Original Article
Middle East Journal of Cancer; July 2018; 9(3): 223-227

A Precisely Calculated Neutrophil to Lymphocyte Ratio could Predict Overall Survival in Multiple Myeloma Patients

Hasan Jalaeikhoo*, Morteza Sharifzadeh*, Mohsen Rajaeinejad*, Manoutchehr Keyhani, Mohammad Zokaasadi*♦**

*AJA Cancer Epidemiology Research and Treatment Center (AJA- CERTC), AJA University of Medical Sciences, Tehran, Iran
**Hematology and Oncology Research Center, Vali Asr Hospital, Tehran University of Medical Sciences, Tehran, Iran

Abstract

Background: Neutrophil to lymphocyte ratio was initially used as a low cost prognostic marker in a group of solid tumors and subsequently hypothesized to have a role in multiple myeloma. This retrospective analysis aimed to report the prognostic importance of the neutrophil to lymphocyte ratio in multiple myeloma.

Methods: Between November 2003 and February 2016, we included 175 patients from two centers in this study. CBC differentials were primarily checked by a Sysmex analyzer in both centers. In one center, differentials were rechecked by light microscopy. Analysis of survival was performed using a Kaplan-Meier estimate and we assessed the effects of prognostic factors by Cox proportional hazards model.

Results: Patients had a mean age of 63.22±10.89 years. Although mean lymphocyte percent did not differ between the two centers, mean neutrophil percent and mean neutrophil to lymphocyte ratio were higher at the center that manually checked the CBC differentials. After adjustments for age and gender, we noted that the hazard ratio for elevated neutrophil to lymphocyte ratio when stratified for the centers was 1.07 (95% CI: 1.01–1.15, *P*=0.034).

Conclusion: A precisely checked neutrophil to lymphocyte ratio could act as a potentially inexpensive, accessible prognostic factor for multiple myeloma patients.

Keywords: Multiple myeloma, Neutrophil to lymphocyte ratio, Survival analysis, Prognosis

♦Corresponding Author:
Mohammad Zokaasadi, MD
AJA Cancer Epidemiology Research and Treatment Center (AJA-CERTC), AJA University of Medical Sciences, West Fatemi Avenue, Shahid Etemadzadeh Street, 501 Army Hospital, Tehran, Iran
Tel/Fax: +982143823545
Email: zokaasadi2@gmail.com

Introduction

Multiple myeloma (MM) is one of the most common hematologic disorders that arise from malignant plasma cells. A myriad of factors that include cytogenetic, serologic, immunologic, and molecular markers have been utilized to predict the prognosis of this disease.[1] The neutrophil to lymphocyte ratio (NLR)

Received: September 03, 2017; Accepted: October 28, 2017

is one of the recent, simple, feasible evaluation methods.[2] This index has been initially used in diverse solid tumors - gastrointestinal, genitourinary, and other groups of malignancies.[3,4] It has been postulated to play a role in MM.[5] The role of NLR in the prognosis of MM in hematological oncology is not clear. However, most evidences favor the prognostic significance of NLR,[2,5-8] despite opposing opinions.[9]

We designed the current study to evaluate the role of NLR in prognosis of newly diagnosed MM patients from two institutions.

Materials and Methods

We retrospectively assessed the medical records of 175 MM patients from two hospitals, Arad and 501, between November 2003 and February 2016. Diagnosis of MM was confirmed based on the presence of at least 10% plasma cells in bone marrow biopsies and one of the following disorders (CRAB): hypercalcemia, renal failure, anemia, or lytic bone lesions. Persistence of plasmacytoma and one of the CRAB disorders was enough to confirm a diagnosis of MM. In Arad Hospital, analysis of CBC elements and differentials was initially done by a Sysmex analyzer and rechecked by a laboratory technician. In 501 Hospital, differentials were only analyzed by the Sysmex analyzer. We extracted all demographics, laboratory values, and other relevant variables from the patients' medical files. Survival status of the patients was obtained from records of the follow-up clinic. In cases where patients were lost to follow-up, we obtained the information by telephone contact or speaking with their family members. The patients provided informed consent for use of their medical records. The Ethical Committee of AJA University of Medical Sciences approved the study protocol.

