

# Serum hormone levels in relation to reproductive and lifestyle factors in postmenopausal women (United States)

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**Objectives:** Endogenous sex hormones are thought to be involved in breast and endometrial cancers, but few studies have evaluated the relationships between hormones and risk factors for these diseases.

**Methods:** We related serum hormone and sex-hormone binding globulin (SHBG) levels to reproductive and lifestyle risk factors in a cross-sectional study of 125 postmenopausal women in five geographic regions of the United States.

**Results:** The estrogens were associated positively, while SHBG was associated negatively with body mass index (wt/ht<sup>2</sup>). Estrone, (E1), estrone sulfate, and bioavailable estradiol (BioE2) were inversely associated with height. Androstenedione was positively associated with age at menopause, while androstenedione, E1, estradiol, and BioE2 were inversely associated with age at menarche. Weekly alcohol drinkers had higher hormone levels, and lower SHBG levels than those who abstained. Androstenedione and E1 decreased with increasing levels of nonrecreational activity.

**Conclusions:** Several of these findings support the hypothesis that breast and endometrial cancer risk factors are mediated, in part, through increased endogenous hormone levels. The androstenedione findings are of interest in light of studies relating androstenedione to endometrial and possibly breast cancer. An association of age at menarche with E2, independent of androstenedione, may reflect increased aromatase activity in women with earlier menarche. *Cancer Causes and Control* 1998, 9, 199-207

**Key words:** Body size, lifestyle factors, postmenopausal women, reproductive history, sex hormones, United States.

## Introduction

Endogenous sex hormones are thought to be involved in the development of breast and endometrial cancers.<sup>1,2</sup> Both estrogens and androgens have been suggested to play a role. Positive associations with estradiol (E2),<sup>3,4</sup> and testosterone<sup>3,4</sup> have been reported in several recent prospective studies of serum sex hormones and breast

cancer, and an association of androstenedione with postmenopausal breast cancer has also been suggested.<sup>4,5</sup> Although other prospective studies have failed to confirm these findings,<sup>7,8</sup> these inconsistencies may derive in part from small sample sizes. In two case-control studies of endometrial cancer,<sup>9,10</sup> positive associations with andros-

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enedione, estrone (E1), and albumin-bound or total E2 have been reported.

The effects of several reproductive and lifestyle factors on cancer risk have been hypothesized to be mediated by endogenous hormones. However, little is known about how reproductive and lifestyle risk factors relate to endogenous hormone levels in postmenopausal women. We know of only three studies to date,<sup>11-13</sup> that have related reproductive factors to overall blood-estrogen levels in postmenopausal women. One of these<sup>12</sup> included analyses of estrone sulfate (E1SO<sub>4</sub>) and found an inverse relationship with parity. A few studies<sup>11,13,14</sup> have measured sex-hormone binding globulin (SHBG), hypothesizing an inverse relation to breast cancer risk factors. Inverse associations of SHBG with body mass index (BMI) (wt[kg]/ht[m]<sup>2</sup>) were noted, but no associations of the reproductive factors with SHBG were found. No prior studies have related reproductive and menstrual factors to postmenopausal androgen levels.

Positive associations between estrogen levels and BMI<sup>13,15-17</sup> or percent ideal weight<sup>18,19</sup> have been reported, although two studies<sup>11,20</sup> found no relation of E1 to BMI in postmenopausal women. Several studies have assessed relationships of blood hormone levels to alcohol consumption and physical activity in postmenopausal women, but results have been inconsistent. Positive associations of alcohol with estrone sulfate (E1SO<sub>4</sub>)<sup>16</sup> and estradiol<sup>21</sup> have been reported, although other studies showed no associations with estradiol<sup>11,15,16</sup> or estrone.<sup>11,15,16,20</sup> Cauley *et al*<sup>15</sup> reported inverse relationships of E1 and E2 with physical activity, while Newcomb *et al*<sup>20</sup> found no association with E1.

We evaluated the relationships of serum hormone and SHBG levels to reproductive, menstrual, and lifestyle factors in postmenopausal community-control subjects participating in an endometrial cancer case-control study.

