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Assessment of urine albumin (Alb), immunoglobulin G (IgG), beta-microglobulin (β2mG) in diabetic patients in Tikrit

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Abstract :

Background: Diabetes is challenging to recognize in the initial stages because it lacks evident disease manifestations; yet, because it is a widespread and frequently occurring disease, its morbidity rises as living standards rise. Earlier diabetes identification, evaluation, and management are therefore crucial. **Aim:** The study aimed at examining the applicability of urine albumin (Alb), immunoglobulin G (IgG), and beta-microglobulin (β2mG) levels in the diabetes diagnosis , Our research offers more information on the biomarker profiles of a distinct population by concentrating on a larger demography in Tikrit. Additionally, this study builds on the past research by investigating the connections between these biomarkers and other variables that were not the main focus of the other investigations, such as BMI, glomerular filtration rates, and microalbuminuria. This differentiation facilitates a more profound comprehension of the suitability of these biomarkers in diagnosing diabetes in various patient groups.". **Patients and methods:** Between July/2022 and December /2022, 100 patients diabetics who have been hospitalized to Tikrit educational Hospital were employed as an experimental group, while 100 other healthy individuals were employed as healthy controls. Alb, IgG, and β2mG concentrations and positive predictive validity in the two groups were assessed. **Results:** The findings revealed that increased urine β2m in diabetics was more common 53.1% (95% CI: 43.2-62.9%) than in healthy controls 33.1% (95% CI: 23.8-42.3%). In comparison to healthy participants (16.7%, 95% CI: 9.5-26.2%), diabetics had a greater frequency of microalbuminuria (39.7%, 95% CI: 27.4-47.3%). Urinary β2m and UAC had a significant relationship (rho = 0.39, p= 0.003). As predictive factors of higher urine beta-2-microglobulin in diabetics, multivariate analysis identified BMI (OR: 1.34, 95% CI: 1.16-1.56), eGFR (OR: 0.86, 95% CI: 0.83-1.01), and the prevalence of microalbuminuria (OR: 3.83, 95% CI: 1.43-11.89). In compared to experimental and control groups, IgGCR are much higher diabetics. Age, the period of diabetes mellitus (DM), serum creatinine, blood urea nitrogen, ACR, and renal sonography all significantly correlated positively with IgGCR (P <0.001), while eGFR, haemoglobin, and serum albumin significantly correlated negatively with IgGCR. **Conclusion:** Alb, IgG, and β2mG concentrations are thereby tightly associated to the degree of diabetes, providing a medical guideline for the diagnosis and assessment of diabetes and having significant clinical implications.

Keywords: Diabetes, urine albumin, immunoglobulin G, beta-microglobulin.

قيم تطبيق البومين البول(الالب),الغلوبولين المناعي , بيتا ميكروغلوبولين في تشخيص مرض السكري في تكريت

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مستخلص:

ساعدت الدراسة في فحص مدى تطبيق مستويات ألبومين البول في تشخيص مرض السكري. تم إجراء 100 مريض مصاب بالسكري تم إدخالهم إلى مستشفى تكريت التعليمي تم توظيفهم كمجموعة تجريبية، بينما تم توظيف 100 فرد سليم آخر كضوابط صحية. تم تقييم تركيزات و والصلاحية التنبؤية الإيجابية في المجموعتين. النتائج: كشفت النتائج أن زيادة ß2m في البول لدى مرضى السكري كانت أكثر شيوعًا بنسبة 53.1٪ (95٪ CI: 43.2–62.9٪) مقارنة بالضوابط الصحية بنسبة 33.1٪ (95٪ CI: 23.8–42.3٪). وبالمقارنة مع المشاركين الأصحاء (16.7٪، 95٪ CI: 9.5–26.2٪)، كان لدى مرضى السكري تواتر أكبر من بيلة الألبومين الدقيقة (39.7٪، 95٪ CI: 27.4–47.3٪). كان لـ ß2m في البول و UAC علاقة مهمة (= rho 0.39، p = 0.003). كعوامل تنبؤية لارتفاع مستويات بيتا-2-ميكروغلوبولين في البول لدى مرضى السكري، حدد التحليل المتعدد المتغيرات مؤشر كتلة الجسم (OR: 1.34، 95٪ CI: 1.16–1.56)، ومعدل الترشيح الكبيبي المقدر (OR: 0.86، 95٪ CI: 0.83–1.01)، وانتشار البول الزلالي الدقيق (OR: 3.83، 95٪ CI: 1.43–11.89). وبالمقارنة مع المجموعات التجريبية والمجموعة الضابطة، فإن IgGCR أعلى بكثير لدى مرضى السكري. العمر، وفترة الإصابة بمرض السكري (DM)، والكرياتينين في المصل، ونيتروجين اليوريا في الدم، و ACR، وتصوير الكلى بالموجات فوق الصوتية، كلها مرتبطة بشكل إيجابي مع IgGCR (P <0.001)، في حين أن eGFR، والهيموجلوبين، والألبومين في المصل يرتبطان بشكل سلبي مع IgGCR. الاستنتاج: وبالتالي، فإن تركيزات Alb وIgG وß2mG مرتبطة ارتباطًا وثيقًا بدرجة مرض السكري، مما يوفر إرشادات طبية لتشخيص وتقييم مرض السكري

