ejpmr, 2025, 12(3), 581-602

SJIF Impact Factor 7.065

EUROPEAN JOURNAL OF PHARMACEUTICAL AND MEDICAL RESEARCH

www.ejpmr.com

Review Article

ISSN 2394-3211

EJPMR

HE-4 IS ADMIRABLE AND APPRECIABLE THAN CA-125 IN MONITORING OF EPITHELIAL OVARIAN CARCINOMA CANDIDATES

Shinymol[1]* and Umesh Verma[2]

[1]Assistant Professor, Department of Pharmacy, Smt. Manjira Devi University, Rukmani Nagar, Hitanu Dhanari, Dunda, Uttarkashi, Uttarakhand 249151, India.

[2]Assistant Professor, Faculty of Pharmacy, Maharaja Agrasen Himalayan Garhwal University, Pokhra, Pauri Garhwal-246169, Uttarakhand (India).

***Corresponding Author: Shinymol**

Assistant Professor, Department of Pharmacy, Smt. Manjira Devi University, Rukmani Nagar, Hitanu Dhanari, Dunda, Uttarkashi, Uttarakhand 249151, India.

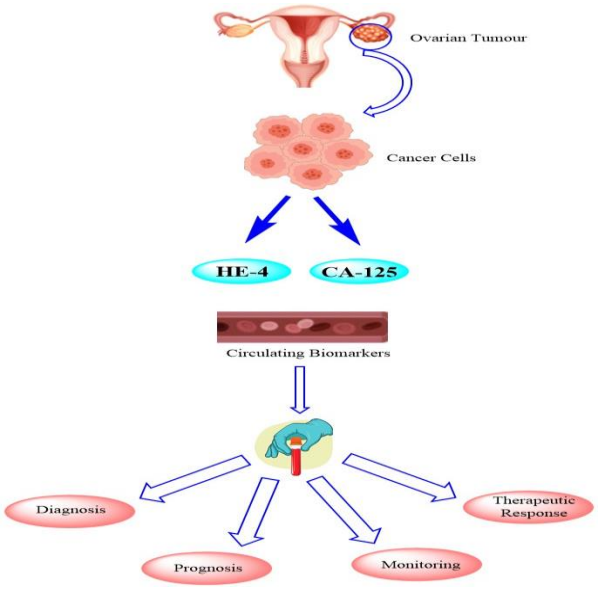
Article Received on 28/01/2025 **Article Revised on 18/02/2025** **Article Accepted on 08/03/2025**

ABSTRACT

The deadliest type of gynecological carcinoma is still EOC, which is identified by its late-stage identification and silent proliferation. To increase survival rates, it is essential to identify the illness early on and with accuracy. The use of tumor markers in the diagnosis and therapy of OC has become increasingly important. This paper explains whether serum concentrations of cancer antigen 125 (CA-125) and human epididymis protein 4 (HE-4) could predict the surgical outcome of EOC. It also examines the diagnostic significance of tumor markers such as CA-125 and HE-4. We also talked about the tumor marker's (HE-4) value in ovarian cancer (OC)patients, both during and after treatment. When used alone, the tumor marker CA-125 is related to poor specificity and low sensitivity, particularly in premenopausal women, for early or stage I disease. Serum HE-4 is a useful biomarker for differentiating benign ovarian disease from ovarian cancer, although it can be influenced by a number of variables, such as age, smoking, and pregnancy. Combining these indicators, or at least two or three of them, is advised for a high sensitivity and specificity early stage EOC diagnosis. HE-4 might be an efficient marker to monitor both during and following OC treatment. For follow-up observations, a complementary role for HE-4 and CA-125 measures was proposed. Numerous researches assessed the predictive role of HE-4 for surgical outcome in primary cytoreductive surgery. Based on results of all studies, HE-4 a high potential biomarker for surgical outcome may be useful in predicting primary treatment.

KEYWORDS: Epithelial ovarian cancer**,** CA-125, HE-4, follow-up.

GRAPHICAL ABSTRACT



HE-4 andCA-125 in relation to ovarian carcinoma.

1. INTRODUCTION

Cancer is any of a large number of diseases characterized by the development of abnormal cells that divide uncontrollably and can infiltrate and destroy normal body tissues. It is one of the causes of death in the developed Nations.[1-3] Despite the availability of several cancer medications for both prevention and control, cancer remains uncontrollable.[3-7] Selective therapies are based on a better understanding of the biology and molecular genetics in the tumor progression utilized for the prospective treatments. These are in addition to common cancer treatments including surgery, radiation therapy, chemotherapy, combination therapy, and laser therapy. Chemotherapy remains a viable therapeutic option for cancer today, despite these advancements. Currently, the invasion and metastasis phases account for 90% of treatment failures.[8] Initial identification of cancer increases survival. But when they are discovered, almost 50% of tumors are already advanced. Early intervention is possible to try to reduce or prevent the growth and lethality of cancer. Before all cancers can be identified at an early stage, a number of challenges must be overcome.[9]

The study in 2022 investigated the cancerincidence rate in India.(Fig. (1) & Fig. (2)) The purpose of this study is to present a report on the anatomical sites, age groups, and sex-specific cancer incidence predicted for India in 2022. Theincidence of cancer for the years 2012–2016 from 28 PBCRs was reported in the National Cancer Registry Programme Report 2020. The aim of assigning PBCRs to the nation's States and regions was to improve our comprehension of the epidemiology of cancer. To calculate the cancer case count in India for 2022, the incidence rate for each age group distinct anatomical site of cancer was applied to the expected population. In India, the prevalence of cancer is still rising. Planning initiatives for early identification, risk reduction, and treatment of cancer will be made easier with the aid of these updated estimations.[10,11]

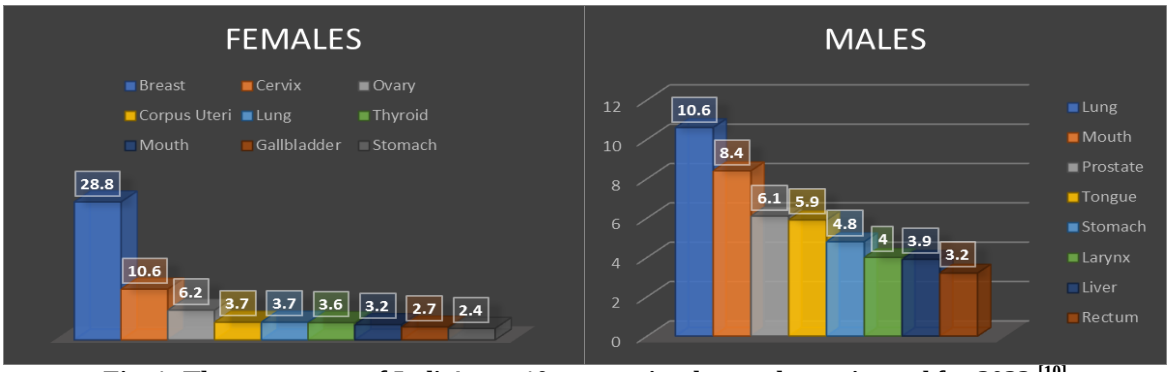


Fig. 1: The percentage of India's top 10 cancer sites by gender, estimated for 2022.[10]

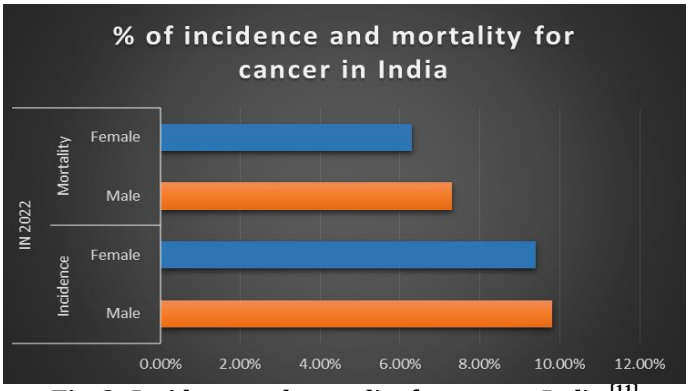


Fig. 2: Incidence and mortality for cancer: India.[11]

2. EPITHELIAL OVARIAN CANCERS

The most typical kind of cancer in ovaries is epithelial. Ninety percent of ovarian tumors are epithelial in nature.[12,13] Ovarian cancer that began in the outer layer of the ovary is known as epithelial ovarian cancer. The majority of ovarian cancers are carcinomas, which arise from the surface epithelium of the ovary.[14,15]

It has been suggested in the past that postovulatory inclusion cysts or the ovarian surface epithelium, which arise following follicular rupture and healing, are the source of the majority of ovarian carcinomas.[16,17] Regarding the incidence of OC in women, numerous hypotheses exist. The "incessant ovulation" theory states that every ovulation creates a wound, which causes an increase in cell proliferation to repair the epithelial cells. This could lead to a higher threat of malignant mutation and DNA damage.[18,19] According to a different theory involving gonadotropin-based stimulation, gonadotropin levels rise after menopause, increasing the prevalence of ovarian cancer.[20-22] The inflammatory hypothesis postulates that inflammation may contribute to the

ovulation process, which is strongly linked to OC.[23] However, progesterone stimulation has shown a protective effect and lowers the probability of carcinoma. In the meantime, the hormonal hypothesis suggests that excess androgen stimulates the ovarian surface epithelium, increasing the hazard of OC.[21]

2.1 Ovarian epithelial carcinoma: Types

Thinking about their histopathology and molecular genetic alterations, the subgroups of EOCs are as follows: (1) High-grade serious, (2) Endometrioid, (3) clean cellular, (4) Mucinous, (5) low-grade serous carcinomas. (Fig. (3)) EOCs represent more than 95% of all OC instances.[24,25]

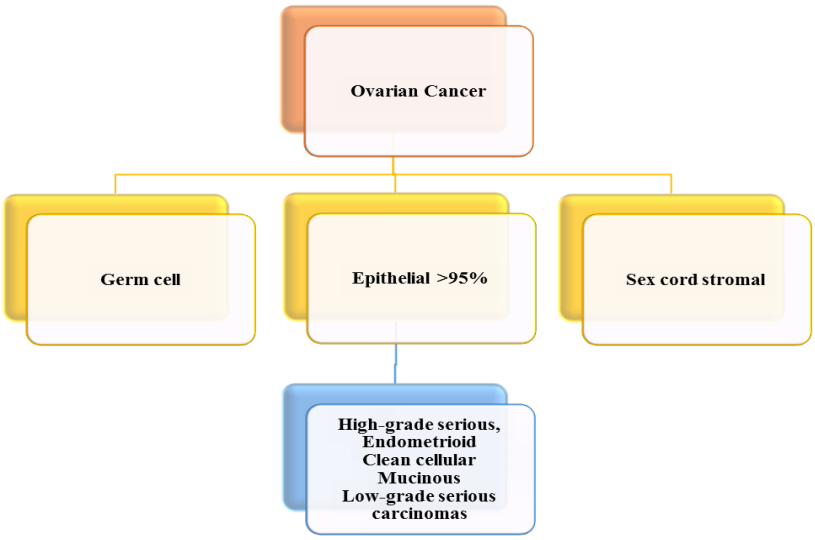


Fig. 3: Classification of ovarian malignancies.[24]

EOC is divided into two categories by the dualistic model, which are called type I and type II.[26,27] Additional histopathological, molecular, and there has been genetic study made possible by an improved model of the two main kinds of ovarian carcinogenesis, known as type-I and type-II. (Fig. (4))[28] Type-I cyst are indolent, slow-growing neoplasms that develop from a distinct precursor called atypical hyperplasia. At the time of diagnosis, these tumors were limited to the ovary and did not exhibit TP53 mutations within a stable genome. Conversely,somatic alterations are often linked to specific genes when it comes to type I tumors.[29] Low-grade serous, mucinous, clear cell carcinoma, and endometrioid adenocarcinoma are examples of type I tumors. Clinically, type II tumors are higher grade, more aggressive neoplasms with extremely unstable genetic makeup. Most of these cancers have TP53 mutations and are detected at an advanced stage. Type II tumors, which include high-grade serous malignancy, have demonstrated their origins in the ovarian surface epithelium and/or the fimbrial part of the fallopian tube.[30-32]

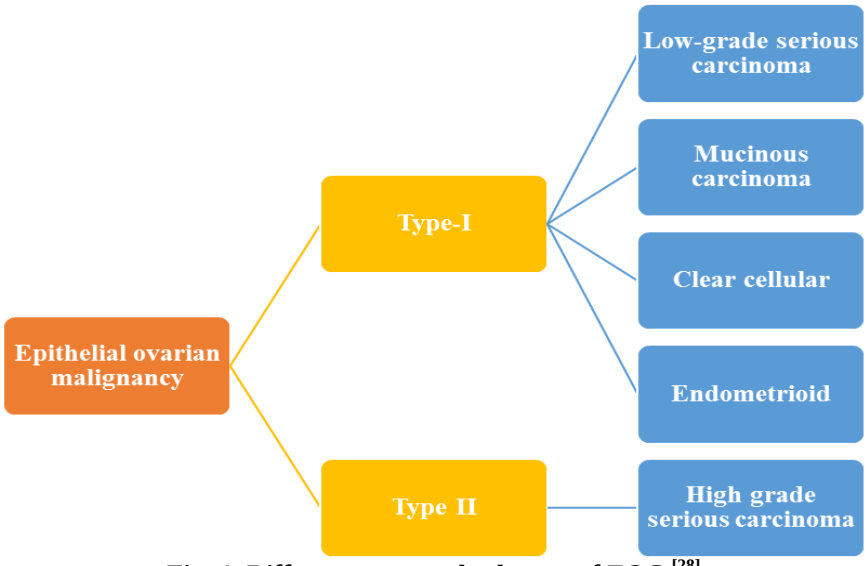


Fig. 4: Different types and subtypes of EOC.[28]

Two stages of OC development are possible. First-stage inclusion cysts, which are thought to arise as a result of repetitive injury and remodeling of the ovarian epithelial surface brought on by regular ovulations, entrap the ovarian surface epithelium in the stroma. In the second stage, hormones cause the inclusion cysts to progressively change into tumor cells. Excessive stimulation by luteinizing hormone is one hormonal component that is strongly implicated. This factor can function directly by activating genes that respond to luteinizing hormone, or indirectly by overstimulating the production of androgens by the ovaries. Women with polycystic ovarian syndrome—who often have higher pituitary luteinizing hormone secretion had a higher hazard of developing ovarian cancer.[33]

