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ORIGINAL ARTICLE

Evaluation of Sialic Acid Level as a Marker for Diabetic Nephropathy in Type II Diabetes

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ABSTRACT

Background: One serious consequence of diabetes mellitus is diabetic nephropathy. Increase serum sialic acid, an acute phase reactant signifies the underlying inflammatory process before albuminuria develops. The purpose of this study was to evaluate the predictive usefulness of "serum sialic acid" in type 2 diabetic patients' early nephropathy identification.

Methods: Seventy-four patients with type 2 diabetes were included in this case-control study and split into two groups: Group I consisted of 37 individuals with type 2 diabetes who did not have diabetic nephropathy; their average age was mean±sd =60.8±5. Group 2 comprises 37 individuals with diabetic nephropathy and type 2 diabetes, with an average age of 60±9.1 years. This group was further classified into: group 2(a); diabetic nephropathy patients with microalbuminuria. group 2(b); diabetic nephropathy patients with macroabluminuria. Serum creatinine, serum urea, urine albumin/creatinine ratio, hemoglobin A1c, extremely sensitive CRP, lipid profile, Glomerular Filtration Rate, serum sialic acid, and fasting and postprandial blood sugar were all measured in blood samples.

Results: Compared to the group of diabetics without nephropathy, the group with nephropathy had considerably more sialic acid. There was no significant correlation between serum sialic acid and Hb A1c, HDL–C, AST, ALT, or alkaline phosphatase in the diabetic nephropathic group, but there was a significant negative correlation with estimated GFR and a significant positive correlation with BMI, triglycerides, total cholesterol, LDL-C, uric acid, urea, creatinine, CRP, and UACR.

Conclusions: In diabetic nephropathy, sialic acid can be utilized as a marker of renal dysfunction, reducing morbidity and death.

Keywords: Diabetic nephropathy; Microalbuminuria; Sialic acid

INTRODUCTION

The most prevalent major chronic endocrine illness, diabetes mellitus (DM), is typified by metabolic problems and hyperglycemia. Among minority groups in the former, as well as in both developed and developing nations, it has become a major worldwide health concern [1].

Chronic hyperglycemia associated with diabetes is associated with long-term damage, dysfunction, and failure of many organs, especially the heart, blood vessels, kidneys, nerves, and eyes. More than 30% of diabetics have diabetic nephropathy, one of the primary causes of end-stage renal disease. Furthermore, severe diabetic nephropathy is the leading cause of glomerulosclerosis and end-stage renal disease worldwide [2].

One of the main long-term effects of diabetes mellitus is diabetic kidney damage, often known as diabetic nephropathy (DN). This condition is characterized by abnormal levels of urine albumin excretion, diabetic glomerular lesions, and a decrease in glomerular filtration rate (GFR) in diabetics. The activation of the innate immune response in type 2 diabetes has been linked to elevated levels of acute phase reactants, including sialic acid and C-reactive protein (CRP), which have been suggested as indicators of the disease's risk [3].

Microalbuminuria is a predictor of incipient nephropathy in diabetics and the first sign of diabetic nephropathy. Microalbuminuria, or elevated albumin excretion between 30 and 300 mg/l, is a defining feature of diabetic nephropathy. At this stage of renal illness, urine tests negative on normal dipsticks. To stop the advancement of diabetic nephropathy, it may be helpful to utilize a sensitive test, such as SA, to identify the condition in its early stages [4].

Sialic acid (SA), a broad term for a group of acetylated derivatives of neuraminic acid, is necessary for glycoproteins and glycolipids. SA is essential for both cell membranes and vascular permeability. Serum sialic acid, which is present in acute phase proteins, is one kind of "glycoprotein" that is increased in diabetes. One of the key regulators of membrane permeability, SA keeps the renal glomerular basement membrane negatively charged. As a result, elevated levels in diabetic nephropathy signify substantial harm to kidney vascular cells [5].

In uncontrolled diabetic patients, their serum total sialic acid level indicates excessive glycation, which can lead to diabetic nephropathy [6]. The purpose of this study was to evaluate the predictive usefulness of "serum sialic acid" in kind 2 diabetic patients' early nephropathy identification.

