infected host cells without any need for dendritic cells or signals other than recognition of antigen (see Chapter 6).

Now that we know how protein antigens are captured, transported to, and concentrated in peripheral lymphoid organs, the next question is how are these antigens displayed to T lymphocytes? To answer this question, we first need to understand what MHC molecules are and how they function in immune responses.

The Structure and Function of Major Histocompatibility Complex Molecules

MHC molecules are membrane proteins on APCs that display peptide antigens for recognition by T lymphocytes. The MHC was discovered as the genetic locus that is the principal determinant of acceptance or rejection of tissue grafts exchanged between individuals. In other words, individuals that are identical at their MHC locus (inbred animals and identical twins) will accept grafts from one another, and individuals that differ at their MHC loci will reject such grafts. Graft rejection is, of course, not a natural biologic phenomenon, and therefore MHC genes, and the molecules they encode, could not have evolved only to mediate graft rejection. We now know that the physiologic function of MHC molecules is to display peptides derived from protein antigens to antigen-specific T lymphocytes. This function of MHC molecules is the explanation for the phenomenon of MHC restriction of T cells, which was mentioned earlier.

The collection of genes that make up the MHC locus is found in all mammals (Fig. 3-6) and includes genes that encode MHC and other proteins. Human MHC proteins are called **human leukocyte antigens (HLAs)** because these proteins were discovered as antigens of leukocytes that could be identified with specific antibodies. In all species, the MHC locus contains two sets of highly polymorphic genes, called the class I and class II MHC genes. These genes encode the class I and class II MHC molecules that display peptides to T cells. In addition to the polymorphic genes, the MHC locus contains many nonpolymorphic genes. Some of these nonpolymorphic genes code for proteins involved in antigen presentation, and others code for proteins whose function is not known.

Class I and class II MHC molecules are membrane proteins that each contains a peptide-binding cleft at the amino-terminal end. Although the two classes of molecules differ in subunit composition, they are very similar in overall structure (Fig. 3-7).

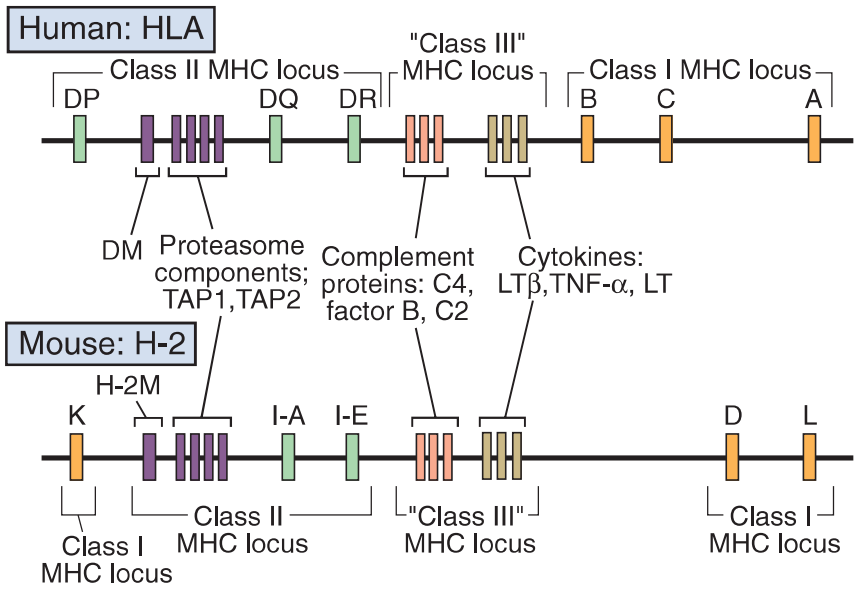


FIGURE 3-6 The genes of the major histocompatibility complex (MHC) locus. Schematic maps of the human MHC (called the HLA complex) and the mouse MHC (called the H-2 complex) are shown, illustrating the major genes that code for molecules involved in immune responses. Sizes of genes and intervening DNA segments are not shown to scale. Class II loci are shown as single blocks but each consists of at least two genes. Class III MHC locus refers to genes that encode molecules other than peptide display molecules; this term is not used commonly. There are also multiple class I–like genes and pseudogenes *(not shown)*. HLA, human leukocyte antigen; LT, lymphotoxin; TAP, transporter associated with antigen processing; TNF, tumor necrosis factor.

