Original Article

The Predictive Role of Neutrophil to Lymphocyte ratio in Chronic Obstructive Pulmonary Disease

Bulent Bilir[1], Nejat Altıntaş[2], Murat Aydın[3], Mustafa Oran[1], Savas Özsu[4], Ümit Tutar[5]

ABSTRACT

Objective: Recently neutrophil-to-lymphocyte ratio (NLR) -the level of neutrophil reflecting the severity of inflammation and lymphocyte occurring after physiological stress has been gaining popularity, which was, along with other inflammatory markers, commonly accepted as an accurate marker of the inflammatory status.In this multi-centered study, an early, rapid, and low-cost diagnosis method was investigated. To this end, the correlation between chronic obstructive pulmonary disease and inflammation was planned to be utilized and whether neutrophil-to-lymphocyte ratio can be used as a valid tool in the diagnosis of acute exacerbations of chronic obstructive pulmonary disease was investigated. *Method:* We retrospectively enrolled the 467 patients. Control group included sex and age-matched healthy people.C-Reactive protein, forced expiratory volume-1, forced vital capacity, complete blood count and clinical data A receiver-operating characteristic curve analysis was performed to determine the best cut-off value of N/L ratio and C-Reaktif protein to predict the exacerbation. Two-sided p values <0.05 were considered statistically significant. *Results:* By spearman analysis, there was a strong correlation between.C-Reactive protein and N/L ratio in both stable group (r=0.436, p<0.001) and exacerbation group (r=0.534, p=0.001). *Conclusion:* Neutrophil/lymphocyte ratio may bea useful predictor of inflammation in chronic obstructive pulmonary disease and acute exacerbation of chronic obstructive pulmonary disease patients.

Key words: Acute exacerbation, chronic obstructive pulmonary disease, neutrophil-to-lymphocyte ratio.

Kronik Obstrüktif Akciğer Hastalığında Nötrofil-Lenfosit Oranının Öngörücü Rolü

ÖZET

Son zamanlarda "nötrofil-lenfosit oranı (NLR)" denen, inflamasyon ciddiyetini yansıtan nötrofil ile fizyolojik strese bağlı olarak değişen lenfositin oranlanmasından elde edilen belirteç popülerlik kazanmakta olup diğer inflamatuvar belirteçlerle birlikte inflamatuvar durumun gerçek bir göstergesi olarak kabul edilir. Bu çok merkezli çalışmada, erken, hızlı, düşük maliyetli bir teşhis yöntemi araştırıldı. Kronik obstrüktif akciğer hastalığı ile inflamasyon arasındaki korelasyonun kullanımı planlandı ve kronik obstrüktif akciğer hastalığının akut alevlenmelerinin teşhisinde nötrofil-lenfosit oranının doğru bir araç olarak kullanılabilirliği araştırıldı. *Yöntem:* Retrospektif olarak 467 vaka alındı. Sağlıklı kontrol grubu yaş ve cinsiyet açısından benzerdi. C-reaktif protein, forse ekspiratuvar volüm-1, forse vital kapasite ve tam kan sayımı yapıldı. Nötrofil-lenfosit oranı ve C-reaktif proteinin alevlenmeyi gösterebilecek en iyi sınır değerini bulmak üzere işlem karakteristiği (ROC) eğrisi analizi yapıldı. *Bulgular:* İki-taraflı p değerinin 0,05'in altında olması istatistiki olarak anlamlı kabul edildi. Spearman analizi sonucunda, C-reaktif protein ile nötrofil-lenfosit oranı arasında hem stabil (r=0.436, p<0.001) hem de alevlenme (r=0.534, p=0.001) grubunda kuvvetli bir ilişki bulundu. *Sonuç:* Nötrofil-lenfosit oranı, kronik obstrüktif akciğer hastalığı vakalarında inflamasyonun ve akut alevlenmenin iyi bir göstergesi olabilir.

Anahtar kelimeler: Akut alevlenme, kronik obstrüktif akciğer hastalığı, nötrofil lenfosit oranı.

Namik Kemal University School of Medicine, Department of Internal Medicine[1], Pulmonary[2] and Clinical Biochemistry[3] Tekirdağ, Karadeniz Technical University School of Medicine, Department of Pulmonary Disease[4], Trabzon, Republic of Turkey Ministry of Health, State of Pulmonary Disease[5] of Hospital, Samsun, Turkey.

