Chapter Six
Immunopathology

This is concerned with the pathological changes that occur in the tissues as a result of improper immune response. Body immune responses are normal defense mechanisms designed to combat the effect of invasion by various environmental factors such as microorganisms & toxic chemicals. It usually works effectively.

Immunologic diseases may result from:
- Inadequate immune response.
- Excessive immune response.
- Inappropriate immune response.

Inadequate immune responses:
These can result from immuno-deficiency states. There are two classes of immunodeficiency syndromes:

Primary: which is present at birth & often the result of a genetic disorder.
Secondary: which is much more common than the primary. It can be secondary to:
- Drugs.
- Diseases:
  1. Old age.
  2. Chronic malnutrition.
  3. Widespread malignancy.
  4. Metabolic diseases (diabetes, chronic liver failure, chronic renal failure).
  5. Drug therapy (cytotoxic therapy, steroid therapy).
  7. AIDS.

Primary immunodeficiency
- X-linked agammaglobulinemia: This is one of the more common forms of primary immunodeficiency. It is due to failure of pre-B-cells to differentiate into B-cells that result in the absence of gamma globulin in the blood (agammaglobulinemia).
- Isolated IgA-deficiency: This is the most common of all primary immunodeficiency states. There is marked reduction in the level of serum IgA but other immunoglobulins are normal. In most cases it is asymptomatic & detected accidentally, but some patients have recurrent respiratory infections & diarrhea. There is also a significant, unexplained association with autoimmune diseases.
- Hyper-IgM syndrome: Patients with this syndrome produce normal or even above normal levels of IgM antibodies to antigens but fail to produce other antibody classes (IgG, IgA, or IgE isotypes).
- Thymic hypoplasia: Affected infants show failure of T-cells to form, with hypoplasia of the thymus gland; the result of failure of development of the third and fourth pharyngeal pouches. In 90% of cases there is a deletion affecting chromosome 22q11.
- Severe combined immune deficiency (SCID): This condition may be inherited as a recessive disorder either autosomal or an X-linked. The condition is due to failure of development of both B-cell & T-cell precursors from primitive stem cells. Therefore the thymus is small or absent & lymphoid tissues in lymph nodes & gut are also greatly
reduced. There is a very low blood lymphocyte count & low blood immunoglobulin levels.

**Acquired immunodeficiency syndrome (AIDS):** AIDS is a retroviral disease caused by human immunodeficiency virus (HIV) & is characterized by:
1. Profound immuno-suppression leading to opportunistic infections.
2. Secondary neoplasms.
3. Neurological manifestations.

**Etiology:** AIDS is caused by HIV. This is a human retrovirus belonging to the lentivirus family.

**Pathogenesis:** The two major targets of HIV infection are the immune system & the central nervous system.

**Immunopathogenesis of HIV disease:** AIDS leads to severe impairment of the cell-mediated immunity system. Infection by HIV leads to destruction of CD4 lymphocytes & a decreased helper/suppressor T-cell ratio in the blood. The virus gains entry to T-cells by attaching to surface CD4 molecules. The CD4 molecule is a high-affinity receptor for HIV. This explains the selective tropism of the virus for CD4+T-cells & its ability to infect other CD4+cells, particularly macrophages & dendritic cells. However binding to CD4 is not sufficient for infection; the HIV envelope gp120 must also bind to other cell surface molecules (co-receptors) to facilitate cell entry. The virus core containing the HIV genome enters the cytoplasm of the cell. The viral genome then undergoes reverse transcription, leading to formation of complementary DNA (cDNA). In the dividing T-cells, the cDNA enters the nucleus & integrates into the host genome. After integration, the provirus may remain non-transcribed for months or years & the infection becomes latent; alternatively, proviral DNA may be transcribed to form complete viral particles that bud from the cell membrane, leading to cell death.

HIV colonizes the lymphoid organs (spleen, lymph nodes, and tonsils). Infected T-cells, macrophages, & dendritic cells represent reservoirs of infection. Initially, the immune system can vigorously proliferate to replace the dying T-cells, thus masking the massive cell death occurring primarily in the lymphoid tissues. Due to loss of CD4+ cells, patients will have an inversion of the CD4/CD8 ratio in the peripheral blood; normally it is about 2, while in AIDS patients the ratio \( \leq 0.5 \).

ADIS patients will also have qualitative defects in T-cell function.

**Pathogenesis of central nervous system involvement:** The nervous system is a major target of HIV infection. Macrophages & their equivalents in the CNS; the microglia, are mainly infected with HIV. The virus is mostly carried into the brain by infected monocytes. The mechanism of HIV-induced damage of the brain remains obscure. It is believed that neurologic deficit is caused indirectly by viral products & soluble factors e.g. cytokines produced by macrophages/microglia.

