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Disease severity and serum endocan levels in fibromyalgia patients

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Abstract

The purpose of this study was to look into the link between serum endocan levels and disease severity in fibromyalgia syndrome (FMS).We evaluated 45 patients with FMS according to the 2010 ACR FMS criteria and 28 controls. Disease severity was evaluated with the symptom severity scale (SSS), the widespread pain index (WPI), and the visual analog scale (VAS). Serum endocan, neutrophil-lymphocyte ratio (NLR), platelet-lymphocyte ratio (PLR), and CRP values of the patient and control groups were measured. Endocan levels were significantly higher in the FMS group compared to controls. Serum endocan levels were 0.99 ± 0.28 ng/mL (range: 0.1-2.2) in FMS patients and 0.63 ± 0.17 ng/mL (range: 0.4-1.1) in controls ($p<0.05$). In our study, endocan levels that were higher in patients with FMS compared to controls supported the use of endocan as an important potential marker for FMS.

Keywords: fibromyalgia syndrome, endocan, symptom severity scale, inflammation, pain

1. Introduction

Fibromyalgia syndrome (FMS) is a chronic rheumatic disease characterized by widespread pain and different clinical comorbidities. Fibromyalgia syndrome can be seen as a comorbidity with other rheumatic diseases. FMS may be associated with clinical conditions such as pain, sleep disturbance, depression, genitourinary disorders, and irritable bowel syndrome (1, 2). What are the causes of FMS? The answer to the question is still under investigation. Genetic, environmental, and neurohormonal factors and inflammation are thought to play a role in the etiopathogenesis of FMS (3, 4). Inflammatory mediators cause the inflammatory response, which is one of the defense processes required for human survival (5). Although FMS is thought to be a non-inflammatory rheumatic disease, the potential role of inflammation in the pathogenesis of FMS is being investigated (5-7). Recent studies have found a relationship between oxidative stress, inflammation, and endothelial dysfunction in FMS (8, 9).

Endocan is a biomarker of endothelial activation and is secreted from endothelial cells as a soluble proteoglycan (10-12). Studies investigating endocan as a blood and tissue-based biomarker for cancer and inflammation have found results supporting this view. (13-15). Most patients admitted to the hospital have a complete blood count (CBC) and laboratory tests as part of their routine evaluation. The blood test results are important in inflammatory processes (16). Neutrophil-lymphocyte ratio (NLR) and platelet-lymphocyte ratio (PLR) are non-invasive, promising, and cost-effective prognostic and diagnostic biomarkers for cardiovascular disease (18), cancer (19, 20), rheumatological diseases (21), and some neurological diseases (22, 23). This study evaluated inflammatory biomarkers in FMS patients; NLR, PLR, and serum endocan levels. The presence of inflammation in FMS, the availability of endocan as a biomarker, and the relationship between endocan levels and disease symptom severity were investigated.

2. Material Methods

Individuals aged ≥18 years old and over who applied to the Physical Medicine and Rehabilitation Clinic between October 2020 and October 2021. The study was conducted by including 45 patients who met the 2010 ACR (24) FMS criteria, and 28 controls without a diagnosis of FMS. Smokers and patients with diagnosed arterial hypertension, inflammatory disease, diabetes mellitus, metabolic syndrome, thyroid dysfunction, anemia, local or systemic infection, kidney or liver dysfunction, malignancy, pregnancy, and coronary artery disease were excluded from the study. The study complied with the Declaration of Helsinki, and informed consent was obtained from all participants. Endocan and laboratory parameters were measured in all subjects. The symptom severity scale (SSS), the widespread pain index (WPI), and the visual analog scale (VAS) were evaluated.

Symptom Severity Scale (SSS): Fatigue, restless sleep, cognitive symptoms, a six-month headache, abdominal pain,

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depression, and other somatic symptoms were questioned (25). Scoring is between 0 and 12. The continuation of the symptoms for at least 3 months is necessary for the diagnosis.

Widespread Pain Index (WPI): Right and left; there is pain in at least 4 of the following: the jaw, shoulder, upper arm, forearm, hip (trochanter, gluteal region), upper-lower leg, neck, back, waist, chest, and abdomen. For each region, areas of continuous pain in the last seven days are marked. The score is between 0 and 19. Widespread pain index (WPI) ≥7, symptom severity scale (SSS) 5, or WPI = 3-6, and SSS ≥ 9 are required for diagnosis (26).