الكلمات الرئيسية: مرض السكري، ألبومين البول، الغلوبولين المناعي ، بيتا ميكروغلوبولين .

Introduction

Chronic hyperglycemia brought on by problems with the metabolism of proteins, fats, and carbohydrates characterises diabetes, a multifactorial, progressive metabolic illness [1]. Long-term harm, malfunction, and failure of many organs, particularly the kidneys, heart, nerves, blood vessels and eyes are linked to persistent hyperglycemia [2].

Because of its increasing incidence and status as a major cause of death and morbidity, diabetes mellitus is a serious worldwide health concern. It is impossible to overestimate the significance of early diabetes diagnosis and treatment because a delayed diagnosis frequently results in serious complications, especially with regard to the kidneys, eyes, and cardiovascular system. The evaluation of biomarkers that have been investigated as indications of kidney impairment in diabetes, including urine albumin (Alb), immunoglobulin G (IgG), and beta-microglobulin (β2mG), is the main focus of this work. Finding these indicators early on can help with better disease management and prompt intervention, which may slow the development of diabetic complications.

The pathophysiology of type 1 diabetes is not entirely known, regardless of the amount of study that has been done on the condition over the years; nonetheless, it is believed to be caused by a number of causes, including genetic defects and/or environmental variables, resulting in either a reduction in insulin production or a loss in insulin release [3-5]. Insulin resistance, poor control of hepatic glucose production, and diminished β cell activity, which ultimately results in β cell failure, are the pathogenesis of type 2 diabetes [6].

A parallel rise in the occurrence of diabetic nephropathy is linked to the rising rates of diabetes mellitus. As in the developed world, diabetic nephropathy might soon become the most significant reason of end-stage renal disease [7, 8]. This has previously happened in other regions of Africa, such as Iraq, where the incidence of diabetes end-stage renal disease (ESRD) climbed significantly from 8.9% in 1996 to 14.5% in 2001 along with a concomitantly higher mortality hazard [9, 10].

For several years, serum creatinine and different estimates of the glomer-

ular filtering rates utilising creatinine-based equations had been used in clinical assessment of renal function in diabetics [11]. Due to the fact that it can only identify more serious conditions of diabetic nephropathy, this has certain accuracy limits. For usage in a clinical environment, other GFR measurement techniques are either too time-consuming or too pricey [12]. The discovery of characteristic Kimmelstiel-Wilson lesions and other structural lesions from biopsies material, such as glomerular basement membrane thickness, can actually signal an advanced disease phase because renal biopsy is not commonly performed [13, 14].

Early research on microalbuminuria ascribed production of >30 mg/day of albumin in urine to enhanced glomerular filtration of albumin. Most subsequently, focus has been placed on the management of persistent microalbuminuria to identify the existence of incipient diabetics [15]. Although investigations on humans and rats have demonstrated that poor tubular reabsorption of albumins at the proximal convoluted tubular contributes to microalbuminuria, the glomerular genesis of the condition has not been disputed [16, 17].

Numerous glomerular or tubular indicators, such as transferrin, type IV collagens, cystatins C, ceruloplasmins, immunoglobulins M (IgM), related lipocalins, and IgG, will manifest before the onset of microalbuminuria [18]. There are four IgG subcategories in type 2 diabetes mellitus (T2DM), however IgG1 was shown to be the most prevalent IgG in blood and urine [19]. It was found to have both anti-inflammatory and pro-inflammatory mediator-releasing properties. It was discovered at the pre - diabetes phase in the urine [20].

The aforementioned shows that studies focusing on diabetic patients' tubular function could be extremely helpful in identifying early diabetic nephropathy, probably before the development of chronic microalbuminuria [21]. The study aimed at examining the applicability of urine albumin (Alb), immunoglobulin G (IgG), and beta-microglobulin (β2mG) levels in the diabetes diagnosis.

