Lecture VI

Major histocompatibility complex

T-cell receptor

Mouse H-2 complex

Complex	H-2								Tla	
MHC class	I	II		III		I			I	I
Region	K	IA	IE	S		D			Qa	Tla
Gene products	H-2K	IA αβ	IE αβ	C′ proteins	TNF-α TNF-β	H-2D	H-2L		Qa	Tla, Qa

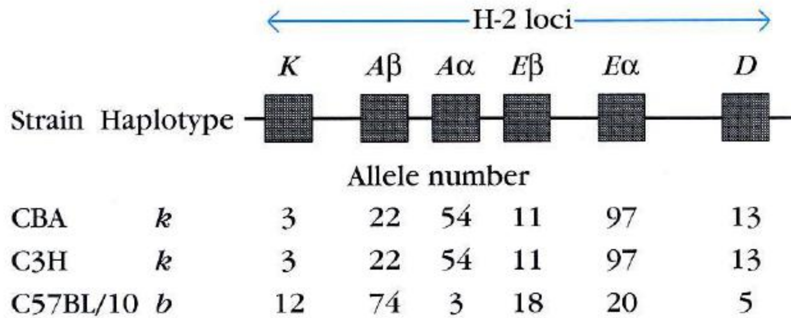
Human HLA complex

Complex	HLA							
MHC class	II			III		I		
Region	DP	DQ	DR	C4, C2, BF		B	C	A
Gene products	DP αβ	DQ αβ	DR αβ	C′ proteins	TNF-α TNF-β	HLA-B	HLA-C	HLA-A

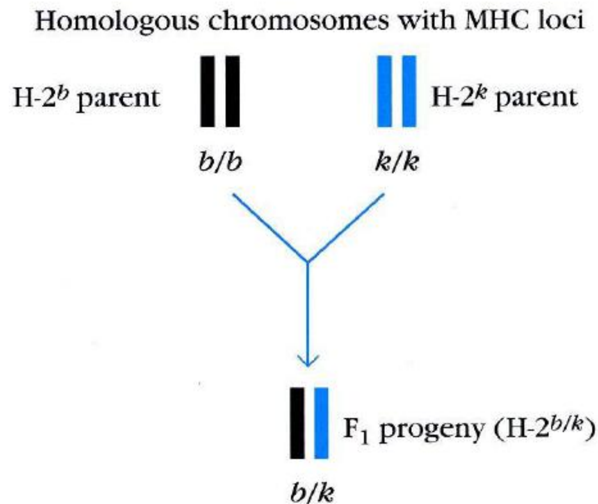
FIGURE 9-1

Simplified organization of the major histocompatibility complex (MHC) in the mouse and human. The MHC is referred to as the H-2 complex in mice and as the HLA complex in humans. In both species the MHC is organized into a number of regions encoding class I (pink), class II (blue), and class III (green) gene products. The class I and class II gene products shown in this figure are considered to be the classical MHC molecules. The class III gene products include complement (C′) proteins and the tumor necrosis factors (TNF-α and TNF-β). The IEβ gene is actually located in the IA region but for pedagogical reasons is shown in the IE region.

(a) Hypothetical allelic composition of mouse MHC haplotypes



(b) Mating of inbred mouse strains with different MHC haplotypes



(c) Mating of outbred mouse strains with different MHC haplotypes

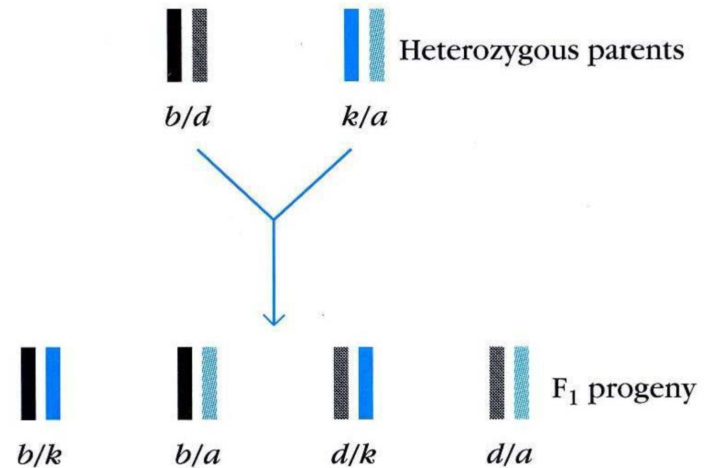


FIGURE 9-2

(a) Illustration of MHC haplotype designation. Strain CBA inherits a particular allele, indicated by an arbitrary number, at each MHC locus. The complete set of alleles is arbitrarily designated as the H-2^k haplotype. Any inbred strain (e.g., C3H) that has the same set of MHC alleles is designated as an H-2^k strain. Strains with a different set of MHC alleles (e.g., C57BL/10) are given a different haplotype designation. (b, c) Because the MHC loci are closely linked and inherited as a set, the MHC haplotype of F_1 progeny from mating of inbred and outbred strains can be predicted easily.

