OUTLINE OF PRESENTATION

• Epidemiology of endometrial cancer
• Purpose, methods, and findings of EDGE Study
• Findings on variants in estrogen biosynthesis genes
  – review of literature on association of genotypes and serum hormone levels
  – genotypes and risk of endometrial cancer from EDGE Study
OUTLINE OF PRESENTATION

• Epidemiology of endometrial cancer
• Purpose, methods, and findings of EDGE Study
• Findings on variants in estrogen biosynthesis genes
  – review of literature on association of genotypes and serum hormone levels
  – genotypes and risk of endometrial cancer from EDGE Study
GYNECOLOGIC CANCERS IN THE U.S.

- Uterus: 41,200 cases per year
- Ovary: 20,180 cases per year
- Cervix: 9,710 cases per year

ACS: Cancer Facts and Figures 2006
INCIDENCE OF ENDOOMETRIAL CANCER

Incidence per 100,000 women age 30-74

Racial/Ethnic Group

Overall, White, Black, Hispanic

Sherman et al., JNCI 2005
INCIDENCE OF ENDOMETRIAL CANCER ADJUSTED FOR HYSSTERECTOMY

Sherman et al., JNCI 2005
A DISEASE OF POST-MENOPAUSAL WOMEN

Incidence per 100,000 women*

*Corpus & Uterus, NOS Cancer (Invasive)

SEER Cancer Statistics Review 1975-2003
SEER 17 areas. Rates are per 100,000 and are adjusted to the 2000 US Std Population
NUMBER OF OLDER WOMEN IS GROWING RAPIDLY

MORTALITY FROM ENDOMETRIAL CANCER

ACS: Cancer Facts & Figures 2006

Number of deaths per year

- Uterus: 7,350
- Ovary: 15,310
- Cervix: 3,700
HORMONES AS RISK FACTORS

• Unifying risk factor: Long-term exposure to estrogens unopposed by progesterone
  – Estrogen has a proliferative effect on endometrial tissue
  – Progesterone counteracts effects of estrogen
  – Also some evidence that estrogen may be directly carcinogenic
SOURCES OF ESTROGENS

- Unopposed estrogen replacement therapy
- Excess weight
INCIDENCE 1975-2003
Corpus & Uterus, NOS, age ≥ 50

*Age-adjusted to 2000 U.S Standard population
EXCESS WEIGHT

• Pre-menopausal women:
  – excess weight leads to irregular cycles and lack of progesterone

• Post-menopausal women:
  – main source of estrogen is conversion of androstenedione to estrone in adipose tissue

• Heavier women also have lower levels of sex-hormone binding globulin
WOMEN ARE GETTING HEAVIER

Centers for Disease Control & Prevention, NCHS, NHES & NHNES . Health United States, 2005
OTHER “LIFESTYLE” RISK FACTORS

- No / few children
- Early menarche / late menopause
- No oral contraceptive use
- Not smoking
- Diet?
- Physical activity?
- Diabetes?
GENETIC SUSCEPTIBILITY

- Endometrial cancer is rare even in women with risk factors

- Do differences in genetic make-up help explain why some women get disease and others do not?
OUTLINE OF PRESENTATION

• Epidemiology of endometrial cancer
• Purpose, methods, and findings of EDGE Study
• Findings on variants in estrogen biosynthesis genes
  – review of literature on association of genotypes and serum hormone levels
  – genotypes and risk of endometrial cancer from EDGE Study
EDGE STUDY

SPECIFIC AIM

• To investigate the role of weight, diet, and genetic susceptibility in risk of endometrial cancer
STUDY PARTICIPANTS

- **487 Cases**: endometrial cancer up to one year before contact (2001-2005)
- **467 Controls**: no hysterectomy; matched to cases by age group
- Aged 21 and over
- Resident of selected counties in northern NJ
- Speak English or Spanish
- Able to sign informed consent
New Jersey
LOCATING CASES

- Rapid case ascertainment: NJDHSS representative reviewed pathology records at hospitals
- Physician contacted and approval given
- Patient contacted to obtain informed consent and for interview
LOCATING CONTROLS