Visual Analog Scale (VAS) It is a table used for digitizing values that cannot be measured numerically. Marks where a patient's condition is appropriate on a 100 mm line. It is a common, reliable test accepted in the literature. It can be applied easily (27).

After 12 hours of fasting, samples taken from the 5 ml antecubital vein from patients for a complete blood count were analyzed by dropping them into vacuum tubes containing ethylene diamine tetraacetic acid (EDTA)-anticoagulation tubes (BD Vacutainer K2E; Becton Dickinson, UK). CBC parameters were evaluated with the Beckman Coulter DxH 800 hematology analyzer (Beckman-Coulter, Brea, CA). CRP levels were assessed with IMAGE 800 (Beckman, USA). Blood samples from the control and patients. The blood materials of the participants were placed in tubes with aprotinin (BD Vacutainer SST II Advance, BD, Plymouth, UK) to determine endocan levels and centrifuged at 4000 rpm for 10 minutes. The resulting plasma (containing Endocan) was placed in small-volume Eppendorf tubes for analysis and stored at -80 °C until runtime. Plasma endocan levels were assessed using the Endothelial Cell-Specific Molecule 1 (ESM1) ELISA Kit (CLOUD-CLONE CORP. (CCC, USA, Wuhan), Item No.: SEC463Hu) in accordance with the working procedure outlined in the kit's catalog. Absorbance was evaluated using the Chromate 4300 Microplate Reader (Awareness Technology, Palm City, USA). The minimum detection limit of Endocan was 0.065 ng/mL. The intra-assay coefficient of variation for Endocan was <10%, and the inter-assay coefficient of variation was <12%.

2.1. Statistical analysis:

All statistical analyses were performed using SPSS version 23.0 (SPSS Inc., Chicago, IL). The results were given as median (range), and mean ± SD. Data that showed normally distributed differences between the fibromyalgia and control groups were evaluated using the independent samples t-test. A $p < 0.05$ was considered statistically significant.

3. Results

The patient and control subjects were similar in age and body mass index (BMI). Participants with FMS had significantly higher serum endocan levels. Endocan mean serum levels in FMS patients were 0.99± 0.28 ng/mL (range: 0.1-2.2) and 0.63± 0.17 ng/mL (range: 0.4- 1.1) in control subjects (p :0.05). The mean age of FMS patients was 44.71± 8.8 years, while the control group's mean age was 43.22 9.3 years (p: 0.49). The neutrophil/lymphocyte ratio in patients was 1.93±0.7 in FMS patients and 2.1±0.63 in controls (p = 0.261). The relationship between inflammatory markers and VAS, WPI, and SSS, which show the severity of the disease, is shown in Table 1. The ROC analysis shown in Fig. 1. [AUC (%95 GA): 0.89 (0.81-0.97), sensitivity: % 68.9, specificity: %96.

Table 1. Clinical and laboratory values in the patient and control groups

	FMS	Control	p
Age	44.08±9	43.8±9.2	0.9
CRP (mg/dl)	5.2±4.5	5.6±6.7	0.7
Monocyte/Lymphocyte	0.2±0.08	0.2±0.14	0.7
Platelet/Lymphocyte	133.1±47.2	133.7± 41.7	0.9
Endocan (mg/dl)	0.9±0.2	0.6±0.1	0.00
VAS	6.4±2.9	0.03±0 .1	0.00
Widespread pain index	11.8±5.9	0	0.00
Symptom severity scale	7.3±3.6	0	0.00
BMI	27.6±4.8	28.7±3.7	0.3

FMS: Fibromyalgia Syndrome, CRP: C-Reactive Protein VAS: Visual Analog Scale, BMI: Body Mass Index, mg: milligram, dl: deciliter, p: p value

