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Influence of CA125, platelet count and neutrophil to lymphocyte ratio on the immune system of ovarian cancer patients

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HIGHLIGHTS

- Immune profile in HGSOC is correlated to CA125, NLR and thrombocytosis at diagnosis.
- A high NLR is correlated to poor prognosis and an immunosuppressive profile.
- IL-10 is increased in patients with CA125 > 500 kU/L or thrombocytosis (>400 × 10^9/l).

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ABSTRACT

Objective. The effect of CA125, neutrophil to lymphocyte ratio (NLR) and thrombocytosis on survival has been studied in ovarian cancer. This study explores the link between these variables and serum markers of ovarian cancer patients, such as signaling proteins and cytokines.

Methods. Serum samples of 39 patients with high-grade serous ovarian cancer (HGSOC) were collected at diagnosis and were retrospectively analysed for clinical characteristics, clinical parameters (NLR, CA125, platelet count) and immune profile [IL-4 (interleukin), IL-10, IL-13, IL-17, transforming growth factor-β, Arginase-1, Interferon gamma), vascular endothelial growth factor (VEGF), galectin-1 and chemokine (C-C) motif ligand 2.

Results. CA125 correlates negatively with VEGF ($p = 0.02$) and if CA125 rises above 500 kU/L, IL-10 is significantly increased ($p = 0.01$). NLR > 6 ($p < 0.01$) was significantly correlated with decreased overall survival. Thrombocytosis was significantly correlated with IL-10 ($p < 0.01$) and a platelet count > 400 × 10^9/l led to an improvement in progression free survival ($p < 0.01$).

Conclusions. A correlation, at the time of diagnosis, of HGSOC between CA125, NLR and thrombocytes and an immunosuppressive cytokine-profile in serum is shown, and correlates with survival.

1. Introduction

Ovarian cancer is the second most lethal type gynecological tumor in women and has an incidence rate of 12.5 per 100,000 women [1]. It is a silent killer, metastasizing throughout the abdomen before causing symptoms. Consequently, most patients are diagnosed at a late stage, which leads to a poor prognosis. The most common histological subtype is high-grade serous ovarian cancer (HGSOC). Surgery in combination with platin-based chemotherapy is the cornerstone of therapy, but treatment is evolving towards targeted therapies. In the search for biomarkers in general, an extensive research has shown the correlation of variables such as CA125, platelet count and neutrophil to lymphocyte ratio (NLR) with survival, in various cancer types including in ovarian cancer (Table 1 [2–17]).

CA125, also known as MUC16, is a high molecular weight glucoprotein, expressed by 80% of epithelial ovarian cancers [18]. Serum CA125 levels are commonly determined in the follow-up of ovarian cancer to detect relapse or evaluate the response to treatment. In addition to this, we know from retrospective analysis of the Gynecological Oncology Group (GOG) 111, 114, 132, 152, 158, 162, and 172 trials, that doubling of CA125 is an independent prognostic factor for progression free survival (PFS) in a multivariable analysis with a hazard ratio (HR) of 1.07 and a 95% confidence interval (95% CI) of 1.04–1.10 [$p < 0.001$] [19].

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Table 1
Overview of neutrophil to lymphocyte ratio (NLR) and the results concerning survival and mortality in multiple tumor types.

Tumortype	Moment of serum sample*	NLR cut-off point	Univariate analysis**	Multivariate analysis***	Conclusion
Non-small cell lung cancer [2]	Diagnosis	/	$p = 0.001$	$p = 0.005$	NLR is an independent predictor for survival in patients with completely resected NSCLC
Hepatocellular carcinoma [3]	Diagnosis	NLR ≥ 5	$p < 0.0001$ (a) $p < 0.001$ (b)	$p < 0.0001$ (a) $p < 0.0001$(b)	NLR is a prognostic factor for recurrence and death.
Pancreatic cancer [4]	Diagnosis	NLR ≥ 5	/	/	NLR < 5 had a significant increased median survival compared with NLR ≥ 5 and it was the only significant peripheral blood marker. NLR can be used as a prognostic factor in pancreatic cancer patients
Breast cancer [5]	Diagnosis	NLR > 3.3	$p < 0.0001$	$p = 0.0138$/ $p = 0.0018$ (c)	NLR > 3.3 is an independent predictor of short and long term mortality
Gastric cancer [6]	No exact timing. In patients with stage IV gastric cancer, treated by S1 (oral fluoropyrimidine)	NLR ≥ 2.5	$p < 0.001$	$p = 0.03$	NLR is an independent prognostic factor in advanced gastric cancer and can be useful for patient survival
Uterine sarcomas [7]	Diagnosis	NLR > 2.12	/	/	Compared to CA125, NLR was a more powerful predictor for diagnosis of uterine sarcomas NLR was a more accurate predictor of recurrence and progression compared to CA125
Colorectal liver metastasis [8]	Diagnosis	NLR > 5	$p = 0.019$	$p = 0.005$	NLR > 5 is an independent predictor of worse survival
Metastatis castration - resistant prostate cancer [9]	From day 1 of the first cycle in TROPIC trial (prior docetaxel-containing chemotherapy)	Continuous NLR > 3 (d)	$p < 0.0001$	$p = 0.0001$	NLR is an independent predictor of overall survival
Ovarian cancer (all types) [17]	Diagnosis	NLR ≥ 2.6	$p = 0.004$ (b)	$p = 0.041$ (b)	NLR is a significant predictor for overall survival
Epithelial ovarian cancer [10]	Diagnosis	NLR ≥ 2.6	$p = 0.019$ (a) $p = 0.052$ (b)	p not significant	NLR tend to be a prognostic factor more for progression free than overall survival
Ovarian cancer (all types) [11]	Diagnosis	Continuous	$p = 0.003$ (b)	$p = 0.02$ (b)	NLR is an independent predictor of worse survival
Serous ovarian cancer [12]	Diagnosis	NLR in quartiles (4th quartile NLR > 3.77)	$p < 0.001$ (a) $p < 0.001$ (b)	$p < 0.001$ (a) $p < 0.001$ (b)	High NLR is an independent predictor of progression free and overall survival
High grade serous ovarian cancer [13]	Diagnosis	NLR ≥ 3.24	$p < 0.001$ (a) $p < 0.001$ (b)	$p = 0.011$ (a) $p = 0.148$ (b)	NLR is an independent prognostic factor for progression free survival, not for overall survival
Epithelial ovarian cancer and neoadjuvant chemotherapy [14]	Diagnosis	NLR ≥ 3.02	/	$p = 0.002$ (a) $p = 0.007$ (b)	NLR is a predictive factor for chemotherapeutic response and for survival in epithelial ovarian cancer patients with platinum-based chemotherapy
Epithelial ovarian cancer [15]	Diagnosis	NLR ≥ 0.89 (PFS) NLR ≥ 2.96 (OS)	$p = 0.0002$ (a) $p = 0.02$ (b)	$p = 0.002$ (a) $p = 0.11$ (b)	NLR is an independent predictive factor for progression free survival, but not for overall survival
Ovarian cancer (all types) [16]	Peri-operatively	NLR ≥ 5.25	$p < 0.001$	$p = 0.002$	NLR was a significant predictor of overall survival