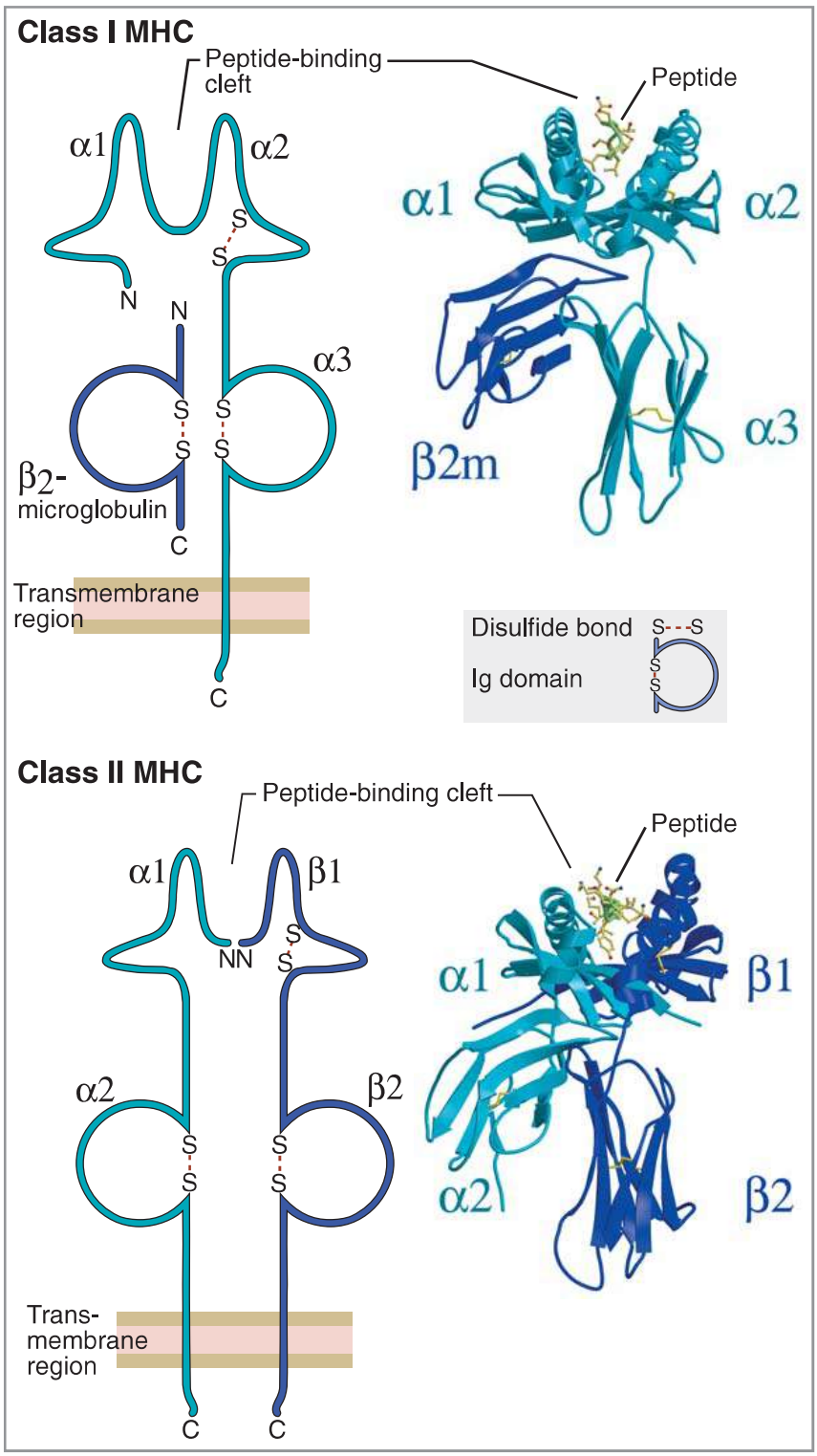


FIGURE 3-7 The structure of class I and class II major histocompatibility complex (MHC) molecules. The schematic diagrams (at *left*) and models (at *right*) of the crystal structures of class I MHC and class II MHC molecules illustrate the domains of the molecules and the fundamental similarities between them. Both types of MHC molecules contain peptide-binding clefts and invariant portions that bind CD8 (the α3 domain of class I) or CD4 (the β2 domain of class II). β2m, β_2-microglobulin; Ig, immunoglobulin. (Crystal structures courtesy of Dr. P. Bjorkman, California Institute of Technology, Pasadena, California.)

Each class I molecule consists of an α chain noncovalently attached to a protein called β_2-microglobulin that is encoded by a gene outside the MHC. The amino-terminal α1 and α2 domains of the class I MHC molecule form a peptide-binding cleft, or groove, that is large enough to accommodate peptides of 8 to 11 amino acids. The floor of the peptide-binding cleft is the region that binds peptides for display to T lymphocytes, and the sides and tops of the cleft are the regions that are contacted by the T cell receptor (which, of course, contacts part of the displayed peptide as well) (see Fig. 3-1). The polymorphic residues of class I molecules, that is, the amino acids that differ among different individuals' MHC molecules, are located in the α1 and α2 domains of the α chain. Some of these polymorphic residues contribute to variations in the floor of the peptide-binding cleft and thus in the ability of different MHC molecules to bind peptides. Other polymorphic residues contribute to variations in the tops of the clefts and thus influence recognition by T cells. The α3 domain is invariant and contains the binding site for the T cell co-receptor CD8. As we shall see in Chapter 5, T cell activation requires recognition of MHC-associated peptide antigen by the TCR and simultaneous recognition of the MHC molecule by the co-receptor. Therefore, $CD8^+$ T cells can only respond to peptides displayed by class I MHC molecules, the MHC molecules to which the CD8 co-receptor binds.

Each class II MHC molecule consists of two chains, called α and β. The amino-terminal regions of both chains, called the α1 and β1 domains, contain polymorphic residues and form a cleft that is large enough to accommodate peptides of 10 to 30 residues. The nonpolymorphic β2 domain contains the binding site for the T cell co-receptor CD4. Because CD4 binds to class II MHC molecules, $CD4^+$ T cells can only respond to peptides presented by class II MHC molecules.

Several features of MHC genes and molecules are important for the normal function of these molecules (Fig. 3-8):

MHC genes are codominantly expressed, meaning that the alleles inherited from both parents are expressed equally. Because there are three polymorphic class I genes, called HLA-A, HLA-B, and HLA-C in humans, and each person inherits one set of these genes from each parent, any cell can express six different class I molecules. In the class II locus, every individual inherits one pair of HLA-DP genes (called DPA1 and DPB1, encoding the α and β chains), one pair of HLA-DQ genes (DQA1 and DQB1, encoding the α and β chains), one HLA-DRα gene (DRA1), and one or two HLA-DRβ genes (DRB1 and DRB3, -4 or -5). Thus, a heterozygous individual can inherit six or eight class II MHC alleles, three or four from each parent (one set each of DP and DQ, and one or two of DR). Because of the extra DRβ genes, and because some DQα molecules encoded on one chromosome can associate with DQβ molecules encoded from the other chromosome, the total number of expressed class II molecules may be considerably more than 6.

The set of MHC alleles present on each chromosome is called an **MHC haplotype.** In humans, each HLA allele is given a numerical designation. For instance, an HLA haplotype of an individual could be HLA-A2, HLA-B5, HLA-DR3, and so on. All heterozygous individuals, of course, have two HLA haplotypes, one from each chromosome.

MHC genes are highly polymorphic, meaning that many different alleles are present among the different individuals in the population. The polymorphism is so great that any two individuals in an outbred population are unlikely to have exactly the same MHC genes and molecules. Because the polymorphic residues determine which peptides are presented by which MHC molecules, the existence of multiple alleles ensures that there are always some members of the population that will be able to present any particular microbial protein antigen. The evolution of MHC polymorphism ensures that a population will be able to deal with the diversity of microbes and will not succumb to a newly encountered or mutated microbe, because at least some individuals will be able to mount effective immune responses to the peptide antigens of these microbes. The variations in MHC molecules (accounting for the polymorphism) result from inheritance of distinct DNA sequences and are not induced by gene recombination (as they are in antigen receptors; see Chapter 4).

Class I molecules are expressed on all nucleated cells, but class II molecules are expressed mainly on dendritic cells, macrophages and B lymphocytes. The physiologic significance of this strikingly different expression pattern is described later in the chapter.

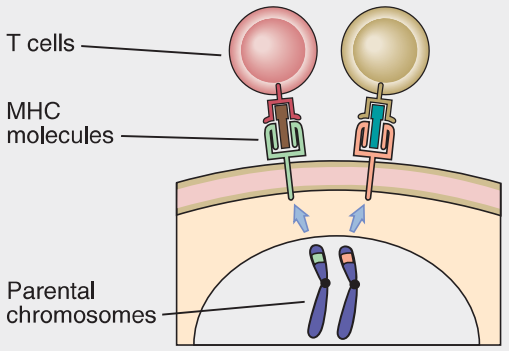
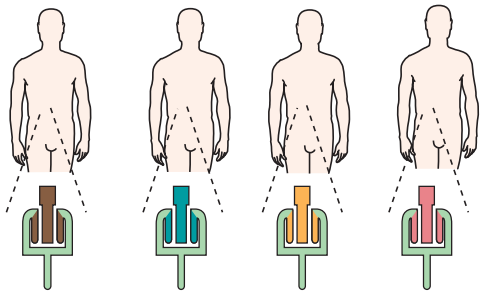
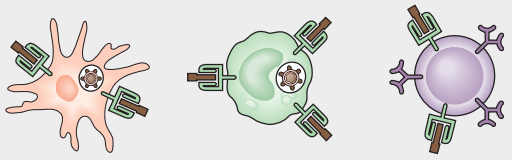
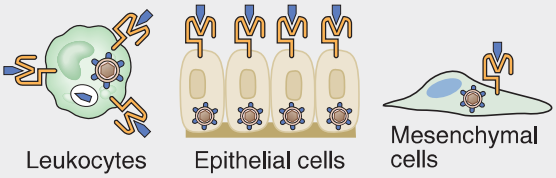
Feature	Significance	
Codominant expression: Both parental alleles of each MHC gene are expressed	Increases number of different MHC molecules that can present peptides to T cells	
Polymorphic genes: Many different alleles are present in the population	Ensures that different individuals are able to present and respond to different microbial peptides	
MHC-expressing cell types: Class II: Dendritic cells, macrophages, B cells	CD4+ helper T lymphocytes interact with dendritic cells, macrophages, B lymphocytes	
Class I: All nucleated cells	CD8+ CTLs can kill any virus-infected cell	

FIGURE 3-8 Properties of major histocompatibility complex (MHC) molecules and genes. Some of the important features of MHC molecules are listed, with their significance for immune responses. CTLs, cytotoxic T lymphocytes.

