

The relationship between fibrosis level and blood neutrophil to lymphocyte ratio in chronic HBV patients

Hepatit B hastalarında nötrofil/lenfosit oranı ile fibrosis arasındaki ilişki

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Background and Aims: In recent years, the neutrophil to lymphocyte ratio has been popular as a simple, inexpensive and effective marker associated with various inflammatory and neoplastic diseases. This marker has not yet been investigated in chronic hepatitis B patients. In our study with hepatitis B patients, we aimed to investigate the relationship between fibrosis levels and the neutrophil to lymphocyte ratio for determining the disease severity and making treatment decisions using HBV-DNA and liver biopsy results. **Materials and Methods:** 486 hepatitis B patients subjected to liver biopsy between 2005 and 2010 were screened retrospectively from patient charts. Only 268 naive patients fit the criteria for this study. Naive patients were included while patients treated with antiviral drugs and with a positive anti-Delta result were excluded from the study. **Results:** The study consisted of 268 (age: 38±13 years) patients with liver biopsies [163 (60.8%) males (age: 38±13), 105 (39.2%) females (age: 37±14)]. Fibrosis score distribution of the patients was as follows: 0/6: 122 patients (45.5%), 1/6: 50 patients (18.7%), 2/6: 37 patients (13.8%), 3/6: 25 patients (9.3%), 4/6: 5 patients (1.9%), 5/6: 6 patients (2.2%), and 6/6: 23 patients (8.6%). There was a significant correlation between fibrosis score and histological activity index ($r: +0.478$ $p: 0.000$), while HBV-DNA levels showed no significant correlation ($r: +0.035$, $p: 0.556$). **Conclusions:** The neutrophil to lymphocyte ratio is predicted to be useful in deciding the disease severity and histopathological grade of liver disease in chronic hepatitis B patients who are first seen in the outpatient clinic.

Key words: Hepatitis B virus (HBV), neutrophil to lymphocyte ratio.

Giriş ve Amaç: Son yıllarda ilgi odağı haline gelen, değişik inflamatuvar ve neoplastik hastalıklar ile ilişkilendirilen basit, ucuz ve efektif bir marker olan nötrofil-lenfosit oranı öne çıkmakta olup kronik hepatit B hastalarında çalışılmamıştır. Biz çalışmamızda hepatit B tanısı ile hastalık düzeyini belirleme, tedavi kararı alma gibi nedenlerle HBV DNA bakılan ve karaciğer biyopsisi uygulanan hastaların fibrosis düzeyi ile nötrofil-lenfosit oranı arasındaki ilişkiyi araştırdık. **Materyal ve Metod:** 2005-2010 yılları arasında biyopsi alınmış 486 hepatit B hastasının dosyası retrospektif tarandı ve bu hastalardan çalışma kriterlerine uyan 268 naiv hasta bulundu Naiv hastalar çalışmaya alınırken, daha önceden antiviral tedavi almış olan hastalar ve anti-Delta pozitif olanlar çalışma dışı bırakıldılar. **Bulgular:** Biyopsi alınmış olan çalışma popülasyonu 163 (%60,8) erkek (yaş: 38±13), 105 (%39,2) kadın (yaş: 37±14) olmak üzere 268 (yaş: 38±13) kişiden oluşmaktaydı. Hastaların fibrosis skoruna göre dağılımlarına bakıldığında 0/6=122 hasta (%45,5), 1/6=50 hasta (%18,7), 2/6=37 hasta (%13,8), 3/6=25 hasta (%9,3), 4/6=5 hasta (%1,9), 5/6=6 hasta (%2,2), 6/6=23 hasta (%8,6) şeklinde dağılmaktaydı. Fibrosis skoru ile nötrofil-lenfosit oranı ($r: +0,510$ $p: 0,000$), histolojik aktivite indeksi ($r: +0,478$ $p: 0,000$) arasında anlamlı korelasyon saptanırken, HBV DNA düzeyi arasında anlamlı bir korelasyon izlenmedi ($r: +0,035$ $p: 0,556$). Fibrosis skoru 0-1 olan hastaların nötrofil-lenfosit oranı değeri 1,6±0,5 iken fibrosis skoru 2-6 olanlarda 2,4±0,8 saptandı ($p=0,000$). **Tartışma:** Kronik hepatit B hastasını poliklinikte ilk kez gördüğümüzde hastalığın evresi ve histopatolojik olarak karaciğerin durumu hakkında klinisyen olarak bir öngöründe bulunmak için nötrofil-lenfosit oranının yararlı olacağını düşünmekteyiz.

