C-Reactive Protein Concentrations and Subsequent Ovarian Cancer Risk

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OBJECTIVE: To estimate the association between prediagnostic levels of C-reactive protein (CRP), a marker of chronic systemic inflammation, and subsequent development of ovarian cancer.

METHODS: A multicenter, nested, case–control study was conducted, including women who developed ovarian cancer (case patients) and women who were cancer-free (controls) from the following cohorts: CLUE ("Give us a CLUE to cancer and heart disease") cohorts of Washington County, Maryland, the Columbia, Missouri Serum Bank, and the Island of Guernsey Prospective Study, United Kingdom. A total of 167 incident invasive epithelial ovarian cancer cases were identified and each matched to an average of two controls on cohort, age, race, menopausal status, time since last menstrual period, current hormone use, date of recruitment, and time of day of blood draw. Baseline serum samples were assayed for CRP concentrations, and estimates of risk associated with CRP levels were assessed using conditional logistic regression.

RESULTS: Ovarian cancer risk was positively associated with increasing CRP concentrations. The risk of developing ovarian cancer among women in the highest third of the distribution of CRP compared with those in the lowest third was 1.72 (95% confidence interval 1.06–2.77), with evidence of an increasing risk with increasing concentration of CRP (*P* trend=0.02). Similar associations were observed using established clinical CRP cutpoints for heart disease risk (odds ratio 2.03, 95% confidence interval 1.20–3.47 for 3–10 mg/L compared with less than 1 mg/L, *P* trend=.008). If this association is causal, roughly 23% of ovarian cancer cases are attributed to chronic inflammation as indicated by elevated CRP concentrations.

CONCLUSION: Higher circulating CRP concentrations in women who subsequently developed ovarian cancer support the hypothesized role of chronic inflammation in ovarian carcinogenesis.

LEVEL OF EVIDENCE: II

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Inflammation has been proposed to play a role in the pathogenesis of ovarian cancer,[1] but there is little direct evidence to support this hypothesis. Indirect support is provided by the observations linking conditions associated with inflammation such as pelvic inflammatory disease,[1] endometriosis,[1] and polycystic ovary syndrome[2] to the development of ovarian cancer. Ovulation is an inflammatory process[3] involving cyclic wound healing and repair and incessant ovulation has long been proposed as an underlying factor leading to ovarian cancer.[4] Consistent with this

hypothesis, factors known to inhibit ovulation, including oral contraceptive use, pregnancy, and lactation, have been consistently associated with reduced ovarian cancer risk.[1] Additionally, tubal ligation and hysterectomy, interventions that may limit transmission of local irritants to the ovary, may also confer risk reduction.[1]

The study hypothesis is that chronic, systemic inflammation as measured by C-reactive protein (CRP) levels precedes onset of disease. To directly assess the potential association between systemic inflammation and the development of ovarian cancer, a multicenter nested case–control study was conducted. C-reactive protein is an acute phase protein released into the circulation in response to tissue damage and inflammation and is a biologic marker of chronic systemic inflammation. Modest elevations in CRP are associated with an increased risk of heart disease[5] and colon cancer.[6] C-reactive protein levels could also represent inflammation relevant to ovarian carcinogenesis, because CRP is involved in several mechanisms of immunologic response that are linked to cancer progression in animal models[7] and are believed to be important in peritoneal progression.[8,9] Furthermore, chronic inflammation has been proposed to play a role in tumor promotion,[10–12] specifically in the accumulation of cellular mutations, proliferation, and in angiogenesis. The association between CRP and subsequent development of ovarian cancer was assessed prospectively using biologic specimens collected years before the diagnosis of cancer, combining the resources of several ongoing cohort studies.