Patients and methods:

Materials

The EIA 3609 beta-2-microglobulin ELISA kits (DRG Diagnostics International Inc., USA). An immunoturbidimetric test called MICROALBUMIN™ (Fortress Diagnostics Limited, Antrim Technology Park, UK). Colorimetric glycohemoglobin reagent set (TECO Diagnostics, Anaheim, California 92807, USA). Human enzyme-linked immunosorbent assay kits and RANDOX kits.

Study design and setting:

Diabetes mellitus type 2 patients treated at the medical outpatient clinic at the University of Tikrit , participated in this single-center, observational cross-sectional analysis.

Inclusion criteria:

Simple random sampling was used to enrol 100 diabetic individuals who matched the inclusion criteria but did not meet any of the exclusions listed below in the study. One hundred other people without diabetes who did not meet any of the exclusion criteria were also included in the recruitment. Inclusion criteria are: Patients with type 2 diabetes who are older than 30 years old and do not have any additional medical conditions that can cause proteinuria.

Exclusion criteria:

End-stage kidney disease patients, those who have experienced a urinary tract infections in the month before the interview, those who have renal ultrasonography findings that point to structural abnormalities in the urinary tract, and those who have illnesses that are related to elevated serum beta-2-microglobulin levels (connective tissue disease, multiple myeloma, HIV disease and lymphomas), and a history of using an aminoglycoside within the two weeks prior to the interview day. People whose samples were not completely obtained were not included in the research.

Data and laboratory samples collection:

Evaluated height, weight, hip and waist circumferences, blood pressure, and other standard measurements were all assessed.

The beta-2-microglobulin ELISA kits, with a precision of 0.1 μg/mL, was used to measure the subjects' urinary β2-microglobulin (β2m) levels. The plate control means' average vari-

ation coefficient was 10.4%. The percentage of subjects whose urine β2-microglobulin levels were determined to be raised by an ELISA kit to be >0.3 µg/mL (the absolute limit of normal for urinary β2m). Utilizing MICRO-ALBUMIN™, albuminuria was detected and quantified. Above a period of three months, the participants were to have three urine samples obtained each month. Persistent microalbuminuria was defined as having at least 2 of the 3 samples.

Jaffe's alkaline picrate kinetics technique was used to estimate creatinine. The formulas for the Cockroft-Gault, 4-variable MDRD, and CKD-EPI were used to calculate the glomerular filtrations rates [22-24]. Using the glycohemoglobin reagents colorimetric sets, HbA1c levels were calculated. The Friedewald equation was used to calculate LDL while RANDOX kits were used to measure total cholesterol, triglycerides, and HDL [25].

Humans enzyme-linked immunosorbents test kits were used to quantify urinary IgG (the typical value is <8.8 mg/l).

Ethical considerations

Ethical considerations were provided from the Tikrit Research Ethics Committee. The local ethics board granted its permission, and the research was carried out in conformity with the guide of the Helsinki Declaration. An informed consent statements were collected from the participant to take part in the study.

Data management and statistical analysis:

STATA 10 (StataCorp, College Station, TX, USA) was used for data processing. Subjects' initial sociodemographic and clinical details were examined. For continuously distributed parameters with a normal distribution, the means ± standard deviations were determined, and for parameters with a normal distribution, the median and corresponding interquartile ranges were determined. Evaluating categorical data was done using Pearson's Chi-square test (or Fisher's exacts test), while evaluating quantitative parameters was done using Student's -test (or its nonparametric counterpart). To determine the variables connected to increased urine β2-microglobulin, we employed univariate logistic regression. The multivariate logistic model comprised variables that had Wald sta-

tistic values of less than 0.25 and those that were identified as risk variables for diabetic. The resulting model's polynomial features of the continuous data were used to test the model's linear association. To evaluate the model's fit, a Receiver Operator Characteristic (ROC) curve has been utilized.

Results

A total of 111 controls and 112 diabetics were included in the research. Twenty-three were missed at follow-up (12 (12%) of the diabetics and 11(11%) of the controls). After removing participants who were unable to be reached for follow-up, data from 100 diabetes and 100 controls were ready for analysis. Tables 1 and 2 display information on the study subjects' socioeconomic and demographic, clinical, and investigations features.

Table 1: Assessment of the clinical and sociodemographic details of the diabetics and the controls.

