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Association of Neutrophil to Lymphocyte Ratio with Disease Severity and Joint Effusion in Patients with Early Stage Knee Osteoarthritis

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Abstract: *The aim of the article was to determine the association between neutrophil to lymphocyte-ratio (NLR), clinical disease severity and joint effusion in patients with early knee osteoarthritis. We evaluated 79 patients with mild knee degeneration. We obtained NLR from routine hematology panels. As controls, 1260 hematology panels were also obtained. Clinical severity was evaluated from patient reported outcome scores: International Knee Documentation Committee Subjective Knee Evaluation Form, Knee Disability And Osteoarthritis Outcome Score For Joint Replacement, Tegner-Lysholm scale and Euroqol EQ-5D-5L questionnaire. Joint effusion was determined by ultrasound. For the statistics software R, version 3.4.4 was used. There were differences in demographics but not in NLR between the study patients and controls (2.03 versus 1.98, p=0.606). There was no correlation between NLR and joint effusion (r2= - 0.121) or between NLR, joint effusion and any other parameter recorded (r2 from - 0.134 to 0.190). In the multiple regression model, NLR was minimally influenced by age (p=0.059) and KOOSJR (p=0.023). Neutrophil to lymphocyte ratio is within normal limits in patients with early stage knee osteoarthritis and is not associated with clinical severity or with joint effusion.*

Keywords: *knee joint, osteoarthritis, ultrasonography, neutrophil to lymphocyte ratio, patient reported outcome measures*

1. Introduction

Osteoarthritis (OA) is one of the most prevalent diseases in the world, and the knee is the most affected large joint [1]. Diagnosis is mostly based on clinical presentation. Patient reported outcomes (PROs) are the most common measure for quantification [2].

Recent evidence showed that degenerative changes may begin many years before evident radiographic changes. The mechanisms of degeneration are not fully understood but inflammation is incriminated in the pathogenesis of OA. Macrophages and proinflammatory cytokines like IL-6, IL8, IL-1, and TNF-α are known as part of this process [3]. Most of these cytokines are used as biological markers detecting OA but they are used for scientific investigations. There is a need for a simple, inexpensive laboratory test for clinical practice. Inflammation and knee effusion are correlated. Knee effusion and synovial hypertrophy are two of the most common manifestations of knee inflammation and they can be easily determined by ultrasound [4,5]. There is an increased interest in using biomarkers for osteoarthritis to determine early disease, and to identify risk of progression [6]. Neutrophil to lymphocyte ratio (NLR) is a nonspecific marker of inflammation.

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It has been proven as a negative outcome predictor in gastrointestinal cancers, cardiovascular events [7,8]. However, studies on fractures, OA and inflammatory arthritis did not bring consistent results [9-14].

The aim of our study was to determine the association between NLR, clinical disease severity and joint effusion in patients with early stage knee OA.

2. Materials and methods

We included 79 subjects with atraumatic knee pain examined in our department, many of which underwent arthroscopy according to current clinical practice guidelines [15,16]. Incipient degenerative changes (OA) were observed from clinical presentation, imaging studies (magnetic resonance imaging, MRI) and available operative reports [1,2,15].

NLR was determined as the ratio between the absolute number of neutrophils and lymphocytes obtained from routine hematology panels retrieved from the hospital's electronic records [8,9]. Tests were determined using Nihon Kohden Celltac 6500, Sysmex XT-4000i or ADVIA 2120 analyzers. For the control group, all standard hematology panels coded as routine control for hospital employees were retrieved anonymously from the electronic medical records over a period of 2 years.

Patients were evaluated using the international knee documentation committee subjective knee evaluation form (IKDC), the knee disability and osteoarthritis outcome score for joint replacement (KOOSJR), Tegner-Lysholm scale and Euroqol EQ-5D-5L Index (converted using the UK tariff) and visual analogue scale (VAS) [2,17,18].

