Impulse Oscillometry for Leukotriene D_4 Inhalation Challenge in Asthma

Wei Jie Guan PhD, Jin Ping Zheng MD, Yi Gao MMed, Cai Yu Jiang, Xu Shi PhD, Yan Qing Xie PhD, Qing Xia Liu MMed, Mei Jiang PhD, Jia Ying An MT, Xin Xin Yu MT, Wen Ting Liu MT, Li Ping Zhong MT, Zhong Ping Wu MT, and Nan Shan Zhong MD

BACKGROUND: The value of impulse oscillometry (IOS) for bronchial provocation testing is poorly defined. We investigated the positive threshold derived from the parameters and diagnostic power of IOS for asthma with the leukotriene D_4 bronchial provocation test. METHODS: We enrolled 62 subjects with asthma and 21 healthy subjects. IOS was employed to perform the leukotriene D_4 bronchial provocation test, followed by spirometry. The positive threshold was determined based on the cutoff point in the receiver operating characteristic curve, from which the parameters with the highest diagnostic power were obtained. RESULTS: Airway impedance at 5 Hz (Z_5), resistance at 5 Hz (R_5), and resonance frequency had the highest diagnostic power (areas under curve 0.82, 0.82, and 0.81, respectively), with increases of 57%, 43%, and 63%, corresponding to a 20% decrease in FEV_1, respectively. IOS indices yielded assay sensitivity and specificity similar to that of spirometry. The positive threshold for IOS, defined as either a 57% increase in Z_5 or a 63% increase in resonance frequency in the bronchial provocation test, yielded an assay accuracy of 0.6 in subjects with asthma. CONCLUSIONS: IOS during the leukotriene D_4 bronchial provocation test has a diagnostic power similar to that of spirometry. Either a 57% increase in Z_5 or a 63% increase in resonance frequency may be regarded as a surrogate of FEV_1 decrease to determine airway hyper-responsiveness in asthma. *Key words: impulse oscillometry; airway resistance; resonance frequency; leukotriene D_4; bronchial provocation test; asthma.* [Respir Care 2013; 58(12):2120–2126. © 2013 Daedalus Enterprises]

Introduction

Cysteinyl leukotrienes are potent inflammatory mediators that elicit bronchoconstriction, leading to air-flow limitation,[1-3] which could be blocked by leukotriene receptor antagonists.[4] Unfortunately, a simple and feasible measure for predicting the efficacy in asthma is lacking. Bronchial provocation test has been extensively applied for measurement of airway responsiveness.[5] Conventional stimuli (ie, methacholine) could not specifically discriminate the ef-

The authors are affiliated with the State Key Laboratory of Respiratory Disease, China Clinical Research Center for Respiratory Disease, First Affiliated Hospital of Guangzhou Medical University, Guangzhou, Guangdong, China, with the exception of Cai Yu Jiang, who is affiliated with Sichuan People's Hospital, Chengdu, Sichuan, China.

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Correspondence: Jin Ping Zheng MD, State Key Laboratory of Respiratory Disease, First Affiliated Hospital of Guangzhou Medical University, 151 Yanjiang Road, Guangzhou, Guangdong, China, 510120. E-mail: jpzhenggy@163.com.

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ficacy of leukotriene receptor antagonist, which might be assessed, at least theoretically, by using leukotriene D_4 for the bronchial provocation test.[6-15]

Although spirometry has been the recommended method for assessing bronchial provocation, repetitive forced inhalation might result in bronchodilation, leading to false negative results. Impulse oscillometry (IOS), a novel lung function testing method characterized by shorter test duration,[16,17] minimal requirements to subjects, and low likelihood of bronchodilation, does not influence airway smooth muscle tone. Theoretically, IOS parameters correlate negatively with airway caliber, as evidenced by the fact that bronchospasm significantly increases airway impedance, resistance, and resonance frequency, and reduces airway capacitance,[18] which suggests that IOS may be a surrogate for spirometry. However, the lack of gold standard test has rendered it difficult to be clinically applied. We investigated the discriminative and significant thresholds of IOS and its diagnostic potential in the detection of asthma with the leukotriene D_4 bronchial provocation test.

QUICK LOOK

Current knowledge

The diagnostic power of impulse oscillometry for bronchial provocation tests, compared with spirometry, remains elusive in asthma. There is a need to define the positive threshold of impulse oscillometry parameters with high diagnostic power.

What this paper contributes to our knowledge

Impulse oscillometry showed acceptable diagnostic power, compared with spirometry, for bronchial provocation testing. A 57% increase in airway impedance at 5 Hz or a 63% increase in resonance frequency may be regarded as a surrogate of an FEV_1 decrease to determine airway hyperresponsiveness in asthma.

