The Immunology of Allergy
and its clinical implications

By Dr Priya Bowry Sikand
MBBS MRCGP DFFP DIC MSc(Allergy)
Understanding allergies

Back to the Basics....
Objectives

- Understand immunological mechanisms behind Type 1 Hypersensitivity reactions
- Relate the science to the patient and the available tests
- Relate the science to therapeutics
Body must **defend** itself against invasion by foreign substance and **eliminate** it – uses the **IMMUNE SYSTEM**

Employs two tactics – *innate immune response* (**non-specific** response) and *acquired immune response* (**specific** response to specific foreign substances, with **memory**).
Acquired Immune Response

• Specific response: ability to recognise small structures (antigens/allergens) on surface of foreign invader by army of immunocompetent cells - T and B cells.

• B cells (humoral response) produce antibodies against foreign substances (neutralizers, activate complement system)

• T cells (cell-mediated immunity): respond to cells displaying ‘surface markers’. T cytotoxic cells – “NINJAS!”, T helper cells – release Cytokines - stimulate B cells; and regulatory T cells - suppress T helper cells and maintain self tolerance

• Uses memory (T helper cells): to improve responses to subsequent exposures i.e. more prompt and powerful response
<table>
<thead>
<tr>
<th></th>
<th>INNATE IMMUNITY</th>
<th>ADAPTIVE IMMUNITY</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>SPEED OF ONSET</strong></td>
<td>Immediate (within minutes)</td>
<td>~ 3d lag</td>
</tr>
<tr>
<td><strong>SPECIFICITY TO ANTIGEN</strong></td>
<td>Lower</td>
<td>Higher</td>
</tr>
<tr>
<td><strong>DIVERSITY OF RESPONSE</strong></td>
<td>Lower</td>
<td>Higher</td>
</tr>
<tr>
<td><strong>POTENCY</strong></td>
<td>Lower</td>
<td>Higher</td>
</tr>
<tr>
<td><strong>MEMORY (reacts quicker to subsequent exposures)</strong></td>
<td>No</td>
<td>Yes</td>
</tr>
</tbody>
</table>
Extracellular pathogen is present

- Antigen binds to B cell receptor
  - B cell presents the antigen on MHC
    - Activate T helper CD4
      - Activate B cell to make plasma cell
        - Antibody production

Intracellular pathogen is present

- Pathogen is phagocytosed by APC
  - Presentation of antigen by APC
    - Activate T helper CD4
      - Activate macrophage
        - Phagocytosis of Pathogen

- Infected cell presents antigen on MHC
  - Activate cytotoxic CD8 T cell
    - Cell mediated cytotoxicity
Why is the immunology important?
These patients are all ATOPIC

A severe type of allergic reaction that involves two or more body systems (e.g. hives and difficulty breathing).
ATOPY

Atopy is the *genetic* predisposition to develop allergic diseases. It is characterised by a $T_H2$ driven environment resulting in IgE antibody responses. (remember this also occurs in materno-foetal phase to prevent rejection and for immunity against parasitic diseases)

Normal individuals have $T_H1$ driven responses and do not react to innocent foreign substances in the same way as an atopic.
The Allergic Reaction

• Allergic diseases occur when the body makes an *inappropriate, exaggerated* response to seemingly *innocent* and common environmental or dietary substances, known as **allergens**.

• Production of **IgE antibodies** against these allergens result in **inflammation** (swelling, redness, itching, irritation and watery discharge) and hence, disease.
The Battlefield

Allergen specific

IL-4

Allergen specific
THE END RESULT......
Key Players of Allergic Responses

- The Epithelial barrier
- Antigen Presenting Cells eg dendritic cells
- T cells – especially $T_{\text{H}2}$ cells
- B cells and plasma cells
- Mast cells
- Endothelial cells
- Eosinophils and Basophils
- Effector cells eg smooth muscle and nerve cells
The Target and Weaponry

• The target: allergens

• Cell surface receptors

• MHC Class 2 molecules

• Mediators (Cytokines, Interleukins, Chemokines), Co-stimulatory molecules and Adhesion molecules