## Materials and methods

### Study population

The case-control study of endometrial cancer from which the current study population is drawn is described in detail elsewhere.<sup>22</sup> Briefly, cases were accrued from seven hospitals in five geographic regions of the United States: Chicago, Illinois; Hershey, Pennsylvania; Irvine and Long Beach, California; Minneapolis, Minnesota; and Winston-Salem, North Carolina. Cases newly diagnosed between 1 June 1987 and 15 May 1990, between ages 20 and 74 years, and living in defined geographic areas were eligible for the study.

The community control subjects were matched to endometrial cancer cases according to age (five-year age group), race, and area of residence.<sup>22</sup> For cases who were

between ages 20 and 64, random digit dialing (RDD) procedures were used to select women with similar telephone exchanges to cases. Older controls were selected from US Health Care Financing Administration (HCFA) data tapes by matching subjects to cases on zip code. Only subjects with intact uteri were eligible. Overall, 65.6 percent ( $n = 313$ ) of 477 eligible community-control subjects agreed to be interviewed: 76.3 percent of the RDD and 46.8 percent of HCFA controls. The 16 control subjects matched to sarcoma cases were excluded from analyses.

Interviewers trained in a standard manner conducted in-person interviews, usually in subjects' homes, to obtain information on a variety of characteristics, including those listed below. Fasting blood samples were collected at the time of the interview, or on a scheduled day after the interview. Blood samples were obtained for 73.6 percent ( $n = 217$ ) of the remaining 297 interviewed controls.

We studied 125 women who reported that their menstrual periods stopped naturally. Excluded from the analysis were women who were premenopausal or who reported a menstrual period within the last year ( $n = 58$ ), whose periods stopped for a reason other than natural menopause ( $n = 5$ ), with a personal history of breast cancer or current tamoxifen use ( $n = 2$ ), who reported current (within the last six months) exogenous hormone use ( $n = 24$ ), whose age at menarche was missing ( $n = 2$ ), or who had an outlying measured value for E1SO<sub>4</sub>, suggestive of exogenous estrogen use ( $n = 1$ ). Nine additional subjects were excluded only from analyses of past estrogen replacement therapy (ERT) due to missing information on past use, or use for less than four months. Six subjects were excluded only from analyses of family income due to missing information.

### Exposure data

Characteristics studied in this analysis are: age; BMI based on reported weight and height; height; years since menopause; age at menopause; age at menarche; nulliparity; number of births; age at first birth; smoking status; current amount smoked; current weekly alcohol intake; current physical activity; past estrogen replacement therapy (ERT); years of education; and family income. Physical activity was evaluated using two composite measures of activity (recreational and non-recreational) as previously described,<sup>23</sup> for the subject's current decade of life. These measures were derived from questions regarding frequencies (daily, weekly, sometimes, rarely, or never) of specific types of activities.

### Laboratory analysis

Blood samples were analyzed at Nichols Institute, Inc. (San Juan Capistrano, CA). Levels of E2, estrone (E1), and androstenedione were measured by an in-house method of radioimmunoassay following extraction with

20 percent ethyl acetate in hexane and separation by celite chromatography.<sup>24</sup> Estrone sulfate (E1SO<sub>4</sub>) was measured by radioimmunoassay after extraction with an organic solvent,<sup>25</sup> enzymatic hydrolysis, and celite chromatography.<sup>24</sup> A commercially available radioimmunoassay kit manufactured by Diagnostic Systems Laboratories, Inc., Webster, Texas (catalog #DSL 6300), was used to measure SHBG levels. Ammonium sulfate precipitation was used to determine the percent of E2 that was not bound to SHBG.<sup>26</sup> The amount of non-SHBG-bound E2, which we refer to as bioavailable E2 (BioE2), was then calculated by multiplying this percent by the total amount of E2. Complete hormone results were available for all subjects, except one postmenopausal subject for whom serum volume was insufficient to obtain the level of BioE2.