Sociodemographic characteristics			
	Controls (n=100)	**Diabetics (n=100)**	**p-value**
Age in years (mean ± SD)	54.3 ± 13.1	55.9 ± 11.2	0.41*
Six			0.54†
• Female	60 (60)	65 (65)	
• Male	40 (40)	35 (35)	
Educational status, n (%)			0.03†
• No formal education	19 (19)	9 (9)	
• Primary	25 (25)	20 (20)	
• Secondary	4 (4)	12 (12)	
• Tertiary	52 (52)	59 (59)	
Marital status, n (%)			0.78†
• Married	94 (94)	91 (91)	
• Single	6 (6)	9 (9)	
†Chi-square analysis, *Student's t-test.			
Clinical Data			
	Controls (n=100)	**Diabetics (n=100)**	**p-value**
History of hematuria	2 (2%)	8 (8%)	0.02†
History of frothy urine	6 (6%)	47 (47%)	0.0001

Sociodemographic characteristics			
	Controls (n=100)	**Diabetics (n=100)**	**p-value**
Family history of hypertension	27 (27%)	28 (28%)	0.83
Family history of kidney disease	4 (4%)	9 (9%)	0.0001
Family history of diabetes mellitus	12 (12%)	44 (44%)	0.0001
mABP (mmHg) $	98.4 (87.4–111.4)	101.6 (94.4–110.4)	0.26
Diastolic blood pressures (mmHg)**	81.5 ± 14.8	83.4 ± 12.6	0.38
Systolic blood pressures (mmHg)**	140.4 ± 23.2	145.3 ± 23.5	0.21
Waist-to-hip ratios$	0.92 (0.87–0.98)	0.95 (0.88–1.23)	0.28*
Hip circumferences (cm)**	95.1 ± 13.5	98.5 ± 12.5	0.08
Waist circumferences (cm)**	88.1 ± 16.4	94.7 ± 13.7	0.006$^{\Delta}$
BMI (body mass index) (kg/m^2)**	27.5 ± 5.78	26.8 ± 4.4	0.23$^{\Delta}$
mABP: mean arterial blood pressure, **Mean (SD), †Fisher's exact testing. *Wilcoxon rank sum testing. $^{\Delta}$Student's -testing. $Median (IQR).			

Table 2: Evaluation of the two groups' laboratory results.

	Diabetics (n=100)	**Controls (n=100)**	**p-value**
LDL-cholesterol (mg/dL)$	3.2 (1.5–3.8)	1.3 (1.13–1.8)	0.0002
Immunoglobulin G creatinine ratio (IgGCR) (mg/g)	3.67±1.75	88.67±43.62*	0.0001
Hemoglobin (g/dL) $	14.7 (12–14.7)	14.3 (12.9–14.9)	0.25
Random plasma glucose (mmol/L)**	13.1 ± 3.8	6.2 ± 1.7	0.0001
Urinary beta-2-microglobulin (β2m) (μg/mL)$	0.52 (0.2–1.01)	0.2 (0.2–0.39)	0.003
Estimated GFRCKD-EPI (mL/min/1.73 m2) $	83.6 (68.5–98.4)	91.1 (76.8–110.9)	0.003
Total cholesterol (mg/dL)**	5.12 ± 1.5	3.6 ± 1.4	0.0001
Estimated GFRMDRD (mL/min/1.73 m2) $	81.6 (68.2–98.8)	87.6 (77.3–128.9)	0.02
Estimated GFRCG (mL/min)$	67 (55–88)	82 (59–115)	0.03
urinary albumin concentration (UAC) (mg/L)$	24 (11–112)	0.47 (0.4–11.1)	0.0002
Serum uric acid (mmol/L)**	5.7 ± 1.6	5.1 ± 1.6	0.02
Atherogenic ratio (total cholesterol/HDL)$	4.5 (3.3–6.8)	3.4 (2.5–4.7)	0.0004
HDL-cholesterol (mg/dL)$	1.2 (0.95–1.3)	0.97 (0.75–1.4)	0.72
Triglycerides (mg/dL)**	2.24 ± 0.82	1.52 ± 0.72	0.0001
Serum urea (mmol/L) $	4.7 (3.6–5.2)	3.8 (2.7–5.01)	0.002
Glycated hemoglobin (%)$	7.2 (5.2–8.2)	4.7 (3.8–5.3)	0.0001
Serum creatinine (μmol/L)$	92 (75–110)	85 (70–97)	0.04

** Mean ± SD. $ Median (IQR) , + indicates chi-square analysis

In comparison with the controls, the patients (diabetics) had a considerably higher percentage of people with secondary and tertiary education. Both groups' percentages of age and gender were comparable, and both groups' proportions of marital status were as well. The percentage of those having a family history of diabetes mellitus was substantially greater in the diabetics (44% versus 12%, p =0.0001). The percentage of people in both groups who had a positive family histories of hypertension was almost the same (28% for the diabetics versus 27% for the controls, p = 0.83), but the percentage of people in the diabetics who had a family history of kidney disorder was higher (9% in diabetes vs. 4% for the controls).

