Dendritic Cell Based Immunotherapy for Cancer

Edgar G. Engleman, M.D.
Two main DC subsets

- **Myeloid (myDC)**
  - Derived from monocytes
  - Capture/process/present Ag to T cells
  - Activate NK cells and B cells

- **Plasmacytoid (pDC)**
  - Ontogeny unclear
  - Secrete IFN\(\alpha\) in response to viruses
myDC are extremely efficient APC

- Express mannose-R, which bind and deliver Ag intracellularly and rapidly recycle to cell surface
- Express high levels of MHC-I and –II, CD80 and CD86, CD40 and multiple adhesion molecules
- Secrete IL-6, IL-12, CCL-18
- Morphology enhances cell-cell interaction
DC are Present in Blood and Most Tissues

Blood DC

Skin Langerhans cells

Heart DC

Kidney DC

Murine Tracheal DC

Rat Tracheal DC

Human Bronchiole DC
Bone Marrow

Stem cell CD34+

1. DC progenitor production in the bone marrow

Epidermis

Langerhans cells

Dermal DC

2. Migration to peripheral tissue

Dermis

Skin

Lymph node

CD8

CD4

NK

B

Lymphatics

Antibodies

3. Sampling of the environment

4. Migration Ag processing

5. Induction of an immune response

PAMPs, dead or dying cells and other “danger signals”

DC: Center of the “Immuniverse”
Human DC Circa 1992

- Isolated from peripheral blood progenitors using density gradient separation

- Develop dendritic morphology with high expression of HLA-DR, CD80, CD86, CD40, CD54 following in vitro culture; blocked by anti-GM-CSF antibody

- Present Ag to naïve CD4$^+$ and CD8$^+$ T cells leading to the generation of Ag specific T cell lines, in vitro

Markowicz et al, J Clin Invest 85:955, 1990
Tumors and the Immune System

• Tumors present a unique challenge because they consist mainly of normal tissue components (“self”) and the immune system is trained to avoid attacking self

• The goal of immunotherapy is to overcome the reluctance of the immune system to attack the tumor (“active” immunotherapy), or produce tumor-specific antibodies or white blood cells outside of the body and inject them into patients (“passive” immunotherapy)
Using Vaccines to Direct the Immune System Against Cancer

• Old view: the inability of the immune system to recognize and attack tumors is inherent and irreversible

• New view: introducing tumor antigens and cellular activation stimuli directly to DC enables potent anti-tumor immunity
DC as Therapeutic Vaccines: Original Concept (circa 1994)

• **Goal**: Induce anti-tumor (or antiviral) immunity using autologous DC pulsed with tumor Ag

• **Methods**
  – Generate DC in vitro from circulating precursors
  – Load DC with Ag
  – Return DC to patients
Stanford University Clinical Trials with Ag Pulsed DC

- Non-Hodgkin’s Lymphoma
- Multiple Myeloma
- Prostate Cancer
- HIV Infection
- Colorectal Cancer
- Non-Small Cell Lung Cancer
- Metastatic Liver Cancer
Malignant Lymphoma Clinical Trial

- Patients with progressive low-grade non-Hodgkin’s B cell lymphoma

- Autologous tumor-derived immunoglobulin idiotype (Id) protein as Ag

- Patients’ DC obtained from blood, pulsed with Id + KLH and returned via i.v. infusion
Preparation and Administration of DC Vaccine
Lymphoma Trial Results

• 10 patients with measurable disease
  – 8 Id-specific T-cell proliferative responses
  – 2 complete clinical responses (progression free: 2 and 3 years)
  – 1 partial clinical response (progression free: 12 months)
  – 1 molecular response (progression free: 6 years)

• 25 patients in first remission: adjuvant therapy
  – 65% T cell or humoral anti-id response
  – 70% no tumor progression (median 2 years)
  – 30% (6 patients) tumor progression; 3 clinical responses (2 CR) to sc Id-KLH

Timmerman et al, Blood 99:1517, 2002
Factors Limiting DC-mediated Immunity

- DC number
- DC access to Ag
Flt3-Ligand (FL)

- Induces proliferation of hematopoietic progenitors (Lyman, Cell 1993)
- Preferentially expands DC in mice (Maraskovsky, J Exp Med 1995)
FL Expanded, Ag-pulsed DC for Colorectal Cancer

- 12 HLA-A*0201 patients with advanced colorectal cancer and rising carcinoembryonic antigen (CEA) blood levels
- Synthetic CEA derived peptide (9 amino acids) with single amino acid substitution
- Treatment with recombinant FL followed by DC isolation, CEA+ KLH loading and activation of DC, and return of the loaded DC
FL Administration Increases DC Yield
Identification of CEA-Specific CD8 T Cells with MHC/Peptide Tetramers
Correlation of Clinical Response with CEA Specific CD8⁺ T Cell Expansion

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<th>% Tetramer+ Post vaccine</th>
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Summary of Results

- Percentage of tetramer⁺ CD8⁺ cells correlated with clinical response (P=0.002)

- All 5 subjects who failed to develop CTL or CD8⁺ T cell expansion had rapidly progressive disease (P<0.001)

Fong et al., PNAS 98:8809-14, 2001
DC Vaccination for the Treatment of Prostate Cancer

Problems with Customized DC Vaccines

• Variable efficacy
• Best tumor Ags, DC activation method, route of delivery all unknown
• Cost and complexity high
• DC biology incompletely understood
Next Generation DC Therapy: Loading and Activating DC In Vivo

1. Combining systemic DC expansion with Ag loading and activation, in situ

2. Attracting DC to tumors with chemokine

3. Combining local tumor treatment with intratumoral DC and/or TLR agonist injection
Combining DC Expansion with Ag Loading and Activation, In Situ
Effect of FL+CpG+TRP2 on B16 Melanoma

Okano, F et al., J Immunol 174:2645-52, 2005
Attracting and Activating Intratumoral DC

CCL20 +/- CpG
CCL20 Chemokine Expression in Tumors Attracts DC
Intratumoral CCL20 Expression Induces Regression of CT26 but Not B16 Tumors
CCL20 + CpG Induces a Therapeutic Anti-tumor Response against B16 Melanoma

Furumoto, K et al., J Clin Invest 113:774–83, 2004
Summary of Results from Mouse Tumor Model Studies

• The lack of spontaneous anti-tumor immunity in tumor bearing hosts may be due to insufficient numbers or inadequate activation of pro-inflammatory DC at the tumor site

• These deficiencies can be overcome (in animal models), resulting in induction of systemic anti-tumor immunity
Conclusions: DC Immunotherapy

- Active immunotherapy, including DC vaccines, for the treatment of cancer has a bright future, but has not yet been proven to be clinically effective
- Suboptimal Ag, inadequate number and activation of tumoral DC and functional DC heterogeneity may explain the variable results from DC clinical trials
- Targeting and activating pro-inflammatory tumoral DC should be a high priority
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