The Egyptian Journal of Hospital Medicine (July 2024) Vol. 96, Page 2494-2499

Vaspin Levels and Diabetic Nephropathy in Elderly Patients with Type 2 Diabetes Mellitus

Mayada M. Mousa[1], Samia Hussein[2*], Reda M. Khodry[1], Atef G. Hussain[2], Ahmad I. Elagrody[1], Azza H. Abd Elfatah[1]

Departments of [1]Internal Medicine and [2]Medical Biochemistry and Molecular Biology, Faculty of Medicine, Zagazig University, Egypt

***Corresponding author:** Samia Hussein, **Mobile:** (+20) 01062725981, **E-mail:** samiahussein82@hotmail.com

ABSTRACT

Background: Diabetic nephropathy (DN) is a highly prevalent major chronic complications of DM. DN is diagnosed depending on clinical background with existence of albuminuria and/or declined eGFR and hypertension without any other primary renal disease. Vaspin is an adipokine, which acts as anti-atherogenic.

Objective: This study aimed to correlate vaspin levels to the biochemical findings in DM and DN, and to investigate the significance of vaspin in predicting the development of DN among type 2 diabetic patients.

Subjects and methods: Our research involved 90 subjects (above 65 years old) who were divided into 3 groups. Group I with 30 elderly healthy individuals, group II with 30 elderly uncomplicated type 2 diabetic patients and group III included 30 elderly type 2 diabetic patients with DN.

Results: Statistically significant decreases in vaspin levels among group III compared to groups I and II were detected. Also, group II had a statistically significant rise compared to group I. Statistically significant positive correlations were detected between vaspin levels and each of fasting insulin (r=0.909, P<0.001), HOMA-IR (r=0.902, P<0.001), C-peptide (r=0.721, P<0.001), and GFR (r=0.892, P<0.001). However, statistically significant negative correlations were detected between vaspin levels and each of albumin/creatinine ratio (ACR) (r=-0.600, P=0.002), creatinine (r=-0.551, P=0.002), and blood urea nitrogen (BUN) (r=-0.451, P=0.031) among group III. Receiver operating curve (ROC) showed that vaspin had a significant validity in the prediction of DN among DM cases with sensitivity of 87%, specificity of 78.3% and accuracy of 82.6%.

Conclusion: Vaspin could be a potential marker in the prediction of DN development among DM cases.

Keyboards: Vaspin levels, DN, Type 2 diabetes mellitus.

INTRODUCTION

Diabetic nephropathy (DN) or diabetic kidney disease (DKD) is a highly prevalent major chronic complications of DM. It is a long-term structural, functional, and clinical abnormalities of the kidneys that are produced by DM [(1)]. This is applicable for the two types of DM. One of the strongest predictors of DN is microalbuminuria [(2)]. DKD develops in 20-40% of diabetic patients. It usually occurs after about 10 years in type 1 DM but can appear at diagnosis of type 2 DM. In both types of DM, CKD development significantly raises the risk of cardiovascular events [(3)].

DKD is a heterogeneous condition. Retinopathy with a urinary ACR of 300 mg/g refer to DN. Hyperglycemia and inflammation change glomerular endothelium with subsequent apoptosis and aberrant angiogenesis leading eventually to loss of glomerular permeability and selectivity [(4)].

DKD is diagnosed based on a clinical background. The typical presentation of DKD includes a prolonged DM, retinopathy, albuminuria without gross hematuria, a progressive decline of eGFR, and hypertension without any other primary renal disease. Neither recent DM nor retinopathy absence can rule out DKD in T 2 DM. Renal biopsy may be useful when there is diagnostic uncertainty [(3)].

DN pathogenesis can be explained by the production of free radicals, hyperglycemia, and stimulation of signaling pathways including protein kinase C. DN involves multiple renal structural and functional alterations including nodular glomerulosclerosis formation. Podocyte loss is also observed, which is the best predictor of albuminuria [(4)]. DN also involves the stimulation of angiotensin II (ATII), which acts as a vasoconstrictor of the glomerular efferent arteriole with raised glomerular capillary pressures. Also, activation of ATII type I receptors causes mesangial matrix growth, podocyte injury with subsequent fibrosis and eventually nephron loss [(5)]. In early DM, raised glomerular filtration rate (GFR) and intrarenal hypertension are observed [(5)].

