NON-SPECIFIC BRONCHIAL HYPERRESPONSIVENESS

J. Sastre.
Allergy Service
Asthma: definition

• Asthma is a syndrome defined on basis of three main aspects:
  1- Symptoms due to variable and reversible airway obstruction
  2- Bronchial inflammation
  3- Hyperresponsiveness to certain stimuli
Types of Bronchial Hyperresponsiveness

- **NON-SPECIFIC**: methacholine, histamine, AMP(adenosine), hypertonic saline, acetaldehyde, dry air, exercise

- **SPECIFIC**: allergens, chemicals
Non-specific bronchial hyperresponsiveness in asthma:

- Diagnosis of asthma vs other diseases
- Diagnosis of exercise induced asthma
- Follow-up (clinical)
- Mode action of drug medications
- Evaluate changes after specific challenges
- Epidemiological studies
ASTHMA CONTROL AND BHR


mild exacerbation rates
ASTHMA CONTROL AND BHR

The graph shows the dosage of inhaled steroids in micrograms (µg) over the course of a 24-month follow-up period. Two strategies are compared:

- **AHR-strategy**: This strategy shows a significant increase in dosage from baseline to the first month, followed by a steady decline over the next 24 months. The dosage fluctuates slightly but remains below the baseline level throughout the follow-up period.

- **Reference-strategy**: This strategy shows a gradual decrease in dosage from baseline to the 18th month, after which it stabilizes. The dosage remains below the baseline level throughout the follow-up period.

The error bars indicate the variability in the dosage measurements.
Bronchial Challenge with gluteraldehyde

\[
PG_{20} > 16 \text{ mg/ml} \quad PG_{20} = 0.74 \text{ mg/ml}
\]

\begin{align*}
\text{Descenso del VEMS} (\%) \\
\text{Tiempo tras la provocacion}
\end{align*}

- **Dia 1:** Glutaraldehído 2% 10 min
- **Dia 2:** Glutaraldehído 2% 10 min
## Increase of HBR after SIC

<table>
<thead>
<tr>
<th>Study</th>
<th>Cases related to negative SIC</th>
<th>% related to negative SIC</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vandenplas et al</td>
<td>5/41</td>
<td>12%</td>
</tr>
<tr>
<td>Thorax 1996</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Kopfersmichmitt et al.</td>
<td>3/11</td>
<td>27%</td>
</tr>
<tr>
<td>Respir Med 1998</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sastre et al.</td>
<td>3/16</td>
<td>19%</td>
</tr>
<tr>
<td>Chest 2003</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Site of action of stimuli that act directly and indirectly to cause airway narrowing

Modified from
Spicuzza L, Bonfiglio C, Polosa R. TiPS 2003
Non-specific bronchial hyperresponsiveness

Direct Agonists

➡ Methacholine:
- The most commonly used worldwide
- Very good sensitivity
- Asthmatic range < 8-16 mg/ml PC20

➡ Histamine:
- Similar to methacholine.
- Asthmatic range < 8-16 mg/ml PC20
Non-specific bronchial hyperresponsiveness

Indirects (induce mast cells degranulation, more related to bronchial inflammation)

- AMP-adenosine-: acts on receptors A2b.
  - No parallel to methacholine.
  - More influenced by steroids.
  - Asthmatic range <400 mg/ml. PC20-PC15
- Mannitol
- Eucapnic hyperventilation
- Exercise: in the lab or in the field (low sensitivity)
- Hypertonic saline (low sensitivity)
- Acetaldehyde (<40 mg/ml) (low sensitivity)
Steps in the response to various challenge tests

Respiratory Water Loss from airway surface
  ↓
Mucosal Dehydration
  ↓
Increase in [Na⁺], [Cl⁻], [Ca²⁺], [K⁺]
  ↓
Increase in osmolarity

Airway surface liquid  Epithelial Cells  Submucosa

Exercise / Dry Air Hyperpnea

Hyperosmolar aerosols
  ↓
AMP
  ↓
Methacholine

Presence of Airway inflammation (eosinophils, mast cells)
  ↓
Mediator Release from Inflammatory Cells
  ↓
Bronchial smooth muscle contraction

SD Anderson AAAAI 2007 Session 3301
¿Tidal volume and dosimeter, are they the same?

Figure 1. Comparison of tidal-breathing PC_{20} and dosimeter PC_{20} values in 17 subjects with measurable PC_{20} values for at least one method. Horizontal lines = geometric means; dashed lines = the five subjects with unmeasurable PC_{20} by dosimeter (in these subjects, PC_{20} values were censored to 400 mg/mL).