Received: 01.07.2015, Accepted: 22.07.2015

Correspondence: Bulent Bilir
Namik Kemal University School of Medicine, Department of Internal Medicine, Tekirdağ, Turkey

E-mail: bulentbilir@yahoo.com

INTRODUCTION

Chronic obstructive pulmonary disease (COPD) is a chronic and progressive inflammatory disease affecting respiratory system, which is accompanied by acute exacerbations (AE). It is characterized by progressive airflow limitation that is not fully reversible. Inhaled particles and gases such as tobacco smoke lead to inflammatory response in the lungs and airways (1,2).

In elderly population 10% is affected world wide (2).World Health Organization describes COPD as a leading cause of mortality and morbidity in elderly population (3,4). Since the disease is complicated with adverse outcomes, high cost, it is critical to treat the disease by preventing acute exacerbation and controlling the inflammation.

Various inflammatory markers such as C-reactive protein (CRP), and white blood cell (WBC) count increase in COPD (5). Additionally, elevated inflammatory markers have longly been used to determine the acute exacerbations of COPD (AECOPD) and prognosis. Neutrophil-lymhocyte ratio (NLR) is a novel inflammatory biomarker which indicates the balance of the immune system and reflecting systemic inflammation. Morever, NLR has widely been studied in various disorders complicated with acute and chronic inflammation, located in US National Library of Medicine National Institutes of Health (PUBMED). Neverthless, the association of this biomarker, with COPD and AECOPD is almost not evaluated.

In this multi-centered study, evaluation of NLRas an early, rapid, and low-cost diagnostic methodin COPD and AECOPD was aimed.

MATERIAL AND METHODS

This multicenter study involved 467 patients withCOPD or AECOPD between June 2013 and February 2014. The patients' demographic and clinical data were retrieved from the hospitals' electronic database and spirometry measurement results listed in the form. CRP, complete blood count (CBC), and clinical data of the patient and control group were recorded in this form. Control group included 215 age and gender matched healthy subjects, who admitted to the hospital for checkup. CBC of 215 sex-, age- and BMI- matched healthy control subjects were evaluated. Diagnosis of COPD was based on Global Initiative for Chronic Obstructive Lung Disease (GOLD) criteria for the diagnosis, classification, and severity of COPD. At the same time NLR and CRP levels were classified according to COPD GOLD Stage (6).

Ethical committee

The study complied with the Declaration of Helsinki and was approved by the ethics committee of the Republic of Turkey Ministry of Health Okmeydani Training and Research Hospital (Date: 27/03/2014-Number: 5559).
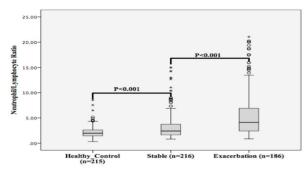
Eligible Criteria

This multicenter cross-sectional retrospective study was conducted in the Internal Medicine Clinics and Pulmonary Disease Clinics. Medical records of 467 patients admitted to these centers between June 2013 and february 2014 were analyzed.patients withother coexisting pulmonary diseases, with any acute inflammatory disease (n:20) other than AECOPD, with any chronic inflammatory disorder (n:22) except COPD, malignity (n:3) and insufficient records (n:20) were excluded. Finally, 402 patients enrolled.

Study design

Patients were divided into two subgoups due to their hospital records stable COPD and AECOPD. Stable COPD was described as having no change in sympthoms as well as no addditional medical treatment or extra doses of daily inhaler treatment for consecutive three months (7). Acute exacarbation of COPD was described as a sudden worsening of COPD symptoms of shortness of breath,increasedquantity and augmentation of sputum which is prolonged more than 48 hours (h) and raised use of maintenance medications and/or need for additional treatment (8). Subsequently, both groups were staged according to the severity criteria of GOLD (6); stage I [FEV1≥80%,], stageII [50%≤FEV1<80%], stage III [30%≤FEV1<50 %] and stage IV [FEV1<30 %].
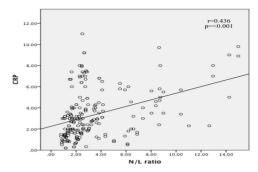
Statistical analysis

Descriptive statistics were computed for all factors. Continuous variables were presented as mean (standard deviation) or median (25th, 75th percentiles) and categorical variables as frequency (percentage). For comparisons among groups, the chi-square test (or Fisher's exact test when any expected cell count was <5 for a 2x2 table) was used for categorical variables and the unpaired Student's t test or the Mann–Whitney rank sum test for continuous variables after testing for normality which (was performed using histograms and the Shapiro-Wilk test).