METHODS

Seventy-four patients with type 2 diabetes who visited the internal medicine outpatient clinics and clinical pathology department at Zagazig University Hospitals between October 2023 and March 2024 were the subjects of this case-control research. They were divided into 37 patients with type 2 diabetes who did not have diabetic nephropathy (16 males and 20 females; ages 60.8 ± 5 years) and 37 patients with type 2 diabetes who did have diabetic nephropathy (18 males and 19 females; ages 60 ± 9.1 years). The group with diabetic nephropathy was further divided into patients with microalbuminuria (group 2(a) & group 2(b)) and patients with macroalbuminuria and diabetic nephropathy. The local ethics commission at Zagazig University gave its approval to the study (ZU-IRB # 10900/25-6-2023). From all patients participating in this study an informed consent was obtained. The study follows the Helsinki Declaration, which is the World Medical Association's guideline of ethics for research involving human subjects.

Inclusion criteria included type 2 diabetes in an adult patient. The study group excludes participants with cancer, autoimmune illnesses, liver diseases, active bacterial or viral infections, current urinary tract infections, and other renal problems.

Every research participant underwent a comprehensive history-taking interview, which included the duration of their diabetes, along with anthropometric measurements, such as height, weight, and body mass index, a clinical examination, and an ultrasound investigation. Laboratory tests include serum creatinine, serum urea, urine albumin/creatinine ratio, lipid profiles, glomerular filtration rate (GFR), hemoglobin A1c (HbA1c), liver enzymes (AST-ALT-alkaline phosphatase), highly sensitive CRP (Hs CRP), and serum sialic acid by ELISA.

Each individual had a vein punctured in an entirely aseptic manner to obtain venous blood samples, which were then divided Three milliliters of the blood sample were placed in a sterile, plain vacutainer tube, allowed to clot for twenty minutes at 37°C, and then centrifuged for ten minutes at 2000–3000 r.p.m. to separate the serum and remove the supernatant. The serum was then used for lipid profiles (patients were asked to fast for 12-14 hours), CRP, liver, and kidney function. Until the sialic acid level was determined using an enzyme-linked immunosorbent assay kit, 1 milliliter of each sample's serum was stored in a deep freezer at -20 degrees Celsius. Two milliliters of the blood sample were put into a sterile tube with ethylene diamine tetra acetic acid (EDTA) in order to measure the HbA1C level. A fresh, random midstream urine sample was collected in a sterile container in order to calculate the urine albumin/creatinine ratio.

Using specific reagents from the manufacturer (Roche Diagnostics, Germany), spectrophotometry was used to quantify LFT, KFT, CRP, urine creatinine, and lipid profile for each participant in the study on a Roche Cobas 8000 (c702 module). Using specific reagents provided by the manufacturer (Roche Diagnostics, Germany), urine albumin was quantified turbidimetrically using a Roche Cobas 6000 (c501 module). HbA1c was measured with the Roche Cobas 6000 (c501 module) utilizing particular reagents supplied by the manufacturer (Roche Diagnostics, Germany).

GFR in mL/min per 1.73 m^2 = 175 x $SerumCr^{-1}$.154 x age^{-0}.203 x 1.212 (if the patient is Black) x 0.742 (if female) [7].

ELISA was used to assess sialic acid in serum samples. SunRed Biotechnology Company (China) supplied the kit (Catalog No. 201-12-1621).

Statistical Analysis:

SPSS version 27 was used for data management and statistical analysis (IBM, Armonk, NY, USA). In order to compare ordinal data between two groups, categorical variables were characterized using their absolute frequencies, and the trend test was performed using chi-square. The Shapiro-Wilk test was used to validate assumptions for use in parametric testing. Depending on the type of data, means, standard deviations, medians, and interquartile ranges were used to characterize quantitative variables. The independent sample t-test is used to compare quantitative data between two groups. When comparing the means of more than two sets of quantitative data, an ANOVA is utilized. Spearman's correlation was used for correlation analysis. The symbol for the correlation coefficient is "r." It falls between -1 and +1. A value of -1 indicates perfect negative correlation, a value of +1 indicates perfect positive correlation, and a value of 0 indicates no correlation. The best cutoff of a certain quantitative parameter in the diagnosis of a particular health issue was found using the ROC curve. Multiple regression analysis is used to model the relationship between the independent variable and the dependent or response variables. Every P value had two sides. P values were deemed significant if they were less than 0.05.