Statistical analysis

Categorical variables were described as frequencies and percentages. Continuous values were expressed as means and standard deviations. We used the t-test to compare means. The overall survival (OS) rate was calculated using the Kaplan-Meier estimate. We defined OS as the time from diagnosis to death from any cause. Median follow-up time was obtained by reverse Kaplan-Meier. The effect of possible factors on outcome was primarily analyzed by univariate analysis. After selection of applicable factors, we used multivariate analysis to calculate the hazard ratio (HR) and significance of relevant factors. No cut-off level for NLR has been suggested. Previous studies used a wide spectrum that ranged from <2 to ≥5 as the cut-off values; therefore, we considered NLR as a continuous variable in the hazards model and did not label it as high and low. All analyses were done in SPSS software for windows version 23. *P*-values <0.05 were considered statistically significant

Results

This study included 175 patients with a mean age of 63.22±10.89 years. There were 119 (68%) males and 56 (32%) females. The mean NLR was 2.80±2.91. Table 1 summarizes the basic characteristics of the study patients.

A comparison of means between the two centers showed significant differences in neutrophil percent, NLR, creatinine, ESR, and hemoglobin. There were no differences in age, WBC, lymphocyte percent, platelets and calcium levels between the two centers.

Cases at 501 Hospital had more severe features. The mean NLR, a probable prognostic factor for poorer outcome, was lower in 501 Hospital patients. Both centers utilized the same set of instruments. However, in Arad Hospital, a skilled technician rechecked differentials of all cases. Therefore, NLRs at Arad hospital were more accurate.

Median follow-up time was 45 months. Survival analysis showed the following OS for patients at one (76.60%), three (53.8%), and five (37.8%) years.

The HR for NLR in univariate analysis for all cases combined was 1.059 (95% CI=0.995-1.126, *P*=0.070). The HR for NLR in univariate analysis was 1.274 (95% CI=0.964-1.684, *P*=0.089) for 501 Hospital and 1.081 (95% CI=1.011-1.156,

Table 1. Basic characteristics of the patients in both centers.

Parameter	Mean±SD		*P*-value
	Arad Hospital	501 Hospital	
Age (years)	63.52±9.94	62.71±12.37	0.652
WBC	6.47±2.89	6.15±2.45	0.448
Lymphocyte (%)	30.52±13.34	33.74±12.39	0.114
Neutrophil (%)	64.32±14.69	54.97±13.36	<0.001
Neutrophil to lymphocyte ratio (NLR)	3.21±3.36	2.00±1.42	0.002
Creatinine	1.67±1.19	2.88±4.39	0.032
Hb	9.99±2.03	9.08±2.41	0.008
Platelets	195.32±81.82	185.05±89.89	0.439
Calcium	9.78±1.61	9.63±1.47	0.584
ESR	91.08±40.03	109.65±41.79	0.011

P=0.023) for Arad Hospital. The HR for NLR for all cases after stratification for the method of analysis was 1.090 (95% CI=1.022-1.161, *P*=0.008). Multivariate analysis showed that the HR for NLR after adjustments for age and gender was 1.07 (95% CI: 1.01-1.15, *P*=0.034). Table 2 shows the results of multivariate analysis for NLR on OS.

Discussion

This study primarily aimed to evaluate the role of NLR as a feasible, economical index in the prognosis of MM. Results of our multivariate analysis revealed an HR of 1.07 for NLR which meant that for each unit increase in NLR as a predictor of prognosis, the patients would face a 7% increase in hazard. We concluded that NLR acted as an independent, appropriate prognostic factor. In the current study, we analyzed NLR as a continuous variable; the same statistical technique was performed in previous studies.[10-13]

Numerous markers are used in this field. However, most are expensive compared to NLR. A brief explanation of the immunology of MM would clarify the rationale behind this index. Studies on alterations of the immune system in MM have suggested that T regulatory cells (Treg), which normally work as self-tolerance inducers, increase in MM patients. Therefore an anti-myeloma immunity may be one of the disturbances in MM patients.[14] A clinical study has reported that MM patients who survived for more than 10 years had lower Treg cells compared to patients with shorter survival. There was significantly greater clonal expansion of cytotoxic T cells in long-term survivors.[15] Hence, the study concluded that long-term survival in MM was linked to specific immunologic features which led to a more normal anti-tumor immune response rather than myeloma induced immunosuppression and tolerance. A number of studies observed the presence of a relatively lower lymphocyte count in MM patients compared to healthy controls.[5] Possibly the presence and severity of lymphopenia, which lead to escalation of NLR, could reflect disease prognosis, an idea supported by the majority of former studies. Wongrakpanich et al. conducted a retrospective analysis of 175 MM patients. They reported that higher NLR (study cut-off: 2.78) was associated with an inferior median OS time. The median OS time in high NLR groups was 45 months versus 62 months in the low NLR group (*P*=0.077). Multivariate analysis showed a HR of 2.89 (95% CI: 1.31-6.40) for high NLR after adjustments for type of immunoglobulin, stem cell transplantation, and beta-2 microglobulin.[2] Shi et al., in a controlled study of 559 MM patients and 123 healthy individuals, reported comparable results. They found an association between higher NLR and poorer outcome regarding OS and progression-free survival (PFS). Median OS was 43.2 months, whereas PFS was 24.03 months for the high NLR group (NLR>4 was considered high). The low NLR group had an OS of 56.0 months and PFS of 37.46 months. The differences

