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Identifying post-menopausal women at elevated risk for epithelial ovarian cancer

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Abstract

OBJECTIVE—We developed and validated a hybrid risk classifier combining serum markers and epidemiologic risk factors to identify post-menopausal women at elevated risk for invasive fallopian tube, primary peritoneal, and ovarian epithelial carcinoma.

METHODS—To select epidemiologic risk factors for use in the classifier, Cox proportional hazards analyses were conducted using 74,786 Women's Health Initiative (WHI) Observational Study (OS) participants. To construct a combination classifier, 210 WHI OS cases and 536 matched controls with serum marker measurements were analyzed; validation employed 143 cases and 725 matched controls from the WHI Clinical Trial (CT) with similar data.

RESULTS—Analyses identified a combination risk classifier composed of two elevated-risk groups: 1) women with CA125 or HE4 exceeding a 98% specificity threshold; and 2) women with intact fallopian tubes, prior use of menopausal hormone therapy for at least two years, and either a first degree relative with breast or ovarian cancer or a personal history of breast cancer. In the WHI OS population, it classified 13% of women as elevated risk, identifying 30% of ovarian cancers diagnosed up to 7.8 years post-enrollment (Hazard Ratio [HR]=2.6, $p<0.001$). In the WHI CT validation population, it classified 8% of women as elevated risk, identifying 31% of cancers diagnosed within 7 years of enrollment (HR=4.6, $p<0.001$).

CONCLUSION—CA125 and HE4 contributed significantly to a risk prediction classifier combining serum markers with epidemiologic risk factors. The hybrid risk classifier may be useful to identify post-menopausal women who would benefit from timely surgical intervention to prevent epithelial ovarian cancer.

Keywords

ovarian cancer; risk prediction; CA125; HE4

Introduction

We describe a hybrid risk classifier combining serum markers with epidemiologic risk factors, designed to identify post-menopausal women at elevated risk for epithelial ovarian cancer (EOC) independent of the risk associated with known mutations. The classifier could aid decision-making in post-menopausal women regarding opportunistic bilateral salpingo-oophorectomy (BSO), follow-up for EOC in symptomatic women (1), and periodic screening in asymptomatic women. We were interested in defining a clinically accessible

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way to identify subgroups of women for whom these interventions might be considered. Statistical models were used to help identify important predictors but the actual classifier is based on a simple assessment of presence or absence of selected risk factors.

In a meta-analysis of 22 studies (3), the average cumulative risks for EOC by age 70 years were 39% (18%-54%) and 11% (2.4%-19%) for *BRCA1* and *BRCA2* mutation carriers respectively. In a meta-analysis of 10 studies, risk-reducing salpingo-oophorectomy (RRSO) reduced future risk of EOC in these women by >80% (4). Among women with a significant family history (FH), including those with deleterious mutations in high penetrance cancer susceptibility genes such as *BRCA1* and *BRCA2*, RRSO reduces risk of EOC by at least 69%, as well as risk of breast cancer by at least 37% (5).

Opportunistic BSO might similarly prevent EOC in women without deleterious mutations; this is important because over 75% of EOC cases occur in these women. Many women elect BSO at the time of surgery for benign gynecologic conditions such as hysterectomy, but many others do not due to a reluctance to lose natural hormonal function. Post-menopausal women below the age of 65 may avoid BSO because of its potential association with cardiovascular disease, hip fracture, dementia and Parkinson's disease (7).

Recent evidence suggests that bilateral salpingectomy with ovarian retention (BSOR) may be an alternative to BSO for women who wish to retain ovarian function (8). The fallopian tubes, especially the native serous epithelium at the fimbria, are increasingly recognized as a site of origin of high grade serous EOC (9, 10), suggesting that bilateral salpingectomy may be both necessary and sufficient for EOC risk reduction. The addition of BSOR to hysterectomy in women who do not carry *BRCA1/2* mutations was recently reported to show no negative effects on ovarian function or perioperative complications (8, 11). Efficacy of this approach remains to be demonstrated (12).

Post-menopausal women having hysterectomy for benign conditions must choose among prophylactic BSO, prophylactic BSOR, or retention of both ovaries and fallopian tubes. Recent recommendations state that women at high risk for EOC undergo BSO at hysterectomy (13), but criteria for identifying high-risk women are not well defined. A woman with a significant FH suggesting inherited susceptibility may be considered high risk (13) in the absence of a negative mutation test in the proband from her high-risk family EOC (14, 15).

A reliable tool to assess EOC risk associated with factors other than deleterious mutations in cancer susceptibility genes, in addition to FH, is needed to inform a post-menopausal woman's decision-making regarding EOC prevention and early detection. Use of serum markers in a risk classifier is novel but is strongly supported by the literature (16) including some evidence that CA125 levels signal EOC precursor lesions (17). The serum marker component of our combination classifier relies on CA125 and human epididymis 4 (HE4) protein. CA125 is a predictive marker for EOC that becomes increasingly sensitive with proximity to diagnosis (16). HE4 similarly predicts EOC and is used clinically in women with a pelvic mass (18); it is more specific than CA125 in women with benign tumors (19). Both CA125 and HE4 show promise as risk and early detection markers (16, 20-23).

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Material and Methods

Overview

Using data from participants in the Women's Health Initiative (WHI) Observational Study (OS) and Clinical Trial (CT), we defined and validated a risk prediction classifier based on a combination of epidemiologic risk factors and serum markers. Our goal was to achieve the best sensitivity for acceptable specificity. Traditional epidemiologic risk factors (24) for which WHI data were available were considered for inclusion in the epidemiologic component of the classifier, in addition to FH based on its widespread use clinically, and use of menopausal hormone therapy (HT) based on recent reports of its association with increased risk of EOC in post-menopausal women (25-27).

We first assessed epidemiologic risk factors in univariate and multivariate Cox proportional hazards models in the WHI OS population, selecting the risk factors most associated with risk of EOC for inclusion in candidate classifiers. Serum markers CA125 and HE4 were selected for inclusion based on predictive performance reported previously (16). For ease of clinical application, each risk factor was defined as present or absent; candidate classifiers were defined using simple "and/or" combinations of the risk factors. The performance of each candidate risk classifier was evaluated in the WHI OS study populations, and subsequently validated in the WHI CT study populations in terms of (1) the percent of women later diagnosed with EOC that the classifier correctly identified as elevated risk (sensitivity); and (2) the percent of the unaffected population it erroneously classified as elevated risk (specificity). We determined statistical significance for each classifier by coding it as a yes/no, time-dependent variable and fitting a univariate Cox proportional hazards model. We report the hazard ratios and p-values from these models.

