

RESEARCH ARTICLE

MMP3 in Comparison to CA 125, HE4 and the ROMA Algorithm in Differentiation of Ovarian Tumors

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Abstract

Ovarian cancer is a highly malignant neoplasm with high mortality rates. Research to identify markers facilitating early detection has been pursued for many years. Currently, diagnosis is based on the CA 125 and HE4 markers, as well as the ROMA algorithm. The search continues for new proteins that meet the criteria of good markers. A total of 90 patients were included in the present study, allocated into: group 1, ovarian cancer, with 29 patients; group 2, endometrial cysts, with 30s; and group 3, simple ovarian cysts, with 31. Following histopathological verification, the CA 125, HE4, and metalloproteinase 3 (MMP3) levels were determined and the ROMA algorithm was calculated for all patients. The mean concentrations of all determined proteins, CA 125, HE4, and MMP3, as well as the ROMA values, were significantly higher in group 1 (ovarian cancer) compared to group 3 (simple ovarian cysts). The highest significant differences for the CA 125 levels ($p < 0.000001$) and ROMA ($p < 0.000001$) values were observed in postmenopausal women. For HE4, statistical significance was at the level of $p = 0.00001$ compared to $p = 0.002$ for MMP3. For the differentiation between ovarian cancer and endometrial cysts, the respective AUC ratios were obtained for CA 125, HE4, and MMP3 levels, as well as the ROMA values (0,93 / 0,96 / 0,75 / 0,98). After removing the post-menopausal patients, the MMP3 AUC value for ovarian cancer vs. benign ovarian cysts increased to 0.814. For post-menopausal women, the MMP3 AUC value for ovarian cancer vs. endometrial cysts was 0.843. As suggested by the results above, both the CA 125 and HE4 markers, as well as the ROMA algorithm, meet the criteria of a good diagnostic test for ovarian cancer. MMP3 seems to meet the criteria of a good diagnostic test, particularly in postmenopausal women; however, it is not superior to the tests used to date.

Keywords: MMP3 - ROMA algorithm - HE4 - CA 125 - ovarian cancer.

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Introduction

Ovarian cancer is one of the most common neoplastic diseases responsible for high mortality in women. This is mainly because 2/3 of all cases are diagnosed too late, when the disease is at FIGO stage III or IV. The search for markers of appropriate specificity and sensitivity to detect early-stage ovarian cancer has continued for many years.

To date, the CA 125 protein, characterized by high sensitivity but poor specificity, was routinely used as a marker. Starting from 2003, much hope was pinned on the determination of HE4 glycoprotein levels. Elevated serum HE4 levels were predicted to enable the early detection of ovarian cancer. In addition to the search for a specific and sensitive marker, the ROMA algorithm was developed to facilitate the classification of ovarian cancer patients into high- or low-risk groups (Chudecka-Głaz, 2015). The search for novel proteins that may facilitate the early detection and monitoring of treatment in ovarian cancer

patients continues.

Metalloproteinases are oncogenic factors. Many metalloproteinases have been tested and those with the greatest angiogenic potential are MMP1, 2, 3, and 9. Angiogenesis is a critical condition for growth and metastasis (Folkman, 1976). Tumour invasion is initiated by the destruction of cellular wall proteins by proteases (Li, 2006). Bodey et al. (2001) proposed that metalloproteinases 1, 3, 9 and 13 are responsible for the intensified invasiveness of breast, pancreatic and prostate cancers. The correlation between matrix metalloproteinase-3 (MMP3, stromelysin) and cellular ras oncogene expression and tumour progression potential was observed (Smolarz, B. 2003). Sheu et al. highlighted the role of metalloproteinases as mediators of immunosuppression in cancer diseases. MMP3 down-regulates the proliferation of T-cells by destroying IL-2 receptors (Sheu, 2003). In addition, some researchers highlighted the role of metalloproteinases in apoptotic

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dysfunction, which undoubtedly contributes to the intensification of pathological tumour proliferation (Menon, 2004).

The overexpression of MMP2 within ovarian tumours in patients with histopathologically confirmed ovarian cancer was correlated with shorter survival (Fu, 2015).

In addition to metalloproteinases 2 and 9 in ovarian cancer, MMP3 most frequently has higher expression.

