

LECTURE NOTES

HYPERSENSITIVITY

(Introduction, Types, Pathogenesis and Examples)

Hypersensitivity is defined as the *inappropriate/misdirected, undesirable, exaggerated 'immune-mediated response'* leading to 'immune-mediated tissue injury' and is caused by **perfectly normal** innate and/or adaptive immune responses to an Antigen.

All hypersensitivity reactions are characterized by (i) **Sensitization** and (ii) **Effector** phases. The **SENSITIZATION** phase requires that the host must have had either a previous exposure or a prolonged exposure to the antigen so that he or she can develop an immune response to the inciting antigen. However, the pathologic response associated with hypersensitivity reactions occurs in the **EFFECTOR PHASE** and is most commonly manifested as an inflammatory reaction or as cell lysis.

It is already known that, foreign antigens are trapped and processed by antigen presenting cells (APCs) and then presented to **helper T cells**.

Each T cell is covered by about 30,000 identical antigen receptors. If these **RECEPTORS** bind sufficient antigen in the correct manner, a **helper T cell** will respond and initiate an immune response. It does this, by secreting multiple cytokines (IL, TNF etc), dividing and differentiating exponentially, increasing its *Effector Response*. (*it is well known facts that the other antigen-responsive T-cell populations, like the B-cells and cytotoxic T cells, cannot respond to antigens unless they too are stimulated by helper T cells*).

Further, the **T-cell antigen Receptors** only recognize antigens associated with **MHC molecules (Major Histocompatibility Complex)**. They do not recognize or respond to free antigen molecules. Since over the T-helper cells, these antigen receptors are generated in a random way from exiting repertoire (not specific to any antigen), therefore, the strength of binding between an antigen and its receptors (its affinity) will also vary. Thus an antigen may be bound **strongly by some receptors and weakly by others**. If this binding strength is too weak, this may be insufficient to activate a T-cell.

T-helper cells and their response to Antigens

- ✓ **Four major Population of T-cells:** (i) helper, (ii) regulatory, (iii) cytotoxic or effector T-cells and (iv) B-cells
- ✓ **Four Key-Antigen Receptor** (over T-cells) – TCRs, BCRs, MHC-I and MHC-II
- ✓ **Effector Response:** based on interactions between two different members of this immunoglobulin superfamily, like TCRs & MHCs
- ✓ **CD3-Complex**
- ✓ **CD4 and CD8 proteins**
- ✓ **Co-stimulation**
- ✓ **Adhesion Molecules**
- ✓ **Cytokines**

CD4 and CD8

These are Two **additional proteins** closely associated with the TCR are called **CD4** and **CD8**. **CD4** is a single chain of 55 kDa, and **CD8** is a dimer of 68 kDa. These proteins CD4 and CD8 **determine the class of MHC molecule that can be recognized by the T cell**.

- **CD4⁺**, is usually found only on **helper T-cells**, and usually binds to **MHC class II** molecules on antigen-presenting cells.
- **CD8⁺**, in contrast, is found only on **cytotoxic T-cells** and binds **MHC class Ia** molecules on virus-infected or other abnormal cells.

Both CD4 and CD8 enhance TCR **signal transduction** by linking the T cell to an antigen presenting cell (APC) through the MHC.

ON the basis of (i) **Immunological mechanism** initiating the disease AND (ii) **Time-taken for the reaction**, the Hypersensitivity are classified into 4 types i.e. Type I, II, III and type IV hypersensitivity.

Type I, II, and III are variations of Antibody-mediated or Humoral-mediated while the fourth one type IV hypersensitivity is Cell-mediated.

TYPE-I Hypersensitivity

(also k.a. ANAPHYLACTIC / IMMEDIATE Type)

Type I hypersensitivity, is result of an immunoglobulin **IgE** (reaginic antibody) mediated immune-response directed against **environmental or exogenous antigens** (also k.a. Allergens) and also **parasitic antigens** causing the release of vasoactive mediators from IgE-sensitized mast cells and an acute inflammatory response. Primary cellular components involved are MAST CELLS and BASOPHILS.

It can have either systemic (e.g., anaphylaxis [bee sting]) and localized (e.g., allergic dermatitis) forms.