Study population

The WHI was a national prospective study of post-menopausal women's health. In total, over 161,000 women aged 50-79 were enrolled between 1993 and 1998, including 93,676 in the WHI Observational Study (OS) and 68,132 in the Clinical Trial (CT). After excluding participants reporting prior BSO at baseline, 74,786 women were eligible for these analyses from the OS and similarly 55,467 participants were eligible from the CT. Mean (maximum) follow-up at the time of these analyses was 12.3 (17.5) years for the OS and 13.2 (17.0) years for the CT; these analyses were based on a mean 12.3 years of follow-up (maximum 17.5 years). Details of the WHI design and implementation have been published (28, 29). Women in the WHI OS and CT can be assumed to be from the same reference population because they met very similar eligibility criteria, had similar data collected, and lived in the same communities; their assays were conducted with the same methods, and their blood samples were stored for similar periods.

Cancer outcomes—We define EOC as invasive ovarian, fallopian tube and primary peritoneal cancer. All incident ovarian cancers were documented and centrally reviewed at the WHI Clinical Coordinating Center according to SEER guidelines (29), including 461 cases of invasive EOC in the OS and 334 cases of invasive EOC in the CT; unconfirmed cases of EOC (n=68 in the OS, n=31 in the CT) and diagnoses of LMP or non-epithelial

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tumors of the ovary (n=53 in the OS and n=80 in the CT) were censored at time of event. Due to low mortality rates, LMP tumors are not considered invasive EOC (30-33). We excluded stromal and germ cell tumors because they also have a different biology and are seldom diagnosed in post-menopausal women; they are generally excluded in validation studies of CA125 and HE4 for early detection of EOC (21, 22).

Data collection—Information on epidemiologic risk factors was obtained from baseline self-administered questionnaires. Most items were collected in parallel in the two cohorts. Because information was limited regarding age of personal history of breast cancer and FH of breast cancer, cancer in aunts, Ashkenazi ethnicity, and lineage, significant FH was defined as any of the following conditions: Personal history of breast cancer diagnosed before age 55; one or more first-degree relatives (mother, sister, daughter) with breast cancer diagnosed before or at age 45; two breast cancers in first or second (grandmother) degree relatives, same lineage, with at least one diagnosed before or at age 45; three or more first or second degree relatives, same lineage, with breast cancer diagnosed at any age; one or more ovarian cancers diagnosed at any age in first or second degree relatives; Jewish religion and either a personal history of breast cancer, or a first or second degree relative also of Jewish religion, with breast cancer diagnosed at any age. Because EOC risk is elevated in breast cancer survivors (37), we counted a personal history of breast cancer as equivalent to a first-degree relative (FDR) with breast cancer in our definition of modest FH of breast or ovarian cancer.

Nested case-control studies—Serum markers and selected epidemiologic risk factors were evaluated in nested case-control studies performed within each cohort. In the OS, serum markers CA125 and HE4 were measured in 210 cases and 536 controls selected in 2003 from the cohort of 74,786 women described above. Cases were included if they had centrally confirmed diagnoses or locally confirmed death from EOC diagnosed before July 30, 2002, and adequate serum available from at least one blood sample for measurement of serum markers. Unconfirmed cases and confirmed LMP cases were excluded. Two controls were selected for each EOC case with matching on age at baseline (±5 years), race/ethnicity, clinical center, BSO status and minimum length of follow-up (±12 months). Average follow-up in years at the time of case-control selection was 3.4 (max: 7.8) for cases and 5.3 (max: 8.2) for controls. Serum markers were measured in blood samples obtained at enrollment and at a clinic visit 3 years later in the WHI OS.

Similarly, a nested case-control validation study was performed in the CT in 143 EOC cases and 725 controls for which CA125 and HE4 serum marker data were measured. For consistency with the OS case-control population, only cases diagnosed within 7 years of enrollment were included. Five controls, rather than two as in the OS study, were selected for each case to improve precision of specificity estimates. Controls were matched on age at baseline (±5 years), race/ethnicity, clinical center, BSO status, and trial arm (28). Control selection was based on risk-set sampling, selecting from eligible controls for each case from the risk set at the time of the case's event, resulting in the same number of blood draws for most case-control pairs. Average follow-up in years at the time of sampling was 3.4 (max:

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7.0) for cases and 12.3 (max: 16.1) for controls. Blood samples were obtained at the time of enrollment and again 1 year later in the WHI CT.

Laboratory analyses

In both OS and CT samples, CA125 and HE4 were measured on the Abbott Architect™ automated platform in a Clinical Laboratory Improvement Amendments (CLIA)-approved laboratory using FDA-approved kits. CV's for these CA125 and HE4 assays are 4% and 6% respectively.

Statistical analyses

Relevant characteristics of all study participants were reported for the eligible WHI OS and CT populations and for the WHI OS and CT case-control studies. Cases were further characterized by histology and grade for study population comparison purposes.

Markers were considered elevated if their levels were above the 98th percentile of the age-specific parametric empirical Bayes (PEB) distribution in healthy women (34-36). Using the PEB method, when blood samples were obtained both at enrollment and again at a subsequent clinic visit, the first measurement was used to tailor each marker threshold to the individual woman as previously described (36). Unaffected women in the WHI CT case-control study were used as the reference population for calculating the PEB distribution for the WHI OS case-control study, and vice versa. Blood samples obtained after diagnosis of EOC are not included in analyses.

Classifier development

Candidate risk factors were: <1 year of oral contraceptive use, nulliparity, no breastfeeding, no tubal ligation, talc use (24), prior hysterectomy and use of HT including use of estrogen alone and estrogen with progestin (25). HT was defined as use of pills or patches. Risk factors were evaluated in univariate and multivariate Cox proportional hazards models with time measured in days from enrollment with censoring at the time of death, loss to follow-up, or last visit, respectively. The multivariate model was adjusted for variables age and race. These models were not used to obtain weights (coefficients) for a classifier based on hazard ratios; rather they were used to identify risk factors most associated with EOC risk.

Using the WHI OS cohort, we first evaluated four risk classifiers using only epidemiologic risk factors, including two using only FH, one using only other epidemiologic risk factors and one using FH in combination with other epidemiologic risk factors. The first FH-alone classifier was included as a reference: it classifies as elevated-risk all women with a significant pedigree suggestive of a *BRCA1* or *BRCA2* mutation; these women are generally considered to be at high risk for EOC in the absence of a negative mutation test (13). The second FH-alone classifier classifies as elevated-risk all women with any first-degree relative with breast or ovarian cancer or a personal history of breast cancer (38); this is a simpler and more inclusive approach that is not directly related to risk for carrying a deleterious mutation. The two other classifiers using epidemiologic risk factors were developed on the basis of the results of the Cox regression analyses described above.

