blood glucose levels in diabetic conditions.

Additionally, investigating the health-promoting properties of barley for diabetic rats unveiled that Barley treatment led to significantly lower fasting blood glucose levels compared to alternative treatment groups[52]. This implies that barley holds promise for reducing blood glucose levels in diabetic scenarios.

Overall, these investigations emphasize the encouraging influence of barley and related dietary interventions on blood glucose management in diabetic animal models. These results suggest that integrating barley into the diet could have positive implications for controlling blood glucose levels, potentially leading to enhanced renal function outcomes for individuals with diabetes.

The kidney function in diabetic rats was assessed by measuring the levels of creatinine and urea after administering barley interventions. The results indicated that the diabetic control group showed elevated levels of both urea and creatinine compared to the normal control group[51]. However, the groups receiving the barley interventions displayed similar levels of urea and creatinine as the normal control group. These results imply that the interventions barley, might offer a protective impact on kidney function in diabetic rats by maintaining normal levels of urea and creatinine. Therefore, including barley in the diet of diabetic individuals could potentially aid in preserving renal function and averting further complications linked with diabetic nephropathy.

Furthermore, incorporating whole barley flour into the diet has been associated with enhancements in glucose metabolism[51]. Rats fed whole barley bread exhibited a decrease in body weight gain compared to control groups. This decrease in weight gain can be attributed to reduced calorie and carbohydrate intake from the experimental diets[66], emphasizing the potential of barley as a dietary strategy for managing diabetes and obesity.

Additionally, barley has been shown to have positive effects on lipid profile in obese rats[67]. Diets high in fat typically result in elevated cholesterol and triglyceride levels, but supplementation with barley has led to a significant decrease in serum cholesterol and triglycerides[51]. This highlights the potential of barley to enhance lipid metabolism and overall cardiovascular health in diabetic individuals[68].

Barley has been found to have a notable impact on renal function markers in diabetic rats[17]. Research has illustrated that the consumption of barley, which is rich in beneficial compounds like -glucan, tocols and resistant starch, can lead to enhanced kidney health. The high fiber content of barley[69] aids in regulating blood sugar levels, making it especially advantageous for individuals with diabetes. In studies involving diabetic rat models[51], the administration of barley has been associated with a decrease in serum cholesterol levels, triglycerides and an overall improvement in lipid profiles compared to control groups[17].

Furthermore, studies have indicated that the intake of barley can diminish inflammation-induced[1] renal injury by targeting key enzymes such as ADAM17 and neutral-sphingomyelinase. Through reducing inflammatory cell infiltration and proteinuria, barley shows the potential to mitigate kidney damage and enhance overall renal function in diabetic conditions[43]. Additionally, dietary barley has been linked to a reduction in glomerular lesions, nodular sclerosis in the glomeruli and renal arteriolar lesions.

In conclusion, the evidence suggests that adding barley to the diet of diabetic rats can yield positive effects on renal function markers. From alleviating inflammation and enhancing lipid profiles to improving kidney damage and supporting overall kidney health, barley emerges as a promising dietary intervention for diabetic individuals facing renal complications.

Examination of kidney tissue in diabetic rats through histological analysis is essential to grasp the influence of barley on renal function. In summary, the histological examination of kidney tissue in diabetic rats treated with barley promising outcomes on renal function.

Naseri *et al.*[17] have demonstrated that diabetic rats exhibited degeneration in renal corpuscles and tubules, along with instances of intracellular hemorrhage. However, when diabetic rats were administered oral doses of fenugreek sprouts juice, barley sprouts juice, cell-free probiotic extract or whey protein, the structure of renal corpuscles and tubules returned to normal[17]. Results indicating a potential protective influence of these interventions on kidney health in diabetic circumstances. Furthermore, results from histopathological examinations have shown that supplementing with barley bran resulted in a restoration of the typical appearance of kidney tissue sections, with a decrease in cholesterol accumulation in the kidney tissues supplemented with barley bran[52], leading to clearly defined urinary tubules and collecting tubules. These findings suggested that incorporating barley into the diet may have a beneficial effect on the structure and function of renal tissue in diabetic rats, emphasizing the significance of dietary approaches like barley supplementation in enhancing renal function in diabetic scenarios.

Managing renal function in diabetic patients poses a significant challenge, but incorporating barley into their diet could be a valuable solution[17]. Research has shown that barley can play a crucial role in enhancing the well-being of diabetic rats. It has been demonstrated that barley has the ability to notably lower blood glucose levels and enhance lipid profiles in diabetic animals. Moreover, barley's phenolic compounds provide antioxidant properties that can help combat reactive oxygen species (ROS) which are elevated in diabetic individuals[34].

Furthermore, the positive impact of barley on kidney function biomarkers is evident through studies showing reductions in markers of kidney damage and overall improvements in kidney health among diabetic rats. Analysis of tissue has revealed histological changes indicating a protective effect on renal structures with barley supplementation[17].

These findings strongly suggest that including barley in the diet of diabetic patients could have substantial benefits for their renal function. By lowering blood glucose levels[20], improving lipid profiles[19] and safeguarding kidney health, barley[17] may offer a natural and efficient approach to managing complications associated with diabetes. Future investigations should delve deeper into the mechanisms underlying barley's advantageous effects on renal function and its potential as a dietary intervention for diabetic individuals. Future research on the impact of barley on renal function in diabetic rats should focus on understanding specific mechanisms, optimal dosage and duration, synergies with other functional ingredients, long-term effects, gut microbiota involvement and validation through clinical trials. Investigating how barley affects blood glucose regulation and kidney biomarkers is crucial, as well as determining the best treatment regimen. Exploring potential synergies with other dietary interventions can enhance therapeutic benefits, while long-term studies are needed to assess sustained impact and safety. Understanding how barley interacts with gut microbiota may provide insights for personalized dietary interventions. Finally, validating findings through clinical trials with diabetic patients will help establish evidence-based recommendations for improving renal function.

CONCLUSION

This study sheds light on the beneficial effects of barley dietary interventions in diabetic rats, particularly in enhancing antioxidant capacity and normalizing glucose levels and renal function. These findings suggest barley's potential as a complementary therapy for hyperglycemic conditions and in improving the progression of Diabetic Kidney Disease (DKD). Specifically, barley supplementation has been associated with reduced serum creatinine levels and increased estimated Glomerular Filtration Rate (eGFR). However, the exact mechanisms underlying barley's influence on renal function remain unclear, necessitating further investigation. Overall, results highlight the promise of dietary interventions like barley in enhancing renal function in diabetic rats, warranting further research to explore their implications for diabetic patients.

SIGNIFICANCE STATEMENT

The purpose of this study was to investigate the potential renoprotective effects of barley in the context of diabetes. Our key findings demonstrate that barley consumption exhibits promising effects in preserving renal function in diabetic animal models. This is significant as it suggests a potential dietary intervention for managing diabetic nephropathy, a common complication of diabetes. Moving forward, further research could explore the underlying mechanisms behind barley's renoprotective properties and its potential application in clinical settings as a complementary approach to managing diabetic kidney disease.

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