INTRODUCTION

Type 2 diabetes mellitus has become a serious threat to global human health because of its vascular complications which are associated with increased disability, frailty and reduced life expectancy, including cardiovascular and cerebrovascular diseases (CVD), diabetic nephropathy, diabetic retinopathy, diabetic foot. India is the diabetes capital of the world, with diabetes and pre-diabetes, prevalence of 9% and 11-14%, respectively. This increased burden of diabetes in Indians is a grim precursor of an exponential increase in diabetes-related end-organ damage and associated morbidity in the next few decades. There is an urgent need for cheap and easy to measure predictors of the occurrence of diabetes related end-organ damage in Indians. This would help in preventing vascular complications in specific individuals to improve long term clinical outcomes.

Chronic inflammation has been considered the potential pathogenesis responsible for the development of diabetic complications. Increased white blood count (WBC) is an easily available inflammatory marker, which correlates with several cardiovascular disease risk factors and its sequel with diabetes. Apart from WBC count, inflammatory markers...
such as interleukin 1(IL-1), IL-6, IL-8, transforming growth factor β1, tumor necrosis factor-α have been linked to end organ damage in diabetes.12-16 Limitations of these markers include lack of availability in routine clinical practice compounded by the associated increased expenses and assay standardization.17,18 Among multiple parameters complete blood count, neutrophil lymphocyte ratio (NLR) has been studied extensively for the association with cardiovascular risk factors like myocardial infarction, heart failure and stroke.19,20 NLR stands out as a novel marker of chronic inflammation that reflects a balance between 2 complementary components of the immune system- Neutrophils being the active nonspecific mediator of inflammation, whereas Lymphocytes acting as the protective or regulatory component of inflammation.21,22

Data are lacking of NLR as a predictor of end-organ damage in Indians with T2DM. This study, therefore, aimed to evaluate the role of NLR has a role in predicting diabetic vascular complications such as retinopathy, neuropathy, nephropathy, diabetic foot, cardiovascular and cerebrovascular complications.

**Aims and objectives**
To compare Neutrophil-Lymphocyte ratio in diabetics with and without vascular complications.

**MATERIALS AND METHODS**

**Study design**
Cross sectional type.

**Study area**
The present study was conducted on patients admitted in hospitals affiliated to Bangalore Medical College and Research Institute, Bangalore.

**Inclusion criteria**
1. Age >18 years
2. Patient willing to give informed consent
3. Diagnosis as per American diabetes association 2018 guidelines23

**Exclusion criteria**
1. Patients who did not give informed consent

**Study population**
A study population of 111 diabetic patients with and without vascular complications was taken with 56 patients with complications and 55 without complications.

**Study period**
November 2018 to May 2020.

**Methodology of data collection**
This study was conducted among patients admitted under medicine department between November 2018 to May 2020 at Victoria hospital and Bowring & Lady Curzon hospital, Bangalore Medical College and Research Institute, Bangalore, Karnataka, India. Approval and clearance were obtained from the institutional ethics committee on 15/11/2018-BMCR/I/PG/124/2018-19. Information was collected from the patients on their duration of T2DM, treatment history, age and sex. Data were collected on the anthropometric parameters and vitals of the patients (height, weight, BMI), Pulse rate and Blood pressure. Both fasting and post prandial blood samples of 5 ml each were collected in plain vacutainer and processed immediately for routine biochemical analysis. Spot urine sample was collected and looked for presence of albuminuria. Such patients underwent ultrasonography abdomen for the evaluation of the kidney echotexture and size. Digital fundoscopy was done to assess diabetic retinopathy. Diabetic retinopathy was diagnosed using The Early Treatment Diabetic Retinopathy Study criteria (ETDRS).24 Patients who had neuropathic symptoms were clinically examined for diabetic neuropathy. Patients who had symptoms and signs of lower limb ischemia were evaluated with respective limb arterial and venous doppler. Patients were screened and evaluated clinically for macrovascular complications and relevant investigations like ECG, 2D ECHO for cardiovascular complications and neuroimaging for cerebrovascular complications. All relevant investigations were done on out-patient and in-patient basis.

