Bronchial responsiveness to histamine and methacholine measured with forced expirations and with the forced oscillation technique


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The objective of this study was to compare bronchial challenge tests with two substances [histamine (H) and methacholine (M)] and two methods of measuring the effect parameter FEV₁ and pulmonary impedance [with the forced oscillation technique (FOT)] in order to determine which test is the shortest, and gives the least (drug) load to the patient. Furthermore, it was considered whether the result of one type of challenge test could be transferred to the result of another type of test. It was hypothesized that, since the FOT technique requires no forced manoeuvres of the subjects and therefore does not affect the airway patency, there must be differences in the provocation concentrations for reaching the conventional thresholds of 20% decrease in FEV₁ (PC₂₀ FEV₁) and 40% increase in airway resistance measured at 8 Hz oscillation frequency (PC₄₀ Rrs₈). It was further hypothesized that the interindividual correlations between thresholds for both drugs will be low, because both drugs set off different mechanisms for bronchoconstriction. Bronchial challenge tests were performed in 23 stable asthmatics (15 males and 8 females; mean ± sd age 30.3 ± 11.6 years). Their mean control FEV₁ was 85.2 ± 12.6% predicted. For both drugs, PC₄₀ Rrs₈ was three-fold lower than PC₂₀ FEV₁.

The within-drug correlation between log PC₂₀ FEV₁ (H,M) and log PC₄₀ Rrs₈ (H,M) was quite good [r(H) = 0.73, r(M) = 0.68]. The between-drug correlation of log PC₂₀ FEV₁ (H) and log PC₂₀ FEV₁ (M) was equally good. However, the ‘between-drug’ correlation of log PC₄₀ Rrs₈ (H) and log PC₄₀ Rrs₈ (M) was low (r = 0.36).

It is concluded that the PC₄₀ Rrs₈ for histamine is the shortest test for bronchial responsiveness, with the lowest drug load for the patient. The results from one type of challenge test cannot be recalculated into the result of another type of test.

Introduction

Bronchial responsiveness can be measured by means of the bronchial challenge test to inhaled histamine (H) or methacholine (M). These tests are usually quantified by indices obtained from forced expirations. They require full co-operation of the patient. The deep inspirations necessary for these forced expiratory manoeuvres may influence the bronchial tone (1). A bronchial provocation test for measuring responsiveness is time-consuming, and may be burdensome for the patient.

Induced bronchoconstriction can also be assessed by measuring the respiratory impedance (Rrs) with the pseudo random noise forced oscillation technique (FOT) (2-5). The method is simple, requires only passive co-operation and no forced expiratory manoeuvres. Forced oscillation parameters appear to be sensitive indicators of airway calibre, especially the resistance at lower frequencies such as 6 Hz and 8 Hz (Rrs₆ and Rrs₈). A 40% increase in resistance (PC₄₀ Rrs) is conventionally accepted to be diagnostic for a positive provocation test (2-4).

The aim of this study was, to find answers to the following questions:

(1) what method of detection of bronchial responsiveness gives the lowest burden to the patient, consequently using the lowest concentrations of both challenging agents?

(2) what is the correlation between H and M hyper-responsiveness, using the pulmonary impedance and forced expirations? Can the outcome of one be recalculated into the result of another test?
Methods

PATIENTS

Twenty-three patients with asthma [according to the ATS criteria (6)], eight females and 15 males; mean age 30.3 years ± 11.6 with a previously determined bronchial responsiveness (PC_{20} \text{FEV}_1 \leq 8 \text{ mg ml}^{-1}) volunteered for the study. The (geometric) mean PC_{20}H was 1.9 ± 2.2 mg ml^{-1}. The mean pre-challenge FEV_1 was 85.2 ± 12.6% predicted and the mean maximum expiratory flow measured from maximum flow – volume curve when 50% of FVC remains to be exhaled (MEF_{50}) was 64.4 ± 22.0% predicted (7). The mean reversibility of FEV_1 after inhalation of salbutamol (400 μg) was 10 ± 11.2% of the prebronchodilator value. At the time of the study, all patients were in a stable clinical condition and had abstained from β-sympathomimetics and anti-cholinergics for at least 8 h. Inhaled corticosteroids were continued and no patients used theophyllines or oral corticosteroids. All patients were non-smokers. Patients with recent (<6 weeks) exacerbations or respiratory infections were excluded from the study. All patients had normal values of oscillatory resistance (Rrs) and reactance (Xrs), at the start of the investigation (8).