Vaspin is a serine protease inhibitor [(6)]. Visceral adipose tissues are a rich source in comparison with the subcutaneous ones [(7)]. It is an adipokine, which acts as anti-atherogenic [(8)].

This study was conducted to correlate vaspin levels to the biochemical findings in DM and DN, and to investigate the significance of vaspin in predicting the development of DN among diabetic patients.

SUBJECTS AND METHODS

This case-control study included 90 subjects from both sexes (above 65 years old) who were divided into 3 groups: Group I with 30 elderly healthy individuals, group II with 30 elderly uncomplicated type 2 diabetic patients and group III that included 30 elderly type 2 diabetic patients with DN. The American Diabetes Association (ADA) criteria were used to select the patients with type 2 DM [(3)].

Received: 30/01/2024
Accepted: 01/04/2024

Exclusion criteria: Patients with type 1 DM, severe infections, acute complications of DM, inflammatory or malignant diseases, genetic or autoimmune disease, liver diseases, kidney diseases other than DN, heart diseases, patients who were using hormonal preparations or immune inhibitors.

Full history taking and complete clinical examination were performed for all participants. Fundus examination was performed and GFR was estimated. eGFR was calculated for all participants by the MDRD equation of the National Kidney Foundation mobile application.

Five mL of peripheral blood were drawn from each fasting participant. The collected venous blood was separated into 2 portions. The 1st was obtained in EDTA-containing tubes to obtain whole blood and the other was collected in plain tubes to obtain serum.

Hemoglobin, FBG, 2h-PPBG and HbA1c were measured. Renal function tests were performed including serum creatinine and serum urea. HOMA-IR, GFR and ACR were calculated. Estimations of serum vaspin, c-peptide, and fasting insulin were performed by ELISA.

Ethical approval: Zagazig Faculty of Medicine and Mansoura Medical Ethics Committees approved this study. After being informed of all the details, each participant provided written consent. Throughout the course of the investigation, the Helsinki Declaration was adhered to.

Statistical analysis

SPSS program version 20.0 was used. Mean ± SD and X^2-test and independent T-test were used to calculate the difference between qualitative and quantitative data respectively. Pearson correlation coefficient was used to assess the correlation between quantitative measures. Significant results were considered at P value ≤ 0.05.

RESULTS

Our findings found no significant differences between the three groups in age or gender (P=0.96 and 0.76 respectively). On the other side, there was a statistically significant rise in HbA1c, FBG, fasting insulin, HOMA-IR, and C peptide among groups II and III compared to the control group and also among group III compared to group II (P < 0.05 for each). However, there was a significant rise in 2 HPPG among group II compared to group I only with no significant difference between groups II and III. Moreover, a statistically significant increase in creatinine, BUN and ACR and a statistically significant decrease in GFR among group III compared to groups I and II were detected. However, no significant differences in renal indicators were detected between group II and control group. Concerning vaspin, there was a statistically significant decrease in its levels among group III compared to groups I and II. Also, group I had a statistically significant decrease compared to group II (Table 1).

Table (1): Demographic and biochemical data of the studied groups

	Group I (Control) (*n*=30)	**Group II (DM) (*n*=30)**	**Group III (DN) (*n*=30)**	**P**
Age (years)	75.87±6.55	75.09±6.72	74.83±6.4	0.96
Sex	10 (43.5%) 13 (56.5%)	12 (52.2%) 11 (47.8%)	10 (43.5%) 13 (56.5%)	0.76
$HbA1_C$ (%)	4.48±0.62	7.23±0.37 [a]	7.91±0.63 [a,b]	<0.001**
FBG (mg/dL)	83.22±5.62	137.04±16.25[a]	158.96±19.2 [a,b]	<0.001**
2HPPG (mg/dL)	113.3±8.03	241.09±31.92[a]	246.91±26.53[a]	<0.001**
Fasting insulin (uIU/mL)	3.55±0.74	6.52±1.59 [a]	8.74±2.11 [a,b]	<0.001**
HOMA-IR	0.74±0.14	2.28±0.55 [a]	3.52±0.86 [a,b]	<0.001**
C-peptide (ng/mL)	0.63±0.17	1.05±0.25 [a]	1.89±0.46 [a,b]	<0.001**
Blood urea nitrogen (mg/dl)	13.83±3.44	13.65±3.40	58.39±11.09 [a,b]	<0.001**
Serum creatinine (mg/dL)	0.76±0.10	0.75±0.10	3.5±0.64 [a,b]	<0.001**
GFR (ml/min/1.73 m^2)	88.79±7.27	89.66±7.47	47.88±6.75 [a,b]	<0.001**
Albumin creatinine ratio (mg/g)	19.48±3.76	20.35±4.41	683.43±155.22 [a,b]	<0.001**
Vaspin (ng/mL)	1.54 ± 0.19	3.57 ± 0.25[a]	0.70 ± 0.15 [a,b]	<0.001**

