# National Academy of Clinical Biochemistry Guidelines for the Use of Tumor Markers in Ovarian Cancer

Daniel W. Chan[1]*, Ie-Ming Shih[1], Lori J. Sokoll[1], Robert C Bast, JR[2]

[1]Departments of Pathology and Oncology, Johns Hopkins Medical Institutions, Baltimore, MD and [2]The University of Texas M.D. Anderson Cancer Center, Houston, Texas, USA.

***Sub-Committee Chair**, to whom all comments should be addressed *via* e-mail to dchan@jhmi.edu, with copies to C.Sturgeon@ed.ac.uk and ediamandis@mtsinai.on.ca

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**Abbreviations:** hCGβ, free beta-subunit of human chorionic gonadotropin; hCGβcf, beta core fragment of hCG; TVUS, transvaginal ultrasound

**INTRODUCTION**

In the United States, ovarian cancer is among the top four most lethal malignant diseases in women, who have a life-time probability of developing the disease of 1 in 59 [2004 American Cancer Society Cancer Facts (http://www.cancer.org/docroot/STT/stt_0.asp)]. Worldwide, the incidence of ovarian cancer has recently been estimated (2002) at 204,499 cases per year with corresponding 124,860 deaths (http://www-dep.iarc.fr/).

The overall mortality of ovarian cancer has remained unchanged despite new chemotherapeutic agents, which have significantly improved the five-year survival rate (1). The main reason is lack of success in diagnosing ovarian cancer at an early stage, as the great majority of patients with advanced stage of ovarian carcinoma die of the disease. In contrast, if ovarian cancer is detected early, 90% of those with well-differentiated disease confined to the ovary survive. Furthermore, biomarkers that can reliably predict clinical behavior and response to treatment are generally lacking. The search for tumor markers for the early detection and outcome prediction of ovarian carcinoma is therefore of profound importance and represents one of the critical subjects in the study of ovarian cancer.

Although ovarian cancer is often considered to be a single disease, it is composed of several related but distinct tumor categories including surface epithelial tumors, sex-cord stromal tumors, germ cell tumors, and metastatic tumors (2). Within each category, there are several histological subtypes. Of these, epithelial tumors (carcinomas) are the most common and are divided, according to Federation of Gynecology and Obstetrics (FIGO) and World Health Organization (WHO) classifications, into five histologic types: serous, mucinous, endometrioid, clear cell, and transitional (3). The different types of ovarian cancers are not only histologically distinct but are characterized by different clinical behavior, tumorigenesis, and pattern of gene expression. Based on prevalence and mortality, the serous carcinoma is the most important, representing the majority of all primary ovarian carcinomas with a dismal clinical outcome (4). Therefore, unless otherwise specified, serous carcinoma is what is generally thought of as “ovarian cancer.”

The search for more effective biomarkers depends on a better understanding of the pathogenesis of ovarian cancer, i.e. the molecular events in its development. Based on a review of recent clinicopathological and molecular studies, a model for the development of ovarian carcinomas has been proposed (5). In this model, surface epithelial tumors are divided into two broad categories designated Type I and Type II tumors which correspond to two main pathways of tumorigenesis. Type I tumors tend to be low-grade neoplasms that

arise in a stepwise fashion from borderline tumors whereas Type II tumors are high-grade neoplasms for which morphologically recognizable precursor lesions have not been identified, so-called "*de novo*" development. As serous tumors are the most common surface epithelial tumors, low-grade serous carcinoma is the prototypic Type I tumor and high-grade serous carcinoma is the prototypic Type II tumor. In addition to low-grade serous carcinomas, Type I tumors are composed of mucinous carcinomas, endometrioid carcinomas, malignant Brenner tumors and clear cell carcinomas. Type I tumors are associated with distinct molecular changes that are rarely found in Type II tumors, such as *BRAF* and *KRAS* mutations for serous tumors, *KRAS* mutations for mucinous tumors, and β-catenin and *PTEN* mutations and microsatellite instability for endometrioid tumors. Type II tumors include high-grade serous carcinoma, malignant mixed mesodermal tumors (carcinosarcoma) and undifferentiated carcinoma. There are very limited data on the molecular alterations associated with Type II tumors, except frequent p53 mutations in high-grade serous carcinomas and malignant mixed mesodermal tumors (carcinosarcomas). This model of carcinogenesis provides a molecular platform for the discovery of new ovarian cancer markers.

