The association between neutrophil to lymphocyte ratio and demographic and clinical data in diabetic peripheral neuropathy patients in Jeddah.

Abstract:

Introduction:

Diabetes mellitus (DM) is a major chronic systemic health concern thatcould develop macro- and microvascular complications. One of the commonest microvascular complications is diabetic peripheral neuropathy (DPN) which is a type of nerve damage caused by uncontrolled hyperglycemia of diabetic patients. DPN is one of the most common causes of reduction in life quality and disability in DM patients, as well as; it overload the national health care system. Finding a cost-effective, easy prognostic marker to early detect DPN could be an important step in prevention DM complications. Our research aimed to assess the association between neutrophil to lymphocyte ratio and demographic and clinical data in diabetic neuropathy patients with average age of 60 years.

Methodology

A retrospective studywas conducted on 133 patients by reviewing the data records over the period of 10 years from January2012 to July 2022. The study was done at king Abdul-Aziz University Hospital, Jeddah, in the western region of KSA amongst diabetic peripheral neuropathy patients who are using either oral hypoglycemic agents or insulin with regular follow ups.

Results:

The study included a total of 133 patients with most of them being male i.e., 81 (61%). The mean age of the patients was 60(SD13.5) years. Approximately two-third of the patients 86(64.7%) had diabetes for more than 10 years. There was no significant association identified for gender, age group, nationality, and medication (p >0.05) with HbA1c or NLR. Moreover, there was no significant association between NLR and HbA1c categories.

Conclusion:

In conclusion our study showed non-significant relationship between NLR and DPN among elderly diabetic patient at King Abdu al-aziz university hospital (KAUH).

Kye words:

Type 2 diabetes, Antidiabetic medications, neutrophil to lymphocyte ratio (NLR), peripheral neuropathy.

Introduction:

Diabetes mellitus (DM) is a major chronic systemic health concern in 2015[1], and by 2040 one in ten adults would develop diabetes [2]. In Saudi Arabia, the prevalence of diabetic patients significantly increased[3], and the prevalence of DM in Jeddah was 18.3%[4,5].

One of the commonest microvascular complications of DM is diabetic peripheral neuropathy (DPN)[6,7]. Diabetic neuropathy is a type of nerve damage caused by uncontrolled hyperglycemia of diabetic patients[8]. Many studies have been showed that DPN is not developed only because of Microvascular complications of DM[9]; but inflammation plays a crucial role in progressing DPN [10].

Early microvascular abnormalities [11] and inflammatory processes [12] have been linked to the onset and progression of DPN, according to research. Neutrophil-to-lymphocyte ratio (NLR), a marker of inflammation that has recently attracted a lot of attention, is closely associated with cancer [13] cardiovascular disease [14] and diabetic microvascular complex disorders [15]. Additionally, it is a predictive factor for individuals with a variety of operable malignancies and an independent risk factor for cerebral haemorrhage in T2DM patients [16,17].NLR has been used to predict the prognosis of other diseases such as end-stage renal failure[18], Atherosclerosis[19], and cancer[20]. NLR is used for screening diseases and as a drug monitoring tool. It is preferable over other inflammatory markers because of its wide availability, low cost, reliability, easy lab detection [21]. The relationship between DPN and NLR were observed in several studies, but none focused on elderly patients. The purpose of this retrospective study was to assess the neutrophil-to-lymphocyte ratio (NLR) in diabetic peripheral neuropathy (DPN) patients and its relation to patients' demographic and clinical data.

Methodology:

Study design and setting

A retrospective study was conducted by reviewing the data records over the period of 10 years from January 2019 to December 2021 done at king Abdul-Aziz University Hospital, Jeddah, in the western region of KSA. The study was done on diabetic peripheral neuropathy patients who are using either oral hypoglycemic agents or insulin with regular follow ups.

After ruling out other potential causes, DPN was defined as the existence of symptoms and/or indications of peripheral nerve dysfunction in diabetics. Pinprick, temperature, vibration perception (using a 128-Hz tuning fork), 10-g monofilament pressure sensation at the distal hallux, and ankle reflexes all together have more than 87% sensitivity in detecting DPN [22].

All diabetic patients younger than 18 years old and those having gestational diabetes, those who have a history of malignancy, autoimmune disease, hematological disorder, other DM complications and patientsdiagnosedwithinfectiousdiseasesatthetimeofcollectinglabresults were excluded. In addition to patients onimmunosuppressant'sorcorticosteroidswithsinglevisitorincompletefollowupwereexcludedfr omthestudy.