Anahtar Kelime: Hepatit B virüsü, nötrofil lenfosit oranı

INTRODUCTION

In the approach to hepatitis B virus (HBV) treatment, the main indicator of this algorithm is viral load level (1-4). Although liver biopsy is still the gold standard in order to determine disease activity in chronic hepatitis B, this method is invasive, and physicians and patients have concerns about its complications (5). Therefore, alternative approaches and non-invasive tests have been investigated for a considerable time (6-9).

In recent years, the neutrophil to lymphocyte ratio (NLR) has become popular as a simple, inexpensive, and effective marker associated with various inflammatory and neoplastic diseases (10-13). This marker has not yet been investigated in the case of chronic hepatitis B patients. In our study, we aimed to investigate the relationship between fibrosis level and NLR for determining disease severity and making treatment decisions.

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MATERIALS and METHODS

Patients hospitalized at the Ministry of Health Haydarpaşa Numune Training and Research Hospital, Gastroenterology Clinic, between December 2005 and December 2010 who had liver biopsy due to hepatitis B were retrospectively determined from patient charts. Treatment-naïve patients were included in the study. The exclusion criteria were previous treatment with antiviral drug therapy, anti-Delta positivity, increased echo in hepatic parenchyma conforming to hepatosteatosis on ultrasonography, suspicious hepatocellular carcinoma (HCC) lesion image, and increased level of alpha-fetoprotein (AFP). Data records included gender, record number, age at biopsy date, serum HBV DNA level determined by quantitative polymerase chain reaction (PCR) within a maximum of three months prior to biopsy date, and absolute neutrophil and lymphocyte counts and their ratio. Ishak fibrosis evaluation and histological activity index (HAI) were also recorded. Pathology results in true carriers showed zero fibrosis and HAI. Fibrosis score was 6 in the case of pathology results as active macro- and micronodular cirrhosis, and HAI score was left blank. In case of re-hospitalization of unevaluated patients due to insufficient biopsy specimen and availability of a second biopsy result, these data were recorded. Patients with steatohepatitis according to biopsy results were excluded. All data were entered in the Statistical Package for the Social Sciences (SPSS) 16,0 program. Parametric data were expressed as arithmetical mean and standard deviation (SD), and non-parametric data were expressed as number and percentage. Normal distribution of parametric data was determined by Shapiro-Wilk test ($p>0,05$). Comparison of non-parametric data was done by Pearson χ^2 test ($p<0,05$). Spearman's correlation analysis was used for correlation.

RESULTS

A chart of 486 hepatitis B patients subjected to biopsy during the period 2005-2010 were investigated retrospectively. Only 268 naïve patients met the criteria for this study. The study population included patients with liver biopsy, consisting of a total of 268 (age: 38 ± 13 years) patients, including 163 (60,8%) males (age: 38 ± 13) and 105 (39,2%) females (age: 37 ± 14). Table 1 summarizes the demographics and parameters according to gender. Fibrosis score distribution of the patients was as follows: 0/6: 122 patients (45.5%), 1/6: 50 patients (18,7%), 2/6: 37 patients (13.8%), 3/6: 25 patients (9.3%), 4/6: 5 patients (1.9%), 5/6: 6 patients (2,2%), and 6/6: 23 patients (8,6%) (Table 2).