Table 2. Results of stratified univariate and multivariate analyses.

Covariate	Univariate		Multivariate	
	HR (95% CI)	*P*-value	HR (95% CI)	*P*-value
Age (years)	1.02 (1.00–1.05)	0.016	1.17 (1.00–1.04)	0.112
Gender (F/M)	0.72 (0.47–1.11)	0.135	0.75 (0.48–1.18)	0.207
NLR	1.09 (1.02–1.16)	0.008	1.07 (1.01–1.15)	0.034*

HR: Hazard ratio; NLR: Neutrophil to lymphocyte ratio.

were statistically significant. The study also found out that in comparison to healthy individuals MM patients had relative lymphopenia. The absolute lymphocyte count in the MM group was 1.677±0.03167 versus 2.104±0.05161 for the healthy group (P<0.0001).[5] Romano et al., in a study on 309 newly diagnosed MM patients treated with novel agents from the time of diagnosis, showed that higher NLR had an association with poorer OS and PFS. The predictive value was independent of disease stage based on an international staging system (ISS). They selected a cut-off of 2 for categorizing NLR into high and low. That study showed that in a subgroup with ISS stage I and NLR<2 the 5-year OS was markedly good and approached 90%.[8] The study of Onec et al. on 52 MM patients in a single center discovered that higher NLR (>1.72) had a significant link to worse performance status, disease stage, and poorer OS. In that study multivariate analysis did not attain a statistical level of significance.[7] A controlled study conducted by Kelkitli et al. on 151 patients and the same number of matched healthy individuals reported the same results. A higher NLR (>2) had a significant association with poorer OS and event-free survival (EFS). The relation remained significant in univariate and multivariate analyses. Additionally the MM patients evidently had higher NLR (2.79±1.82) than the control group (1.9±0.61, P<0.0001).[6] Li et al. investigated 315 MM patients and found that higher NLR (≥2) led to inferior OS and PFS independent of age, disease stage and serum creatinine, calcium and LDH levels. The 95% CI for HR of NLR for OS was 1.045-1.131 (P<0.001); for PFS, the 95% CI was 1.032-1.118 (P<0.001).16 Although there are disagreements in the literature, a study by Azab et al. on 96 patients did not show any prognostic significance for NLR - as a continuous variable nor in each one of the NLR quarterlies.[9] That study was conducted on a relatively smaller sample size compared to others. Therefore, lack of statistical significance might just simply arise from lack of adequate power.

The analyzing method had a significant influence on NLR. Differences between the results of studies could be related to different methods of evaluation. The use of precise methods or stratification of NLR values based on methods of analysis would increase accuracy of the assessments. Elevated NLR is associated with an inferior OS in MM patients. Therefore, NLR could be used as an inexpensive prognostic factor in MM patients.

The main limitation of this study was its retrospective nature which made it difficult to reach a stronger interpretation of the findings. Because of cost considerations, a group of patients were not able to undergo more accurate, expensive evaluations at the time of diagnosis, such as β2-microglobulin. Therefore we could not calculate and compare our results with standard prognostic staging methods such as ISS. The latter limitation also ironically acted as a potential motivation for conducting this types of research. Studies on both NLR and the ISS system suggested an independent prognostic value for NLR.[8]

Hence, NLR could be used as one of the available prognostic indices in MM patients. Differences between techniques of evaluation of CBC differentials should be noted in order to reach an accurate judgment.

Acknowledgement

The authors would like to sincerely thank Mr. Kalahroodi and Mr. Moradi at 501 Hospital and Ms. Mirakhori and Ms. Aarianfar at Arad Hospital.

Conflict of Interest

No conflict of interest is declared.