**STATISTICAL ANALYSIS**

**Statistical methods**
Data was entered into Microsoft excel data sheet and was analyzed using SPSS 22 version software. Categorical data was represented in the form of Frequencies and proportions. Chi-square test was used as test of significance for qualitative data. Continuous data was represented as mean and standard deviation. Independent t test was used as test of significance to identify the mean difference between two quantitative variables. ANOVA (Analysis of Variance) was the test of significance to identify the mean difference between more than two groups for quantitative data.

| Table 1: Age distribution of subjects in the study |
|----------------|----------|--------|
| Age            | Count    | %      |
| <40 years      | 17       | 15.3%  |
| 41 to 50 years | 27       | 24.3%  |
| 51 to 60 years | 27       | 24.3%  |
| 61 to 70 years | 25       | 22.5%  |
| 71 to 80 years | 11       | 9.9%   |
| >80 years      | 4        | 3.6%   |
Gowda, et al.: Association of Neutrophil- Lymphocyte ratio with vascular complications in Type-2 Diabetes

Table 2: Glycemic Profile of subject’s comparison with respect to duration of diabetes

<table>
<thead>
<tr>
<th>Duration of Diabetes</th>
<th>Newly Detected</th>
<th>&lt;5 years</th>
<th>6 to 10 years</th>
<th>&gt;10 years</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean</td>
<td>SD</td>
<td>Mean</td>
<td>SD</td>
<td>Mean</td>
</tr>
<tr>
<td>FBS (mg/dl)</td>
<td>202.77</td>
<td>70.93</td>
<td>242.5</td>
<td>85.35</td>
<td>230.44</td>
</tr>
<tr>
<td>PPBS (mg/dl)</td>
<td>265.67</td>
<td>63.67</td>
<td>281.38</td>
<td>78.14</td>
<td>276.29</td>
</tr>
<tr>
<td>HbA1c (%)</td>
<td>9.42</td>
<td>1.56</td>
<td>9.86</td>
<td>1.93</td>
<td>9.58</td>
</tr>
</tbody>
</table>

Graphical representation of data
MS Excel and MS word was used to obtain various types of graphs such as bar diagram, Pie diagram and ROC Curve

p value (Probability that the result is true) of <0.05 was considered as statistically significant after assuming all the rules of statistical tests.

Statistical software
MS Excel, SPSS version 22 (IBM SPSS Statistics, Somers NY, USA) was used to analyze data.

RESULTS

This study was conducted among 111 patients admitted in Victoria hospital and Bowring & Lady Curzon hospital attached to Bangalore Medical College and Research Institute.

Age distribution
In this study Mean age was 55.66 ± 13.675 years (Table 1).

Sex distribution
Out of 111 patients 63 patients (56.8%) were Male and 48 patients (43.2%) were Female. In this study male patients were more than female.

Distribution of cases according to duration of diabetes
In the study, duration of diabetes was newly detected in 16.2%, <5 years in 37.8%, 6 to 10 years in 30.6% and >10 years in 15.3%.

Glycemic profile of subject’s comparison with respect to duration of diabetes
There was no significant correlation of FBS, PPBS and HbA1C with the duration of diabetes (Table 2).

Correlation of NLR with respect to duration of diabetes
There was significant correlation of N (%) and NL ratio with duration of diabetes but L (%) did not show significant correlation with duration of diabetes (Table 3).

Vascular complications in diabetic subjects
In the study patients had no vascular complications in 49.5%, had micro-vascular complications in 22.5% and macro-vascular complications in 28.8%. Out of which patients had Diabetic retinopathy in 34%, diabetic nephropathy in 34.2%, diabetic neuropathy in 0.9%, cerebrovascular Accident in 17.1%, ischemic heart disease in 12.6%, diabetic foot in 6.3% of the individuals (Table 4).

NLR comparison with complications and glycemic profile in type 2 diabetics
In the study N (%) was 70.9 ±13.6, L (%) was 19 ± 8.1 and NL Ratio was 4.8 ± 3 among subjects involved in the study.

In this study, mean N (%) in no vascular complications was 61.7 ± 10.6, in micro complications was 80.6 ± 9.4 and in macro complications was 79.5 ± 9.7. There was a significant difference in mean N (%) comparison with respect to complications.

Mean L (%) in no vascular complications was 23.7 ± 5.8, in micro complications was 14.1 ± 6.9 and in macro
Table 5: NLR Comparison with respect to complications

<table>
<thead>
<tr>
<th>Complications</th>
<th>No Vascular Complications</th>
<th>Micro complications</th>
<th>Macro complications</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean ± SD</td>
<td>Mean ± SD</td>
<td>Mean ± SD</td>
</tr>
<tr>
<td>N (%)</td>
<td>61.7 ± 10.6</td>
<td>80.6 ± 9.4</td>
<td>79.5 ± 9.7</td>
</tr>
<tr>
<td>L (%)</td>
<td>23.7 ± 5.8</td>
<td>14.1 ± 6.9</td>
<td>14.8 ± 8.0</td>
</tr>
<tr>
<td>NL Ratio</td>
<td>2.8 ± 0.7</td>
<td>7.1 ± 3.5</td>
<td>6.5 ± 2.7</td>
</tr>
</tbody>
</table>

