Cell-Mediated Immunity

Case 54

HPI: IL is a 45-year-old diabetic man **who had a kidney transplant** 2 weeks ago. IL now presents with **shortness of breath, severe hypertension, rapid weight gain,** and **oliguria** (decreased urine production). He has gained 10 pounds over the last 2 days and his blood pressure has increased from 130/85 to the 190s/100s. He has also had a progressive decline in urine output to 500 mL/day. He reports increased shortness of breath with new onset of **orthopnea.** The patient is taking **cyclophosphamide for immunosuppression.**

PE: T 38.0°C HR 85 BP **190/105** RR 22 SaO_2 96% on room air
On exam, he is in mild respiratory distress. His exam is significant for diffusely **increased skin turgor, bibasilar crackles** to the midchest, a **cardiac flow murmur,** a **distended abdomen** with left lower quadrant tenderness to palpation, and significant lower extremity **pitting edema.**

Labs: He has a normal CBC but an elevated potassium. His serum **creatinine is elevated** at 5.0. UA reveals **3+ protein** and WBC. Blood cultures are negative.

Thought Questions

- What is cell-mediated immunity and how does it defend against invading pathogens and foreign cells?
- What is the major histocompatibility complex and its involvement in antigen presentation? What are cytokines and how are they involved?
- How does tolerance to self-antigen develop among T-lymphocytes?
- How are foreign cells recognized and eliminated? What implication does this have on allogeneic transplant rejection?
- What is the pathogenesis of the different forms of transplant rejection and their clinical manifestations? How is transplant rejection medically suppressed?

Basic Science Review and Discussion

Cell-mediated immunity, an arm of the adaptive immune system, is involved in the surveillance of not only the extracellular but also the **intracellular compartment** for the elimination of pathogens. Its mediators not only help B-cells produce antibodies to neutralize extracellular pathogens but also eliminate intracellular pathogens by killing cells that harbor these pathogens. Targets of cellular immunity include mycobacteria, fungi, and cells viewed as defective or foreign, such as tumor cells and transplanted cells. Like the humoral immune system, the cellular immune system must be capable of (1) **specific recognition** of its targets, (2) processing and **presenting antigen** to effector cells involved in the immune response, (3) activating the most appropriate components of the immune response to optimize **pathogen elimination,** and (4) **establishing memory** of these pathogens for more rapid elimination upon re-exposure.

T-Lymphocyte Development T-lymphocytes are the primary effectors of cellular immunity. They arise from lymphoid progenitors in the bone marrow that mature and eventually migrate to the thymus where they undergo further development. On the next page is a diagram of T-cell development and the key receptors present in each stage. (See Figure 54-1.)

Antigen recognition in T-cells is mediated by the **T-cell receptor (TCR).** TCR is similar to the B-cell receptor in that it consists of two subunits, α and β, homologous to immunoglobulin heavy and light chains. The α-chain genes include multiple alleles for the V, J, and C regions. The β-chain genes include multiple alleles for the V, D, J, and C regions. The final TCR product consists of recombinations of these alleles within each region of the α- or β-chains. This rearrangement during development allows for the production of an almost infinite array of antigen-recognizing TCRs using relatively few genes.

Following migration to the thymus, T-cells undergo a process whereby they develop **the ability to distinguish self from nonself.** Within the thymus, T-cells are exposed to thymic epithelial cells and antigen-presenting cells (macrophages and dendritic cells) that expose them to the majority of self-peptides. Immature T-cells that recognize self-antigen are destroyed by **negative selection.** Those that successfully recognize MHC-I or MHC-II receptors, and not self antigens, undergo **positive selection.** Those that fail to recognize MHC at all die of attrition.

CD4+ T-cells become **T-helper cells,** which become restricted to recognizing antigen complexed to **class II MHC.** These cells support T- and B-cells in orchestrating the adaptive immune response. **CD8+ T-cells** become **cytotoxic T-cells,** restricted to recognizing antigen bound to **class I MHC** and to the lysis of virus-infected, tumor, or foreign cells. CD4+ and CD8+ T cells are released from the thymus into the bloodstream to mediate cellular immunity.