Class II molecules also are expressed on thymic epithelial cells and endothelial cells and can be induced on other cell types by the cytokine interferon-γ.

The peptide-binding clefts of MHC molecules bind peptides derived from protein antigens and display these peptides for recognition by T cells (Fig. 3-9). There are pockets in the floors of the peptide-binding clefts of most MHC molecules. The side chains of amino acids in the peptide antigens fit into these MHC pockets and anchor the peptides in the cleft of the MHC molecule. Peptides that are anchored in the cleft by these side chains (also called

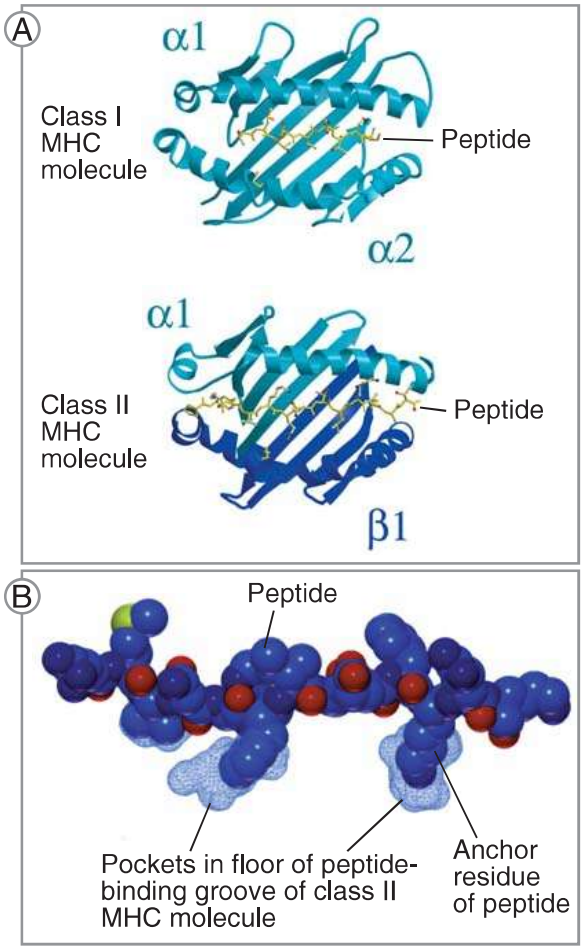


FIGURE 3-9 Binding of peptides to major histocompatibility complex (MHC) molecules. A, These top views of the crystal structures of MHC molecules show how peptides (in *yellow*) lie on the floors of the peptide-binding clefts and are available for recognition by T cells. (Courtesy of Dr. P. Bjorkman, California Institute of Technology, Pasadena, California.) **B,** The side view of a cut-out of a peptide bound to a class II MHC molecule shows how anchor residues of the peptide hold it in the pockets in the cleft of the MHC molecule. (From Scott CA, Peterson PA, Teyton L, Wilson IA: Crystal structures of two I-Ad-peptide complexes reveal that high affinity can be achieved without large anchor residues. Immunity 8:319-329, 1998. © Cell Press; with permission.) These structures are the basis for the schematic view of peptide recognition by T cells shown in Figure 3-1.

anchor residues) contain some residues that bow upward and are recognized by the antigen receptors of T cells.

Several features of the interaction of peptide antigens with MHC molecules are important for understanding the peptide display function of MHC molecules (Fig. 3-10).

Each MHC molecule can present only one peptide at a time, because there is only one cleft, but each MHC molecule is capable of presenting many different peptides. So long as the pockets of the MHC molecule can accommodate the anchor residues of the peptide, that peptide can be displayed by the MHC molecule. Therefore, only one or two residues in a peptide have to fit into an MHC molecule's cleft. Thus, MHC molecules are said to have a "broad" specificity for peptide binding: Each molecule can bind many but not all possible peptides. This feature is essential for the antigen display function of MHC molecules, because each individual has only a few different MHC molecules that must be able to present a vast number and variety of antigens.

MHC molecules bind only peptides and not other types of antigens. This is why MHC-restricted $CD4^+$ T cells and $CD8^+$ T cells can recognize and respond to only protein antigens, the natural source of peptides.

MHC molecules acquire their peptide cargo during their biosynthesis and assembly inside cells. Therefore, MHC molecules display peptides derived from microbes that are inside host cells, and this is why MHC-restricted T cells recognize cell-associated microbes and are the mediators of immunity to intracellular microbes. Of importance, class I MHC molecules acquire peptides from cytosolic proteins and class II molecules from proteins in intracellular vesicles. The mechanisms and significance of these processes are discussed later in the chapter. Only peptide-loaded MHC molecules are stably expressed on cell surfaces. The reason for this is that MHC molecules must assemble both their chains and bound peptides to achieve a stable structure, and "empty" molecules are degraded inside cells. This requirement for peptide binding ensures that only "useful" MHC molecules, that is, those that are displaying peptides, are expressed on cell surfaces for recognition by T cells. Once peptides bind to MHC molecules and are displayed on the cell surface, they stay bound for a long time, even up to days. The slow off-rate ensures that after an MHC molecule has acquired a peptide, it will display the peptide long enough

