Hypersensitivity

Stephen Canfield, MD PhD
Assistant Professor
Division of Pulmonary, Allergy
and Critical Care Medicine

Timeline

• 1893 - Emil von Behring
  - Working with diphtheria toxin
    noted that animals would suffer enhanced responses and even death following a second dose of toxin too small to injure normal untreated animals
  - Described this phenomenon as “hypersensitivity”

Timeline

• 1902 - Charles Richet and Paul Portier
  - Set sail on the yacht of the Prince of Monaco to study the effects of marine toxins in mammals
  - Attempted to protect dogs from the effects of toxins by innoculating them at low doses
  - Re-exposure to innocuous doses resulted in a rapid shock and suffocation
  - Coined the term “ana-phylaxis” to emphasize its antithesis to the familiar “prophylaxis”

Timeline

• 1903 - Maurice Arthus
  - Described a stereotypical response in rabbits following repeated intradermal injection of protein antigens
  - The response, characterized by local erythema, induration, hemorrhage and necrosis became known as the “Arthus Reaction”
Timeline

• 1906 - Clemens von Pirquet and Bela Schick
- Coined the term "serum sickness" to describe strange systemic symptoms suffered by some patients weeks after receiving diphtheria or tetanus anti-toxin horse serum
- Postulated for the first time that these hypersensitivity reactions might be the product of immune response
- Named these responses "allergic" from the Greek *allos ergos*, altered reactivity.

Definitions

• Hypersensitivity:
  - Broadest (Abbas) - Disorders caused by immune responses
  - Dysregulated response to foreign antigen
  - Failure of tolerance to self-antigen
  - Practical - Used clinically to refer to aberrant or excessive immune responses generated against foreign antigens, although the same immune processes apply in many autoimmune disease

• Allergy:
  - Symptoms elicited by encounter with foreign antigen in a previously sensitized individual
### Manifestations of Hypersensitivity

• Symptoms frequently are localized to the anatomical site of antigen exposure:

<table>
<thead>
<tr>
<th>Site of Exposure</th>
<th>Syndrome</th>
<th>Common Allergens</th>
<th>Symptoms</th>
</tr>
</thead>
<tbody>
<tr>
<td>Respiratory Mucosa</td>
<td>Allergic Rhinitis</td>
<td>Nasal Pruritis, Rhinorrhea, Congestion</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Asthma</td>
<td>Bronchospasm Chronic Airway Inflammation</td>
<td></td>
</tr>
<tr>
<td>G.I. Mucosa</td>
<td>Food Allergy</td>
<td>Cramping, Vomits/Diarrhea, Hives, Anaphylaxis</td>
<td></td>
</tr>
</tbody>
</table>

### Manifestations of Hypersensitivity

<table>
<thead>
<tr>
<th>Site of Exposure</th>
<th>Syndrome</th>
<th>Common Allergens</th>
<th>Symptoms</th>
</tr>
</thead>
<tbody>
<tr>
<td>Skin</td>
<td>Contact Urticaria</td>
<td></td>
<td>Hives, Pruritis</td>
</tr>
<tr>
<td></td>
<td>Contact Dermatitis</td>
<td></td>
<td>Rash, Pruritis</td>
</tr>
<tr>
<td>Blood</td>
<td>Systemic Allergy</td>
<td></td>
<td>Hives, Edema, Abd. Cramping, Bronchospasm, Hypotension</td>
</tr>
</tbody>
</table>
### Hypersensitivity: Gell & Coombs Classification

<table>
<thead>
<tr>
<th>Immune reactant</th>
<th>Type I</th>
<th>Type II</th>
<th>Type III</th>
<th>Type IV</th>
</tr>
</thead>
<tbody>
<tr>
<td>Antigen</td>
<td>IgE</td>
<td>IgG</td>
<td>IgG</td>
<td>T_{h1} cells</td>
</tr>
<tr>
<td>Effector mechanism</td>
<td>Mast-cell activation</td>
<td>Complement, FcR(^+) cells, phagocytes, NK cells</td>
<td>Complement, Phagocytes</td>
<td>Macrophage activation</td>
</tr>
<tr>
<td>Example of hypersensitivity reaction</td>
<td>Allergic rhinitis, asthma, systemic anaphylaxis</td>
<td>Some drug allergies (e.g., penicillin)</td>
<td>Serum sickness, Arthus reaction</td>
<td>Contact dermatitis, tuberculin reaction</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Cytotoxicity</td>
</tr>
</tbody>
</table>

