Immunotherapy for Breast Cancer Clinical Development

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Rationale to develop immunotherapy in BC

**Features of BC**

- TIL infiltration
- Mutation Load
- PD-L1 expression
- Responses in early phase trials
Clinical significance of TIL infiltration in BC

TIL have prognostic and predictive value in early stage BC, particularly in HER2+ and TNBC
Rationale to develop immunotherapy in BC

Features of BC

- TIL infiltration
- Mutation Load
- Responses in early phase trials
- PD-L1 expression
Clinical significance of mutation load

- TIL can recognize somatic mutations and are correlated to the density of predicted mutant epitopes.

BC has a moderate mutational load

- Higher mutation load in TNBC and HER2 BC

Total mutations and survival

- Number of predicted immunogenic mutations and survival

Ton N. Schumacher, and Robert D. Schreiber Science 2015
Scott D. Brown et al. Genome Res. 2014
Rationale to develop immunotherapy in BC

**Features of BC**

- TIL infiltration
- Responses in early phase trials
- Mutation Load
- PD-L1 expression
PD-L1 expression in early stage BC

<table>
<thead>
<tr>
<th>PD-L1 IHC expression on</th>
<th>N = 110 (%)</th>
<th>Median (% cells-positive cases)</th>
<th>25th-75th percentile (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tumor cells</td>
<td>6 (5.5)</td>
<td>4</td>
<td>1-16.25</td>
</tr>
<tr>
<td>Immune cells</td>
<td>22 (20)</td>
<td>6</td>
<td>1-10.5</td>
</tr>
<tr>
<td>Stromal cells</td>
<td>4 (3.6)</td>
<td>7.5</td>
<td>2-17.5</td>
</tr>
<tr>
<td>Any cells</td>
<td>26 (23.6)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*PD-L1 positivity: ≥1% expression on tumor or immune or stromal cells*
PD-L1 expression in early stage BC

PD-L1 expression in metastatic BC

111 metastases from 11 sites including skin (40), ipsilateral breast relapse (23), liver (12), soft tissues (7), pleura (6), bone (6), brain (5), peritoneum (3), colon (1), lung (1), nodes (7)

<table>
<thead>
<tr>
<th>PD-L1 IHC expression on</th>
<th>N = 111 (%)</th>
<th>Median (% cells-positive cases)</th>
<th>25th-75th percentile (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tumor cells</td>
<td>3 (2.7)</td>
<td>1</td>
<td>1-5</td>
</tr>
<tr>
<td>Immune cells</td>
<td>12 (10.8)</td>
<td>5</td>
<td>5-10</td>
</tr>
<tr>
<td>Stromal cells</td>
<td>9 (8.1)</td>
<td>5</td>
<td>5-10</td>
</tr>
<tr>
<td>Any cells</td>
<td>17 (15.3)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

PD-L1 positivity: ≥1% expression on tumor or immune or stromal cells

Rationale to develop immunotherapy in BC

- TIL infiltration
- Mutation Load
- PD-L1 expression
- Responses in early phase trials

Features of BC
Single Agent Activity of PD-1/PD-L1 Blockade in Relapsed/Refractory Cancer

- MPDL3280A/Atezolizumab
- Pembrolizumab
- Nivolumab

TNBC: the candidate for immunotherapy

- Worse prognosis than other BC subtype
- Limited treatment options
- Cancer immunotherapy represents a promising treatment approach for TNBC
  - Higher TIL infiltration
  - Higher mutation rate → immunogenic neoantigens
  - Higher PD-L1 expression → inhibit T-cell antitumor responses

Strategies to modulate the immune system in breast cancer

**Active:** priming of the immune system

- **Antigen-specific**
  - Peptide vaccine
  - DC-vaccine
  - DNA-vaccine
  - Whole cell vaccine

- **Non antigen-specific**
  - Checkpoint inhibitors
  - Cytokines

**Passive:** delivery of compounds that may use immune system

- **Monoclonal antibodies**
  - Trastuzumab
  - Pertuzumab

- **Adoptive cell transfer**
  - CAR T cells

**Immune modulators**

**Targeted antibodies**

**Cellular immunotherapy**
IMMUNE CHECKPOINT INHIBITORS
Phase Ib of pembrolizumab in mTNBC
KEYNOTE 012

- Recurrent or metastatic ER⁻/PR⁻/HER2⁻ breast cancer
- ECOG PS 0-1
- PD-L1⁺ tumor
- No systemic steroid therapy
- No autoimmune disease (active or history of)
- No active brain metastases

Pembro 10 mg/kg Q2W

- Complete Response
- Discontinuation Permitted
- Partial Response or Stable Disease
- Treat for 24 months or until progression or intolerable toxicity
- Confirmed Progressive Disease
- Discontinue

- **PD-L1 positivity:** 58% of all patients screened had PD-L1-positive tumors
- **Treatment:** 10 mg/kg IV Q2W
- **Response assessment:** Performed every 8 weeks per RECIST v1.1

---

*PD-L1 expression was assessed in archival tumor samples using a prototype IHC assay and the 22C3 antibody. Only patients with PD-L1 staining in the stroma or in ≥1% of tumor cells were eligible for enrollment.

