OVERVIEW OF GENE EXPRESSION-BASED TESTS IN EARLY BREAST CANCER

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Disclosures

• Advisory role for Nanostring Technologies
Breast Cancer Gene Expression Profiling Tests Include:

1. The PAM50 Intrinsic Subtypes: LumA, LumB, Basal-like, HER2-enriched (Wallden BMC 2015)
2. The PAM50 Risk of Recurrence (ROR) (Wallden BMC 2015)
3. OncotypeDX Recurrence Score (Paik et al., NEJM, 2004)
4. Mammaprint (van de Vijver et al., NEJM, 2002)
5. EndoPredict (Filipits et al., CCR 2011)
7. Genomic Grade Index (Sotiriou et al. JNCI 2006)
Clinical Implementation of Drugs

**DRUG**

- Phase I
- Phase II
- Phase III

**BIOMARKER**

- Analytical Validation
- Clinical Validation
- Clinical Utility

READY FOR PRIME TIME
Clinical Implementation of Biomarkers

**BIOMARKER**

**Analytical Validation**
- Accuracy and Precision in measurement of analyte.
- Robustness.

**Clinical Validation**
- Correlation of score/classifier with clinical state or outcome.
  - e.g. biomarker identifies 2 prognostic groups.

**Clinical Utility**
- Actionable (could affect treatment).
- Use results for patient benefit.
### Evaluation of Prognostic and Predictive Biomarkers. Levels of Evidence

<table>
<thead>
<tr>
<th>Level</th>
<th>Characteristics</th>
<th>Use?</th>
</tr>
</thead>
</table>
| I     | • Prospective Clinical Trial (PCT) designed to test marker  
       • Consistent results from ≥ 2 PCTs not designed to test marker, but biomarker is tested in preplanned manner on both trials | Yes |
| II    | • 1 PCT not designed for marker; biomarker analyses preplanned  
       • ≥ 2 consistent results from Prospective Observational Cohorts (POC); preplanned analyses | Usually no |
| III   | • 1 analysis POC; biomarker preplanned analyses | No |
| IV-V  | • Unplanned biomarker analyses  
       • Retrospectively ascertained cohorts | No |

Richard M. Simon, Soonmyung Paik, Daniel F. Hayes, JNCI 2009
Are they analytically validated?

YES
Customized mini-array reproducibility vs. original Agilent Arrays

71/78 = 91%  Kappa Value = 0.82

Customized mini-array reproducibility

Correlation Coeff. > 0.99

FDA 510(k)

RS (0-100) SD < 2.0 units
## MAMMAPRINT FFPE Assay

**FDA 510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION**

### FF vs FFPE

#### Table 14 Second Validation Dataset – Agreement Analysis

<table>
<thead>
<tr>
<th></th>
<th>Point Estimate</th>
<th>N</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>PPA</strong></td>
<td>86.6%</td>
<td>136/157</td>
<td>80.4% - 91.1%</td>
</tr>
<tr>
<td><strong>NPA</strong></td>
<td>91.5%</td>
<td>172/188</td>
<td>86.7% - 94.7%</td>
</tr>
<tr>
<td><strong>Overall Concordance</strong></td>
<td>89.3%</td>
<td>308/345</td>
<td>85.4% - 92.2%</td>
</tr>
</tbody>
</table>

PPA: positive percent agreement
NPA: negative percent agreement
Analytical Validation of Decentralized Gene Expression-based tests (EndoPredict and PROSIGNA)

- EP (0-15) SD (from RNA) = 0.14
- Subtype concordance= 97-100%
- ROR (0-100) SD (from RNA) = 0.82
- ROR (0-100) SD (from tissue) = 2.90

Kronenwett et al. BMC Cancer 2012

Torsten et al. BMC Genomics 2014
Can these tests help us identify patients who do not need adjuvant chemotherapy because of their low risk of relapsing?

YES
Predicting Baseline Prognosis

Identification of patients with HR+/HER2-negative disease (T1-2/0-3 N+):

- Who can be spared adjuvant multi-agent chemotherapy due to their low risk (<10%) of distant recurrence at 10-years with endocrine therapy-only.