Methods

The study protocol was approved by the ethics committee of First Affiliated Hospital of Guangzhou Medical University. All subjects gave written informed consent prior to participation in the study.

Subjects

Asthma was diagnosed based on the Global Initiative for Asthma 2006 guideline.[19] Consecutive patients with asthma who visited the clinics at First Affiliated Hospital of Guangzhou Medical University between March and August 2010 underwent screening. The inclusion criteria were age 18–65 years, no respiratory-tract infection in the past 2 weeks, normal chest radiography, and baseline $FEV_1 > 60\%$ of predicted. We excluded patients whose FEV_1 decreased $\geq 20\%$ after ethanol dilution challenge, who had other confirmed lower airway or severe systemic diseases, and who had poor adherence.

We asked the subjects not to use leukotriene receptor antagonist for 5 days, oral corticosteroid or antihistamine for 3 days, oral xanthenes or long-acting bronchodilators for 2 days, inhaled corticosteroid or long-acting bronchodilator for 24 hours, and short-acting bronchodilator for 4 hours prior to the measurements.

The healthy subjects were 18–65 years old, and had no upper-respiratory-tract infection for the previous 2 weeks, no allergic or systemic diseases, normal chest radiograph, and regular spirometry values.

Study Protocol

IOS was performed prior to spirometry. Airway responsiveness was assessed as the cumulative dose of leukotriene D_4 that caused a 20% decrease in FEV_1, and a positive response was defined as ≤ 4.800 nmol of leukotriene D_4. The diagnostic power of IOS parameters was evaluated with a receiver operating characteristic curve, from which we obtained the cutoff points for the sensitivities and specificities of the IOS parameters for asthma diagnosis.

Changes in IOS parameters corresponding to a 20% decrease in FEV_1 were calculated with the following formula:

$$R = \frac{(\ln D - \ln D_1)(R_2 - R_1)}{\ln D - \ln D_1} + R_1$$

where D_1 (or D_2) is the cumulative dose that causes an $R_1\%$ (or $R_2\%$) increase in IOS parameter, R_1 (or R_2) is the variation (%) corresponding to the cumulative dose of X_1 (or X_2), and D is the cumulative dose that causes R% increase in IOS parameter.

Impulse Oscillometry

IOS (MS-IOS, Erich Jaeger, Friedberg, Germany) was conducted 5 min following leukotriene D_4 inhalation challenge, immediately prior to spirometry, based on the methods previously described.[20] The subject was seated upright with head straight and in slight extension. A nose clip was applied and the lips were tightly sealed around the mouthpiece to avoid gas leakage, and the subject firmly supported his or her cheeks with the palms of both hands. The

Table 1. Protocol of Leukotriene D_4 Inhalation Challenge Test

Step	Concentration (nmol/L × 10^4*)	Inhalation Time (s)	Inhalation Duration (s)	Dose (nmol)	Cumulative Dose (nmol)
1	1.6	1	0.315	0.075	0.075
2	1.6	3	0.315	0.225	0.300
3	3.2	5	0.375	0.900	1.200
4	3.2	10	0.750	3.600	4.800

* 1 nmol leukotriene D_4 = 0.5 μg leukotriene D_4.

IOS maneuver lasted for 40–60 seconds. The measured IOS parameters were respiratory impedance at 5 Hz (Z_5), airway resistance at 5 Hz (R_5), central airway resistance (R_{20}), lung resistance at 5 Hz (X_5), and resonance frequency.

Spirometry

Spirometry (Quark PFT, Cosmed, Rome, Italy) was performed per the joint recommendations of the American Thoracic Society and European Respiratory Society.[21] At least 3 (not more than 8) maneuvers were performed, with the variation between the best 2 maneuvers of < 5% or 150 mL in FVC and FEV_1. The maximum FVC, FEV_1, and PEF values are reported. The predicted values were calculated based on the normative values recommended by Zheng and Zhong.[22]

Inhalation Challenge of Leukotriene D_4

The method for the leukotriene D_4 bronchial provocation test has been described previously.[23,24] Briefly, dilutions were delivered from a nebulizer (646, DeVilbiss Healthcare, Somerset, Pennsylvania) powered by compressed oxygen at 37.0 psi at 900 μL/min. Inhalation challenge using 16% ethanol dilution was performed for excluding the subjects hypersensitive to ethanol. Leukotriene D_4 challenge was started when FEV_1 decrease was < 20% and was restored to < 10% within 1 min. The initial and final cumulative dose of leukotriene D_4 was 0.075 nmol and 4.800 nmol, respectively, based on the 4-fold incremental steps.[23,24] Repetitive inhalation challenges were performed at an interval of 5 min (Table 1). These procedures were terminated in case of ≥ 20% FEV_1 decrease.