MATERIALS AND METHODS

A multicenter nested case–control study was conducted within a consortium of cohorts[6,13–15] using a standardized protocol for case and control selection. All samples were assayed together in a single laboratory. Consortium participants include the CLUE I ("Give us a CLUE to cancer and heart disease") and CLUE II ("Campaign Against Cancer and Heart Disease") cohorts of Washington County, Maryland, the Columbia, Missouri Serum Bank, and the Island of Guernsey Prospective Study, United Kingdom. The CLUE I cohort was initiated to learn more about risk factors for cancer and CLUE II was initiated as follow-up for further cancer inquiries and cardiovascular disease risk. Both the Guernsey studies and the Columbia, MO Serum Bank were originally established to study hormonal and other risk factors for breast cancer among a cohort of women who were cancer free at study entry. The main characteristics of each study group are presented in Table 1. Data available from all five cohorts included questionnaire information obtained at study entry, as well as stored biologic samples (either sera or plasma). During the follow-up period, 167 case patients with invasive epithelial ovarian cancer were identified across all cohorts. Invasive epithelial ovarian cancer (*International Classification of Diseases*, Ninth Revision, code 183) was the first cancer diagnosed with the possible exception of nonmelanoma skin cancer or cervical cancer in situ.

Community-based volunteers from Washington County, Maryland were enrolled in CLUE I over a 4-month period in 1974 and from May to October 1989 for CLUE II. Participants are followed up for cancer outcome by linkage to the Washington County Cancer Registry (estimated to be at least 90% complete) and the Maryland State Cancer Registry. Participants in the Columbia, Missouri Serum Bank were volunteers drawn from the Women's Cancer Control Program at the Cancer Research Center, University of Missouri Hospital, and the Ellis Fischel Cancer Center in conjunction with the Breast Cancer Detection Demonstration Project. The Columbia, Missouri Serum Bank recruited 6,720 women as part of the National Cancer Institute's Biological Markers Project. Participants responded to follow-up question-

Table 1. Selected Characteristics of Collaborative Cohorts

Cohort	Recruitment Period	Total Cohort Population*	Average Follow-up (y)	Age at Enrollment Mean (range)	Case Patients	Control Participants	Median CRP Levels Among Case Patients (IQR)	Median CRP Levels Among Controls (IQR)
CLUE I	1974	14,139	27.8	50.5 (26–86)	52	117	3.48 (1.2–6.0)	2.43 (1.13–6.15)
CLUE II	1989	14,622	13.0	60.9 (36–87)	47	93	3.41 (1.34–6.9)	2.14 (0.75–5.34)
Columbia, MO	1977–1987	6,720	19.4	53.8 (17–89)	34	63	3.14 (2.02–5.92)	2.28 (0.59–5.15)
Guernsey Phase I	1977–1985	5093	20.0	46.0 (26–88)	17	31	1.48 (0.45–3.66)	0.83 (0.27–1.99)
Guernsey Phase II	1986–1990	5163	11.5	51.6 (32–88)	17	31	0.93 (0.36–3.10)	0.88 (0.31–3.40)

CRP, C-reactive protein; IQR, interquartile range.
* CLUE I and CLUE II base populations are comprised of men and women, counts reflect women only.

naires regarding health status as part of case ascertainment. Loss to follow-up among women not known to be deceased was less than 10%. Case patients were identified by self-report and by linkage to the Missouri Cancer Registry, the Breast Cancer Detection Demonstration Project Cohort files, and the National Death Index Plus. Recruitment of community-based participants for the Island of Guernsey Prospective Study occurred in two phases, between 1977 and 1985 (phase I) and 1986 and 1990 (phase II). Case patients were identified through pathology reports, general practitioners, the Wessex Cancer Registry, and from death certificates. Pathology was not independently reviewed in this investigation, because tumor blocks were no longer available for most of the women identified as ovarian cancer patients in the participating cohorts.

Written informed consent was provided by all participants in each of the cohorts at the time of participation. The institutional review boards at the Johns Hopkins University Bloomberg School of Public Health, the University of Missouri, the National Cancer Institute, and the Guernsey Board of Health reviewed and approved the study.