*If clinically stable, patients are permitted to remain on pembrolizumab until progressive disease is confirmed on a second scan performed ≥4 weeks later. If progressive disease is confirmed, pembrolizumab is discontinued. An exception may be granted for patients with clinical stability or improvement after consultation with the sponsor.*
Patients Evaluable for Response

- Overall response rate: 5 (18.5%)
- Best overall response:
  - Complete response: 1 (3.7%)
  - Partial response: 4 (14.8%)
  - Stable disease: 7 (25.9%)
  - Progressive disease: 12 (44.4%)
  - No assessment: 3 (11.1%)

Phase Ib of pembrolizumab in mTNBC
KEYNOTE 012

Nanda R et al. JCO 2016
# Immune checkpoint inhibitors

<table>
<thead>
<tr>
<th>Drug</th>
<th>Phase</th>
<th>Subtype</th>
<th>PD-L1</th>
<th>Nb pts</th>
<th>ORR</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Pembrolizumab</strong> (anti-PD-1)</td>
<td>Ib</td>
<td>TNBC</td>
<td>≥ 1% TC Stroma+ (58% of screened pts)</td>
<td>32</td>
<td>27</td>
<td>18.5%</td>
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<tr>
<td></td>
<td></td>
<td>PDL1+</td>
<td></td>
<td></td>
<td></td>
<td>1 CR 4 PR</td>
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<td></td>
<td>KEYNOTE 012 Nanda et al. SABC 2014</td>
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<td></td>
<td></td>
<td>JCO 2016</td>
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<td><strong>Atezolizumab</strong> (anti-PD-L1)</td>
<td>Ia</td>
<td>TNBC</td>
<td>≥ 5% IC</td>
<td>115</td>
<td>112</td>
<td>10%</td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>3 CR 8 PR</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Schmid et al. AACR2017</td>
</tr>
<tr>
<td><strong>Pembrolizumab</strong> (anti-PD-1)</td>
<td>Ib</td>
<td>ER+/HER2-PDL1+</td>
<td>≥ 1% TC Stroma+ (19% of screened pts)</td>
<td>25</td>
<td></td>
<td>12%</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>0 CR 3 PR</td>
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<tr>
<td></td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td>KEYNOTE 028 Rugo et al. SABC 2015</td>
</tr>
<tr>
<td><strong>Avelumab</strong> (anti-PD-L1)</td>
<td>Ib</td>
<td>All</td>
<td>≥ 1% TC (58%)</td>
<td>168</td>
<td>153</td>
<td>4.8%</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td>≥ 5% TC (16%)</td>
<td></td>
<td></td>
<td>1 CR 7 PR</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td>≥ 10% IC (9%)</td>
<td></td>
<td></td>
<td>JAVELIN Dirix et al. SABC 2015</td>
</tr>
<tr>
<td></td>
<td></td>
<td>TNBC</td>
<td></td>
<td>58</td>
<td>72</td>
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<tr>
<td></td>
<td></td>
<td>ER+/HER2-</td>
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<td>2.8%</td>
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</table>
Phase Ia: Atezolizumab, an anti-PD-L1 antibody, in patients with locally advanced or metastatic TNBC

ORR according to PD-L1 expression

Schmid P. et al. AACR 2017
## ORR according to PD-L1 expression

### Phase Ib: Avelumab, an anti-PD-L1 antibody, in patients with locally advanced or metastatic BC

<table>
<thead>
<tr>
<th>PD-L1 expression</th>
<th>All patients (N=136)</th>
<th>TNBC (N=48)</th>
</tr>
</thead>
<tbody>
<tr>
<td>≥ 1% TC</td>
<td>3/85 (3.5%)</td>
<td>2/33 (6.1%)</td>
</tr>
<tr>
<td>≥ 5% TC</td>
<td>1/23 (4.3%)</td>
<td>1/13 (7.7%)</td>
</tr>
<tr>
<td>≥ 25% TC</td>
<td>0/3 (0)</td>
<td>0/2 (0)</td>
</tr>
<tr>
<td>≥ 10% IC</td>
<td>4/12 (33.3%)</td>
<td>4/9 (44.4%)</td>
</tr>
</tbody>
</table>

Dirix L et al. SABC 2015
Long lasting responses

<table>
<thead>
<tr>
<th>Change From Baseline, %</th>
<th>On treatment</th>
<th>Discontinued treatment</th>
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</thead>
<tbody>
<tr>
<td>100</td>
<td></td>
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</tr>
<tr>
<td>80</td>
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<td>60</td>
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<td>10</td>
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<td>-90</td>
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<tr>
<td>-100</td>
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</tr>
</tbody>
</table>

3 "exceptional" responders

Nanda et al. SABC 2014
Metastatic TNBC treated with pembrolizumab
Metastatic TNBC treated with pembrolizumab

Images provided by Henri Maisonnier
Metastatic TNBC treated with pembrolizumab

2 months later → PR

4 months later → CR

3 years later → CR!
Survival benefit for responders

Phase Ia: Atezolizumab, an anti-PD-L1 antibody, in patients with locally advanced or metastatic TNBC

