Rationale for Combining Immunotherapy with Chemotherapy or Targeted Therapy

Qing Yi, MD, PhD
Staff and Chair,
Department of Cancer Biology
Betsy B. DeWindt Endowed Chair for Cancer Research
Lerner Research Institute
Cleveland Clinic
Why Immunotherapy for Cancers

• Current therapeutics unable to cure cancers
• Vaccines are the best defense against infectious diseases
• Powerful and yet specific immune system:
  – Able to reject mismatched organs
  – Immunological memory
  – Polyclonal immune responses
  – Target different antigens
Idiotype: Unique Amino Acid Sequences in CDRs

Variable Region - Heavy Chain

1 31 35 50 65 95 102 113

S

NH₂

FW1 FW2 FW3 FW4

CDR1 CDR2 CDR3

Variable region

Light chain

Heavy chain

Constant region

B cell lymphoma

Multiple Myeloma

B cell lymphoma
Preclinical Studies of Id Vaccines

- Active immunization with idiotype was first demonstrated to induce resistance to tumor challenge in the early 1970’s – *Lynch et al. Proc Natl Acad Sci, 1972.*

- Optimal immunization required conjugation to a strongly immunogenic carrier protein, such as keyhole-limpet hemocyanin (KLH) – *Kaminski et.al. J Immunol, 1987.*

- Use of GM-CSF as an adjuvant to the Id-KLH vaccine facilitated the induction of tumor-specific CD8+ T cell responses. – *Kwak et.al. PNAS, 1996.*

- Dendritic cells present Id fragments and induce type-1 T cell immunity – *Yi et.al. Br J Haematol, 1998.*
## Phase III Id-KLH+GM-CSF Vaccine Trials in FL

<table>
<thead>
<tr>
<th>Sponsor</th>
<th>Induction therapy</th>
<th>Randomize</th>
<th>Vaccination</th>
<th>DFS/PFS/TTP</th>
</tr>
</thead>
<tbody>
<tr>
<td>NCI/Biovest</td>
<td>PACE</td>
<td>CR/CRu</td>
<td>Id-KLH+GM-CSF or KLH+GM-CSF</td>
<td>Significant</td>
</tr>
<tr>
<td>Genitope</td>
<td>CVP</td>
<td>CR/CRu, PR</td>
<td>Id-KLH+GM-CSF or KLH+GM-CSF</td>
<td>Not significant</td>
</tr>
<tr>
<td>Favrille</td>
<td>Rituximab</td>
<td>CR/CRu, PR/SD</td>
<td>Id-KLH+GM-CSF or Placebo+GM-CSF</td>
<td>Not significant</td>
</tr>
</tbody>
</table>
NCI/Biovest Phase III trial: DFS from Randomization

\[ N = 117 \]
Id-KLH (BiovaxID) N = 76
Control vaccine N = 41

Median Follow-up
56.6 mo (range 12.6 – 89.3)

Median DFS
Id-KLH (BiovaxID) = 44.2 mo
Control vaccine = 30.6 mo

Vaccination with Id Proteins in MM

• Bergenbrant, Yi et al, 1996
  – Id/Alu vaccines in 5 (4 untreated, 1 after HDT); IFN-γ in 3, all SD

• Ősterborg, Yi et al, 1998
  – Id-Alu/GM-CSF in 5 (2 untreated, 3 after HDT); IFN-γ in 5; 4 SD and 1 partial remission (65% reduction)

• Massaia et al, 1999
  – Id-KLH/GM-CSF in 12 (CR after HDT); DTH in 8/10 and T-cell proliferation in 2; no favorable clinical outcome

• Mellstedt et al, 2003
  – Id/IL-12+GM-CSF in 6 (stage I MM); T cells in 3, tumor reduction in 4 (one with complete molecular remission)
Intranodal Id-DC vaccination

• To date 10 (7 IgG and 3 IgA) indolent or smoldering MM enrolled
• CD4 count > 600/µl
• Each received, per injection, 14.6 x 10^6 (range 1.2-35.6 x 10^6), Id- and KLH-pulsed, CD40L(Immunex)-matured DCs
• ELISPOT assays, proliferation and DTH
DC Vaccines Induce Th1-Type Response

IFN-γ Response

IL-4 Response

Yi et al, Br J Haematol 2010
Id-Specific CTLs Detected 4 Weeks After Vaccination

Yi et al, Br J Haematol 2010
Vaccination Induces PR or SD in Most Patients

Yi et al, Br J Haematol 2010
Potential Reasons for Low Clinical Responses with Vaccine Therapy

**Afferent Phase**
- Magnitude of T cell response
- High avidity T cells

**Efferent Phase**
- Trafficking
- Immunosuppressive mechanisms

**Vaccination**
- Dendritic cell vaccines
- Target multiple tumor antigens
- DNA vaccines

**Stromal factors**
- Immunosuppressive cytokines
- Immunosuppressive enzymes
- Co-inhibitory ligands

**Tumor factors**
- Antigen loss
- Ag processing defects
- HLA loss or down-regulation
- Co-inhibitory ligands
- Immunosuppressive cytokines
- Immunosuppressive enzymes

**CD4+ T cells**

**CD8+ T cells**

**Co inhibitory receptors**
- eg: PD-1

**Macrophage**

**Tregs**

**Tolerant DCs**

**Follicular Lymphoma**
Approach

Combinational therapy of vaccines and chemotherapy to break immune suppression
Lenalidomide and IMiDs

