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# Comparison of CA-125 and HE4 in ovarian cancer recurrence detection

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**ABSTRACT**

**Purpose:** This study aims to comprehensively evaluate CA-125 and HE4 as predictors of ovarian cancer (OC) recurrence in the same patient population.

**Methods:** We systematically searched the WOS, PubMed, and Scopus databases on May 8, 2024, for studies investigating both tumor markers CA-125 and HE4 in the same patient population of ovarian cancer recurrence. We calculated pooled values of AUC, sensitivity, specificity, and univariate or multivariate hazard ratios (HR) for both tumor markers in serum using a random effects model and StataMP 17.0 software.

**Results:** Thirteen articles comprising 1026 patients satisfied the inclusion criteria. Liquid-biopsy-based HE4 and CA-125 measurements were both proven to have high predictive value for detecting OC recurrence with comparable AUC values ($AUC_{HE4}$:0.78, 95% CI=0.73-0.83; $AUC_{CA125}$:0.80, 95% CI=0.73-0.88). While sensitivity of HE4 tests was higher than their specificity ($Sensitivity_{HE4}$=80.7%; 95% CI=73-88.4; $I^2$=77.05%; p<0.001; $Specificity_{HE4}$=77.8%; 95% CI=68.9-86.6; $I^2$=83.88%; p<0.001) in detecting OC recurrence, specificity was comparably higher for CA-125 analyses ($Sensitivity_{CA-125}$=71.4%; 95% CI=60.2-82.7; $I^2$=85,67%; p<0.001; $Specificity_{CA-125}$=94.5%; 95% CI=91.9-97.1; $I^2$=10.64%; p=0.34). Pooled HR values indicate that increased values of HE4 and CA125 increase the risk for worse progression-free survival by 3.1 (95% CI=1.3–5.0, $I^2$=0.00 %, p=0.38) and 2.4-fold (95% CI=1.3–3.5, $I^2$=0.00 %, p=0.93) respectively. HE4 indicates worse overall survival (HR=6.9, CI=0.8–12.6, $I^2$=0.00 %, p=0.7).

**Conclusions:** We suggest that HE4 is valuable as a recurrence tracker, with its higher sensitivity, while CA-125 can be used as a validator due to its higher specificity. Further prospective studies analyzing both biomarkers together are required for complete validation.

**Keywords:** ovarian cancer, recurrence, HE4, CA-125

**ÖZET**

**Amaç:** Bu çalışmanın amacı, over kanseri (OK) rekürrensinin izleminde, CA-125 ve HE4'ü aynı hasta popülasyonunda kapsamlı şekilde değerlendirmektir.

**Yöntemler:** OK rekürrensinin tespitinde CA-125 ve HE4 tümör belirteçlerinin etkinliğini araştıran çalışmalar, 8 Mayıs 2024'te WOS, PubMed ve Scopus veri tabanlarında sistematik olarak arandı. Serumdaki her iki tümör belirteci için AUC, duyarlılık, özgüllük ve tek değişkenli veya çok değişkenli tehlike oranlarının (HR) bileşik değerleri, StataMP 17.0 yazılımı kullanarak, rastgele etkiler modeli ile hesaplandı.

**Bulgular:** 1026 hastayı kapsayan on üç makale, dahil etme kriterlerini karşıladı. Sıvı biyopsi bazlı HE4 ve CA-125 ölçümlerinin, birbirine yakın bileşik AUC değerleri ile, OK rekürrensini yüksek prediktif etkinlik ile tespit ettiği gösterilmiştir ($AUC_{HE4}$:0.78, 95% CI=0.73-0.83; $AUC_{CA125}$:0.80, 95% CI=0.73-0.88). HE4 testlerinin OC rekürrensini saptamadaki duyarlılığı, özgüllüğünden daha yüksek iken ($Sensitivite_{HE4}$=80.7%; 95% CI=73-88.4; $I^2$=77.05%; p<0.001; $Spesifite_{HE4}$=77.8%; 95% CI=68.9-86.6; $I^2$=83.88%; p<0.001), CA-125 analizlerinde özgüllük karşılaştırmalı olarak daha yüksektir ($Sensitivite_{CA-125}$=71.4%; 95% CI=60.2-82.7; $I^2$=85,67%; p<0.001; $Spesifite_{CA-125}$=94.5%; 95% CI=91.9-97.1; $I^2$=10.64%; p=0.34). Bileşik HR değerleri; HE4 ve CA125 değerlerindeki artışın, daha kötü progresyona sahip sağkalım riskini sırasıyla 3,1 (95% CI=1,3–5,0, $I^2$=0,00 %, p=0,38) ve 2,4 kat (95% CI=1,3–3,5, $I^2$=0,00 %, p=0,93) artırdığını göstermektedir. HE4 yüksekliği, daha kötü genel sağkalımı göstermektedir (HR=6,9, CI=0,8–12,6, $I^2$=0,00 %, p=0,7).

**Sonuçlar:** Bu çalışmanın sonucunda elde edilen veriler ışığında; daha yüksek duyarlılığa sahip HE4'ün, OK rekürrensinin taranmasında etkinliğinin daha yüksek olduğu; özgüllük değeri daha yüksek olan CA-125'in ise rekürrensin doğrulanmasında sekonder biyobelirteç olarak daha etkin kullanılabileceği önerilmektedir. Her iki tümör biyobelirtecini birlikte analiz eden yeni prospektif çalışmalara ihtiyaç vardır.

**Anahtar Kelimeler:** over kanseri, rekürrens, HE4, CA-125

Globally, ovarian cancer (OC) ranks as the eighth most common malignancy in women (1) and has the highest mortality rate among all gynecological neoplasms (2). The overall 5-year survival rate is below 40% (3). The standard initial treatment regimen consists of extensive cytoreductive surgery, systemic chemotherapy incorporating taxanes and platinum agents, and subsequent individualized maintenance therapies (4). The absence of robust predictive biomarkers, limited understanding of tumor biology, and the development of chemotherapy resistance exacerbate the poor prognosis as well as the high rates of recurrence (5). Therefore, implementing enhanced follow-up is crucial for the early detection of ovarian cancer recurrence.

CA-125 and HE4, which are diagnostic biomarkers that have successfully been used for the differentiation of benign and malignant OC cases, are currently being investigated for their prognostic roles in identifying disease recurrence. CA-125 is a biomarker bound by a monoclonal antibody generated using an ovarian cell line and its concentration in serum is widely used as a valuable tool for OC detection and surveillance (6,7). CA-125 levels at the time of disease relapse have been shown to predict overall survival regardless of treatment approach (8). The well-documented limitations of CA-125, including its poor sensitivity for early-stage disease, persistence as minimal residual disease, and elevation in various benign conditions, significantly hinder its clinical utility (9). HE4, which was initially identified in epididymal epithelium as a potential protease inhibitor involved in sperm maturation, is later observed to be abnormally elevated in OC tissue compared to healthy ovarian tissue. While HE4 demonstrates better prognostic value by predicting disease recurrence earlier than that of CA-125 (10–12), additional factors such as age, smoking, and renal disease were also shown to increase HE4 levels (13). Despite FDA approval of the combined use of HE4 and CA-125 in post-treatment monitoring, their efficacy in detecting recurrent disease remains understudied (14–18). Furthermore, there is substantial variation in the utilization of HE4 as a diagnostic and prognostic marker across European countries and worldwide (15).

This meta-analysis aims to evaluate the prognostic potential and recurrence-predictive value of serum HE4 and CA-125 by pooling the available data in the literature that is published until May 8, 2024.

## Materials and Methods

### *Search strategy*

We followed the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) 2020 Statement to identify, select, appraise, and synthesize the studies included in this meta-analysis (19). Web of Science, PubMed, and Scopus databases were systematically searched up to May 8, 2024, for studies reporting the prognostic values of both CA-125 and HE4 tumor markers in detecting ovarian cancer recurrence. No date restriction was applied. The MeSH terms and additional keywords used for each database were: ("ovarian cancer" OR "ovarian carcinoma" OR "ovarian tumor" OR "ovarian tumour" OR "ovarian neoplasms" OR "ovarian malignancy") AND ("recurrence" OR "relapse") AND ("HE4" OR "human epididymis protein 4" OR "HE-4") AND ("CA125" OR "CA-125").

### *Screening and selection of studies*

Documents shortlisted by initial keyword search were downloaded as BibTex documents from Web of Science, PubMed, and Scopus databases individually, and BibTex files were uploaded to the Mendeley 2.99.0 (Elsevier Ltd.) reference manager. Article duplications were initially removed by Mendeley's automatic duplication tracker. The remaining articles were then aligned by the document name, and additional duplications were removed manually.