Table 6: Correlation between NLR with Glycemic Profile

<table>
<thead>
<tr>
<th>NLRATIO</th>
<th>Pearson Correlation</th>
<th>P value</th>
<th>N</th>
</tr>
</thead>
<tbody>
<tr>
<td>FBS (mg/dl)</td>
<td>0.38</td>
<td>&lt;0.000039</td>
<td>111</td>
</tr>
<tr>
<td>PPBS (mg/dl)</td>
<td>0.49</td>
<td>&lt;0.00001</td>
<td>111</td>
</tr>
<tr>
<td>HBA1C (%)</td>
<td>0.29</td>
<td>.002019</td>
<td>111</td>
</tr>
</tbody>
</table>

Table 7: Validity of NLR in predicting Vascular Complications

| Area under the ROC curve (AUC) | 0.924 |
| Standard Error                | 0.0315 |
| 95% Confidence interval       | 0.858 to 0.966 |
| z statistic                   | 13.441 |
| Significance level P (Area=0.5) | <0.0001 |

| Youden index J               | 0.8565 |
| 95% Confidence interval      | 0.7326 to 0.9282 |
| Associated criterion         | >3.9989 |
| 95% Confidence interval      | 3.477203178 to 4.2334 |

DISCUSSION

Diabetes is a growing health problem associated with increased risk of micro and macro-vascular complications. With the easy availability of various blood tests such as complete blood count (CBC) efforts are made to identify and prove their utility to act as bio-markers for early detection of diabetic complications.

In the study there was significant positive correlation between NLR and glycemic profile i.e., with increase in FBS, PPBS, HBA1C there was increase in NLR and vice versa, (Table 6).

NLR of >3.998 had highest sensitivity of 89.29%, Specificity of 96.36%, PPV of 96.2% and NPV of 89.8% in predicting vascular complications among diabetics (Figure 2, Table 8).
of NLR with vascular complications in Type 2 DM. 111 patients were included in this study, where 56 patients were diabetics with vascular complication and 55 patients were diabetics without vascular complications.

**Age**
In this study the most common age group was 51-60 (22 patients) with mean age of 55.66±13.67. Most of the patients were of 40-70 years. The IDF Diabetes Atlas which tracks the global impact of diabetes also shows the more common age group of diabetes is between 20-79 years of age. This is important because of increasing risk within population and failure to control factors such as obesity and poor diets. This would further cause a rising prevalence as each person getting diagnosed at an earlier age stays longer.

**Sex**
In this study there was male predominance i.e., 56.8% of patients and female patients were 43.2%, which was similar to study done by Mohammed Haghighatpanah et al, Correlation of glycosylated hemoglobin levels with fasting and postprandial glucose in South Indian Type 2 Diabetic patients.

**Duration of diabetes**
In this study, Duration of Diabetes was Newly Detected in 16.2%, <5 years in 37.8%, 6 to 10 years in 30.6% and >10 years in 15.3%. Ours being a tertiary care center most of the patients were incidentally diagnosed patients and duration more than 6 years who were on regular follow up for their medications.

There was no significance in mean FBS, mean PPBS, mean HBA1C compared with the duration of the diabetes.

In this study, Duration of diabetes had significant correlation with N (%) with p value (0.035) and NL ratio with p value (0.045) but the L (%) did not show significant correlation.

Study done by Sachin chittawar et al. showed that patients in higher NLR quartiles had significantly higher diabetes duration.

**Glycemic profile**
In this study, there was a significant positive correlation found in between NLR and FBS, NLR and PPBS and NLR and HBA1C.

Study done by Mazhar hussain et al. also showed that NLR was significantly high in diabetic patients with poor glycemic control.

**NLR with respect to vascular complications in type-2 diabetes**
Study done by Moursy et al. showed NLR is an important predictor for the presence of microvascular complications like diabetic nephropathy, retinopathy and neuropathy.

In this study there was a significant difference in NLRatio comparison with respect to vascular complications in patients with Type 2 DM. Thus, this study highlighted the importance of a routine assessment of Neutrophil-Lymphocyte ratio which can be easily calculated from a simple peripheral blood count in patients with diabetes to predict the vascular complications.

**CONCLUSION**
From this study we can conclude that NLR is significantly increased in diabetic patients with complications compared to diabetics without complications showing that increment is more in diabetics with poor glycemic control and longer duration of the disease. NLR would be a useful marker of vascular complications in diabetes both micro and macro-vascular. Hence, it can be shown that NLR can be used as a simple and cost-effective tool to monitor the progression and control of DM and its vascular complications.

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**REFERENCES**


Gowda, et al.: Association of Neutrophil- Lymphocyte ratio with vascular complications in Type-2 Diabetes


Author's Contribution:
STA - Concept and design of the study; coordination; SHK - Interpreted the results; reviewed the literature and manuscript preparation; RKR - Review of literature and manuscript preparation; coordination; RMG - Prepared first draft of manuscript Statistically analyzed and interpreted, preparation of manuscript and revision of the manuscript.

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