**Supplementary Material**

**Table S1: Coefficients of variation for the sex steroid hormones and SHBG**

|  |  |  |
| --- | --- | --- |
| **Biomarker\*** | **IQC level** | Average within laboratory (total) CV (%) |
| **Estradiol** | Low | 15.26 |
| Medium | 8.68 |
| High | 6.47 |
| **Testosterone** | Low | 8.34 |
| Medium | 3.66 |
| High | 4.15 |
| **SHBG** | Low | 5.67 |
| Medium | 5.25 |
| High | 5.22 |
| **Albumin** | Low | 2.09 |
| Medium | 2.20 |
| High | 2.13 |

Limit of detection values\*: Estradiol= <175 pmol/L: Testosterone= <0.35 nmol/L, SHBG= <0.33 nmol/L, albumin= <15g/L

|  | No. of cases | HR (95% CI) |  | No. of cases | HR (95% CI)† |  | No. of cases | HR (95% CI) |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Testosterone (nmol/L)\*** |  |  |  |  |  |  |  |  |
| Per SD increase |  | 1.37 (1.01-1.86) |  |  | 1.18 (0.87-1.59) |  |  | 1.17 (0.82-1.68) |
| Q1 | 49 | 1.00 |  | 48 | 1.00 |  | 27 | 1.00 |
| Q2 | 41 | 0.93 (0.62-1.41) |  | 43 | 0.86 (0.57-1.31) |  | 28 | 0.94 (0.55-1.59) |
| Q3 | 54 | 1.35 (0.91-1.99) |  | 57 | 1.13 (0.77-1.66) |  | 40 | 1.19 (0.73-1.94) |
| Q4 | 42 | 1.24 (0.82-1.88) |  | 54 | 1.07 (0.72-1.59) |  | 46 | 1.13 (0.70-1.82) |
| Ptrend |  | 0.123 |  |  | 0.450 |  |  | 0.440 |
| Pinteraction |  | 0.899 | | | | | | |
| **FT (ng/mL)\*** |  |  |  |  |  |  |  |  |
| Per SD increase |  | 1.42 (1.06-1.91) |  |  | 1.22 (0.91-1.64) |  |  | 1.18 (0.82-1.70) |
| Q1 | 46 | 1.00 |  | 34 | 1.00 |  | 23 | 1.00 |
| Q2 | 56 | 1.34 (0.91-1.98) |  | 41 | 1.11 (0.70-1.75) |  | 13 | 0.56 (0.28-1.11) |
| Q3 | 38 | 1.24 (0.80-1.91) |  | 59 | 1.48 (0.97-2.26) |  | 33 | 0.91 (0.54-1.56) |
| Q4 | 34 | 1.84 (1.17-2.87) |  | 53 | 1.37 (0.88-2.11) |  | 58 | 1.00 (0.62-1.63) |
| Ptrend |  | 0.020 |  |  | 0.081 |  |  | 0.458 |
| Pinteraction |  | 0.452 | | | | | | |
| **SHBG (nmol/L)\*** |  |  |  |  |  |  |  |  |
| Per SD increase |  | 0.75 (0.59-0.95) |  |  | 0.99 (0.78-1.25) |  |  | 0.94 (0.72-1.24) |
| Q1 | 21 | 1.00 |  | 52 | 1.00 |  | 63 | 1.00 |
| Q2 | 44 | 1.01 (0.60-1.71) |  | 52 | 0.90 (0.61-1.32) |  | 42 | 1.13 (0.76-1.67) |
| Q3 | 48 | 0.72 (0.43-1.20) |  | 53 | 1.02 (0.69-1.49) |  | 17 | 0.83 (0.48-1.42) |
| Q4 | 62 | 0.65 (0.39-1.07) |  | 30 | 0.78 (0.50-1.23) |  | 8 | 0.83 (0.40-1.74) |
| Ptrend |  | 0.017 |  |  | 0.462 |  |  | 0.526 |
| Pinteraction |  | 0.394 | | | | | | |