- 1-y OS: 100%
- 2-y OS: 100%
- 3-y OS: 100%
- 1-y OS: 51%
- 1-y OS: 33%

**irRC Response**
- CR/PR (n = 15)
- SD (n = 19)
- PD (n = 55)

Median survival follow-up (range) was 15.2 mo (0.4+ to 36.7) in all patients, 17.0 mo (0.43+ to 36.7) in IC2/3 patients and 12.8 mo (0.8+ to 16.9) in IC0/1 patients. Data cutoff: March 31, 2016.
Immune checkpoint inhibitors in BC

- Single agent response rate: 5-20%
- Higher response rate in:
  - TNBC
  - PD-L1 positive cases
- Long-lasting responses in a subset of responders
- Survival benefit for responders patients
- Acceptable safety profile in early phases trials in metastatic setting
Future directions for immune checkpoint inhibitors in BC

- Phases II & III clinical trials are ongoing
- Combined strategies
- Neoadjuvant/Adjuvant setting
- Identification of biomarkers of response for a better selection of patients
- Development of next generation immunoregulatory antibodies
KEYNOTE-119: A Randomized Phase III Study of Single-Agent Chemotherapy per Investigator’s choice for mTNBC

Patients
- Recurrent mBC
- 1 or 2 prior systemic treatments for mBC
- Documented disease progression on/after most recent therapy
- Previous treatment with an anthracycline and/or a taxane in the (neo)adjuvant or metastatic setting
- ECOG PS 0-1

Stratification by:
- PD-L1 tumor status (positive vs negative)
- Prior (neo)adjuvant therapy vs de novo metastatic disease at initial diagnosis

Randomize 1:1 N = 600

Pembrolizumab
200 mg Q3W

Follow-up for safety (≤90 days)
Follow-up for survival (every 3 months)

Investigator’s choice of one of the following:
- Capecitabine
- Enbulin
- Gemcitabine
- Vinorelbine

ECOG PS = Eastern Cooperative Oncology Group performance status; mBC = metastatic breast cancer; PD-L1 = programmed death ligand 1; Q3W = every 3 weeks.

Maximum enrollment cap of 60% of total enrollment for each chemotherapy drug.
Future directions for immune checkpoint inhibitors in BC

- Phases II & III clinical trials are ongoing
- Combined strategies
- Neoadjuvant/Adjuvant setting
- Identification of biomarkers of response
- Development of next generation immunoregulatory antibodies
Combined strategies with immune checkpoint inhibitors

- Development of multiple rationale combinations with compatible mechanisms that act synergistically to:
  - Increase anti-tumor efficacy
  - Reduce on-target side effects
Combined strategies with immune checkpoint inhibitors in BC

- **Immunotherapies**
  - Anti-PD-L1 & cancer vaccine

- **Chemotherapy**
  - Paclitaxel & Nab-Paclitaxel
  - Eribuline
  - Doxorubicin

- **Targeted therapies**
  - Trastuzumab, Pertuzumab
  - Bevacizumab

- **Radiotherapy**
Combination of checkpoint inhibitors

**Phase I/II**: agonist OX40 + radiation – mBC
NCT01862900

**Phase Ib/II**: anti-PD-L1 + agonist 4-1BB(CD137) or + agonist OX40 - mTNBC
NCT02554812

**Phase I**: anti-PD-1 + anti-B7-H3- mTNBC
NCT02475213

**Phase I**: anti-CTLA-4 + anti-B7-H3- mTNBC
NCT02381314

**Phase I**: CA-170 (anti-PD-L1/PD-L2, anti-VISTA) – mTNBC
NCT02812875

**Phase IIb**: anti-LAG-3 + Paclitaxel –mBC
NCT02614833

T cell targets for immunoregulatory antibody therapy.

*I Mellman et al. Nature 480, 480-489 (2011) doi:10.1038/nature10673*
Combined strategies with immune checkpoint inhibitors in BC

- **Immunotherapies**
  - Anti-PD-L1 & cancer vaccine

- **Chemotherapy**
  - Paclitaxel & Nab-Paclitaxel
  - Eribuline
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- **Targeted therapies**
  - Trastuzumab, Pertuzumab
  - Bevacizumab

- **Radiotherapy**
## Combination with Chemotherapy

### ORR according to treatment lines setting

<table>
<thead>
<tr>
<th>Drug</th>
<th>Ph</th>
<th>Subtype</th>
<th>Lines setting</th>
<th>Nb of pts</th>
<th>ORR</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Atezolizumab + nab-Paclitaxel</td>
<td>Ib</td>
<td>TNBC</td>
<td>1&lt;sup&gt;st&lt;/sup&gt; line</td>
<td>32</td>
<td>42%</td>
<td>Adams et al. ASCO 2016</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>2&lt;sup&gt;nd&lt;/sup&gt; line</td>
<td>24</td>
<td>67%</td>
<td></td>
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<tr>
<td></td>
<td></td>
<td></td>
<td>3+ line</td>
<td>9</td>
<td>25%</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>8</td>
<td>29%</td>
<td></td>
</tr>
<tr>
<td>Pembrolizumab + Eribuline</td>
<td>Ib/II</td>
<td>TNBC</td>
<td>1&lt;sup&gt;st&lt;/sup&gt; line</td>
<td>89</td>
<td>33.3%</td>
<td>Tolaney et al. SABC 2016</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>2-3&lt;sup&gt;rd&lt;/sup&gt; line</td>
<td>39</td>
<td>41.2%</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>17</td>
<td>27.3%</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>22</td>
<td></td>
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</table>
Combination with Chemotherapy