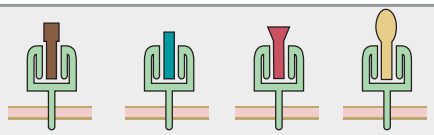
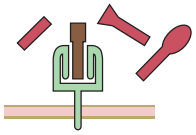
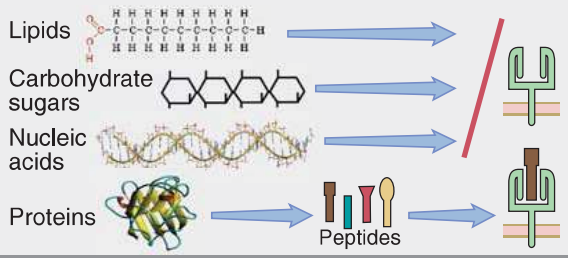
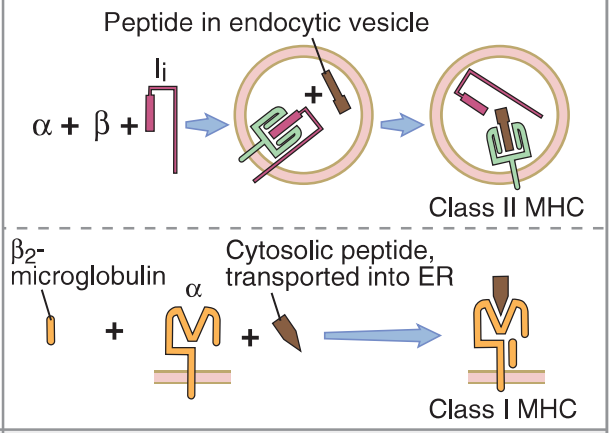
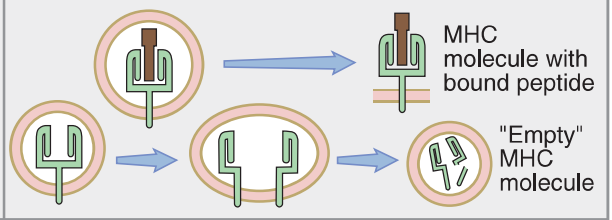
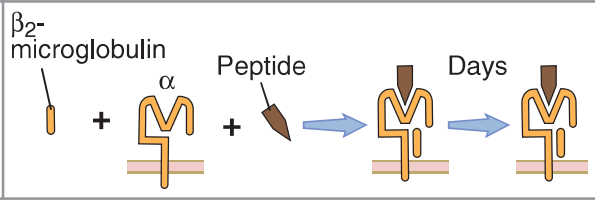
Feature	Significance	
Broad specificity	Many different peptides can bind to the same MHC molecule	
Each MHC molecule displays one peptide at a time	Each T cell responds to a single peptide bound to an MHC molecule	
MHC molecules bind only peptides	MHC-restricted T cells respond only to protein antigens, and not to other chemicals	
Peptides are acquired during intracellular assembly	Class I and class II MHC molecules display peptides from different cellular compartments	
Stable surface expression of MHC molecule requires bound peptide	Only MHC molecules that are displaying peptides are expressed for recognition by T cells	
Very slow off-rate	MHC molecule displays bound peptide for long enough to be located by T cell	

FIGURE 3-10 Features of peptide binding to major histocompatibility complex (MHC) molecules. Some of the important features of peptide binding to MHC molecules are listed, with their significance for immune responses. ER, endoplasmic reticulum.

to maximize the chance that a particular T cell will find the peptide it can recognize and initiate a response.

In each individual, the MHC molecules can display peptides derived from foreign, that is, microbial, proteins as well as peptides from that individual's own proteins. This inability of MHC molecules to discriminate between foreign antigens and self antigens raises two questions. First, at any time, the quantity of self proteins is certain to be much greater than that of any microbial antigens. Why, then, are the available MHC molecules not constantly occupied by self peptides and unable to present foreign antigens? The likely answer is that new MHC molecules are constantly being synthesized, ready to accept peptides, and they are adept at capturing any peptides that are present in cells. Also, a single T cell may need to see a peptide displayed by only as few as 0.1% to 1% of the approximately 10^5 MHC molecules on the surface of an APC, so that even rare MHC molecules displaying a peptide are enough to initiate an immune response. The second problem is that if MHC molecules are constantly displaying self peptides, why do we not develop immune responses to self antigens, so-called autoimmune responses? The answer to this question is that T cells specific for self antigens are either killed or inactivated; this process is discussed in Chapter 9. Although it seems puzzling that MHC molecules present self peptides, this is actually the key to the normal surveillance function of T cells. Thus, T cells are constantly patrolling the body looking at MHC-associated peptides, not reacting to peptides derived from self proteins but able to respond to rare microbial peptides.

MHC molecules are capable of displaying peptides but not intact microbial protein antigens. It follows that there must be mechanisms for converting naturally occurring proteins into peptides able to bind to MHC molecules. This conversion, called **antigen processing**, is described next.

Processing and Presentation of Protein Antigens

Extracellular proteins that are internalized by specialized APCs (dendritic cells, macrophages, and B cells) are processed in vesicles and displayed by class II MHC molecules, whereas proteins in the cytosol of any nucleated cell are processed in the cytoplasm and displayed by class I MHC molecules (Fig. 3-11). These two pathways of antigen processing involve different cellular organelles and proteins (Fig. 3-12). They are designed to sample all of the proteins present in the extracellular and intracellular environments. The segregation of antigen-processing pathways also ensures that different classes of T lymphocytes recognize antigens from different compartments, as is discussed later.

PROCESSING OF INTERNALIZED ANTIGENS FOR DISPLAY BY CLASS II MAJOR HISTOCOMPATIBILITY COMPLEX MOLECULES

APCs may internalize extracellular microbes or microbial proteins by several mechanisms (Fig. 3-13). Microbes may bind to surface receptors specific for microbial products or to receptors that recognize antibodies or products of complement activation that are attached to the microbes. B lymphocytes internalize proteins that specifically bind to the cells' antigen receptors (see Chapter 7). Some APCs may phagocytose microbes or pinocytose proteins without any specific recognition event. After internalization into APCs by any of these pathways, the microbial proteins enter acidic intracellular vesicles, called endosomes or phagosomes, which may fuse with lysosomes. In these vesicles the proteins are broken down by proteolytic enzymes, generating many peptides of varying lengths and sequences.

APCs constantly synthesize class II MHC molecules in the endoplasmic reticulum (ER). Each newly synthesized class II molecule carries with it an attached protein called the invariant chain, which contains a sequence (called the class II invariant chain peptide [CLIP]) that binds tightly to the peptide-binding cleft of the class II molecule. Thus, the cleft of the newly synthesized class II molecule is occupied. This "inaccessible" class II molecule begins its transport to the cell surface in an exocytic vesicle, which then fuses with the endosomal vesicle containing peptides derived from ingested extracellular proteins. The same endosomal vesicle contains a class II–like protein called DM, whose function is to remove CLIP from the class II MHC molecule. After removal of CLIP, the cleft of the class II molecule becomes

