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ORIGINAL ARTICLE

Value of neutrophil/lymphocyte ratio in the diagnosis of diabetic neuropathy

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Abstract

Background Peripheral diabetic neuropathy (PDN) had been demonstrated as a chronic inflammation state and one of the most common complications of type 2 diabetes mellitus (T2DM). Neutrophil-to-lymphocyte ratio (NLR) is a novel marker to reflect many kinds of chronic inflammation disease including diabetes. We aim to evaluate the association between NLR and PDN and to determine whether NLR could be a new indicator of PDN in T2DM.

Methods Hospital records of the patients who underwent electroneuromyography studies with the diagnosis of T2DM in the Neurology Outpatient Clinic between 01/01/2018 and 01/04/2021 were divided into two groups as those with normal results and those with polyneuropathy. The NLR was calculated from the hemogram tests.

Results Eighty-nine (52.7%) normal and 80 (47.3%) PDN patients included in the study, of them 77 (45.6%) were male and 92 (54.4%) were female. The mean age of the patients was 58.92±13.88 years. According to the records examined, the mean NLR value was significantly higher in patients with PDN (2.70±1.99) than in those with normal results (1.98±0.80). According to the ROC analysis, the sensitivity is 0.875 and the specificity is 0.292 for 1.46, which is determined as the optimal cut-off value for the NLR value in the diagnosis of PDN.

Conclusion The results of our study have shown that there was a significant correlation between NLR and PDN, implying that inflammation and endothelial dysfunction could be an integral part of PDN. NLR was significantly and independently raised in patients with T2DM.

Keywords Diabetes · Peripheral diabetic neuropathy · Neutrophil-to-lymphocyte ratio · ROC analysis

Background

Peripheral neuropathy is a term used to describe general diseases of the nervous system and is seen with motor, sensory and autonomic symptoms. Approximately 50% of all polyneuropathies are associated with pain. Peripheral diabetic neuropathy (PDN) is a common serious complication of diabetes and negatively affects the daily lives of patients.

Diabetes is an increasingly common disease. The World Health Organization (WHO) estimates that by 2025 the total number of diabetics will reach 300 million. Its prevalence is approximately 0.3–0.5%. Type 2 diabetes mellitus (DM) is usually observed over the age of 40. It progresses with insulin deficiency and insulin resistance in peripheral tissues. The prevalence of diabetes mellitus varies with age [1, 2].

Complications begin in the first years following the diagnosis of diabetes or patients are affected by complications when the diagnosis is made. Although many factors cause the development of PDN, the most effective method in the development of complications and prognosis is to keep the blood glucose level under good control [3, 4].

PDN may present clinically as neuropathic pain, trophic changes, motor symptoms, and autonomic dysfunction. Although glycemic control reduces the risk of neuropathy in diabetic patients, unfortunately, there is no effective treatment for diabetic neuropathy yet. Nerve damage in PDN occurs as a result of metabolic factors, oxidative stress, ischemic factors, and inflammation. Neuroinflammation is a physiological process necessary for regeneration and healing. As a result of deterioration of neuroinflammation, chronic pain occurs [5, 6].

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Neutrophil-to-lymphocyte ratio (NLR) is an inexpensive and easily calculated index that correlates with the prognosis of systemic inflammatory diseases. It can be used especially in inflammatory, cardiovascular, and cancer diseases [7, 8].

PDN is a common complication of diabetes that may be associated with inflammation and vascular diseases. PDN is a disease that significantly impairs quality of life and prevents activities of daily living. Increased levels of peripheral inflammation may play a role in the pathogenesis of PDN. Differential diagnosis of this disease involves some difficulties. NLR and similar markers are easily accessible parameters and can be used in differential diagnosis and can guide clinicians in diagnosis. In this study, the neutrophil-to-lymphocyte ratios of patients with proven PDN by electroneurophysiological tests and the use of NLR ratio in the differential diagnosis were clarified by comparing them in patients with diabetes and without neuropathy.

Materials and methods

The study was carried out on the patient registration system data of Çanakkale Onsekiz Mart University Hospital. Ethics committee approval was obtained for the study from Çanakkale Onsekiz Mart University Ethics Committee for Clinical Studies with decision number 05-19 dated May 06, 2021. The study population consisted of the patients included in the hospital registration system, and the study sample consisted of the records selected from here within the study inclusion criteria.