![Graphs showing disease recurrence and survival data for MammaPrint, OncotypeDX, PAM50 ROR, and EndoPredict.](Image)

- MammaPrint
- OncotypeDX
- PAM50 ROR
- EndoPredict

Vijver NEJM 2002
Paik NEJM 2006
Dowsett JCO 2013 (includes tumor size)
Filipits CCR 2011 (includes tumor size and nodal status)
Comparisons between different polychemotherapy regimens for early breast cancer: meta-analyses of long-term outcome among 100 000 women in 123 randomised trials

- 1/3 breast cancer mortality reduction
- Depend on absolute risks without chemotherapy.
- Proportional risk reductions were little affected by age, nodal status, tumor size, estrogen receptor status, or tamoxifen use.
- However, gene expression-based tests were not evaluated.

<table>
<thead>
<tr>
<th>10-year Absolute Risk without chemo</th>
<th>10-year Absolute Benefit from chemo</th>
<th>10-year Risk with chemo</th>
</tr>
</thead>
<tbody>
<tr>
<td>10%</td>
<td>3%</td>
<td>7%</td>
</tr>
<tr>
<td>20%</td>
<td>6%</td>
<td>14%</td>
</tr>
<tr>
<td>30%</td>
<td>9%</td>
<td>21%</td>
</tr>
</tbody>
</table>

EBCTCG: Early Breast Cancer Trialists’ Collaborative Group

Lancet 2012
HR+/HER2-neg and NODE-negative

<table>
<thead>
<tr>
<th>INDICATION</th>
<th>Evidence Quality</th>
<th>Recommendation Strength</th>
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</thead>
<tbody>
<tr>
<td>OncotypeDX</td>
<td>YES</td>
<td>HIGH</td>
</tr>
<tr>
<td>PAM50 ROR</td>
<td>YES</td>
<td>HIGH</td>
</tr>
<tr>
<td>EndoPredict</td>
<td>YES</td>
<td>INTERMEDIATE</td>
</tr>
<tr>
<td>MammaPrint</td>
<td>NO</td>
<td>INTERMEDIATE</td>
</tr>
</tbody>
</table>

New data!  
TransATAC  
MINDACT
MINDACT

The primary analysis population

Enrolled
N = 6,693

Clinical risk (c)
Adjuvant Online!

Genomic risk (g)
70-gene signature or MammaPrint®

Discordant
N = 592

- c-Low/g-Low
  \[N = 2745\]

- c-Low/g-High

- c-High/g-Low

R-T

- No Chemotherapy

- Chemotherapy

Primary endpoint: Distant metastasis free survival (DMFS) at 5 years

Null hypothesis: 5-year DMFS rate in PT population = 92%

ha: 2.5% (1-sided)

Power: 80% when true 5-year DMFS rate = 95%

Cardoso et al. NEJM 2016
## MINDACT – Clinical Risk Definition

<table>
<thead>
<tr>
<th>ER status</th>
<th>HER2 status</th>
<th>Grade</th>
<th>Nodal status</th>
<th>Tumor Size</th>
<th>Clinical Risk in Mindact</th>
</tr>
</thead>
<tbody>
<tr>
<td>ER positive</td>
<td>HER2 negative</td>
<td>well differentiated</td>
<td>N-</td>
<td>≤ 3 cm</td>
<td>C-low</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>N-</td>
<td>3.1-5 cm</td>
<td>C-high</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1-3 positive nodes</td>
<td>N-</td>
<td>≤ 2 cm</td>
<td>C-low</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>N-</td>
<td>2.1-5 cm</td>
<td>C-high</td>
</tr>
<tr>
<td></td>
<td></td>
<td>moderately differentiated</td>
<td>N-</td>
<td>≤ 2 cm</td>
<td>C-low</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>N-</td>
<td>2.1-5 cm</td>
<td>C-high</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1-3 positive nodes</td>
<td>N-</td>
<td>≤ 2 cm</td>
<td>C-low</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>N-</td>
<td>2.1-5 cm</td>
<td>C-high</td>
</tr>
<tr>
<td></td>
<td></td>
<td>poorly differentiated or undifferentiated</td>
<td>N-</td>
<td>≤ 1 cm</td>
<td>C-low</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>N-</td>
<td>1.1-5 cm</td>
<td>C-high</td>
</tr>
<tr>
<td></td>
<td>HER2 positive</td>
<td>well differentiated OR moderately differentiated</td>
<td>N-</td>
<td>≤ 2 cm</td>
<td>C-low</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>N-</td>
<td>2.1-5 cm</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td>N-</td>
<td>1.1-5 cm</td>
<td>C-high</td>
</tr>
</tbody>
</table>

