Immunology

Def'n: the branch of biomedical science concerned with the response of the organism to ***antigenic challenge***, the recognition of self from non-self, and all of the biological (*in vivo*), serological (*in vitro*), and physical chemical aspects of immune phenomena

• ***defence mechanisms*** include all physical, chemical and biological properties of the organism which reduce it's susceptibility to foreign organisms, material, etc.
• functionally these may be divided into those which are static, or ***innate*** to the organism, and those which are responsive, or ***adaptive*** to a potential pathogen or foreign substance

Functional Division

1. ***innate system***
   - evolutionary older system
   - first line of defence
   - non-specific
   - resistance is static, ie. doesn't improve with repeated exposure, ***no memory***
   - often sufficient to prevent disease

   i. physical defences
      - skin & epithelial surfaces, cilia
      - commensual flora
      - acidic gastric contents
      - fever

   ii. biochemical defences
      - soluble - lysosyme, acute phase reactants
        - complement, fibronectin, interferon
      - cellular - natural killer cells, RES phagocytes

2. ***adaptive system***
   - second line of defence
   - activated once the innate system has been penetrated/overwhelmed
   - is ***specific*** to the infective agent
   - exhibits ***memory*** with an enhanced response to subsequent challenge

   i. soluble factors

   ii. cellular factors

THE INNATE SYSTEM

Physical Defences

1. skin
2. epithelial surfaces, cilia
3. gastric acid secretion
4. commensual flora
5. inflammatory circulatory response
6. fever

Soluble Factors

- ***Lysozymes***

• distributed widely in secretions
• act by cleaving bacterial cell wall proteoglycans

- ***Fibronectin***

• family of closely related ***glycoproteins***
• synthesised by endothelial cells & fibroblasts
• involved with,

a. non-specific ***opsonization***
b. facilitation of phagocytosis
c. wound healing and tissue repair

• levels are decreased in patients following,

a. major burns
b. major surgery
c. trauma
d. sepsis, MODS
e. DIC

• controlled trials using ***cryoprecipitate***, or purified human fibronectin have ***failed*** to demonstrate an improvement in organ function, or a reduction in mortality in patients with septic shock

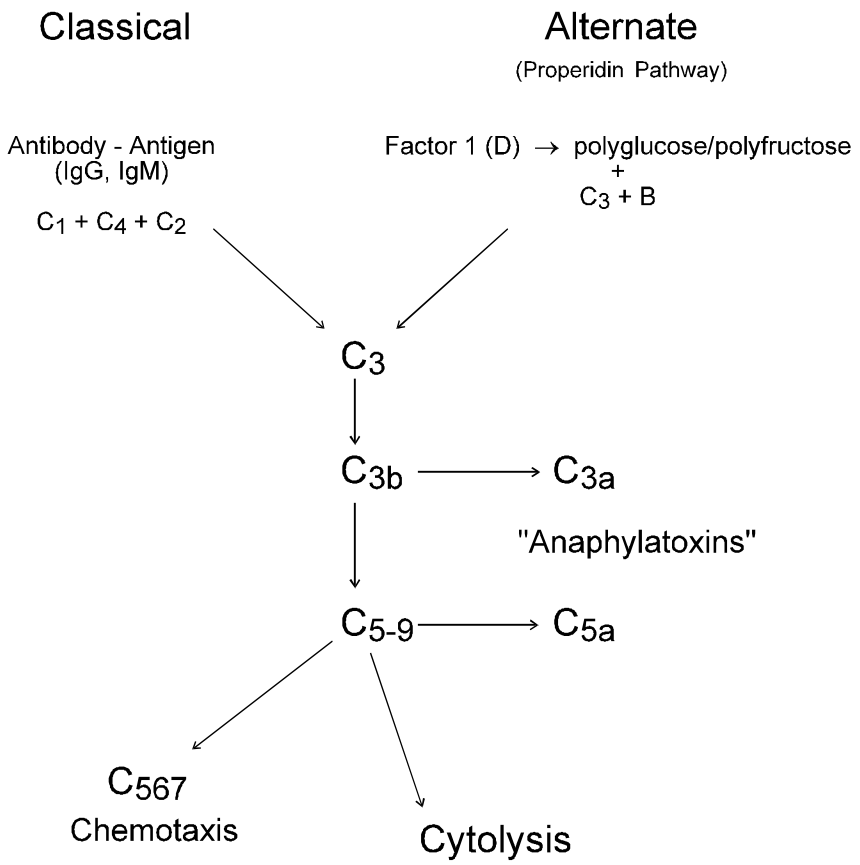
Complement

- series of > 15 plasma proteins, functions including,

1.	chemotaxis	C_{5a}, C_{567}	
2.	mast-cell degranulation	C_{3a}, C_{5a}	(histamine release)
3.	opsonization	C_{3b}	
4.	cytolysis	C_{56789}	(membrane attack complex)
5.	kinin-like activity	C_2 frag.	
6.	viral neutralisation	C_1, C_4	

