Sarcoma Immunotherapy: The Future Is Near!

Seth M. Pollack, MD
Assistant Member
Outline

1. Introduction
2. Immune Response to Sarcoma Subtypes
3. Targeting NY-ESO-1
4. Looking into tumors
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Sarcoma is a heterogeneous group of diseases

Sarcoma (1% of all cancer - percentages include children and adults)

<table>
<thead>
<tr>
<th>Bone Sarcomas (10%):</th>
<th>Soft Tissue Sarcoma (STS)</th>
</tr>
</thead>
<tbody>
<tr>
<td>• Osteosarcoma</td>
<td>GIST (18%)</td>
</tr>
<tr>
<td>• Ewings Sarcoma</td>
<td>RMS</td>
</tr>
<tr>
<td>• Chondrosarcoma</td>
<td>Other “special” STS:</td>
</tr>
<tr>
<td>• Giant Cell Tumor</td>
<td>Kaposi’s DFSP etc.</td>
</tr>
<tr>
<td>• Other</td>
<td></td>
</tr>
</tbody>
</table>

Non-GIST
Non-RMS
Not special STS:

Or, in other words, what I usually call: STS

Ducimetriere et al 2011
Is STS really that rare?

Rare cancers are more than 25% of all adult cancers.

Approximately 12,000 Americans are diagnosed with STS annually (non-gist, per American Cancer Society).
5000 people die annually.

Rare cancers lead to important scientific breakthroughs:

- Merkel Cell Carcinoma – 1500 patients/year
- ALL – 6590 patients/year
- Hodgkins Lymphoma – 8,500 patients/year
- Allogeneic transplant - 8000 patients/year
- Testicular cancer – 8720 patients/year

Greenlee et al. et al 2010
There are over 50 STS subtypes

UPS – very highly mutated subtypes
LMS – less than UPS, but also highly mutated
SS – most common translocation associated sarcoma
Liposarcoma – genetically “simple”

Undifferentiated Pleomorphic Sarcoma (UPS)
Liposarcoma
Leiomyosarcoma
Other
Synovial

Total=438
“Liposarcoma” is actually at least 3 diseases

Well-differentiated (WD)

- WD/DD
- MRCL
- Pleomorphic
- Other/NOS

De-differentiated (DD)

- 77.06 mm
- 83.98 mm
- 49.79 mm
Basic Sarcoma Vocabulary

- Localized Disease
- Locally recurrent disease
- Metastatic Disease
- Neoadjuvant Therapy
- Adjuvant Therapy
- Palliative Therapy
What are the FDA approved drugs for sarcoma?

• First line therapy: Single Agent Doxorubicin (Average 4.6 month PFS)
• Recent approval: olaratumab (may improve OS combined with dox, phase III results pending)
  • Votrient (3 month PFS, no proven OS benefit – approved for STS other than liposarcomas)
  • Eribulin (2 month OS benefit, approved for liposarcoma only)
  • Trabectedin (3 month PFS improvement liposarcoma and leiomyosarcoma)
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Different Types of Immunotherapy

- Cytokine
- Checkpoint inhibitor
- T cell therapy
- Vaccine
- Other
Checkpoint Inhibitors

Checkpoint Inhibition in Melanoma

A

Nivolumab plus Ipilimumab
Median Change: Decrease of 68.1%

Ipilimumab
Median Change: Increase of 5.5%

Best Change from Baseline in Target-Lesion Volume (%)
**Death or Disease Progression**

- Nivolumab plus Ipilimumab: 25/37
- Ipilimumab: 30/72

**Median Progression-free Survival**

- Nivolumab plus Ipilimumab: 4.4 mo (95% CI 2.8–5.7)
- Ipilimumab: NR

Hazard ratio: 0.40 (95% CI 0.23–0.68)
P < 0.001

**No. at Risk**

<table>
<thead>
<tr>
<th>Treatment</th>
<th>72</th>
<th>54</th>
<th>45</th>
<th>38</th>
<th>20</th>
<th>1</th>
<th>0</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nivolumab plus Ipilimumab</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ipilimumab</td>
<td>37</td>
<td>20</td>
<td>9</td>
<td>6</td>
<td>2</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

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C Patient with Melanoma

TCR recognition of tumor requires MHC
Retrospective Analysis

- Two highly mutated, genetically complex STS types: UPS and LMS
- Two genetically “simple” STS types liposarcoma (WD/DD and MRCL) and SS