<table>
<thead>
<tr>
<th>Drug</th>
<th>Ph</th>
<th>Subtype</th>
<th>PD-L1</th>
<th>Nb of pts</th>
<th>ORR</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Atezolizumab + nab-Paclitaxel</td>
<td>Ib</td>
<td>TNBC</td>
<td>IC 0 IC 1/2/3 Unknown</td>
<td>7 9 8</td>
<td>57.1% 77.8% 75%</td>
<td>Adams et al. ASCO 2016</td>
</tr>
<tr>
<td>Pembrolizumab + Eribuline</td>
<td>Ib/II</td>
<td>TNBC</td>
<td>PD-L1 pos PD-L1 neg</td>
<td>17 18</td>
<td>29.4% 33.3%</td>
<td>Tolaney et al. SABC 2016</td>
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</table>

ORR according to PD-L1 expression
## Combination with chemotherapy

<table>
<thead>
<tr>
<th>Phase</th>
<th>Drug</th>
<th>Chemotherapy</th>
<th>BC</th>
<th>Trials and status</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Nivolumab</td>
<td>Nab-paclitaxel</td>
<td>HER2-neg</td>
<td>NCT02309177 recruiting</td>
</tr>
<tr>
<td>II</td>
<td>Nivolumab</td>
<td>Low dose CT</td>
<td>TNBC</td>
<td>NCT024999367 recruiting</td>
</tr>
<tr>
<td></td>
<td></td>
<td>- Doxorubicine</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>- Cyclophosphamide</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>- cisplatin</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ib</td>
<td>Atezolizumab</td>
<td>Doxorubicin &amp; cyclophosphamide</td>
<td>HER2-neg</td>
<td>NCT02605915 recruiting</td>
</tr>
<tr>
<td>I</td>
<td>Durvalumab</td>
<td>Paclitaxel</td>
<td>TNBC</td>
<td>NCT02628132</td>
</tr>
</tbody>
</table>
Combination with chemotherapy

**Phase III trials**

**Pembrolizumab**
- **KEYNOTE-355**
  - First-line metastatic TNBC PD-L1+
  - pembrolizumab + chemo*
  - chemo* + placebo
  - Primary endpoints: safety & PFS, OS

**Atezolizumab**
- **Impassion 130**
  - First-line metastatic TNBC
  - atezolizumab + nabpaclitaxel
  - nabpaclitaxel+ placebo
  - Primary endpoints: PFS & OS

*Chemotherapy: one of the three regimens: Nab-paclitaxel, paclitaxel, gemcitabine/carboplatin

*Chemotherapy: one of the three regimens: Nab-paclitaxel, paclitaxel, gemcitabine/carboplatin
Combined strategies with immune checkpoint inhibitors in BC

- **Immunotherapies**
  - Anti-PD-L1 & cancer vaccine

- **Chemotherapy**
  - Paclitaxel & Nab-Paclitaxel
  - Eribuline
  - Doxorubicin

- **Targeted therapies**
  - Trastuzumab, Pertuzumab
  - Bevacizumab

- **Radiotherapy**
## Combination with targeted therapies

**HER2**

<table>
<thead>
<tr>
<th>Drugs</th>
<th>Phase</th>
<th>Setting</th>
<th>Trials and status</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Pembrolizumab</strong> + Trastuzumab</td>
<td>Ib/II</td>
<td>Trastuzumab resistant PD-L1+ Up to 3 lines of anti-HER2</td>
<td>PANACEA NCT02129556 Recruiting</td>
</tr>
<tr>
<td><strong>Atezolizumab</strong> + Trastuzumab, Pertuzumab +/- Paclitaxel Or TDM-1</td>
<td>Ib</td>
<td>Metastatic HER2+</td>
<td>NCT02605915 Recruiting</td>
</tr>
<tr>
<td><strong>Atezolizumab</strong> + Trastuzumab, Pertuzumab, Paclitaxel</td>
<td>IIA</td>
<td>1&lt;sup&gt;st&lt;/sup&gt; line treatment HER2+</td>
<td>NCT03125928 Not yet recruiting</td>
</tr>
<tr>
<td><strong>Durvalumab</strong> + Trastuzumab</td>
<td>Ib</td>
<td>HER2+ mBC</td>
<td>NCT02649686 Ongoing, not recruiting</td>
</tr>
</tbody>
</table>
Combined strategies with immune checkpoint inhibitors in BC

