Innate Core and Adaptive Shell

Cytokine-based immunotherapies and other molecular bispecifics, other immunotherapies

Michael T. Lotze, MD—Nurix Therapeutics, Chief Cellular Therapy Officer San Francisco and Pittsburgh

- TNF and IL-1/FGF/HMGB1 Family Members (Leaderless cytokines)
- The Interferons, IL-10 Family
- IL-2 Family Members
- IL-12 Family Members
- Pegylated or Muteinized Cytokines
- Anti-cytokines (TNF, IL-17, etc.)
- Cytokine-antibody conjugates
Disclosures-Company & Consultant

- Clinigen (IL-2)
- Nurix (NextACT) Chief Cellular Therapy Officer
- Checkmate, Inc.
- iRepertoire, Inc. (Hudson Alpha Institute)

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Finally, any statements I may make about Nurix, Nurix’s product candidates and Nurix’s business are not intended to contradict or modify Nurix’s existing public disclosures.
Cytokines (1728) and Bispecific (216) Clinical Trials for Cancer clinicaltrials.gov

https://www.fda.gov/media/123313/download Activating and non-activating (The minimum anticipated biological effect level-MABEL dose)
# Bispecific antibodies in cancer immunotherapy

Eva Dahlen, Niina Veitonmäki and Per Norlén

<table>
<thead>
<tr>
<th>Class</th>
<th>Description</th>
<th>Targets</th>
<th>Examples</th>
<th>Stage*</th>
</tr>
</thead>
<tbody>
<tr>
<td>T-cell redirectors</td>
<td>Redirects T cells to malignant cells by targeting a tumor antigen and CD3</td>
<td>CD19 × CD3</td>
<td>Binatumomab</td>
<td>Marketed</td>
</tr>
<tr>
<td></td>
<td></td>
<td>EpCAM × CD3</td>
<td>Catumaxomab</td>
<td>Marketed (withdrawn)</td>
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<tr>
<td></td>
<td></td>
<td>CD20 × CD3</td>
<td>XmAb13676, BTCT4465A, R07082859</td>
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<tr>
<td></td>
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<td>CD123 × CD3</td>
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<tr>
<td></td>
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<td>BCMA × CD3</td>
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<td>B7H3 × CD3</td>
<td>MG0009</td>
<td>I</td>
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<td></td>
<td>CEA × CD3</td>
<td>ROI198688, MT111</td>
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<tr>
<td></td>
<td></td>
<td>PSMA × CD3</td>
<td>Pasotuximab, Es414/MOR209</td>
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<tr>
<td>NK-cell redirectors</td>
<td>Redirects NK cells to malignant cells by targeting a tumor antigen and CD16A</td>
<td>CD30 × CD15A</td>
<td>AFM13</td>
<td>II</td>
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<tr>
<td></td>
<td></td>
<td>EGFR × CD16A</td>
<td>AFM24</td>
<td>PC</td>
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<td></td>
<td></td>
<td>BCMA × CD16A</td>
<td>AFM26</td>
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</tbody>
</table>

| Tumor-targeted immunomodulators | TA × CD40 | ABBV-428 | I     |
| Directs potent costimulation to the tumor-infiltrating immune cells by targeting a tumor antigen and costimulatory molecules such as CD40 or 4-1BB |
| HER2 × 4-1BB | PRS343 | I     |
| FAP × 4-1BB  | 4-1BB agonist | PC   |
| ST4 × 4-1BB  | ALG-APIV-527 | PC   |

| Dual immunomodulators | PD-L1 × TGF-β | M7824 | I     |
| Simultaneous targeting of two immunomodulating targets, resulting in blockade of inhibitory targets, depletion of suppressive cells or activation of effector cells |
| PD-1 × LAG-3 | MGD013 | I     |
| PD-1 × TIM-3 | MCLA-134 | PC   |
| PD-1 × CTLA-4 | XmAb20717 | PC   |
| CTLA-4 × 0X40 | ATOR-1015 | PC   |
Classes Of Bispecific Antibodies In Cancer Immunotherapy

Based on the types of biological targets and modes of action, bispecific immunotherapies can be divided into three main categories:

1. Cytotoxic effector cell redirectors, including
   a. T-cell redirectors
   b. NK-cell redirectors
2. Tumor-targeted immunomodulators
3. Dual immunomodulators.
## Cytotoxic Effector Cell Redirectors