Cardoso et al. NEJM 2016
Clinical outcome of the MINDACT population at 5y median follow-up

B) DISCORDANT RISK GROUPS: PRIMARY TEST

Risk group
clinical High / genomic Low

<table>
<thead>
<tr>
<th>N</th>
<th>med. age</th>
<th>T size &gt; 2cm</th>
<th>Node positive</th>
<th>Luminal 90% HER2+ 8% triple - 1%</th>
</tr>
</thead>
<tbody>
<tr>
<td>1550</td>
<td>55 y</td>
<td>58%</td>
<td>48%</td>
<td>90%</td>
</tr>
</tbody>
</table>

Randomization

No chemotherapy Compliance = 89%
Chemotherapy Compliance = 85%

Received E.T.: 94%
Received Trastuzumab: 5%

The primary statistical test
(DMFS at 5Y)

Null Hypothesis: set at 92%
Observed 5Y DMFS = 94.7%
95% CI ≈ 92.5 – 96.2%

from Placart et al., AACR 18th April 2016 on behalf of the TRANSBig Consortium/MINDACT Investigators

Cardoso et al. NEJM 2016
MINDACT
clinical low risk / genomic low risk

Risk group
clinical Low / genomic Low

- N = 2745, med. age = 55y
- T size < 2cm, 96%
- Node negative, 94%
- Luminal 96%, HER2+ 4%
- Grade 1 or 2, 98%

Assigned:
NO CHEMOTHERAPY

Compliance = 99%
(Received Endocrine therapy: 79%)

DMFS

% at 5y (95% CI)
cL/gL 97.6 (96.9 – 98.1)

Cardoso et al. NEJM 2016
**HR+/HER2-neg and NODE-negative**

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<td>YES</td>
<td>HIGH</td>
</tr>
<tr>
<td>EndoPredict</td>
<td>YES</td>
<td>INTERMEDIATE</td>
</tr>
<tr>
<td>MammaPrint</td>
<td>NO</td>
<td>INTERMEDIATE</td>
</tr>
</tbody>
</table>

**HR+/HER2-neg and NODE-positive**

<table>
<thead>
<tr>
<th>INDICATION</th>
<th>Evidence Quality</th>
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</tr>
</thead>
<tbody>
<tr>
<td>OncotypeDX</td>
<td>NO</td>
<td>INTERMEDIATE</td>
</tr>
<tr>
<td>PAM50 ROR</td>
<td>NO</td>
<td>INTERMEDIATE</td>
</tr>
<tr>
<td>EndoPredict</td>
<td>NO</td>
<td>INSUFFICIENT</td>
</tr>
<tr>
<td>MammaPrint</td>
<td>NO</td>
<td>INTERMEDIATE</td>
</tr>
</tbody>
</table>
PROSIGNA within HR+/HER2-neg and 1 positive node

- TransATAC + ABCSG08 combined analysis
- N=331 1N+ and 212 2-3N+
- 5-years of endocrine therapy and no chemotherapy

Gnant et al. Annals Oncol 2015
Should we use Ki67 IHC to identify low risk outcome patients who do not need adjuvant chemotherapy?

NO
IHC for Ki-67 analysis **lacks reproducibility across laboratories** and, therefore, cannot be consistently interpreted when performed in a broad range of laboratories.
Can these tests help us determine the benefit of adjuvant chemotherapy?

Maybe
NSABP-B20 subanalysis  
*Paik et al. JCO 2006*

OncotypeDX RS HIGH RISK  
N=651 ; ER+/NODE-negative

---

SWOG8814 subanalysis  
*Albain et al. Lancet Oncol 2010*

OncotypeDX RS HIGH RISK  
N=367 ; ER+/NODE+

---

NSABP-B20 data are confounded by the dataset originally used to generate the assay.