<table>
<thead>
<tr>
<th>Sarcoma Type</th>
<th>N</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Liposarcoma</td>
<td>27</td>
<td>33%</td>
</tr>
<tr>
<td>WD/DD</td>
<td>15</td>
<td>56%</td>
</tr>
<tr>
<td>Myxoid/round cell</td>
<td>12</td>
<td>44%</td>
</tr>
<tr>
<td>LMS</td>
<td>19</td>
<td>23%</td>
</tr>
<tr>
<td>Non-uterine</td>
<td>17</td>
<td>89%</td>
</tr>
<tr>
<td>Uterine</td>
<td>2</td>
<td>11%</td>
</tr>
<tr>
<td>Pleomorphic</td>
<td>20</td>
<td>25%</td>
</tr>
<tr>
<td>SS</td>
<td>15</td>
<td>19%</td>
</tr>
<tr>
<td>Monophasic</td>
<td>13</td>
<td>87%</td>
</tr>
<tr>
<td>Biphasic</td>
<td>1</td>
<td>7%</td>
</tr>
<tr>
<td>Unknown</td>
<td>1</td>
<td>7%</td>
</tr>
</tbody>
</table>
Focused Clustering Analysis

- Genes included if $p \leq 0.05$ difference for at least one subtype
- 367/760 genes
- Regions defined based on dendrogram clustering
- Most striking separation for genes related to antigen presentation and T cell infiltration

Pollack et al., *Cancer*. In press
Class I MHC Molecules

Pollack et al., *Cancer*. In press
Genes Related to T cell Infiltration

Pollack et al., *Cancer*. In press
TCR Vβ Sequencing

Pollack et al., *Cancer*. In press
Maybe those STS types rely more on PD-L1/L2?

Pollack et al., *Cancer*. In press
PD-1 Immunohistochemistry

4+ (above) and 5+ (below)
Staining showing infiltration with PD-1+ cells

Pollack et al., *Cancer*. In press
**PD-L1 Immunohistochemistry**

Very high PD-L1 Staining in a UPS tumor

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Pollack et al., *Cancer*. In press
Pollack et al., *Cancer.* In press
PD-1 Inhibition in STS

Burgess/Tawbi ASBO 2016: SARC28 – 40 patients (4 of 10 UPS responses, LMS – 0/10, SS – 1/10, Lipo 1/10)

Suzanne George ASBO 2016: 12 Uterine LMS - no responses
TAM impact for STS But are Killed by Trabectedin

TAM Markers in Uterine LMS

Ganjoo et al., Am J Clin Oncol 2011

Germano et al. Cell 2013
**FHCRC 9717: Avelumab (anti-PD-L1) + trabectedin**

For patients with liposarcoma and leiomyosarcoma.

<table>
<thead>
<tr>
<th>Drug</th>
<th>Week</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
<th>8</th>
<th>9</th>
<th>10</th>
<th>11</th>
<th>12*</th>
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</thead>
<tbody>
<tr>
<td>Avelumab</td>
<td>●</td>
<td></td>
<td>●</td>
<td></td>
<td></td>
<td>●</td>
<td></td>
<td>●</td>
<td></td>
<td>●</td>
<td></td>
<td></td>
<td>●</td>
</tr>
<tr>
<td>Trabectedin</td>
<td>●</td>
<td>●</td>
<td></td>
<td></td>
<td></td>
<td>●</td>
<td></td>
<td>●</td>
<td></td>
<td>●</td>
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</tbody>
</table>

PI: Pollack
FHCRC 9624: Doxorubicin + Pembrolizumab

Cycle 1:
Pembrolizumab
200mg
single agent

Cycle 2-7:
Pembrolizumab
200mg
+ Doxorubicin

Cycle 8:
Pembrolizumab
200mg
single agent

Phase I
3+3 design:
Part 1: Doxorubicin 45 mg/m²
Part 2: Doxorubicin 75 mg/m²

Phase II
2-stage design:
Part 1: 20 patients (requires ≥ 2 responses)
Part 2: 15 patients

PI: Pollack
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NY-ESO-1 is a CT Antigen Expressed by SS

Cancer Testis Antigens (CTA) are well established self-antigens

- Not expressed at the protein level in most normal tissues
- Unknown biologic function
- Epigenetically regulated
- NY-ESO-1, PRAME, MAGE family antigens included
- NY-ESO-1 frequently and homogenously expressed by SS
- Positive 20/25; homogenous 14/20

Jungbluth et al., Int J Cancer 2001
Screening Tumors for Tissue Bank for CTA Expression
MRCL Expresses NY-ESO-1 Homogenously

- 25 cases tested, all positive
- >70% of cases, homogenous.
- MRCL cell lines can be lysed by NY-ESO-1 specific effectors
- No other disease expresses NY-ESO-1 with this frequency

Pollack et al., *Cancer* 2012
Culturing NY-ESO-1 Specific Cells

SS and MRCL Leukapheresis products lack clear tet+ populations.