All cohorts collected data on age, race, current oral contraceptive and hormone replacement use at time of blood draw, menopausal status, and date of last menstrual period. Information varied by cohort on smoking, height, weight, reproductive history, prior hormone use, medical history, and current medication use. Because we did not have information on diabetes at baseline, which is associated with inflammation, C-peptide concentrations were assayed as a surrogate measure of diabetes. Case patients were each matched to an average two control participants on cohort of origin, age, race, menopausal status, days since last menstrual period (LMP; if premenopausal), current oral contraceptive use, current use of other hormone replacement therapy (HRT), date of recruitment (within 6 months), and time of day of blood draw (AM or PM). For 12 case–control sets, only one suitable control participant was identified (7% of total). Control participants were cancer free and alive at the time of case diagnosis. All control participants were matched within 5 years of the age at which each case patient was diagnosed with ovarian cancer, and 85% of control participants were matched within 3 years. Premenopausal normally cycling (LMP within 35 days) control participants were matched within menstrual cycle phase (0–12, 13–15, and 16–35 days). Seven controls not matched within phase were matched within 2 days of case LMP when the case patient's LMP fell on the borderline of a phase cutoff point. A subset of CLUE I case–controls sets were not matched on time of day of blood draw[14] but CRP does not have significant circadian variation,[16] and this did not affect overall results. Ninety percent of case–control sets were matched within 6 months of case patient entry date. One case from the Guernsey cohort was excluded from analysis due to inability to find a suitable control. One control had insufficient serum for CRP assays.

Serum was stored from CLUE I, the Columbia, Missouri Serum Bank, and the Guernsey cohorts; plasma from CLUE II. All samples were stored at –70°C with the exception of the Guernsey cohort, where serum samples were stored at –20°C. High-sensitivity CRP and C-peptide concentrations were assayed in duplicate using an enzyme-linked immunosorbent assay (Diagnostic Systems Laboratories, Inc., Webster, TX). The upper and lower limits for the CRP assay were 10–500 ng/mL and were 0.13–13 ng/mL for C-peptide. Sixty masked quality control samples (approximating a 10% sample) aliquotted from pooled plasma or serum were arranged in triplet clusters and interspersed among the case–control sets. The mean intraset coefficient of variation among quality control samples was 11.0% for serum CRP and 5.6% for plasma CRP. Intraset coefficients of variation for C-peptide were 2.4% in serum and 16.1% in plasma. Because C-peptide is not stable in long-term storage at –20°C, C-peptide measures for the Guernsey cohort were excluded from the analysis of C-peptide, and ovarian cancer. Measures of CA 125, a tumor marker, were available for a subset of 33 cases and 53 controls (31 intact case–control sets) from CLUE I as part of a prior study.[17]

The distribution of circulating CRP was right skewed, and the data were log transformed to reduce departures from normality. Geometric means of CRP were compared between cases and controls using a paired *t* test with robust standard errors to account for correlations due to matching among controls, using the Huber/White/sandwich estimate of variance to account for the violation of the independent errors assumption[18] Case patients and matched control participants were also ranked by CRP concentrations within matched sets, and rankings were compared using Wilcoxon signed rank test for paired data. Case–control differences in categorical variables were compared using the Kruskal-Wallis test for ranked data. Odds ratios (ORs) and corresponding 95% confidence intervals (95% CI) were computed as estimates of the relative risk, using conditional logistic regression to account for the matched design. Increasing thirds of circulating CRP concentrations were

assigned using the distribution among control group. The cohorts differed on type of blood sample stored and the length and conditions of storage. For this reason, cutpoints by thirds were assigned within individual cohorts and by menopausal status and given a quantitative ranking score (0, 1, or 2). These within-cohort rankings were pooled across cohorts and menopausal groups. Odds ratios for thirds of CRP were estimated using the lowest third as the referent. Using the quantitative scores for the pooled thirds of CRP in the model as an ordinal variable, the presence of a trend in risk was assessed by calculation of Cuzick's nonparametric test for trend.[19]

Several factors are known to be associated with higher circulating CRP levels. These factors could either operate through CRP and thus represent a causal pathway or they may be confounders of the association between CRP and ovarian cancer. These include body mass index (BMI, kg/m^2), hormone use, smoking, age or menopause, metabolic syndrome or diabetes,[20] and nonsteroidal anti-inflammatory drugs (NSAID) or aspirin use.[21] Confounding was assessed using multiple conditional logistic regression analyses or by stratifying by those factors which were also part of the matching criteria. These adjustments for confounding showed no appreciable effect on risk estimates. Therefore, the most parsimonious models are presented. Stratification of results by quartiles of age at entry or year of entry excluded the likelihood of age- or time-period effects. For a specific variable, the Wald statistic was used to test for heterogeneity in the association between subgroups.[22]

Analyses were also conducted using CRP cutpoints defined as clinically relevant for heart disease risk by the American Heart Association (AHA) and Centers for Disease Control and Prevention (CDC) recommendations (1 mg/L or less, more than 1 mg/L, and 3 mg/L or less, more than 3 mg/L)[5] and excluding values corresponding to CRP levels characteristic of acute inflammation (more than 10 mg/L).[5]