Ovarian cancer ranks 3rd in inflicting gynecological cancer losses, and it remains the lethal form of gynecological cancer.[34,35] According to estimates from the American Cancer Society, 22,430 women are expected to get an ovarian cancer diagnosis in 2007, with 15,280 deaths will report due to the disease. The majority of these females will present with an adnexal mass, with or without confirmation of metastatic disease. specially, an adnexal mass or an ovarian cyst will be diagnosed in about 20% of all females at some point in their life, yet only a small probability of these masses represents an ovarian malignancy.The circumstance of EOC is about 60–90 % of all malignant ovarian cancer; the five years of survival for EOC has changed from 30 % to 40 %.[36] The poor rate of survival to occurrence in EOC results from the high probability of cases diagnosed at an advanced stage.[17]

Ovarian cancer is considered to cause around 114,000 deaths and 190,000 new cases annually. The topmost rates are documented in the USA, Scandinavia, Eastern Europe, and Canada.Asia and Africa have lower rates. The threat of epithelial tumours increases with age, being generally in peri- and postmenopausal women. Tumours of germinal or embryonic root are more frequent in adolescent.[12]

Ovarian carcinoma (Fig. (6) & Fig. (7))is a diverse set of neoplasms that make up the seventh most deadly cancer in the world for women. In the western world, it is a leading cause of mortality from gynecological carcinoma. It has been suggested in past that postovulatory inclusion cysts or the ovarian surface epithelium, which develop after follicular rupture and healing, are the source of the majority of ovarian carcinomas. (Fig. (5)).[16]

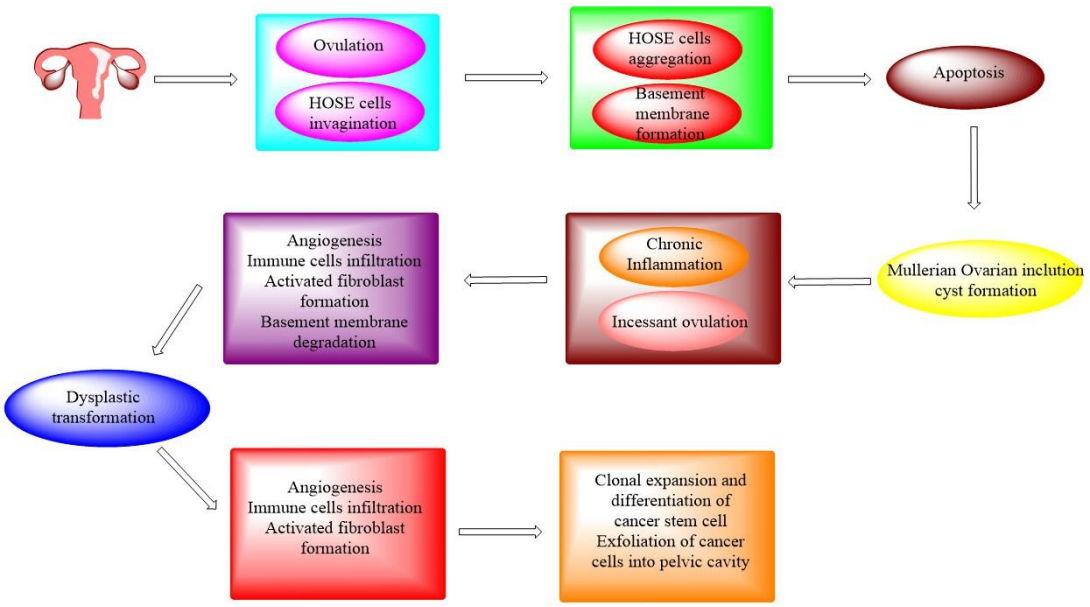


Fig. 5: Beginning of an EOC and the microenvironment of OC.[16]

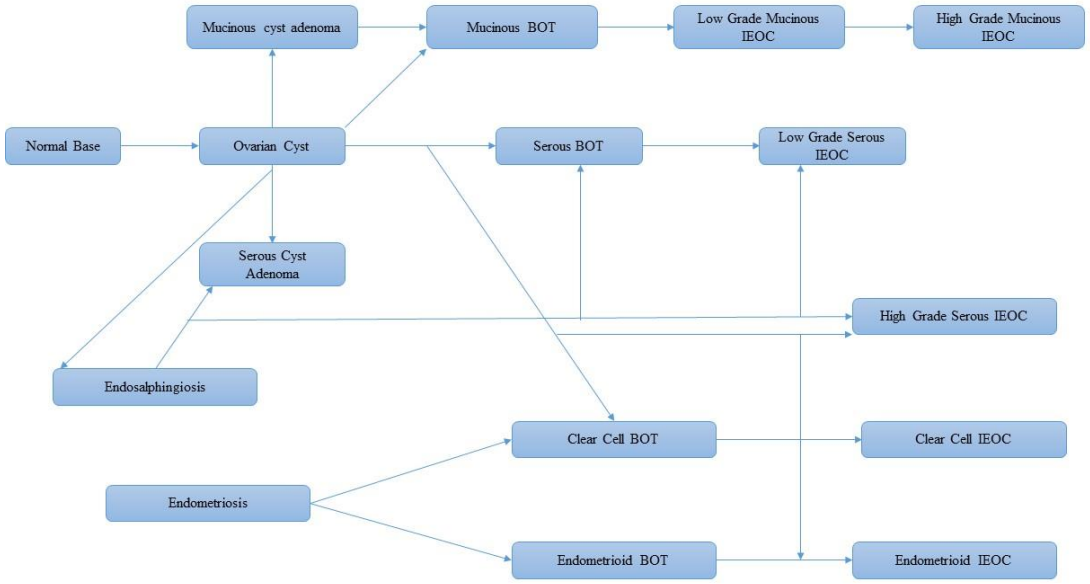


Fig. 6: Pathogenetic pathways of EOC of different histologic types.[16]

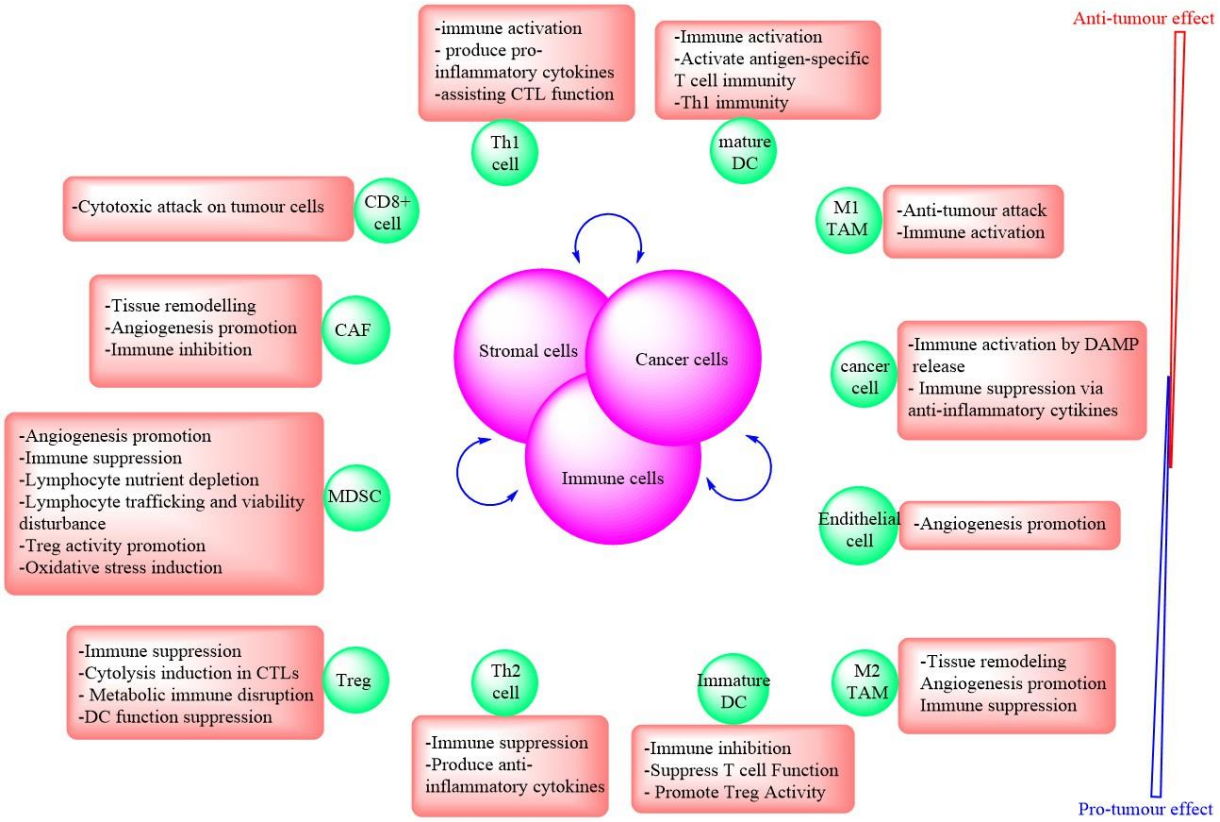


Fig. 7: The components and roles of cells inside the tumor microenvironment (TME).[37,38]

The two main characteristics of ovarian cancer (OC) are silent progression and late-stage diagnosis. Astronomically, there are three divisions of OC grounded on the types of ovarian cells involved. Surface epithelial cells are the cell type in division one, and can cover the ovary and be subdivided into numerous subtypes. The alternate division consists of germ cells, which are the cells that ultimately transform into ova. Yolk sac tumors, immature teratoma, and dysgerminoma are OC subtypes associated with germ cells. Eventually, sex cord – stromal cells comprise the third division. These tumors include malignant granulosa cells and Sertoli – Leydig cells.(Fig. (8)) Among all forms of ovarian cancer, EOC occurs constantly.[27]

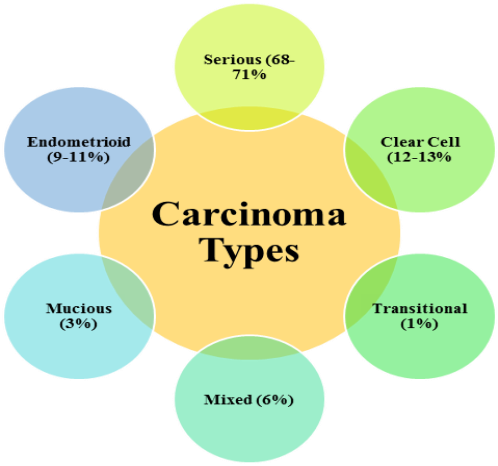


Fig. 8: Relative frequencies of ovarian carcinoma sub-types.[27]

In the USA in 2020, there were 1,806,590 new instances of tumor and 606,520 cancer-related deaths. Specifically, OC seems to be the primary motive of death for female reproductive tract cancers. There were 21,750 new cases and 13,940 deaths related to OC in 2020. Post-menopausal womens are taken into consideration at a high hazard of developing OC due to the fact the likelihood of developing a sophisticated level ailment will increase with age.[2,39,40]

OCs are diagnosed with a complicated stage for around 70% of the instances, resulting in a five-year survival rate of 30%. The 5-year survival rate can exceed 90% while OC is detected early. It is crucial to gain deep knowledge of the molecular causes of OC in the final 25 years. Crucially, new biomarkers ought to aid in the timely diagnosis pathway.[41,42]

2.2. Significance of Early Diagnosis

Among gynecological malignancies, malignant epithelial tumors are the fatal forms of OC. Presently, the only criteria used to categorize ovarian epithelial tumors was the tumor cells' morphology.6 to 9 cases per 100,000 women is the global incidence rate of these cancers.[43]

Up to 90% of individuals can be treated with currently available surgery and chemotherapy if their ovarian cancer is stage I, meaning it has not progressed beyond of their ovaries. On the other hand, approximately 30% or less of patients with illness that has progressed from the pelvis (stages III–IV) can be treated. Only 25% of ovarian tumors are currently identified as stage I. A higher percentage of patients at stage I may be found, which would be beneficial for survival.[44] The absence of early detection strategies and the restricted effectiveness of widespread chemotherapy are the principal elements contributing to this vulnerability.**(**Fig. (9))[39,40] There are strict requirements for an efficient screening technique because OC is so common. A positive predictive value of 10% indicates that there will be ten surgeries for every instance of OC that is discovered, since the disease is typically diagnosed during surgery. A high sensitivity of 75% or higher for early-stage disease and a very high specificity of 99.6% are needed to obtain a positive predictive value of 10% with a prevalence of 1 in 2,500.[12]

OC is a complex and various group of sicknesses. Even though its occurrence is less than that of breast cancers, the outcome of OC is disproportionally higher with large number of deaths. OC proves deadly for the great majority of patients having the diagnosis of advanced (stage III) ovarian tumors. OC is regarded as the deadliest gynecological cancer worldwide. By improving the efficacy of screening methods, consisting of checks for specific biomarkers, the chances of detecting OC at early stage can elevated.[24]

There are strict criteria for an efficient screening technique since ovarian cancer has become so prevalent. As diagnosis of ovarian cancer is generally made at surgery, a positive predictive value of 10% implies ten operations for each case of ovarian cancer diagnosed. A high sensitivity of 75% or higher for early-stage disease and a very high specificity of 99.6% are needed to obtain a positive predictive value of 10% with a prevalence of 1 in 2,500.[44]

Biomarkers may want to have the finest impact on survival in 4 areas: screening, diagnosis, monitoring, and prognostication. During the last 30 years, Cancer Antigen 125 (CA-125) has exhaustively evaluated in most of these regions of EOC.[45]

In the past decade, Human Epididymis Protein 4 (HE-4) has emerged as a precious biomarker for EOC. Compared with CA-125, HE-4 is a superior biomarker to differentiate benign from malignant ovarian cancer.(Table 1& Table 2) Many studies propose that HE4 has the advantage in terms of diagnosis and detecting recurrent ovarian cancer.Moreover, combining CA-125 and HE-4 was a superior predictor of malignancy than a single biomarker.(Fig. (9))[46-49]

