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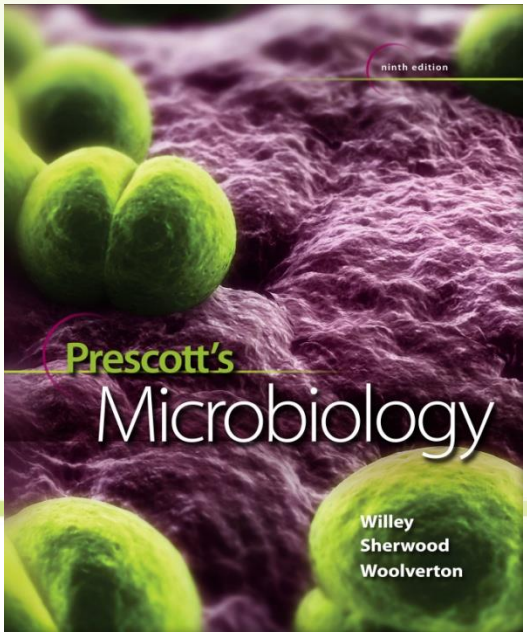


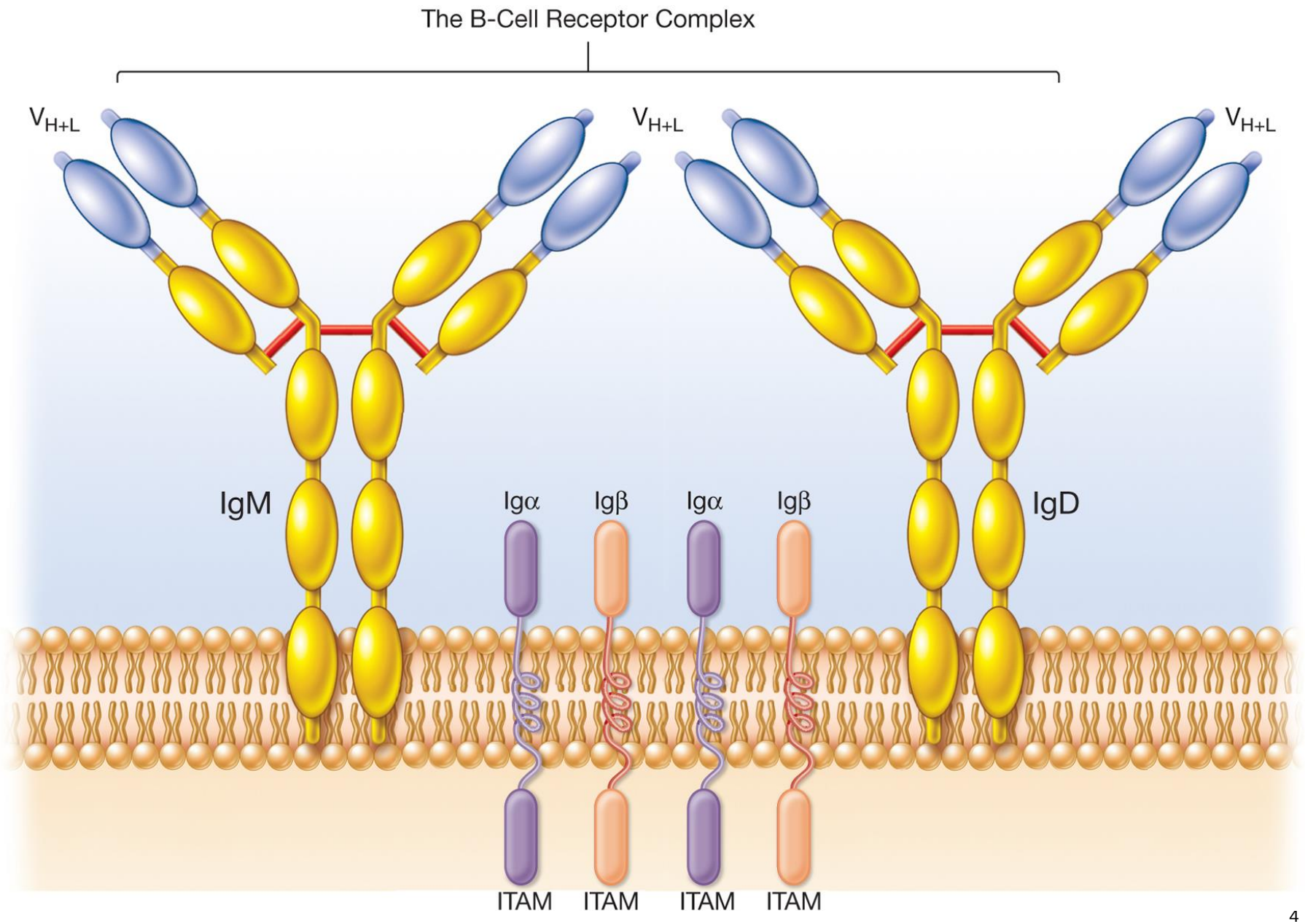
Adaptive Immunity

34.6 B-cell Biology

1. Describe the B-cell receptor structure and function
2. Illustrate the B-cell maturation process in response to antigen triggering
3. Compare T-dependent and T-independent B-cell activation
4. Build a model of the molecular events resulting in B-cell activation

B-Cell Biology

- B cells must be activated by a specific antigen to continue mitosis
  - cells then replicate and differentiate into ***plasma cells*** which secrete antibodies
- B cells have immunoglobulin receptors for the specific antigen that will activate that particular B cell
  - these receptors associate with other proteins and are called B-cell receptors (BCRs)
- Interaction with that antigen is communicated to the nucleus via a signal transduction pathway similar to that described for T cells

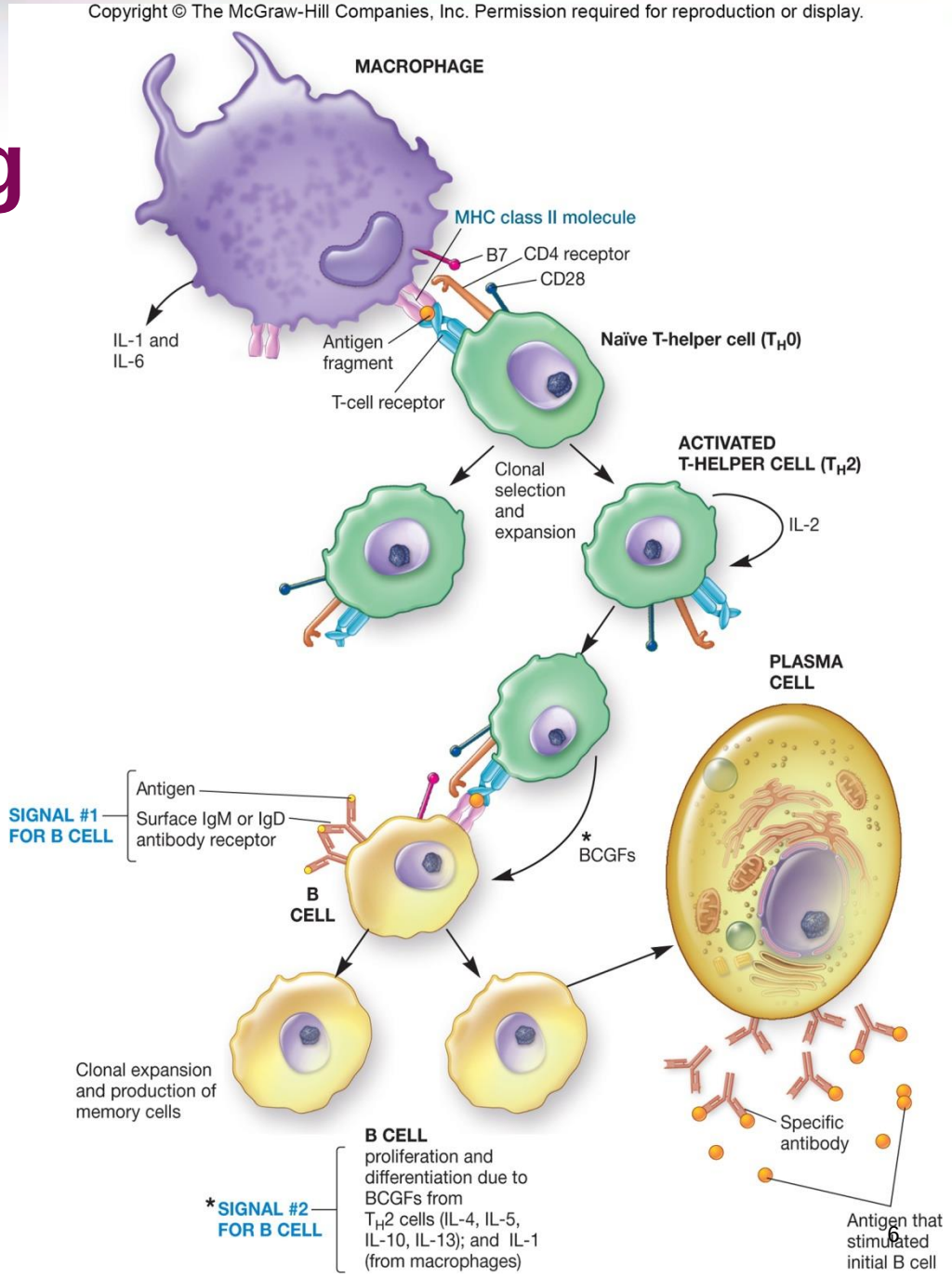


B-Cell Activation

- Leads to proliferation and differentiation into plasma cells
  - some cytokines produced by helper T cells can act on B cells and assist in growth and differentiation
- Typically antigen-specific
- Two mechanisms for antigen-specific activation
  - T dependent
  - T independent

T-Dependent Antigen Triggering

- Like T cells, require two signals
  - antigen-BCR specific interaction
  - activated T helper 2 binds B cell presented antigen and secretes B cell growth factors
- B cell differentiates into plasma cell and memory cell