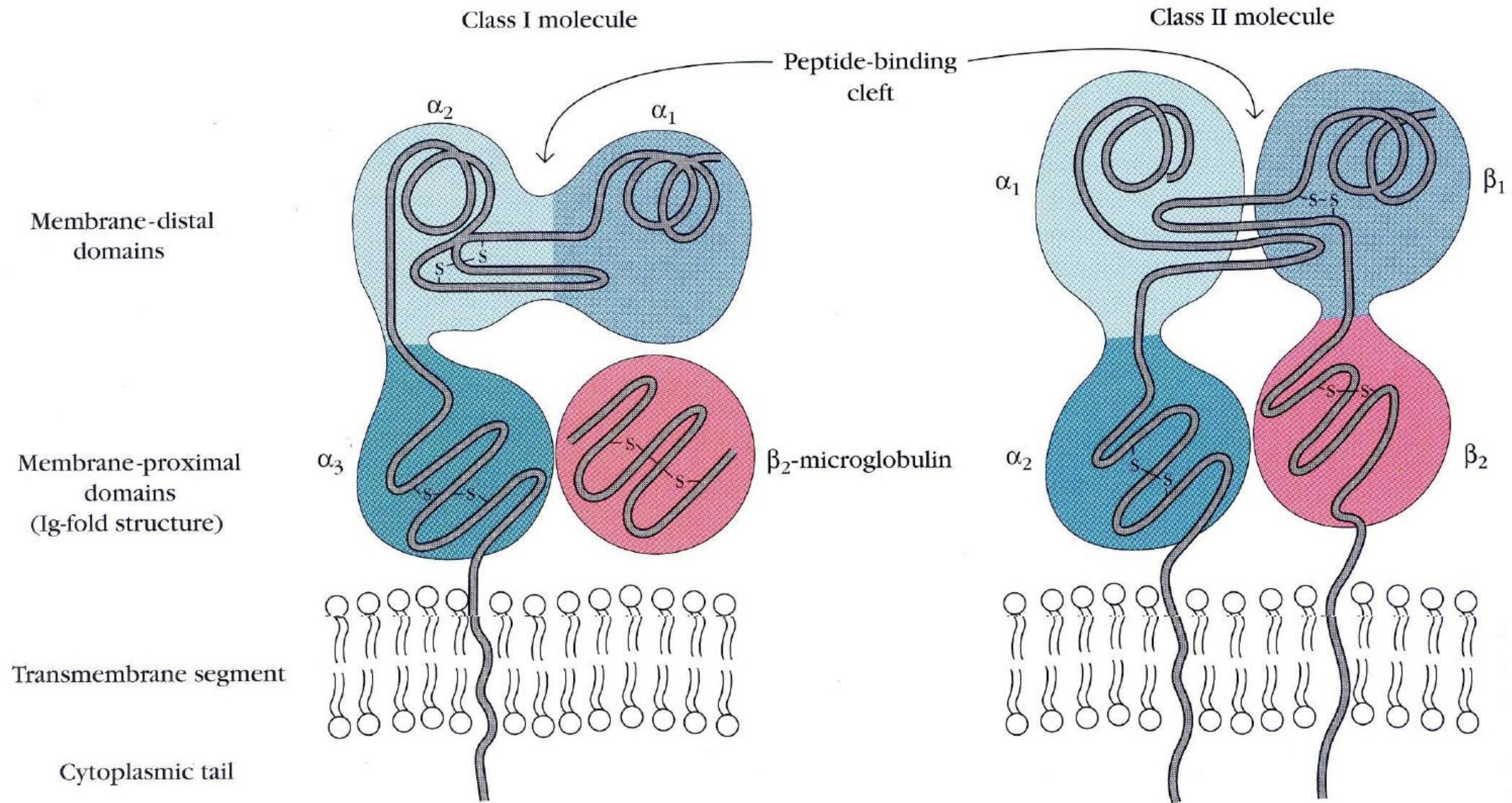
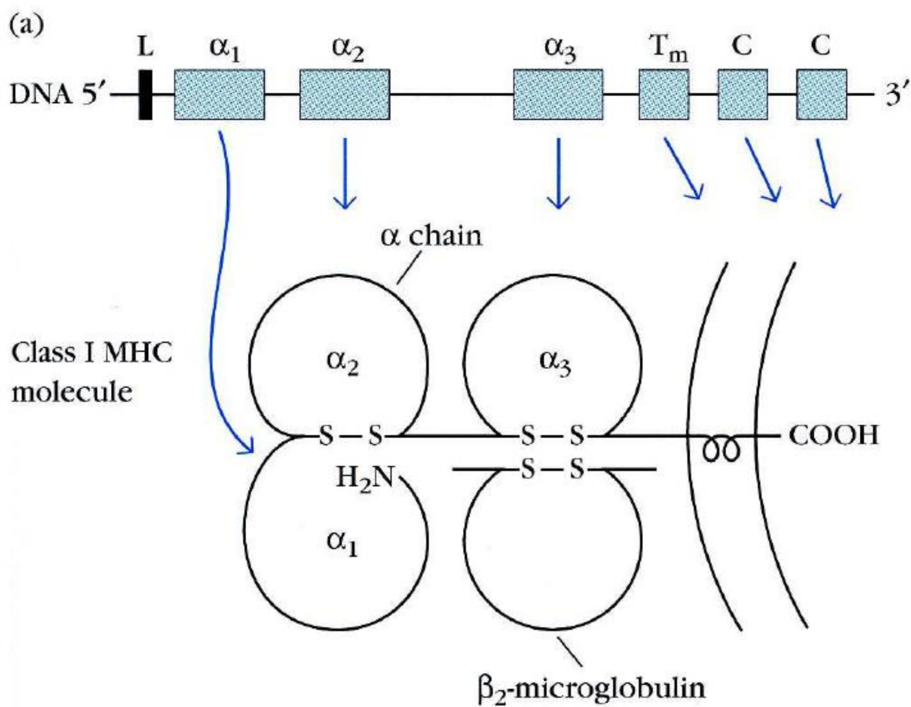


FIGURE 9-5

Schematic diagrams of a class I and class II MHC molecule showing the external domains, transmembrane segment, and cytoplasmic tail. The peptide-binding cleft is formed by the membrane-distal domains in both class I and class II molecules. The membrane-proximal domains possess the basic immunoglobulin-fold structure; thus class I and class II MHC molecules are classified as members of the immunoglobulin superfamily.



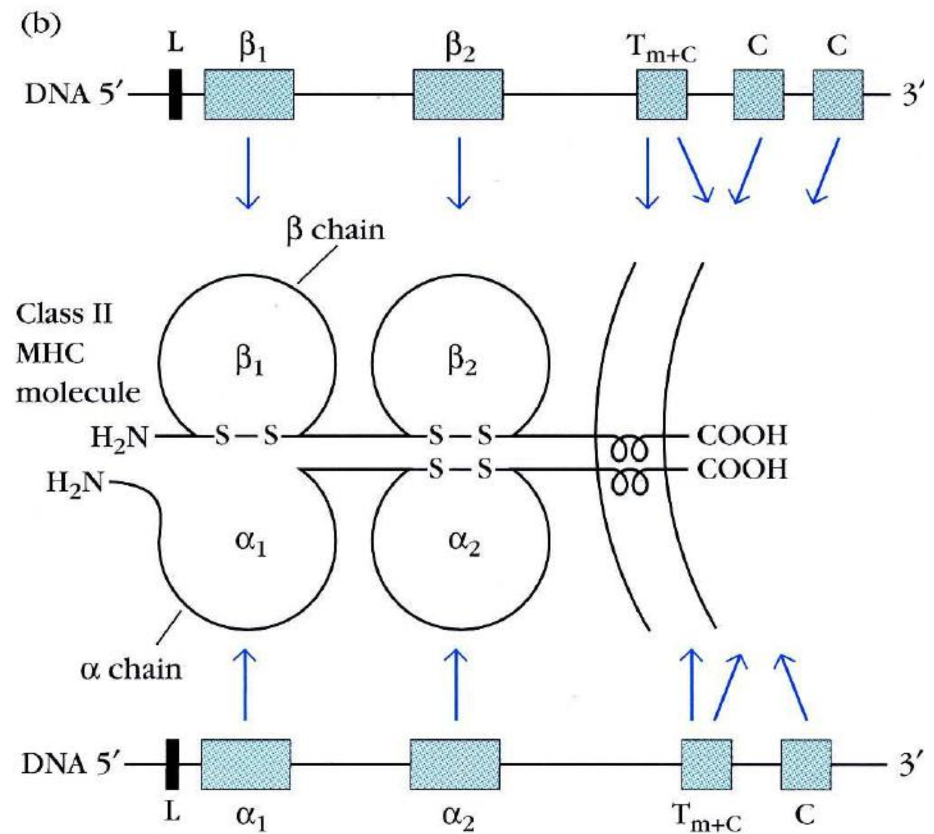


FIGURE 9-9

Schematic diagram of (a) class I and (b) class II MHC genes and molecules showing the correspondence between exons (blue) and the domains in the gene products. Each exon, with the exception of the leader (L) exon, encodes a separate domain of the MHC molecule. The gene encoding β_2-microglobulin is located on a different chromosome. Tm = transmembrane; C = cytoplasmic.

PEPTIDE BINDING BY CLASS I AND CLASS II MHC MOLECULES

	CLASS I MOLECULES	CLASS II MOLECULES
Peptide-binding domain	$\alpha 1/\alpha 2$	$\alpha 1/\beta 1$
Nature of peptide-binding cleft	Closed at both ends	Open at both ends
General size of bound peptides	8–10 amino acids	13–18 amino acids
Peptide motifs involved in binding to MHC molecule	Anchor residues at both ends of peptide; generally hydrophobic carboxyl-terminal anchor	Anchor residues distributed along the length of the peptide
Nature of bound peptide	Extended structure in which both ends interact with MHC cleft but middle arches up away from MHC molecule	Extended structure that is held at a constant elevation above the floor of MHC cleft

