



# **IMMUNOLOGY SUMMARY SHEETS**

**DR HARISH THAMPY**

# LYMPHATIC SYSTEM

At any one moment, most of the lymphocytes are not in blood but in the lymphoid organs.

## Primary lymphoid organs

- 1) Thymus
- 2) (Bursa of Fabricius – in birds)

## Secondary lymphoid organs

- 1) Lymph nodes (filters lymph)
- 2) Spleen (filters blood)
- 3) MALT eg. Peyer's patches in ileum of SI – deal with pathogens on overlying epithelium
- 4) Tonsils – deal with pathogens on overlying epithelium

**Lymphoid organs** – made up of lymphoid tissue (is a tissue with huge masses of lymphocytes). They all have a delicate skeleton of fibroblasts and reticular fibres (fine collagen fibres) that support the lymphocyte.

## T- lymphocyte: Cell-mediated Immunity – Thymus dependant

- Made in RBM, migrates to thymus, matures/ programmed, then moves to 2<sup>o</sup> lymphoid organs
- Rare Condition: Di George's syndromes: baby born without thymus → so: T-lymphocytes are not mature so lymphoid organs do not contain T-lymphocytes

## B- lymphocyte: Antibody (Humoral) Response

- Produced AND programmed in RBM
- NB: RBM not really a primary lymphoid organ since is mostly made up of blood precursors and other blood cells with relatively few lymphocytes compared to the other lymphoid organs. It is however sometimes thought of a primary since it is making/programming the B-lymphocytes

*Lymphocytes can only populate secondary lymphoid organs once they have matured/ programmed in primary*

Most of lymphocytes in secondary organs are NOT originating from primary since a mature lymphocyte can migrate/ programmed from primary to secondary and then proliferate: ie. clone population. But all lymphocytes in secondary organs have ancestors that once were matured/ programmed in primary.

Only lymph nodes (out of all the lymphoid organs) are part of lymphatic system.

- Are concentrated near groin and mammary glands. Quite small. Bean shaped structures

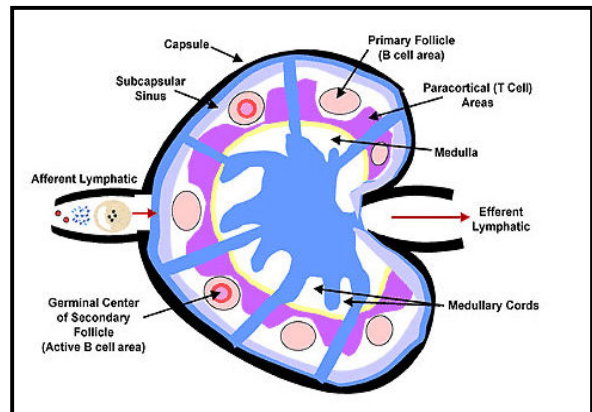
1 or 2 efferent lymphatics per node

The blood vessels in/out are just for nutrient/ oxygen supply

- 1) **Outer cortex** = cortex nearest capsule. Contains B-lymphocytes
- 2) **Deep cortex** (aka paracortex) = cortex nearest medulla. Contains T-lymphocytes

Lymph flows from afferent lymphatics to subcapsular sinus and

- a. Around subcapsular sinus to medullary spaces to efferent OR
- b. From subcapsular sinus into trabecular sinuses which extend through cortex and run parallel to trabeculae to medullary spaces to efferent



## How does node filter lymph?

### 1) Mechanical Filtration:

- Fluid leaving narrow tube (aff lymphatic) is entering broad space (subcapsular sinus)
- So:
- Decrease flow rate
- So: any debris (bacteria, carbon particles etc) settles out onto lymphocytes and cells of cortex
- Then from broad space to medullary spaces
- Further decrease of flow rate
- Settling out of debris onto cells of medullary cords

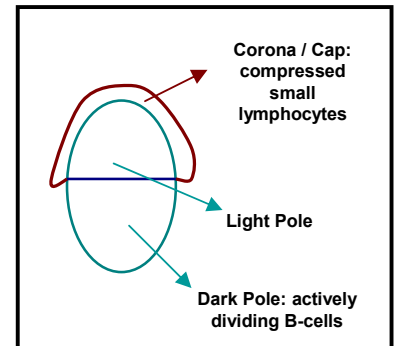
(Analogous to septic tank)

### 2) Biological filtration:

- Phagocytosis by macrophages (are star-shaped cells with processes that attach to other macrophages and to medullary cords ie 'fixed')
- If there is an antigen in the filtered debris: presented to immune system – response mounted

### LYMPHOID NODULES:

- aka germinal centres/ lymphoid follicles
- Found in cortex. Oval shaped. Made up of rapidly dividing B-lymphocytes.
- Thus, if found: indicates body is having an antibody-mediated immune response.
- Since made up of B-lymphocytes then only found in *outer cortex*.
- It is thought that the lymphatic nodules are not consisting of plasma cells but rather memory cells
  - The plasma cells are:
    - ⇒ Populating the medullary cords
    - ⇒ Leave in efferent lymphatics to rest of body → has local effect there



- As a general rule: the cap/ corona faces the source of the antigen. Thus in the lymph nodes, this is usu the subcapsular sinus.
- NB: if viral infection: then rapid T- lymphocyte proliferation but does not produce these characteristic nodule structures.

### SPLEEN:

- Filters blood. Size of clenched fist. In left hypochondrium. Easily damaged. Has indentations from many surrounding organs. Has a hilus where splenic artery, vein and the efferent lymphatic vessels enter/ leave.
- Made up of:
  - 1) Red pulp – with the splenic cords and splenic sinusoids. Blood is filtered here.
    - Functions:
      - ⇒ Macrophages in the red pulp take out debris, damaged/ neoplastic cells, bacteria etc
      - ⇒ Stores platelets (upto 1/3 of body's total)
      - ⇒ Haemopoiesis in fetal life
  - 2) White pulp – lymphoid tissue (and macrophages). So, if antigen is present in blood, then immune response can be mounted. Again has reticular fibre skeleton. The white pulp forms a sleeve around a small 'central artery' (branch of splenic artery) → if Ab response, lymphatic nodules may be seen in white pulp. This time cap faces the red pulp (antigen source).

### MALT:

- Mostly in submucosa of eg, GI, resp tracts
- If Ab-response, then cap of nodule points to lumen of tract

### TONSILS:

- Are in a sense MALT but considered as a separate secondary lymphoid organ.
- Protects mainly the resp tract → Waldeyer's Ring:
  - 2 x palatine, 2x lingual and 2x pharyngeal (adenoid) tonsils
- In the tonsils are crypts that can fill up with debris/ pus. Seen macroscopically as white spots on tonsils. Reason for crypts: allows lymphocytes to recognise and mount an immune response to any present antigen.
- Lymphatic nodules of tonsil are very big – cap faces to source ie. to crypt.

### THYMUS:

- In mediastinum. Made up of 2 lobes. Big in baby. After puberty fills with fatty connective tissue.
- Has thin capsule
- Divided into lobules
- Has cortex and medulla (NOT same as lymph node). Lymphocytes more closely packed in cortex than medulla.
- Is made up of T-lymphocytes undergoing maturation/ programming thus NO lymphatic nodules seen.
- Programming occurs in cortex
- Function of medulla is unclear
- Also present are supporting cells (aka thymus nurse cells) → secretes chemicals to help maturation of T-lymphocytes
- These degenerate and forms clumps called thymic corpuscles (aka Hassall's corpuscles)