<table>
<thead>
<tr>
<th>Class</th>
<th>Description</th>
<th>Targets</th>
<th>Examples</th>
<th>Stage*</th>
</tr>
</thead>
<tbody>
<tr>
<td>T-cell redirectors</td>
<td>Redirects T cells to malignant cells by targeting a tumor antigen and CD3</td>
<td>CD19 × CD3</td>
<td>Blinatumomab</td>
<td>Market</td>
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<td>EpCAM × CD3</td>
<td>Catumaxomab</td>
<td>Marketed</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td>(withdrawn)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Tumor-specific T-cell</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Non-tumor-specific T-cell</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

### Examples

- **B7H3 × CD3**: MGD009
- **CEA × CD3**: RO6958688, MT111
- **PSMA × CD3**: Pasotuximab, ES414/MOR209
Blinatumomab versus Chemotherapy for Advanced Acute Lymphoblastic Leukemia


Overall Survival Censored at Time of Stem-Cell Transplantation

<table>
<thead>
<tr>
<th></th>
<th>Median Overall Survival (mo)</th>
<th>95% CI</th>
<th>Hazard ratio</th>
<th>P-value</th>
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</thead>
<tbody>
<tr>
<td>Blinatumomab</td>
<td>6.9</td>
<td>5.3–8.8</td>
<td>0.66</td>
<td>0.004</td>
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<tr>
<td>Chemotherapy</td>
<td>3.9</td>
<td>2.8–4.9</td>
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</table>

Hazard ratio, 0.66 (95% CI, 0.50–0.88) P=0.004

No. at Risk

<table>
<thead>
<tr>
<th></th>
<th>Blinatumomab</th>
<th>Chemotherapy</th>
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<tbody>
<tr>
<td>No. at Risk</td>
<td>271</td>
<td>134</td>
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<tr>
<td>12 months</td>
<td>163</td>
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<td>18 months</td>
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<td>42 months</td>
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<td>54 months</td>
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<td>60 months</td>
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Cytotoxic Effector Cell Redirectors-NK

<table>
<thead>
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<th>Class</th>
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<tbody>
<tr>
<td>CD4⁺ T cells</td>
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<td>CD30 × CD16A</td>
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<tr>
<td>T_{H1}</td>
<td>T-bet</td>
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<td>T_{H2}</td>
<td>GATA-3</td>
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<tr>
<td>T_{H17} and T_{H22}</td>
<td>RORγt</td>
<td>IL-17, IL-22</td>
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<tr>
<td>Helper-like ILCs</td>
<td>IFN-γ</td>
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<td></td>
<td></td>
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<tr>
<td>ILC1</td>
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<td>ILC2</td>
<td>GATA-3</td>
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<tr>
<td>ILC3</td>
<td>RORγt<em>NKp46</em></td>
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<tr>
<td>B cells</td>
<td>CLP</td>
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<tr>
<td>Killer T cells</td>
<td>Eomes</td>
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<tr>
<td></td>
<td>IFN-γ</td>
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<td></td>
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<tr>
<td>Killer ILCs</td>
<td>Eomes</td>
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<td>NK</td>
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Immunomodulators

Other potential pathways

Class

Tumor-targeted immunomodulators

Dual immunomodulators

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<td>428</td>
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<td>3</td>
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<td>agonist</td>
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<td>13</td>
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<td>134</td>
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<td>10717</td>
<td>PC</td>
</tr>
<tr>
<td>1015</td>
<td>PC</td>
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</table>
Tumor Targeted Immunomodulators

(a) Dendritic cell
- CD40
- Dispecific antibody
- Tumor antigens
- Non-tumor-specific T-cell
- Tumor-specific T-cell
- TCR / CD3

(b) Tumor-specific T-cell
- CD188
- Tumor antigen
- MHC
- Tumor
Dual Tumor Targeted Immunomodulators
Macrophages

CD40 Agonists Alter Tumor Stroma and Show Efficacy Against Pancreatic Carcinoma in Mice and Humans
Gregory L. Beatty, et al.
Science 331, 1612 (2011);
DOI: 10.1126/science.1198443

