Manipulation of the Immune Response
- Immunomodulation -

Immune system is made of components, cells and organs that act together to defend the host from microbes.
Aim of immunomodulation

Hyperimmune response

E.g.
- Autoimmune diseases
- Allergies
- Transplant rejection

Hypoimmune response

E.g.
- Infections
- Carcinomas

Regulation

Immunity

tolerance
response
Example: Targets of immune intervention in type 1 diabetes
Immunomodulation methods

IMMUNOSUPPRESSION:
• Immunosuppressive drugs
• Monoclonal antibodies
• Antitumor therapy
• Gene manipulation (CRISPR-Cas9, siRNA)

IMMUNOSTIMULATION:
• BCG
• Interferons
• Talidomide, levamisol
• IL-2

- IMMUNISATION
  • Vaccination
  • Immunoglobulins
Immunosuppression

When?
- Autoimmune diseases
- Organ transplantation
- Allergies

Problems:
• Lifetime usage of drugs
• Infections, tumors
• Nephrotoxicity
• Diabetogens
Immunosuppressive drugs

- Anti-inflammatory (NSAIDs, corticosteroids)
- Cytotoxic (azathioprine, cyclophosphamide)
- Noncytotoxic (cyclosporin A, tacrolimus, rapamycin)
Immunosuppressive drugs: Nonsteroidal anti-inflammatory drugs - NSAIDs

Phospholipids

\[ \text{Phospholipase A}_2 \]

\[ \text{Arachidonic acid} \]

- Cyclooxygenase (COX)-1 or -2
- Lipoxigenase

\[ \text{NSAIDs} \]

\[ \uparrow \text{inflammation} \]

Prostaglandins

Thromboxans

Leukotriens
**Immunosuppressive drugs: steroids**

- **Corticosteroids** – anti-inflammatory drugs
  - **Prednisone** (synthetic cortisol analog)
  - Used in transplantations, autoimmune diseases, allergies
  - Activated steroid receptors act as transcription factors

<table>
<thead>
<tr>
<th>Effect on</th>
<th>Physiological effects</th>
</tr>
</thead>
<tbody>
<tr>
<td>↓ IL-1, TNF-α, GM-CSF</td>
<td>↓ Inflammation caused by cytokines</td>
</tr>
<tr>
<td>↓ IL-3, IL-4, IL-5, CXCL8</td>
<td></td>
</tr>
<tr>
<td>↓ NOS</td>
<td>↓ NO</td>
</tr>
<tr>
<td>↓ Phospholipase A₂</td>
<td>↓ Prostaglandins</td>
</tr>
<tr>
<td>↓ Cyclooxygenase type 2</td>
<td>↓ Leukotrienes</td>
</tr>
<tr>
<td>↑ Annexin-1</td>
<td></td>
</tr>
<tr>
<td>↓ Adhesion molecules</td>
<td>Reduced emigration of leukocytes from vessels</td>
</tr>
<tr>
<td>↑ Endonucleases</td>
<td>Induction of apoptosis in lymphocytes and eosinophils</td>
</tr>
</tbody>
</table>

Figure 16.3 Janeway's Immunobiology, 9th ed. (© Garland Science 2017)
Immunosuppressive drugs: steroids

- Lymphocyte cytokines (IL-2, IL-3, IL-6, TNF-α, GM-CSF, IFN-γ)
- Endothelial cell adhesion molecules (ICAM-1, E-selection), cytokines (IL-1, GM-CSF), arachidonic acid metabolites
- Monocyte cytokines (IL-1, TNF-α), prostaglandins/leukotrienes synthesis and release, Fc and C3 receptors
- Basophils histamine and leukotriene release
Corticosteroids – physiology

Corticotropin-releasing hormone (CRH)

Adrenocorticotropic hormone (ACTH)

cortisol

STRESS

+ + +

- - -
Corticosteroids – physiology

- Possible multiple side effects
- Used in combination with other drugs to reduce toxicity

from P. Stewart, Williams Textbook of Endocrinology, 2003
Immunosuppressive cytostatics

- Azathioprine (AZA), cyclophosphamide
  - Interfere with DNA synthesis (dividing cells)
  - Primarily planned to be used for anti-tumor therapy
  - Used in low dosage for autoimmune diseases (combination with corticosteroids)
  - Used in high dosage only before bone marrow transplantation to eliminate all lymphocytes
  - Cyclophosphamide (more toxic) – developed as chemical weapon
Immunosuppressive cytostatics

• **Cyclosporin A, tacrolimus (FK506)**
  – Less toxic
  – Bacterial/fungal origin
  – Interfere with clonal expansion of activated lymphocytes
  – Used in transplanted patients
  – Block calcineurin (cyclosporin A & tacrolimus)

• **Rapamycin (sirolimus)**
  – inhibits lymphocyte proliferation and increases the number of Treg
Cyclosporin A and tacrolimus

With calcium bound to it, calmodulin can activate the enzyme calcineurin to dephosphorylate NFAT, which can then enter the nucleus to stimulate IL-2 transcription.