The final article list for the initial screening phase was transferred from Mendeley to an Excel spreadsheet. Both authors (ŞO and CCS) independently reviewed the abstract of each article on this list according to PECOS criteria listed in (Supplementary Table 1). Descriptive information including article title, publication type/year/journal, author name, study type, tumor marker information, and information about the detection of recurrence were extracted from each abstract, entered in individual Excel sheets named "Initial Screening Results (ISR)" separately by ŞO and CCS, and used for the initial elimination process. Book chapters, case reports, conference proceedings, comments, dissertations, editorials, guidelines, meeting abstracts, meta-analyses, reviews, and technical reports were excluded. Original articles in English that investigated both biomarkers CA-125 and HE4 in the serum of patients with histologically confirmed recurrent ovarian carcinoma were included in the study. Articles lacking patient data (cell culture studies, studies performed on model organisms), studies examining cancers other

than ovarian cancer, studies that did not address recurrent cases, and those that did not evaluate both tumor markers consecutively in the same patient population were excluded from the study. The study population was restricted to studies that exclusively use recurrent OC patients as the study group, and non-recurrent OC patients as controls. Two investigators crosschecked each other's ISR sheet; discrepancies regarding article selection were resolved by discussion; and documents to be included in the detailed evaluation were shortlisted as a "Detailed Screening (DS) list".

### *Data extraction*

During the secondary selection and data extraction process, the full-text contents of each article that was shortlisted in the DS list and their supplementary materials were further examined. Comprehensive data from the eligible studies were extracted as detailed in Table 1. Respective data from the human studies examining both tumor markers, CA-125 and HE4, in ovarian cancer recurrence which are compatible with the PECOS criteria (Supplementary Table 1) were extracted as Eligible Data (ED) by both authors (ŞO and CCS) separately. Data extraction sheets created by each author were cross-checked by the other investigator. Disagreements were reconciled through collaborative review, and the consolidated data was subjected to statistical analysis. Quality assessment of the included studies was performed according to quality appraisal guidelines (20).

**Supplementary Table 1:** PECOS criteria for inclusion and exclusion of studies

| Criteria | Inclusion Criteria | Exclusion Criteria |
|---|---|---|
| Participants | Studies that involve<br>- Female ovarian cancer patients whose recurrence is tracked following cancer treatment<br>- Treatment regimes that are clinically valid for ovarian cancer treatment such as "surgery only", "surgery + chemotherapy", "NACT + surgery", "NACT + surgery + chemotherapy", etc. | - Patients with other types of cancer<br>- Studies that do not examine ovarian cancer patients<br>- Studies that do not examine humans |
| Exposure | Studies in which<br>- CA-125 and HE4 are measured from patients' sera.<br>- Measurements are performed from sera that are collected during the follow-up period | - Studies that do not involve measurement of CA-125 and HE4 from patients' sera.<br>- Studies only involving HE4 and CA-125 measurements from the serum samples that are collected before the treatment, instead of the follow-up period. |
| Comparison | Studies in which<br>- The recurrence prediction efficiencies of the following biomarkers are compared: HE4, CA125, and HE4 + CA125 combination | - Studies comparing the recurrence prediction efficiencies of markers other than HE4 and CA-125.<br>- Studies comparing the efficiency of HE4 and CA-125 for cancer diagnosis or post-surgery success, but do not compare the efficiency of these markers in the detection of recurrence during the follow-up period. |
| Outcome | Studies in which<br>- AUC (Area Under the Curve value for ROC analysis),<br>- Sensitivity and specificity values<br>- HR (Hazard Ratio) values<br>are provided for<br>- Detection of recurrence<br>- Disease Free Survival (DFS)<br>- Progression Free Survival (PFS)<br>- Overall Survival (OS)<br><br>with sufficient statistical details enabling meta-analysis | Studies that do not<br>- Provide data regarding recurrence statistics,<br>- Provide AUC, Sensitivity / Specificity, and HR values for<br>- Provide 95% CI values if AUC and HR are analyzed<br>- Provide 95% CI values for sensitivity and specificity OR positive predictive and negative predictive values OR number of patients with/without recurrence having low/high values for HE+ and CA-125. |
| Study Design | - Case-control<br>- Prospective Cohort<br>- Retrospective Cohort | - Review, editorial notes, book chapters<br>- Studies that examine the biomarkers only on model organisms or cell culture, but provide no human patient data<br>- Case Reports, technical reports<br>- Conference proceedings, meeting abstracts, comments, dissertations, editorials<br>- Guidelines |

### *Statistical evaluation*

StataMP 17.0 software (Stata Corp LLC, College Station, TX, USA) was used for statistical analyses. The AUC values of both tumor markers were analyzed individually and in combination to predict OC recurrence. Sensitivity and specificity were calculated for each tumor marker separately and in combination. Unadjusted hazard ratios (UHRs) presented as univariate HRs in the original studies, and adjusted hazard ratios (AHRs) reported as multivariate HRs were analyzed separately. Forest plots including the respective confidence intervals were generated to visualize pooled UHR or AHR estimates for PFS and OS, in addition to AUC, sensitivity, and specificity values. The homogeneity and heterogeneity of the pooled studies were assessed using the Q ($p<0.05$ indicates heterogeneity, $p>0.05$ indicates homogeneity) and $I^2$ statistics ($I^2<25$ % indicating unimportant heterogeneity; 25 %< $I^2<50$ % indicating moderate heterogeneity; 50 %< $I^2<75$ % indicating substantial heterogeneity; $I^2>75$ % indicating considerable heterogeneity), respectively (21). While the Regression-based Egger test was used for small-study effects, the random-effects model (DerSimonian – Laird estimation) was chosen for pools with remarkable heterogeneity. Publication bias was investigated using Begg's funnel plots and Egger's linear regression test.

**Supplementary Table 2:** Evaluation of the quality of the eligible articles included in the meta-analysis.

| Article Ref | Name of First Author | Study Participation | Study Attrition | Prognostic Factor Measurement | Outcome Measurement | Confounding Measurement and Account | Analysis |
|---|---|---|---|---|---|---|---|
| (22) | Chen L. | YES | YES | YES | YES | PARTLY | PARTLY |
| (26) | Gong Z | YES | YES | YES | YES | PARTLY | YES |
| (27) | Han JJ | PARTLY | YES | PARTLY | PARTLY | PARTLY | YES |
| (28) | Innao P | YES | YES | PARTLY | YES | PARTLY | YES |
| (29) | Kotowicz BU | YES | YES | YES | YES | PARTLY | YES |
| (30) | Li R | PARTLY | YES | PARTLY | YES | PARTLY | YES |
| (5) | Nassir M | YES | YES | YES | YES | YES | YES |
| (31) | Rong Y | YES | YES | YES | YES | PARTLY | YES |
| (32) | Salminen L | YES | YES | YES | PARTLY | YES | YES |
| (33) | Shen ZY | YES | PARTLY | YES | YES | PARTLY | YES |
| (23) | Steffensen KD | YES | YES | YES | YES | PARTLY | YES |
| (24) | Sun J | YES | YES | YES | YES | PARTLY | YES |
| (25) | Uno M | YES | YES | YES | YES | YES | PARTLY |

## Results

An initial keyword search in the Web of Science, PubMed, and Scopus databases yielded 102 articles accepted by publishers as of May 8, 2024 (Figure 1). In the first screening step, the abstracts (and full-text documents if accurate elimination was compromised by lack of sufficient information in the abstract) of these articles were thoroughly reviewed and 49 articles were shortlisted for in-depth full-text screening. Only the studies where both CA-125 and HE4 were analyzed in recurrent OC cases were included in the final analysis. The flowchart depicted in Figure 1 provides a detailed overview of the selection process. At the end of the detailed full-text screening, 13 articles comprising 1026 patients were eligible for this meta-analysis, which reported the results for both CA-125 and HE4 measurements in patients with recurrence provided that these studies included the required statistical data. The risk assessment for bias performed according to quality appraisal guidelines (20) is shown in Supplementary Table 2. Key details from each article, such as study design, tumor marker information, sample characteristics, and patient demographics, were compiled and presented in Table 1 (5,22–33).