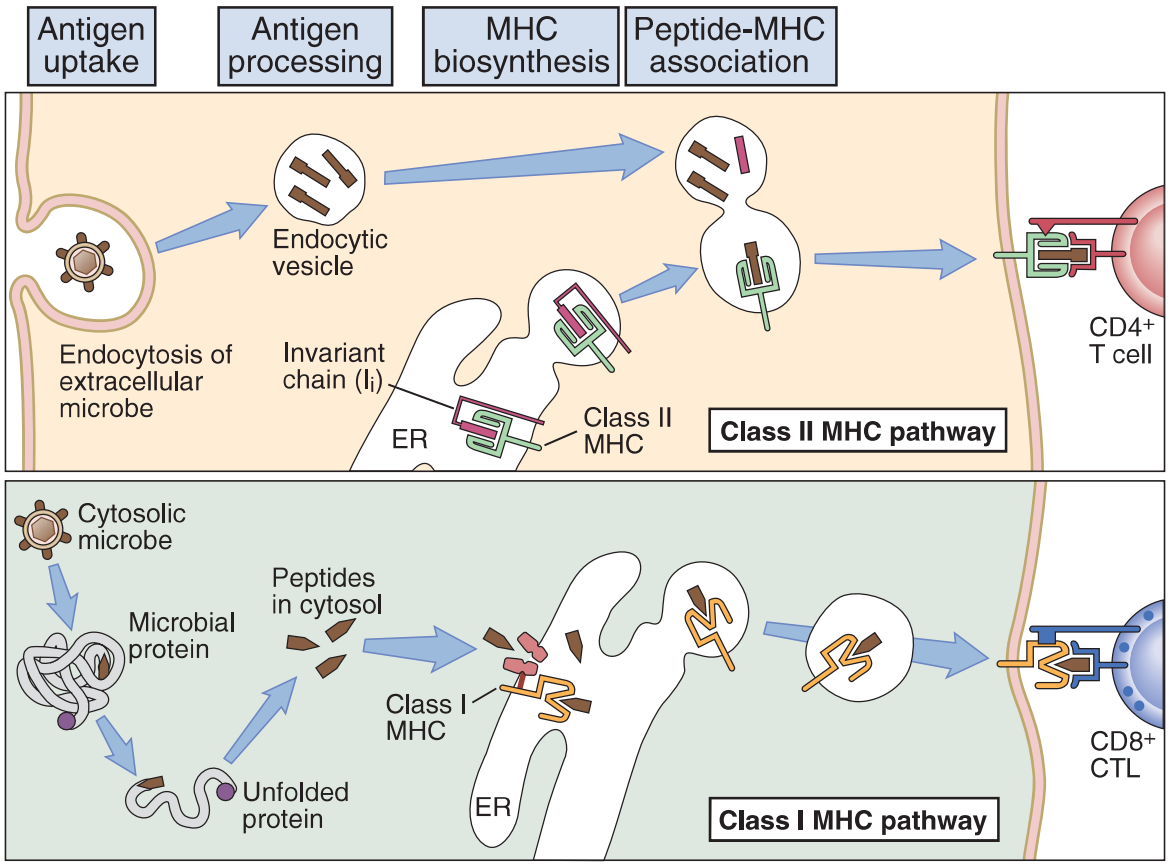


FIGURE 3-11 Pathways of intracellular processing of protein antigens. The class II MHC pathway converts protein antigens that are endocytosed into vesicles of antigen-presenting cells into peptides that bind to class II MHC molecules for recognition by $CD4^+$ T cells. The class I MHC pathway converts proteins in the cytoplasm into peptides that bind to class I MHC molecules for recognition by $CD8^+$ T cells. CTL, cytotoxic T lymphocyte; ER, endoplasmic reticulum; MHC, major histocompatibility complex.

available to accept peptides. If the class II MHC molecule is able to bind one of the peptides generated from the ingested proteins, the complex becomes stable and is delivered to the cell surface. If the MHC molecule does not find a peptide it can bind, the empty molecule is unstable and is degraded by proteases in the endosomes. One protein antigen may give rise to many peptides, only a few of which (perhaps only one or two) may bind to the MHC molecules present in the individual. Therefore, only these peptides derived from the intact antigen stimulate immune responses in that individual; such peptides are said to be the **immunodominant epitopes** of the antigen.

PROCESSING OF CYTOSOLIC ANTIGENS FOR DISPLAY BY CLASS I MAJOR HISTOCOMPATIBILITY COMPLEX MOLECULES

Antigenic proteins may be produced in the cytoplasm from viruses that are living inside infected cells, from some phagocytosed microbes that may leak from, or be transported out of, the vesicles into the cytoplasm, and from mutated or altered host genes, as in tumors. All of these proteins, as well as the cell's own nonfunctional cytoplasmic proteins, are targeted for destruction by proteolysis. These proteins are unfolded, covalently tagged with multiple copies of a small peptide called ubiquitin, and "threaded" through a

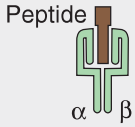
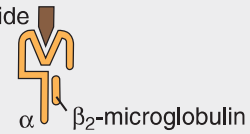
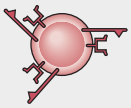
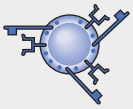
Feature	Class II MHC Pathway	Class I MHC pathway
Composition of stable peptide-MHC complex	Polymorphic α and β chains, peptide 	Polymorphic α chain, β_2-microglobulin, peptide 
Types of APCs	Dendritic cells, mononuclear phagocytes, B lymphocytes; some endothelial cells, thymic epithelium	All nucleated cells
Responsive T cells	$CD4^+$ T cells (helper T cells) 	$CD8^+$ T cells (CTLs) 
Source of protein antigens	Endosomal/lysosomal proteins (mostly internalized from extracellular environment)	Cytosolic proteins (mostly synthesized in the cell; may enter cytosol from phagosomes)
Enzymes responsible for peptide generation	Endosomal and lysosomal proteases (e.g., cathepsins)	Cytosolic proteasome
Site of peptide loading of MHC	Specialized vesicular compartment	Endoplasmic reticulum
Molecules involved in transport of peptides and loading of MHC molecules	Invariant chain, DM	TAP

FIGURE 3-12 Features of the pathways of antigen processing. APCs, antigen-presenting cells; CTL, cytotoxic T lymphocyte; MHC, major histocompatibility complex; TAP, transporter associated with antigen processing.

proteolytic organelle called the proteasome, where the unfolded proteins are degraded by enzymes (Fig. 3-14). Some classes of proteasomes efficiently cleave cytosolic proteins into peptides with the size and sequence properties typical of class I MHC–binding peptides. But the cell faces another challenge: The peptides are in the cytoplasm, while the MHC molecules are being synthesized in the ER, and the two have to come together. This problem is overcome by a specialized transport molecule, called transporter associated with antigen processing (TAP), which is located in the ER membrane. TAP binds peptides from the cytoplasm and actively pumps them across the ER membrane into the interior of the ER. (This, of course, is the reverse of the normal direction of protein traffic, which is from the site of synthesis in the ER out into the cytoplasm or to the plasma membrane.) Newly synthesized class I MHC molecules are loosely attached to the interior face of the TAP molecule. Thus, as peptides enter the ER, they can be captured by the class I molecules. (Recall that in the ER, the class II MHC molecules are not able to bind peptides because of the invariant chain.) If a class I molecule finds a peptide with the right fit, the complex is