RESULTS

The nephropathy of diabetes in comparison to the Diabetic without nephropathy group, the patient's levels for sialic acid, HDL-C, LDL-C, total cholesterol, triglycerides, CRP, uric acid, and Hb A1c are significantly higher. However, compared to diabetic patients without nephropathy, those with diabetic neuropathy had a much higher serum sialic acid level **(Table 1).** Additionally, DN patients had significantly higher levels of triglycerides, total cholesterol, HDL-C, LDL-C, urea, creatinine, eGFR, and urine albumin creatinine ratio than diabetics without nephropathy **(Table 2).** Serum sialic acid was found to have a strong negative link with estimated GFR and a large positive correlation with BMI, triglycerides, total cholesterol, LDL-C, uric acid, urea, creatinine, CRP, and UACR in the diabetic nephropathic group. Sialic acid did not, however, significantly correlate with alkaline phosphatase, HDL-C, AST, ALT, or Hb A1c in the same group. **(Table 3).** Serum sialic acid was found to have a strong negative link with estimated GFR and a large positive correlation with BMI, triglycerides, total cholesterol, LDL-C, uric acid, urea, creatinine, CRP, and UACR in the diabetic nephropathic group. **(Table 4).** A Roc curve analysis shows that sialic acid has a good AUC =0.927, 91.7% sensitivity, and 91.9% specificity, making it a significant predictor of whether a patient has microalbuminuria or normoalbuminuria at a cutoff point >1.23. (**Figure 1)**. At a cutoff point >3.4, sialic acid is a significant predictor of microalbuminuria versus macroalbuminuria patients, with a strong AUC = 0.928, 92.3% sensitivity, and 95.8% specificity (**Figure 2)**.

Table 1: Comparison between two main studied groups in laboratory data.

	Diabetic without nephropathy (n = 37)	**Diabetic nephropathy Patients (n = 37)**	**P**
Hb A_{1c} (%)	5.24 ± 0.3	8.99 ± 2.47	**P< 0.001***
Triglycerides (mg/dl)	94.3± 7.67	154± 58	**P< 0.001***
Total cholesterol (mg/dl)	89± 12.85	201.9 ± 38.27	**P< 0.001***
HDL –C (mg/dl)	53 ± 10	38 ± 6	**P< 0.001***
LDL -C (mg/dl)	70.4± 14	123 ± 25	**P< 0.001***
Uric acid (mg/dl)	4.2 ± 0.5	7.1 ± 1.4	**P< 0.05***
CRP (mg/dl)	5 ± 0.8	44.5± 10.8	**P< 0.001***
Sialic Acid (mmol/l)	**1.4 ± 0.3**	**3.6 ±0.5**	**P< 0.001***

Data are represented as Mean± SD, n= number of subjects, * = significant.

Table (2): Comparison between studied groups in laboratory data

	Diabetic without nephropathy (n = 37)	Diabetic nephropathy Patients		P
		Microalbuminuric n= 24	Macroalbuminuric n= 13	
Urinary albumin creatinine ratio	6.8±1.09	265±25	1444±249	P< 0.001*
Urea (mg/dl)	24.40± 4.23	28.64 ± 5.34	47.92 ± 21.86	P< 0.05*
Creatinine (mg/dl)	0.8± .13	0.7 ± 0.1	1.59 ± 1.13	P < 0.001*
eGFR (ml/min)	95 ± 10	43.9 ±5.16	14.6 ± 7.8	P < 0.001*
Triglycerides (mg/dl)	94.3± 7.67	164.88 ± 70.52 (100-300)	225 ± 48.57 (153-310)	P < 0.001*
Total cholesterol (mg/dl)	89± 12.85	201.9 ± 38.27 (155-285)	254 ± 45.65 (176-320)	P < 0.001*
HDL –C (mg/dl)	38 ± 6	45.32 ± 10 (34-59)	37.36 ± 6.34 (30-50)	P < 0.001*
LDL -C (mg/dl)	70.4± 14	123.37 ± 35.48 (75.4-198)	172.14±43.39 (99-229.8)	P < 0.001*

Data are represented as Mean± SD, n= number of subjects, * = significant.
Anova test was used to compare three studied group in laboratory data.