Prieto, et al. Chest 2006;130;1448-1453
Pharmacological 'direct' challenge showing effect of different doses at start of study

PD_{20} FEV_{1}, histamine, µmol

0 8 16 24 32 40 48 56 64 72

Weeks of Budesonide treatment

AHR remission
Mild AHR
Moderate AHR
Severe AHR

- 3200 µg Wks 1-8
- 1600 µg Wks 1-8


n=61
Demonstrates that in asthmatics the AHR to AMP diminishes more rapidly than to methacholine in response to 6 wks Rx with Budesonide.

Properini G. et al. JACI 2002
AHR to AMP is reduced more than AHR to methacholine

Properini G et al. JACI 2002
Response to AMP predicts failure to ICS reduction

Group with -ve PC20AMP results

Group with +ve results

PC20AMP < 400 mg/ml

O.R. = 8.17

% subjects without exacerbations

Weeks after steroid reduction

LFT and eNO did not have predictive value at any time points!

Prieto L et al. CHEST 2003; 124: 1325–33
Kaplan-Meier curve: AHR to both histamine & to mannitol at baseline (solid line) better predicts a failure to halve steroid dose than AHR to only one test (dashed line).

The odds ratio was 4.38 (1.03 -18.66) p<0.05 to predict failure at or before the 2nd ICS reduction.

BIOMARKERS

Comparison of Bronchial Hyperresponsiveness to Methacholine and Adenosine and Airway Inflammation Markers in Patients with Suspected Asthma

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Background. Bronchial hyperresponsiveness is usually measured by bronchial challenge test with direct (e.g., methacholine) and indirect (e.g., adenosine) agonists. There are few studies comparing both types of agents and they have had conflicting concordance. Objective. We sought to compare the results of both tests in a population with symptoms suggestive of asthma so as to determine their relationship with bronchial inflammatory markers. Methods. Seventy-nine patients whose age ranged from 14 to 81 years were recruited for this study. Challenge tests were performed using the tidal volume method. PC20 methacholine and PC15 and PC20 adenosine were calculated. Induced sputum and fraction of exhaled nitric oxide measurements were also performed. Results. Atopy was found in 69% of the patients. Methacholine PC20 and adenosine PC15 were positive in 32 patients (40.5%), both having a sensitivity of 73%. Percentage of agreement was 45.45% and κ index was only 0.369. Adenosine PC20 elicited lower sensitivity and agreement. No correlation between methacholine PC20 and adenosine PC15 was observed. Higher fraction of exhaled nitric oxide values and sputum eosinophil counts were seen in patients with positive adenosine challenge results. The use of adenosine PC15 or PC20 did not alter the association with inflammatory markers. Conclusions. The concordance between both techniques was low. Methacholine is not a reliable predictor of hyperresponsiveness to adenosine, leading us to conclude that the two tests are complementary but not interchangeable in clinical practice. Additionally, responsiveness to the two bronchoconstrictor stimuli does not indicate presence of the same airway abnormality. Indirect stimuli provide a better reflection of bronchial inflammation.

Keywords adenosine, asthma, bronchial challenge, bronchial hyperresponsiveness, fraction of exhaled nitric oxide, induced sputum, methacholine
Comparison of bronchial hyperresponsiveness to Methacholine and Adenosine in clinical practice.

Results
- 46 patients with a mean age of 40.6±13 y.o.
- 36 patients were atopic with at least one positive skin prick test.
- In 20 (43%) both tests were negative.
- MethacT (PC20<16 mg/ml) was positive in 22(46%),
- 15(32.6%) had a positive AMP-PC20 (<512mg/ml) and 17 (36%) positive AMP-PC15 (<512mg/ml).
- In 11 patients MethacT was positive and AMP-PC20 negative and,
- in 5 AMP-PC20 was positive but with negative MethacT.
- In the latter group, 4 out of 5 had high FENO values. Presence of atopy, ACT score, FENO and %FEV1 predicted values was not associated with positive results to one or either test.