Sample size and sampling procedure

The sample size calculated for this study was 133 patients with 95% confidence level and a 5% margin of error. The calculations were made using the Raosoft sample size calculator [23].

Data collection instruments

The data was collected via the electronic documentation system used at KAUH (phoenix system), and it was categorized into five sections. Section one was about the demographic data which include age, gender, nationality. Section two consisted of data about medications as Insulin, OHG or both. Section three wasto collect data about HbA1c which was divided into excellent and poor. Section four contained the duration of DM (≤5 years, 6-10 years and >10 years). Section five was to assess the NLR level (Low/Medium or High).

Analysis

Microsoft Excel version 2016 was used for data entry. Regarding data analysis, data were coded, checked, and entered into the Statistical Package for the Social Sciences (IBM® SPSS®, Armonk, NY) version 26.
The categorical variables, consisting of demographic information were expressed as frequencies and percentages. Measures of central tendency were calculated for the continuous variables, including means ± standard deviations, range, median and dispersion. Chi-squared test was used to compare between varies variables with a p-value <0.05 to indicate statistically significant.

Data was available only to the principal investigator to ensure the privacy and confidentiality of participants.

Results:

The study included a total of 133 patients with the majority of them being male i.e., 81 (61%). The mean age of the patients was 60(SD13.5) years. The median value for NLR and HbA1c was 2.8(IQR18.86-5.06) and 8.9(IQR7.14-10.60) respectively. The median duration

for being diabetic was 15(IQR10-24.5) years. More than half 73(54.9%) were non-Saudi and almost all the patients (98.5%) were older than 30 years as shown in Table 1. There were 13 (9.8%) patients who were taking both Insulin and oral hypoglycemic, and another 74 (55.6%) were taking Insulin alone. The HbA1c was excellent in 29 (21.8%), poor in 104(78.2%) of the patients. Approximately two-third of the patients 86(64.7%) had diabetic for more than 10 years as well as Neutrophil-Lymphocyte Ratio.

Table 2 compares the association between the demographic variables and duration of diabetes with the HbA1c categories. There was no significant association identified for gender, age group, nationality, and medication (p >0.05). The duration of diabetes mellitus was found to be associated with the HbA1c categories with 86% of patients having duration of diabetes more than 10 years being in the poor category as compared to 50% in those having diabetes for ≤5 years (p=0002).

Table 3 shows the comparison of the demographic variables with Neutrophil-Lymphocyte Ratio. There was no significant association between any of the demographic variables and duration of diabetes with NLR (p>0.05).

Table 4 shows that there was also no significant association between NLR and HbA1c categories. Figure 1 shows the scatter plot between NLR and HbA1c and it was found that there is weak positive correlation with no significant correlation between the two $r_s=0.073$, $p=0.402$, $N=133$.

Table 1: The sociodemographic and medical characteristics of the study population

	Mean	SD*
Age	60.01	13.52
	Median	**IQR**
Duration Of DM** (Y)	15.0	10-24.5
NLR^	2.8	1.87-5.05

Hb1AC	8.9	7.14-10.6
	N	**%**
Gender		
Male	81	60.9
Female	52	39.1
Nationality		
Saudi	60	45.1
Non-Saudi	73	54.9
Medication		
Insulin	74	55.6
OHGɬ	46	34.6
Both	13	9.8
Age		
18-30 years	2	1.5
>30 years	131	98.5
HbA1c		
Excellent	29	21.8
Poor	104	78.2
Duration		
≤5 Years	18	13.5
6-10 Years	29	21.8
>10 Years	86	64.7
NLR		
Low/medium	47	35.3
High	86	64.7

**DM: Diabetes mellitus, *SD: Standard deviation

^NLR: Neutrophils lymphocytes ratio,ɬOHG: Oral hypoglycemic agents

Table 2: Comparative statistics of HgA1c between the subgroups of the study population