**Table 1:** Article Information, Study Characteristics

| Study Details | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Article No | Ref | Year of publication | Name of First Author | Country of study | Study Type | Treatment Regime | Follow-Up Duration | Sample Size | Sample Type | Method for HE4 & CA125 Measurement |
| 1 | (22) | 2018 | Chen L | China | Cross-sectional | Surgery + Platin-based chemotherapy | 4 years | 103 | Serum | ELISA |
| 2 | (26) | 2022 | Gong Z | China | Cross-sectional | N/A | 1-3 years | 73 | Serum | ELISA (HE4) ECLIA (CA125) |
| 3 | (27) | 2011 | Han JJ | USA | Prospective Cohort | Surgery + platinum/ taxane adjuvant chemotherapy | Recruitment: From 2000-2005 Follow Up: Until 2009 (max 5 years) | 23 | Plasma | ELISA (HE4) N/A (CA125) |
| 4 | (28) | 2016 | Innao P | Thailand | Prospective Cohort | Surgery + platinum/ paclitaxel adjuvant chemotherapy | Recruitment: From June 2014 - March 2016 Follow-Up: For 22 months | 47 | N/A | N/A |
| 5 | (29) | 2022 | Kotowicz BU | Poland | Cross-sectional | (Group I) surgical treatment + standard systemic treatment (Group II) NACT + surgery | 2.5 year | 64 | Serum | ECLIA - COBAS e601 |
| 6 | (30) | 2022 | Li R | China | Cross-sectional | (Group I) NACT + surgery (Group II) Surgery + carboplatin + paclitaxel | Recruitment: From 2016 and 2019 Follow Up: To March 2022 | 159 | Serum | N/A |
| 7 | (5) | 2016 | Nassir M | Europe-Multicenter | Retrospective Cohort | radical cytoreductive surgery + platinum-based chemotherapy | Until the day of the first recurrence | 62 | Serum | EIA (HE4) Luminex (CA125) |
| 8 | (31) | 2021 | Rong Y | China | Cross-sectional | Staging surgery or optimal cytoreductive surgery + Platinum-based combined chemotherapy | Recruitment: From July 2012 to December 2018 Follow-Up: To the end of 2019 (median follow-up: 35 months) | 89 | Serum | ECLIA |
| 9 | (32) | 2020 | Salminen L | Finland | Prospective Cohort | (Group I) Surgery + Chemotherapy (Group II) NACT + Surgery + Adjuvant Chemotherapy | Recruitment: Between 2009-2019 Follow-up: from 1.5 months to 10.2 years, median 2.5 years | 143 | Serum | EIA (HE4) ECLIA or EIA (CA125) |
| 10 | (33) | 2019 | Shen ZY | China | Cross-sectional | Surgery + Chemo (paclitaxel-cisplatin) | Recruitment: From July 2014 - Dec 2019 Follow Up: Every 3 months (Exact duration N/A in the text) | 58 | Serum | EIA (HE4) CLIA (CA125) |
| 11 | (23) | 2016 | Steffensen KD | Denmark | Prospective Cohort | radical cytoreductive surgery + carboplatin + paclitaxel | 26-86 month | 88 | Serum | EIA |
| 12 | (24) | 2020 | Sun J | China | Cross-sectional | Surgery + Chemotherapy | Recruitment: From January 2014 - December 2016 Follow Up: 6-60 month | 69 | Serum | ECLIA |
| 13 | (25) | 2023 | Uno M | Japan | Cross-sectional | (Group I) NACT + Surgery (Group II) Surgery + carboplatin + paclitaxel | 20.8 months (5.6-.43.9 months) | 48 | Serum | CMIA - ARCHITECT |

***ELISA:*** *Enzyme-Linked Immunosorbent Assay;* ***EIA:*** *Enzymatic Immuno Assay;* ***ECLIA:*** *Electrochemiluminescence;* ***CLIA:*** *Chemiluminescent immunoassay;* ***CMIA:*** *Chemiluminescent Microparticle Capture Immunoassay;* ***NACT:*** *Neoadjuvant Chemotherapy,* ***N/A:*** *Not available*