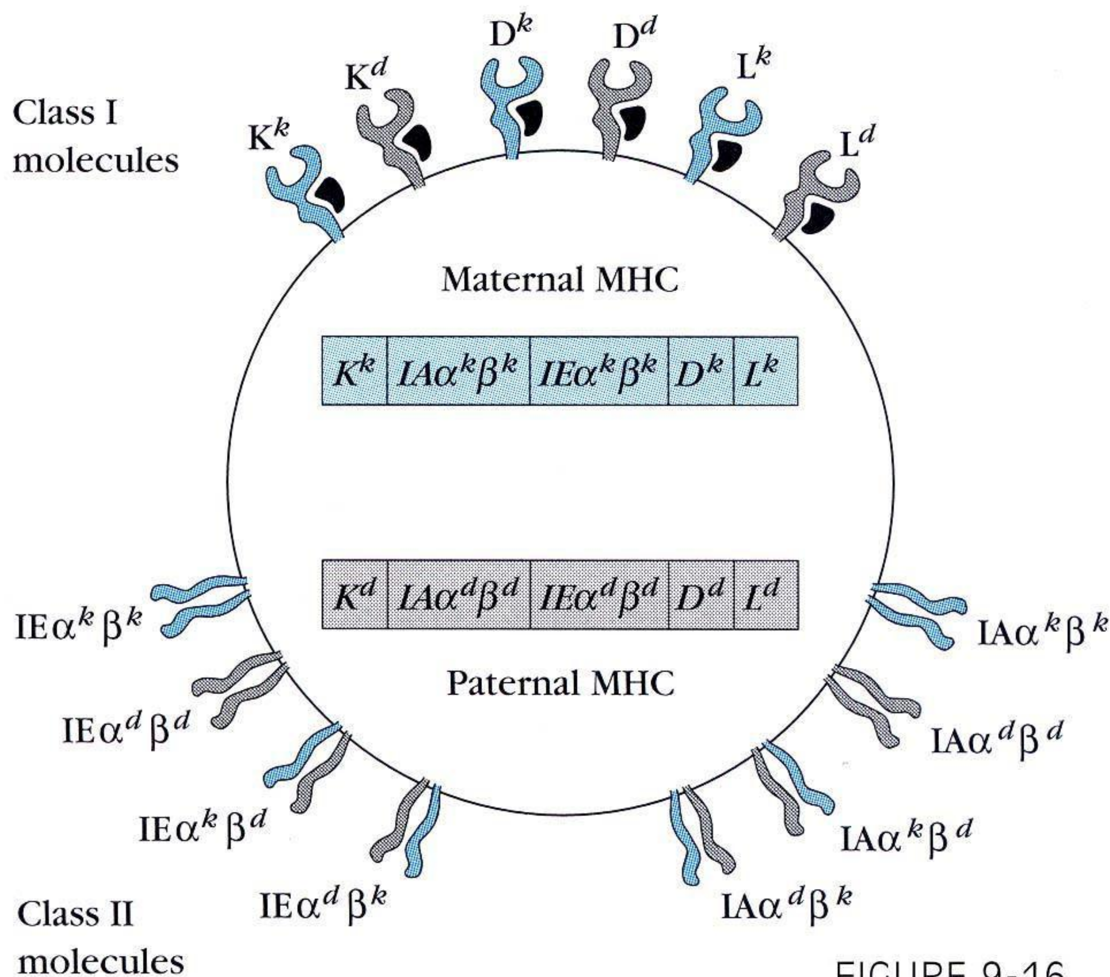


FIGURE 9-16

Diagram illustrating various MHC molecules expressed on antigen-presenting cells of a heterozygous H-$2^{k/d}$ mouse. Both the maternal and paternal MHC genes are expressed. Because the class II molecules are heterodimers, heterologous molecules containing one maternal-derived and one paternal-derived chain are produced. The α_2-microglobulin component of class I molecules (black) is encoded by a gene on a separate chromosome and may be derived from either parent.

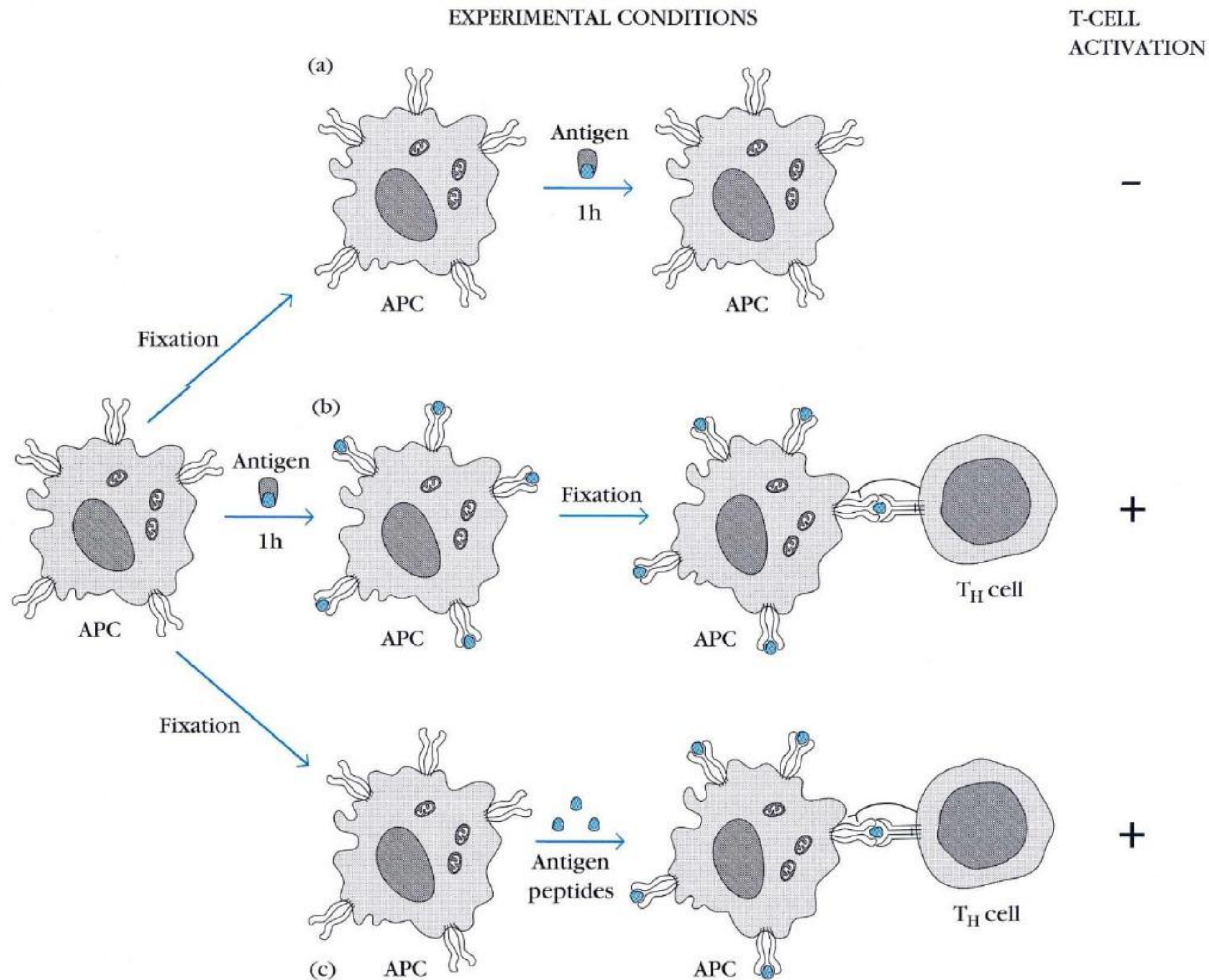


FIGURE 10-3

Experimental demonstration that antigen processing is necessary for T_H-cell activation. (a) When antigen-presenting cells (APCs) are fixed before exposure to antigen, they are unable to activate T_H cells. (b) In contrast, APCs fixed at least 1 h after antigen exposure can activate T_H cells. (c) When APCs are fixed before antigen exposure and incubated with peptide digests of the antigen (rather than native antigen), they also can activate T_H cells. T_H-cell activation is determined by measuring a specific T_H-cell response (e.g., cytokine secretion).