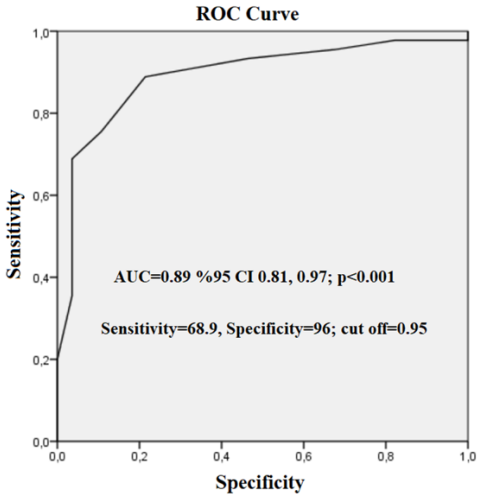

Fig. 1. ROC analysis for endocan level in the FMS group

4. Discussion

In our study, we determined significantly higher endocan levels in FMS patients compared with healthy individuals. Endocan is an essential immunomodulatory protein secreted by human vascular endothelial cells that has been proposed as a biomarker to demonstrate endothelial dysfunction. (12,13). According to our reviews, there is only one study in the literature that demonstrates elevated serum endocan levels in patients with FMS (28). Unlike the current study, the effects of endocan levels on FMS severity were examined in our study. We evaluated the subclinical inflammation suggested to exist in FMS with the endocan level, CRP, platelet-lymphocyte ratio

(PLR), and neutrophil-lymphocyte ratio (NLR) (16). CRP, PLR, and NLR values were similar in patients and controls. CRP is typically only significantly elevated during acute inflammation or infection. High-sensitivity CRP (hs-CRP) measurement is useful in low-grade and chronic conditions as it allows accurate measurement at low levels (29). Focusing on chronic inflammation in FMS may explain the similarity of CRP values. Studies in which hs-CRP measurements can be made will help in this regard. On the other hand, endocan levels were significantly different between groups. Our findings support previous studies showing that these markers alone cannot help us determine the pathophysiology of the disease in FMS, but may be supportive in the diagnosis (30–32). The low number of our patients may explain this situation. We found no evidence that PLR and NLR changed disease severity (SSS and WPI). Similarly, Jayakrishnan et al. reported that PLR and NLR were not associated with disease severity. Our study is consistent with these findings (31). High serum endocan levels have been associated with subclinical inflammation and endothelial cell dysfunction in cancer, sepsis, diabetes, psoriasis (33), Behçet's syndrome (34), and cardiovascular diseases (35), as in FMS.

Studies have shown that increased IL-6 and IL-8 levels correlate with the severity of symptoms in FMS cases (2). A recent review highlighted high levels of cytokines in FMS. All these results clearly show that inflammation is effective in the pathogenesis of FMS. The relationship between the chemokine-cytokine network and FMS is important for determining effective treatment modalities (36-38). Endocan had a sensitivity and specificity for FMS diagnosis in our study's ROC analysis. [AUC (%95 GA): 0.89 (0.81-0.97), sensitivity: % 68,9, specificity: %96. Mertoğlu et al. evaluated the possibility of endocan as a diagnostic marker in FMS, the sensitivity was 88.5% and the specificity was 89.7% (39).

We determined a significant relationship between endocan levels and SSS and WPI, reflecting cognitive impairment, pain, fatigue, and mood disorders in FMS. This demonstrated the link between disease severity and endocan levels. In our literature search, we did not find any studies evaluating the effects of endocan levels on disease severity in FMS. Taylor et al. In their study evaluating inflammation and pain in FMS, they stated that high levels of inflammation increase perceived pain (40). The relationship we found between endocan levels and the parameters we evaluated for the severity of FMS; supports the hypothesis that subclinical inflammation affects the severity of the disease in the pathophysiology of FMS. Our study also supported the utility of endocan as an important potential marker for FMS. More comprehensive, multicenter studies are needed to explain the inflammation network in FMS and its possible role in the diagnosis and/or treatment of FMS. Recent research should look into the levels of biomarkers, cytokines, and autoantibodies that are more specific to inflammatory processes in patients with pre-diagnosed FMS.

Conflict of interest
The authors declared no conflict of interest.

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None to declare.

Authors' contributions
Concept: N.P.T., R.A.B., P.Ö., Design: N.P.T., P.Ö., Data Collection or Processing: N.P.T., Ö.A., F.F.Ş Analysis or Interpretation: N.P.T., R.AB., Literature Search: N.P.T., R.AB., Ö.A.,F.F.Ş Writing: N.P.T., R.A.B

Ethical Statement
Approval was obtained from Fırat University Noninterventional Research Ethics Committee, the study started. The ethics committee decision date is 19/04/2019 and the number of ethical committee decisions is 31.

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