	HgA1c				
	Excellent (n=29)		**Poor (n=104)**		**P-value**
	N	**%**	**N**	**%**	
Gender					
Male	19	23.5%	62	76.5%	0.565*
Female	10	19.2%	42	80.8%	
Age					
18-30 years	1	50.0%	1	50.0%	0.390**
>30 years	28	21.4%	103	78.6%	
Nationality					
Saudi	11	18.3%	49	81.7%	0.379*

	n	%	n	%	P-value
Non-Saudi	18	24.7%	55	75.3%	
Duration					
≤5 years	9	50.0%	9	50.0%	0.002*
6-10 years	8	27.6%	21	72.4%	
>10 years	12	14.0%	74	86.0%	
Medication					
Insulin	16	21.6%	58	78.4%	0.804*
OHG^	11	23.9%	35	76.1%	
Both	2	15.4%	11	84.6%	
	n	**Mean Rank**	**Mann-Whitney U**	**Z**	**P-value**
NLR#			1355	-0.834	
Excellent	29	72.28			0.404ꜰ
Poor	104	65.53			

^OHG: Oral Hypoglycemic agents, #NLR: Neutrophils lymphocytes ratio, *Chi-square test

**Fisher exact test, ꜰMann-Whitney U test

Table 3: Comparative statistics of neutrophils lymphocytes ratio between the subgroups of the study population

	NLR^				
	Low/Medium(n=47)		High(n=86)		p
	n	%	n	%	
Gender					
Male	25	30.9%	56	69.1%	**0.178***
Female	22	42.3%	30	57.7%	
Age					
18-30 years	0	0.0%	2	100.0%	**0.540****
>30 years	47	35.9%	84	64.1%	
Nationality					
Saudi	22	36.7%	38	63.3%	**0.771***
Non-Saudi	25	34.2%	48	65.8%	
Duration					
=<5 years	9	50.0%	9	50.0%	
6-10 years	10	34.5%	19	65.5%	**0.369***
>10 years	28	32.6%	58	67.4%	
Medication					
Insulin	29	39.2%	45	60.8%	
OHG#	13	28.3%	33	71.7%	**0.462***
Both	5	38.5%	8	61.5%	

	n	Mean Rank	Mann-Whitney U	Z	p
Gender					
Male	81	71.93	1707	1.840	**0.066ɫ**
Female	52	59.33			
Nationality					
Saudi	60	64.43	2036	0.696	**0.486ɫ**
Non-Saudi	73	69.11			
Age					
18-30 years	2	65.50	128	0.055	**0.956ɫ**
>30 years	131	67.02			

	n	Mean Rank	Chi-Square	df	p
Duration					
=<5 years	18	58.44			
6-10 years	29	65.00	1.317	2	**0.518ɧ**
>10 years	86	69.47			
Medication					
Insulin	74	65.54	0.591	2	**0.744ɧ**
OHG	46	70.41			

| | Both | 13 | 63.23 | |

Table 4: Association of Hga1C and neutrophils lymphocytes ratio

	NLR₸				OR	95% CI	P-Value
	Low/medium		High				
	N=47	%	N=86	%	OR	95% CI	P-Value
HgA1c							
Excellent	8	27.6%	21	72.4%	0.635	0.257-1.571	0.323*
Poor	39	37.5%	65	62.5%			

₸NLR: Neutrophils lymphocytes ratio, **Chi-square test*
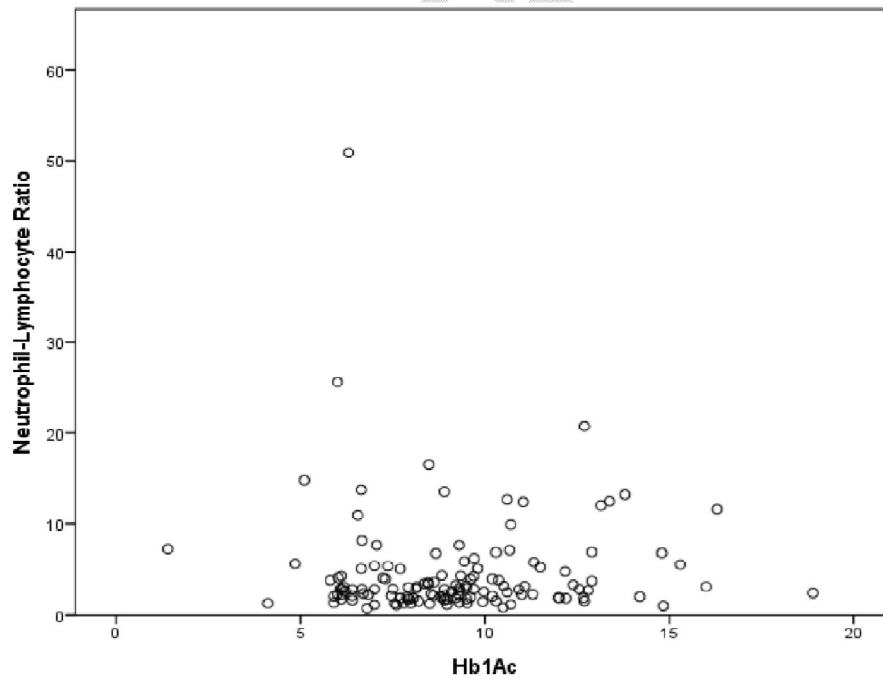
Figure 1: Scatter plot showing a week positive correlation between Neutrophils lymphocytes ratioand HbA1c.