Three sources of controls:

1. Random digit dialing for women <65
2. Medicare lists for women aged ≥65
3. Neighborhood sampling for women aged ≥60
MEASUREMENTS

• Main questionnaire
  – Known and potential risk factors
• Diet questionnaire
  – Block food frequency
  – Supplement for phytoestrogens
• Mouthwash sample for DNA (87%)
• Hip/waist measurement
• Genotyping of 10 variants in 7 genes
• Centralized pathology review at MSKCC
LITERATURE REVIEW

• Studies investigating relation between genotypes of genes in estrogen biosynthesis pathway and serum hormone levels
  – Seven genes
  – Serum levels of progesterone, androgens, estrogens
• In normal women
• Summarized these as a HuGE Review
## RISK FACTORS

<table>
<thead>
<tr>
<th></th>
<th>Cases (430)</th>
<th>Controls (402)</th>
<th>Odds ratio* (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Mean age (SD)</strong></td>
<td>61.9 (9.6)</td>
<td>64.8 (11.0)</td>
<td></td>
</tr>
<tr>
<td><strong>Body Mass Index</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Normal (&lt;25)</td>
<td>26%</td>
<td>47%</td>
<td>1</td>
</tr>
<tr>
<td>Overweight (25-&lt;30)</td>
<td>27</td>
<td>31</td>
<td>1.2 (1.2-2.3)</td>
</tr>
<tr>
<td>Obese (30-&lt;34)</td>
<td>16</td>
<td>16</td>
<td>1.8 (1.2-2.8)</td>
</tr>
<tr>
<td>Very obese(≥35)</td>
<td>30</td>
<td>6</td>
<td>7.8 (4.8-12.6)</td>
</tr>
<tr>
<td><strong>Mean BMI (SD)</strong></td>
<td>31.1 (8.8)</td>
<td>26.0 (5.4)</td>
<td></td>
</tr>
</tbody>
</table>

*adjusted for age
# RISK FACTORS

<table>
<thead>
<tr>
<th></th>
<th>Cases (345)</th>
<th>Controls (301)</th>
<th>Odds ratio* (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Parity</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0-1</td>
<td>35%</td>
<td>23%</td>
<td>1</td>
</tr>
<tr>
<td>2</td>
<td>34</td>
<td>36</td>
<td>0.62 (0.43-0.87)</td>
</tr>
<tr>
<td>&gt;= 3</td>
<td>30</td>
<td>41</td>
<td>0.52 (0.36-0.74)</td>
</tr>
<tr>
<td><strong>Age at last birth</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nulliparous</td>
<td>20%</td>
<td>12%</td>
<td>1</td>
</tr>
<tr>
<td>&lt;35</td>
<td>68</td>
<td>66</td>
<td>0.64 (0.43-0.95)</td>
</tr>
<tr>
<td>&gt;=35</td>
<td>12</td>
<td>22</td>
<td>0.38 (0.23-0.63)</td>
</tr>
</tbody>
</table>

*adjusted for age
## RISK FACTORS

<table>
<thead>
<tr>
<th>Age at menarche</th>
<th>Cases (430)</th>
<th>Controls (402)</th>
<th>Odds ratio* (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>≥13</td>
<td>57%</td>
<td>49%</td>
<td>1</td>
</tr>
<tr>
<td>&lt;13</td>
<td>43</td>
<td>51</td>
<td>1.3 (1.0-1.7)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Age at menopause</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;50</td>
<td>26%</td>
<td>33%</td>
<td>1</td>
</tr>
<tr>
<td>≥50</td>
<td>47</td>
<td>42</td>
<td>1.5 (1.0-1.7)</td>
</tr>
<tr>
<td>Post, age unknown</td>
<td>11</td>
<td>13</td>
<td>1.1 (0.7-1.8)</td>
</tr>
<tr>
<td>Premenopausal</td>
<td>16</td>
<td>12</td>
<td>1.2 (0.68-2.1)</td>
</tr>
</tbody>
</table>

