Mouse Models of Inflammatory Lung Diseases

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Outlines

- Basic lung structure, function, and inflammatory lung diseases
- Methods, techniques, and approaches to studying lung diseases
- Mouse models of inflammatory lung diseases

Lung: Basic Structure
**Lung: Basic Structure**

Lung Histology  
H&E (100x)

- Airways
- Alveoli
- Blood vessel

**Lung Function and Diseases**

- Gas exchange: $O_2$ from atmosphere to bloodstream and $CO_2$ from bloodstream to atmosphere
- Diseases
  - COPD
  - Asthma
  - Pneumonia
  - Pulmonary fibrosis
  - Sarcoidosis
  - Cystic fibrosis
  - Lung cancer
  - ...

**Studying the Lung: Methods and Techniques**

- Pulmonary Function Test (PFT)
  - Airway resistance
  - Lung compliance
- Bronchoalveolar Lavage (BAL)
  - Inflammatory cells in the airway: total and differential
  - Mediators: cytokines, chemokines
- Lung tissues
  - Histology: H&E, Alcian Blue, PAS, Trichrome
  - Proteins
  - RNA
  - Immunohistochemistry (IHC)
  - FACS analysis of cell populations
  - Culture of specific cell types
Studying the Lung: Approaches

- Stimulation
  - Allergens
  - Cigarette smoke
- Transgenics
  - Inflammatory cytokines
- Knock-out & knock-in
  - Cytokines & receptors
  - Siglecs
- Mutant strains
  - Phosphatases

Allergen Induced Allergic Asthma

Protocol for ovalbumin (OVA) induced allergic asthma

Day 0
Immunization with OVA
50 µg OVA + 1 mg alum in 200 µL

Day 7
PFT & Sacrifice

Day 14
Day 16
Intranasal 50 µg OVA

OVA Induced Airway Inflammation

BAL Cells

PBS
OVA

Macrophages
Eosinophils
**OVA Induced Lung Inflammation**

- **Trichrome (400x)**

  - PBS
  - OVA

**Elastase Induced Emphysema**

- **Protocol for elastase induced emphysema**

  - **Porcine Pancreatic Elastase**
    - 1u/mouse, I.T. once

  - **Day 0 1 2 3 7 14 21 28**

  - **Sacrifice & Collect BAL, lung tissue**

**Elastase Induced Airway Inflammation**

- **BAL Cells**

  - **Day 2**
  - **Day 21**
Elastase Induced Emphysema

Lung Histology (H&E 20x)

Day 3  Day 21

SHP-1

Src Homology 2 domain-containing protein tyrosine phosphatase-1

Encoded by the hcp gene
Primarily expressed in hematopoietic-derived cells
Negative regulator of a variety of growth factor/cytokine signaling pathways
Activated SHP-1 → binds to its target proteins → dephosphorylates the phospho-tyrosine residues → terminates the signaling

SHP-1 Regulation of Signaling

Inactive  Active
SHP-1 Deficiency in Mice

- Motheaten mice (me/me) – a natural mutant strain
- Viable motheaten mice (me/me<sup>+</sup>) – another spontaneous mutant strain

⇒ The mutations cause aberrant splicing of the SHP-1 transcript, resulting in diminished production of functional SHP-1 protein

Phenotype
inflammatory lesions of the skin, increased neutrophils in the peripheral blood, glomerulonephritis, abnormal lymphoid tissues, pneumonitis with macrophage infiltration in the alveoli, increased production of immunoglobulins, depressed immune responses, profound NK-cell deficiency, and a shortened life span (average 22 days and 61 days, respectively)

IL-4/IL-13 Signaling Systems

Type I
IL-4

Type II
IL-4/IL-13

SHP-1 Expression and Activation in Lung Epithelial Cells
Hypothesis

SHP-1 is an important regulator of
• Immunological homeostasis of the lung
• Allergic inflammatory responses

SHP-1 Deficiency and Phenotype

mev/+: Phenotypically Normal
mev/mev: Inflammatory phenotype

Pulmonary Inflammation
Mucus Hyperproduction

A. PAS Alcian Blue

WT

me

B. RT-PCR

MUC5AC

β-Actin

WT

me

Immunoblot

MUC5AC

Pulmonary Fibrosis

Trichrome

RT-PCR

ELISA

Airway Resistance and Hyperresponsiveness

Airway Resistance and Hyperresponsiveness
Upregulation of Th2 Cytokines

Role of Th2 Cytokines in Pulmonary Inflammation

Increased Mast Cells in the Lung
Mast Cells and Lung Inflammation

Crossbreeding to mast cell deficient K\textsuperscript{mev} mice

- Spontaneous and progressive pulmonary inflammation rich in macrophages, lymphocytes, and eosinophils
  - ↑ Th2 cytokines IL-4, IL-5, and IL-13
  - ↑ Th2 chemokines Eotaxin and MCP-1
- Mucus hyperplasia  ▲ Mucin genes
- Subepithelial and parenchymal fibrosis  ▲ TGF-β
- Increased airway resistance and AHR
- Mast cell-dependent

Summary

mev mice:

- Spontaneous and progressive pulmonary inflammation rich in macrophages, lymphocytes, and eosinophils
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