Immunotherapies for virus-associated cancers: A new paradigm in personalized medicine
Acknowledgements

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Hallmarks of Cancer
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Sustaining proliferative signaling

Resisting cell death

Evading growth suppressors

Inducing angiogenesis

Activating invasion and metastasis

Enabling replicative immortality

Cell (2000) 100: 57-70
Hallmarks of Cancer: The viral link

HTLV1, HCV, EBV, HBV, HPV & KSHV

Sustaining proliferative signaling

HTLV1, HCV, EBV, HBV, HPV, CMV & KSHV

Resisting cell death

HTLV1, HCV, EBV, HBV, HPV, CMV & KSHV

Inducing angiogenesis

HTLV1, HCV, EBV, HBV, HPV, CMV & KSHV

Enabling replicative immortality

HTLV1, HCV, EBV, HBV, HPV & KSHV

Evading growth suppressors

HTLV1, HCV, EBV, HBV, HPV & KSHV

Activating invasion and metastasis

Modified from Cell (2000) 100: 57-70
Hallmarks of Cancer: The viral link

Emerging Hallmarks

- Deregulating cellular energetics
  - HTLV1, HCV, EBV, HBV, HPV, CMV & KSHV

- Avoiding immune destruction
  - HTLV1, HCV, EBV, HBV, HPV, CMV & KSHV

- Genome instability and mutation
  - HTLV1, HCV, EBV, HBV, HPV, CMV & KSHV

- Tumor-promoting Inflammation
  - HTLV1, HCV, EBV, HBV, HPV, CMV & KSHV

Enabling Characteristics
Hallmarks of Cancer: The viral link

EBV
- Immune Modulation
- Apoptosis Regulation
- Proliferation
- Genetic Instability

HBV
- Immune Modulation
- Apoptosis Regulation
- Proliferation
- Genetic Instability

HPV
- Immune Modulation
- Apoptosis Regulation
- Proliferation
- Genetic Instability

KSHV
- Immune Modulation
- Apoptosis Regulation
- Proliferation
- Genetic Instability
Virus-associated cancers: Potential targets for immunotherapy

<table>
<thead>
<tr>
<th>VIRUS</th>
<th>ASSOCIATED CANCER</th>
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</thead>
<tbody>
<tr>
<td>HBV</td>
<td>Hepatocellular carcinoma</td>
</tr>
<tr>
<td>HCV</td>
<td>Hepatocellular carcinoma</td>
</tr>
<tr>
<td>HPV</td>
<td>Cervical cancer; Vaginal cancer; vulvar cancer; Oropharyngeal cancer; Anal cancer; Penile cancer; Squamous cell carcinoma of the skin</td>
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<tr>
<td>EBV</td>
<td>Burkitt lymphoma; Non-Hodgkin lymphoma; Hodgkin lymphoma; Nasopharyngeal carcinoma</td>
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<tr>
<td>KSHV (HHV8)</td>
<td>Kaposi sarcoma</td>
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<tr>
<td>HTLV1</td>
<td>Adult T-cell leukaemia/lymphoma</td>
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<tr>
<td>CMV</td>
<td>Glioblastoma</td>
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</table>
Epstein-Barr virus-associated malignancies

- Post-transplant lymphoproliferative disease (PTLD)
- Nasopharyngeal Carcinoma
- Hodgkin’s Lymphoma

Cytomegalovirus-associated malignancies

- Glioblastoma multiforme
EBV-associated post-transplant lymphoproliferative disease (PTLD)

Organ Transplant

Intensive Immunosuppression

EBV-infected cells proliferate

Polyclonal lymphomas

Loss of Virus-specific CD8+ T cells
Genesis of EBV-associated lymphomas in Transplant Patients

**Stem Cell Tx**
- EBV transferred in the graft
- Includes EBV+ B Cells
- Tx recipient EBV-specific CTL immunity
- PTLD emerges of donor origin

**Solid Organ Tx**
- EBV transferred from the graft
- Solid organ
- EBV donated with graft is released
- Infects recipient B cells
- EBV CTL immunity
- PTLD emerges of recipient origin
Pre & Post autologous CTL immunotherapy for PTLD in SOT recipient

Preinfusion  Postinfusion
Liver CT scan Pre & Post autologous CTL immunotherapy for PTLD in SOT recipient

Preinfusion

Postinfusion

2 weeks

20 weeks
Summary of clinical outcome of T cell immunotherapy for EBV PTLD

(data collated from studies carried out by multiple groups)
Alternative strategy for adoptive immunotherapy to treat PTLD

EBV-specific T cells from unrelated HLA matched healthy donors?

Safe - Yes
GvHD - No
Therapeutic - Yes
Prophylactic - Yes

Haque and Crawford, 2003 Lancet Oncology
Haque and Crawford, 2010 Transplantation
Gandhi et. al. 2007 Amer. J. Transplant
Heslop et. al. 2010 Blood
Allogeneic EBV-specific T cells to treat EBV-associated PTLD
Viral Gene Expression of EBV-associated type II malignancies

- **Latency I**: EBNA1, EBERs
  - Burkitt’s Lymphoma

- **Latency II**: EBNA1, EBERs
  - Hodgkin’s lymphoma
  - Nasopharyngeal carcinoma
  - LMP1
  - LMP2

- **Latency III**: EBNA1-6, EBERs
  - Post-transplant lymphomas
World-wide incidence of EBV-associated nasopharyngeal carcinoma
NPC immunotherapy: multiepitope
Technology: E1-LMPpoly™