After 2 stimulations, three wells were identified with clear tet+ populations.

Final product contained $57 \times 10^9$ cells and was over 94% CD8+, tet+.

Pollack et al. JITC 2014
These Cells Kill Tumor Lines

Pollack et al. JITC 2014
However, because of safety issues with this trial, this regimen was abandoned.
LV305 is a novel hybrid viral vector gene delivery system (ZVex™) that expresses NY-ESO-1 RNA designed to target DCs *in vivo* and stimulate CD8 T cell responses against this cancer testis antigen.

| ZVex |
|---|---|---|
| **Envelope:** | **Genome:** |
| DC Targeting: | Safety: |
| • Sindbis virus envelope targets the Dendritic Cell receptor, CD209 (DC-SIGN) | • 3rd generation lentiviral vector backbone |
| ZVex Envelope: | • Modified to be replication incompetent and integration deficient |
| DC Targeting: | Specificity: |
| • Sindbis virus envelope targets the Dendritic Cell receptor, CD209 (DC-SIGN) | • Vpx prevents degradation within DCs |

Slide courtesy of Immune Design
Increased NY-ESO-1 Tet+ Cells in A0201+ Patients

Tetramer Staining in A2+ Patients (Percent of CD8+ Cells)

PBMC Elispot

Pollack et al. Submitted
Pre and Post Tx TCR sequencing

1.68% of Pre-tx T cells

2.51% of Post-tx T cells

Clonality 0.11

Clonality 0.18
Durable Tumor Regression Following LV305

Percent Change by RECIST From Baseline (%) vs. Days Post First Dose of LV305

RECIST PR

Pollack et al. Submitted
Durable Tumor Regression Following LV305

Pollack et al. Submitted
Survival on the LV305 Vaccine Trial

All Sarcomas
N = 24 (progressed = 18)
95% CL: 2.6, 14.4

Proportion Progression Free

All Sarcomas
N = 24 (deaths = 4)

Proportion Surviving

Pollack ASCO 2016
## Outcomes on LV305 and Approved Drugs For Sarcoma

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Indication</th>
<th>PFS</th>
<th>OS</th>
</tr>
</thead>
<tbody>
<tr>
<td>LV305</td>
<td>STS</td>
<td>4.6 mo</td>
<td>Not reached</td>
</tr>
<tr>
<td>Pazopanib</td>
<td>STS (not MRCL)</td>
<td>4.7 mo</td>
<td>12.5 mo</td>
</tr>
<tr>
<td>(Votrient)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Trabectedin</td>
<td>MRCL after Anthracycline (all STS in EU)</td>
<td>4.2 mo</td>
<td>12.4 mo</td>
</tr>
<tr>
<td>(Yondelis)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Eribulin</td>
<td>MRCL after Anthracycline</td>
<td>2.6 mo</td>
<td>13.5 mo</td>
</tr>
<tr>
<td>(Halaven)</td>
<td></td>
<td></td>
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</tbody>
</table>
CMB +/- Atezolizumab

**Combination Arm**
Sequential regimen of LV305 and G305 for 3 months with atezolizumab q3w up to 2 yrs

**Control Arm**
Atezolizumab q3w up to 2 yrs

Screen, biopsy, safety run-in
- ≤ 80 pts with locally advanced, relapsed, or metastatic synovial sarcoma or MRCL

Randomize

Biopsy, then follow up to 2 years
-Endpoints of PFS, safety, PFR (at 6 mo), ORR, duration of response, and OS

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Sarcomas Can Be Quite Large and Are Relatively Accessible
Presage’s Civo Device

1. Device Loading

2. Ultrasound Measurements

3. Microinjection

4. Confirmation and Skin Marking

PI: Pollack
Slide courtesy of Presage
FHCRC 9145: Intratumor TLR4 Agonist + Radiation

TLR4 agonist, GLA

conversion of immunosuppressive (M2) TAM with high IL-10, TGFβ to immune-activating (M1) TAM with high MHC, IL-12

TAM

T cell

tumor cell

release of tumor proteins

radiation

increased antitumor T cell immunity

PI: Pollack
FHCRC 9145: Schema

• Two, 6-patient cohorts treated: 5 mcg and 10 mcg.
• All patients were required to have unresectable of metastatic disease.
• GLA was injected directly into a tumor.
• Radiation was started to the injected tumor during the first week of treatment.
• Patients received high dose (generally 50 Gy) in few fractions (generally 5 or less).
• Current cohort using 20 mcg.

Weekly GLA-SE injections through week 8.
TLR4 Agonism Can Convert M2 to M1
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