T-Independent Antigen Triggering

- T-independent antigens
  - polymeric antigens with large number of identical epitopes (e.g., bacterial lipopolysaccharides)
- Less effective than T-dependent B cell activation
  - antibodies produced have a low affinity for antigen
  - no memory B cells formed

34.7 Antibodies

1. Describe the structure of the B-cell receptor that is secreted as antibody
2. Compare and contrast the five classes of antibody
3. Diagram the antibody changes, induced by antigen binding, that facilitate antigen capture and removal from the host
4. Integrate antibody secretion with antigen exposure
5. Create a model of genetic diversity that results from recombination, alternative splicing, and somatic hypermutation
6. Predict antibody specificity resulting from clonal selection

Antibodies

- Antibody
  - immunoglobulin (Ig)
  - glycoprotein made by activated B cells (plasma cells)
  - serves as antigen receptor (BCR) on B cell surface
- Found in blood serum, tissue fluids, and mucosal surfaces of vertebrate animals
  - an antibody can recognize and bind antigen that caused its production

Table 34.2 Physicochemical Properties of Human Immunoglobulin Classes

	IMMUNOGLOBULIN CLASSES				
Property	**IgG[1]**	**IgM**	**IgA[2]**	**IgD**	**IgE**
Heavy chain	γ_1	μ	α_1	δ	ε
Mean serum concentration (mg/ml)	9	1.5	3.0	0.03	0.00005
Percent of total serum antibody	80–85	5–10	5–15	<1	<1
Valency	2	5(10)	2(4)	2	2
Mass of entire molecule (kDa)[3]	146	970	160[3]	184	188
Placental transfer	+	−	−	−	−
Half-life in serum (days)[4]	23	5	6	3	2
Complement activation					
Classical pathway	++	+++	−	−	−
Alternative pathway	−	−	+	−	−
Induction of mast cell degranulation	−	−	−	−	+
% carbohydrate	3	7–10	7	12	11
Major characteristics	Most abundant Ig in body fluids; neutralizes toxins; opsonizes bacteria	First to appear after antigen stimulation; very effective agglutinator; expressed as membrane-bound antibody on B cells	Secretory antibody; protects mucous membranes	Present on B-cell surface; B-cell recognition of antigen	Anaphylactic-mediating antibody; resistance to helminths

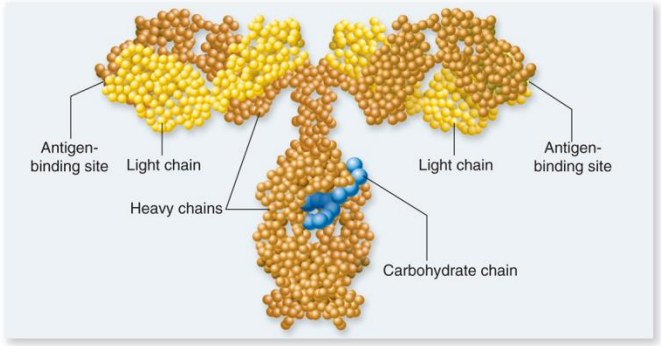
1 Properties of IgG subclass 1.
2 Properties of IgA subclass 1.
3 sIgA = 360 − 400 kDa.
4 Time required for half of the antibodies to disappear.

Immunoglobulin Structure

- All immunoglobulin molecules have the same basic structure
  - four polypeptide chains
    - two identical heavy chains
    - two identical light chains
    - heavy and light chains connected to each other by disulfide bonds
  - both chains contain two different regions
    - constant (C) regions (CL and CH)
    - variable (V) regions (VL and VH)

Immunoglobulin Structure

- Four chains are arranged in form of a flexible Y with a hinge region
  - stalk of Y is the crystallizable fragment (Fc)
    - composed of only constant region
  - top of Y is two antigen binding fragments (Fab)
    - composed of both constant and variable regions



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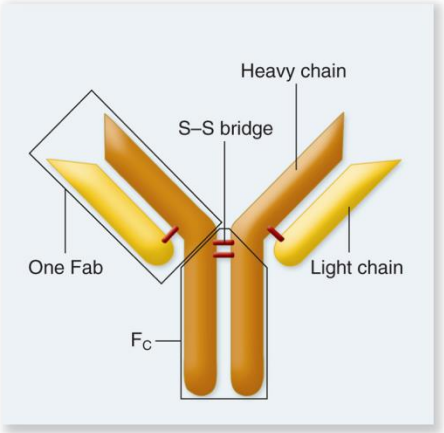

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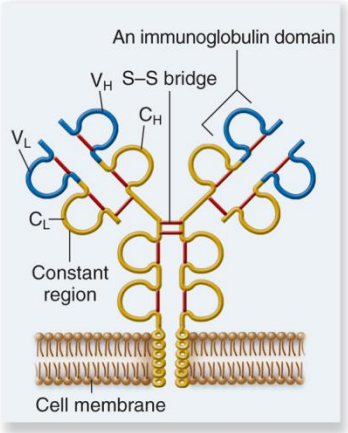

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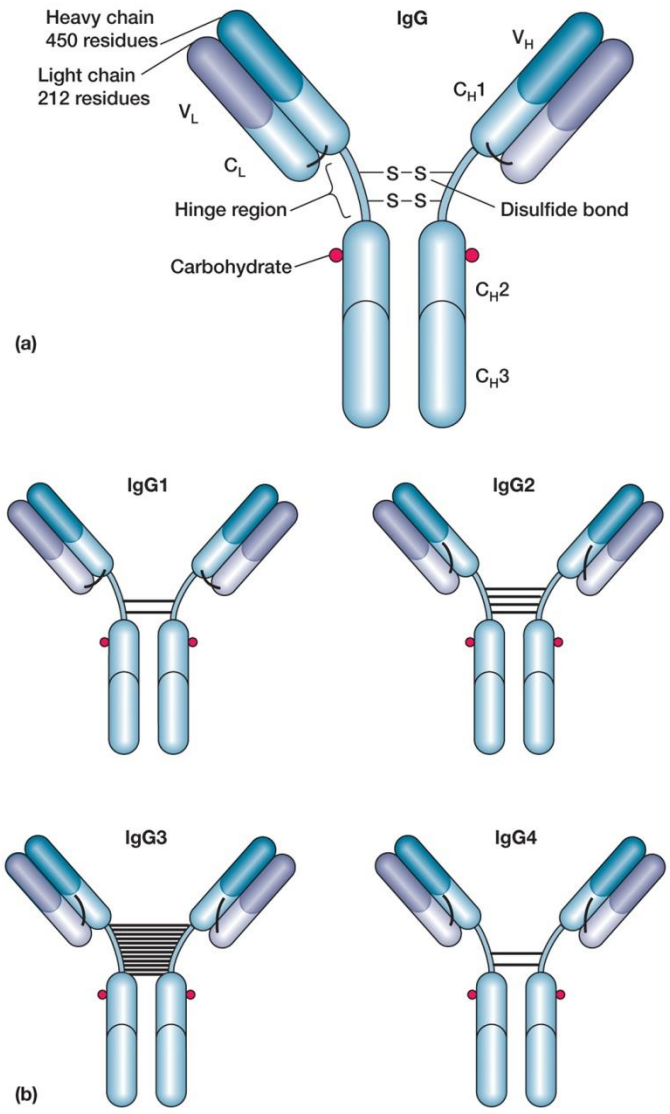
Immunoglobulin Function

- Fab binds antigen specifically
  - marks antigen for immunological attack
  - activates nonspecific defense mechanisms that can destroy antigen
    - e.g., opsonization for enhanced phagocytosis
- Fc mediates binding to:
  - host tissue
  - various cells of immune system
  - first component of complement system

Immunoglobulin Classes



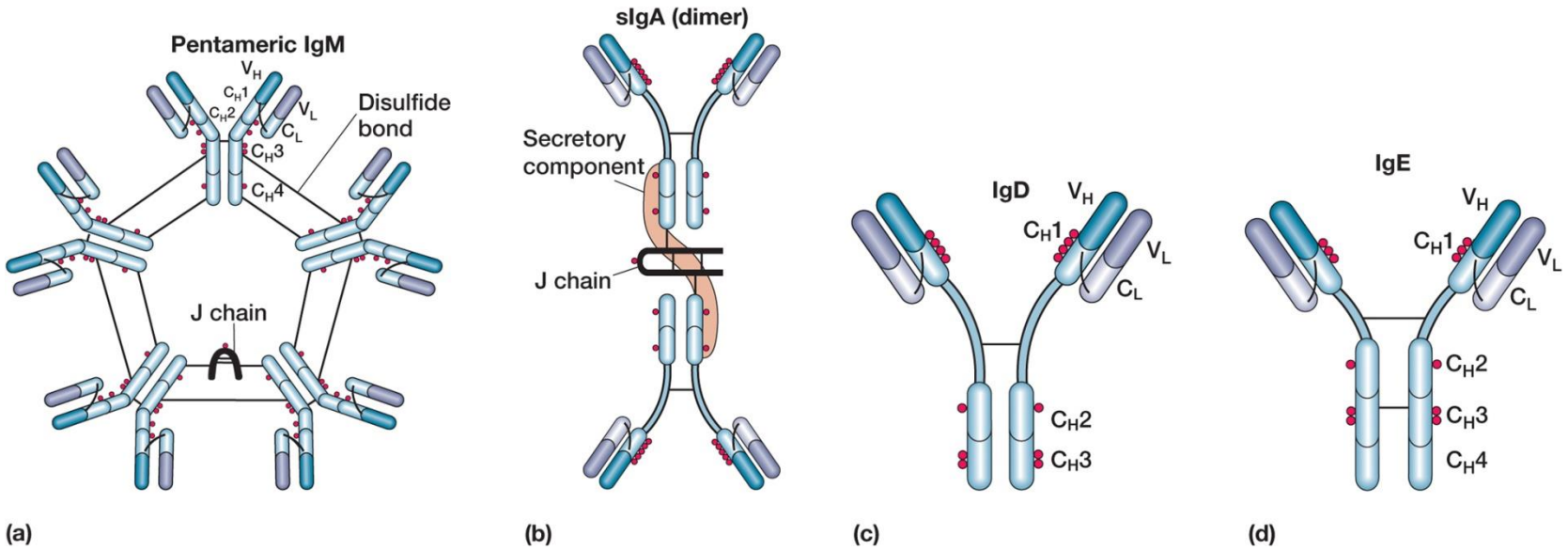
- IgG
  - 80% of serum immunoglobulin
  - opsonization, neutralization, activates complement
  - only Ig that can cross the placenta for natural passive immunity to neonate
- IgD
  - part of the B cell receptor complex
  - signals B cells to start antibody production