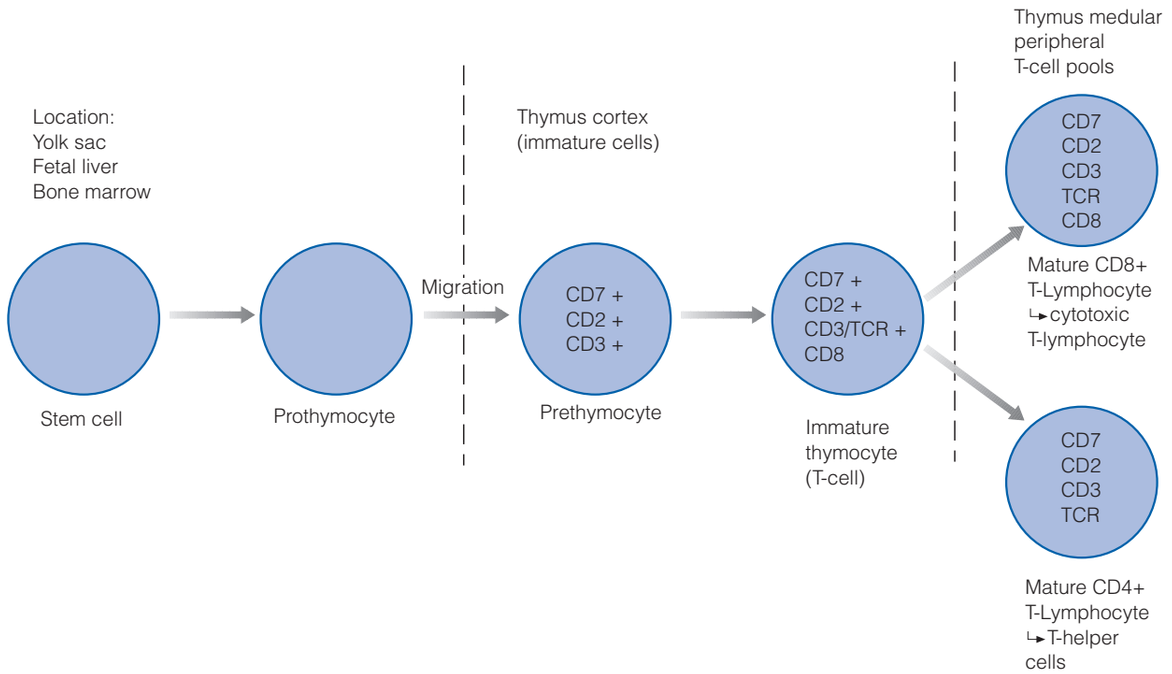

Figure 54-1 T-cell development begins at a primary hematopoietic site such as the bone marrow and ends in the thymus. In the thymus, T-cells undergo T-cell antigen receptor (TCR) rearrangement. The T-cell population is then selected for tolerance to self antigen and for reactivity to self-MHC receptors that will eventually bear foreign antigen. The immature thymocyte expresses TCR as well as CD4 and CD8 concurrently. T-cells are fully mature when they express either CD4 or CD8. CD4+ cells differentiate into T-helper cells that orchestrate the adaptive immune response. CD8+ cells develop into cytotoxic T-lymphocytes that kill infected cells as well as neoplastic and foreign cells.

Cell-Mediated Immunity Cell-mediated immunity is most easily demonstrated by the process of eliminating viral infection. Viruses have both extracellular and intracellular phases. An antigen-presenting cell (APC) may take up virus in two ways—through **phagocytosis** and through **direct viral infection.** Viruses that are phagocytized are fused with vesicles containing digestive enzymes. Viral peptides are then processed, bound to class II MHC molecules, and presented on the APC cell surface to CD4+ T-lymphocytes. The T-cells recognize the antigen-MHC complex via the TCR, stabilized by the CD4 receptor. A **costimulatory signal** between the APC's **B7 receptor** and the T-helper cell's **CD28 receptor** is required as a "confirmation" signal for T-cell activation. This up-regulates **CD40 ligand** expression in T-helper cells, which serves to drive B-cell **isotype switching,** a key step in antibody **affinity maturation** (see Figure 55-2 in Humoral Immunity section).