Analyses were performed excluding women who were diagnosed with ovarian cancer within 2 and 5 years of blood draw to examine the possible effect of the presence of latent tumors on the association of CRP concentration with risk. In addition, correlations between log transformed CRP to both time to diagnosis and CA 125, an established tumor marker for ovarian cancer, were examined using Spearman rank correlation coefficients to address the possibility of latent tumor effects on study findings. All analyses were conducted using STATA 8.0 (College Station, TX). All significance testing was two-tailed, and *P* values of .05 or less were considered significant.

RESULTS

Baseline characteristics of case patients and control patients are shown in Table 2. Case patients and control participants were similar in age, menopausal status, current oral contraceptive use, and current hormone use because they were matched on these variables. All but two ovarian cancer case patients were white, and matching accounted for an equal proportion of nonwhite control participants. The mean BMI was slightly higher among case patients than control participants, but this was not statistically significant. The prevalence of current smoking and aspirin or NSAID use was similar between case patients and control participants (Table 2). Case patients and control participants were similar in parity among the 65 case–control sets with information available at study entry. Of 166 case patients included in this study, 124 had details on pathology; the remainder was confirmed by death certificate only. Among these, 11.3% were serous type, 10.8% endometrioid, 9.7% mucinous, 1.7% clear cell, and 66.1% were designated (adeno)carcinoma, not otherwise specified. Staging was available for only a limited number of case patients (n=54). Of these, 74 presented with stage III or stage IV disease, which is consistent with the presentation of ovarian cancer in the general population. Grading information was available on 31 case patients, 58% of whom were designated to have poorly differentiated tumors.

The association between selected characteristics and CRP concentrations among control participants is shown in Table 3. Covariate baseline characteristics associated with higher concentrations of CRP among cancer -free control participants include higher BMI (*P* for trend<.01), C-peptide (*P* for trend<.01), and current hormone use (oral contraceptives and HRT combined, *P* for trend<.01). C-reactive protein concentration was not associated with age, nulliparity (yes or no), or NSAID use among the control participants (Table 3).

Analysis of differences between case patients and controls, adjusted only for matching criteria, showed that the geometric mean of circulating CRP was 22% higher among cases (2.3 mg/L) than controls (1.9 mg/L, *P*=.05). Using the Wilcoxon signed rank test for paired data, women who developed ovarian cancer were, on average, statistically significantly more likely to have higher ranked CRP concentrations than their matched control participants (*P*=.02). Ovarian cancer risk increased with higher concentrations of circulating CRP (Table 4). Compared with women in the lowest third of CRP concentration, the odds ratio

Table 2. Baseline Characteristics of Incident Ovarian Cancer Case Patients and Matched Control Participants

Characteristic	Case Patients (n=166)	Control Participants (n=335)	*P*
Age at enrollment*	53.6 (±12.3)	53.6 (±12.7)	.89
Menopausal Status*			
Premenopausal	63 (38.0)	125 (37.3)	.89
Postmenopausal	103 (62.1)	210 (62.7)	
Current hormone use*			
Current oral contraceptive use	4 (2.4)	7 (2.1)	.64
Current hormone replacement therapy	16 (9.6)	32 (9.6)	.98
No current hormone use	146 (88.0)	296 (88.4)	
Nulliparous			
Yes	55 (82.1)	105 (84.0)	.74
No	12 (17.9)	20 (16.0)	
Missing (% of total)	99 (59.6)	210 (62.7)	
Smoking status			
Current	28 (17.4)	70 (21.5)	.28
Not current	133 (82.6)	255 (78.5)	
Missing	5 (3.0)	10 (3.0)	
BMI	26.3 (±5.8)	25.7 (±4.6)	.70
Missing	53 (31.9)	117 (34.9)	
NSAID and aspirin use, combined			
Current	27 (20.5)	68 (25.0)	.31
Not current	105 (79.6)	204 (75.0)	
Missing	34 (20.5)	63 (18.8)	
C-Peptide	1.77 (±2.41)	1.85 (±2.19)	.80
Missing	39 (23.5)	70 (20.9)	

BMI, body mass index; NSAID, nonsteroidal anti-inflammatory drug.
Data are number of persons (%) or mean (±standard deviation) unless otherwise specified.
* Matching criteria.