Immunoglobulin Classes

- IgM
  - pentamer arranged in pinwheel
  - first Ig in all immune responses
  - agglutination, activates complement
- IgA, secretory IgA (sIgA)
  - monomers and dimers
  - secreted across mucosal surfaces
  - tears, saliva, breast milk, MALT
  - immune exclusion

Immunoglobulin Classes

- IgE
  - lowest Ig serum level, elevated in parasitic infection and allergic reactions
  - opsonization (then binds to dendritic cells/macrophages)
  - mast cells bind Fc portion, activated to degranulate vasoactive granules when Fab portion binds allergens

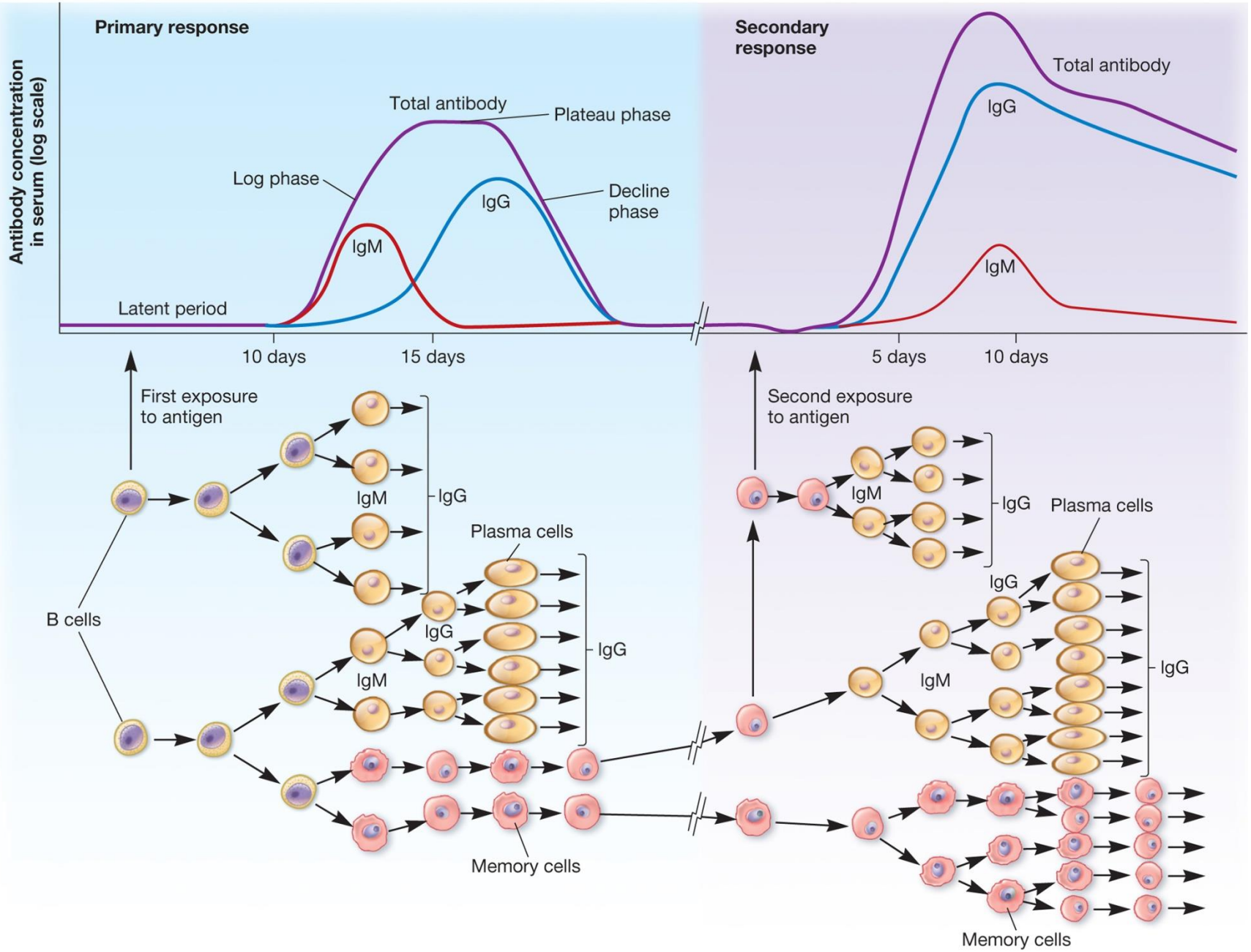


Antibody Kinetics

- Antibody synthesis and secretion can also be evaluated as a function of time
  - monomeric IgM is the B cell receptor for antigen whereas after B cell activation, pentameric IgM is secreted
  - class switching
    - change in antibody class secreted by plasma cells under the influence of T helper cells
    - event unfolds with time

Primary Antibody Response

- Several days to weeks lag or latent period after initial exposure to antigen
  - no antibody detectable in blood
- After B cell differentiation into plasma cells, antibody is secreted
  - antibody titer
    - is measure of serum antibody concentration
    - reciprocal of highest dilution of antiserum that gives positive reaction
- IgM appears first, followed by IgG



Secondary Antibody Response

- Upon secondary exposure to same antigen, B cells mount a heightened, memory response
- Characterized as having a shorter lag, a more rapid log phase, longer persistence, a higher IgG titer and production of antibodies with a higher affinity for the antigen

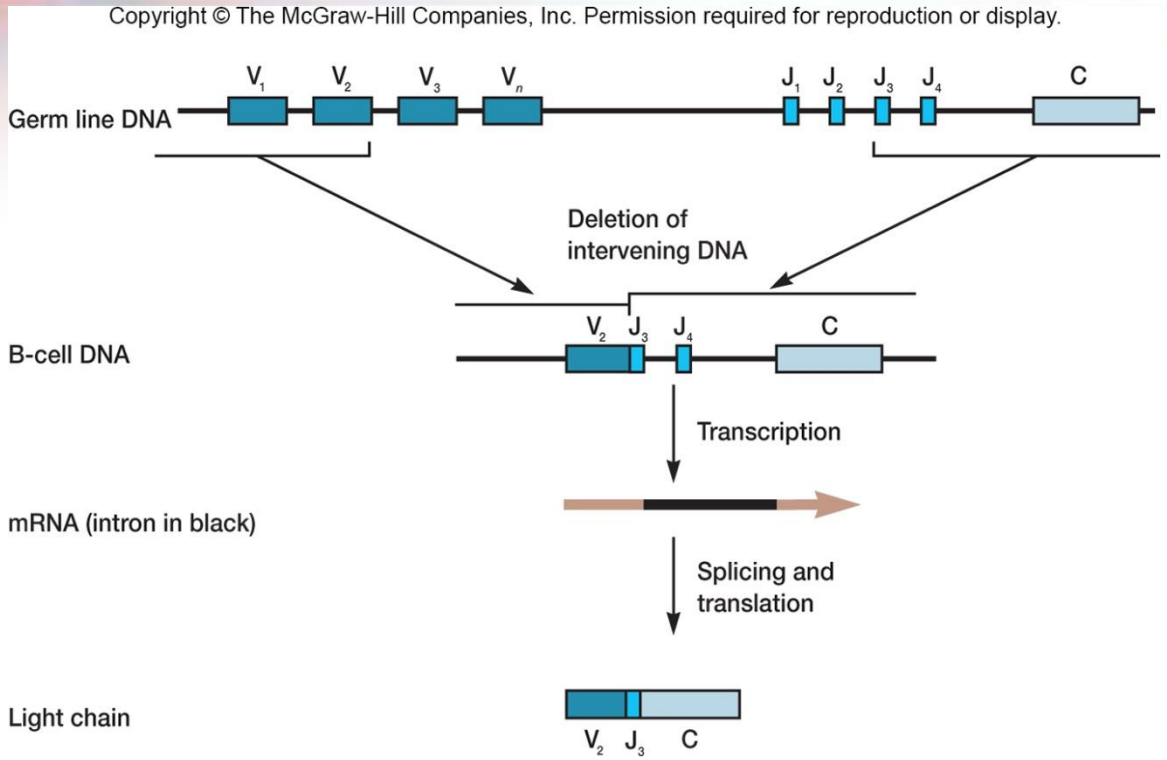
Diversity of Antibodies

- Three mechanisms contribute to generation of antibody diversity
  - rearrangement of antibody gene segments (combinatorial joining)
    - genes are split or interrupted into many gene segments
  - generation of different codons during antibody gene splicing
  - somatic mutation

Combinatorial Joining

- Segments clustered separately on same chromosomes
  - exons that code for constant regions
  - exons that code for variable regions
- Exons for constant region are joined (spliced together) to one segment of the variable region
- RAG-1 and RAG-2 are recombination enzymes
  - process still not fully understood
  - multiple enzymes involved
- Occurs on heavy and light chains