Discussion:

DPNis a microvascular complication of T2DM which is caused by chronic inflammatory proses and dysfunction of immune system. Many inflammatory mediators are associated with developing DPN, like NLR, platelet-lymphocytic ratio (PLR) and lymphocyte-monocyte ratio (LMR) [24].

Regarding NLR, it is a traditional marker which has various advantages over other inflammatory markers because of its wide availability, low cost, reliability, easy lab detection. Moreover, NLR can be used as population screening, disease and drug monitoring tool on large scale basis. Even patients with increase NLR but normal TLC count could have increased risk of atherosclerosis related diseases.[20,25,26]Most of the past studies have showed a significant relationship between high levels of NLR and neurovascular complication, such as cardiomyopathies, retinopathies, chronic kidney disease and DPN [1,27,28].

In this study, the average age was 60 years with sample size of 133 patients. More than half 73(54.9%) were non-Saudi and approximately two-third of the patients 86(64.7%) had diabetic for more than 10 years.There was no significant association identified for gender, age group, nationality, and medication (p >0.05). Also,there was no significant association found between any of the demographic variables and duration of diabetes with NLR (p>0.05). Moreover, there was no significant association between NLR and HbA1c categories.

Unexpectedly, our results were opposite to past studies that showed significant relationship between high NLR and DPN in T2DM [29] or other long-term diabetes related complications[22,27,30,31]. Even though NLR has lots of advantages, it can be affected by some common physical conditions, such as dehydration. Furthermore, physical exercise and release of catecholamine (CA) can cause a drop in neutrophilic granulocyte and lymphocyte[32]. In addition to NLR, there are other inflammatory markers that can be used for early detection and assisting the severity of neurovascular complications in T2DM. For example, research conducted in Turkey, Erzincan University found that PLR is a good predictive marker for chronic inflammation in diabetic patient[33]. Poorly controlled diabetes mellitus, as reflected by elevated HbA1c values and disease complications, is associated with a small but statistically significant elevation of ANC and ALC values[34]. High levels of HbA1C and diabetic complications reflecting a poor control statues of diabetes mellitus, is related with significant elevation of absolute neutrophil counts (ANC) and absolute lymphocyte counts (ALC) according to a study done in Kuwait[34].

The nature of retrospective design relies on the hospital record system which may lack some information regarding the latest laboratory results and loss of follow-up for some patients. Moreover, conducting the study in a single center has resulted in a small sample size which may reduce the power to reveal the true association between NLR and other variables.

Conclusion

The present study investigated neutrophil-to-lymphocyte ratio (NLR) in diabetic peripheral neuropathy (DPN) patients and its association with patients' demographic and clinical data. While not statistically significant, the NLR had a weak positive correlation with the results of HgA1c. We recommend conducting future multi-center studies with larger sample size to assess these associations.

Ethical approval:
The study was approved by the Medical Ethics Committee of King Abdul-Aziz University in December 2919 (Ethical approval reference No 652-19).

Data and material availability:
All data associated with this study are present in the paper.