---

SWOG8814 data is hypothesis generating: small sample set and no additional prediction beyond 5 years.
TAILORx Study Design
ECOG/Inter-group
PI: J. A. Sparano

Accrual completed on Oct 25th 2010
Target: 10,000

ER+/HER2-/Node-

ONCOTYPE DX ASSAY

REGISTER
Specimen Banking

Primary Study Group
RS 11-25
~44% of Population

Secondary Study Group 1
RS < 11
~29% of Population

Secondary Study Group 2
RS > 25
~27% of Population

Primary Objective:
- Non-inferiority study within the intermediate group
- Null hypothesis: no treatment differences
- No chemotherapy arm (>87% 5-year DFS)
- Chemotherapy arm (90% 5-year DFS).
- Type 1 error: 10%; Type 2 error: 5%
- 95% power

N=1,626
5-year 99.3% DMFS (98.7-99.6)
69% <1.9cm ; 93% Grade 1-2

Sparano et al. NEJM 2015
A Phase III, Randomized Clinical Trial of Standard Adjuvant Endocrine Therapy +/- Chemotherapy in Patients with 1-3 Positive Nodes, HR+/HER2-negative and HER2-Negative Breast Cancer With Recurrence Score (RS) of 25 or Less.

ClinicalTrials.gov Identifier: NCT01272037
Opened 2011, Estimated Accrual = 4000

Primary Objective:
- To find a significant interaction between Recurrence Score (as a continuous variable) and treatment
- DFS
- Type 1 error: 5% ; 80% power
• N=4,500
• HR+/HER2-negative
• pN1-2 or pT ≥3 cm
• Non-inferiority (delta 3%, 85% power)
  • 5-year DFS 82% without chemotherapy
• PROSIGNA will be used (cutpoint 60)
  • High risk
  • Low/Intermediate risk

http://www.nets.nihr.ac.uk/projects/hta/1034501
Clinical outcome of the MINDACT population at 5y median follow-up
DMFS IN ALL 4 RISK GROUPS

Distant Metastasis Free Survival

% at 5 year
- cL/gL: 97.6 (96.9, 98.1)
- cL/gH: 94.8 (92.4, 96.4)
- cH/gL: 95.1 (93.8, 96.2)
- cH/gH: 90.6 (89.0, 92.0)

Number of patients at risk:
- cL/gL: 2745, 2628, 2331, 735, 33
- cL/gH: 592, 550, 484, 136, 2
- cH/gL: 1550, 1457, 1317, 311, 9
- cH/gH: 1806, 1689, 1462, 395, 11

Discordant risk groups

from Plascart et al., AACR 18th April 2016 on behalf of the TRANSBiG Consortium/MINDACT Investigators
MINDACT – secondary endpoints

Efficacy: CT vs no CT in discordant risk group c-Low/g-High per protocol analysis

<table>
<thead>
<tr>
<th>c-Low/g-High</th>
<th>DMFS</th>
<th>DFS</th>
<th>OS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Treatment received</td>
<td>CT</td>
<td>no CT</td>
<td>CT</td>
</tr>
<tr>
<td>Patients Observed Events</td>
<td>224</td>
<td>11</td>
<td>96.1 (92.4, 98.1)</td>
</tr>
<tr>
<td>% at 5 Year(s) (95% CI)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hazard Ratio (adjusted Cox model) (95% CI)</td>
<td>0.90 (0.40, 2.01)</td>
<td>0.74 (0.40, 1.39)</td>
<td>0.72 (0.23, 2.24)</td>
</tr>
<tr>
<td>p-value (adjusted logrank)</td>
<td>0.798</td>
<td>0.355</td>
<td>0.572</td>
</tr>
<tr>
<td>no CT</td>
<td>254</td>
<td>14</td>
<td>93.9 (89.6, 96.5)</td>
</tr>
</tbody>
</table>

from Piccart et al., AACR 103rd April 2016 on behalf of the TRANSBIG Consortium/MINDACT investigators

Cardoso et al. NEJM 2016

Trial not powered for the comparisons of yes or no chemotherapy
MINDACT – secondary endpoints

