Shift Work and Breast Cancer: need for mechanisms

R Stevens
University of Connecticut
Breast Cancer Incidence/100,000 women/year age-adjusted

<table>
<thead>
<tr>
<th>REGION</th>
<th>Incidence</th>
</tr>
</thead>
<tbody>
<tr>
<td>North America</td>
<td>86.30</td>
</tr>
<tr>
<td>Australia/NZ</td>
<td>71.69</td>
</tr>
<tr>
<td>Temp. S. America</td>
<td>69.14</td>
</tr>
<tr>
<td>Northern Europe</td>
<td>68.31</td>
</tr>
<tr>
<td>Western Europe</td>
<td>67.35</td>
</tr>
<tr>
<td>Micro/Poly</td>
<td>51.73</td>
</tr>
<tr>
<td>Southern Europe</td>
<td>49.51</td>
</tr>
<tr>
<td>Trop. S. America</td>
<td>39.07</td>
</tr>
<tr>
<td>Eastern Europe</td>
<td>35.95</td>
</tr>
<tr>
<td>Caribbean</td>
<td>33.50</td>
</tr>
<tr>
<td>Southern Africa</td>
<td>31.46</td>
</tr>
<tr>
<td>Japan</td>
<td>28.61</td>
</tr>
<tr>
<td>Central America</td>
<td>25.46</td>
</tr>
<tr>
<td>Northern Africa</td>
<td>24.99</td>
</tr>
<tr>
<td>Western Asia</td>
<td>24.29</td>
</tr>
<tr>
<td>Melanesia</td>
<td>23.93</td>
</tr>
<tr>
<td>S.E. Asia</td>
<td>22.51</td>
</tr>
<tr>
<td>S. Central Asia</td>
<td>21.24</td>
</tr>
<tr>
<td>Western Africa</td>
<td>19.02</td>
</tr>
<tr>
<td>Eastern Africa</td>
<td>18.56</td>
</tr>
<tr>
<td>Other E. Asia</td>
<td>17.85</td>
</tr>
<tr>
<td>Middle Africa</td>
<td>13.64</td>
</tr>
<tr>
<td>China</td>
<td>11.77</td>
</tr>
</tbody>
</table>

25 years ago we knew more about the causes of breast cancer than we do today.

At that time, most of us believed the high-fat ‘Western’ diet explained the high risk.

But decades of intense research has so far failed to find an obvious connection.

So where are we now?
Electric Light
a hallmark of modern life

- **Our Evolutionary Past**
  - bright, full-spectrum days
  - dark nights

- **Modern Life**
  - dim, spectrum-restricted days inside buildings
  - lighted nights ('light pollution')
Light at Night - U.S. - the Present
Light at Night - U.S. - the Past

http://www.earthview.mars/safe/
Human Studies
Breast Cancer and ‘Light-at-Night’

- **Theory:** light-at-night alters hormones, increasing risk, and thereby explains some of the high risk in industrialized societies

- **Predictions (i.e., 'hypotheses'):**
  - shift workers at higher risk
  - blind women at lower risk
  - lighted bedrooms at night increase risk
  - long sleep lowers risk
November 13, 1987

Walter Willett, M.D.
Channing Laboratory
180 Longwood Avenue
Boston, MA 02115

Dear Walter:

We met once about 4 years ago when I was applying for a job in the department of Epidemiology. I admire your work, and appreciate your efforts in understanding the tools of epidemiology (e.g. food frequency questionnaires) as well as making advances in understanding disease causation.

My purpose in writing is to suggest a look at shift work and breast cancer in your nurse cohort. I wrote a little thing about electric power and breast cancer (Am J Epi, 1987;125:556), and a letter with Bob Hiatt about your alcohol study that is supposed to appear in New Engl J Med soon. The underlying notion to both is that disruption of pineal function, and consequent reduction in melatonin production might increase the turnover of the normal breast epithelial stem cells at risk, and result in the reduction of an oncostatic agent in serum and tissue (i.e. melatonin). Disruption of circadian rhythms by exposure to shift work might also have this effect. Your cohort of nurses undoubtedly offers a powerful data set for investigating this possibility. Such analyses would be complicated, I suspect, by association of shift work with age, parity, marital status, and the like.

Anyway, I hope to meet you again in the future.

Sincerely,

Richard Stevens
IARC: shift workers at higher risk


“On the basis of ‘limited evidence in humans for the carcinogenicity of shift-work that involves nightwork’, and ‘sufficient evidence in experimental animals for the carcinogenicity of light during the daily dark period (biological night)’, the Working Group concluded that ‘shift-work that involves circadian disruption is probably carcinogenic to humans’ (group 2A).”
Circadian Disruption and Cancer: Genes & Mechanisms

understanding will be crucial for intervention and mitigation
points of impact of CD

pre-initiation

initiation/promotion

progression

pre-initiation     e.g., early effects on mammary tissue development
initiation/promotion     e.g., CD affects on cyclin D1 expression
progression     e.g., oncostatic effects of melatonin

Moolgavkar, Day, Stevens, *JNCI*, 65:559-569, 1980
Circadian Disruption Example

all cells in tissue start here in BRCA1 hereditary cases

○ normal cell
Ø intermediate cell
⊗ cancer cell

A = normal tissue growth
B = intermediate lesion
C = clinical behavior

\[ \begin{align*}
\int_1 &= \text{oxidative stress} \\
\int_2 &= \text{DNA repair} \\
A &= \text{altered reproductive endocrinology} \\
B &= \text{altered cell cycle regulation (CLOCK)} \\
C &= \text{depleted melatonin}
\end{align*} \]