Type I hypersensitivity occurs in a previously sensitized host and is initially manifested as acute inflammatory process that occurs within minutes ("immediate hypersensitivity") of exposure to the specific antigen.

The basic pathogenesis involves a Sensitization phase and an Effector phase. The sensitization phase occurs during the initial exposure to an antigen (allergens), stimulating IgE production, antigen-specific IgE response, resulting in sensitization of the host by binding of antigen-specific IgE to Fc_ε receptors on the surface of mast cells.

On re-exposure (second exposure) or prolonged initial exposure to the IgE-specific antigen-in any sensitized host, cross-linking of 2 or more IgE molecules on surface of mast cells, activation of IgE and further degranulation of Mast cells / basophils take place, resulting in release of preformed and newly synthesized vasoactive amines (mediators) like histamine, prostaglandins, heparin, other chemotactic factors etc, responsible for effector phase & clinical manifestation.

The pathology is primarily acute, due to smooth muscle contractions, vascular leakages, hypotension, influx of inflammatory cells, mucus secretions, itching and rashes on skin. The late-phase and chronic reactions, often associated with repeated or prolonged antigen exposures, are largely the result of a more intense inflammatory cell infiltration (primarily eosinophils, neutrophils, macrophages, and T lymphocytes) and tissue damage.

Important disease examples includes:

- Systemic Anaphylaxis (Penicillin sensitivity)
- ATOPY (Hay fever, Asthma, Atopic Dermatitis, Eczema)
- ALLERGIES (Drug, food, parasitic allergens)

Anaphylaxis: refers to an acute systemic hypersensitivity type-I reaction to an antigen that is mediated by IgE and involves mast cell activation, resulting in a shock-like state often involving single to multiple organ systems. The clinical signs and pathologic changes attributable to a systemic anaphylactic reaction vary by species and often correlate to the primary shock organ in its most severe manifestation—death. The primary target tissues are blood vessels and smooth muscle.

In a **Localized type I Hypersensitivity** reaction, the clinical signs and pathologic findings are restricted to a specific tissue or organ. Localized reactions most commonly occur at epithelial surfaces such as the surfaces of the skin and mucosa of the respiratory and gastrointestinal tract. Species differences on the location of mast cells, the mediators contained within them, and the histamine receptor distribution on target tissue may explain the difference in manifestation of diseases seen among different species.

TYPE- II Hypersensitivity

(also k.a. CYTOTOXIC, antibody-mediated cytotoxic Hypersensitivity)

Type II, also known as Antibody-mediated Cytotoxic hypersensitivity, most often occurs as the result of the development of **IgG or IgM antibodies** directed against antigens on the surface of a cell or in a tissue, causing destruction of the tissue or cell by (i) Antibody-dependent Cellular Cytotoxicity (ADCC) or complement-mediated lysis or (2) altered cellular function without evidence of tissue or cell damage.

Antigens may be either endogenous (normal cellular or tissue protein) or exogenous (e.g., a drug or microbial protein adsorbed to the cell). The pathogenesis of many immune-mediated and **autoimmune diseases** is centered on the development of antireceptor or anti-surface antigen antibodies and a type II hypersensitivity reaction. The largest group of “cytotoxic” hypersensitivity reactions involves the **hematologic diseases**, with antibodies directed against antigens present on the surface of red blood cells and platelets.

Type II hypersensitivity reactions most frequently involve IgM and IgG Antibodies, that likely react to target cells like ERYTHROCYTES, LEUCOCYTES & PLATELETS, resulting in (i) Activation of Complement, (ii) Increased Phagocytic activity by OPSONIZATION or (iii) Stimulation of Killer Cell Activity-leading to destruction of target cells.

So, the Tissue destruction is by 3 mechanisms:

- Complement Activation: Complement-dependent reactions occur as a result of the **complement-activating capability of IgG and IgM**. Complement activation mediates cytotoxicity by either the (a) formation of the membrane attack complex (MAC) resulting in cell lysis, or (b) the fixation of **C3b fragments** (opsonization) to the surface, facilitating phagocytosis.
- Increased Phagocytosis by OPSONIZATION: Opsonization of cells by antibody makes them susceptible to destruction by macrophages, neutrophils, NK cells, and eosinophils.
- Antibody-dependent Cellular Cytotoxicity (ADCC)

Diseases, syndromes, auto-immune disorders for Type-II hypersensitivity / ADCC includes:

- Blood Transfusion Reactions
- Autoimmune Hemolytic Anaemia (Foals)
- Erythroblastosis foetalis and PEMPHIGUS
- Penicillin-induced Hemolytic Injury

TYPE- III Hypersensitivity

(also k.a. IMMUNE-COMPLEX MEDIATED Hypersensitivity)

TYPE- III Hypersensitivity is caused by the formation of insoluble antibody-antigen complexes (immune complexes) resulting in activation of the complement system and the development of an inflammatory reaction at the sites of immune complex deposition. Here "*Frustrated Phagocytes*" are encountered as the neutrophils and macrophages that are unable to phagocytise (eat) the immune complexes, so try to digest outside cell. As a result, these cells would degranulate in area of immune complex deposition and further trigger inflammation.

The immune-complexes are formed in such large quantities, that it cannot be adequately removed by R.E. System of body. As result, such complexes are precipitated around blood vessels, particularly in glomerular tuft. These immune-complexes fix & activate complement at sites of precipitation, activating complement factors C3a & C5a, release of vasoactive amines, resulting in increased vascular permeability and chemotactic attraction accumulation of neutrophils/PMN leucocytes & subsequent tissue damage. In presence of C3a, neutrophils release Hydrolytic enzymes as site of immune-complex deposition, that destroys ground substance, basement membranes & elastic tissue around blood vessels, resulting in severe inflamm changes like Oedema, Vasculitis, Haemorrhages, & necrosis.

Primarily, **IgG** plays role in this Hypersensitivity. Central role in immune-complex mediated injury is played by Activated COMPLEMET and NEUTROPHILS.

Based upon pattern of tissue-reactions, Type-III hypersensitivity can have TWO forms: It may have (1) **LOCALISED Form / ARTHUS Reaction** (e.g., cutaneous Arthus reaction; hypersensitivity pneumonitis in cattle; blue eye [anterior uveitis in dogs]) and (2) **GENERALISED or Systemic Form** (e.g., SERUM SICKNESS, Rheumatoid arthritis and systemic lupus erythematosus).

ARTHUS REACTION: is a localised hypersensitivity, occurs with introduction of antigen into an individual with high titer antibody (already sensitized). This event requires both complement & phagocytes.

Immune complexes are deposited beneath endothelium, where they activate Complement, attract Neutrophils and platelets. Neutrophils attempt phagocytosis, but saturate due to non-phagocytale basement membranes, hence releasing enzymes like proteases, collagenase, elastase so to damage digest basement membrane & surrounding tissue.

It peaks at 3-6 hours after exposure and histologically involve massive influx of neutrophils, edema, sometimes necrosis

Such localised reactions also seen in Occupational hazards like:

- Farmer's lung (thermophilic actinomycetes)
- Malt worker's lung (Aspergillus spores)
- Pigeon fancier's disease (avian proteins)
- Cheese washer's lung (Penicillium spores)
- Furrier's lung (fox fur)
- Laboratory technician's lung (rat urine proteins)

SERUM SICKNESS is a generalised systemic reaction that occurs from conditions like large amounts of antigen in blood circulation (possessing high-level of precipitating antibodies), eg. in cases such as injection of foreign serum. So immune-complexes are formed in circulation, most large sized complexes are phagocytised while many small/soluble complexes are deposited in various tissues/organis like skin, arteries, Glomeruli, heart, joints etc resulting in arteritis, urticarial skin rashes, glomerulonephritis, endocarditis , arthritis etc.

Serum sickness is usually ACUTE and transient immune complex disease which can be handled with removal of antigen source. However Chronic forms have been observed rarely.

TYPE- IV Hypersensitivity

(also k.a. DELAYED or CELL-MEDIATED Hypersensitivity)

It is purely a cell-mediated Immune reaction mediated by Specifically SENSITIZED T-Cells, without involvement of antibody or complement.

Type IV, also known as delayed-type hypersensitivity, is the result of activation of sensitized T lymphocytes to a specific antigen. The resulting immune response is either mediated by direct cytotoxicity by lymphocytes or by the release of cytokines that act primarily through macrophages to produce chronic inflammation. It is the underlying mechanism for tuberculin testing in cattle for bovine tuberculosis (*Mycobacterium bovis*) and for allergic contact hypersensitivity and granulomatous inflammatory responses.

