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Predicting short cervix: Can neutrophil/lymphocyte ratio be used as a first-line screening method for short cervix?

Kısa serviks öngörüsü: Nötrofil/lenfosit oranı servikal kısalmayı öngördürücü bir birinci basamak tarama yöntemi olabilir mi?

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SUMMARY

Aim: Preterm birth is the leading cause of neonatal and infant mortality. It is recommended to measure the length of the cervix with transvaginal ultrasound in order to detect the patients at risk for spontaneous preterm delivery and to offer preventive treatment options. However, there is no definite consensus on cost-effectiveness of universal screening with this method in low- risk patients particularly in the absence of follow-up algorithms for those cases with 26-29 mm cervical length. The relationship between spontaneous preterm delivery, cervical insufficiency and intraamniotic infection/inflammation is well-known. In this study, the relationship of neutrophil/lymphocyte ratio, which is clinically used as an inflammation marker, with short cervix was investigated.

Material and Methods: This study is a retrospective observational study. 115 patients with short cervix detected by transvaginal ultrasound between 16-34 weeks were recruited as study group, and 94 patients with normal cervical length were taken as the control group. Multiple pregnancies, the patients who did not have a blood count performed one week before or after the ultrasound date, the patients with a positive cervicovaginal culture, the ones who had an active infection clinic according to the examination notes were excluded. Demographic data, hemogram parameters and neutrophil/lymphocyte ratios of the study and control groups were compared.

Results: There was no significant difference in terms of maternal age and parity in the study and control groups. Body mass index, number of pregnancies and number of previous preterm deliveries were higher in the short cervix (study) group than in the control group. Cervical length, gestational week at birth and birth weight were significantly lower in the study group than in the control group. Mean week of birth was 33.7 (±5.2) and birth weight was 2374.1 (±1027.5) in the study group, while it was 38.2 (±3.1) and 3410 (±760) in the control group. While the neutrophil and lymphocyte counts were not different between the groups, the neutrophil/lymphocyte ratio was found to be significantly higher in the study group than in the control group.

Conclusion: : Elevated neutrophil/lymphocyte ratio in mid-trimester is associated with short cervix. The gold standard in the diagnosis of cervical insufficiency is cervical length measurement with transvaginal ultrasound. However, this examination is not applied to everyone in daily practice. It should be emphasized to patients that this examination should not be abandoned in patients with a high neutrophil/lymphocyte ratio. In the low-risk group, the neutrophil/lymphocyte ratio can be used as a first-line screening method.

Keywords: Annulus fibrosus, curcumin, cytotoxicity, nucleus pulposus, primary cell culture, proliferation

ÖZET

Amaç: Preterm doğum, neonatal ve infant mortalitesinin en önde gelen nedenidir. Spontan preterm doğum açısından riskli hastaları yakalamak ve önleyici tedavi seçenekleri sunmak için transvajinal ultrason ile serviks uzunluğunun ölçülmesi önerilmektedir. Ancak düşük riskli hastalarda, özellikle servikal uzunluk ölçümü 26-29 mm çıktığında takip algoritmalarının olmaması nedeniyle, bu yöntemle evrensel taramanın maliyet etkinliği konusunda kesin bir fikir birliği yoktur. Spontan preterm doğum ve servikal yetmezlikle intraamniyotik infeksiyon veya steril inflamasyon ilişkisi bilinmektedir. Bu çalışmada inflamasyon belirteci olarak klinik kullanımı olan nötrofil/lenfosit oranının servikal kısalıkla ilişkisi araştırılmıştır.

Materyal ve Metotlar: Bu çalışma, retrospektif bir gözlemsel çalışmadır. Kliniğimizde takip edilmekte olup 16-34 haftalar arasında transvajinal ultrason ile servikal kısalık tespit edilen 115 hasta çalışma grubu, servikal uzunluğu normal olan 94 hasta kontrol grubu olarak alındı. Ultrason tarihinden 1 hafta öncesine ve sonrasına kadar kan sayımı yaptırmamış olan, servikovajinal kültüründe üreme olan, aktif enfeksiyon kliniği olan ve çoğul gebelikler dışlandı. Çalışma ve kontrol gruplarının demografik verileri, hemogram parametreleri ve nötrofil/lenfosit oranları karşılaştırıldı.

