INTRODUCTION

Ovarian cancer is a common and fatal gynecological malignancy with high morbidity and mortality rates. Ovarian cancer is the second most common cancer among women accounting 13.6% female malignancy.\(^1\) Both vague symptoms and location cause ovarian cancer to be diagnosed in advance stages.\(^2\) The long term survival rate is <30% in advance stages while 5 years survival rate exceeds 90% in localised tumors. Unfortunately, only <20% of ovarian cancers are localised to the ovaries at the time of diagnosis.\(^3\) Current practices for the diagnosis of ovarian masses are transvaginal ultrasounds and other imaging techniques. Serum biomarkers like CA 125 and HE4, histopathology and proteomics are also available. Suspicion of malignancy is based largely on imaging appearance and experience of physician for distinguishing benign from malignant.\(^4\) The accuracy of ultrasonography is still low for detecting ovarian cancer and highly false positive rates have been reported by using other imaging technologies.\(^5\) In comparison with histopathology, which is taken as gold standard for the diagnosis and also an invasive procedure, serum tumor markers are rather cost-effective, non-subjective, and non-invasive technique.

Nowadays, among several tumor markers, CA 125 is being used for the diagnosis of ovarian cancer but CA 125 levels may also be raised in other benign reproductive disorders like endometriosis, benign ovarian cysts, first trimester of pregnancy and pelvic inflammatory diseases, fibromas, dermoid cysts etc. New tumor markers are also emerging and in combination with CA 125, they can improve the specificity for diagnosis. HE4 (Human Epididymis Protein 4) is a new tumor marker and has been proposed for ovarian cancers as it is frequently over-expressed in ovarian cancers and its levels are less likely to be elevated in benign conditions as is the case with CA125.\(^6\)

Risk of ovarian malignancy algorithm (ROMA) is a scoring system based in corporation of CA-125 and HE4 with menopausal status and shows excellent diagnostic performance for the differentiation of ovarian cancers from benign masses. ROMA stratifies these patients into high risk group or low risk group.\(^7\) This stratification of risk of having malignancy is becoming important when laparoscopy or non-invasive management is being considered in benign cases or very extensive cytoreductive surgeries are being mandatory by gynecologic oncologist.\(^5,7,8\)

ABSTRACT

Objective: To determine the diagnostic accuracy of ROMA in postmenopausal women with history of ovarian mass.

Study Design: Observational study.

Place and Duration of Study: Dr. Ziauddin University Hospital, Karachi, from May 2014 to June 2015.

Methodology: Two hundred and sixty postmenopausal women of 40-65 years of age with ovarian masses, planned for surgery, were included in the study. Their samples were obtained preoperatively and analysed on Abbot Architect i1000 SR immunoassay analyser for quantitative estimation of tumor markers, i.e. HE4 and CA125. By combination of these two tumor markers, ROMA scores were calculated and studied after histopathological verification of masses.

Results: Total number of patients were 260, out of which 122 (46.9%) were diagnosed as having ovarian cancer, while 138 (53.0%) were diagnosed as benign condition. Median ROMA score levels in patients with malignant masses were 95.58 (IQR=44.4) as compared to 20.6 (IQR=14) in benign masses. ROMA had sensitivity 92.6% (CI=86.47-96.04), specificity 78.3% (CI=70.09-83.82), positive predictive value 79% (CI=70.87-84.29), negative predictive value 92.3% (CI=86.02-95.9) and positive likelihood ratio 4.26, while negative likelihood ratio 0.1. Diagnostic accuracy of ROMA was 85%, based on ROC curve analysis. ROMA had the highest sensitivity in detecting ovarian carcinoma.

Conclusion: ROMA is a very useful diagnostic tool for the preoperative stratification of patients with ovarian masses showing 85% diagnostic accuracy. However, there is need of more studies with homogenous laboratory procedures for HE4 and CA125 assays as well as patients, selection criteria, so we can draw firm conclusion about utility of ROMA in clinical setups.

Key Words: Diagnostic accuracy. ROMA. Post-menopausal. Ovarian mass. HE4. Tumor markers.
The objective of the study was to determine the diagnostic accuracy of risk of ovarian malignancy algorithm (ROMA) in our population, as there is no data available in Pakistan as well as there are limited studies which show discrepancy regarding diagnostic accuracy of ROMA.