EQUIPMENT

Histamine solutions were made from histamine di-phosphate powder, in phosphate-buffered saline, according to prescriptions of the European Respiratory Society (7). Methacholine chloride was dissolved in saline. The FEV_1 and MEF_{50} were obtained by flow–volume measurements (Discom, Chest C, Tokyo, Japan), and the integrated pneumotachograph-signal was calibrated with a 3-litre syringe, at three levels of flow. Oscillatory resistance and Xrs were determined by means of a pseudorandom forced oscillation technique (9). Briefly, a pseudorandom noise signal, containing all harmonics of 4 Hz–52 Hz (4, 8, 12, . . . 52 Hz) was applied for 8 s at the mouth of the seated, quietly breathing subject, who firmly supported the cheeks and submental regions with the hands. The impedance of the respiratory system, obtained from pressures and flows measured at the mouth for all investigated oscillatory frequencies, is divided into real (resistance, Rrs) and imaginary (reactance, Xrs) sections. Only values of Rrs and Xrs with a coherence function equal to or exceeding 0.95 were retained (9). Since many measurements at 6 Hz were rejected by this procedure, the MEF_{40} Rrs_{80} were used as the threshold parameter. The FOT-apparatus was calibrated daily with a fixed external resistance of 0.2 kPa 1^{-1} s^{-1}.

STUDY DESIGN

Studies were performed on each subject in a randomized, single-blind, cross-over manner on two consecutive days, at the same time of day. On the first day, the mean baseline values of Rrs and Xrs were measured by averaging three consecutive measurements. The best of three consecutive flow–volume curves (greatest sum of FVC and FEV_1) was used.

After a period of 5 min, the bronchial challenge test was carried out. Doubling concentrations of histamine phosphate or methacholine chloride from 0.03–8.0 mg ml^{-1} were inhaled for 2 min during tidal breathing from a DeVilbiss 646 nebulizer operated by oxygen. The nebulizer was previously calibrated to give an output of 0.13 ml min^{-1} (9). Thirty seconds after each nebulization, Rrs and Xrs were measured. Ninety seconds after the nebulization, FEV_1 was measured. The inhalation of a new concentration was started 5 min after the start of the previous inhalation. The test was stopped when FEV_1 had dropped by at least 20%, or when a concentration of 8 mg ml^{-1} H or M was reached.

DATA ANALYSIS

The characteristics of the subjects such as the mean FEV_1 and MEF_{50} were taken from the best baseline values of the 2 days. The provocations concentration causing a 20% fall of FEV_1 (PC_{20} \text{FEV}_1) and provocation concentration causing a 40% increase at the Rrs at 8 Hz (PC_{40} \text{Rrs}_{80}) were used as indices (4,6) for bronchial responsiveness. If a threshold value was reached after inhalation of saline, the arbitrary PC value of 0.01 was used, in order to enable logarithmic transformations. If the threshold was not reached at 8 mg ml^{-1}, then the value of 16 was attributed. Each of these extremes occurred only once.

The correlation coefficients between the log PC_{20} \text{FEV}_1 and the log PC_{40} \text{Rrs}_{80} values for H and for M were calculated (Pearson’s correlation). The increase of resistance and the decrease of reactance at the frequencies of 4–52 Hz were determined when the PC_{20} \text{FEV}_1 and the PC_{40} \text{Rrs}_{80} were reached. Changes in the various parameters were considered to be significant at a P-level of 0.05 (paired t-test). Mean PC values were calculated as geometric means; t-tests were performed on log PC values. The study was approved by the Hospital Ethical Committee.
Bronchial responsiveness with FEV₁ and respiratory impedance

Results

All patients who entered the study, reached completion, no patients dropped out. The mean baseline lung function values were the same on both days, when measured before the H and M challenge tests — FEV₁ (H), 78.7 ± 10.8% predicted; FEV₁ (M), 84.8 ± 12.6% predicted. The baseline values of Rs and Xrs, as well as the values at PC20 FEV₁ and at PC40 Rs8, are shown in Figs 1 and 2 for H and M respectively. The threshold values for the individual subjects are given in Table 1. A pattern characteristic for obstructive lung disease developed during the challenges (9,11).

The geometric mean value of the provocation concentrations for PC20 FEV₁ (H) was 0.78 mg ml⁻¹ (95% CI, 2.1-10.37), PC20 FEV₁ (M) was 1.67 mg ml⁻¹ (95% CI, 3.02-0.93), PC40 Rs8 (H) was 0.20 mg ml⁻¹ (95% CI, 0.43-0.09), and PC40 Rs8 (M) was 0.53 mg ml⁻¹ (95% CI, 1.09-0.26). Both values for PC40 Rs8 were significantly lower than those for PC20 FEV₁ (P<0.01). In all patients, the PC40 Rs8 (M,H) was lower or equal to the PC20 FEV₁ (M,H).

The indices, PC20 FEV₁ and PC40 Rs8, which are commonly accepted to measure bronchial responsiveness, correlated partially between both drugs (Table 2, Fig. 3). The dose-response curve for H and M is shown in Fig. 4 for both indices. The dose-response curve of Rrs8 values was steeper than the decrease of FEV₁ for both drugs.

The correlation between log PC40 Rs8 (H) and log PC40 Rs8 (M) was low (r=0.36, P=0.09) in contrast with the good correlation between log PC20 FEV₁ (H) and log PC20 FEV₁ (M) (r=0.72, P<0.001). The difference between log PC40 Rs8 values for H and M was significant (P=0.03).