- activation may be via,

1. ***classical pathway***
   - involves Ag-Ab interaction (IgG, IgM)
   - effectively a part of the ***adaptive*** system
2. ***alternate pathway***
   - recognition of repetitive sugar moieties, ie. bacterial cell walls
   - part of the ***innate*** system



Complement Disturbances

1. predominantly classical activation
   - ***all*** early factors low - low C_3, C_1q, C_4 and C_2
   - active SLE
   - SBE, PAN, HBV also result in activation, but plasma C-levels normal
2. predominantly alternate activation
   - low C_3, the remainder of the early factors are normal
   - post-streptococcal GN, mesangioproloferative GN
3. inherited C-factor deficiency - most often C_2, C_6, C_7, C_9
4. deficiency of C_1-esterase inhibitor
   - non-specific ***serine esterase*** inhibitor - C_1r, C_1s, kallikrein, plasmin, XI_a, $XIII_a$
   - ***autosomal dominant*** and results in hereditary angioneurotic oedema,
   i. type I - deficient inhibitor, ~ 75% of patients
   ii. type II - abnormal/inactive inhibitor
   - produces sporadic activation of the classical pathway via C_2
   - management includes ***danazol*** ($\uparrow$ C_1E inhibitor), fibrinolytic inhibitors, but ***not*** FFP

Interferons

- produced by virally infected cells
- transmit information to adjacent cells, making them resistant to viral replication, thereby impeding the spread of infection
- also activate ***natural killer cells*** and enhance cytotoxic action

Acute Phase Reactants

- a group of plasma proteins which increase rapidly following infection
- eg. CRP, which is probably produced by the liver, recognises and binds to a wide variety of bacteria & fungi
- acts as an ***opsonin***, enhancing phagocytosis, and activates complement

Cellular Factors

NB: all are derived from the ***myeloid series*** in the bone marrow

- ***Large Granular Lymphocytes Natural Killer Cells***

• ***non-thymic*** derived lymphocytes with no antigenic surface markers of T/B-cells
• bind to altered surface markers on virally infected or tumour cells
• ***do not*** require complement or Ab for recognition, but are activated by interferons
• actively regulated by T-cells as well as interferon, therefore ***innate*** as well as ***adaptive***
• function is depressed by,

a. cyclosporin
b. cytotoxics
c. cimetidine
d. malnutrition

- ***Phagocytes***

• these are cells of the ***reticuloendothelial system***, ie. monocytes & macrophages

a. alveolar macrophages
b. splenic macrophages
c. lymph node macrophages
d. kidney mesangial macrophages
e. blood monocytes
f. brain microglia
g. hepatic Kupffer cells
h. synovial A cells

• ***macrophages*** can engulf particles & destroy them, or represent the antigen in a more "active" form on their cell surface
• their ability to recognise foreign particles is enhanced if the antigen is coated with,

1. complement
2. antibody
3. antibody + complement

• ***monocytes*** are produced in the bone marrow, circulate for a short period then localise in various tissues becoming specific macrophages

▪ ***Neutrophils***

• comprise ~ 80-90% of the circulating polymorphs
• diameter ~ 10-20 µm
• contain,

a. lysozymes
b. ingested organisms - phagosomes
c. phagolysosomes

• able to penetrate endothelial surfaces under the influence of chemotactic factors

→ ***diapedesis***

▪ ***Eosinophils***

• also capable of phagocytosis
• release granular contents adjacent to large foreign bodies which would otherwise be impossible to phagocytose, eg. worms (helminths)
• attracted by ***eosinophil chemotactic factor***
• attach to immunoglobulins on foreign particles & release,

a. ***major basic protein***, which is toxic to a wide variety of pathogens
b. eosinophil cationic protein
c. eosinophil derived neurotoxin
d. ***anti-inflammatory enzymes*** - histaminase, arylsulphatase, phospholipase D
   • may have a role in down-regulating the immune response

▪ ***Basophils & Mast Cells***

• small numbers of basophils in the circulation
• more commonly associated with epithelial surfaces, then termed mast cells
• granular contents include,

a. histamine
b. SRSA - leukotrienes, LTC_4, $\mathbf{LTD_4}$, LTE_4
c. ECFA → central to anaphylactic response

• may also have a role in immunity to parasitic infections and as enhances of the inflammatory response

▪ ***Platelets***

• also myeloid derivatives & participate in the inflammatory response

THE ADAPTIVE SYSTEM

Soluble Factors

▪ *Immunoglobulins*

• two broad actions,

1. antigen recognition
2. outcome determination

• these functions are subserved by different regions of the immunoglobulin moiety,

1. ***fraction antigen binding*** **Fab**
   • this is the highly variable area which determines specificity
2. ***fraction crystalline*** **Fc**
   • which determines what happens once the Ab-Ag interaction has occurred

• consist of 2 types of peptide chains, H (heavy) and L (light) chains
• immunoglobulin functions,

1. direct neutralisation - bacterial toxins
   - viral particles
2. opsonization - enhancing phagocytosis
3. complement activation - classical pathway
4. activation of cellular elements