There was a significant correlation between fibrosis score

and NLR ($r: +0,510$, $p: 0.000$). NLR was $1,6\pm 0,5$ in patients with fibrosis score 0-1, and $2,4\pm 0,8$ in patients with fibrosis score 2-6 ($p=0,000$). There was a significant correlation between fibrosis score and HAI ($r: +0,478$, $p: 0,000$), while HBV-DNA levels showed no significant correlation ($r: +0,035$, $p: 0,556$).

DISCUSSION

When patients with chronic hepatitis B are first seen in the outpatient clinic, we determine an algorithm. Generally, the main indicator of this algorithm is the viral load level, and patients with low viral load are usually monitored, while patients with high viral load are more commonly considered for liver biopsy. However, it is speculated that this may not always be true and that there may be cirrhotic patients with low viral load due to severe fibrosis and reduced number of hepatocytes. In recent years, the NLR has become popular as a simple, inexpensive, and effective marker associated with various inflammatory and neoplastic diseases (10-13). In light of these studies, we aimed to investigate the relationship between fibrosis level and NLR for determining disease severity and making treatment decisions.

For this purpose, in our study, we retrospectively examined the records of about 500 patients who were subjected to liver biopsy for hepatitis B. Two hundred and sixty-eight patients were eligible for the study; patients with a history of prior treatment and comorbidities were

Table 1. Gender, age and analyzed parameters of all patients

	Male (n: 163)	Female (n: 105)	p
Age	38 ± 13	37 ± 14	$p=0,54$
HBV DNA (log)	$6,1\pm 1,7$	$6,1\pm 1,8$	$p=0,99$
Fibrosis score	$1,5\pm 1,8$	$1,3\pm 1,8$	$p=0,56$
HAI level	$4,5\pm 2,4$	$4,4\pm 2,9$	$p=0,84$
NLR	$1,9\pm 0,7$	$1,8\pm 0,7$	$p=0,62$

Table 2. Fibrosis score distribution of the patients (Mean-median)

Fibrosis	Mean \pm SD	Median (Range)
0	$1,65 \pm 0,44$	1,66 (0,78-3.30)
1	$2,04 \pm 0,62$	1,97 (0,93-3.75)
2	$2,30 \pm 0,45$	2,34 (1,08-2.99)
3	$2,47 \pm 0,86$	2,44 (1,19-3.64)
4	$2,94 \pm 1,06$	3,44 (1,11-3.76)
5	$2,86 \pm 1,05$	2,80 (1,62-4.32)
6	$2,81 \pm 0,94$	2,71 (1,31-4.27)

excluded. As biopsy was rare in patients with end-stage liver disease, we searched patients with a cirrhosis diagnosis without biopsy in order to increase the number of patients with cirrhosis in our study population. We excluded those patients who consumed alcohol or were diabetic.

A significant correlation was determined between fibrosis score and NLR ($r: +0.510$ $p: 0.000$); NLR was 1.6 ± 0.5 in patients with fibrosis score 0-1, and 2.4 ± 0.8 in patients with fibrosis score 2-6 ($p=0.000$). We demonstrated that there was a positive relationship between degree of fibrosis and NLR. A correlation between HAI and fibrosis was also demonstrated in our study.

Several studies have investigated non-invasive tests to predict the severity and histopathological grade of fibrosis in liver diseases (14-17). In a recent study, Alkhouri *et al.* reported that the NLR is higher in patients with nonalcoholic steatohepatitis (NASH) and advanced fibro-

sis (14). They determined higher NLR in NASH patients compared with controls and also reported that this ratio is correlated with liver inflammation and fibrosis. In addition to this study, Dogan *et al.* confirmed that NLR correlated with the main histological features of fatty liver disease including fibrosis (16). Celikbilek *et al.* explained these findings by arterial hypertension, which may lead to activation of pro-inflammatory pathways (17). All these well-designed studies confirmed our findings that there was a positive relationship between fibrosis and NLR, with NLR increasing in the presence of fibrosis progression.