**Table S2:** Association between sex steroid hormones, SHBG and risk of ductal carcinoma in situ of the breast by BMI categories among postmenopausal women in the UK Biobank

* Abbreviations: FT= free testosterone; SHBG= sex hormone binding globulin
* Models were adjusted for age at recruitment (years; continuous), socioeconomic status (based on quintiles of Townsend deprivation index), ethnicity (European descent; other); age at menarche (years; <12, 12-13, 14+, missing), parity/age at first live birth (nulliparous, age at first birth ≥25 and parity<3, age at first birth ≥25 and parity≥3, age at first birth ≤25 and parity<3, age at first birth ≤25 and parity≥3, missing), mammogram screening (yes/no), age at menopause (years; <45, 45-52, >52, missing), HRT use (yes/no), family history of breast cancer in a first degree relative (yes/no/missing), physical activity (continuous, Mets-hr/week), alcohol consumption (never, >0-<1/week, 1-2 times/week; 3-4 times/week; daily or almost daily). For analyses involving women of European descent, the models were not adjusted for ethnicity.
* \*Cut-points: testosterone: ≤0.70, 0.71-0.98, 0.99-1.33, >1.33; FT: ≤0.0024, 0.0025-0.0036; 0.0037-0.0052, >0.0052; SHBG: ≤38.85, >38.85-54.07, >54.07-72.87, >72.87 for Q1, Q2, Q3 and Q4, respectively

**Table S3:** Association between sex steroid hormones, SHBG and risk of ductal carcinoma in situ of the breast by waist circumference categories among postmenopausal women in the UK Biobank

|  | <88cm | |  | ≥88cm | |
| --- | --- | --- | --- | --- | --- |
|  | No. of cases | HR (95% CI) |  | No. of cases | HR (95% CI) |
| **Testosterone (nmol/L)\*** |  |  |  |  |  |
| Per SD increase |  | 1.21 (0.96-1.54) |  |  | 1.32 (0.99-1.77) |
| Q1 | 80 | 1.00 |  | 44 | 1.00 |
| Q2 | 69 | 0.89 (0.64-1.23) |  | 43 | 0.95 (0.62-1.45) |
| Q3 | 86 | 1.15 (0.85-1.56) |  | 65 | 1.37 (0.93-2.01) |
| Q4 | 75 | 1.11 (0.81-1.52) |  | 67 | 1.25 (0.85-1.84) |
| Ptrend |  | 0.272 |  |  | 0.100 |
| Pinteraction | 0.530 | | | | |
| **FT (ng/mL)\*** |  |  |  |  |  |
| Per SD increase |  | 1.21 (0.96-1.52) |  |  | 1.44 (1.07-1.93) |
| Q1 | 69 | 1.00 |  | 34 | 1.00 |
| Q2 | 85 | 1.25 (0.91-1.72) |  | 25 | 0.73 (0.44-1.23) |
| Q3 | 79 | 1.37 (0.99-1.90) |  | 51 | 1.11 (0.71-1.71) |
| Q4 | 58 | 1.38 (0.97-1.96) |  | 87 | 1.35 (0.91-2.02) |
| Ptrend |  | 0.047 |  |  | 0.025 |
| Pinteraction | 0.593 | | | | |
| **SHBG (nmol/L)\*** |  |  |  |  |  |
| Per SD increase |  | 0.93 (0.77-1.13) |  |  | 0.83 (0.67-1.03) |
| Q1 | 41 | 1.00 |  | 95 | 1.00 |
| Q2 | 73 | 1.08 (0.74-1.59) |  | 65 | 1.01 (0.74-1.39) |
| Q3 | 90 | 1.04 (0.72-1.51) |  | 28 | 0.70 (0.46-1.06) |
| Q4 | 88 | 0.86 (0.59-1.25) |  | 12 | 0.57 (0.31-1.05) |
| Ptrend |  | 0.241 |  |  | 0.028 |
| Pinteraction | 0.135 | | | | |

