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The Relationship Between Nephrotoxic Serum Trace Elements and Renal Function Tests in Diabetic Nephropathy

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Abstract

Background

Diabetic nephropathy (DN) is a leading cause of chronic kidney disease (CKD) and end-stage renal disease (ESRD), with oxidative stress and trace element imbalances playing a significant role in its progression. This study investigates the relationship between nephrotoxic serum trace elements and renal function tests (RFT) in DN patients.

Methods

A cross-sectional study was conducted on 123 participants divided into four groups: diabetic patients without nephropathy (n=25), kidney disease patients without diabetes (n=22), diabetic nephropathy patients (n=20), and healthy controls (n=56). Serum levels of essential trace elements (iron, zinc, copper, magnesium) and nephrotoxic elements (arsenic, lead, mercury, cadmium) were analyzed using inductively coupled plasma mass spectrometry (ICP-MS). Renal function markers, including urea, creatinine, and blood urea nitrogen (BUN), were assessed. Statistical analyses included ANOVA, Pearson's correlation, and multivariate regression.

Results

Diabetic nephropathy patients exhibited significantly higher creatinine (92.8 ± 44.34 µmol/L, $p<0.0001$) and urea levels compared to other groups. Magnesium showed significant positive correlations with sodium ($r=0.490$, $p<0.05$), potassium ($r=0.605$, $p<0.01$), and calcium ($r=0.539$, $p<0.05$), while iron negatively correlated with uric acid ($r=-0.520$, $p<0.05$).

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Conclusion
Magnesium and iron play a crucial role in DN pathophysiology, with their dysregulation linked to renal dysfunction. Monitoring trace element levels could aid in DN management and early intervention strategies.

Keywords
Diabetic nephropathy, trace elements, renal function, oxidative stress, magnesium, iron, nephrotoxicity

Introduction

Diabetes mellitus (DM) is a chronic metabolic disorder affecting millions worldwide, with an increasing prevalence that poses a major public health concern. Type 2 diabetes mellitus (T2DM) accounts for over 90% of all diabetes cases and is characterized by insulin resistance and progressive pancreatic beta-cell dysfunction, leading to chronic hyperglycemia and metabolic derangements [1]. Among the long-term complications of diabetes, diabetic nephropathy (DN) is a leading cause of chronic kidney disease (CKD) and end-stage renal disease (ESRD), necessitating dialysis or kidney transplantation [2, 3]. Despite advances in glycemic control and antihypertensive therapies, the incidence of DN continues to rise, emphasizing the need for novel biomarkers and mechanistic insights into its pathogenesis [4].

Trace elements play crucial roles in cellular metabolism, enzymatic reactions, and antioxidant defense systems, yet their involvement in DN remains an area of intense investigation. Essential trace elements such as zinc (Zn), copper (Cu), iron (Fe), and magnesium (Mg) contribute to antioxidant enzyme function and glucose metabolism, whereas nephrotoxic trace elements—including arsenic (As), lead (Pb), cadmium (Cd), and mercury (Hg)—have been implicated in oxidative stress, inflammation, and renal toxicity [5]. The balance between these elements is critical, as deficiencies in protective elements or excess accumulation of nephrotoxic metals may accelerate renal dysfunction in diabetic patients. However, whether diabetic nephropathy arises primarily due to a deficiency in protective trace elements or an overload of nephrotoxic elements remains an unresolved question [6].

The Role of Oxidative Stress and Nephrotoxicity in Diabetic Nephropathy

The pathophysiology of DN is multifactorial, involving hyperglycemia-induced oxidative stress, inflammatory responses, endothelial dysfunction, and dysregulated mineral metabolism [7, 8]. Chronic hyperglycemia promotes the production of reactive oxygen species (ROS) via multiple pathways, including glucose auto-oxidation, advanced glycation end-product (AGE) formation, and mitochondrial dysfunction [8]. Excessive ROS disrupt cellular homeostasis, leading to lipid peroxidation, protein oxidation, and DNA damage, which contribute to renal injury [9].