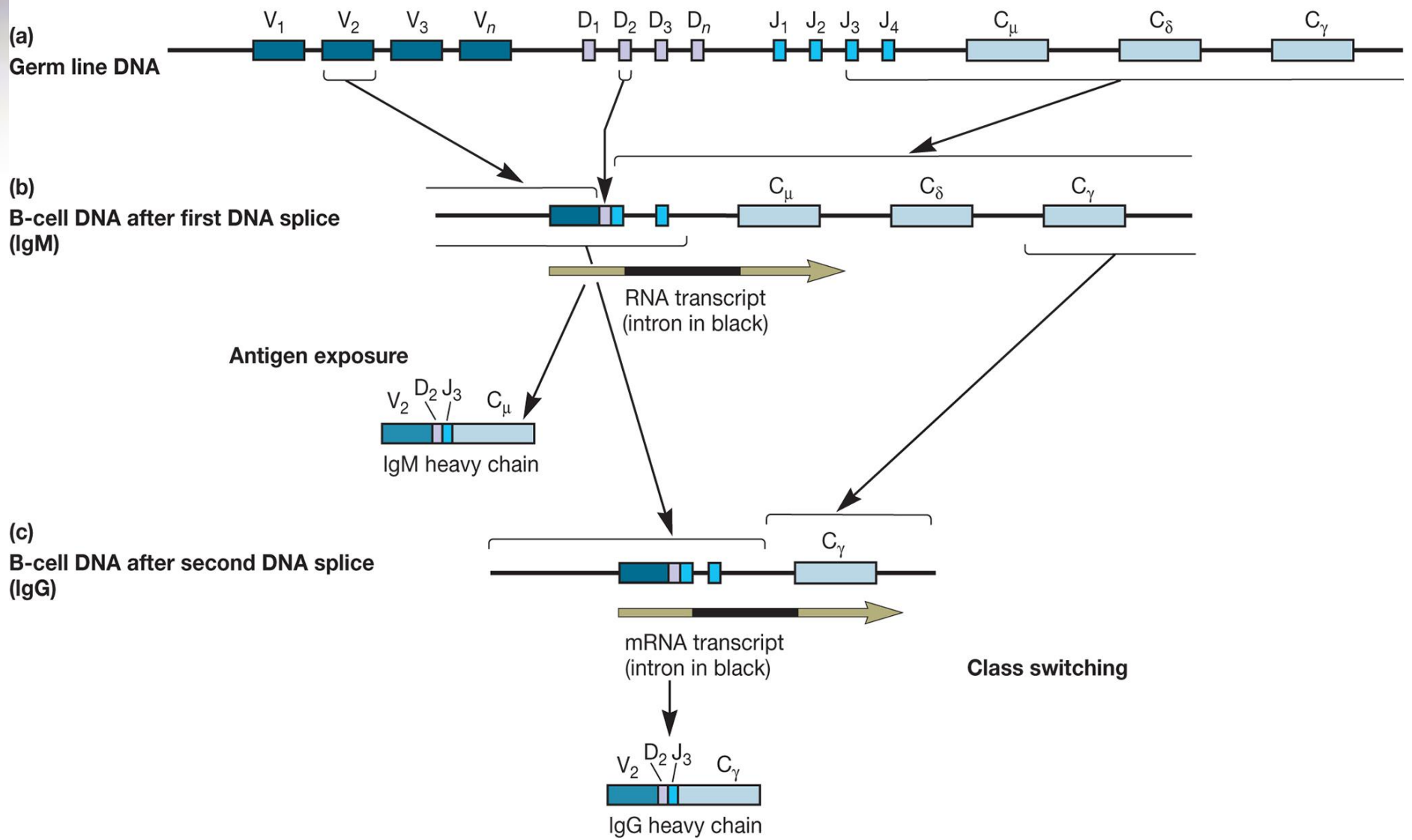
Light Chain



- Germ line DNA for light chain contains multiple coding sequences, V and J (joining)
- In B cell development
  - one V is joined with one J region
  - many possible combinations formed
  - VJ joined with C (constant) exon after transcription

Heavy Chain

- V and J regions are joined to 3rd coding region called D (diversity) sequences
- VDJ joined to C region after transcription (RNA level)
- Antibody class switch
  - initial C region results in IgM but changes as the immune response progresses and B cells proliferate
    - Region containing initial IgM C region is deleted, along with other intervening sequence
      - Occurs at DNA level, not RNA level (pre-transcription)
      - Process not fully understood yet, but depends on activation-induced cytidine deaminase (AID) enzyme and other enzymes



Antibody Diversity

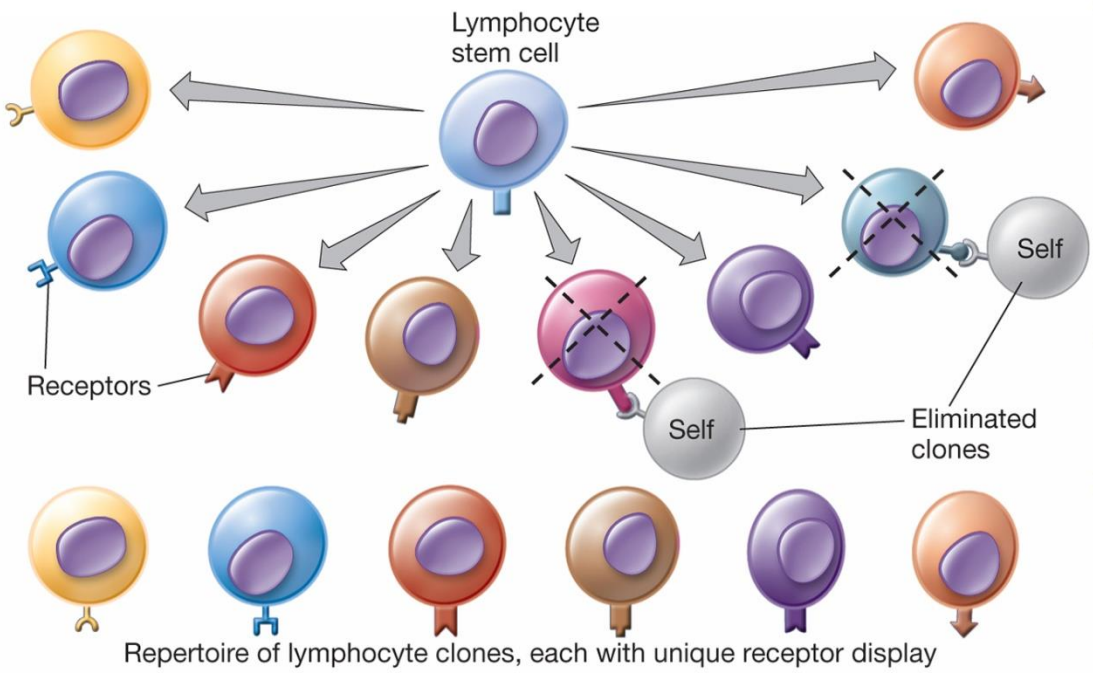
Table 34.3	Theoretical Antibody Diversity Resulting from Combinatorial Joining of Germ-Line Genes[1]
λ light chains	V regions = 2 J regions = 3 Combinations = 2 × 3 = 6
κ light chains	V_κ regions = 250 − 350 J_κ regions = 4 Combinations = 250 × 4 = 1,000 = 350 × 4 = 1,400
Heavy chains	V_H = 250 − 1,000 D = 10–30 J_H = 4 Combinations = 250 × 10 × 4 = 10,000 = 1,000 × 30 × 4 = 120,000
Diversity of antibodies	κ-containing: 1,000 × 10,000 = 10^7 1,400 × 120,000 = 2 × 10^8 λ-containing: 6 × 10,000 = 6 × 10^4 6 × 120,000 = 7 × 10^5

1 Approximate values.

- Splice site variability
  - VJ joining can produce polypeptides with different amino acid sequences
- Somatic mutation of V regions
  - V regions are susceptible to high rate of somatic mutation during an antigen challenge
  - produce antibodies with different epitope recognition

Clonal Selection Theory

- Body forms large, diverse B lymphocyte pool that can bind to large range of antigenic epitopes
- Self-reactive cells are eliminated at an early stage of development (clonal deletion)
- Encounter with antigen stimulates only those B cells that recognize and bind antigen
- Stimulated B cells proliferate to produce B cell clone (all have same antigen specificity)
- B cell clone differentiates to form two cell populations
  - plasma cells and memory B cells



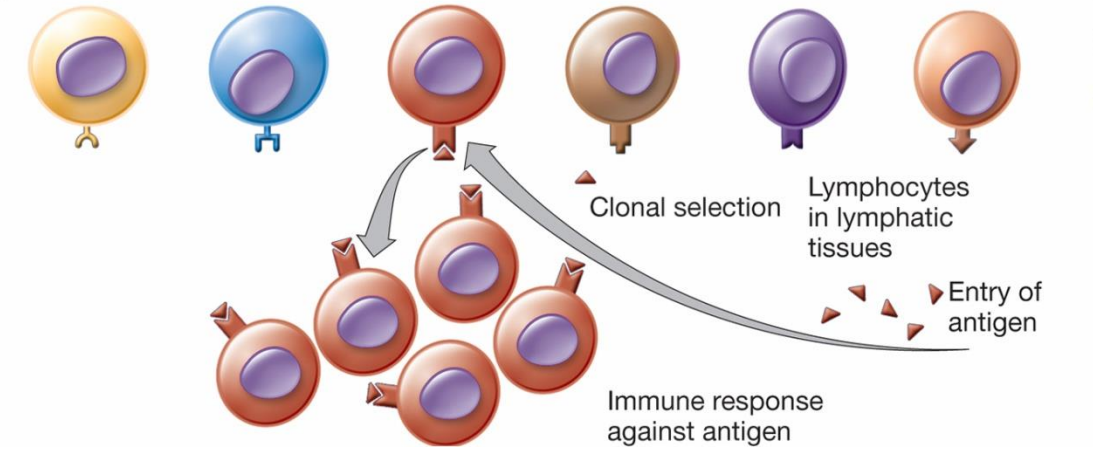
(a) Antigen-Independent Period

1. During development of early lymphocytes from stem cells, a given stem cell undergoes rapid cell division to form numerous progeny.

