Evaluation of osteopontin and CA125 in detection of epithelial ovarian carcinoma

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Summary

The objective of this study was to determine the potential of osteopontin (OPN) and OPN + CA125 (cancer antigen 125) combination in differential diagnosis of the ovarian cancers and non-malignant ovarian disease. Serum and plasma samples were obtained preoperatively from 79 women undergoing surgery for pelvic mass; 48 of them had ovarian carcinoma, and 31 had benign cyst. The samples were analyzed for the levels of OPN and CA125 (using ELISA and CMIA methods) and then compared with the final pathologic results. The median plasma level of OPN in patients with benign and malignant cysts was 356.33 ng/ml and 865.15 ng/ml, respectively ($p < 0.001$). Receiver operating characteristic (ROC) analysis for plasma OPN revealed the area under the curve (AUC) of 0.838. At the predefined specificity of 90%, OPN showed sensitivity of 62.5%, whereas the combination of OPN + CA125 reached 74.9% at the same specificity.

Key words: Ovarian cancer; Osteopontin; CA125; Tumor markers.

Introduction

It is well-known that an ovarian cyst may be diagnosed in a large number of women, even 20%, at some point in their lifetime. Accurate diagnostics aim to differentiate benign cysts from malignant ovarian tumors and provide adequate and fast treatment of patients. It is believed that 5% - 10% of women present for surgical interventions due to suspected ovarian neoplasm, out of which ovarian malignancy is verified in only 13% - 21% of these [1]. Conversely, if ovarian cancer (OC) is not detected at an early stage, the odds for successful treatment would be very low [2]. Most commonly used and currently best-studied tumor marker for diagnosis of ovarian neoplasms is cancer antigen 125 (CA125), but its utilization is limited by insufficient sensitivity and specificity.

Osteopontin (OPN) is another potential cancer biomarker. It is secretory glyco-phosphoprotein present in all body fluids and in the extracellular matrix. OPN is involved in cellular signaling pathways related to adhesion, cell migration, prevention of apoptosis, and neovascularization [3]. Its role in tumor genesis and progression of metastases has been confirmed.

This prospective study was designed to evaluate the significance of osteopontin in diagnostics of women presenting with suspected cystic pelvic mass and to summarize the combination of osteopontin and CA125 in detection of ovarian cancer.

Materials and Methods

The study included 79 subjects whose age ranged from 25 to 80 ($\bar{x} = 59.3 \pm 13.8$) years, and treated at the Clinic of Gynecology and Obstetrics, Clinical Center of Serbia, between January 2008 and December 2010 (Table 1). The study group included women with an ovarian cyst or pelvic mass, who were scheduled for surgery to ensure the correct diagnosis of possible epithelial ovarian cancer. Histopathological analysis, presented in Table 2, showed that, out of a total number of patients, 31 (39.2%) had benign cysts and 48 (60.8%) had epithelial ovarian cancer, [12 (15.2%) - Stage I, six (7.6%) - Stage II, 23 (29.1%) - Stage III and 7 (8.9%) - Stage IV].

Plasma OPN was measured by enzyme-linked immunosorbent assays (ELISA Kit), and serum CA125 using a chemiluminescent enzyme immunoassay (CMIA) test on the Architect i System.

Statistical methods used ANOVA for testing the differences between the benign and malignant group, histopathological subgroups of malignant group, and FIGO stages. The applied non-parametric tests were as follows: Kruskall-Wallis test and Mann-Whitney *U* test. Linear regression model was used to determine the relationship between the biomarkers. Receiver operator characteristic (ROC) curves were plotted for each biomarker in relation to benign and malignant tumors. SPSS Statistics software (ver. 15) was used for all statistical analyses. Statistical significance was set at value of $p < 0.05$.