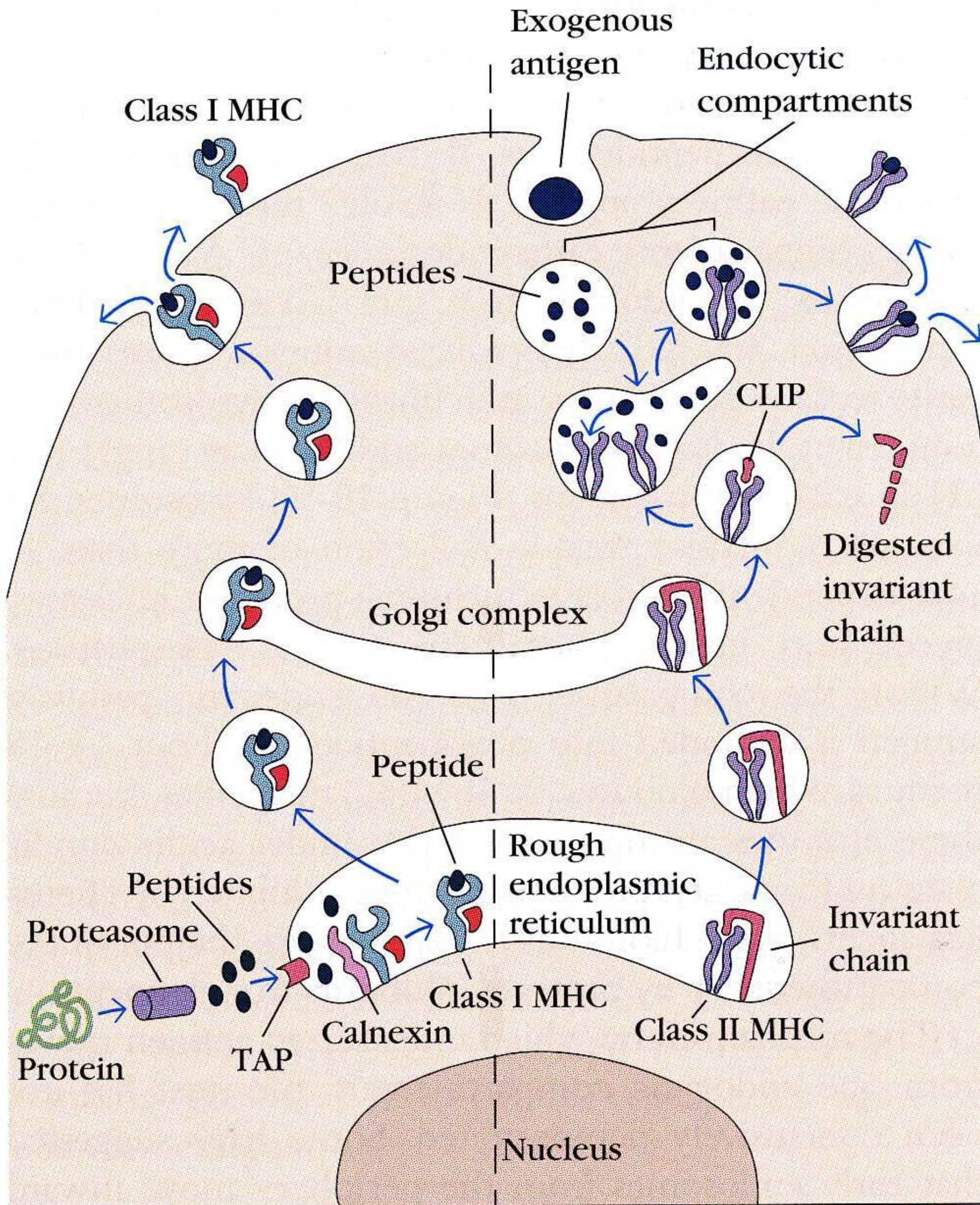
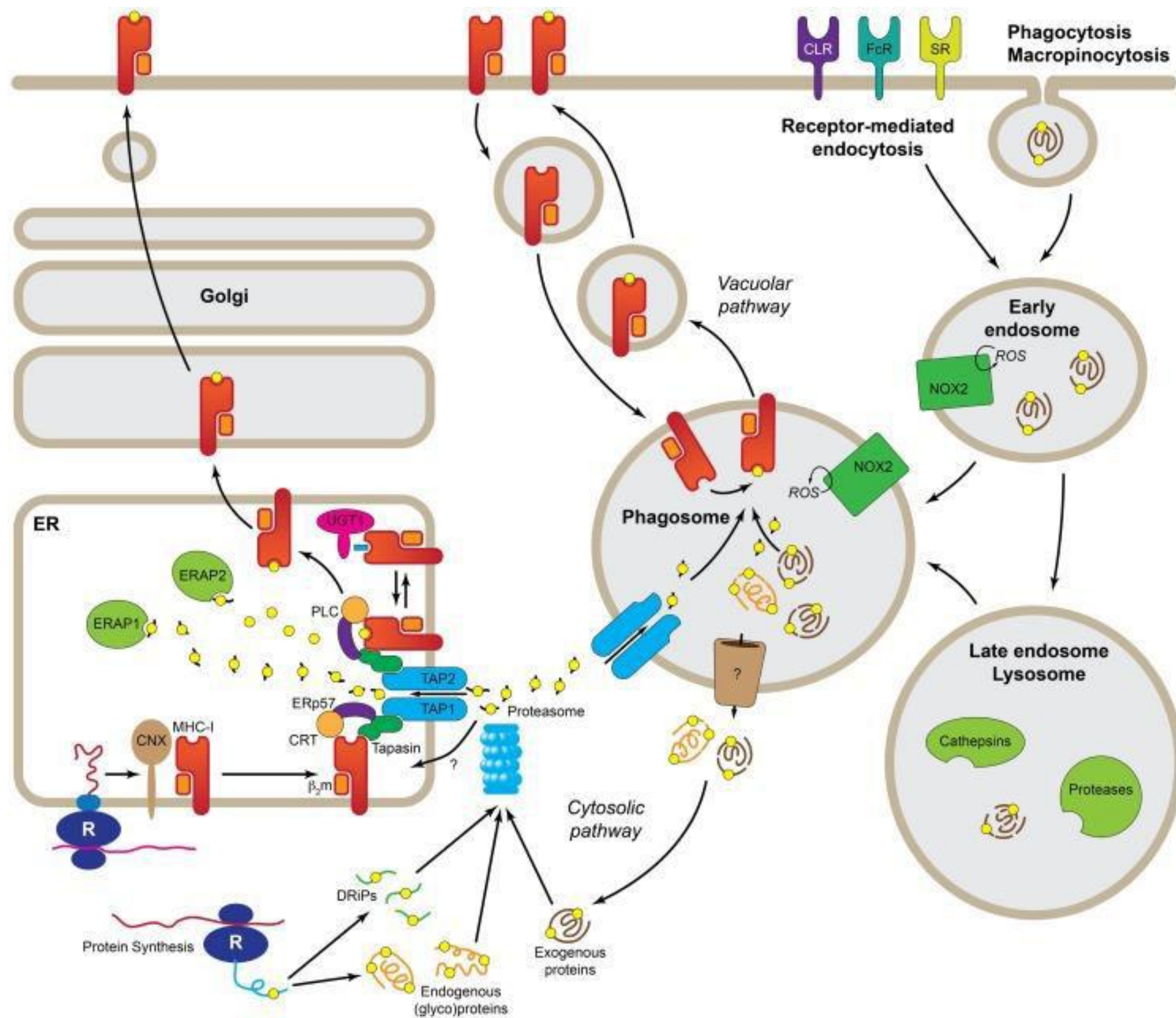


FIGURE 10-8

Model of separate antigen-presenting pathways for endogenous and exogenous antigens. The mode of antigen entry into cells and the site of antigen processing appear to determine whether antigenic peptides associate with class I MHC molecules in the rough endoplasmic reticulum or with class II molecules in endocytic compartments. Some elements of this model have not been experimentally demonstrated.