**: highly significant difference (P<0.001); [a]: significant versus Group I; [b]: significant versus Group II

Concerning correlation studies, group II had a significant positive correlation between vaspin level and each of fasting insulin (r=0.785, P <0.001), HOMA IR (r=0.798, P<0.001), C-Peptide (r=0.782, P<0.001), and a statistically significant negative correlation between vaspin level and duration of DM (r=-0.510, P=0.013). Moreover, there was a statistically significant positive correlation between vaspin level and each of fasting insulin (r=0.909, P<0.001), HOMA-IR (r=0.902, P<0.001), C-peptide (r=0.721, P<0.001) and GFR (r=0.892, P<0.001) and a significant negative correlation between vaspin level and disease duration (r=-0.617, P=0.002), ACR (r=-0.600, P=0.002), creatinine (r=-0.551, P=0.002) and BUN (r=-0.451, P=0.031) among group III (Table 2 and figures 1 & 2).

Table (2): Correlations between vaspin levels and clinical data among Groups II (DM) and Group III (DN)

		Group II	Group III
Age (years)	**r**	0.055	0.187
	P	0.804	0.393
Duration (years)	**r**	-0.510	-0.617
	P	0.013*	0.002*
Creatinine (mg/dL)	**r**	-0.093	-0.551
	P	0.672	0.002*
BUN (mg/dL)	**r**	0.196	-0.451
	P	0.370	0.031*
GFR (mL/min/1.73 m^2)	**r**	0.129	0.892
	P	0.558	<0.001**
ACR (mg/g)	**r**	-0.205	-0.600
	P	0.349	0.002*
HbA1c (%)	**r**	0.248	0.015
	P	0.254	0.945
FBG (mg/dL)	**r**	0.059	0.262
	P	0.788	0.228
2HPPG (mg/dL)	**r**	0.023	0.057
	P	0.918	0.794
F. Insulin (uIU/mL)	**r**	0.785	0.909
	P	<0.001**	<0.001**
HOMA- IR	**r**	0.798	0.902
	P	<0.001**	<0.001**
c-peptide (ng/mL)	**r**	0.782	0.721
	P	<0.001**	<0.001**

*: a significant difference (P<0.05); **: a highly significant difference (P<0.001), r: Pearson's correlation coefficient;