T-helper cells differentiate and are activated to secrete signaling peptides, or **cytokines,** depending on the nature of the pathogen to be eliminated. T-helper cells universally secrete **IL-2** upon activation, which stimulates proliferation of many immune cells including B-cells, cytotoxic lymphocytes (CTL), and T-helper cells. T-helper cells differentiate into two types: T_h1-cells and T_h2-cells. **T_h1-cell** differentiation is stimulated by APC-secreted **IL-12. T_h1-cells primarily promote immunity against small extracellular pathogens** such as viruses and bacteria, eliminated through phagocytosis and cell killing. Both CD40L and the cytokine **interferon-γ (IFN-γ)** promote B-cell isotype switching toward IgG production. IFN-γ also superactivates macrophage phagocytosis and antigen presentation by up-regulating their oxidative killing machinery and MHC expression, respectively. T_h1-cells also secrete **tumor necrosis factor (TNF),** which activates neutrophil phagocytosis and CTL-mediated elimination of virally infected cells.

T_h2-cells are differentiated toward eliminating large extracellular pathogens, such as parasites. T_h2-cell differentiation is driven by **IL-4,** secreted by mast cells and basophils after a parasite encounter. T_h2-cells in turn secrete more cytokines like IL-4 itself, which promotes B-cell isotype switching to IgE, the primary anti-parasitic antibody. **IL-5** promotes eosinophil function, the primary parasite killer cell. Eosinophils recognize the Fc portion of IgE via **Fcε receptors,** which activate the extracellular release of **major basic protein** and other enzymes that attack the parasite's cell membranes. See Figures 54-2 and 54-3.

To initiate intracellular compartment surveillance, APCs take up virus through direct infection. Viral peptides produced during replication are processed differently than