Results

Biomarkers osteopontin and CA125 were determined in collected blood samples and it was found that the values of both biomarkers were significantly higher in patients with malignant cysts in comparison to the benign group ($p < 0.001$). There was also a significant difference between the median value of these two markers in the

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Table 1. — *Characteristics of patients (n = 79).*

	No. of patients	%	*p* value
Diagnostic category			
Benign	31	39.2%	0.056
Malignant	48	60.8%	
Age at diagnosis (years)			
< 30	4	5.06%	< 0.001
31 - 40	5	6.33%	
41 - 50	11	13.92%	
51 - 60	17	21.52%	
61 - 70	26	32.92%	
> 70	16	20.25%	

Table 2. — *Histology of cancer.*

Cancer Stage	FIGO I	FIGO II	FIGO III	FIGO IV	Total
Undifferentiated			1		1
Mucous	4		3	2	9
Serous	4	3	11	4	22
Mixed	2		1		3
Clear cell		1	1		2
Endometrioid	2	2	6		10
Brenner tumor				1	1
Total	12	6	23	7	48

Table 3. — *Descriptive statistics of serum OPN and CA 125 in benign and cancer patients.*

Biomarker	Groups	n	Median	Min	Max	*p* value
OPN	Benign	31	356.33	56.70	1,000.80	$p < 0.001$
	EOC	48	865.15	89.07	4,512.30	
	Stage I / II	18	415.25	89.07	1,220.00	$p < 0.001$
	Stage III / IV	30	1,445.50	379.75	4,512.30	
CA125	Benign	31	30.10	5.90	141.80	$p < 0.001$
	EOC	48	997.85	15.40	2,972.30	
	Stage I / II	18	94.85	15.40	2,600.32	$p < 0.001$
	Stage III / IV	30	1,447.96	125.40	2,972.30	

early phase of disease (Stage I/II) and late phase (III/IV) of epithelial ovarian cancer (EOC) ($p < 0.001$), (Figure 1). Table 3 illustrates the descriptive statistics of OPN and CA125 in blood of patients with the benign cysts, as well as patients with various stages of ovarian cancer.

Mann-Whitney test confirmed that there was no significant difference of OPN values between different histological types of tumors (Figure 2).

Linear regression analysis showed that there was a low correlation coefficient between the OPN and CA125 values in the group with malignant cysts ($r^2 = 0.1489$), and slightly higher correlation coefficient between the OPN and CA125 values in the group with benign cysts ($r^2 = 0.1522$) (Figure 3). The low correlation coefficient indicated that a combination of these two biomarkers would improve their individual abilities for cancer detection.

ROC analysis of preoperative plasma OPN and CA125 for all patients are presented in Figure 4. The area under the curve (AUC) for OPN was slightly inferior [AUC = 83.8%; 95% CI (75 - 92.5%)] to area under the CA125 [AUC = 90.3%; 95% CI (83.7 - 96.8%). At the predefined specificity of 90%, OPN and CA125 showed sensitivity of 62.5% and 72.6%, respectively, whereas the combination of OPN + CA125 reached 74.9% at the same specificity. Arbitrary cut-off level for plasma osteopontin was 650 ng/ml.

Discussion

Ovarian cancer is one of the most common reproductive cancers and has the highest mortality rate among gynecologic cancers. Most of ovarian cancer diagnoses occur in the late stages of the disease and five-year survival rates fall below 20%. To overcome the significant mortality associated with ovarian cancer, research on the clinical significance of new sensitive and specific biomarkers/panels of biomarkers are still very important.

In this paper, the authors reported that plasma OPN could augment CA125 detection, providing higher sensitivity and specificity in predicting ovarian cancer. With a sensitivity level of 62.5% alone (specificity 90%) OPN may have a lower potential than CA125 to accurately detect the presence of ovarian cancer. High sensitivity was achieved, reaching 74.9% (specificity 90%) when OPN was combined with CA125 in a biomarker screening panel. The obtained results show better characteristics of OPN as a tumor marker from the one that was given from Nakae *et al*. [4]. Regarding the present samples, there was no significant difference of plasma OPN concentration in different histological types of tumors, suggesting that all histological EOC types have increased plasma level of OPN. This is in agreement with the findings of Tiniakos *et al*. [5]. However, the authors proved that plasma OPN was significantly elevated during advanced stages of the disease, but there was also border significance between benign patients and early stage of disease. All these results suggest the potential use of plasma OPN and CA125 serum values for ovarian cancer diagnostic.

Ovarian cancer is known as the "silent killer", with very weak, nonspecific symptoms. For this reason, using a non-invasive approach, such as tumor markers for detection of the disease, is still very attractive. A number of proteins present in either blood or urine have been identified as specific markers for epithelial ovarian cancer [6, 7]. However, no single protein has provided adequate sensitivity and specificity for distinguishing malignant from benign pelvic masses. Some recent studies described panels of biomarkers that beside OPN had four [8], five [9], or more biomarkers [10] with high sensitivity and specificity for ovarian cancer detection. This present study suggests that measuring of preoperative plasma OPN and CA125 could provide a cost-effective and sensitive test in triage women with pelvic mass and therefore, reduce disease-associated mortality.

