Reframing tumor heterogeneity
gene mapping & precision treatments

Nicola J Camp, PhD
University of Utah
Common diseases are heterogeneous

- Locus
- Allelic
- Tumor

- Impacts
  - Gene discovery
  - Risk stratification
    - Prevention
    - Clinical management
  - Drug response – precision treatments
Tumor heterogeneity
Universal approach = highest impact

• Goal – approach relevant for research across the translational spectrum
  • Discover inherited susceptibility
  • Pre-clinical modeling
  • Clinical trials

• ‘Natural characteristics’ -- variability across patients
  • Gene expression
Breast tumors

• natural groupings
  • 2 epithelial cell lines
    • Luminal / ER
    • Basal (myoepithelial) KRT 5, 17
  • Erb

• Fine-t
  • Luminal-A
  • Luminal-B
  • HER2-enriched
  • Basal-like
  • Normal-like

Could intrinsic subtype be the key to inherited susceptibility?

BRCA1 carriers associated with the basal tumor subtype

Sørlie et al PNAS 2003
# Intrinsic subtypes in Utah high-risk pedigrees

<table>
<thead>
<tr>
<th>Pedigree</th>
<th>BC cases</th>
<th>Tumors</th>
</tr>
</thead>
<tbody>
<tr>
<td>1817</td>
<td>138</td>
<td>35</td>
</tr>
<tr>
<td>1822</td>
<td>159</td>
<td>31</td>
</tr>
<tr>
<td>1819</td>
<td>114</td>
<td>26</td>
</tr>
<tr>
<td>1808</td>
<td>112</td>
<td>24</td>
</tr>
<tr>
<td>1800</td>
<td>66</td>
<td>20</td>
</tr>
<tr>
<td>1818</td>
<td>111</td>
<td>20</td>
</tr>
<tr>
<td>1820</td>
<td>68</td>
<td>20</td>
</tr>
<tr>
<td>1821</td>
<td>81</td>
<td>18</td>
</tr>
<tr>
<td>1801</td>
<td>57</td>
<td>17</td>
</tr>
<tr>
<td>1812</td>
<td>43</td>
<td>17</td>
</tr>
<tr>
<td>1809</td>
<td>50</td>
<td>15</td>
</tr>
<tr>
<td><strong>TOTAL</strong></td>
<td><strong>243</strong></td>
<td></td>
</tr>
</tbody>
</table>

Melissa Cessna, MD

Phil Bernard, MD
General population Utah non-BRCA1/2 pedigrees

Sweeney et al. CEBP 2014
Single dimension: sufficient to describe a tumor?

- Intrinsic subtype = 1 categorical variable
- ROR = 1 quantitative score

- More complex?
- Does the data require more dimensions to describe variability?
Newton’s Color Theory, ca. 1665

Red Orange Yellow Green Blue Indigo Violet
Multiple quantitative dimensions

• Principal component analysis

Sweeney et al CEBP 2014
PC1, PC2, and PC5 quantitative framework for subtypes
Any evidence for familial clustering?