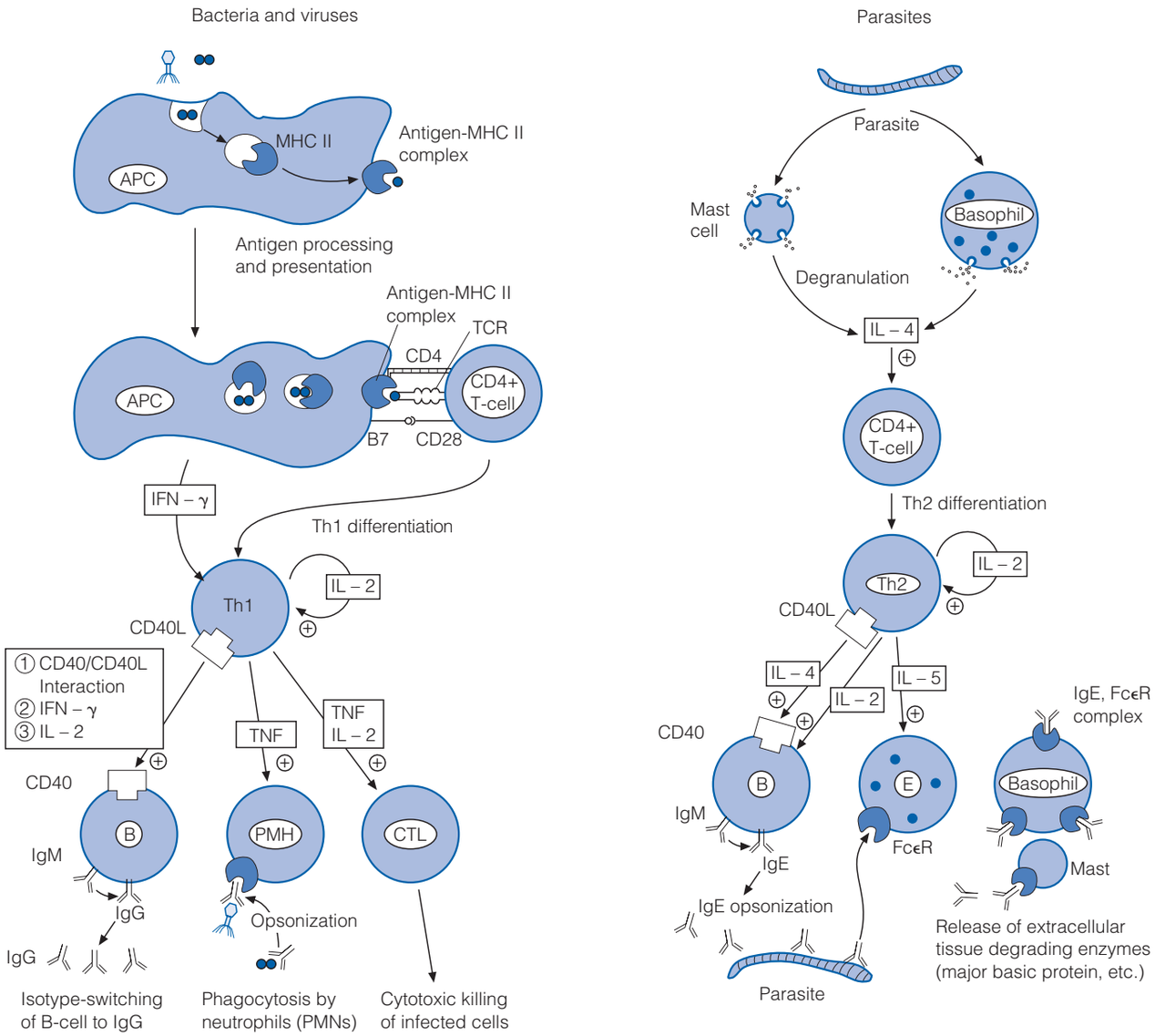

Figure 54-2 Cell-mediated immunity against intracellular pathogens, such as viruses. This pathway uses class I MHC to present antigens that are found in the cytosolic compartment of any cell. CD8+ T-lymphocytes are subsequently activated and mature into cytotoxic T-cells, which kill infected cells. T-helper cells are also involved in augmenting this immune response.

those that are phagocytized; intracellularly replicated viral particles are ultimately associated with class I MHC molecules. **This antigen-MHC I complex specifically activates CD8+ T-lymphocytes, which are then stimulated to differentiate into cytotoxic lymphocytes.** One important molecular interaction includes the up-regulated CD40L/CD40 receptors.

The CD40L-CD40 interaction activates CTLs to produce several cell-killing mediators. **Fas ligand** surface expression is up-regulated, which binds to **Fas** on infected cells and thereby induces cell **apoptosis.** The CD40L-CD40 interaction also promotes the production of **perforin, granzymes,** and **caspases** that kill cells by inserting into the plasma membrane of the infected cell, forming a pore that leads to cell lysis. CTLs recognize virus-infected cells by their TCRs binding to class I MHC molecules, presenting the specific viral peptide to which they are sensitized. This activates the cytotoxic machinery described above. Finally, the T-helper cell's CD40L also binds CD40 on B-cells, promoting activation and antibody production.

Immunologic memory to a pathogen is established through the differentiation of a subpopulation of activated T-helper, B-, and CD8+ T-cells into their own respective memory cells. If the body is re-exposed to the antigen, it