Gregory L. Beatty,1,2,6 Elena G. Chiorean,3 Matthew P. Fishman,1 Babak Saboury,5 Ursina R. Teitelbaum,2,6 Weijie Sun,2,6 Richard D. Huhn,4 Wenru Song,6 Dongguang Li,4 Leslie L. Sharp,4 Drew A. Torigian,2,5 Peter J. O'Dwyer,2,6 Robert H. Vonderheide1,2,6*

1Abramson Family Cancer Research Institute, University of Pennsylvania School of Medicine, 421 Curie Boulevard, Philadelphia, PA 19104, USA. 2Abramson Cancer Center, University of Pennsylvania School of Medicine, Philadelphia, PA 19104, USA. 3Division of Hematology/Oncology, Department of Medicine, Indiana University School of Medicine, Indianapolis, IN 46202, USA. 4Pfizer Corporation, New London, CT 06320, USA. 5Department of Radiology, Department of Medicine, University of Pennsylvania School of Medicine, Philadelphia, PA 19104, USA. 6Division of Hematology-Oncology, Department of Medicine, University of Pennsylvania School of Medicine, Philadelphia, PA 19104, USA.

*To whom correspondence should be addressed. E-mail: rhv@exchange.upenn.edu
Fig. 1. Agonist CD40 mAb in combination with gemcitabine induces clinical responses in patients with surgically incurable PDA.

A

Percentage change from baseline

Patient number

10031022 10031004 10061006 10061001 10031020 10031018 10031012 10031001 10031008 10031019 10031002 10031005 10031007 10031006 10031003 10031010 10031017 10031002 10031016 10061003
General properties of cytokines
General properties of cytokines

Autocrine action

Paracrine action

Endocrine action

Nearby cell

Circulation

Distant cell
General Properties Of Cytokines

<table>
<thead>
<tr>
<th>Target Cell</th>
<th>Effect</th>
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<tbody>
<tr>
<td>B cell</td>
<td>Activation, Proliferation, Differentiation</td>
</tr>
<tr>
<td>Thymocyte</td>
<td>Proliferation</td>
</tr>
<tr>
<td>Mast cell</td>
<td>Proliferation</td>
</tr>
</tbody>
</table>

**PLEIOTROPY**
- Activated $T_H$ cells
- IL-4

**REDUNDANCY**
- Activated $T_H$ cells
- IL-2, IL-4, IL-5

**SYNERGY**
- Activated $T_H$ cells
- IL-4, IL-5

**ANTAGONISM**
- Activated $T_H$ cells
- IL-4, IL-5

Induces class switch to IgE
General Properties Of Cytokines
General properties of cytokines