*adjusted for age
# RISK FACTORS

<table>
<thead>
<tr>
<th></th>
<th>Cases (430)</th>
<th>Controls (402)</th>
<th>Odds ratio* (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>OC use</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Never used</td>
<td>54%</td>
<td>50%</td>
<td>1</td>
</tr>
<tr>
<td>Ever used</td>
<td>46</td>
<td>50</td>
<td>0.65 (0.48-0.87)</td>
</tr>
<tr>
<td><strong>Smoking</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Never smoked</td>
<td>55%</td>
<td>52%</td>
<td>1</td>
</tr>
<tr>
<td>Past smoker</td>
<td>39</td>
<td>39</td>
<td>0.95 (0.71-1.3)</td>
</tr>
<tr>
<td>Current smoker</td>
<td>6</td>
<td>9</td>
<td>0.52 (0.30-0.89)</td>
</tr>
</tbody>
</table>

*adjusted for age
OUTLINE OF PRESENTATION

• Epidemiology of endometrial cancer
• Purpose, methods, and findings of EDGE Study
• Findings on variants in estrogen biosynthesis genes
  – review of literature on association of genotypes and serum hormone levels
  – genotypes and risk of endometrial cancer from EDGE Study
ESTROGEN BIOSYNTHESIS

- Cholesterol
- Pregnenolone
- 17α-Hydroxypregnenolone
- Dehydroepiandrosterone
- Androstenedione
- Testosterone
- Estrone
- Estradiol
- Estrone Sulfate
- DHEAS
- 5-androstenediol
- 17α-Hydroxyprogesterone
- Progesterone
- 17α-Hydroxyprogesterone
- Androstenedione
- CYP11A1
- CYP17A1
- CYP17A1
- HSD3β
- HSD17β
- CYP19A1
- HSD17β
- HSD3β
ESTROGEN BIOSYNTHESIS

Cholesterol \[ \xrightarrow{\text{CYP11A1}} \] Pregnenolone \[ \xrightarrow{\text{CYP17A1}} \] 17\(\alpha\)-Hydroxypregnenolone \[ \xrightarrow{\text{CYP17A1}} \] Dehydroepiandrosterone \[ \xrightarrow{\text{HSD3} \beta} \] 5-androstenediol

Progesterone \[ \xrightarrow{\text{CYP17A1}} \] 17\(\alpha\)-Hydroxyprogesterone \[ \xrightarrow{\text{CYP17A1}} \] Androstenedione \[ \xrightarrow{\text{HSD3} \beta} \] Estrone

5-androstenediol \[ \xrightarrow{\text{HSD3} \beta} \] Estrone Sulfate

DHEAS

Testosterone

Estradiol
CYP11A1 GENOTYPE AND HORMONE LEVELS

- Pentanucleotide repeat in promoter region
  -528 [TTTTA]_n
  - 4 to 10 repeats, 4 most common
  - commonly studied in relation to polycystic ovarian syndrome
  - no relation between genotype and serum levels of progesterone, androgens or estrogens in normal women
  - 6 studies, 18 comparisons
CYP11A1 REPEAT AND RISK OF ENDOMETRIAL CANCER

<table>
<thead>
<tr>
<th></th>
<th>Cases (430)</th>
<th>Controls (402)</th>
<th>Odds ratio* (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>4 / 4</td>
<td>29%</td>
<td>28%</td>
<td>1</td>
</tr>
<tr>
<td>4 / longer</td>
<td>51</td>
<td>50</td>
<td>0.89 (0.65-1.24)</td>
</tr>
<tr>
<td>Both longer</td>
<td>24</td>
<td>22</td>
<td>1.05 (0.72-1.54)</td>
</tr>
</tbody>
</table>

*adjusted for age, BMI, menopausal status, ERT use
CONCLUSIONS
CYP11A1

• No association between this genotype and either serum hormone levels or risk of endometrial cancer
ESTROGEN BIOSYNTHESIS