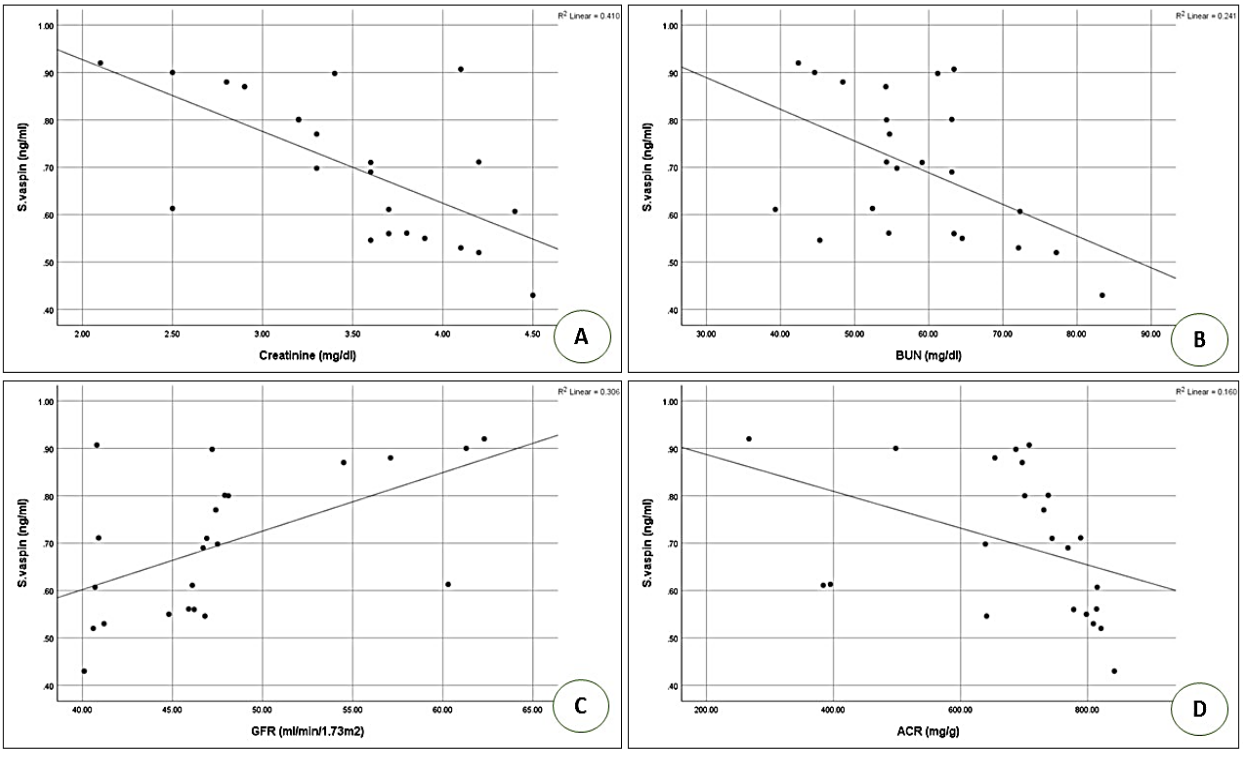

Figure (1): The correlations between vaspin and renal functions in group III

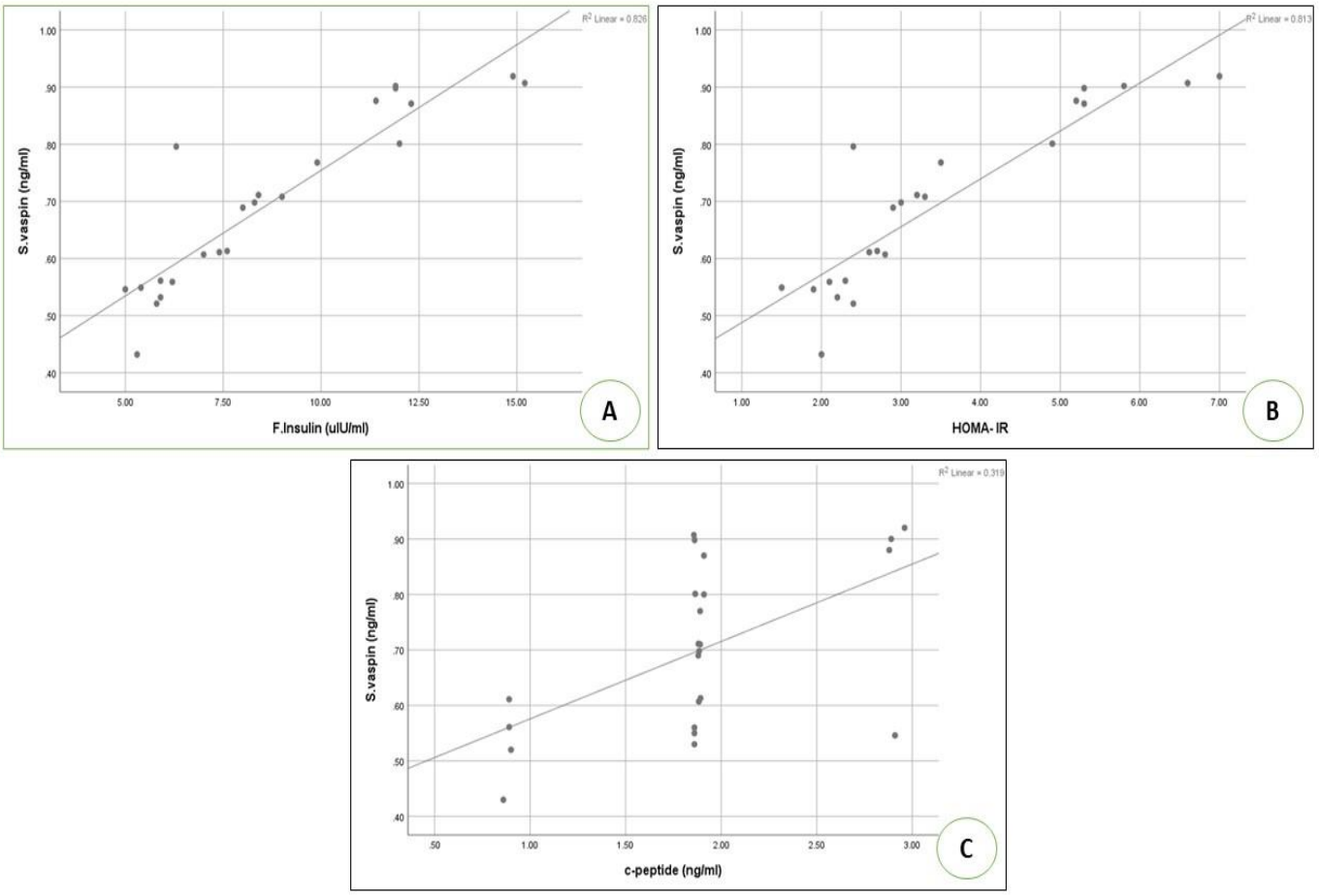

Figure (2): The correlations between vaspin and biochemical parameters in group III A**:** The correlation between vaspin and fasting insulin, **B:** The correlation between vaspin and HOMA IR & **C:** The correlation between vaspin and C peptide.

Receiver operating characteristic (ROC) curve demonstrated that vaspin had a significant validity in the prediction of DN among DM cases with sensitivity of 87%, specificity of 78.3% and accuracy of 82.6% at a cut off level >1.62 (Table 3 and figure 3).