<table>
<thead>
<tr>
<th>Pedigree</th>
<th>n</th>
<th>PC3</th>
<th></th>
<th>PC5</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>1800</td>
<td>20</td>
<td>p&lt;10^{-12}</td>
<td>ns</td>
<td>p=7.6 x 10^{-8}</td>
<td>-1.609</td>
</tr>
<tr>
<td>1801</td>
<td>17</td>
<td>ns</td>
<td>2.03</td>
<td>ns</td>
<td>-2.311</td>
</tr>
<tr>
<td>1808</td>
<td>24</td>
<td>0.006</td>
<td>2.76</td>
<td>0.0129</td>
<td>-3.674</td>
</tr>
<tr>
<td>1809</td>
<td>15</td>
<td>0.0778</td>
<td>3.13</td>
<td>ns</td>
<td>-2.468</td>
</tr>
<tr>
<td>1812</td>
<td>17</td>
<td>ns</td>
<td>2.42</td>
<td>ns</td>
<td>-0.305</td>
</tr>
<tr>
<td>1817</td>
<td>35</td>
<td>0.0076</td>
<td>5.55</td>
<td>0.0133</td>
<td>-3.508</td>
</tr>
<tr>
<td>1818</td>
<td>20</td>
<td>0.0147</td>
<td>3.72</td>
<td>ns</td>
<td>-0.230</td>
</tr>
<tr>
<td>1819</td>
<td>26</td>
<td>0.201</td>
<td>2.58</td>
<td>ns</td>
<td>-1.437</td>
</tr>
<tr>
<td>1820</td>
<td>20</td>
<td>ns</td>
<td>1.60</td>
<td>0.0033</td>
<td>-4.352</td>
</tr>
<tr>
<td>1821</td>
<td>18</td>
<td>0.1014</td>
<td>2.92</td>
<td>ns</td>
<td>-1.735</td>
</tr>
<tr>
<td>1822</td>
<td>31</td>
<td>0.00004</td>
<td>5.54</td>
<td>ns</td>
<td>-0.184</td>
</tr>
</tbody>
</table>

Comparing to the population (LACE/Pathways). Bonferroni correction already performed.

Potentially promising for gene mapping!
12q15

p=4.0×10⁻⁹, LOD=7.2

Madsen et al. CEBP 2018
Polygenic risk?

- 38 SNPs (from GWAS hits)
- Multivariate $r^2=0.93$

- Precision risk?
- Prevention trials?
Gene expression – prognosis and treatment modality
Poor prognosis disease. Informative for treatment modality

Precision therapy elusive  Unable to identify interactions with specific drugs
PC1 is recurrence risk... prognosis
In a best fitting model:
- PC1 \( (p=1.3 \times 10^{-6}) \)
- Nodal status \( (p=2.7 \times 10^{-4}) \)
- Tumor size \( (p=1.0 \times 10^{-3}) \)
- PC5 \( (p=3.0 \times 10^{-3}) \)
- Overall fit \( p=2.5 \times 10^{-12} \)

**PC1 and Disease-Free Survival**

- Logrank test:
  - \( \text{chisq} = 24.7 \) (3 df)
  - \( p = 1.74 \times 10^{-5} \)

- \( n=819 \)

- Number at risk

<table>
<thead>
<tr>
<th>Strata</th>
<th>Q1</th>
<th>Q2</th>
<th>Q3</th>
<th>Q4</th>
</tr>
</thead>
<tbody>
<tr>
<td>Q1</td>
<td>205</td>
<td>194</td>
<td>177</td>
<td>163</td>
</tr>
<tr>
<td>Q2</td>
<td>205</td>
<td>187</td>
<td>162</td>
<td>146</td>
</tr>
<tr>
<td>Q3</td>
<td>204</td>
<td>174</td>
<td>140</td>
<td>127</td>
</tr>
<tr>
<td>Q4</td>
<td>205</td>
<td>155</td>
<td>130</td>
<td>122</td>
</tr>
</tbody>
</table>

- Survival probability

- Years: 0, 2.5, 5, 7.5, 10, 12.5
PC5 and clinical-pathological characteristics
Main trial result: paclitaxel extends disease-free interval

FEC: 80% survive to ~3 years
FEC-P: 80% survive to ~5 years

DFS by treatment arm

Strata: FEC-P, FEC

(b) Taxane-plus-anthracycline-based regimen (taxane courses given alone) vs MORE (but < doubled) non-taxane cytotoxic chemotherapy