Trace elements are key modulators of oxidative stress. Zn, for instance, plays a fundamental role in insulin metabolism, enhances the activity of antioxidant enzymes like superoxide dismutase (SOD), and stabilizes cell membranes against oxidative damage [10]. Conversely, nephrotoxic metals such as Pb, As, and Cd exert their toxic effects by displacing essential metals, disrupting enzyme function, and generating free radicals [11]. The accumulation of these elements in renal tissue has been linked to tubular toxicity, glomerular damage, and impaired filtration capacity, exacerbating the progression of DN [12].

A growing body of evidence suggests that nephrotoxic trace elements contribute to the pathogenesis of DN by inducing renal inflammation and fibrosis [13, 14]. As is a potent disruptor of cellular function; it inhibits mitochondrial oxidative phosphorylation, depletes cellular glutathione levels, and alters calcium homeostasis, all of which contribute to renal endothelial dysfunction [15]. Pb, another widespread environmental pollutant, has been shown to interfere with nitric oxide bioavailability, promote renal vasoconstriction, and induce

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apoptosis in proximal tubular cells [16]. Cd, a known nephrotoxin, accumulates in the renal cortex, leading to chronic inflammation and fibrosis, thereby exacerbating the decline in renal function [17].

Emerging Paradigm: Dual Hypothesis of Diabetic Nephropathy Pathogenesis

Historically, DN has been attributed to hyperglycemia-induced damage, but recent perspectives highlight the role of trace element imbalances in exacerbating renal dysfunction. The "dual hypothesis" suggests that DN may result from both a deficiency in antioxidant trace elements (such as Zn and Mg) and the accumulation of nephrotoxic trace elements (such as As, Pb, and Cd) [18]. This model proposes that oxidative stress alone may not fully explain DN progression, but rather an intricate interplay between protective and toxic elements influences disease severity [19].

Recent studies indicate that elevated serum Pb levels correlate with increased serum creatinine and reduced glomerular filtration rate (GFR) in diabetic patients, suggesting a direct nephrotoxic impact [20]. Similarly, higher urinary As levels have been linked to proteinuria and kidney damage, further reinforcing the toxic metal hypothesis [21]. However, research on the combined effects of multiple trace elements on DN remains limited, necessitating further investigation.

Rationale for the Study

Despite extensive research on the metabolic and inflammatory aspects of DN, the role of trace elements—particularly nephrotoxic metals—has not been fully elucidated. Most studies focus on individual elements rather than exploring the combined influence of protective and toxic elements on renal function [22]. Given that trace elements interact dynamically within biological systems, a comprehensive analysis of their correlation with renal function tests (RFT) could provide valuable insights into the pathogenesis of DN [23].

Saudi Arabia has one of the highest global prevalence rates of diabetes, with an increasing burden of DN contributing to ESRD cases [24]. Identifying novel biomarkers for early detection and risk stratification is crucial for preventing disease progression and reducing healthcare costs. By investigating the relationship between nephrotoxic serum trace elements and renal function, this study aims to bridge a critical knowledge gap and potentially inform future therapeutic strategies for DN [25].

Objective of the Study

The primary objective of this study is to investigate the association between serum levels of nephrotoxic trace elements (As, Pb, Cd, Hg) and renal function tests (urea, creatinine, and BUN) in diabetic patients with nephropathy. This study also aims to explore whether nephrotoxic elements could serve as potential biomarkers for DN progression and whether their accumulation correlates with declining renal function.

By addressing these questions, this study seeks to contribute to the growing field of trace element research in diabetes-related kidney disease and offer novel insights into the complex interplay between environmental exposures and metabolic disorders.

Methods

Study Design

This study was conducted as a cross-sectional observational study to investigate the association between nephrotoxic serum trace elements and renal function tests (RFT) in patients with diabetic nephropathy (DN). A cross-sectional design was chosen to allow for the simultaneous measurement of exposure (trace element levels) and outcomes (renal function parameters) in a defined population at a single point in time. This approach is particularly effective for identifying correlations and patterns between trace element concentrations and renal function markers without requiring prolonged follow-up periods.

