SLIDE 1 T Cell Effector Mechanisms. This week’s presentation will further describe the specific mechanisms employed by activated T cells to target microbes after antigen presentation and recognition. In addition, we will discuss the importance of the helper T cell class in the regulation of the immune response. Activated T cells produce many cytokines that serve to tip the scales of the immune system towards either a humoral immune response or a cell-mediated response.

SLIDE 2 Learning objectives for Week Twelve: T cell effector functions.

SLIDE 3 This diagram illustrates how both CD4+ and CD8+ T cells recognize antigens that are presented to them by professional antigen presenting cells. These native cells then will differentiate into effector cells that will also recognize antigen at the site of infection. In a cell-mediated immune response, T cells can recognize antigen in both their native state in the lymphoid organs, and in the effector state anywhere in the body to eliminate microbes.
**Slide 4**

*T Cell Homing*
- T cells exit nodes & are attracted to site of infection with adhesion molecules (homing).
- Activated T cells that recognize antigen are retained to battle the infection.
- Other T cells that are not antigen specific continue to circulate.

SLIDE 4 Adhesion molecules (such as integrins, selectins) and cytokines (produced as part of the innate immune response) at the site of infection act to “home” the activated T cells to the battle. See Figure 6-3 in the textbook (page 109) for more information on these homing receptors. T cells are then able to leave the blood & lymphatic vessels to migrate to the site of infection. This homing of effector T cells to the site of infection is not limited to only T cells that have recognized the antigen, but once there, if the T cell does recognize the antigen (microbe) it is preferentially selected to stay. This allows for a specific T cell response to the antigen to perform effector functions.

**Slide 5**

*Focus on CD8+ cells*
- CD8 cells primarily respond to intracellular pathogens (restricted to MHC I).
- Once activated, CD8 cells proliferate into antigen specific effector cells.
- Effector cells leave the peripheral lymphoid organs to migrate to the site of infection.
- Major effector function: recognize & kill infected host cells.
- CD8+ cells provide the major cellular response to viral infections.

SLIDE 5 CD8+ cells usually respond to intracellular pathogens, and are very important in controlling these infections. Once these cells are activated, they proliferate into antigen specific effector cells (remember, acquired immunity is specific!). These cells then leave the peripheral lymphoid organs on a mission to find & destroy infected cells.

**Slide 6**

*CD8+ T cells: Differentiation*
- First signal: recognize antigen peptide on surface of host cell displayed by MHC I.
- Second signal: need costimulators (B7 – CD28) to trigger activation.
- Differentiation into effector cells leads to specific targeting of any other cell infected with same microbe (ag specificity).

SLIDE 6 CD8+ T cells allow the immune system to specifically kill infected cells & eliminate reservoirs of infection throughout the body. As discussed last week, CD8+ T cells must have antigenic peptides presented to them by infected host cells that serve as the APC. It is especially important that CD8+ T cells are restricted and can only be activated if costimulators are present on the APC because they can directly kill other cells.
**Slide 7**

**CD8+ T cells: Deadly Consequences**
- CD8+ cells then release granules that kill the organism
- Perforin punches holes through the targeted cell membrane
- Granzymes then enter the cell, activate caspases which induce apoptosis
- Infected cell is killed, CD8 T cell also can produce IFNγ to recruit macrophages
- Apoptotic (dead) cells are quickly phagocytosed & removed

Watch an excellent animation on how effector Tc cells kill targets with perforin.

**Slide 8**

**Focus on CD4+ cells**
- CD4 cells primarily respond to extracellular antigens (restricted to MHC II)
- Once activated, CD4 cells differentiate into effector cells
- Primarily function to release cytokines that activate B cells & macrophages
- Two subsets of CD4 cells:
  - Th1
  - Th2
  **Also, Th0 (Treg cells – discussed in Block 5)**

CD4+ cells are especially susceptible to infection with HIV. The image on the top are healthy T cells, compared to those below that have been infected.

