Post-menopausal hormone therapy and colorectal cancer risk by molecularly-defined subtypes and tumor location

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Abbreviations:

BMI, body mass index

CCFR, Colon Cancer Family Registry

CPS-II, Cancer Prevention Study II

CRC, colorectal cancer

CI, confidence interval

CIMP, CpG island methylator phenotype

DACHS, Darmkrebs: Chancen der Verhütung durch Screening Study

DALS, Diet, Activity and Lifestyle Study

EPIC, European Prospective Investigation into Cancer

GECCO, Genetics of Epidemiology of Colorectal Cancer Consortium

HT, post-menopausal hormone therapy

IHC, immunohistochemistry

MCCS, Melbourne Collaborative Cohort Study

MSI, microsatellite instability; -H, high; -L, low

MSS, microsatellite stable

NHS, Nurses' Health Study

NSHDS, Northern Sweden Health and Disease Study

OR, odds ratio

PCR, polymerase chain reaction

PMR, percent of methylated reference

ABSTRACT

Background: Post-menopausal hormone therapy (HT) is associated with a decreased colorectal cancer (CRC) risk. As CRC is a heterogeneous disease, we evaluated whether the association of HT and CRC differs across etiologically-relevant, molecularly-defined tumor subtypes and tumor location.

Methods: We pooled data on tumor subtypes (microsatellite instability (MSI) status, CpG island methylator phenotype (CIMP) status, *BRAF* and *KRAS* mutations, pathway: adenoma-carcinoma, alternate, serrated), tumor location (proximal colon, distal colon, rectum), and HT use among 8,220 post-menopausal women (3,898 cases and 4,322 controls) from eight observational studies. We used multinomial logistic regression to estimate odds ratios (OR) and 95% confidence intervals (CI) for the association of ever versus never HT use with each tumor subtype compared with controls. Models were adjusted for study, age, body mass index, smoking status, and CRC family history. All statistical tests were two-sided.

Results: Among post-menopausal women, ever HT use was associated with a 38% reduction in overall CRC risk (OR[95%CI]=0.62[0.56–0.69]). This association was similar according to MSI, CIMP, *BRAF*, or *KRAS* status. However, the association was attenuated for tumors arising through the serrated pathway (0.81[0.66–1.01]) compared with the adenoma-carcinoma pathway (0.63[0.55–0.73]; P_{het}=.04) and alternate pathway (0.61[0.51–0.72]). Additionally, proximal colon tumors had a weaker association (0.71[0.62–0.80]) compared with rectal (0.54[0.46–0.63]) and distal colon (0.57[0.49–0.66]; P_{het}=.01) tumors.

Conclusions: We observed a strong inverse association between HT use and overall CRC risk, which may predominantly reflect a benefit of HT use for tumors arising through the adenoma-

carcinoma and alternate pathways as well as distal colon and rectal tumors.

Colorectal cancer (CRC) is a heterogeneous disease that evolves through multiple pathways defined by genetic and epigenetic events.[1, 2] Four tumor markers have been commonly used to better characterize this heterogeneity: microsatellite instability (MSI), CpG island methylator phenotype (CIMP), somatic mutations in *BRAF*, and somatic mutations in *KRAS*. Together, these tumor markers approximate three distinct molecular pathways of colorectal carcinogenesis: adenoma-carcinoma ("traditional"), "alternate," and serrated.[1, 3, 4] These pathways are established early in disease pathogenesis and can be identified within precancerous lesions by microscopy.[3, 5-8] Research has shown that these tumor types have distinct appearances, predilections for locations within the colon, and biologic behaviors.[8-10] As such, it is plausible that the epidemiologic factors underlying their etiologies could also differ.

Multiple lines of evidence, including randomized controlled trials, show that post-menopausal hormone therapy (HT) is associated with a decreased risk of CRC.[11-20] The reduction in risk, about 20-40% in recent analyses, has been observed in users of estrogen alone as well as combined estrogen plus progestin. Few studies have evaluated whether the association of HT use and CRC risk differs by molecularly defined CRC subtypes; however, such information might increase the understanding of the mechanisms for this beneficial effect. Current literature suggests that HT use is associated with a lower risk of MSI-low or -stable tumors (MSI-L/MSS) and possibly with a lower risk of CIMP-negative and *BRAF*-wildtype tumors.[21, 22] HT use has only been associated with *KRAS*-wildtype tumors in the distal colon in one previous study.[23] Regarding tumor location, the association of HT use and CRC is reportedly stronger among tumors of the distal colon compared with the proximal colon.[22, 24]

To our knowledge, no study has evaluated both tumor markers and location in relation to HT use in order to provide a comprehensive understanding of subtype-specific CRC risk.