The binding of cyclosporin A to an immunophilin creates a complex that inhibits calcineurin activation by calmodulin, thus preventing the dephosphorylation of NFAT.

Figure 16.5 Janeway's Immunobiology, 9th ed. © Garland Science 2017
# Cytotoxic immunosuppressive drugs

<table>
<thead>
<tr>
<th>Cell type</th>
<th>Effects</th>
</tr>
</thead>
</table>
| **T lymphocyte** | Reduced expression of IL-2, IL-3, IL-4, GM-CSF, TNF-α  
Reduced proliferation following decreased IL-2 production  
Reduced Ca²⁺-dependent exocytosis of granule-associated serine esterases  
Inhibition of antigen-driven apoptosis |
| **B lymphocyte** | Inhibition of proliferation secondary to reduced cytokine production by T lymphocytes  
Inhibition of proliferation following ligation of surface immunoglobulin  
Induction of apoptosis following B-cell activation |
| **Granulocyte** | Reduced Ca²⁺-dependent exocytosis of granule-associated serine esterases |

Figure 16.4 Janeway’s Immunobiology, 9th ed. (© Garland Science 2017)
Antibodies in therapy

- Monoclonal antibody therapy: transplantations, autoimmune diseases, tumors
  - **Cytotoxic** (antibody-mediated cytotoxicity)
  - **Neutralizing** (block the function of target molecule)
  - Usually produced in mice – problems?!?!!?

Examples:
- anti-CD3
- anti-TNF-a
- anti-IL-2
- anti-LFA1, ...

100% mouse  30% mouse  10% mouse  100% human

Figure 16.7 Janeway's Immunobiology, 9th ed. (© Garland Science 2017)
<table>
<thead>
<tr>
<th>Monoclonal antibodies developed for immunotherapy</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Generic name</strong></td>
</tr>
<tr>
<td>-------------------</td>
</tr>
<tr>
<td>Rituximab</td>
</tr>
<tr>
<td>Alemtuzumab (Campath-1H)</td>
</tr>
<tr>
<td>Muromonab (OKT3)</td>
</tr>
<tr>
<td>Daclizumab</td>
</tr>
<tr>
<td>Basiliximab</td>
</tr>
<tr>
<td>Infliximab</td>
</tr>
<tr>
<td>Certolizumab</td>
</tr>
<tr>
<td>Adalimumab</td>
</tr>
<tr>
<td>Golimumab</td>
</tr>
<tr>
<td>Tocilizumab</td>
</tr>
<tr>
<td>Canakinumab</td>
</tr>
<tr>
<td>Denosumab</td>
</tr>
<tr>
<td>Ustekinumab</td>
</tr>
<tr>
<td>Efalizumab</td>
</tr>
<tr>
<td>Natalizumab</td>
</tr>
<tr>
<td>Omalizumab</td>
</tr>
<tr>
<td>Belimumab</td>
</tr>
<tr>
<td>Ipilimumab</td>
</tr>
<tr>
<td>Raxibacumab</td>
</tr>
</tbody>
</table>

Figure 16.8 Janeway’s Immunobiology, 9th ed. (© Garland Science 2017)
Anti-CD4 Ab & graft tolerance

Figure 14-6 Immunobiology, 6/e. (© Garland Science 2005)
Anti-TNFα Ab in autoimmune diseases

- Works well for: Rheumatoid arthritis, Crohn’s disease, ankylosing spondylitis
- Does not work for: multiple sclerosis

Figure 16.9 Janeway’s Immunobiology, 9th ed. (© Garland Science 2017)
Anti-integrin Ab im MS

Figure 16.10 (part 1 of 2) Janeway's Immunobiology, 9th ed. (© Garland Science 2017)
Immunosuppression

DOI: 10.1056/NEJMra033540
Immunostimulation

When?
- Tumors
- Prevention of pathogenic infections

• Specific immunostimulants
  – antibodies or antigens
  – Vaccines

• Non-specific immunostimulants
  – adjuvants
  – non-specific immunostimulators

Problems:
  – Unknown effects (novel methods and approaches)
  – Autoimmunity?
Immune responses and tumors

- Immunize mouse with irradiated tumor cells
  - Irradiated tumor cells

  - Inject viable cells of the same tumor
    - Response to unique tumor rejection antigens eliminates tumor
  - Inject viable cells of a different tumor
    - Response to irradiated tumor does not eliminate unrelated tumors of a different cell type

Figure 16.12 Janeway's Immunobiology, 9th ed. (© Garland Science 2017)
### Potential tumor rejection antigens have a variety of origins