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We also evaluated three classifiers that included serum markers, one using serum markers alone and two hybrid classifiers that combined serum markers with epidemiologic risk factors and/or FH. The three classifiers that include use of serum markers were examined in the nested case-control population only; the four classifiers based only on epidemiologic risk factors use the entire cohort.

Results

Characteristics of women at the time of enrollment are reported in **Table 1** by study. Women in the eligible populations and in the nested case-control studies were similar with respect to age, ethnicity, parity, use of HT, tubal ligation, and hysterectomy. Women on a low fat diet and women with a personal history of breast cancer were ineligible for the CT; accordingly, more WHI OS than CT women had low BMI or reported a significant or modest relevant FH.

Histology and grade of cases included in each study are reported in **Table 2**. Serous and not otherwise specified (NOS) cases that are not well-differentiated are classified as high-grade serous (bolded). High-grade serous cancers predominate in both the WHI OS and CT cohorts, consistent with the distribution observed in the US population (39), and are the higher clinical priority.

Results of univariate Cox regression analyses (**Table 3**) suggest that in the eligible WHI OS population, prior use of HT for over 2 years, and no tubal ligation are significant predictors of EOC. In a multivariate Cox proportional hazards analysis prior use of HT for over two years was the only statistically significant risk factor, with an estimated hazard ratio (HR) of 1.50 (95% confidence interval: 1.23-1.83; $p<0.001$). Remaining candidate risk factors conferred small, non-significant increases in risk. Having a modest FH conferred a 19% increase in risk (HR=1.19; $p=0.13$) and intact fallopian tubes (no tubal ligation) had an estimated HR of 1.24 ($p=0.13$).

Based on these results, we evaluated an epidemiologic classifier combining use of HT for over 2 years with no tubal ligation. Tubal ligation was included despite its borderline statistical significance in recognition of the fact that intact fallopian tubes play a role in EOC risk, and that in the future, women may undergo BSOR. We also examined the impact of adding modest FH to the hybrid classifier, in recognition of its borderline statistical significance, its importance to women and physicians, and its associated improvement in specificity.

In the OS eligible population, a “significant FH” classifier selected 8.9% of the unaffected population as elevated risk and identified 8.5% of cases diagnosed during the follow-up period of up to 17 years; a more modest FH selected 20.4% of the unaffected population as elevated risk and identified 22.6% of cases diagnosed during follow-up (**Figure 1A**). The HR was not statistically significant for either classifier.

The classifier using other epidemiologic risk factors performed better than those using FH alone. Use of HT for over two years in women with intact fallopian tubes selected 32.5% of the unaffected population as elevated risk and identified 44.5% of cases diagnosed during

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the follow-up period (HR 1.59, p<0.001), yielding good sensitivity but poor specificity. Requiring that the woman have a modest FH, in addition to intact fallopian tubes and use of HT for over two years, improved specificity at substantial cost in sensitivity, classifying 5.8% of the unaffected population as elevated risk and identifying 8.5% of cases diagnosed during the follow-up period (HR 1.50, p=0.018).

In the OS case-control study, the classifier using serum markers alone performed better than either the FH or the other epidemiologic risk factor classifiers (**Figure 1B**). An elevated marker (CA125 or HE4) without consideration of epidemiologic risk factors selected 7.1% of the unaffected population as elevated risk and identified 23.3% of cases diagnosed up to 7.8 years post-enrollment (HR 3.19, p<0.001). Marker elevation alone yielded reasonably good sensitivity in combination with good specificity.

Sensitivity was improved by combining epidemiologic risk factors and serum markers to select women with an elevated marker or epidemiologic risk factors inclusive or exclusive of FH. The combination classifier using serum markers *or* HT use for over 2 years in women with intact fallopian tubes selected 40.5% of the unaffected population as elevated risk and identified 58.2% of cases diagnosed during the follow-up period (HR 1.81, p<0.001). Sensitivity of this classifier was good, but specificity of 60% may be inadequate for many clinical decisions. Improvement in specificity was achieved as before by restricting selection based on epidemiologic risk factors to women with modest FH.

The classifier that offered the best sensitivity for acceptable specificity in the WHI OS case-control study was selected as the “best” classifier. It selected as elevated risk: 1) women with CA125 or HE4 exceeding a 98% specificity threshold; and 2) women with intact fallopian tubes, >2 years prior use of menopausal HT, and either a first degree relative with breast or ovarian cancer or a personal history of breast cancer (modest FH). A hybrid classifier combining these two groups selected 12.6% of the unaffected population as elevated risk and identified 30.1% of cases diagnosed during the follow-up period (HR 2.60, p<0.001).

To validate the best combination classifier, the performance of all classifiers (including the best combination classifier and the serum markers only classifier) was estimated in the WHI CT study (**Figure 1).** In the WHI CT case-control study, the “best” classifier selected 8.0% of the unaffected population as elevated risk and identified 30.8% of cases diagnosed up to 7 years post-enrollment (HR 4.63, p<0.001), performing at least as well as in the WHI OS case-control study. Other classifiers performed similarly in the two populations.

The classifiers performed very similarly when the validation population was restricted to a subset that might be relevant for a specific intended use, such as non-hysterectomized women regarding decisions about opportunistic BSO. Of women with an intact uterus, 7% of the unaffected population and 34% of women diagnosed with EOC were classified as elevated risk. The classifier also performed similarly when outcome was restricted to high-grade serous disease, identifying 32% of high-grade serous cases as elevated risk. These results are reported in Supplemental Figures 1 and 2 respectively. We also considered the

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effect of adjustment for matching variables age and race in the WHI OS and CT case-controls studies. The results were unchanged.

Discussion

Lifetime risk for EOC is 1 in 70 in an unselected population; a *BRCA2* mutation carrier faces a risk of over 1 in 10. The hybrid classifier that selected women based on either 1) CA125 or HE4 exceeding a 98% specificity threshold, or 2) a family or personal history of ovarian or breast cancer, intact fallopian tubes, and prior use of menopausal HT for at least two years is associated with a hazard ratio between 4 and 5 in the validation population. While the available data do not allow us to estimate lifetime risk, the hazard ratio represents a significant increase in the instantaneous risk of EOC over the course of the study. The classifier designated 8% of post-menopausal women in the CT validation study as elevated risk, correctly predicting 31% of women later diagnosed with EOC. The classifier may be useful to identify elevated-risk women for clinical intervention when 92% specificity is tolerable.