In this study, we examined HT use in relation to molecularly-defined CRC subtypes using available data from the Colon Cancer Family Registry (CCFR)[21, 25, 26] and seven studies contributing to the Genetics of Epidemiology of Colorectal Cancer Consortium (GECCO).[27, 28] Specifically, we evaluated each of the four common tumor markers (MSI, CIMP, *BRAF*, and *KRAS*) separately, as well as three pathways of carcinogenesis defined by combinations of those markers and tumor location.

METHODS

Study Populations

Data from eight observational studies of CRC were pooled: the Cancer Prevention Study-II (CPS-II),[29] the German Darmkrebs: Chancen der Verhutung durch Screening Study (DACHS),[30, 31] the Diet Activity and Lifestyle Study (DALS),[32] the Swedish population of the European Prospective Investigation into Cancer (EPIC),[33] the Melbourne Collaborative Cohort Study (MCCS),[34] the Nurses' Health Study (NHS),[35, 36] the Northern Sweden Health and Disease Study (NSHDS),[37] and three population-based sites from the Colon Cancer Family Registry.[21, 25, 26] Each study included women diagnosed with incident invasive CRC and contemporaneous unaffected controls. Only women with tumor marker data were eligible for inclusion in this analysis. Study-specific details are described in the Supplementary Methods. All participants provided informed consent for participating in this research, and studies were approved by their respective Institutional Review Boards.

Data collection and harmonization

The harmonization procedure and ascertainment of HT use are described in more detail in the Supplementary Methods. Information on demographics and environmental risk factors was collected via telephone or in-person interviews and/or structured self-completed questionnaires.[24, 38, 39] HT use was generally ascertained as any self-reported use at baseline survey. Additionally, ever use of formulation-specific (estrogen-only or estrogen plus progestin) HT use was derived from three studies (CCFR, CPSII, and NHS). HT non-users at reference time were used as the comparison group. Post-menopausal status was harmonized as either 1) study-derived menopausal status, if available; 2) self-reported menopausal status, if study-derived data were not available; or 3) age $\geq$55, if study-derived and self-report data were not available.[40]

Tumor characteristics and molecular subtyping

Tumor marker testing was conducted using DNA extracted from formalin-fixed paraffin-embedded tumor tissue specimens. Individual study protocols varied, as outlined below and further detailed in the Supplementary Methods.

MSI testing was primarily conducted using polymerase chain reaction (PCR) following the National Cancer Institute Bethesda Consensus Panel (CCFR, CPS-II, MCCS, NHS).[41] Typically, $\geq$4 interpretable markers were required to classify tumors, with some variation across studies outlined in Supplementary Table 1. Additional methods used include immunohistochemistry (NSHDS, EPIC and a subset of CCFR and MCCS) and mononucleotide marker panels (DACHS, DALS) (Supplementary Methods). Tumors were classified as MSI-high (MSI-H) if $\geq$30% of the markers showed instability and MSI-low/microsatellite stable (MSI-

L/MSS) if <30% of the makers showed instability. MSI status could be determined for 3,639 CRC cases (93.4%).

Most studies used Methylight[42] methylation analysis to determine CIMP status, classified as positive or negative based on either an eight- (CPS-II, EPIC, NHS, NSHDS)[43, 44] or five-gene (CCFR, MCCS)[45-47] panel. The percent of methylated reference (PMR) value was calculated to determine whether each gene was positive for methylation (generally PMR>10). DACHS used a different five-gene panel[48, 49] to determine CIMP status, and based methylation on the presence or absence of the methylation-specific PCR product. DALS[50] determined CIMP status using a classic panel of CpG islands.[51, 52] Specific genes included in each panel, details of calling methylation status, and number of methylated genes present to classify a tumor as CIMP-positive are outlined in Supplementary Table 2. CIMP status could be determined for 3,453 CRC cases (88.6%).