Choi et al. exhibited increased mRNA expression of MMP3 in cancerous ovarian of chickens (Choi, 2011).

The study used the family of microRNA 200 inhibitors of MMP3 and showed invasion inhibition of the ovarian cancer cell lines SKOV3 and OVCAR3 (Sun, 2014).

Taking under consideration the role which seems to metalloproteinase 3 plays in the formation and metastasis of ovarian cancer, we would like to trace the output concentration of MMP3 in the blood serum of patients with ovarian cancer before treatment commenced. In addition, we want to see if MMP3 protein meets the criteria of a good diagnostic test in comparison to previously used markers: CA 125, HE4 and ROMA algorithm.

Materials and Methods

Included in the study were 90 patients hospitalized in 2014 at the Gynecological Surgery Clinic for Adults and Adolescents. A transvaginal ultrasound scan was performed for all patients before the procedure. CT scans were also performed if ovarian cancer was suspected.

After histopathological results were obtained, the patients were divided into 3 groups as follows: group 1, ovarian cancer with 29 patients; group 2, endometrial cysts with 30 patients; and group 3, simple ovarian cysts with 31 patients. Detailed data for the number of patients in each group, hormonal status, tumour stage and degree of tumour differentiation and the presence of ascites is shown in Table 1.

All patients gave written informed consent to participate in the study. Blood samples (5 mL) were collected from each patient into two test tubes. Detailed distribution of the concentration of markers for each group is presented in tables 2, 3, and 4. CA 125 and HE4 levels were determined at the hospital central laboratory, whereas MMP3 levels were determined at the laboratory of the Department of General Pathology. Patients with elevated creatinine levels were not included in the study because this coincides with elevated concentrations of the HE4 marker.

Assay analysis. CA 125 was determined using ARCHITECT CA 125 II assay [Reagent Kit No 2K45], Abbott Diagnostics, Illinois, USA. Analytical sensitivity was 1.0 U/mL. Precision Intra-assay - 2.4, precision Inter - assay - 3.9, normal range was 1-35 U/mL.

Serum HE4 concentrations were measured using the Elecsys ECLIA assay [Kit No 05950929190], Roche Diagnostics Ltd, Rotkreuz, Switzerland running on the cobas e 601 analyzer. Detection limit - 15.0 pmol/L.

The normal range was below 70 pmol/L for premenopausal women and below 140 pmol/L for postmenopausal women.

CA125 and HE4 assays were performed according

to manufacturers' instructions, with appropriate controls testing within the normal ranges. The predictive index of ROMA was calculated separately for pre- and postmenopausal patients using the following formulas: *i*). Premenopausal $PI = -12.0 + 2.38 * LN[HE4] + 0.0626 * LN[CA125]$, *ii*). Postmenopausal $PI = -12.0 + 1.04 * LN[HE4] + 0.732 * LN[CA125]$, where LN is a natural logarithm. Final ROMA values were calculated by inserting the obtained PI into the following formula: $\%ROMA = \exp(PI) / [1 - \exp(PI)] * 100$.

MMP3 concentrations were quantified in the serum using multiplex fluorescent bead-based immunoassays (Luminex Corporation, Austin, TX, USA) with the commercial Human MMP Magnetic Bead Panel 1 (Merck Millipore, Billerica, MA, USA Minimum Detectable Concentration - 45 pg/mL. Precision Intra-assay - 3.0. Precision Inter-assay - 4.5, normal range - 45-26000 pg/mL).

Statistical analysis was performed using STATICA 9.1 PL software. Descriptive characteristics of the examined population of patients were prepared, including the minimum, maximum, mean and median values. Because the distributions of the study traits were not normal, the mean values were compared using positional parameters (median) and non-parametric tests with Kruskal-Wallis test and Dunn's post-hoc test for comparisons between three groups and Mann-Whitney's U-test for comparison between two groups. For the selected groups, the receiver operating characteristic (ROC) curves were obtained and the area under curve (AUC) was calculated with 95% confidence intervals according to the nonparametric method of DeLong. A p value of <0.05 was considered as statistically significant. Sensitivity and specificity was calculated using the formula:

$$\text{Sensitivity} = TP / TP + FN$$

$$\text{Specificity} = TN / FP + TN$$

Results

The mean patient age and serum concentrations of the particular proteins are presented in tables 2, 3, and 4.