**REDUNDANCY**

Activated TH cells → IL-2, IL-4, IL-5

**SYNERGY**

Activated TH cells → IL-4 + IL-5

Proliferation

B cell

Induces class switch to IgE

B cell
General Properties Of Cytokines

**Redundancy**

Activated TH cells

IL-2

IL-4

IL-5

B cell

**Synergy**

Activated TH cells

IL-4 + IL-5

B cell

**Induces class switch to IgE**

**Antagonism**

Activated TH cells

IL-4

IFN-γ

B cell

**Blocks class switch to IgE induced by IL-4**

**Proliferation**
### Role of Cytokines in Promoting Cancer (Dranoff Review)

<table>
<thead>
<tr>
<th>Cytokine</th>
<th>Cellular sources</th>
<th>Role in tumour formation</th>
</tr>
</thead>
<tbody>
<tr>
<td>IL-1</td>
<td>Macrophages, dendritic cells, B cells, natural killer cells, keratinocytes</td>
<td>Required for tumour invasion and angiogenesis</td>
</tr>
<tr>
<td>IL-6</td>
<td>Macrophages, T cells, B cells, endothelial cells, fibroblasts</td>
<td>Required for chemically induced lymphoma</td>
</tr>
<tr>
<td>IL-12</td>
<td>Macrophages, dendritic cells, neutrophils</td>
<td>Inhibits chemical carcinogenesis</td>
</tr>
<tr>
<td>IL-15</td>
<td>Macrophages, dendritic cells</td>
<td>Promotes natural killer T cell leukaemias</td>
</tr>
<tr>
<td>IFN-γ</td>
<td>Natural killer cells, natural killer T cells, T cells, B cells, macrophages, dendritic cells</td>
<td>Inhibits chemical carcinogenesis; inhibits lymphomas (especially with perforin); Stat1 and Rag2 inhibit carcinomas</td>
</tr>
<tr>
<td>M-CSF</td>
<td>Macrophages, endothelial cells, fibroblasts, bone-marrow stroma</td>
<td>Promotes breast cancer invasion</td>
</tr>
<tr>
<td>GM-CSF</td>
<td>Respiratory epithelial cells, T cells, natural killer cells, natural killer T cells, macrophages, eosinophils, endothelial cells, fibroblasts</td>
<td>Inhibits lymphomas and carcinomas (with IFN-γ and IL-3)</td>
</tr>
<tr>
<td>TNF-α</td>
<td>Macrophages, natural killer cells, B cells, T cells, neutrophils, fibroblasts, keratinocytes</td>
<td>Required for chemically-induced skin cancer</td>
</tr>
<tr>
<td>MIF</td>
<td>Macrophages, T cells, eosinophils, fibroblasts, keratinocytes, pituitary</td>
<td>Inhibits p53 tumour-suppressor functions</td>
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<tr>
<td>TGF-β</td>
<td>T cells, B cells, macrophages, platelets, bone-marrow stroma, eye, testis</td>
<td>Inhibits colon carcinomas (with Rag2)</td>
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<tr>
<td>Fas/Fas ligand</td>
<td>B cells, T cells, hepatocytes, colon, ovary, respiratory epithelial cells</td>
<td>Inhibits lymphomagenesis</td>
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Clinical Administration of Cytokines (Dranoff)

<table>
<thead>
<tr>
<th>Cytokine</th>
<th>Therapeutic actions</th>
<th>Clinical administration</th>
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<tbody>
<tr>
<td>IL-2</td>
<td>Enhances NK cell and CD8+ T-cell function; increases vascular permeability</td>
<td>Systemic, local</td>
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<tr>
<td>IL-3</td>
<td>Enhances tumour antigen presentation</td>
<td>Systemic</td>
</tr>
<tr>
<td>IL-4</td>
<td>Enhances eosinophil function and T-cell activation</td>
<td>Systemic, local</td>
</tr>
<tr>
<td>IL-6</td>
<td>Enhances T-cell and B-cell function; inhibition of IL-6 reduces lymphoproliferation</td>
<td>Systemic, local</td>
</tr>
<tr>
<td>IL-7</td>
<td>Enhances T-cell function</td>
<td>Local</td>
</tr>
<tr>
<td>IL-10</td>
<td>Inhibits tumour antigen presentation</td>
<td>Pending</td>
</tr>
<tr>
<td>IL-12</td>
<td>Enhances Th1 immunity and cytotoxicity; inhibits angiogenesis</td>
<td>Systemic, local</td>
</tr>
<tr>
<td>IL-13</td>
<td>Inhibits cytotoxicity against viral neoplasms</td>
<td>Pending</td>
</tr>
<tr>
<td>IL-15</td>
<td>Enhances cytotoxicity</td>
<td>Pending</td>
</tr>
<tr>
<td>IL-18</td>
<td>Enhances Th1 immunity and cytotoxicity; inhibits angiogenesis</td>
<td>Pending</td>
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<tr>
<td>M-CSF</td>
<td>Enhances macrophage function</td>
<td>Systemic</td>
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<tr>
<td>GM-CSF</td>
<td>Enhances tumour antigen presentation</td>
<td>Systemic, local</td>
</tr>
<tr>
<td>IFN-α</td>
<td>Enhances tumour antigen presentation and cytotoxicity</td>
<td>Systemic</td>
</tr>
<tr>
<td>IFN-γ</td>
<td>Enhances tumour antigen presentation and cytotoxicity</td>
<td>Systemic, local</td>
</tr>
<tr>
<td>TNF-α</td>
<td>Induces tumour-cell apoptosis; activates endothelium and granulocytes</td>
<td>Systemic</td>
</tr>
<tr>
<td>TRAIL</td>
<td>Induces tumour-cell apoptosis</td>
<td>Pending</td>
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<tr>
<td>FLT3 ligand</td>
<td>Stimulates dendritic-cell and NK-cell function</td>
<td>Systemic</td>
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<tr>
<td>Lymphotoxin</td>
<td>Enhances T-cell recruitment</td>
<td>Systemic</td>
</tr>
<tr>
<td>TGF-β</td>
<td>Inhibits T-cell effector function</td>
<td>Pending</td>
</tr>
</tbody>
</table>
Cytokines in the Treatment of Cancer