Studies used PCR, sequencing, and immunohistochemistry (IHC) techniques to assess *BRAF* and *KRAS* mutations, as detailed in the Supplementary Methods. The majority of studies evaluated *BRAF* via V600E mutations in exon 15 and *KRAS* via mutations in codons 12 and 13, though a few evaluated additional loci. *BRAF* and *KRAS* status could be determined for 3,564 (91.4%) and 3,435 (88.1%) CRC cases, respectively.

Tumor pathways were defined as follows, consistent with previously suggested classifications[3, 8]: 1) "Adenoma-carcinoma (traditional) pathway" (MSS/MSI-L, CIMP-negative, *BRAF*-wildtype, *KRAS*-wildtype), 2) "Alternate pathway" (MSS/MSI-L, CIMP-negative, *BRAF*-wildtype, *KRAS*-mutated), 3) "Serrated pathway" (CIMP-positive, *BRAF*-mutated, *KRAS*-wildtype). Tumor pathway could be classified for 2,401 CRC cases (61.6%).

Tumor location was obtained from registry and pathology reports. Location was grouped based on ICD-9 codes as follows: 1) "Proximal" (153.0/Hepatic flexure, 153.1/Transverse colon, 153.4/Cecum, 153.6/Ascending colon), 2) "Distal" (153.2/Descending colon, 152.3/Sigmoid colon, 153.7/Splenic flexure), 3) "Rectal" (154.0/Rectosigmoid junction, 154.1/Rectum). Tumor location could be classified for 3,808 CRC cases (97.7%).

Statistical Analysis

We excluded women who were pre- or peri-menopausal at study baseline (934 cases, 760 controls) and those with missing data on HT use (208 cases, 209 controls). After exclusions, 3,898 CRC cases and 4,322 controls were included in our analyses (Figure 1).

Odds ratios (ORs) and 95% confidence intervals (CIs) from logistic regression models were used to approximate the relative risks for the association of HT use and CRC. Separate models were evaluated for each tumor-specific outcome using multinomial logistic regression with tumor marker status vs. control as the outcome (e.g., *BRAF*-mutated or *BRAF*-wildtype vs. control). All models included study site as well as covariates selected *a priori* based on known associations with both HT and CRC. These included age in years, body mass index (normal or underweight [BMI <25]/overweight [BMI 25-30]/obese [BMI ≥30]/unknown), smoking status (current/former/never/unknown), and first-degree relative with CRC (yes/no/unknown). Secondary analyses were conducted for estrogen-only therapy and combined estrogen plus progestin therapy. For multinomial logistic regression models, Wald chi-square tests were used to evaluate heterogeneity in ORs by tumor marker status.[53]

Additionally, sensitivity analyses were conducted excluding 1) women aged ≤45 years (n=131) and 2) women with probable Lynch syndrome based on four tumor markers (defined as

MSI-H, CIMP-negative, *BRAF*-wildtype, *KRAS*-wildtype; n=89), as both populations may have unique factors altering their CRC risk. We also performed a meta-analysis of the association of any HT use and CRC risk to evaluate heterogeneity by study site.

All analyses were conducted using R version 3.5.2 with a two-sided *P*-value <.05 considered statistically significant.

RESULTS

Baseline population characteristics of the 8,220 post-menopausal women in our study are shown in Table 1. Compared to controls, cases were more likely to have a family history of CRC and be current or former smokers. Cases were less likely to be HT users than controls (32.4% vs. 42.8%). Among those with formulation-specific data, cases were less likely than controls to use both estrogen-only (22.2% vs. 29.7%) and estrogen plus progestin formulations (14.3% vs. 17.8%).

Multivariable-adjusted associations between HT use, HT formulation, and overall- and tumor marker specific-CRC risk are presented in Figure 2 and Supplementary Table 3. Ever use of HT was associated with a 38% reduction in CRC risk (OR=0.62, 95% CI=0.56–0.69). Both use of estrogen-only (OR=0.71, 95% CI=0.62–0.83) and estrogen plus progestin (OR=0.76, 95% CI=0.64–0.91) formulations were associated with reduced CRC risk, although the effect estimates were attenuated compared to any HT use in this subsample of the study population. Few differences in baseline characteristics were noted between women with and without formulation-specific data (Supplementary Table 4).