Table 1: Test effectiveness formalevolent disease.[47]

	N (34)	Mean	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)
CA-125 >35U/mL	12	29.68	35.29	58.82	46.15	47.62
HE-4 >70pmol/L	25	144.56	73.53	100	100	79.07
HE-4 >150 pmol/L	9		26.47	100	100	57.63

Table 2: CA-125 + HE-4 accuracy in detecting recurrent ovarian cancer.[47]

	Sensitivity (%)	Specificity (%)
CA-125 >35 U/mL + HE-4 >70 pmol/L	76.47	100
CA-125 >35 U/mL + HE-4 >150 pmol/L	44.11	100

2.3 Evaluation and management of ovarian epithelial cancer

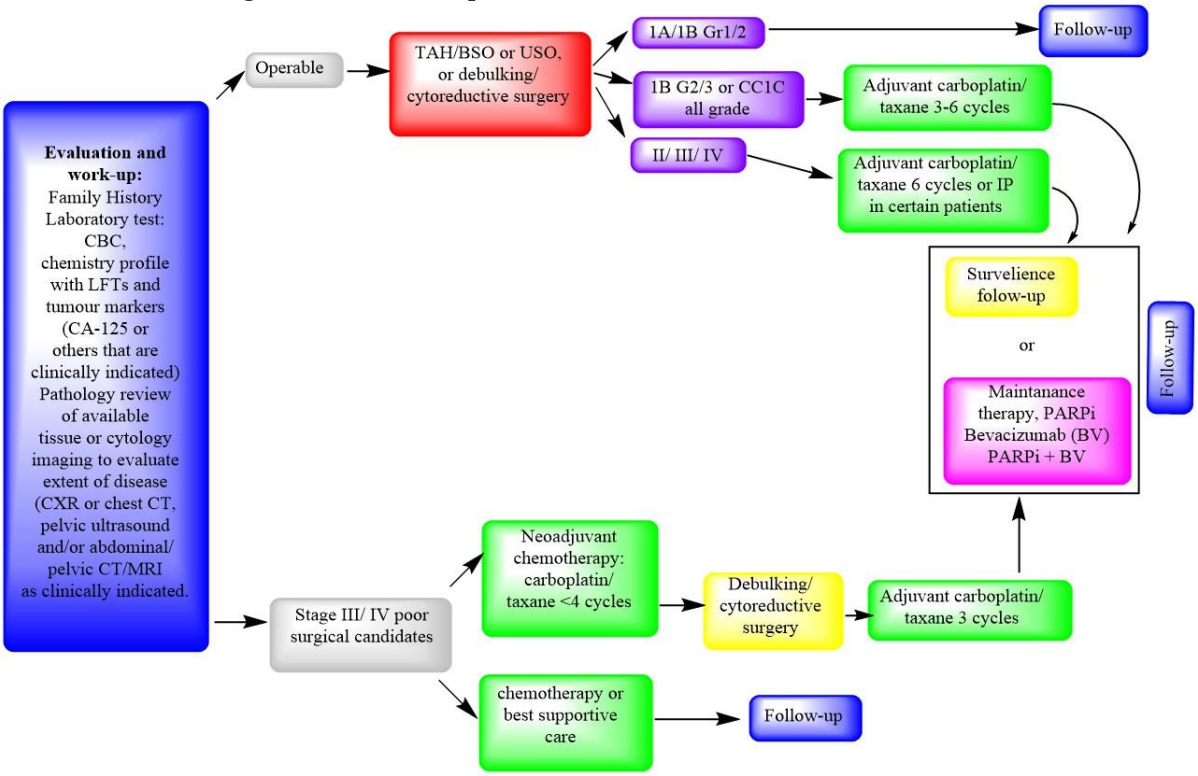


Fig. 9: Evaluation and management of EOC.[50,51]

3. TUMOR MARKERS

A tumor marker is[52]

- A substance produced via a tumor or by the host in response to most cancers mobile;
- Found in bodily fluids, tissues, or cells; and
- Measured qualitatively or quantitatively by way of methods inclusive of chemical, immunological, molecular, and mass spectrometry to pick out the presence of cancer.

Biomarkers, also known as oncomarkers, play a crucial role in cancer studies and treatment. These molecular signatures encompass genes, proteins, and different molecular functions that could function as objective clinical signs. (Table 3.) Biomarkers serve two primary purposes: firstly, they assist in verifying the chance of disorder development or pathological process, and secondly, they aid in evaluating the response to healing interventions. Cancer biomarkers are molecules produced by neoplasm cells or cells of their place and can quantified in frame fluids and blood in cancer screening, diagnosis, and treatment tracking. Biomarkers include antigens, cytoplasmic proteins, enzymes, hormones, receptors, oncogenes, and their byproducts.[42,43]

According to NACB, tumor markers are Tumor markers are "surrogate indicators that increase or decrease the clinician's suspicion that future clinically important events, such as cancer development, recurrence, or development or death of a patient, will or will not happen, and/or that a specific treatment will decrease the risk of such events". These molecules can act as a signal of the existence of a tumor by being produced and discharged by the tumor host cells and found in serum or other bodily fluids.[53]

3.1 Tumor markers classification

Table 3: Tumor markers classification.[52]

Class	Examples
Enzymes, isoenzymes	PSA, LDH, and neuron-specific enolase
Hormones	hCG, calcitonin, ACTH, gastrin, and VIP
Proteins/peptides	β2-Microglobulin, NMP22, progastrin-releasing peptide, and thyroglobulin
Oncofetal antigens	AFP and CEA
Carbohydrates	CA 125, CA 15-3, and CA 27.29
Blood group antigens	CA 19-9 and CA 72-4
Receptors	Estrogen and progesterone
Gene mutations and overexpression	BRCA1, BRCA2, and HER-2/neu
Other	Circulating tumor cells, and cell-free nucleic acids

3.2 FDA Approved Biomarker

For many years, CA125 was the only ovarian cancer marker that FDA provided permission for use in monitoring treatment outcomes and identifying recurrence of the disease. But in recent years, three novel serum-based tests/algorithms for the treatment of OC have been approved owning the explosion of high-throughput technology-driven biomarker discovery trials. The FDA authorized HE4 in 2009 in order to monitor treatment and identify recurrences of disease. Soon after, the FDA approved the OVA1™ and the Risk of Ovarian Malignancy Algorithm (ROMA) tests to assess the risk of malignancy in women who were postmenopausal and premenopausal and who presented with an adnexal tumor.[54]

3.3 The following list of characteristics represents what a perfect tumor biomarker should have[55]

- It should be very specific, meaning it should only target a single kind of tumor.
- Needs to be highly sensitive; benign or physiological tumors shouldn't be detected up on.
- Levels should correspond with the size and features of the tumor.
- It is important to understand the prognostic and predictive value of tumor biomarkers.
- There should be frequent, short half-lives, and serial monitoring is possible.
- Simple to use and inexpensive.
- Suitable for a screening examination.
- It should be simple to take samples.

Considering their nature, the potential biomarkers for OC can be classified as gene, protein, metabolite, or miRNA-based biomarkers.[55]

Perfect biomarkers own ideal standards like high sensitivity and specificity to a particular tumor type, affected person recognition, positive and negative predictive values for predictive and prognostic benefits, and clinical validation through potential trials. (Fig. (10)) However, presently, no biomarker fulfils all these ideal standards. Biomarkers are categorized primarily based on their application, including screening, detection of tumor presence or absence, analysis, and identification of molecular goals for novel treatments.[56,57]

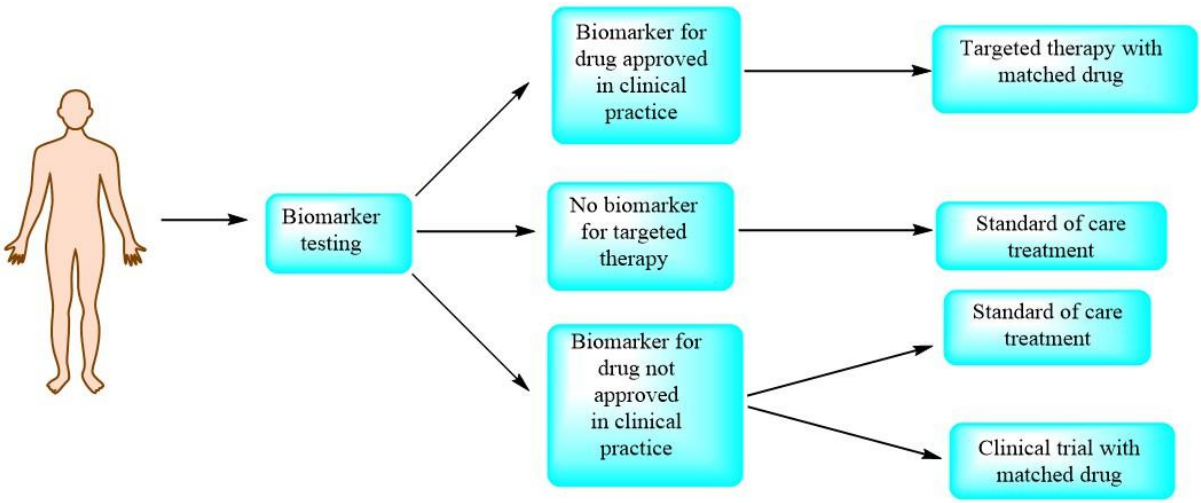


Fig. 10: The use of biomarker testing to inform treatment choices for cancer patients with metastases.[57]

The exploration for tumor biomarkers is more suitable via the evaluation of frame fluids similarto saliva, urine, and blood/serum/plasma using minimally invasive and non-invasive ways. Presently, there could be a selected emphasis on urine as an important waste material this is effortlessly accessible, gives a bigger volume, and possesses a lower proteome complexity compared to blood.(Fig. (11)).[58-60]

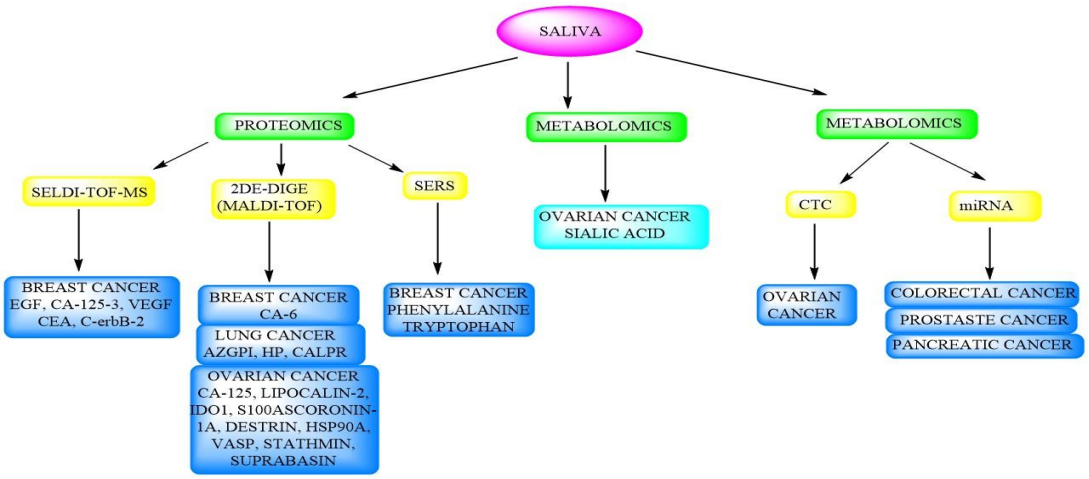


Fig. 11. Recent and advanced technology have revealed biomarkers for a variety of cancers from saliva.[58,61]

Those urine-based biomarkers maintain promising prospects for the detection and monitoring of OC, force possibilities for more desirable diagnostics,and greater powerful control of the disorder.[62,63]

3.4 Technologies for detecting biomarkers in OC

Numerous cancer forms have their gene expression linked to different characteristics of disease through high-throughput cellular transcriptome analysis approaches. Technologies including expression microarrays, CGH, real-time PCR, and next-generation sequencing (NGS), which arebeing used at present in ovarian cancer research, enable genome-wide scanning and the discovery of changed genes linked to cancer.[55]

Depending on the type of analyte, tumor markers in bodily fluids or tissue can be quantified or qualitatively assessed using a variety of techniques. In addition to molecular approaches, they could be enzyme assays, immunoassays, immunohistochemistry, receptor assays, flow cytometry, or mass spectrometry. The invention of monoclonal antibodies and the RIA and ELISA techniques in the 1960s and 1970s greatly aided in the identification and study of tumor markers as well as their clinical application. Therefore, immunoassay is utilized to measure the most of the tumor markers now utilized in urineand serum. (Fig. (12)).[52]

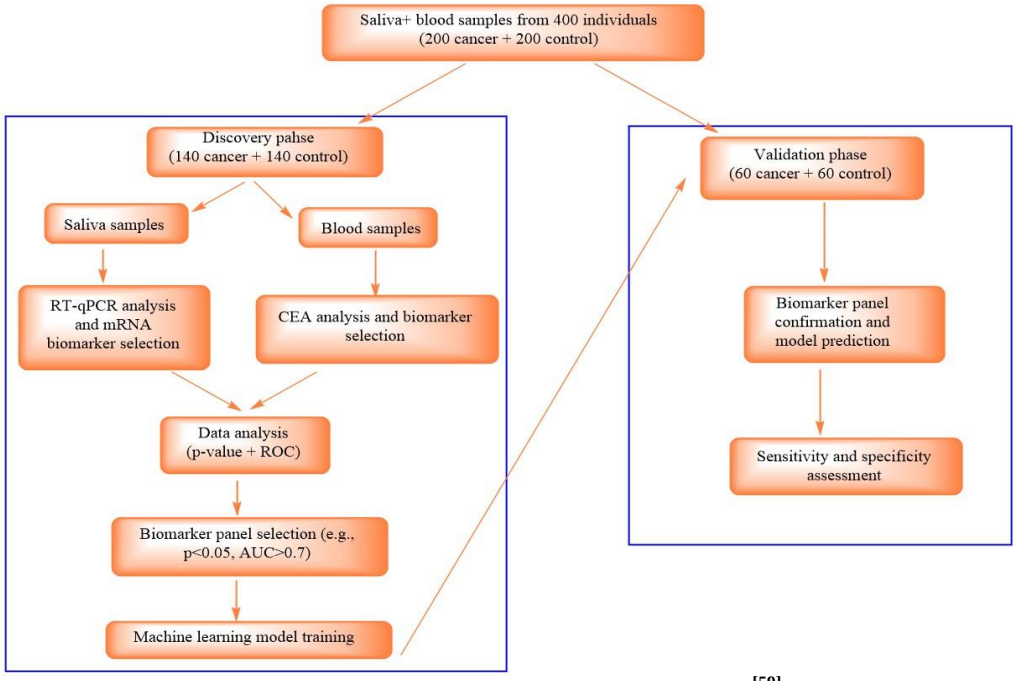


Fig. 12: Flowchart to identify ovarian cancer.[59]

