Research Article

Study of neutrophil lymphocyte ratio in patients with type 2 diabetes mellitus and its correlation with glycemic control

Devamsh G. N.*, Parvathi M., Madhumathi R., Leela Raghavan

Department of Medicine, Bangalore Medical College and Research Institute, KR ROAD, Bengaluru, Karnataka, India

*Correspondence:
Dr. Devamsh G. N.,
E-mail: chdev1990@gmail.com

ABSTRACT

Background: Neutrophil lymphocyte ratio (NLR) is an indicator of subclinical inflammation. Subclinical inflammation may be associated with increased cardiovascular risk. Raised NLR is associated with metabolic syndrome and is found to be a predictor of cardiovascular disorders. There are only few studies assessing the correlation between NLR and glycemic control. The aim of the present study is to investigate the relationship between NLR and glycemic control in type 2 diabetes patients.

Methods: This observational study was conducted in Department of Medicine, Bangalore medical college. 100 patients diagnosed to have type 2 diabetes mellitus were assessed. They were divided into three groups based upon HbA1c levels: Group 1, HbA1c ≤7%; group 2, HbA1c 7-9%, and group 3, HbA1c>9%. Complete blood count and other relevant investigations were performed. SPSS software was used for statistical analysis. T test was used for continuous variables and chi square test for categorical variables. ANOVA test was used to compare three groups. A p value of <0.05 was considered statistically significant.

Results: Out of 100 patients, the white blood cell count (WBC count) was higher in group 3 as compared to group 1 and group 2(p 0.008). Similarly, the absolute neutrophil count was higher in group 3 as compared to group 1 and group 2(p.017). The neutrophil lymphocyte ratio (NLR) was significantly higher in group 3 as compared with group 1 and group 2(p.009). NLR had a positive correlation with HbA1c and was found to be an independent predictor of poor glycemic control in patients with type 2 diabetes mellitus

Conclusions: Our study found a significant positive correlation between NLR and glycemic control. Increased NLR is associated with elevated HbA1c and poor glycemic control. Type 2 diabetes mellitus patients with raised NLR should be evaluated for cardiovascular, renal and ocular complications of diabetes.

Keywords: Neutrophil lymphocyte ratio, HbA1c, Glycemic control

INTRODUCTION

Diabetes mellitus (DM) refers to a group of common metabolic disorders that share the phenotype of hyperglycemia.[1] Several distinct types of DM are caused by a complex interaction of genetics and environmental factors.[1] It is characterized by high blood glucose levels resulting from defects in insulin production, insulin action, or both.[2,3]

Type 2 diabetes mellitus is the predominant form of diabetes worldwide. The incidence of diabetes is increasing day by day. The epidemic of diabetes is under way in both developing and developed countries. According to IDF, some 425 million people worldwide, or

8.8% of adults aged 20-79, are estimated to have diabetes. About 75% live in low and middle income countries. If these trends continue, by 2040 some 642 million people, or one adult in ten, will have diabetes.

It has been estimated by IDF that globally as many as 193 million people, or close to half (46.5%) of all people with diabetes, are unaware of their disease. Most of these cases are type 2 diabetes. Many of these cases present with diabetes related complications at the time of diagnosis. Diabetes and its complications are major causes of early death in most countries. Cardiovascular disease is one of the leading causes of death among people with diabetes and can account for 50% or more of deaths due to diabetes in some populations. Several steps have to be taken in order to control the epidemic of diabetes and to recognize the complications of diabetes at an early stage thereby reducing morbidity and mortality due to diabetes related complications.

HbA1c is used to measure the long term glycemic control in patients with diabetes. HbA1c has been a major tool in assessing the adequacy of therapy and need for intervention in patients with poorly controlled diabetes mellitus. However, HbA1c usually does not predict ongoing inflammation and diabetes associated complications accurately.

Clues to the involvement of inflammation in diabetes date back to more than a century ago, when high doses of sodium salicylate (5.0-7.5 g/d) were first demonstrated to diminish glycosuria in diabetic patients having "the milder form of the disease," presumably type 2 diabetes. Neutrophils, the key effector cells of the innate immune system, play a pivotal role in T2DM and cardiovascular disease pathogenesis.[4] Various neutrophil dysfunctions have been reported in T2DM patients including cell stiffening, impaired chemotaxis and phagocytosis which lead to increased susceptibility to bacterial infections.[5-9] Despite the adverse changes of leukocytes in diabetes, there are currently no specific measurements to assess patient's leukocyte phenotypes or inflammatory status.