Moolgavkar, Day, Stevens, JNCI, 65:559-569, 1980
Phototransduction (retina)

Neuronal Signaling (SCN)

Neuroendocrine Transduction (pineal, pituitary, ovary)

Mammary Tissue (altered normal tissue development and/or enhanced tumor growth)

- melanopsin variants
- clock gene variants

- cell cycle regulatory genes
- melanotin, other hormones?
- melatonin receptor variants

Stevens, *Epidemiology*, 2005
Mechanism: Progression
Phototransduction (retina)

Neuronal Signaling (SCN)

Neuroendocrine Transduction (pineal, pituitary, ovary)

Mammary Tissue (altered normal tissue development and/or enhanced tumor growth)

- melanopsin ipRGCs
- melanopsin variants
- clock gene variants
- 9 core clock genes
- cell cycle regulatory genes
- melatonin, other hormones?
- melatonin receptor variants

Blask et al., Cancer Res, 2005
The Circadian Clock: clock-controlled genes

- 5-10% of all mammalian genes are clock controlled
- Among these are genes for the key regulators of cell-cycle progression and apoptosis (e.g., cyclins and caspases)
Circadian Genes and Cancer

"When you're thinking about something that you don't understand, you have a terrible, uncomfortable feeling called confusion."
- Richard Feynman, 1963
map of NYC subway system; tough for an out-of-towner
Circadian Loop

Positive (transcriptional activators?): CLOCK (or NPAS2) and BMAL1 are basic helix-loop-helix PAS-domain containing transcription factors that activate transcription of the Per and Cry genes.

Negative (transcriptional repressors?): The resulting PER and CRY proteins heterodimerize, translocate to the nucleus and interact with the CLOCK–BMAL1 complex to inhibit their own transcription. After a period of time, the PER–CRY repressor complex is degraded and CLOCK–BMAL1 can then activate a new cycle of transcription.

The entire cycle takes approximately 24 hours to complete

Takahashi et al., Nat Rev: Genet, October, 2008
# Epidemiology of Cancer and Circadian Gene Polymorphisms

(Yong Zhu and his student Aaron Hoffman, Yale)

<table>
<thead>
<tr>
<th>gene</th>
<th>cancer</th>
<th>finding</th>
<th>role in feedback loop; comment</th>
</tr>
</thead>
<tbody>
<tr>
<td>PER3 length (Zhu 2005)</td>
<td>breast</td>
<td>structural variants 5-peat/4-peat OR = 1.7</td>
<td>negative; predicted due to lark/owl</td>
</tr>
<tr>
<td>NPAS2 Ala394Thr (Zhu 2008)</td>
<td>breast</td>
<td>heteros/wt homozygotes OR = 0.61</td>
<td>positive; hetero advantage? role in DNA repair (Hoffman et al., Mol Cancer Res, 2008)</td>
</tr>
<tr>
<td>CRY2, 140147:G&gt;C NPAS2, Per1, Per3 (Chu 2008)</td>
<td>prostate</td>
<td>for CRY2, OR = 1.7 NS for others including Per3 structural</td>
<td>CRY2, negative;</td>
</tr>
<tr>
<td>gene</td>
<td>cancer</td>
<td>finding</td>
<td>comment</td>
</tr>
<tr>
<td>--------------</td>
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<td>--------------------------------</td>
<td>----------------------------------------------------------</td>
</tr>
<tr>
<td>CRY2 3 SNPs</td>
<td>NHL</td>
<td>ORs of 2.3, 2.4, and 3.0</td>
<td>MCF-7 functional analysis affects on immune function</td>
</tr>
<tr>
<td>(Hoffman 2009)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NPAS2 Ala394Thr</td>
<td>NHL</td>
<td>heteros/wt homozygotes OR = 0.69</td>
<td>differs from BrCa result in that homozygote variant also at reduced risk</td>
</tr>
<tr>
<td>(Zhu 2006)</td>
<td></td>
<td></td>
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<tr>
<td>CLOCK several variants</td>
<td>breast</td>
<td>ORs range from 0.78 to 2.25</td>
<td>stronger in ER-/PR-cancers</td>
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Are these gene findings replicable, and if so, what do they mean?

as with the larger area of GWAS looking for common but low penetrance SNPs for cancer causation, there is as yet no clear message for screening, treatment, or prevention

e.g., Easton (Nature 2007) GWASed 227,876 SNPs and found 5 to be highly significantly associated with breast cancer in a two stage case control study design: 4,398 cases in first stage and 21,860 in the second

odds ratios ranged from 1.08 to 1.23
Mechanism:
circadian gene function
Cyclin D1
(one tenuous thread; Stevens & Rea, 2001)

- Cyclin D1 over expressed in breast cancer (Arnold and Papanikolaou, 2005)
- maybe the CLOCK gene product affects cyclin D1 function and thereby risk (Stevens and Rea, 2001)
- CLOCK is also an enzyme that has histone acetyltransferase (HAT) activity and might affect cyclin D1 (Sahar and Sassone-Corsi, 2007)
- disruption and over-expression of CLOCK might increase breast cancer risk
- many other possibilities (Matsuo et al., Science, 2003; Haus & Smolensky, Cancer Cause Control, 2006)
“CLOCK in Breast Tumorigenesis”
(Hoffman et al., Cancer Research, 2010;70:1459-68)

- case-control study in CT (441 cases)
- 80 cases before adjuvant therapy
- hypomethylation strongly associated with risk
Thank You