   During this period of cell differentiation, random rearrangements of the genes that code for cell surface protein receptors occur. The result is a large array of genetically distinct cells, called clones, each clone bearing a different receptor that is specific to react with only a single type of foreign molecule or antigen.

2. At the same time, any lymphocyte clones that have a specificity for self molecules and could be harmful are eliminated from the pool of diversity. This is called immune tolerance.

3. The specificity for a single antigen molecule is programmed into the lymphocyte and is set for the life of a given clone. The end result is an enormous pool of mature but naïve lymphocytes that are ready to further differentiate under the influence of certain organs and immune stimuli.



(b) Antigen-Dependent Period

4. Lymphocytes come to populate the lymphatic organs, where they will finally encounter antigens. These antigens will become the stimulus for the lymphocytes' final activation and immune function. Entry of a specific antigen selects only the lymphocyte clone or clones that carry matching surface receptors. This will trigger an immune response, which varies according to the type of lymphocyte involved.

Monoclonal Antibody Technology

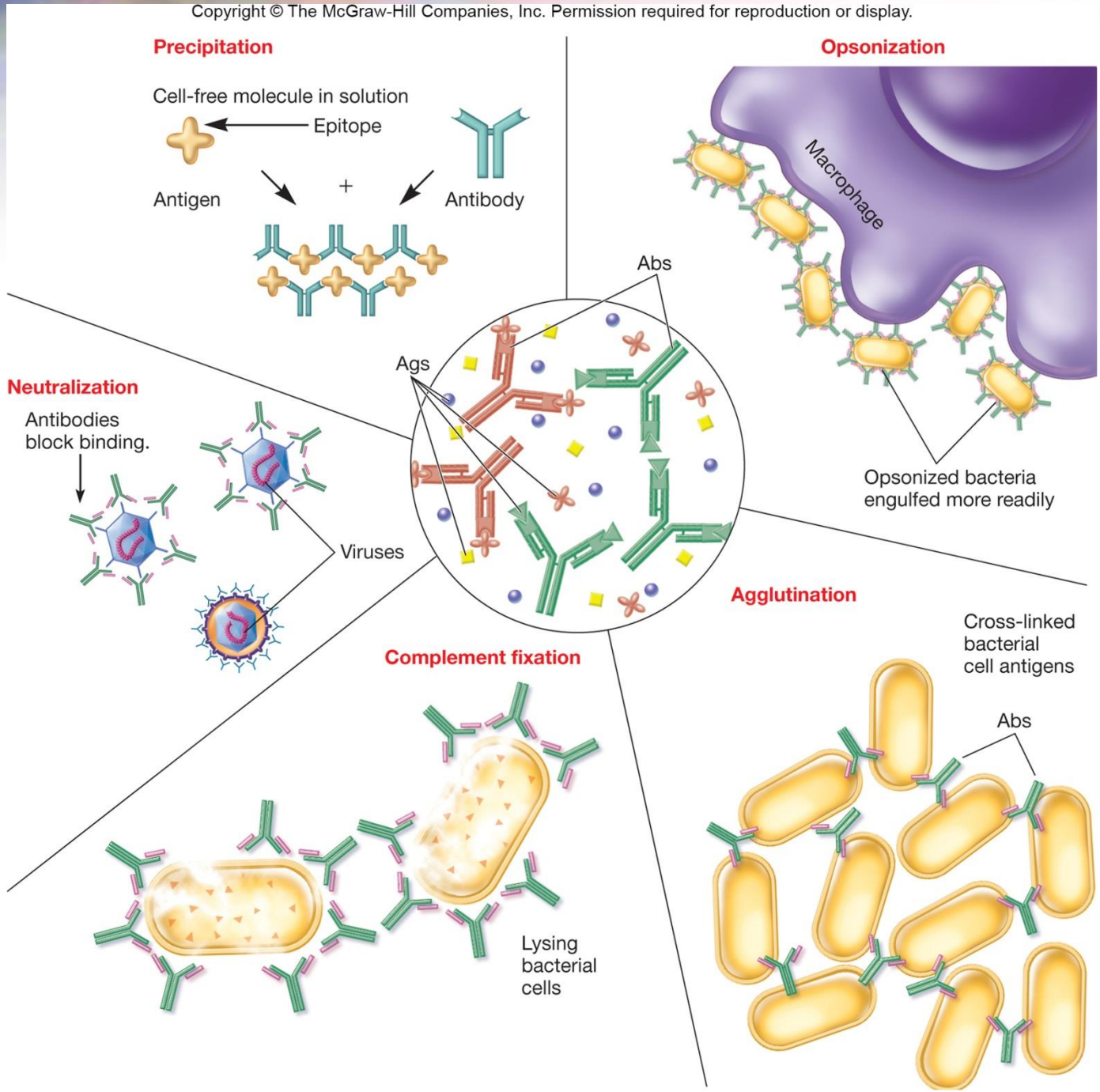
- Hybridomas
  - overcome some limitations of antisera as a source of antibodies
  - used to produce large quantities of monoclonal antibodies (MAb) that recognize one epitope
- Potential for numerous biomedical applications
  - most of current applications involve in vitro diagnostic testing and research
  - MAbs can be tagged with markers or even small drug molecules for delivery to specific sites

34.8 Action of Antibodies

1. Explain the consequences of antibody binding of antigen
2. Assess the effectiveness of antigen removal by antibody
3. Predict which antigens will be most susceptible to antibody action

Action of Antibodies

- Bind antigens with great specificity
  - can occur within animal body (in vivo)
    - essential for the protection of animal from viruses, microbes, and cancer cells
- Antibody coats foreign invading material
  - marks it for recognition by components of the innate and adaptive immune systems
  - neutralization, opsonization, and immune complex formation



Toxin Neutralization

- Inactivation of toxins resulting from interaction between toxin and specific antitoxin antibodies
- Complexing toxin with antibodies
  - can prevent the toxin from attaching to host cells
  - can prevent toxin from entering host cells
  - can result in ingestion by macrophates

Viral Neutralization

- IgG, IgM, and IgA antibodies can bind to some extracellular viruses and inactivate them
- Fixation of complement component C3b, from classical complement pathway, helps in the neutralization process
- Viral infection is prevented because neutralization of viruses prevents them from binding and entering target cells

Opsonization

- Microorganisms or other foreign particles become coated with antibodies and/or complement
- Opsonizing antibodies bind Fc receptors on macrophages and neutrophils, creating bridge between phagocyte and antigen

Immune Complex Formation

- Antigens and antibodies can crosslink, producing immune complexes
- Precipitation (precipitin) reaction occurs when antigens are soluble molecules and the immune complex settles out of solution
- Agglutination reaction occurs when cells or particles are cross-linked
  - the immune complex formed is more readily phagocytosed in vivo than are free antigens
  - caused by agglutinin antibodies
- Antibody:antigen ratio is in equivalence zone when their concentration is optimal for formation of the immune complex
- Is the basis for many immunological assays

34.9 Acquired Immune Tolerance

1. Evaluate the impact of improper B-cell activation
2. Contrast the outcome of B-cell and T-cell exposure to self antigens

Acquired Immune Tolerance

- How do we ‘know’ to respond to foreign but not self antigens?
  - Three proposed mechanisms
    - Negative selection (deletion) of autoreactive lymphocytes
      - Central tolerance if performed in bone marrow or thymus
    - Induction of anergy (peripheral tolerance)
      - Partial or incomplete stimulation (no 2nd signal) of lymphocytes induces them to become nonreactive
        - » B cells receive only self Ag signal through BCR
        - » T cells receive only self Ag signal from an APC
    - Inhibition of anti-self reactivity by Treg cell activity and cytokine release

34.10 Immune Disorders

1. Illustrate differences between hypersensitivity, autoimmunity, graft rejection, and immunodeficiency
2. Relate types of hypersensitivity to the root biological cause
3. Predict disease potential due to the presence in the body of “self-reactive” T and B cells
4. Explain graft rejection as a function of cells responding to the graft
5. Propose the impact on immunity when T cells and B cells become deficient