**Opportunistic infections:**
These are responsible for about 80% of deaths in patients with AIDS.
1. Pneumocystis carinii pneumonia is the presenting feature in many cases.
2. Recurrent mucosal candidiasis
3. Disseminated cytomegalovirus infection (particularly enteritis & retinitis)
4. Herpes simplex; especially severe ulcerating oral & perianal infections
5. Mycobacterium tuberculosis & atypical mycobacteria (mycobacterium ovium-intracellular), usually disseminated infections
6. Toxoplasmosis, which is the most common secondary infection of the central nervous system
7. Cryptococcal meningitis is also quite frequent.
8. Cryptosporidium or isospora belli infections are often the cause of the so common persistent diarrhea. However, bacterial pathogens such as Salmonella & Shigella species may also be seen.

Secondary Neoplasms: the basis of increased risk of malignancy is multifactorial and include:
1. Profound defects in T-cell immunity
2. Dysregulated B-cell & monocyte functions
3. Infections with known viruses e.g. Human herpes virus type 8, EBV, human papilloma virus & unknown viruses.

Patients with AIDS have a high incidence of certain tumors as:
● Kaposi sarcoma (KS): this vascular tumor is the most common neoplasm in AIDS patients. KS in AIDS patients is usually multicentric & tends to be aggressive. It can affect the skin, mucous membranes, GIT, lymph nodes, & lungs. KS is associated with human herpes virus 8 infections in AIDS patients.
● Non-Hodgkin lymphomas: the second most common neoplasm in AIDS patients. These tumors are highly aggressive, & involve many extra-nodal sites, commonly the brain; so primary lymphoma of the brain is considered as an AIDS-defining condition. 30%-40% of these lymphomas are associated with EBV infection.
● Cervix uteri carcinoma: this due to human papilloma virus infection in ADIS patients. This virus is intimately associated with squamous cell carcinoma of the cervix & its precursor lesions (cervical dysplasia & carcinoma in situ); therefore, gynecologic examination should be routinely done for HIV-infected women.

Central nervous system involvements:
This is common in AIDS patients. In addition to opportunistic infection & neoplasms, patients may have a progressive encephalopathy clinically called AIDS-dementia complex.

Immunologically Mediated Tissue Injury: Immune responses not only protect against invasion by foreign organisms but may also cause tissue damage. An immune response that leads to tissue injury or disease is broadly called a hypersensitivity reaction.
Hypersensitivity reactions are classified according to the type of immune mechanism.
● In most type I, or immediate-type hypersensitivity reactions, IgE antibody is formed and binds to high-affinity receptors on mast cells and/or basophils via its Fc domain. Subsequent binding of antigen and cross-linking of IgE triggers rapid (immediate) release of products from these cells, leading to the characteristic manifestations of such diseases as urticaria, asthma and anaphylaxis.
● In type II hypersensitivity reactions, IgG or IgM antibody is formed against an antigen, usually a protein on a cell surface. Less commonly, the antigen is an intrinsic structural component of the extracellular matrix (e.g., part of the basement
membrane). Such antigen–antibody coupling activates complement, which in turn lyses the cell (cytotoxicity) or damages the extracellular matrix. In some type II reactions, other antibody-mediated effects are operative.

- **In type III hypersensitivity reactions**, the antibody responsible for tissue injury is also usually IgM or IgG, but the mechanism of tissue injury differs. The antigen circulates in the vascular compartment until it is bound by antibody. The resulting immune complex is deposited in tissue. Complement activation at sites of antigen–antibody deposition leads to leukocyte recruitment, which is responsible for the subsequent tissue injury. In some type III reactions, antigen is bound by antibody in situ.
- **Type IV reactions**, or cell-mediated or delayed-type, hypersensitivity reactions, do not involve antibodies. Rather, antigen activation of T lymphocytes, usually with the help of macrophages, causes release of products by these cells, thereby leading to tissue injury.

*Inappropriate immune response:*

**Transplant rejection:** Organ transplantation is used increasingly to treat irreversible diseases of the kidney, liver, heart, lung, & bone marrow. Unfortunately, the action of the immune system of the recipient can lead to destruction of the transplanted tissue a process termed "transplant rejection". This is a complex immunologic phenomenon involving both cell & humoral-mediated hypersensitivity responses of the host, directed against histocompatibility antigens, human leukocytes antigens (HLA) on the donor allograft. The endothelial cells that line the blood vessels of the graft are particularly rich in both HLA & blood group antigens, thus blood vessels are important targets of the host's immune response to a transplanted allograft.

