The long-term predictive value of the neutrophil-to-lymphocyte ratio in Type 2 diabetic patients presenting with acute myocardial infarction

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Summary

Background: Patients with diabetes mellitus have worse long-term outcomes after acute myocardial infarction (AMI) than non-diabetics. This may be related to differential contribution of neutrophil and lymphocyte to inflammation during AMI in diabetics vs. non-diabetics. We aim to determine the predictive value of neutrophil-to-lymphocyte ratio (NLR) for major adverse events post-AMI in Type 2 diabetics vs. non-diabetics.

Methods and Results: A total of 2559 consecutive patients admitted for AMI (61 ± 14 years, 73% male and 43% diabetic) were analyzed. A complete blood count was obtained and the NLR computed for each patient on admission. Across the cohort, the 1-year reinfarction rate was 8.4% (n = 214) and 1-year mortality was 14.5% (n = 370). Univariate determinants of the composite endpoint included age, hypertension, hyperlipidemia, smoking, revascularization and NLR (P < 0.001 for all). The cohort was divided into NLR quartiles. Admission NLR was significantly higher in the diabetic group, 5.2 ± 5.8 vs. 4.6 ± 5.4 (P = 0.007). A step-wise increase in the incidence of the composite endpoint was noted across NLR quartiles for diabetic subjects; hazard ratio (HR) was 2.41 for fourth vs. first quartile (95% confidence interval = 1.63–3.53, P < 0.001). Multivariate analysis of the diabetic group showed that NLR remains an independent predictor of the composite endpoint (adjusted HR = 1.53, 95% confidence interval = 1.00–2.33, P = 0.048). However, in non-diabetics, HR for NLR was not significant (P = 0.35).

Conclusions: Increased NLR post-AMI is an independent predictor of major adverse cardiac events in diabetics. Monitoring this easily obtainable new index allows prognostication and risk stratification.

Introduction

Inflammation is a cornerstone in the pathogenesis of acute myocardial infarction (AMI) and has been shown to be a strong predictor of adverse events post-AMI. For example, raised inflammatory markers such as C-reactive protein have been shown to predict increased long-term mortality and heart failure post-AMI;¹ elevated white blood cell (WBC) count has good predictive value for the development of coronary artery disease²–³ and is a strong independent predictor of adverse outcomes in patients presenting with AMI.⁴,⁵ The neutrophil-to-lymphocyte ratio (NLR) is an emerging marker for
both cardiac and non-cardiac disorders, and recent studies have demonstrated the prognostic value of NLR in stable coronary artery disease, \(^6\) acute coronary syndromes, \(^7\)–\(^10\) heart failure \(^11\)–\(^12\) as well as patients undergoing percutaneous coronary interventions (PCI) \(^13\) and coronary artery bypass grafting. \(^14\)

Diabetes mellitus portends a poorer prognosis after AMI. \(^15\) Large studies have shown that patients with diabetes have higher 30-day \(^16\) and 1-year \(^17\) mortality rates post-AMI. The severity and duration of diabetes correlate significantly with the prognosis. \(^16\)–\(^17\) Patients with diabetes are more likely to have diffuse disease, microvascular dysfunction and impaired cardiac myocyte contractility. This may be, in part, related to differential contribution of neutrophil and lymphocyte to inflammation during AMI in diabetic vs. non-diabetic subjects. One study has shown the NLR to be an important predictor of macrovascular disease in diabetic subjects, \(^18\) but to our knowledge there has been no study evaluating the prognostic value of NLR in this group post-AMI. In this study, we aim to determine the predictive value of NLR for major adverse events post-AMI in diabetic compared with non-diabetic subjects.