Multiple, masked quality-control blood samples were assayed with the blood samples from study subjects. Coefficients of variation (CV) were 17 percent or less for SHBG and all hormone measures except androstenedione.<sup>9</sup> Since exclusion of one outlying batch for androstenedione reduced the CV from 45.1 percent to 10.8 percent, we repeated analyses in which statistically significant relationships with androstenedione were found, excluding samples from that batch. Results were unchanged; therefore, reported results include data from that batch.

#### Statistical analysis

Hormone and SHBG serum concentrations were logarithm-transformed for all analyses; results are presented as geometric means. Analyses of covariance (ANCOVA), and two-way ANOVAS were conducted to evaluate the relationships of SHBG and steroid hormones to the reproductive and lifestyle factors, while controlling for BMI and years since menopause, if these were related to SHBG or the hormone being analyzed. In these models, years since menopause was modeled as a categorical variable, but BMI was modeled as a continuous variable to conserve power. For instances in which a hormone was statistically significantly associated with more than one characteristic, independence of each relationship was assessed by examining geometric-mean hormone levels by categories of each characteristic with inclusion of the other characteristic in the model. All relationships persisted.

The statistical independence of the relationships of the hormones (e.g., androstenedione) to the reproductive and lifestyle factors (e.g., age at menarche) was assessed by including associated hormones (e.g., E2) as independent variables in the ANCOVA and examining whether the patterns of mean hormones, with and without adjustment, were similar. When correlations between hormones were greater than 0.65, we did not attempt to establish whether each was statistically independently related to the repro-

ductive and lifestyle factors. Linear regression analyses were also performed using the continuous data to test for trend.

A previous small study<sup>27</sup> reported a significant diurnal variation in androstenedione levels, but no difference between samples drawn at 8 am and 12 noon. Because a diurnal variation might obscure associations with risk factors, we examined mean androstenedione levels by hour of blood draw. We found no significant differences (data not shown), perhaps because most samples were obtained before 1 pm.

All analyses were conducted using SAS/Stat, Version 6 software.<sup>28</sup>

## Results

The women ranged in age from 45 to 78 years (mean, 62.8), and had been menopausal for at least one year and up to 36 years (mean = 14.0) (Table 1). Their mean BMI was 26.4 kg/m<sup>2</sup>. Only 12 women (9.6 percent) were nulliparous. Twenty percent were current smokers, and 22 percent were past smokers. Weekly alcohol intake in the study population was low: 40 percent of the women did not drink at all; 42 percent drank less than 20 grams/week; and 18 percent drank more. Fifty-four percent of the women finished high school; college graduates comprised 19 percent more. Ten percent had taken exogenous menopausal estrogen in the past. Mean levels and ranges of the serum hormones and SHBG are included in Table 1.

**Table 1.** Descriptive characteristics and serum hormone and SHBG levels in 125 postmenopausal US women

Characteristic	Mean	(SD) <sup>a</sup>
Age (yrs)	62.8	6.6
Years since menopause	14.0	8.3
Body mass index (kg/m <sup>2</sup> )	26.4	6.1
Age at menarche (yrs)	13.0	1.3
Weekly alcohol consumption (grams)	18.7	50.7
Number of children <sup>b</sup>	3.5	1.9
Age at first birth (yrs)	23.6	4.0
Serum hormone	Geometric mean	Range
Androstenedione (ng/dL)	56.8	8-172
E1 (pg/mL)	29.1	10-92
E1SO <sub>4</sub> (pg/mL)	287.1	25-2,173
E2 (pg/mL)	6.3	1-42
SHBG (nmol/L)	38.1	2.5-112
BioE2 (pg/mL)	1.5	0.1-9.3

<sup>a</sup> Standard deviation.

<sup>b</sup> Among parous women only.

**Table 2.** Pearson correlations among serum hormone and SHBG levels in 125 postmenopausal US women<sup>a</sup>

	E1	E1SO <sub>4</sub>	E2	SHBG	BioE2
Androstenedione	0.57	0.38	0.43	-0.15	0.42
E1		0.76	0.82	-0.19	0.66
E1SO <sub>4</sub>			0.65	-0.37	0.68
E2				-0.29	0.84
SHBG					-0.69

<sup>a</sup>  $P \leq 0.05$  for all correlations except between SHBG and androstenedione ( $P = 0.09$ ).