Joint effusion has been evaluated by ultrasound according to previously reported recommendations [4,5]. The knee was examined in approximately 30 degrees of flexion (patient supine on with a cloth rolled under the examined knee). A 12 MHz linear transducer (GE Healthcare, Venue 40) was placed longitudinally over the quadriceps tendon. Effusion was defined as a hypo/anechoic, displaceable and compressible collection in the suprapatellar recess [4, 5]. Effusion size was determined as an average of 2 screens captures as shown in Figure 1. Results were recorded as three categories, as follows: "0" if no effusion was present, "1" if there was an effusion of 0-4 mm thickness and "2" if the effusion thickness was over 4 mm.

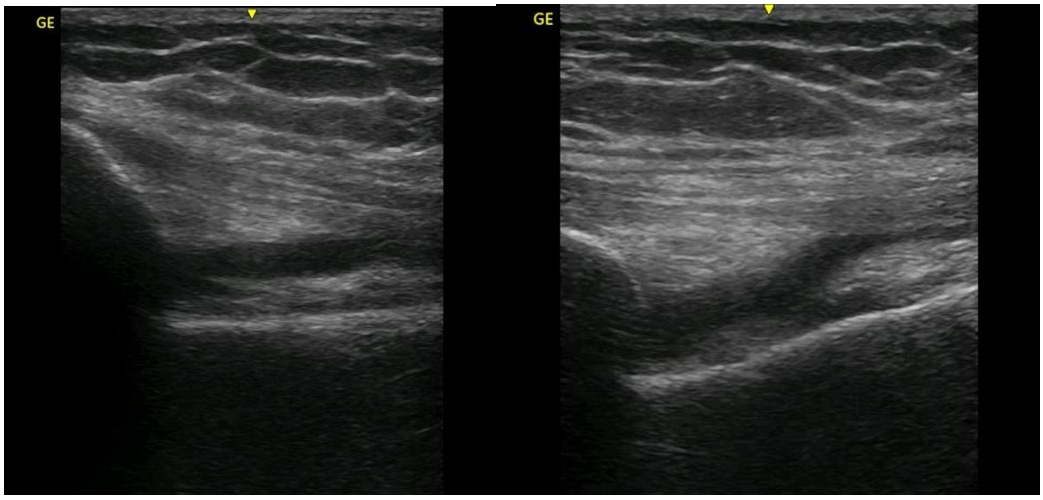

Figure 1. Ultrasound determination of suprapatellar effusion (thickness of 8mm, category 2)

The study protocol was approved by the Local ethics committee for scientific research and was conducted in accordance with the Declaration of Helsinki.

For the statistics software R, version 3.4.4 was used, with a significance level for alpha being 0.001. Unpaired t-test was used to determine the difference between two means. The backward selection procedure was used to optimize a multiple linear regression model with respond (dependent)

variable NLR, for all sets of data using R version 3.5.3 statistical software (https://www.r-project.org). Predictor variables were age, gender, Tegner-Lysholm scoring scale, IKDC, effusion, KOOSJR and EQ-5D Index and VAS.

3. Results and discussions

There were significant differences in demographics but not in NLR between the study patients and controls: age 54.1 versus 42.4 years old ($p<0.001$); M:F ratio 1:1.8 versus 1:5.2 ($p<0.001$); average NLR 2.03 versus 1.98 ($p=0.606$). There were 15 patients with no effusion (category 0), 52 with category 1 effusion and 12 with significant joint fluid (category 2). The control group consisted of 1260 subjects: 83.96% females, average age 42.4 years, (range 18-78). In Table 1 higher values of Tegner-Lysholm score,VAS and IKDC score indicate better health. Lower value of KOOSJR score indicate better health. Higher values of NLR (neutrophil to lymphocyte ratio) indicate more inflammation.

Table 1. Demographics and mean values ± SD of scores
(Tegner -Lysholm, VAS, IKDC, NLR, KOOSJR, effusion)

Age	54.1±11.6
M:F ratio	1:1.8
Tegner-Lysholm	46.8±18.89
IKDC	31.9±14.4
Effusion	0.96±0.58
KOOSJR	15.6±4.78
Index	0.51±0.26
VAS	63.4±19.5
NLR	2.03±0.81

Correlations between NLR and Age, Gender, Tegner-Lysholm knee rating scale, IKDC, Effusion, KOOSJR, EQ-5D-5L Index and VAS are presented in Table 2 with $p<0.01$

There was no correlation between NLR and joint effusion nor between NLR, joint effusion and any other parameter recorded in Table 2.