Kevin C. Conlon, Milos D. Miljkovic, and Thomas A. Waldmann

**Type I Interferon α + β**
- Plasmacytoid dendritic cells
- Fibroblasts, macrophages
- Endothelial cells

**Type II Interferon**
- IFNα1-12
- IFNγ

**GM-CSF**
- Monocytes, T cells
- Fibroblasts, macrophages
- Stromal cells

**IL-12**
- Monocytes
- DCs

**IL-2**
- T cells
- DCs and DCS

**IL-7**
- Stromal cells
- Epithelial cells

**IL-15**
- Monocytes
- CD4+ T cells
- NKT cells

**IL-21**
- Monocytes
- CD4+ T cells
- NKT cells

And IL-4 and IL-9 (mast cells)

**Target of Anti-cancer**
- Hairy Cell
- Kaposis Sarcoma
- Chronic Myelogenous Leukemia

**Fungoide**
- Melanoma
- Renal Cell
- Follicular leukemia
- Adult T-cell Leukemia

**Mycosis**
- Melanoma
- Renal Cell

**Hairy Cell**
- Kaposis Sarcoma
- Chronic Myelogenous Leukemia
- Melanoma
- Renal Cell

**Trials (Bold approved by FDA)**

**STAT4**
Cytokines are medically relevant endogenous small (~15kDa) proteins

Cytokine-based therapies in human disease

<table>
<thead>
<tr>
<th>Cytokine</th>
<th>Brand name</th>
<th>Status</th>
<th>Indication</th>
<th>Year of 1st FDA Approval</th>
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<tbody>
<tr>
<td>IL-2</td>
<td>Proleukin</td>
<td>Approved</td>
<td>Cancer</td>
<td>1992</td>
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<tr>
<td>IL-11</td>
<td>Neumega</td>
<td>Approved</td>
<td>Thrombocytopenia</td>
<td>1994</td>
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<td>EPO</td>
<td>Epogen</td>
<td>Approved</td>
<td>Anemia</td>
<td>1989</td>
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<tr>
<td>GCSF</td>
<td>Neupogen</td>
<td>Approved</td>
<td>Myelosuppression from chemo</td>
<td>1991</td>
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<tr>
<td>GM-CSF</td>
<td>Leukine</td>
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<td>IFN-α</td>
<td>Intron-A</td>
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<td>IFN-β</td>
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<td>Multiple sclerosis</td>
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<td>Granulomatosis</td>
<td>1990</td>
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<tr>
<td>IL-7</td>
<td>Clin dev</td>
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<td>Cancer, anti-viral</td>
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<tr>
<td>IL-10</td>
<td>Clin Dev</td>
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<td>Cancer, anti-inflammatory</td>
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<td>IL-12</td>
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<td>IL-15</td>
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<tr>
<td>IL-21</td>
<td>Clin dev</td>
<td></td>
<td>Cancer</td>
<td></td>
</tr>
</tbody>
</table>
IL-23 promotes tumour incidence and growth

John L. Langowski¹*, Xueqing Zhang¹*, Lingling Wu¹, Jeanine D. Mattson¹, Taiying Chen¹, Kathy Smith¹, Beth Basham¹, Terrill McClanahan¹, Robert A. Kastelein¹ & Martin Oft¹

Figure 1 | Overexpression of IL-23 but not of IL-12 in human cancer.
The Beginning of Molecular Therapeutics - 1978

Will Interferon Kill Cancer? Finnish Dr. Kari Cantell Is Helping the World Find Out

But Cantell and the Finnish Red Cross, now producing 250 billion units (5,285 quarts) a year, have provided the great bulk of pure interferon used for clinical studies on humans, including a $2 million batch bought last year by the American Cancer Society. “Production is the bottleneck,” says Cantell, who finds it “stupid and irritating” that until recently nobody else has tried to produce the substance in large-scale volume.
E1684, E1690, and E1694: Durable and significant Impact upon relapse-free * and overall survival **

Meta-analysis of all trials of IFN confirm RFS and OS impact

Interferon Alpha

• IFNa and Peginterferon alpha 2b are approved as adjuvant treatment for patients with completely resected stage III or IV high-risk melanoma

• Adjuvant in Melanoma (Kirkwood/ECOG); On February 15, 2019, the Food and Drug Administration approved pembrolizumab (KEYTRUDA, Merck) for the adjuvant treatment of patients with melanoma with involvement of lymph node(s) following complete resection.

