Clinical performance of a multivariate index assay for detecting early-stage ovarian cancer

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OBJECTIVE: We sought to analyze the effectiveness of a multivariate index assay (MIA) in identifying early-stage ovarian malignancy compared to clinical assessment, CA 125-II, and modified American Congress of Obstetricians and Gynecologists (ACOG) guidelines among women undergoing surgery for an adnexal mass.

STUDY DESIGN: Patients were recruited in 2 related prospective, multinational trials involving 44 sites. All women had preoperative imaging and biomarker analysis. Preoperative biomarker values, physician assessment of ovarian cancer risk, and modified ACOG guideline risk stratification were correlated with surgical pathology.

RESULTS: A total of 1016 patients were evaluable for MIA, CA 125-II, and clinical assessment. Overall, 86 patients (8.5%) had primary-stage I/II primary ovarian malignancy, with 70.9% having stage I disease and 29.1% having stage II disease. For all early-stage ovarian malignancies, MIA combined with clinical assessment had significantly higher sensitivity (95.3%; 95% confidence interval [CI], 88.6–98.2) compared to clinical assessment alone (68.6%; 95% CI, 58.2–77.4), CA 125-II (62.8%; 95% CI, 52.2–72.3), and modified ACOG guidelines (76.7%; 95% CI, 66.8–84.4) (P < .0001). Among the 515 premenopausal patients, the sensitivity for early-stage ovarian cancer was 89.3% (95% CI, 72.8–96.3) for MIA combined with clinical assessment, 60.7% (95% CI, 42.4–76.4) for clinical assessment alone, 35.7% (95% CI, 20.7–54.2) for CA 125-II, and 78.6% (95% CI, 60.5–90.9) for modified ACOG guidelines. Early-stage ovarian cancer in postmenopausal patients was correctly detected in 98.3% (95% CI, 90.9–99.7) of cases by MIA combined with clinical assessment, compared to 72.4% (95% CI, 59.8–82.2) for clinical assessment alone, 75.9% (95% CI, 63.5–85.0) for CA 125-II, and 75.9% (95% CI, 63.5–85.0) for modified ACOG guidelines.

CONCLUSION: MIA combined with clinical assessment demonstrated higher sensitivity for early-stage ovarian malignancy compared to clinical assessment alone, CA 125-II, and modified ACOG guidelines with consistent performance across menopausal status.

Key words: detection, early stage, ovarian cancer

Over 3000 women are diagnosed with early-stage ovarian cancer in the United States annually.¹ For this group, comprehensive surgical staging and selected use of adjuvant chemotherapy are associated with improved survival.²⁻⁴ Gynecologic oncologists are significantly more likely to perform proper surgery and administer indicated chemotherapy.⁵⁻⁷ As a result, accurate referral to subspecialty care is an important determinant of clinical outcomes for women with early-stage ovarian cancer.⁸ To assist primary care providers and general gynecologists in
the triage of women with an adnexal mass, the American Congress of Obstetricians and Gynecologists (ACOG) and the Society of Gynecologic Oncology (SGO) jointly published updated referral guidelines in 2011.9 These guidelines have been shown to perform well in predicting advanced-stage ovarian cancer but have limited accuracy in predicting early-stage disease, especially in premenopausal women.10,11 Similarly, CA 125, the most widely used tumor marker in ovarian cancer, is only expressed by 50% of early-stage tumors.12-14 Consequently, efforts to aid in the triage of women with an adnexal mass have focused on algorithms utilizing novel serum biomarkers.

The multivariate index assay (MIA) (OVA1; Vermillion, Inc., Austin, TX) is a multiple biomarker test that was approved by the US Food and Drug Administration (FDA) in 2009 as a diagnostic triage aid in the prediction of ovarian cancer in women with an adnexal mass. In the pivotal clinical utility trial reported by Ueland et al15 in 2011, OVA1 combined with physician assessment was found to have a higher sensitivity and negative predictive value than physician assessment alone and CA 125. The consistent performance of OVA1 combined with physician assessment was confirmed in an intended-use-population validation trial reported by Bristow et al.16 in 2013. Both studies had identical selection criteria (women aged ≥18 years with a pelvic mass scheduled to undergo surgery) with prospectively collected clinical and pathological data. The object of the current study was to examine the performance of OVA1 in identifying early-stage ovarian malignancy compared to clinical assessment, CA 125-II, and modified ACOG guidelines among women undergoing surgery for an adnexal mass using the combined dataset of >1000 subjects from the aforementioned prospective clinical trials.