Study Setting

The study was conducted at King Abdulaziz Air Base Hospital in Dhahran, Saudi Arabia, a tertiary healthcare facility that provides specialized care for patients with chronic conditions, including diabetes and kidney disease. Patients were recruited from the outpatient, maternity, urology clinics, and emergency department between December 2021 and May 2022. The hospital's diverse patient population allowed for the selection of cases with varying degrees of renal impairment, ranging from diabetic patients without nephropathy to those with established diabetic nephropathy.

Sample and Sampling

A total of 123 participants were recruited using a randomized stratified sampling method to ensure representative distribution across different study groups. The participants were divided into four distinct groups:

- Group I (Diabetic Patients Without Nephropathy): 25 patients with type 2 diabetes mellitus (T2DM) but no clinical evidence of nephropathy.
- Group II (Kidney Disease Group - Non-Diabetic CKD Patients): 22 patients with chronic kidney disease (CKD) unrelated to diabetes.
- Group III (Diabetic Nephropathy Group): 20 patients with T2DM and diagnosed diabetic nephropathy based on urinary albumin excretion and renal function parameters.
- Group IV (Control Group): 56 healthy individuals without diabetes or kidney disease, recruited as the reference group.

The inclusion criteria for the diabetic groups were based on the American Diabetes Association (ADA) diagnostic criteria for T2DM, which include fasting plasma glucose (FPG) ≥126 mg/dL, hemoglobin A1c (HbA1c) ≥6.5%, or a 2-hour plasma glucose ≥200 mg/dL during an oral glucose tolerance test (OGTT). Patients in the diabetic nephropathy group were diagnosed based on increased urinary albumin excretion (UAE >20 μg/min) and a history of persistent albuminuria (>0.5 g/24 h) without other renal diseases.

The exclusion criteria included patients with type 1 diabetes mellitus (T1DM), those with a duration of diabetes of less than three years, individuals on mineral or vitamin supplements, pregnant women, patients with chronic liver or endocrine disorders, and those with active infections or inflammatory diseases.

Data Collection Tools

Data collection was carried out using a structured questionnaire and laboratory analyses. The questionnaire gathered demographic data (age, sex, duration of diabetes, medical history), while blood and urine samples were collected for biochemical and trace element analysis. The laboratory tests included serum glucose, urea, creatinine, uric acid, and HbA1c, measured using the ARCHITECT Clinical Chemistry Analyzer (Abbott, Wiesbaden, Germany). Serum trace element concentrations (Zn, Cu, Fe, Mg, As, Pb, Cd, Hg) were measured using inductively coupled plasma mass spectrometry (ICP-MS) and atomic absorption spectroscopy (AAS).

Data Collection Procedure

All participants provided informed consent before sample collection. After a 10-hour overnight fast, a 10 mL venous blood sample was drawn from each participant. Blood was collected into two separate tubes:

- One tube containing EDTA for HbA1c measurement.
- Another plain tube for serum separation and trace element analysis.

The blood samples were immediately centrifuged at 3000 rpm for 10 minutes to separate the serum, which was then stored at -70°C until analysis. Serum trace elements were quantified using ICP-MS (Thermo X-Series 2, Bremen, Germany) following a rigorous sample digestion protocol. The digestion process involved microwave-assisted acid digestion using nitric acid (HNO_3) and hydrogen peroxide (H_2O_2) to ensure complete mineralization of organic components. The microwave digestion program was standardized at

200°C with a ramp time of 7 minutes and a hold time of 9 minutes. All samples were analyzed in triplicate, and appropriate quality control measures, including reagent blanks and standard reference materials, were employed to validate the results.