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Fig. 1

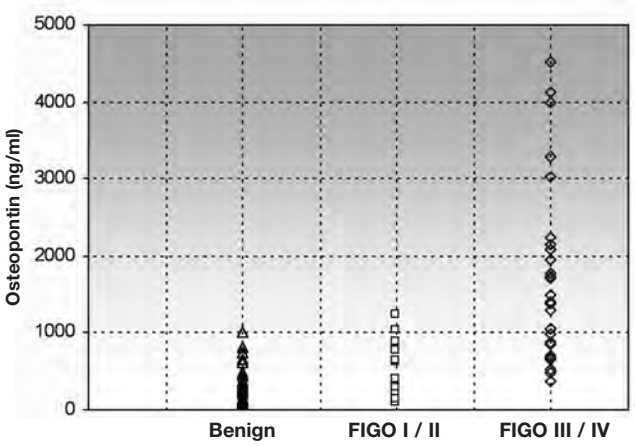

Fig. 2

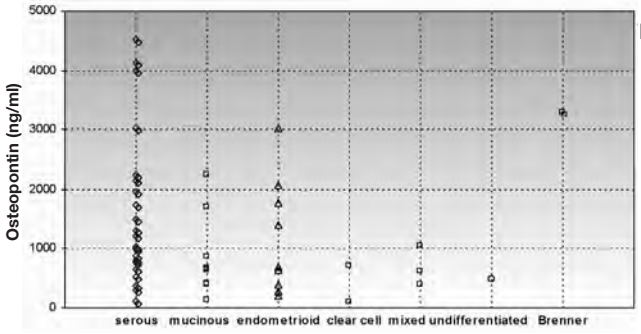

Fig. 3

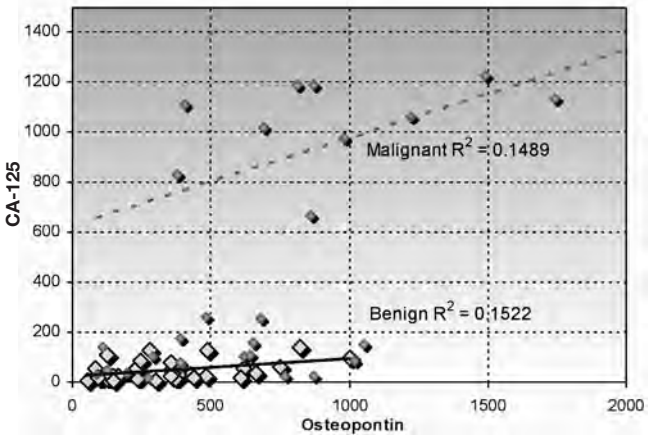

Fig. 4

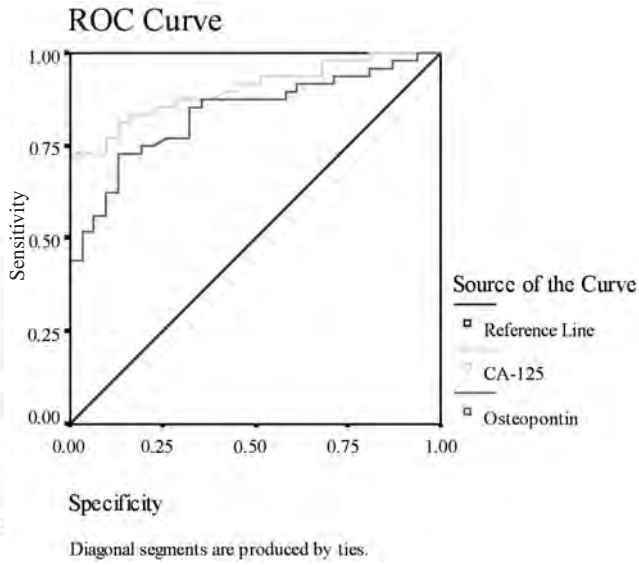

Figure 1. — Preoperative levels of osteopontin in patients with benign and malignant ovarian mass.
Figure 2. — Osteopontin levels in patients within histology of cancer.
Figure 3. — Linear regression curves.
Figure 4. — ROC analysis of OPN and CA125 for patients with the benign and malignant cysts.

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