▪ *Immunoglobulin G*

• most abundant & has the broadest role
• four subclasses,

a. IgG_1 & IgG_3 - activate complement
b. IgG_2 & IgG_4 - interact with Ig receptors on phagocytes

• crosses the placenta,

a. provides immunity for the neonate
b. produces rhesus incompatibility disease

- ***Immunoglobulin A***

• predominant Ig found in secretions,

a. respiratory tract
b. GIT
c. urinary tract
d. tears
e. saliva
f. colostrum

• exists as a monomer in serum, but as the dimer "secretory-IgA" in secretions
• 2 units are joined by a ***J-chain*** (MW ~ 15,000) then extruded from ***plasma cells*** in the submucosa
• taken-up by epithelial cells and the ***secretory piece*** (MW ~ 70,000) is added
• this effectively makes the molecule resistant to enzymatic degradation
• congenital absence of IgA occurs **~ 1:900**, with plasma expression of anti-IgA **~ 20-60%**

- ***Immunoglobulin M***

• a ***pentamer*** comprising ~ 10% of circulating Ig
• capable or forming a spontaneous pentamer configuration, but usually with ***J-chain***
• also exists as a monomeric form in mature B-cells
• predominantly intravascular & involved in the early immune response
• major class of antibodies involved in,

a. blood groups - A & B
b. autoimmune disease - rheumatic fever
   - rheumatoid arthritis, etc.
c. cold agglutinins

- ***Immunoglobulin D***

• predominantly intravascular
• associated with the surface of resting ***B-cells***, with IgM
• may be important in B-cell Ag binding and subsequent differentiation to ***plasma cells***

- ***Immunoglobulin E***

• major role in ***hypersensitivity*** reactions
• bind to mast cells via the Fc fragment
• Ag must bind & cross-link 2 antibodies to initiate mast cell degranulation

Immunoglobulin Subclasses					
	IgG	IgA	IgM	IgD	IgE
MW	150K	160K	900K	180K	190K
Serum g/l	12	2.3	1	0.03	< 0.00025
Half life	23	5.5	5.1	2.8	2.3
C' fixation	**classic**	**alternate**	**classic**	none	none
Reaginic properties	no	no	no	no	**yes**
Secretion by mucous membranes	no	**yes**	no	no	no
Placental transfer	**yes**	no	no	no	no
Macrophage binding	**yes**	no	no	no	no

- ***Tumour Necrosis Factor TNF-α***

• cachectin is a macrophage polypeptide hormone

a. induces the release of ***IL-1*** from monocytes and endothelial cells
b. induces fever through direct effects on the hypothalamus
c. enhances PMN adhesion and phagocytosis
d. directly toxic to endothelial cells - DIC, ARDS, GIT ischaemia, ARF

• ***endotoxin*** is the most potent known stimulus to production & release
• closely related TNF-β is produced by T-lymphocytes following specific Ag challenge

▪ ***Cytokines***

• a generic term applied to,

a. ***lymphokines*** - IL-1...IL-8, interferon, B-cell growth & differentiation factors

b. ***monokines***, or

c. other cell products influencing the behaviour of other cells

• ***interleukin*** is a term applied to lymphokines and monokines which influence the behaviour of other lymphocytes (IL-1 to IL-10)
• react with specific cell surface receptors & are active at low concentrations (10^{-9}-10^{-12} mmol/l)

1. **IL-1**
   - polypeptide, MW ~ 17,000
   - produced primarily by phagocytic cells (monocytes, tissue macrophages)
   - stimulated by wide variety of inflammatory processes
   - results in
     - fever
     - bone marrow release of PMN's
     - T-cell and PMN chemotaxis
     - B-cell proliferation and Ab production
     - * T_4-cell production of **IL-2**
     - increased skeletal muscle catabolism
     - ↑ hepatic production of acute phase reactants
     - ↓ hepatic production of albumin, prealbumin, transferrin
     - slow-wave sleep
2. IL-2
   - polypeptide growth factor which stimulates the proliferation of activated B-cells, T-cells, and NK cells

• ***interferons*** have antiviral & antitumor activity,

a. alpha-interferon ~ 17 subtypes
   - secreted by blood mononuclear cells

b. beta-interferon - secreted by fibroblasts & epithelial cells

c. gamma-interferon - secreted by lymphoid cells

• alpha & beta-IF have similar characteristics, cf. gamma-IF which has more of an immunoregulatory role
• react with specific ***cell surface*** receptors, resulting in,

1. inhibition of viral attachment, transcription, translation, protein synthesis & budding from the cell surface
2. inhibition of malignant cell growth
3. enhanced cytotoxicity of T-cells & NK cells
4. increased monocyte & PMN chemotaxis
5. increased lymphokine production

- in general, interferons have been of little use in the management of viral infection
- exceptions include,

1. alpha-interferon - chronic hepatitis B & C
   - condyloma accuminatum
2. alpha-interferon as a ***cytotoxic agent***
   - hairy cell leukaemia, CML
   - HIV related Kaposi's sarcoma
3. ? gamma-interferon in wound sepsis- under trial