Significant correlations were observed between the patient age and HE4 protein levels ($r=0.5752$, $p=0.0009$) and patient age and ROMA values ($r=0.663$, $p=0.00007$) without dividing the group according to the histopathological diagnosis.

For the analysis of CA 125 marker levels in the group of patients with ovarian cancer and the group of patients with endometrial cysts, significant differences were observed between the groups (significance level $p=0.00003$). The median concentration the CA 125 marker was significantly higher compared to the CA 125 median concentration in patients with benign ovarian cysts ($p=0.00003/p=0.00001$). Significant differences in the ROMA values were observed between the ovarian cancer group and the endometrial cysts group ($p=0.000001$).

Medians concentrations of all determined proteins, CA 125, HE4, and MMP3, as well as the ROMA values, were significantly higher in the ovarian cancer group compared to the benign ovarian cysts group. For the comparison of average marker values between the

groups of cancer patients and benign cyst patients, the highest significant differences for the CA 125 levels ($p=0.000000$) and ROMA ($p=0.000000$) values were observed in postmenopausal women. For HE4, statistical significance was at the level of $p=0.00001$ compared to $p=0.002$ for MMP3.

Significant differences were observed between patients with endometrial cysts and those with benign cysts only for the CA 125 marker in premenopausal women. The median concentrations of HE4 and MMP3, as well as the ROMA values, were higher in patients with benign ovarian cysts compared to patients with endometrial cysts; however, this difference was not statistically significant.

No differences were observed in the median concentration of HE4 levels or ROMA values for different tumour gradings in the ovarian cancer patient group. By contrast, significant differences in the mean marker levels were observed for tumours of FIGO I, II versus FIGO III, IV staging (CA 125, $p=0.01$; HE4, $p=0.0005$; MMP3, $p=0.04$; ROMA, $p=0.004$). More detailed results are presented in table 5.

In the analysis of the group of ovarian cancer patients,

correlations between the CA 125 levels and HE4 levels ($r=0.419$, $p=0.0369$) and between the CA 125 levels and MMP3 levels ($r=0.454$, $p=0.0226$) were observed. Correlations between the patient age and CA 125 levels ($p=0.141$), age and MMP3 levels ($p=0.0273$), and age and ROMA values ($p=0.0072$) were also observed in this group of patients. A correlation was also observed between the age and Cramer clinical staging ($V=0.388$, $\chi^2=4.43$).

The ROC curves in figure 1 present the levels of individual markers and ROMA values as diagnostic tests. The AUC values were obtained for individual parameters as follows: CA 125, 0.97888; HE4, 0.93; ROMA, 0.9646; and MMP3: 0.6890. In the differentiation between ovarian cancer and benign ovarian cysts, the differences between the areas under the ROC for CA 125 and HE4, CA 125 and ROMA, or ROMA and HE4 were not statistically significant. Significant differences were observed between the areas under the curve for MMP3 and ROMA ($p=0.04$, $AUC=0.689$ for MMP3 and $AUC=0.96$ for ROMA).

The ROC curves in figure 2 illustrate the individual markers for the differentiation of ovarian cancer from endometrial cysts. The obtained AUC values were as

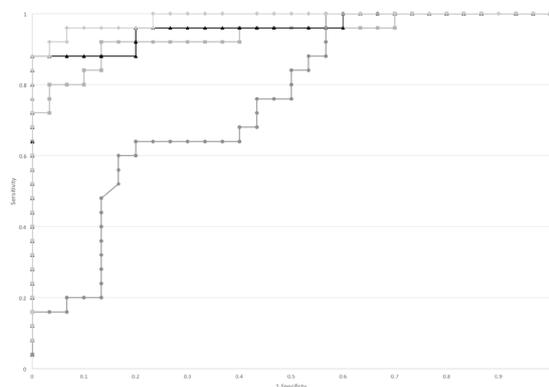


Figure 1. ROC Curves for Levels of HE4, CA 125, MMP3 and ROMA between Ovarian Cancer and Benign Ovarian Cysts