References:

1. Asher Fawwad and others, 'Neutrophil-to-Lymphocyte Ratio and Microvascular Complications in Subjects with Type 2 Diabetes: Pakistan's Perspective', *Turkish Journal of Medical Sciences*, 48.1 (2018), 157–61 <https://doi.org/10.3906/sag-1706-141>
2. CumaMertoglu and Murat Gunay, 'Neutrophil-Lymphocyte Ratio and Platelet-Lymphocyte Ratio as Useful Predictive Markers of Prediabetes and Diabetes Mellitus', *Diabetes and Metabolic Syndrome: Clinical Research and Reviews*, 11 (2017), S127–31https://doi.org/10.1016/j.dsx.2016.12.021>.
3. David R. Whiting and others, 'IDF Diabetes Atlas: Global Estimates of the Prevalence of Diabetes for 2011 and 2030', *Diabetes Research and Clinical Practice*, 94.3 (2011), 311–21https://doi.org/10.1016/j.diabres.2011.10.029>.
4. James Bentham and others, 'Worldwide Trends in Body-Mass Index, Underweight, Overweight, and Obesity from 1975 to 2016: A Pooled Analysis of 2416 Population-Based Measurement Studies in 128·9 Million Children, Adolescents, and Adults', *The Lancet*, 390.10113 (2017),2627–42 <https://doi.org/10.1016/S01406736(17)32129-3>.

5. Linda Tran and others, 'Pharmacologic Treatment of Type 2 Diabetes: Oral Medications', *Annals of Pharmacotherapy*, 49.5 (2015), 540–56 <https://doi.org/10.1177/1060028014558289>.
6. Yan Zheng, Sylvia H. Ley, and Frank B. Hu, 'Global Aetiology and Epidemiology of Type 2 Diabetes Mellitus and Its Complications', *Nature Reviews Endocrinology*, 14.2 (2018), 88–98https://doi.org/10.1038/nrendo.2017.151>.
7. Basel Alzahrani and others, 'Prevalence and Risk Factors for Diabetic Nephropathy in Type 2 Diabetic Patients, Taif City, Saudi Arabia', *International Journal of Medicine in Developing Countries*, 3.January (2019), 167–72 <https://doi.org/10.24911/ijmdc.51-1541336905>.
8. Mark Davies and others, 'The Prevalence, Severity, and Impact of Painful Diabetic Peripheral Neuropathy in Type 2 Diabetes', *Diabetes Care*, 29.7 (2006), 1518–22https://doi.org/10.2337/dc05-2228>.
9. N. E. Cameron and others, 'Vascular Factors and Metabolic Interactions in the Pathogenesis of Diabetic Neuropathy', *Diabetologia*, 44.11 (2001), 1973–88https://doi.org/10.1007/s001250100001>.
10. N Janahi, '" THE ROLE OF PRO -INFLAMMATOY CYTOKINES AND AUTOIMMUNE ANTIBODIES IN DIABETIC PERIPHERAL NEUROPATHY " NOOR JANAHI A Thesis Submitted in Partial Fulfilment of the Requirements for the Degree of Professional Doctorate of Health and Social Sciences', 2014, 287.
11. Tesfaye S and others, 'Vascular risk factors and diabetic neuropathy' N Engl J Med, 352, (2005),341–350.
12. Kampoli AM, and others, ''otential pathogenic inflammatory mechanisms of endothelial dysfunction induced by type 2 diabetes mellitus. Curr Pharm Des,17 (2011), 4147–4158.
13. Cho H and others, 'Pre-treatment neutrophil to lymphocyte ratio is elevated in epithelial ovarian cancer and predicts survival after treatment' Cancer Immunol Immunother,58, (2009),15–23.
14. Bhat T and others, 'Neutrophil to lymphocyte ratio and cardiovascular diseases: a review', Expert Rev Cardiovasc Ther,11 (2013), 55–59.
15. Huang W and others, 'Neutrophil-lymphocyte ratio is a reliable predictive marker for early-stage diabetic nephropathy' ClinEndocrinol (Oxf) 82 (2015), 229–233.
16. Luo P and others, 'The relationship between neutrophil-to-lymphocyte ratio and intracerebral hemorrhage in type 2 diabetes mellitus' J Stroke Cerebrovasc Dis, 26 (2017), 930–937.
17. Yuan ZY and others, 'Prognostic value of preoperative neutrophil-lymphocyte ratio is superior to platelet-lymphocyte ratio for survival in patients who underwent complete resection of thymic carcinoma' J Thorac Dis, 8(2016), 1487.
18. Kultigin Turkmen and others, 'The Relationship between Neutrophil-to-Lymphocyte Ratio and Inflammation in End-Stage Renal Disease Patients', *Renal Failure*, 34.2 (2012), 155–59https://doi.org/10.3109/0886022X.2011.641514>.
19. SevketBalta, TurgayCelik, and others, 'The Relation between Atherosclerosis and the NeutrophilLymphocyte Ratio', *Clinical and Applied Thrombosis/Hemostasis*, 22.5 (2016), 405–11 https://doi.org/10.1177/1076029615569568>.
20. S. R. Walsh and others, 'Neutrophil-Lymphocyte Ratio as a Prognostic Factor in Colorectal Cancer', *Journal of Surgical Oncology*, 91.3 (2005), 181–84 https://doi.org/10.1002/jso.20329>.