Among cases with respective tumor marker data, 19.8% were MSI-H (n=719), 24.3% were CIMP-positive (n=841), 18.4% were *BRAF*-mutated (n=654), and 32.0% were *KRAS*-

mutated (n=1,098). Ever use of HT was associated with reduced risk of almost all tumor marker subtypes of CRC, with some variation across subtypes (Figure 2, Supplementary Table 3). The association of ever HT use and CRC was attenuated among CIMP-positive cases (OR=0.74, 95% CI=0.63–0.87) compared to CIMP-negative cases (OR=0.62, 95% CI=0.55–0.69) ($P_{het=}$.04). This trend was consistent across HT formulations, although the difference in ORs was not statistically significant. HT use was inversely associated with both *KRAS*-mutated and wildtype individuals. This association was consistent for estrogen-only use; however, estrogen plus progestin formulations were not statistically significantly associated with *KRAS*-mutated individuals (OR=0.90, 95% CI=0.70–1.14; P_{het}=0.09). No differences were observed for MSI or *BRAF* mutation status.

Of 2,401 tumors (61.6%) that we were able to be classified by pathway, the majority were classified as adenoma-carcinoma pathway tumors (48.4%; n=1,162), with 32.9% classified as alternate pathway (n=790) and 18.7% as the serrated pathway (n=449). No major differences in baseline characteristics were noted between women who were and were not able to be classified by pathway (Supplementary Table 4). The effect estimates for HT use in both the adenoma-carcinoma and alternate pathways were similar to that seen for HT overall (adenoma-carcinoma OR=0.63, 95% CI=0.55–0.73; alternate OR=0.61, 95% CI=0.51–0.72). However, the effect estimate was attenuated and no longer statistically significant for tumors that arose via the serrated pathway (OR=0.81, 95% CI=0.66–1.01 $P_{het\ vs.\ adenoma\text{-}carcinoma}$=.04). This difference was not consistent across HT formulation: for estrogen-only formulations, ever use was statistically significantly inversely associated with all three pathways (adenoma-carcinoma OR=0.71, 95% CI=0.57–0.88; alternate OR=0.60, 95% CI=0.47–0.77; serrated OR=0.72, 95% CI=0.54–0.98) and there was no statistical difference between pathways. However, for estrogen plus progestin

formulations, ever use was only statistically significantly associated with tumors that arose via the adenoma-carcinoma pathway (OR=0.70, 95% CI=0.55–0.91).

Most tumors were located in the proximal colon (47.3%), with tumors of the distal colon (29.4%) only slightly more common than rectal tumors (23.3%). Compared to distal colon (OR=0.57, 95% CI=0.49–0.66) and rectal tumors (OR=0.54, 95% CI=0.46–0.63), the effect estimate for the association of HT use and proximal colon tumors was attenuated (OR=0.71, 95% CI=0.62–0.80, $P_{het\ vs.\ distal}$=.01). This trend was consistent across HT formulations, although there was no statistical difference between proximal and distal colon tumors for estrogen only formulation (P_{het} =.32).

No substantial changes in results were noted after removing 131 women $\leq$45 years of age or 89 women with molecularly-defined Lynch Syndrome (Supplementary Tables 5 and 6). Meta-analysis results were consistent with our pooled main analysis (summary OR=0.64, 95% CI=0.58–0.71; P_{het}=.10).

DISCUSSION

In this large pooled study of post-menopausal women, HT use, regardless of formulation type, was associated with a decreased risk of CRC, consistent with prior research.[11-20] In general, this inverse association was observed irrespective of MSI, CIMP, *BRAF*, or *KRAS* status. However, when considering all tumor markers together and grouping cases by common pathways and tumor location, the association was attenuated for tumors arising via the serrated pathway and for proximal colon tumors.

Our results do not support the hypothesis that the association of HT and CRC differs by the individual tumor markers MSI, *BRAF* and *KRAS*. Strong inverse associations were observed

for HT use and CRC, regardless of *BRAF* and *KRAS* status. Prior studies found a nearly 20% reduced risk among ever HT users irrespective of *BRAF* and *KRAS* mutation status, although effect estimates did not reach statistical significance.[22, 23] These studies had substantially smaller samples sizes than ours, contributing to reduced power to detect difference in effect. We additionally observed strong inverse associations for HT use and both MSI-L/MSS and MSI-H CRC. Prior research is somewhat conflicting regarding the association of HT and MSI status, with most studies suggesting an association only among MSI-L/MSS patients.[22, 24, 54] There are many possible explanations for this discrepancy, including sample size, study design, reference period used for ascertaining HT use, and panels used to classify MSI status. Since prior studies have indicated high concordance across MSI panels,[41] we suspect the latter had the least influence.