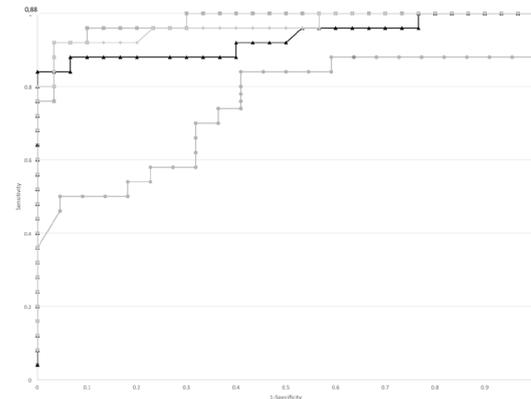


Figure 2. ROC Curves for Levels of HE4, CA 125, MMP3 and ROMA between Ovarian Cancer and Endometrial Ovarian Cysts

Table 1. Characteristics of the Study Group

Group	n person	Age	Hormonal status		Figo I, II	Figo III, IV	G1	G2, G3	Ascites	No Ascites
			PM	M						
Ovarian cancer	25	62,5	6	19	6	19	4	21	21	4
Endometrial ovarian cysts	35	34,46	27	8	-	-	-	-	-	-
Benign ovarian cysts	30	51,47	21	9	-	-	-	-	-	-

Table 2. Descriptive Statistics of Endometrial Ovarian Cysts Group

	n person	mean	-95% confidence interval	95% confidence interval	median	min	max	SD	coefficient of variation	standard error
age	30	34,46	31,40	37,53	33,00	23,0	58,00	8,20	23,80	1,49
CA 125 [U/mL]	30	47,25	32,04	62,47	32,15	13,9	210,90	40,73	86,20	7,43
HE 4 [pmol/L]	30	44,88	41,18	48,58	42,40	30,9	74,50	9,91	22,08	1,81
MMP 3 [pg/mL]	30	9951,94	6524,04	13379,83	10196,50	71,53	35608,00	9180,06	92,24	1676,04
ROMA	30	7,25	5,00	9,50	5,49	2,68	31,61	6,03	83,16	1,10

Table 3. Descriptive Statistics of Ovarian Carcinoma Group

	n person	mean	-95% confidence interval	95% confidence interval	median	min	max	SD	coefficient of variation	standard error
age	25	62,52	57,69	67,34	62,00	40,00	82,00	11,68	18,68	2,33
CA 125 [U/mL]	25	1302,88	399,79	2205,97	536,00	26,40	10000,00	2187,82	167,92	437,56
HE 4 [pmol/L]	25	917,08	303,08	1531,08	396,10	41,40	7000,30	1487,48	162,19	297,49
MMP 3 [pg/mL]	25	19381,12	13578,71	25183,52	15668,00	7084,00	66673,00	14056,89	72,52	2811,37
ROMA	25	79,73	67,79	91,67	96,12	8,13	99,89	28,92	36,27	5,78

Table 4. Descriptive Statistics of Benign Ovarian Cysts

	n person	mean	-95% confidence interval	95% confidence interval	median	min	max	SD	coefficient of variation	standard error
age	35	51,47	45,10	57,84	50,00	20,00	77,00	13,21	25,67	3,03
CA 125 [U/mL]	35	27,80	10,34	45,26	20,00	5,40	167,00	36,23	130,33	8,31
HE 4 [pmol/L]	35	52,86	46,36	59,35	50,40	37,60	78,90	13,47	25,48	3,09
MMP 3 [pg/mL]	35	13951,00	10806,73	17095,27	11926,00	5563,00	28635,00	6523,58	46,76	1496,61
ROMA	35	13,56	8,52	18,60	11,73	4,23	50,06	10,45	77,14	2,40

Table 5. Concentration Behaviour of the Various Markers Depending on FIGO Grading and the Presence or Absence of Ascites

Bio markers	FIGO I, II	FIGO III, IV	p	G1	G2, G3	p	No ascites	Ascites	p
CA 125 [U/mL]	328,6	1628,2	0,01	881,4	1443,3	0,02	421,3	1421,8	0,01
HE 4 [pmol/L]	163,2	826,4	0,0005	231,6	318,4	NS	211,3	632,1	0,003
ROMA	82	99	0,004	86	88	NS	82	97	0,04
MMP3 [pg/mL]	14688,2	19846,3	0,04	12434,2	17696,1	0,03	11112,1	20102,8	0,003