## CURRENTLY AVAILABLE MARKERS FOR OVARIAN CANCER

The most widely studied ovarian cancer body fluid-and tissue-based tumor markers are listed in Table 1, which also summarizes the phase of development of each marker and the level of evidence (LOE) for its clinical use. The LOE grading system is based on a previous report describing the framework to evaluate clinical utility of tumor markers (6). The following discussion will focus mainly on CA125, which is the only marker that has been accepted for clinical use in ovarian cancer. The NACB panel does not recommend clinical utilization of other biomarkers in diagnosis, detection, or monitoring of ovarian cancer as all other markers are either in the evaluation phase or in the research/discovery phase.

## TUMOR MARKERS IN OVARIAN CANCER: NACB RECOMMENDATIONS

Several organizations including the European Group on Tumor Markers (EGTM) (7), The American College of Physicians (8), The European Society for Medical Oncology (9), and the National Comprehensive Cancer Network (10) have developed guidelines for the use of CA125 as a tumor marker for ovarian cancer. In addition, an NIH Consensus Conference on screening, prevention, diagnosis, and treatment of ovarian cancer was held in 1994 (11). Recommendations from these groups are summarized in Table 2. The table

also includes previous recommendations from the NACB as well as current recommendations based on the information below and other established guidelines.

**CA125**

In 1981, Bast *et al* identified the CA125 antigen with the development of the OC 125 murine monoclonal antibody from cell line OVCA 433, which was derived from a patient with ovarian serous carcinoma (12). The CA125 molecule has since been cloned using a partial cDNA sequence originating from the peptide core of the molecule identified (13). This new mucin molecule has been designated CA125/MUC16 (gene MUC16) and consists of a 156-amino-acid tandem repeat region in the *N*-terminus and a possible transmembrane region and tyrosine phosphorylation site in the *C*-terminus.

The first immunoassay for CA125, commercialized in 1983, used the OC 125 antibody for both capture and detection (14, 15). A second-generation assay (CA125 II) was subsequently developed, incorporating M11 and OC 125 antibodies, which have distinct non-overlapping epitopes. Assays for CA125 have since been adapted to automated platforms and although the majority of manufacturers quote a similar reference interval, concentrations of CA125 may vary among manufacturers due to differences in assay design and reagent specificities. Thus, values from different methods are not interchangeable and patients who are serially monitored should be re-baselined if there is a change in methodology (16). Laboratory reports should also indicate the specific assay used.

The cut-off of 35 U/mL for the CA125 and CA125II assays was determined from the distribution of values in healthy individuals. Values tend to decline with menopause and aging. It has recently been reported that CA125II concentrations vary 20–50% by race in postmenopausal women, with concentrations in African and Asian women lower than in Caucasian women (17). Menstrual cycle variations can also be found, with increases during the follicular phase. Elevated values may be found in 1–2% of normal healthy individuals, 5% of those with benign diseases, and 28% of those with non-gynecologic cancers. (14, 15, 18).

It is recommended that analysis be performed shortly after the prompt centrifugation of the specimen and separation of serum from the clot, and that specimens be stored at either 4°C (1–5 days) or –20°C (2 weeks–3 months) in the short term or –70°C in the long term to ensure stability (18). Plasma is an acceptable specimen type for some assays. As in other immunoassays, assay interferences may be observed if heterophilic antibodies are present in the serum, particularly following therapeutic or diagnostic use of monoclonal antibodies.

The recommendations of the current NACB panel and other groups with respect to the potential clinical utility for CA125 are summarized in Table 2 and described below.

*Screening / early detection*. In women with epithelial ovarian cancer, 80% have CA125 levels >35 U/mL, with elevations of 50% in clinically detected stage I disease, 90% in stage II, and >90% in stages III and IV (15). Concentrations correlate with tumor burden and stage. Due to the lack of sensitivity and specificity for a single determination of the marker, CA125 is not recommended for use in screening asymptomatic women by the NACB panel as well as other authoritative organizations (7-11). A National Cancer Institute (NCI) consensus development panel concluded that neither CA125 nor transvaginal ultrasonography effectively reduces mortality from ovarian cancer (11). The same panel did recommend annual CA125 determinations, in addition to pelvic and ultrasound examinations, in women with a history of hereditary ovarian cancer who have an estimated lifetime risk of 40%.

