RESEARCH ARTICLE

Predictive Role of the Neutrophil Lymphocyte Ratio for Invasion with Gestational Trophoblastic Disease

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Abstract

Purpose: The objective of this study was to assess the predictive role of the neutrophil/lymphocyte ratio (NLR) for invasion of gestational trophoblastic disease (GTD). Materials and Methods: A retrospective analysis was conducted on 127 women who were managed at our clinic for GTD. Of all patients, 8 showed invasion according to histological examination. The clinical parameters of patients with invasive GTD (Group 1; n=8) were compared with patients who showed no invasion (Group 2; n=119). All underwent a prior uterine evacuation and followed up by regular assessment of β-hCG titers. Results: Demographic and obstetric history and pre-evacuation hCG levels of the patients showed no statistically significantly difference between the groups (p>0.05). The mean gestational weeks (GW), size of the GTD and NLR levels were statistically significantly higher in the invasive GTD group (p<0.05). Correlations between invasion and gestational weeks, size of GTD, post-evacuation chemotherapy and NLR were evident. ROC curve analysis demonstrated that GW, size of GTD and NLR may be discriminative parameters in predicting invasion of GTD. Conclusions: To the best of our knowledge, this is the first study evaluating the predictive role of NLR in invasion of GTD. In conclusion ,we think that pretreatment NLR can be used as a biomarker of invasion in GTD.

Keywords: Gestational trophoblastic disease - neutrophil lymphocyte ratio - marker - invasion

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Introduction

Gestational trophoblastic disease (GTD) is a spectrum of pregnancy related trophoblastic abnormalities ranging from the premalignant complete and partial hydatiform mole to the malignant invasive mole, choriocarcinoma and placental site trophoblastic tumor (Shanbhogue et al., 2013). The treatment modalities include suction dilation and curettage and combination chemotherapy in case of malignancy (Manopunya et al., 2012; Oranratanaphan et al.,2014). Invasive mole that follows complete and partial hydatiform mole is characterized with invasion of the molar villi to the myometrium. The ratio of invasion following complete hydatiform mole is approximately 15 % and 3 to 5 % in partial mole hydatiform. The diagnosis of invasion generally depends on a plateau or elevation of hCG levels after molar evacuation but invasion may also be detected by hystopathological examination (Goldstein et al., 2012). Previous studies reported different markers to predict malignant transformation of GTD such as; E-cadherin and integrin β-1 (Shu et al., 2013), interleukine 12 (Zhang et al., 2012) and CLIC1 protein (Shi et al., 2011).

NLR is a simple and easily calculated marker obtained from the differantial white blood cellcount. NLR has been reported as predictive markers in outcomes of endometrial precancerous and cancerous lesions in patients with abnormal uterine bleeding, other cancers and coronary artery disease (Duffy et al., 2006; Halazun et al., 2009;Dirican et al., 2013; Karaman et al., 2013; Unal et al., 2013; Yucel et al., 2013; Acmaz et al., 2014).

To the best of our knowledge, there is no study in the literature evaluating the predictive role of NLR in invasion of GTD. Therefore, we conducted this retrospective analysis to determine if pretreatment NLR may be a diagnostic marker in invasion of GTD.

Materials and Methods

Ethical approval for the entire study was obtained from the Ethics committee of Dr. Zekai Tahir Burak Women's Health Education and Research Hospital. This is a tertiary referral research and education hospital in Ankara, Turkey. Due to the retrospective design, informed consent was not obtained.

The study included a total of 127 women with GTD managed at our gynecological oncology department. Of all women; 99 had complete hydatiform mole, 20 partial

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hydatiform mole and 8 invasive mole. The data of the cases were collected from hospital records and patient files. The clinical characteristics evaluated were age, gravidity, parity, size of the GTD, pre-evacuation β-hCG levels, NLR, post-evacuation chemotheraphy and histopathology of the evacuated specimens.

All of the patients after the initial evaluation that included a general and a gynecological and obstetric history, vital signs were recorded. The patients were also assessed clinically with a Doppler ultrasound scan (Aloka Co., Tokyo, Japan) of the pelvis, a chest X-ray (CXR) and an updated serum hCG level pretreatment. All of the patients underwent a prior uterine evacuation and followed up by regular β-hCG titers. Invasive mole was assessed by pre-evacuation Doppler sonography and corrected by hystopathological examination.