Cellular Factors

Def'n: ***cell mediated immunity*** is any immune response in which antibodies play a subordinate role

- CMI is far more persistent than ***humoral immunity***, lasting ≥ 10 years or for life
- major importance is in host defence against,

1. TB
2. fungi
3. protozoans
4. viruses
5. intracellular organisms
6. tumour cells
7. allografts

- the classical example is ***delayed hypersensitivity*** in skin, intradermal injection in a sensitised person resulting in,

1. rapid onset of erythema
2. induration maximal at 2 days, which may proceed to necrosis

- CMI may be transferred between individuals via ***cells*** but not via serum
- ***transfer factor*** is a low MW material derived from sensitised lymphocytes
- important cells types include,

1. macrophages
   - endocytosis of antigen and presentation to T-cells
   - T-cell & B-cell stimulation via IL-1
   - effector cells in inflammatory response
2. T-lymphocytes
   - recognition of antigen - ie. self versus non-self
   - central modulation of CMI
   - principle effector cells
3. B-lymphocytes
   - produce Ag-specific Ab
   - "cellular" components with "soluble" product
4. large granular lymphocytes - null cells, or "third population"
   - large granular cells with neither T-cell nor B-cell antigenic specificity

   i. natural killer cells - innate, Ab independent

   ii. Ab-dependent cytotoxic cells - adaptive
   - act via IgG/Fc receptor

T Lymphocytes

Def'n: ***thymus derived*** lymphocytes,
actually originate in the bone marrow but migrate to the thymus late in utero and early neonatal life

- main effectors of CMI and comprise ~ 70-80% of circulating lymphocytes in blood
  ~ 90% of lymphocytes in the thoracic duct
- circulate as long-lived "small lymphocytes"
- predominant cell types in,
  1. deep cortical areas of lymph nodes
  2. periarteriolar white matter of the spleen

- cell surface possesses receptors for sheep rbc's, enabling identification → "rosettes"
- differentiation & maturation occurs in utero & neonatal period under the influence of ***thymopoietin***
- separated into ***subtypes*** (T_1-T_{10}) with the use of ***monoclonal Ab's*** to surface antigens →
  1. T_4 helper inducers ~ 65% of circulating lymphocytes
  2. T_8 cytotoxic suppressers ~ 25% of circulating lymphocytes

■ ***T_4 Helper Inducers***

- involved in a number of cell-cell interactions
  1. T-cell / T-cell → stimulate mitosis of,
     i. cytotoxic cells
     ii. macrophage activating T-cells
  2. T-cell / B-cell → ***"co-operation"*** with mitosis & differentiation to,
     i. plasma cells
     ii. memory cells
  3. T-cell / macrophage → induce,
     i. migration
     ii. proliferation
     iii. activation

- when stimulated, T_4-cells,
  1. elaborate ***lymphocytic mitogenic factor*** which stimulates all classes of lymphocytes
  2. express surface antigens which become recognition sites for T-cells, B-cells & macrophages

- ***T_8 Cytotoxic Suppressers***

1. when stimulated, suppress B-cell production of antibodies
2. with the aid of T_4 helper-inducers may be cytotoxic
3. via the Fc fraction of IgG can recognise antigen with attached Ab

• over/underactivity of this class can lead to corresponding hypo/hypergammaglobulinaemia

B-Lymphocytes

• derived from the ***bursa of Fabricius***, or its equivalent,

1. ~ 10% of peripheral circulating lymphocytes
2. ~ 50% of splenic lymphocytes
3. ~ 75% of bone marrow lymphocytes

• differentiation occurs in the 3rd trimester in utero & the neonatal period
• all have membrane bound immunoglobulins, which attach via their Fc portion,

a. IgM & IgD ~ 75%
b. IgG ~ 24%
c. IgA ~ 1%

• clonal expansion and differentiation to,

1. ***plasma cells*** - which produce specific antibody, and
2. ***memory cells*** - which readily produce plasma cells to repeat challenge, requires,
   i. Ig-Fab portion attaching specific antigen
   ii. antigen presented & pre-processed by macrophages
   iii. modulatory signals from other cells, especially T_4-cells

NB: plasma cells produce only one class of Ab which is specific for one antigen

→ theory of ***clonal specificity*** McFarlane Burnet

• Ab production may occur as above, with the ***co-operation*** of T-cells

→ the antigen is said to be ***thymus dependent antigen***

• most antigens are of this class, and the Ab's produced include all classes

• ***thymus independent antibodies*** are exclusively of the IgM class

THE IMMUNE RESPONSE

• introduction of a foreign substance may produce,

1. no reaction
2. specific antibody
3. cell mediated immunity

Def'n: an ***immunogen*** is a substance which initiates an immune response

immunogenicity is the ability to produce an immune response

an ***antigen*** is a substance which reacts with - available antibodies
- sensitised lymphocytes

haptens are smaller molecules (usually < 1000 MW) which cannot induce an immune response in their own right, but may do so when combined with a carrier molecule