- Novel drugs for treatment of myeloma and B-cell lymphomas
- Inhibiting angiogenesis and TNF-\(\alpha\)
- Immunomodulatory
  - Activating NK cells and increasing NK numbers
  - Enhancing T cell proliferation and cytokine production
  - Polarizing T-cell immunity toward Th1 responses
  - Repairing T-cell immunologic synapse dysfunction
Lenalidomide Enhances Vaccine Responses in Myeloma

- Myeloma patients receiving lenalidomide randomized to A or B
- Vaccine: pneumococcal 7-valent conjugate vaccine (PCV)
- PCV-specific humoral and cellular responses greater in Cohort B than Cohort A

Lenalidomide Enhances Vaccine Responses in Lymphoma

DNA vaccination (100 ug/mouse)

Day 0 1 14 28 35 42

A20 Lymphoma

Lenalidomide: 5 mg/kg i.p. for 35 consecutive days

Collect tumor-free mice A20 Lymphoma

(100 ug/mouse)

Day 0 1 14 28 35 42

A20 Lymphoma (re-challenge)

Day after tumor re-challenge

Overall survival (%)

Vac+Len Len Vac PBS

P = 0.046 c/w vac
P < 0.001 c/w PBS

P < 0.01 c/w PBS

P < 0.01 c/w saline

Overall survival (%)

Vac+Len Len Vac PBS

Day after tumor re-challenge

Days after tumor challenge
Lenalidomide Enhances Vaccine Responses in Lymphoma A20 Lymphoma Challenge

Lenalidomide: 5 mg/kg i.p. for 35 consecutive days

DNA vaccination (100 ug/mouse)

Day 0 | 1 | 14 | 28 | 35 | 37 | 39 | 42 | 56

T cell depletion T cell depletion

Days after tumor challenge

Overall survival (%)

Vac + Len
CD4 depletion
CD8 depletion
CD4CD8 depletion
PBS

P=0.0008 c/w CD8 depletion
P<0.001 c/w CD4/CD8 depletion or PBS

Sakamaki et al, Leukemia, in press
Lenalidomide Enhances Vaccine Responses in Lymphoma

Sakamaki et al, Leukemia, in press
Cyclophosphamide

- An alkylating agent to treat cancers
- Immunosuppression
- Immunopotentiation
  - Selective inhibition of Treg
  - T-cell activation by regulating type-1 interferons
  - Action of NK cells and their activity
Cyclophosphamide in Cancer Patients

Depleting Treg

Activating T and NK cells

Gemcitabine

• An anti-metabolite analogue of nucleoside
• Induction of cancer apoptosis by dysfunctional DNA synthesis
• Immunopotentiation
  – Selectively eliminating MDSCs
  – Promoting T-cell immunity
  – Inhibiting B-cell proliferation
Gemcitabine in Tumor-Bearing Mice

Depleting MDSCs

Reducing immunosuppression

Approach

Combinational therapy of vaccines and novel agents to break immune suppression
Myeloma Cells Express B7-H1 and PD-1

- **MOPC-21** (Murine) - B7-H1: 100, PD-1: 358
- **MPC-11** (Murine) - B7-H1: 528, PD-1: 329
- **HOPC-1F/12** (Murine) - B7-H1: 82, PD-1: 358
- **MOPC-315** (Murine) - B7-H1: 49, PD-1: 60.7
- **5TGM1** (Murine) - B7-H1: 358, PD-1: 88
- **C1.18.4** (Murine) - B7-H1: 329, PD-1: 88
- **U266** (Human) - B7-H1: 20.4, PD-1: 88.4
- **RPMI-8226** (Human) - B7-H1: 88.4, PD-1: 57.3
- **MM.1S** (Human) - B7-H1: 42.3, PD-1: 60.7
- **ARP-1** (Human) - B7-H1: 1.7, PD-1: 88
- **XG-1** (Human) - B7-H1: 49.2

B7-H1 and PD-1 expression levels are shown for both murine and human myeloma cell lines.
PD-1+ T cells and Treg Are Increased in Tumor-Bearing Mice

**A**

<table>
<thead>
<tr>
<th></th>
<th>Spleen</th>
<th>Tumor</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tumor-bearing</td>
<td>32%</td>
<td>52%</td>
</tr>
<tr>
<td>Tumor</td>
<td></td>
<td>69%</td>
</tr>
</tbody>
</table>

**B**

<table>
<thead>
<tr>
<th></th>
<th>Normal</th>
<th>Tumor-bearing</th>
</tr>
</thead>
<tbody>
<tr>
<td>FOXP3</td>
<td>3.9%</td>
<td>9.8%</td>
</tr>
<tr>
<td>CD25</td>
<td>14%</td>
<td>24%</td>
</tr>
</tbody>
</table>
Blocking B7-H1 or Neutralizing IL-10 Enhance Vaccine Effects

Qian et al, Blood 2009
Combination Immunotherapy

Afferent phase
- Magnitude of T-cell response
- High avidity T cells

Vaccination
- Novel or multiple tumor antigens
- Novel adjuvants
- Vaccinate SCT donors

Combination anti-cancer immunotherapy

Efferent phase
- Trafficking
- Tolerance mechanisms

Tumor and stromal immunosuppressive factors
- Co-inhibitory ligands
- Immunosuppressive cytokines
- Indoleamine 2,3-dioxygenase (IDO)
- Arginase
- Nitric-oxide synthase (NOS)