• First-line treatment for patients with mRCC (alfa-2a, and alfa-2b in combination with bevacizumab),

• AIDS-related Kaposi’s sarcoma (alfa-2b), follicular lymphoma (alfa-2b), HCL (alfa-2a, alfa-2b), chronic myelogenous leukemia (Philadelphia chromosome-positive alfa-2a),

• Condyloma acuminata (alfa-2b), and cervical intraperitoneal neoplasms (alfa-2b) (Gutterman and others 1980; Kirkwood and Ernstoff 1984; Windbichler and others 2000).

• Pegylated for Hepatitis

• Hairy cell leukemia (BRAF mutant; purine analogues)
Interferon-gamma (IFN-γ)

- Only type II IFN
- Produced mainly by NK cells and T lymphocytes
- Works primarily as an immunomodulator
  - 100-10K x more active than Type I interferons
- Functions
  - Regulate MHC expression
  - Activates differentiation and function of phagocytes
  - Augments interactions between macrophages and T-cells
  - Key role in regulating T-cell subsets to determine the type of immune effector function during a specific immune response

SOUNDS LIKE A GREAT ANTI-CANCER DRUG!!!! FAILED!!!!!
Cytokines IFNα, IL-1, IL-2, IL-4, IL-7, IL-10, IL-12, IL-15, IL-18, IL-21, IL-24….IL-41

• IL-2 Discovered in 1976 and described as a protein that stimulated growth of T cells¹
• Jurkat IL-2 in 1983 [Lotze]
• Recombinant IL-2 first cloned in 1983¹
• First phase I studies of rIL-2 in malignant disease in 1984²
• Phase II clinical trials began in 1985³

High Dose IL-2 Immunotherapy

- Approved in patients with melanoma and kidney cancer.
- Significant ‘toxicity’.
- Associated with ‘cytokine storm’.
- iNOS blockers, sTNF-R or IL-1Ra have yielded limited reduction in side effects.
- IL-2 treatment is associated with a ‘systemic autophagic syndrome’ and temporally limited tissue dysfunction.

Ultra-low dose IL-2 expands regulatory T cells and NK cells in healthy donors

Increased $T_{\text{reg}}$ suppression

Enhanced IFN$\gamma$ production in NK

ULD IL-2 for GVHD prophylaxis: rationale

- **Ultra low dose IL-2**
  - Expands \( T_{\text{regs}} \) and NK cells
  - Effective for steroid refractory GVHD
  - Has been used for GVHD prophylaxis in matched donor SCT

- Quality and quantity of *regulatory* \( T_{\text{reg}} \) and NK cells impact haplo-SCT outcomes

The Hallmark of IL-2 Therapy

CR: 6% (17 pts)
OR: 16% (33 pts)
PR: 10% (26 pts)
Renal Cancer Response Rate = 25% (n=118)
May 27, 2010 — Two white-coated cancer researchers are among the luminaries picked for *TIME* magazine's 2010 list of the 100 most influential people in the world. Larry Kwak, MD, PhD, and Doug Schwartzentruber, MD, FACS, join Sarah Palin, James Cameron, Steve Jobs, & Lady Gaga on this year's "influentials" list.

Dr. Doug Schwartzentruber

Melanoma gp100 2092M +IL-2

BiovaxID patient-specific vaccine for follicular lymphoma

Dr. Larry Kwak
gp100 Peptide Vaccine and Interleukin-2 in Patients with Advanced Melanoma

Douglas I. Schwartzentruber, M.D., David H. Lawson, M.D.