A number of approaches have been proposed to improve the specificity of CA125 for early detection as very high specificity (99.7%) is needed to achieve an acceptable positive predictive value of 10% with a prevalence of disease of 40 per 100,000 in women over age 50 (19). Strategies have included combining with or employing a two-stage strategy of CA125 with ultrasound, longitudinal measurements of CA125, and combining measurement of CA125 with other markers such as OVX1, M-CSF or other new biomarkers discovered using proteomic profiling approaches (14, 19-21). A large clinical trial is currently underway in the United Kingdom that will involve 200,000 women and will be adequately powered to detect a significant improvement in survival among women screened with serial CA125 measurements and transvaginal sonography.

*Discrimination of pelvic masses.* In contrast to its use in early detection, CA125 is more widely accepted as an adjunct in distinguishing benign from malignant disease in women, particularly in post-menopausal women presenting with ovarian masses (10, 11). Benign conditions resulting in elevated CA125 levels may be a confounding factor in pre-menopausal women. Sensitivities of 71–78% and specificities of 75–94% have been reported in different studies. Elevated concentrations of CA125 >95 U/mL in post-menopausal women can discriminate malignant from benign pelvic masses with a positive predictive value of 95% (14). Therefore, based on current evidence, CA125 is

recommended as an adjunct in distinguishing benign from malignant pelvic masses, particularly in postmenopausal women.

*Post-operative use.* Early studies on CA125 indicated that it was useful post-operatively in predicting the likelihood that tumor would be found at a second-look operation, therefore CA125 assays were initially approved by the Food and Drug Administration (FDA) for this indication (15, 22). Elevations of CA125 greater than 35 U/mL after debulking surgery and chemotherapy indicate that residual disease is likely (>95% accuracy) and that chemotherapy will be required. Second-look laparotomy is now considered to be controversial and suggested only for patients enrolled in clinical trials or in situations when surgical findings would alter clinical management. Monitoring with CA125 testing in women with elevated pre-operative CA125 concentrations, along with a routine history and physical, and recto-vaginal pelvic examination, has been advocated instead of surgery for asymptomatic women after primary therapy (11). Elevated, rising, or doubling CA125 concentrations predict relapse. However, negative values do not exclude disease presence.

Although monitoring intervals are as yet undefined, current practice suggests following patients every two to four months for two years and then less frequently (10). Elevations in CA125 can precede clinical or radiological evidence of recurrence with a median time of two to six months, although there is no evidence to date that initiating salvage chemotherapy prior to clinical recurrence improves survival (23). Early detection of recurrent disease, however, permits the timely evaluation of the multiple drugs available for salvage therapy. As only a fraction of patients will respond to any single drug and as reliable predictive tests are not yet available, chemotherapeutic agents are generally used individually and sequentially to identify those drugs that are active against a particular patient's cancer. Given the modest difference between time to recurrence and overall survival, early detection of recurrence provides time in which to find effective palliative therapy. Therefore, measurement of CA125 at follow-up visits is recommended if values were initially elevated.

*Monitoring Treatment.* Serial measurement of CA125 may also play a role in monitoring response to chemotherapy. Declining CA125 concentrations appear to correlate with treatment response even when disease is not detectable by either palpation or imaging. In a meta-analysis, serial CA125 concentrations in 89% of 531 patients correlated with clinical outcome of disease (22, 24, 25). There is general consensus among current guidelines in recommending that CA125 be used to monitor therapeutic response. The Gynecologic

Cancer Intergroup (GCIC) defines a response as a reduction of 50% or more in pre-treatment CA125 level that is maintained for at least 28 days (26-28). The pretreatment sample must be at least twice the upper limit of the reference range. The first sample is recommended within 2 weeks prior to treatment with subsequent samples at 2-4 weeks during treatment and at intervals of 2 to 3 weeks during follow-up. The same assay method is required throughout and patients who received immunotherapy (mouse antibodies) cannot be evaluated. In addition to monitoring initial chemotherapeutic regiments, CA125 measurements may be useful in monitoring salvage therapy, because a doubling of values is associated with disease progression and treatment failure in more than 90% of cases. However, disease progression may also occur without an increase in CA125, and therefore the presence of tumor should also be assessed by physical examination and imaging (18). Serial measurement of CA125 to aid in the monitoring response to therapy is a second FDA indicated use for the marker.