*Patterns of transplant rejection:*

Rejection reactions have been classified as
- Hyperacute.
- Acute.
- Chronic.

**Hyperacute rejection:** This occurs within a very short time from the moment the organ is perfused by the host's blood (minutes to a few hours). In this form there is a widespread intravascular thrombosis in small vessels, with focal necrosis. It is the result of pre-formed humoral host antibodies reacting with antigens in the graft. The hyperacutely rejected kidney rapidly becomes cyanotic, mottled, & flaccid & may excrete only a few drops of blood-stained urine. In contrast to a non-rejected kidney graft that regains a normal pink coloration & tissue turgor & promptly excretes urine. Histologically the rejected kidney show acute arteritis & arteriolitis, vessel thrombosis & ischemic necrosis.

**Acute rejection:** It occurs within days or weeks of transplantation, but may also appear after cessation of immunosuppressive therapy that is given to the recipient to prevent rejection. It is mediated by both humoral & cell-mediated mechanisms. Acute cellular rejection is mediated by T-cells reacting against donor HLA antigens, particularly class II. It is accompanied by signs of renal failure. Histologically there is extensive interstitial CD₄+ & CD₈+T-cell infiltration. The humoral component of acute rejection is characterized by vasculitis with endothelial necrosis, neutrophils infiltration of vessel walls, & damage to the intima and elastic lamina of the larger arteries in the graft.
**Chronic rejection:** Chronic rejection occurs slowly & progressively after transplantation (months to years). It is the result of slow breakdown of the host's tolerance to the graft and may be due to inadequate immune suppression. Histologically, there is intimal fibrosis mainly in arteries & arterioles, leading to secondary ischemic damage to the parenchyma manifested by hyalinization and loss of glomeuli, interstitial fibrosis, & tubular atrophy. Chronic rejection does not respond to standard immunosuppressant regimens.

**Autoimmune diseases:** Autoimmune diseases are the result of immune reactions against self-antigens i.e. against body tissue or individual tissue components. Sometimes the immune response is an antibody response (autoantibody), or it is a cell-mediated immune response. In autoimmune diseases, the normal mechanisms ensuring tolerance for self-antigens have broken down. (Self-tolerance indicates lack of immune responsiveness to one's own tissue antigens). Some autoimmune diseases have a genetic component; e.g. certain diseases are associated with particular HLA histocompatibility types. In other situations, an autoimmune disease can be triggered by a microbial infection.

**Mechanisms of Autoimmunity:** Autoimmunity arises from a combination of the inheritance of susceptibility genes, which may contribute to the breakdown of self-tolerance, and environmental triggers, such as infections and tissue damage, which promote the activation of self-reactive lymphocytes. In general, these genetic and environmental influences conspire to create an imbalance between control mechanisms that normally function to prevent self-reactivity and pathways that lead to the generation and activation of pathogenic effector lymphocytes. **Role of Susceptibility Genes:** Most autoimmune diseases are complex multigenic disorders. Among the genes known to be associated with autoimmunity, the greatest contribution is that of HLA genes. It is postulated that the presence of particular MHC alleles affects the negative selection of T cells in the thymus or the development of regulatory T cells, but there is little proof for either possibility. It should be pointed out that many normal individuals inherit the MHC alleles that are disease-associated in patient populations, and normal MHC molecules are capable of presenting self-antigens. Therefore, the presence of particular MHC alleles is not, by itself, the cause of autoimmunity. Studies have shown that multiple non-MHC genes are associated with various autoimmune diseases. Some of these genes are disease-specific, but many of the associations are seen in multiple disorders, suggesting that the products of these genes affect general mechanisms of immune regulation and self-tolerance.

**Role of Infections:** Many autoimmune diseases are associated with infections, and clinical flare-ups are often preceded by infectious prodromes. Two mechanisms have been postulated to explain the link between infections and autoimmunity.

First, infections may up-regulate the expression of costimulators on APCs. If these cells are presenting self-antigens, the result may be a breakdown of anergy and activation of T cells specific for the self-antigens.

Second, some microbes may express antigens that have the same amino acid sequences as self-antigens. Immune responses against the microbial antigens may result in the activation of self-reactive lymphocytes. This phenomenon is called
molecular mimicry. A clear example of such mimicry is rheumatic heart disease, in which antibodies against streptococcal proteins cross-react with myocardial proteins and cause myocarditis. But more subtle molecular mimicry may be involved in classical autoimmune diseases as well. Microbes may induce other abnormalities that promote autoimmune reactions. Some viruses, such as Epstein-Barr virus (EBV) and HIV, cause polyclonal B-cell activation, which may result in production of autoantibodies. The tissue injury that is common in infections may release self-antigens and structurally alter self-antigens so that they are able to activate T cells that are not tolerant to these new, modified antigens. Infections may induce the production of cytokines that recruit lymphocytes, including potentially self-reactive lymphocytes, to sites of self-antigens. **General Features of Autoimmune Diseases:** Diseases caused by autoimmunity have some important general features.