**Table 3.** Geometric mean serum hormone and SHBG levels (and 95% confidence intervals) by age, relative weight, and height, 125 postmenopausal US women

Characteristic	No.	Androstenedione ng/dL	E1 pg/mL	E1SO <sub>4</sub> pg/mL	E2 pg/mL	SHBG nmol/L	BioE2 pg/mL
<b>Age<sup>a,b</sup></b>							
< 55	17	61 (47-81)	32 (26-38)	425 (299-603)	7.6 (6.1-9.3)	39 (28-54)	1.7 (1.2-2.5)
55-59	21	56 (45-70)	28 (24-33)	271 (198-372)	5.9 (4.8-7.1)	30 (23-39)	1.5 (1.1-2.1)
60-64	39	59 (51-69)	27 (24-31)	263 (208-331)	6.1 (5.3-7.0)	39 (32-47)	1.4 (1.1-1.7)
65-69	27	51 (42-63)	29 (25-34)	252 (191-333)	5.8 (4.9-6.9)	43 (33-55)	1.4 (1.0-1.8)
70+	21	59 (45-78)	31 (27-37)	311 (227-426)	6.7 (5.6-8.1)	40 (29-56)	1.9 (1.3-2.8)
<i>P</i> for trend		0.88	0.57	0.50	0.72	0.66	0.57
<b>BMI (kg/m<sup>2</sup>)<sup>a</sup></b>							
< 21	16	49 (39-61)	24 (19-28)	217 (150-315)	3.3 (2.6-4.1)	65 (51-84)	0.5 (0.4-0.7)
21 to < 23	21	70 (57-86)	28 (23-33)	265 (192-367)	5.6 (4.6-6.7)	35 (28-44)	1.6 (1.2-2.0)
23 to < 25	23	55 (45-66)	25 (22-30)	229 (168-312)	5.4 (4.5-6.4)	49 (40-61)	1.1 (0.9-1.4)
25 to < 29	34	55 (48-65)	28 (24-32)	277 (215-357)	6.5 (5.6-7.5)	38 (32-45)	1.6 (1.3-1.9)
29+	31	58 (49-67)	39 (34-45)	433 (332-566)	10.3 (8.8-12.1)	26 (21-31)	3.2 (2.6-4.0)
<i>P</i> for trend		0.63	0.0001	0.0001	0.0001	0.0001	0.0001
<b>Height (cm)<sup>a,b,c</sup></b>							
≤ 154.94	19	56 (45-69)	31 (26-37)	361 (257-508)	6.0 (4.9-7.4)	29 (23-38)	1.7 (1.3-2.3)
157.48	21	60 (49-73)	33 (28-39)	376 (275-515)	7.9 (6.6-9.5)	39 (30-49)	1.8 (1.4-2.4)
160.02-162.56	36	61 (53-71)	29 (25-32)	278 (219-353)	6.0 (5.2-6.9)	38 (32-46)	1.6 (1.3-2.0)
165.1-167.64	29	55 (47-65)	28 (24-32)	261 (200-341)	6.4 (5.4-7.5)	42 (34-51)	1.4 (1.1-1.8)
170.18-177.8	20	52 (42-64)	26 (22-31)	213 (154-295)	5.5 (4.5-6.6)	42 (33-54)	1.2 (0.9-1.6)
<i>P</i> for trend		0.33	0.07	0.05	0.21	0.10	0.05

<sup>a</sup> Means for androstenedione, SHBG, and BioE2 are adjusted for years since menopause; BMI = body mass index.

<sup>b</sup> Means for SHBG and all hormones except androstenedione are adjusted for BMI.

<sup>c</sup> Height was reported in inches and converted to centimeters, hence the gaps between ranges.