Immune Disorders

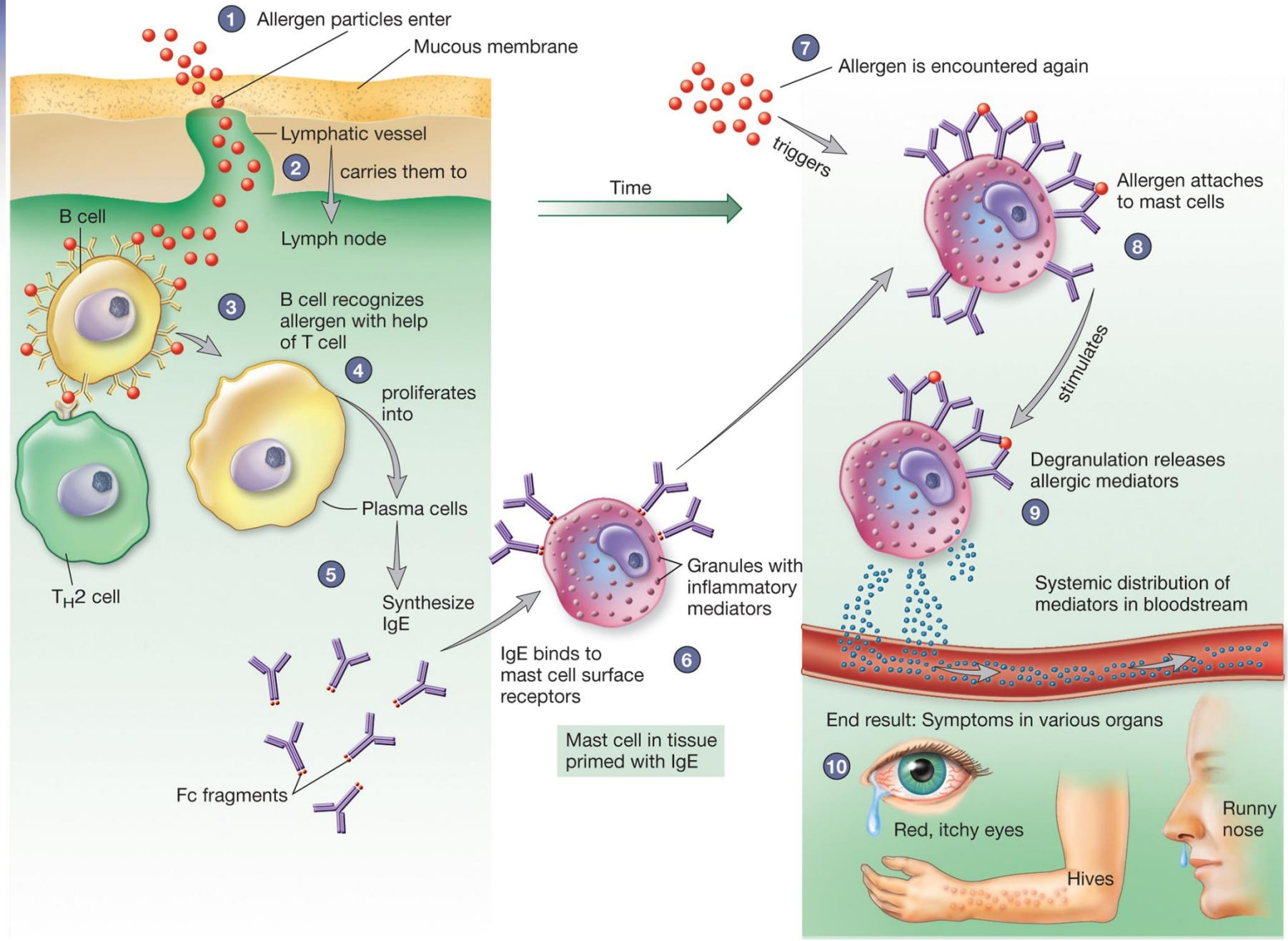
- Hypersensitivities
- Autoimmune diseases
- Transplantation (tissue) rejection
- Immunodeficiencies
  - congenital
  - acquired

Hypersensitivities

- Exaggerated immune response upon second or subsequent contact with antigen
- Causes tissue damage
- Reactions classified as immediate or delayed
- Gell-Coombs classification into four different types of hypersensitivity: I, II, III, and IV

Type I Hypersensitivity

- Allergy
  - one kind of Type I hypersensitivity
- Allergen
  - antigen that causes allergic reaction
- Occurs immediately following second contact with allergen
- Involves production and action of IgE (sometimes called reagin) and mast cells
  - basophils or eosinophils may be involved as well



(a) Sensitization/IgE production

(b) Subsequent exposure to allergen

Anaphylaxis

- Release of physiological mediators in response to allergen cause
  - smooth muscle contraction
  - vasodilation
  - increased vascular permeability
  - mucous secretion
- Can be systemic or localized

Systemic Anaphylaxis

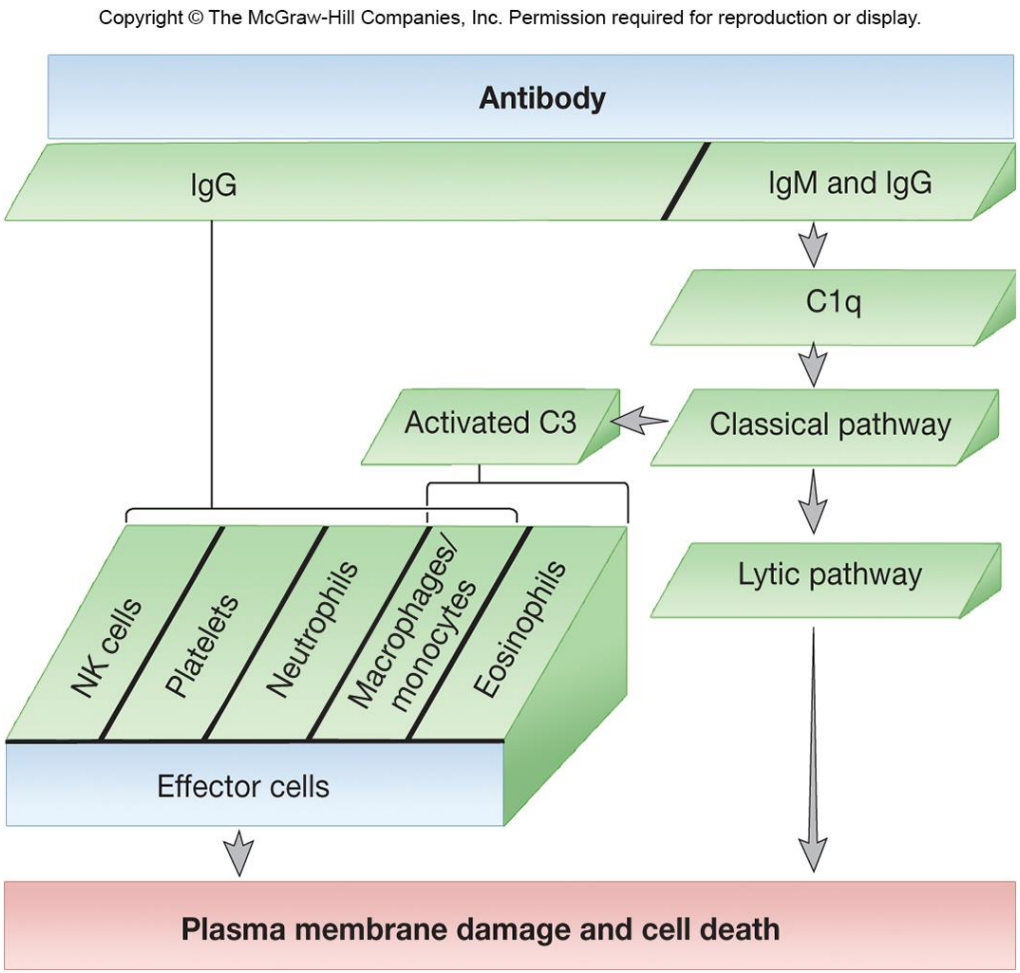
- Results from massive release of mast cell mediators in a short time
- Usually results in respiratory impairment, decreased blood pressure, and circulatory shock
- Can cause death within a few minutes

Localized Anaphylaxis

- An atopic (“out of place”) reaction
  - symptoms depend on route by which allergen enters body
- Hay fever
  - upper respiratory tract
- Bronchial asthma
  - lower respiratory tract
- Hives
  - skin
  - common with true food allergies

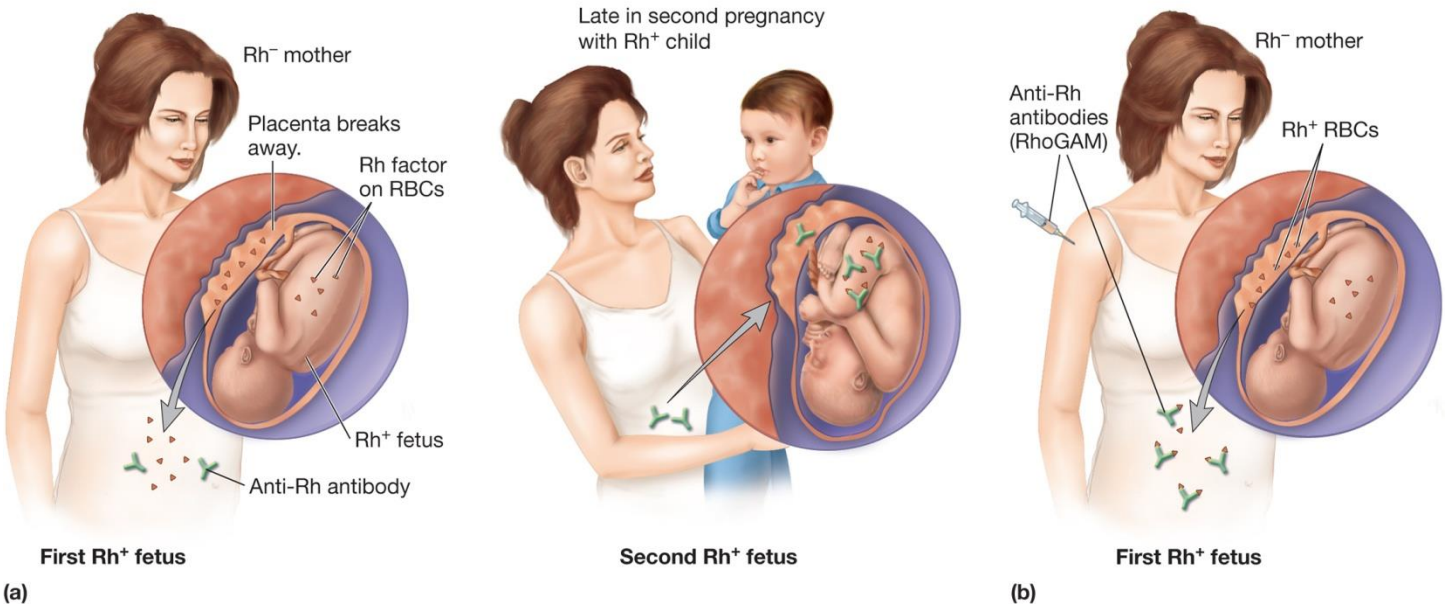
Type II Hypersensitivity

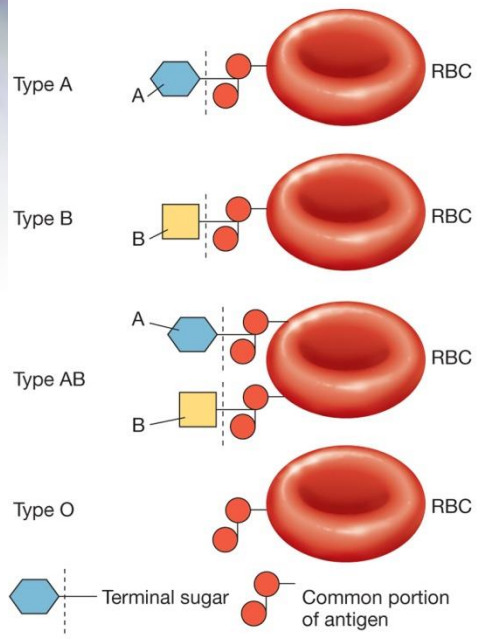
- Cytolytic or cytotoxic reaction
- Involves IgG and IgM antibodies
  - directed against cell surface or tissue antigens
  - stimulate complement pathway and effector cells