## CELLS OF THE IMMUNE SYSTEM

- 1) Macrophages
  - a. From monocytes → part of reticuloendothelial system (RES)
  - b. Function: phagocytosis → have receptors for IgG (Fc<sub>γ</sub> receptors I/ II/ III = CD64/32/16) and receptor for C3b (CD35)
  - c. Monocytes → 10% of all WBCs, large, mainly nucleus/ little cytoplasm → become macrophages
  - d. Phagocytosis greatly enhanced by coating of microbe with Abs/ C3b (latter is main one)
- 2) Polymorphonuclear leucocytes (neutrophils)
  - a. 70% of all WBCs – major type of granulocyte – last hours-days (BM continually produces)
  - b. Also has Fc<sub>γ</sub> receptors
- 3) Eosinophils
  - a. 3% of all WBCs
  - b. Function is aimed to target parasitic infections (helminths) but also involved in hypersensitivity
- 4) Mast Cells
  - a. Have a circulating counterpart called basophils (but are diff cells/ diff origins)
  - b. Function is aimed to target parasitic infections (helminths) but also involved in hypersensitivity
  - c. Cross-linking of membrane bound IgEs by Ag/ allergen causes degradation of preformed mediators eg histamine and later on get phospholipase A2 activation and thus get AA breakdown into newly synthesised mediators → attracts eosinophils that release proteases, free-radicals etc (accounts for most of damage seen in hypersensitivity)
- 5) Lymphocytes
  - a. 15% of all WBCs (70% T-cells, 15% B-cells, 15% NK cells)
  - b. NK Cells:
    - i. Distinct type of lymphocyte – are Ab-dependent killer cells (has Fc<sub>γ</sub>-III receptor)
    - ii. More info on this cell can be found in Innate Immunity Section
  - c. B-cells:
    - i. Have molecules of specific Ab on surface (sIgM/D) → upon stimulation get clonal expansion and transformation of plasma cells → makes Abs (IgM then IgG)
    - ii. Plasma cells make only one type of Ab (κκ or λλ)
  - d. T-cells:
    - i. Big nucleus, little rim of cytoplasm
    - ii. Transform into large blast cells
    - iii. CD25 (IL-2) receptor → growth and expansion of T-cell population (IL-2 is a T-cell GF)
    - iv. CD4:CD8 (2/3 : 1/3) → both have T-cell receptor
- 6) Dendritic Cells
  - a. Are scattered around body eg Langerhan cells of dermis
  - b. Are APCs with long cytoplasmic processes (incrs SA for Ag uptake)
  - c. Are crucial in processing and presenting (p/p) of Ags to T<sub>H</sub> cell in association with MHC-II
  - d. These cells decide whether to make immune response or not, determines type of resp (T<sub>H</sub>1 vs T<sub>H</sub>2 and CD4/ CD8)
  - e. Once in lymph node, meets with T-cells – activation – effectors cells (eg T<sub>C</sub> cells) acquire specific adhesion molecules that allow them to home to specific site that DC came from → DCs control the pattern of homing receptors on T-cells

### MAJOR HISTOCOMPATIBILITY COMPLEX ANTIGENS (MHC) ANTIGENS:

- All plasma membrane surfaces of all body cells (except RBCs) are 'self-antigens', the **major histocompatibility complex antigens**
- Are integral membrane glycoproteins
- Also called *human leukocyte associated (HLA) antigens* since first identified on WBCs.
- Except for identical twins, MHC antigens are unique.
- MHC antigens → reason that transplanted tissues may be rejected
- Normal function: to hep T-cells to recognize antigen is foreign, not self
- **Two** types of MHC antigens: class I and class II

**MHC Class I:** built into plasma membranes of all body cells (except RBCs)

- Function: all cells sample their internal contents and presents them to immune sys in assoc with MHC-I
- If contents of the cell are purely self then no attraction of T cells occurs
- If eg. virally infected cell, then the T<sub>C</sub> cells can recognize the viral antigens (9-11 amino acids) presented by the Class-I MHC and the cells are killed. Unfortunately foreign MHC is seen by the immune system so that the cells in transplants activate the killer cells in a similar manner as virally infected cells. The end result is the same, cell death (and transplant rejection).
- There are 6 MHC-1 antigens: 2A, 2B, 2C → many different types of each. One A from mum and one from dad etc.

**MHC-Class II:** appears only on surface of antigen presenting cells (APC) – B-cells, macrophages, dendritic cells

- The function of these proteins is to **present an antigen to T helper cells** to activate an immune response, which will provide both humoral (antibody) and cell mediated immunity
- The class II MHC consists 3 pairs again: 2 of DP, 2 of DQ and 2 of DR
- The processed antigen consists of a small peptide.
- Every MHC class II on the surface of a cell contains an antigen fragment because the MHC II is not expressed on the surface until it has antigen in the cleft. MHC-II picks up the antigen that has been ingested (phagocytosed) by the APC. It does not present MHC-2 alone.

## INNATE IMMUNITY / NON-SPECIFIC IMMUNITY

For immediate protection regardless of type of invader → Splits into two:  
1<sup>st</sup> line of defence and 2<sup>nd</sup> lines of defence

### FIRST LINE OF DEFENCE (SKIN AND MUCOUS MEMBRANES):

- are mechanical & chemical barriers to discourage pathogens from entering body

- 1) Keratinised **epidermis**: physical barrier. Periodic shedding of skin removes microbes on surface.
- 2) Epithelial layer of **mucous membranes** (line body cavities) secretes fluid **mucus** to lubricate/ moisten cavity. Its viscosity traps microbes/ foreign species.
- 3) The mucous membrane of nose: mucous coated **hairs** – trap and filter microbes, pollutants & dust from inhaled air.
- 4) Mucous membrane of upper respiratory tract contains **cilia** – microscopic hairlike projections used to move mucous to throat. Coughing/ sneezing accelerates movement of mucociliary escalator.
- 5) **Lacrimal apparatus**: makes and drains away tears in response to irritants. Blinking spreads tears over eyeball surface – continual washing action of tears → dilutes microbes; does not allow them to settle
- 6) **Saliva**: produced by salivary glands. Washes microbes from the teeth and the mucous membrane of the mouth.
- 7) Urethra cleansed by **flow of urine**: retards microbial settling (colonisation). Vaginal secretions does equivalent in females. **Vaginal secretions**: slightly acidic.
- 8) **Defecation and vomiting**: expels microbes. Example: microbial toxins irritate lining of lower gastrointestinal tract; smooth muscle contracts violently; diarrhoea expels microbes.
- 9) Sebaceous (oil) glands of the skin produce **sebum**: forms protective film over skin surface – unsaturated fatty acids in sebum inhibit growth of some pathogenic bacteria and fungi
- 10) **Acidity** of skin (pH 3-5) caused partly by fatty acids and lactic acid secretion.
- 11) **Perspiration**: helps flush microbes from surface of skin. Also contains **lysosyme**: enzyme capable of breaking down some cell walls of some bacteria.
- 12) **Lysosyme** also found in tears, saliva, nasal secretions & tissue fluids.
- 13) **Gastric juice**: mix of HCl, enzymes and mucous. Strong acid (pH: 1.2 → 3.0) kills many bacteria & their toxins.

### SECOND LINE OF DEFENCE:

- If microbe has penetrated 1<sup>st</sup> line of defence

- 1) **Antimicrobial proteins – recently coined term is Defensins (group of proteins with structural similarities that are naturally capable of combating infection):**
  - a. **Interferons (IFNs)**: are cytokines produced and released from most cells infected by viruses. Diffuses to neighbouring infected/ uninfected cells and binds to surface receptors inducing pathways that prevent protein synthesis in that cell. So viral proteins cannot be made, though nor can it make host cell proteins – ie cell is sacrificed to save surrounding cells. Also IFN has autocrine effect and some diffuses into blood stream to act on further away cells – ie endocrine agent too.
    - o **NB**: IFNs cannot stop attachment and penetration – only replication. Viruses only cause disease by replication.
    - o Three types of INF:  $\alpha$ ,  $\beta$ ,  $\gamma$ . Latter is mainly a T-cell product though is also made by NK cells.
  - b. **Complement**: group of normally inactive proteins in blood plasma and on plasma membranes make up **complement system**. When activated: enhances certain immune/ allergic/ inflammatory reactions.
  - c. **Transferrins**: iron-binding proteins – inhibit growth of some bacteria by reducing amount of available iron.
- 2) **Natural killer (NK) cells:**
  - Are lymphocytes that lack membrane molecules that identify B-cells and T-cells
  - Have ability to kill wide variety of infectious microbes. Make up 15% of lymphocytes
  - Found in blood but also present in spleen, lymph nodes and red bone marrow too
  - NK cells attack cells displaying no MHC-1 antigens (viruses down-regulate MHC-1 to ensure own survival)
  - NK cells achieve cell destruction by emperipolesis (migrates into cell it is going to kill) and releases perforins from the inside (cytolysis)
  - They also secrete IFN- $\gamma$ .
- 3) **Phagocytosis:**
  - Ingestion of microbes/ particulate matter → greatly enhanced by Abs and C3b (ie opsonisation)
  - **Two** major cells: **neutrophils** and **macrophages**. Latter – scavenger cells; develop from monocytes
  - Macrophages can be: **wandering macrophages** or can be **fixed macrophages** (latter stand guard in specific tissues eg alveolar macrophages, Kupffer cells of liver etc)
  - Stages: chemotaxis, adherence, ingestion, digestion, killing, expelling/ exocytosis of residual bodies
    - o Vesicle: phagosome and lysosome fuse → phagolysosome
    - o Also: phagocytes produces lethal oxidants in process called **oxidative/ respiratory burst** → reduction of O<sub>2</sub> to free oxygen radicals esp superoxide ion that gets converted to H<sub>2</sub>O<sub>2</sub> by superoxide dysmutase.
    - o Both of these are bactericidal but not in sufficient quantities to kill so instead MPO (myeloperoxidase) found in neutrophil granules react a halide eg Cl with H<sub>2</sub>O<sub>2</sub> to make HOCl → potent antimicrobial (all occurs in localised fashion within phagolysosome).
  - Macrophages have receptors for IgG (Fc $\gamma$ - receptors I/ II/ III = CD64/32/16) and receptor for C3b (CD35)