Renal function tests were assessed by measuring serum urea, creatinine, and blood urea nitrogen (BUN). The estimated glomerular filtration rate (eGFR) was calculated using the Modification of Diet in Renal Disease (MDRD) equation to evaluate kidney function.

Data Analysis

Statistical analysis was conducted using IBM SPSS Statistics (Version 27) to evaluate the relationship between nephrotoxic serum trace elements and renal function tests (RFT) in diabetic nephropathy. Descriptive statistics, including means, standard deviations, and 95% confidence intervals, were used to summarize the demographic data and laboratory findings. The Kolmogorov-Smirnov test was applied to assess the normality of data distribution, and appropriate parametric or non-parametric tests were chosen accordingly. To compare mean serum trace element levels and renal function parameters across the four study groups, one-way analysis of variance (ANOVA) was used, followed by Tukey's post hoc test for pairwise comparisons. For normally distributed variables, independent t-tests were employed to compare differences between two specific groups, whereas the Mann-Whitney U test was applied for non-normally distributed data. Correlations between serum trace elements and renal function markers (urea, creatinine, and blood urea nitrogen) were analyzed using Pearson's correlation coefficient to determine the strength and direction of associations. Additionally, multivariate linear regression analysis was performed to adjust for potential confounders, such as age, sex, and duration of diabetes, ensuring robust findings. A p-value of less than 0.05 was considered statistically significant in all analyses, and results were presented with corresponding effect sizes and confidence intervals to enhance interpretability.

Ethical Considerations

This study was conducted in compliance with the ethical principles of the Declaration of Helsinki and was approved by the Ethical Committee of the Deanship of Scientific Research, King Faisal University (Approval Reference: EA000627). All participants provided written informed consent before enrollment. Confidentiality was maintained by anonymizing participant data, and samples were coded to ensure privacy. Patients were informed about their right to withdraw from the study at any time without any consequences.

Additionally, all laboratory procedures were conducted following biosafety protocols to prevent contamination and ensure sample integrity. Proper disposal of biohazardous waste, including blood samples and chemical reagents, was carried out in accordance with hospital and Saudi Ministry of Health (MOH) regulations.

Results

Demographic Data

The demographic characteristics of the study participants are shown in **Table 1**. A total of 123 individuals were enrolled and classified into four study groups: diabetic patients without nephropathy (Group I, n=25), chronic kidney disease (CKD) patients without diabetes (Group II, n=22), diabetic nephropathy (DN) patients (Group III, n=20), and healthy controls (Group IV, n=56). The diabetic group had the highest mean age (48.76 ± 12.72 years), while the control group was the youngest (33.78 ± 18.76 years).

Table 1: Demographic Characteristics of Study Participants

Group	N	Mean Age (years) ± SD	Age Range (years)

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Diabetic (I)	25	48.76 ± 12.72	25 - 77
Kidney Disease (II)	22	37.59 ± 18.23	16 - 81
Diabetic Nephropathy (III)	20	42.50 ± 25.14	5 - 84
Control (IV)	56	33.78 ± 18.76	0.67 - 77

Laboratory Findings

The laboratory findings in Table 2 highlight significant differences in renal function parameters and glycemic control across the study groups. The diabetic nephropathy (DN) group exhibited the highest creatinine levels (92.8 ± 44.34 µmol/L) compared to all other groups, indicating impaired glomerular filtration and reduced renal clearance. Similarly, urea (BUN) levels were markedly elevated in the kidney disease group (32.46 ± 10.66 mmol/L), significantly higher than in diabetic and control groups ($p < 0.0001$), suggesting progressive renal dysfunction and nitrogenous waste accumulation in CKD patients.

The BUN/Creatinine ratio was substantially higher in the kidney disease group (56.98 ± 19.14), reflecting the chronic impact of kidney impairment on nitrogen balance. However, in the diabetic and diabetic nephropathy groups, this ratio remained within normal limits, suggesting a primary renal excretory dysfunction rather than prerenal azotemia.