3.5 Measurement of serum CA-125 and HE4 levels

The HE4 and CA125 electrochemiluminescent immunoassays were utilized to measure the serum levels of HE4 and CA125, respectively. The required controls were within the manufacturers' ranges, and all assays were conducted and collected according to the manufacturer's instructions.[48]

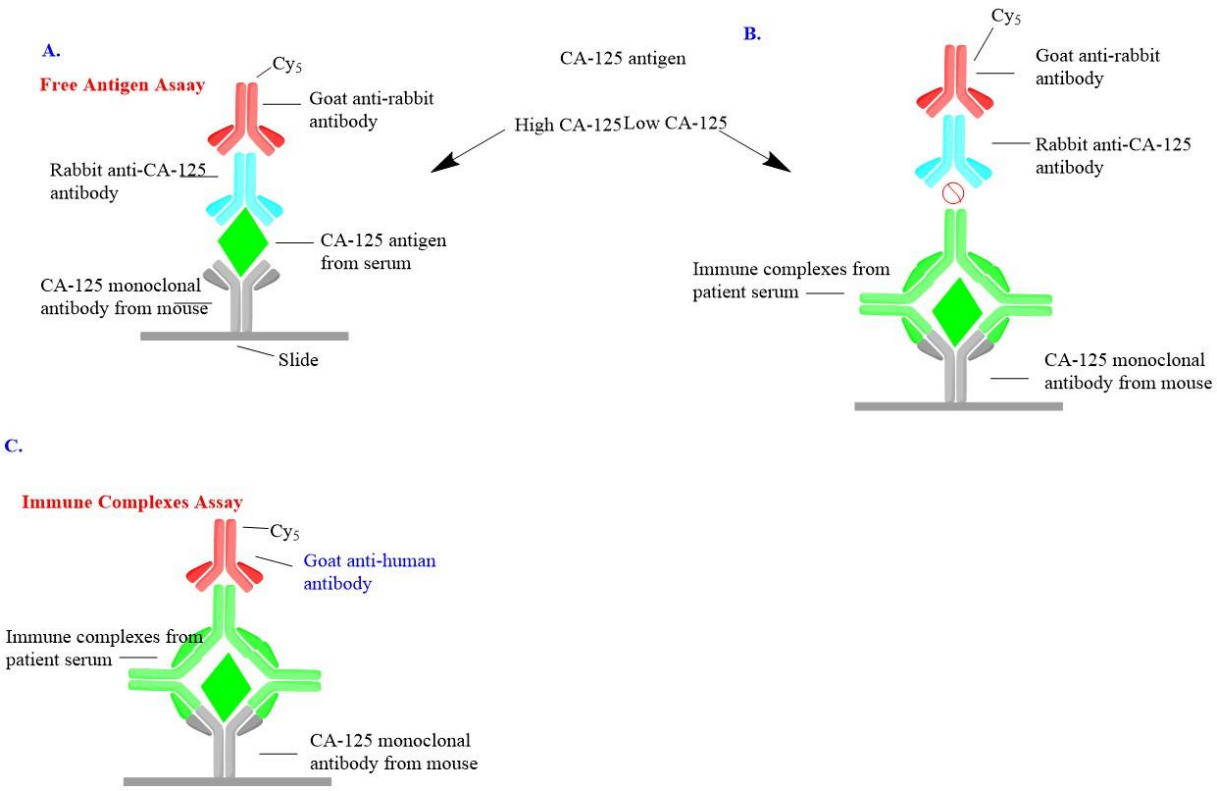


Fig. 13: Evaluation procedure for CA125 in immune complexes and CA125 free antigen based on antigen captureprinciple.[64,65]

A nanoparticle slide is coated with mouse mAbs against CA125, and patient serum is then added. Goat anti-rabbit antibody with Cy5 tag is added after washing, then rabbit anti-CA125 antibody.(Fig. (13))

(A) Rabbit anti-CA125 antibodies may be blocked when CA125 binds to human anti-CA125 in an immunological complex.

(B) Goat antihuman IgG Abswhich have been Cy5-tagged are added to CA125 immune complexes right away the sera are incubated.

(C) When comparing an invasive serous instance with a CA125 concentration of 3331 kilounits/L to another with a CA125 concentration of 26.7 kilounits/L, the insets of panels A–C show the array-based antigen and immune complex reactions. For the high-CA125 case, the free-antigen assay is greatly positive; for the low-CA125 case, it is just marginally positive. With the CA125 CIC test, the opposite is true. Green areas serve as orientation marks and are BSA controls. A mouse mAb to GSTA1 was then introduced into the array as a negative control, enabling the determination of a "normalized" intensity for the CA125 CIC.

4. CA-125 (Carbohydrate Antigen 125)

CA-125, sometimes named as Cancer Antigen-125 or Tumor Antigen 125, is a glycoprotein produced by the mucin 16 (MUC16) genes(Fig. (15)) and may be recognized using OC-125 monoclonal antibodies in cancerous ovarian tissues. It has been applied in the early 1980's when Bast et al. especially separated the monoclonal antibody OC-125 in cancerous ovarian tissue compared to healthy ovarian tissue. The upper limit for CA-125 is 35.0 U/mL in each premenopausal and postmenopausal patient.[66,67]CA125 is used clinically to monitor cancer progression and is a prime candidate for a screening biomarker.[68]

The FDA guidelines advise CA-125 as a valuable protein biomarker for evaluating treatment action and monitoring ovarian cancer patients. CA-125 levels correlate with medical stage and survival outcomes, presenting insights for scientific choice-making. However, CA-125 alone does not accurately replicate tumor burden owning to potential secretion by non-tumor cells in an inflammatory surrounding.[69]

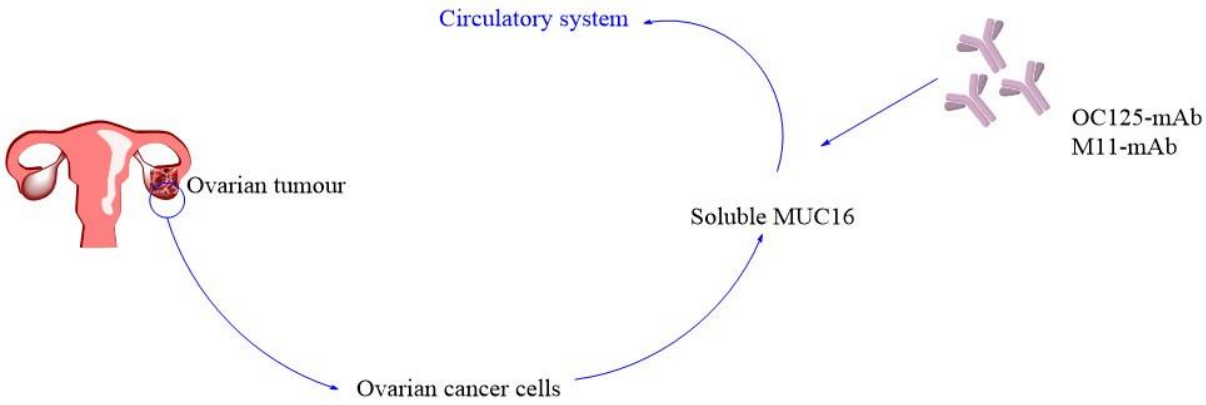


Fig. 14: MUC16(CA-125) structure and its role in ovarian cancer.[66]

MUC16 has various O- and N-glycosylation sites in its cytoplasmic, transmembrane, and extracellular components. MUC16's peptide component has about 22,152 amino acids. MUC16's 12,000 amino acid N-terminal domain was the only region where O-glycosylation is present. The tandem repeat region, which consists of more than 60 repeats of 156 amino acids, makes up a sizable amount of the peptide component. About 56 sea-urchin, enterokinase, and agrin (SEA) domains are present in MUC16. Mucins all share the SEA domain, which is essential for the cleavage and binding of MUC16 subunits. A cytoplasmic tail consisting of 32 amino acids and perhaps phosphorylation sites come after the transmembrane domain.About 12 amino acids from the cytomembrane are required for MUC16 to be cleaved from an extracellular location.(Fig. (14)& (15)).[66,70]

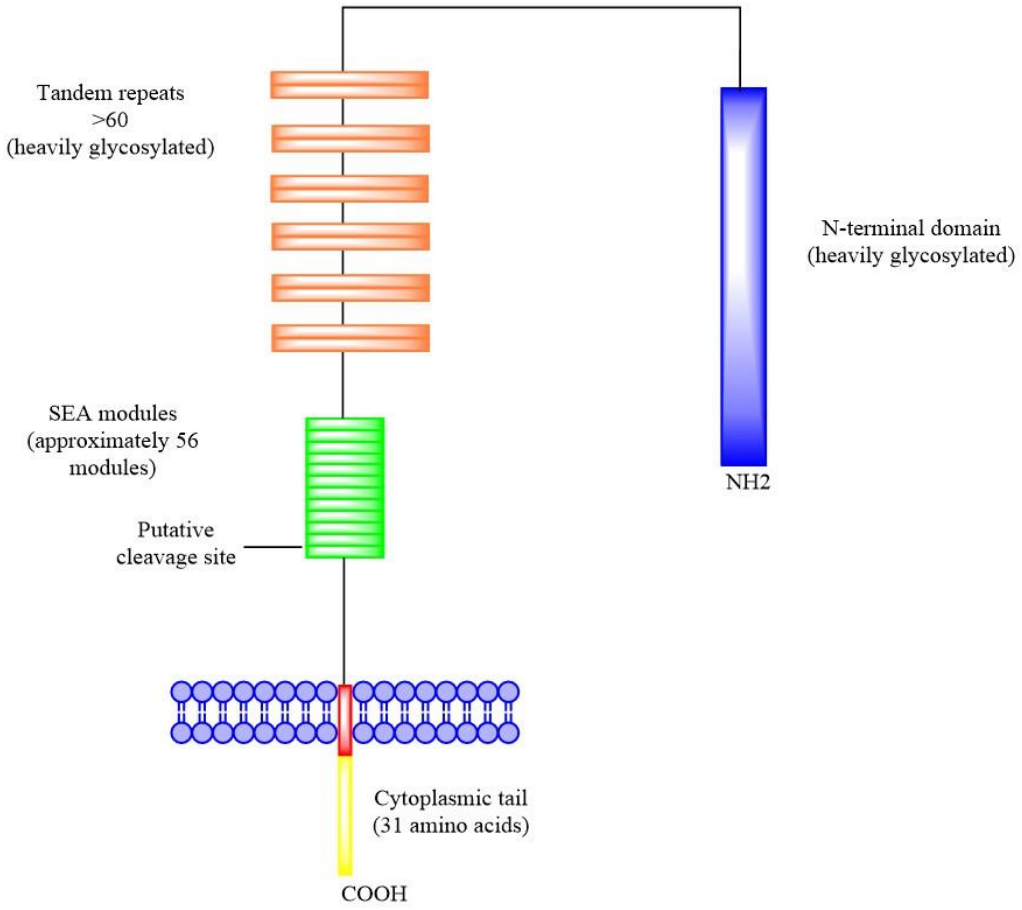


Fig. 15: Schematic structure of MUC16 mucin.[71,72]

Ovarian cancer cells or healthy coelomic epithelial cells produce CA-125. Although used to predict pelvic mass, using CA-125 alone has significant drawbacks. Initially, low sensitivity for stage I illness. Alternatively, it lack of specificity, especially in women with pelvic mass who are not yet menopausal.[73,74]

Approximately 20% of EOC patients do not exhibit elevated CA-125 levels, while lower CA-125 situations are connected with previous stages and enhanced outcomes. Circulating immune complexes (CICs) by binding antibodies may contribute to lower CA-125 concentrations and inhibiting accurate discovery.[24]

The PLCO trial proved that combining CA-125 screening with ultrasound didn't significantly improve early discovery or mortality issues compared to routine care. Also, false-positive results led to severe post-operative complications in 15% of cases. Also, the UKCTOCS trial set no significant mortality benefit in the CA-125 screening group assimilated to the control group.[24,75]

Post-surgery, an elevated CA-125 level (>35 U/mL) indicates residual ailment, reduced chemotherapy sensitivity, and better tumor malignancy. The Gynecologic Cancer Intergroup (GCIG) proposes standards for assessing tumor remission and recurrence based on CA-125 level. A minimal 50% decrease sustained for four weeks classifies patients as responders, while complete responders have CA-125 levels in the regular range (<35 U/mL). Ovarian cancer development or recurrence is indicated using CA-125 level doubling with a one-week interval. Appreciably, chronic CA-125 levels below 35 U/mL do not rule out residual ailment and recurrence.[24,76]

The sensibility of CA-125 in identifying ovarian cancer is limited. About 50 percentage of individuals experiencing early-stage OC have increased levels of CA 125; this indicates that CA 125 has a particularly low sensitivity for OC that has not yet manifested symptoms. Additionally, 90% of individuals with second stage cancer and over 90% of patients in stages III and IV disease had increased CA 125. Tumor size and stage are correlated with the concentration of CA 125. When ovarian cancer is detected with CA 125, particularly in its early stages, it commonly results in false negative results that have significant clinical implications. It follows that individual who receive false negative results may not receive the necessary care or the right medication for their condition. The measurement of CA 125 may therefore be helpful in assessing the state of the disease in individuals with advanced endometriosis, but it is not helpful in OC screening in the population that don't exhibit any symptoms.[76]