**Methods**

We conducted a retrospective study of 2559 consecutive patients admitted to the National University Hospital, Singapore, a 928-bed academic medical center, for AMI from January 2001 to September 2005. A complete blood count was obtained on presentation to the emergency department and the NLR was computed for each patient. Other baseline characteristics obtained on interviewing the patient or from the electronic medical record included age, history of diabetes mellitus, history of hypertension, history of hyperlipidemia, smoking status, history of prior myocardial infarction, prior PCI and prior coronary artery bypass grafting. We measured peak serum creatine kinase (CK), creatine kinase – myocardial band (CK-MB) and troponin levels as well as obtained serial electrocardiograms for each patient. AMI was diagnosed in accordance with the universal definition of myocardial infarction, \(^19\) and CK-MB was assumed to be a surrogate marker of infarct size. We obtained data on revascularization with either thrombolytic therapy or PCI during the admission. The patients were subsequently followed up for 1 year. Study endpoints were the 1-year reinfarction and mortality rates. Patients readmitted for another AMI within 1 year of the index event were considered to sustain a reinfarction. These data were obtained from electronic medical records and/or the Registry of Births and Deaths.

Patients were considered diabetic if they had a history of diabetes, diagnosed with diabetes during the index admission using generally accepted criteria or were on anti-diabetic medications at presentation. We included only Type 2 diabetic patients in our analysis. The cohort was divided into diabetic and non-diabetic groups, and the mean NLR was computed between the two groups. Both the diabetic and non-diabetic groups were subsequently divided into four quartiles with equal numbers of subjects based on their NLR. The proportions of patients meeting the endpoints were computed for each quartile and compared for both the diabetic and non-diabetic groups. The hazard ratios (HRs) of NLR between the first and fourth quartiles for predicting study endpoints were computed for diabetic and non-diabetic patients. Univariate predictors of study endpoints were determined, and the HRs were adjusted for these univariate predictors.

Categorical variables are expressed as frequencies and percentages. For continuous variables with normal distributions, the results are expressed as mean ± standard deviation. The Student’s t-test and one-way analysis of variance test with post hoc multiple comparisons were used to compare means for continuous variables; the \(\chi^2\) test was used to compare proportions. A \(P\)-value of <0.05 indicated statistical significance. A multivariate logistic regression model was used to assess the independent association of NLR with 1-year reinfarction and mortality rates after adjusting for variables found to predict study endpoints. All analyses were performed using SPSS version 16.0 for Windows.

**Results**

The mean age of our cohort was \(61 \pm 14\) years, 73\% of subjects were male and 43\% were diabetic. Baseline characteristics are described in Table 1; diabetic subjects were significantly older, had higher incidences of hypertension, hyperlipidemia, prior AMI, prior coronary artery bypass grafting, were less likely to be smokers and were less likely to be revascularized. In-hospital, 1-month, 6-month and 1-year mortality rates increased significantly across the second to fourth NLR quartiles (\(P<0.001\) for all). Across the cohort, the 1-year reinfarction rate was 8.4\% (\(n=214\)) and 1-year mortality was 14.4\% (\(n=370\)); 21.5\% (\(n=553\)) of subjects met the composite endpoint of 1-year reinfarction and mortality rates. Thirty-one patients had reinfarction events and subsequently died. A significantly higher proportion of diabetic subjects met the
composite endpoint compared with non-diabetic subjects (28.0% vs. 16.6%, \( P < 0.001 \)). Univariate determinants of the composite endpoint included age, hypertension, hyperlipidemia, smoking, revascularization and NLR (\( P < 0.001 \) for all). The cohort was divided into NLR quartiles for both the diabetic and non-diabetic groups. Admission NLR was significantly higher in the diabetic group, 5.2 \( \pm \) 5.8 vs. 4.6 \( \pm \) 5.4 (\( P = 0.007 \)). A significant increase in mean neutrophil percentage and decrease in mean lymphocyte percentage across the four quartiles was noted in both the diabetic and non-diabetic groups (\( P < 0.001 \) for both) (Table 2). There was a significant difference between the proportions of neutropenic patients in the first quartiles of the non-diabetic and diabetic groups (\( P < 0.001 \) for both) (Table 2). There was a significant difference between the proportions of neutropenic patients in the first quartiles of the non-diabetic and diabetic groups (\( P < 0.001 \) for both) (Table 2). There was a significant difference between the proportions of neutropenic patients in the first quartiles of the non-diabetic and diabetic groups (\( P < 0.001 \) for both) (Table 2). There was a significant difference between the proportions of neutropenic patients in the first quartiles of the non-diabetic and diabetic groups (\( P < 0.001 \) for both) (Table 2).