Figure 1. A graph of neutrophil-to-lymphocyte ratio in chronic obstructive pulmonary disease groups and healthy controls. Data are expressed as box plots, in which the horizontal lines illustrate the 25th, 50th and 75th percentiles of the values of Neutrophil-to-Lymphocyte ratios.
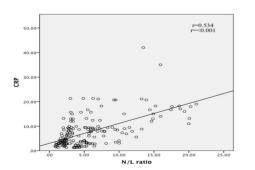

Figure 2. Correlation between C-reactive protein and neutrophil-to-lymphocyte ratio in chronic obstructive pulmonary disease patients with stable period.

One way analysis of variance (ANOVA) with Bonferroni adjustments were used to compare more than two groups, if tests of normality were met and the Kruskal-Wallis test when tests of normality failed. Bivariate relationships between variables were determined by Pearson's or Spearman rank correlation coefficients (rho). Receiver operating characteristics curve (ROC) analysis was used to evaluate the role of NLR and CRP in distinguishing subjects with exacerbation. Cut-off values that maximized both sensitivity and specificity were chosen. Two-sided p values <0.05 were considered statistically significant. All statistical analyses were performed using the Statistical Package for the Social Sciences for Macintoch version 20.0 (SPSS Inc., Chicago, IL, USA; serial number = 10229569).
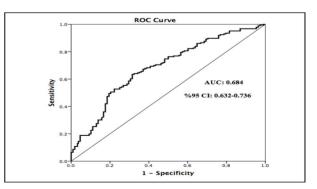
RESULTS

The hospital records of 467 patients COPD patients with a stable period and acute exacerbation (216 and 186 patients, respectively) and leukocyte profiles of 215 sex, age and BMI matched healthy control subjects were evaluated. The characteristics and laboratory findings of involved subjects in three groups and PFTs of COPD groups are outlined in Table 1. There was no statistical difference between the groups with respect to age and sex. The current findings indicated that the NLR in COPD patients with exacerbation was significantly higher compared with stable period and healthy controls (p<0.001). The NLR was higher in COPD patients with stable period than healthy control (p<0.001) (Figure 1). CRP levels were significantly different in three groups (p<0.001). CRP levels had a tendency to increase with exacerbation. Pulmonary function test was not different

Figure 3. Correlation between C-reactive protein and neutrophil-to-lymphocyte ratio in chronic obstructive pulmonary disease patients with exacerbation.

Figure 4. Receiver Operating Curve for neutrophil-to-lymphocyte ratio.

Table 1. Clinical characteristic of studied subjects.

	Control (n:215)	COPD (Stable) (n:216)	COPD (Exacerbation) (n:186)	p value[§]
Mean age, yr (SD)	54.6 (13.8)	53.2 (13.2)	56.9 (14.9)	0.178[†]
Male n (%)	181(84.2%)	175(81%)	163(87.6%)	0.194[‡]
Laboratory finding				
Leukocyte, ×103 µl	7.0(1.9) [a,b]	7.6(2.0)[a,c]	8.3(1.9)[b,c]	<0.001*
Neutrophil, ×103 µl	4.1(1.4)[a,b]	4.7(1.9)[a,c]	5.7(2.0)[b,c]	<0.001*
Lymphocyte, ×103 µl	2.2(0.8)[a,b]	1.9(0.8)[a,c]	1.4(0.8)[b,c]	<0.001*
NLR	1.9(1.1)[a,b]	2.41(2.9)[a,c]	4.1(4.6)[b,c]	<0.001*
Haemoglobin, g/dl	13.4(0.9)	13.2(2.1)	13.1(2.3)	0.922*
Platelets, ×103 µl	242.3(57.9)	234.63(80.0)	244.5(96.8)	0.163*
CRP, mg/dl	1.7(1.1)[a,b]	3.3(2.3)[a,c]	7.6(6.2)[b,c]	P<0.001*
FEV$_1$	NA	54.1(8.43)	52.0(9.74)	0.656[¥]
FEV$_1$/FVC	NA	60.1 (7.7)	58.7 (7.5)	0.355[¥]

CRP; C-reactive protein, FEV1 forced expiratory volume in the first second, FVC forced vital capacity, MPV mean platelet volume, NLR; neutrophil-to- lymphocyte ratio, Data are Median (interquartile range) unless otherwise indicated. [§] As compared among the three groups; Where the P value is significant, values within a row with the same superscript letter are significantly different.
[†]One-way analysis of variance (ANOVA), [‡]Chi-square test, * Kruskal-Wallis test with Benforoni correction, [¥] Student T test.

in COPD groups (Table 1). NLR and CRP levels were classified according to COPD GOLD stage in Table 2. There was a linear increase in NLR with COPD severity in stable group, however this increase was not linear in patients with exacerbation, in a way that NLR tended to be high in stage-I patients even this elevation was not statistically significant and   NLR was higher in stage 4 patients than stage-2 and -3 patient. By spearman correlation, there was a strong correlation between CRP and NLR in both stable group (r=0.436, p<0.001) (Figure 2) and exacerbation group (r=0.534, p=0.001) (Figure 3).