Neutrophil lymphocyte ratio (NLR) is a potential marker to determine inflammation in various cardiac and non-cardiac disorders because it has a superior predictive, diagnostic and discriminative ability than total WBC count. It is used to predict the prognosis of diseases such as acute myocardial infarction (MI), stroke, and heart failure. However, there haven't been many studies directly correlating HbA1c and NLR in patients with type 2 diabetes mellitus. The present study was conducted to assess the correlation between NLR and glycaemic control in patients with type 2 diabetes mellitus.

Aims and objectives

- To measure neutrophil lymphocyte ratio in patients with type 2 diabetes mellitus.

- To study the correlation between neutrophil lymphocyte ratio and glycemic control in type 2 diabetes patients.

METHODS

Study population

This observational study was carried out at Department of General Medicine, Bangalore Medical College and Research Institute, Bengaluru between November 2017 and December 2018. The study was conducted after getting the institutional ethical committee clearance. All the patients gave informed written consent for the study. 100 patients with type 2 diabetes mellitus according to ADA criteria who were aged more than 18 years were included in the study. Patients were included in the study by simple random sampling. Patients with acute infections, leukocytosis or leukocytopenia, chronic cardiac, renal or hepatic disorders were excluded from the study. Patients who were a known case of autoimmune disorders, malignancy or hematological proliferative disorders were also excluded from the study. Patients who had received anti-inflammatory or immunosuppressive therapy were also excluded from the study.

Patients were divided into three groups. Group A with HbA1c ≤7, Group B with 7< HbA1c <9 and group C with HbA1c ≥9. A detailed history and general physical examination were conducted.

Blood parameters

After overnight fast, venous samples were collected and total leukocyte count (TLC), neutrophil and lymphocyte count were measured by automated hematology analyzer. Neutrophil lymphocyte ratios (NLR) were estimated by dividing the absolute neutrophil ratio to absolute lymphocyte ratio. HbA1c level was measured by high performance liquid chromatography using automated ion exchange method.

Statistical analysis

Data were presented as mean±standard deviation(SD). SPSS software was used for statistical analysis. T test was used for continuous variables and chi square test for categorical variables. ANOVA test was used to compare three groups. A p value of <0.05 was considered statistically significant. Linear regression analysis was performed to investigate any direct relationship between HbA1c and NLR, WBC count, age, sex, BMI and duration of diabetes.

RESULTS

The sample size in our study was 100 patients. The baseline demographic characteristics of study population is given in table 1. The mean age of patients included in group A, B and C was 55.222±4.3, 55.527±9.6 and

54.472±10.8 respectively (Figure I). There was no statistically significant difference between the three groups (p<0.218). Similarly there was no statistically significant difference between the three groups in terms of demographic characteristics including gender, BMI and duration of diabetes (Table 1, Figures 2). 5 patients (2 from group B and 3 from group C) were on insulin therapy. The rest of the study population was on oral hypoglycemic agents. 38 patients in the study population had hypertension as a co-morbidity.

Table 1: Demographic data of study population.

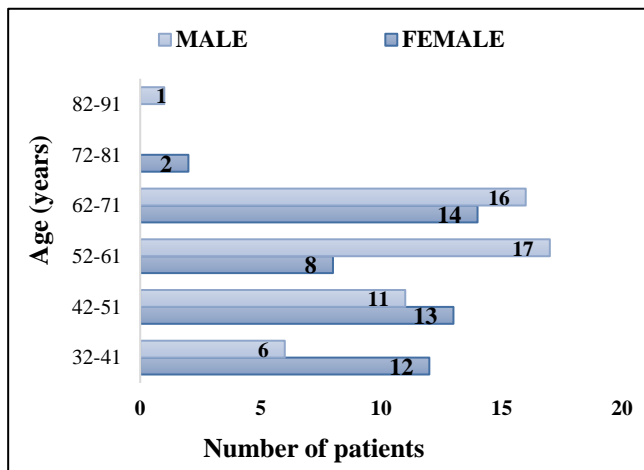| Parameters	Group I (HbA1c <7%)	Group II (HbA1c 7-9%)	Group III (HbA1c >9%)	p values
Age(years)	55.22±4.3	55.52±9.6	54.47±10.8	0.218
BMI(kg/m^2)	21.07±4.1	20.55±5.2	21.10±5.3	0.246
Duration of diabetes	6.93±5.3	6.84±3.6	6.78±4.5	0.535

Figure 1: Age distribution.