<table>
<thead>
<tr>
<th></th>
<th>Overall ( (n = 2559) )</th>
<th>Diabetic subjects ( (43%, n = 1097) )</th>
<th>Non-diabetic subjects ( (57%, n = 1462) )</th>
<th>( P )</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>61 ( \pm ) 14</td>
<td>64 ( \pm ) 13</td>
<td>60 ( \pm ) 15</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Hypertension</td>
<td>61%</td>
<td>76%</td>
<td>50%</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Hyperlipidemia</td>
<td>73%</td>
<td>75%</td>
<td>71%</td>
<td>0.019</td>
</tr>
<tr>
<td>Smoker</td>
<td>51%</td>
<td>38%</td>
<td>60%</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Prior MI</td>
<td>14%</td>
<td>18%</td>
<td>11%</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Prior PCI</td>
<td>7%</td>
<td>8%</td>
<td>6%</td>
<td>0.076</td>
</tr>
<tr>
<td>Prior CABG</td>
<td>4%</td>
<td>5%</td>
<td>3%</td>
<td>0.006</td>
</tr>
<tr>
<td>Revascularization</td>
<td>58%</td>
<td>51%</td>
<td>63%</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

CABG = coronary artery bypass grafting.

Discussion

Our findings are significant in that the complete blood count on admission, an inexpensive and routinely performed investigation, carries important prognostic information regarding 1-year reinfarction and mortality risks for AMI. In our study, the admission NLR was found to be an independent predictor of 1-year reinfarction and mortality in diabetic patients admitted for AMI.

Neutrophils secrete many mediators that are responsible for the inflammatory processes occurring during AMI including elastase,\(^5\) myeloperoxidase,\(^20\) oxygen free radicals\(^21\) and various hydrolytic enzymes such as acid phosphatases\(^22\) that are associated with further tissue damage and plaque disruption.\(^9\) Indeed, activated neutrophils have been found in eroded plaque specimens in patients presenting with AMI,\(^23\) indicating their possible role in the pathogenesis of AMI. Increased neutrophil counts in AMI are possibly part of a maladaptive process and are associated with platelet–leukocyte aggregation in the microcirculation leading to the no-reflow phenomenon,\(^24\) larger infarct sizes,\(^25\) adverse angiographic outcomes\(^26\) and short-term prognosis in patients with non-ST-segment acute coronary syndromes\(^27\) and ST-segment elevation myocardial infarction.\(^28\) On the other hand, lymphocytes, particularly the CD4+ count, represent the regulatory arm of the immune system; low lymphocyte counts are associated with poorer prognosis and increased mortality post-AMI.\(^29\) An initially low CD4+ count and a CD4+ count that remained low were associated with higher reinfarction and mortality rates.\(^30\) Moreover, the number and function of CD4+/CD25+ regulatory T cells, believed to modulate the inflammatory and cytotoxic response during AMI, are decreased in patients...
presenting with AMI, possibly in response to increased oxidized low-density lipoprotein.31 The relative lymphopenia observed in AMI may also be attributed to the acute stress response mediated by increased endogenous cortisol secretion.29 Therefore, in the NLR, we have a readily available independent prognostic indicator that integrates the two opposing but interrelated arms of the immune system. Although WBC counts have been shown to predict adverse outcomes in patients with AMI, common physiologic conditions (e.g. dehydration) and in vitro handling of blood specimens may affect the accuracy of this reading. For example, exercise and catecholamine release may result in increases in both the individual neutrophil and lymphocyte count,32 but affect the NLR to a lesser extent. Indeed, there is evidence to suggest that the NLR has superior predictive value to WBC count for post-discharge mortality.33