Common to All Types

- Products of the adaptive immune system
  - Require at least one exposure for sensitization to occur
  - Sensitization can be long lived in the absence of re-exposure (>10 years) due to immunologic memory
Type I (Immediate) Hypersensitivity

- **Antigens:**
  - Classically exogenous, as opposed to “self” (autoimmune)
  - Contact via mucous membranes and at low dose appears to favor type I sensitization

- **Reactions:**
  - Occur within seconds-minutes of exposure
  - Severity ranges from irritating to fatal

- **Immune Effect**
  - Initial antigen contact leads to IgE production
  - On re-exposure, antigen-specific IgE initiates the reaction

IgE Production

- Occurs as part of a secondary immune response (generally multiple or persistent exposures)
- Class switch to IgE is directed by IL-4 and IL-13 (Th2 cytokines), and requires T cell help (CD40L)
- The propensity to make an IgE response to environmental antigens varies among individuals
- “Atopic” individuals are those genetically predisposed to form IgE responses. That is, atopy is heritable
Genetics of Atopy

- Complex, multigenic heritability. Candidate genes:
  - Chrom. 11q - β-subunit of the high affinity Fc,RI
  - Chrom. 5q - Cytokine cluster: IL-3, IL-4, IL-5, IL-9, IL-13
  - TIM (T-cell, Ig domain, Mucin domain) - surface protein, variation assoc. with IL-4/IL-13 prod.
  - IL-12 p40 subunit (assoc. with asthma and AD)

- Variation in IgE response to specific allergens is associated with MHC II genetics
  - DRB1*1501 is associated with IgE responses to specific ragweed pollen proteins

Allergy Epidemic

- Type I Hypersensitivity diseases, including asthma and allergic rhinitis, have been increasing in prevalence in the economically “advantaged” parts of the world for 30 years

- The “hygiene hypothesis” attributes increased allergic disease rates to generally decreasing microbial exposure in early life which would normally provide a Th1-promoting effect
  - Neonatal bias: ↓IL-12 (DC) and ↓IFN-γ (T cells)
  - Birth order: ↓allergy rates among 3rd- and 4th-born children
  - Protective effect of day care
  - 1990 - East/West Berlin immediately after the wall fell: East had ↓vaccination rates, ↑prev. childhood infection, but ↓‘ed asthma
  - Hx of measles or HAV infection, or +PPD ↑allergy rates
Allergy Epidemic

• Weighing against the Hygiene Hypothesis:
  - Despite this epidemiologic data, some evidence is hard to reconcile
  - Previous infection with helminths, which generates a strong Th2 response, is also associated with protection against allergy
  - Early life exposure to pathogens is also associated with decreased risk of autoimmune disease (e.g., type I diabetes), a classic Th1-mediated condition
  - Revised hygiene hypothesis - early life exposure to microbial pathogens influences the balance of immune responsive vs. immune modulating influences

Allergy: Sensitization Phase

• Serum IgE produced by plasma cells has a short $T_{1/2}$ (serum $T_{1/2}$ IgG≈30 days; for IgE≈2 days)
• Rapidly taken up by FcεRI on tissue mast cells and circulating basophils
Allergy: Effector Phase

• *Early Phase Response:* within seconds-minutes
  - IgE crosslinking by antigen \(\Rightarrow\) release of preformed mediators
  - histamine \(\Rightarrow\) smooth muscle constriction, mucous secretion, ↑vascular permeability, ↑GI motility, sens. nerve stimulation
**Allergy: Effector Phase**

*Early Phase Response*: within seconds-minutes
- IgE crosslinking by antigen → release of preformed mediators
- Histamine → smooth muscle constriction, mucous secretion, ↑vascular permeability, ↑GI motility, sens. nerve stimulation

---

**Allergy: Effector Phase**

*Late Phase Response*: 6-24 hours after exposure
- Mast cell production of newly synthesized mediators
  - Leukotrienes → smooth mm. contraction, vasodil., chemotaxis
  - Cytokines → recruitment of PMN and eosinophils
Mast Cell Degranulation