Examples

- Blood transfusion reaction in which donated blood cells are attacked by recipient's antibodies
- Erythroblastosis fetalis
  - mother can be passively immunized with anti-Rh factor antibodies or RhoGam to control this disease which is potentially fatal for newborn





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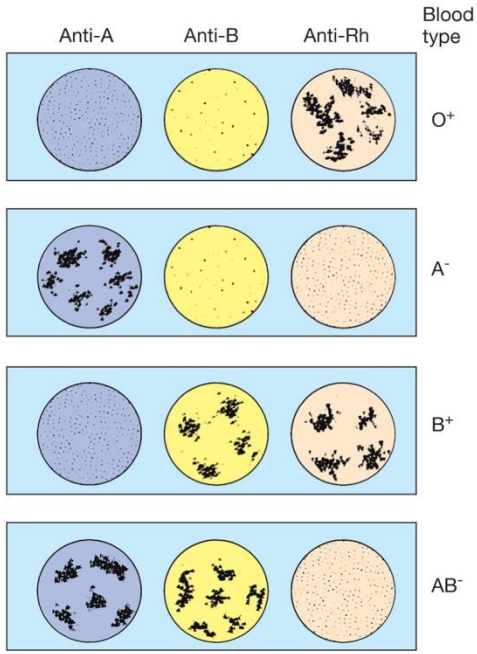

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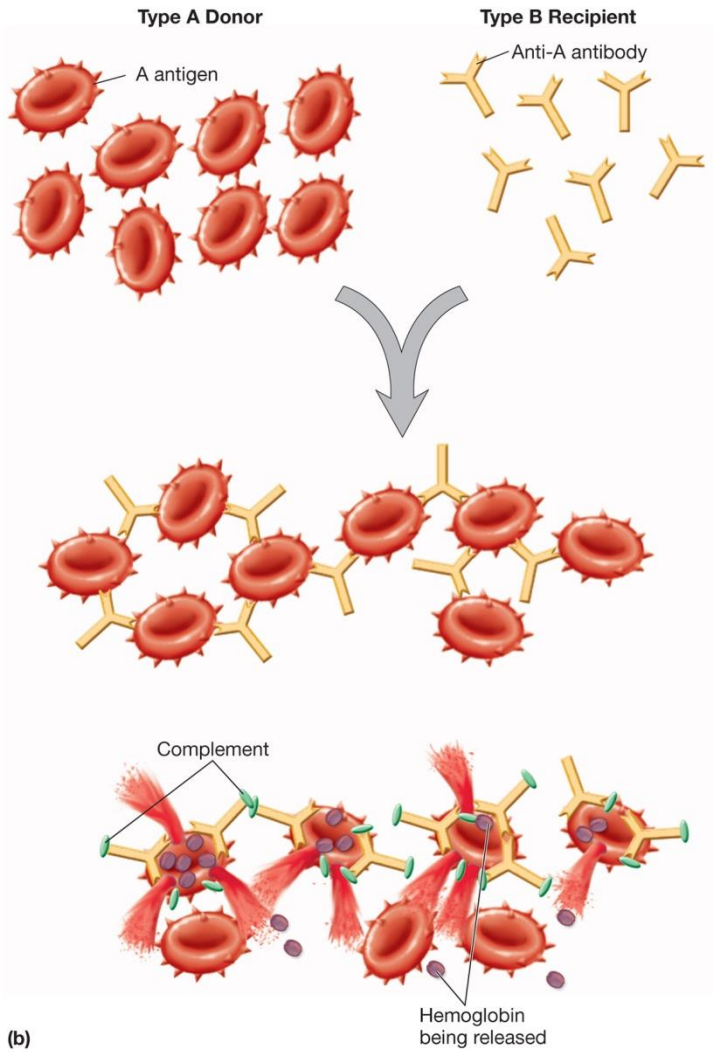
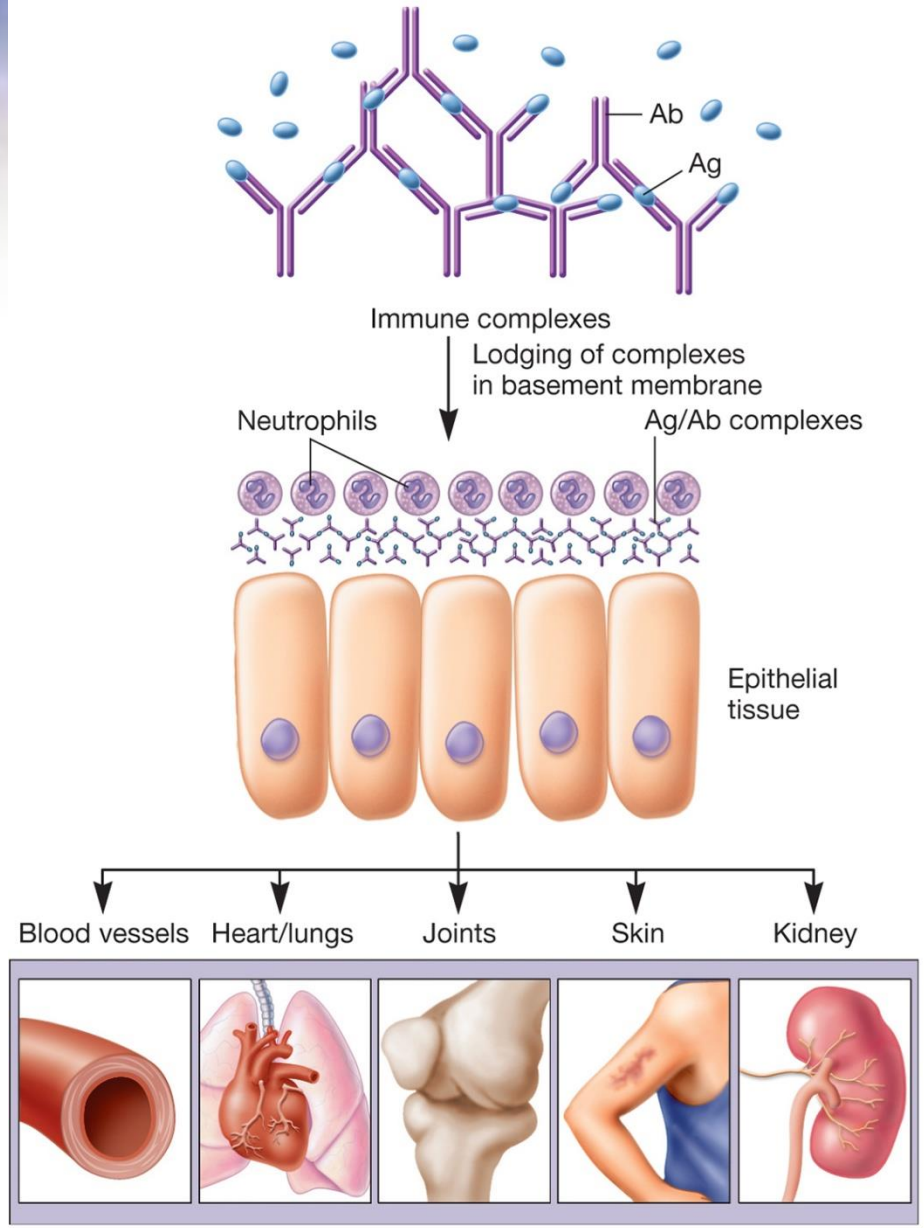


(b)

Type III Hypersensitivity

- Involves formation of immune complexes
  - usually removed by monocytes and macrophages
  - if accumulate, leads to hypersensitivity reaction
    - resulting inflammation causes tissue damage
- e.g., vasculitis, glomerulonephritis, arthritis, and systemic lupus erythematosis