Patient's records of who underwent electroneuromyography (ENMG) with the diagnosis of DM in the Neurology Outpatient Clinic of Çanakkale Onsekiz Mart University Hospital between January 2018 and April 2021 were collected from the patient registry system. All records available between the specified dates were included in the study. The study method did not involve any patient contact.

The NLR was calculated from the hemogram test results. The oldest hemogram test result was taken if more than one test present. Records of patients with diseases known to seriously impair hemogram parameters (oncological diseases, hematological diseases, acute vascular diseases, end-stage renal disease, inflammatory diseases (e.g., ankylosing spondylitis, ulcerative colitis), and acute infections) were excluded. Hemogram measurements were studied using the Coulter method on the hospital's Beckman Coulter DXH800 device.

PDN was diagnosed according to nerve conduction study (NCS) recordings done as part of ENMG performed with a Nihon-Kohden device (NihonKohden-Neuropack®) and interpreted by an experienced neurologist (first author). NCS was performed on three extremities of each subject including motor components of the peroneal, posterior tibial, median, and ulnar nerves and sensory components of the sural, median, and ulnar nerves; nerve conduction velocities; distal latencies; and amplitudes were recorded. Demyelinating neuropathy was diagnosed in prolonged distal motor latency, slowed conduction velocity, conduction blocks, and prolonged or absent F-wave latency while axonal neuropathy was diagnosed when low or loss of motor and sensory action potential was detected.

Patients with normal or mixed-type sensorimotor polyneuropathy in the NCS results were included in the study; other subtypes of PDN and mononeuropathy were excluded. Patients were divided into two groups according to their NCS as PDN and normal results.

NCS results in 89 (52.7%) were normal. Out of 90 NCS reflecting pathological results, 74 (82.2%) were diffuse mixed-type sensorimotor polyneuropathy, 6 (6.7%) were mixed-type sensorimotor polyneuropathy in the lower extremities, 7 (7.8%) were carpal tunnel syndrome, 1 (1.1%) was sensory polyneuropathy, 1 (1.1%) was demyelinating polyneuropathy, and 1 (1.1%) was ulnar neuropathy. When all NCS results were evaluated, it was determined that there were 80 (47.3%) patients with mixed-type sensorimotor polyneuropathy as a complication of DM. Patients with mononeuropathy and subtypes of PDN other than mixed-type sensorimotor polyneuropathy were excluded from the study and the study was conducted on 169 outcomes.

Statistical analysis

Collected data were digitalized and corrected; descriptive information is presented as the frequencies and percentages for categorical data, and with mean and standard deviation for ordinal data. Since the sample number was larger than 30, the normal distribution assumption of parametric tests was ignored based on the central limit theorem. In the evaluation of the strength of the correlations, $r < 0.30$ was classified as weak, $0.30 \leq r < 0.50$ moderate, and $r \geq 0.50$ strong. Chi-square, Student t, and Pearson correlation tests were used for statistical analysis. When the expected value was less than 5, Fisher's exact test result was reported. Test constants and absolute p values are presented for all analyses and $p < 0.05$ was accepted as the general significance limit. ROC analysis was performed to determine the diagnostic power of the NLR value for PDN. The optimal cut-off value was selected and reported in the ROC curve obtained.

Results

Of the 169 recordings included in the study, 89 (52.7%) of the NCS results were normal and 80 (47.3%) were mixed-type sensorimotor PDN. Of the 169 records included in the study, 77 (45.6%) were male and 92 (54.4%) were female patients. The mean age of the patients was 59.05 ± 14.02 (minimum 20 and maximum 87). The mean age of men (58.92 ± 13.88) was

not significantly different from that of women (59.15 ± 14.20) ($t = 0.106$; $p = 0.916$).

The rate of women with PDN (38.0%) was significantly lower than that of men (58.4%) ($X^2 = 6{,}996$; $p = 0.008$). The mean age of those with PDN (62.14 ± 12.79) was significantly higher than that of the normal (56.27 ± 14.55) ($t = 2.771$; $p = 0.006$).

