ALLERGIC BRONCHOPULMONARY ASPERGILLOSIS

How could we diagnose it earlier?

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Take home message

Rare

When do you need to think about it?

1)  

2) Pulmonary infiltrate or clinical deterioration do not respond to one week of antibiotherapy

3) Major increase of the total serum IgE upon annual screening (even if not specific)
ABPA: introduction

• **Definition**
  - Complex hypersensitivity reaction, often in patients with asthma or cystic fibrosis (CF),
  - Occurs when the bronchi become colonized by *Aspergillus fumigatus* (AF)

• **Incidence**
  - Comprise 2-10% of the subjects with CF (rare)
  - Not all CF patients colonized with AF will develop ABPA

• **Symptoms: non-specific**
  - Repeated episodes of bronchial obstruction, inflammation, and mucoid impaction
  - Lead to bronchiectasis, fibrosis, and respiratory compromise

• **Gaps in clinical management**
  - **Tests are not optimal:** skin test, specific IgE, sputum culture
  - Therapies not optimal: steroids, antifungal, anti-IgE
Aspergillus fumigatus

is widespread in nature: soil and decaying organic matter.

Spores are ubiquitous in the atmosphere.

Everybody inhales several hundred spores each day quickly eliminated in HC.

No relation between the intensity of exposure and the rate of sensitization to the fungus as measured by skin test.

Not much can be done to reduce exposure.
A. fumigatus and human disease: 3 distinct entities

**Chronic pulmonary Aspergillosis**

In patients with preexisting lung-cavities or damage

- Hemoptysis, cough
- Low grade fever

**Invasive infection**

In mildly immunodeficient patients

- Most commonly kidney, liver, spleen, and central nervous system
- Pulmonary, nasal involvement

**ABPA**

In CF or asthma

- Lung damage, fibrosis, bronchiectasis
ABPA: therapeutic management (2003 consensus)

- **Anti-inflammatory: Glucocorticoids**
  0.5-2 mg/kg/day for 1-2 weeks
  0.5-2 mg/kg/evryother day for 1-2 weeks
  Taper off within 2-3 months

- **Antifungal: Itraconazole-Sporanox**
  Slow or poor response to corticosteroids, relapse, corticodependent or toxicity
  5 mg/kg/day, max 400 mg/day for 3-6 months

- **+/- Anti-IgE-Omalizumab**
  If poor response to corticosteroids, relapse, corticodependent or toxicity
ABPA: radiographic criteria

- New or recent changes in chest radiograph/CT (infiltrate, mucous plugging)

CXR: bronchial wall thickening and impressive central bronchiectasis

CT: varicoid and cystic central bronchiectasis in all 5 lobes and mucous plugging
• Criteria for CF-ABPA were updated in 2003 (CFF)

Acute or sub acute clinical deterioration (cough, expectoration of brownish mucous plugs, hemoptysis)
• Criteria for CF-ABPA were updated in 2003 (CFF)

- Acute or sub acute clinical deterioration (cough, expectoration of brownish mucous plugs, hemoptysis)

- Serum total IgE > 1,000 UI/ml

- Immediate positive skin test to *A. fumigatus* >3 mm or positive specific IgE to *A. fumigatus*

- *A. fumigatus* positive precipitins or presence of anti-*A. fumigatus* antibodies

- New or recent changes in chest radiograph/CT (infiltrate, mucous plugging)

- Should be suspected if patient with pulmonary infiltrate or clinical deterioration that do not subside after one week of antibiotherapy
ABPA: questions

• Unmet need for better diagnostic test, ideally:
  • Blood-based (minimally invasive)
  • Will discriminate CF subjects with ABPA from CF subjects with stable Aspergillosis colonization
  • Will provide objective, quantitative assessment of treatment response in patients under therapy
A basophil assay for CF-ABPA?

- Mediates the hypersensitivity type I: ALLERGY which is a key element in ABPA
- Represent less than 1% of total leukocytes (white cells in blood).
- Originate and develop in bone marrow from hematopoietic CD34+ stem cells
- Are released into circulation as mature cells (# from mast cells).
- Survival: 2-3 days (<< mast cells).
FACS analysis of basophils

Dead cells were excluded

Gernez et al. Int Arch Allergy Immunol 2010
Basophil CD203c and CD63 in blood from patients with nut allergy could discriminate the allergic patient from the HC and could identify the offending food allergen.