The NLR is a novel non-invasive, inexpensive test that can be used to predict fibrosis in HBV patients. Especially in cirrhotic patients with low viral load due to severe fibrosis and a reduced number of hepatocytes, this finding can be useful to predict liver fibrosis and roadmap in HBV treatment.

REFERENCES

- Liaw YF. Natural history of chronic hepatitis B virus infection and long-term outcome under treatment. *Liver Int* 2009; 29 (Suppl 1): 100-7.
- Ceylan B, Fincanci M, Yardimci C, *et al.* Can mean platelet volume determine the severity of liver fibrosis or inflammation in patients with chronic hepatitis B? *Eur J Gastroenterol Hepatol* 2013; 25: 606-12.
- Ahn SH, Chan HL, Chen PJ, APPROACH Working Group. Chronic hepatitis B: whom to treat and for how long? Propositions, challenges, and future directions. *Hepatol Int* 2010; 4: 386-95.
- European Association for the Study of the Liver. EASL clinical practice guidelines: management of chronic hepatitis B virus infection. *J Hepatol* 2012; 57: 167-85.
- Lee IC, Huang YH, Chan CC. Impact of body mass index and viral load on liver histology in hepatitis B e antigen-negative chronic hepatitis B. *Clin Nutr* 2011; 30: 647-52.
- Poynard T, Morra R, Ingiliz P, *et al.* Assessment of liver fibrosis: noninvasive means. *Saudi J Gastroenterol* 2008; 14: 163-73.
- Ucar F, Sezer S, Ginis Z, Ozturk G. APRI, the FIB-4 score, and Forn's index have noninvasive diagnostic value for liver fibrosis in patients with chronic hepatitis B. *Eur J Gastroenterol Hepatol* 2013 Mar 18 [Epub ahead of print].
- Liang B, Li Y, Zhao A, *et al.* Clinical utility of serum matrix metalloproteinase-2 and tissue inhibitor of metalloproteinase-2 concentrations in the assessment of liver fibrosis due to chronic hepatitis B. *J Int Med Res* 2012; 40: 631-9.
- Martínez SM, Crespo G, Navasa M, Forn X. Noninvasive assessment of liver fibrosis. *Hepatology* 2011; 53: 325-35.
- Zhou K, Gao CF, Zhao YP, *et al.* Simpler score of routine laboratory tests predicts liver fibrosis in patients with chronic hepatitis B. *J Gastroenterol Hepatol* 2010; 25: 1569-77.
- Wang D, Wang Q, Shan F, *et al.* Identification of the risk for liver fibrosis on CHB patients using an artificial neural network based on routine and serum markers. *BMC Infect Dis* 2010; 10: 251.
- Guthrie GJ, Charles KA, Roxburgh CS, *et al.* The systemic inflammation-based neutrophil-lymphocyte ratio: experience in patients with cancer. *Crit Rev Oncol Hematol* 2013 Apr 16 [Epub ahead of print].
- 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.
- Alkhouri N, Morris-Stiff G, Campbell C, *et al.* Neutrophil to lymphocyte ratio: a new marker for predicting steatohepatitis and fibrosis in patients with nonalcoholic fatty liver disease. *Liver Int* 2012; 32: 297-302.
- Ozturk ZA, Kuyumcu ME, Yesil Y, *et al.* Is there a link between neutrophil-lymphocyte ratio and microvascular complications in geriatric diabetic patients? *J Endocrinol Invest* 2013 Mar [Epub ahead of print].
- Dogan S, Celikbilek M, Yagbasan A, Yücesoy M. Noninvasive assessment of the effect of treatment in nonalcoholic fatty liver disease: neutrophil to lymphocyte ratio. *Liver Int* 2012; 32: 1189.
- Celikbilek M, Dogan S, Yurci A, Ozbakir O. Neutrophil to lymphocyte ratio: a hopeful marker predicting the disease severity in patients with nonalcoholic fatty liver disease. *Liver Int* 2012; 32: 1188.