To our knowledge, the prognostic value of the NLR following AMI in diabetic subjects has not been systematically explored. Our study is the first to isolate the relationship between NLR and adverse outcomes post-AMI solely to diabetic patients. A similar (albeit less pronounced) relationship is seen in quartiles 2 to 4 of the non-diabetic cohort. A possible explanation for the anomalous quartile 1 in non-diabetic patients is the inclusion of neutropenic patients who may sustain increased adverse events; this U-shaped relationship may not be reflected in the diabetic cohort given that the mean absolute neutrophil count in quartile 1 of the diabetic cohort is higher. Indeed, our study found a statistically significant difference between the proportions of neutropenic patients in quartile 1 of the non-diabetic and diabetic cohorts. Neutropenia is associated with increased mortality in certain situations such as post-renal transplant patients,34 but no study has evaluated the impact of neutropenia on outcome in patients with an AMI. Another possible explanation may be a synergism between NLR and diabetes in affecting prognosis. To our knowledge, no prior study has addressed the prognostic implications of NLR in diabetics vs. non-diabetics. Prior studies on the subject describe a higher prevalence of diabetics as the NLR increases;7,35 this may mean that the poorer prognosis in those groups with high NLR may actually be driven to some extent by diabetes. Also, these studies divided their cohorts only into tertiles, which may mask this U-shaped association.

In our study, diabetic subjects have a significantly elevated NLR compared with non-diabetic subjects. This is consistent with the findings of the study by Tamhane et al.7 Diabetes represents a state of altered metabolic and immunologic function, and the worse outcomes in diabetic patients post-AMI

<table>
<thead>
<tr>
<th>Quartile</th>
<th>Non-diabetic subjects</th>
<th>Diabetic subjects</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>NLR</td>
<td>White blood cell (×109/l)</td>
</tr>
<tr>
<td>Quartile 1</td>
<td>1.2±0.4</td>
<td>10.1±3.4</td>
</tr>
<tr>
<td>Quartile 2</td>
<td>2.4±0.4</td>
<td>12.4±3.7</td>
</tr>
<tr>
<td>Quartile 3</td>
<td>4.4±0.8</td>
<td>17.4±8.1</td>
</tr>
<tr>
<td>Quartile 4</td>
<td>11.5±8.0</td>
<td>26.8±3.2</td>
</tr>
</tbody>
</table>

Table 2: Selected laboratory results for each quartile in diabetic and non-diabetic subjects.
may be related to the differential contributions of neutrophils and lymphocytes to the inflammatory response in diabetic compared with non-diabetic patients. Mechanisms contributing to neutrophilia in diabetic subjects include increased plasma cortisol, leptin and insulin. Advanced glycation end products, oxygen free radicals and other cytokines may play a part in the priming of neutrophils. These activated neutrophils secrete many inflammatory mediators, contributing to increased levels of oxidative stress, inflammation, necrosis with resultant worsening prothrombotic states, endothelial dysfunction, plaque rupture and infarct size. It is also reasonable to assume that PCI or thrombolysis in the setting of increased oxidative stress places the patient at higher risk of revascularization injury. It is therefore likely that the adverse outcomes in diabetic subjects post-AMI may in part be due to increased numbers and activation of neutrophils. In our study, we found that the worse outcomes seen in diabetic patients post-AMI were independent of infarct size; the lack of correlation between NLR and infarct size is, however, a surprising finding that is not in agreement with previous studies. A possible reason for this is the heterogeneity in the timing of sample collection with respect to the index event because this is not a standardized clinical trial setting.

Figure 1. The incidence of the composite endpoint for each NLR quartile in both non-diabetic and diabetic subjects.