Post-initial cycle measurement and subsequent normalization of CA-125 beneath 35 U/mL by the 3rd cycle are crucial for analysis. Decreased CA-125 levels and quicker normalization suggest a good chemotherapy reaction and extended progression-free survival. Throughout first-line chemotherapy, routine CA-125 monitoring aids in identifying patients with decreased drug sensitivity, allowing for prompt treatment modifications. CA-125 predicts disease progression following chemotherapy but has no impact on survival afterward. Insulin signaling-induced CA-125 oversecretion suggests capability in predicting chemo-resistance.[77-79] The most important predictor factors for predicting OS and PFS in patients with metastaticOC were absolute CA125 levels evaluated after the first chemotherapy cycleand restoration of CA125 levels to normal until the third cycle of treatment.[70]Reliability in predicting the progression of the disease during first-line treatment monitoring was provided by CA 125.[79]

5. HE4 (Human epididymis protein 4)

HE-4 is a glycoprotein produced by the WFDC2 (WAP four-disulfide core domain protein 2)gene(Fig. (16)) and acts as a serine proteinase inhibitor. It serves as a putative biomarker for OC and detected in blood and urine of cases using enzyme immunoassay.[24,80] It was detected by Kirchhoff et al. in 1991 in the epididymal duct. (Table 4.)[74,81]

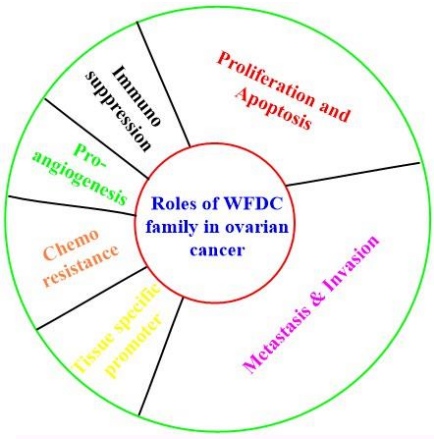


Fig. 16: Roles of WFDC family in overian cancer.[82]

HE-4 exhibits over-expression in specific OC subtypes, with a 100% occurrence in endometroid tumors and 93% in serious OC. This characteristic enables its usefullness in distinguishing between various tumor types, supporting in process differential diagnosis.[24]

Women with OC had a high concentration of HE4, while lung adenocarcinoma showed a moderate level of HE4. Additionally, gastric, pancreatic, breast and transitional cell carcinomas had the lowest levels of HE4.[74]

Women who were pregnantshow reduced levels of HE4 than non-pregnant women. In addition, the HE4 concentration were considerably higher among older women, smokers, and women who menarched later than the control group. Moreover, the use of E and P contraceptives, endometriosis, and the menstruation did not alter the HE4 serum level.[83]

HE4 protein value explosively depends on the case's age and smoking. The serum attention of HE4 marker increases with the duration of gestation. Understanding the normal range of HE4 protein enables the exact interpretation of marker measures. This may affect in an earlier and further effective diagnosis of OC.[84] It is

regarded as among the most promising biomarkers in oncology and gynecology.[85,86]

5.1 Functions of HE-4

HE4's exact functions are unknown as of yet, latest studiespoint to its involvement in the immune system's proinflammatory response and resistance against proteolytic enzymes.Crucially, overexpression of the HE4 gene has been shown to advance ovarian cancer. (fig. (17) and fig.(18)) Furthermore, through a No. of oncogenic signalling channels, such as ERK/mitogen-activated protein kinase (MAPK), hypoxia-inducible factor 1 alpha (HIF1α), and matrix metalloproteinases, HE4 drives EOC progression, cancer cell motility and adhesion, invasion, and metastasis. (Table 4.) Additionally, HE4 is involved in estrogen signalling.[80]

Table 4: Pathways and functions that connect OC oncogenesis to HE4.[87]

Pathway	HE4 Influence
ERK/MAPK (extracellular signal-regulated kinases/ mitogen-activated protein kinase)	• Regulation of proliferation and invasionof SOC cells • ERK activation with HE4 overexpression • Decrease in proliferation when HE4 wassilenced in SKOV3 cells. • ERK/MAPK pathway activation following to HE4-EGFR/EGGF interaction
PI3K/AKT (phosphoinositide 3- kinases/ Protein kinase B)	• AKT increase promoting cell growth in OVCAR3 cells when HE4 is overexpressed. • When HE4 is knocked down, AKT decreases and OVCAR3 cells grow less rapidly.
HDAC3 (histone deacetylase 3)	• HDAC3 expression or knockdown lead to a corresponding increase or decrease in HE4 expression • HE4 and HDAC3 binding activates the PI3K/AKT signaling pathway • Potential therapeutic benefits might result from inhibiting the interaction between HDAC3 and HE4.
HIF1α (hypoxia-inducible factor 1-alpha)	• HE4-HIF1α interaction is yet not well understood • Decrease in HE4 levels in SKOV3 cells treated with HIF1α sirna or with HIF1α inhibitors
JAK/STAT (Janus kinases/signal transducer and activator of transcription proteins)	HE4 knockdown inhibits the action of the JAK/STAT3 pathway in-vivo and in-vitro HE4 knockdown inhibits ovarian cancer's malignant development and proliferation of cells.

Fig. 17: HE-4 as a disease checkpoint.[87]

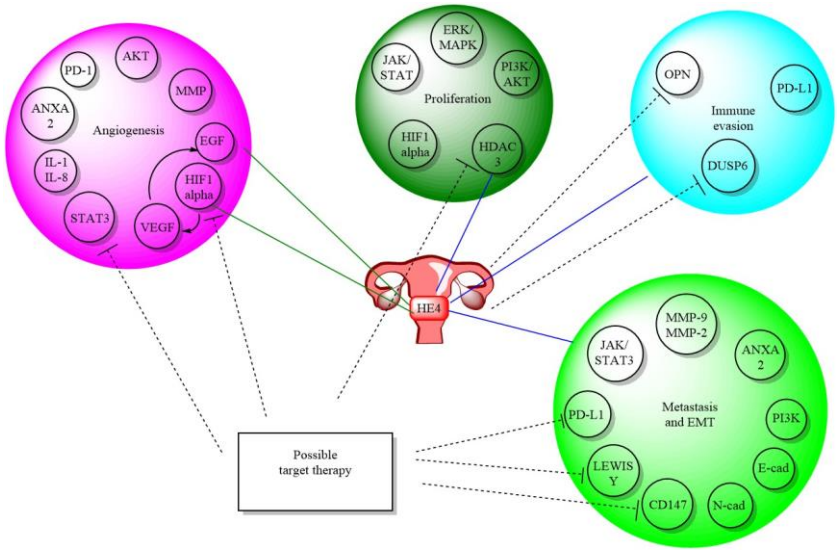


Fig. 18: HE4 interaction network in OC oncogenesis: Schematic representation.[87]

5.2 Diagnostic value of HE-4

In 2018, a study examined the predictive value of HE-4 marker readings in patients receiving first-line treatment for ovarian cancer. It was discovered that HE4 levels predicted platinum sensitivity and were linked to overall survival (OS), progression-free survival (PFS), and surgical outcome. HE4 showed promise as a relevant biomarker for OC treatment response evaluation and outcome prediction. (Table 5 and Fig. (18)).[88]

Table 5: Median range of serum HE-4 and CA-125.[88]

Prognostic factor	HE-4 [pmol/1]	CA-125 [U/ml]
Age		
Premenopausal, n = 12	172 [35.8–1116.3]	114.2 [33–4638]
Postmenopausal, n = 78	311 [41.1–3608]	323.5 [11.3–14199]
FIGO stage		
I and II, n = 17	120.7 [41.1–345]	74.5 [11.3–1441]
III and IV, n = 73	543 [35.8–3608]	535.1 [15.9–14199]
Tumor grade		
1 and 2, n = 41	226 [35.8–1500]	198 [11.3–1639]
3 n = 49	521 [41.1–2556]	521.8 [15.9–14199]
Surgery		
Optimal, n = 65	226 [35.8–3608]	198.5 [11.3–14199]
Suboptimal, n = 25	543 [53.9–2556]	543 [20.4–10000]
2-year survival		
Yes, n = 65	239 [35.8–1500]	227.6 [11.3–10000]
No, n = 25	385.2 [53.9–3608]	536 [20.4–14199]

In order to determine how well the preoperative plasma tumor markers HE-4 and CA-125 predicted cancer death in women experiencing EOC, a new study was carried out at the University Hospital of Quebec City. Significant relationships between HE-4 levels and key prognostic variables were seen during both training and validation cohorts. In the training cohort, HE-4 outperformed CA-125 in terms of predicting mortality, and in the validation cohort, a significant correlation was seen.Nevertheless, the connection lost significance when preoperative predictive factors were taken into account. There was a stronger link founds between HE-4 and death in females diagnosed with serous ovarian cancer. In particular, in situations of serous ovarian cancer, HE-4 and other prognostic variables might provide helpful details for predicting death in EOC.[60]

Higher levels of HE-4 at diagnosis, during cytoreductive surgery, and during first-line chemotherapy were linked to an increased risk of recurrence, according to a single-center scrutiny involving 188 individuals with ovarian cancer. Patients who experienced platinum resistance and those who possesses larger residual tumors following first surgery both showed elevated HE-4 levels. Furthermore, when the second recurrence diagnosis, patients with neoplastic leftovers larger than 10 mm had noticeably higher levels of HE-4.[89]

The predictive power of HE4, CA-125, the ROMA, and the RMI for OC in patients having pelvic masses was examined in a different diagnostic investigation. The models with the highest overall accuracy were HE4, CA-125, RMI, and ROMA. When compared to CA-125, HE4 and ROMA demonstrated superior detection of benign tumors. HE4 and ROMA demonstrated greater specificity and negative predictive value in premenopausal women, while HE4 showed the highest specificity in postmenopausal women.[90]

Preoperative blood HE4 levels above 500 pM were substantially related with a decreased 5-year overall survival rate (27% vs. 59%), according to a retrospective analysis of 89 EOC patients. These outcomes show the potential of HE-4 as a predictor for forecasting treatment response, survival rates, and OC recurrence.[91]

In ovarian cancer, the diagnostic utility of serum concentration ofHE-4, carbohydrate antigen-125 (CA-125), the ROC curve, and the ROMA index were investigated. The HE-4 serum level and the ROMA index are significant markers in discovering ovarian cancer, according to the study of 158 individuals. Nevertheless, the ROMA index is very helpful in enhancing the diagnostic efficacy of ovarian cancer in addition to HE-4 and CA-125 detection.[92]

In 2019, an investigation was carried out tostudy the function of HE4 in prognosticating and monitoring ovarian cancer recurrence by predicting 149 patients' OS and PFS. Of the patients, 68.5% (n = 102) showed recurrence. The recurrence rate was detected with 85.3% sensitivity and 91.5% specificity using serum HE4. When compared to CA125, HE4 levels are more specific but have a similar sensitivity for detecting recurrent ovarian cancer.[93]

6. Dual Biomarkers

The effectiveness of using multiple biomarkers to obtain high specificity and sensitivity has been demonstrated by numerous researches.

Important roles are played by CA125, HE4, and CA125 in combination with HE4 in the diagnosis of OC, especially EOC. Compared to HE4 in combination with CA125, HE4 is more sensitive, making it useful for diagnosis. On the other hand, in some particular histological forms of cancer, combined HE4 and CA125 identification of late-stage ovarian cancers may have substantial clinical diagnostic importance.[94]

Analysing EOC individual's serum HE4 concentration revealed performance indicators that were similar to CA125. (Table 7) Compared to utilizing either marker alone, the combination of HE4 and CA125 increased negative predictive value, sensitivity, and accuracy. According to the study's findings, HE4 and CA125 are comparable for tracking EOC patients, and using both markers together offer better tracking potential.[95]

Table 6: Performance of CA125, HE4 and HE4+CA125.[95]

	CA-125	HE-4	CA-125 + HE-4
Sensitivity	58.6% (95%CI: 48.8–67.8%)	54.5% (95%CI: 44.8–63.9%)	70.5% (95%CI: 61.2–78.8%)
Specificity	92.5% (95%CI: 87.3–96.1%)	95.0% (95%CI: 90.4–97.8%)	88.8% (95%CI: 82.8–93.2%)
PPV	84.4% (95%CI: 74.4–91.7%)	88.4% (95%CI: 78.4–94.9%)	81.4% (95%CI: 72.3–88.6%)
NPV	76.3% (95%CI: 69.7–82.1%)	74.9% (95%CI: 68.3–80.7%)	81.1% (95%CI: 74.5–86.6%)
Accuracy	78.6% (95%CI: 73.2–83.3%)	78.3% (95%CI: 72.9–83.1%)	81.3% (95%CI: 76.1–85.7%)

The serum levels of CA125 and HE-4 were measured in 30 EOC patients and healthy women in a study comparing the two tests for the identification of benign gynecological disorders and epithelial ovarian cancer (EOC). When compared to the healthy controls, the serum concentrations of CA-125 and HE-4 were considerably greater in ovarian cancer patients. When HE4 and CA125 were combined, the sensitivity and PPV increased to 96.7% and 97%, respectively. HE4 also exhibited greater specificity (95% vs. 85%), NPV (92.7% vs. 87.2%), PPV (93.1% vs. 80.7%), and sensitivity (90% vs. 83.3%).[96]

In a multicentred prospective analysis, 531 pelvic mass patients planned for surgery were included. Serum levels of HE4 and CA125 were assessed before to surgery to categorize individualswith varying levels of risk for EOC. Patients with benign tumors, EOC, tumors with LMP, non-EOC tumors, and non-ovarian malignancies were incorporated into the research. The postmenopausal and premenopausal groups both showed good specificity and sensitivity for the model. It was successful in classifying patients into high- and low-risk groups, properly identifying a sizable percentage of EOC cases as high-risk.[97]

When compared to healthy controls, HE4 concentrationhaveconsiderably higher inovarian and endometrial cancer patients, serous carcinomas showing the highest values. When distinguishingindividuals who haveOC from healthy controls and those with ovarian endometriosis, the hybrid of HE4 and CA125 yielded the best results in terms of accuracy and sensitivity.[98]