Table 6. Sensitivity and Specificity for the Different Markers in Differentiating Benign and Malignant Lesions in the Ovaries

	Sensitivity	Specificity
CA 125	88,7%	82,2%
HE 4	85,6%	88,1%
ROMA	88,3%	88,1%
MMP3	66,2%	68,8%

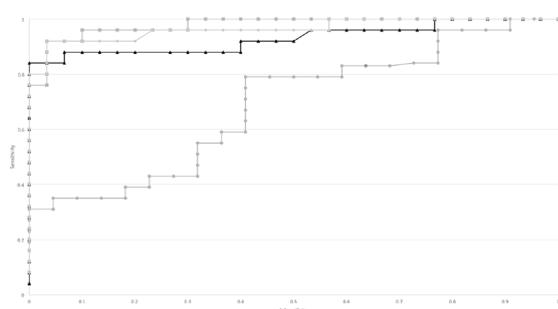


Figure 3. ROC Curves for Levels of HE4, CA 125, MMP3 and ROMA after Selecting Only Postmenopausal Patients between Ovarian Cancer and Benign Ovarian Cysts

follows. CA 125 AUC=0.93; HE4 AUC=0.96; MMP3 AUC=0.7526; and ROMA AUC=0.9866. No statistically significant differences were observed between the areas under the curves for markers CA 125 and HE4 levels as well as the HE4 levels and ROMA values in the groups

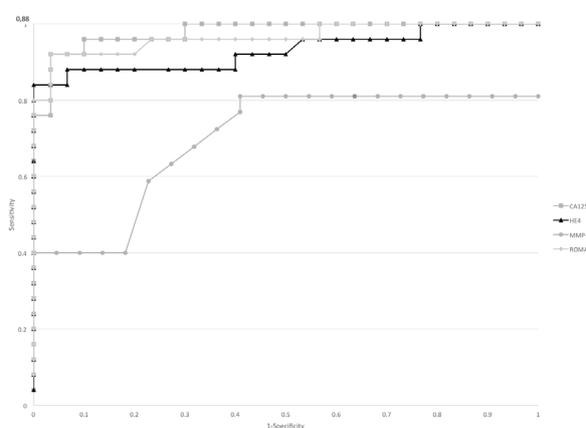


Figure 4. ROC Curves for Levels of HE4, CA 125, MMP3 and ROMA after Selecting Only Postmenopausal Patients between Ovarian Cancer and Endometrial Ovarian Cysts

of patients with ovarian cancer and endometrial cysts. Significant differences were observed between the areas under the curves for the HE4 and MMP3 levels (p=0.02), as well as the ROMA values and MMP3 levels (p=0.01) in the groups of patients with ovarian cancer and endometrial cysts.

After removing the post-menopausal patients, the MMP3 AUC value for ovarian cancer vs. benign ovarian cysts increased to 0.814. For post-menopausal women, the MMP3 AUC value for ovarian cancer vs. endometrial cysts

was 0.843 as shown in figures 3 and 4. For differentiating benign cysts from cancer, the EOC was calculated for the sensitivity and specificity of the different markers for the entire group. These results are presented in Table 6.

Discussion

The identification of a marker to facilitate the detection of ovarian cancer has continued for over a decade. The discovery of such a marker would enable the earlier detection and more effective treatment of ovarian cancer. For many years, CA 125 was an established protein marker for detecting and monitoring the treatment of ovarian cancer. Our study confirmed that the median concentration CA 125 levels differed significantly between ovarian cancer patients and benign ovarian cyst patients ($p=0.00001$) and between ovarian cancer patients and endometrial ovarian cyst patients ($p=0.00035$). The median concentration CA 125 levels in endometrial cyst patients were significantly higher than in the benign cyst patients.

So far, studies show us that the serum levels of CA 125 are slightly elevated during ovulation, significantly elevated during menstruation and pregnancy, and also following peritoneal irritation or infection. The median concentration CA 125 levels may be significantly elevated in cases of endometrial cysts or deep endometriosis (Muyldermans et al., 1995). In our study, we observed statistically higher CA 125 levels in patients with endometriosis compared to patients with benign ovarian cysts, which confirms previous reports.