#### 4) Inflammation:

##### A. ACUTE INFLAMMATION

- The immediate/ early non-specific response of living tissue to injury
- Traps microbes in localised area and brings in phagocytes etc
- Aetiology:
  - ⇒ Infective, physical (temp, radiation), chemical, immunological (eg hypersensitivity), ischaemic
- 3 main features:
  - 1) Vascular changes → vasodilation and incrs permeability
  - 2) Because of (1) get formation of fluid exudates
  - 3) And then formation of cellular exudates (leukocyte emigration and accumulation)
    - Then get tissue repair after (not really part of inflammation)
- (1) (2)
  - ⇒ Initial transient arteriolar vasoconstriction seen but then vasodilation due to mediators (so get erythema and warmth)
  - ⇒ So get ↑ intravasc press so ↑ fluid filtration – so transudate (little protein)
  - ⇒ But then get ↑ permeability so get proteins out ie exudate (Abs, fibrinogen, complement, cells etc leave) and water follows so get oedema
    - Permeability increases transiently by EC contraction (widened intercellular gaps) due to histamine/ LTs/ kinins and increases in a more sustained fashion by direct EC injury (necrosis/ detachment)
- (3)
  - ⇒ Ultimate aim of (1)(2) is to bring in WBCs – mainly phagocytes (initially are neutrophils then macrophages >24hrs)
  - ⇒ 4 stages:
    1. Margination and Rolling → vasodilation causes ↓ blood flow and so cells settle to periphery of blood column (usu is cell-poor) and sticks transiently to ECs (rolling) due to selectins on WBC binding to addressins on EC
    2. Adhesions and Transmigration → more firm adhesions b/w VCAMs on EC and integrins on WBC – then get diapedesis (WBC moves b/w EC cells and degrades b memb by collagenases to go subendothelialy)
    3. Chemotaxis and Activation → WBC follows chemical gradient – chemokines bind to receptors on WBC – get signal transduction → synthesis of contractile elements
    4. Phagocytosis (see before)

Substances that contribute to vasodilation and increased permeability and inflammation in general:

- Blood mediators → end-products of 3 cascades: kinin system, complement system (C3a/ C5a potent chemokines), fibrinolytic system (get FDPs), coagulation system
  - All 4 systems started by activation of Hageman factor (XII) which occurs in acute inflamm
- Tissue Mediators:
  - Histamine (mast cells) and 5-HT (basophils/ platelets) → SMC contraction (extrvasc) and SMC dilation (vasc)
  - Arachidonic Acid metabolites → COX (PGs, TXA, PC) or LOC (LTs)
  - Lysosomal components
  - Other lymphokines (IL-1, TNF, IFN) and misc (NO, PAF, free-radicals)
- 5 characteristics of inflammation:
  1. Redness – increased blood flow (rubor)
  2. Heat – increased blood flow and increased metabolic reactions (calor)
  3. Pain – injury/ toxic irritation of nerve endings by toxins, kinins, pressure of oedema (dolor)
  4. Swelling – due to increased permeability (tumour)
  5. May also result in loss of function (functio laesia)

##### B. CHRONIC INFLAMMATION

- 3 key features:
  1. Infiltration by ch inflamm cells (lymphocytes, macrophages, plasma cells)
  2. Tissue destruction
  3. Proliferation/ repair – angiogenesis, granulation tissue/ fibrosis
- (1) and (2) occur simultaneously – called ORGANISATION
- Aetiology:
  1. After acute infection → acute response cannot be resolves since a) healing process disturbed (eg recurrent bouts of injury in duodenal ulcer) or b) persistent/ prolonged infections (eg mycobacteria, viruses (eg Hep B), protazoa (amoebiasia), fungi...)
  2. Prolonged exposure to toxic agents eg silicone
  3. Autoimmune diseases eg RA, thyroiditis
- Granulomatous Inflammation: distinctive histological type – get formation of granulomas
  - Can get (but not always) central caseating necrosis with surrounding activated macrophages (Langhan Giant cells/ epithelioid cells), cuff of surrounding lymphocytes (mainly CD8), then general connective tissue
  - Seems to be an immune response to indigestible material → can be infection, foreign material, idiopathic (eg sarcoidosis, Crohn's)
  - Tends to cause destruction and accompanied scarring and so deformity cf acute inflamm where can get complete healing

- 5) **Fever:**
- See Acute Phase Response section

**ACUTE PHASE RESPONSE:**

- Constellation of systemic responses which accompany trauma and inflammation of all causes
- Allows inflammation and healing to progress optimally and thus increase survival
- Triggers: infection, burns, surgery, necrosis → ie any kind of serious insult
- There is a **primary cellular reaction** → stimulation of macrophages, ECs, epith cells, fibroblasts etc
- There is also release of a number of **chemical mediators** → main ones are IL-1, IL-6, TNF, IFN- $\gamma$
- This results in **secondary systemic reactions viz:**

**1. Fever**

- Acts to enhance many cellular functions integral to inflammation and host survival
  - Enhanced bactericidal activity of neutrophils
  - Enhanced viricidal effects (intensifies action of IFN)
  - Enhanced B and T-cell function
  - Increases rate of reactions of tissue repair
- Mechanism:
  - Occurs when phagocytes kill bacteria → endotoxins released.
  - These stimulates phagocyte and other inflamm cells to make endogenous pyrogens → are cytokines incl IL-1, IL-6, TNF, IFN- $\gamma$
  - These act on OVLT in brain → this in turn activates pre-optic hypothalamus which releases PGs esp PGE<sub>2</sub> (inhibited by NSAIDs)
  - PGs act on endogenous pyrogen (EP) receptors on anterior hypothalamus to reset the 'thermostat'

**2. Leucocytosis:**

- Esp if cause of APR is infection
- Mainly get neutrophilia
  - IL-1 ↑ release from BM stores
  - IL-6 ↑ GM-CSF production and ↑receptors for GM-CSF on neutrophils ie maintains the increase

**3. Glucocorticoid Levels Rise:**

- Due to:
  - IL-1 ↑ ACTH release from pituitary and g/c release from adrenals
  - IL-6 ↑ g/c release from adrenals
- Imp to host survival eg Addisonian pts may succumb to infections due to poor glucocorticoid response

**4. Amino Acid Response:**

- Incrs proteolysis (initially from muscle)
- AAs used to fuel repair, cell proliferation, Ab synthesis, acute phase protein synthesis etc

**5. Iron/ Zinc Response:**

- Rapid fall in plasma iron (1hr) due to sequestration in damaged tissues
  - Due to lactoferrin released by neutrophils in response to IL-1
  - Decrs iron: anti-microbial effects
- Also get fall in plasma zinc due to hepatocyte sequestration (binds to intracellular metallothionein which is produced in resp to IL-6)

**6. Acute Phase Protein Response:**

- Involves many plasma proteins (~30)
- Can divide into 3 main groups based on magnitude of increase
 

○ 50% Increase	48-72hrs	C3, C4, caeruloplasmin
○ 2-4 Fold Increase	10-24hrs	$\alpha$ 1AT, $\alpha$ 1ACT, fibrinogen
○ 100 Fold Increase	6-10hrs	CRP, SAA
- CRP is good thing to measure as it increases rapidly by large amount and can monitor recovery by its decrease
- SAA = serum amyloid protein
- Can also divide APPs into 3 functional groups → mediators (eg CRP, thrombin, clotting factors), inhibitors (eg anti-thrombin, anti-trypsin, anti-chymotrypsin), and scavengers (SAA, haptoglobin etc)
- Overall effects of APPs are to balance up to mediate inflammation and promote healing
- Made quantitatively most in liver but qualitatively more extra-hepatically (targeted production by eg macrophage at site)

## ADAPTIVE IMMUNITY/ SPECIFIC IMMUNITY

- The '3<sup>rd</sup> line of defence'

### 'THIRD LINE OF DEFENCE' (SPECIFIC INTERNAL DEFENCES):

- Called **specific resistance/ immunity**.
- Substances recognised as foreign and provoke immune response: called **antigens (Ags)**
- Differences between this defence line and those above: (1) *specificity* for particular antigens – also involves distinguishing self from non-self molecules; (2) *memory* for most previously encountered antigens – a second encounter prompts a more rapid and vigorous response; (3) *diversity* of lymphocytes to recognise diversity of antigens; (4) *adaptivity*: the best response is chosen to eliminate pathogen

### TYPES OF IMMUNE RESPONSES:

- Two kinds of closely allied responses, both triggered by antigens:
- **Cell-mediated immune response**: CD8+ T<sub>C</sub>-cells proliferate into 'killer' T-cells – directly attack invading antigen.
- **Antibody-mediated (humoral) immune responses**: B-cells transform into plasma cells, which synthesise and secrete specific proteins called **antibodies (Abs)** or **immunoglobulins**.
- To some extent – each type of response specialised to deal with certain invaders:
- Cell-mediated immunity particularly effective against: (1) intracellular pathogens residing in host cell (primarily fungi, parasites and viruses); (2) some cancer cells; and (3) foreign tissue transplants
- Thus CMI always involves **cells attacking cells**.
- Antibody-mediated immunity works mainly against: (1) antigens present in body fluids and (2) extracellular pathogens that multiply in body fluids but rarely enter body cells (**primarily bacteria**).
- Often a pathogen may provoke both types of immune response.