Glycemic control, as reflected by HbA1c levels, was significantly elevated in the diabetic nephropathy group (7.49 ± 1.99%), indicating poor long-term glucose regulation, which is a major contributing factor to nephropathy progression. In contrast, the control and kidney disease groups had significantly lower HbA1c values (~5.1%), reflecting normal glucose metabolism. The significantly higher HbA1c levels in the DN group compared to the diabetic group (7.49% vs. 7.26%) suggest a possible link between worsening glycemic control and the progression of nephropathy.

Table 2: Laboratory Findings in Study Groups

Parameters (Normal Range)	Diabetic (I)	Kidney Disease (II)	Diabetic Nephropathy (III)	Control (IV)	p-value
Urea (BUN) (3.5-6.7 mmol/L)	4.44 ± 0.88	32.46 ± 10.66*	5.93 ± 3.59	4.28 ± 1.04	< 0.0001
Creatinine (50-98 µmol/L)	68.89 ± 12.45	67.3 ± 51.94	92.8 ± 44.34*	64.2 ± 14.98	< 0.0001
BUN/Creatinine Ratio (6.0-25 mmol/L)	6.63 ± 1.73	56.98 ± 19.14*	6.42 ± 2.35	6.89 ± 2.01	< 0.0001
HbA1c (%)	7.26 ± 2.25	5.16 ± 0.51	7.49 ± 1.99*	5.06 ± 0.39	0.001

*Significant difference compared to all other groups ($p < 0.0001$).

Serum Trace Elements Levels

Table 3 show The analysis of serum trace elements across the study groups reveals significant differences in magnesium (Mg) levels, whereas iron (Fe), zinc (Zn), and copper (Cu) did not show statistically significant variations. Magnesium levels were significantly lower in the diabetic nephropathy (DN) group (0.73 ± 0.09 mg/dL) compared to the control group (0.79 ± 0.08 mg/dL, $p = 0.026$), suggesting a possible role of

magnesium deficiency in the pathophysiology of diabetic nephropathy. Magnesium is a crucial cofactor in enzymatic antioxidant systems and glucose metabolism, and its deficiency has been associated with insulin resistance, oxidative stress, and endothelial dysfunction, all of which contribute to renal impairment in diabetic patients.

Although iron (Fe) levels were slightly higher in the diabetic group (17.26 ± 6.89 µmol/L) compared to other groups, the difference was not statistically significant. Iron plays a vital role in oxidative metabolism, but excessive iron accumulation can contribute to oxidative damage in diabetic nephropathy. However, in this study, iron levels remained within a comparable range across all groups, indicating no major alterations due to diabetes or nephropathy.

Zinc (Zn) and copper (Cu) levels also showed no significant differences among the groups, despite a slightly higher mean zinc level in the diabetic nephropathy group (20.2 ± 12.88 µg/dL) compared to other groups. Zinc is essential for antioxidant enzyme function and insulin metabolism, and while its levels appear elevated in diabetic nephropathy patients, the lack of statistical significance suggests that the variation may not be clinically relevant. Similarly, copper (Cu) levels were highest in the diabetic nephropathy group (26.84 ± 18.74 µg/dL) but did not reach statistical significance, indicating that copper metabolism remains relatively stable across the study populations.

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Table 3: Serum Trace Elements Levels in Study Groups

Trace Element	Diabetic (I)	Kidney Disease (II)	Diabetic Nephropathy (III)	Control (IV)	p-value
Fe (µmol/L)	17.26 ± 6.89	14.92 ± 5.01	15.77 ± 5.25	14.09 ± 5.71	NS
Mg (mg/dL)	0.78 ± 0.06	0.75 ± 0.09	**0.73 ± 0.09**	**0.79 ± 0.08**	0.026
Zn (µg/dL)	16.29 ± 10.9	15.75 ± 7.46	20.2 ± 12.88	15.99 ± 6.35	NS
Cu (µg/dL)	24.51 ± 18.74	22.73 ± 11.68	26.84 ± 18.74	23.63 ± 10.72	NS

NS: Not Significant, **p < 0.05** (significant vs. control group).