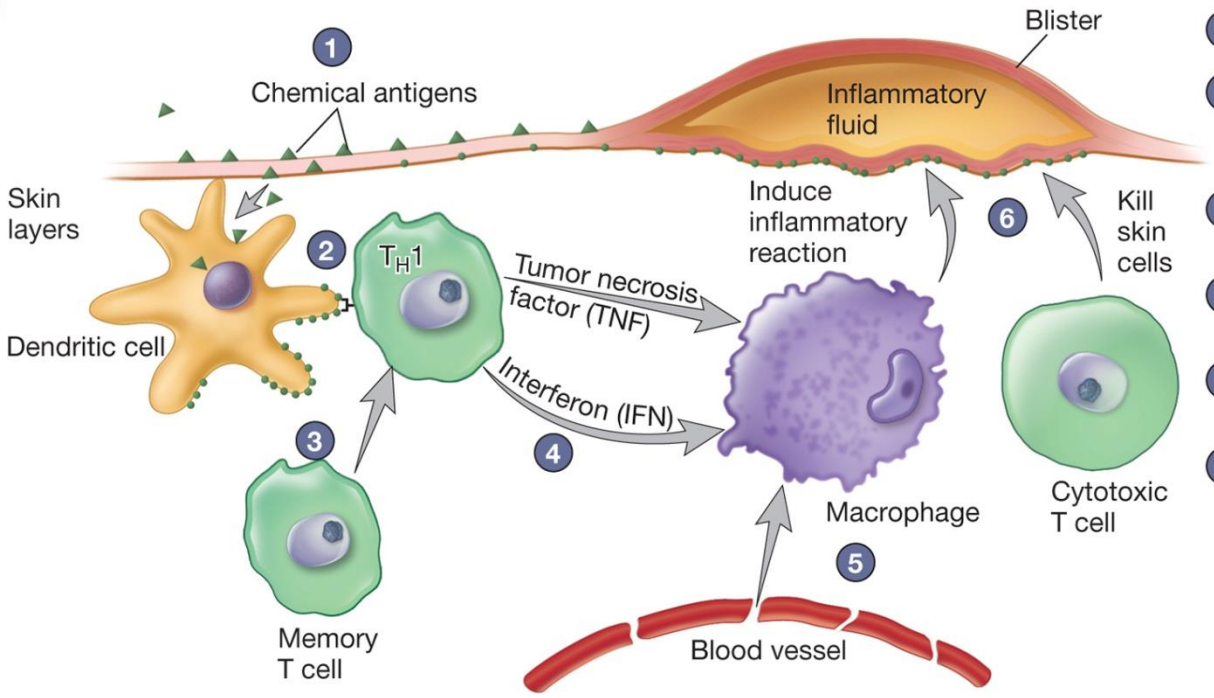


Major organs that can be targets of immune complex deposition

1. Antibody combines with excess soluble antigen, forming large quantities of Ab/Ag complexes.
2. Circulating immune complexes become lodged in the basement membrane of epithelia in sites such as kidney, lungs, joints, skin.
3. Fragments of complement cause release of histamine and other mediator substances.
4. Neutrophils migrate to the site of immune complex deposition and release enzymes that cause severe damage to the tissues and organs involved.

Type IV Hypersensitivity

- Involves delayed, cell-mediated immune reactions
- Important factor is time required for T cells to reach and accumulate near antigens
- T_H and CTL cells can elicit type IV reactions
- Examples
  - tuberculin hypersensitivity
  - some autoimmune diseases
  - transplantation rejection
  - cancer cell killing
  - allergic contact dermatitis



1. Lipid-soluble catechols are absorbed by the skin.
2. Dendritic cells close to the epithelium pick up the allergen, process it, and display it on MHC proteins.
3. Previously sensitized T_H1 ($CD4^+$) cells recognize the presented allergen.
4. Sensitized T_H1 cells are activated and secrete cytokines (IFN, TNF).
5. These cytokines attract macrophages and cytotoxic T cells to the site.
6. Macrophages release mediators that stimulate a strong, local inflammatory reaction. Cytotoxic T cells directly kill cells and damage the skin. Fluid-filled blisters result.

Autoimmune Diseases

- Autoimmunity
  - presence of serum antibodies that react with self antigens (autoantibodies)
  - often benign
  - natural consequence of aging
  - reversibly induced by numerous stimuli (e.g., infectious organisms, drugs)
- Autoimmune disease
  - results from activation of self-reactive T and B cells
  - leads to tissue damage

Table 34.4 Some Autoimmune Diseases in Humans

Disease	Autoantigen	Pathophysiology
Acute rheumatic fever	Streptococcal cell wall antigens mimic self antigens and induce antibodies that cross-react with antigens on cardiomyocytes and other cells.	Type II hypersensitivity leads to myocarditis, heart valve scarring, and arthritis.
Autoimmune hemolytic anemia	Rh blood group antigen induces antibody to Rh antigen on red blood cells.	Type II hypersensitivity leads to anemia when red blood cells are destroyed by complement and phagocytosis.
Goodpasture's syndrome	Damage to kidney basement membrane exposes cryptic collagen protein, inducing anti-collagen antibody.	Type II hypersensitivity results in glomerulonephritis and pulmonary hemorrhage.
Graves' disease	Antibody to thyroid-stimulating hormone (TSH) receptor mimics TSH.	Overstimulation of TSH receptor leads to hyperthyroidism.
Multiple sclerosis	Antibody and activated T cells to several nervous system antigens	Types II and IV hypersensitivities alter nerve communications, leading to numbness, weakness, spasm, and loss of motor and cognitive function.
Myasthenia gravis	Antibody to acetycholine receptor in skeletal muscle	Antibody blockade of neurotransmitter receptors results in progressive muscular weakness.
Rheumatoid arthritis	IgG antibody (to synovial joint cartilage antigen) recognized as foreign	Type III hypersensitivity from immune complexes of antibodies to antibodies results in joint inflammation and destruction.
Systemic lupus erythematosus	Antibodies to various cellular (DNA, nucleoprotein, cardiolipin) and blood clotting components	Type III hypersensitivity results in immune complex-induced arthritis, glomerulonephritis, vasculitis, and rash.
Insulin-dependent diabetes mellitus	Antibody and activated T cells to pancreatic beta cell antigens	Types II and IV hypersensitivities destroy beta cells, resulting in insulin deficiency.

Autoimmune Diseases

- Examples
  - rheumatoid arthritis
  - insulin-dependent diabetes mellitus
- Facts that influence the development of autoimmune disease
  - infection
  - genetic
  - viral
  - hormones
  - influence of stress and neurochemicals on the immune response

Transplantation (Tissue) Rejection

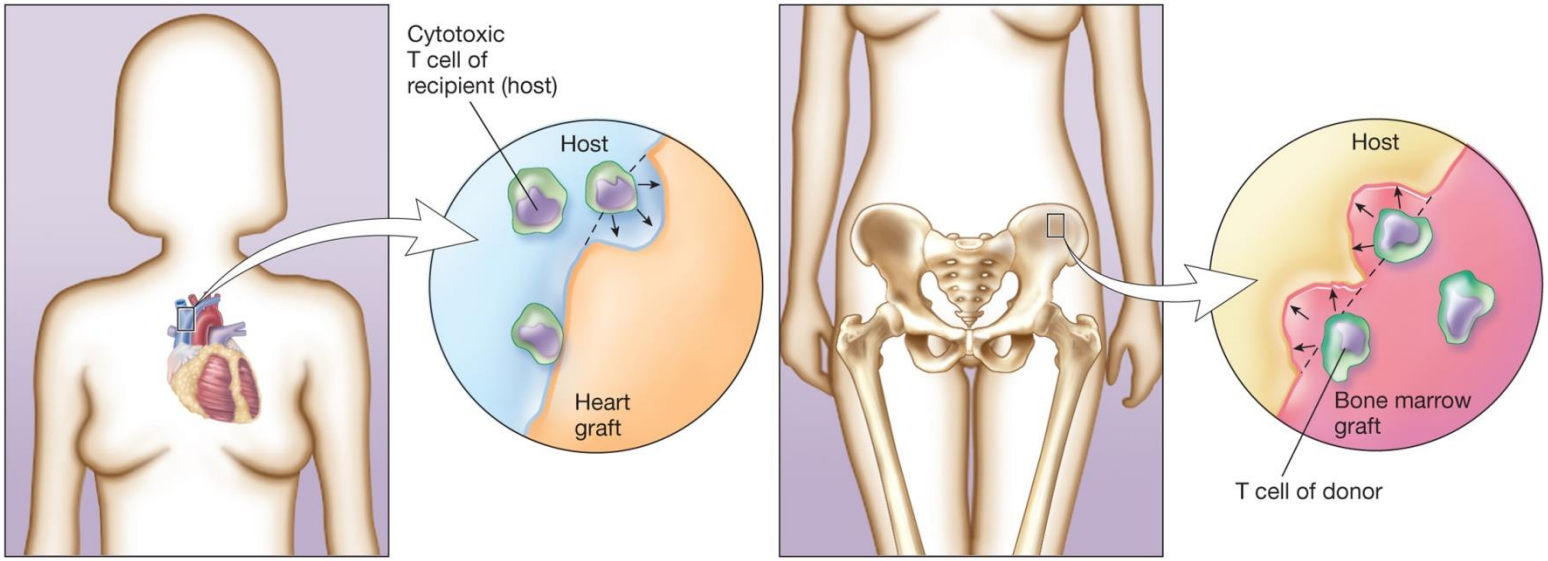
- Types of transplants
  - allograft
    - transplants between genetically different individuals within a species
  - xenograft
    - donor and recipient are different species
- Two mechanisms can occur
  - foreign MHC molecules on transplanted tissue (graft) recognized by host T_H cells which aid T_C cells in destroying graft
  - T_H cells react to graft by releasing cytokines which stimulate destruction of graft by macrophages

Tissue Transplantation

- To be successful, the ABO blood group and the MHC molecules of the donor and recipient must be as closely matched as possible
  - after matching blood types of donor and recipient, the identification of Human Leukocycte Antigens (HLA) is done
  - family members are the first choice for a close match because HLA genes are usually inherited as a complete set from parents

Graft-Versus-Host Disease

- Can occur in organ transplant recipients
- Immunocompetent cells in donor tissue reject host
- e.g., in bone marrow transplants
  - disease prevented by treating donor with immunosuppressive drugs to deplete marrow of mature T cells



Immunodeficiencies

Table 34.5 Some Congenital Immune Deficiencies in Humans

Condition	Symptoms	Cause
Chronic granulomatous disease	Defective monocytes and neutrophils leading to recurrent bacterial and fungal infections	Failure to produce reactive oxygen intermediates due to defective NADPH oxidase
X-linked agammaglobulinemia	Plasma cell or B-cell deficiency and inability to produce adequate specific antibodies	Defective B-cell differentiation due to loss of tyrosine kinase
DiGeorge syndrome	T-cell deficiency and very poor cell-mediated immunity	Lack of thymus or a poorly developed thymus
Severe combined immunodeficiency disease (SCID)	Both antibody production and cell-mediated immunity impaired due to a great reduction of B- and T-cell levels	Various mechanisms (e.g., defective B- and T-cell maturation because of X-linked gene mutation; absence of adenosine deaminase in lymphocytes)

- Failure to recognize and/or respond to foreign Ag
- Primary (congenital) immunodeficiencies
  - result from genetic disorder
- Acquired immunodeficiencies
  - result from infection by immunosuppressive microbes (e.g., HIV)