The hybrid classifier outperformed FH alone. In the WHI eligible cohorts, neither FH nor most traditional epidemiologic risk factors were significantly associated with increased risk of EOC. Significant FH suggestive of a deleterious *BRCA1* or *BRCA2* mutation, sometimes used to select women for preventive surgery or screening in the absence of mutation testing (13, 40, 41), was not highly associated with EOC in either of these post-menopausal WHI populations. Only serum markers, use of HT for over two years and no tubal ligation were statistically significant univariate predictors of EOC. Intact fallopian tubes and a modest FH contributed some risk, but were not statistically significant in the WHI OS population.

Serum markers contributed importantly. The best specificity (96.6%) and largest hazard ratio (7.2) were obtained in the CT validation study using a classifier that depended only on serum markers, which identified 25% of cases occurring within 7 years of enrollment. However, serum markers rise most dramatically in the 3 years prior to clinical diagnosis, as opposed to germ-line mutations that confer high lifetime risk from birth. Common low penetrance ovarian cancer genetic risk variants that can be assayed using germline DNA samples might also contribute to a predictive model that includes serum markers and epidemiological risk factors (42). Women identified as elevated risk by CA125 or HE4 may actually have subclinical disease. More research is needed to characterize the fallopian tubes of women with elevated serum markers. Screening is not recommended because it has not been shown to reduce mortality (20), but prophylactic surgery in women already scheduled for abdominal or pelvic surgery is recommended in post-menopausal women. In such women, suspicious lesions may be found at BSO or BSOR; such lesions are identified in about 8% of mutation carriers at prophylactic surgery (43).

Strengths of this study include its prospective design as well as its use of two large independent well-characterized study populations with identical protocols for data collection as well as for blood sample collection, processing and storage. However, because women with prior breast cancer were not eligible for the CT, it was necessary to combine personal with FH of breast cancer; non-significant hazard ratios for these variables in the WHI OS

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were 1.07 and 1.19 respectively. Other limitations include absence of data on *BRCA1* and *BRCA2* mutation status and Ashkenazi Jewish heritage as well as nested case-control subsets that include a modest number of cases for which marker levels were measured. It should be noted that the sampling for the OS case-control study was performed several years before the CT case-control study. For both, follow-up time and case-control designation reflect conditions at the time of sampling. Analyses were also performed in which controls that went on to develop cancer were censored, using follow up time from latest data release; results were similar and conclusions were not affected (data not shown). Also note that marker elevation thresholds are age-specific and tailored to the individual woman; age-specific thresholds on the original scale have been published (37).

Our observed association between HT and EOC diagnosis suggests that HT exposure may drive disease progression or otherwise contribute to EOC diagnosis in post-menopausal women. All WHI participants were post-menopausal and most were over 60 years of age. Postmenopausal EOC may have a different biology from either pre-menopausal or *BRCA* mutation-driven EOC. More research is needed to elucidate the relationship between HT use and post-menopausal EOC, and prospective validation of the risk classifier is needed to better understand how it might contribute to better outcomes for women at risk of developing EOC.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Appendix 1: Short List of WHI Investigators

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References

1. Andersen MR, Goff BA, Lowe KA, Scholler N, Bergan L, Drescher CW, et al. Use of a Symptom Index, CA125, and HE4 to predict ovarian cancer. Gynecol Oncol. 2010; 116(3):378–83. Epub 2009/12/01. [PubMed: 19945742]
2. Ramus SJ, Gayther SA. The contribution of BRCA1 and BRCA2 to ovarian cancer. Mol Oncol. 2009; 3(2):138–50. Epub 2009/04/23. [PubMed: 19383375]
3. Antoniou A, Pharoah P, Narod S, et al. Average risk of breast and ovarian cancer associated with BRCA1 or BRCA2 mutations detected in case series unselected for family history: a combined analysis of 22 studies. Am J Hum Genet. 2003; 72(5):1117–30. [PubMed: 12677558]
4. Rebbeck TR, Kauff ND, Domchek SM. Meta-analysis of risk reduction estimates associated with risk-reducing salpingo-oophorectomy in BRCA1 or BRCA2 mutation carriers. J Natl Cancer Inst. 2009; 101(2):80–7. Epub 2009/01/15. [PubMed: 19141781]
5. Nelson, HD.; Fu, R.; Goddard, K.; Mitchell, JP.; Okinaka-Hu, L.; Pappas, M., et al. Risk Assessment, Genetic Counseling, and Genetic Testing for BRCA-Related Cancer: Systematic Review to Update the US Preventive Services Task Force Recommendation. Rockville (MD): 2013.
6. Plusquin C, Fastrez M, Vandromme J, Rozenberg S. Determinants of the decision to perform prophylactic oophorectomy in association with a hysterectomy for a benign condition. Maturitas. 2012; 73(2):164–6. Epub 2012/09/12. [PubMed: 22964073]
7. Parker WH, Shoupe D, Broder MS, Liu Z, Farquhar C, Berek JS. Elective oophorectomy in the gynecological patient: when is it desirable? Curr Opin Obstet Gynecol. 2007; 19(4):350–4. Epub 2007/07/13. [PubMed: 17625417]
8. McAlpine JN, Hanley GE, Woo MM, Tone AA, Rozenberg N, Swenerton KD, et al. Opportunistic Salpingectomy: Uptake, Risks and Complications of a Regional Initiative for Ovarian Cancer Prevention. Am J Obstet Gynecol. 2014 Epub 2014/01/15.
9. Folkins AK, Jarboe EA, Roh MH, Crum CP. Precursors to pelvic serous carcinoma and their clinical implications. Gynecol Oncol. 2009; 113(3):391–6. Epub 2009/02/25. [PubMed: 19237187]
10. Dietl J, Wischhusen J, Hausler SF. The post-reproductive Fallopian tube: better removed? Hum Reprod. 2011; 26(11):2918–24. Epub 2011/08/19. [PubMed: 21849300]
11. Morelli M, Venturella R, Mocciaro R, Di Cello A, Rania E, Lico D, et al. Prophylactic salpingectomy in premenopausal low-risk women for ovarian cancer: Primum non nocere. Gynecol Oncol. 2013 pii: S0090-8258(13):00178-9. Epub 2013/04/06.
12. Gilks CB, Miller D. Opportunistic salpingectomy for women at low risk for development of ovarian carcinoma: The time has come. Gynecol Oncol. 2013; 129(3):443–4. Epub 2013/05/15. [PubMed: 23664100]
13. Berek JS, Chalas E, Edelson M, Moore DH, Burke WM, Cliby WA, et al. Prophylactic and risk-reducing bilateral salpingo-oophorectomy: recommendations based on risk of ovarian cancer. Obstet Gynecol. 2010; 116(3):733–43. Epub 2010/08/25. [PubMed: 20733460]
14. Kauff ND, Mitra N, Robson ME, Hurley KE, Chuai S, Goldfrank D, et al. Risk of Ovarian Cancer in BRCA1 and BRCA2 Mutation-Negative Hereditary Breast Cancer Families. J Natl Cancer Inst. 2005; 97(18):1382–4. [PubMed: 16174860]
15. Metcalfe KA, Finch A, Poll A, Horsman D, Kim-Sing C, Scott J, et al. Breast cancer risks in women with a family history of breast or ovarian cancer who have tested negative for a BRCA1 or BRCA2 mutation. Br J Cancer. 2009; 100(2):421–5. Epub 2008/12/18. [PubMed: 19088722]
16. Anderson GL, McIntosh MW, Wu L, Barnett M, Goodman G, Thorpe JD, et al. Assessing Lead Time of Selected Ovarian Cancer Biomarkers: A Nested Case–Control Study. J Natl Cancer Inst. 2010; 102(1):26–38. [PubMed: 20042715]