Table 4 presents the serum levels of nephrotoxic trace elements (arsenic, lead, mercury, and cadmium) across the study groups. Arsenic (As) levels were significantly higher in diabetic patients (0.61 ± 1.41 µg/dL) compared to all other groups, particularly the control group (0.04 ± 0.18 µg/dL, p = 0.010), suggesting a possible link between arsenic exposure and diabetes. The diabetic nephropathy group had lower arsenic levels than the diabetic group, indicating that arsenic may play a role in the early stages of diabetes but might not be a direct contributor to nephropathy progression.

Lead (Pb) levels were also significantly elevated in the diabetic group (8.58 ± 3.61 µg/dL) compared to kidney disease (4.4 ± 2.59 µg/dL), diabetic nephropathy (3.92 ± 2.38 µg/dL), and control (1.64 ± 1.83 µg/dL) groups (p < 0.0001). This suggests a potential relationship between lead exposure and diabetes, possibly through oxidative stress or altered insulin metabolism. The lower lead levels in the diabetic nephropathy group may indicate increased renal excretion or sequestration in tissues as kidney function declines.

Mercury (Hg) levels showed an inverse trend, with the highest mean concentration in the control group (1.14 ± 0.81 µg/dL), followed by diabetic nephropathy (1.04 ± 0.38 µg/dL), kidney disease (0.8 ± 0.35 µg/dL), and the lowest in the diabetic group (0.54 ± 0.50 µg/dL, p = 0.002). This finding suggests that diabetes may be associated with lower mercury levels, possibly due to altered renal handling or dietary differences. The

slightly higher mercury levels in the nephropathy group compared to diabetics without nephropathy could indicate impaired renal mercury clearance as kidney function worsens.

Cadmium (Cd) levels were below detectable limits in all groups, indicating minimal exposure or effective renal excretion in the study population. This is consistent with previous research suggesting that cadmium accumulation primarily occurs in chronic environmental exposure rather than acute diabetes-related processes.

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Table 4: Serum Nephrotoxic Trace Elements Levels in Study Groups

Element	Diabetic (I)	Kidney Disease (II)	Diabetic Nephropathy (III)	Control (IV)	p-value
As (µg/dL)	**0.61 ± 1.41**	0.22 ± 0.44	0.12 ± 0.29	**0.04 ± 0.18**	0.010
Pb (µg/dL)	**8.58 ± 3.61**	4.4 ± 2.59	3.92 ± 2.38	**1.64 ± 1.83**	< 0.0001
Hg (µg/dL)	0.54 ± 0.50	0.8 ± 0.35	1.04 ± 0.38	**1.14 ± 0.81**	0.002
Cd (µg/dL)	0	0	0	0	-

Correlation Between Serum Trace Elements and Renal Function

The data presented in Table 5 highlights significant correlations between select serum trace elements and renal function markers in diabetic nephropathy (DN) patients. Iron (Fe) showed a significant negative correlation with uric acid ($r = -0.520$, $p < 0.05$), suggesting that as serum iron levels increase, uric acid levels tend to decrease. This inverse relationship may indicate an altered iron metabolism affecting purine degradation pathways in DN patients, potentially influencing oxidative stress and renal impairment. Conversely, Fe exhibited a positive correlation with chloride ($r = 0.467$, $p < 0.05$), which may reflect iron's role in electrolyte homeostasis and kidney function regulation.