Molecular pathways leading to cross-presentation in DCs. DCs take up Ag by three general mechanisms, receptor-mediated endocytosis, phagocytosis, or macropinocytosis. Once the Ag reaches the endolysosomal pathway, depending of the specific routing, it may be degraded by the concourse of the mild pH and different types of cathepsins and other proteases. At this point, properly degraded Ag can be directly loaded into recycling MHC-I in the phagosome (Vacuolar pathway). Ag that still needs further processing must be transported to the cytosol (Cytosolic pathway) where it is degraded, together with endogenous proteins and DRiPs, by the proteasome. The peptides generated by the proteasome are transported by TAP or a yet uncharacterized transporter into the ER where they are loaded into MHC-I with the help of the peptide-loading complex. Further trimming in the ER prior to loading, it is possible by the presence of ER-localized endopeptidases (ERAP1 and 2). R, ribosome; CNX, calnexin; CRT, calreticulin; b2m, b2microglobulin; UGT1, UDP-glucose:glycoprotein glucosyltransferase 1; ERAP1/2, ER-aminopeptidases 1/2; PLC, peptide-loading complex; ERp57, protein disulfide isomerase 3; TAP1/2, transporter associated with antigen-presenting 1/2; DRiPs, defective ribosomal products; ROS, reactive oxygen species; NOX2, NADPH oxidase 2; CLR, C-type lectins; FcR, Fc receptors; SR, scavenger receptors.

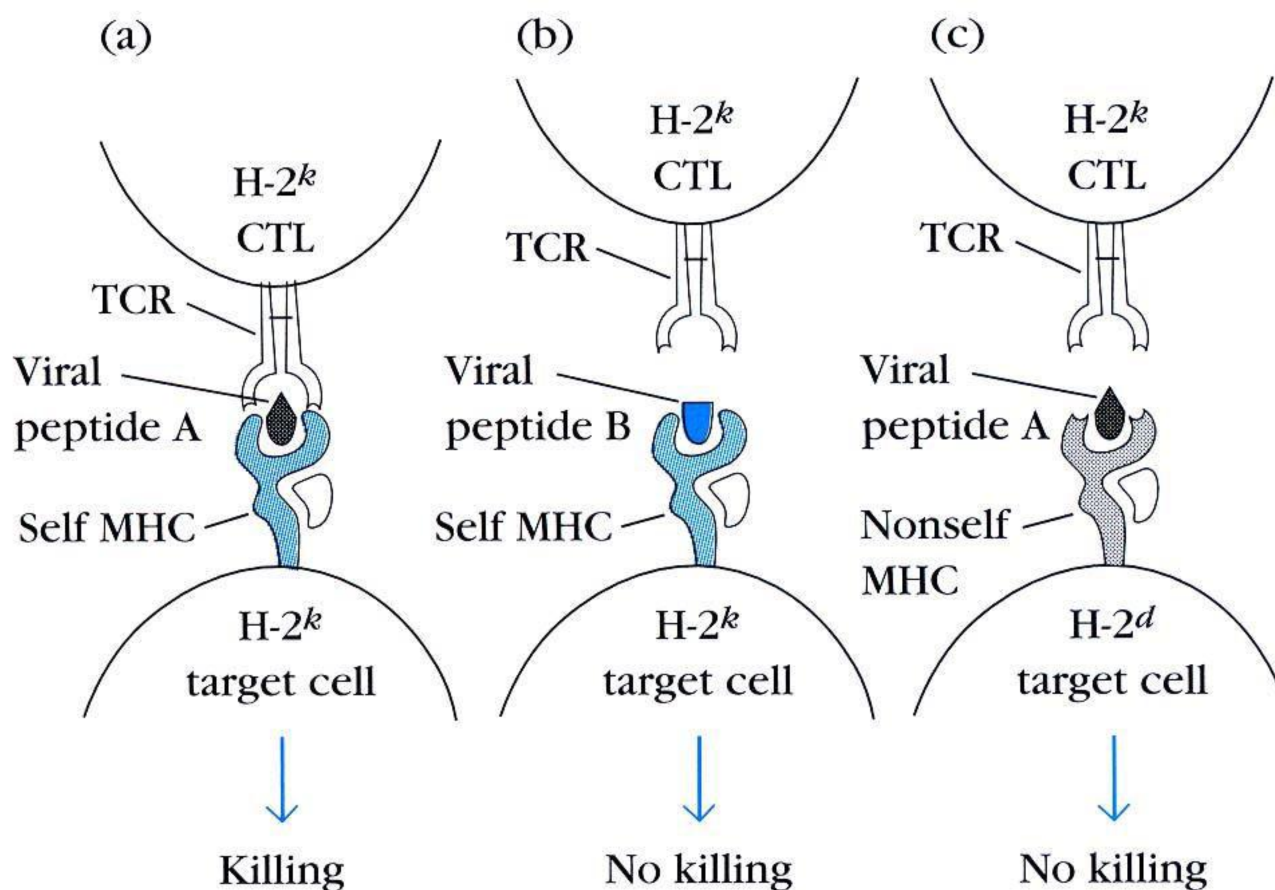


FIGURE 11-1

Self-MHC restriction of the T-cell receptor (TCR). A particular TCR is specific for both an antigenic peptide and a self-MHC molecule. In this example, the H-2^k CTL is specific for viral peptide A presented on an H-2^k target cell (a). Antigen recognition does not occur when peptide B is displayed on an H-2^k target cell (b) nor when peptide A is displayed on an H-2^d target cell (c).

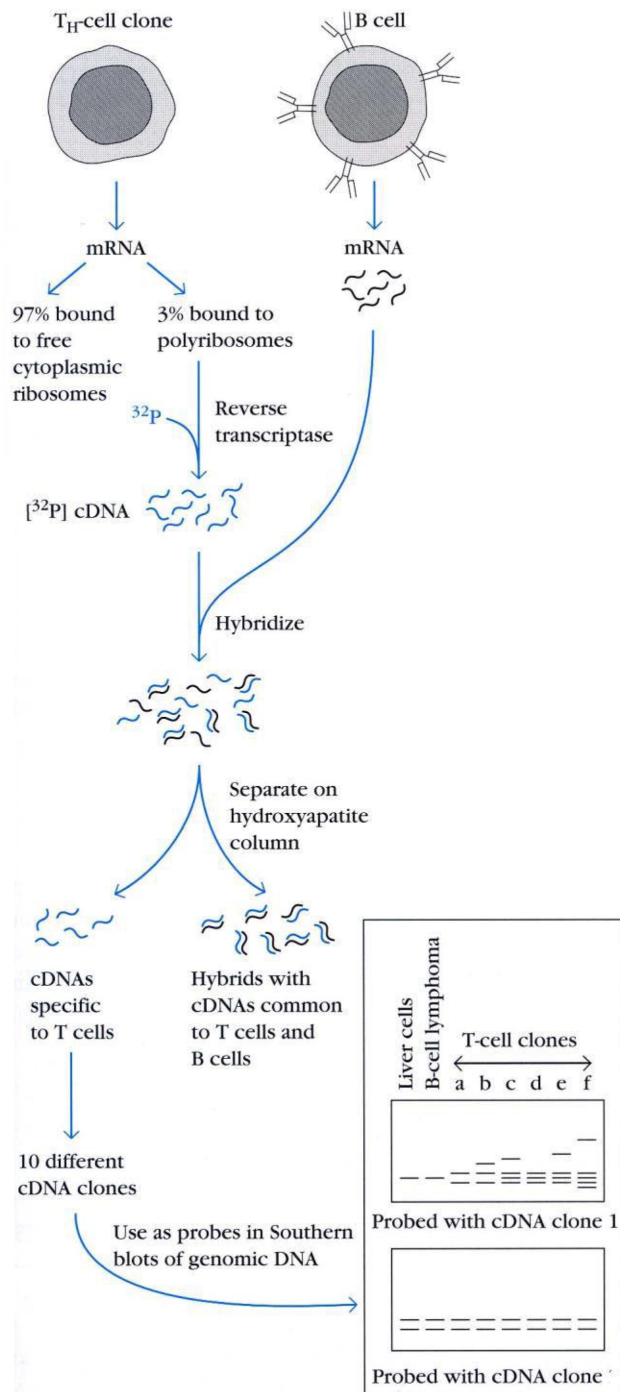
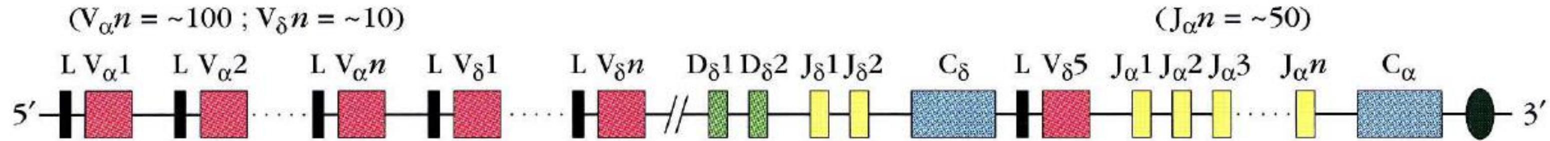