Figure 2. The incidence of the composite endpoint for each NLR quartile in non-diabetic subjects.
As with any novel prognostic indicator, there are many unknowns surrounding the NLR. First, the question as to what the normal reference range for NLR has not been systematically explored. The reference ranges for absolute neutrophil and lymphocyte counts are wide, making it difficult to establish a ‘normal’ NLR range for our population. The question as to whether the NLR is merely a marker of poor prognosis or whether it truly plays a substantial part in the pathogenesis of the adverse outcomes remains to be established. This question is relevant because it has implications on whether therapeutic modifications of the NLR during and after hospitalization for AMI affect outcomes. Commonly used drugs such as clopidogrel\(^{42}\) and statins\(^{43}\) have been shown to exert anti-inflammatory (including NLR-lowering) effects, but to our knowledge, there has not been a study evaluating this relationship. The short life of neutrophils in the circulation (around 7 h) and the brief steady kinetic state of neutrophils may warrant repeated measurements of the complete blood count and utilization of the mean NLR for better prognostication.\(^{44}\) However, one study has shown that there was a statistically significant correlation between long-term mortality and the initial, last, maximum and average NLR readings.\(^{9}\) Recommended cut-points for NLR are unclear as well; nonetheless our study found an optimal cut-point of 5.0 for predicting reinfarction and death, which seems to be in general agreement with prior studies (albeit with somewhat wide variations).\(^{6,7,9,10,13,14}\) This is likely due to the NLR being subject to confounding factors such as laboratory techniques and concomitant infection, making it a less meaningful clinical tool. Although the NLR has been shown to have incremental predictive value for mortality over the Global Registry of Acute Coronary Events risk profiles for in-hospital\(^{45}\) and 6-month mortality,\(^{46}\) it is perhaps premature to include it in such existing risk scoring systems. A large prospective study that uses a clinically significant NLR cut-point is required to incorporate NLR into a prognostic score.\(^{9}\) Also, the question as to why the prognostic information that the NLR carries is limited to the diabetic subgroup has yet to be explored, although there is a possibility that the raised NLR in diabetic subjects may play a role in their poor outcomes post-AMI.

Our study has several limitations. First, this is a retrospective study with use of electronic medical records which may not be standardized. However, we are careful to include consecutive patients and have supplement records with subject interviews to achieve complete datasets. Second, the observational nature, the relatively small sample size of our study and the limited follow-up period of 1-year render our study subject to selection bias and various unaccounted confounders inherent to such an analysis. Given the enhanced survival rates of AMI patients in the era of early revascularization, the 1-year follow-up period may be insufficient to determine the long-term prognostic value of NLR. Nonetheless, some of the studies on NLR have comparable or smaller sample sizes\(^{7,9}\) and follow-up period.\(^{7}\) Additionally, we were unable to compare the prognostic value of NLR with other inflammatory markers such as C-reactive protein, fibrinogen and myeloperoxidase because they were not routinely obtained as part of the AMI workup. NLR trends and averages over the hospital stay were not

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available for analysis in our study, posing a limitation in terms of reliability on an individual basis. We adjusted for five confounding factors in our multivariate model, but the limited data on other potential confounders has to be acknowledged. Second, this is a single-center study in one locality. There is a need for multiethnic population studies for longer duration to demonstrate the relationship between increased NLR and worse outcomes in diabetic patients.

**Conclusions**

Our study has shown that an increased NLR post-MI is an independent predictor of major adverse cardiac events in Southeast Asian diabetic subjects. Monitoring this easily obtainable index above and beyond usual clinical parameters could aid in the risk stratification of such patients. This is an independent predictor of major adverse cardiac events in Southeast Asian diabetic subjects. Our study has shown that an increased NLR post-MI is an independent predictor of major adverse cardiac events in diabetic patients. Need for multiethnic population studies for longer duration to demonstrate the relationship between increased NLR and worse outcomes in diabetic patients.

**Conflict of interest:** None declared.

**References**