## HUMORAL RESPONSES

### B-CELLS:

- Develop from pluripotent stem cells originating from red bone marrow
- B-cells complete development into mature, immunocompetent cells in bone marrow. Process continues throughout lifetime
- Before B-cells leave marrow they acquire several distinctive surface proteins.
- Some function as **antigen receptors**: molecules capable of recognising specific antigens
- When a B cell is stimulated by an antigen that it encounters in the body fluids, it transforms, with the aid of a helper T cell, into a larger cell called a blast cell. (ie no need for processing and presenting of antigen)
- The blast cell begins to divide rapidly, forming a clone of identical cells.
- Some of these transform further into **plasma cells** → in essence, antibody-producing factories.
- These plasma cells produce a single type of antigen-specific antibody at a rate of about 2,000 antibodies per second. The antibodies then circulate through the body fluids, attacking the triggering antigen.
- NB: plasma cells rarely found in plasma since out in tissues by then
- Not all of the cells from the clone formed from the original B cell transform into antibody-producing plasma cells; some serve as so-called **memory cells**.
- These closely resemble the original B cell, but they can respond more quickly to a second invasion by the same antigen than can the original cell.
- NB. B-cells (and to an extent, plasma cells) are fixed in tissues – it is the Abs that circulates

### STRUCTURE AND FUNCTION OF ANTIGENS:

- Antigen: aka '*immunogens*' – 2 important characteristics: immunogenicity and reactivity.
- **Immunogenicity**: ability to provoke immune response by stimulating production of specific antibodies and/or the proliferation of specific T-cells (Antigen = *antibody generator*)
- **Reactivity**: ability of antigen to react specifically with antibodies/ cells it provoked.
- Strictly speaking, antigens only have reactivity. **Complete antigens** have both reactivity and immunogenicity.
- Entire microbes or part of microbes may act as antigens.
- Bacterial structures such as flagella, capsules and cell walls are antigenic as are their toxins.
- Non-microbial examples of antigens: pollen, egg white, incompatible blood cells, transplanted tissues/ organs.
- Once antigens get past innate defence system, follow one of three routes:
  - ⇒ Most antigens enter bloodstream (eg. through injured blood vessel) and deposited in spleen
  - ⇒ Antigens that penetrate skin enter lymphatic vessels and reach lymph nodes
  - ⇒ Antigens that penetrate mucous membranes lodge in the mucosa-associated lymphoid tissue (MALT).
- Antigen: large, complex molecules, most frequently protein.
- Nucleic acids, lipoproteins, glycoproteins and some large polysaccharides may also act as antigens.
- T-cells only respond only to antigens that include protein.
- B-cells respond to variety of antigens, made of proteins, nucleic acids, lipids and carbohydrates.
- Large molecules with simple repeating units are not antigenic (eg. cellulose, plastics). This is why plastics are used for artificial heart valves/ joints.
- **Hapten**: smaller substance that lacks immunogenicity but has reactivity.

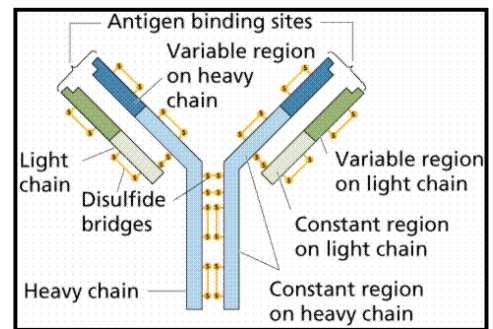
- Hapten – can stimulate immune response only when attached to large carrier molecule. Example: small lipid toxin in poison ivy, triggers immune response after combining with a body protein.
- Some drugs (eg. penicillin) may combine with proteins in body to form immunogenic complexes. Such hapten-stimulated immune responses responsible for allergic reactions.

#### EPITOPES:

- Are small specific portions of antigen molecules – trigger immune responses.
- Also called **antigenic determinants**.
- Most antigens have many epitopes, each induces production of specific antibody or activates specific T-cell.
- As rule: antigens are foreign substances, not usually in body tissue.
- However: **autoimmune disorder** – when immune system fails to distinguish self from non-self. Result: self molecules attacked as if they were foreign.
- **Genetic recombination**: There are billions of different possible epitopes for the human immune system to recognise. The immune system **can** recognise them even if the epitope is man-made and does not exist in nature. This is because of shuffling of gene segments that code for antigen receptors giving equally high diversities of antigen receptors in both B-cells and T-cells.

#### STRUCTURE AND FUNCTION OF ANTIBODIES:

- Combines specifically with the epitope on the antigen as its structure complements the epitope of the Ag
- Belongs to a special group of blood proteins called **immunoglobulins**.
- Most antibodies have four polypeptide chains:
  - 2 chains are identical: **heavy (H) chains**. Each consists about 450 amino acids  
⇒ Short carbohydrate chains attached to each heavy polypeptide chain
  - Other 2 chains are identical: **light (L) chains**. Each consists about 220 amino acids
  - Disulphide bonds (S-S) links light chain to heavy chain.
  - Two disulphide bonds link mid-region of the two heavy chains.



- This part of 2 chains are identical: **heavy (H) chains**. Each consists about 450 amino acids
- Has great flexibility, called **hinge region**.
- Antibody arms can move to T-shape or Y-shape
- Tips of H and L chains: **variable (V) regions**. Are the **antigen-binding sites**.
- Most antibodies – 2 binding sites → termed **bivalent**.
- Flexibility at hinge: antibody can bind to two epitopes far apart eg. on microbe
- Remainder of H and L chains: **constant (C) regions**.
- Nearly same in all antibodies of same class.
- Constant Heavy region in IgG also has the complement activation binding site and the Fc binding site.
- Constant region of H chain differs between classes of antibodies.
- **Five different classes** of antibodies: IgG, IgA, IgM, IgD and IgE
- Each class → distinct chemical structure; specific role
- IgM: appear first – short-lived – presence indicates recent invasion
- Resistance of foetus and newborn to infection → IgG cross placenta before birth & IgA antibodies absorbed from breast milk after birth.
- All classes of antibodies act to disable antigens by:
  - 1) **Activation of complement**: initiates classical pathway of complement → THE MAIN FUNCTION OF IgG ANTIBODIES IS TO CALL ON **COMPLEMENT TO CAUSE PHAGOCYTOSIS**
  - 2) **Neutralisation of antigens**: antibody-antigen reaction blocks/ neutralises damaging effects of bacterial toxins and prevents attachment of some viruses to body cells.
  - 3) **Immobilisation of bacteria**: if antibodies form against antigens on cilia/ flagella of bacteria, reaction may make bacteria lose mobility → limits spreading
  - 4) **Agglutination & precipitation of antigen**: since 2 binding sites, antibody-antigen reaction may cross-link pathogens together → agglutination. Also, soluble antigens may precipitate when cross-linked by antibodies → easily phagocytised.

**Enhancement of phagocytosis**: a process called **opsonisation**. This causes agglutination, precipitation, activates complement and coats microbes so that they are more susceptible to phagocytosis.

Antibody	Abundance & Function
IgG	80% → monomer; provides immunity for newborns; protects against bacteria and viruses by enhancing phagocytosis, neutralising toxins and triggering complement system; 2 <sup>o</sup> imm resp
IgA	10-15% → dimer; in secretions (sweat, tears, saliva, mucous, milk, gastrointestinal secretions) to provide localised protection on mucous membranes. Cannot call upon complement since not in blood but instead 'smothers' Ag
IgM	5-10% → pentamers but valency is only 5 since not all Ag-binding sites are accessible. First antibody class to be secreted by plasma cells; activates complement; causes agglutination; lysis of microbes; are the anti-A / anti-B antibodies in plasma
IgD	0.2% → monomer; Are the antigen receptors on B cell
IgE	<0.1% → monomer; bound to mast cells and basophil membrane (ie cytophilic). Involved in allergic reactions. Protection against parasitic worms.

### THE COMPLEMENT SYSTEM:

- A series of around 25 serum proteins that work to "complement" the activity of antibodies in destroying bacteria → either by enhancement of phagocytosis/ or by puncturing the bacterial cell membrane via the MAC.
- Other functions: gets rid of Ag-Ab complexes and also brings in inflamm cells.
- Heat-inactivated serum destroys complement
- Complement proteins circulate in the blood in an inactive form. When the first of the complement substances is triggered (usually by Ab interlocked with an Ag) sets up a precise sequence of carefully regulated steps known as the "**complement cascade**."