- **Once an autoimmune disease has been induced it tends to be progressive,** sometimes with sporadic relapses and remissions, and the damage becomes inexorable. 

  One reason for this is that the immune system contains many intrinsic amplification loops that allow small numbers of antigen-specific lymphocytes to accomplish their task of eradicating complex infections. When the response is inappropriately directed against self-tissues, the very same amplification mechanisms exacerbate injury.

  Another reason for the persistence and progression of autoimmune disease is the phenomenon of epitope spreading. Infections, and even the initial autoimmune response, may damage tissues, release self-antigens and expose epitopes of the antigens that are normally concealed from the immune system. The result is continuing activation of lymphocytes that recognize these previously hidden epitopes; since these epitopes were not expressed normally, the lymphocytes did not become tolerant to them. The activation of such autoreactive T cells is referred to as epitope spreading because the immune response “spreads” to epitopes that were initially not recognized.

- **The clinical and pathologic manifestations of an autoimmune disease are determined by the nature of the underlying immune response.** T₉₁ responses are associated with destructive macrophage-rich inflammation and the production of antibodies that cause tissue damage by activating complement and binding to Fc receptors. T₉₁₇ responses are believed to underlie inflammatory lesions dominated by neutrophils as well as monocytes.

- **Different autoimmune diseases show substantial clinical, pathologic, and serologic overlaps.** For this reason, precise phenotypic classification of these disorders is often a challenge.

**Autoimmune diseases may be either:**

- Organ specific (response directed against a single component of a single tissue).
- A non-organ-specific autoimmune disease (response directed against a component that present in many tissues & organs throughout the body).

**Systemic lupus erythematosus (SLE):** SLE is an autoimmune disease and one of the "connective tissue disorders".

**Etiology & pathogenesis:** SLE is a complex disease of multifactorial origin including genetic, hormonal, & environmental factors. These insults lead to the apoptosis of
cells. Inadequate clearance of the nuclei of these cells results in a large burden of nuclear antigens. An underlying abnormality in B and T lymphocytes is responsible for defective tolerance, because of which self-reactive lymphocytes survive and remain functional. These lymphocytes are stimulated by self-nuclear antigens, and antibodies are produced against the antigens. Complexes of the antigens and antibodies bind to Fc receptors on B cells and dendritic cells, and may be internalized. The nucleic acid components engage Toll-like receptors (TLRs) and stimulate B cells to produce autoantibodies and activate dendritic cells to produce interferons and other cytokines, which further enhance the immune response and cause more apoptosis. The net result is a cycle of antigen release and immune activation resulting in the production of high-affinity autoantibodies.

**Mechanisms of Tissue Injury:** Regardless of the exact mechanisms by which autoantibodies are formed, they are clearly the mediators of tissue injury.

Most of the visceral lesions are caused by immune complexes (type III hypersensitivity).

Autoantibodies specific for red cells, white cells, and platelets opsonize these cells and promote their phagocytosis and lysis.

**Malar skin rash:** direct immunofluorescence microscopy reveals deposition of immunoglobulins (IgG, IgM) & complement at the dermo-epidermal junction. Histologically: there is characteristic liquefactive degeneration of the basal layer of epidermis, edema at the dermo-epidermal junction & mononuclear infiltrates around blood vessels & skin appendages.

**Renal disorder:** The basis of the glomerular damage is the deposition of immune complexes within glomeruli. Involvement of the heart showing mainly pericarditis, myocarditis & vascular lesions called Libman-Sacks endocarditis, which represent a nonbacterial verrucous endocarditis.

**Rheumatoid Disease (Rheumatoid Arthritis) (RA):** RA is a multi-system connective tissue disease in which the dominant effects are on the joints. It is characterized by the presence of a circulating autoantibody, "**Rheumatoid Factors**".

**Pathogenesis:** Although much remains uncertain, it is currently believed that rheumatoid arthritis is triggered by exposure of a genetically susceptible host to an arthritogenic antigen resulting in a breakdown of immunological self-tolerance and a chronic inflammatory reaction. In this manner, an acute arthritis is initiated, but it is the continuing autoimmune reaction, the activation of CD4+ helper T cells, and the local release of inflammatory mediators and cytokines that ultimately destroys the joint. About 80% of the patients have **rheumatoid factors (RF)** in their serum & synovial fluid. RF represents an autoantibody mainly of IgM class directed against the Fc portion of IgG. RF & IgG form immune complex that fix complement, attract neutrophils, & lead to injury by a type III hypersensitivity reaction.