* Abbreviations: FT= free testosterone; SHBG= sex hormone binding globulin
* Models were adjusted for age at recruitment (years; continuous), socioeconomic status (based on quintiles of Townsend deprivation index), ethnicity (European descent; other); age at menarche (years; <12, 12-13, 14+, missing), parity/age at first live birth (nulliparous, age at first birth ≥25 and parity<3, age at first birth ≥25 and parity≥3, age at first birth ≤25 and parity<3, age at first birth ≤25 and parity≥3, missing), mammogram screening (yes/no), age at menopause (years; <45, 45-52, >52, missing), HRT use (yes/no), family history of breast cancer in a first degree relative (yes/no/missing), physical activity (continuous, Mets-hr/week), alcohol consumption (never, >0-<1/week, 1-2 times/week; 3-4 times/week; daily or almost daily). For analyses involving women of European descent, the models were not adjusted for ethnicity.
* \*Cut-points: testosterone: ≤0.70, 0.71-0.98, 0.99-1.33, >1.33; FT: ≤0.0024, 0.0025-0.0036; 0.0037-0.0052, >0.0052; SHBG: ≤38.85, >38.85-54.07, >54.07-72.87, >72.87 for Q1, Q2, Q3 and Q4, respectively

**Table S4: Associations between sex steroid hormone concentration and risk of ductal carcinoma in situ of the breast among premenopausal women from the UK Biobank (excluding cases with a diagnosis within two years of recruitment)**

| **Markers** | HR (95% CI)a |
| --- | --- |
| **Estradiol (pmol/L)b** |  |
| 1st tertile | 1.00 |
| 2nd tertile | 1.09 (0.70-1.69) |
| 3rd tertile | 1.40 (0.94-2.10) |
| **P trend** | 0.094 |
| **Unit increase in estradiol concentrationc** | 1.31 (0.97-1.77) |
| **FE2 (pg/mL)b** |  |
| 1st tertile | 1.00 |
| 2nd tertile | 1.14 (0.71-1.85) |
| 3rd tertile | 1.55 (1.00-2.42) |
| **P trend** | 0.050 |
| Unit increase in FE2 concentrationc | 1.44 (1.03-2.03) |
| **Testosterone (nmol/L)b** |  |
| 1st tertile | 1.00 |
| 2nd tertile | 1.24 (0.85-1.83) |
| 3rd tertile | 0.97 (0.64-1.46) |
| **P trend** | 0.900 |
| Unit increase in testosterone concentrationc | 1.38 (0.90-2.11) |
| **FT (pg/mL)** |  |
| 1st tertile | 1.00 |
| 2nd tertile | 1.11 (0.73-1.69) |
| 3rd tertile | 0.93 (0.59-1.48) |
| **P trend** | 0.795 |
| Unit increase in FT concentrationc | 1.27 (0.84-1.91) |
| **SHBG (nmol/L)b** |  |
| **Per unit increase** |  |
| 1st tertile | 1.00 |
| 2nd tertile | 1.04 (0.66-1.64) |
| 3rd tertile | 1.09 (0.68-1.75) |
| **P trend** | 0.706 |
| Unit increase in SHBG concentrationc | 1.19 (0.89-1.59) |