<table>
<thead>
<tr>
<th>Epitope Sequence</th>
<th>Antigen</th>
<th>HLA Restriction</th>
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<tbody>
<tr>
<td>PYLFWLAA</td>
<td>LMP2A</td>
<td>A23, A24, A30</td>
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<tr>
<td>SSCSSCPLSKL</td>
<td>LMP2A</td>
<td>A24</td>
</tr>
<tr>
<td>TYGPVFMC</td>
<td>LMP2A</td>
<td>A24</td>
</tr>
<tr>
<td>RRRWRRRLTV</td>
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<td>B27</td>
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<tr>
<td>LLSAWILTA</td>
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<td>A2.01</td>
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<td>WTLVLLI</td>
<td>LMP2A</td>
<td>B63</td>
</tr>
<tr>
<td>CPLSKILL</td>
<td>LMP2A</td>
<td>B8</td>
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NPC immunotherapy Production Process

[Diagram showing the production process of NPC immunotherapy, including a patient, injection, E1-LMPpoly, white blood cells, and killer T cells.]
NPC Therapy Production Process

Cellular Therapy Production Suite (Q-Gen)
T cell expansions from stage IV NPC patients using E1-LMPpoly™

<table>
<thead>
<tr>
<th>11ChLe E1-LMPpoly mediated T cell expansion</th>
<th>Control</th>
<th>LMP-1/2</th>
<th>EBNA-1</th>
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<tbody>
<tr>
<td>Day 0</td>
<td>0.37%</td>
<td>0.27%</td>
<td>0.24%</td>
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<tr>
<td>Day 14</td>
<td>0.26%</td>
<td>27.9%</td>
<td>3.22%</td>
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</table>

(IFN-γ) CD8
E1-LMPpoly™ is highly efficient in expanding LMP1/2 and EBNA1-specific T cells from stage IV NPC patients.

10-200 fold expansion of LMP1/2 and EBNA-1-specific T cells
E1-LMPpoly T cell therapy is safe with minimal side effects

<table>
<thead>
<tr>
<th></th>
<th>G0</th>
<th>G1</th>
<th>G2</th>
<th>G3</th>
<th>G4</th>
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<tr>
<td>Flu-like Sx</td>
<td>14/15</td>
<td>1/15</td>
<td>-</td>
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<tr>
<td>Malaise</td>
<td>12/15</td>
<td>3/15</td>
<td>-</td>
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<td>2/15</td>
<td>-</td>
<td>-</td>
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<td>LBP</td>
<td>14/15</td>
<td>1/15</td>
<td>-</td>
<td>-</td>
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</table>

- One SAE occurred for subject 17TaHo
Impact of EBV-specific T cell therapy on OS and TTP of stage IV NPC patients

- Treated (n = 14)

  - Median survival: 523.0 days
  - Range of TTP: 4.5-18.2m
  - Mean TTP: 10.2m
  - Median TTP: 9.2m

Graph showing survival and times to progression (TTP) for treated patients.
EBV-specific T cell numbers and time to diagnosis of progressive disease

Spearman r = 0.09934
p = ns
Future Direction: Combining EBV T cell immunotherapy with chemo/radiotherapy

Phase I/II open-label clinical trial of autologous Epstein-Barr virus- specific T cell therapy as consolidative treatment following chemotherapy for metastatic EBV-associated nasopharyngeal carcinoma.
CMV and Glioblastoma

A Viral Link to Glioblastoma?

Circumstantial evidence hints that cytomegalovirus, a common herpesvirus, may play a role in the aggressive brain cancer, but big questions remain.
Consensus on the role of human cytomegalovirus in glioblastoma

The human cytomegalovirus (HCMV) and glioma symposium was convened on April 17, 2011 in Washington, DC, and was attended by oncologists and virologists involved in studying the relationship between HCMV and gliomas. The purpose of the meeting was to reach a consensus on the role of HCMV in the pathology of gliomas and to clarify directions for future research. First, the group summarized data that describe how HCMV biology overlaps with the key pathways of cancer. Then, on the basis of published data and ongoing research, a consensus was reached that there is sufficient evidence to conclude that HCMV sequences and viral gene expression exist in most, if not all, malignant gliomas, that HCMV could modulate the malignant phenotype in glioblastomas by interacting with key signaling pathways; and that HCMV could serve as a novel target for a variety of therapeutic strategies. In summary, existing evidence supports an oncomodulatory role for HCMV in malignant gliomas, but future studies need to focus on determining the role of HCMV as a glioma-initiating event.
CMV-specific T cell responses in Glioblastoma patients

Quantitative Analysis

Qualitative Analysis

Healthy Donors vs GBM Patients

p=0.0196

p=0.0186

Healthy Donors

GBM Patients

VTE (pp50) HLA A*0101

12.5%

QIK (IE-1) HLA B*0801

12.8%

TPR (pp65) HLA B*0702

8.04%

TRA (pp65) HLA Cw*0602

21.6%
In vitro stimulation with CMV antigen and γC cytokines restores T cell function.

**A**

Pre-stimulation

Post-stimulation

**B**

A diagram showing the percentage of CD8+ T cells with IFN-γ expression before and after stimulation.

**C**

Pie charts for GBM01, GBM02, GBM03, and GBM04 showing the distribution of cytokine combinations before and after stimulation.
CMV T cell therapy for recurrent GBM patients

Total number of patients recruited: 15
Number of patients who have completed: 9
T cell therapy
Number of patients undergoing T cell therapy: 1
Number of patients who were unable to complete T cell therapy: 2
Number of patients who were excluded due to progressive disease: 3