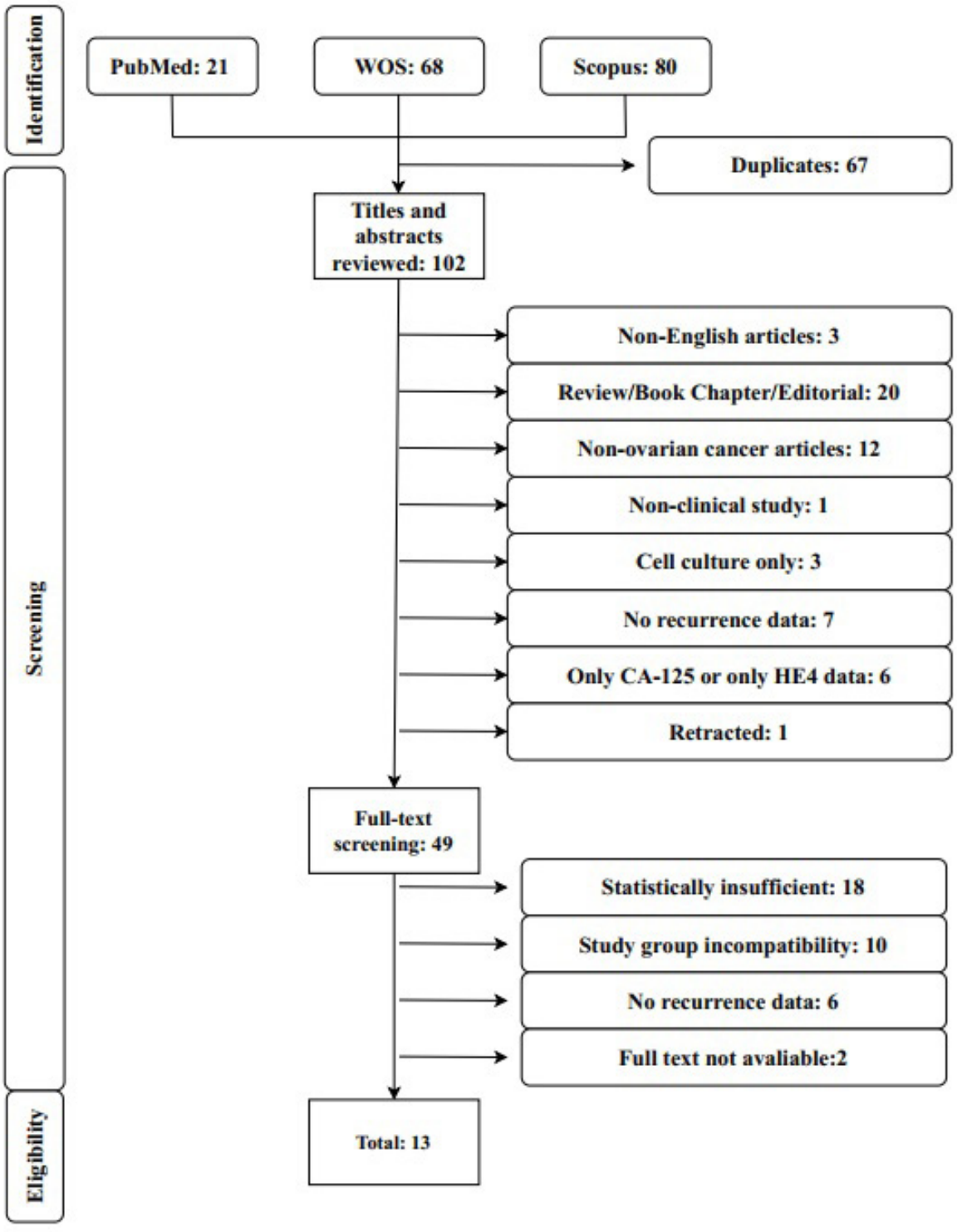



**Figure 1:** Flowchart of included articles.

**Supplementary Table 3:** AUC Data

| Name of First Author | Year of publication | Sampling Time/ Recurrence Time | Biomarker | CutOff | Analysis Criteria | AUC | SE of AUC | Total Nb of Patients | # of patients with recurrence | # of patients without recurrence |
|---|---|---|---|---|---|---|---|---|---|---|
| Gong Z | 2022 | | HE4 | 157.004 pmol/L | predicting poor prognosis | 0.77 | 0.057 | 73 | 33 | 40 |
| Gong Z | 2022 | | CA125 | 175.243 kU/L | predicting poor prognosis | 0.764 | 0.057 | 73 | 33 | 40 |
| Kotowicz BU | 2022 | (rec6)* | HE4 (6 months) | N/A | predicting poor prognosis | 0.785 | 0.063 | 64 | 24 | 40 |
| Kotowicz BU | 2022 | (rec6)* | CA125 (6 month) | N/A | predicting poor prognosis | 0.913 | 0.042 | 64 | 24 | 40 |
| Kotowicz BU | 2022 | (rec12)* | HE4 (12 months) | N/A | predicting poor prognosis | 0.739 | 0.063 | 64 | 31 | 33 |
| Kotowicz BU | 2022 | (rec12)* | CA125 (12 month) | N/A | predicting poor prognosis | 0.844 | 0.050 | 64 | 31 | 33 |
| Li R | 2022 | PTFM | HE4 | 64.14 pmol/L | detecting recurrence | 0.87 | 0.028 | 159 | 85 | 74 |
| Li R | 2022 | PTFM | CA125 | 24.3 U/mL | detecting recurrence | 0.94 | 0.019 | 159 | 85 | 74 |
| Nassir M | 2016 | (rec12)* | HE4 (recurrence in 12 months) | 49.5 pmol/L | diagnosis of recurrence in responders after 1st line chemotherapy | 0.81 | 0.065 | 62 | 19 | 43 |
| Nassir M | 2016 | (rec12)* | CA125 (recurrence in 12 months) | 20 U/mL | diagnosis of recurrence in responders after 1st line chemotherapy | 0.884 | 0.053 | 62 | 19 | 43 |
| Nassir M | 2016 | (rec12)* | HE4 & CA125 (recurrence in 12 months) | 49.5 pmol/L + 20 U/mL | diagnosis of recurrence in responders after 1st line chemotherapy | 0.93 | 0.042 | 62 | 19 | 43 |
| Rong Y | 2021 | (m6 rec6)* | HE4 (analyte measurement in 6th cycle) | 70 pmol/L | diagnosis of recurrence in 6 months | 0.73 | 0.076 | 89 | 16 | 73 |
| Rong Y | 2021 | (m6 rec6)* | CA125 (analyte measurement in 6th cycle) | 35 U/mL | diagnosis of recurrence in 6 months | 0.705 | 0.078 | 89 | 16 | 73 |
| Rong Y | 2021 | (m3/1 rec6)* | 3rd cycle of HE4 &1st cycle of CA125 | 70 pmol/L + 35 U/mL | diagnosis of recurrence in 6 months | 0.723 | 0.077 | 89 | 16 | 73 |
| Rong Y | 2021 | (m6 rec24)* | HE4 (analyte measurement in 6th cycle) | 70 pmol/L | DFS in 2 years | 0.666 | 0.063 | 89 | 30 | 59 |
| Rong Y | 2021 | (m6 rec24)* | CA125 (analyte measurement in 6th cycle) | 35 U/mL | DFS in 2 years | 0.6 | 0.065 | 89 | 30 | 59 |
| Rong Y | 2021 | (m3/1 rec24)* | 3rd cycle of HE4 &1st cycle of CA125 | 70 pmol/L + 35 U/mL | DFS in 2 years | 0.625 | 0.064 | 89 | 30 | 59 |
| Shen ZY | 2019 | PTFM | HE4 | 105 pmol/L | diagnosis of recurrence | 0.737 | 0.067 | 58 | 26 | 32 |
| Shen ZY | 2019 | PTFM | CA125 | 35 U/mL | diagnosis of recurrence | 0.825 | 0.057 | 58 | 26 | 32 |
| Steffensen KD | 2016 | (m6)* | HE4 (analyte measurement after 1st line treatment) | 41 pmol/L | diagnosis of recurrence | 0.6976 | 0.056 | 88 | 55 | 33 |
| Steffensen KD | 2016 | (m6)* | CA125 (analyte measurement after 1st line treatment) | 1 U/mL | diagnosis of recurrence | 0.6079 | 0.061 | 88 | 55 | 33 |
| Steffensen KD | 2016 | (m6)* | HE4 & CA125 (analyte measurement after 1st line treatment) | 41 pmol/L + 1 U/mL | diagnosis of recurrence | 0.7395 | 0.052 | 88 | 55 | 33 |
| Sun J | 2020 | PTFM | HE4 | 184 pmol/mL | diagnosis of recurrence | 0.858 | 0.046 | 69 | 54 | 15 |
| Sun J | 2020 | PTFM | CA125 | 57.5 U/L | diagnosis of recurrence | 0.847 | 0.048 | 69 | 54 | 15 |

***AUC:*** *Area Under the Curve;* ***SE:*** *Standard Error;* ***PTFM:*** *Post Treatment Follow-Up Analyte Measurement;* ***T1:*** *Treatment regime 1;* ***T2:*** *Treatment regime 2;* ***m3/1:*** *HE4 is analyzed from the serum sample collected at 3rd month of the treatment, CA-125 is analyzed from the serum sample collected at 1st month of the treatment;* ***m6:*** *respective biomarkers that are analyzed from the serum sample collected at 6th month of the treatment;* ***rec6:*** *detection of recurrence at the 6th month of the follow-up;* ***rec12:*** *detection of recurrence at the 12th month of the follow-up;* ***rec24:*** *detection of recurrence at the 24th month of the follow-up;* ***DFS:*** *Disease-Free Survival*

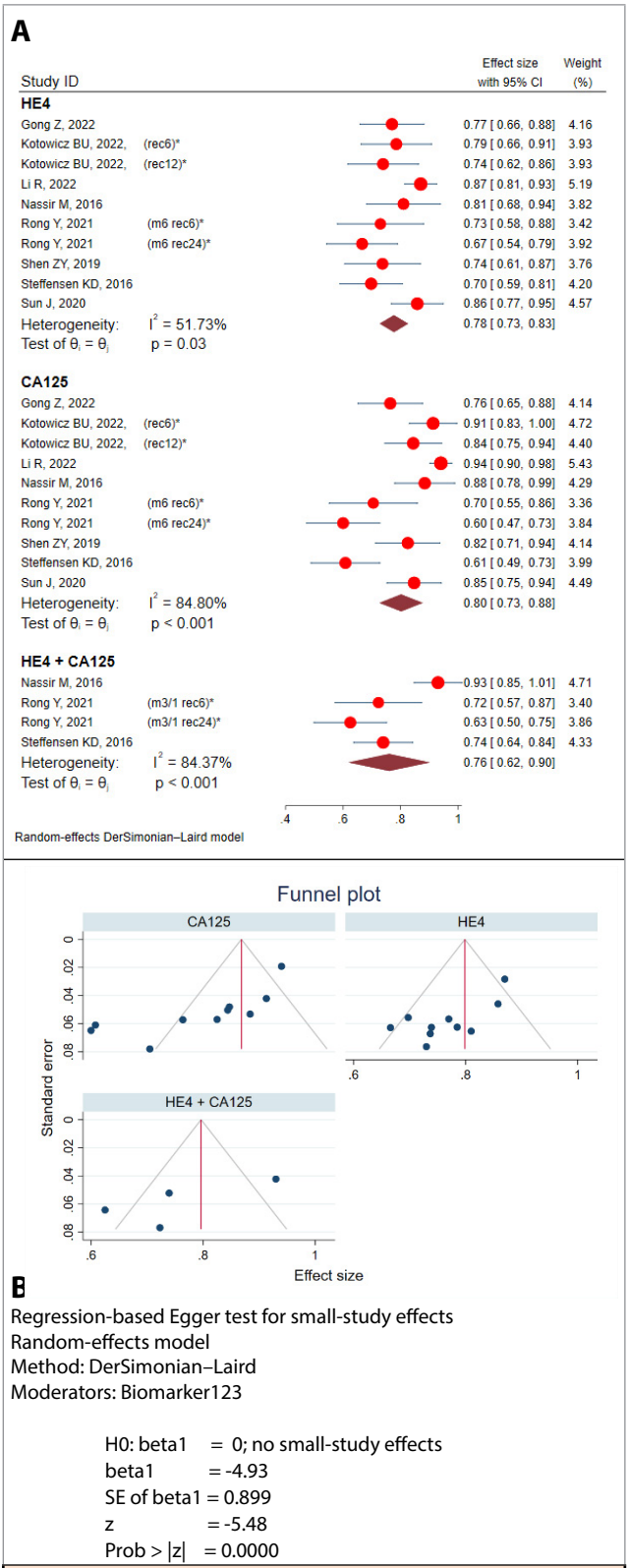
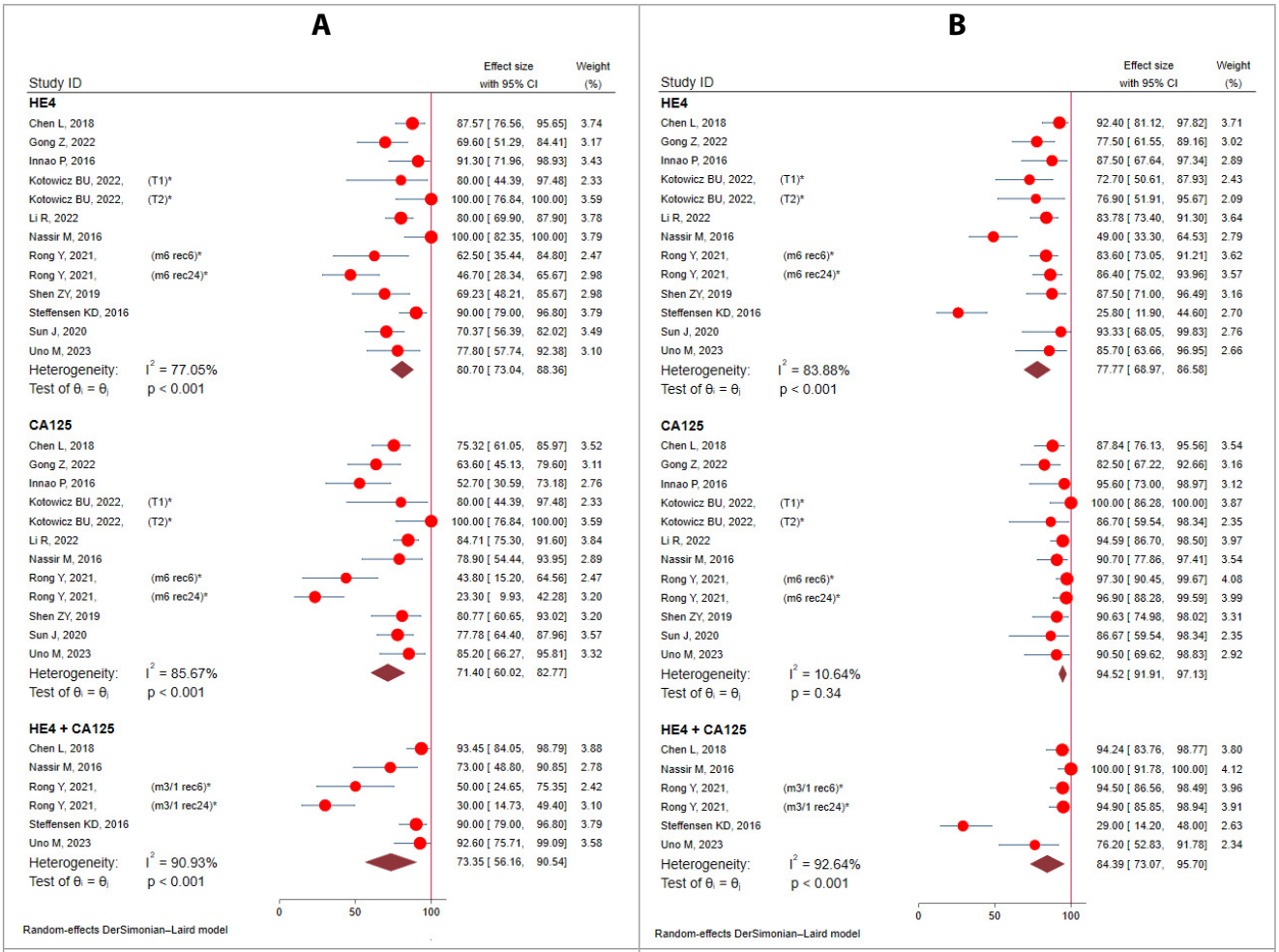