Table 3. Association Between C-reactive Protein and Covariates Among Control Participants at Baseline*

	Ranked Thirds of C-Reactive Protein			
Variable	Lowest (n=110)	Middle (n=114)	Highest (n=110)	*P* for Trend
Age at enrollment	53.0 (±13.2)	53.9 (±12.8)	53.2 (±12.3)	.72
BMI	22.7 (±2.2)	26.1 (±3.9)	28.5 (±5.2)	<.001
Current smokers	18 (16.7)	22 (20.0)	30 (28.3)	.10
Current hormone use (OC and HRT)	7 (6.4)	7 (6.1)	24 (21.8)	<.001
NSAID or aspirin use	21 (23.6)	26 (28.3)	21 (23.3)	.69
C-Peptide (ng/mL)†	1.56 (±2.17)	1.83 (±2.16)	2.21 (±2.16)	<.01
Parity (ever or never)†	32 (78.1)	35 (81.4)	37 (92.5)	.18

Abbreviations: BMI, body mass index; OC, oral contraceptive; HRT, hormone replacement therapy; NSAID, nonsteroidal anti-inflammatory drug.
* Data are mean (±standard deviation) or number (%) unless otherwise specified.
† C-peptide (Lowest, n=87; Middle, n=89; Highest, n=89), Parity (Lowest, n=41; Middle, n=43, Highest n=40).

was 1.26 (95% CI 0.78–2.04) in the middle third and increased to 1.72 (95% CI 1.06–2.77) in the highest third (*P* for trend=.02).

To assess for the potential influence of occult cancers undetected at study entry, women who were diagnosed with ovarian cancer within 2 and 5 years of blood draw were excluded, along with their respective control participants. The observed associations persisted after these exclusions. In addition to exclusion of case patients diagnosed within 2 or 5 years of study entry, no correlation was observed between log-transformed CRP and time to diagnosis among case patients (Spearman $r=-0.06$), nor was CRP correlated with CA 125 among the subset measured in CLUE I (Spearman $r=-0.02$), providing further evidence that increased risk of ovarian cancer with higher levels of CRP is likely not due to the effect of undiagnosed cancers at study entry.

Table 4. Ovarian Cancer Risk by Ranked Thirds of Baseline C-reactive Protein

	Ranked Thirds of C-reactive Protein			
	Lowest	Middle	Highest	*P* for Trend
All Participants*				
Cases	41	54	71	
Controls	110	114	110	
Odds ratio	1.0	1.26 (0.78-2.04)	1.72 (1.06-2.77)	.02
Excluding diagnoses within 2 y				
Cases	40	51	67	
Controls	105	106	107	
Odds ratio	1.0	1.26 (0.77-2.06)	1.65 (1.01–2.68)	.03
Excluding diagnoses within 5 y				
Cases	33	43	52	
Controls	84	87	88	
Odds ratio	1.0	1.25 (0.73–2.15)	1.52 (0.88-2.61)	.08
Nonsmokers				
Cases	34	46	58	
Controls	92	92	80	
Odds ratio	1.0	1.51 (0.86–2.67)	2.10 (1.18–3.72)	.01
Excluding diagnoses within 2 y among nonsmokers				
Cases	34	43	55	
Controls	88	84	77	
Odds ratio	1.0	1.48 (0.83–2.64)	1.95 (1.09–3.48)	.02
Excluding diagnoses within 5 y among nonsmokers				
Cases	27	36	43	
Controls	68	67	61	
Odds ratio	1.0	1.58 (0.82–3.05)	1.98 (1.03–3.83)	.04

Data are n or odds ratio (95% confidence interval).
* Case patients and controls were matched on age, race, menopausal status, last menstrual period, current hormone use, date and time of day of blood draw, and cohort of origin.

Smoking may increase levels of circulating CRP and analyses were stratified to parse out effects on results. The associations with CRP were strengthened after excluding current smokers (Table 4), such that the OR in the highest compared with lowest tertile was 2.10 (95% CI 1.18–3.72; *P* for trend=.01). The association between CRP and ovarian cancer risk also remained statistically significant after excluding case patients diagnosed within either 2 or 5 years of follow-up among nonsmokers only.