#### Classical Pathway:

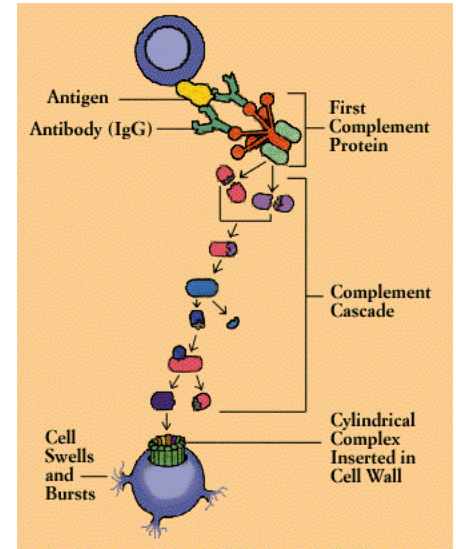
- Relies on an Ab to initiate (complement fixation by antibody)
- First component of pathway, C1q binds to binding site on Constant Heavy Chain of Ab – Nb the Ab **MUST** be bound to an Ag in order to set up cascade (reason why complement system not continually activated)
- Results in formation of MAC that is inserted into membrane and causes cell swelling and so lysis
- Is imp to remember that the MAC is effective for small cells like RBCs but not that effective against eg bacteria
- Thus MAC formation is not primary aim of complement system → instead it is the enhancement of phagocytosis by coating the bacteria/ microbe with C3b (remember phagocytes have C3b receptors/ CD35) by activation of C3 convertase enzyme
- Host cells have certain molecules that prevent bystander lysis esp host RBCs
- Various fragments flung off during the course of the cascade can produce other consequences. Many are potent chemoattractants for other inflamm cells eh neutrophils
- Others cause degranulation of mast cells and basophils → redness, heat, warmth
- The C3b fragment as well as opsonizing the microbe also helps prevent the formation of large and insoluble (and thus potentially damaging) immune aggregates. Moreover, receptors for C3b are also present on red blood cells, which appear to use the receptors to pick up complement-coated immune complexes and deliver them to the Kupffer cells in the liver.

#### Alternative Pathway:

- Occurs when spontaneously activated complement component binds to surface of pathogen
- Again results in coating of microbe with C3b (ie enhances phagocytosis) and MAC

#### MB-Lectin Pathway:

- Initiated by binding of a serum protein (Mannin-Binding lectin) to mannose-containing carbohydrates on bacteria/ viruses
- MB-Lectin v similar in structure to first complement component C1q so sets up classical pathway
- Deficiency of MB-Lectin incrs infection rates in early childhood (ie before child's own imm sys fully working and after loss of protective effects of mother's Ab)



## CELL-MEDIATED IMMUNITY

### T-CELLS:

- Develop from pluripotent stem cells originating from red bone marrow
- T-cells develop from pre-T cells that migrate from bone marrow into thymus.
- Most T-cells arise before puberty – some continue to mature through life (cells (thymus-derived cells).
- Before T-cells leave thymus it acquire several distinctive surface proteins.
- Some function as **antigen receptors**: molecules capable of recognising specific antigens
- T-cells exit thymus as either: **CD4+** or **CD8+** cells (displaying either protein CD4 or CD8 on plasma membrane)
- There are **two** major classes of T-cells produced in the thymus: helper T-cells and cytotoxic, or killer T-cells

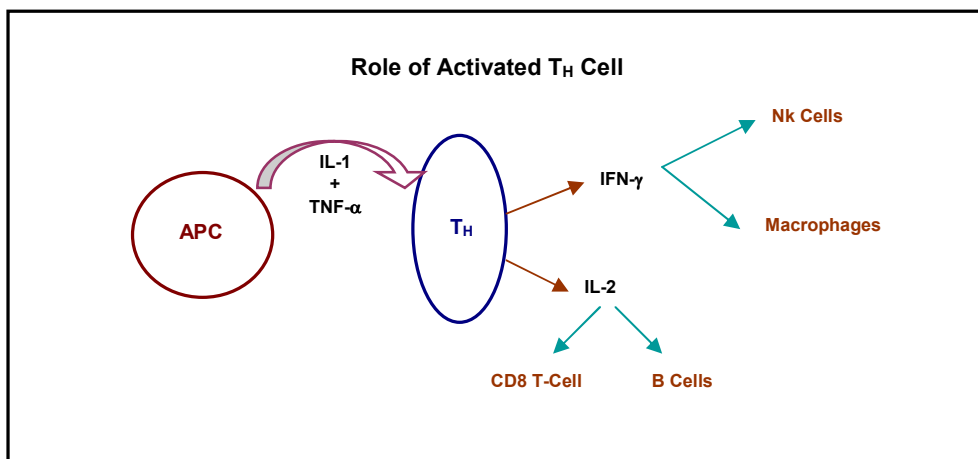
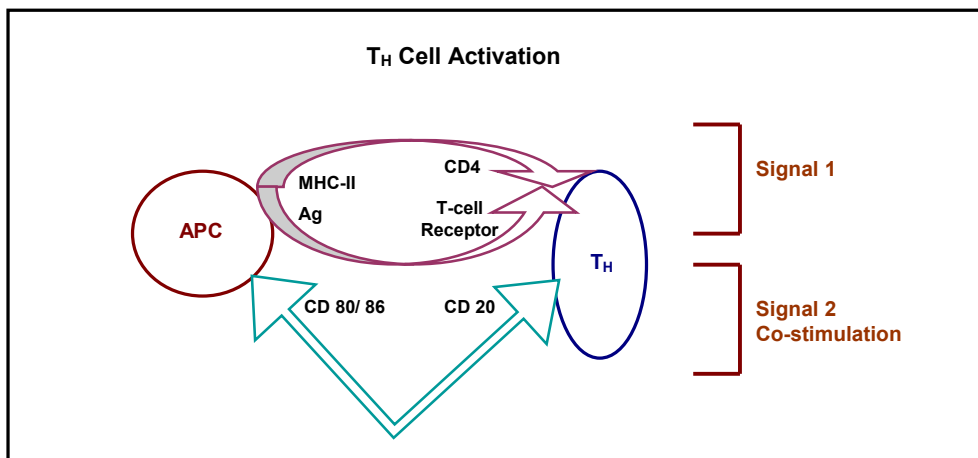
#### Helper T-cells:

- **Helper T-cells** → display CD4. Also termed ( $T_H$ ) cells or T4 cells
- Interacts with APCs – recognises foreign Ag in association with MHC-II
- Secrete molecules called **cytokines** that promote development of B cell into plasma cells and activates  $T_C$  cells (later)
- Nb. The cytokines that are secreted by lymphocytes are also called lymphokines
- The cytokines that are secreted by other monocytes and macrophages are called monokines
- Main class of cytokines are the **interleukins** (IL)
- Some eighteen different interleukins are known: IL-1, IL-2, etc, interferon, lymphotoxin, and **tumour necrosis factor** (TNF- $\alpha$ ). Each interleukin has complex biological effects

#### Cytotoxic T Cells:

- **Cytotoxic T-cells** (Killer T cells) → display CD8
- Destroy cells infected with viruses and other pathogens and may also destroy cancerous cells
- The receptors of T-cells are different from those of B-cells because they are able to recognize fragments of foreign antigens combined with MHC-I molecules on the surfaces of all the body's cells

- As T-cells circulate through the body, they scan the surfaces of body cells for the presence of foreign antigens that have been picked up by the MHC molecules. This function is sometimes called immune surveillance.



- Nb – need both interaction of MHC-II/CD4 with the Ag/TcR for Signal 1. Also need costimulation by interaction of CD80/86 on APC with CD20 on T<sub>H</sub>
- Once Signal 1 and 2 are made then APC secretes IL-1 and TNF-α which stimulates the T<sub>H</sub> cell to make IFN-γ and IL-2
- IL-2 is said to be a T-cell growth factor → it acts on the IL-2 receptor on the T<sub>C</sub> cell (CD25) and causes clonal proliferation of T<sub>C</sub> cell
- Imp to Remember: **Cytotoxic T cells require antigen to be associated to MHC-I whereas Helper T cells require antigen to be associated to MHC-II**

#### ANTIGEN PROCESSING:

- B cells can only bind to antigens in extracellular fluids
- T cells can only recognise fragments of antigenic proteins that have **first been processed and presented** along with a MHC self-antigens.
- As proteins in body cell are broken down – some peptide fragments associate with a peptide-binding **groove** of newly synthesised MHC molecules
- This association: stabilises MHC molecules + aids proper folding so that MHC molecule can be inserted into plasma membrane
- T-cells **cannot recognise** this association of MHC antigen with self-protein peptide fragments nor can it recognise the MHC-I alone of foreign peptide alone
- When fragment is from foreign-protein, T-cells recognise intruder → immune response
- Preparation of foreign antigen at cell surface: processing and presenting of an antigen

Occurs in two ways

##### 1) Processing of exogenous antigens:

- Foreign antigens present in fluid outside body cells.
- Include: bacteria, bacterial toxins, parasites etc