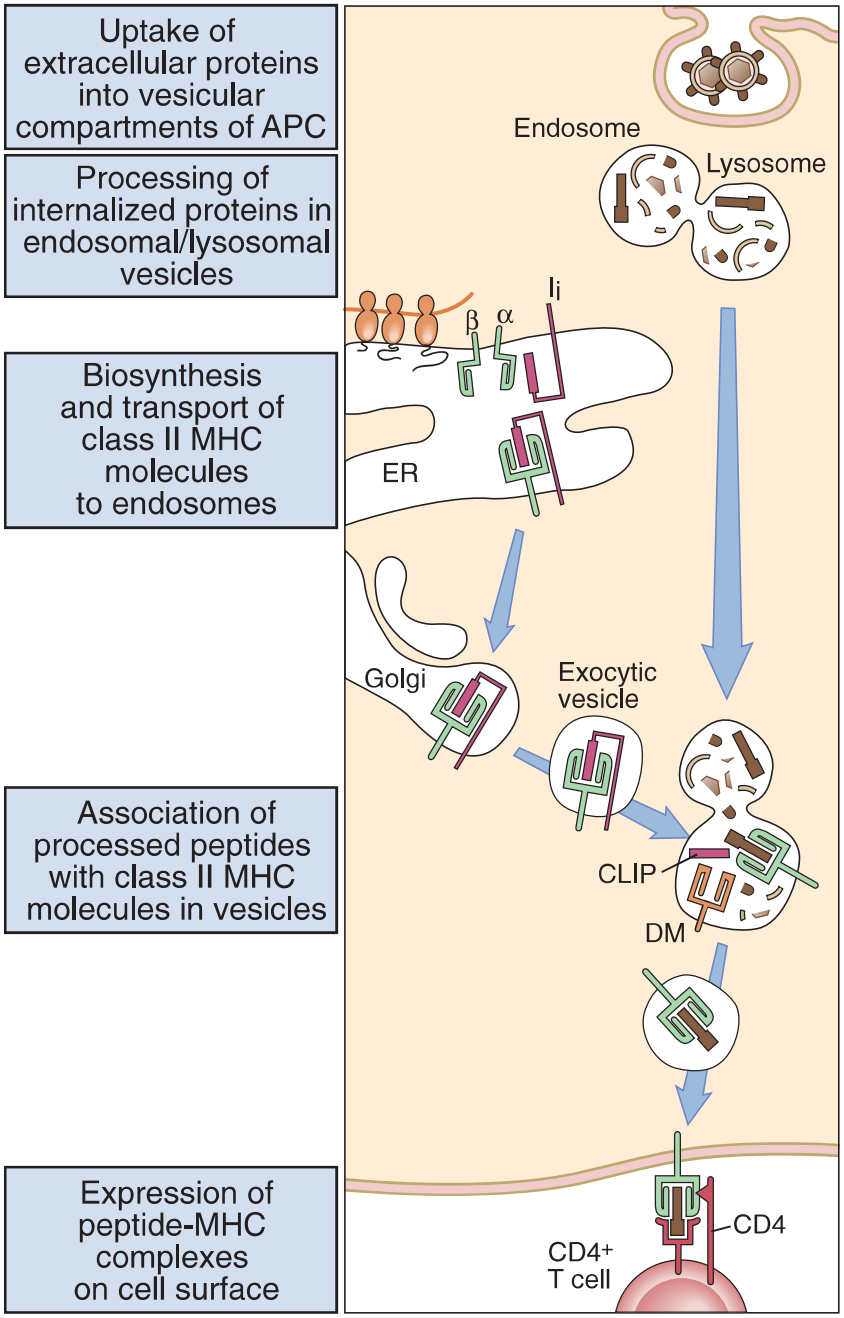


FIGURE 3-13 The class II major histocompatibility complex (MHC) pathway of processing of internalized vesicular antigens. Protein antigens are ingested by antigen-presenting cells (APCs) into vesicles, where they are degraded into peptides. Class II MHC molecules enter the same vesicles and lose the CLIP peptide that occupies the cleft of newly synthesized class II molecules. These class II molecules are able to bind peptides derived from the endocytosed protein. The DM molecule facilitates the removal of CLIP and subsequent binding of the antigenic peptide. The peptide–class II MHC complexes are transported to the cell surface and are recognized by $CD4^+$ T cells. ER, endoplasmic reticulum.

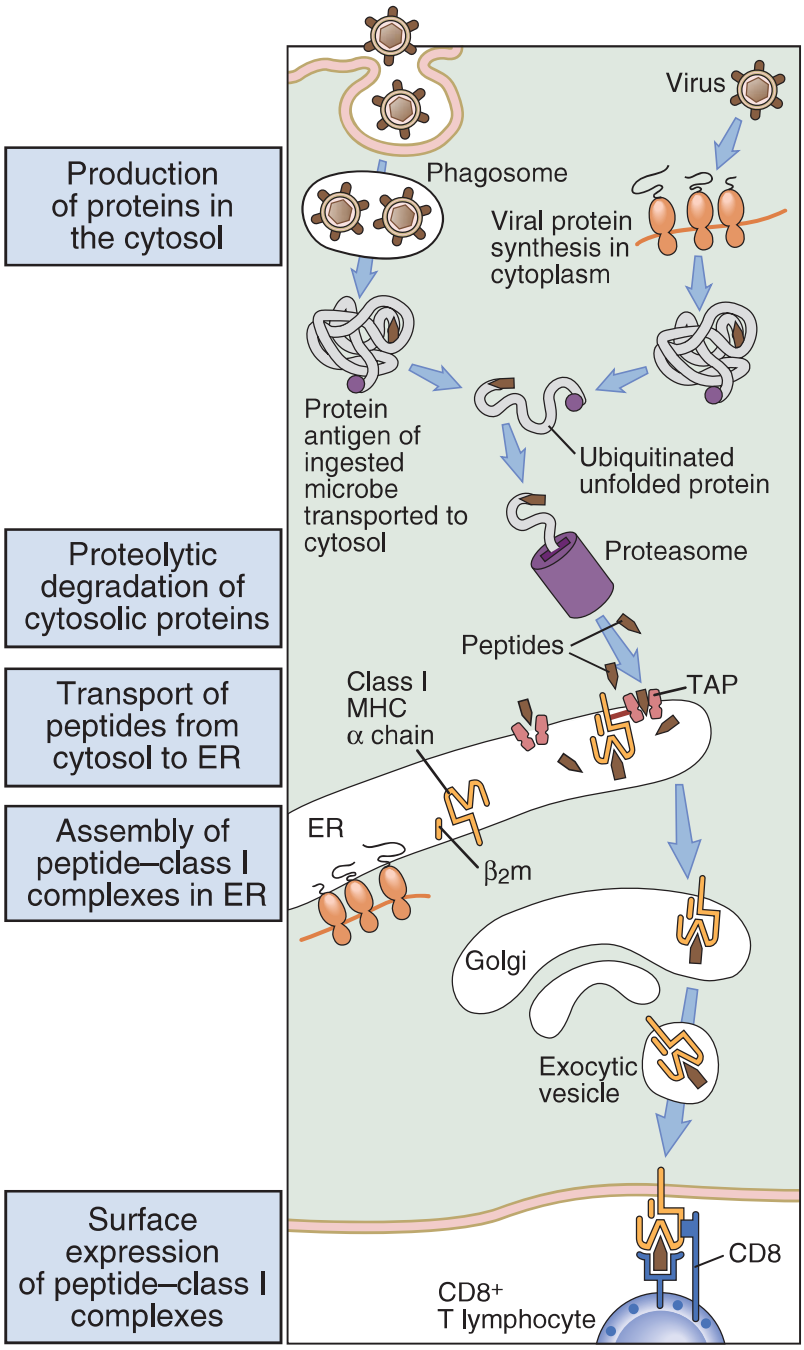


FIGURE 3-14 The class I major histocompatibility complex (MHC) pathway of processing of cytosolic antigens. Proteins enter the cytoplasm of cells either from phagocytosed microbes or from endogenous synthesis by microbes, such as viruses, that reside in the cytoplasm of infected cells. Cytoplasmic proteins are unfolded, ubiquitinated, and degraded in proteasomes. The peptides that are produced are transported by the transporter associated with antigen processing (TAP) into the endoplasmic reticulum (ER), where the peptides may be further trimmed by an ER-resident aminopeptidase and then bind to newly synthesized class I MHC molecules. The peptide–class I MHC complexes are transported to the cell surface and are recognized by $CD8^+$ T cells.

stabilized and transported to the cell surface. During this transport, the class I MHC–peptide complex may intersect endosomes, but now the class I molecule is not available to bind peptides, and, being stable, it is able to resist proteolysis by endosomal proteases. If a class I MHC molecule does not find a peptide in the ER, the molecule becomes unstable and is degraded by proteases.