Statistical Analysis

Statistical analysis was performed with statistics software (SPSS 16.0, SPSS, Chicago, Illinois). Data are expressed as mean ± SD for normal distribution data, and as median (IQR) for non-normal distribution data. Two-sided independent *t* tests for comparison on baseline levels were performed when normal distribution was detected; otherwise the 2-sided Mann-Whitney U test was employed. The diagnostic power of IOS parameters was assessed with the receiver operating characteristic curve, from which the area under the curve and 95% CI were derived. To study the diagnostic power of the combination of IOS parameters we calculated the assay sensitivity and specificity. A positive response was defined if either IOS parameter reached the positive threshold. $P < .05$ was considered as statistical significance.

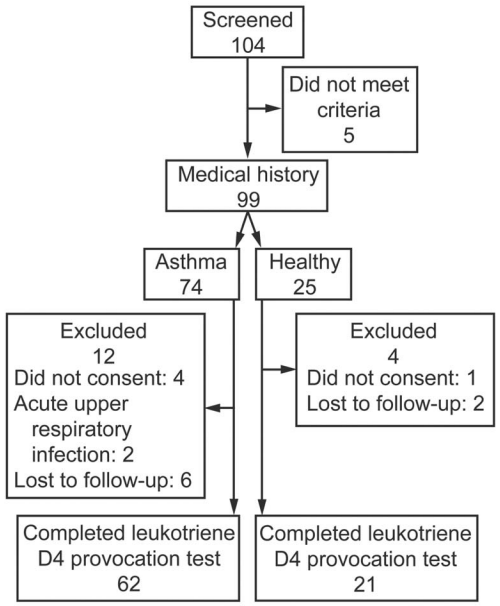

Fig. 1. Subject enrollment.

Results

The recruitment of subjects is shown in Figure 1. Of 104 subjects who underwent screening, 74 with physician-diagnosed asthma and 25 healthy subjects were enrolled, and 62 subjects with asthma and 21 healthy subjects completed the assessment.

The anthropometric parameters, FVC (3.52 ± 0.81 L vs 3.61 ± 0.76 L, $P = .53$), and percent-of-predicted FVC (FVC%) (110.33 ± 15.71% vs 107.03 ± 12.97%, $P = .43$) did not differ significantly between subjects with asthma and normal subjects. The normal subjects had significantly higher FEV_1 (3.03 ± 0.61 L vs 2.53 ± 0.62 L, $P = .001$) and FEV_1% (107.30 ± 11.54% vs 94.56 ± 15.20%, $P = .001$) at baseline. Compared with the healthy subjects, the subjects with asthma had significantly higher IOS values: Z_5 4.54 (2.12) vs 3.10 ± 0.87 ($P < .001$), resonance frequency 10.61 (2.83) vs 8.63 ± 1.18 ($P = .01$),

Table 2. Changes in Major Impulse Oscillometry Parameters Prior to and Following Leukotriene D_4 Challenge

	Subjects With Asthma $n = 62$ (cm H_2O/L/s)				Healthy Subjects $n = 21$ (cm H_2O/L/s)			
	Pre	Post	Z	*P*	Pre	Post	Z	*P*
Z_5	4.69 ± 2.18	7.97 ± 6.02	−6.075	< .001	3.10 ± 0.87	3.62 ± 1.82	−3.441	.001
R_5	4.33 ± 2.34	8.30 ± 3.58	−6.712	< .001	2.95 ± 0.82	3.50 ± 1.81	−3.615	< .001
R_{20}	3.77 ± 1.18	5.08 ± 1.60	−6.213*	< .001	2.36 ± 1.10	3.25 ± 0.98	−3.424	.001
Resonance frequency	9.34 ± 2.83	22.49 ± 14.29	−6.811	< .001	8.63 ± 1.18	9.32 ± 5.41	−2.850	.004
X_5	−1.10 ± 0.74	−2.76 ± 3.25	−6.641	< .001	−0.84 ± 0.51	−1.10 ± 0.50	−1.929	.054

± Values are mean ± SD.
* Denotes t value.