The serum sex hormones were moderately to highly correlated (Table 2). Correlations among the estrogens ranged from 0.65 to 0.84, and correlations between androstenedione and the estrogens ranged from 0.38 to 0.57. SHBG was negatively correlated with BioE2, E1SO<sub>4</sub>, E1, and E2; correlations ranged from -0.19 to -0.69.

Age was not related to SHBG or any of the hormones (Table 3). All of the estrogen measures were associated positively with BMI, and SHBG was related inversely to BMI. The relationships of SHBG and E1 to BMI persisted after mutual adjustment. Androstenedione was not asso-

ciated with BMI. E1SO<sub>4</sub> and BioE2 decreased with increasing height; a similar trend for E1 was suggested.

Androstenedione was associated positively with age at menopause. Mean levels were 45, 58, 58, and 68 ng/dL ( $P$  trend = 0.003) for ages at menopause of less than 45, 45 to 49, 50 to 54, and 55 or older. An association persisted (means = 48, 60, 58, and 65 ng/dL) after adjustment for years since menopause. No associations of age at menopause with SHBG and the estrogen measures were observed (data not shown).

SHBG levels were higher, and androstenedione, E1SO<sub>4</sub>, and BioE2 levels were lower in the later post-

**Table 4.** Geometric mean serum hormone and SHBG levels (and 95% confidence intervals) by menstrual characteristics, 125 postmenopausal US women

Characteristic	No.	Androstenedione ng/dL	E1 pg/mL	E1SO <sub>4</sub> pg/mL	E2 pg/mL	SHBG nmol/L	BioE2 pg/mL
<b>Years since menopause<sup>a</sup></b>							
< 5	19	57 (47-70)	28 (24-33)	332 (237-465)	6.7 (5.4-8.1)	36 (28-46)	1.7 (1.3-2.3)
5-9	26	69 (58-82)	31 (26-36)	319 (239-425)	6.7 (5.6-7.9)	31 (26-39)	1.9 (1.5-2.4)
10-14	28	61 (52-73)	29 (25-33)	298 (226-393)	6.5 (5.5-7.7)	31 (26-38)	1.9 (1.5-2.4)
15-19	23	54 (45-64)	28 (24-33)	232 (171-316)	5.5 (4.6-6.6)	49 (40-62)	1.0 (0.8-1.3)
20+	29	47 (40-55)	29 (25-34)	272 (208-357)	6.2 (5.3-7.3)	46 (38-56)	1.3 (1.1-1.7)
<i>P</i> for trend		0.0003	0.78	0.24	0.22	0.007	0.006
<b>Age at menarche<sup>a,b</sup></b>							
< 12	12	68 (52-88)	32 (25-39)	331 (217-505)	7.5 (5.9-9.6)	41 (29-56)	1.9 (1.3-2.7)
12	33	55 (47-65)	31 (27-35)	316 (244-408)	6.9 (6.0-8.0)	41 (33-49)	1.7 (1.3-2.1)
13	39	61 (53-70)	29 (26-33)	271 (214-343)	6.5 (5.7-7.4)	37 (31-45)	1.6 (1.3-1.9)
14	23	55 (46-66)	27 (23-32)	305 (225-415)	5.6 (4.7-6.7)	34 (27-42)	1.4 (1.1-1.9)
15+	18	50 (40-62)	26 (22-31)	232 (164-327)	5.0 (4.1-6.1)	40 (31-52)	1.2 (0.9-1.6)
<i>P</i> for trend		0.05	0.04	0.12	0.0008	0.63	0.02

<sup>a</sup> Means for SHBG and all hormones except androstenedione are adjusted for BMI (wt/ht<sup>2</sup>).

<sup>b</sup> Means for androstenedione, SHBG and BioE2 are adjusted for years since menopause.