Table (3): Correlation between sialic acid and other parameters in studied groups.

	Correlation coefficient (r)	P
BMI	**0.28**	P < **0.05***
Hb A_{1c}	**0.21**	P >**0.05**
Triglycerides	**0.3**	P <**0.05***
Total cholesterol	**0.33**	P <**0.05***
HDL –C	**0.13**	P >**0.05**
LDL -C	**0.23**	P <**0.05***
Uric acid	**0.32**	P <**0.05***
Urea	**0.54**	P <**0.001***
Creatinine	**0.69**	P <**0.001***
eGFR	**- 0.86**	P <**0.001***
CRP	**0.47**	P <**0.001***
AST	**0.22**	>**0.05**
ALT	**0.17**	P >**0.05**
Alkaline phosphatase	**0.21**	P >**0.05**
UACR	**0.78**	P <**0.001***

* = significant

Table (4): Multiple regression analysis for study relation of sialic acid and other parameters in studied groups

	Coefficients	P
BMI	-0.04123	P>0.05
S.creatinine (mg/dl)	-0.15527	P>0.05
S.urea (mg/dl)	-0.00722	P>0.05
ALT (U/L)	0.015134	P>0.05

	Coefficients	P
AST(U/L)	0.01787	P>0.05
Alkaline phosphatase(U/L)	-0.00405	P>0.05
CRP (mg/l)	0.008742	P<0.001*
HbA1c (%)	0.017303	P>0.05
S.uric acid(mg/dl)	0.031068	P>0.05
cholesterol(mg/dl)	-0.02314	P>0.05
Triglyceride(mg/dl)	0.002242	P>0.05
HDL(mg/dl)	0.031713	P>0.05
LDL(mg/dl)	0.02572	P>0.05
U.ACR	0.000303	P<0.001*
GFR	-0.03017	P<0.001*

* = significant

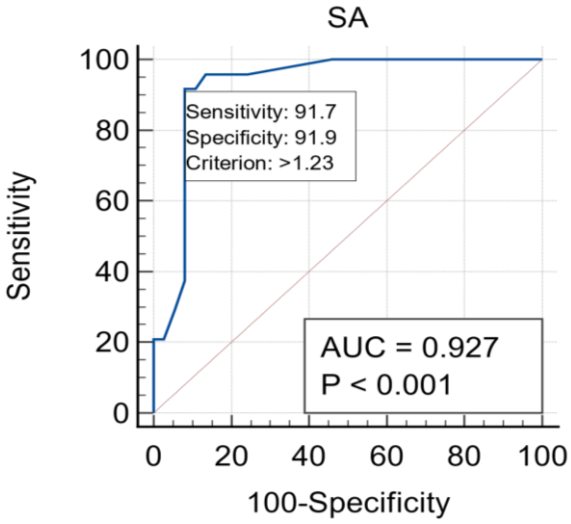

Figure (1): Roc curve in differentiation between diabetic patients without nephropathy and diabetic nephropathy patients

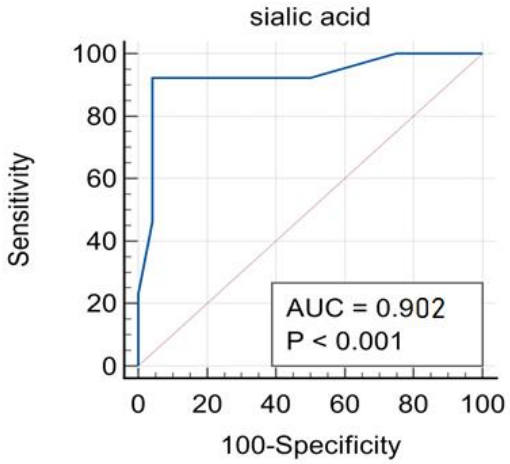

Figure (2): Roc curve for sialic acid in differentiation between microalbuminuric and macroalbuminuria patients**.**

DISCUSSION

One type of "glycoprotein" that is elevated in diabetes is serum sialic acid, which is found in acute phase proteins. Maintaining the negative charge of the renal glomerular basement membrane, one of the key regulators of membrane permeability, is the basic mechanism linked to SA's action. Diabetic nephropathy is the result of

severe damage to the kidney's vascular cells, which is indicated by elevated levels in diabetes [5]. Type 1 and type 2 diabetes were linked to microvascular problems and serum sialic acid levels before microalbuminuria and clinical proteinuria [6].