- Class of cells called **antigen-presenting cells (APCs)**: process and present these antigens
  - APCs: include macrophages, B-cells (this is the B-cell's 2<sup>nd</sup> function after producing Abs), dendritic cells
  - APCs located at areas where penetration of non-specific defences is likely eg. dermis of skin (Langerhans cells: type of dendritic cell); mucous membranes of respiratory system, GI tract...
  - Steps:
    1. **Ingestion of antigen:**
      - By phagocytosis/ endocytosis. Ingestion could occur anywhere in body
    2. **Digestion of antigen into peptide fragments:**
      - Inside phagosome/ endosome: digestive enzymes split large antigens into small peptide fragments, also, APC is making MHC-II molecules & packing them into vesicles (the MHC is anchored to inside membrane of vesicle)
    3. **Fusion of vesicles:**
      - Vesicles containing fragments and MHCs merge and fuse
    4. **Binding of peptide fragments to MHC-II:**
      - After fusion, antigen peptide fragment (ie epitope) bind to MHC-II molecules
    5. **Insertion of antigen-MHC-II complex into plasma membrane:**
      - Combined vesicle undergoes exocytosis. Antigen-MHC-II complex inserted into plasma membrane
  - After antigen processing, APCs migrate to lymphatic tissue & present antigens to T<sub>H</sub>-cells
- 2) **Processing of endogenous antigens:**
- Foreign antigens that are synthesised inside a body cell
  - Eg. viral proteins after viral infection or abnormal synthesised proteins as a result of a cancer
  - Fragments of endogenous antigens associate with MHC-I inside the affected cell
  - The complex moves to plasma membrane, where it is displayed at cell surface
  - Most body cells can process/present endogenous antigens

**T<sub>c</sub> activation:**

- T<sub>c</sub> recognises the foreign peptide on MHC-I complex → this is the first step of activation
- Is then costimulated by cytokines (IL-2) from T<sub>H</sub> (ie. need both recognition and costimulation to activate → prevents accidental immune response from occurring)
- Cell then enlarges and proliferates → clone of identical cells that recognises same antigen
- Leaves lymphatic tissues, migrates to infection site/ tumour site etc.
- 2 mechanisms of action:
  - 1) Releases perforins → cell lysis
  - 2) Secretes lymphotoxins → activates enzymes inside target cell so cell dies

# IMMUNOLOGICAL ABNORMALITIES

## Three Groups:

- Hypersensitivity / Allergy
- Immunodeficiency
- Autoimmune

## HYPERSENSITIVITY

### Hypersensitivity/ Allergy:

- A state of increased reactivity of the host to a normally innocuous antigen and implies that the reaction is damaging to the host
- It results from the interaction between the antigen and the antibody or T-cells produced by earlier exposure to the same antigen

Lay people → will tend to use 'allergy' to describe all four types of hypersensitivity.

Medical professionals → use allergy for Type I sensitivity and hypersensitivity to describe the rest

**Allergen:** an antigen capable of eliciting an allergic state

**Atopy:** describes the hypersensitivity reactions to non-pathological antigens (allergens)

- People who exhibit these reactions are more prone to varying sorts of allergies and are described as **atopic**
- 4 classes of hypersensitivity → 'Gell and Coombs Classification'

### TYPE I HYPERSENSITIVITY (IMMEDIATE HYPERSENSITIVITY)

- IgE: Y-shaped antibody (protein molecule). Evolved as a means to combat large parasitic organisms. Is usu present in small amounts → in atopic pple: can be 10,000 x > than normal
- The Fc regions of IgE are bound to **mast cells** (in tissues) and to **basophils** (circulating)
- Immune response triggered when IgE binds to antigen resulting in cross-linking of IgE molecules → this triggers **degranulation** of the mast cell/ basophil
  - NB – allergen must be big enough to be able to cross-link but not too big that it cannot penetrate mucosal surface
- This involves releases of stored potent mediators → esp histamine, leukotrienes, cytokines etc and proteases directed at parasite (or the allergen)
- Rapid release and effects
- Preformed Mediators:
  - Histamine → immediate effect
    - ⇒ Causes sm muscle in immediate vicinity to contract eg bronchoconstriction
    - ⇒ Acts on blood vessels to cause vasodilation, vascular leakage (increased permeability) and decreased blood flow
    - ⇒ (Also stimulates gastric secretion – H receptor on parietal cell)
- The cytokines and leukotrienes → later effect – LATE PHASE RESPONSE
  - Prolong the reaction
  - LTs made by phospholipases converting membrane phospholipids into arachidonic acid → converted by LOX into LTs
  - Attract inflammatory cells to area (occurs >6hrs after exposure) – mainly eosinophils that release toxic granule proteins and free-radicals that are supposed to kill parasite but instead causes most of the damage seen in allergy

### Mechanism of Type I hypersensitivity:

Stage 1: sensitisation → 1<sup>st</sup> year of exposure

- Innocuous antigen eg inhaled
- B-lymphocyte combines to allergen for first time – with help of Th cell becomes plasma cell → makes IgE. Memory cells made too
- Why is IgE made? The T<sub>H</sub> cell reacting with dendritic cell/ APC is usually T<sub>H0</sub> ie naïve – it is stimulated to become T<sub>H1</sub> or T<sub>H2</sub> by cytokines secrete from APC that act on the T<sub>H</sub> (IL-4 favours T<sub>H2</sub> development while IL-12 favours T<sub>H1</sub> development)
- T<sub>H2</sub> cells switch the Ab isotype from IgM to IgE

Stage 2: allergic reaction and symptoms → subsequent year

- Enhanced immune response to allergen if re-encountered
- So: IgE made earlier and in greater quantities than before → binds to mast cell etc etc as before

### Anaphylaxis:

- Describes the s/s resulting from this IgE mediated allergic reaction → often used to refer to the more severe s/s
- Due to massive release of vasoactive mediators
- Key features: Hypotension --- Airway obstruction --- Dyspnoea
- It is not always the case that anaphylaxis only occurs in pts who have experienced previous clinical signs of atopy due to sensitisation
- Even though a pt may have many allergies, it is only usu one of them that can cause a potential anaphylaxis

### Clinical Features of Anaphylaxis:

- General: feeling of warmth, light-headedness, feeling of impending doom
- Resp: hoarseness, stridor, wheeze, dyspnoea, lump in throat (oedema of epiglottis and larynx → potentially fatal)
- CV: hypotension, acute myocardial ischaemia
- Skin: urticaria, (weal and flare reaction) angioedema
- GI: nausea, vomiting, abdominal pain
- Uterus: uterine cramps, diarrhoea, vomiting (all due to sm muscle contraction)

### Treatment of Anaphylaxis:

- Most imp drug in emergency Rx of anaphylaxis is im **adrenaline** (may have to give iv or endotracheal if severe)
  - Reverses vasodilation, reverses increased permeability, reverses shock, relaxes bronchial sm muscle

Other Rx:

- Give O<sub>2</sub>, iv fluids
- Anti-histamines: affect the early phase of reaction – relieve s/s
- Corticosteroids: to counter effects of later stages ie. leukotriene and cytokine release
  - Relieves slower-onset urticaria, hypotension etc
- Anti-histamines and corticosteroids not as effective as adrenaline
- Monitor pt – for at least 24 hrs

### Management of Type I Hypersensitivity:

- Accurately identify offending substance
- Avoid that substance
- Have rescue medicine – ‘epipen’ → Know how to use rescue medicine → practice – ie pt education
- Can use **immunological desensitisation**: only for certain types hypersensitivity eg stings
  - Give v small does of allergen → so recruits high levels of IgG producing cells
  - So, when faced with high does of allergen then IgG binds it before gets chance to bind to IgE on mast cell/ basophil

### Diagnosis of Type I Hypersensitivity:

- History:
  - Detailed account of worst ever reaction, type and quantity of food etc
  - Time between ingestion and symptoms etc etc
- Investigations:
  - Main technique: Skin Prick Testing:
    - ⇒ Based on weal and flare reaction (oedema and erythema). Introduce variety of allergens onto skin by pricking epidermis. A positive result may produce a weal after 10-15 mins.
    - ⇒ Use with controls eg. Histamine → see how reactive skin and discovers interfering medicines
    - ⇒ Are easy and safe to perform. Done in allergy clinic with trained staff and adrenaline available
    - ⇒ Used to demonstrate an IgE mediated mechanism is involved and identify possible allergens
  - Other Ix:
    - ⇒ RAST (radioallergosorbent test) – blood testing using radio-immunoassay. Measure either total circulating IgE levels or specific IgE to specific allergens. Expensive and needs experience to interpret results.