Statistics

Means and standard deviations (SD) were calculated for continuous variables. Subject characteristics and demographics were analyzed descriptively. The normal distribution of the variables was analyzed by the Kolmogorov-Smirnov test. The Chi-square (χ^2) test and the Student's t test were used to evaluate associations between the categorical and continuous variables. Logarithmic transformation (log10) was performed to correct the variance of β-hCG levels as the range of those distributions was large. Pearson correlation analysis was used to find the correlation between invasion of GTD and gestational weeks, size of GTD, post-evacuation chemotherapy and NLR. ROC curve analysis was used to assess the discriminative role of gestational weeks, size of GTD and NLR levels. All variables were included in the backward stepwise procedure. Two-sided P values were considered statistically significant at P\0.05. Statistical analyses were carried out using the statistical package SPSS 15.0 for Windows (SPSS Inc., Chicago, IL, USA).

Results

Table 1 showed the demographic and clinical features of the patients between the groups. The mean age of the patients in group 1 was 27.25±5.97 years old and in group 2 30.25±8.59 years old. The median gravidity and parity of the cases were similar between the groups. There was no statistically significantly difference between the groups in terms of pre-evacuation β-hCG levels (p>0.05). The mean gestational weeks was 9.75±3.24 weeks in group 1 and 7.57±1.85 in group 2 and the mean size of GTD was 12.62±4.20 cm in group 1 and 8.93±3.75 cm in group 2 and there were statistically significantly difference between the groups. The mean NLR levels was 6.43±4.31 in group 1 and 3.38±1.92 in group 2 (p<0.05).

There was a correlation between invasion of GTD and gestational weeks, size of GTD, post-evacuation chemotherapy and NLR as shown in Table 2.

ROC curve analysis (Figure 1) demonstrated that GW, size of GTD and NLR may be discriminative parameters for invasion of GTD. The area under curve (AUC), cut off values and sensitivity and specificity of ROC curve are depicted in Table 3. The AUC (Cut off value) for GW, size of GTD and NLR was; 0.561 (5.5), 0.745 (14.5) and 0.711 (8.96); respectively.

Table 1. The Demographic and Clinical Characteristics of the Patients in Invasive GTD and Non-Invasive GTD Group

	Invasive GTD (n=8)	Non-invasive GTD (n=119)	P
Age (mean±SD)	27.25±5.97	30.25±8.59	0.334
Gravidity	2 (1-8)	3 (1-6)	0.269
Parity	1 (0-6)	1 (0-4)	0.150
Gestational weeks	9.75±3.24	7.57±1.85	0.003
Log β- hCG levels	5.13±0.30	5.05±0.36	0.421
Size of GTD (cm)	12.62±4.20	8.93±3.75	0.008
NLR	6.43±4.31	3.38±1.92	<0.001

GTD: Gestational trophoblastic disease, SD: Standard deviation, hCG: human chorionic gonadotropine, NLR: neutrophil lymphocyte ratio

Table 2. Correlation between Invasion of GTD and Clinical Parameters

	GW	Size of GTD	Post-evacuation chemotherapy	NLR
Invasion of GTD	CC=0.263 p 0.003	CC=0.233 p 0.008	CC=0.304 p=0.001	CC=-0.332 p<0.001

GTD: Gestational trophoblastic disease, GW: Gestational weeks, NLR: neutrophil lymphocyte ratio

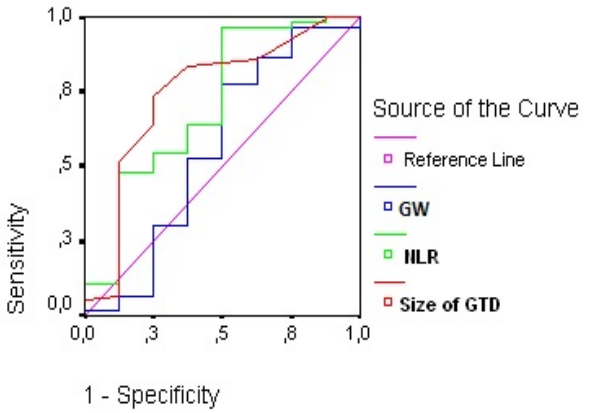

Figure 1. ROC Curve Demonstrating the AUC of GW, Size of GTD and NLR for Invasion of GTD