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17. Hermsen BBJ, von Mensdorff-Pouilly S, Berkhof J, van Diest PJ, Gille JJP, Menko FH, et al. Serum CA-125 in Relation to Adnexal Dysplasia and Cancer in Women at Hereditary High Risk of Ovarian Cancer. J Clin Oncol. 2007; 25(11):1383–9. [PubMed: 17416858]
18. Karlsen MA, Sandhu N, Hogdall C, Christensen IJ, Nedergaard L, Lundvall L, et al. Evaluation of HE4, CA125, risk of ovarian malignancy algorithm (ROMA) and risk of malignancy index (RMI) as diagnostic tools of epithelial ovarian cancer in patients with a pelvic mass. Gynecol Oncol. 2012; 127(2):379–83. Epub 2012/07/28. [PubMed: 22835718]
19. Moore RG, Miller MC, Steinhoff MM, Skates SJ, Lu KH, Lambert-Messerlian G, et al. Serum HE4 levels are less frequently elevated than CA125 in women with benign gynecologic disorders. Am J Obstet Gynecol. 2012; 206(4):351. e1-8. Epub 2012/01/31. [PubMed: 22284961]
20. Buys SS, Partridge E, Black A, Johnson CJ, Lamerato L, Isaacs C, et al. Effect of Screening on Ovarian Cancer Mortality: The Prostate, Lung, Colorectal and Ovarian (PLCO) Cancer Screening Randomized Controlled Trial. JAMA. 2011; 305(22):2295–303. [PubMed: 21642681]
21. Cramer DW, Bast RC Jr, Berg CD, Diamandis EP, Godwin AK, Hartge P, et al. Ovarian Cancer Biomarker Performance in Prostate, Lung, Colorectal, and Ovarian Cancer Screening Trial Specimens. Cancer Prev Res (Phila). 2011; 4(3):365–74. [PubMed: 21372036]
22. Zhu CS, Pinsky PF, Cramer DW, Ransohoff DF, Hartge P, Pfeiffer RM, et al. A Framework for Evaluating Biomarkers for Early Detection: Validation of Biomarker Panels for Ovarian Cancer. Cancer Prev Res (Phila). 2011; 4(3):375–83. For the PLCO Project Team. [PubMed: 21372037]
23. Urban N, Thorpe JD, Bergan LA, Forrest RM, Kampani AV, Scholler N, et al. Potential role of HE4 in multimodal screening for epithelial ovarian cancer. J Natl Cancer Inst. 2011; 103(21): 1630–4. Epub 2011/09/16. [PubMed: 21917606]
24. Vitonis AF, Titus-Ernstoff L, Cramer DW. Assessing ovarian cancer risk when considering elective oophorectomy at the time of hysterectomy. Obstet Gynecol. 2011; 117(5):1042–50. Epub 2011/04/08. [PubMed: 21471855]
25. Morch LS, Lokkegaard E, Andreasen AH, Kjaer SK, Lidegaard O. Hormone therapy and different ovarian cancers: a national cohort study. Am J Epidemiol. 2012; 175(12):1234–42. Epub 2012/04/21. [PubMed: 22517811]
26. Koskela-Niska V, Lyytinen H, Riska A, Pukkala E, Ylikorkala O. Ovarian cancer risk in postmenopausal women using estradiol-progestin therapy - a nationwide study. Climacteric : the journal of the International Menopause Society. 2013; 16(1):48–53. Epub 2012/05/30. [PubMed: 22640598]
27. Koskela-Niska V, Pukkala E, Lyytinen H, Ylikorkala O, Dyba T. Effect of various forms of postmenopausal hormone therapy on the risk of ovarian cancer--a population-based case control study from Finland. Int J Cancer. 2013; 133(7):1680–8. Epub 2013/03/26. [PubMed: 23526244]
28. Anderson GL. Implementation of the Women's Health Initiative Study Design. Ann Epidemiol. 2003; 13(9 Suppl):S5–17. [PubMed: 14575938]
29. Curb JD, McTiernan A, Heckbert SR, Kooperberg C, Stanford J, Nevitt M, et al. Outcomes ascertainment and adjudication methods in the Women's Health Initiative. Ann Epidemiol. 2003; 13(9 Suppl):S122–8. Epub 2003/10/25. [PubMed: 14575944]
30. Morris CR, Liu L, Rodriguez AO, Cress RD, Snipes K. Epidemiologic features of borderline ovarian tumors in California: a population-based study. Cancer Causes Control. 2013; 24(4):665–74. Epub 2013/01/15. [PubMed: 23314731]
31. Kurman RJ, Trimble CL. The behavior of serous tumors of low malignant potential: are they ever malignant? Int J Gynecol Pathol. 1993; 12(2):120–7. Epub 1993/04/01. [PubMed: 8463035]
32. Leake JF, Currie JL, Rosenshein NB, Woodruff JD. Long-term follow-up of serous ovarian tumors of low malignant potential. Gynecol Oncol. 1992; 47(2):150–8. Epub 1992/11/01. [PubMed: 1468692]
33. Sherman ME, Mink PJ, Curtis R, Cote TR, Brooks S, Hartge P, et al. Survival among women with borderline ovarian tumors and ovarian carcinoma: a population-based analysis. Cancer. 2004; 100(5):1045–52. Epub 2004/02/26. [PubMed: 14983501]
34. McIntosh M, Urban N. A parametric empirical Bayes method for cancer screening using longitudinal observations of a biomarker. Biostatistics. 2003; 4(1):27–40. [PubMed: 12925328]