Bulgular: Çalışma ve kontrol gruplarında maternal yaş ve parite açısından anlamlı fark yok idi. Kısa serviks (çalışma) grubunda vücut kitle indeksi, gebelik sayısı ve önceki preterm doğum sayısı kontrol grubundan yüksek saptandı. Çalışma grubunda servikal uzunluk, doğumda gebelik haftası ve doğum kilosu kontrol grubundan anlamlı düşüktü. Çalışma grubunda ortalama doğum haftası 33,7 (±5,2) doğum ağırlığı 2374,1 (±1027,5) iken kontrol grubunda 38,2 (±3,1) ve 3410 (±760) idi. Nötrofil ve lenfosit sayısı gruplara arasında farksız iken, çalışma grubunda nötrofil/lenfosit oranının kontrol grubuna göre anlamlı olarak yüksek olduğu tespit edildi.

Sonuç: Gebelik ortası yüksek nötrofil/lenfosit oranı servikal kısalık ile ilişkilidir. Servikal yetmezliğin tanısında altın standart transvajinal ultrason ile servikal uzunluk ölçümüdür. Ancak bu muayene günlük pratikte herkese uygulanmamaktadır. Nötrofil/lenfosit oranı yüksek olan hastalarda bu muayeneden vazgeçilmemesi gerektiği hastalara vurgulanmalıdır. Düşük riskli grupta nötrofil/lenfosit oranı birinci basamak tarama yöntemi olarak kullanılabilir.

Anahtar kelimeler: Kısa serviks, nötrofil/lenfosit oranı, preterm doğum, servikal yetmezlik

INTRODUCTION

Preterm birth (PTB) is one of the leading causes of neonatal death and morbidity. It accounts for 70% of neonatal deaths and 36% of infant deaths. It is also the reason of 25-50% of the long-term neurologic impairment of the children (1). 12 to 18 millions of babies are born prematurely each year (2). The incidence is 10%, varying between the countries. Detection of the women who have high risk for spontaneous preterm delivery offers the therapeutic options such as progesterone therapies, cerclage, or pessary which are proved to reduce the PTB risk (3). However, in the absence of a prior spontaneous PTB, or cervical surgery, it is difficult to predict who is going to give a preterm delivery. Cervical length measurement with transvaginal ultrasonography (TVUSG) is recommended to detect those patients at risk of spontaneous preterm delivery. For those without a prior spontaneous PTB, universal cervical length measurement with TVUSG before 24th gestational weeks is the only recommended screening tool (4). Intraamniotic inflammation secondary to either intraamniotic infection or sterile intraamniotic inflammation is a very well-established cause of spontaneous preterm delivery (5). There are inflammation markers in amniotic fluid like interluekin-6 (IL-6), interleukin-8 (IL-8), interleukin-1α (IL-1α) and high mobility group box 1 (HMGB1) reported to be useful for the prediction of PTB (5). However, they are not available in all maternity units, and they necessitate invasive procedure which is not acceptable except for those who are already scheduled for an emergency cerclage. Therefore, even if they are highly accurate predictors for intraamniotic inflammation, they cannot be used as a routine test. On the other hand, neutrophil/lymphocyte ratio (NLR) has recently been shown a good marker for subclinical inflammatory diseases. There are studies reporting that it can be used in the prediction of spontaneous preterm delivery (6,7).

Considering the relationship between intraamniotic inflammation and cervical insufficiency, we aimed to investigate the utility of midtrimester maternal NLR in predicting short cervix.