Diagnostic characteristics of NLR and CRP for prediction of Exacerbation: A receiver-operating characteristic curve (ROC) analysis was pe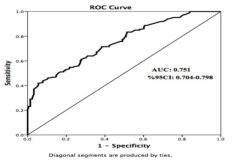rformed to determine the best cut-off value of NLR and CRP to predict the exacerbation. The best cutoff for NLR was taken as 3.35. The area under the curve (AUC) was 0.684 (95% CI 0.632 to 0.736, p<0.001) and the sensitivity, specificity, positive predictive value, negative predictive value, and diagnostic accuracy were 69%, 59%, 59%, 69%, and 64%, respectively (Figure 4).

The best cutoff for CRP was taken as 3.35. The area under the curve (AUC) was 0.751 (95% CI 0.704 to 0.798, p<0.001) and the sensitivity, specificity, positive predictive value, negative predictive value, and diagnostic accuracy were 68%, 62%, 60%, 69%, and 65%, respectively (Figure 5).

DISCUSSION

COPD is a progressive disease which is complicated with acute exececrbations. It has longly been a prevelant health problem as well as economical burden.The economical burden, the prevalance of COPD and AECOPD are expected to raise in the future, as a result of increasing survival age and exposure to the risk factors such as tobacco smoking, occupational and air pollution, wood and other biomass fuels (4-9). AECOPD is a major factor of increasing costs in COPD (10), and are an important COPD-related mortality (11). Although few biomarkers have been widely used in the clinical practice, many efforts have been made to

Figure 5. Receiver operating curve for C-reactive protein.

Table 2. Neutrophil-lymhocyte ratio and C- reactive protein levels during stable and exacerbation period, according to Global Initiative for Chronic Obstructive Lung Disease Stage.

Parameters	Group	Stage I	Stage II	Stage III	Stage IV	p value
NLR	Stable	1.64(1.60)[a]	2.33(2.09)[b]	2.60(3.58)[a]	2.66(3.86)[a,b]	<0.001*
	Exacerbation	6.19(2.94)	4.18(4.48)[a]	3.47(3.53)[b]	6.29(6.38)[a,b]	0.028*
CRP	Stable	2.20(2.38)	2.94 (2.48)	2.70 (3.20)	3.7 (4.4)	0.053*
	Exacerbation	6.90 (8.51)	4.00 (5.87) [a]	6.00 (6.00)[b]	9.92 (9.48) [a,b]	0.01*

NLR: Neutrophil-to-lymphocyte ratio; CRP: C-reactive protein. Data are Median (interquartile range) unless otherwise indicated. As compared among the four groups; Where the P value is significant, values within a row with the same superscript letter are significantly different. * Kruskal-Wallis test with Benforoni correction.

explore new biomarkers that have shown promise as indicators of disease severity, acute execerbations of the disease as well as treatment response (12). Recently, it has been suggested that NLR could be used as a marker of inflammation in patients with Familial Mediterranean Fever, cancer, cardiovacular diseases, kidney diseases, ulcerative colitis (13-17). Taylan et al. (18); evaluate the NLR in 100 COPD patients. They investigate the data of the patients in acute exacerbation period retrospectively and than they repeted the measurements three mounts later in the stable period.

They found a significant corelation of NLR with CRP (r= 0.415, p<0.001), WBC (r= 0.304, p= 0.002) and ESR (r= 0.275, p= 0.035). For an NLR cutoff of 3.29, sensitivity of detecting AECOPD was 80.8% and specificity was 77.7%. As a conclusion they suggested that NLR can be used as a marker like WBC, ESR, CRP in the AECOPD fort he early diagnose also who have normal levels of tradiotional markers.

Although there has been many studies evaluating the usefulness of this marker in some disorders that is complicated by systemic inflammation, at the best of our knowledge there is only one study evaluating the NLR in patients with COPD. In the study of Günay et al. (19); it has been demonstrated that there had been a significant increase in NLR in stable COPD patients compared to healthy controls. Additionally, a further increase in NLR was observed in AECOPD compared to stable period.

In the present study a larger number of COPD patients and control subjects were included. Similar to findings of Günay et al. NLR found to be higher in COPD and AECOPD groups compared to control groups. Different from their study desingn, COPD and AECOPD patients were classified according to the GOLD criteria. A linear increase was

observed in NLR with COPD severity, which may show the relationship between inflammation and increased NLR, in stable group.However, increase in NLR was not linear in AECOPD. Although NLR was suggested to be used in determination of inflammation, however a cutoff level was not determined in COPD patients.Our findings demonstrated that NLR higher than 3.35 would be a good predictor of inflammation. NLR may be a useful predictor of inflammation in COPD patients. Prospective studies would be useful to assess the predictive value of this marker for determination of inflammation.