**Figure 2: The association of HE4 and CA-125 with recurrence detection of ovarian cancer patients.** Pooled AUC values of serum HE4, CA-125, and their combination regarding recurrence detection of ovarian cancer are seen as Forest graphs (**A**) and Funnel graphs together with the Egger-test scores (**B**).

While the potential of serum tumor markers CA-125 and HE4 for detecting the recurrence of OC was analyzed through ROC analysis in most of the eligible studies, only eight studies documented statistically sufficient AUC data (5,23,24,26,29–31,33); presented in detail in Supplementary Table 3. Three of these studies assessed AUC across different recurrence intervals (recurrence in 6 months, 12 months, or 24 months) or patient subgroups (5,29,31). Pooled AUC analysis of CA-125 and HE4 indicated comparable efficacy in detecting OC recurrence, with relatively higher heterogeneity index for CA-125 ($AUC_{HE4}$=0.78; 95% CI=0.73–0.83; $I^2$=51.73%; p=0.03; $AUC_{CA-125}$=0.80; 95% CI=0.73–0.88; $I^2$=84.80%; p<0.001 Figure 2). The performance of the two biomarkers together as a combined prognostic tool was analyzed only in three of these articles, one of which evaluated the combination in two different recurrence intervals (5,23,31). The high level of heterogeneity among the studies precludes a conclusion that the combination outperforms individual markers ($AUC_{HE4+CA-125}$=0.76; 95% CI=0.62–0.90; $I^2$=84.37%; p<0.001) (Figure 2).

Eleven of the eligible studies evaluated the sensitivity and specificity of serum tumor markers CA-125 and HE4 in predicting OC recurrence (5,22–26,28–31,33), comprehensive data is presented in Supplementary Table 4. Two of these studies assessed sensitivity and specificity across different recurrence intervals or patient subgroups (29,31). Pooled analysis performed here revealed an increased risk of recurrence in OC patients with higher serum HE4 ($Sensitivity_{HE4}$=80.7; 95% CI=73-88.4; $I^2$=77.05%; p<0.001; $Specificity_{HE4}$=77.8; 95% CI=68.9-86.6; $I^2$=83.88%; p<0.001), and higher serum CA-125 levels ($Sensitivity_{CA-125}$=71.4; 95% CI=60.2-82.7; $I^2$=85,67%; p<0.001; $Specificity_{CA-125}$=94.5; 95% CI=91.9-97.1; $I^2$=10.64%; p=0.34). Except for $Specificity_{CA-125}$, high heterogeneity was observed for all parameters due to the small sample size of the study pool, as shown in the funnel plot and indicated by the Egger test results. The usage of both tumor markers as a combination was evaluated only in five studies, one of which assessed two distinct recurrence intervals. The pooled analysis of the combination revealed sensitivity and specificity values comparable to each of the tumor markers measured individually ($Sensitivity_{HE4+CA-125}$=73.35; 95% CI=56.16-90.54; $I^2$=90.93%; p<0.001; $Specificity_{HE4+CA-125}$=84.39; 95% CI=73.07-95.70; $I^2$=92.64%; p<0.001) (Figure 3).




Regression-based Egger test for small-study effects
Random-effects model
Method: DerSimonian–Laird
H0: beta1 = 0; no small-study effects
beta1 = -4.04
SE of beta1 = 0.912
z = -4.43
Prob > |z| = 0.0000

Regression-based Egger test for small-study effects
Random-effects model
Method: DerSimonian–Laird
H0: beta1 = 0; no small-study effects
beta1 = -3.83
SE of beta1 = 0.714
z = -5.37
Prob > |z| = 0.0000

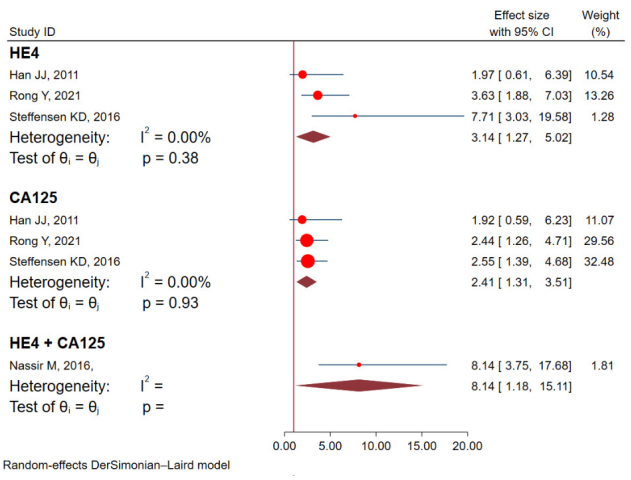
**Figure 3: Sensitivity and Specificity of HE4 and CA-125 biomarkers for detection of recurrence in ovarian cancer patients.** Pooled Sensitivity (**A** and **C**) and Specificity (**B** and **D**) values of serum HE4, CA-125, and their combination regarding recurrence detection of ovarian cancer are seen as Forest graphs (**A** and **B**) and Funnel graphs together with the Egger-test scores (**C** and **D**).