Elevated levels were found in baseline samples of patients with advanced high-grade serious EOC, a prospective study assessing CA125 and HE4 measurements in the blood and ascites found that these indicatorswere not able to distinguish between patients with complete and incomplete resection or residual disease. Tumor markers decreased after surgery, most likely as a result of the lengthy half-life of CA125 and the reduction in ascites volume. However, previous studies have shown that evaluations of CA125 and HE4 both prior to and during the start of chemotherapy can forecast treatment outcome and survival.[99]

Blinded tests were performed in 2003 on sera from 65 healthy asymptomatic controls, 19 people with benign ovarian illness, and 37 ovarian cancer patients (7 early stage and 30 late stage). Serous ovarian carcinoma was the most prevalent histology seen in the patients with ovarian cancer (21 cases), and stage III was the most common stage (24 instances). When applying a 95% specificity criterion for positivity, HE4 was only able to detect 7 cases of ovarian cancer, while CA125 was only able to identify 8 cases. These results demonstrate the limitations of both markers as ovarian cancer predictors.[100]

In a prospective analysis, HE-4 outperformed CA-125 in terms of specificity for benign illness, and the combination of the two markers yielded the best sensitivity for differentiating between benign ovarian neoplasms and invasive epithelial ovarian malignancies.[36]

Using the ECLIA immunological approach, Chen et al.obtained a specificity of 65.7% with a cut-off value for HE4 of 140 pmol/L. In an alternative investigation employing an alternative method to evaluate serum HE4, the correlation between CA125 and HE4 had a significantly higher specificity (80%). The correlation between HE4 and CA125 is a helpful diagnostic method for ovarian cancer.[101,102]

In cases of suspected benign ovarian tumors, to assess both markers: an increasing value of the two markers is predictive of ovarian cancer. It appears better to use limits of 70 and 140 pmol/l based on menopausal status and 35 UI/ml for CA125, as proposed in a recent study by Goff et al.Additionally, as demonstrated by Urban et al., the use of this combination HE4 and CA125 assay may also be very beneficial for ovarian cancer screening in the general population.[103,104]

7. Usefulness of tumor marker during and after treatment in OCpatients

Research done in 2023 on Japanese patients treated at a hospital between 2014 and 2021with an OC diagnosis. The clinical utility of the tumor marker HE4 in the follow-up of OC patients was assessed in this study. They examined 48 individuals, comprising 31 postmenopausal and 17 premenopausal patients, 27 with recurrence and 21 without, with a median age of 57 years (range 42-80) and a median observation time of 20.8 months.(Table 8) In brief, variations in the values of the tumor marker HE4 during follow-up, either during or after OC treatment, were in accordance with the clinical assessment of the disease state in Japanese patients. Therefore, HE4 may be used in addition to CA125 for follow-up evaluation both during and after OC treatment.[105]

Table 7: Sensitivity, specificity, PPV and NPV in follow-up period.[105]

Criteria	N	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)
Post-treatment follow-up					
HE4 > 2-fold elevation	96	39.4	93.7	76.5	74.7
HE4 >70 pmol/L	140	77.8	75.8	60.3	87.8
HE4 >140 pmol/L	140	46.7	92.6	75.0	78.6
CA125 >35 U/mL	140	86.7	82.1	69.6	92.9
Follow-up during drug treatment					
HE4 >70 pmol/L	163	83.3	49.0	16.9	95.9
HE4 >140 pmol/L	163	77.8	75.2	28.0	96.5
HE4 >25% elevation	163	44.4	96.6	61.5	93.3
HE4 >14% elevation	163	50.0	92.4	45.0	93.7
CA125 >35 U/mL	163	100.0	51.7	20.5	100.0
Recurrence (patients)					
HE4 >70 pmol/L	48	77.8	85.7	87.5	75.0
HE4 >140 pmol/L	48	44.4	100.0	100.0	58.3
CA125 >35 U/mL	48	85.2	90.5	92.0	82.6
HE4 >70 pmol/L or CA125 >35 U/mL	48	92.6	76.2	83.3	88.9

A prospective study that investigated at the predictive significance of serum HE4 in advanced peritoneal, fallopian tube, and ovarian cancerpatients in 2020. Based on the results, advanced patients had noticeably greater serum concentration of HE4 and CA125 than controls with benign diseases. While HE4 was less sensitive than CA125, it was more specific. Additionally, there was a strong correlation between blood HE4 and both treatment response and recurrence; HE4 demonstrated a higher effective response rate to therapy than CA125 did.[106]

Plotti et al. in 2019 conducted a retrospecific study aimed to evaluate the importance of HE4 in OC patients with negative CA125 at diagnosis for recurrence detection. The study comprised eight patients in total. 53 was the average age (with a range of 40 to 75). Upon diagnosis, they all had normal CA125 values, however seven (87.5%) had abnormal HE4 levels. seven patients are recurred. HE4 levels in patients initially diagnosed with normal CA125 may be employed as a recurrence marker. Subsequent research is required to assess the importance of HE4 levels in the early identification of recurrent ovarian cancer.[107]

According to a retrospecific study conducted in 2010 by Anastasi et al., HE4 was discovered to have a sensitivity of 96.9%, whereas CA125 had a sensibility of 85.7%. HE4 was investigated as a novel prospective early biomarker for the OC recurrence. Furthermore, for 5-8 months, a greater HE4 might detect an ovarian cancer recurrence more quickly than CA125.[46]

Each patient provided three serum samples, each obtained three months apart, for the Manganaro et al. study: time interval I (1–3 months after surgery), time interval II (4–6 months after surgery), and time interval III (7–10 months after surgery). A rise in HE4 was observed in 22, 78, and 89% of patients with EOC recurrence within time periods I, II, and III, respectively. Only 44% of patients had positive levels for CA125, and these were observed later in the disease (at time interval III). As a result, the authors came to the conclusion that elevated HE4 levels may occur around three months before an increase in CA125 in the event of a disease recurrence.[108]

In the Angioli et al. study, CA125 and HE4 were assessed at three distinct intervals: prior to the first

chemotherapy cycle, at the third and sixth cycles, and every three months following the sixth cycle until the sixth month of follow-up. While the CA125 readings did not appear during chemotherapy to be statistically significant in terms of predicting the platinum action, they find that the HE4 profile was strongly related with the response to platinum-based chemotherapy. They deduced that the time required for HE4 normalization during initial chemotherapy may allow the identification of non-responders after the third cycle.[109]

With a higher sensitivity in the early stages, specificity, and efficiency than CA 125, HE4 is the preferred tumor marker in ovarian cancer. The primary way to enhance this method would be to apply it to patients whose results are CA 125 positive but HE4 negative. Utilizing this combination results in an increased sensitivity of 90.1% (95% in non-mucinous tumors) and specificity of 82.1% for tumor marker use in the identification of pelvic masses.[110]

A validated supplementary biomarker for HGSC to cancer antigen 125 (CA125) is human epididymis protein 4 (HE4). For the purpose of prognostic stratification and therapy monitoring in patients with HGSC, HE4 is a viable biomarker. In particular, patients' survival was connected with their seerum level of HE4 at the time of their first relapse, could be a helpful supplementary measure for choosing second-line therapies.[111]

In a cohort of 30 EOC patients (260 samples), Havrilesky et al. carried out a prospective pilot study with the goal of determining the prognostic efficacy of a panel of three biomarkers, including HE4, MMP7, and Glycodelin, as compared to the conventional CA-125 alone. This biomarker panel achieved 100% sensitivity as compared to CA-125 (96%) alone, according to a longitudinal examination of 27 patients who experienced illness recurrence. It is noteworthy that section of the enrolled patients (n=14), minimum of three biomarkers, including HE4, was shown to be elevated prior to CA-125 positive and the identification of clinical recurrence by 6 to 69 weeks.[112]

Schummer et al. evaluated the utility of four biomarkers (HE4, CA-125, Mesothelin, and MMP7) to track the recurrence of EOC in a prospective trial with n=23 participants. HE4 outperformed CA-125 in identifying recurrences in patients both before and after they show this antigen.[113] After the first round of therapy, blood levels of HE4 and CA125 dropped, but they spiked another time resulting from a relapse.[114]

The highest risk of disease occurrence was shown to occur in people between the ages of 51 and 60, according to a study using HE4 to predict the recurrences of EOCinstances for a period of 22 months. The most prevalent illness that recurred was serous carcinoma. There were 47 instances are participated. A total of 23 cases, 48.9% of recurrent cases. HE-4 was able to predict recurring cases after treatment and had a higher sensibility than CA125. The efficiency of CA125 and HE4in predicting repeated instances, however, did not differ (p value=0.8314), however for a period of three to six months, HE4 might identify recurring episodes more quickly than CA125. Furthermore, a greater HE-4 could identify an OC recurrence more quickly than CA-125 for 5 to 8 months.[50,115]

The integrate of HE4 and CA125 may aid in identifying individuals who are at disease risk reoccurring, as perresearch by Nassir et al. They also came to the conclusion that HE4 seems to be a useful biomarker for anticipating recurrence following the end of second-line treatment. (Fig. (19) & (20))[116]

The HE4 gene is not exclusive to ovarian cancer. It could contribute to the diagnosis of certain illnesses, such as cancer. However, it is still unknown how its biology, genetics, and pathological condition are related. The review's most significant finding advises caution when utilizing HE4 to diagnose ovarian cancer and to keep in mind any additional conditions that may affect our assessment. In addition, given the growing field of HE4 application in various illnesses, we may wonder if it applicable to syndromes that are currently unidentified.[117,118]

CONCLUSION

Although tumor markers,CA-125 in particularare widely utilized to diagnose ovarian cancer, their sensitivity and specificity are limited, especially when the disease is still in its early stages. The FDA has already approved the biomarker HE-4 for monitoring the progression of the disease in EOC patients. However, HE-4 is still not as popular in cancer practice as CA-125, despite having superior follow-up accuracy.

A novel EOC biomarker called HE-4 has been used to identify ovarian cancer in its early stages and in cases of recurrence. The primary sources of HE-4 are the tumor environment and cancerous ovarian cells. Tumor elimination should be correlated with a drop in HE-4 after surgery, and HE-4 itself should serve as a biomarker for predicting surgical outcome. The tumor marker CA-125 has low sensitivity and poor specificity when used alone. A helpful biomarker for distinguishing between benign ovarian illness and ovarian cancer is serum HE-4. In follow-up evaluations, HE-4 and CA-125 indicators that were positive prior to treatment could be employed in addition to one another. Combining CA-125 with HE-4 improves risk stratification and diagnostic accuracy, increasing the usefulness of tumor markers in the diagnosis of EOC.

In ovarian cancer, HE-4 is the preferred tumor marker due to its superior sensitivity, specificity, and efficacy over CA-125. Cytoreductive surgery is the current standard treatment for EOC, and it is followed with systematic chemotherapy. Numerous studies assessed

HE-4's ability to predict surgical outcome in primary cytoreductive surgery. HE-4, a highly promising biomarker for surgical outcome, may be helpful in predicting primary therapy. When it comes to predicting the surgical result of EOC cytoreductive surgery, HE-4 outperforms CA-125. It is anticipated that this biomarker's potential for clinical use will increase. Therefore, further prospective research is needed to strengthen these findings.

LIST OF ABBREVIATIONS

EOC- Epithelial ovarian cancer
OC- Ovarian cancer
CA-125- Cancer antigen 125
HE-4- Human epididymis protein 4
PBCR- Population-Based Cancer Registries
TME- Tumor microenvironment
PPV-Positive predictive value
NPV- Negative predictive value
NACB-National Academy of Clinical Biochemistry Laboratory Medicine
ROMA- Risk of Ovarian Malignancy Algorithm
mAb- Monoclonal antibody
CIC- Circulating immune complexes
GCIG- Gynecologic Cancer Intergroup
PFS- Progression-free survival
WFDC2- WAP four-disulfide core domain protein 2
E and P contraceptives- Estrogen and progesterone contraceptives
OS- Overall survival.

CONFLICTS OF INTEREST: The authors have no conflicts of interest, financial or otherwise.