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35. McIntosh M, Urban N, Karlan B. Generating Longitudinal Screening Algorithms Using Novel Biomarkers for Disease. Cancer Epidemiol Biomarkers Prev. 2002; 11(2):159–66. [PubMed: 11867503]

36. Urban N, Thorpe JD, Karlan BY, McIntosh MW, Palomares MR, Daly MB, et al. Interpretation of single and serial measures of HE4 and CA125 in asymptomatic women at high risk for ovarian cancer. Cancer Epidemiol Biomarkers Prev. 2012; 21(11):2087–94. Epub 2012/09/07. [PubMed: 22962406]

37. Schonfeld SJ, Berrington de Gonzalez A, Visvanathan K, Pfeiffer RM, Anderson WF. Declining second primary ovarian cancer after first primary breast cancer. J Clin Oncol. 2013; 31(6):738–43. Epub 2013/01/04. [PubMed: 23284037]

38. Obermair A, Youlden DR, Baade PD, Janda M. The impact of risk-reducing hysterectomy and bilateral salpingo-oophorectomy on survival in patients with a history of breast cancer--a population-based data linkage study. Int J Cancer. 2014; 134(9):2211–22. Epub 2013/10/16. [PubMed: 24127248]

39. SEER Cancer Statistics Review, 1975-2011, National Cancer Institute. Bethesda, MD, http://seer.cancer.gov/csr/1975_2011/browse_csr.php?sectionSEL=21&pageSEL=sect_21_table.16.html, 2015. [cited 2015 4/7/15]

40. Piver MS. Prophylactic Oophorectomy: Reducing the U.S. Death Rate from Epithelial Ovarian Cancer. A Continuing Debate. Oncologist. 1996; 1(5):326–30. Epub 1996/01/01. [PubMed: 10388011]

41. Rosenthal AN, Fraser L, Manchanda R, Badman P, Philpott S, Mozersky J, et al. Results of annual screening in phase I of the United Kingdom familial ovarian cancer screening study highlight the need for strict adherence to screening schedule. J Clin Oncol. 2013; 31(1):49–57. Epub 2012/12/06. [PubMed: 23213100]

42. Pearce CL, Stram DO, Ness RB, Stram DA, Roman LD, Templeman C, et al. Population distribution of lifetime risk of ovarian cancer in the United States. Cancer Epidemiol Biomarkers Prev. 2015; 24(4):671–6. Epub 2015/01/28. [PubMed: 25623732]

43. Powell CB, Swisher EM, Cass I, McLennan J, Norquist B, Garcia RL, et al. Long term follow up of BRCA1 and BRCA2 mutation carriers with unsuspected neoplasia identified at risk reducing salpingo-oophorectomy. Gynecol Oncol. 2013; 129(2):364–71. Epub 2013/02/09. [PubMed: 23391663]

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Highlights

- We define and validate an ovarian cancer risk classifier for post-menopausal women using data from the Women's Health Initiative study.
- Serum markers and epidemiologic factors classified 13% of women as elevated risk and identified 30% of ovarian cancers (HR=2.6, p-value<0.001).

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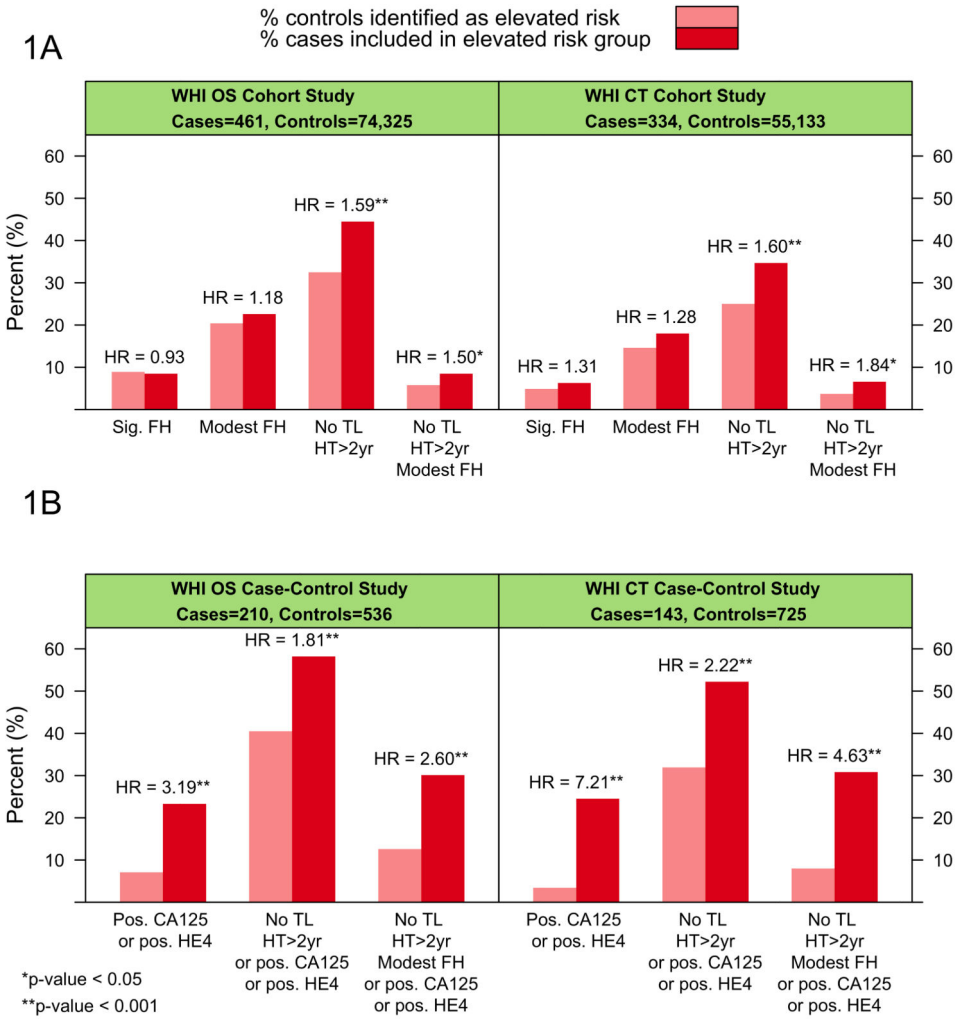