High-density lipoprotein and haemoglobin values were comparable across the two groups. It was discovered that the other laboratory variables varied significantly. In comparison to the control group, the diabetics had increased levels of serum creatinine, urea, uric acid, glycated haemoglobin, and randomly plasma glucose. In comparison to the controls, the diabetics' total cholesterol, LDL cholesterol, atherogenic ratio and triglycerides were all significantly higher. Eighty-five percent of the individuals had at least one aspect of their lipid profiles that was abnormal. Amongst diabetics with high urinal beta-2-microglobulin, this rate rose to 87.9%. This was made up of high triglycerides (71.7%), high total cholesterol (41.8%), high LDL cholesterol (62.1%), and insufficient HDL cholesterol (49.3%). It was determined that 64 (64%) of the diabetics had poor long-term glycemic control when their glycated haemoglobin level exceeded 6.7%.

Increased Urinary β2m Rate

The controls scored a median urine β2m of 0.1 μg/mL, whereas the diabetics had a median urinary β2m of 0.52 μg/mL (interquartile range: 0.2-1.01 μg/mL) (IQR: 0.2–0.39). The result showed a statistically significant difference (p=0.003). In comparison to the 33.1% (95% CI: 23.8-42.3%) of the controls, 53.1% (95% CI: 43.2-62.9%) of the diabetics showed increased urine β2m, p=0.003.

High urine β2m among the participants didn't vary by gender [51.4% (18) of male diabetes against (34)

52.3% of female diabetics, p = 0.87]. In comparison to 13 (21.7%) female controls, there were 18 (45%) male controls with increased urinary β2m (p = 0.02).

There wasn't any variation between diabetics and hypertension and those with DM alone in the percentage of diabetes group with high urinary β2m. In the "DM only" group, 14 people (46.7%) had increased urinary β2m, in contrast to 35 people (51.5%) in the group of 68 individuals who had diabetes and furthermore hypertensive (p = 0.58). Similarly, among the controls, 19 (39.6%) of the 48 people with hypertension and 15 (28.8%) of the 52 people without hypertension had high urine β2m (p = 0.06).

The interactions between β2m and UAC, estimated GFR, systolic and diastolic blood pressure, the elements of lipids profile and serum uric acids are shown in Table 3 along with their p values.

Table 3: Relation between selected variables in the diabetics and the controls for β2m.

	Total (n=200)		**Diabetics (n=100)**		**Controls (n=100)**	
	Rho	**P**	**Rho**	**P**	**Rho**	**p**
Age	0.03	0.68	0.07	0.52	−0.02	0.79
Glycated haemoglobin	0.21	0.003	0.26	0.23	0.07	0.46
HDL-cholesterol	−0.02	0.69	−0.19	0.25	0.03	0.69
Serum creatinine	**0.54**	**0.0003**	0.47	0.0001	0.38	0.002
LDL-cholesterol	0.29	0.003	0.32	0.28	−0.07	0.75
Serum urea	0.25	0.03	0.14	0.29	0.12	0.39
Triglycerides	0.32	0.003	0.33	0.01	0.008	0.85
Total cholesterol	0.21	0.0002	0.45	0.002	0.25	0.26
Estimated glomerular filtration rate (eGFR) (CG)	−0.18	0.02	−0.33	0.003	−0.03	0.95
Serum uric acid	0.17	0.03	0.28	0.09	0.05	0.82
Urinary albumin concentration	0.39	0.0002	0.36	0.003	0.30	0.003

Variables with p < 0.05 are considered statistically significant

Risk Factors for Elevated β2m in Diabetics

Table 4 demonstrated the incidence of increased β2m in the diabetics using both univariate and multivariate regression models. Microalbuminuria in diabetics increased their risk of high β2m by fivefold at the univariate level. At the univariate level, BMI (7% higher risk), eGFR (3% lower risk), LDL (48% higher risk), triglycerides (94% higher risk), and enhanced atherogenic ratio (22% higher risk) all had significant relationships with increased urine β2m. The area under the Receiver Operator Characteristic (ROC) curve of the prototype was 0.77.