We found some evidence that the association of HT differs by CIMP status, with an attenuated effect estimate observed for CIMP-positive tumors. A previous study had similar findings, reporting a borderline inverse association among CIMP-negative tumors and no association for CIMP-positive tumors.[22, 47] This finding should be interpreted with caution since CIMP is not consistently defined across studies and CIMP prevalence may be affected by detection method and sample quality.

Our results suggest that a comprehensive approach of considering tumor markers together as pathways may reveal otherwise nebulous patterns. Our findings indicate that the association of HT use and CRC was largely driven by tumors arising via the adenoma-carcinoma and alternate pathway. These tumors make up the majority of CRC cases, whereas serrated tumors represent about 20-30% of CRC.[1, 3, 8, 55-57] Serrated tumors, characterized as CIMP-positive, *BRAF*-mutated, and *KRAS*-wildtype, tend to behave more aggressively, with faster progression and

poorer prognosis.[3, 8, 10, 56, 58-60] Based on the different biologic behavior, appearance, distribution of tumor markers, and genetic susceptibility of serrated tumors, it is plausible that HT may indeed play a lesser role in their pathogenesis.

We also observed a weaker association for tumors of the proximal colon, consistent with prior studies.[22, 24] There is evidence that serrated tumors are more likely to develop in the proximal colon,[8, 61, 62] so it is unclear whether these are independent associations. In our study, most serrated tumors (n=391) were in the proximal colon. The association of HT use and proximal tumors was similar (OR=0.68, 95% CI=0.60–0.78) after removing serrated tumors from analysis, suggesting an independent association. However, 43.1% (n=776) of proximal tumors could not be classified by pathway due to incomplete tumor marker data, so this analysis is limited. The proximal and distal colon have different embryologic origins, microbiomes, and microenvironments.[62-66] As such, they appear to be predisposed to different tumor types. For instance, proximal colon tumors are more likely to be MSI-H, CIMP-positive, and mucinous and occur more commonly in women and older individuals.[67-71] Further research is needed to better elucidate whether differences in the proximal colon make it less sensitive to the effects of estrogens (i.e. less receptors, different microbiota), whether pre-cancerous lesions in the proximal colon are estrogen insensitive based on differences in the carcinogenic pathway, or some combination of factors.

Our results indicate that both estrogen-only and estrogen plus progestin formulations reduce CRC risk. In general, effect estimates were attenuated for estrogen plus progestin formulations compared to estrogen-only formulations, perhaps reflecting smaller exposure frequencies. However, overall trends were similar. Two main exceptions were present. First, while estrogen-only and any HT use were associated with about a 40% reduction in tumors

arising via the alternate pathway, estrogen plus progestin use was not statistically significantly associated with these tumors. This may indicate alternate pathway tumorigenesis is specifically modified by estrogen and not progestin. Likewise, there was a null association between estrogen plus progestin use and proximal colon tumors despite a 24-29% reduction in risk with estrogen-only or any HT respectively.

To our knowledge, this is the largest study to assess whether the association of HT use and CRC differs by individual tumor markers and location. In addition, it is one of few investigations that combines multiple tumor markers to evaluate tumor pathway-specific associations. Some limitations should be considered in interpreting our results. First, all exposure and epidemiologic covariate information included in this analysis was based on self-report, which could lead to exposure misclassification. Second, HT use was assessed only during the reference period, and detailed information on dose, frequency, and duration of use was not routinely available. Third, we were not able to assess endogenous hormones that may reflect age at menarche, parity, or breast feeding, which may also influence CRC risk. Fourth, there is some evidence that HT users may be more likely to undergo CRC screening.[72, 73] It is unclear how this may impact our results since this relationship may be complicated by differences in sensitivity of screening detection for specific CRC subtypes. Temporal trends and regional differences in screening and HT use may also influence observed associations. Finally, while this study includes populations in many locales, the participants were predominantly white and therefore, these findings may not be generalizable to other racial/ethnic groups.

In this large, multi-site study, we observed a strong inverse association between HT use and CRC risk, regardless of individual tumor markers and HT formulation. The decreased risk may predominantly reflect tumors of the distal colon or rectum and those arising via the

adenoma-carcinoma (traditional) pathway, as the association was relatively weaker among proximal colon tumors and those arising via the serrated pathway. Further investigation into the mechanisms underlying these differences may add to our understanding of subtype-specific CRC risk and pathways of tumorigenesis.