**METHODOLOGY**

This observational study was conducted from May 2014 to June 2015 in the Department of Chemical Pathology, Dr. Ziauddin University Hospital. Two hundred and sixty postmenopausal women with ovarian mass (>2 cm) on pelvic ultra sound examination, attending gynecology clinics, planned for surgical intervention, were informed about the study and consent was taken for participation in it. Permission from the Ethical Committee of the Hospital was taken. Questionnaire was filled. Menopausal status was determined by history of the patients and age group, as inclusion criteria. Those who did not give consent were excluded from the study.

Blood samples were collected from all the patients into the serum separator tube, before their surgical procedures, and centrifuged at 4000g for 15 minutes then part of the sample was transferred to aliquot and stored at -30° centigrade until testing. Serums were analysed for the quantification of CA125 and HE4 on automated immunoassay analyser, Abbot ARCHITECT i1000 by chemiluminescent microparticle immunoassay method. All manufacturer recommendations for maintenance, calibration, and internal quality assessment were followed for both assays.

ROMA was calculated using CA 125 and HE4 results by using ROMA calculator, which is based on predictive index. Predictive index was calculated from the equation; post-menopause PI = -8.09 + 1.04*LN (HE4) + 0.732*LN (CA125),\(^9\) where LN is the natural logarithm. ROMA was determined using the equation: ROMA (%)= exp (PI)/[1-exp (PI)]*100.\(^10\)

A score of 27.7% was adopted as the cutoff point for ROMA as recommended for postmenopausal patient.\(^7\)

The patients were labelled as either malignant or benign, according to histopathological findings. Accuracy of the ROMA was assessed by measuring specificity, sensitivity, positive predictive value (PPV) and negative predictive value (NPV). Confounding variables were controlled by strictly following the inclusion and exclusion criteria.

Median with interquartile values were calculated for quantitative variables like age, size, CA125 and HE4 levels; while frequencies with percentage were calculated for qualitative variables. HE4, CA125, and ROMA represented diagnostic variables acting as stimulants which increase the probability of ovarian cancer proportionally to their rising value. Asymmetrically (skewed) distributed quantitative variables were compared by Mann-Whitney test for two disease groups.

Effect modifiers like age, and size of the tumor were controlled by stratification. Diagnostic accuracy was represented in terms of sensitivity, specificity, positive predictive value and negative predictive value, and positive and negative likelihood ratio derived from contingency table to evaluate the diagnostic performance of ROMA. ROC curve analysis and 95% CI was used to validate the ROMA performance, while p-value less than 0.05 was taken as significant. All analyses were performed on SPSS version 20.

**RESULTS**

A total of 260 postmenopausal women, diagnosed for having ovarian mass on ultrasound were included in this study. The average age of the patients was 49.28 ±6.26 years. The mean size of tumors, CA125 and HE4, are given in Table I. Standard cutoff values for HE4, and CA125 were already established, i.e. 150 pmol/L and 35 U/L, respectively. The most common benign tumor was serous cyst adenomas which accounted for 30% (n=42), followed by mucinous cyst adenomas which accounted for 21% (n=29). Dermoid cysts were 18% (n=25), followed by endometriotic cysts and stromal cell tumor (10%, n=14 each); while 11.6% (n=16) were of other category. Of malignancies, there were 51% (n=62) endometrioid carcinoma, 30% (n=37) serous cyst carcinoma, 7.3% (n=9) clear cell carcinoma, 4% (n=5) mixed epithelial tumor and mucinous adenocarcinoma 2% (n=3), 5% (n=6) papillary carcinoma.

The median serum levels for CA125 was 49.45 IU/ml (IQR=761.5) while HE4 median level was 101.55 pmol/L (IQR=456.3) as shown in Table I. CA125 and HE4 levels were significantly high in ovarian malignancies. CA125 and HE4 median levels were 685.7 (IQR=2605.3) IU/ml and 543.9 (IQR=1256) pmol/L, respectively as compared to benign disorders where CA125 and HE4 median levels were 25 (IQR=43.9) IU/ml and 57 (IQR=41) pmol/L, respectively (p<0.001). CA125 levels were markedly increased in fibromas and some dermoid cysts. HE4 levels were not falsely elevated in these conditions. ROMA median levels in patients with malignant masses were 95.58 (IQR=44.4) as compared to 20.6 (IQR=14) in benign masses (p<0.001).