According to the records examined, the mean neutrophil count in mm^3 blood was 4.80 ± 2.09, and the lymphocyte count was 2.33 ± 0.83. The mean NLR value was 2.32 ± 1.53. While the mean lymphocyte count of women (2.17 ± 0.75) was higher than that of men (2.52 ± 0.88) ($t = 2.801$; $p = 0.006$), there was no significant difference between the sexes in terms of mean neutrophil counts and NLR ratio ($t = 0.034$, $p = 0.973$ and $t = 0.968$, $p = 0.335$ respectively). While there was a weak negative correlation between age and mean lymphocyte count ($r = -0.152$; $p = 0.049$), there was no significant correlation with neutrophil count and NLR ($r = 0.002$; $p = 0.975$, $r = 0.082$; $p = 0.287$, respectively).

Table 1 shows the neutrophil count, lymphocyte count, and NLR values in patients with PDN and normal EMG. The appearance of NLR values in patients with normal NCS results and PDN is presented in Figure 1.

The ROC analysis curve for the comparison of the NLR values of those with PDN and those with normal NCS results is presented in Figure 2. According to the results of the analysis, the area under the curve was calculated as 0.593 (95% confidence interval 0.508–0.679) (standard error = 0.044, $P = 0.036$). According to the results of the analysis, the optimal cut-off value for the NLR value in the diagnosis of PDN was determined as 1.46, and the sensitivity for this value was 0.875 and the specificity was 0.292. For the cut-off value of 2.485, the sensitivity was 0.338 and the specificity was 0.775.

Discussion

Involvement of the peripheral nervous system in diabetic patients affects the quality of life. Diabetes can affect various parts of the nervous system. PDN is the most common peripheral nervous system complication and defined as the presence of symptoms of peripheral nerve dysfunction in diabetic patients after excluding other causes of neuropathy [9]. The diagnosis of PDN is recommended on the basis of neuropathic symptoms, signs, and nerve conduction studies, and there is no gold standard [10].

Inflammatory processes play a key role in DM like other chronic diseases including cardiovascular disease, cancer, and chronic kidney disease [11]. Peripheral nerve inflammation leads to oxidative stress, and increased reactive oxygen concentrations have been associated with the development of microvascular complications of diabetes, including neuropathy. Total antioxidant levels were low and total oxidative status increased in diabetic patients with neuropathy [12, 13]. In a prospective study, biomarkers of inflammation were associated with the onset and progression of neuropathy in an elderly population with diabetes [14].

Chronic inflammation accompanying metabolic dysfunction in type 2 DM supports the development and acceleration of micro- and macro-angiopathic complications [15]. Inflammatory molecules (such as adipokines, chemokines, adhesion molecules, and cytokines) and endothelial dysfunction play a role in disease processes [16].

NLR, a new marker of chronic inflammation, reflects the balance of two interconnected components of the immune system. The first of these is neutrophils, which form the first line of defense as active nonspecific inflammatory mediators, and the other is lymphocytes, which are the regulatory or protective component of inflammation [17]. It has been demonstrated that NLR calculated from blood cell counts can be used as an indicator of systemic inflammation [18]. Systemic inflammation reflected by NLR has been associated with chronic diseases such as hypertension and diabetes [19]. In addition to being associated with glucose intolerance and insulin resistance, NLR has been shown to be used as a prognostic marker for macro- and microvascular complications in diabetic patients [20].

Inflammation has a significant impact on the development and progression of PDN [21]. Inflammatory molecules and endothelial dysfunction play an important role in the PDN

Table 1 The status of neutrophil and lymphocyte counts and NLR values in those with normal NCS results and PDN

	EMG result		
	Normal ($n = 89$)	PDN ($n = 80$)	Analyses*
Neutrophil count (per mm^3)	4.48 ± 1.53	5.16 ± 2.55	$t = 2.133$; $p = 0.034$
Lymphocyte count (per mm^3)	2.46 ± 0.85	2.18 ± 0.79	$t = 2.177$; $p = 0.031$
NLR value	1.98 ± 0.80	2.70 ± 1.99	$t = 3.145$; $p = 0.002$

*Independent samples *t* test

NLR neutrophil-to-lymphocyte ratio, *NCS* nerve conduction studies, *PDN* peripheral diabetic neuropathy