**Table 5.** Geometric mean serum hormone and SHBG levels (and 95% confidence intervals) by alcohol intake and non-recreational physical activity,<sup>a</sup> 125 postmenopausal US women

Characteristic	No.	Androstenedione ng/dL	E1 pg/mL	E1SO <sub>4</sub> pg/mL	E2 pg/mL	SHBG nmol/L	BioE2 pg/mL
<b>Current alcohol intake (g/wk)</b>							
0	50	53 (47-60)	27 (25-30)	265 (215-326)	6.0 (5.3-6.8)	39 (33-45)	1.4 (1.2-1.7)
< 20	53	58 (52-66)	28 (25-31)	278 (228-339)	6.2 (5.5-7.0)	40 (34-46)	1.5 (1.3-1.8)
20+	22	65 (54-79)	36 (31-43)	376 (273-517)	7.1 (5.9-8.6)	33 (26-42)	1.9 (1.5-2.5)
<i>P</i> for trend		0.39	0.02	0.37	0.17	0.45	0.39
<b>Non-recreational physical activity</b>							
<b>Activity score<sup>b</sup></b>							
0-2	28	67 (57-79)	33 (29-38)	361 (274-476)	6.3 (5.3-7.4)	36 (29-44)	1.6 (1.2-2.0)
3-4	37	56 (49-65)	30 (26-34)	285 (224-363)	6.6 (5.7-7.6)	43 (36-51)	1.6 (1.3-1.9)
5-6	37	58 (50-67)	28 (25-32)	260 (205-331)	6.2 (5.4-7.2)	37 (31-45)	1.5 (1.2-1.9)
7-9	23	47 (39-56)	25 (21-29)	260 (191-353)	5.8 (4.9-7.0)	36 (28-45)	1.5 (1.1-1.9)
<i>P</i> for trend		0.01	0.003	0.15	0.32	0.84	0.50

<sup>a</sup> Means for SHBG and all hormones except androstenedione are adjusted for BMI. Means for androstenedione, SHBG and BioE2 are adjusted for years since menopause.

<sup>b</sup> Increasing scores represent increasing activity levels.

menopausal years (Table 4). The relationship of androstenedione to years since menopause was similar after adjustment for BioE2, but the relationship of BioE2 to years since menopause disappeared after adjustment for androstenedione.

Age at menarche was related inversely to androstenedione, E1, E1SO<sub>4</sub>, E2, and BioE2 (Table 4). The inverse associations between age at menarche and both androstenedione and E1 disappeared after mutual adjustment (data not shown). An inverse relation of E2 to age

at menarche persisted after adjustment for androstenedione (mean E2 levels became 6.8, 6.9, 6.4, 5.7, and 5.4 pg/ml for categories of increasing age at menarche), although the range of means decreased; the inverse relation of androstenedione to age at menarche became equivocal (mean A levels became 63, 53, 60, 58, and 55 ng/dL for categories of increasing age at menarche) after adjustment for E2.

E1 was significantly higher among women drinking more than 20 grams/week of alcohol compared with

abstainers (Table 5). The median intake in the group drinking at least 20 g/week was 63.5 g/week, or about five drinks per week; only eight women drank 15 or more g/day, precluding more detailed analysis at higher consumption levels. The associations with the other hormones followed similar patterns, and SHBG was lower with higher alcohol intake.

In an analysis of recreational physical activity, we initially examined mean SHBG and hormone levels without adjusting for BMI, since activity may affect hormones and SHBG through its influence on BMI. The least active group had higher levels of all of the estrogen measures, but there were no significant trends, and this suggested effect disappeared after adjustment for BMI (data not shown). No associations of SHBG and androstenedione with recreational physical activity were found. Androstenedione, E1 and E1SO<sub>4</sub> were associated inversely with non-recreational physical activity (Table 5). The relationship of E1 to non-recreational physical activity persisted after adjustment for androstenedione, but the relationship of androstenedione to activity disappeared after adjustment for E1 (data not shown).

An association of androstenedione to smoking (past or current) was suggested. The mean androstenedione levels for current smokers, past smokers, and nonsmokers were 65 (95 percent confidence intervals [CI] = 54-77), 60 (CI = 51-70), and 54 (CI = 48-60) ng/dL. Women smoking 10 to 19 cigarettes per day had a similar mean androstenedione to those smoking more (68 ng/dL each); only two current smokers smoked less than 10 cigarettes per day. No associations of any of the smoking measures were found with the estrogens or SHBG.