Table 3. Change in Major Impulse Oscillometry Parameters Corresponding to a 20% Decrease in FEV_1

	Change	
	Mean ± SD	Range
Z_5	75.60 ± 46.23	−2.17 to 237.98
R_5	65.56 ± 37.28	−0.88 to 172.35
Resonance frequency	88.88 ± 58.19	5.73 to 226.00
	Median	(IQR)
R_{20}	26.83	(−17.59 to 168.56)
R_5–R_{20}	209.12	(−2,152.41 to 17,400.00)
X_5	−137.66	(32.64 to 1,008.16)

Table 4. Diagnostic Power of Major Impulse Oscillometry Parameters

	Area Under the Curve	Standard Error	*P*	95% CI
Z_5	0.821	0.052	< .001	0.720–0.923
R_5	0.806	0.056	< .001	0.696–0.915
R_{20}	0.595	0.067	.20	0.464–0.727
R_5–R_{20}	0.717	0.069	.003	0.581–0.852
X_5	0.764	0.066	< .001	0.635–0.893
Resonance frequency, %	0.815	0.054	< .001	0.709–0.922

R_5 4.40 (2.29) vs 2.95 ± 0.82 ($P < .001$), and R_{20} 2.58 (1.10) vs 3.77 ± 1.18 ($P < .001$).

A significant change in IOS parameters was revealed (all $P < .001$) following leukotriene D_4 bronchial provocation test, when compared with the baseline levels, particularly in subjects with asthma, except for X_5 in healthy subjects (−0.84 ± 0.51 vs −1.10 ± 0.50), $P = .054$) (Table 2). There were significant outlier percentages observed in the increase of (R_5–R_{20}) and reduction of X_5 (Table 3).

The overall diagnostic power of IOS parameters, as evidenced by the area under the curve of the receiver operating characteristic curve, was the highest for Z_5, followed by resonance frequency, R_5, X_5 (R_5–R_{20}), and R_{20}. Of all these parameters, Z_5, resonance frequency, and R_5 showed the highest and acceptable diagnostic power (area under the curve 0.82, 0.82, and 0.81, respectively), while X_5 and (R_5–R_{20}) were excluded because of significant outlier values (Fig. 2, Table 4).

Based on the cutoff points obtained from the receiver operating characteristic curve, the positive thresholds of a 57%, 45%, 62%, 63% and 43% increase in Z_5, R_{20} (R_5–R_{20}), resonance frequency and R_5, as well as a 77% reduction in X_5, respectively, were defined. The 3 most sensitive indices, Z_5, R_5, and resonance frequency, were associated with assay accuracies of 0.64, 0.61, and 0.57, respectively.

Overall, the IOS parameters yielded acceptable assay sensitivity and specificity, compared with FEV_1. When the combinations of Z_5, R_5, and resonance frequency were employed for analysis, the assay sensitivity was increased, when compared with either individual IOS parameter. The tests were regarded positive as soon as the first single parameter reached the critical threshold. The 57% increase in Z_5 or 63% increase in resonance frequency yielded an improved assay accuracy of 0.60 (sensitivity 0.74, specificity 0.86) (Table 5).

Discussion

This was the first study, to our knowledge, that has investigated the positive threshold of IOS parameters and compared these indices with FEV_1, a well established spirometric parameter, in subjects with asthma who underwent leukotriene D_4 inhalation challenge. Our data evidence that Z_5, R_5, and resonance frequency were characterized by the highest diagnostic power, with an increase of 57%, 43%, and 63% corresponding to a 20% decrease in FEV_1, respectively. IOS was associated with a similar diagnostic power, compared with spirometry. The positive

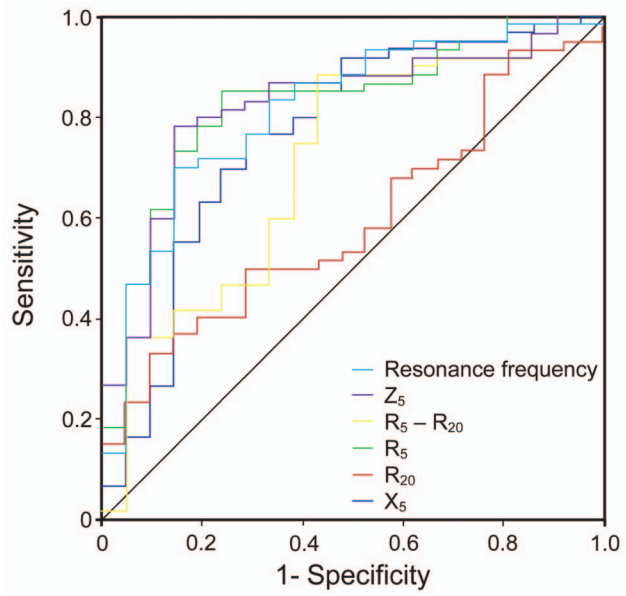

Fig. 2. Receiver operating characteristic curves for impulse oscillometry parameters. Z_5, R_5, R_{20}, and X_5 are defined in the text. Z_5, R_5, and resonance frequency had the highest diagnostic power, as reflected by the area under curve of the receiver operating characteristic curve.

response of leukotriene D_4 bronchial provocation test, defined as a 57% increase in Z_5 or 63% increase in resonance frequency, was associated with an assay accuracy of 0.6.