• IgE

• Histamine and other inflammatory mediators

• Eosinophil products
ALLERGENS

• INNOCIOUS AND INNOCENT
• Soluble & extracellular
• Usually proteinous
• Epitopes (allergenic determinants) on their surface are recognised by the immune system
• Different conformations of epitopes which subsequently affect the allergenicity
• Epitope structure can change conformation with different processes eg heating, digestion
**Allergy in a nutshell**

In allergic responses, B cells produce allergen-specific IgE when allergen is presented by $T_H^2$ cells. Memory $T_H^2$ cells proliferate for future use. When re-exposed to the allergen, Allergen-IgE complex binds to primed Mast cells which release histamine and other inflammatory mediators resulting in the acute clinical symptoms.
$T_H^2$ cells promote proliferation and release of eosinophils which are responsible for chronic symptoms. Eosinophils release powerful tissue damaging inflammatory mediators.
Antigen Presentation

I. Antigen is up-taked by APC after crossing epithelial barrier

II. Antigen is processed and digested into peptide fragments

III. Peptide is loaded onto MHC and presented to TCR on the membrane of T-cell

Naïve T cell ($T_{H0}$) then differentiates to $T_{H2}$ if IL 4 present

APC cell travels to lymphatic system where it meets naïve T cell $T_{H0}$
**Naïve T (T\textsubscript{H0}) cell differentiation:** T\textsubscript{H2} differentiation needs **IL-4** in the cellular environment/milieu when antigen is presented. T\textsubscript{H2} subsequently release cytokines (IL 4, 5, 10, 13) which act on B cells. If IL-12 present, then T\textsubscript{H0} differentiate into T\textsubscript{H1}.
In presence of IL 2, 4 & 5, T_H2 cells interact with B cell, activating it leading to class switching to produce allergen-specific IgE. Thereafter, allergen-specific IgE producing B cells (plasma cells) proliferate.

The TCR and MHC Class 2 receptors and Co-stimulatory molecules participate in the cell to cell interaction promoting the Class Switching of B cells from IgM/G producing to IgE production.

Class switching : T cell - B cell cooperation!
Mast Cell Degranulation

When in contact with allergen, two allergen specific IgE-Abs crosslink with high affinity IgE receptors (FcεRI) on mast cells and basophils leading to mast cell degranulation. Release: histamine, leukotrienes, tryptase, prostaglandins and cytokines.
Acute Inflammation!!!!!!
Immediate (acute) to Late (chronic) reactions
Early and Late Responses

- **Histamine**
  - Predominant mediator types
  - Early-phase allergic response (within minutes)
  - Cysteiny1 leukotrienes
- **Cytokines**
  - Late-phase allergic response (4+ hours)

**Most commonly associated allergy symptoms**
- Sneezing
- Nasal itching
- Rhinorrhea
- Nasal obstruction (congestion)
- Urticaria
- Wheeze
- ANAPHYLAXIS

**Further wheeze**
EØ: $T_H^2$ cells also release *IL 5, IL 13*, together with other chemokines *promote eosinophil proliferation, recruitment and chemo-attractant* to the tissues. There, eosinophils release enzymes which promote tissue damage – late response.
Eosinophils release the following proteins:

- Major basic protein
- Eosinophil peroxidase
- Eosinophil derived neurotoxin
- Eosinophil cationic protein

Eosinophils meant to destroy large pathogens – parasites, so their enzymes are incredibly destructive to tissues when microscopic allergens present!!!
**Effector Sites**

**Mast-cell activation and granule release**

**Skin:**
- Urticaria,
- Angioedema,
- Itching

**Nerves:**
- Itching,
- Irritation

**Gastrointestinal tract**
- Increased fluid secretion, increased peristalsis
- Expulsion of gastrointestinal tract contents (diarrhea, vomiting)

**Airways**
- Decreased diameter, increased mucus secretion
- Congestion and blockage of airways (wheezing, coughing, phlegm)
- Swelling and mucus secretion in nasal passages

**Blood vessels**
- Increased blood flow, increased permeability
- Increased fluid in tissues causing increased flow of lymph to lymph nodes, increased cells and protein in tissues, increased effector response in tissues
Clinical implications

A severe type of allergic reaction that involves two or more body systems (e.g., hives and difficulty breathing).