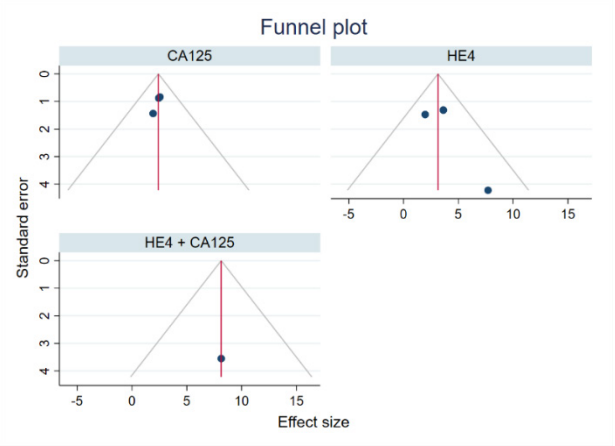
**Supplementary Table 4:** Sensitivity & Specificity Data

| | | | | | | Sensitivity | | | Specificity | | | | | With Recurrence | | Without Recurrence | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Name of First Author | Year of publication | Sampling Time/ Recurrence Time | Biomarker | CutOff | Analysis Criteria | % | 95% CI Min | 95% CI Max | % | 95% CI Min | 95% CI Max | # of OC patients with recurrence | # of OC patients without recurrence | Biomarker > Cutoff | Biomarker < Cutoff | Biomarker > Cutoff | Biomarker < Cutoff |
| Chen L | 2018 | PTFM | HE4 | 70 pmol/L | diagnosis of recurrent ovarian cancer | 87.6 | 76.6 | 95.6 | 92.4 | 81.1 | 97.8 | 52 | 51 | 46 | 6 | 4 | 47 |
| Chen L | 2018 | PTFM | CA125 | 35 IU/ml | diagnosis of recurrent ovarian cancer | 75.3 | 61.1 | 86.0 | 87.8 | 76.1 | 95.6 | 52 | 51 | 39 | 13 | 6 | 45 |
| Chen L | 2018 | PTFM | HE4 + CA125 | 70 pmol/L + 35 IU/ml | diagnosis of recurrent ovarian cancer | 93.5 | 84.1 | 98.8 | 94.2 | 83.8 | 98.8 | 52 | 51 | 49 | 3 | 3 | 48 |
| Gong Z | 2022 | | HE4 | 157 pmol/L | predicting poor prognosis | 69.6 | 51.3 | 84.4 | 77.5 | 61.5 | 89.2 | 33 | 40 | 23 | 10 | 9 | 31 |
| Gong Z | 2022 | | CA125 | 175 kU/L | predicting poor prognosis | 63.6 | 45.1 | 79.6 | 82.5 | 67.2 | 92.7 | 33 | 40 | 21 | 12 | 7 | 33 |
| Innao P | 2016 | PTFM | HE4 | 200 % | Prediction of Recurrence | 91.3 | 72.0 | 98.9 | 87.5 | 67.6 | 97.3 | 23 | 24 | 21 | 2 | 3 | 21 |
| Innao P | 2016 | PTFM | CA125 | 200 % | Prediction of Recurrence | 52.7 | 30.6 | 73.2 | 95.6 | 73.0 | 99.0 | 23 | 24 | 12 | 11 | 2 | 22 |
| Kotowicz BU | 2022 | (T1)* | HE4 | 79.1 pmol/L | detecting recurrence | 80.0 | 44.4 | 97.5 | 72.7 | 50.6 | 87.9 | 10 | 25 | 8 | 2 | 7 | 18 |
| Kotowicz BU | 2022 | (T1)* | CA125 | 30.7 IU/mL | detecting recurrence | 80.0 | 44.4 | 97.5 | 100.0 | 86.3 | 100.0 | 10 | 25 | 8 | 2 | 0 | 25 |
| Kotowicz BU | 2022 | (T2)* | HE4 | 90.4 pmol/L | detecting recurrence | 100.0 | 76.8 | 100.0 | 76.9 | 51.9 | 95.7 | 14 | 15 | 14 | 0 | 3 | 12 |
| Kotowicz BU | 2022 | (T2)* | CA125 | 25.6 IU/mL | detecting recurrence | 100.0 | 76.8 | 100.0 | 86.7 | 59.5 | 98.3 | 14 | 15 | 14 | 0 | 2 | 13 |
| Li R | 2022 | PTFM | HE4 | 64.14 pmol/l | detecting recurrence | 80.0 | 69.9 | 87.9 | 83.8 | 73.4 | 91.3 | 85 | 74 | 68 | 17 | 12 | 62 |
| Li R | 2022 | PTFM | CA125 | 24.3 IU/ml | detecting recurrence | 84.7 | 75.3 | 91.6 | 94.6 | 86.7 | 98.5 | 85 | 74 | 72 | 13 | 4 | 70 |
| Nassir M | 2016 | (rec12)* | HE4 | 49.5 pmol/l | diagnosis of recurrence in responders after 1st line chemotherapy | 100.0 | 82.4 | 100.0 | 49.0 | 33.3 | 64.5 | 19 | 43 | 19 | 0 | 22 | 21 |
| Nassir M | 2016 | (rec12)* | CA125 | 20 IU/ml | diagnosis of recurrence in responders after 1st line chemotherapy | 78.9 | 54.4 | 94.0 | 90.7 | 77.9 | 97.4 | 19 | 43 | 15 | 4 | 4 | 39 |
| Nassir M | 2016 | (rec12)* | HE4 + CA125 | 49.5 pmol/l + 20 IU/ml | diagnosis of recurrence in responders after 1st line chemotherapy | 73.0 | 48.8 | 90.9 | 100.0 | 91.8 | 100.0 | 19 | 43 | 14 | 5 | 0 | 43 |
| Rong Y | 2021 | (m6 rec6)* | HE4 | 70 pmol/L | diagnosis of recurrence in 6 months | 62.5 | 35.4 | 84.8 | 83.6 | 73.0 | 91.2 | 16 | 73 | 10 | 6 | 12 | 61 |
| Rong Y | 2021 | (m6 rec6)* | CA125 | 35 U/ml | diagnosis of recurrence in 6 months | 43.8 | 15.2 | 64.6 | 97.3 | 90.5 | 99.7 | 16 | 73 | 6 | 10 | 2 | 71 |
| Rong Y | 2021 | (m3/1 rec6)* | HE4 + CA125 | 70 pmol/L + 35 U/ml | diagnosis of recurrence in 6 months | 50.0 | 24.7 | 75.3 | 94.5 | 86.6 | 98.5 | 16 | 73 | 8 | 8 | 4 | 69 |
| Rong Y | 2021 | (m6 rec24)* | HE4 | 70 pmol/L | DFS in 2 years | 46.7 | 28.3 | 65.7 | 86.4 | 75.0 | 94.0 | 30 | 59 | 14 | 16 | 8 | 51 |
| Rong Y | 2021 | (m6 rec24)* | CA125 | 35 U/ml | DFS in 2 years | 23.3 | 9.9 | 42.3 | 96.9 | 88.3 | 99.6 | 30 | 59 | 7 | 23 | 2 | 57 |
| Rong Y | 2021 | (m3/1 rec24)* | HE4 + CA125 | 70 pmol/L + 35 U/ml | DFS in 2 years | 30.0 | 14.7 | 49.4 | 94.9 | 85.9 | 98.9 | 30 | 59 | 9 | 21 | 3 | 56 |
| Shen ZY | 2019 | PTFM | HE4 | 105 pmol/L | diagnosis of recurrence | 69.2 | 48.2 | 85.7 | 87.5 | 71.0 | 96.5 | 26 | 32 | 18 | 8 | 4 | 28 |
| Shen ZY | 2019 | PTFM | CA125 | 35 U/ml | diagnosis of recurrence | 80.8 | 60.6 | 93.0 | 90.6 | 75.0 | 98.0 | 26 | 32 | 21 | 5 | 3 | 29 |
| Steffensen KD | 2016 | (m6)* | HE4 | 41 pmol/L | diagnosis of recurrence | 90.0 | 79.0 | 96.8 | 25.8 | 11.9 | 44.6 | 55 | 33 | 50 | 5 | 24 | 9 |
| Steffensen KD | 2016 | (m6)* | HE4 + CA125 | 41 pmol/L + 1 U/ml | diagnosis of recurrence | 90.0 | 79.0 | 96.8 | 29.0 | 14.2 | 48.0 | 55 | 33 | 50 | 5 | 23 | 10 |
| Sun J | 2020 | PTFM | HE4 | 184 pmol/mL | diagnosis of recurrence | 70.4 | 56.4 | 82.0 | 93.3 | 68.1 | 99.8 | 54 | 15 | 38 | 16 | 1 | 14 |
| Sun J | 2020 | PTFM | CA125 | 57.5 U/L | diagnosis of recurrence | 77.8 | 64.4 | 88.0 | 86.7 | 59.5 | 98.3 | 54 | 15 | 42 | 12 | 2 | 13 |
| Uno M | 2023 | PTFM | HE4 | 70 pmol/mL | diagnosis of recurrence | 77.8 | 57.7 | 92.4 | 85.7 | 63.7 | 97.0 | 27 | 21 | 21 | 6 | 3 | 18 |
| Uno M | 2023 | PTFM | CA125 | 35 U/L | diagnosis of recurrence | 85.2 | 66.3 | 95.8 | 90.5 | 69.6 | 98.8 | 27 | 21 | 23 | 4 | 2 | 19 |
| Uno M | 2023 | PTFM | HE4 + CA125 | 70 pmol/mL + 35 U/L | diagnosis of recurrence | 92.6 | 75.7 | 99.1 | 76.2 | 52.8 | 91.8 | 27 | 21 | 25 | 2 | 5 | 16 |