Level of education was associated positively with E1 ( $P$  trend = 0.03), E1SO<sub>4</sub> ( $P$  trend = 0.01), E2 ( $P$  trend = 0.005), and BioE2 ( $P$  trend = 0.02).

There was a suggestion that past ERT users had lower levels of E1SO<sub>4</sub> than nonusers (211 [CI = 141-317] of 314 [CI = 274-361] pg/ml); however this result was based on only 12 past users. Smaller differences in the same direction for E2, androstenedione, and in the opposite direction for SHBG, were also suggested.

Nulliparity, number of births, age at first birth, and income were unrelated to SHBG and all of the hormones measured (data not shown), although statistical power for assessment of nulliparity was limited by the small number ( $n = 12$ ) of nulliparous subjects.

Analyses were also conducted in the subset of normal weight women (BMI  $\leq 27$  kg) for relations in which statistically significant associations were found for all women combined. Generally similar trends were found employing a more liberal significance test ( $P < 0.1$ ) to compensate for loss of statistical power. However, the relationships of E1 with age at menarche and BMI were not quite significant ( $P_{\text{trend}} = 0.11$  for both relationships).

## Discussion

We noted associations of several female-cancer risk factors with androstenedione, the estrogens, and SHBG in this analysis. Our finding of associations of BMI with the estrogens and inversely with SHBG is consistent with most studies of estrogen<sup>13,15-19</sup> and SHBG,<sup>11,13,14,17,20,29,30</sup> however, associations with E1<sup>11,20</sup> and estradiol<sup>11</sup> were not found in other studies. Higher hormone levels also were associated with earlier menarche, later menopause, weekly alcohol consumption, less non-recreational physical activity, shorter stature, and more years of education, and SHBG was associated in a positive direction with years since menopause. The increase in E1SO<sub>4</sub> and BioE2 with shorter height is not consistent with the observed relationship of greater height with breast cancer risk being mediated through increased estrogen levels.<sup>31</sup> Hankinson *et al*<sup>16</sup> found no relationship, nor did two studies of urine estrogens.<sup>32,33</sup>

The associations of several characteristics with androstenedione levels are of particular interest in light of related findings: androstenedione was associated positively with endometrial cancer risk in two case-control studies,<sup>9,10</sup> and a positive association with breast cancer was suggested in two cohort studies.<sup>43</sup> Further, there is evidence that androstenedione can be converted to estrogen in adipose tissue by the enzyme aromatase.<sup>34</sup> Whether greater androstenedione or other hormone levels underlie the association of recognized risk factors with disease is a topic for further research.

We observed inverse trends for androstenedione and BioE2, and a positive trend for SHBG, with years since menopause. Previous results have been mixed regarding the effect of time since menopause and current age on androstenedione levels.<sup>15,18,19,35-39</sup> A study in oophorectomized women showed declining androstenedione with age.<sup>40</sup> Although current age was not related to androstenedione in the present study after adjustment for years since menopause, the possibility that androstenedione declines with age cannot be ruled out given the high correlation of current age with years since menopause. To our knowledge, no studies have presented mean androstenedione levels stratified concurrently by current age and time since menopause, which may explain some of the discrepancies in findings; a larger study than ours would be required to do this. Our finding of a positive association of years since menopause with SHBG agrees with previous studies,<sup>14,20</sup> although again an association with age cannot be ruled out.

We also found that androstenedione increased with age at menopause, an association which appeared to be independent of time since menopause. Our finding of no association of estradiol<sup>11,13,32</sup> or estrone<sup>11,32</sup> with age at menopause is consistent with other studies.