NA: patients with nut allergy
HC: Healthy controls

Gernez et al. Int Arch Allergy Immunol 2010
Anti-IgE therapy decreases basophil CD203c
Development of a FACS-based blood basophil assay for CF-ABPA

ONE DROP OF WHOLE BLOOD

Baseline

In vitro stimulation within 10/30 minutes

Surface staining for basophil CD203c and CD63

Fixation

FACS

- 4 groups of patients:
  1) CF and ABPA
  2) CF with A. Fumigatus in their sputum
  3) CF patients (without ABPA/A. fumigatus colonization)
  4) Healthy controls
Basophil CD203c and CD63 in blood from CF subjects with ABPA, following a stimulation with the fungus

Blood basophil CD203c levels were significantly increased in CF patients with ABPA following 10-minute of *ex vivo* activation with *A. Fumigatus* compared to the 2 other groups.
Level of basophil CD203c, following stimulation with the fungus could distinguish CF subjects with ABPA from CF subjects with *Aspergillus* in their sputum.

**ROC Curve:** Basophil CD203c could distinguish patients with CF and ABPA from CF patients with *A. Fumigatus* in their sputum (P=0.0039).
In the group of CF patients with ABPA, was the increase of basophil CD203c specific to *A. fumigatus*?
Basophil CD203c and CD63 response in blood from 8 CF subjects with ABPA

Blood basophil CD203c and CD63 levels were specifically increased in the sample from CF patients with ABPA following 10-minute of *ex vivo* activation with *A. fumigatus*

Ag1: offending allergen
Ag2: non offending allergen
High unmet needs for blood assays to both diagnose and monitor response to therapy and for new targeted therapies in patients with CF-ABPA

Our blood basophil CD203c assay could improve:
- the diagnosis of ABPA in Cf patients (CD203c following *ex vivo* stimulation)
- the monitoring of responses to therapy

Most important: **FAST, SAFE, EASY, REPRODUCIBLE**, appropriate for all ages,
ABPA project: Summary

Future goals

Study the interaction Fungus-host immune response (sputum)

Collect additional samples to establish the value of this assay as a new diagnostic blood assay, which, hopefully will be beneficial to CF patients
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THANK YOU SO MUCH
  to Kriss Benson

For her review

THANK YOU SO MUCH
  to all the patients :)

It could not be done without you
Table 9. Treatment recommendations for allergic bronchopulmonary aspergillosis (ABPA) in cystic fibrosis (CF).

<table>
<thead>
<tr>
<th>Total serum IgE, IU/mL</th>
<th>Pulmonary symptoms and/or worsening PFT results</th>
<th>New infiltrates on CR or CT</th>
<th>Positive serology&lt;sup&gt;a&lt;/sup&gt;</th>
<th>Treatment recommendation(s)</th>
</tr>
</thead>
<tbody>
<tr>
<td>&gt;1000 or &gt;2-fold rise from baseline</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Treat for ABPA</td>
</tr>
<tr>
<td>&gt;1000 or &gt;2-fold rise from baseline</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
<td>No treatment; monitor IgE, CR, PFT</td>
</tr>
<tr>
<td>&gt;1000 or &gt;2-fold rise from baseline</td>
<td>No</td>
<td>Yes</td>
<td>Yes</td>
<td>Treat for CF-related infection; consider treatment for ABPA if no response</td>
</tr>
<tr>
<td>&gt;1000 or &gt;2-fold rise from baseline</td>
<td>Yes</td>
<td>No</td>
<td>Yes</td>
<td>Consider treatment for ABPA, CF-related infection, and/or asthma</td>
</tr>
<tr>
<td>&gt;500 in the past; no change from baseline</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Treat for CF-related infection; consider treatment for ABPA or asthma if no response</td>
</tr>
<tr>
<td>500–1000</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Treat for ABPA</td>
</tr>
</tbody>
</table>

NOTE. CR, chest radiography; PFT, pulmonary function testing.

<sup>a</sup> Aspergillus-specific IgG or IgE or presence of precipitins to *Aspergillus fumigatus*. Because these test results may not be available quickly, they are not required for initiation of therapy but should be obtained.
What about the sputum?

- Measurement of the blood and airway neutrophils might provide a better understanding of the pathology.
Mutant CFTR

Passive release of HNE

Massive neutrophil recruitment to airways

Neutrophil necrosis

Airway obstruction

Opportunistic infections

Massive neutrophil recruitment to airways

Neutrophil necrosis

Passive release of HNE

Chronic airway disease

Current therapies

Key assumptions

• Lung inflammation occurs late in the course of the disease

• Inflammation/obstruction is due to neutrophil death in the lung

• CF airway neutrophils are not functional

NEW DISEASE PARADIGM

Tirouvanziam R.
Large numbers of viable neutrophils are present in CF sputum (airway)

1) Single events
2) Live non apoptotic cells
3) Granulocyte subset
4) Neutrophil subset

Blood

Airway

Fsc A
Fsc H

Gsb
AnnV

Ssc
CD66b

Fsc A
CD45

Tirouvanziam R.