21. Mazhar Hussain and others, 'Neutrophil Lymphocyte Ratio (NLR): A Well Assessment Tool of Glycemic Control in Type-2 Diabetic Patients', *Pakistan Journal of Medical Sciences*, 33.6 (2017), 1366–70 <https://doi.org/10.12669/pjms.336.12900>.

22. Korean Diabetes Association, 'Treatment guideline for diabetes' J Korean Diabetes 12, (2015), S109–S112.

23. http://www.raosoft.com/samplesize.html

24. Tuna Demirdal and Pinar Sen, 'The Significance of Neutrophil-Lymphocyte Ratio, Platelet-Lymphocyte Ratio and Lymphocyte-Monocyte Ratio in Predicting Peripheral Arterial Disease, Peripheral Neuropathy, Osteomyelitis and Amputation in Diabetic Foot Infection', *Diabetes Research and Clinical Practice*, 144 (2018), 118–25 <https://doi.org/10.1016/j.diabres.2018.08.009>.

25. Fauzia Imtiaz and others, 'Neutrophil Lymphocyte Ratio as a Measure of Systemic Inflammation in Prevalent Chronic Diseases in Asian Population', *International Archives of Medicine*, 5.1 (2012), 2https://doi.org/10.1186/1755-7682-5-2>.

26. SevketBalta, ErtugrulKurtoglu, and others, 'Neutrophil-Lymphocyte Ratio as an Important Assessment Tool', *Expert Review of Cardiovascular Therapy*, 12.5 (2014), 537–38https://doi.org/10.1586/14779072.2014.902309>.

27. SagarAshokraoKhandare and others, 'Study of Neutrophil-Lymphocyte Ratio as Novel Marker for Diabetic Nephropathy in Type 2 Diabetes', *Indian Journal of Endocrinology and Metabolism*, 21.3 (2017), 387–92 <https://doi.org/10.4103/ijem.IJEM_476_16>.

28. G. K. Lee and others, 'The Long-Term Predictive Value of the Neutrophil-to-Lymphocyte Ratio in Type 2 DiabeticPatients Presenting with Acute Myocardial Infarction', *Qjm*, 105.11 (2012), 1075–82https://doi.org/10.1093/qjmed/hcs123>.

29. Siying Liu and others, 'Neutrophil-to-Lymphocyte Ratio Is Associated with Diabetic Peripheral Neuropathy in Type 2 Diabetes Patients', *Diabetes Research and Clinical Practice*, 130 (2017), 90–97 <https://doi.org/10.1016/j.diabres.2017.05.008>.

30. Pace NP and others, 'Association Between Neutrophil-Lymphocyte Ratio and Gestational Diabetes-A Systematic Review and Meta-Analysis; J EndocrSoc, 5.7, (2021),5(7):bvab051.

31. Luo WJ and others, 'The relationship of blood cell-associated inflammatory indices and diabetic retinopathy: a Meta-analysis and systematic review, 'Int J Ophthalmol, 12.2 (2019), 312-323.

32. Meiqin Lou and others, 'Relationship between Neutrophil-Lymphocyte Ratio and Insulin Resistance in Newly Diagnosed Type 2 Diabetes Mellitus Patients', *BMC Endocrine Disorders*, 15.1 (2015), 4–9 <https://doi.org/10.1186/s12902-015-0002-9>.

33. Mertoglu C, Gunay M. Neutrophil-Lymphocyte ratio and Platelet-Lymphocyte ratio as useful predictive markers of prediabetes and diabetes mellitus. Diabetes MetabSyndr. 2017 Nov;11 Suppl 1:S127-S131. doi: 10.1016/j.dsx.2016.12.021. Epub 2016 Dec 12. PMID: 28017281.

34. Fatima Ali and others, 'White Blood Cell Subpopulation Changes and Prevalence of Neutropenia among Arab Diabetic Patients Attending Dasman Diabetes Institute in Kuwait', *PLoS ONE*, 13.3 (2018), 1–12 <https://doi.org/10.1371/journal.pone.0193920>.