Cholesterol → Pregnenolone → 17α-Hydroxypregnenolone → Dehydroepiandrosterone → 5-androstenediol

Progestrone → 17α-Hydroxyprogesterone → Androstenedione → Testosterone

Estradiol

CYP11A1

CYP17A1

CYP17A1

CYP19A1

HSD3β

HSD3β

HSD3β

HSD17β

HSD17β
CYP17A1 GENOTYPE AND HORMONE LEVELS

• CYP17A1 T-34C change in promoter region
  – No relationship with progesterone
    • 4 studies, 7 comparisons
  – No relationship with androgens
    • 9 studies, 20 comparisons
  – Most studies show no association of variant C allele with serum estrogen levels
    • Out of 12 studies (22 comparisons), 3 showed the C allele related to higher levels of estrogens but rest did not
EXAMPLES: STUDIES OF CYP17A1 T-34C GENOTYPE AND SERUM ANDROGENS

- N=1708
  Dunning, 2004
  Post-Menopausal
  p-Value 0.97

- N=440
  Haiman, 2001
  Post-Menopausal
  p-Value 0.5

- N=192
  Garcia-Closas, 2002
  Post-Menopausal
  p-Value 0.5

- N=171
  Tworoger, 2004
  Pre-Menopausal
  p-Value 0.20

EXAMPLES: STUDIES OF CYP17A1 T-34C GENOTYPE AND SERUM ANDROGENS

- N=1708
  Dunning, 2004
  Post-Menopausal
  p-Value 0.97

- N=440
  Haiman, 2001
  Post-Menopausal
  p-Value 0.5

- N=192
  Garcia-Closas, 2002
  Post-Menopausal
  p-Value 0.5

- N=171
  Tworoger, 2004
  Pre-Menopausal
  p-Value 0.20
POSITIVE STUDIES OF CYP17A1 T-34C GENOTYPE AND SERUM ESTROGENS

N=440
Haiman, 2001
Post-Menopausal

N=173
Hong, 2004
Post-Menopausal

N=83
Feigelson, 1998
Pre-Menopausal

POSITIVE STUDIES OF CYP17A1 T-34C GENOTYPE AND SERUM ESTROGENS
EXAMPLES: LATER STUDIES OF CYP17A1 T-34C GENOTYPE AND SERUM ESTROGENS

- N=1708, Dunning, 2004
- N=524, Travis, 2004
- N=220, Lurie, 2005
- N=171, Tworoger, 2004

Post-Menopausal

- p-Value 0.5
- p-Value 0.6
- p-Value 0.14
- p-Value 0.43

EXAMPLES: LATER STUDIES OF CYP17A1 T-34C GENOTYPE AND SERUM ESTROGENS
CYP17A1 T-34C GENOTYPE AND RISK OF ENDOMETRIAL CANCER

<table>
<thead>
<tr>
<th></th>
<th>Cases (429)</th>
<th>Controls (400)</th>
<th>Odds ratio* (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>TT</td>
<td>36%</td>
<td>31%</td>
<td>1</td>
</tr>
<tr>
<td>TC</td>
<td>50</td>
<td>52</td>
<td>0.75 (0.54-1.04)</td>
</tr>
<tr>
<td>CC</td>
<td>14</td>
<td>18</td>
<td>0.63 (0.40-0.99)</td>
</tr>
</tbody>
</table>

*adjusted for age, BMI, menopausal status, ERT use
CONCLUSIONS

CYP17A1

- CYP17A1 T/C variant does not seem to be related to hormone levels

- But does appear to be related to risk
  - C allele may be protective

- Consistent with several small studies
ESTROGEN BIOSYNTHESIS

- Cholesterol 
  - CYP11A1 
  - Pregnenolone 
  - CYP17A1 
  - 17α-Hydroxypregnenolone 
  - CYP17A1 
  - Dehydroepiandrosterone 
  - HSD3β 
  - 5-androstenediol 
  - HSD3β 
  - 17α-Hydroxyprogesterone 
  - CYP17A1 
  - Androstenedione 
  - HSD3β 
  - Testosterone 
  - HSD3β 
  - Estrone 
  - CYP19A1 
  - Estradiol 
  - Estrone Sulfate 
  - HSD17β 
  - DHEAS
CYP19A1 GENOTYPES

• aromatase converts androstenedione to estrone in adipose tissue, as well as converting testosterone to estradiol in ovarian granulosa cells.