*Prognosis.* CA125 is recommended during primary therapy as a potential prognostic marker since CA125 concentrations, both pre-operative and post-operative, may be of prognostic significance (29-31). After primary surgery and chemotherapy, declines in CA125 concentrations during chemotherapy have generally been observed to be independent prognostic factors, and in some studies the most important indicator. Persistent elevations indicate a poor prognosis. In patients who had a pre-operative CA125 concentration >65 U/mL, the five-year survival rates were significantly lower and conferred a 6.37-fold risk of death compared to patients who had values less than 65 U/mL (15, 25). In addition to the measured level, the half-life of the CA125 marker indicates prognosis after chemotherapy. A half-life of less than 20 days was associated with significantly improved survival (28 months vs. 19 months) as compared to greater than 20 days (14, 32). Improved survival also correlates with normalization of CA125 after three cycles of combination chemotherapy.

**Other markers for ovarian cancer**

Several other potential tumor-associated markers have been reported in body fluid and tissue of ovarian cancer patients. Although these experimental markers could represent promising new biomarkers for future ovarian cancer screening, diagnosis, and monitoring, it is uncertain whether they will become viable clinical tools, i.e., their clinical usefulness needs to be validated by assessing the sensitivity and specificity in larger groups of patients with stage I disease.

*The kallikrein family.* Kallikreins are a subgroup of the serine protease enzyme family that play an important role in the progression and metastasis of human cancers (33). Kallikreins 4,5, 6, 7, 8, 9, 10, 11, 13, 14 and15 in ovarian cancer have been shown to have value in detection, diagnosis, prognosis prediction and monitoring of ovarian cancer (34-51). Kallikrein 4, for example, is expressed in the majority of serous carcinomas but rarely in normal ovarian surface epithelium (37, 38). Kallikrein 4 expression is associated with higher clinical stage and tumor grade in ovarian cancer: a univariate survival analysis revealed that patients with ovarian tumors positive for kallikrein 4 expression had an increased risk for relapse and death (38). Similarly, kallikrein 5 has been suggested to be a useful independent prognostic indicator in patients with stage I and II diseases (39). Assessment of kallikrein 5 expression could help oncologists determine those who are at higher risk of relapse. Kallikrein 7 expression in ovarian cancer tissue is associated with poorer prognosis of ovarian cancer patients, especially those with lower grade disease and those who have been optimally debulked (52). In contrast, kallikrein 8 (neuropsin or ovasin) (40), kallikrein 9 (53) and kallikrein 11 (50) are favorable prognostic markers in ovarian cancer. Patients with higher kallikrein 8 expression in their tumors have lower-grade disease, lower residual tumor, longer survival, and low rate of recurrence. In a multivariate analysis, higher kallikrein 8 expression was significantly associated with longer disease-free survival. As well as their roles as tissue markers, kallikrein 6, 10, 11 can be detected in serum, and are potential serological markers of the disease (34, 36, 54).

*Osteopontin.* Osteopontin was first identified by a cDNA microarray approach used to identify up-regulated genes in ovarian cancer cells and osteopontin has been found as a potential diagnostic biomarker for ovarian cancer (55). In the original report, osteopontin expression was higher in invasive ovarian cancer than in borderline ovarian tumors, benign ovarian tumors and normal ovarian surface epithelium (55). Plasma levels of osteopontin were significantly higher in patients with epithelial ovarian cancer when compared to healthy controls, patients with benign ovarian disease and patients with other gynecologic cancers. In a more recent report (56), osteopontin has been shown to be less sensitive than CA125 in predicting clinical response to therapy. However, osteopontin increased earlier than CA125 in 90% of the study patients who developed recurrent disease, indicating that osteopontin may be a clinically useful adjunct to CA125 in detecting recurrent ovarian cancer.

*Prostasin.* Using gene expression profiling by cDNA microarrays, Mok *et al*. have identified an overexpressed gene called prostasin that produces a secretory product (57). Prostasin was originally isolated from human seminal fluid and its highest levels are found in the prostate gland (58). Prostasin was detected more strongly in ovarian carcinoma than in normal ovarian tissue. The mean level of serum prostasin was 13.7 μg/mL in patients with ovarian cancer and 7.5 μg/mL in control subjects. In a series of patients with non-mucinous ovarian carcinoma, the combination of prostasin and CA125 gave a sensitivity of 92% and a specificity of 94% for detecting ovarian cancer. Although the above finding is promising, prostasin should be investigated further as a screening or tumor marker, both alone and in combination with CA125.