**Pathologic changes:** In the early stage there will be rheumatoid synovitis. The synovium is swollen with prominent villous pattern. There is a great increase in chronic inflammatory cells mainly lymphocytes, plasma cells & macrophages with formation of lymphoid follicles. There is marked synovial hypertrophy & hyperplasia, often with increased vascularity due to angiogenesis. There is often fibrinous effusion in the joint space; the fibrin gets deposited on the synovial surfaces.
With time there is articular cartilage destruction with replacement by vascular granulation tissue (pannus). The latter grows across the surface of the articular cartilage from the edge of the joint. The inflammatory pannus causes focal destruction of the subjacent bone; this is manifested as "erosions" on radiographs. Following destruction of the articular cartilage & erosion of the subarticular bone, the pannus fills the joints space. Subsequent fibrosis & classification may cause permanent ankylosis of the affected joint.

**Amyloidosis:** Amyloid is the generic term for a variety of proteinaceous materials that are abnormally deposited in tissue interstitium causing clinical disorders.

**Morphology:** The diagnosis depends on identification by light microscopy of the material in biopsy. H & E stain shows amyloid as an amorphous, eosinophilic hyaline extracellular substance. It also takes up certain special stains; the most widely known of these is congo red stain, which gives pink to red color under ordinary light microscopy but characteristically green birefringence under polarizing microscopy. Electron microscopy of amyloid shows that it is composed of fibrils in a β-pleated sheet.

**Effects:** Progressive accumulation causes pressure atrophy at adjacent cells.

**Composition:** Amyloid is not a single chemical entity, there major & several minor biochemical forms could be found.

**Physical nature of amyloid:** 95% of amyloid component is nonbranching fibrils, with characteristic crossed β-pleated sheet conformation. The minor component is a nonfibrillar pentagonal glycoprotein (P-component) & proteoglycans, which form the remaining 5%.

**Chemical nature of amyloid:**

- **AL (amyloid light chain):** derived from plasma cell & contains Ig light chain. It is encountered with some forms of monoclonal B-cell proliferation.
- **AA (amyloid-associated):** derived from serum precursor protein synthesized by the liver (serum amyloid associated) (SAA). It is non-immunoglobulin protein and deposited in the setting of chronic inflammatory states.

Amyloidosis may be systemic (generalized) or may be localized to a single organ.

- **Systemic Amyloidosis:** may be:
  - Primary which is associated with immunocyte dyscrasia, or
  - Secondary as a complication at chronic diseases.

- **Primary Amyloidosis:** AL type, usually systemic. Examples include amyloidosis which is associated with multiple myeloma (a malignant neoplasm of plasma cells).

- **Reactive Systemic Amyloidosis (AA):** the distribution of the amyloid deposition in this pattern is systemic. It tends to be associated with chronic inflammation caused by autoimmune states such as rheumatoid arthritis and inflammatory bowel disease.

- **Heredofamilial Amyloidosis (AA):** examples include Familial Mediterranean Fever, which is an autosomal recessive febrile illness of unknown cause, associated with serosal inflammation such as the peritoneum, pleura & synovium.

- **Localized Amyloidosis:** (AL) limited to one organ or tissue that may produce detectable nodular masses or be evident only through microscope examination.

- **Endocrine Amyloid:** this form is found in medullary carcinoma of the thyroid, islet cell tumor of pancreas and pheochromocytoma.

- **Amyloid of Aging:** usually occurs in the age group 70-80 years and is called Senile Systemic Amyloidosis. The heart is predominantly involved.
Pathogenesis: Long standing tissue injury & inflammation cause macrophage activation & lead to elevated SAA levels through the influence of cytokines (IL₁, IL₆) on liver cells. Elevation of SAA levels alone does not lead to amyloidosis. It is believed that SAA is normally degraded to soluble end products by action of monocyte-derived enzymes. So individuals who develop amyloidosis have an enzyme defect that results in the incomplete breakdown of SAA, thus generating insoluble AA molecules.

Morphological effects on various organs

Kidneys: become large, pale, gray and firm. Amyloid is deposited in the glomeruli, peritubular tissue and in the wall of blood vessels.

Spleen: becomes firm, enlarged, pale and waxy on cut section.

Liver: is enlarged, pale and waxy. The deposition occurs in the space of Disse and surrounding blood vessels.