Efficacy: CT vs no CT in discordant risk group c-High/g-Low per protocol analysis

<table>
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<tr>
<th>c-High/g-Low</th>
<th>Treatment received</th>
<th>CT vs no CT per protocol population</th>
<th>Hazard Ratio (adjusted Cox model) (95% CI)</th>
<th>p-value (adjusted logrank)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Patients</td>
<td>Observed Events</td>
<td>% at 5 Year(s) (95% CI)</td>
<td></td>
</tr>
<tr>
<td>DMFS</td>
<td>CT</td>
<td>592</td>
<td>22</td>
<td><strong>96.7</strong> (94.7, 98.0)</td>
</tr>
<tr>
<td></td>
<td>no CT</td>
<td>636</td>
<td>37</td>
<td><strong>94.8</strong> (92.6, 96.3)</td>
</tr>
<tr>
<td>DFS</td>
<td>CT</td>
<td>592</td>
<td>39</td>
<td><strong>93.3</strong> (90.7, 95.2)</td>
</tr>
<tr>
<td></td>
<td>no CT</td>
<td>636</td>
<td>66</td>
<td><strong>90.3</strong> (87.6, 92.4)</td>
</tr>
<tr>
<td>OS</td>
<td>CT</td>
<td>592</td>
<td>10</td>
<td><strong>98.8</strong> (97.4, 99.5)</td>
</tr>
<tr>
<td></td>
<td>no CT</td>
<td>636</td>
<td>18</td>
<td><strong>97.3</strong> (95.6, 98.4)</td>
</tr>
</tbody>
</table>

from Piccart et al. AACR 10th April 2016 on behalf of the TRANSBIG Consortium/MINDACT Investigators

Trial not powered for the comparisons of yes or no chemotherapy

Cardoso et al. NEJM 2016
Can these tests identify patients that may be spared extended endocrine therapy?

Maybe
Predicting Late Recurrence

To identify a group of patients with HR+/HER2-negative disease (T1-2/0-3 N+):

- That may be spared extended endocrine therapy (5-10 years) due to their low risk of recurrence

**BC Index**

Sgroi Lancet Oncol 2013

**PAM50 ROR**

Sestak JCO 2015

(includes tumor size)

**EndoPredict**

Dubsky BJC 2013

(includes tumor size and nodal status)
The ROR score was the strongest molecular prognostic factor in the late follow-up period, whereas IHC4 and OncotypeDX RS were only weakly prognostic in this period.
The EndoPredict score provides prognostic information on late distant metastases in ER+/HER2− breast cancer patients

P Dubsky1,2, J C Brase3, R Jakesz4, M Rudas5, C F Singer6, R Greil7, O Dietze8, I Lusser7, E Klug8, R Sedivy9, M Bachner10, D Mayr11, M Schmidt12, M C Gehrmann13, C Petry12, K E Weber2, K Fisch2, R Kronenwett2, M Gnant1 and M Filipits14 on behalf of the Austrian Breast and Colorectal Cancer Study Group (ABC6G)

N=1,702 (ABC6G6/8)

Table 1. Multivariate Cox proportional hazard models for estimating the contribution of variables to predict distant recurrence in the time interval 0–5 years and after 5 years (1702 ER+/HER2− tumours, ABC6G6/8)

<table>
<thead>
<tr>
<th>Variable</th>
<th>0–5 years unit HR (95% CI)</th>
<th>P-value</th>
<th>&gt; 5 years unit HR (95% CI)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>EP</td>
<td>1.20 (1.10–1.31)</td>
<td>&lt;0.001</td>
<td>1.28 (1.10–1.48)</td>
<td>0.001</td>
</tr>
<tr>
<td>Age</td>
<td>1.03 (1.00–1.06)</td>
<td>0.032</td>
<td>0.97 (0.93–1.02)</td>
<td>0.264</td>
</tr>
<tr>
<td>Nodal status</td>
<td>2.15 (1.67–2.77)</td>
<td>&lt;0.001</td>
<td>2.45 (1.58–3.81)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Tumour size</td>
<td>1.26 (0.94–1.70)</td>
<td>0.121</td>
<td>1.11 (0.67–1.86)</td>
<td>0.679</td>
</tr>
<tr>
<td>Ki67</td>
<td>1.01 (0.99–1.03)</td>
<td>0.171</td>
<td>1.01 (0.97–1.05)</td>
<td>0.761</td>
</tr>
<tr>
<td>Grade</td>
<td>1.21 (0.77–1.90)</td>
<td>0.414</td>
<td>0.64 (0.32–1.28)</td>
<td>0.210</td>
</tr>
<tr>
<td>Treatment arm</td>
<td>0.95 (0.61–1.48)</td>
<td>0.807</td>
<td>0.91 (0.40–2.09)</td>
<td>0.827</td>
</tr>
</tbody>
</table>