*Tissue polypeptide antigen (TPA).* TPA is a single chain polypeptide which may represent proteolytic fragments of the cytokeratins (59). Production of TPA may be associated with rapid cell turnover, and elevated TPA levels in serum have been reported in patients suffering from cancers and probably other disease (60). In ovarian cancers of serous and mucinous type, TPA levels correlate with FIGO stage. Thirty-three to 50% of patients with stage I–II disease, and 88–96% of patients with stage III–IV disease, presented with elevated serum TPA. Serial TPA measurements correlated with the clinical course of ovarian cancer in 42–79% of the matched event. These findings suggest that TPA may be a potential marker for following ovarian cancer in patients.

*Lysophosphatidic acid (LPA).* LPA was first identified in ascites of ovarian cancer patients and has since been demonstrated to play a biological role in ovarian cancer cell growth (61-64). In a preliminary study in a small number of patients (61), plasma LPA concentrations were elevated in 90% of patients with stage I disease and 100% of patients with advanced and recurrent disease compared to controls without apparent diseases, although 80% of women with other gynecologic cancers also had elevated levels. CA125 concentrations appeared to complement LPA levels.

*Tumor-associated trypsin inhibitor (TATI).* TATI was first identified from the urine of patients with ovarian cancer (65). The amino acid sequence and biochemical properties of TATI are identical to those of pancreatic secretory trypsin inhibitor (66). Elevated serum and urinary concentrations of TATI are frequently observed in post-operative patients, in severe inflammatory diseases, and in various types of cancer, especially gynecological and

pancreatic cancer (60). Increased concentrations of TATI can be observed in ovarian cancers, especially the mucinous types. The elevated serum levels of TATI appear to correlate with higher stages of disease. In one report, the sensitivity is only 8% in patients with stage I-II and 62% of patients with stage III-IV (67). Several reports suggest that TATI is not a good marker for monitoring disease during therapy, as TATI had a lower sensitivity for residual tumor than CA125, and less than 50% of the matched clinical events are observed to correlate serum levels of TATI.

*Carcinoembryonic Antigen (CEA).* CEA is an oncofetal antigen (60) and elevated serum levels of CEA are frequently found in a variety of benign diseases and cancers, including ovarian carcinoma. The frequency of elevated concentration in ovarian carcinoma varies with the histological type and disease stage, generally being higher in patients with mucinous ovarian cancers and with metastatic disease. The sensitivity of CEA as a marker to detect ovarian cancer is approximately 25%, and the positive predictive value of an elevated CEA concentration is only 14% (60). Although CEA is not a marker for early diagnosis due to its low sensitivity, CEA can be useful in determining treatment response in ovarian cancer patients.

*Cancer-Associated Serum Antigen (CASA).* CASA was initially defined by a monoclonal antibody that bound to an epitope on the polymorphic epithelial mucin (68). Elevated CASA levels in serum were found in individuals in the later stage of pregnancy, in the elderly, in smokers and in patients with cancers. CASA is expressed in all histological types of ovarian cancer and appears to have a sensitivity of 46-73% in patients with ovarian cancer (60). Only a few studies have indicated that CASA is a potentially useful marker in monitoring ovarian cancer. Ward *et al* reported that inclusion of CASA in a diagnostic tumor panel might improve the detection of residual disease by increasing the sensitivity from 33% to 62% and the negative predictive value from 66% to 78% (69, 70). One study has demonstrated that CASA can detect more cases with small volume disease than CA125, and that 50% of patients with microscopic disease are detected by CASA alone (60). Another study has shown that the prognostic value of post-operative serum CASA level is superior to CA125 and other parameters including residual disease, histological type, tumor grade, and the cisplatin-based chemotherapy (71)

*Plasminogen activator inhibitor-1 and -2 (PAI-1 and -2).* Fibrinolytic markers include PAI-1 and PAI-2, for which diagnostic and prognostic values have recently been reported in ovarian cancer (72). In this pilot study, PAI-1 appeared to be a poor prognostic factor (73). as plasma levels of PAI-1 are significantly higher in patients with ovarian cancer, and their levels correlate with the diseases at higher clinical stages. Whether PAI-1 can be used clinically for screening and/or monitoring ovarian cancer awaits further studies, including correlation with clinical treatment events and comparison with CA125. In contrast, expression of PAI-2 in tumors has been shown to be a favorable prognostic factor in ovarian cancer patients (72).