HbA1c, triglycerides, total cholesterol, HDL-C, LDL-C, uric acid, CRP, urea, creatinine, and urine albumin creatinine ratio were all considerably higher in patients with DN than in the group of diabetics without nephropathy, according to the current study. Furthermore, eGFR was substantially lower in the group with diabetic nephropathy. Individuals with diabetic nephropathy have a much higher serum sialic acid level than diabetic individuals without nephropathy.

Consistent with our findings, Kumar and Rafi [6] found that the group of diabetic patients without nephropathy and the group of patients with nephropathy had a statistically significant increase in sialic acid. Additionally, Subzwari and Qureshi [8] found that patients with diabetic nephropathy had significantly higher serum sialic acid levels than those without nephropathy. Additionally, compared to diabetic people without nephropathy, Roozbeh et al. [9] discovered that patients with diabetic nephropathy consistently had abnormally increased serum levels of SA and neuraminidase activity.

Our study found that the groups with and without diabetic nephropathy had mean serum sialic acid levels of 3.6±0.5 and 1.4±0.3, respectively. These conclusions were supported by Ramana's research [5]. The mean serum sialic acid levels among diabetics with nephropathy and those without were 3.01±0.75 mmol/L and 1.89±0.48 mmol/L, respectively, indicating a highly significant association between the two groups.

Because both type 1 and type 2 diabetes are known to have elevated levels of serum acute phase proteins, the elevated blood sialic acid level in diabetics with nephropathy can be explained. Furthermore, it was shown that elevated serum sialic acid levels in diabetic patients with nephropathy were caused by abnormalities of the red blood cell membrane, which in turn resulted in the release of SA. Inflammatory indicators have been associated with adult diabetes, supporting the notion that inflammation contributes to the genesis of the illness.

Our current research demonstrated a substantial positive correlation between serum sialic acid and BMI, triglycerides, total cholesterol, and LDL-C in the diabetic nephropathic group.

This was in line with the results of El Badawy et al. [10], who found a positive correlation between serum sialic acid levels and cholesterol, TG, and BMI. According to Englyst et al. [11], the amount of body fat is a predictor of the level of sialic acid, which is controlled by it. Additionally, this was in line with the findings of Subzwari and Qureshi[8], who discovered that parameters like cholesterol, LDL, and TG were linked to elevated blood sialic acid concentrations, as well as who discovered a substantial correlation between SA and metabolic syndrome.

Serum sialic acid was found to have a substantial negative association with estimated GFR and a significant positive correlation with uric acid, urea, creatinine, CRP, and UACR.

Kumar and Rafi [6] and El Badawy et al. [10] came to similar conclusions. They found a positive significant correlation between sialic acid level and DN measurements (creatinine, urea, and 24-hour urine albumin) and that sialic acid level was a significant predictor of serum creatinine and urinary albumin (nephropathic parameters). Furthermore, consistent with the findings of Ghosh et al. [12], who found a very strong positive correlation between serum sialic acid and urinary microalbumin, suggesting that serum sialic acid contributes to renal damage as microalbumin excretion increases. Additionally, they discovered that individuals with diabetes who also had nephropathy had much greater serum sialic acid levels than those without nephropathy. These findings imply a relationship between elevated serum sialic acid and the severity of diabetic renal issues.

Sialic acid is a significant predictor to distinguish between individuals with normoalbuminuria and those with microalbuminuria at a cutoff point >1.23, according to the current study. It has a good AUC of 0.927, 91.7% sensitivity, and 91.9% specificity. At a cutoff point >3.4, sialic acid is a significant predictor to distinguish individuals with microalbuminuria from those with macroalbuminuria, with a strong AUC = 0.902, 92.3% sensitivity, and 95.8% specificity.