Figure 1.
Performance and validation of candidate risk classifiers

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Table 1

Characteristics of women included in analyses by study population

	WHI OS eligible population*	WHI OS Case-Control study population	WHI CT eligible population*	WHI CT Case-Control study population
N	74786	746	55467	868
Cases	461 (0.62%)	210 (28.2%)	334 (0.6%)	143 (16.5%)
Controls	74325 (99.38%)	536 (71.8%)	55133 (99.4%)	725 (83.5%)
Age at baseline, y				
49-59	24238 (32.4%)	182 (24.4%)	19772 (35.6%)	261 (30.1%)
60-69	32611 (43.6%)	370 (49.6%)	25292 (45.6%)	404 (46.5%)
70-81	17937 (24.0%)	194 (26%)	10403 (18.8%)	203 (23.4%)
Race/ethnicity				
American Indian	336 (0.4%)	3 (0.4%)	223 (0.4%)	1 (0.1%)
Asian/Pac Islander	2147 (2.9%)	14 (1.9%)	1231 (2.2%)	10 (1.2%)
African-American	5942 (7.9%)	32 (4.3%)	5497 (9.9%)	32 (3.7%)
Hispanic	2975 (4%)	20 (2.7%)	2398 (4.3%)	12 (1.4%)
White	62336 (83.4%)	677 (90.8%)	45359 (81.8%)	811 (93.6%)
Other	835 (1.1%)	0 (0%)	636 (1.1%)	0 (0%)
Missing	215 (0.3%)	0 (0%)	123 (0.2%)	0 (0%)
BMI (kg/m2)				
< 25	30716 (41.6%)	303 (40.9%)	15529 (28.1%)	230 (26.5%)
25-30	24986 (33.8%)	264 (35.6%)	19719 (35.7%)	339 (39.1%)
30+	18172 (24.6%)	174 (23.5%)	19955 (36.1%)	297 (34.4%)
Jewish Religion				
No	68916 (92.2%)	691 (92.7%)	N/A	N/A
Yes	5812 (7.8%)	54 (7.3%)	N/A	N/A
Significant Family History suggestive of *BRCA1/2* Mutation**				
No	68169 (91.2%)	681 (91.3%)	52764 (95.1%)	819 (94.4%)
Yes	6617 (8.8%)	65 (8.7%)	2703 (4.9%)	49 (5.6%)
Modest Family History of Breast and/or Ovarian Cancer***				
No	59510 (79.6%)	590 (79.1%)	47348 (85.4%)	720 (82.9%)
Yes	15276 (20.4%)	156 (20.9%)	8119 (14.6%)	148 (17.1%)
Parity				
1+ live births	65325 (87.4%)	652 (87.4%)	49690 (89.6%)	778 (89.6%)
Never pregnant/no term pregnancy	9381 (12.6%)	94 (12.6%)	5754 (10.4%)	90 (10.4%)
Breast Feeding				
Never breast fed > 28 days	39021 (52.2%)	398 (53.4%)	29525 (53.3%)	464 (53.5%)
Breast fed >28 days	35685 (47.8%)	348 (46.6%)	25919 (46.7%)	404 (46.5%)
OC use				
Never	44463 (59.5%)	471 (63.1%)	31095 (56.1%)	502 (57.8%)

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	WHI OS eligible population*	WHI OS Case-Control study population	WHI CT eligible population*	WHI CT Case-Control study population
<1 year	9332 (12.5%)	94 (12.6%)	7334 (13.2%)	130 (15%)
1-5 years	9795 (13.1%)	83 (11.1%)	7696 (13.9%)	112 (12.9%)
5-10 years	6703 (9.0%)	52 (7.0%)	5643 (10.2%)	67 (7.7%)
10+ years	4473 (6.0%)	46 (6.2%)	3682 (6.6%)	57 (6.6%)
Tubal ligation				
Yes	13342 (17.9%)	107 (14.3%)	10541 (19%)	137 (15.8%)
No	61364 (82.1%)	639 (85.7%)	44903 (81%)	731 (84.2%)
Talc use				
<10 years	57544 (77.0%)	587 (78.8%)	N/A	N/A
>10 years	17184 (23.0%)	158 (21.2%)	N/A	N/A
Prior oral hormone therapy (HT) use (any type)				
None	34303 (45.9%)	311 (41.7%)	29992 (54.1%)	418 (48.2%)
(0-1] year	6336 (8.5%)	60 (8.0%)	5202 (9.4%)	83 (9.6%)
(1-2] years	3947 (5.3%)	42 (5.6%)	2866 (5.2%)	50 (5.8%)
(2-5] years	8473 (11.3%)	75 (10.1%)	5590 (10.1%)	88 (10.1%)
(5-10] years	9801 (13.1%)	97 (13.0%)	5717 (10.3%)	99 (11.4%)
10+ years	11926 (15.9%)	161 (21.6%)	6100 (11%)	130 (15%)
Prior Hysterectomy				
No	54074 (72.4%)	535 (71.8%)	39449 (71.1%)	647 (74.5%)
Yes	20639 (27.6%)	210 (28.2%)	16016 (28.9%)	221 (25.5%)
Elevated CA125 or HE4****				
No	N/A	659 (88.3%)	N/A	807 (93%)
Yes	N/A	87 (11.7%)	N/A	61 (7%)

* Women reporting BSO at baseline as well as LMP ovarian cancer and non-adjudicated ovarian cancer cases were excluded.

** Significant family history defined as: Personal history of breast cancer diagnosed before age 55; One or more first-degree relatives (mother, sister, daughter) with breast cancer diagnosed before or at age 45; Two breast cancers in first or second degree (grandmother) relatives, same lineage, with at least one breast cancer diagnosed before or at age 45 *(Note that WHI data include only grandmother not aunt)*; Three or more first or second degree relatives, same lineage, with breast cancer diagnosed at any age; One or more ovarian cancers diagnosed at any age in first or second degree relatives; Jewish religion and personal history of breast cancer diagnosed at any age (*Note that WHI data do not include information on Ashkenazi ethnicity);* Jewish religion and one first or second degree relative with breast cancer diagnosed at any age in the same lineage (*Note that WHI data do not provide perfect specification of lineage).*

*** Any first-degree relative with a history of breast or ovarian cancer *or* personal history of breast cancer

**** Elevated CA125 or HE4 based on 98% specificity cutoffs estimated using the parametric empirical Bayes method on serum marker data from the WHI OS/CT.