• the response to a foreign substance depends upon,

1. the route of entry
2. the dose - very high or low levels may induce ***tolerance***
3. genetic factors - response to a given immunogen
   - major ***histocompatibility gene locus***
   - genes code for initiation, stimulation, suppression, etc
4. cell co-operation - thymus dependent immunogen
   - thymus independent immunogen
5. other factors
   i. foreign surfaces
   ii. presence of coexisting infection, or disease affecting immune status
   iii. fever
   iv. nutritional status of the host
   v. immunomodulatory agents administered to the host

■ ***Primary Response***

1. thymus dependent
   - IgM is first Ab to appear, with a peak ~ 2 weeks
   - switch from IgM $\rightarrow$ IgG / IgA / IgE requires T-cell co-operation
2. thymus independent
   - IgM is the ***only*** Ab to appear

Secondary Response

- occurs earlier than the primary response, usually within 4-5 days
- marked proliferation of Ab producing and effector T-cells
- Ab is usually IgG and has a higher affinity, and therefore more specific
- requires immunological memory in both T-cells and B-cells

Tolerance

Def'n: an active physiological process producing ***immunological unresponsiveness*** to an otherwise immunogenic substance

both humoral and CMI must be inhibited

1. depends upon both ***dose*** and ***presentation***,
   i. high dose produces tolerance in T-cells and B-cells
   ii. low dose produces tolerance in T-cells only
   iii. monomeric solutions may produce tolerance where macromolecules are immunogenic
2. requires repeat exposure
3. is easier to produce in the neonate than in the adult

- mechanism involves the presence of T-suppressor cells which are ***antigen specific***, or the presence of antibodies which alter self-antigens such that they are no longer susceptible to an immune response

Autoimmune Disease

- may be due to,

1. failure of suppression - ie. a T-cell defect
2. tissue damage altering self-antigens with a sustained response
   - classical example is post-streptococcal GN
3. infection altering cell surface markers in a genetically susceptible individual
   - IDDM probably included in this group

HYPERSENSITIVITY REACTIONS

• classified by Gell & Coombs according to,

1. antibody involved
2. type of cell mediating the response
3. nature of the antigen
4. duration of the reaction

• although classified as distinct entities, reactions may involve more than one type
• penicillin may invoke type I, II, or type III reactions

■ *Type I Immediate Hypersensitivity*

• also termed "anaphylactic", involves antigen interacting with IgE on mast cells or basophils
• degranulation results from cross-linking of IgE molecules with release of mediators of inflammation (see later)
• not all type I reactions are anaphylactic

→ ***extrinsic asthma*** & ***allergic rhinitis*** are examples of ***local immediate hypersensitivity***

■ *Type II Cytotoxic Reactions*

• involve either IgG or IgM and ultimately results in target ***cell lysis***
• Ab's bind immune specific antigens and activate ***complement*** via the classical pathway
• in addition, complement fragments result in mast cell degranulation and a systemic "anaphylactoid" response
• the antigen may be either a cell wall component, or molecular components, eg.

a. ABO incompatibility
b. Rh disease
c. Goodpasteur's disease
d. drug induced autoimmune haemolytic anaemia - eg. methyldopa

■ *Type III Immune Complex Disease*

• involves either IgG or IgM
• antigens and Ab's form ***insoluble complexes*** which are too small, or too numerous to be filtered by the reticuloendothelial system (ie. relative ***antigen excess***)
• these are then deposited in the microcirculation and activate ***complement*** at their site of deposition, especially in the joints, skin and kidney
• complement fragments, ***anaphylatoxins***, attract inflammatory cells with a resulting ***vasculitis***
• ***serum sickness*** is the classic reaction, seen in repeat exposure to foreign antisera
• other examples include,

a. penicillin induced vasculitis
b. drug induced SLE

Type IV Cell Mediated / Delayed Hypersensitivity

- independent of antibody production
- T-cells become activated by cellular antigens or circulatory proteins
- these cells can directly mediate the response, or liberate ***lymphokines*** which stimulate other mediators of CMI
- the time course of the reaction is slow,
    1. appearing within 18-24 hours
    2. maximal response at ~ 48 hours
    3. frequently subsiding by 96 hours

- common examples include,
    1. Mantoux test
    2. graft rejection

Mechanisms of Immunological Injury		
Mechanism	Pathophysiology	Disease types
Type I • immediate hypersensitivity • IgE mediated	• basophil & mast cell degranulation • histamine, SRSA, ECFA, NCF • immediate weal & flare	• anaphylaxis • atopy
Type II • cell cytotoxicity • IgG, IgM mediated	• direct phagocytosis or cell lysis • activation of complement, classical • tissue deposition of complement	• blood transfusions • Goodpasteur's syndrome • autoimmune cytopaenias
Type III • immune complex • IgG, IgM, IgA mediated	• tissue deposition of Ag-Ab complexes • accumulation of PMN's, macrophages & complement	• SLE • serum sickness • necrotising vasculitis
Type IV • delayed hypersensitivity • T-cell mediated	• T-cell induced mononuclear cell accumulation • release of lymphokines & monokines • often with granuloma formation	• TB, sarcoid • Wegener's granulomatosis • granulomatous vasculitis