*Interleukin-6 (IL-6).* High levels of IL-6 have been detected in the serum and ascites of ovarian cancer patients (74). IL-6 correlates with tumor burden, clinical disease status, and survival time of patients with ovarian cancer, implying that this marker may be useful in diagnosis. Based on a multivariate analysis, investigators have found serum levels of IL-6 to be of prognostic value, but less sensitive than CA125 (75, 76).

*Human chorionic gonadotropin (hCG).* hCG normally is produced by the trophoblast, and clinically has been used as a serum or urine marker for pregnancy and gestational trophoblastic disease (77). Ectopic hCG production, however, has been detected in a variety of human cancers. Recent studies have demonstrated that the immunoreactivity of total hCG in serum and urine (urinary β-core fragment, hCGβcf) provides a strong independent prognostic factor in ovarian carcinoma, and its prognostic value is similar to that of grade and stage (78, 79). When serum hCG is normal, the five-year survival rate can be as high as 80%, but it is only 22% when hCG is elevated (78). In patients with stage III or IV and minimal residual disease, the five-year survival is 75% if hCG is not detectable compared to 0% if hCG is elevated. Similarly, hCGβcf can be detected in urine in 84% of ovarian cancer patients (79). The incidence of positive urinary hCGβcf correlates with disease progression (a higher proportion of patients is seen in advanced clinical stages). Although the availability of this marker before surgery could facilitate selection of treatment modalities, the clinical application of hCG and its free beta-subunit (hCGβ) for screening and diagnosis is limited. Since several different types of tumors can produce hCG±hCGβ and only a small proportion of ovarian tumors express these, detection of serum hCG±hCGβ or urinary hCGβcf will not provide a specific or sensitive tool for screening or diagnosis in ovarian cancer.

*Her-2/neu.* The c-erbB-2 oncogene expresses a transmembrane protein, p185, with intrinsic tyrosine kinase activity, also known as Her-2/neu. Amplification of Her2/neu has been found in several human cancers, including ovarian carcinoma. In ovarian cancer, 9% to 38% of patients have elevated levels of p105, the shed extracellular domain of the HER-2/neu protein (80-82). According to one report, measurement of Her2/neu alone or in combination with CA125 is not useful for differentiating benign from malignant ovarian tumors (82). However, elevation of p105 in serum or the over-expression immunohistochemically of Her2/neu in tumors has correlated with an aggressive tumor type, advanced clinical stages, and poor clinical outcome (83). Screening for increased p105 levels might therefore make it possible to identify a subset of high-risk patients (81). Furthermore, the test could be potentially useful for detecting recurrent disease.

*AKT2 gene.* The AKT2 gene is one of the human homologues of v-akt, the transduced oncogene of the AKT8 virus, which experimentally induces lymphomas in mice. AKT2, which codes for a serine-threonine protein kinase, is activated by growth factors and other oncogenes such as v-Ha-ras and v-src through phosphatidylinositol 3-kinase in human ovarian cancer cells (84, 85). Studies have shown that the AKT2 gene is amplified and overexpressed in approximately 12–36% of ovarian carcinomas (86-88). In contrast, AKT2 alteration was not detected in 24 benign or borderline tumors.

Ovarian cancer patients with AKT2 alterations appear to have a poor prognosis. Amplification of AKT2 is more frequently found in histologically high-grade tumors or tumors at advanced stages (III or IV), suggesting that AKT2 gene overexpression, like c-erbB-2, may be associated with tumor aggressiveness (87).

*Mitogen activated protein kinase (MAPK).* Activation of MAPK occurs in response to various growth stimulating signals and as a result of activating mutations of the upstream regulators, KRAS and BRAF, which can be found in many types of human cancer. Activation of MAPK activates downstream cellular targets (89, 90) including a variety of cellular and nuclear proteins. Two studies have reported that expression of active MAPK in ovarian cancer tissue or ascites cells correlates with better prognosis in the advanced stage ovarian cancer (91, 92).