The co-evolution of microbes and their hosts is well illustrated by the numerous strategies that viruses have developed to block the class I MHC pathway of antigen presentation. These strategies include removing newly synthesized MHC molecules from the ER, inhibiting the transcription of MHC genes, and blocking peptide transport by TAP. By inhibiting the class I MHC pathway, viruses reduce presentation of their own antigens to $CD8^+$ T cells and are thus able to evade the adaptive immune system. These viral evasion strategies are partly counterbalanced by the ability of natural killer cells of the innate immune system to recognize and kill virally infected cells that have lost class I MHC expression (see Chapter 2). We will discuss the mechanisms of immune evasion by viruses in more detail in Chapter 6.

THE PHYSIOLOGIC SIGNIFICANCE OF MAJOR HISTOCOMPATIBILITY COMPLEX-ASSOCIATED ANTIGEN PRESENTATION

It is expected that such a precisely regulated system for protein antigen processing and presentation plays an important role in stimulating immune responses. In fact, many fundamental features of T cell–mediated immunity are closely linked to the peptide display function of MHC molecules.

The restriction of T cell recognition to MHC-associated peptides ensures that T cells see and respond only to cell-associated antigens. This is partly because MHC molecules are cell membrane proteins and partly because peptide loading and subsequent expression of MHC molecules are dependent on intracellular biosynthetic and assembly steps. In other words, MHC molecules can be loaded with peptides only inside cells, where the antigens of phagocytosed and intracellular pathogens are present. Therefore, T lymphocytes can recognize the antigens of only phagocytosed and intracellular microbes, which are the types of microbes that have to be combated by T cell–mediated immunity.

By segregating the class I and class II pathways of antigen processing, the immune system is able to respond to extracellular and intracellular microbes in different ways that are best able to combat these microbes (Fig 3-15). Extracellular microbes are captured by APCs, including B lymphocytes and macrophages, and are presented by class II molecules, which, of course, are expressed mainly on these APCs (and on dendritic cells). Because of the specificity of CD4 for class II, class II–associated peptides are recognized by $CD4^+$ T lymphocytes, which function as helper cells. These helper T cells help B lymphocytes to produce antibodies, and they help phagocytes to destroy ingested microbes, thereby activating the two effector mechanisms best able to eliminate extracellular and ingested microbes. Neither of these mechanisms is effective against viruses and other pathogens that survive and replicate in the cytoplasm of host cells. Cytosolic antigens are processed and displayed by class I MHC molecules, which are expressed on all nucleated cells—again, as expected, because all nucleated cells can be infected with some viruses. Class I–associated peptides are recognized by $CD8^+$ T lymphocytes, which differentiate into CTLs. The CTLs kill the infected cells and eradicate the infection, this being the most effective mechanism for eliminating cytoplasmic microbes. Thus, the nature of the protective immune response to different microbes is optimized by linking several features of antigen presentation and T cell recognition: the pathways of processing of vesicular and cytosolic antigens, the cellular expression of class II and class I MHC molecules, the specificity of CD4 and CD8 co-receptors for class II and class I molecules, and the functions of $CD4^+$ cells as helper cells and of $CD8^+$ cells as CTLs. This function of MHC-associated antigen processing pathways is important because T cells themselves cannot distinguish between extracellular and intracellular microbes. In fact, as mentioned at the beginning of this chapter, the same virus can be extracellular early after infection and becomes intracellular once the infection is established. During its extracellular life, it is combated by antibodies and phagocytes activated by helper T cells, but once the virus has found a haven

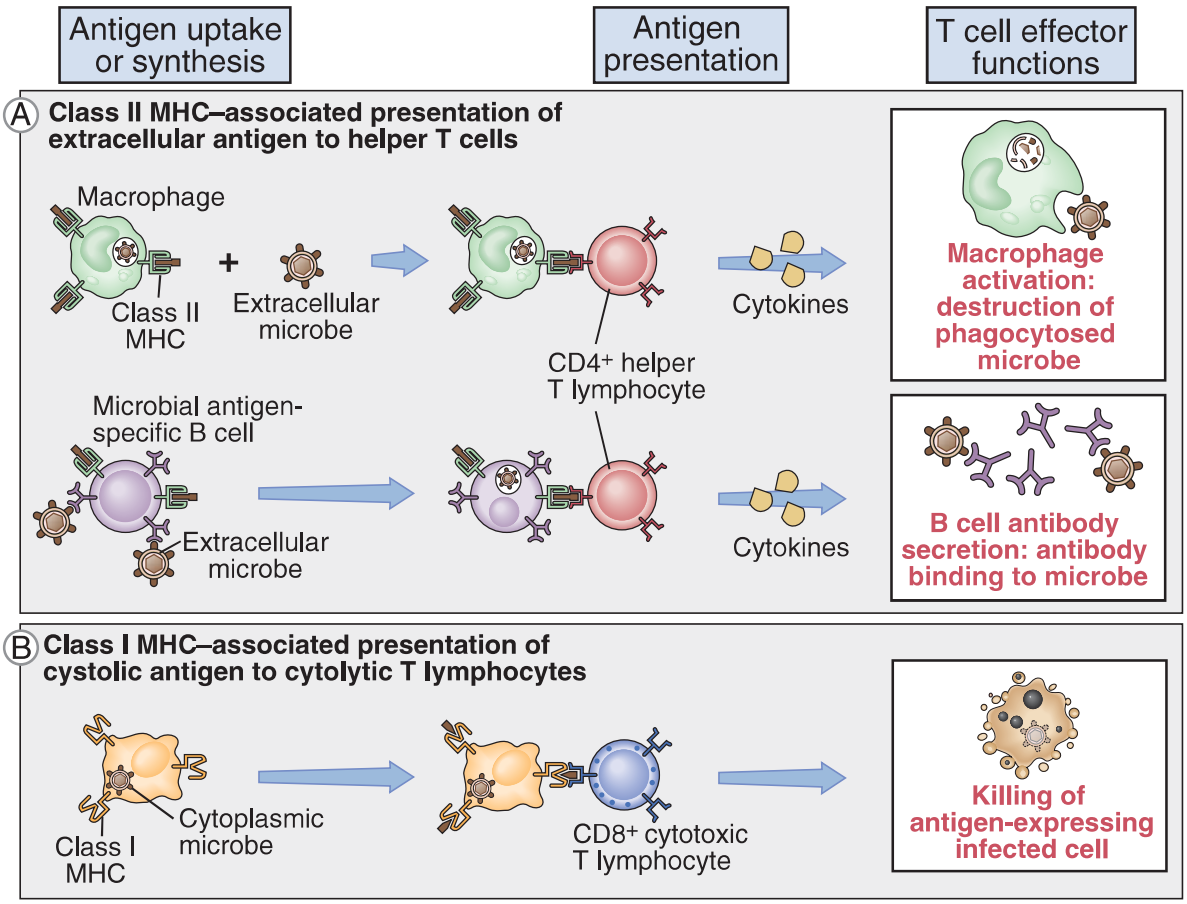


FIGURE 3-15 The role of major histocompatibility complex (MHC)-associated antigen presentation in the recognition of microbes by CD4+ and CD8+ T cells. A, Protein antigens of microbes that are endocytosed from the extracellular environment by macrophages and B lymphocytes enter the class II MHC pathway of antigen processing. As a result, these proteins are recognized by CD4+ helper T lymphocytes, whose functions are to activate macrophages to destroy phagocytosed microbes and activate B cells to produce antibodies against extracellular microbes and toxins. **B,** Protein antigens of microbes that live in the cytoplasm of infected cells enter the class I MHC pathway of antigen processing. As a result, these proteins are recognized by CD8+ cytotoxic T lymphocytes, whose function is to kill infected cells.