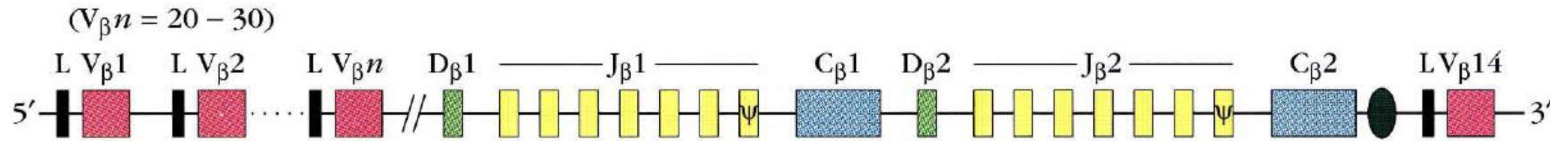
FIGURE 11-4

Production and identification of a cDNA clone encoding the T-cell receptor. The flow chart outlines the procedure used by S. Hedrick and M. Davis to obtain [^{32}P]cDNA clones corresponding to T-cell–specific mRNAs. The technique of DNA subtractive hybridization enabled them to isolate [^{32}P]cDNA unique to the T cell. The labeled T_H-cell cDNA clones were used as probes (*Inset*) in Southern-blot analyses of genomic DNA from liver cells, B-lymphoma cells, and six different T_H-cell clones (a–f). Probing with cDNA clone 1 produced a distinct blot pattern for each T-cell clone, whereas probing with cDNA clone 2 did not. Assuming that liver cells and B cells contained unrearranged germ-line DNA, and that each of the T-cell clones contained different rearranged TCR genes, the results using cDNA clone 1 as the probe identified the T-cell receptor of clone 1. The cDNA of clone 2 identified another T-cell membrane molecule encoded by DNA that does not undergo rearrangement. [Based on S. Hedrick et al., 1984, *Nature* **308**:149.]

Mouse TCR α-chain and δ-chain DNA (chromosome 14)



Mouse TCR β-chain DNA (chromosome 6)



Mouse TCR γ-chain DNA (chromosome 13)

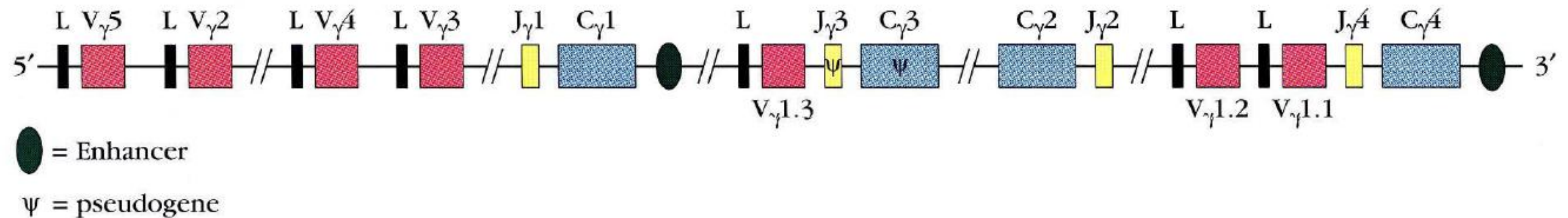


FIGURE 11-5

Germ-line organization of the mouse TCR α-, β-, γ-, and δ-chain gene segments. Each C gene segment is composed of a series of exons and introns, which are not shown. The organization of TCR gene segments in humans is similar, although the number of the various gene segments differs in some cases (see Table 11-1). [Adapted from D. Raulet, 1989, *Annu. Rev. Immunol.* **7**:175 and M. Davis, 1990, *Annu. Rev. Biochem.* **59**:475.]

COMPARISON OF POSSIBLE DIVERSITY IN MOUSE IMMUNOGLOBULIN AND TCR GENES

MECHANISM OF DIVERSITY	IMMUNOGLOBULINS H CHAIN	IMMUNOGLOBULINS κ CHAIN	αβ T-CELL RECEPTOR α CHAIN	αβ T-CELL RECEPTOR β CHAIN	γδ T-CELL RECEPTOR γ CHAIN	γδ T-CELL RECEPTOR δ CHAIN
ESTIMATED NUMBER OF SEGMENTS						
Multiple germ-line gene segments						
V	300	300	100	25	7	10
D	12	0	0	2	0	2
J	4	4	50	12	3	2
POSSIBLE NUMBER OF COMBINATIONS*						
Combinatorial V-J and V-D-J joining	$300 \times 12 \times 4 = 1.4 \times 10^4$	$300 \times 4 = 1.2 \times 10^3$	$100 \times 50 = 5 \times 10^3$	$25 \times 2 \times 12 = 6 \times 10^2$	$7 \times 3 = 21$	$10 \times 2 \times 2 = 40$
Alternative joining of D gene segments	–	–	–	+ (some)	–	+ (often)
Junctional flexibility	+	+	+	+	+	+
N-region nucleotide addition †	+	–	+	+	+	+
P-region nucleotide addition	+	+	+	+	+	+
Somatic mutation	+	+	–	–	–	–
Total estimated diversity ‡	**~ 10^{11}**		**~ 10^{15}**		**~ 10^{18}**	

* A plus sign (+) indicates mechanism makes a significant contribution to diversity but to an unknown extent. A minus sign (–) indicates mechanism does not operate.

† See Figure 11-8d for theoretical number of combinations generated by N-region addition.

‡ Total estimated diversity includes contribution from combinatorial association of chains.