The hallmarks of Type-IV Sensitivity are:

- i. Its initiation by T-cells (as distinct from antibody)
- ii. The Delay required for reaction to develop; and,
- iii. Recruitment of MACROPHAGES and/or Cytotoxic T-Lymphocytes (instead of neutrophils/eosinophils etc) as primary cellular components of infiltrate that surrounds the site of inflammation.

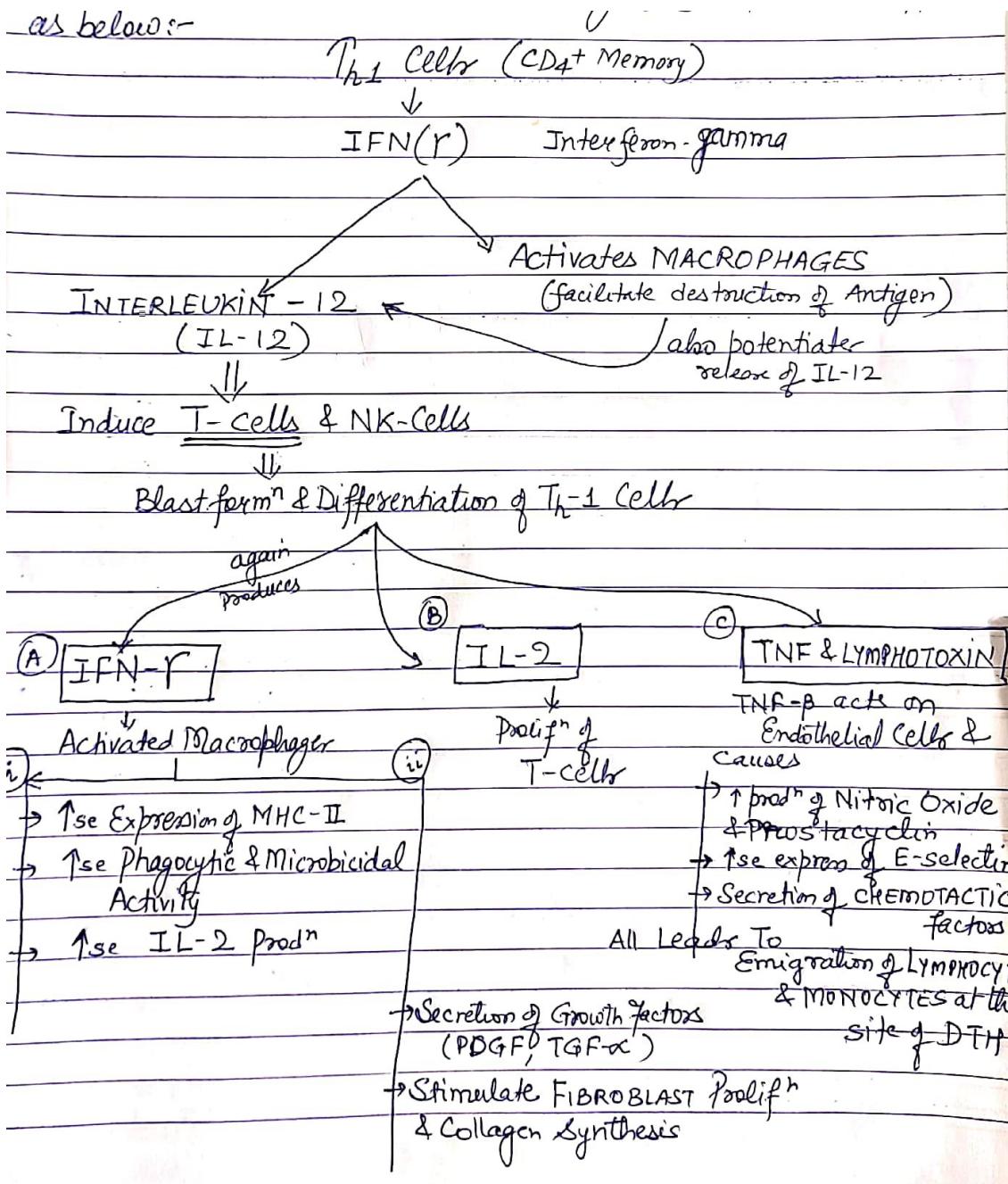
Type-IV Hypersensitivity are basically of TWO Types:

- I. **DELAYED TYPE HYPERSENSITIVITY (DTH)**: initiated by **CD⁴⁺ T-helper** cells, which secrete **Cytokines**, that further recruit Major Effector Cells (**Macrophages**), which induces the changes.