MATERIAL AND METHODS

This study was designed as a retrospective observational study and conducted in a tertiary maternal fetal medicine center with the approval of Health Sciences University, Hamidiye Faculty of Medicine ethics committee (17.08.2023, no:29) The records of the pregnant women who were examined for cervical length on TVUSG between 16-34 gestational weeks were assessed on computerized hospital records and patient charts. It is our center's protocol to perform universal cervical screening to the patients during the fetal anomaly screening, or an opportunistic cervical screening to the women who first presented in the outpatient clinics for other reasons after the mid-trimester anatomic screening period. Singleton pregnancies who had been drawn blood sample within the two weeks of cervical length assessment were recruited and included in the study. Those with multiple pregnancies, Müllerian anomalies, and cervical operations were excluded. Symptomatic cervico-vaginal/uterine infection at the time of assessment and blood sampling, any symptoms, examination, or laboratory findings suggesting any other infections at the time of assessment, premature rupture of membranes (PPROM) before assessment and known systemic inflammatory diseases were also the reasons for exclusion. Demographic data and clinical information were all obtained from the clinical records and the patients with lack of anamnesis and clinical examination details were not included in the study to avoid any active infections which might have impacted on the cervical length. The patients were allocated into two groups as 'short cervix group' (those with a cervical length 25 mms) and 'normal cervical length' (those with cervical length >25 mm). Maternal age, gestational weeks at the time of assessment, gravidity, parity, number of previous spontaneous PTB or second trimester abortions, body mass index (BMI), white blood cell count, neutrophyl/lymphocyte ratio, gestational age (GA) at birth and neonatal birth weight (NBW) were compared between the two groups.

The normality of the data was confirmed using the Shapiro–Wilk test. The significance of between-group differences was evaluated using the independent-samples t-test. P value which one was below <0.05 considered significant. Statistical analyses were conducted with SPSS software (ver. 22.0; IBM Corp., Armonk, NY, USA).

RESULTS

A total of 94 patients were allocated to normal cervical length group and 115 patients were allocated to short cervix group. Mean maternal age was 29.7 (±5.4) and 30.7 (±5.25), respectively. Number of parities did not differ between the groups. Short cervix group had significantly higher BMI (28.7±4.92 vs 26.8±4.59); higher gravidity and higher rates of spontaneous PTB history (Table 1).

Table 1: Demographic features of the study and control groups

	Normal cervical length (G1) N=94	Short cervix (G2) N=115	*p*
Age	29.7 (± 5.4)	30.7 (± 5.25)	0.15
BMI	26.8 (± 4.59)	28.7 (± 4.92)	<0.01
Gravidity	2.43 (± 1.25)	3.06 (± 1.80)	<0.01
Parity	1.17 (± 0.94)	0.97 (± 1.08)	0.31
Previous PTB number	0.27 (± 0.55)	0.90 (± 0.87)	<0.01

BMI: body mass index, PTB: pretrem birth, p: significance value

There was no significant difference between the groups regarding the gestational weeks at which cervical length assessment and blood count performed (Table 2).

Table 2: Transvaginal cervical examination and pregnancy outcomes

	Normal cervical length N=94	Short cervix N=115	*p*
GA at TVUSG	24.4 (± 4.2)	23.0 (± 4.5)	0.02
Cervical length on TVUSG	35.5 (± 5.1)	13.1 (± 7.1)	<0.01
GA at birth	38.2 (± 3.1)	33.7 (± 5.2)	<0.01
BW	3410 (± 760)	2374.1 (± 1027.5)	<0.01

GA: gestational age, TVUSG: transvaginal ultrasonography, BW: birth weight

Not surprisingly, the gestational age at birth and neonatal birth weight were significantly lower in the short cervix group. When the inflammation markers in blood count were evaluated, neutrophil and lymphocyte count did not differ between the groups, however, total white blood cell count and neutrophil/lymphocyte ratio were significantly higher in the short cervix group (4.72±2.46) vs 3.93 (±1.19), p<0.01, 95% CI) (Table 3).