<table>
<thead>
<tr>
<th>Class of antigen</th>
<th>Antigen</th>
<th>Nature of antigen</th>
<th>Tumor type</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tumor-specific</td>
<td>Cyclin-dependent kinase 4</td>
<td>Cell-cycle regulator</td>
<td>Melanoma</td>
</tr>
<tr>
<td>mutated oncogene</td>
<td>β-Catenin</td>
<td>Relay in signal transduction pathway</td>
<td>Melanoma</td>
</tr>
<tr>
<td>or tumor</td>
<td>Caspase 8</td>
<td>Regulator of apoptosis</td>
<td>Squamous cell</td>
</tr>
<tr>
<td>suppressor gene</td>
<td>Surface Ig/idiotype</td>
<td>Specific antibody after gene rearrangements in B-cell clone</td>
<td>Lymphoma</td>
</tr>
<tr>
<td>Cancer-testis</td>
<td>MAGE-1</td>
<td>Normal testicular proteins</td>
<td>Melanoma</td>
</tr>
<tr>
<td>antigens</td>
<td>MAGE-3</td>
<td></td>
<td>Breast</td>
</tr>
<tr>
<td></td>
<td>NY-ESO-1</td>
<td></td>
<td>Glioma</td>
</tr>
<tr>
<td>Differentiation</td>
<td>Tyrosinase</td>
<td>Enzyme in pathway of melanin synthesis</td>
<td>Melanoma</td>
</tr>
<tr>
<td>Abnormal</td>
<td>HER-2/neu</td>
<td>Receptor tyrosine kinase</td>
<td>Breast</td>
</tr>
<tr>
<td>gene expression</td>
<td>WT1</td>
<td>Transcription factor</td>
<td>Ovary</td>
</tr>
<tr>
<td>Abnormal</td>
<td>MUC-1</td>
<td>Underglycosylated mucin</td>
<td>Breast</td>
</tr>
<tr>
<td>post-translational</td>
<td>NA17</td>
<td>Retention of introns in the mRNA</td>
<td>Melanoma</td>
</tr>
<tr>
<td>modification</td>
<td>HPV type 16, E6 and E7</td>
<td>Viral transforming gene products</td>
<td>Cervical carcinoma</td>
</tr>
<tr>
<td>modification</td>
<td>proteins</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Figure 16.17 Janeway's Immunobiology, 9th ed. (© Garland Science 2017)
Malignant cells are monitored by immune system

When tumors arise in a tissue, a number of immune cells can recognize and eliminate them.

Variant tumor cells arise that are more resistant to being killed.

Over time a variety of different tumor variants develop.

Eventually, one variant may escape the killing mechanism, or recruit regulatory cells to protect it, and so spread unchallenged.

Figure 16.13 Janeway's Immunobiology, 9th ed. (© Garland Science 2017)
Tumors can avoid immune recognition

### Mechanisms by which tumors avoid immune recognition

<table>
<thead>
<tr>
<th>Low immunogenicity</th>
<th>Tumor treated as self antigen</th>
<th>Antigenic modulation</th>
<th>Tumor-induced immune suppression</th>
<th>Tumor-induced privileged site</th>
</tr>
</thead>
</table>
| No peptide: MHC ligand  
No adhesion molecules  
No co-stimulatory molecules | Tumor antigens taken up and presented by APCs in absence of co-stimulation  
tolerize T cells | T cells may eliminate tumors expressing immunogenic antigens, but not tumors that have lost such antigens | Factors (e.g., TGF-β, IL-10, IDO) secreted by tumor cells inhibit T cells directly. Expression of PD-L1 by tumors | Factors secreted by tumor cells create a physical barrier to the immune system |

Figure 16.14 Janeway's Immunobiology, 9th ed. (© Garland Science 2017)
mAbs and tumors

- **Tumor-specific antibody:**
  - Antibodies bind to the tumor cell
  - NK cells with Fc receptors (CD16) are activated to kill the tumor cells

- **Tumor-specific antibody (or antibody fragment) conjugated to toxin:**
  - Antibody–toxin conjugates bind to the tumor cell
  - Conjugates are internalized, killing the cell

- **Tumor-specific antibody (or antibody fragment) conjugated to radionuclide or chemotherapeutic drug:**
  - Radioactive antibody binds to the tumor cell
  - Radiation kills the tumor cell and neighboring tumor cells

Figure 16.19 Janeway's Immunobiology, 9th ed. (© Garland Science 2017)
Nonspecific immunostimulative drugs

- **Levamisole**: antihelmintic, leukocyte activator (colon cancer)
- **Thalidomide**: increases cytokine production, activates NK cells (multiple myeloma)
- **BCG**: live microbes (bladder cancer)
- **Interferons**: antiviral effects (hepatitis, melanoma, Kaposi sarcoma)
- **IL-2**: (kidney carcinoma, melanoma)