**Slide 9**

**A Delicate Balance**

**CD4+ Cells Polarize the Immune System**
- **Th1 Cells**
  - Cytokines: Ifnγ, IL-12, IL-2
  - IgG2a antibodies
  - Pro-inflammatory
  - Cell mediated Immunity
- **Th2 Cells**
  - Cytokines: IL-4, 9, 10, 13
  - IgG4 and IgE antibodies
  - Anti-inflammatory
  - Humoral Immunity

CD4+ cells polarize the immune system. Th1 cells and Th2 cells secrete different cytokines to elicit either a cell-mediated or humoral immune response. Th1 cytokines are pro-inflammatory, whereas Th2 cytokines are anti-inflammatory. These T cells work together to stimulate or control the immune response, and act upon each other. If a Type 1 response is under way, then Th2 cells are down-regulated. And the reverse is also true: a Type 2 response regulates Th1 cells, so that the immune response is anti-inflammatory. These cells work in a continual feedback loop during an immune response to control T cell activity.
SLIDE 10 Another illustration that shows how CD4+ helper T cells serve a dual purpose and impact both the cell-mediated immune response & the humoral immune response. For this reason, the helper cells have the ability to influence either a pro-inflammatory response by secreting cytokines that stimulate cell-mediated immunity, or by secreting other cytokines that promote anti-inflammatory responses and B cell activation.

SLIDE 11 Th1 helper cells stimulate cell-mediated immunity, primarily by releasing IFN V to activate macrophages. They also increase MHC expression on APCs to amplify the T cell response.

SLIDE 12 This diagram illustrates the tremendous influence Th1 cells have on other cell types to generate a large & multiple cell-mediated attack on an antigen. Watch the brief video to see how the smaller cytotoxic T cell can destroy a much larger cell that has been infected with influenza virus.
SLIDE 13 This slide shows the interaction a naive Tc cell and a Th1 cell. Notice the production of interleukin-2 by the Th1 cells and its effect on the Tc cell. Also, understand that T-cells must recognize antigen in conjunction with the MHC molecule.

SLIDE 14 Macrophage activation is an important component of the cell-mediated immune response. Macrophages also work on a 2 signal system to destroy microbes that they have “eaten”. These signals are not as specific as those required for T cell activation, but they must both bind the CD40 ligand on the Th1 cell & receive a cytokine stimulus (IFN gamma) for activation. Once activated, macrophages can generate an oxidative burst with nitric oxide & reactive oxygen intermediates to kill the ingested organism. Activated macrophages also further amplify a cell-mediated response by secreting cytokines to induce inflammation, activate more T cells, and express more costimulators and MHC molecules on their surface to present more antigen to T cells & further the cycle. For this reason, a cell-mediated immune response must be extremely specific and highly regulated because it is difficult to slow down once it has started & can lead to damage to other cells.
Th1 Cells & Leprosy

- M. leprae bacteria lives inside macrophages (immune evasion strategy) & causes leprosy
- Destructive (lepromatous form) can occur in individuals that do not mount a strong cell-mediated immune response
- Defect in Th1 cell activation prevents macrophages from becoming activated to destroy bacteria

Photos of active lepromatous leprosy cases, with significant physical disfigurement.

Leprosy is an ancient disease closely associated with biblical times that is chronic and can lead to severe physical disfigurement. In 2005, 500,000 new cases were diagnosed with millions already infected worldwide, despite being completely treatable. In fact, patients with disfiguring nodules can often clear the infection & decrease the size of the nodules with multi-drug therapy with antibiotics. Unfortunately, disfigurement that has caused severe physical damage by eating away at the bone and connective tissue cannot be reversed. Leprosy is caused by M. leprae, which infects the skin, and has a strong affinity for nerves. In "tuberculoid" leprosy, the less severe version of the disease, an active cell-mediated immune response controls the bacteria and few organisms are present. In contrast, "lepromatous" leprosy is the much more severe version of the disease because of an impaired cell-mediated immune response. This type of leprosy allows a high number of organisms to live in the body and suppress the immune response.