REFERENCES

1. Salvi SS, Barnes PJ. Chronic obstructive pulmonary disease in non-smokers. Lancet 2009;374:733-43.

2. Hogg JC, Chu F, Utokaparch S, Woods R, Elliott WM, Buzatu Let al. The nature of small airway obstruction in chronic obstructive pulmonary disease. N Engl J Med 2004;350:2645-53.

3. Halbert RJ, Natoli JL, Gano A, Badamgarav E, Buist AS, Mannino DM.Global burden of COPD: systematic review and meta-analysis. Eur Respir J2006;28:523-32.

4. Mathers C, Boerma T, Fat DM. The Global Burden of Disease: 2004 Update. Geneva: World Health Organization; 2008. [Accessed 15.06.2015]. Available from:http://www.who.int/healthinfo/global_burden_disease/GBD_report_2004update_full.pdf.

5. Bestall JC, Paul EA, Garrod R, Garnham R, Jones PW, Wedzicha JA.Usefulness of the Medical Research Council (MRC) dyspnoea scale as a measure of disability in patients with chronic obstructive pulmonary disease. Thorax 1999;54:581-6.

6. Global Initiative for Chronic Obstructive Lung Disease The Global Strategy for the Diagnosis, Management, and Prevention of Chronic Obstructive Pulmonary Disease.2013. [Accessed April 4, 2015]. Available from: http://www.gold-copd.org/uploads/users/files/GOLD_Report_2013_Feb20.pdf.

7. Celli BR, MacNee W, ATS/ERS Task Force. Standards for the diagnosis and treatment of patients with COPD: A summary of the ATS/ERS position paper. Eur Respir J 2004;23:932-46.

8. Anthonisen NR, Manfreda J, Warren CP, Hershfield ES, Harding GK, Nelson NA. Antibiotic therapy in exacerbationsof chronic obstructive pulmonary disease. Ann Int Med 1987;106:196-204.

9. Mathers CD, Loncar D. Projections of global mortalityand burden of disease from 2002 to 2030. PLoS Med 2006;3: e442.

10. Hertel N, Kotchie RW, Samyshkin Y, Radford M, Humphreys S, Jameson K. Cost-effectiveness of available treattreatment options for patients suffering from severe COPD in the UK: a fully incremental analysis. Int J Chron Obstruct Pulmon Dis 2012;7:183-99.

11. Soler-Cataluña JJ, Martínez-García MA, Román Sánchez P, Salcedo E, Navarro M, Ochando R.Severe acute exacerbations and mortality in patients with chronic obstructive pulmonary disease. Thorax 2005;60:925-31.

12. Leung JM, Sin DD. Biomarkers in airway diseases. Can Respir J 2013;20:180-2.

13. Ahsen A, Ulu MS, Yuksel S, et al. As a new inflammatory marker for familial Mediterranean fever: neutrophil-to-lymphocyte ratio. Inflammation 2013;36:1357-62.

14. Guthrie GJ, Charles KA, Roxburgh CS, Horgan PG, McMillan DC, Clarke SJ.The systemic inflammation-based neutrophil-lymphocyte ratio: experience in patients with cancer. Crit Rev Oncol Hematol 2013;88:218-30.

15. Bhat T, Teli S, Rijal J, et al.Neutrophil to lymphocyte ratio and cardiovascular diseases: a review.Expert Rev Cardiovasc Ther 2013;11:55-9.

16. Turkmen K, Erdur FM, Ozcicek A, et al. Platelet-to-lymphocyte ratio better predicts inflammation than neutrophil-to-lymphocyte ratio in end-stage renal disease patients. Hemodial Int 2013;17:391-6.

17. Celikbilek M, Dogan S, Ozbakir O, et al. Neutrophil-lymphocyte ratio as a predictor of disease severity in ulcerative colitis. J Clin Lab Anal 2013;27:72-6.

18. Taylan M, Demir M, Kaya H, et al. Alterations of the neutrophil-lymphocyte ratio during the period of stable and acute exacerbation of chronic obstructive pulmonary disease patients. Clin Respir J 2015;doi:10.1111/crj.12336.

19. Günay E, Sarınç US, Akar O, et al. Neutrophil to lymphocyte ratio in chronic obstructive pulmonary disease:a retrospective study. Inflammation 2014;37:374-80.