Conclusions
- MethacT appears to be a more sensitive than adenosine to demonstrate bronchial hyperresponsiveness in clinical practice. Although, isolated hyperresponsiveness to adenosine may be found and associated to high FENO levels.
Comparison of Methacholine and Adenosine Inhalation Challenge in Patients with Suspected Asthma

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Objective. Bronchial hyperresponsiveness is usually measured by bronchial challenge with direct (e.g. methacholine) and indirect (e.g. adenosine) agonists. A prospective, randomized, crossover, single-blind study was performed to compare both methods in the first diagnosis of asthma. Patients and methods. Fifty-seven patients, in which asthma was suspected, were selected (21 male, 36 female). Fifty-four underwent both challenges following the five-breath dosimeter protocol. PC20 was calculated according to ATS guidelines. Data of symptoms developed during the challenge, PC20 methacholine and adenosine, and FEV1 improvement after bronchodilator therapy were recorded. Results. Symptoms at consultation were consistent with asthma in 68.4% patients, asthma and rhinitis in 29.8% and exercise-induced asthma in 1.8%. Atopy was reported in 93%; 49.1% had family history of atopy and 26.3% of asthma. The most frequent symptoms developed during the challenge were cough (40.4% with adenosine and 20.4% with methacholine) and wheezing (26.3% and 7.4% respectively), statistically significant differences. Bronchial challenge with MCh resulted positive in 44.4% of the patients and positive with AMP in 50%. Every patient with negative result to adenosine, was also negative to methacholine. In 94.4% subjects the result of both challenges was concordant (kappa index = 0.889). PC20 in both challenges showed poor linear correlation (Pearson r = 0.43, p < 0.05). Positivity of both challenges was only associated with having a positive skin prick test to dandrs (p = 0.001). Percentage of improvement after bronchodilator was 34.9% (SD12.2%) with adenosine challenge and 33.9% (SD17.9%) with methacholine (differences non statistically significant). Conclusions. Concordance in the result of both techniques is very high. Cough and wheezing are more frequent with adenosine, though not severe. PC20 with both techniques shows poor linear correlation.
Exercise Induce Asthma - Diagnosis

- Free running more effective than cycling or running on a treadmill.
- For standarized provocations, running on a motor-driven treadmill is employed.
- A standarized protocol*:
  - Increase the speed on a treadmill with 5% incline over 6-8 min. period to achieve a constant heart rate of ~95% of maximum (calculated by subtracting the age from 220). Room temperature ~20°C and relative humidity ~40%. Or dry air < 5mgH2O/L (air bottle).
  - If negative EUCAPNIC HYPERVERVENTILATION TEST IS RECOMMENDED, SPECIALLY IN ATHLETES (IS MORE SENSITIVE THAN METACHOLINE TEST).

(*>35 y.o. Rule out cardiac diseases)
EIA - Diagnosis

- Lung function (PEF or FEV\textsubscript{1}) is measured before running, immediately after running and 3, 6, 10, 15 and 20 min post exercise.
- 10-15\% fall in PEF or FEV\textsubscript{1} from baseline is diagnostic.
- Exercise-induced laryngeal stridor can occur (usually in female athletes) and is reflected by flattening of maximal inspiratory flow-volume curve, the presence of stridor and symptoms during exercise. In contrast, EIA usually occurs after exercise and is expiratory.
**Procedure for inhaled mannitol challenge**

<table>
<thead>
<tr>
<th>Dose #</th>
<th>Dose mg</th>
<th>Cum Dose mg</th>
<th>Capsules/dose</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>2</td>
<td>5</td>
<td>5</td>
<td>1</td>
</tr>
<tr>
<td>3</td>
<td>10</td>
<td>15</td>
<td>1</td>
</tr>
<tr>
<td>4</td>
<td>20</td>
<td>35</td>
<td>1</td>
</tr>
<tr>
<td>5</td>
<td>40</td>
<td>75</td>
<td>1</td>
</tr>
<tr>
<td>6</td>
<td>80</td>
<td>155</td>
<td>2 x 40</td>
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<tr>
<td>7</td>
<td>160</td>
<td>315</td>
<td>4 x 40</td>
</tr>
<tr>
<td>8</td>
<td>160</td>
<td>475</td>
<td>4 x 40</td>
</tr>
<tr>
<td>9</td>
<td>160</td>
<td>635</td>
<td>4 x 40</td>
</tr>
</tbody>
</table>

**FEV$_1$ fall > 15%**
Airway response to mannitol in asthmatic & non-asthmatic subjects

Anderson SD et al AJRCCM 1997 156:758
Comparison of mannitol and methacholine to predict exercise-induced bronchoconstriction and a clinical diagnosis of asthma