CD4: Type 2 Helper Cells (Th2)

- Th2 cells release interleukin 4 (IL-4), which stimulates B cell responses
- Th2 cells also activate eosinophils to defend against parasites via IgE antibodies
- Can also dampen the Th1 response to limit tissue damage (anti-inflammatory)
- Improves humoral immunity

SLIDE 16 Th2 helper cells stimulate humoral immunity, primarily by releasing interleukin 4, which activates B cells. The best humoral immune response (memory) occurs only with T cell help, so this subset of cells is extremely important to defend the body. The Th2 response can also dampen a Type 1 response to limit injury to the body (as cytotoxic T cells can kill self cells & result in tissue damage).
Slide 17

This diagram shows that CD4+ Helper (Th2) cells especially promote B cell growth and differentiation to stimulate antibody production. Production of IgG1, IgE, and IgA antibody classes are favored by Th2 cytokines. And, two signals are required for the B cell to become a plasma cell (this is the reason why T cells are especially important to the humoral immune response).

Slide 18

This slide shows the interactions between Th1 cells and Th2 cells. Notice the inhibitory effects of IL-10 and interferon gamma on each Th cell type.

Slide 19

The different roles of the CD4+ helper T cells. The Th1 subset produces different cytokines that promote cell-mediated immunity, primarily activation of macrophages and antibodies that improve opsonization and phagocytosis by macrophages. The Th2 subset secrete other cytokines (such as IL-4 & IL-5) that enhance antibody-mediated destruction of microbes. This subset promotes humoral immunity and these cytokines provide negative feedback to the Th1 subset to inhibit activation of macrophages & suppress Th1 cell-mediated responses.
SLIDE 20 An overview of the different methods by which the immune response can be modulated.

- Regulation by antigen
- Regulation by Antigen Presenting Cell (APC)
- Regulation by antibody
- Regulation by lymphocytes
- Regulation by neuroendocrine modulation
- Genetic control of immune response

SLIDE 21 The nature of an antigen as well as its amount and route of administration can affect the immune response. The interactions of the immune components with the antigen will also affect the eventual outcome. For example, if an antigen enters through the bloodstream (by a mosquito for instance), then it will travel through the circulatory system until it gets filtered by the spleen. The specific receptors on the T & B cells influence activation and differentiation of those cells as they can only recognize certain antigenic determinants.

SLIDE 22 Antigen-presenting cells can also regulate the immune response via the MHC proteins. Activated T cells can produce cytokines that stimulate antigen presenting cells into increasing the number of MHC proteins on the APC surface so that they can present more antigen to the T cells. This serves to amplify the cell-mediated immune response to target the organism.
SLIDE 23 Antibodies may block receptor sites for antigens and may inhibit or augment the formation of immune complexes. In addition, cross-linking between the Fc and antigen receptors may inhibit antibody formation.

SLIDE 24 As discussed previously, T-cells can have a strong modulatory effect on the immune system. Th1 and Th2 cells determine the nature of immune responses as to whether cell-mediated immunity or humoral immunity is the dominant response. Further, there can be cross-regulation of the Th1 and Th2 responses via cytokine release. In addition, CD4+ cells can prevent the induction of autoimmunity; and CD8+ cells can be immunosuppressive.

SLIDE 25 There can also be neuroendocrine modulation via direct sympathetic innervation of lymphoid tissues. In addition, lymphocytes have hormonal receptors for corticosteroids (etc.) which can produce immunosuppression during stress.
### Slide 26
**Genetic Control of Immune Responses**

- Inherited ability to make immune responses to given Ag
- Influence of MHC haplotypes
- Influence of non-MHC linked genes
- T cells recognize Ag only in MHC context
- Thymus selection for self-recognition
- MHC linked (inflammatory bowel disease, psoriasis, diabetes, etc) and autoimmune disease
- Non-MHC linked genes and susceptibility to infection

**SLIDE 26** Genetic variability can also affect immune responses. Immune responses to certain antigens can be inherited (for example, allergies). Both MHC-linked and non-MHC linked genes can also affect the immune response and the likelihood of autoimmunity development and disease susceptibility. In addition, thymus selection for self-recognition can also affect the immune response.