Adjusting for other factors known to affect circulating CRP levels, including NSAID or aspirin use, parity, BMI, log-transformed C-peptide as a marker for diabetes, or residual confounding by age, did not appreciably change the observed associations. Restricting analyses to women not currently using hormones resulted in a slightly stronger association (OR 1.81, 95% CI 1.10–2.98), highest compared with lowest tertile (data not shown). Exclusion of women not using NSAIDs or aspirin at time of blood donation did not appreciably change the results. The OR in the highest compared with lowest tertile among overweight women (BMI more than 25) was 1.93 (95% CI 0.75–4.97) compared with 1.53 (95% CI 0.32–7.32) among nonoverweight women (BMI 25 or less). Tests for heterogeneity between strata by overweight status were not statistically significant. The estimated relative risks were similar among premenopausal and postmenopausal women. Data were also stratified by age at study entry (younger than 55 years or 55 years or older) and by year of study entry to assess age and time period effects, with no significant differences observed between strata.

C-reactive protein levels above 10 mg/L are indicative of acute inflammation,[5] which has been proposed to represent physiologic activities differing from those believed to be involved in carcinogenesis as a result of chronic inflammation.[10] After excluding either case patients or control participants with CRP levels above this threshold (18 case–control sets), the odds ratio was 1.31 (95% CI 0.80–2.15) in the middle third and increased to 1.89 (95% CI 1.12–3.19) in the highest third (*P* for trend=.02).

To frame these findings within a clinical context, additional analyses were conducted using the AHA and CDC–recommended CRP cutpoints (1 mg/L or less, more than 1 and 3 mg/L or less, more than 3 and 10 mg/L or less) for cardiovascular disease risk.

Findings using these clinical cutpoints were similar to the increased risks observed in analyses using tertile cutpoints (OR 2.03, 95% CI 1.20–3.47, *P* for trend<.01), comparing women with CRP levels more than 3 and 10 or less mg/L to those with levels less than 1 mg/L (*P* for trend=.008). Although the CRP distribution was significantly different in the Guernsey population than among the other cohorts, excluding these observations (34 case–control sets) did not affect the association (OR 1.97, 95% CI 1.06–3.24). Adjusting for log-transformed C-peptide or BMI, for participants with this information available, did not appreciably alter the findings. If this proves to be a causal association, an estimated 23% of ovarian cancer cases are attributed to chronic inflammation as indicated by CRP levels more than 3 mg/L and less than 10 mg/L.

DISCUSSION

The associations observed in this study support the hypothesis that inflammation *may* contribute to the development of ovarian cancer.[1] A trend of increased risk with increasing serum CRP levels, measured years before the diagnosis of ovarian cancer was observed, with a nearly two-fold risk among those in the highest category of CRP levels. Using the clinical cutpoints recommended by the AHA and CDC, a two-fold risk of developing ovarian cancer was observed among women with CRP levels within a range of 3 to 10 mg/L, as compared with women with CRP concentrations less than 1 mg/L. There was a positive trend (*P*=.008) in increasing risk observed with increasing increments of the AHA and CDC cutpoints. Should further studies find this to be a causal association, an estimated 23% of ovarian cancer cases are attributed to chronic inflammation as indicated by CRP concentrations falling within the highest range defined by the CDC and AHA cutpoints, (3–10 mg/L).

Several aspects of the results suggest this may be a true association. A significant dose-response relationship was observed across increasing thirds of CRP concentrations and persisted after adjusting for known confounders. The increased risk was observed in each of the participating study populations from three different sites, suggesting consistency in association. A strength of the prospective study design is that the measurement of CRP concentration was based on blood samples drawn before diagnosis of ovarian cancer. The associations observed do not seem to be due to the presence of occult cancers undiagnosed at baseline based on the observation that the association persisted after excluding case patients diagnosed within 5 years of follow-up. Further, CRP was not correlated with CA 125, a recognized tumor marker for ovarian cancer, nor was CRP correlated with time to diagnosis, providing further evidence for independence of associations from occult cancer at baseline. This is a large prospective study that evaluated the association between serum CPR concentrations measured before disease onset, so temporality of the association is assured.