REFERENCES

1. Vijay Gyanani, Jeffrey C. Haley, Roshan Goswami. Challenges of Current Anticancer Treatment Approaches with Focus on Liposomal Drug Delivery Systems. Pharmaceuticals, 2021 Aug 24; 14(9): 835.
2. P Rani, D Pal, R R Hegde and SRH. Acetamides: Chemotherapeutic agents for inflammation associated cancers. J Chemother, 2016; 28(4): 255–65.
3. P Rani, D Pal RRH and SRH. Anticancer, Anti-inflammatory, Analgesic activities of Synthesized 2-(Substituted phenoxy) acetamide derivatives. Biomed Int., 2014; 1–9.
4. Bhupendra Kaushik DP and SS. Gamma Secretase Inhibitor: Therapeutic Target via NOTCH Signaling in T cell Acute Lymphoblastic Leukemia. Curr Drug Targets, 2021; 22(15): 1789–98.
5. DilipkumarPal, In-ho Song, Shrikant Dashrath Warkad, Keum-soo Song, Gyu Seong Yeom, Supriyo Saha, Pramod B. Shinde SBN. Indazole-Based Microtubule-Targeting Agents as Potential Candidates for Anticancer Drugs Discovery. Bioorg Chem, 2022; 105735.
6. Dilipkumar Pal, Pooja Sahu, Goutam Sethi CEW and AB. Gossypol and its Natural Derivatives: Multi-targetted phytochemicals as potential drug candidates for oncologic diseases. Pharmaceutics, 2022; 14: 2624–49.
7. Dilipkumar Pal, Khushboo Raj, Shyam Sundar Nandi, Surajit Sinha, Abhishek Mishra, Arijit Mondal, Ricardo Lagoa JTB and AB. Natural Compounds as Novel Histone Deacetylase Inhibitors for the Treatment of Hematological Malignancies. Cancers (Basel), 2023; 15(2808).
8. Behzad Mansoori, Ali Mohammadi, Sadaf Davudian, Solmaz Shirjang, Behzad Baradaran. The Different Mechanisms of Cancer Drug Resistance: A Brief Review. Adv Pharm Bull., 2017 Sep 25; 7(3): 339–48.
9. Crosby D, Bhatia S, Brindle KM, Coussens LM, Dive C, Emberton M, et al. Early detection of cancer. Science (80-), 2022 Mar 18; 375(6586).
10. Krishnan Sathishkumar, Meesha Chaturvedi, Priyanka Das, S Stephen, Prashant Mathur. Cancer incidence estimates for 2022 & projection for 2025: Result from National Cancer Registry Programme, India. Indian J Med Res., 2022; 156: 598–607.
11. Mayank Singh, Ravi Prakash Jha, Neha Shri, Krittika Bhattacharyya, Priyanka Patel, Deepak Dhamnetiya. Secular trends in incidence and mortality of cervical cancer in India and its states, 1990-2019: data from the Global Burden of Disease 2019 Study. BMC Cancer., 2022; 22(149): 1–12.
12. Bernard W. Stewart Paul Kleihues. World Health Organization International Agency for Research on Cancer. Bernard W. Stewart Paul Kleihues, editor. WHO chronicle. IARC Press Lyon, 2003; 2003: 215–222.
13. Rong Y, Li L. Early clearance of serum HE4 and CA125 in predicting platinum sensitivity and prognosis in epithelial ovarian cancer. J Ovarian Res., 2021; 14(2): 1–9.
14. Christine H. Holschneider, Jonathan S. Berek. Ovarian Cancer: Epidemiology, Biology, and. Semin Surg Oncol., 2000; 19(3): 3–10.
15. Phool Chandra, Neetu Sachhan DP. Glycogen Synthase Kinase-3 (GSK-3) Inhibitors as new lead to Treat Breast and Ovarian Cancer Current Drug Target. Bentham Sci., 2021; 22(13): 1548–54.
16. Samuel C. Mok, Joseph Kwong, William R. Welch, Goli Samimi, Laurent Ozbun, Tomas Bonome, et al. Etiology and pathogenesis of epithelial ovarian cancer. Dis Markers., 2007; 23(5–6): 367–76.
17. Charles N. Landen, Michael J. Birrer, Anil K. Sood. Early Events in the Pathogenesis of Epithelial Ovarian Cancer. J Clin Oncol., 2008; 26(6): 995–1005.
18. Fathalla M.F. Incessant ovulation- A factor in ovarian neoplasia? Lancet., 1971 Jul; 298(7716): 163.
19. Guillermo Tortolero-Luna, Michele Follen Mitchell. The epidemiology of ovarian cancer. J Cell Biochem, 1995; 59(S23): 200–7.

20. B.R.Rao, Berend J. Slotman. Endocrine Factors in Common Epithelial Ovarian Cancer*. Endocr Rev., 1991 Feb; 12(1): 14–26.
21. H. A. Risch. Hormonal Etiology of Epithelial Ovarian Cancer, with a Hypothesis Concerning the Role of Androgens and Progesterone, J Natl Cancer Inst., 1998; 90(23): 1774–86.
22. Masaki Mandai, Ikuo Konishi, Hideki Kuroda, Shingo Fujii. LH/hCG action and 42 developmentsof ovarian cancer—A short review on biological and clinical/epidemiological aspects. Mol Cell Endocrinol., 2007; 269(1–2): 61–4.
23. Nigel Bonello, Kylie McKie, Melinda Jasper, Lucy Andrew, Nicki Ross, Emily Braybon, et al. Inhibition of Nitric Oxide: Effects on Interleukin-lβ-Enhanced Ovulation Rate, Steroid Hormones, and Ovarian Leukocyte Distribution at Ovulation in the Rat1. Biol Reprod., 1996; 54(2): 436–45.
24. Alkis Matsas, Dimitrios Stefanoudakis, Theodore Troupis, Konstantinos Kontzoglou, Makarios Eleftheriades, Panagiotis Christopoulos, et al. Tumor Markers and Their Diagnostic Significance in Ovarian Cancer. Life., 2023; 13(1689): 1–18.
25. Robert J. Kurman, Ancel Blaustein. Blaustein's pathology of the female genital tract. 5th ed. Kurman RJ, editor. new york, 2002; 1391.
26. C. I. Amos, J. P. Struewing. Genetic epidemiology of epithelial ovarian cancer. Cancer, 1993; 71(S2): 566–72.
27. Yogita Lugani, Smita Asthana, Satyanarayana Labani. Ovarian carcinoma: An overview of current status. Adv Mod Oncol Res., 2016; 2(5): 261–70.
28. Masafumi Koshiyama, Matsumura N, Ikuo Konishi. Recent Concepts of Ovarian Carcinogenesis: Type I and Type II. Biomed Res Int., 2014; 1–11.
29. Robert J. Kurman, Ie-Ming Shih. Molecular pathogenesis and extraovarian origin of epithelial ovarian cancer—Shifting the paradigm. Hum Pathol., 2011; 42(7): 918–31.
30. Jurgen M.J. Piek, Peter Kenemans, René H.M. Verheijen. Intraperitoneal serous adenocarcinoma: A critical appraisal of three hypotheses on its cause. Am J Obstet Gynecol., 2004; 191(3): 718–32.
31. Jurgen M. J. Piek, Paul J. van Diest, Ronald P. Zweemer, Jan W. Jansen, Ria J. J. Poort-Keesom, Fred H. Menko, et al. Dysplastic changes in prophylactically removed Fallopian tubes of women predisposed to developing ovarian cancer. J Pathol., 2001; 195(4): 451–6.
32. Christopher G. Przybycin, Robert J. Kurman, Brigitte M. Ronnett, Ie-Ming Shih, Russell Vang. Are All Pelvic (Nonuterine) Serous Carcinomas of Tubal Origin? Am J Surg Pathol., 2010; 34(10): 1407–16.
33. Andrew M Hanby, Clare Walker. The WHO, and what of the breast and female genital organs: part IV in the WHO classification of tumours series. Vol. 6, Breast Cancer Research, 2004; 133–134.
34. Ahmedin Jemal, Rebecca Siegel, Elizabeth Ward, Jiaquan Xu. Cancer Statistics, 2010. CA Cancer J Clin., 2010; 60(5): 277–300.
35. Kristina Lindemann, G. Kristensen, M. R. Mirza, L. Davies, F. Hilpert, I. Romero, et al. Poor concordance between CA-125 and RECIST at the time of disease progression in patients with platinum-resistant ovarian cancer: Analysis of the AURELIA trial. Ann Oncol., 2016; 27(8): 1505–10.
36. Richard G. Moore, Amy K. Brown, M. Craig Miller, Steven Skates, W. Jeffrey Allard, Thorsten Verch, et al. The use of multiple novel tumor biomarkers for the detection of ovarian carcinoma in patients with a pelvic mass. Gynecol Oncol., 2008; 108(2): 402–8.
37. Yanfei Yang, Yang Yang, Jing Yang, Xia Zhao, Xiawei Wei. Tumor Microenvironment in Ovarian Cancer: Function and Therapeutic Strategy. Front Cell Dev Biol., 2020; 8(758): 1–30.
38. Douglas Hanahan, Robert A. Weinberg. Hallmarks of cancer: The next generation. Cell., 2011; 144(5): 646–74.
39. Rebecca L. Siegel, Kimberly D. Miller, Ahmedin Jemal. Cancer statistics, 2020. CA Cancer J Clin., 2020; 70(1): 7–30.
40. Rebecca L. Siegel, Kimberly D.Miller, Nikita Sandeep Wagle, Ahmedin Jemal. Cancer statistics, 2023. CA Cancer J Clin., 2023; 73(1): 17–48.
41. Ahmedin Jemal, Rebecca Siegel, Elizabeth Ward, Taylor Murray, Jiaquan Xu, Michael J. Thun. Cancer Statistics, 2007; 2007; 57(1): 43–66.
42. P Rani, D Pal RRH and SRH. Leuckart synthesis and pharmacological assessment of novel acetamide derivatives, Anticancer agent in Medicinal Chemistry. Anticancer agent Med Chem., 2016; 16(7): 898–906.
43. Jacques Ferlay, Hai Rim Shin, Freddie Bray, David Forman, Colin Mathers, Donald Maxwell Parkin. Estimates of worldwide burden of cancer in 2008: GLOBOCAN 2008. Int J Cancer., 2010; 127(12): 2893–917.
44. Robert C. Bast Jr., Molly Brewer, Changping Zou, Mary A. Hernandez, Mary Daley, Robert Ozols, et al. Prevention and early detection of ovarian cancer: mission impossible? Vol. 174, Recent results in cancer research, 2007; 91–100.
45. Jiwen Wang, Jia Gao, Hongwen Yao, Zongyong Wu, Minjie Wang, Jun Qi. Diagnostic accuracy of serum HE4, CA125 and ROMA in patients with ovarian cancer: A metaanalysis. Tumor Biol., 2014; 35(6): 6127–38.
46. Emanuela Anastasi, Giulia Giovanna Marchei, Valentina Viggiani, Giuseppina Gennarini, Luigi Frati, Maria Gabriella Reale. HE4: A new potential early biomarker for the recurrence of ovarian cancer. Tumor Biol., 2010; 31(2): 113–9.
47. Francesco Plotti, Stella Capriglione, Corrado Terranova, Roberto Montera, Alessia Aloisi, Patrizio Damiani, et al. Does HE4 have a role as biomarker in the recurrence of ovarian cancer? Tumour Biol., 2012; 33(6): 2117–23.

48. Ying Shen, Li Li. Serum HE4 superior to CA125 in predicting poorer surgical outcome of epithelial ovarian cancer. Tumor Biol., 2016; 37(11): 14765–72.
49. Shuai Zhen, Li-Hong Bian, Li-Li Chang, Xin Gao. Comparison of serum human epididymis protein 4 and carbohydrate antigen 125 as markers in ovarian cancer: A meta-analysis. Mol Clin Oncol., 2014; 2(4): 559–66.
50. Pedrada Innao, Methasinee Pothisuwan, Prasit Pengsa. Does human epididymis protein 4 (HE4) have a role in prediction of recurrent epithelial ovarian cancer. Asian Pacific J Cancer Prev, 2016; 17(9): 4483–6.
51. Lindsay Kuroki, Saketh R Guntupalli. Treatment of epithelial ovarian cancer. BMJ., 2020; 371: 1–20.
52. Lori J. Sokoll, Daniel W. Chan. Chapter 44 - Tumor markers. Fourth Edi. Vol. 125, Contemporary Practice in Clinical Chemistry. INC, 2020; 779–793.
53. M. Montagnana, E. Danese, S. Giudici, M. Franchi, G.C. Guidi, M. Plebani, et al. HE4 in ovarian cancer: From Discovery to clinical application. Adv Clin Chem., 2011; 55: 1– 20.
54. Felix Leung, Eleftherios P. Diamandis, Vathany Kulasingam. Ovarian cancer biomarkers: Current state and future implications from high-throughput technologies. 1st ed. Vol. 66, Advances in Clinical Chemistry. Elsevier Inc., 2014; 25–77.
55. Ece Gumusoglu, Tuba Gunel. The Role of Circulating Biomarkers in the Early Diagnosis of Ovarian Cancer. Ovarian Cancer - From Pathogenesis to Treatment, 2018; 157–174.
56. Michael J. Duffy. Tumor markers in clinical practice: A review focusing on common solid cancers. Med Princ Pract, 2012; 22(1): 1–8.
57. Nicola Normanno, Kathi Apostolidis, Francesco de Lorenzo, Philip A. Beer, Raymond Henderson, Richard Sullivan, et al. Cancer Biomarkers in the era of precision oncology: Addressing the needs of patients and health systems. Semin Cancer Biol., 2022; 1–9.
58. Kumari Binita Chandra, Savita Yadav. Biomarkers in ovarian cancer and saliva: An update. J Reprod Healthc Med., 2021; 2(1): 1–11.
59. Jinfang Yang, Cuiping Xiang, Jianmeng Liu. Clinical significance of combining salivary mRNAs and carcinoembryonic antigen for ovarian cancer detection. Scand J Clin Lab Invest., 2020; 81(1): 39–45.
60. Daniela Furrer, Jean Grégoire, Stéphane Turcotte, Marie Plante, Dimcho Bachvarov, Dominique Trudel, et al. Performance of preoperative plasma tumor markers HE4 and CA125 in predicting ovarian cancer mortality in women with epithelial ovarian cancer. Wong KK, editor. PLoS One., 2019; 14(6): 1–12.
61. J.A. Loo, W. Yan, P. Ramachandran, D.T. Wong. Comparative human salivary and plasma proteomes. J Dent Res., 2010; 89(10): 1016–23.
62. Kelly Grayson, Ebony Gregory, Ghazala Khan, Barbara-Ann Guinn. Urine Biomarkers for the Early Detection of Ovarian Cancer – Are We There Yet? Biomark Cancer., 2019; 11: 1–8.
63. Chloe E. Barr, Kelechi Njoku, Gemma L. Owens, Emma J. Crosbie. Urine CA125 and HE4 for the Detection of Ovarian Cancer in Symptomatic Women. Cancers (Basel), 2023; 15(1256): 1–13.
64. Jacobs IJ, Menon U, Ryan A, Gentry-Maharaj A, Burnell M, Kalsi JK, et al. Ovarian cancer screening and mortality in the UK Collaborative Trial of Ovarian Cancer Screening (UKCTOCS): A randomised controlled trial. Lancet., 2015; 387(10022): 945– 56.
65. Daniel W. Cramer, Dennis J. O'Rourke, Allison F. Vitonis, Ursula A. Matulonis, Daniel A. DiJohnson, Patrick M. Sluss, et al. CA125 Immune Complexes in Ovarian Cancer Patients with Low CA125 Concentrations. Clin Chem., 2010; 56(12): 1889–92.
66. Parsa Charkhchi, Cezary Cybulski, Jacek Gronwald, Fabian Oliver Wong, Steven A. Narod, Mohammad R. Akbari. CA125 and Ovarian Cancer: A Comprehensive Review. Cancers (Basel)., 2020; 12(12): 3730.
67. Xiuzhu Hou, Shanshan Liu, Jing Liu, Jiansuo Zhou, Yongming Liang, Liyan Cui. The performance of Carbohydrate Antigen 125-Thomsen-nouveau and anti-Müllerian hormone combined with CA125, Human epididymis protein 4 and Risk of Malignancy Algorithm in diagnosis for patients with Epithelial ovarian cancer. Clin Biochem., 2023; 119(110615): 1–9.
68. Babatunde O. Akinwunmi, Ana Babic, Allison F. Vitonis, Daniel W. Cramer, Linda Titus, Shelley S. Tworoger, et al. Chronic medical conditions and CA125 levels among women without ovarian cancer. Cancer Epidemiol Biomarkers Prev., 2018; 27(12): 1483–90.
69. Brian C. Cooper, Anil K. Sood, Charles S. Davis, Justine M. Ritchie, Joel I. Sorosky, Barrie Anderson, et al. Preoperative CA 125 levels: An independent prognostic factor for epithelial ovarian cancer. Obstet Gynecol., 2002; 100(1): 59–64.
70. Christine L. Hattrup, Sandra J. Gendler. Structure and function of the cell surface (tethered) mucins. Annu Rev Physiol., 2008; 70: 431–57.
71. Alain Piché. Pathobiological role of MUC16 mucin (CA125) in ovarian cancer: Much more than a tumor biomarker. World J Obstet Gynecol, 2016; 5(1): 39–49.
72. Timothy J. O'Brien, John B. Beard, Lowell J. Underwood, Richard A. Dennis, Alessandro D. Santin, Lyndal York. The CA 125 gene: An extracellular superstructure dominated by repeat sequences. Tumor Biol., 2001; 22: 348–66.
73. M. J. Duffy, J. M. Bonfrer, J. Kulpa, G. J.S. Rustin, G. Soletormos, G. C. Torre, et al. CA125 in ovarian cancer: European Group on Tumor Markers guidelines for clinical use. Int J Gynecol Cancer., 2005; 15: 679–91.