Table 4: Models of single- and multiple-variable logistic regression for predicting high β2m.

	Multivariate*		**Univariate**	
	Odds ratio (95% CI)	***p*-value**	**Odds ratio (95% CI)**	***p*-value**
Age (years)	0.89 (0.91–1.12)	0.78	1.12 (0.95–1.15)	0.86
Male gender	1.28 (0.47–3.87)	0.79	0.95 (0.49–1.97)	0.78
HbA1c	1.18 (0.97–1.49)	0.58	1.29 (0.87–1.53)	0.08
Triglycerides	1.76 (0.87–3.65)	0.07	1.82 (1.27–3.33)	0.02
Total cholesterol/HDL	0.84 (0.73–1.13)	0.53	1.18 (1.02–1.52)	0.03
LDL-C	1.43 (0.82–2.54)	0.49	1.57 (1.09–2.01)	0.03
Estimated glomerular filtration rate (eGFR)	0.86 (0.83–1.01)	0.03**	0.87 (0.86–1.01)	0.03
BMI	**1.34 (1.16–1.56)**	**0.02****	**1.17 (0.99–1.27)**	**0.21**
Waist circumference	0.96 (0.94–1.14)	0.41	1.01 (0.99–1.14)	0.86
Positive microalbuminuria	3.83 (1.43–11.89)	0.02**	5.81 (2.41–14.68)	0.0002
Duration of DM	0.97 (0.96–1.12)	0.16	1.02 (0.88–1.12)	0.93
Positive hypertension status	0.59 (0.19–1.79)	0.47	1.4 (0.67–3.01)	0.65

** Statistically significant when other factors are taken into account.

After adapting for the effects of variations in eGFR, the existence of period of diabetes mellitus, HbA1c levels, waist circumference, body mass index, dyslipidemia, gender differences, hypertension and age, diabetics

with continual microalbuminuria had a well almost 4-fold increased likelihood of developing tubular disorder as indicated by altitudes in urinary β2m excretions.

Comparative analysis of Microalbuminuria and Urinary β2m's Usefulness in Identifying Early Diabetics

Comparable association values between urine β2m and eGFR in diabetics and between albumin creatinine ratio and eGFR in controls are shown in Table 5.

Table 5: The association between β2m and eGFR was compared to that between eGFR and UAC in both diabetics and controls.

	Diabetics (n=100)		**Controls (n = 100)**	
	Rho-value	**p-value**	**Rho-value**	**P-value**
β2m	−0.31	0.02	−0.02	0.84
UAC	−0.29	0.04	−0.19	0.06

Increased urine β2m was found in 27 (38.6%) of the normoalbuminuric diabetics while it was only seen in 28.7% of the normoalbuminuric controls. The statistical significance of this difference was $p<0.001$.

Approximately 8 (20.5%) of the diabetics with normal urine β2m had microalbuminuria. Microalbuminuria was present in only 6 (11%) of the controls with normal urine β2m. Table 6 compares the roles of urine β2m and microalbuminuria in the early identification of diabetes.

Table 6: Correlation of urinary β2m and microalbuminuria in identifying possible diabetes

	Microalbuminuria	
	Positively	**Negatively**
Higher urinary β2m		
• Positively	31 (79.5%)	29 (41.4%)
• Negatively	8 (20.5%)	41 (58.6%)

McNemar $X = 9.67$, $p = 0.002$.

As comparing diabetics with normal urine β2m and microalbuminuria, normoalbuminuric diabetics had a considerably larger percentage of high urinary β2m.

Predictors of IgGCR among diabetes

IgGCR had substantial, statistically significant positive relationships with age, glycated haemoglobin, ACR, blood urea, the length of diabetes mellitus (DM) and serum creatinine, as shown in Table 7. IgGCR had statistically significant negative associations with serum albumin, eGFR, and haemoglobin.

Table 7: simple Pearson's correlation between the variables being investigated in the diabetic group and the immunoglobulin G creatinine ratios

Parameters	Immunoglobulin G creatinine ratio (IgGCR)	
	r	P
Age (years)	0.613	0.0002
Serum creatinine (mg/dl)	0.762	0.0003
glycated hemoglobin (HbA1c)	0.86	0.0001
Serum albumin (g/dl)	−0.873	0.0006
Albumin creatinine ratios (ACR)	0.929	0.0002
Blood urea (mg/dl)	0.734	0.0001
Estimated glomerular filtration rate (eGFR) (ml/min/1.73 m2)	−0.893	0.0008
Haemoglobin (Hb) (g/dl)	−0.652	0.0001
Diabetes mellitus (DM) duration (years)	0.936	0.0004

Discussion

The use of urine albumin (Alb), immunoglobulin G (IgG), and beta-microglobulin (β2mG) concentrations in the diagnosis of diabetes is investigated in this research.