## CONCLUSION

The NACB Panel recommends CA125 as the only marker for clinical use in ovarian cancer for the following indications: early detection in combination with trans-vaginal ultrasound (TVUS) in hereditary syndromes, differential diagnosis in suspicious pelvic mass, detection of recurrence, monitoring of therapy and prognosis. The NACB Panel does not recommend CA125 for screening of ovarian cancer. All other markers are either in the evaluation phase or in the research/discovery phase. Therefore the NACB Panel does not recommend these biomarkers for clinical use in ovarian cancer.

**Table 1. Currently available serum markers for ovarian cancer.**

| Cancer Marker | Year discovered | Proposed Uses | Phase of Development | LOE | References |
|---|---|---|---|---|---|
| CA125 | 1981 | Tumor monitoring | Accepted clinical use | I, II | (10, 11, 14, 28, 93-97) |
| Osteopontin | 2002 | Tumor monitoring | Research/Discovery | III, IV | (55, 56, 98, 99) |
| CEA | 1995 | Tumor monitoring | Research/Discovery | IV | (60) |
| Tumor-associated trypsin inhibitor (TATI) | 1991 | Tumor monitoring | Research/Discovery | IV, V | (67) |
| Kallikreins 5,6, 7, 8, 9,10, 11,13,14,15 | 1996 | Differential diagnosis, Tumor monitoring, Prognosis prediction | Research/Discovery | IV, V | (33-53) |
| Lysophosphatidic acid (LPA) | 1995 | Detection | Evaluation | IV, V | (61, 100) |
| Tissue polypeptide antigen (TPA) | 1994 | Tumor monitoring | Research/Discovery | IV | (59, 60) |
| Cancer-associated serum antigen (CASA) | 1991 | Tumor monitoring, Prognosis prediction | Research/Discovery | IV | (60, 69-71, 101) |
| Plasminogen activator inhibitor-1 (PAI-1) | 1995 | Prognosis prediction | Research/Discovery | V | (72, 73, 102) |
| Interleukin-6 (IL-6) | 1995 | Prognosis prediction | Research/Discovery | IV | (74-76) |
| Prostasin | 2001 | Differential diagnosis | Research/Discovery | IV | (57) |
| Urinary β-core hCG (hCGβcf) | 2000 | Prognosis prediction | Evaluation | III, IV | (78, 79) |
| Insulin-like growth factor binding protein–2 (IGFBP-2) | 2004 | Prognosis prediction | Research/Discovery | IV | (103) |
| Tumor released DNA | 2002 | Detection | Research/Discovery | IV | (104-106) |
| HLA-G | 1990 | Differential diagnosis | Research/Discovery | V | (107) |
| Her-2/neu | 1985 | Tissue marker for prognosis prediction and treatment outcome prediction of Herceptin | Evaluation | IV | (108) |
| Akt-2 | 1987 | Tissue marker for prognosis Prediction | Research/Discovery | V | (87) |
| MAPK | 2003 | Tissue marker for prognosis prediction | Research/Discovery | V | (91, 92) |

LOE: Levels of evidence, Hayes, et al (6);

**Table 2. Recommendations for use of CA125 as a tumor marker in ovarian cancer by different Expert Groups**

| Use | American College of Physicians (8) | EGTM (7) | ESMO (9) | NACB and EGTM 2002 (18) | NCCN (10) | NIH Panel (11) | NACB 2005 |
|---|---|---|---|---|---|---|---|
| Screening - no family history or other risk factors | **No** | **No** | None published | **No** | None published | **No** | **No** |
| Early detection in hereditary syndromes - with trans-vaginal ultrasound (TVUS) | **No** | **Yes** | None published | **Yes** | None published | **Yes** | **Yes** |
| Differential diagnosis - suspicious pelvic mass | None published | **Yes** [Post-menopausal women only] | None published | **Yes** [Post-menopausal women only] | **Yes** | **Yes** [Post-menopausal women] | **Yes** [Post-menopausal women] |
| Detection of recurrence | None published | **Yes** | **Yes** | **Yes** | **Yes** | **Yes** | **Yes** |
| Monitoring therapy | None published | **Yes** | **Yes** | **Yes** | **Yes** | None published | **Yes** |
| Prognosis | None published | **Yes** | **Yes** | **Yes** | None published | **Yes** | **Yes** |

EGTM, European Group on Tumor Markers; ESMO, European Society for Medical Oncology; NACB, National Academy for Clinical Biochemistry; NCCN, National Comprehensive Cancer Network; NIH, National Institutes of Health. Recommendation: Yes or No or None published.

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