- **Immunotherapies**
  - Anti-PD-L1 & cancer vaccine

- **Chemotherapy**
  - Paclitaxel & Nab-Paclitaxel
  - Eribuline
  - Doxorubicin

- **Targeted therapies**
  - Trastuzumab, Pertuzumab
  - Bevacizumab

- **Radiotherapy**
## Combination with radiotherapy

<table>
<thead>
<tr>
<th>Phase</th>
<th>Drug</th>
<th>RT</th>
<th>setting</th>
<th>Trials and status</th>
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<tbody>
<tr>
<td>II</td>
<td>Pembrolizumab</td>
<td>6Gyx5</td>
<td>mTNBC</td>
<td>NCT02730130 recruiting</td>
</tr>
<tr>
<td>I</td>
<td>Pembrolizumab</td>
<td>hypofractionated</td>
<td>Metastatic</td>
<td>RADVAX NCT02303990 recruiting</td>
</tr>
<tr>
<td>I</td>
<td>Pembrolizumab</td>
<td>20 Gy x1</td>
<td>Oligometastatic BC</td>
<td>BOSTON II NCT02303366 recruiting</td>
</tr>
<tr>
<td>I</td>
<td>Durvalumab + Tremelimumab</td>
<td>8 Gyx3 vs 17 Gyx1</td>
<td>Metastatic</td>
<td>NCT02639026 recruiting</td>
</tr>
<tr>
<td>II</td>
<td>Nivolumab</td>
<td>20 Gyx1</td>
<td>mTNBC</td>
<td>TONIC NCT02499367 recruiting</td>
</tr>
<tr>
<td>I</td>
<td>Pembrolizumab</td>
<td>SBRT X3-5 fractions</td>
<td>Solid tumors</td>
<td>NCT02608385 Active, not recruiting</td>
</tr>
<tr>
<td>II</td>
<td>Pembrolizumab</td>
<td>Palliative RT</td>
<td>Luminal BC HER2-negative</td>
<td>NCT03051672 recruiting</td>
</tr>
</tbody>
</table>
Future directions for immune checkpoint inhibitors in BC

- Phases II & III clinical trials are ongoing
- Combined strategies
- **Neoadjuvant/ Adjuvant setting**
- Identification of biomarkers of response
- Development of next generation immunoregulatory antibodies
## Neoadjuvant/Adjuvant setting

<table>
<thead>
<tr>
<th>Phase</th>
<th>Setting</th>
<th>BC</th>
<th>Immunotherapy</th>
<th>Primary Endpoints</th>
<th>Trial &amp; Status</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ib</td>
<td>Neoadjuvant</td>
<td>TNBC</td>
<td>Pembrolizumab and chemotherapy</td>
<td>DLTs</td>
<td>KEYNOTE 173 NCT02622074 recruiting</td>
</tr>
<tr>
<td>I/II</td>
<td>Neoadjuvant</td>
<td>TNBC</td>
<td>Durvalumab and weekly nab-paclitaxel &amp; dd AC</td>
<td>pCR</td>
<td>NCT02489448 recruiting</td>
</tr>
<tr>
<td>II</td>
<td>Neoadjuvant &amp; Adjuvant</td>
<td>TNBC</td>
<td>Atezolizumab and nab-paclitaxel</td>
<td>pCR</td>
<td>NCT02530489 recruiting</td>
</tr>
<tr>
<td>Ib</td>
<td>Adjuvant</td>
<td>TNBC</td>
<td>Durvalumab and peptide vaccine (PVX-410)</td>
<td>Safety Immune response</td>
<td>NCT02826434 recruiting</td>
</tr>
<tr>
<td>II</td>
<td>Adjuvant (after neo-adjuvant CT if non pCR)</td>
<td>Luminal inflammatory</td>
<td>Pembrolizumab and endocrine treatment</td>
<td>DFS</td>
<td>NCT02971748 recruiting</td>
</tr>
</tbody>
</table>
Neoadjuvant/Adjuvant setting

- Phase III trials

**Pembrolizumab**
- KEYNOTE-522
- NCT03036488
- Pembrolizumab + chemo (taxanes – anthracyclines)
- Pembrolizumab 9 cycles
- Primary endpoints: pCR & EFS

**Atezolizumab**
- NeoTRIPaPDL1
- NCT02620280
- Atezolizumab + chemo (Carbo-abrax)
- Chemo (anthra)
- Primary endpoints: EFS
Neoadjuvant/Adjuvant setting

**SWOG S1418/NRG BR006**
RPhIII Pembrolizumab for Residual TNBC post NAC

- **TNBC with \( \geq 1 \) cm residual invasive breast cancer or any + LN after neoadjuvant chemotherapy N=1000

**Pembrolizumab 200 mg IV q 3 weeks x 1y**

1:1

**Observation**

- **Hypothesis:**
  - Pembrolizumab reduces IDFS by 33% c/w observation alone

- **Primary Endpoint:**
  - Invasive DFS in PD-L1-positive and overall cohort

- **Secondary Endpoints:**
  - Toxicity
  - OS
  - DRFS
  - QOL (PROMIS, PRO-CTCAE forms, inflammatory markers)
  - Tissue banking

**Pts: Pusztai/Mamounas**

S. Adams. Immunotherapy for mTNBC. 2016
Future directions for immune checkpoint inhibitors in BC

- Phases II & III clinical trials are ongoing
- Combined strategies
- Neoadjuvant/ Adjuvant setting
- **Identification of biomarkers of response**
- Development of next generation immunoregulatory antibodies
Biomarkers of response

- Response to PD-1/PD-L1 blockade has been linked to
  - PD-L1 expression
    - Both by immune and tumor cells
  - TIL infiltration
  - Mutational load
  - Serum LDH levels