Pre-exposure to Ag  Post-exposure to Ag

FcεRI Signaling

- Structure:
  - Alpha, Beta, Gamma-Gamma
- Alpha - binds IgE monomer
- Beta, Gamma - signal
- ITAM’s
  - Conserved sequences within the receptor tail containing tyrosines
  - ITAM Tyr is phosphorylated on ligand binding
  - Serve as docking sites for downstream activating kinases
Eosinophils

- Innate responder cell in Type I hypersensitivity
  - Production in marrow induced by IL-3, IL-5, GM-CSF
  - Chemotaxis to tissue sites: IL-5, Eotaxin-1, 2, 3
  - "Primed" by IL-5, eotaxins, C5a
    - ↑FcγR and C′ receptor expression
    - induce FcεR expression
    - ↓threshold for degranulation
  - On activation, eosinophils secrete
    - Toxic proteins- major basic protein, eos. cationic protein, eos. derived neurotoxin
    - IL-3, IL-5, GM-CSF, IL-8
    - LT's

Evolutionary Role of Type I Response

- Mast cells line all subepithelial mucosa
  - Rapid recruitment of PMN, eosinophils, monocytes to sites of pathogen entry
  - ↑Lymph flow from peripheral sites to lymph node
  - ↑G.I. motility favors expulsion of G.I. pathogens

- Important role in parasite clearance
  - c-kit−/− mice have no mast cells ↑susceptibility to trichinella, strongyloides
  - Eosinophil depletion (Ab-mediated) severity of schistosomal infection
Evolutionary Role of Type I Response

- STAT6:
  - Mediates IL-4/IL-13 signaling
  - Required for IgE class switch
  - STAT6−/− mice have no IgE

- Wild type or STAT6−/− mice were injected with 500 N. brasiliensis larvae

- Worm counts and fecal egg counts were assessed at 13 days

Type I Sensitivity in Allergy

- Type I Hypersensitivity mediates:
  - Allergic Rhinitis/conjunctivitis (Hayfever)
  - Asthma
  - Food/Medication reactions
  - Contact urticaria
  - Some forms of eczema
  - Anaphylaxis - food, bee sting, drug, exercise-induced
Type I Sensitivity in Allergy

• Documenting allergic sensitivity: skin testing
  - Allergenic extract (airborne, food, venom) is introduced by prick or injection intracutaneously
  - Sensitization is evident within 15-20 minutes as a wheal/flare at the allergen introduction site

Anaphylaxis

• Response to systemic circulation of allergen
  - Triggering of mast cells in peri-vascular tissue
  - Circulating histamine, PG’s/LT’s vascular leak, vasodilatation
  - High-output shock (increased cardiac output, ↓↓BP)
  - Other symptoms: urticaria, flushing, wheeze, laryngeal edema with airway compromise, G.I. cramping, diarrhea
• Rapid progression over seconds-minutes
• Treatment -
  - Early administration epinephrine I.M., followed by antihistamines (H1 and H2 blockade) treat early phase
  - Subsequent administration corticosteroids prevent late phase
Type II Hypersensitivity: Antibody (Ab) Mediated

- Target-specific IgM and IgG mediate damage
- Targets:
  - Self-molecules altered by foreign antigen \( \wedge \) neo-epitope
    - penicillin conjugates to RBC surface proteins \( \wedge \) new penicilloated-protein serves as a target for IgM/IgG \( \wedge \) intravascular hemolysis
  - Self-molecules unaltered = breaking of tolerance
    - Group A Strep pharyngitis yields Ab’s to the Strep M protein \( \wedge \) Ab’s cross react with cardiac muscle and valves \( \wedge \) scarring

Type II Hypersensitivity: Ab Functions

- The mechanisms of type II hypersensitivity are exactly the those of normal Ab function, plus some:

<table>
<thead>
<tr>
<th>Ab Function</th>
<th>Target</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>Opsonization</td>
<td>Platelet surface proteins</td>
<td>Splenic clearance, thrombocytopenia</td>
</tr>
<tr>
<td>Neutralization</td>
<td>Acetylcholine receptor</td>
<td>Myasthenia Gravis</td>
</tr>
<tr>
<td>ADCC</td>
<td>Glomerular basement membrane proteins</td>
<td>Goodpasteur’s Disease</td>
</tr>
<tr>
<td>C’ Fixation</td>
<td>Penicilloyl-RBC protein conjugates</td>
<td>Hemolytic anemia</td>
</tr>
<tr>
<td>Non-Physiologic</td>
<td>TSH receptor</td>
<td>Grave’s Disease</td>
</tr>
</tbody>
</table>
Type III Hypersensitivity: Immune Complex Mediated

• First Description: Arthus Reaction
  - Rabbit received horse serum containing anti-toxin antibody
  - After several days, antigen (toxin) was injected subcutaneously
  - Classic Arthus reaction occurs within 5-8 hours:
    » Local erythema/tenderness with edema, necrosis, hemorrhage

Arthus Reaction

• Immune Mechanism
  - Antibody-Antigen complexes form within blood vessel walls
  - Complement fixation generates C5a
    » Neutrophil chemoattractant  PMN infiltration
    » Anaphylatoxin - local mast cell histamine release  tissue edema
  - Neutrophil activation by FcγR's  release of cytotoxic enzymes
  - Platelet aggregation by FcγR's  small vessel thrombosis, necrosis
  - Local macrophage release of IL-1, TNF-α, and IL-8 - propagation
Type III Hypersensitivity: Immune Complex Mediated

- Serum Sickness: Systemic Arthus-like reaction
    - Rash, fever, lymphadenopathy and arthralgias in recipients of anti-diphtheria antiserum made in horses (hint: 2-3 weeks post-infusion)

- Rabbit Model (Dixon and Lambert, 1960’s):
  - Injection of radiolabeled bovine serum albumin (BSA) day zero
  - Serum BSA and anti-BSA antibody levels were tracked
  - Look for serum immune complexes and proteinuria
Importance of C5a in I.C. Disease

• Mouse Model of Immune Complex Disease:
  - Infuse Anti-ovalbumin Ab via trachea; ovalbumin via I.V.
  - I.C.’s form at respiratory capillaries; examine histology at 4 hours

<table>
<thead>
<tr>
<th></th>
<th>Intratracheal Anti-Ova Ab</th>
<th>I.V. Ova</th>
<th>Genotype</th>
</tr>
</thead>
<tbody>
<tr>
<td>+</td>
<td>+</td>
<td>+</td>
<td>C5aR+/+</td>
</tr>
<tr>
<td>+</td>
<td>+</td>
<td>+</td>
<td>C5aR−/−</td>
</tr>
</tbody>
</table>


Importance of FcγR’s in I.C. Disease

• B/W Mouse - spontaneous accumulation of I.C.’s in the glomerulus
• FcγRI and FcγRIII - contain ITAM’s; activating for phagocytes
• γ-chain knockout (γ−/−): Lacks expression of FcγRI and FcγRIII

Immunology Wars

• Epic Immunologic Battle: 1870-1950
  – “Humoralists” (France): Hypersensitivity is mediated by serum factors
  – vs.
  – “Cellularists” (Germany): Hypersensitivity is mediated by phagocytes

• By 1915, the Humoralists appeared to have won
  – Hay fever, asthma, anaphylaxis
  – Drug-induced hemolysis
  – Arthus reaction, serum sickness

Type IV Hypersensitivity: Tuberculin Reaction

• 1892 - Robert Koch
  – Discoverer of tubercle bacillus
  – Attempted to prevent TB by inoculation with bacillus extract
  – Unfortunately:
    -- No protection for naive individ.
    -- Reactivated disease in exposed
  – But: intradermal injection of bacillus extract in previously exposed individuals resulted in a stereotypic indurated lesion within 48-72 hours
Type IV Hypersensitivity: Delayed Type

• 1942 - Karl Landsteiner and Merrill Chase
  - Demonstrated transfer of tuberculin test sensitivity in guinea pigs
  - Sensitivity is transferred from TB-exposed to unexposed animals with leukocyte transfer, but not with serum transfer
  - Redemption for the Cellularists

Delayed Type Hypersensitivity

• Group of related responses to antigen, all dependent on cell-mediated immunity

• Although prior sensitization is required, reactions occur over 1-3 days following re-exposure