Discussion

Our study shows that the FOT gives lower threshold values for determining bronchial responsiveness, than measurements of FEV₁. If one uses PC20 FEV₁ and PC40 Rs8 as accepted thresholds, then bronchial hyper-reactivity is detected at threefold lower concentrations of H and M by measuring respiratory impedance, as compared with forced expiratory manoeuvres. The dose-response curve
(Fig. 4) did show a shift to the left and an increase in slope, for Rrs when compared to FEV₁.

Both parameters Rrs₈ and FEV₁ were measured in a fixed sequence, within the timespan of 1 min. Thus FEV₁ was systematically measured at a later time than Rrs₈. The effect of the challenge drug could have worn off. This may have contributed to the higher value of PC₂₀ FEV₁ as compared to Rrs₈. However, a cumulative effect has been described for M challenge with this protocol, where subsequent doses were given every 5 min (12). Therefore, it seems unlikely that the higher value of PC₂₀ FEV₁, as compared with PC₄₀ Rrs₈ (M), is due to this fixed sequence protocol.

The PC₂₀ FEV₁ for H and M was shown to be highly reproducible (coefficient of determination r²=0.994 and r²=0.990 respectively) (12). The reproducibility of log PC₄₀ Rrs₈ proved to be good. The standard deviation of the reproducibility of log PC₄₀ Rrs₈ to either H and M with a 24 h interval about 0.3 mg ml⁻¹ (2).

The baseline value for Rrs is somewhat high, as compared with normal values of airways resistance from body-plethysmograph measurements, but they were still in the range of the values for normal subjects as described by Låndser et al. (8). Furthermore, the somewhat low FEV₁ value (85.2% predicted) may have contributed to this baseline Rrs value.

The parameter Rrs₈ was sensitive to assess induced bronchoconstriction for both H and M. Similar results were obtained with Rrs₈ in cold air

Table 2 Relationships between bronchial responsiveness parameters

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<tr>
<th>Relationship</th>
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<tr>
<td>log PC₂₀ FEV₁ (M) = 0.5789 × log PC₂₀ FEV₁ (H) + 0.2875</td>
<td>0.73</td>
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<tr>
<td>log PC₄₀ Rrs₈ (H) = 0.7350 × log PC₂₀ FEV₁ (H) − 0.6177</td>
<td>0.71</td>
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<tr>
<td>log PC₄₀ Rrs₈ (M) = 0.4249 × log PC₂₀ FEV₁ (H) − 0.2311</td>
<td>0.39</td>
</tr>
<tr>
<td>log PC₄₀ Rrs₈ (M) = 0.8349 × log PC₂₀ FEV₁ (M) − 0.4648</td>
<td>0.66</td>
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PC₂₀ FEV₁ (mg ml⁻¹); PC₄₀ Rrs₈ (mg ml⁻¹); H, histamine; M, methacholine.
provocation tests (19). The reproducibility of $R_{rsg}$ is good with a coefficient of variation of 9-7% (3). The adequate use of this parameter in bronchial challenge testing was also shown by van Noord (4), Wouters (13) and v.d. Elshout (14).

A hypothesis that the similar bronchoconstrictive effects from both agents lies at the level of smooth muscle cannot be confirmed by our measurements with the FOT (15,16). The difference of effects measured from pulmonary impedance for H and M may be explained by the complexity of the bronchial response to H (17). The response to a challenge with H is mediated by a contraction of the smooth muscle, but vasodilatation, increased permeability of bronchial venules, local oedema and an increase of secretion of bronchial glands (18) also play a role. Methacholine primarily causes a contraction of the smooth muscle (18). This is also in keeping with the results of Duiverman et al. (2,3) and van Noord et al. (4). There was a low correlation between H and M thresholds with the measurement of pulmonary impedance, in contrast to the ones measured with the forced expiratory manoeuvres. These results may suggest that the different mechanisms underlying the increased responsiveness to H and M can probably be detected by measuring airway impedance. The differences were not caused by more coughing during the H challenge.

When pulmonary impedance alone is measured, it is not possible to detect lung volume changes (hyperinflation), which occur during bronchal provocation, and to correct for their effects. This may slightly reduce the sensitivity of the pulmonary impedance measurements (4). On the other hand, the maximal inspirations before every forced expiratory manoeuvre also lower the airway resistance (1). One may presume that the effects of maximal inspirations on lowering airway resistance are much larger than the increase in FRC during tidal breathing. The results of this study support this assumption.

It is concluded that the $PC_{40} R_{rs_g}$ is a useful index for bronchial responsiveness. The method requires only passive co-operation of the patient, and does not necessitate forced manoeuvres which may influence bronchial tone. The $PC_{40} R_{rs_g}$ for both H and M was reached at three-fold lower concentrations than $PC_{20} FEV_1$. This will shorten challenge tests in terms of duration by $3 \times 5 = 15$ min, but more importantly, also in terms of drug loads, with higher concentrations of bronchoconstrictive agents. Provocation concentration values for H and M, measured with forced manoeuvres or pulmonary impedance, are not strongly correlated.
References