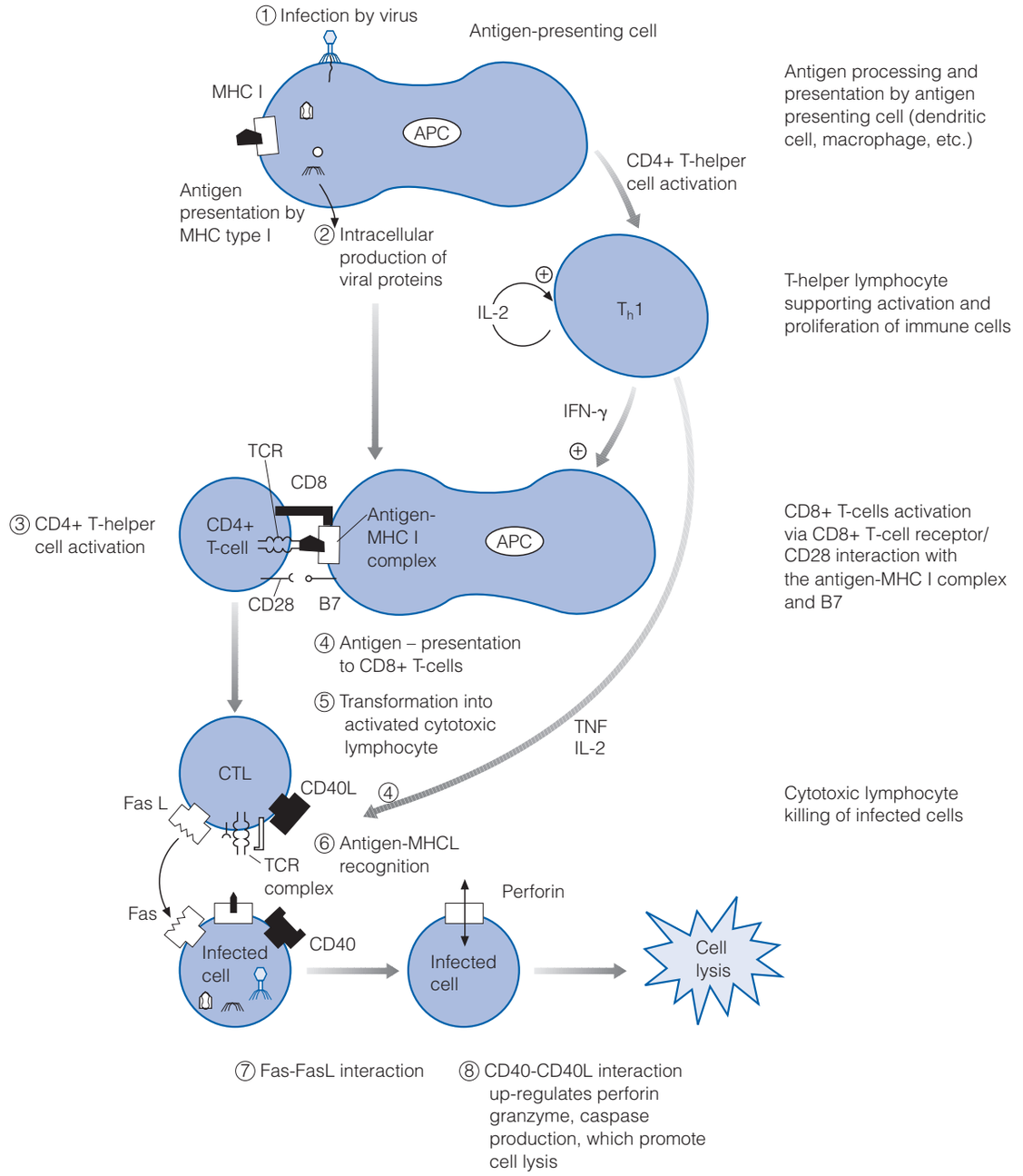

Figure 54-3 Cell-mediated immunity against extracellular pathogens, such as bacteria and fungi. Class II MHC are used to present antigens endocytosed by antigen-presenting cells, such as macrophages to CD4+ cells. These cells are activated and mature into T-helper cells that subsequently promote B-lymphocyte maturation, plasma cell antibody production, and the phagocytic activity of macrophages, neutrophils, and eosinophils in the elimination of bacteria and fungi.

will be able to mount a faster, more effective immune response with more efficient elimination of the pathogen, the process we commonly refer to as "immunity."