The Allergy Clinic
North Diagnostic Laboratory
Allergic Rhinitis

Early symptoms (MINUTES): Sneezing, itching (neural), rhinorrhea (mucus secretion)

Late symptoms (HOURS): nasal blockage (vascular)
Chronic Rhinosinusitis
Asthma
reversible airway obstruction, mucus overproduction, airway thickening & narrowing – bronchoconstriction due to smooth muscle hypertrophy
Asthma: remodelling

Remodelling: Permanent structural changes leading to
• Increased airway wall thickness that involves both smooth muscle and collagen tissue
• Increased mucous glands and mucus production
• Increased vascularity, or blood supply, in the airways

Leads to reduced lung function
a) Acute phase

- Allergen
- IgE
- Mast cell
- IL-4
- IL-5
- Leukotrienes
- Histamine

b) Chronic phase

- TNF-α
- Macrophage
- Goblet cell
- Epithelial cell
- Mucus
- Airway damage/inflammation
- Degranulation
- Eosinophil
- IL-4
- IL-5
- IL-13
- T_{H2}

- Smooth muscle hyperplasia and hypertrophy
- Mucus gland hyperplasia
- Chronic inflammation
- T_{H1}
- T_{H2}
- Neutrophil
- Cytokines
- Chemokines
- Prostanoids
- Collagen deposition
- Fibroblast activation

Nature Reviews | Immunology

The Allergy Clinic
North Diagnostic Laboratory
Changes in remodelled airway: Bronchial wall

(A) H&E stained airway from normal control
(B) H&E stained airway in severe asthma. Note thickened subepithelial collagen below the basement membrane (BM), inflammatory cells between the smooth muscle layer (SM) and goblet cell hyperplasia in epithelium
(C) Collagen IV staining of a control airway to demonstrate basement membrane
(D) Asthmatic airway showing true basement membrane beneath epithelium and vessels (VV) in submucosa

The Allergy Clinic North Diagnostic Laboratory
Eczema - Langerhans' cells, inflammatory dendritic epidermal cells, monocytes, macrophages, lymphocytes, mast cells and keratinocytes, all interact through an intricate cascade of cytokines leading to a predominance of Th2 cells which are increased in the skin, and there is a corresponding decrease in Th1 cytokines.
Chronic AD (microbial toxins act as superantigens) results in the infiltration of inflammatory IDECs, macrophages (Mφ), and eosinophils. IL-12 production by these various cell types results in the switch to a Th1-type cytokine milieu associated with increased IFN-γ expression.
The Atopic March

The apparent progression of atopic diseases from severe atopic dermatitis, food allergy through to asthma and rhinitis.

The multi-factorial nature of this Allergic March is complex with several mechanisms yet to be clearly identified.
Principles of Allergy Tests

• The investigations used in allergies depend on the availability of different services in Africa:

  – **NON-SPECIFIC TESTS** raise allergic disease as a differential diagnosis when evaluated with the clinical findings. They cannot offer exact cause of the allergies.

  – **SPECIFIC TESTS** detect *specific IgE antibodies* directed against causative food, drugs, aeroallergens, cosmetics, insect stings etc.
Non Specific Tests

These tests indicate underlying allergic process but there are other differentials to be considered

- Blood Eosinophilia
- Tissue Eosinophilia
- Total serum IgE
- Serum tryptase test
- Exhaled NO
- Pulmonary function tests
Specific allergy tests.

- **In Vivo tests** Skin prick test (SPT) & Intradermal test
- **In Vitro assays** in blood samples from serum or cells
- **Gold standard tests for confirmation**
  1. Airways- nose, bronchial provocation test
  2. Eyes-conjunctival allergen test
  3. Oral Food Challenge – OFC

The skin prick test and serum specific IgE test both identify presence of specific IgE antibodies to allergens & sensitization. Positive result does not necessarily correlate to clinical allergy.
Applying the immunology to therapeutics

• Antihistamines
• Leukotriene antagonists
• Corticosteroids
• Adrenaline
• Immunomodulators
  • Anti IL-4, 5 etc
  • Anti IgE
• Allergen Specific Immunotherapy
Mechanism of antihistamines
Use 2nd generation antihistamines!!!
Mechanisms of Corticosteroids