ROMA score was elevated in 113 out of 122 ovarian cancer patients, while not elevated in108 out of 138 benign cases as shown in Table II. Sensitivity, specificity, PPV, NPV and diagnostic accuracy of ROMA in detection of ovarian tumors in postmenopausal women was very good as shown in Table II.

The best cutoff point was 28.11%. AUC for ROMA was 91.4% at sensitivity 92.6% and specificity 78.3%, which was higher as compared to CA125 alone, as shown in Figure 1.

Stratification analysis was performed and observed that accuracy of ROMA in detection of ovarian tumors in
postmenopausal women was above 80% in all age groups; except in 56 to 60 years of age, accuracy was 79.3% as shown in Table III. Similarly, with respect to tumor size, accuracy of ROMA was about 91% in cases where size of tumor was 3 to 10 cm and above 15 cm while 67.65% in cases where tumor size was 11 to 15 cm as shown in Table IV.

**Table I:** Descriptive statistics of the patients.

<table>
<thead>
<tr>
<th>95% Confidence Interval for Mean Median IQR</th>
<th>Lower bound</th>
<th>Upper bound</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>48.47</td>
<td>50.09</td>
</tr>
<tr>
<td>Size of tumor (cm)</td>
<td>8.53</td>
<td>9.65</td>
</tr>
<tr>
<td>CA125 IU/ml</td>
<td>765.6</td>
<td>1317.15</td>
</tr>
<tr>
<td>HE4 pmol/L</td>
<td>372.11</td>
<td>669.86</td>
</tr>
</tbody>
</table>

**Table II:** Diagnostic accuracy of ROMA in detection of ovarian tumors in postmenopausal women taking histopathology as gold standard (n=260).

<table>
<thead>
<tr>
<th>ROMA value &gt;27.7% [Malignant]</th>
<th>ROMA value &lt;27.7% [Benign]</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>ROMA value &gt;27.7% [Malignant]</td>
<td>113 (TP)</td>
<td>143 (55%)</td>
</tr>
<tr>
<td>ROMA value &lt;27.7% [Benign]</td>
<td>9 (FN)</td>
<td>117 (45%)</td>
</tr>
<tr>
<td>Total</td>
<td>122 (46.5%)</td>
<td>260</td>
</tr>
</tbody>
</table>

Sensitivity = 113/122 = 92.6%
Specificity = 108/138 = 78.3%
PPV = 113/143 = 79.0%
NPV = 108/117 = 92.3%
Positive likelihood ratio = 4.26; Negative likelihood ratio = 0.1; Accuracy = 85%.

**Table III:** Diagnostic accuracy of ROMA in detection of ovarian tumors in postmenopausal women taking histopathology as gold standard in respect to age of patient.

<table>
<thead>
<tr>
<th>Size of the tumor</th>
<th>3 to 10 cm</th>
<th>11 to 15 cm</th>
<th>&gt;15 cm</th>
</tr>
</thead>
<tbody>
<tr>
<td>True positive</td>
<td>62</td>
<td>37</td>
<td>14</td>
</tr>
<tr>
<td>False positive</td>
<td>8</td>
<td>21</td>
<td>1</td>
</tr>
<tr>
<td>False negative</td>
<td>7</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>True negative</td>
<td>93</td>
<td>9</td>
<td>6</td>
</tr>
<tr>
<td>Sensitivity</td>
<td>89.7%</td>
<td>97.4%</td>
<td>93.3%</td>
</tr>
<tr>
<td>Specificity</td>
<td>92.1%</td>
<td>30%</td>
<td>85.7%</td>
</tr>
<tr>
<td>PPV</td>
<td>88.4%</td>
<td>63.79%</td>
<td>93.3%</td>
</tr>
<tr>
<td>NPV</td>
<td>93%</td>
<td>90%</td>
<td>85.7%</td>
</tr>
<tr>
<td>Diagnostic accuracy</td>
<td>91.1%</td>
<td>67.65%</td>
<td>90.9%</td>
</tr>
</tbody>
</table>