Clinical Discussion

Transplant Rejection There are two types of allograft rejection: humoral (antibody-mediated) and cellular (T-

Table 54-1 Types of rejection and pathogenesis

Rejection Type	Timing	Pathogenesis
Hyperacute rejection	Minutes to < 48 hr	• Classically upon reperfusion of allograft → graft failure • Preformed antibodies to HLA, ABO Ag, and vascular endothelium are in circulation and bind immediately to vasculature, causing complement activation, platelet aggregation, and thrombosis → vasculitis and ischemia
Accelerated rejection	7–10 days	• Humoral: anti-HLA antibodies • Cellular: T-lymphocyte mediated, from prior sensitization • 60% graft loss
Acute rejection	7 days–3 months	• Development of cellular and humoral immunity, often due to inadequate immunosuppression • Signs and symptoms: graft pain, warmth, edema, fever, malaise, fatigue, and signs of specific organ failure
Chronic rejection	Months to years	• Unclear etiology: presumably humoral and cellular • Signs and symptoms of specific organ failure • Diagnosed by biopsy, showing concentric vessel narrowing and ischemic disease of the graft

lymphocyte-mediated). In **humoral rejection,** antibodies are formed by B-cells that recognize foreign antigen on allograft tissue. These antibodies bind allograft tissue and activate complement, which leads to platelet aggregation and thrombosis. Ischemic injury and ultimately transplant rejection ensues.

There are two pathways in **cell-mediated allograft rejection:** the direct and indirect pathways. In the **direct pathway,** recipient CD4+ T-helper cells and CD8+ T-cells recognize foreign class II and class I **human leukocyte antigens** (HLA, the MHC in humans), respectively, on **dendritic cells** (APCs) carried in the donor organ. The T-cells are then activated and cause a local increase in vascular permeability, lymphocyte and macrophage infiltration, and cell lysis of allograft tissue. In the **indirect pathway,** foreign HLA is processed like any foreign antigen and presented by the host's own APCs to T-cells; this eventually leads to transplant rejection.

Therefore, an important clinical issue is **HLA matching** of the donor to the transplant recipient. There are six HLA types to match: HLA-A, HLA-B, and HLA-C (those that contribute to class I HLA) and HLA-DP, HLA-DQ, and HLA-DR (those in class II HLA). Individuals are likely to match with siblings, having a 25% chance of matching. However, the likelihood of matching falls dramatically between unrelated individuals. Matching affords prolonged survival of the graft. With the exception of identical twins, lifelong immunosuppression is necessary to prolong graft survival and is accomplished with such drugs as azathioprine, steroids, cyclosporine, antilymphocyte globulins, and monoclonal anti-T-cell antibodies. Immunosuppression unfortunately also makes the recipient susceptible to opportunistic infections. (See Table 54-1.)

Case Conclusion With the suspicion of acute kidney transplant rejection, our patient is started on prednisone and stabilized hemodynamically with diuretics and ACE inhibitors. His immunosuppressive dosing regimen is increased and his symptoms eventually resolve. At this time, IL continues to have normal functioning of his kidney allograft.