ANAPHYLAXIS

Def'n: ***anaphylaxis:*** symptom complex following exposure of a ***sensitised*** individual to an antigen, produced by immediate hypersensitivity or a type I hypersensitivity reaction, associated with IgE mediated mast cell degranulation

anaphylactoid reactions: are indistinguishable from true anaphylaxis, however the immune nature of the reaction is either unknown, or not due to a type I hypersensitivity reaction

\ ***immediate generalised reaction*** may be a better term

Aetiology

1. ***anaphylaxis***
   - i. prior sensitisation to an antigen, either alone or in combination with a hapten
   - ii. synthesis of antigen specific **IgE**, which attaches to mast cells & basophils
   - iii. subsequent exposure $\rightarrow$
     - mast cell & basophil degranulation
     - release of ***histamine*** + SRS-A (LT - C_4, **D_4**, E_4)
       ECF-A, NCF
       PAF, heparin
     - activation of ***phospholipase A*** with production of prostaglandins, leukotrienes and platelet activating factors
2. ***anaphylactoid reactions***
   - i. exposure & combination of antigen with **IgG, IgM** ± a hapten
   - ii. activation of ***complement*** via the classical pathway (C_{1q}, C_4, C_2)
   - iii. formation of ***anaphylatoxins*** - C_{3a}, C_{5a}
     - mast cell & basophil degranulation $\rightarrow$ ***histamine***, SRSA, etc.
3. direct ***histamine release***
   - many anaesthetic drugs - STP, dTC, ? atracurium, morphine, etc.

- the coupling between Ag-Ab and mediator release is complex
- the pivotal step is an increase in membrane Ca^{++} conductance and $[Ca^{++}]_{ICF}$
- the magnitude of degranulation is determined by,

1. the dose / concentration of Ag
   - receptors are 40-100kD MW and allow a ***graded response***
2. the number of specific IgE Ab's present
3. the affinity of a given drug/molecule for those Ab's
4. the level of intracellular cyclic nucleotides
   - β_2 adrenergic stimulation ***decreases*** mast cell degranulation
   - cholinergic stimulation increases cGMP and ***increases*** degranulation
   - α adrenergic stimulation increases cAMP and may ***increase*** degranulation

■ ***Cellular Elements***

- ***mast cells*** are tissue bound, lying in the perivascular spaces of the lung, GIT and skin
- ***histamine*** is stored in electron dense granules, coupled with ***heparin***
- they posses beta, alpha and probably cholinergic receptors

- ***basophils*** comprise ~ 1% of circulating PMN's
- their role other than in allergic responses is poorly understood
- definite role in ***parasitic infection***, where individuals may display elevated IgE levels

■ ***Preformed Mediators***

1. ***histamine***
   - release is essential & ***all*** the features of anaphylaxis can be produced by histamine

   i. H_1 receptors
      - relaxation of vascular smooth muscle
        $\rightarrow$ short duration of effect but more sensitive
      - increased capillary ***endothelial permeability***
      - contraction of bronchial and GIT smooth muscle
      - $\downarrow$ AV node conduction

   ii. H_2 receptors
      - relaxation of vascular smooth muscle
        $\rightarrow$ longer duration of effect
      - $\uparrow$ GIT secretion and $[H^+]$
      - $\uparrow$ in cardiac - contractility (Ca^{++})
        - automaticity (ventricular)
        - phase 4 δV (SA node)

   - ***NB:*** $\downarrow$ BP $\propto$ $\downarrow$ TPR, ***not*** due to myocardial depression

2. ***chemotactic factors***

   i. eosinophil chemotactic factor of anaphylaxis
      - acid peptide, MW ~ 500
      - role of eosinophils uncertain, but many of their granular contents may act to ***attenuate*** the local tissue response, eg.

        histaminase $\rightarrow$ inactivates histamine
        arylsulphatase $\rightarrow$ inactivates SRSA (leukotrienes)
        phospholipase D $\rightarrow$ inactivates platelet activating factor

   ii. neutrophil chemotactic factor

3. ***enzymes***
   - proteases, hydrolases and peroxidase are stored in granules
   - these suppress coagulation and fibrin deposition

4. ***heparin***
   - MW ~ 50,000 and bound ionically to heparin in mast cells
   - basophils have ***chondroitin*** (MW ~ 300k)
   - commercially produced heparin from animal lungs which are high in mast cells