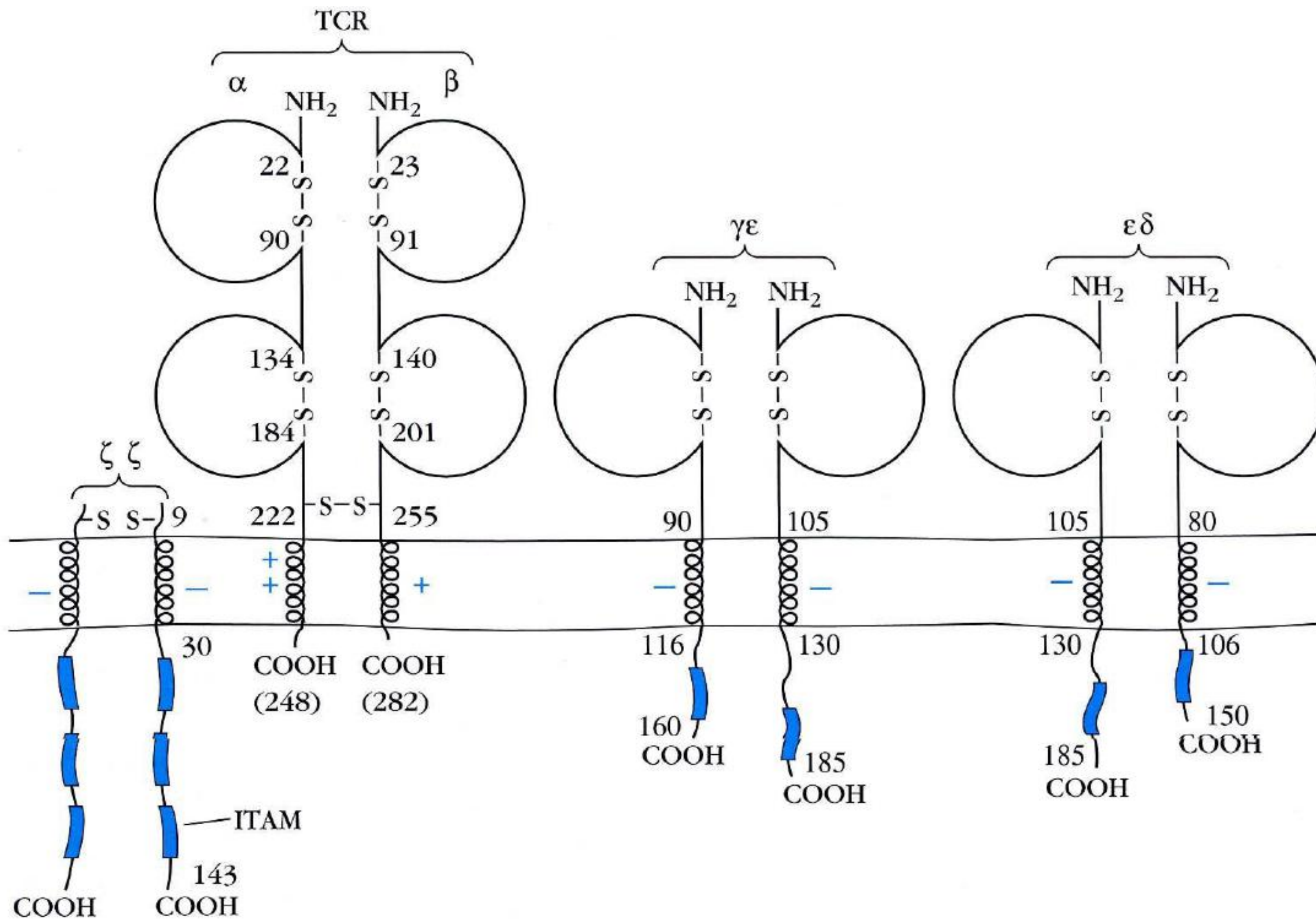


FIGURE 11-9

Schematic diagram of the TCR-CD3 complex, which constitutes the T-cell antigen-binding receptor. The figure shows the $\alpha\beta$ T-cell receptor and CD3 complex consisting of the $\zeta\zeta$ homodimer plus $\gamma\varepsilon$ and $\delta\varepsilon$ heterodimers. The external domains of the γ, δ, and ε chains of CD3 are similar to the immunoglobulin-fold structure, which may facilitate their interaction with the T-cell receptor and each other. Ionic interactions also may occur between the oppositely charged transmembrane regions in the TCR and CD3 chains. The long cytoplasmic tails of the CD3 chains contain a common sequence, the immunoreceptor tyrosine-based activation motif (ITAM), which functions in signal transduction.

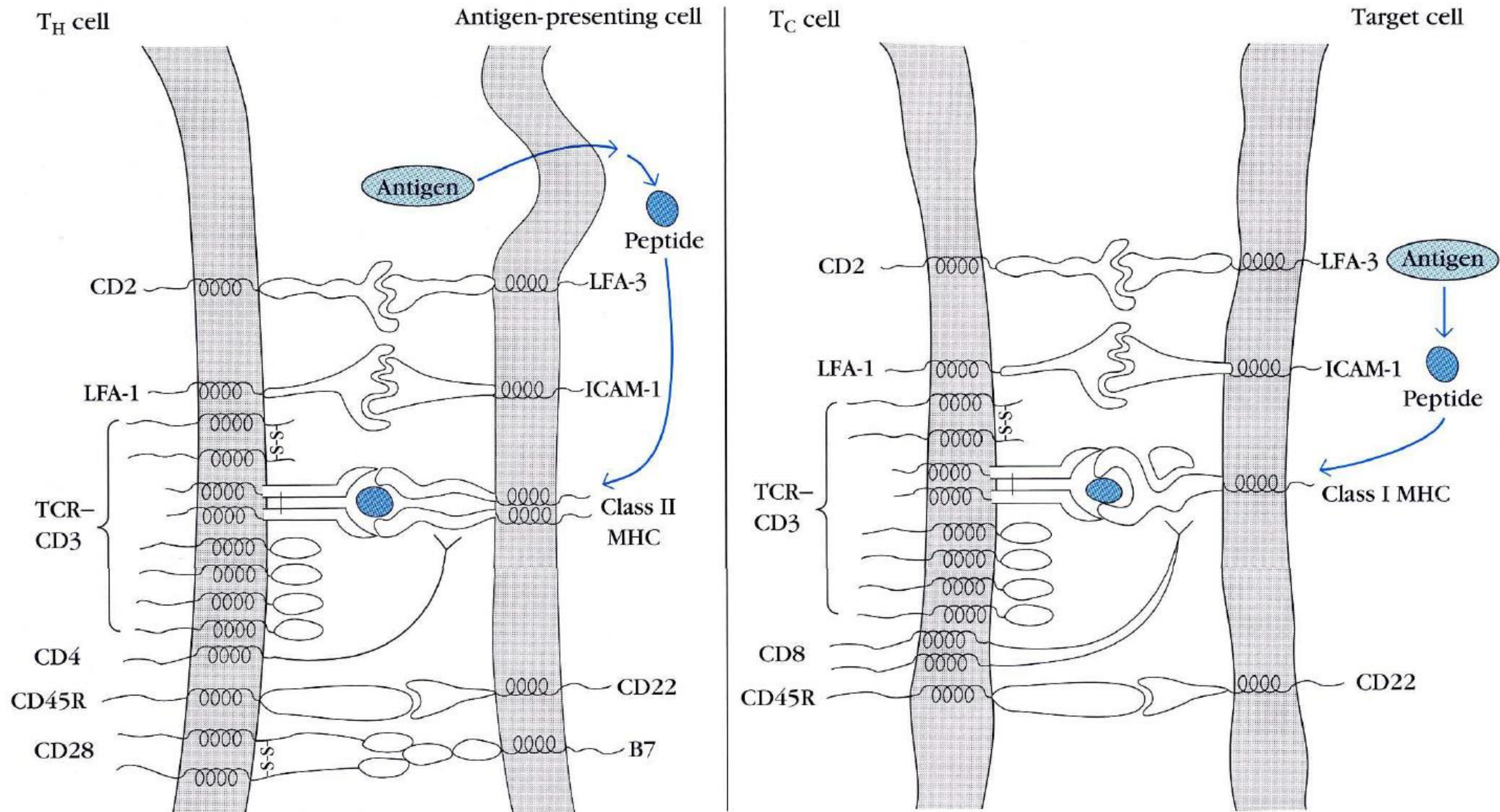


FIGURE 11-14

Schematic diagram of the interactions between the T-cell receptor and various accessory molecules with their ligands on an antigen-presenting cell (*left*) or target cell (*right*). Binding of the coreceptors, CD4 and CD8, and the other accessory molecules to their ligands strengthens the association between the interacting cells and/or aids in the signal transduction leading to T-cell activation.