Mechanism of DTH is classically explained in **Classical Tuberculin Test** or **Tuberculin Reaction**, induced in animal which is already sensitized to a TB antigen due to prior infection.

When Tuberculin Ag is injected to a previously sensitized animal, DTH reaction is characterized by *Hyperemia, Oedema, and Mononuclear Cell Infiltration*, followed by *Induration (Hardening/Swelling)* at site of injection in about 24-72 hrs. Induration is due to deposition of FIBRIN around the blood vessels.

After 1st exposure to TB bacilli, Sensitized CD4⁺ Th₁ Lymphocytes are generated that remain in circulation for years as memory cells. On Subsequent exposure (transdermal tuberculin-TB antigen injection-in case of Tuberculin Test), the memory Th1 lymphocytes (formed in 1st exposure) respond to processed antigen on APCs and are ACTIVATED to further undergo '**blast**' formation and proliferation, that in turn liberate various CYTOKINES and their effects as shown below:



Interleukin-12 (IL-12) produced by macrophages; and causes differentiation of Th1 cells, further producing other cytokines.. IL-12 also induces IFN- γ secretion by T-cells and NK-cells.

Interferon-gamma (IFN- γ): a powerful activator of macrophages and further increases their IL-12 production.

Activated macrophages express more MHC class-II molecules on surface – leading to increase Antigen presentation. They also have increased Phagocytic and Microbial activity. Activated macrophages also secrete growth factors PDGF and TGF-alpha, and these stimulate FIBROBLAST PROLIFERATION and Increased COLLAGEN Synthesis. If macrophage activation sustains, Fibrosis occurs.

Interleukin-12 (IL-2): It causes proliferation of T-cells, accumulated at sites of DTH.

Tumour Necrosis factor (TNF-alpha) and Lymphotoxin (TNF-beta): These produce important effect on Endothelial cells as, (i) it causes increased secretion of **Nitric Oxide and Prostacyclins** and thus induces increased blood flow through local vasodilation, (ii) also cause increase expression of E-selectin adhesion molecules, promoting mononuclear cell attachment. (iii) Further make Endothelial Cells secrete Chemotactic Factors such as IL-8.

II. T-Cell mediated CYTOTOXICITY

Direct Cell-Cytotoxicity By Direct Recognition : prototype example of GRAFT REJECTION.

Here, **CD⁸⁺ T-cell** of host recognise directly the **MHC class-I** molecules on surface of **APCs of Donor**.

The Dendritic Cells are most important APCs in Direct Recognition. Dendritic cells in donor organ - express high MHC-I & MHC-II-but also express **Co-stimulatory molecules B7-1 (CD80) and B7-2 (CD-86)**. In T-cell mediated cytotoxicity, the **CD⁸⁺ T-cell** kills the antigen bearing target cells.

As in few viral infections – viral antigens are processed intracellularly and MHC-class I molecules bind to intracellular antigen and present them to **CD8+ T-cells**. **CD⁸⁺ Lymphocytes** are Main Effector Cells called **Cytotoxic T Lymphocytes** (CTLs) which play critical role in the lysis of infected cells (by viral replication) is completed.

Cytotoxic T-lymphocyte trigger action by 2 mechanisms :-
(Lysis of Infected Cell)

1) PERFORIN - GRANZYME-dependent killing :-

12-18 No. of perforin molecules combine to form Large molecule through polymerization (k.a Membrane Attack Complex) which inserts itself into the target cell & make PORES. These pores allow water into the cell - resulting Osmotic Lysis.

Lymphocytic granules also contain variety of PROTEASES called GRANZYMES. These are also delivered into target cells through Perforin pores. Once inside target cells, Granzyme activate target cell APOPTOSIS.

2) Fas - Fas ligand dependent killing :-

T-lympho also express Fas-ligand that bind to Fas on target cell activating death signals via DISC & lead to APOPTOSIS.