- ***Mediators Synthesised De Novo***

1. ***arachidonic acid*** derivatives
   - *in vivo*, human mast cells produce predominantly **PGD_2, TXA_2, PGF_{2a}**

   i. ***prostaglandins***
   - PGD_2 - major source is human mast cells
     - bronchoconstriction, even in normal individuals
     - mild increase in vascular permeability
   - TXA_2 - potent bronchoconstriction and vasoconstriction
   - $PGF_{2\alpha}$ - produced by mast cells and PMN's
     - produces fever, erythema, & increased in vascular permeability
     - bronchodilator
     - vasodilator in systemic, pulmonary & coronary beds
   - PGI_2 - produced by mast cells *in vitro*
     - vasodilatation and inhibition of platelet aggregation

   ii. ***leukotrienes***
   - SRSA actually consists of LTB_4, LTC_4, **LTD_4**, LTE_4
   - LTB_4 - potent bronchoconstriction, indirect via ↑ TXA_2 production
     - margination of neutrophils
     - ↑ mucus, vascular oedema & ↑ permeability

     * implicated in ***asthma***
   - **LTC_4** - predominant LT produced by IgE stimulated mast cells
     - potent bronchoconstriction ~ 10,000 x histamine
     - slow onset (~ 10 min) with peak at ~ 30 min
     - coronary vasoconstriction & ↓ LV contractility
   - LTD_4 - potent bronchoconstriction ~ 5000 x histamine
     - other actions similar to LTC_4

2. ***non-arachidonic acid*** derivatives

   i. platelet activating factor
   - platelet aggregation & activation
   - activation of leukocytes with inflammatory mediator release
   - profound weal & flare response when given s.c. in man
   - smooth muscle contraction & ↑ capillary permeability
   - ***NB:*** may have central role in anaphylaxis, SIRS, and asthma

   ii. kinins - prekallikrein and bradykinin
   - low MW peptides resulting in vasodilatation and bronchoconstriction
   - implicated in previous reactions to SPPS

- ***"Common" Antigens***

1. blood & blood products*rare
2. XRay contrast media ~ 2-4% to the older I^--based agents, now rare
3. antibiotics - penicillin produces ~ 75% of all reactions in USA
   - may be up to ~ 30% cross-reactivity with cephalosporins
4. sulphonamides

- ***Anaesthetic Agents***

1. thiopentone - true anaphylaxis rare, ~ 1:30,000, but often severe & fatal
2. relaxants - SCh and alcuronium are the worst offenders
   - pancuronium is ~ safest
   - cross-reactivity may occur within a class
   - reaction may occur on "first exposure", NB: NH_4^+ in foodstuffs
3. opioids - principally direct histamine release, anaphylaxis *per se* is rare
   - only 1 documented case each for pethidine & fentanyl
4. local anaesthetics - true allergy < 1% of cases
   - usually either overdose, IV injection, reaction to vasopressor
   - may have allergy to preservative, NB: bisulphite, methylparabenz
5. colloids
   i. plasma protein solutions < 0.003%
      - said to be less with NSA-5% versus SPPS
   ii. ***haemaccel*** ~ 0.146%
      ? due to ***hexamethylene diisocyanate***
   iii. dextran 40 ~ 0.07%
      - reduced by Promit (0.001%)
      dextran 70 ~ 0.008%

- ***Incidence***

1. hospital patients ~ 3:10,000
2. mortality ~ 3-4%

Presentation

NB: variable latent period, but usually within ***30 minutes*** of exposure

1. respiratory
   - dyspnoea, chest tightness
   - stridor, laryngeal obstruction
   - ***bronchospasm*** (*LTD_4)
   - ↑ peak P_{AW}, ↑ slope of alveolar plateau, ↓ $ETCO_2$
   - pulmonary oedema
2. cardiovascular
   - ***hypotension***, tachycardia ± arrhythmias
   - most common and may be sole finding
   - cardiovascular "collapse"
   - pulmonary oedema is a common finding at autopsy
   - ? existence of "myocardial depressant factors"
3. cutaneous
   - erythematous blush, generalised urticaria
   - ***angioedema***
   - conjunctival injection & chemosis
   - pallor & cyanosis
4. gastrointestinal
   - nausea, vomiting, abdominal cramps & diarrhoea

- ***Fisher's Series***[1,2]

1. ***CVS collapse*** ~ 92% (254/276)
   - the predominant cause being a decrease in ***venous return***,
   i. increased venous capacitance
   ii. fluid loss at the capillary level
      - estimated ~ half the plasma volume lost within the first 15 minutes
   iii. the increase in Hct. increasing ***viscosity***, decreasing venous return further
   - apart from 2 cases in the Lancet 1986, there is minimal evidence for a myocardial depressant factor
   - the healthy heart displays a tachycardia, both direct & reflex, with a decrease in CVP/PCWP
   - the only patients with severe IHD had raised filling pressures
   - however this group also had a higher incidence of SVT other than sinus tachycardia

2. ***erythema*** ~ 50% (132/276)
3. ***bronchospasm*** ~ 29% (80/276)
   * severe in only 41 cases
4. ***angioedema*** ~ 20%

- ***Anaesthetic Management***

NB: multiple actions simultaneously / conclude surgery / call for experienced help

1. cease administration of the likely antigen
2. maintain oxygenation
   i. maximal O_2 via face mask
   ii. IPPV via bag/mask
   iii. intubate & 100% O_2 ASAP *cease anaesthetic agents
3. support circulation
   i. CPR if no output
   ii. ***adrenaline***
      - inhibits mast cell degranulation, ↑ SVR & venous return, ↓ bronchospasm
      - hypotension: 10-50 µg boluses prn or infusion if available
      - collapse: 0.5-1.0 mg stat, then infusion
   iii. volume expansion *"whatever is available"
      - Haemaccel, NSA-5%, CSL, N.saline
      - CVP monitoring once situation under adequate control

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1 Fisher & Baldo. MJA 1988 Vol.149. p34-38.