Abbreviations: CI = confidence interval; ER = oestrogen receptor; HR = hazard ratio.
### Distant Late Recurrence Rates of Low Risk Groups

<table>
<thead>
<tr>
<th>ASSAY</th>
<th>STUDY</th>
<th>TOTAL N</th>
<th>10-yr Risk</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>BCI</strong> TransATAC</td>
<td></td>
<td>665</td>
<td>3.5%</td>
<td>2.0%</td>
</tr>
<tr>
<td><strong>PAM50 ROR</strong></td>
<td>TransATAC +ABCsg08</td>
<td>2,137</td>
<td>2.4%</td>
<td>1.6%</td>
</tr>
<tr>
<td><strong>Endo Predict</strong></td>
<td>ABCsg06 +ABCsg08</td>
<td>1,702</td>
<td>3.7%</td>
<td>0.9%</td>
</tr>
</tbody>
</table>

Potential absolute % reduction of distant recurrence with extended endocrine therapy:
- 0.8-3.1% at 10-years
- 2.9-5.6% at 15 years (*assuming 1% annual recurrence hazard rate)

Sgroi Lancet Oncol 2013, Sestak JCO 2015, Dubsky BJC 2013
Withdrawing extended endocrine therapy based on current prognostic gene expression-based assays?

**Treatment Benefit**
- How low should we go?
- Do they benefit more or less?
- How much toxicity is the patient willing to accept?
Prediction of Late Disease Recurrence and Extended Adjuvant Letrozole Benefit by the HOXB13/IL17BR Biomarker


- Retrospective analysis of samples of the MA.17 clinical trial
- Nested case-control design: 83 recurrences vs 166 non-recurrences ∙ N= 249 patients
EXTENDED ENDOCRINE THERAPY

HR+/HER2-neg and NODE-negative

<table>
<thead>
<tr>
<th>INDICATION</th>
<th>Evidence Quality</th>
<th>Recommendation Strength</th>
</tr>
</thead>
<tbody>
<tr>
<td>EndoPredict</td>
<td>NO</td>
<td>INTERMEDIATE</td>
</tr>
<tr>
<td>PAM50 ROR</td>
<td>NO</td>
<td>INTERMEDIATE</td>
</tr>
</tbody>
</table>

HR+/HER2-neg and NODE-positive

No comment
Are these assays the same at the individual patient level?

NO
Comparing Breast Cancer Multiparameter Tests in the OPTIMA Prelim Trial

Only 39.4% were classified uniformly.

Regarding subtype, 40.7% patients had discordant calls (BLUEPRINT vs PROSIGNA).

BLUEPRINT was trained on IHC, while PROSIGNA was trained on natural patterns from gene expression data.

For the individual patient, tests may provide differing risk categorization and subtype information.