***OC:*** *Ovarian Cancer;* ***95 % CI Min:*** *Lower limit of the 95 % confidence interval;* ***95 % CI Max:*** *Upper limit of the 95 % confidence interval;* ***PTFM:*** *Post Treatment Follow-Up Analyte Measurement;* ***T1:*** *Treatment regime 1;* ***T2:*** *Treatment regime 2;* ***m3/1:*** *HE4 is analyzed from the serum sample collected at 3rd month of the treatment, CA-125 is analyzed from the serum sample collected at 1st month of the treatment;* ***m6:*** *respective biomarkers that are analyzed from the serum sample collected at 6th month of the treatment;* ***rec6:*** *detection of recurrence at the 6th month of the follow-up;* ***rec12:*** *detection of recurrence at the 12th month of the follow-up;* ***rec24:*** *detection of recurrence at the 24th month of the follow-up;* ***DFS:*** *Disease-Free Survival*

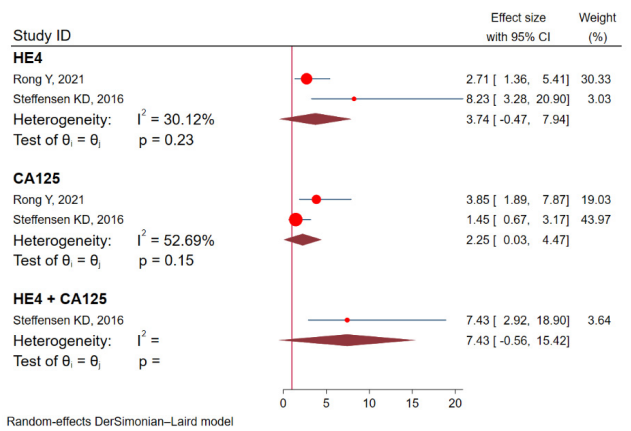
*(The statistical data highlighted in gray, although not explicitly presented in the cited article, are iterated using Medcalc based on the statistical data readily presented within the cited article.*

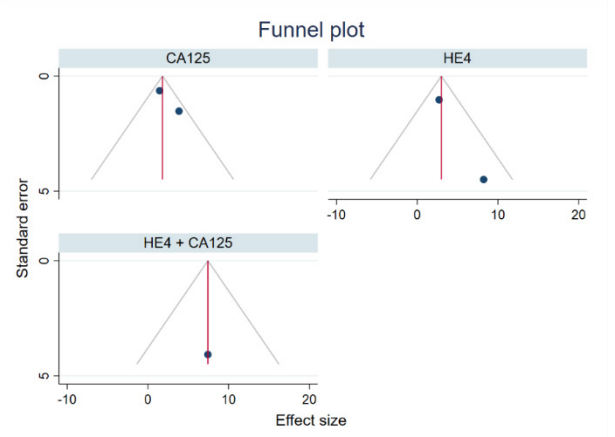



Regression-based Egger test for small-study effects
Random-effects model
Method: DerSimonian–Laird

H0: beta1 = 0; no small-study effects
beta1 = 1.46
SE of beta1 = 0.870
z = 1.68
Prob > |z| = 0.0933

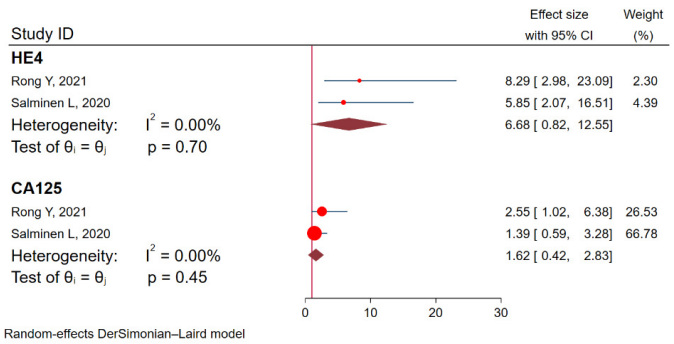


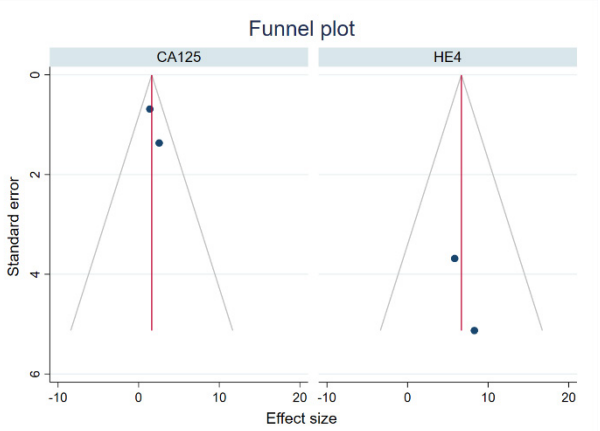




Regression-based Egger test for small-study effects
Random-effects model
Method: DerSimonian–Laird

H0: beta1 = 0; no small-study effects
beta1 = 1.92
SE of beta1 = 0.791
z = 2.43
Prob > |z| = 0.0153

**Figure 4: The association between serum values of HE4 and CA-125 with progression-free survival of ovarian cancer patients.** Pooled univariate hazard ratios (**A, B**) and pooled multivariate hazard ratios (**C, D**) of serum HE4, CA125, or their combination regarding progression-free survival of ovarian cancer patients are seen as Forest graphs (**A, C**) and Funnel graphs together with the Egger-test scores (**B, D**).




Regression-based Egger test for small-study effects
Random-effects model
Method: DerSimonian–Laird

H0: beta1 = 0; no small-study effects
beta1 = 1.54
SE of beta1 = 0.829
z = 1.86
Prob > |z| = 0.0629

**Figure 5: The association between serum values of HE4 and CA-125 with overall survival of ovarian cancer patients.** Pooled hazard ratios of HE4 and CA125 regarding overall survival of ovarian cancer patients are seen as Forest graphs (**A**) and Funnel graphs together with the Egger-test scores (**B, D**).

While three of the eligible studies reported hazard ratios for PFS (23,27,31) either by univariate or multivariate analyses, only two of the studies presented OS data (31,32) as shown in Figures 4 and 5. The univariate analysis of PFS suggests that serum positivity of both HE4 (PFS-UHR$_{HE4}$=3.14, CI=1.27–5.02, $I^2$=0.00 %, p=0.38) and CA-125 (PFS-UHR$_{CA-125}$=2.41, CI=1.31–3.51, $I^2$=0.00 %, p=0.93) increase the HR for PFS (Figure 4A-B). As the sample size for eligible studies was very small, heterogeneity in the studies cannot be significantly assessed through Egger's test and funnel plot analysis (Figure 4B). While the combination of the two biomarkers holds promise as an effective prognostic tool for the prediction of PFS (PFS-HR$_{HE4 + CA125}$=8.14, CI=1.18–15.11), insufficiency in the number of studies providing detailed statistical data hinders conclusive remarks (Figure 4A). Despite the limited number of eligible studies providing sufficient multivariate statistics, the multivariate analysis of PFS also suggests both biomarkers as indicators for worse PFS upon an increase in serum values (Figure 4C-D). The analysis of association between overall survival and the serum levels of HE4 and CA-125 indicate that increase in serum HE4 levels increase the risk for worse overall survival by 6.9 fold (OS-HR$_{HE4}$=6.68, CI=0.82–12.55, $I^2$=0.00 %, p=0.7), while the risk is increased 1.6 fold upon increase in serum CA-125 levels (OS-HR$_{CA-125}$=1.62, CI=0.42-2.83, $I^2$=0.00 %, p=0.45) (Figure 5).