Magnesium (Mg) demonstrated multiple significant positive correlations with renal function markers, particularly with sodium ($r = 0.490$, $p < 0.05$), potassium ($r = 0.605$, $p < 0.01$), and calcium ($r = 0.539$, $p < 0.05$). These findings suggest that lower magnesium levels may contribute to electrolyte imbalances in DN patients, potentially exacerbating kidney dysfunction and metabolic disturbances. The strong correlation with potassium, in particular, indicates a possible role of magnesium in maintaining potassium stability, which is critical for kidney function and cardiovascular health in diabetic patients

Table 5: Correlation Between Serum Trace Elements and RFT

Element	RFT Parameter	Group	Correlation (r)	p-value
Fe	Uric Acid	DN	-0.520	< 0.05
Fe	Chloride	DN	0.467	< 0.05
Mg	Na	DN	0.490	< 0.05
Mg	K	DN	0.605	< 0.01
Mg	Ca	DN	0.539	< 0.05

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Discussion

Diabetic nephropathy (DN) is a progressive renal complication of diabetes mellitus and a leading cause of end-stage renal disease (ESRD) worldwide. Despite significant advancements in glycemic control and renoprotective therapies, DN remains a major challenge due to its multifactorial pathogenesis, which involves oxidative stress, inflammation, and trace element dysregulation [13, 26]. This study investigated the association between nephrotoxic serum trace elements and renal function in diabetic nephropathy patients, revealing key correlations that may enhance our understanding of the disease process and potential biomarkers for early detection and progression monitoring.

Trace elements are essential micronutrients that regulate enzymatic functions, antioxidant defenses, and cellular homeostasis. While essential trace elements such as iron (Fe), zinc (Zn), copper (Cu), and magnesium (Mg) play protective roles in metabolic regulation, nephrotoxic trace elements like arsenic (As), lead (Pb), cadmium (Cd), and mercury (Hg) have been implicated in renal toxicity through oxidative stress and inflammatory pathways [27]. The present study found a significant negative correlation between serum iron (Fe) and uric acid ($r = -0.520$, $p < 0.05$) in DN patients, suggesting an inverse relationship between iron homeostasis and renal function decline. This finding aligns with previous reports indicating that iron dysregulation contributes to renal dysfunction through iron-induced oxidative stress, which leads to endothelial damage and tubular injury [28]. Elevated iron levels can catalyze the formation of reactive oxygen species (ROS) via the Fenton reaction, exacerbating oxidative stress and accelerating the progression of DN (7).

Furthermore, iron exhibited a positive correlation with serum chloride ($r = 0.467$, $p < 0.05$) in DN patients, which may indicate an alteration in electrolyte balance in response to renal impairment. Chloride plays a crucial role in maintaining acid-base homeostasis and tubular reabsorption, and changes in its levels could be a compensatory response to renal dysfunction [29]. Since hyperchloremia has been associated with renal injury and increased mortality risk in chronic kidney disease (CKD) patients, this correlation may further highlight iron's involvement in renal pathophysiology [30].

Magnesium (Mg) is a critical cofactor for enzymatic reactions, insulin metabolism, and antioxidant enzyme activity. The findings of this study revealed significant positive correlations between serum magnesium and multiple electrolytes, including sodium ($r = 0.490$, $p < 0.05$), potassium ($r = 0.605$, $p < 0.01$), and calcium ($r = 0.539$, $p < 0.05$) in DN patients. These associations emphasize the potential role of magnesium in maintaining electrolyte stability in diabetic kidney disease [31]. Magnesium depletion is commonly observed in DN patients, and studies have shown that hypomagnesemia contributes to insulin resistance, increased oxidative stress, and vascular complications in diabetes [32, 33].

The strongest correlation was observed between magnesium and potassium ($r = 0.605$, $p < 0.01$), suggesting that magnesium deficiency may exacerbate potassium dysregulation in DN. Hypomagnesemia has been shown to impair potassium retention, leading to hypokalemia, which further worsens kidney function and predisposes patients to cardiovascular complications [34]. The positive correlation between magnesium and calcium ($r = 0.539$, $p < 0.05$) is also significant, as calcium homeostasis is tightly regulated by renal function, and disturbances in magnesium levels can impact calcium reabsorption and bone health in diabetic patients [35]. These findings align with previous reports suggesting that magnesium supplementation may improve metabolic and renal outcomes in DN patients by mitigating oxidative stress and restoring electrolyte balance [36].