References

1. Anderson KC. Progress and paradigms in multiple myeloma. *Clin Cancer Res.* 2016 ;22(22):5419-27.
2. Wongrakpanich S, George G, Chaiwatcharayut W, Biso S, Candelario N, Mittal V, et al. The prognostic significance of neutrophil-to-lymphocyte and platelet-to-lymphocyte ratios in patients with multiple myeloma. *J Clin Lab Anal.* 2016;30(6):1208-13.
3. Mei Z, Shi L, Wang B, Yang J, Xiao Z, Du P, et al. Prognostic role of pretreatment blood neutrophil-to-lymphocyte ratio in advanced cancer survivors: A systematic review and meta-analysis of 66 cohort studies. *Cancer Treat Rev.* 2017;58:1-13.
4. T Templeton AJ, McNamara MG, Šeruga B, Vera-Badillo FE, Aneja P, Ocaña A, et al. Prognostic role of neutrophil-to-lymphocyte ratio in solid tumors: a systematic review and meta-analysis. *J Natl Cancer Inst.* 2014;106(6):dju124.
5. Shi L, Qin X, Wang H, Xia Y, Li Y, Chen X, et al. Elevated neutrophil-to-lymphocyte ratio and monocyte-to-lymphocyte ratio and decreased platelet-to-lymphocyte ratio are associated with poor prognosis in multiple myeloma. *Oncotarget.* 2017;8(12):18792-801.
6. Kelkitli E, Atay H, Cilingir F, Güler N, Terzi Y, Ozatlı D, et al. Predicting survival for multiple myeloma patients using baseline neutrophil/lymphocyte ratio. *Ann Hematol.* 2014;93(5):841-6.
7. Onec B, Okutan H, Albayrak M, Saribacak Can E, Aslan V, Unver Koluman B, et al. The predictive role of the neutrophil/lymphocyte ratio in survival with multiple myeloma: A single center experience. *J Clin Lab Anal.* 2017;31(2).
8. Romano A, Parrinello NL, Consoli ML, Marchionni L, Forte S, Conticello C, et al. Neutrophil to lymphocyte ratio (NLR) improves the risk assessment of ISS staging in newly diagnosed MM patients treated upfront with novel agents. *Ann Hematol.* 2015;94(11):1875-83.
9. Atallah JP, Azab B, Vivekanandarajah A, Naboush A, Sater HA, Farhan M, et al. Elevated neutrophil: lymphocyte ratio does not predict survival in multiple myeloma patients. *Blood*. 2013;122(21):5336.
10. Lee Y, Kim SH, Han JY, Kim HT, Yun T, Lee JS. Early neutrophil-to-lymphocyte ratio reduction as a surrogate marker of prognosis in never smokers with advanced lung adenocarcinoma receiving gefitinib or standard chemotherapy as first-line therapy. *J Cancer Res Clin Oncol.* 2012;138(12):2009-16.
11. Lee YY, Choi CH, Kim HJ, Kim TJ, Lee JW, Lee JH, et al. Pretreatment neutrophil:lymphocyte ratio as a prognostic factor in cervical carcinoma. *Anticancer Res.* 2012;32(4):1555-61.
12. Smith RA, Bosonnet L, Raraty M, Sutton R, Neoptolemos JP, Campbell F, et al. Preoperative platelet-lymphocyte ratio is an independent significant prognostic marker in resected pancreatic ductal adenocarcinoma. *Am J Surg.* 2009;197(4):466-72.
13. Sarraf KM, Belcher E, Raevsky E, Nicholson AG, Goldstraw P, Lim E. Neutrophil/lymphocyte ratio and its association with survival after complete resection in non-small cell lung cancer. *J Thorac Cardiovasc Surg*. 2009;137(2):425-8.
14. Braga WM, Atanackovic D, Colleoni GW. The role of regulatory T cells and TH17 cells in multiple myeloma. *Clin Dev Immunol.* 2012;2012:293479.
15. Bryant C, Suen H, Brown R, Yang S, Favaloro J, Aklilu E, et al. Long-term survival in multiple myeloma is associated with a distinct immunological profile, which includes proliferative cytotoxic T-cell clones and a favourable Treg/Th17 balance. *Blood Cancer J.* 2013;3:e148.
16. Li Y, Li H, Li W, Wang L, Yan Z, Yao Y, et al. Pretreatment neutrophil/lymphocyte ratio but not platelet/lymphocyte ratio has a prognostic impact in multiple myeloma. *J Clin Lab Anal.* 2017;31(5).