The percentage of diabetics with increased urine β2m in addition to the median value of urine β2m were both considerably greater in diabetics compared to controls. Similarly, increased urineβ 2m in more than half of the diabetics indicated tubulopathy. Other investigations have observed elevated ratios of β2m in diabetics [26, 27], and they have also discovered percentages

of diabetic tubulopathy in type 2 diabetes between 55 and 57% using urine β2m as a marker of tubulopathy [28]. In this diabetes cohort, we found a strong negative association between urine β2m and GFR in addition to a linear correlation between urinary β2m and serum creatinine. These results imply that urine β2m tracks improvements in serum creatinine as renal functions diminishes quite well [29]. Urinary β2m and serum creatinine showed a substantial positive association, and creatinine clearance and GFR as determined by the absorption stage of 99m technetium diethylenetriaminepentacetate renogram (GFR-DTPA) showed a significant negative association as demonstrated by Apakkan Aksun et al. [27]. As eGFR declines and blood creatinine rises, diabetes tubulopathy worsens concurrently, presenting as a rise in urine β2m. As this is a cross-sectional analysis, it is impossible to say if the tubulopathy came first or the glomerulopathy before it, which would explain why serum creatinine increased and GFR decreased. It is also possible that both lesions developed at the same time. Cohort studies are the most effective way to answer both issues.

Upon controlling for all the other components in the model, multivariate regression showed that BMI was a predictor of higher urine β2m values, which resulted in a 24% greater risk of tubular dysfunctions for every one unit rise in BMI among diabetics. Arguments in favour of and against the BMI's contribution to the development of diabetic nephropathy have been made. After a 3-year follow-up duration for type 2 diabetics, a retrospective cohort research study [30] disproved the hypothesis that BMI has a substantial impact on diabetic nephropathy when analysis revealed no correlation between eGFR and BMI. Due to its close ties to albuminuria and nephrosclerosis, several other researchers think that obesity promotes the evolution of nephropathy in the majority of pathophysiology. In this research, waist measurement, a gauge of abdominal obesity, had no discernible effect on the emergence of diabetic tubulopathy. Consequently, it is challenging to draw the conclusion from this research that a higher BMI can promote the development of diabetes.

Microalbuminuria and eGFR were two additional important risk vari-

ables for elevated urinary β2m. There was a 3% decrease in the chance of developing increased urinary β2m for every one unit rise in the eGFR after controlling for the effects of age, gender, time since diagnosis of diabetes mellitus, lipid profile, BMI, waist measurement, hypertension, and glycated haemoglobin levels. This demonstrates that the quantity of urine β2m rises as renal function deteriorates (declining eGFR). This study's univariate analysis levels also revealed this inverse association. It has been suggested that urine β2m might be utilised either alone or as part of a panel of testing in the early detection of diabetes because of the high correlation between GFR and microalbuminuria [27].

After controlling for all other variables in the multivariate modelling, the participants with microalbuminuria had a statistically substantial fourfold greater risk of additionally having elevated urine β2m contrasted to those with normoalbuminuria. Although simultaneous tubular and glomerular dysfunctions in diabetes patients could be the chief reason of the rising urine albumin intensity, an initial tubular dysfunctions that impaired albumin reabsorption at the proximal tubules after it had been filtered through normal glomeruli could also be to blame [31]. This is among the explanations why some believe that in diabetics, tubular dysfunction might manifest before glomerular impairment. The symptoms of diabetic nephropathy include both tubular and glomerular impairment. The main indicator of early diabetic nephropathy has historically been glomerular injury, which is manifested by microalbuminuria. However, there is growing evidence that suggests glomerular alterations may be preceded by tubular dysfunction, which is evidenced by high levels of β2-microglobulin. Increased urine excretion of β2-microglobulin results from tubular dysfunction, which affects the proximal tubules' ability to reabsorb it. Microalbuminuria is a result of increased albumin filtration brought on by glomerular dysfunction. The fact that both dysfunctions coexist indicates that there are several routes involved in the development of diabetic nephropathy. Early management in diabetic kidney disease may be possible if tubular failure is identified early, perhaps even before microalbuminuria appears.[31]