00S WSG/AGO AM-02
4E0cE0; 4D10 vs 0(F00cE0cE00)
†D100x E360 vs E360
E440
42/2003 59/10000 -9.8 24.4
95T HORG Greece
4D10; 4E0cE vs 0(F00cE1cE00)
†D100x E360 vs E440
158/105 145/23 -8.5 43.2
90E FinHer/FBCG 00-01
3D100; 3FEC vs 3Vfb2b; 3(F00cE0cE00)
†D100x E180 vs E180
147/105 145/23 -14.8 23.6
98D1 BIG 02-08
3A15; 3D10; 3CMF-4 vs 4A15; 3(C00c114M023X50203)4
†D100x A270 vs A270
A0
197/4376 245/4200 -22.0 65.0
99K GEICAM 0606 Spain
4FEC; 8(P100x1 vs 0(F00cE0cE00)
†P100x E360 vs E440
E180
73/2365 134/2368 -28.5 46.5
97R HE1097 Greece
3E0a3; 3P200; 3CMF-2 vs 4(E100x10); 4(C00c114M023X50203)4
†P100x E360 vs E440
E110
90/100 100/1233 -3.5 42.3
99F1 NCIC MA.21
6(E120cC00); 3P200; 4(E100x10); 6(C00c114M023X50203)4
†P100x E720 vs E720
E0
74/1798 82/1809 -3.5 36.8

(b) subtotal

0.75 (SE 0.05)
reduction
2p < 0.0001

Number at risk

<table>
<thead>
<tr>
<th>Strata</th>
<th>FEC-P</th>
<th>FEC</th>
</tr>
</thead>
<tbody>
<tr>
<td>0-2.5</td>
<td>402</td>
<td>417</td>
</tr>
<tr>
<td>2.5-5</td>
<td>357</td>
<td>353</td>
</tr>
<tr>
<td>5-7.5</td>
<td>315</td>
<td>294</td>
</tr>
<tr>
<td>7.5-10</td>
<td>291</td>
<td>267</td>
</tr>
<tr>
<td>10-12.5</td>
<td>210</td>
<td>206</td>
</tr>
<tr>
<td>12.5+</td>
<td>4</td>
<td>4</td>
</tr>
</tbody>
</table>
FEC: 80% survive ~3 years

FEC-P: 80% survive ~7 years

Dimension x Drug interactions?

PC3 & PC4 High (48.6% patients)

FEC & FEC-P: 80% survive to ~3 years

Logrank test:
chisq = 0.00457 (1 df)
p = 0.946

n=357

FEC-P: 73%
FEC: 73%

FEC-P: 68%
FEC: 65%

FEC-P: 60%
FEC: 62%

PC3 and PC4
Significant interactions with paclitaxel
Camp et al. BCRT 2019
Patient-derived organoid models

• Preliminary utility as a framework for drug screens

Bryan Welm, PhD
Quantitative tumor dimension framework

- Dimensions add resolution
  - PC1-PC2-PC4 provides framework with increased resolution for categorical intrinsic subtypes

- Inherited susceptibility
  - Pedigree studies, PC3 and PC5 heritable
  - GWAS polygenic studies

- Precision risk
  - Precision prevention

- Survival analysis: Prognosis
  - PC1 and PC5 associated with prognosis
  - PC1 associated with risk of recurrence –Prosigna, Oncotype DX, Mammaprint

- Precision therapeutics
  - PC3 and PC4 proposed drug interaction with paclitaxel

- Preclinical models
  - Dimensions preserved in organoid and xenograft models
Thank you!

University of Utah / HCI
• Phil Bernard
• Rachel Factor
• Bryan Welm
• Carol Sweeney
• Myke Madsen
• John Gardner
• Rob Sargent
• Brandt Jones

Intermountain, UT
• Melissa Cessna
• Stacey Knight
• Kerry Rowe

Kaiser Permanente, CA
• Larry Kushi
• Bette Caan

GEICAM, Spain
• Miguel Martín
• Rosalia Caballero
• Jesús Herranz Valera

Support
• R01 CA163353 (breast)
• UGP (breast)
• CGM (methods)
• CCTS pilot (MM)