* Abbreviation: FE2= free estradiol; FT= free testosterone; SHBG= sex hormone binding globulin
* aAdjusted for age at recruitment (years; continuous), socioeconomic status (based on quintiles of Townsend deprivation index), age at menarche (<12, 12-13, 14+, missing), parity/age at first live birth (nulliparous, age at first birth ≥25 and parity<3, age at first birth ≥25 and parity≥3, age at first birth ≤25 and parity<3, age at first birth ≤25 and parity≥3, missing), mammogram screening (yes/no), family history of breast cancer in a first degree relative (yes/no/missing), BMI (<18.5, 18.5-24.9, 25.0-29.9, ≥30kg/m2), physical activity (continuous, Mets-hr/week), alcohol consumption (never, >0-<1/week, 1-2 times/week; 3-4 times/week; daily or almost daily), menstrual cycle phase.
* bCut-points: estradiol: ≤218.9, 219.0-448.3, >448.3; FE2: ≤1.28, 1.29-2.65, >2.65; testosterone: ≤0.65, 0.65-0.98, >0.98; FT: ≤2.72, 2.73-4.65, >4.65; SHBG: ≤50.5, 50.6-75.4, >75.4 for T1, T2 and T3, respectively
* cequivalent to a doubling in hormone/SHBG concentration

**Table S5: Associations between serum sex steroid hormones, SHBG and risk of ductal carcinoma in situ of the breast among postmenopausal women in the UK Biobank (excluding cases with a diagnosis within two years of recruitment)**

| **Markers** | HR (95% CI)a |
| --- | --- |
| **Testosterone (nmol/L)b** |  |
| 1st quartile | 1.00 |
| 2nd quartile | 0.95 (0.70-1.30) |
| 3rd quartile | 1.33 (1.00-1.77) |
| 4th quartile | 1.33 (0.99-1.77) |
| **P trend** | 0.010 |
| Unit increase in testosterone concentrationc | 1.39 (1.12-1.73) |
| **FT (pg/mL)b** |  |
| 1st quartile | 1.00 |
| 2nd quartile | 1.12 (0.81-1.55) |
| 3rd quartile | 1.42 (1.04-1.95) |
| 4th quartile | 1.70 (1.25-2.32) |
| **P trend** | <0.001 |
| Unit increase in FT concentrationc | 1.43 (1.16-1.78) |
| **SHBG (nmol/L)b** |  |
| 1st quartile | 1.00 |
| 2nd quartile | 1.04 (0.78-1.37) |
| 3rd quartile | 0.90 (0.66-1.21) |
| 4th quartile | 0.68 (0.49-0.96) |
| **P trend** | 0.022 |
| Unit increase in SHBG concentrationc | * 1. (0.74-1.04) |

* Abbreviation: FT= free testosterone; SHBG= sex hormone binding globulin
* aAdjusted for age at recruitment (years; continuous), socioeconomic status (based on quintiles of Townsend deprivation index), age at menarche (<12, 12-13, 14+, missing), parity/age at first live birth (nulliparous, age at first birth ≥25 and parity<3, age at first birth ≥25 and parity≥3, age at first birth ≤25 and parity<3, age at first birth ≤25 and parity≥3, missing), mammogram screening (yes/no), family history of breast cancer in a first degree relative (yes/no/missing), BMI (<18.5, 18.5-24.9, 25.0-29.9, ≥30kg/m2), physical activity (continuous, Mets-hr/week), alcohol consumption (never, >0-<1/week, 1-2 times/week; 3-4 times/week; daily or almost daily), menstrual cycle phase.
* bCut-points: testosterone: ≤0.50, 0.51-0.86, 0.87-1.24, >1.24; FT: ≤1.68, 1.69-3.09; 3.10-4.84, >4.84; SHBG: ≤38.82, >38.83-54.02, >54.03-72.79, >72.79 for Q1, Q2, Q3 and Q4, respectively
* cequivalent to a doubling in hormone/SHBG concentration

Excluded 15,019 women with no exposure information; 29,785 women<55 years with unknown menopausal status whose hysterectomy or oophorectomy status was unknown or who had missing information on days since last period were excluded from the study; 29,350 current HRT/OC users or with unknown HRT/OC; 15,150 prevalent cancer cases (except non-melanoma skin cancer (ICD-10 C44)); 163 with lobular carcinoma in situ

273,402

Analytic cohort

N= 182,935

Non-cases: 182,218

DCIS cases= 717

**Figure S1**: Summary of analytic cohort