**Materials and Methods**

Female subjects with a planned surgical procedure for removal of an adnexal mass were enrolled in 2 prospective OVA1 studies at 44 sites across the United States. Institutional review board approval was obtained at each site. All demographic, clinical, and pathologic data were collected on standardized case report forms and included the specialty of physicians who enrolled and operated on the patient: gynecologic oncology specialist or non-specialist. Inclusion criteria were as follows: consented females age ≥18 years, an adnexal mass documented by imaging (computed tomography, ultrasonography, or magnetic resonance imaging), preoperative phlebotomy, and surgery within 3 months of imaging. Exclusion criteria included a diagnosed malignancy within the previous 5 years (with the exception of nonmelanoma skin cancer), declined phlebotomy, surgery not performed within 3 months as planned, or an incomplete case report that prevented the analysis of test performance. Women were classified as premenopausal if <6 months had passed since last menstruation and as postmenopausal if 12 months had passed since last menstruation. If last menstruation occurred between 6-12 months or if the time was not given, women age ≤50 years were classified as premenopausal and women >50 years as postmenopausal.

The methods for blood collection and specimen handling have been previously reported.15,16 Biomarker measurements were performed according to the OVA1 instructions for use at Quest Diagnostics Inc (Chantilly, VA) or the Division of Clinical Chemistry, Department of Pathology, Johns Hopkins Medical Institutions (Baltimore, MD). OVA1 is an FDA-cleared MIA that incorporates CA125-II, transferrin, transthyretin (prealbumin), apolipoprotein A1, and beta-2-microglobulin. Biomarker values were transformed by the OvaCalc software using a proprietary multivariate algorithm, to generate an ovarian malignancy risk score from 0.0-10.0 as described previously.17 Subjects were stratified as high risk with OVA1 scores ≥5.0 (premenopausal) or ≥4.4 (postmenopausal).

For CA 125-II measurement, the same value used for OVA1 calculation was used for individual analysis and stratified, for comparison with OVA1, as high or low risk using clinical cutoff values in accordance with published ACOG referral criteria ≥200 U/mL (premenopausal) or ≥35 U/mL (postmenopausal).

After assessment by physicians, a clinical prediction of malignancy was documented using the physician’s usual method to establish a benchmark for routine clinical practice. Clinical assessment always included physical examination and imaging, per the study inclusion criteria, and CA 125, if used. Postoperative pathology diagnosis was recorded at each enrolling site, and malignancies were classified by primary site of origin (ovarian or nonovarian), International Federation of Gynecology and Obstetrics stage, and histological subtype. All case report forms, pathology reports, diagnoses, and tumor classifications were independently adjudicated, blinded to OVA1 results.

To assess the effect of adding OVA1 to an established standard method, the Dearking-modified ACOG criteria for consultation with a gynecologic oncologist were used to identify subjects meeting ≥1 of the following criteria11:

- Premenopausal women
  1. Very elevated CA 125 (>67 U/mL).
  2. Ascites.
  3. Evidence of abdominal or distant metastasis.

- Postmenopausal women
  1. Elevated CA 125 (>35 U/mL).
  2. Nodular or fixed pelvic mass.
  3. Ascites.
  4. Evidence of abdominal or distant metastasis.

Case report forms, biomarker values, and OVA1 scores were sent to Applied Clinical Intelligence for statistical analysis. Results were statistically stratified based on the subject’s menopausal status, surgical pathology, and stage of malignancy. Clinical diagnostic performance criteria (sensitivity, specificity, positive and negative predictive values) were calculated for clinical assessment, OVA1, CA 125, and modified ACOG criteria alone or in combination with OVA1. The OVA1 instructions for use indicate that referral should use a Boolean OR between physician assessment and OVA1 risk stratification, so that either criterion will trigger patient evaluation by a gynecologic oncologist. Accordingly, the addition of OVA1 to...