Table (3): Validity of vaspin in predicting DN among DM cases

Cut off	AUC (95%CI)	Sensitivity	Specificity	PPV	NPV	Accuracy	P
>1.62	0.92 (0.84-0.99)	87%	78.3%	80%	85.7%	82.6%	<0.001**

AUC: Area under curve; CI: Confidence interval; PPV: Positive predicted value; NPV: Negative predicted value; **: a highly significant difference (P<0.001)

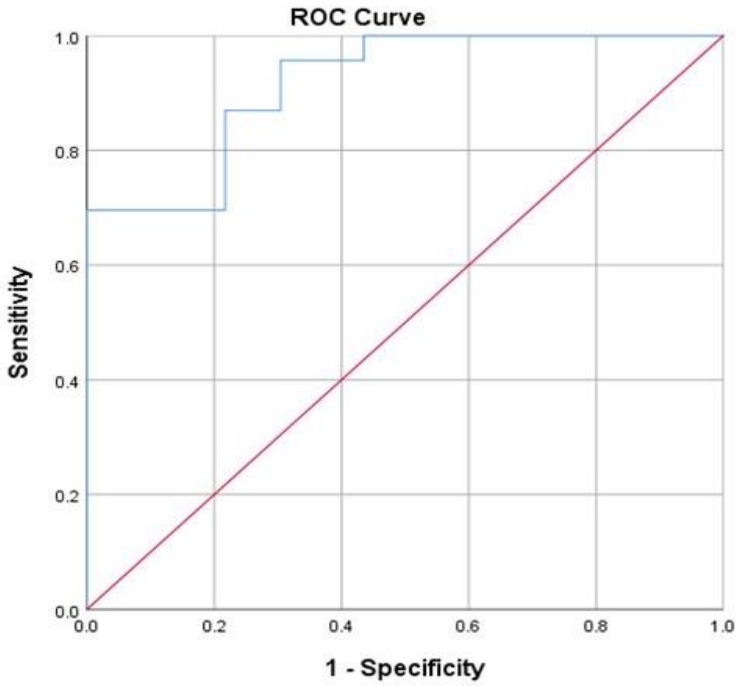

Figure 3: Roc curve for validity of vaspin in prediction of DN among the studied cases groups.

DISCUSSION

Vaspin has a major role in endothelial dysfunction and inflammation, which are the main contributors in the pathophysiology of DM and its consequences [9].