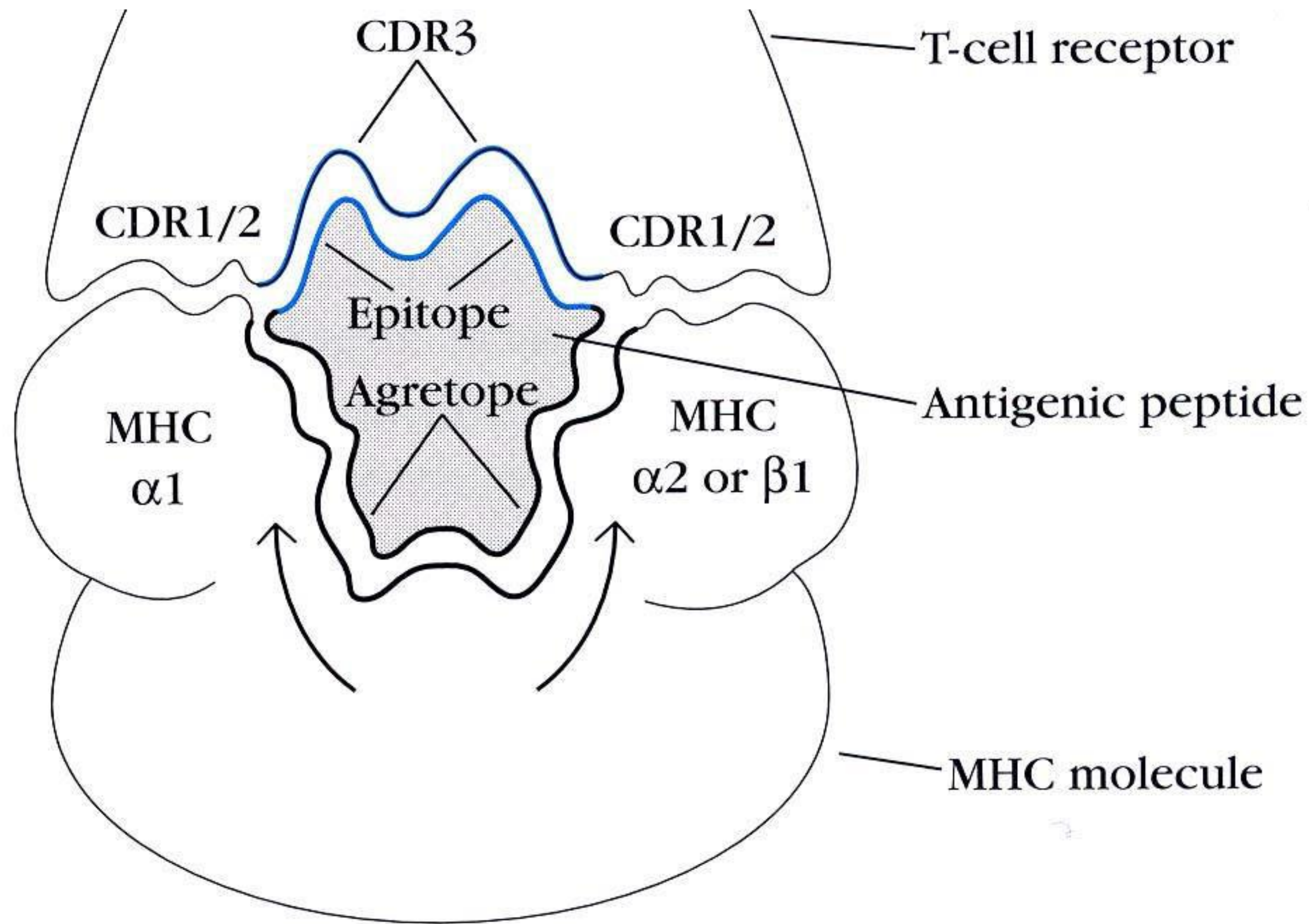


FIGURE 11-16

Schematic diagram showing the various sites in a T-cell receptor, antigenic peptide, and MHC molecule that interact in the TCR-peptide-MHC trimolecular complex. [Adapted from J. McCluskey et al., 1992, in *Antigen Processing and Recognition*, CRC Press.]

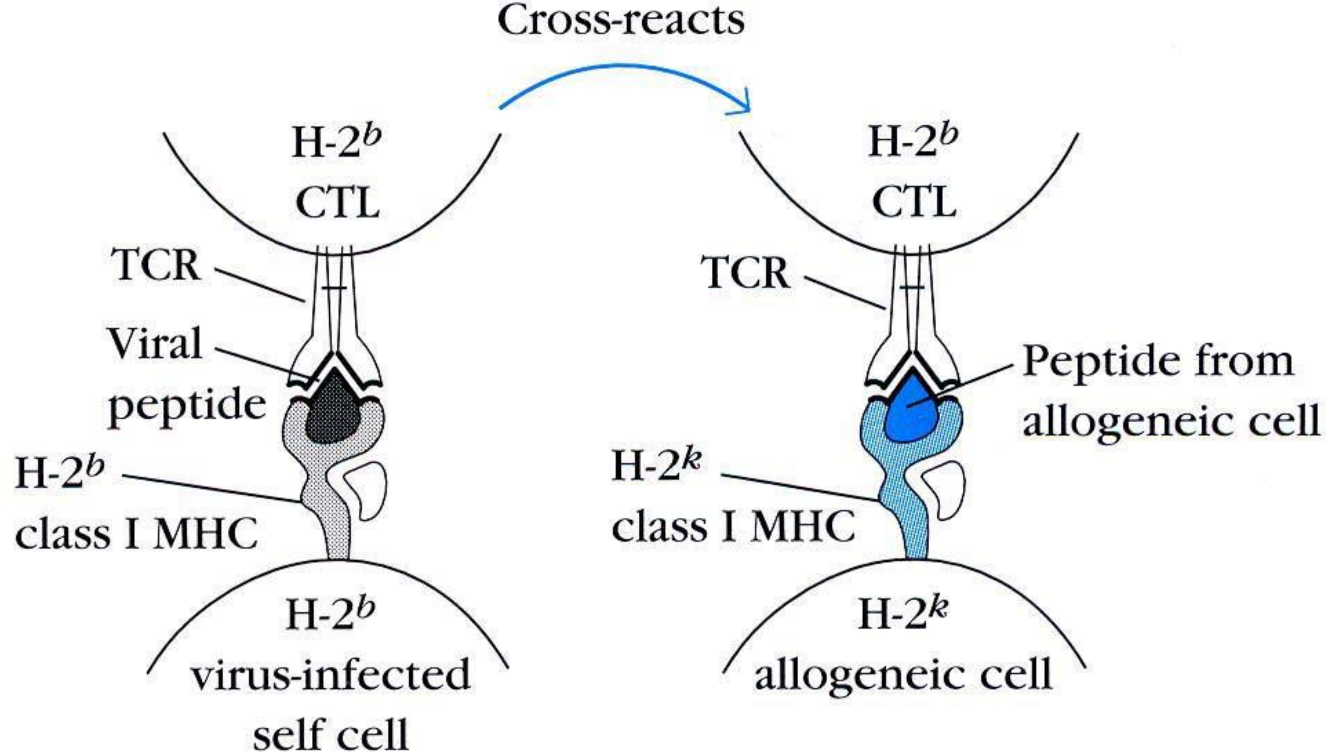


FIGURE 11-17

Possible mechanism of T-cell cross-reactivity that would explain the observed high incidence of alloreactive T cells. As schematically illustrated, a T-cell receptor specific for an H-2^b class I MHC molecule plus viral peptide (*left*) cross-reacts with an allogeneic H-2^k class I molecule plus an allogeneic peptide (*right*). According to this model, the conformation of the allogeneic peptide–MHC complex and foreign peptide–self-MHC complex is sufficiently similar that the same TCR recognizes both complexes.