### Slide 27
**Cell-Mediated Immunopathology**

- Cell-Mediated Immune Response causes tissue damage
- Cytotoxicity: essential cells are killed with a resultant deficit, autoimmunity
- Chronic inflammation: autoantigens, cross-reactive Ags, lysosomal enzyme release
- Space-Occupying lesion: granulomas, impaired functions of organs
- Excessive cytokine release: Toxic Shock Syndrome, Schwartzman Reaction, Hemorrhagic Necrosis (TNF)
- Type 4 Delayed Hypersensitivity

**SLIDE 27** Cell-mediated immunity is capable of producing tissue damage. Examples include cytotoxicity, chronic inflammation, granulomas, excessive cytokine release, and type 4 delayed hypersensitivity. In many instances, these effects are overreactions of a normally controlled process.

### Slide 28
**SLIDE 28** This slide shows the importance of the MHC molecule in the T-cytotoxic cell recognition of cells. If the receptor on the virally infected cell has been altered, the T-cytotoxic cell will not recognize the infected cell and therefore not lyse the cell. Viruses that down-regulate the amount of MHC molecules on the cell surface are then killed by Natural Killer cells. NK cells also recognize altered MHC on tumor cells & can also kill them.
SLIDE 29 This slide shows one example of cell-mediated pathology: chronic inflammation in a joint. Notice the immune cells and immune complexes in the joint.

SLIDE 30 T cells are also important in the pathology associated with other diseases, including diabetes, lupus, and delayed type hypersensitivity which will be covered in Block Five.

SLIDE 31 Cell Mediated Immunity is cell-based and dependent upon T lymphocytes. T cells are antigen specific by their TcR. Major Histocompatibility restriction is especially important to control the type of T cell response and to limit non-specific T cell damage to other cells.
Summary

• AMI & CMI: Two sides of the same coin
• Adaptive immune response mediated by B & T cells
• Both have specific effector functions to protect against different types of pathogens
• Work together to produce a strong response, memory, and elimination of antigens

SLIDE 32 Don’t forget about B cells and antibody mediated immunity! Both B cells & T cells create an adaptive immune response, where they specifically target antigens (microbes) to create immunity. These lymphocyte populations begin as native cells that are either selected by (B cells) or presented (T cells) antigens that are specific to that clone. Upon antigen recognition, these cells rapidly undergo clonal expansion & differentiation into effector and memory cells. These cell types have different effector functions that can improve antibody production or enhance cell-mediated immunity (or both in the case of CD4+ cells). Both B & T cells differentiate into memory cells that can survive for long periods & create immunologic memory. This 2-pronged specific approach allows for the rapid elimination of microbes, and a faster response upon re-exposure, especially if the antigens are T dependent. Consequently, T cells are critical to protect the body as they regulate the immune response and can direct the predominant type of immunity necessary to defeat the enemy. For this reason, acquired immunodeficiency is a fatal disease, as the body can no longer protect itself from “harmless” microbes and memory is compromised because helper T cells counts decrease, which are critical to talk to B cells. The healthy immune response is incredibly complex, highly regulated to prevent recognition of self, specific and effective, with inter-linked pathways to eliminate microbes. Many parts of the immune system are still not completely understood, but are actively researched to further our understanding of the immune response, especially in the area of vaccine design & development that stimulates both antibody & cell mediated immunity!
In Summary

- CD8+ effector mechanisms
- CD4+ effector mechanisms (Th1 & Th2)
- How T cells help B cells
- Macrophage activation
- Cytokines that control the immune response
- Regulation of the immune response
- CMI immunopathology

Self-Test Questions

- How do T cells find the site of injury? Include specific adhesion molecules that attract them to the area & those that keep them at the site (hint: see text).
- Describe how CD8+ T cells kill infected cells. What does granzyme do?
- Describe the effector functions of CD4+ Th1 cells, Th2 cells.
- What cytokines are involved in the regulation of the immune system? What cytokines stimulate CMI? Humoral immunity?
- Name 3 examples of CMI immunopathology.