in the cytoplasm of cells, it can be eradicated only by CTL-mediated killing of the infected cells. The segregation of class I and class II antigen presentation pathways ensures the correct, specialized immune response against microbes in different locations.

This chapter began with two questions: How do rare antigen-specific lymphocytes find antigens, and how are the appropriate immune responses generated against extracellular and intracellular microbes? Understanding the biology of APCs and the role of MHC molecules in displaying the peptides of protein antigens has provided satisfying answers to both questions, specifically for T cell–mediated immune responses.

FUNCTIONS OF ANTIGEN-PRESENTING CELLS IN ADDITION TO ANTIGEN DISPLAY

APCs not only display peptides for recognition by T cells but, in response to microbes, also express "second signals" for T cell activation. The "two-signal" concept of lymphocyte activation was

introduced in Chapters 1 and 2 (see Fig. 2-16), and we will return to this concept when we discuss the responses of T and B cells (see Chapters 5 and 7). Recall that antigen is the necessary signal 1, and signal 2 is provided by microbes or APCs reacting to microbes. Different types of microbial products and innate immune responses may activate APCs to express molecules that are the second signals for lymphocyte activation. For instance, many bacteria produce a substance called lipopolysaccharide (LPS), also called endotoxin. When the bacteria are captured by APCs for presentation of their protein antigens, LPS acts on the same APCs, via a TLR, and stimulates the expression of costimulators and the secretion of cytokines. The costimulators and cytokines act in concert with antigen recognition by the T cell to stimulate the proliferation and differentiation of the T cells.

Antigens Recognized by B Cells and Other Lymphocytes

B lymphocytes use membrane-bound antibodies to recognize a wide variety of antigens, including proteins, polysaccharides, lipids, and small chemicals. These antigens may be expressed on microbial surfaces (e.g., capsular or envelope antigens) or they may be in soluble form (e.g., secreted toxins). B cells differentiate in response to antigen and other signals into cells that secrete antibodies (see Chapter 7). The secreted antibodies enter the circulation and mucosal fluids and bind to the antigens, leading to their neutralization and elimination. The antigen receptors of B cells and the antibodies that are secreted usually recognize antigens in the native conformation, without any requirement for antigen processing or display by a specialized system. Macrophages in lymphatic sinuses may capture antigens that enter lymph nodes and present the antigens, in intact (unprocessed) form, to B lymphocytes in the follicles. However, it is not known if there is a requirement for a specialized population of APCs to present antigens to naive B cells in order to initiate humoral immune responses.

The B cell–rich lymphoid follicles of the lymph nodes and spleen contain a population of cells called follicular dendritic cells (FDCs), whose function is to display antigens to activated B cells. The antigens that FDCs display are coated with antibodies or by complement byproducts such as C3b and C3d. FDCs use receptors for one end of antibody molecules, called Fc receptors, to bind the antigen-antibody complexes, and receptors for complement proteins, to bind antigens with these proteins attached. These antigens are seen by specific B lymphocytes during humoral immune responses, and they function to select B cells that bind the antigens with high affinity. This process is discussed in Chapter 7.

Although our focus in this chapter has been on peptide recognition by MHC-restricted $CD4^+$ and $CD8^+$ T cells, there are other, smaller populations of T cells that recognize different types of antigens. Natural killer T cells (NK-T cells) are specific for lipids displayed by class I–like CD1 molecules, and $\gamma\delta$ T cells recognize a wide variety of molecules, some displayed by class I–like molecules and others apparently requiring no specific processing or display. The functions of these cells and the significance of their unusual specificities are poorly understood.

SUMMARY

- The induction of immune responses to the protein antigens of microbes is dependent on a specialized system for capturing and displaying these antigens for recognition by the rare naive T cells specific for any antigen. Microbes and microbial antigens that enter the body through epithelia are captured by professional APCs, mainly dendritic cells, located in the epithelia and transported to regional lymph nodes, or are captured by APCs resident in lymph nodes and spleen. The protein antigens of the microbes are displayed by the APCs to naive T lymphocytes that recirculate through the lymphoid organs.
- Molecules encoded in the MHC perform the function of displaying peptides derived from protein antigens.
- Proteins that are ingested by APCs from the extracellular environment are proteolytically degraded within the vesicles of the APCs, and the peptides that are generated bind to the clefts of newly synthesized class II MHC molecules. Class II MHC molecules are recognized by CD4, because of which $CD4^+$ helper T cells are specific for class II MHC–associated peptides that are derived mainly from extracellular proteins.

- Proteins that are produced by microbes living in the cytoplasm of infected cells, or enter the cytoplasm from phagosomes, are degraded by proteasomes, transported into the ER by TAP, and bind to the clefts of newly synthesized class I MHC molecules. Class I MHC molecules are recognized by CD8, because of which $CD8^+$ cytotoxic T lymphocytes are specific for class I MHC–associated peptides derived from cytosolic proteins.
- The role of MHC molecules in antigen display ensures that T cells only see cell-associated protein antigens, and the correct type of T cell (helper or cytotoxic cell) responds to the type of microbe that T cell is best able to combat.
- Microbes activate APCs to express membrane proteins (called costimulators) and to secrete cytokines that provide signals that function in concert with antigens to stimulate specific T cells. The requirement for these second signals ensures that T cells respond to microbial antigens and not to harmless, nonmicrobial substances.
- B lymphocytes recognize proteins as well as nonprotein antigens, even in their native conformations. It is not known if a specialized system of antigen display is essential for the induction of B cell responses. FDCs display antigens to germinal center B cells and select the high-affinity B cells during humoral immune responses.

REVIEW QUESTIONS

1. *When antigens enter through the skin, in what organs are they concentrated? What cell type(s) play important roles in this process of antigen capture?*
2. *What are MHC molecules? What are human MHC molecules called? How were they discovered, and what is their function?*
3. *What are the differences between the antigens that are displayed by class I and class II MHC molecules?*
4. *Describe the sequence of events by which class I and class II MHC molecules acquire antigens for display.*
5. *Which functional subsets of T cells recognize antigens presented by class I and class II MHC molecules? What molecules on T cells contribute to their specificity for either class I or class II MHC–associated peptide antigens?*