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## TYPE II HYPERSENSITIVITY

- Is mediated by **complement** and **IgG**
- Involves an antibody directed complement-mediated cytotoxicity
- Many type II reactions are towards small molecular weight molecules eg drugs
- Onset of s/s is brisk – within hours

*Example 1:*

- **Drug** (eg penicillin) behaves as a hapten (too small to be immunogenic)
- In certain individuals, penicillin binds to platelet's cell surface and allows immune system to 'see' the drug → it is now immunogenic
- First encounter: drug binds to platelet → causes IgM production and then eventually IgG (but by that time drug has left system so causes no s/s)
- When re-encountered: memory B cells allow fast production of IgG. Binds to penicillin which is itself bound to platelet surface.
  - Complement system is recruited via classical pathway → membrane attack complex causes platelet lysis
  - Result: reduced clotting efficiency and potentially fatal haemorrhage

*Example 2:*

- **Rhesus disease** (haemolytic disease of newborn). 85% of pple are Rh positive
- Rh antigen is a protein (ABO is a glycolipid)
- RhD negative mother carries RhD positive child
- At birth, RhD foetal cells immunises mother → makes IgG – but baby is born so no adverse effects

- If 2<sup>nd</sup> child: memory cells. If 2<sup>nd</sup> baby is RhD positive then mother will make anti-RhD antibodies (IgG) which can cross placenta, bind to foetal RBC → lysis → HDN
- Prevention: inject mother with anti-RhD antibodies within 48hrs of 1<sup>st</sup> Rh +ve child
  - These 'mop up' any RhD Ags in the mother (it causes lysis of any foetal RhD RBC that has made its way into mother) so does not allow mothers immune system to make her own Abs → so no memory cell ie. mum isn't immunised against RhD

### TYPE III HYPERSENSITIVITY (IMMUNE COMPLEX HYPERSENSITIVITY)

- Like Type II, most of the tissue damage is caused by complement system → so onset of s/s is also similar (2-6 hrs)
- **Immune complexes** form when Ab binds to soluble Ag
- If small amount of Ag: large immune complexes form → easily phagocytosed
- If there is excess of Ag → then small, insoluble precipitates form – tend to deposit on blood vessels wall → tissue damage
- In pple who are prev sensitised to antigen: quick onset of s/s

#### Example 1:

##### Serum sickness

- eg: drug has bound to serum protein (hapten). (Can also be caused by eg. blood products)
- Circulating immune complexes deposit in tissues
- Complement fixed → so direct tissue damage and recruitment of inflammatory cells → so further damage
- Lysosomal enzymes do most damage

So: blockage of vessels: s/s = fever, rash, sore joints

- Drug-induced serum sickness usu abates within days after withdrawal of causative agent
- May persist for longer if drug is long-acting

#### Example 2:

##### Arthus reaction

- A local Type III hypersensitivity reaction which can be triggered in the skin of sensitised individuals who possess IgG Abs against the sensitising antigen
- So: Ag injected into skin
- Circulating IgG Abs from immune complexes locally
- Deposit in local tissues or on vessel walls
- Recruit complement
- Inflammation and tissue damage results → erythema, oedema, necrosis (microthrombi)

### TYPE IV HYPERSENSITIVITY (DELAYED-TYPE HYPERSENSITIVITY: DTH)

- T-cell dependant reaction (the rest are Ab-mediated)
- 12-24 hrs between time of exposure to onset of clinical s/s
  - Can be days-weeks to eg in TB
- DTH can be induced by protein antigens or by small chemicals which modify the structure of the self proteins (eg. dyes and metal ions)
  - Rubber, cosmetics, metals, plants are all potential causative agents

2 phases of pathogenesis:

#### 1) Induction

- Ag is processed and presented by Langerhan cells bound to MHC-II to CD4 T<sub>H</sub> cells

#### 2) Elicitation:

- Re-exposure to Ag
- p/ p by Langerhan cells → CD4 cells activated
- Secretion of cytokines in area of contact with Ag
- Activate local macrophages – these produce further inflammatory cytokines, enzymes, free radicals
- Result: tissue damage/ inflammation – DERMATITIS
- Contact dermatitis → typically due to Ni-plating on jewellery that modify self proteins at the site that the jewellery was worn
  - Modification of proteins
  - Metal-modified protein is recognised as non-self → T-cell attack mounted
  - Lymphocytes can move so if the pt then wore a Ni-plated earring (instead of bracelet), a DTH response would develop at this site

Management of allergic contact dermatitis: prevention (identify and avoid antigen) and Rx

#### NB: Drug allergy/ hypersensitivity:

- Can be due to Type I, II or III
- Most common is III → immune complex: serum sickness

## AUTOIMMUNITY

- Breakdown of tolerance to self antigens with no known external eliciting agent
- Spectrum of AID from organ-specific eg Hashimoto's Thyroiditis to non-organ specific eg SLE/ RA
- Can be classified by mech of injury using same scheme as Hypersensitivity (but no Type I AID)
- Since cannot eliminate self-Ags then get sustained immune response (except eg IDDM where target tissue destroyed completely)

### Normal Tolerance:

- Tolerance to self Ags is induced by:
  1. Early clonal detection → negative selection of reactive B or T-cells
  2. Clonal anergy → non-deleted reactive T-cells undergo clonal anergy
    - Eg. Activated T-cell that doesn't have costimulation signals results in selective inhibition of IL-2 production (remember: IL-2 needed for T-cell proliferation)
  3. Selective migration of lymphocytes → some Ags are in 'immunologically privileged' sites ie not in contact with immune system eg crystalline lens
  4. T-cell mediated suppression – eg IL-10 from T<sub>H2</sub> impairs costimulation signal

### Breakdown of Tolerance:

1. Molecular mimicry eg in rheumatic fever (Abs directed against strep also target Ags of heart, joints etc)
2. Release of sequestered Ags eg tissue trauma
3. Inappropriate MHC expression and cytokine imbalances
4. Polyclonal B-cell activation eg EBV infection → make autoAbs

### Aetiology of AID:

- Major theory to AID is over-expression of MHC-II so ↑ p/p to B-cells → auto-Ab formation
- Genetic Factors:
  - More than one type of AID found in same pt- termed 'clustering'
  - Twin studies: concordance noted
  - Possession of certain HLA types predisposes (mainly class II) eg HLA DR3/4 for IDDM and DR2 for Goodpasture's
  - F>M (poss link to oestrogen) and middle-age onset
- Environmental Factors:
  - Must be involved since eg IDDM has only 50% concordance in identical twins
  - Known that diet may be inv eg fish oils improve RA
  - Organic solvents linked to Goodpasture's (dry cleaners)
  - Drugs – alerted antigenicity
  - Microorganisms → molecular mimicry – grp A Strep

### Types of AID:

- Type II
  - Abs to cell components/ matrix components with fixation of complement
  - Eg Hashimoto's Thyroiditis → Abs against thyroglobulin and peroxidases → fixation of complement so get inflammatory damage incl NK-cell antibody-dependent injury
  - Eg Grave's Disease → Abs to TSH receptor → stimulates excessive thyroid hormone production (-ve feedback results in ↓ TSH production but since auto-Ab is stimulatory then no effect)
  - Eg Goodpasture's syndrome → Abs made against glom b memb and also can cross react with b memb of lung alveoli
- Type III → Immune-Complex Formation
  - Eg SLE: deficiencies in early classical pathway components so fail to solubilise immune complexes
    - Get chronic IgG production directed at ubiquitous self Ags present in all nucleated cells ie auto-Abs formed against common cellular components
    - Due to large quantities of auto-Abs get large number of small immune-complexes forming – these deposit in vasculature, glomerular vessels, glomerular basement membrane
    - So phagocytes are activated via Fc receptors → inflammatory mediated damage
- Type IV → T-Cell Mediated
  - Activated T-cells due to self-peptide/ self-MHC complex
  - These activated T-cells then damage tissue (a) directly and (b) via macrophage activation
  - Eg IDDM – IoL heavily infiltrated with T-cells and activated macrophages
  - RA now also thought to be Type IV AID
- 30% of gen population have low titre ANA (anti-nuclear Abs) – why have Abs to this nuclear component? → due to failure of apoptosis so get exposure of nuclear components to imm sys and Abs to them ie auto-immunity
- ANCAs seen in vasculitis → cANCA specific for Wegener's (detects proteinase 3) and pANCA less specific (detects myeloperoxidase)

## IMMUNODEFICIENCY

- Can be congenital or acquired
- Can occur at any point in imm system:
  1. Cellular Level → missing or non-functional cells
  2. Mediator Level → enzyme defect etc
  3. Receptor Level → missing/ masked
- Main clinical features:
  - Ch infections
  - Recurrent infections
  - Unusual infections → eg PCP, CMV, candida
  - Unusual tumours eg KS

### Environmental/ Acquired:

- Infection – eg malaria, HIV
- Malnutrition (commonest cause w'wide) esp for iron/ proteins eg alcoholics
- Drugs – esp chemotherapy, post-transplants
- Irradiation – X-Rays, UV
- Chronic alloantigen stimulation – eg blood transfusion effect, pregnancy
- Neoplasms of imm sys – eg HD, leukaemias

### Genetic:

- Mainly X-linked recessive
- Can be:
  - Specific defects eg in B-cells or T-cells
  - Non-specific:
    - Complement → components or regulators
    - Phagocytosis → neutrophils or macrophage