• T cells: necessary and sufficient to elicit the reaction
  - Athymic subjects (animal or human) are not sensitizable
  - T cell depletion (via anti-T cell Ab’s) reverses sensitization
  - Transfer of purified T cells confers sensitization
Varieties of DTH Reactions

<table>
<thead>
<tr>
<th>Type</th>
<th>Reaction Time</th>
<th>Clinical Appearance</th>
<th>Histology</th>
<th>Site/ Antigen</th>
</tr>
</thead>
<tbody>
<tr>
<td>Contact</td>
<td>48-72 hours</td>
<td>Eczema</td>
<td>T cells followed by macrophages, edema of the epidermis</td>
<td>Epidermal: organic mols., poison ivy, heavy metals</td>
</tr>
<tr>
<td>Tuberculin</td>
<td>48-72 hours</td>
<td>Local Induration</td>
<td>T cells, monocytes, macrophages, basophils, fibrin deposition/edema</td>
<td>Intradermal: PPD, candida, mumps</td>
</tr>
<tr>
<td>Granuloma</td>
<td>21-28 days</td>
<td>Hardened Nodular</td>
<td>Macrophages, epithelioid giant cells, fibrosis</td>
<td>Skin, viscera: persistent Ag (TB, leprosy)</td>
</tr>
</tbody>
</table>

Common to all DTH Reactions

- **Histology of the DTH reaction:**
  - T Cells - CD4 (Th1); some forms CD8
  - Macrophages/monocytes
  - Basophils
  - Fibrin
  - If persistent antigen: multinucleated giant cells; granulomata

- **Cytokines found at the site of a DTH reaction:**
  - IL-2
  - IFN-γ
  - TNF-α
  - Macrophage chemotactic protein (CCL-2)
Contact Sensitivity: Hapten DTH

• Phase One: Initial Exposure - Sensitization
  - Hapten - small organic molecule, frequently lipophilic crossing epidermal barrier by diffusion, associates with cell proteins
    - Chromates (leather tanning), urushiol (poison ivy), nickel
  - Haptenylated proteins are taken up by Langerhans’ cells - peptides bearing hapten are loaded onto MHC I and MHC II
  - LC’s migrate to regional lymph nodes, activate naive T cells

• Phase Two: Re-exposure - Elicitation
  - Hapten-specific memory T cells bearing the cutaneous lymphocyte antigen (CLA-1) continuously migrate between lymphatics and skin
  - Re-encounter with haptenylated protein may occur on:
    - Langerhans’ cell (MHC II) ▶ Th1 cell secretion of IFN-γ, MCP-1 with macrophage recruitment
    - Keratinocyte (MHC I) (lipophilic hapten) ▶ CD8 CTL activation ▶ release of perforins and granzyme ▶ local tissue damage
Hypersensitivity Progression

• Antigen-specific responses may progress from one type of hypersensitivity to another:
  - Latex allergy among healthcare workers
    ‣ Initial reaction is typically a contact sensitivity (type IV reaction)
    ‣ With recurrent latex contact, sensitivity progresses to latex-specific IgE, imparting risk of anaphylaxis
  - p-aminobenzoic acid (PABA), the active ingredient in many sunscreens, can act as a contact sensitizer
    ‣ PABA DTH reactivity is associated with ↑ed risk of immediate type hypersensitivity to local anesthetics (e.g., benzocaine) due to cross-reactivity of the aromatic core

Penicillin Mediates All Types of Hypersensitivity

• Immune-mediated adverse reactions occur at a rate of 1 per 100 administrations

<table>
<thead>
<tr>
<th>Type</th>
<th>Mechanism</th>
<th>Example</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>IgE-mediated</td>
<td>Acute anaphylaxis, urticaria</td>
</tr>
<tr>
<td>II</td>
<td>C’-mediated cytolysis Opsonization</td>
<td>Hemolytic anemia Thrombocytopenia</td>
</tr>
<tr>
<td>III</td>
<td>Immune Complex Damage</td>
<td>Serum sickness Drug fever, Vasculitis</td>
</tr>
<tr>
<td>IV</td>
<td>T Cell mediated</td>
<td>Contact sensitivity</td>
</tr>
</tbody>
</table>