Many researchers are trying to answer the question, which of the markers has the best detection for ovarian cancer. They are trying to compare what is the diagnostic effectiveness of the CA 125, HE4 and ROMA algorithm in ovarian cancer. One of the reports stated that the median concentration CA 125 levels were elevated in 21% of patients with benign ovarian cysts. The area under the ROC curve for CA 125 is 0.911 (Ortiz-Munoz, B. 2014). In our study, the AUC value for ovarian cancer and benign cysts is 0.98. The development of the new marker HE4 brought hopes for the earlier detection of ovarian cancer. Above cited researchers highlight that the marker is superior to CA 125 in premenopausal patients. HE4 is characterized by an AUC value of 0.92, a sensitivity of 86.2% and specificity of 87.4%. (Ortiz-Munoz, B. 2014) When assessing the diagnostic efficacy of HE4 in patients with type I and II endometrial ovarian cancer, Kristjansdottir et al. determined AUC values of 0.72 ($P<0.001$) and 0.92 ($P<0.001$), for EOC types I and II, respectively (Kristjansdottir et al., 2013).

In our study, the AUC for HE4 was 0.93. The HE4 median concentration levels in patients with FIGO III, IV cancer were significantly higher than in FIGO I, II cancer patients ($p=0.0005$). Similar results were presented by Chen et al. (2014) conducting a study of a group of Chinese patients with ovarian cancer. He demonstrated significant differences in the HE4 concentration level between patients with low and high clinical stage ovarian cancer. Furthermore, he suggested that low HE4 levels were largely correlated with the feasibility of the radical

resection of tumour to achieve R0 status. At the same time, the elevated HE4 levels after the completion of chemotherapy were a superior prognostic factor compared to CA 125.

Most studies evaluating detection sensitivity of early ovarian cancers using existing markers is not satisfactory.

Researchers reported that the AUC in the group of patients with low cancer staging of FIGO I compared to benign lesions was 0.72 for HE4 and 0.76 for CA 125 (Partheen, K. 2011). In our study, the AUC values in these groups of patients were as follows: 0.75 for HE4, 0.94 for CA 125, 0.87 for ROMA and 0.68 for MMP3.

These results suggest that neither the HE4 marker nor the enzyme MMP3 are good markers for early stage ovarian cancer detection. The above study suggests that we still lack a marker that would have both high sensitivity and specificity in detecting low advanced ovarian cancer.

Other position represents Winarto et al. in their assessment of the applicability of biomarkers for the detection of early ovarian cancer in Indonesian women, showed that in premenopausal women, HE4 and ROMA had the same AUC values of 0.85, with the superiority of ROMA versus HE4 observed in postmenopausal women (0.97 vs. 0.93). Attempts to change the biomarker threshold levels by raising the cut-off values led to a marked reduction in the sensitivity of the test (Winarto et al., 2014). Bandiera et al. (2011) demonstrated the high specificity of HE4 compared to CA 125 in differentiating endometrial or benign lesions from EOC. Furthermore, they propose to use HE4 levels and the ROMA algorithm as an independent prognostic factor.

In our studies, for the differentiation between EOC and benign disease, it was revealed that the sensitivity for HE4 in the entire studied group was 85.6% and the specificity was 88.1%. For the CA 125 marker, the sensitivity was 88.7% and the specificity was 82.2%. For the ROMA algorithm, the sensitivity and specificity were similar and adequate at 88.3% and 88.1%, respectively.

In carried out a meta-analysis demonstrated that ROMA is a useful algorithm for the identification of patients at high risk of EOC compared to patients with benign ovarian lesions. The authors report that HE4 is markedly superior to CA 125 for predicting the EOC vs. OC status. Both assays meet the criteria of a good diagnostic marker with no superiority of either (Li, 2012). Our studies revealed no significant differences in the AUC values for HE4, CA 125 and ROMA, with values of 0.93, 0.97, and 0.96, respectively, in ovarian cancer patients versus benign lesion patients and 0.96, 0.93, and 0.98, respectively, in EOC patients versus endometrial cyst patients. These results correspond with the results obtained by Molina et al. in which the AUC values for the ROMA and HE4 levels for determining the benign vs. malignant ovarian tumours was 0.952 and 0.936, respectively. The only exception was the slight difference in the AUC for CA 125 of 0.853 (Molina, 2011). For the studies by Bandiera et al., high preoperative ROMA values were associated with a higher FIGO staging, suboptimum cytoreduction, ascites, the presence of cancer cells within the cytology fluid, and shorter disease-free survival and overall survival (Bandiera et al., 2011).