The mean fasting blood glucose levels of groups A, B and C were 126.88±49.5, 152.33±52.1 and 234.89±83.2 respectively (p<0.05). Similarly post prandial blood glucose levels of groups A, B and C were 229.55±51.3, 250.91±52.8 and 333.92±73.2 respectively (p<0.05). The FBS and PPBS were significantly higher in group C compared to group A and B (Table 2). The mean HbA1c of patients in group A, B and C was 6.36, 7.82 and 11.69 respectively.
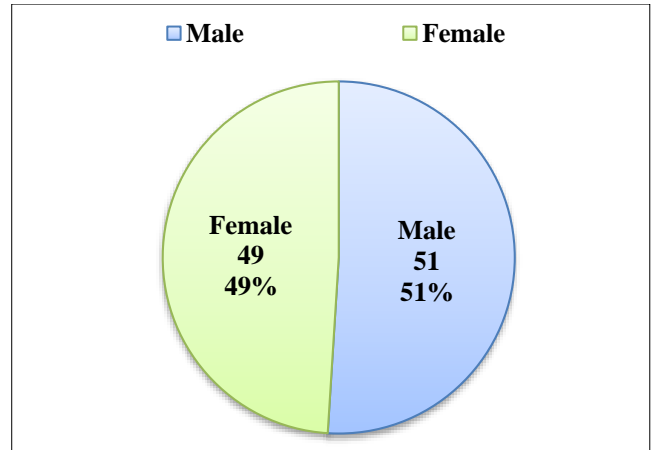

Figure 2: Gender distribution.

The total count was significantly higher in group C as compared to groups A and B (7771.11±2027.5, 8316.02±1625.9 and 9105.455±2310.1 respectively; p<0.05; Figure III). The neutrophil lymphocyte ratio (NLR) was significantly higher in group C as compared to groups A and B (2.496±1.2, 2.701±1.5 and 4.17±2.3; Figure 4). The p value was statistically significant (p<0.05). Linear regression analysis showed that NLR correlated positively with HbA1c.
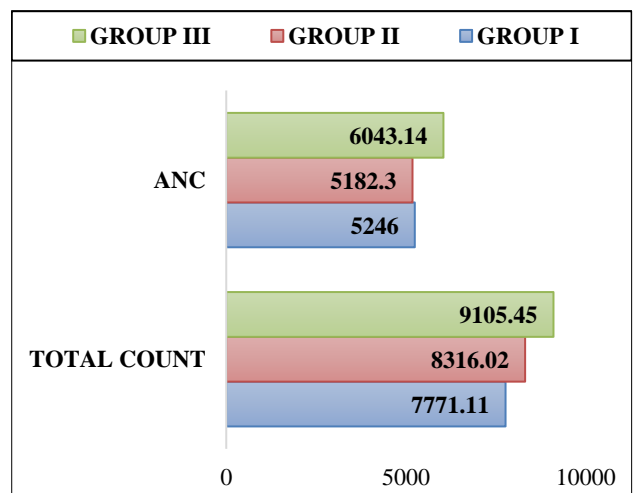

Figure 3: Comparison of total count and ANC among three group.

Table 2: Hematological parameters of study population.

Parameters	Group I (HbA1c<7%)	Group II (HbA1c 7-9%)	Group III (HbA1c >9%)	p values
FBS	126.88±49.5	152.33±52.1	234.89±83.2	6.091E-05
PPBS	229.55±51.3	250.91±52.8	333.92±73.2	0.004
WBC count	7771.11±2027.5	8316.02±1625.9	9105.45±2310.1	0.031
Absolute Neutrophil Count(ANC)	5246±2208.2	5182.3±1475.3	6043.14±2035.9	0.083
Neutrophil Lymphocyte Ratio(NLR)	2.49±1.2	2.701±1.5	4.17±2.3	0.002