The relationships of endogenous postmenopausal hormones to age at menarche are interesting since menarche occurred years earlier. The finding of higher E2 levels with earlier age at menarche in postmenopausal women, independent of androstenedione, suggests that early menarche may be a marker for greater E2 production from precursors, perhaps due to greater aromatase activity. This finding is consistent with two previous studies<sup>32,41</sup> of postmenopausal hormones, although others found no relation.<sup>11-13</sup> It is possible that imprecise recall of age at menarche obscured differences in these latter studies. Results are also mixed for studies of premenopausal women; several studies have found higher plasma or urinary estrogen levels with earlier menarche,<sup>42-43,44</sup> (Japanese women) while others have not.<sup>13,44</sup> (British women),<sup>45-47</sup> A positive association of SHBG with age at menarche in premenopausal women has been reported in several,<sup>11,14,42</sup> but not all<sup>44</sup> studies.

We found no relationships of hormones to nulliparity, number of births, or age at first birth, although we were limited by the small number of nulliparous subjects. Several other studies of postmenopausal blood hormone levels found no associations of these factors with estradiol,<sup>11-13</sup> estrone,<sup>11,12</sup> or SHBG;<sup>11,13,14</sup> one study,<sup>12</sup> however, examined E1SO<sub>4</sub> and found an inverse association with number of births. One study of urinary estrogens<sup>13</sup> showed a nonsignificant positive association with nulliparity.

We observed associations, statistically significant for E1, of alcohol consumption with all of the hormones in women who drank at least 20 g/week (almost all of whom drank less than two drinks [less than 30 g] per day). Hankinson<sup>16</sup> also found an association of alcohol with E1SO<sub>4</sub>. Plasma E1 was not associated with alcohol in other studies;<sup>11,15,16,20</sup> E2 was related positively in one study,<sup>21</sup> but not in others.<sup>11,15,16</sup> It is possible that factors such as imprecision or inaccuracy in reported alcohol consumption, or inaccuracy in hormone assays explain some of the discrepancies among studies. In fact, mean E2 levels vary widely among four of these five studies, and one study was unable to measure E2 in normal weight women.<sup>15</sup> Urinary E1 and E2 were associated in one study<sup>48</sup> but not another.<sup>32</sup>

We found a suggested inverse association of each estrogen measure with recreational activity which disappeared after adjustment for BMI. However, a significant inverse association of E1 to non-recreational physical activity was independent of BMI, and similar associations with androstenedione and E1SO<sub>4</sub> were suggested. Similarly, non-recreational physical activity was associated with decreased endometrial cancer risk in these data, and an apparent inverse relationship with recreational physical activity was explained by BMI.<sup>23</sup> One prior study<sup>15</sup> found an inverse relation between physical activity and

E1 and E2, while another<sup>20</sup> found no association with E1. Possible reasons for the lack of association of the hormones with recreational activity, given a non-recreational activity association, include: (i) measurement error in our assessment of recreational activity; and (ii) non-recreational activity generally may be more sustained than recreational activity.

Smoking has been related inversely to endometrial cancer risk in a number of studies.<sup>49</sup> The suggestion of increased androstenedione in smokers, and no difference in estrogens between smokers and nonsmokers, is consistent with other studies of postmenopausal androstenedione,<sup>15,50-54</sup> and estrogen (E1 and/or E2).<sup>11,15,20,51-55</sup> However, since androstenedione was associated with increased endometrial cancer risk in the parent case-control study,<sup>9</sup> the mechanism underlying the protective effect of smoking on endometrial cancer risk remains unclear.

Two weaknesses of the study should be noted. First, as in most random samples from the community, the response rate to the blood draw was low, which may have limited the generalizability of our results. Second, since a number of comparisons were performed, some of our findings may be due to chance, although most of our findings are consistent with the hypothesis that breast and endometrial cancer risks are mediated, in part, by increased hormone and decreased SHBG levels.

Since testosterone (T) has been associated with breast cancer risk in two recent cohort studies,<sup>3,4</sup> further studies examining the relationship of this hormone to risk factors are of interest. Two studies found no relationships of T to exercise, alcohol consumption, and smoking.<sup>15,20</sup> Associations of T with obesity<sup>15</sup> and body weight<sup>46</sup> have been reported.

In summary, these findings provide evidence that increased serum hormone levels and decreased SHBG levels may explain, in part, the cancer risks related to greater relative weight, early menarche, late menopause, less physical activity, and alcohol consumption.

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