Accumulation of nephrotoxic trace elements such as arsenic, lead, cadmium, and mercury has been implicated in renal toxicity and DN progression [37, 38]. While this study primarily focused on essential trace elements, the significant correlations between iron, magnesium, and renal function markers underscore the potential interplay between nephrotoxic metals and antioxidant trace elements in DN. Arsenic exposure, for example, has been linked to increased proteinuria, oxidative stress, and endothelial dysfunction, all of which contribute to the progression of DN [6]. Similarly, lead has been associated with glomerular damage and accelerated renal function decline in diabetic patients [39].

Although cadmium levels were below detectable limits in this study, previous research has shown that cadmium exposure promotes tubular dysfunction and inflammation in CKD [40]. Given the known interactions between nephrotoxic elements and essential trace elements, it is possible that elevated levels of toxic metals disrupt the bioavailability of protective elements such as magnesium, zinc, and iron, further exacerbating oxidative stress and inflammation in DN patients [41]. Future studies should investigate the combined effects of nephrotoxic and essential trace elements in DN progression, as this could provide novel insights into therapeutic interventions aimed at restoring trace element homeostasis.

The findings of this study suggest that serum iron and magnesium levels may serve as potential biomarkers for DN progression, given their significant correlations with renal function markers. Monitoring iron homeostasis could provide insights into oxidative stress-related damage, while magnesium levels may reflect electrolyte imbalances that contribute to DN complications. Given the observed relationships, magnesium supplementation may represent a potential therapeutic strategy for DN patients with hypomagnesemia, as previous studies have demonstrated its renoprotective effects [42].

Additionally, the observed correlations between magnesium and electrolyte markers (Na, K, Ca) highlight the importance of comprehensive electrolyte monitoring in DN patients to prevent complications such as hypokalemia, metabolic acidosis, and cardiovascular dysfunction [43]. Future clinical trials should explore whether correcting magnesium deficiency can improve renal function and slow DN progression in diabetic patients.

Another important aspect is the need for early detection of trace element imbalances in DN patients. Routine screening for essential and nephrotoxic trace elements in diabetic individuals may allow for personalized treatment strategies aimed at optimizing mineral balance and reducing oxidative damage. The integration of trace element monitoring into standard DN management protocols could enhance risk stratification and help identify high-risk patients who may benefit from nutritional interventions or chelation therapies.

Despite these promising findings, this study has some limitations. The cross-sectional design prevents the establishment of causal relationships between trace element levels and DN progression. Additionally, dietary intake and environmental exposures were not controlled, which may influence trace element variability among participants. Future longitudinal studies are needed to confirm these associations and assess whether modulating trace element levels through dietary or pharmacological interventions can alter DN outcomes.

Conclusion

This study provides compelling evidence that serum iron and magnesium levels are significantly correlated with renal function markers in diabetic nephropathy patients, reinforcing the role of trace elements in DN pathophysiology. The inverse relationship between iron and uric acid suggests a potential link between iron metabolism and renal impairment, while positive correlations between magnesium and multiple electrolytes highlight its role in maintaining electrolyte balance in DN. These findings underscore the need for further

research into trace element-based interventions as potential strategies for mitigating DN progression. Incorporating trace element monitoring into routine DN management could enhance early detection and inform targeted therapies to improve renal outcomes in diabetic patients.

Declarations

Funding

No specific funding was received for this study.

Conflicts of Interest

The authors declare that they have no competing interests.

Ethics Approval and Consent to Participate

The study was approved by the Ethical Committee of the Deanship of Scientific Research, King Faisal University (Approval Reference: EA000627). All participants provided written informed consent.

Consent for Publication

Not applicable.

Availability of Data and Materials

The datasets generated and/or analyzed during the current study are available from the corresponding author upon reasonable request.

Authors' Contributions

All authors contributed substantially to the conception, design, data collection, analysis, manuscript drafting, and approval of the final version.

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