A Progression-free Survival

B Overall Survival
Serum Vascular Endothelial Growth Factor and Fibronectin Predict Clinical Response to High-Dose Interleukin-2 Therapy

Marianna Sebastian, Sunghee Kim-Waide, Monica G. Pusell, David Strumak, Ena Wang, Pest Tabor, Dan Wee Koo, Gal Daffadieh, Zechar Paz, Francesc M. Martinac, and Howard L. Kaufman
Anti-GD2 Antibody with GM-CSF, Interleukin-2, and Isotretinoin for Neuroblastoma

Alice L. Yu, M.D., Ph.D., Andrew L. Gilman, M.D., M. Fevzi Ozkaynak, M.D., Wendy B. London, Ph.D., Susan G. Kreissman, M.D., Helen X. Chen, M.D., Malcolm Smith, M.D., Ph.D., Barry Anderson, M.D., Judith G. Villablanca, M.D., Katherine K. Matthay, M.D., Hiro Shimada, M.D., Stephan A. Grupp, M.D., Ph.D., Robert Seeger, M.D., C. Patrick Reynolds, M.D., Ph.D., Allen Buxton, M.S., Ralph A. Reisfeld, Ph.D., Steven D. Gillies, Ph.D., Susan L. Cohn, M.D., John M. Maris, M.D., and Paul M. Sondel, M.D., Ph.D., for the Children’s Oncology Group
# The Strange Immunobiology of RCC

<table>
<thead>
<tr>
<th>RESPONSE</th>
<th>INTERFERON α</th>
<th>IL-2</th>
<th>CTLA4 AB</th>
<th>PD-1 AB</th>
<th>TIL</th>
</tr>
</thead>
<tbody>
<tr>
<td>MELANOMA</td>
<td>+</td>
<td>+++</td>
<td>++</td>
<td>++</td>
<td>++++</td>
</tr>
<tr>
<td>RCC</td>
<td>+</td>
<td>+++</td>
<td>++</td>
<td>++</td>
<td>-</td>
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</tbody>
</table>

D50 in 29 Advanced Renal Cancer Patients Treated with High Dose Interleukin 2 (IL-2) and Hydroxychloroquine Associated with Clinical Response

3CR, 3PR
NKTR-214 (CD-122-biased agonist) plus nivolumab in patients with advanced solid tumors: Preliminary phase 1/2 results of PIVOT

ClinicalTrials.gov Identifier: NCT02983045

Adi Diab1, Michael Hunwitz2, Daniel Cho3, Vasilis Papadimitriou1, Brendan Curti2, Scott Tykodi2, Igor Puzanov2, Nuhad K. Ibrahim1, Sara M. Tolaney1, Debby Tripathy1, Jianjun Gao1, Arlene O. Steifker-Redtke1, Wendy Clemens3, Mary Tagliafierri4, Scott N. Gettinger2, Harriet Kluger2, James M. G. Larkin5, Giovanni Grignani6, Mario Szmol7, Nizar Tannir1

1The University of Texas MD Anderson Cancer Center, Houston, TX, USA; 2Yale School of Medicine, New Haven, CT, USA; 3NYU Medical Oncology Associates, New York, NY, USA; 4Providence Cancer Center and Earle A. Chiles Research Institute, Portland, OR, USA; 5University of Washington and Fred Hutchinson Cancer Research Center, Seattle, WA, USA; 6Dana Farber Cancer Institute, Boston, MA, USA; 7Nektar Therapeutics, San Francisco, CA, USA; 8Royal Marsden NHS Foundation Trust London, United Kingdom; 9Candiolo Cancer Institute, Turin, Italy, Europe.
NKTR-214 Background: Harnessing the IL-2 Pathway to Increase TILs

- NKTR-214 prodrug design with sustained signaling
- Mitigation of rapid immune stimulation to achieve safe, outpatient regimen administered every 3 week IV dosing
- Biased signaling preferentially activates and expands effector T cells and NK cells over Tregs in the tumor microenvironment
- NKTR-214 increases proliferation of TILs and PD-1 expression on the surface of CD8+ T cells providing a mechanistic rationale for combining with nivolumab
Stage IV IO-Naïve 1L Melanoma Dose Escalation Cohort (N=11)
Deepening of Responses Over Time

Best Overall Response by RECIST: ORR=7/11 (64%); DCR=10/11 (91%)

SITC 2017 (Data Cut: Nov 2, 2017)  
ASCO 2018 (Data Cut: May 29, 2018)

PD-L1 Negative (<1%)  
PD-L1 Positive (≥1%)  
Treatment Ongoing  
Off Study Treatment (maximal clinical benefit achieved)

ORR PD-L1 (-) 3/5 (60%)  
ORR PD-L1 (+) 4/6 (67%)

Horizontal dotted lines indicate the thresholds for PD, PR and CR response according to RECIST (version 1.1) criteria. CR: Complete response, all target and non-target lesions cleared. # Best Overall Response is SD (PR for target lesions, PD per new lesion on confirmatory scan). ++ Best overall response is CR to (CR for target lesions, non-target lesions still present). + 100% is PR (CR for target lesions, non-target lesions still present). *: Unconfirmed.