In Kumar [13] and his colleagues investigated how sialic acid can be used to diagnose diabetic nephropathy. They included both healthy controls and diabetic nephropathy patients in their study. With an area AUC of 0.998 at a cutoff value of 2.65 mmol/L, they showed that the examination of the Receiver Operator Characteristics curve showed 100% specificity and 98% sensitivity in the SA measurements. All research parameters showed 100% specificity on the ROC curve. Since the research parameters (fasting and post-prandial blood sugar, urea, creatinine, serum sialic acid, and urinary microalbumin) were evaluated in cases with diabetic nephropathy that had

previously been established, there was very little overlap between the two distributions of true positives and false positives. This led to a high specificity when comparing patients with diabetic nephropathy to healthy controls.

We came to the conclusion that sialic acid is linked to type 2 diabetes and associated risk factors, indicating higher sensitivity for early identification to microvascular problems. Thus, sialic acid may facilitate both therapeutic activity and early complication diagnosis.

When describing the most recent guidelines for the management of type 2 diabetes, in order to get an early diagnosis of nephropathy in this disease, we recommend considering the assessment of sialic acid.

Conclusion

The current study demonstrated that in diabetic nephropathy, sialic acid can be utilized as a marker of renal dysfunction, reducing morbidity and death.

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Conflict of interest: None.

References

1-Tomic D, Shaw JE, Magliano DJ. The burden and risks of emerging complications of diabetes mellitus. Nat Rev Endocrinol. 2022 Sep;18(9):525-39.

2- Cheng HT, Xu X, Lim PS, Hung KY. Worldwide epidemiology of diabetes-related end-stage renal disease, 2000–2015. Diabetes Care. 2021 Jan 1;44(1):89-97.

3- Sagoo MK, Gnudi L. Diabetic nephropathy: an overview. Diabetic nephropathy: methods and protocols. 2020:3-7.

4- Hu Q, Chen Y, Deng X, Li Y, Ma X, Zeng J, Zhao Y. Diabetic nephropathy: focusing on pathological signals, clinical treatment, and dietary regulation. Biomedicine & Pharmacotherapy. 2023 Mar 1;159:114252.

5- Ramana CV. A study of serum sialic acid levels in diabetic nephropathy. Int J Med Public Health. 2024 Apr;14(2):455-8.

6- Kumar BS, Rafi M. Serum total sialic acid and (HS–CRP) Highly Sensitive C-Reactive Protein) as markers of diabetic nephropathy. Eur J MolClin Med. 2022; 9(3), 11557-73.

7- Levey AS, Stevens LA, Schmid CH, Zhang YL, Castro AF 3rd, Feldman HI et al. CKD-EPI (Chronic Kidney Disease Epidemiology Collaboration). A new equation to estimate glomerular filtration rate. Ann Intern Med. 2009 May 5;150(9):604-12.

8- Subzwari JJ, Qureshi MA. Relationship between sialic acid and microvascular complications in type 2 diabetes mellitus. Biomedica. 2012; 28(2), 130-3.

9- Roozbeh J, Merat A, Bodagkhan F, Afshariani R, Yarmohammadi H. Significance of serum and urine neuraminidase activity and serum and urine level of sialic acid in diabetic nephropathy. Int Urol Nephrol. 2011; 43, 1143-8.

10- El Badawy AM, Mansour AE, Elsayed MS, Abdelmoniem RO. Relationship between serum sialic acid concentration and diabetic nephropathy in Egyptian patients with type 2 diabetes mellitus. J Diabetol. 2021 Jan 1;12(1):70-5.

11- Englyst NA, Crook MA, Lumb P, Stears AJ, Masding MG, Wootton SA et al. Percentage of body fat and plasma glucose predict plasma sialic acid concentration in type 2 diabetes mellitus. Metabolism. 2006 Sep 1;55(9):1165-70.

12- Ghosh J, Datta S, Pal M. Role of sialic acid in prediction of diabetic nephropathy. Al Ameen J Med Sci. 2016; 9(1), 58-64.

13- Kumar BS, Khan SA, Baba KS, Satyanarayana PV, Mohan IK. Sialic acid: an early sensitive marker improves detection of nephropath

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