## NON-SPECIFIC

### Complement Deficiency:

- Loss of early components – seen in SLE (imm complex formation) so get defective phagocytosis and recurrent pyogenic infections
- Loss of late – Neisserial infections
- C1 esterase inhibitor deficiency – ↑ C1 activation → hereditary angiooedema
- Properdin deficiency (component of alternative pathway) → fulminant meningococcal septicaemia
- PIG anchor defect (anchors the molecules that prevent by-stander lysis (during complement activation) to host cell membranes) → so get RBC lysis and anaemia

### Phagocytosis Deficiency:

- Chronic granulomatous disease
  - X-linked recessive
  - No O<sub>2</sub> intermediates made so no killing despite phagocytosis
  - Get recurrent infections, granulomas
  - Rx: IFN- $\gamma$
- Adhesion defects
  - Loss of adhesion molecules so impaired chemotaxis
  - Recurrent bacterial infections
  - Rx: BMT
- Lazy leucocyte syndrome
  - Motility defect – no chemotactic response
- Myeloperoxidase deficiency
  - No hypochlorous acid so no killing
  - Incs candida infections
- Chediak-Higashi Syndrome
  - Giant, non-functional lysosomes

## SPECIFIC

### B-cell Deficiency

- Results in: no Ab (most severe) → decrs Ab → selective loss of Abs
- Indicated by ↓ total serum Ig levels, ↓ B-cell numbers in blood
  - 1) X-linked infantile hypogammaglobulinaemia
    - Most common type of B-cell deficiency
    - Affects 1:100,000 male children – onset at 6 months (mums Abs working till this point)

- Get otitis media, bronchitis, pneumonia etc
- V low IgG and NO IgA/M/D/E
- No B-cells but normal T-cells
- 2) Transient hypogammaglobulinaemia
  - Onset at 6 months → recurrent infections
  - B-cell numbers normal → perhaps T<sub>H</sub> problem
  - Rx: Igs
- 3) Acquired hypogammaglobulinaemia
  - Unknown cause – can occur at any age / both sexes
  - Low serum Ig levels, normal B-cell levels
  - ↑ CD8 and ↓ CD4 cells seen
  - Defective regulation of Ab synthesis
- 4) Selective Ig deficiency
  - IgA: 1:5000 pple → other Igs normal, normal T-cells
    - Unknown cause – poss defect in B-cell development
    - Rx: Abx and NOT IgA (since if give IgA can get imm resp against them since seen as foreign)
  - IgM: other Igs normal → perhaps T<sub>H</sub> problem
  - IgG: deficiencies in all 4 subclasses of IgG documented

### T-cell Deficiency:

1. Di Georges Syndrome:
  - Congenital thymic aplasia – 12 wks gestation
  - So no thymus so low T-cells
  - Poor CMI and due to ↓ T<sub>H</sub> then ↓ B-cell function → so prone to intracellular pathogens eg candida
  - Rx: fetal thymus graft
2. Chronic Mucocutaneous Candidiasis:
  - Onset: 1 yr
  - ↓ CMI response specifically for candida
  - T-cells number and function normal as are B-cells and Igs
  - Defect unknown
3. Wiscott-Aldrich Syndrome
  - Sex-linked recessive – males
  - Small thymus, ↓ T-cells, ↓ IgM, ↑ IgE/A, IgG normal
  - Also get triad of recurrent viral infections, eczema and thrombocytopenia
  - Rx: BMT/ Igs

### Deficiency of Both B-cells and T-cells:

- SCID → severe combined immunodeficiency:
  - X-linked recessive and autosomal forms
  - V low B-cells and T-cells
  - Die by 2yrs old unless get BMT

# TRANSPLANTATION

- Rejection of an allograft is an adaptive imm resp → lymphocyte inv, specific resp, escalating resp, exhibits memory
- 1<sup>st</sup> set rejection: slow (naive)
- 2<sup>nd</sup> set rejection: fast (memory)

## Transplantation Antigens:

- MHC I: A, B, C → all nucleated cell
- MHC II: D, DR/DQ, DP → B-cell, dendritic cell, macrophage

## Immune Response to Allograft:

- Cellular: CD4/CD8, NK, activated macrophages, B-cells → CD4 cells are key to rejection process
- Humoral: cytokines, Ab-dependent cell-mediated cytotoxicity, complement-dependent cell mediated cytotoxicity
- Direct Presentation: donor allograft can contain dendritic cells (DCs) – drain to lymph node once transplanted and get direct allorecognition of MHC present on these 'passenger DCs' and activation of host T cells
  - Is imp early in imm resp and ↓ over time and indirect takes over (with time foreign DCs disappear)
- Indirect Presentation: normal imm resp → recipient's APCs finds graft – samples donor Ags and drains to node where presents with MHC-II and activates T-cells etc etc

## Rejection:

- ① Hyperacute rejection: occurs within mins-hrs of organ being reperfused → no pronounced neutrophil infiltration but instead get Ab deposition, complement and fibrinogen activation and EC activation → thrombotic occlusion (can now be prevented)
  - Occurs due to reaction of pre-existing alloAbs in recipient (From eg prev blood transfusions, pregnancy) to blood grp Ags/ MHC class polymorphisms from donor
- ② Acute rejection: humoral and cellular inv/ lymphocyte driven → occurs days after, relatively rapid – full blown inflamm resp → lytic enzymes made from macrophages, Ab-dependent cell mediated toxicity
- ③ Chronic rejection: for renal transplants get chronic allograft nephropathy (CAN) → b memb destruction and interstitial fibrosis – progressive functional deterioration over mo-yrs caused by vasc obliteration and fibrosis (low grade persistent DTH)
  - Is both an immune and non-immune mechanism:
  - ① Alloreactivity due to minor HLAs, polymorphisms of graft genes and proteins being presented
  - ② Ischaemic-reperfusion injury – effects appear later
  - ③ Chronic toxic effects of ciclosporine and tacrolimus
- ① = pathology of coagulation, ② = pathology of inflammation and ③ = pathology of tissue remodelling
- Rx only target acute rejection – little exist for ② and ③
- Self-tolerance: auto-reactive T cells removed (apoptosis, anergy, suppression) → quest is to achieve donor-specific tolerance so to avoid life-long immunosuppression

## Prevention of Rejection:

- Tissue match recipient with donor
- Tx life expectancy is 10-15yrs so if match for 1<sup>st</sup> Tx then ↓ risk of rejection of 2<sup>nd</sup> (2<sup>nd</sup> set rejection)
- Matching techniques:
  - Class I and II typing → by serology (though requires large no of cells) or molecular techniques (PCR to look at genes)
  - ABO blood grouping of donor and recipient
  - Sensitisation: identify pre-existing Abs in recipient to HLA Ags of donor (flow cytometry/ ELISA)
  - Cross-matching:
    - ⇒ Class I: if complement fixing then predicts likelihood of hyper/acute rejection while non-complement fixing for accel/ chronic rejection
    - ⇒ Class II: not as clear but can be assoc with hyper/acute but more for accel/ chronic rejection

## Approaches to Renal Transplantation:

- Cadaveric: DR> B + A
- Living and related: identical > haplo > spouses > unrelated >un/mismatched cadaveric

## Bone Marrow Transplantation:

- Identical sibling > haploidentical sibling > haploidentical parent > matched unrelated donors

## Immunosuppressive Therapy:

- Overcomes the adaptive (mainly T-cell) response
- ① Calcineurin Inhibitors: eg ciclosporin (fungal metabolite), tacrolimus
  - Acts early (gene transcription) → good at preventing and treating rejection
  - S/e: nephrotoxic, DM, immunopsych problems,
- ② Agents inhibiting T-cell proliferation: eg azathioprine, mycophenolate
  - Act later (T-cell replication/ proliferation)
  - Not nephrotoxic but other s/es eg anaemia
- ③ Monoclonal Abs (Mabs): eg basiliximab (CD25 (IL-2R) antagonist)
  - ↓ First time rejection by 40%

- ④ Corticosteroids eg prednisolone
    - Non-specific immunosuppressive action
  - Usual use ④ plus one of ① or ②
- 

## VACCINATION

- Pre-vaccination: variolation
- Vaccines are Ags
- ① Killed vaccines – use chemical/ heat to kill and inactivate organism eg whole cell pertussis vaccine
- ② Subunit vaccines – toxins are pathogenic fragments of bacteria
  - Chemically inactivated toxins are toxoids
  - Abs to toxins can protect against infection
  - Eg acellular pertussis vaccine

### **Conjugate Vaccines:**

- B-cells can recognise most molecules incl sugars and proteins
- T-cells can only recognise proteins
- The protective Ags on some bacteria are sugars so T-cells can not help B-cell to make Ab → sugars are weak Ags
- For a vaccine link sugar to something that gives good imm resp eg protein
- Eg diphtheria or tetanus toxoids

### **Recombinant Vaccines:**

- Certain viruses/ bacterial/ parasites are v hard to grow – can grow in host cells but not readily cultured