Thumbnail: Cytokines and Their Actions

Cytokine	Cell origin	Cell target	Stimulus for release and subsequent actions
Cytokines Released from Local Insult			
IL-1	Macrophages Infected cells	T- and B-cells Neutrophils Epithelial cells Dendritic cells	Secretion stimulated by infection → • Acts on hypothalamus to induce fever • Stimulates cell growth • T-cell differentiation, IL-2 production
IL-6	Macrophages Neutrophils	Hepatocytes T- and B-cells	Phagocytosis of microbe stimulates → • Production of acute phase proteins (e.g., ESR) • Promotes T- and B-cell growth and differentiation
TNF-α	Macrophages Mast cells/basophils Eosinophils NK cells T- and B-cells	All cells except RBCs	Local tissue damage or infection → • Induces fever, anorexia, shock • Causes capillary leak syndrome • Enhanced cytotoxicity, NK cell function • Acute phase protein synthesis
TNF-β	T- and B-cells	All except RBCs	• Cell cytotoxicity, lymph node, and spleen development
Cytokines of Anti-viral Immunity			
IFN-α/β	Macrophages Infected cells	All cells, especially neighboring cells	Viral infection → induces antiviral state; antitumor activity • Up-regulates MHC class I antigen expression • ↓ Protein synthesis in infected cell • ↑ RNase expression → degrades viral RNA
Cytokines of T-helper Type 1 Cells			
IFN-γ	T_h1 cells NK cells	Macrophage NK cells B-cells Dendritic cells	Bacterial/viral infection → antigen presentation • Promotes T_h1 cell differentiation • Regulates macrophage and NK cell activation • Stimulates B-cell growth and IgG isotype switching
IL-2	T_h1 cells	T-helper cells CD8+ cells B-cells	• Activates all branches of adaptive immunity to proliferate and differentiate (activates cytokine production, effector functions)
IL-12	APC: macrophages, dendritic cells	T-helper cells CD8+ T-cells NK cells	• Promotes T_h1 proliferation • CTL activation • NK cell activation
Cytokines of T-helper Type 2 Cells			
IL-4	T_h2 cells	B-cells T-helper cells	Helminthic infection and allergen exposure → • Stimulates B-cell growth; IgE isotype switching • Recruits eosinophils • Promotes T_h2 cell proliferation • Inhibits macrophages/delayed-type reaction
IL-5	T_h2 cells	B-cells Eosinophils	• Activates eosinophils IgE binding via FcεR • Stimulates B-cell differentiation
IL-10	T_h2 cells	Macrophages	• Inhibits macrophage activation
Cytokines of Mast Cells			
Histamine Serotonin	Mast cells	Vascular endothelium	Helminthic infection and allergen exposure → • Increased vascular permeability → hypotension, edema
Lipid Mediators		Smooth muscle cells	• Bronchiole contraction → bronchospasm • Intestinal hypermotility → diarrhea

Key Points

- Cell-mediated immunity is the arm of the immune system specialized in targeting intracellular pathogens (e.g., viruses, mycobacteria, fungi), as well as tumor cells and foreign cells
- Specific recognition of pathogens is accomplished by antigen presentation on major histocompatibility complexes of antigen-presenting cells to T-lymphocytes
- Activated T-helper cells release cytokines that activate and support all branches of the adaptive immune system
- A subset of T-cells and B-cells persists as memory cells that can respond with great efficacy upon re-exposure of the body to the original pathogen

Questions

1. The following is a series of immunological events that occurred during the course of IL's transplant rejection. Which step did *not* occur?

A. Antigen-presenting cells from the donor and the recipient take up HLA protein from the allograft and present it to recipient T-helper cells on class I and class II HLA receptors

B. B-cells are activated by T_h2 cells to proliferate and isotype switch to producing IgG, which bind to allograft vascular endothelium and activate complement, causing thrombosis and ischemic injury to the graft

C. Activated T-helper cells secrete IL-2 and thereby promote proliferation and maturation of B-cells, CD4+ T-cells, CD8+ T-cells, and monocytes into plasma cells, T_h1 cells, cytotoxic T-lymphocytes, and macrophages, respectively

D. TNF-α is released from activated macrophages, creating symptoms such as fever and malaise, promoting allograft edema by increasing local vascular permeability, and promoting allograft rejection by enhancing NK cell cytotoxicity

E. The CD40-CD40L interaction between allograft dendritic cells and IL's T-helper cells promote cytotoxic lymphocyte activity by up-regulating *Fas* ligand receptor expression, which binds *Fas* on infected cells and induces apoptosis of that cell

2. There is an immunodeficiency of the interferon-γ receptor that leads to serious infections caused by the BCG vaccine for *Mycobacterium tuberculosis* and nontuberculous mycobacteria. What immune machinery would be defective in this deficiency?

A. There is a lack of signal to the hypothalamus to induce enough of a febrile reaction to eliminate mycobacterial infection

B. This deficiency yields a defective mechanism of specific antigen recognition, thereby shielding mycobacterial species from immune recognition

C. This deficiency prevents the up-regulation of MHC class I and RNase expression, thus preventing intracellular mycobacteria from having their antigen presented to T-helper cells as well as preventing degradation of the pathogen's genomic material

D. This deficiency prevents the proliferation of subtype 1 T-helper cells and the subsequent activation of oxidative killing by macrophages required to fully eliminate phagocytized mycobacteria

E. This deficiency prevents the activation of B-cells to isotype switch toward IgE production, preventing the elimination of mycobacteria