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Table 1. Baseline characteristics of 8,220 post-menopausal women by case-control status*

	Overall (n=8,220)	Case (n=3,898)	Control (n=4,322)
Age, mean (standard deviation)	65.28 (9.08)	64.79 (9.54)	65.72 (8.62)
Age group			
<45 years	101 (1.2)	84 (2.2)	17 (0.4)
45-55 years	828 (10.1)	464 (11.9)	364 (8.4)
55-65 years	2780 (33.8)	1246 (32.0)	1534 (35.5)
65-75 years	3309 (40.3)	1561 (40.0)	1748 (40.4)
>75 years	1202 (14.6)	543 (13.9)	659 (15.2)
First-degree relative with CRC			
Yes	1251 (15.2)	722 (18.5)	529 (12.2)
No	6633 (80.7)	2994 (76.8)	3639 (84.2)
Missing	336 (4.1)	182 (4.7)	154 (3.6)
Body mass index			
Normal or underweight	3659 (44.5)	1613 (41.4)	2046 (47.3)
Overweight	2818 (34.3)	1322 (33.9)	1496 (34.6)
Obese	1571 (19.1)	870 (22.3)	701 (16.2)
Missing	172 (2.1)	93 (2.4)	79 (1.8)
Smoking			
Current smoker	948 (11.5)	522 (13.4)	426 (9.9)
Former smoker	2619 (31.9)	1285 (33.0)	1334 (30.9)
Never smoker	4477 (54.5)	2012 (51.6)	2465 (57.0)
Missing	176 (2.1)	79 (2.0)	97 (2.2)
Self-reported race			
White	8077 (98.3)	3780 (97.0)	4297 (99.4)
Other	113 (1.4)	98 (2.5)	15 (0.4)
Missing	30 (0.4)	20 (0.5)	10 (0.2)
Study			
CCFR	1985 (24.1)	1215 (31.2)	770 (17.8)
CPSII	893 (10.9)	412 (10.6)	481 (11.1)
DACHS	2074 (25.2)	872 (22.4)	1202 (27.8)
DALS	891 (10.8)	427 (11.0)	464 (10.7)
EPIC Sweden	129 (1.6)	37 (0.9)	92 (2.1)
MCCS	455 (5.5)	185 (4.7)	270 (6.2)
NHS	1649 (20.1)	686 (17.6)	963 (22.3)
NSHDS	144 (1.8)	64 (1.6)	80 (1.9)
Any post-menopausal hormone therapy use			
Ever	3112 (37.9)	1262 (32.4)	1850 (42.8)
Never	5108 (62.1)	2636 (67.6)	2472 (57.2)
Estrogen-only			
Ever	1160 (14.1)	506 (13.0)	654 (15.1)
Never	3323 (40.4)	1778 (45.6)	1545 (35.7)
Missing	3737 (45.5)	1614 (41.4)	2123 (49.1)
Estrogen plus progestin			
Ever	717 (8.7)	328 (8.4)	389 (9.0)
Never	3758 (45.7)	1961 (50.3)	1797 (41.6)
Missing	3745 (45.6)	1609 (41.3)	2136 (49.4)

*No. (%) shown unless otherwise indicated

†Abbreviations: CRC, colorectal cancer; CCFR, Colon Cancer Family Registry; CPSII, Cancer Prevention Study-II; DACHS, Darmkrebs: Chancen der Verhutung durch Screening Study; DALS - Diet Activity and Lifestyle Study; EPIC, European Prospective Investigation into Cancer; MCCS, Melbourne Collaborative Cohort Study; NHS, Nurses' Health Study; NSHDS, Northern Sweden Health and Disease Study

Figure titles and legends

Figure 1. Overview of participants included in analytic population

Abbreviations: HT, post-menopausal hormone therapy; MSI, microsatellite instability; CIMP, CpG island methylator phenotype; A-C, adenoma-carcinoma

*Estrogen-only and estrogen plus progestin groups are not mutually exclusive

Figure 2. Association between post-menopausal hormone therapy (HT) use and colorectal cancer (CRC), overall and formulation-specific†

Abbreviations: CRC, colorectal cancer; MSI, microsatellite instability; CIMP, CpG island methylator phenotype; A-C, adenoma-carcinoma

*Wald p-value < 0.05. Wald p-values are comparing within-group ORs; reference groups are: *BRAF*-wildtype, *KRAS*-wildtype, CIMP-negative, traditional, distal colon

†Controls are used as reference for all ORs. All ORs are adjusted for age, body mass index, smoking status, and first-degree family history of CRC.