2 Fisher Anaphylaxis, DM 1987; 33; 438-479

4. manage ***bronchospasm***
   - i. slow RR, high E:I ratio ventilation
   - ii. adrenaline ~ 0.5 mg IM if no access
     - IV dependent upon MAP & ECG monitoring
   - iii. aerosol bronchodilators
   - iv. aminophylline - additive effects with adrenaline
     ~ 5-6 mg/kg loading dose over 30-60
   - v. suction ETT
   - vi. volatile agents - if isolated bronchospasm with maintenance of MAP
5. monitoring
   - i. ECG, NIBP, IABP when possible
   - ii. S_pO_2, $ETCO_2$, AGA's
   - iii. CUD, CVP ± PAOP
   - iv. transfer to ICU
6. other therapy
   - i. antihistamines - ***no benefit*** in acute episode
     - H_2 blockers contraindicated acutely
     - may be useful for ongoing angioedema
     - require both H_1 & H_2 for prophylaxis
   - ii. sedation - if intubated & resuscitation successful
   - iii. steroids - marginal benefit in acute episode
     - may be useful for ongoing bronchospasm & angioedema
     - required in addition to antihistamines for prophylaxis
7. follow-up
   - i. blood specimen
     - ***tryptase*** level - released from mast-cells/basophils, stable in plasma
     - complement - levels decreased with anaphylactoid responses
     - re-type screen & cross-match if due to blood reaction
   - ii. return unused blood products to the blood bank
   - iii. intradermal ***skin testing***
     - histamine releasing agents ~ 1:10,000
     - non-histamine releasing agents ~ 1:1,000
     - graded responses of limited value, use ***absolute*** result
   - iv. medic-alert bracelet & accompanying letter(s)
8. ***prophylaxis***
   - i. methylprednisolone ~ 32mg 12 hrs & 2 hrs pre-exposure
   - ii. diphenhydramine ~ 0.5-1.0 mg/kg 2 hrs pre-exposure

Effects of Anaesthesia

- there is a large amount of contradictory information
- all anaesthetic agents result in short-term, reversible depression of ***chemotactic migration***
- ***phagocytosis*** has been reproducibly inhibited by inhalational agents, intravenous agents, opioids and local anaesthetics *in vitro*, however ***no consistent*** pattern has been demonstrated *in vivo*
- bactericidal activity of ***lysosomes*** has been variably depressed, though only *in vitro*

- effects on CMI and ***lymphocyte function*** have been assessed by,
    1. lymphocyte proliferation in response to mitogens
    2. lymphocyte cytotoxicity
    3. T helper / suppresser ratios

- all agents result in reversible depression *in vitro*, however the results *in vivo* are variable
- greater effects may be seen when N_2O is added to volatile anaesthetics

- no specific change in antibody production has been demonstrated either *in vitro* or *in vivo*
- animal studies do show an increase in ***mortality*** from infection in those animals anaesthetised
- however, this is difficult to study in humans

- the ability of leukocytes to kill tumour cells has been shown to be depressed for up to 7 days following halothane anaesthesia
- similar effects have been shown for other agents *in vitro*

- anaesthesia offers no protection against anaphylaxis

- prolonged exposure to analgesic concentrations of N_2O results in neutropenia & megaloblastic anaemia, due to inhibition of methionine synthetase

ASSESSMENT OF IMMUNE FUNCTION

1. history and examination
2. routine investigations
   i. FBE
   ii. quantitative Ig levels
3. B-cell function
   i. assays for naturally occurring antibodies
      - rubella, influenza, tetanus, diphtheria, etc.
   ii. assay response to immunisation
      - typhoid, polio, CDT, HBV, etc
4. T-cell function
   i. skin tests
      - PPD, candida, trichophyton, tetanus toxoid 1/100
      - contact sensitivity to dinitrochlorobenzene
      - absence of skin reactivity $\rightarrow$ ***anergy***
   ii. CXR in children $\rightarrow$ thymus shadow
5. complement
   - individual component assays are difficult
   - therefore use ***total haemolytic complement*** $\mathbf{CH_{50}}$
6. phagocytic function
   - removal of nitroblue tetrazolium dye from intradermal injection
7. lymphocyte cell cultures
   - stimulation tests and induced lymphokine assays
8. mixed lymphocyte cultures
   - used for compatibility testing in transplant work
   - one set of irradiated WBC's is incubated with a second set
   - a ***blastogenic transformation*** occurs if there is sufficient antigenic discrepancy between the two cell sets
   - this may be clinically manifest as either ***graft rejection*** or ***graft versus host disease***