74. Mojgan Karimi–Zarchi, Nadereh Behtash, Azamsadat Mousavi, Fazemeh Ghaem Maghami, Mitra Moddares Gilani, Zohreh Chiti, et al. A survey on the role of cancer 47 antigen 125 (CA125), human epididymis protein 4 (HE4), risk of ovarian malignancy algorithm (ROMA), and risk of malignancy index (RMI) in pelvic mass. Int J Cancer Manag, 2018; 11(12): 1–7.
75. Jillian T. Henderson, Elizabeth M. Webber, George F. Sawaya. Screening for ovarian cancer updated evidence report and systematic review for the US preventive services task force. J Am Med Assoc., 2018; 319(6): 595–606.
76. Patrizia Bottoni, Roberto Scatena. The role of CA 125 as tumor marker: Biochemical and clinical aspects. Adv Exp Med Biol., 2015; 867: 229–44.
77. Maria Lee, Min Young Chang, Hanna Yoo, Kyung Eun Lee, Doo Byung Chay, Hanbyoul Cho, et al. Clinical significance of CA125 level after the first cycle of chemotherapy on survival of patients with advanced ovarian cancer. Yonsei Med J., 2016; 57(3): 580–7.
78. J.A.L. Liefers-Visser, R.A.M. Meijering, A.K.L. Reyners, A.G.J. van der Zee, S. de Jong. IGF system targeted therapy: Therapeutic opportunities for ovarian cancer. Cancer Treat Rev., 2017; 60: 90–9.
79. MK Tuxen, G Sölétormos, P Dombernowsky. Serum tumour marker CA 125 in monitoring of ovarian cancer during first-line chemotherapy. Br J Cancer., 2001; 84(10): 1301–7.
80. Khalid El Bairi, Said Afqir, Mariam Amrani. Is HE4 Superior over CA-125 in the Follow-up of Patients with Epithelial Ovarian Cancer? Curr Drug Targets., 2020; 21: 1026–33.
81. Jinping Li, Sean Dowdy, Tracy Tipton, Karl Podratz, Wei Guo Lu, Xing Xie, et al. HE4 as a biomarker for ovarian and endometrial cancer management. Expert Rev Mol Diagn, 2009; 9(6): 555–66.
82. Chen Zhang, Haoyue Hu, Xiaoyan Wang, Yajuan Zhu, Ming Jiang. WFDC Protein: A Promising Diagnosis Biomarker of Ovarian Cancer. J Cancer., 2021; 12(18): 5404–12.
83. Archana R. Simmons, Keith Baggerly, Robert C. Bast Jr. The emerging role of HE4 in the evaluation of epithelial Ovarian and endometrial carcinomas. Oncology., 2013; 27(6): 548–56.
84. Emilia Gasiorowska, Tomasz Kluz, Dawid Lipski, Wojciech Warchoł, Andrzej 48 Tykarski, Ewa Nowak-Markwitz. Human epididymis protein 4 (HE4) reference limits in polish population of healthy women, pregnant women, and women with benign ovarian tumors. Dis Markers, 2019; 2019: 1–7.
85. Anthony Richards, Unine Herbst, Jane Manalang, Selvan Pather, Samir Saidi, Trevor Tejada-Berges, et al. HE4, CA125, the Risk of Malignancy Algorithm and the Risk of Malignancy Index and complex pelvic masses - A prospective comparison in the preoperative evaluation of pelvic masses in an Australian population. Aust New Zeal J Obstet Gynaecol., 2015; 55: 493–7.
86. Marijn M. Speeckaert, Reinhart Speeckaert, Joris R. Delanghe. Human Epididymis Protein 4 in Cancer Diagnostics. A Promising and Reliable Tumor Marker. 1st ed. Vol. 59, Advances in Clinical Chemistry. Elsevier Inc., 2013; 1–21.
87. Emanuela Anastasi, Antonella Farina, Teresa Granato, Flavia Colaiacovo, Beatrice Pucci, Sara Tartaglione, et al. Recent Insight about HE4 Role in Ovarian Cancer Oncogenesis. Int J Mol Sci., 2023; 24(10479): 1–14.
88. Anita Chudecka-Głaz, Aneta Cymbaluk-Płoska, Małgorzata Wężowska, Janusz Menkiszak. Could HE4 level measurements during firstline chemotherapy predict response to treatment among ovarian cancer patients? PLoS One., 2018; 13(3): 1–16.
89. Anita Chudecka-Głaz, Aleksandra Strojna, Kaja Michalczyk, Sylwia Wieder-Huszla, Krzysztof Safranow, Edyta Skwirczy´nska, et al. Evaluation of He4 Use in the Diagnosis of Ovarian Cancer: First and Second Recurrence, and an Analysis of HE4 Concentration during Second- and Third-Line Chemotherapy. Diagnostics., 2023; 13(452): 1–27.
90. Ruchi Arora, Shilpa M. Patel, Abhilash V, Priyanka Vemanamandi. Diagnostic accuracy of HE4 and risk of ovarian malignancy algorithm in prediction of ovarian cancer in patients with pelvic mass: a regional cancer centre experience. Int J Reprod Contraception, Obstet Gynecol., 2022; 11(5): 1531–8.
91. Moore RG, Hill EK, Horan T, Yano N, Kim KK, MacLaughlan S, et al. HE4 (WFDC2) gene overexpression promotes ovarian tumor growth. Sci Rep., 2014; 4(3574): 1–7.
92. Su Wei, Hui Li, Bei Zhang. The diagnostic value of serum HE4 and CA-125 and 49 ROMA index in ovarian cancer. Biomed Reports, 2016; 5(1): 41–4.
93. Manikandan Lakshmanan, Vijay Kumar, Arun Chaturvedi, Sanjeev Misra, Sameer Gupta, Naseem Akhtar, et al. Role of serum HE4 as a prognostic marker in carcinoma of the ovary. Indian J Cancer., 2019; 56(3): 216–21.
94. Tingting Zhao, Weiping Hu. CA125 and HE4: Measurement Tools for Ovarian Cancer. Gynecol Obstet Invest., 2016; 81(5): 430–5.
95. Alexandra Samborski, M. Craig Miller, Alexandra Blackman, Shannon MacLaughlanDavid, Amanda Jackson, Geralyn Lambert-Messerlian, et al. HE4 and CA125 serum biomarker monitoring in women with epithelial ovarian cancer. Tumor Biol., 2022; 44: 205–13.
96. Elham O Hamed, Hydi Ahmed, Osama B Sedeek, Abeer M Mohammed, Ali A Abd-Alla, Hazem M Abdel Ghaffar. Significance of HE4 estimation in comparison with CA125 in diagnosis of ovarian cancer and assessment of treatment response. Diagn Pathol., 2013; 8(11): 1–8.

97. Richard G. Moore, D. Scott McMeekin, Amy K. Brown, Paul DiSilvestro, M. Craig Miller, W. Jeffrey Allard, et al. A novel multiple marker bioassay utilizing HE4 and CA125 for the prediction of ovarian cancer in patients with a pelvic mass. Gynecol Oncol., 2009; 112: 40–6.
98. Huhtinen K, Suvitie P, Hiissa J, Junnila J, Huvila J, Kujari H, et al. Serum HE4 concentration differentiates malignant ovarian tumours from ovarian endometriotic cysts. Br J Cancer., 2009; 100: 1315–9.
99. Heitz F, Lakis S, Harter P, Heikaus S, Sehouli J, Talwar J, et al. Cell-free tumor DNA, CA125 and HE4 for the objective assessment of tumor burden in patients with advanced high-grade serous ovarian cancer. PLoS One., 2022; 17(2): 1–15.
100. Ingegerd Hellstrom, John Raycraft, Martha Hayden-Ledbetter, Jeffrey A. Ledbetter, Michel Schummer, Martin McIntosh, et al. The HE4 (WFDC2) protein is a biomarker for ovarian carcinoma. Cancer Res., 2003; 63(13): 3695–700.
101. Chen X, Zhou H, Chen R, He J, Wang Y, Huang L, et al. Development of a multimarker assay for differential diagnosis of benign and malignant pelvic masses. Clin Chim Acta., 2015; 440: 57–63.
102. Dochez V, Caillon H, Vaucel E, Dimet J, Winer N, Ducarme G. Biomarkers and algorithms for diagnosis of ovarian cancer: CA125, HE4, RMI and ROMA, a review. J Ovarian Res., 2019; 12(28): 1–9.
103. Goff BA, Agnew K, Neradilek MB, Gray HJ, Liao JB, Urban RR. Combining a symptom index, CA125 and HE4 (triple screen) to detect ovarian cancer in women with a pelvic mass. Gynecol Oncol., 2017 Nov; 147(2): 291–5.
104. Urban N, Thorpe JD, Bergan LA, Forrest RM, Kampani A V., Scholler N, et al. Potential Role of HE4 in Multimodal Screening for Epithelial Ovarian Cancer. JNCI J Natl Cancer Inst., 2011 Nov 2; 103(21): 1630–4.
105. Masaya Uno, Rie Matsuo, Naoki Maezawa, Tomoyasu Kato. Evaluation of follow-up observation using human epididymis protein 4, a tumor marker, in patients with ovarian cancer. Obstet Gynecol Sci., 2023; 66(4): 290–9.
106. Dong Mi, Yuexiang Zhang. Prognostic value of serum HE4 in patients with advanced ovarian, fallopian tube, and peritoneal carcinoma. Arch Gynecol Obstet, 2020; 301(3): 779–85.
107. Plotti F, Guzzo F, Schirò T, Terranova C, De Cicco Nardone C, Montera R, et al. Role of human epididymis protein 4 (HE4) in detecting recurrence in CA125 negative ovarian cancer patients. Int J Gynecol Cancer., 2019; 29: 768–71.
108. Manganaro L, Michienzi S, Vinci V, Falzarano R, Saldari M, Granato T, et al. Serum HE4 levels combined with CE CT imaging improve the management of monitoring women affected by epithelial ovarian cancer. Oncol Rep., 2013; 30: 2481–7.
109. Angioli R, Capriglione S, Aloisi A, Guzzo F, Luvero D, Miranda A, et al. Can HE4 predict platinum response during first-line chemotherapy in ovarian cancer? Tumor Biol., 2014; 35(7): 7009–15.
110. Molina R, Escudero JM, Augé JM, Filella X, Foj L, Torné A, et al. HE4 a novel tumour marker for ovarian cancer: comparison with CA 125 and ROMA algorithm in patients with gynaecological diseases. Tumor Biol., 2011; 32(6): 1087–95.
111. Salminen L, Gidwani K, Grènman S, Carpén O, Hietanen S, Pettersson K, et al. HE4 in the evaluation of tumor load and prognostic stratification of high grade serous ovarian carcinoma. Acta Oncol (Madr), 2020; 59(12): 1461–8.
112. Havrilesky LJ, Whitehead CM, Rubatt JM, Cheek RL, Groelke J, He Q, et al. Evaluation of biomarker panels for early stage ovarian cancer detection and monitoring for disease recurrence. Gynecol Oncol., 2008; 110(3): 374–82.
113. Schummer M, Drescher C, Forrest R, Gough S, Thorpe J, Hellström I, et al. Evaluation of ovarian cancer remission markers HE4, MMP7 and Mesothelin by comparison to the established marker CA125. Gynecol Oncol.,s 2012; 125(1): 65–9.
114. Gong S, Quan Q, Meng Y, Wu J, Yang S, Hu J, et al. The value of serum HE4 and CA125 levels for monitoring the recurrence and risk stratification of endometrial endometrioid carcinoma. Heliyon., 2023; 9(7): 1–9.
115. Amitava Dasgupta, Amer Wahed. Tumor Markers. In: Clinical Chemistry, Immunology and Laboratory Quality Control. Elsevier, 2014; 229–47.
116. Nassir M, Guan J, Luketina H, Siepmann T, Rohr I, Richter R, et al. The role of HE4 for prediction of recurrence in epithelial ovarian cancer patients—results from the OVCAD study. Tumor Biol., 2015; 37(3): 3009–16.
117. Maksimiuk M, Budzik MP, Deptała A, Badowska-Kozakiewicz AM. HE4 – not only an ovarian cancer biomarker – a brief review. Nowotw J Oncol., 2019; 69(3–4): 142–5.
118. Zhang Y, Jin Q, Zhao Z, Zhao Q, Yu X, Yan L, et al. Carbohydrate Antigen 125 Is a Biomarker of the Severity and Prognosis of Pulmonary Hypertension. Front Cardiovasc Med., 2021; 8: 1–10.