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<table>
<thead>
<tr>
<th>Age</th>
<th>Intent-to-Treat</th>
<th>Excluded and Safety</th>
<th>Per-Protocol</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Number</td>
<td>Percent</td>
<td>Number</td>
</tr>
<tr>
<td>Total</td>
<td>391</td>
<td>1.5%</td>
<td>509</td>
</tr>
<tr>
<td>6-7</td>
<td>6</td>
<td>1.5%</td>
<td>9</td>
</tr>
<tr>
<td>8-9</td>
<td>7</td>
<td>1.8%</td>
<td>11</td>
</tr>
<tr>
<td>10-11</td>
<td>20</td>
<td>5.1%</td>
<td>25</td>
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<tr>
<td>12-15</td>
<td>38</td>
<td>9.7%</td>
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<tr>
<td>16-18</td>
<td>44</td>
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<tr>
<td>19-24</td>
<td>113</td>
<td>28.9%</td>
<td>135</td>
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<tr>
<td>25-30</td>
<td>69</td>
<td>17.6%</td>
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<tr>
<td>31-35</td>
<td>28</td>
<td>7.2%</td>
<td>44</td>
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<tr>
<td>36-40</td>
<td>31</td>
<td>7.9%</td>
<td>44</td>
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<tr>
<td>41-45</td>
<td>19</td>
<td>4.9%</td>
<td>25</td>
</tr>
<tr>
<td>46-50</td>
<td>16</td>
<td>4.1%</td>
<td>27</td>
</tr>
</tbody>
</table>
The scatter plot shows the relationship between the percentage fall in mannitol (% Fall Mannitol) and the percentage fall in methacholine (% Fall Methacholine). The data points are color-coded to indicate two categories:

- Blue dots represent Fall < 10%.
- Pink triangles represent Fall ≥ 10%.

The correlation coefficient (r) is 0.41, with a p-value < 0.0001, indicating a statistically significant positive correlation between the two variables.
Sensitivity and specificity of challenge at different cut points for a positive test.

<table>
<thead>
<tr>
<th>Exercise Positive Cut-Points - % fall from baseline</th>
<th>10%</th>
<th>15%</th>
<th>20%</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Mannitol</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sensitivity</td>
<td>58.6</td>
<td>69.4</td>
<td>78.6</td>
</tr>
<tr>
<td>Specificity</td>
<td>65.2</td>
<td>62.0</td>
<td>60.8</td>
</tr>
<tr>
<td>n = 372</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Excluding those with challenge &gt;35 min</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sensitivity</td>
<td>64.1</td>
<td>75.3</td>
<td>82.7</td>
</tr>
<tr>
<td>Specificity</td>
<td>59.9</td>
<td>57.0</td>
<td>55.4</td>
</tr>
<tr>
<td>n = 319</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Methacholine 16 mg/ml</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sensitivity</td>
<td>55.2</td>
<td>67.4</td>
<td>80.3</td>
</tr>
<tr>
<td>Specificity</td>
<td>68.9</td>
<td>66.1</td>
<td>65.2</td>
</tr>
<tr>
<td>n = 375</td>
<td></td>
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</table>
Methacholine is more sensitive than mannitol for evaluation of bronchial hyperresponsiveness in children with asthma

JACI, 2010

Our results confirm that bronchoprovocation with a directly acting stimulus such as methacholine is extremely sensitive, demonstrating BHR as a result of the presence of current asthma symptoms more reliably than mannitol in this population of children with asthma. Therefore, methacholine is a poor predictor of hyperresponsiveness to mannitol. However, indirect stimulus may be a clinically relevant marker for assessing the clinical course of asthma, leading us to conclude that the 2 tests are complementary but not interchangeable in clinical practice.

We thank Oliver Shaw for editorial assistance.

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Methacholine is more sensitive than mannitol for evaluation of bronchial hyper-responsiveness in youth athletes with exercise-induced bronchoconstriction.

Eucapnic Voluntary Hyperventilation (VEH)

• Voluntary hyperventilation for 6 min dry air (from a bottle) that contains 5% CO₂, 21% O₂ and rest of N₂. The gas is inhaled from a bag (as reservoir with at least 90 L) with an unidirectional valve. Volume per minute is measured by a rotameter. The volume inhaled must be 30 times the FEV₁ in 6 min.
Eucapnic Voluntary Hyperventilation

- Lung function (FEV$_1$) is measured before and immediately after the test and 3, 6, 10, 15 and 20 min post exercise.

- 10% fall in FEV$_1$ from baseline is considered as positive

Non-specific bronchial hyperresponsiveness in asthma

- Are all tests equivalent?: NO
- Are all tests measuring same parameters: NO
- Indirect tests are more related to bronchial inflammation: YES
- Which has more sensitivity: DRY AIR
- Recommended in clinical practice: METHACHOLINE IN TIDAL VOLUME
- In case of Methacholine (-) and clinical symptoms: AMP or DRY AIR (always in athletes)