To the extent possible with the available data, factors that may confound the association were carefully accounted for CRP; values characteristic of major acute inflammation (more than 10 mg/L) were excluded, and associations were adjusted for current smoking, hormone use, NSAID or aspirin use, and overweight status. Adjusting for C-peptide or BMI did not appreciably affect the estimated relative risk, suggesting the association is independent of diabetes or metabolic syndrome.

The association between CRP concentrations and ovarian cancer was stronger among nonsmokers when assessing risk along increasing thirds of CRP levels. Prior studies have shown that CRP levels are higher among smokers and current HRT users.[20] However, studies of cardiovascular disease risk have shown that despite increased CRP concentrations among current HRT users, there is an equivalent or stronger association of CRP levels to disease risk among nonusers.[23,24]

C-reactive protein is a nonspecific marker of systemic inflammation, and moderate increases in circulating concentrations, relative to high levels observed with acute inflammatory processes, indicate a state of chronic inflammation. Proinflammatory agents, such as CRP, act in an antitumorigenic manner in the acute phase response to inflammatory stimuli and are counterbalanced by anti-inflammatory agents as healing occurs. A state of chronic inflammation ensues when the system is dysregulated or when proinflammatory agents are persistently stimulated.[8] In this state, inflammatory agents may act in tumor promotion by enabling tumor cells to act opportunistically in exploiting extravasation mechanisms used by leukocytes,[25] or in promoting angiogenesis.[10] As such, inflammation would presumably play a role in tumor progression rather than initiation; providing the necessary impetus for advancement of a small group of recently transformed cells, as may frequently occur in postovulatory healing of the epithelium, to a frank tumor. The underlying cause of the inflammation may be of less importance, because the systemic environment provides proangiogenic, prometastatic mechanisms to the transformed cell, or population of

such cells, in a manner not specific to the initial source of inflammation. Results of the present study may thus represent a link between a generalized inflammatory state and ovarian carcinogenesis.

C-reactive protein may have usefulness as a factor in assembling risk profiles and in the identification of those that may benefit from anti-inflammatory drugs or weight loss. However, replication of these findings in other study groups is necessary before clinical recommendations should be implemented. The strength of the association observed using the CDC and AHA cutpoints are within the same range as that observed in meta-analyses of CRP and risk of heart disease. Similarly, these cutpoints may be useful in identifying women at high risk of ovarian cancer who may benefit from more intensive screening. Measures to reduce inflammation, such as weight loss or use of statins, may be useful in reducing ovarian cancer risk and deserve further study. For example, a recent case–control study showed that statin-users had a reduced risk of colon cancer.[26] Statins are known to reduce CRP levels,[27] and colon cancer has been associated with elevated prediagnostic CRP concentrations.[5] However, this is the first study to examine the role of CRP as a prediagnostic factor in ovarian cancer risk, and as such, it is premature to recommend changes in clinical practice without further study.

The study design is a multicenter study and presents challenges in pooling across sites with differences in handling of specimens. These challenges were addressed by matching within cohort and carefully pooling across cohorts. The Guernsey cohorts stored samples at a higher temperature than the others. Excluding the results from that cohort did not alter the inferences drawn from the study results. In addition, cohorts varied by collection of serum compared with plasma. Although CRP was measured more precisely in plasma, concentrations of CRP did not differ statistically between serum and plasma measures. Ascertainment of outcome status varied across the cohorts. To assess whether these differences influenced the overall results, analyses were conducted by systematically excluding case–control sets by cohort of origin and study location. Results were similar to that observed in the entire study population combined. Steps in the analysis were taken to reduce the effects of inconsistencies in questionnaire data collected, including limiting analyses to observations with data available, performing sensitivity analyses using imputed values for missing BMI information, and using all positive or all negative values for missing information regarding NSAID or aspirin use. Additional studies are needed to replicate these finding among other racial or ethnic groups.

In summary, this prospective study demonstrated that, among women who were cancer-free at baseline, moderately elevated CRP concentrations consistent with chronic inflammation were associated with subsequent development of ovarian cancer. The main contribution of this study is to further elucidate the cause of ovarian cancer and provide a forum for further directions in research and prevention. If this association is, in fact, causal, then chronic systemic inflammation may explain as much as 23% of ovarian cancer cases. These findings provide direct evidence in support of the hypothesis that inflammation contributes to ovarian carcinogenesis.

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