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Table 2

Tumor characteristics for 210 WHI OS Case-Control Study EOC cases, 461 WHI OS Cohort Study EOC cases, 143 WHI CT Case-Control Study EOC cases, and 334 WHI CT Cohort Study EOC cases included in analyses (High-grade serous cancers shown in bold).

WHI OS Case-Control Study	**Clear Cell**	**Endometrioid**	**Mucinous**	**Serous**	**NOS**	**Other**
Well Differentiated	0 (0%)	0 (0%)	5 (38.5%)	5 (4.3%)	1 (2.4%)	0 (0%)
Moderately Differentiated	1 (7.7%)	3 (17.7%)	1 (7.7%)	**18 (15.5%)**	**2 (4.9%)**	**0 (0%)**
Poorly Differentiated	5 (38.5%)	13 (76.5%)	4 (30.8%)	**79 (68.1%)**	**17 (41.5%)**	**7 (70%)**
Unknown	7 (53.9%)	1 (5.9%)	3 (23.1%)	**14 (12.1%)**	**21 (51.2%)**	**3 (30%)**
WHI OS Cohort Study	**Clear Cell**	**Endometrioid**	**Mucinous**	**Serous**	**NOS**	**Other**
Well Differentiated	0 (0%)	2 (5.7%)	7 (33.3%)	7 (2.5%)	1 (1.2%)	0 (0%)
Moderately Differentiated	1 (5%)	13 (37.1%)	2 (9.5%)	**39 (14.1%)**	**3 (3.5%)**	**1 (4.3%)**
Poorly Differentiated	9 (45%)	19 (54.3%)	6 (23.8%)	**190 (68.8%)**	**29 (34.1%)**	**15 (65.2%)**
Unknown	11 (50%)	1 (2.9%)	6 (23.8%)	**40 (14.5%)**	**52 (61.2%)**	**7 (30.4%)**
WHI CT Case-Control Study	**Clear Cell**	**Endometrioid**	**Mucinous**	**Serous**	**NOS**	**Other**
Well Differentiated	0 (0%)	3 (17%)	0 (0%)	1 (1%)	0 (0%)	0 (0%)
Moderately Differentiated	0 (0%)	7 (39%)	2 (67%)	**13 (16%)**	**1 (3%)**	**0 (0%)**
Poorly Differentiated	4 (57%)	7 (39%)	1 (33%)	**49 (60%)**	**19 (63%)**	**4 (100%)**
Unknown	3 (43%)	1 (6%)	0 (0%)	**18 (22%)**	**10 (33%)**	**0 (0%)**
WHI CT Cohort Study	**Clear Cell**	**Endometrioid**	**Mucinous**	**Serous**	**NOS**	**Other**
Well Differentiated	0 (0%)	4 (11%)	0 (0%)	2 (1%)	0 (0%)	0 (0%)
Moderately Differentiated	2 (14%)	12 (33%)	4 (44%)	**24 (12%)**	**4 (6%)**	**1 (92%)**
Poorly Differentiated	6 (43%)	16 (44%)	3 (33%)	**133 (68%)**	**38 (56%)**	**11 (8%)**
Unknown	6 (43%)	4 (11%)	2 (22%)	**36 (18%)**	**26 (38%)**	**0 (0%)**

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Table 3

Hazard ratios, 95% confidence intervals and associated p-values from Cox proportional hazards models applied to the WHI OS Cohort Study population; 461 EOC cases and 74,325 controls (risk factors measured at the time of enrollment)

	Univariate Analysis			Multivariate Analysis***		
Risk Factors (WHI OS population)	**Hazard Ratio**	**95% CI**	**P-value**	**Hazard Ratio**	**95% CI**	**P-value**
OC use < 1 year	1.19	(0.97, 1.46)	0.101	1.12	(0.89, 1.39)	0.336
Nulliparity	1.10	(0.83, 1.44)	0.512	1.12	(0.82, 1.52)	0.478
Breast Feeding <28 days	0.99	(0.82, 1.19)	0.883	0.97	(0.79, 1.19)	0.768
No tubal ligation	**1.35**	**(1.04, 1.77)**	**0.027**	1.24	(0.94, 1.65)	0.134
Talc use >10 years	0.99	(0.79, 1.24)	0.914	0.97	(0.78, 1.22)	0.822
Prior Hysterectomy	1.16	(0.95, 1.42)	0.157	1.08	(0.87, 1.34)	0.487
Prior HT use <1 year (Ref=No HT use)	0.94	(0.63, 1.39)	0.740	N/A	N/A	N/A
Prior HT use 1-2 years (Ref=No HT use)	0.96	(0.60, 1.55)	0.880	N/A	N/A	N/A
Prior HT use 2-5 years (Ref=No HT use)	**1.37**	**(1.02, 1.85)**	**0.039**	**1.50** ^	**(1.23, 1.83)**	**<0.001**
Prior HT use 5-10 years (Ref=No HT use)	**1.37**	**(1.03, 1.82)**	**0.028**			
Prior HT use >10 years (Ref=No HT use)	**1.74**	**(1.36, 2.23)**	**<0.001**			
Significant Family History suggestive of *BRCA1/2* Mutation*	0.93	(0.66, 1.30)	0.665	N/A	N/A	N/A
Modest Family History of Breast and/or Ovarian Cancer**	1.18	(0.95, 1.48)	0.140	1.19	(0.95, 1.50)	0.126

* Significant family history defined as: Personal history of breast cancer diagnosed before age 55; One or more first-degree relatives with breast cancer diagnosed before or at age 45; Two breast cancers in first or second degree (grandmother) relatives, same lineage, with at least one diagnosed before or at age 45 *(Note that WHI data include only grandmother not aunt)*; Three or more first or second degree relatives, same lineage, with breast cancer diagnosed at any age; One or more ovarian cancers diagnosed at any age in first or second degree relatives; Jewish religion and personal history of breast cancer diagnosed at any age (*Note that WHI data do not include information on Ashkenazi ethnicity);* Jewish religion and one first or second degree relative with breast cancer diagnosed at any age in the same lineage (*Note that WHI data do not provide perfect specification of lineage).*

** Any first-degree relative with a history of breast or ovarian cancer *or* personal history of breast cancer

*** Adjusted for age at baseline (continuous) and race (white/non-white)

^ In the multivariate model, Prior HT was recoded to > 2 years of exposure (yes/no) for parsimony, due to the similarity of the point estimates for the corresponding subintervals in the univariate model.