Legend: NLR: neutrophil to lymphocyte ratio; NSCLC: non-small cell lung cancer.
* Diagnosis (before surgery, (neo)adjuvant chemotherapy or radiotherapy).
** Survival/short term and long term mortality/risk of death in cancer patients.
*** Survival/Short-term and long-term mortality/risk of death in cancer patients. a. Significant *p*-values concerning disease-free survival. b. Significant *p*-values concerning overall survival. c. The researchers used two separate Cox proportional hazard multivariate models to avoid combining T stage and AJCC stage (American Joint Committee of Cancer) in one model because there are highly correlated with each other. d. The study also calculated a valid cut-off for NLR based on comparing c-statistic values of NLR values representing approximately the median (NLR3) and first (NLR2).

NLR is a ratio for inflammation, determined by dividing the number of neutrophils by the number of lymphocytes in a peripheral blood sample. Recently two meta-analyses were performed independently investigating the prognostic role of NLR at diagnosis of ovarian cancer [20,21]. Both studies concluded that NLR above the cut-off (range = 1.90 to 4.68) was correlated with poor overall survival with a HR of 1.53 [20,21]. NLR remained an important variable throughout the disease course, predicting survival upon treatment with checkpoint-inhibitors in phase I studies for solid malignancies, including ovarian cancer. In a multivariate analysis NLR > 6 was significantly correlated with poor survival ($p < 0.034$) after treatment with immune checkpoint-inhibitors with a HR of 1.75–95% CI 1.04–2.94 [22].

Thrombocytosis is identified in 20–50% of ovarian cancer cases. A high number of platelets (>400 × 10^9/l) is correlated with poor overall survival (OS) in a retrospective series from the Vanderbilt University Medical Center with a HR 2.01 (95% CI 1.25–3.23). Preclinical evidence also suggests a link between thrombocytosis and response to chemotherapy in ovarian cancer. In a xenograft model for ovarian cancer, both depletion of platelets and administration of Docetaxel resulted in a significant reduction in tumor volume. However, the effect of Docetaxel on the *in vivo* tumor growth of A2780 and SKOV3 in nude mice was lost ($p = 0.55$) when platelet transfusions were administered to the mice by decreasing tumor cell apoptosis [23]. This suggests a role for platelets in the *in vivo* efficacy of taxane-based chemotherapy in ovarian cancer.

In addition to this, the tumor microenvironment proves also to be an important biomarker in ovarian cancer [24,25]. The presence of intratumoral T cells correlates significantly with both PFS and OS, even after correction for disease response or extent of residual disease after cytoreductive surgery [26]. Furthermore, Curiel et al. showed that infiltration of the tumor by regulatory T cells (Treg) correlates significantly with survival, even after controlling for tumor stage and cytoreduction [27]. More recently, a correlation between MDSC ($CD33^+$) infiltration and overall survival was observed HR = 2.87, 95% CI: 1.11–7.42 [28]. These studies have all characterized the intratumoral immune phenotype of ovarian cancer. However, this is an important disadvantage of these studies as ovarian cancer is a widespread metastatic disease and displays a large variety of heterogeneity both genetically and immunologically [29,30]. Therefore it might be advantageous to use immune markers in peripheral blood to characterize the immune profile of patients [31]. Our group analysed cytokines in serum produced by immune cells and linked them to survival in ovarian cancer patients at

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