IMMUNOSUPPRESSIVE AND ANTI-INFLAMMATORY EFFECTS

Acts on intracellular glucocorticoid receptors in cytoplasm. The complex enters nucleus where it binds to the DNA regulatory site and alters gene expression by transcription of mRNA. Affects phospholipase A2 synthesis, arachidonic acid pathway & prostaglandin production thereby preventing inflammation.
Corticosteroids

- Macrophage
  - Apoptotic eosinophil
    - Promotion of clearance
    - Enhanced phagocytic capacity of macrophages

- Eosinophil
  - Increased spontaneous cell death

- T cell

- Fibroblast

- Epithelial cell

- IL-5
- IL-3
- GM-CSF

Inhibition of expression of survival factors

Nature Reviews | Drug Discovery

The Allergy Clinic
North Diagnostic Laboratory
Mechanism of Leukotriene Antagonists
Block leukotriene receptors

Leukotrienes in asthma
Potential sites and effects of cysteiny1 leukotrienes

- Increased mucus secretion
- Decreased mucus transport
- Cationic proteins (epithelial cell damage)
- Increased release of tachykinins
- Sensory C fibers
- Smooth muscle contraction and proliferation

Adapted from Hay DW, et al.\textsuperscript{15}

Figure 2 - Leukotriene actions on airway structures
Adrenaline in Anaphylaxis

- Direct acting sympathomimetic, agonist at alpha and beta adrenergic receptors
- Causes vasoconstriction to counteract the vasodilation & hypotension associated with anaphylaxis
- Bronchodilatory effects
- Downregulates release of mast cell and basophil mediators
Mechanism of anti-IgE monoclonal antibody forms complexes with free IgE and reduces FcεRI expression on mast cells and dendritic cells.
Mechanism of Allergen Specific Immunotherapy (SIT)

SIT represents the only curative & specific method of Rx for allergic diseases by inducing a state of immunological tolerance by modulation of:

1. T&B cell responses and their antibody isotopes
2. effector cells of allergic inflammation eg eosinophils, basophils, mast cells
Increasing doses of antigen drive Tregs to produce IL10 & TGFβ and Th1 cells to produce IFNγ. These downregulate Th2 differentiation and upregulate Th1. IL10 also drives B cells to produce IgG4 (stabilizes mast cells by blocking IgE crosslinking).
Allergen-specific immunotherapy

- Tissue numbers
- Mediator release

- Allergen-specific proliferation
- Numbers in late-phase reaction
- $T_{h1}$ and $T_{h2}$ cytokines in blood
- $T_{h1}$ cytokines in tissues
- $T_{h2}$ cytokines in tissues
- Regulatory T cells, IL-10 and TGFβ

- Allergen-specific IgE
- Seasonal increases in IgE
- IgE-facilitated antigen presentation
- Blocking antibodies: IgG1, IgG4 and IgA
- IL-10

Clinical parameters
- Quality of life
- Symptoms and drug usage
- Allergen-challenge test responses
- Size of, and number of cells in, late-phase reaction
- Skin-prick-test reactivity
  - Prevention of progression
  - Prevention of new sensitization

Copyright © 2006 Nature Publishing Group
Nature Reviews Immunology

The Allergy Clinic
North Devonic Laboratory
In Conclusion....
Immunological mechanisms of allergic diseases are complex with multiple pathways which are self perpetuating and powerful in continued presence of the allergens.....

...WITH SIGNIFICANT CLINICAL IMPLICATIONS!
• Tests chosen must be relevant, ideally to identify specific allergens causing disease so that appropriate avoidance measures may be advised
• Therapeutic drugs which only focus on one aspect of these processes are unlikely to be successful except in some conditions eg anti-IL5 in eosinophilic asthma
• Specific Immunotherapy is the ONLY therapy which is induces immunological tolerance.
And of course, please have a healthy respect for the seemingly innocent allergens...
Any Questions?

THAT IS ALL

THANK YOU FOR YOUR ATTENTION!
Prof T R Bowry & Dr P Bowry Sikand
1st Floor, Upper Hill Medical Centre,
Ralph Bunche Road
P.O.Box 45549 - 00100
Nairobi, Kenya

Telephone: 0716956173
Telephone: 0725645569

E-mail: info@theallergyclinic.co.ke
www.theallergyclinic.co.ke