DISCUSSION

In our study, there is a positive correlation between NLR and HbA1c. There are not many studies directly correlating NLR and glycemic control although several studies have observed a link between NLR and insulin resistance, metabolic syndrome and atherosclerosis. To our knowledge, there are only few studies directly correlating NLR and glycemic control and this study is the second in south Indian population.
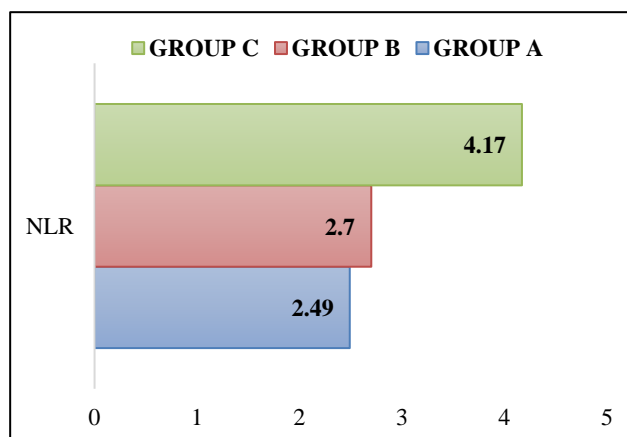

Figure 4: Comparison of NLR among three groups.

Neutrophil lymphocyte ratio NLR is a potential marker to determine inflammation in various cardiac and non-cardiac disorders because it has a superior predictive, diagnostic and discriminative ability than total WBC count. It is used to predict the prognosis of diseases such as acute myocardial infarction (MI), stroke, and heart failure. Predictive value of NLR is comparable to various other inflammatory markers such as C-reactive protein (CRP), tumor necrosis factor (TNF-α) and interleukin (IL-6) in the detection of subclinical inflammation and endothelial dysfunction in various clinical studies.[10] Unlike the total WBC count, NLR is a dynamic parameter and appears to possess a superior predictive value over total leukocyte count.[11] NLR has various advantages over other inflammatory markers because of its wide availability, low cost, reliability and easy lab detection. Even patients with increase NLR but normal TLC count could have increased risk of atherosclerosis related diseases.[12-14] HbA1c is commonly advised to measure the long term glycemic control and it also tells about the severity of hyperglycemia.[15] However HbA1c usually do not predict ongoing inflammation and diabetes associated complications accurately. NLR has a potential to be used to assess inflammatory state and the complications associated with diabetes.

Among the studies conducted outside India, Sefil et al, conducted a study in Turkey in 2013 and concluded that increased NLR may be associated with elevated HbA1c in type 2 diabetes patients.[16] Similarly Mazhar et al, conducted a study in Pakistan in 2016-17 and concluded that increased NLR is associated with elevated HbA1c and poor glycemic control in type 2 diabetes patients.[17]

Shiny et al, revealed that increased NLR has strong association with glucose intolerance and insulin resistance in type 2 diabetic patients.[18] Oh et al, and Demirtas et al, also came to similar conclusions as our study.[19,20] Of note is that several studies have consistently shown that NLR is also associated with diabetic nephropathy and diabetic retinopathy.[21-23]

Drawbacks of our study include, small sample size, effects of different anti diabetic drugs used by patients could not be assessed and hypertension was a comorbidity in 38% of the study population.

CONCLUSION

NLR is an easily available, safe, cost effective and simple test. NLR is not only a marker of subclinical inflammation but also an indicator of poor glycemic control in patients with type 2 diabetes. NLR can be used as a screening test to identify type 2 diabetes patients more likely to develop diabetes related complications and initiate preventive measures and avoid/delay life threatening complications. NLR can also guide the physician in the resource limited settings like primary health centres (PHC) when to evaluate a patient with type 2 diabetes for microvascular and macrovascular complications.

REFERENCES

1. Fauci AS, Braunwald E, Kasper DL, Hauser SL, Longo DL, Jameson JL, et al. Harrison's principles of internal medicine. 19th edition, New York: McGraw-Hill; 2015;2,16:3178-3209.
2. Centers for Disease Control and Prevention. National Diabetes Fact Sheet: General Information and National Estimates on Diabetes in the United States. Atlanta, GA. US Department of Health and Human Services, Centers for Disease Control and Prevention; 2007.
3. The Expert Committee on the Diagnosis and Classification of Diabetes Mellitus. Report of the expert committee in the diagnosis and classification of diabetes mellitus. Diabetes Care. 2003;26 Suppl 1:S5-20.
4. Bagdade JD, Root RK, Bulger RJ. Impaired Leukocyte Function in Patients with Poorly Controlled Diabetes. Diabetes. 1974;23(1):9-15.
5. Ernst E, Matrai A. Altered red and white blood cell rheology in type II Diabetes. Diabetes. 1986;35(12):1412-535(12):1412-5.
6. Pécsvárády Z, Ficher TC, Darwin CH, Fabok A, Maqueda TS, Saad MF, et al. Decreased polymorphonuclear leukocyte deformability in NIDDM. Dia Care 1994 Jan 1;17(1):57-63.
7. Mowat AG, Baum J (1971) Chemotaxis of polymorphonuclear leukocytes from patients with diabetes mellitus. N Engl J Med 284(12):621-27.