Table 3: Comparison of white blood cell count and neutrophyl/lymphocyte ratio between the patients with short cervix and normal cervical length

	Normal cervical length N=94	Short cervix N=115	*p*
WBC	10.47 (± 2.24)	11.67 (± 2.50)	<0.01
Neutrophyl	7.71 (± 2.00)	9.37 (± 2.35)	<0.25
Lymphocyte	1.98 (± 0.43)	2.22 (± 0.72)	<0.25
NLR	3.93 (± 1.19)	4.72 (± 2.46)	<0.01

WBC: white blood cell count, NLR: neutrophil/lymphocyte ratio.

DISCUSSION

Leading maternal-fetal health organizations have been dealing for improving the prevention strategies against PTB for many years. These strategies include reducing the number of high-order pregnancies as a result of artificial reproductive techniques, recommendations about optimal elective delivery timing for high-risk pregnancies, progesterone therapies for high-risk women. However, spontaneous preterm delivery is still a huge burden on health care. Cervical length measurement is shown to be a good predictor of those who have higher risk for spontaneous preterm delivery (8). Transvaginal ultrasound is a superior method for optimal measurement of cervical length comparing with transabdominal and translabial route (9) and has been recommended over a decade by guidelines (3). As it is reported that the sensitivity and positive predictive value of transvaginal cervical length measurement for predicting PTB is lower for those who don't have a prior PTB history, and due to the conflicting results of the studies about cost- effectiveness of universal screening with TVUSG in low-risk group, ACOG and SMFM recommends visualization of cervix during the fetal anatomic assessment at 18-23 weeks, but not strictly recommends universal screening in low-risk group (10). However, this group constructs the largest population of spontaneous PTB. Moreover, the acceptance of TVUSG among the low-risk patients is questionable as most women find the process uncomfortable (11,12).

Beyond the efforts to identify the high-risk pregnant for spontaneous PTB, the underlying etiology still remains indefinite. There are various complex hypotheses for the etiology of spontaneous preterm labor, including genetic predisposition, hormonal and environmental factors, trauma, tissue mechanics, immune factors, and microbial factors (13). One of the most prominent and well- proved mechanism is intraamniotic infection. Intraamniotic infection is present in 50% of the women with cervical insufficiency (14). Besides the microbial or infectious inflammation of amniotic cavity, there are several theories about the maternal and fetal immune mechanisms leading to preterm birth. These mechanisms trigger both maternal systemic inflammatory response and inflammation in maternal-fetal interface, leading to the onset of preterm labor (5). Neutrophils are shown to be the first to respond to the amniotic infection by phagocytosis, release of immune mediators and antimicrobial products (4,5). Neutrophil/lymphocyte ratio is an emerging marker of the relationship between various diseases and human immune system (15). It is also reported to be a sensitive marker combined with the cervical length for identifying the women at risk of spontaneous preterm delivery (16). In this study, the exclusion of the women with active cervico-vaginal infection or chorioamnionitis clinic and the ones with positive cervico-vaginal culture supported the suggestion that non- microbial sterile inflammation has an important role in cervical insufficiency, NLR is capable to detect those who do not have obvious infection, and it worths to be screened in all midtrimester cases.

The most accurate test to diagnose intraamniotic infection is microbial culture, IL-6, white blood cell count and gram stain of the amniotic fluid (17). However, as these tests require invasive procedure, they cannot be an option as a routine method. Therefore, it can be beneficial to use a noninvasive, cheap, widespread, and simple marker of intraamniotic inflammation. This study showed that midtrimester maternal blood NLR is significantly higher in the pregnant women with short cervix compared to the ones without short cervix. It did not reach to any conclusion about the specifity of NLR. It is reasonable to assume that it cannot be a specific marker. However, as the recommendations in favor of universal cervical length screening consolidate, and patients are not that volunteer for TVUSG, it is important to at least identify the ones who should not refrain to have TVUSG assessment as the gold standard examination.

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