In this research, over than a third of the normoalbuminuric diabetes sufferers had already experienced high urine β2m values. On the contrary, microalbuminuria was seen in only 16.3% of diabetics with normally urine β2m. This statistically significant proportional variation indicates that improvements in urine β2m could happen before microalbuminuria. This discovery seems to corroborate the more recent pathogenetic hypotheses of diabetics, which contend that diabetic tubulopathy develops before diabetic glomerulopathy [31]. According to a research, increased urine β2m could be present in up to 55% of normoalbuminuric diabetes patients [28]. One reason is the significant association between glomerular hyperfiltrations, one of the first signs of glomerular failure, and sodium fractional reabsorptions at the proximal tubules in individuals with normoalbuminuria [32]. Second, the observation that those with higher kidney volume—primarily as a result of tubular hypertrophy and interstitial expansion—have an increased probability of developing microalbuminuria from normoalbuminuria. Thirdly, glomerular hyperfiltrations as an outcome of enhanced proximal tubule reabsorption brought on by glomerulotubular feedbacks. Earlier diabetes' decreased GFR following an initial increase might well be explained by glomerulotubular feedback, but tubulointerstitial damage (particularly atrophic proximal tubules) causes decreased salt reuptake and thus decreased GFR.

The comparative lesser molecular weight of β2m compared to albumin (one-sixth of albumin's molecular weight) should be taken into account in the association between 2m and microalbuminuria. This characteristic promotes free filtering of approximately 2 metres over the intact filtering barrier. Greater amounts of albumin might necessitate glomerular injury as smaller amounts are filtered over the "normal" filtering barrier before being detected in urine. Earlier on in the development of diabetes mellitus, urinary β2m rises when tubulopathy is present, impairing β2m reabsorption. Additionally, for microalbuminuria to develop, proximal tubules need to have been exposed to significant amounts of albumin due to glomerulopathy in order for the tubular procedures for reabsorptions of proteins to be overwhelmed [33]. This

suggests that there is a tubular element to microalbuminuria in addition to glomerular failure.

In addition to a significant rise in IgGCR with age, type 2 diabetes individuals who had macroalbuminuria compared to those who had microalbuminuria and normoalbuminuria also saw a considerable rise in IgGCR with DM duration. This shows that having diabetes for a prolonged period of time has been linked to a higher risk of developing and progressing DM. This finding is corroborated by Assal et al. [34] who discovered a substantial differences in the duration of diabetes between the normoalbuminuric and microalbuminuric groups.

This research observed no significant significant differences in sex or BMI across the four categories of type 2 diabetic individuals, which is consistent with findings made by Aly et al. [35] who identified no significant differences in BMI between type 2 diabetic individuals at varying phases of diabetes. Amin et al. [36], who identified a substantial rise in BMI in macroalbuminuric individuals with T2DM compared to micoalbuminuric and normoalbuminuric patients with T2DM, disagreed with the current findings. Their sufferers were susceptible to hyperglycemia that was out of control. This suggests a role for obesity in the emergence of microvascular problems, and it may be explained by the diverse research design, patient populations, or clinical features. In this study, IgGCR and glycosylated haemoglobin showed a strong positive connection. Additionally, it is considerably higher in type 2 diabetic patients with microalbuminuria especially in comparison to those with normoalbuminuria, which is consistent with Kundu et al.'s study [37] of 50 T2DM patients and 50 normal individuals, which found that diabetics with poor glycemic control seemed to have greater levels of microalbuminuria than diabetics with good glycemic control.

Limitations

Our capacity to record the temporal profile of the investigated indicators in the development of diabetes has been hampered by the cross-sectional design of this investigation. This subject would have been better addressed by a prospective cohort study of originally normoalbuminuric diabetic individuals.

Conclusion

However, one can confidently conclude that increased urinary β2m happens at least as early as microalbuminuria in the course of diabetics and there is a recommendation that urine β2m altitude could happen earlier. It could be challenging to conclude from the outcomes of the current study that increased urinary β2m occurs earlier than microalbuminuria. As a result, it has been suggested that a panel of urine indicators, particularly beta-2-microglobulin, be used to improve the likelihood of identifying early diabetes. Therefore, in patients with normoalbuminuria, urine IgG can be employed as an indicator for the early diagnosis of DM. Alb, IgG, and β2mG concentrations are thereby tightly associated to the degree of diabetes, providing a medical guideline for the diagnosis and assessment of diabetes and having significant clinical implications.

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