8. Delamaire M, Maquendre D, Moreno M, Legoff MC, Allanic H, Genetet B. Impaired leucocyte functions indiabetic patients. Diabet Med.1997 Jan;14(1):29-34.

9. Shah BR, Hux JE (2003) Quantifying the risk of infectious diseases for people with diabetes. Diabetes Care.2003 Feb 1;26(2):510-13.

10. Azab B, Jaglall N, Atallah JP, Lamet A, Raja-Surya V, Farah B, et al. Neutrophil-lymphocyte ratio as a predictor of adverse outcomes of acute pancreatitis. Pancreatology 2011;11(4):445-52.

11. Bhutta H, Agha R, Wong J, Tang TY, Wilson YG, Walsh SR. Neutrophil-lymphocyte ratio predicts medium-term survival following elective major vascular surgery: a crosssectional study. Vasc Endo Surg 2011;45(3):227-31.

12. Ozturk C, Balta S, Balta I, Demirkol S, Celik T, Turker T, et al. Neutrophil–lymphocyte ratio and carotid–intima media thickness in patients with Behcet disease without cardiovascular involvement. Angio. 2015 Mar;66(3):291-6.

13. Balta S, Celik T, Mikhailidis DP, Ozturk C, Demirkol S, Aparci M, et al. The relation between atherosclerosis and the neutrophil–lymphocyte ratio. Clinical applied thrombosis/hemostasis. 2016 Jul;22(5):405-11.

14. Balta S, Kurtoglu E, Kucuk U, Demirkol S, Ozturk C. Neutrophil–lymphocyte ratio as an important assessment tool. Expert review cardiova ther. 2014 May 1;12(5):537-8.

15. Lyons TJ, Basu A. Biomarkers in diabetes: hemoglobin A1c, vascular and tissue markers. Translational Research. 2012 Apr 1;159(4):303-12.

16. Selfil F, Ulutas KT, Dokuyucu R, Sumbul AT, Yengil E, Yagiz AE, et al. Investigation of neutrophil lymphocyte ratio and blood glucose regulation in patients with type 2 diabetes mellitus. J Int Med Res. 2014;42(2):581-8.

17. Hussain M, Babar MZM, Akhtar L, Hussain MS. Neutrophil lymphocyte ratio (NLR): A well assessment tool of glycemic control in Type- 2 diabetic patients. Pak J Med Sci. 2017;33(6):1366-70.

18. Shiny A, Bibin YS, Shanthirani CS, Regin BS, Anjana RM, Balasubramanyam M, et al. Association of neutrophil lymphocyte ratio with glucose intolerance: an indicator of systemic inflammation in patients with type 2 diabetes. Diabetes Technol Ther. 2014;16(8):524-30.

19. Oh Y, Kwon GC, Koo SH, Kim J. Association between glycemic control and hematolical indices in type 2 diabetic patients. Lab Med Online. 2016;6(3):134-9.

20. Demirtas L, Degirmenci H, Akbas EM, Ozcicek A, Timuroglu A, Gurel A, et al. Association of haematological indices with diabetes, impaired glucose regulation and microvascular complications of diabetes. Int J Clin Exp Med. 2015;8(7):11420-7.

21. Chittawar,S., Khandare,S., Nahar, N., Dubey,T., Qureshi,Z.. Study of neutrophil-lymphocyte ratio as novel marker for diabetic nephropathy in type 2 diabetes. Ind J Endocr Metab. 2017;21(3):387-92.

22. Srinivas B, Maldar A, Bellad A, Mukhi N. Relation of neutrophilic lymphocyte ratio to microvascular complications of diabetes mellitus. Int J Health Sci Res. 2015;5(12):34-8.

23. Kahraman C, Kahraman NK, Aras B, Coşgun, S, Gülcan E. The relationship between neutrophil-to-lymphocyte ratio and albuminuria in type 2 diabetic patients: a pilot study, Arch Med Sci. 2016;12(3):571-5.