Our study showed a significant rise in HbA1c and FBG, fasting insulin, HOMA-IR and C-peptide among all diabetic cases compared to the control. These findings are similar to that found by **Kumari *et al.*** [10], **Rashad *et al.*** [11] and **Baig *et al.*** [12]**.** Also, there was a statistically significant increase in HbA1c, FBG, fasting insulin, HOMA-IR and C-peptide among diabetic cases with DN (Group III) compared to diabetic cases without complications (Group II). These results are in the same line to that found by **Hiammohammedsalih and Idan** [13]**.**

In our results, there was a significant rise in creatinine, BUN and ACR and a significant decrease in GFR among diabetic patients with DN (Group III) compared to uncomplicated diabetic patients (Group II) and controls (Group I). These findings are in the same line to that detected by **Mihanfar *et al.*** [14]**.**

In this research, vaspin levels were significantly higher in uncomplicated diabetic patients in comparison with controls. These findings agree with **Kumari *et al.*** [10] and **Rashad *et al.*** [11]**.** The raised vaspin levels could be a possible antagonizing mechanism to the increased proteases in obesity and insulin resistance. This impact was explained by inhibition of kallikrein 7 by vaspin. There is a possible relationship of vaspin with the persistent inflammation in DM, which explains its significance in the development of complications of type 2 DM [15]**.**

Our results disagree with **Baig *et al.*** [12] and **Hosseini *et al.*** [16] who observed that vaspin levels decreased in the DM subjects in comparison with the healthy subjects. Also, **Taşdemir and Şermet** [17] detected that plasma vaspin levels in diabetic rats were less than those of control. **Bozaci *et al.*** [18] detected vaspin levels in diabetes patients and controls were not different.

Our findings revealed that diabetic individuals with DN had considerably lower vaspin levels compared to healthy persons. Our results disagree with **Tony *et al.*** [19] who found that serum vaspin levels were significantly higher in T 2 DM patients with reduced renal function in comparison with healthy subjects. Also, **Hiammohammedsalih and Idan** [13] showed that patients with DN had a significantly raised levels of vaspin in comparison with controls. However, lower vaspin levels in patients on hemodialysis can be explained by the low molecular mass of vaspin compared to other adipokines, which are higher in patients on hemodialysis. So, it may not be completely removed by hemodialysis. Declined serum vaspin levels in hemodialysis patients suppose the removal from the circulation possibly by cell surface receptors in multiple tissues [20, 21]**.**

Consistent with our findings, **Kadoglu *et al.*** [22] showed that vaspin levels declined with diabetic microvascular complications. Also, **Mihanfar *et al.*** [14] found that vaspin levels significantly decreased in DN in comparison with diabetic patients with normal renal functions. They considered that reduced serum vaspin levels could be a possible indicator to the development of DN in type 2 diabetic patients. **Dimova and Tankova** [23] showed that serum vaspin levels in

complicated diabetic patients including DN were less than in uncomplicated diabetics.

CONCLUSION

Vaspin could be a potential marker in the prediction of DN among DM cases.

Conflict of interests: None.
Funding: Not applicable.