In the multiple regression model, NLR of patients with mild knee degeneration was only minimally influenced by age ($p=0.059$) and KOOSJR ($p=0.023$).

Table 2. Correlation matrix

	Age	Gender	Tegner	IKDC	Effusion	KOOSJR	Index	VAS	NLR
Age	1.000	-0.132	-0.332	-0.290	-0.010	0.372	-0.211	-0.179	-0.124
Gender	-0.132	1.000	0.188	0.307	0.138	-0.342	0.259	0.412	0.066
Tegner	-0.332	0.188	1.000	0.527	-0.068	-0.500	0.530	0.476	-0.090
IKDC	-0.290	0.307	0.527	1.000	0.102	-0.631	0.635	0.433	-0.134
Effusion	-0.010	0.138	-0.068	0.102	1.000	0.017	0.066	0.061	-0.121
KOOSJR	0.372	-0.342	-0.500	-0.631	0.017	1.000	-0.457	-0.483	0.190
Index	-0.211	0.259	0.530	0.635	0.066	-0.457	1.000	0.573	-0.031
VAS	-0.179	0.412	0.476	0.433	0.061	-0.483	0.573	1.000	-0.039
NLR	-0.124	0.066	-0.090	-0.134	-0.121	0.190	-0.031	-0.039	1.000

The aim of the article was to determine the association between neutrophil to lymphocyte-ratio (NLR), clinical disease severity and joint effusion in patients with early knee osteoarthritis. Studies have shown that NLR levels can be used as indicators of systemic inflammation supporting the inflammatory hypothesis of the pathogenesis of osteoarthritis [19]. A correlation between NLR levels,

clinical severity and joint effusion in early osteoarthritis could be of use in developing new anti-inflammatory treatments to slow down the progression.

In our analysis NLR was not associated with PROs or suprapatellar joint effusion. KOOSJR and age were the only independent predictors of NLR in the multivariate analysis. NLR was age dependent for both study subjects and controls.

Several studies found that age and NLR over 2.1 were predictive of advanced OA [11-14]. Tasoglu et al found that blood NLR ≥ 2.1 had 50 % sensitivity and 77 % specificity in predicting severe knee OA and in the multivariate analysis, age and blood NLR ≥ 2.1 were independent predictors of severe knee OA [11]. Mild to moderate OA was radiographically evaluated as Kellgren Lawrence stages 1 to 3, and severe OA stage 4. However, for mild knee degeneration there is limited radiographic correlation. Furthermore, new MRI protocols have been able to determine changes in cartilage composition even before evident morphological signs [1,2].

There are several limitations of our study. The main one is the low number of subjects. Different associations may become apparent or more robust with large, multicentric cohorts. We hypothesized that joint effusion would be the most likely clinical factor to be associated with inflammatory markers such as NLR. It was measured using ultrasound and divided into 3 categories. The cutoff value of 4 mm was arbitrarily chosen, based on previously published data [4,5]. The normal interval for NLR has been reported in the range of 0.78-3.53, with an average of 1.65 [20]. Our control group has a higher female ratio but is still within the range at average of 1.98.

Treatment options for early stages of OA are limited and evidence supporting them varies. Current options range from intraarticular instillations, filling of focal cartilage lesions to lower limb mechanical axis realignment. Rehabilitation procedures with stability and balance exercises may be applied for increasing mobility, strength and equilibrium [21]. A biomarker for early osteoarthritis highly corelated with clinical severity would be of great use for developing new treatments in early stages and for treatment follow up. Physical and chemical structure of joint have a great influence in disease progression and in the development of new types of treatments [22- 24].

4. Conclusions

Neutrophil to lymphocyte ratio is within normal limits in patients with early stage knee osteoarthritis and is not associated with clinical severity or with joint effusion.

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