‡Formulation-specific data were only available for a subset of women (n=4,483 for estrogen-only, n=4,475 for estrogen plus progestin).

Figure 1

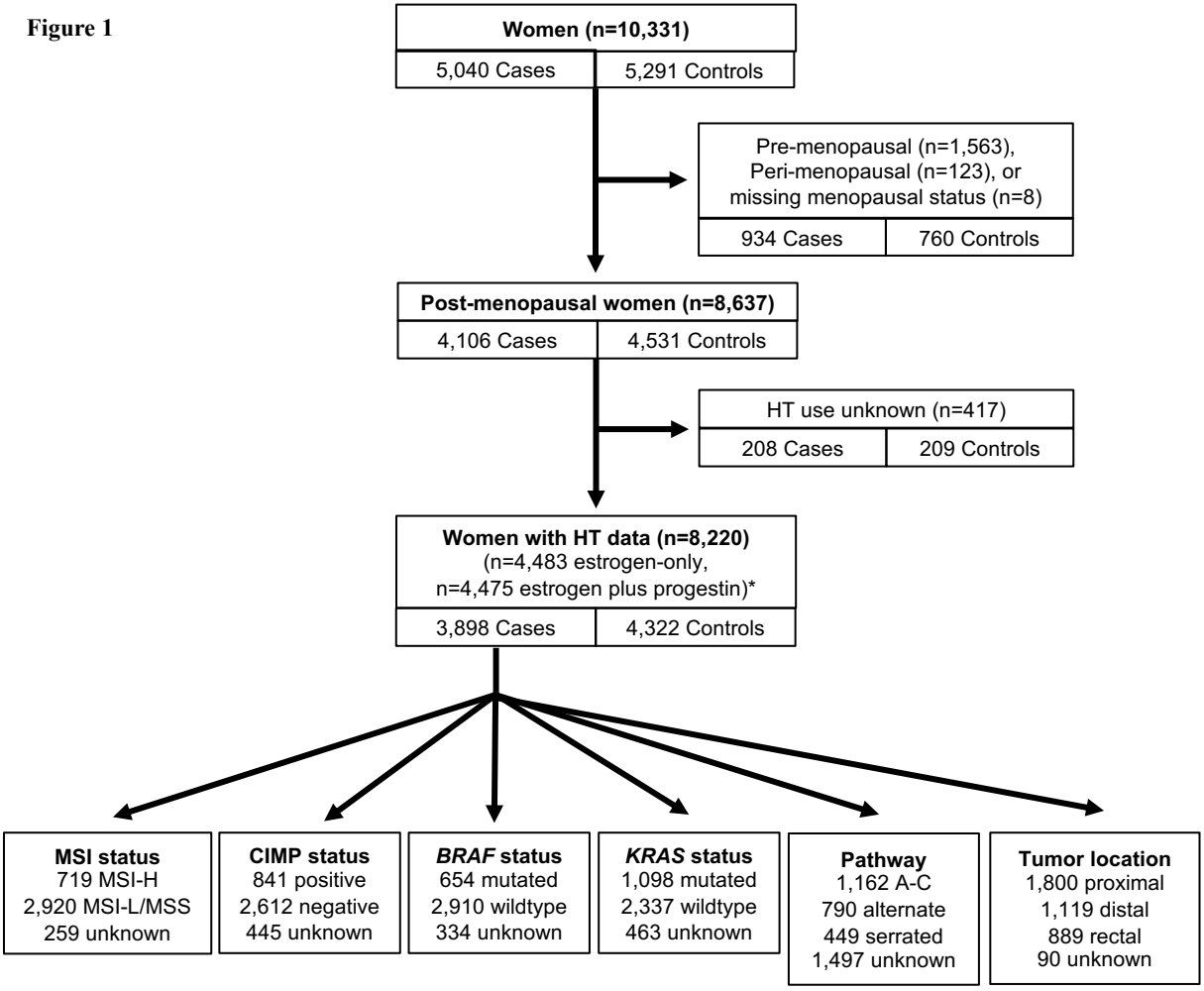

Figure 2