Topalian S et al. Safety, Activity, and Immune Correlates of Anti—D-1 Antibody in Cancer. NEJM2012
Limitations in defining PD-L1 as the biomarker

- Expression is dynamic and focal
  - < Biopsies / full sections
  - < location of the metastases
- Expression depend of the antibody used
- Responses in PD-L1-negative cases
  - 5-20% objective response rate in PD-L1 negative tumors (melanoma & NSCLC)
- Studies have used different threshold of positivity / different cells type
- Stratification < PD-L1 status in clinical trials

TIL infiltration

**Amplify:**
existing anti-tumor immunity (immunogenic chemo, radiation and/or targeted therapy?)

**Boost & Expand:**
developing anti-tumor immunity (immune checkpoint inhibitors; plus immunogenic or targeted therapies?)

**Induce:**
nascent anti-tumor immunity; break tolerance (vaccines, adoptive cell therapy, cytokines?)
Possible trial design using TIL as a biomarker

- TNBC or HER2-positive early-stage breast cancer
- Neoadjuvant chemotherapy ± trastuzumab

- High TILs
  - pCR
    - Excellent prognosis
      - Standard therapy
  - No pCR
    - Ineffective immunity
      - Immuno-modulators (IDO, CD73, adenosine)
      - T-cell checkpoints (PD-1/PL-L1, CTLA-4, TIM-3, LAG-3)

- Low TILs
  - pCR
    - Absent immunity
      - Oncogenic signalling pathways (MEK)
      - Personalized vaccines
      - Adoptive T-cell therapy
  - No pCR
    - Poor prognosis
      - Aggressive therapy

Nature Reviews | Clinical Oncology
Future directions for immune checkpoint inhibitors in BC

- Phases II & III clinical trials are ongoing
- Combined strategies
- Neoadjuvant/ Adjuvant setting
- Identification of biomarkers of response
- Development of next generation immunoregulatory antibodies
Next generation immunoregulatory antibodies

- Main families of immunoregulatory targets
  - TNF receptor superfamily
    - CD40, OX40, 4-1BB, …
  - B7/CD28 family
    - ICOS, VISTA, B7-H3, B7-H4, …
  - Nectin family
    - TIGIT, CD96
  - Butyrophilin family
  - KIR family
  - …
OTHER IMMUNOMODULATORS
Immunomodulators under clinical development in BC

- **Cytokines**
  - Combination Therapy of F16IL2 and Paclitaxel in Solid Tumour Patients (NCT01134250)
  - A Study of Ad-RTS-hIL-12 With Veledimex in Subjects With Breast Cancer (NCT02423902)

- **IDO inhibitors**
  - Study of Chemotherapy in Combination With IDO Inhibitor in Metastatic Breast Cancer (NCT01792050)

- **TLR agonists**
  - Toll-like Receptor (TLR) 7 Agonist, Cyclophosphamide, and Radiotherapy for Breast Cancer With Skin Metastases (NCT 01421017)
Regulated intratumoral expression of IL-12 promotes activation of tumor-infiltrating lymphocytes to drive a cytotoxic immune response
CANCER VACCINES
## BC Tumor-associated antigens

<table>
<thead>
<tr>
<th>Target</th>
<th>Description</th>
</tr>
</thead>
</table>
| HER2                | Human epidermal growth factor  
 Overexpressed in 20-30% of BC                                            |
| CEA                 | Glycoprotein involved in cell adhesion, normally expressed  
 during fetal development                                                  |
| MUC-1               | Membrane glycoprotein involved in immunologic and cell signaling functions  
 Overexpressed in 70% of BC                                                |
| hTERT               | Component of the telomerase complex, a ribonucleoprotein  
 that maintain chromosome integrity during cell proliferation  
 and division                                                            |
| Mammaglobin-A       | Glycoprotein overexpressed in 80% of mBC                                    |
| Cancer testis antigens | Proteins expressed in normal germ cells of the testis and  
 embryonic ovaries and in certain types of cancer                           |

Adapted from Milani et al. Recent advances in the development of breast cancer vaccines. 2014
## Therapeutic BC vaccines