N=313 ONCOTYPEDX, MAMMAPRINT and PROSIGNA

Table 4. Kappa statistics for tests providing risk predictions*  

<table>
<thead>
<tr>
<th>Test</th>
<th>MammaPrint (low), Kappa statistic (95% CI)</th>
<th>Prosigna (low/intermediate), Kappa statistic (95% CI)</th>
<th>IHC4 (low/intermediate), Kappa statistic (95% CI)</th>
<th>IHC4-AQUA† (low/low-mid), Kappa statistic (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Oncotype DX</td>
<td>0.40 (0.30 to 0.49)</td>
<td>0.44 (0.33 to 0.54)</td>
<td>0.53 (0.41 to 0.65)</td>
<td>0.40 (0.30 to 0.51)</td>
</tr>
<tr>
<td>(recurrence score ≤25)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>MammaPrint</td>
<td>–</td>
<td>0.53 (0.43 to 0.63)</td>
<td>0.33 (0.21 to 0.44)</td>
<td>0.42 (0.30 to 0.53)</td>
</tr>
<tr>
<td>Prosigna (low/intermediate)</td>
<td>–</td>
<td>–</td>
<td>0.39 (0.27 to 0.50)</td>
<td>0.43 (0.31 to 0.54)</td>
</tr>
<tr>
<td>IHC4 (low/intermediate)</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>0.60 (0.50 to 0.70)</td>
</tr>
</tbody>
</table>

Bartlett et al. JNCI 2016
Comparing PAM50/Prosigna ROR vs. OncotypeDX RS

- N=1,017 patients with ER+ disease treated with 5-years of adjuvant endocrine therapy
- PROSIGNA ROR provides more prognostic information
Comparing EndoPredict (EPclin) vs. OncotypeDX RS

- N=928 patients with ER+ disease treated with 5-years of adjuvant endocrine therapy
- EPclin provides more prognostic information
- This was partly but not entirely because of EPclin integrating molecular data with nodal status/tumor size

TransATAC

Buus et al. JNCI 2016
Take-home messages

• At least 4 tests based on gene expression are available in Europe.

• All are standardized/highly reproducible:
  • MammaPrint and PAM50 \(\dagger\) FDA/510(k) cleared.
  • A 10% discordant rate is expected between MammaPrint FF vs. FFPE.
  • EndoPredict and PAM50 can be performed at local labs.

• These tests help identify patients who do not need adjuvant chemotherapy because of their low risk of relapsing at 10 years if treated with endocrine therapy-only:
  • IMPORTANT: use them with clinical-pathological variables, mostly tumor size and nodal status.
  • EndoPredict and PAM50 ROR integrate molecular data with tumor size and nodal status.
  • In patients with 2-3 high-risk clinical features treated with endocrine therapy-only, MammaPrint has shown prospectively (MINDACT) that:
    • The low-risk groups has a DMFS >92% at 5-years
    • The DMFS at 10 years of the low-risk is likely to be <90%. More follow-up is needed.
    • A clinically meaningful chemotherapy benefit in this group cannot be excluded.
In terms of predicting the degree of adjuvant chemotherapy benefit:

- Evidence exists regarding the predictive ability of OncotypeDX in the high-risk group. However:
  - NSABP-B20 data are confounded by the dataset originally used to generate the assay.
  - SWOG8814 data is hypothesis generating: small sample set and no additional prediction beyond 5 years.

- Two large phase III prospective clinical trials (TailorX and RxPonder) are evaluating the clinical utility of OncotypeDX as a predictive test in the following scenarios:
  - TAILORX: Patients with node-negative, HR+/HER2-negative disease with Intermediate RS (11-25).
  - RXPONDER: Patients with 1-3 N+, HR+/HER2-negative disease with Low/Intermediate RS (≤25).

- One large phase III prospective clinical trial (OPTIMA) will evaluate the clinical utility of Prosigna as a predictive test in the following scenario:
  - Patients with pN1-2 or pT2(>3cm)pN0 HR+/HER2-negative disease.

Take-home messages
Take-home messages

- EndoPredict and Prosigna predict late recurrence in HR+/HER2-negative breast cancer.
  - These assays might identify patients who can be spared extended endocrine therapy beyond 5 years due to their low risk of relapsing between period 5-10.
  - However, for this indication, the community and the patients might need to establish where to draw the cutoff based on risk, benefit, toxicity and cost (in some countries).

- Gene expression-based tests should not be considered to be the same.
  - Head-to-head comparisons for outcome and treatment benefit are needed.
  - To date,
    - Both EndoPredict and Prosigna provided more prognostic information than OncotypeDX in TransATAC.
      - This is only partially explained by the fact that EndoPredict and Prosigna include tumor size and nodal status.
When possible, use these tests!
They are helpful, reproducible and valuable!

THANK YOU!