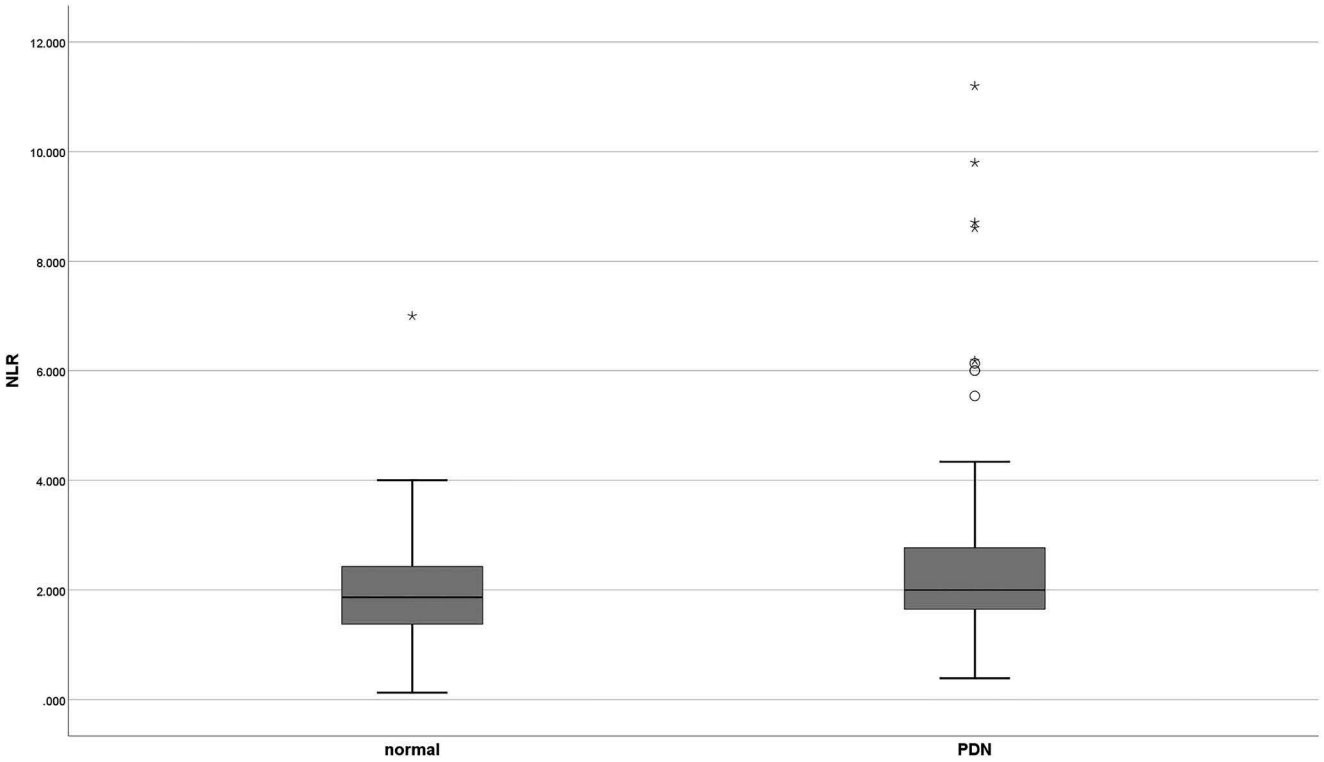

Fig. 1 Neutrophil-to-lymphocyte ratio (NLR) values in patients with peripheral diabetic neuropathy (PDN) and normal NCS results

settings [22]. NLR has also been proposed as a predictor to evaluate the development of microvascular complications of diabetes. Ulu et al. showed that NLR is a reliable prognostic marker for the presence and severity of diabetic retinopathy [23] and a predictive and prognostic marker for sensorineural hearing loss [24].