<table>
<thead>
<tr>
<th>Metastatic setting</th>
<th>Vaccines</th>
<th>Phase</th>
<th>Targets</th>
<th>Nb patients</th>
<th>Clinical activity</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>HER2 peptide + GM-CSF</td>
<td>I</td>
<td>HER2</td>
<td>64</td>
<td>NR</td>
<td>Disis 1999</td>
</tr>
<tr>
<td></td>
<td>HER2 peptide (E75) + GM-CSF</td>
<td>I</td>
<td>HER2</td>
<td>14</td>
<td>NR</td>
<td>Murray 2002</td>
</tr>
<tr>
<td></td>
<td>HER2 ICD protein + GM-CSF</td>
<td>I</td>
<td>HER2</td>
<td>29</td>
<td>NR</td>
<td>Disis 2004</td>
</tr>
<tr>
<td></td>
<td>hTERT peptide + montanide + GM-CSF</td>
<td>I</td>
<td>hTERT</td>
<td>19</td>
<td>NR</td>
<td>Domchek 2007</td>
</tr>
<tr>
<td></td>
<td>Survivin-2B peptide</td>
<td>I</td>
<td>survivin</td>
<td>14</td>
<td>NR</td>
<td>Tsuruma 2008</td>
</tr>
<tr>
<td></td>
<td>HER2 peptide + GM-CSF + trastuzumab</td>
<td>I</td>
<td>HER2</td>
<td>22</td>
<td>NR</td>
<td>Disis 2009</td>
</tr>
<tr>
<td></td>
<td>Theratope (STn-KLH + Cy vs KLH+Cy)</td>
<td>III</td>
<td>MUC-1</td>
<td>1028</td>
<td>Negative trial</td>
<td>Miles 2011</td>
</tr>
<tr>
<td></td>
<td>HER2 protein + AS15 + lapatinib</td>
<td>I</td>
<td>HER2</td>
<td>12</td>
<td>NR</td>
<td>Hamilton 2012</td>
</tr>
<tr>
<td></td>
<td>MAM-A DNA vaccine</td>
<td>I</td>
<td>Mammaglobin-A</td>
<td>14</td>
<td>Possible benefit</td>
<td>Tiriveedhi 2014</td>
</tr>
<tr>
<td></td>
<td>PANVAC (poxviral-based) + docetaxel</td>
<td>II</td>
<td>MUC-1 + CEA</td>
<td>48</td>
<td>Possible benefit</td>
<td>Heery 2015</td>
</tr>
<tr>
<td></td>
<td>dHER2 protein + AS15</td>
<td>I/II</td>
<td>HER2</td>
<td>40</td>
<td>Possible benefit</td>
<td>Curigliano 2016</td>
</tr>
<tr>
<td>Cell-based vaccines</td>
<td>Lapuleucel-T</td>
<td>I</td>
<td>HER2</td>
<td>18</td>
<td>SD in 16.7%</td>
<td>Park 2007</td>
</tr>
<tr>
<td></td>
<td>p53-DC</td>
<td>I</td>
<td>p53</td>
<td>26</td>
<td>SD in 42%</td>
<td>Svane 2007</td>
</tr>
<tr>
<td></td>
<td>Allogenic GM-CSF-secreting breast tumor cells + low dose CY and DOX</td>
<td>I</td>
<td>HER2</td>
<td>28</td>
<td>NR</td>
<td>Emens 2009</td>
</tr>
<tr>
<td></td>
<td>Allogenic GM-CSF-secreting breast tumor cells + low dose CY and trastuzumab</td>
<td>II</td>
<td>HER2</td>
<td>20</td>
<td>NR</td>
<td>Chen 2014</td>
</tr>
</tbody>
</table>

Adapted from Cimino-Mathews et al. Oncology 2015
Theratope vaccine

**Phase III trial:**
- Largest double-blind, randomized vaccine study in mBC
- 1028 patients (525 vaccinated)
- MUC-1 peptide vaccine
  - carbohydrate epitope: Sialyl-Tn (STn) conjugated to a protein carrier (keyhole limpet hemocyanin (KLH))
- Low dose of cyclophosphamide to deplete Treg
- Concomitant hormone therapy allowed
- Median TTP: 3.4 months vs 3 months
- Median OS: 23.1 vs 22.1 months

Miles et al. *The Oncologist*. 2011
Theratope vaccine

Phase III, Randomized, prospective trial

Retrospective post-hoc analysis

→ No survival benefit!

→ Survival advantage for patients treated with endocrine therapy
## Preventive BC vaccines

<table>
<thead>
<tr>
<th>Vaccines</th>
<th>Phase</th>
<th>target</th>
<th>Nb patients</th>
<th>Clinical activity</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>HER2 peptide (E75) + GM-CSF</td>
<td>I</td>
<td>HER2</td>
<td>53</td>
<td>possible benefit</td>
<td>Peoples 2005</td>
</tr>
<tr>
<td>Oxidized mannan-MUC-1</td>
<td>Pilot</td>
<td>MUC-1</td>
<td>31</td>
<td>possible benefit</td>
<td>Vassilaros 2013</td>
</tr>
<tr>
<td>HER2 peptide (E75) + GM-CSF</td>
<td>I/II</td>
<td>HER2</td>
<td>187</td>
<td>5y DFS: 89.7% vs 80.2%</td>
<td>Mittendorf 2014</td>
</tr>
<tr>
<td>HER2 peptide (GP2) + GM-CSF</td>
<td>II</td>
<td>HER2</td>
<td>180</td>
<td>5y DFS: 94% vs 85%</td>
<td>Mittendorf 2016</td>
</tr>
<tr>
<td>HER2 peptide (AE37) + GM-CSF</td>
<td>II</td>
<td>HER2</td>
<td>298</td>
<td>5y DFS: 80.8% vs 79.5%</td>
<td>Mittendorf 2016</td>
</tr>
<tr>
<td>dHER2 + AS15</td>
<td>I</td>
<td>HER2</td>
<td>61</td>
<td>NR</td>
<td>Limentani 2016</td>
</tr>
<tr>
<td>HER2 pulsed DC</td>
<td>I/II</td>
<td>HER2</td>
<td>54</td>
<td>NR</td>
<td>Lowenfeld 2016</td>
</tr>
<tr>
<td>Triple peptide</td>
<td>I/II</td>
<td>MUC-1+ HER2+CEA</td>
<td>14</td>
<td>possible benefit</td>
<td>Morena 2016</td>
</tr>
</tbody>
</table>