• large area of this gene is in linkage disequilibrium; includes several variants:
  – tetranucleotide repeat \([\text{TTTA}]_n\) (7 to 13)
  – 3 base-pair deletion found only with 7 repeats
  – C/T change in exon 10 (5’ UTR)
CYP19A1 GENOTYPES AND HORMONE LEVELS

• No consistent association of estrogens or androgens with these polymorphisms
  – 12 studies, 69 comparisons

• Fairly consistent findings that variants are associated with increased ratios of estrogens to androgens
  – 6 studies, 11 comparisons
EXAMPLES: STUDIES OF CYP19A1 INS/DEL AND RATIO OF ESTRONE TO ANDROSTENEDIONE

N=1708
Dunning, 2004
Post-Menopausal

N=136
Baghaei, 2003
Post-Menopausal

p-Value 0.01
CYP19A1 3 BASE PAIR INS/DEL AND RISK OF ENDOMETRIAL CANCER

<table>
<thead>
<tr>
<th></th>
<th>Cases (429)</th>
<th>Controls (401)</th>
<th>Odds ratio* (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ins/Ins</td>
<td>44%</td>
<td>44%</td>
<td>1</td>
</tr>
<tr>
<td>Ins/Del</td>
<td>48</td>
<td>43</td>
<td>1.15 (0.84-1.57)</td>
</tr>
<tr>
<td>Del/Del</td>
<td>8</td>
<td>13</td>
<td>0.48 (0.28-0.81)</td>
</tr>
</tbody>
</table>

*adjusted for age, BMI, menopausal status, ERT use
CYP19A1 TTTAn GENOTYPE AND RISK OF ENDOMETRIAL CANCER

<table>
<thead>
<tr>
<th></th>
<th>Cases (428)</th>
<th>Controls (301)</th>
<th>Odds ratio* (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>7 / 7</td>
<td>27%</td>
<td>29%</td>
<td>1</td>
</tr>
<tr>
<td>7 / longer</td>
<td>50</td>
<td>47</td>
<td>1.0 (0.70-1.45)</td>
</tr>
<tr>
<td>Both longer</td>
<td>24</td>
<td>25</td>
<td>0.81 (0.54-1.22)</td>
</tr>
</tbody>
</table>

*adjusted for age, BMI, menopausal status, ERT use
CONCLUSIONS

CYP19A1

- CYP19A1 variants in linked sites appear to be associated with conversion of androgens to estrogens
- These variants also appear to be associated with risk
- Consistent with other smaller studies
SUMMARY

• Relatively large study
• Collected extensive data on risk factors
• Confirmed importance of weight, other factors
• One of first studies to look at genetic risk factors for endometrial cancer
• Focused on genes in estrogen biosynthesis pathway
• Variants in CYP17A1 and CYP19A1 likely to affect risk
NEXT STEPS

• Diet data
• Genes in hormone metabolism pathway
• Investigate whether effects of genetic variants are stronger in certain subgroups of women
• Pool data from this and other ongoing studies for adequate power for gene-gene and subgroup analysis
  – E2C2 Consortium
COLLABORATORS

• NJDHSS
  – Tara Blando, Betsy Kohler, Kevin Masterson, Lisa Roche, Helen Weiss, Homer Wilcox

• CINJ
  – Elisa Bandera, Dina Considine

• MSKCC
  – Sharon Bayuga, Shameka Faulkner, Irene Orlow, Katie Pulick, Louise Salant, Camelia Sima, Robert Soslow, Michelle Sriprasert, Diana Tomassi, Ann Zauber
  – Field staff: Silvia Brendel, June Kittredge, Mathilde Saxon, Elizabeth Ward, Doreen Wass, Kay Yoon