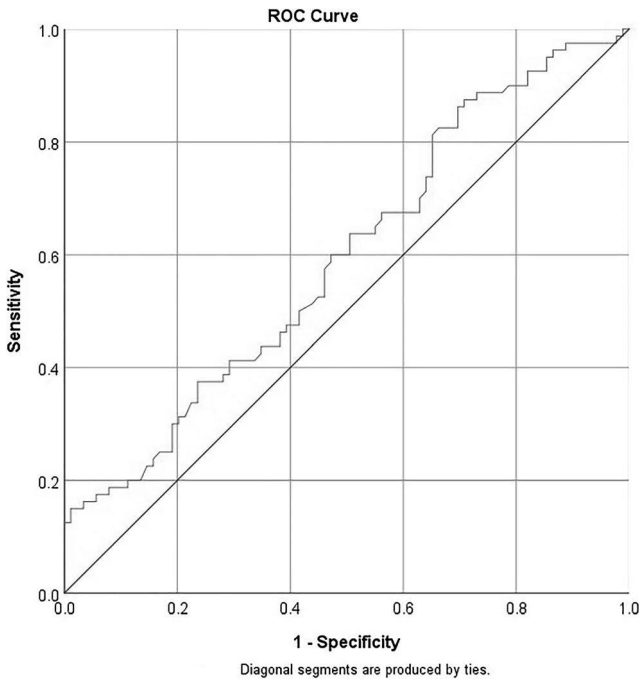

Fig. 2 ROC curve for the comparison of neutrophil-to-lymphocyte ratio values of those with peripheral diabetic neuropathy and normal NCS results

In our study, we investigated the relationship between NLR and PDN in patients with electroneurophysiologically detectable sensorimotor polyneuropathy. There are studies investigating the relationship between PDN and NLR but this study focused on a specific subtype of diabetic neuropathy. We concluded that NLR has a significant predictive value and can be used as a prognostic marker for diabetic mixed-type sensorimotor polyneuropathy.

In a recent study, T1 and T2 DM patients were followed up for 18 months and newly developed PDN cases were separated and examined [25]. According to the results of the study, NLR and platelet-to-lymphocyte ratio (PLR) for T1DM and NLR for T2DM were significant markers for the development of PDN. The sensitivity for NLR at a cut-off value of 2.485 was calculated as 0.380 and specificity 0.790 for demonstrating the development of PDN in T2DM.

Our results have very close sensitivity (0.338) and specificity (0.775) values with the cut-off value (2.485) reported in this study. Although the figures have not been reported, it is observed that very close values have been reached in the ROC graph of this study for the optimal cut-off value (1.46) that we suggest according to our study results. When the results are interpreted together, it can be concluded that the NLR results between the two specified cut-off values will be less reliable in clinical decision-making.

Although high diagnostic values of NLR values in other diabetic complications such as nephropathy and retinopathy have been reported in various studies, it should be kept in mind that cut-off values and diagnostic powers will be different for each clinical situation.

The primary limitation of our study is that it was conducted retrospectively. However, the effects of this will not be very serious because the diagnoses of the patients were made by electroneurophysiological tests and the hemogram values performed at the diagnosis were used. In fact, unlike other vascular complications of diabetes, it is difficult to clearly determine when PDN begins. Another limitation is that only large fiber neuropathies are included. Nerve conduction studies, which are widely used for peripheral neuropathies, are primarily based on large nerve functions and are often found to be normal in small fiber neuropathies. Finally, the clinical signs and symptoms of the patients were not evaluated in our study. In accordance with the aim and method of the study, only electroneurophysiological diagnosis was focused. The mechanism of formation of neuropathic pain, which is the most important symptom, differs from neuropathy and its presence or absence is not correlated with neuropathy. Although other screening tests based on symptoms and findings may aid in the diagnosis of clinical neuropathy, they will not be of additional benefit for patients with a definitive electroneurophysiological diagnosis.

Conclusion

The results of our study showed that there is a significant correlation between NLR and PDN. NLR can be considered as a predictor of PDN and a prognostic risk marker. NLR is an easy parameter to calculate using blood cell counts. This test is simple, inexpensive, and routinely performed. NLR can be an alternative to other expensive inflammatory markers when laboratory facilities and financial constraints exist. More research with prospective design should be conducted on the role of NLR as a marker of inflammation and a risk factor for PDN.

Author contribution All authors contributed to the study conception and design, material preparation, data collection and analysis, and draft writing of the manuscript. All authors read and approved the final manuscript.

Declarations

Ethics approval Approval was obtained from the Çanakkale Onsekiz Mart University Ethics Committee for Clinical Studies with decision number 05-19 dated May 06, 2021. The procedures used in this study adhere to the tenets of the Declaration of Helsinki.

Competing interests The authors declare no competing interests.

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