Adapted from Cimino-Mathews et al. Oncology 2015
REDEFINING THE STANDARD OF CARE

RECEIVES PRIMARY TREATMENT

- Surgery
- Chemotherapy
- Radiation

Disease free "survivor"

HER2 Status

- HER2, 3+ (20-25% of patients)
- HER2, 1+/2+ (50-60% of patients)
- HER2, 3+ High Risk (20-25% of patients)

Standard of Care

- Multiple, including Herceptin®
- No FDA approved therapies
  - PRESENT trial
  - NeuVax + trastuzumab
  - NeuVax + trastuzumab
SN-33 PHASE 2 HER2 IHC 1+/2+ (N=45)

NeuVax vaccine

- HER2-derived peptide, HLA-A2/A3+
- MHC class I epitope – CD8⁺ T cell response
- Phase III trial: Prevention of Recurrence in Early-Stage, node-positive BC with low of Intermediate HER2 Expressions with NeuVax™ Treatment (PRESENT)
  - HER2 1+/2+ (IHC)
  - NeuVax + Sargramostim, GM-CSF
  - 758 patients included
NeuVax vaccine

- HER2-derived peptide, HLA-A2/A3+
- MHC class I epitope – CD8+ T cell response

**Phase III trial:**
- Prevention of recurrence in Early-stage, node-positive BC with low or Intermediate HER2 expressions with NeuVax™ Treatment (PRESENT)
- HER2 1+/2+ (IHC)
- NeuVax + Sargramostim, GM-CSF

758 patients included

**Press Release – June 2016**
- No survival advantage (interim analysis)
- The trial is stopped!
Cancer vaccines in BC

- HER2 is the most used antigen
- Disappointing results in the metastatic setting
- Higher potential for secondary prevention?
- Combining BC vaccines with other strategies
Future directions: personalized Immunotherapy

- Neoantigen-based cancer immunotherapy

  - Identification of tumor-specific neo-antigens
    - Sequencing (RNAseq & Whole exome sequencing)
    - In silico HLA binding prediction tools
    - Prioritization of potential unique tumor antigens

- Generation of highly personalized DNA-based vaccine or mature DC-based vaccine
Personalized vaccine against mTNBC

- **Phase I trial**: Safety and Immunogenicity of a Personalized Synthetic Long Peptide Breast Cancer Vaccine Strategy in Patients With Persistent TNBC Following Neoadjuvant Chemotherapy

  - Started in September 2015 at Siteman Cancer Center (Washington University School of Medicine) - (NCT02427581)

  - Primary outcome:
    - Safety of the vaccine regimen

  - Secondary outcome:
    - Immunogenicity of the vaccine regimen
ADOPTIVE T-CELL THERAPIES
Adoptive T-cell therapies

- Administration of autologous T cells with genetic material transferred into the cell to redirect them to target breast cancer cells rather than their usual target.

- T cells are activated by
  - Primary signal: CAR (Chimeric Antigen Receptor)
  - Second signal: costimulatory domains (CD28 or CD137)
Adoptive T-cell therapies

- Majority of early-phase trials are being performed to treat **B cell malignancies**
- Minority of trials targeting solid cancers
  - Ongoing studies including BC patients with CAR T cells targeting:
    - HER2 (NCT02547961)
    - CEA (NCT 02349724)
    - Mesothelin (NCT02792114)
    - MUC-1 (NCT02587689)
    - NKG2D-ligands (NCT03018405)
Conclusions

- Growing interest to identify immunotherapeutic approaches to treat BC
  - Multiple ongoing trials !!!
  - Promising results in TNBC
  - Potential future success of synergistic combinations

- Remaining challenges
  - Identification of the adequate strategy for the appropriate disease
  - Identification of biomarkers of response → selection of patients
Thank you for your attention
Classifying Cancers Based on T-cell Infiltration and PD-L1
### Classifying Cancers Based on T-cell Infiltration and PD-L1

<table>
<thead>
<tr>
<th>Tumor Microenvironment</th>
<th>Early BC</th>
<th>Ovarian</th>
<th>Melanoma</th>
</tr>
</thead>
<tbody>
<tr>
<td>Type I TIL+/PD-L1+</td>
<td>21%</td>
<td>57.4%</td>
<td>38%</td>
</tr>
<tr>
<td>Type II TIL-/PD-L1-</td>
<td>24%</td>
<td>5.1%</td>
<td>41%</td>
</tr>
<tr>
<td>Type III TIL-/PD-L1+</td>
<td>2%</td>
<td>0%</td>
<td>1%</td>
</tr>
<tr>
<td>Type IV TIL+/PD-L1-</td>
<td>53%</td>
<td>37.4%</td>
<td>20%</td>
</tr>
</tbody>
</table>

**References**
- Buisseret 2016
- Webb 2016
- Teng 2015

Type I: Adaptive immune resistance  
Type II: Immunological ignorance  
Type III: Intrinsic Induction  
Type IV: Tolerance