REFERENCES

1. **Woodhams L, Sim T, Chalmers L *et al.* (2021):** Diabetic kidney disease in type 2 diabetes: a review of pathogenic mechanisms, patient-related factors and therapeutic options. Peer J., 9: e11070. doi:10.7717/peerj.11070
2. **Sagoo M, Gnudi L (2020):** Diabetic Nephropathy: An Overview. Methods Mol Biol., 2067: 3-7. doi:10.1007/978-1-4939-9841-8_1
3. **American Diabetes Association Professional Practice Committee (2022):** Pharmacologic Approaches to Glycemic Treatment: Standards of Medical Care in Diabetes-2022. Diabetes Care, 45 (1): 125-143.
4. **Vasanth Rao V, Tan S, Candasamy M *et al.* (2019):** Diabetic nephropathy: An update on pathogenesis and drug development. Diabetes Metab Syndr., 13 (1): 754-762.
5. **Wenstedt E, Vogt L (2019):** Microvascular Damage and Hemodynamic Alterations in Diabetic Nephropathy. In: Roelofs J, Vogt L (eds), Diabetic Nephropathy. Springer, Cham., Pp: 255-276. https://doi.org/10.1007/978-3-319-93521-8_16
6. **Mansour S, Tawfiq M, Khalefa A *et al.* (2019):** Effect of Diet Regimen on Serum Vaspin Level in Obese Diabetic Female Patients. Zagazig University Medical Journal, 25 (5): 699-707.
7. **Kurowska P, Mlyczyńska E, Dawid M *et al.* (2021):** Review: Vaspin (SERPINA12) Expression and Function in Endocrine Cells. Cells, 10 (7): 1710. doi:10.3390/cells10071710
8. **Ibrahim A, Mustafa A, El-halim W *et al.* (2020):** Assessment of Serum Level of Vaspin in Patients with Acne Vulgaris and its Relation with Insulin Resistance. Benha Journal of Applied Sciences, 5 (7): 43-48.
9. **Su Y, Ming Q, Li W *et al.* (2016):** Serum vaspin: a reliable independent predictor for major adverse cardiac events. Int J Clin Exp Med., 9: 18133–18141.
10. **Kumari P, Bansal S, Sharma D *et al.* (2020):** Correlation of Serum Vaspin with Cardiovascular Risk Factors in T2DM. International Journal of Current Research and Review, 12 (5): 13-18.
11. **Rashad N, Ahmed H, Ashour W *et al.* (2020):** Association of vaspin gene expression and its serum level on the risk of ischemic stroke in type 2 diabetic Egyptian patients: Prospective case-control study. Biotechnology and Applied Biochemistry, 67 (6): 912-919.
12. **Baig M, Gazzaz Z, Bakarman M *et al.* (2021):** Correlation of Serum Vaspin, Omentin-1, and adiponectin with metabolic phenotypes in Type-2 diabetes mellitus patients. Pak J Med Sci., 37 (7): 1762-1767.
13. **Hiammohammedsalih M, Idan S (2021):** Eleveted Vaspin Level S in Diabetic Nephropathy. Annals of the Romanian Society for Cell Biology, 25 (6): 8362-8374.
14. **Mihanfar A, Rahmati-Yamchi M, Mota A *et al.* (2018):** Serum levels of vaspin and its correlation with nitric oxide in type 2 diabetic patients with nephropathy. Current Diabetes Reviews, 14 (2): 162-167.
15. **Heiker J, Klöting N, Kovacs P *et al.* (2013):** Vaspin inhibits kallikrein 7 by serpin mechanism. Cell Mol Life Sci., 70 (14): 2569-2583.
16. **Hosseini M, Nezhadali M, Hedayati M (2021)** Association of vaspin rs2236242 gene polymorphism with serum vaspin level, insulin resistance and diabetes in an Iranian diabetic/pre-diabetic population. Journal of Medical Biochemistry, 40 (1): 33-40.
17. **Taşdemir E and Şermet A (2019):** The Relationship of Adipokines with Glucose Metabolism and Diabetes. Haydarpasa Numune Med J., 59 (2): 95–103.
18. **Bozacı İ, Özkan O, Özkan A *et al.* (2020):** The Relationship Between Serum Vaspin Levels and the Degree of Coronary Involvement in Patients with Stable Angina Pectoris. Medical Bulletin of Haseki/Haseki Tip Bulteni., 58 (2): 176-82.
19. **Tony E, Saleem T, Zakhary M *et al.* (2016):** Levels of serum vaspin and tumor necrosis factor-α in patients with type 2 diabetes mellitus in relation to kidney function and glycemic control. Journal of The Egyptian Society of Nephrology and Transplantation, 16 (4): 131–143.
20. **Hida K, Wada J, Eguchi J *et al.* (2005):** Visceral adipose tissue-derived serine protease inhibitor: a unique insulin-sensitizing adipocytokine in obesity. Proc Natl Acad Sci USA., 102: 10610-10615.
21. **Inoue J, Wada J, Teshigawara S *et al.* (2012):** The serum vaspin levels are reduced in Japanese chronic hemodialysis patients. BMC Nephrology, 13: 163-163.
22. **Kadoglou N, Vrabas I, Kapelouzou A *et al.* (2011):** Impact of atorvastatin on serum vaspin levels in hypercholesterolemic patients with moderate cardiovascular risk. Regulatory Peptides, 170 (1-3): 57-61.
23. **Dimova R, Tankova T (2015):** The role of vaspin in the development of metabolic and glucose tolerance disorders and atherosclerosis. BioMed Research International, 6: 823481. doi:10.1155/2015/823481.