Table 3. The Area Under Curve (AUC), Cut off Values and Sensitivity and Specificity for Gestational Weeks, Size of GTD and NLR in Invasive GTD Patients

	AUC	SE	95%CI	Cut off value	Sensitivity% -specificity%
Size of GTD (cm)	0.745	0.104	.542-.948	14.5	85.7-62.5
NLR	711	0.109	.497-.925	8.96	96.6-75.0
GW (weeks)	0.561	0.131	.304-.818	5.5	92.2-87.5

GTD: Gestational trophoblastic disease, GW: Gestational weeks, NLR: neutrophil lymphocyte ratio

Discussion

This study has some important aspects. First, it is the first study to evaluate the NLR as a predictive factor for invasion in patients with GTD. Second, it was found that NLR ratio has high sensitivity and relatively high spasticity in prediction of GTD invasion. Third, by combining NLR with preevacuation size of GTD, the

clinicians can categorize patients due to their invasion probabilities with high sensitivity.

Invasive GTD comprises a group of aggressive fertilization disorders characterized by invasion of the uterine endometrial and myometrial layers by malignant trophoblastic cells. The exact pathogenesis of this process is still unrevealed. Some investigators have suggested that immunologic factors have important role invasion mechanism. Zhang et al. (2012) reported that IL-12 inhibited cell invasion through regulating the expression of matrix metalloproteinases (MMP)-9 and tissue inhibitors of metalloproteinases (TIMP)-1 in choriocarcinoma. In another study by Prabha et al. (2001), it was indicated that increased expression of interleukin-1 beta in the villous cytotrophoblasts and the stromal Haufbaur cells in molar placentae, was associated with persistence of the disease and invasion in complete hydatidiform moles. These kind of cytokines including growth factors or interleukines may also contribute to the accumulation of neutrophils (Hotchkiss et al., 2003). Increased neutrophils levels inhibit the lymphocyte activity and stimulate lymphopenia by increasing lymphocytes apoptosis (Yoon et al., 2013). This is the physiological immune response of circulating leukocytes to various stressful events such as inflamation or malignancy which is characterized by an increased neutrophil count and decreased lymphocyte count (Wu et al., 2011).

Increased neutrophil counts have been observed in patients with solid tumors (Gabrilovic et al., 2009). Neutrophils have ability to suppress T-cell function (Movahadi et al., 2008;). In addition to this immune suppression, neutrophils may have additional tumor-promoting ability. CXCL1/MIP-2, an angiogenic chemokine, is associated with neutrophil recruitment and induces vascular endothelial growth factor production in neutrophils, resulting in angiogenesis in vivo and stimulates neutrophil recruitment (Scapini et al., 2004). Finally, infiltration with large numbers of peritumoral neutrophils is associated with progression of angiogenesis at the edge of hepatocellular carcinoma (Kuang et al., 2011). These observations support that neutrophils may participate in GTN invasion, angiogenesis, and metastasis as in cancer process.

We found that NLR was significantly higher in patient with invasive GTD than in patient with non-invasive GTD. And also NLR was a significant discriminative parameter in predicting GTD invasion with high sensitivity and relatively high spesifity. These results may reflect the hidden connection between the NLR and immunological pathogenesis of GTD invasion.

In our study, we also found that preevacuation GTD size should be considered as a significant factor for GTD invasion. However, GTD size has less predictive value than NLR. We believe that further studies are needed to decide the exact effect of GTD size in prediction of GTD invasion. However, we have some limitations. The retrospective nature of the study design is the main limitation. In addition the short follow-up period, and the relatively small sample size of the test participants are the other ones.

In conclusion, NLR derived from a single blood sample during the initial diagnostic stage of GTD is a very useful laboratory marker for discriminating patients with invasive GTD from patients with non invasive GTD. This simple, available parameter can be easily used in clinical practice.

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