Presented By Adi Diab at 2018 ASCO Annual Meeting
IOV-3001 is an engineered IL-2 CDR graft which targets IL2R beta-gamma-expressing cells and limits IL2R alpha-beta-gamma-dependent Treg activation. The protein has an improved half-life leading to a better exposure while minimizing Cmax possibly reducing the side effects associated with IL-2 protein. Iovance will focus on GMP manufacturing of IOV-3001 during 2020 and may initiate IND-enabling activities as early as 2021.
Interleukin 7 (IL-7)

- Required for T cell development (lymphoid precursor to memory T cell) and peripheral T cell homeostasis.
- Enhances T cell reconstitution in HSCT recipients in mouse models (increased thymopoiesis and homeostatic proliferation of transferred and de novo generated mature T cells, and decreased peripheral T-cell apoptosis).
- Dose-dependent expansion of CD4+ and CD8+ T cells in initial clinical trials (patients with solid tumors or HIV infection).

Administration of interleukin-7 after allogeneic bone marrow transplantation improves immune reconstitution without aggravating graft-versus-host disease

Onder Alpdogan, Cornelius Schmaltz, Stephanie J. Muriglan, Barry J. Kappel, Miguel-Angel Perales, Jimmy A. Rotolo, Jens A. Halm, Benjamin E. Rich, and Marcel R. M. van den Brink

Alpdogan et al, Blood 2001;98:2256-226

IL-7 enhances peripheral T cell reconstitution after allogeneic hematopoietic stem cell transplantation

Önder Alpdogan, Stephanie J. Muriglan, Jeffrey M. Eng, Lucy M. Willis, Andrew S. Greenberg, Barry J. Kappel, and Marcel R.M. van den Brink

Department of Medicine, Memorial Sloan-Kettering Cancer Center, New York, New York, USA

Phase I evaluation of intravenous recombinant human interleukin 12 in patients with advanced malignancies.

The T1/2 elimination of rhIL-12 was calculated to be 5.3-9.6 h. Biological effects included dose-dependent increases in circulating IFN-gamma, which exhibited attenuation with subsequent cycles. Serum neopterin rose in a reproducible fashion regardless of dose or cycle. There was one partial response (renal cell cancer) and one transient complete response (melanoma), both in previously untreated patients.
Durable responses and immune activation with intratumoral electroporation of pIL-12 plus pembrolizumab in actively progressing anti-PD-1 refractory advanced melanoma: KEYNOTE 695 interim data. SITC 2020

Figure 2. Best Overall Response

In patients with M1c/M1d disease, the ORR was 35.3% (n=6/17)

In patients with prior exposure to ipilimumab, the ORR was 40% (n=6/15)
## Interleukin 15 Adverse Event Summary

<table>
<thead>
<tr>
<th>Dose Level (mcg/kg)</th>
<th>Chills Rigors</th>
<th>Fever</th>
<th>Capillary Leak</th>
<th>Nausea</th>
<th>Vomiting</th>
<th>Hypotension</th>
<th>Edema</th>
<th>Hypoalbuminemia</th>
<th>Aspartate transaminase</th>
<th>Alanine transaminase</th>
<th>Lymphopenia</th>
<th>Leukopenia</th>
<th>Thrombocytopenia</th>
<th>Neutropenia</th>
<th>Anemia</th>
</tr>
</thead>
<tbody>
<tr>
<td>Percentage of Patients with Adverse Event</td>
<td>0.3</td>
<td>0.3</td>
<td>0.3</td>
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</tbody>
</table>
Cytokine Release and Adverse Events 3 mcg patients

**Mean Arterial Blood Pressure**

- mm Hg
- Time in Minutes After Treatment
- Days: Day 1 to Day 12

**Temperature**

- Degrees Centigrade
- Rigors
- Time in Minutes After Treatment

**Cytokine Level in Mean Fold Increase Above Baseline**

- Fold Increase Above Baseline
- Cytokines: IFNγ, IL-6, IL-10, IL-8, IL-1β, TNFa