Considering the routes of ovarian cancer spread into adjoining organs, as well as the role of metalloproteinases in the development of metastases, it appears that appropriate markers should be sought in this group of enzymes. As noted by Chambers and Matrisian (1997), concentrations of individual MMP family members increase with the staging of cancer. Higher concentrations of these proteases are detected after encroachment of the basal membrane when tumour spread is observed locally or as distant metastases.

The expression of several metalloproteinases, specifically 1, 3, 9 and 13, was demonstrated in breast cancer (Balduyck et al., 2000). Baruch et al. (2001) observed that MMP-3 gene expression was correlated with cellular ras oncogene expression and tumour progression potential. Other researchers demonstrated that metalloproteinases play a role as mediators of immunosuppression in cancer diseases by destroying IL-2 receptors and down-regulating the proliferation of T-cells following contact with neoplastic cells (Sheu et al., 2003). In addition some of the scholars highlight the role of metalloproteinases in apoptotic dysfunction, which also contributes to the intensification of pathological tumour proliferation (Menon et al., 2004).

Choi et al. (2011) observed the increased expression of MMP3 mRNA in early-stage chicken ovarian cancer compared to healthy chicken ovaries $p < 0.05$. In our study, comparison of the median concentration MMP3 levels between groups of patients with ovarian cancer and benign ovarian cysts yielded significant differences, with $p = 0.04$. In post-menopausal patients, the differences were significant at $p = 0.002$. The MMP3 AUC value between low-stage ovarian cancer patients and benign ovarian cysts was 0.68.

We also observed significantly higher MMP3 levels in FIGO III, IV compared to FIGO I, II ovarian cancer patients ($p = 0.04$). Furthermore, we observed significantly higher MMP3 levels in patient with G2, G3 and with confirmed ascites. High concentrations of MMP3 in advanced forms of ovarian cancer affirm the role of metalloproteinases in cancer metastasis.

The correlation between serum MMP3 and the age of patients is important ($p = 0.0273$). Researchers of the Boston team compared the mean urine MMP2 and MMP9 levels in non-CA 125 elevated (marker-negative) ovarian cancer patients. They observed that although the mean urine levels of MMP2 and MMP9 were not significantly different in cancer and non-cancer subjects, consideration of the patient age and multivariate logistic regression increased the statistical significance of the assay, and the AUC value ratios increased to 0.88 (Coticchia et al., 2011). As demonstrated by Ziyi Fu et al. (2015) in ovarian cancer patients, overexpression of MMP2 within the tumour was correlated with shorter survival and was an independent factor of a poor prognosis.

In our studies, after removing the post-menopausal patients, the MMP3 AUC value for ovarian cancer vs. benign ovarian cysts increased from 0.689 to 0.814. For post-menopausal women, the MMP3 AUC value for ovarian cancer vs. endometrial cysts was 0.843. Analysing data for the sensitivity and specificity of individual

markers shows that the MMP3 protein parameters do not exceed the currently used markers. The sensitivity for metalloproteinase 3 is 66.2% and the specificity is 68.8%. Due to the significant differences in the median concentrations of MMP3, it has been implemented in the group of patients with advanced ovarian cancer compared to the concentrations of MMP3 in patients with a low stage. In the future, we will determine whether the initial level of MMP3 is correlated with the survival time of patients. Sun et al. conducted a study on two ovarian cancer cell lines, SKOV3 and OVCAR3, and found that overexpression of the miR200 family member significantly correlated with the inhibition of MMP3 secretion, leading to a reduction of the invasiveness and metastasis of ovarian cancer cells (Sun, 2014).

In conclusion, as suggested by the results presented here, both CA 125 and HE4 markers, as well as the ROMA algorithm, meet the criteria of a good diagnostic test. MMP3 meets the criteria for a good diagnostic test, particularly in postmenopausal women; however, it is not superior to CA 125 or HE4 as a marker. Therefore, MMP3 protein cannot replace the tests used thus far and is also not useful for diagnosing ovarian carcinoma. Further studies with larger patient groups are required to verify whether higher serum MMP3 concentrations correlate with a poorer prognosis for patients.

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