Ovarian cancer is the most frequent cause of death from gynecological cancer. It has highest fatality ratio of all gynecological malignancies, which is characterised by the early widespread metastasis and high grade malignancy at the time of diagnosis. Studies are being available since many years for a novel, more sensitive, and more specific tumor marker or diagnostic algorithm to provide the stratification of patients with a pelvic mass and for screening in ovarian cancer. Much hope has been attached recently to the ROMA algorithm, developed by Moore et al. By combining serum CA125 with HE4 levels and menopausal status of patients, ROMA was considered helpful as it reduces the number of false positive cases with elevated CA125. Some studies have claimed superiority of ROMA over other biomarkers of ovarian malignancies and algorithms; but also suggested that further studies are also required to prove their claim.

Total number of patients selected in this study were 260; out of which, 46.9% were diagnosed as having ovarian cancer both clinically as well as proven histopathologically. In this study, there was a significant difference between benign and malignant disease with respect to serum CA125, HE4 and ROMA levels. ROMA was sensitive and specific marker as our results were consistent with other studies. Studies conducted by Hellstrom et al. in 2003 and Montagnana et al. in 2009 showed similar performances of CA125, HE4 and ROMA. In this study, ROMA misclassified 22.3% benign tumors and showed high risk for malignancy, i.e. false positivity among stromal cell tumors like fibromas and adenofibromas, which showed its lack of specificity in non-epithelial tumors when compared with endometriotic cyst, and dermoid cyst; while 78.3% were classified as true negative, i.e. low risk for malignancy in serous and...
mucinous cyst adenomas. Among malignant tumors, ROMA misclassified 7.43% cases as falsely negative, although by histopathology they were malignant. ROMA score were significantly high in epithelial ovarian cancers rather than in non-epithelial tumors. These results were consistent with the study performed by T Van Gorp et al. which showed elevated ROMA values in fibroma / thecomas and cystadenofibromas.\(^9\)

The high negative predictive value in this study provides a strong reassurance that a pelvic mass is benign. The sensitivity, specificity, PPV, NPV values and accuracy of ROMA were consistent with other studies.\(^{13,17,20}\) A study, conducted by Moore, concluded that dual marker combination successfully classify pelvic mass into benign and malignant as shown in our study.\(^9\) By using area under the receiver operator characteristic curves analysis (AUC ROC) as a measure of test performance, among CA125, HE4 and ROMA, ROMA achieved a higher AUC ROC (0.914, CI=0.876-0.952, \(p<0.05\)), and therefore, had increased sensitivity than either marker alone. These findings also validate the performance of ROMA.

In 2009, FDA approved ROMA and recommended its use in women over 18 years of age with a pelvic tumor or cyst qualified for surgery, emphasizing that ROMA must always be interpreted against clinical and radiological findings. ROMA is a very useful diagnostic tool for stratification of patients with a pelvic mass.\(^{14}\)

Several multinational studies were published on validation of ROMA for ovarian mass risk stratification. When combined these studies, which performed in the United States, Europe and Asia over 4,000 women with ovarian mass, the range of sensitivity for ROMA test was found from 75% - 94%, at specificity from 75% - 95%. As in this study, ROMA demonstrates consistent and reliable performance for classifying ovarian mass into high risk and low likelihood groups for epithelial ovarian cancers. ROMA may help physicians in identifying as well as triage women to specialising unit in the care and management of ovarian cancer.\(^{7,21}\)

Limitations of this study were absence of multicenter data collection and also lack of availability of comorbidity data. However, strengths are that sample size was appropriate and very few studies have been conducted on ROMA in our part of the world.

**CONCLUSION**

It is concluded from this study that ROMA is a very useful diagnostic tool for the preoperative stratification of patients with ovarian mass showing 85% diagnostic accuracy, as the combination of CA125 and HE4 gives better differentiation between benign and malignant lesions as compared to being used alone.

**REFERENCES**

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