## Discussion

Recognizing the importance of timely intervention in recurrent OC for improving patient outcomes, gynecologic oncologists strongly emphasize recurrence monitoring during follow-up. Compared to pelvic and imaging examinations which suffer from subjectivity, low accuracy, and limited ability to detect small lesions, tumor markers provide a substantial benefit in monitoring patients for detection of OC recurrence.

Despite FDA approval of the combined use of HE4 and CA-125 in post-treatment monitoring (34), HE4 is a biomarker with limited clinical adoption across European countries and worldwide (35). European Society of Gynaecological Oncology (ESGO), the European Society for Medical Oncology (ESMO), and the European Society of Pathology (ESP) recommend routine oncological follow-up including imaging and/or CA-125 according to local practice and after discussion with the patient (35). To our knowledge, this meta-analysis which pooled data from 1026 patients across thirteen studies, is the first to examine the prognostic efficacy of CA-125 and HE4 in the detection of ovarian cancer recurrence simultaneously in the same patient cohort, enabling their direct comparison with one another, in addition to evaluation of their performance as a combined prognostic tool.

Here, pooled analyses of the eligible data confirm the findings of the previous studies, indicating that an increase in serum HE4 and CA-125 during the follow-up period is associated both with poor prognosis and poor survival in ovarian cancer.

Pooled AUC analysis of CA-125, HE4, and their combination indicated comparable efficacy in detecting OC recurrence ($AUC_{HE4}$=0.78; 95% CI=0.73–0.83; $I^2$=51.73%; p=0.03; $AUC_{CA-125}$=0.80; 95% CI=0.73–0.88; $I^2$=84.80%; p<0.001; $AUC_{HE4+CA-125}$=0.76; 95% CI=0.62–0.90; $I^2$=84.37%; p<0.001 Figure 2). The high level of heterogeneity among the studies examining these biomarkers individually or as a combined prognostic tool results in large and overlapping confidence intervals, defying the prominence of HE4-alone, CA-125-alone, or their combination as a better prognostic tool outperforming the others.

Pooled analysis of sensitivity and specificity of HE4 and CA-125 measurements indicate HE4 as a biomarker with higher sensitivity ($Sensitivity_{HE4}$: 80.7 [73-88.4]; $Sensitivity_{CA-125}$: 71.4 [60.2-82.7]) while indicating CA-125 as a biomarker with higher specificity ($Specificity_{HE4}$: 77.8 [68.9-86.6]; $Specificity_{CA-125}$: 94.5 [91.9-97.1]) in detecting recurrence in OC. With its higher sensitivity, HE4 may be able to identify recurrence in patients with negative CA-125 test results. Considering these results, it might be argued that HE4 has a higher potential as a recurrence tracker during the follow-up period, utilized in the initial screening phase limiting the false-negative results, and CA-125 might prove useful as a secondary parameter acting as a recurrence validator, decreasing the false-positive rate. Due to the limited number of available studies and high heterogeneity among eligible studies, the pooled analysis of the HE4 + CA-125 combination demonstrates limited improvement from individual biomarkers in terms of sensitivity or specificity ($Sensitivity_{HE4+CA-125}$=73.35 [56.16-90.54[; $Specificity_{HE4+CA-125}$=84.39 [73.07-95.70]). Additional data from new studies examining both biomarkers in parallel in the same patient population will be required to support this improvement to suggest the combination as a better indicator in predicting OC recurrence.

While pooled HR results suggest that the risk for worse progression-free survival (PFS) was greater than 3-fold upon increasing serum HE4, and close to 2.5-fold upon increase in serum CA-125, the limiting number of the eligible studies result in large and overlapping confidence intervals, precluding comprehensive comparison of their prognostic efficacy with one another.

The heterogeneity among the studies is attributable to several factors including variation in sample size, lack of standardization in the methods and cutoff values used to measure the analytes, variation in the recurrence intervals tracked, heterogeneity in the methods used during follow-up period for validation of cancer recurrence, etc.

Cao et al demonstrated a 2.6-fold increased risk of recurrence in OC patients with higher serum HE4 levels through a subgroup meta-analysis of five studies (36). However, the authors did not conduct subgroup analyses to assess the risk associated with increased CA-125 levels. Gu et al conducted a meta-analysis of 34 studies, demonstrating pooled specificity and sensitivity values of (0.93, 95% CI: 0.89–0.95) and (0.69, 95% CI:0.65–0.72), respectively, for CA-125 (37).

By utilizing EIA with a lower cutoff point (HE4: 41 pmol/L; CA-125: 1 U/ml), Steffensen et al. (23) achieved high sensitivity (90%) for HE4 and both tumor markers in combination in the early phase of OC (the first 6 months after first-line therapy), however, this approach compromised the specificity of the tests. In addition to the primary analysis, a secondary analysis was conducted using a cutoff value determined by a 50% increase after the first-line treatment in a six-month follow-up. They found that patients with elevated HE4 levels at the 3- and 6-month follow-up points experienced significantly shorter progression-free survival compared to those with elevated CA-125 levels, as demonstrated by substantially higher hazard ratios for HE4. They suggested in summary that a >50% early increase in HE4 post-treatment strongly predicts recurrence risk.

Rong et al. analyzed tumor markers using the ECLIA method (cut-off values HE4: 70 pmol/L; Ca125: 35 U/ml) during the initial six-month postoperative period while patients were undergoing first-line chemotherapy treatment (31). While they demonstrated that HE4 had a higher sensitivity than CA-125, their reported sensitivity was lower than our meta-analysis. This difference might be attributed to their analysis being restricted to the initial six months following surgery. They claimed that HE4 is a better predictor of platinum sensitivity than CA-125. Their data revealed a maximum AUC of 0.779 (p<0.001) for HE4 alone in predicting platinum sensitivity after the third chemotherapy cycle, compared to the maximum AUC=0.731 of CA125 (p=0.004) after the first cycle. Combining both biomarkers, they found that either HE4 clearance after the third cycle or CA-125 clearance after the first cycle yielded AUC, sensitivity and specificity were 0.788, 100, and 57.5% respectively. Notably, the absence of HE4 clearance after the third cycle and CA-125 clearance after the first cycle perfectly identified all platinum-resistant patients.

While the current study represents a pioneering contribution to the field, by evaluating statistically relevant and sufficient data from the studies examining both tumor markers in the same study population for the first time, it is afflicted by the limitations present in the studies currently available in the literature. A significant limitation is attributable to the high heterogeneity among studies regarding study periods, cut-off values, patient populations, and treatment regimens. Available literature had utilized diverse methods for the assessment of tumor markers and studies using varying cut-off levels had to be combined in the pooled analyses. Variation in the cut-off levels utilized in different studies results from the fact that the analysis methods used in the studies for measurement of either of the tumor markers are not yet standardized, traceable, or harmonized. Once the measurement methods become traceable, standardized, and harmonized, the homogeneity among the studies will increase, enabling the utilization of a mutual cut-off. Another limitation was caused due to the limited number of available studies. As the number of studies examining both biomarkers in parallel in the same patient population increases, it will be possible to evaluate the prognostic efficacy of these markers based on tumor stage or type. Larger prospective studies comparing both tumor markers in the same patient population are needed.

## Conclusion

Frequent and meticulous surveillance of patients following cancer treatment is essential for early detection of recurrence. Our meta-analysis indicates comparable efficacy of both tumor markers in detecting OC recurrence, with HE4 as a feasible complementary tool in tracing OC recurrence and CA-125 as a recurrence validator. Given the substantial heterogeneity among studies, additional prospective trials assessing both biomarkers within the same patient cohort are imperative for definitive conclusions. In addition, the imperative to discover biomarkers with superior prognostic potential remains paramount. Earlier detection of cancer recurrence with higher accuracy will open possibilities for advanced therapeutic approaches.

## Declarations

### *Funding*

Not applicable

### *Conflict of Interest*

The authors state no conflict of interest.

### *Ethics Approval*

No ethical approval and patient consent were required for this study as all of the included studies had recruited patients that provided informed consent.

### *Availability of Data and Material*

The raw data can be obtained on request from the corresponding author.

### *Author Contributions*

Both authors contributed to the study conception and design, screening and selection of the studies, and data extractions. Statistical evaluation was performed by Ceyhan Ceran Serdar. The first draft of the manuscript was written by Şeyma Osmanlıoğlu, and both authors read and approved the final manuscript.

### *Acknowledgement*

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