A reduced airway caliber in consequence of bronchospasm is linked to increase of airway resistance and decrease of capacitive lung resistance, which cannot be clinically measured by invasive approaches. Although spirometry has been well validated for bronchial provocation tests, IOS is expected to reflect changes in airway resistance, capacitive lung resistance, and inertance in a more spontaneous and physiologic manner. The test signals of IOS technique at low frequency (ie, 5 Hz) represent the total airway resistance. This is because the vibration energy may readily transmit throughout the respiratory tract. Contrarily, the energy at high frequency (ie, 20 Hz) represents central airway resistance, as the signal is shunted by the proximal airways. Furthermore, the difference between total and central airway resistance (R_5–R_{20}) reflects peripheral resistance, and the capacitive lung resistance at low frequency denotes the compliance of the lungs and chest wall. These suggest that IOS technique might be feasible for assessment of leukotriene D_4 bronchial provocation test.

The leukotriene D_4 inhalation challenge led to a significant change ($P < .01$) in IOS parameters. The evidence that a significant reaction was shown following inhalation challenge, except for R_{20} and airway capacitance (X_5), was consistent with our previous findings,[25] suggesting that stronger bronchoconstriction is associated with a higher airway resistance, as a consequence of reduced airway caliber. The FEV_1 decrease was postulated to vary approximately proportional to the increase in IOS parameters, except for X_5.

The positive threshold of IOS parameters among different studies remains largely inconclusive. Bohadana et al[16] reported a lower cumulative dose of carbachol and higher positive rate when using a 47% increase in mean resistance when the dose-response curve was employed. By using the average reaction in IOS parameters and linear regression model, Wang et al[25] reported that an 80% increase in resonance frequency, 50% increase in Z_5 or R_5, or 100% increase in X_5 could be regarded as significant. Pairon et al[26] reported an assay sensitivity of 0.75 and specificity of 0.76 for a 65% increase in total resistance for methacholine bronchial provocation test in active workers with normal baseline spirometry. They also showed that the positive threshold was 65% and 50% for elastance at

Table 5. Diagnostic Power of Major Impulse Oscillometry Parameters and FEV_1

	Sensitivity	Specificity	Youden Index	Positive Likelihood Ratio	Negative Likelihood Ratio
PD_{57} Z_5	0.6290	0.8571	0.4861	4.4017	0.4329
PD_{43} R_5	0.7258	0.7619	0.4877	3.0483	0.3599
PD_{27} R_{20}	0.4677	0.4762	−0.0561	0.8929	1.1178
PD_{62} (R_5–R_{20})	0.7581	0.7143	0.4724	2.6535	0.3387
PD_{63} resonance frequency	0.6129	0.8571	0.4700	4.2890	0.4516
PD_{77} X_5	0.7097	0.7143	0.4240	2.4841	0.4064
R_5 + Z_5	0.7419	0.7619	0.5038	3.1160	0.3388
R_5 + resonance frequency	0.7742	0.7619	0.5361	3.2516	0.2964
Z_5 + resonance frequency	0.7419	0.8574	0.5993	5.2027	0.3010
PD_{20} FEV_1	0.8710	0.9048	0.7758	9.1492	0.1426

PD = provocational dose that causes the subscript value percent change.

10 Hz and resonance frequency: results similar to our study. Snashall et al[27] documented that, when compared with FEV_1, Z_{10} yielded a 2.7-times increase in the cumulative dose that caused a 20% decrease in Z_{10} and the cumulative dose that caused a 20% decrease in FEV_1 might be regarded as equivalent in subjects who underwent histamine inhalation challenge. However, the airway capacitance was shown to have a greater diagnostic power in another study. By studying preschool children who underwent methacholine inhalation challenge, Jee et al[28] reported an assay sensitivity of 0.80 and a specificity of 0.83, corresponding to an 80% decrease in X_5, particularly when compared using the area under curve with R_5 (0.87 vs 0.75). These data at least suggest that the various thresholds could have stemmed from different populations, settings, and the stimuli for the bronchial provocation tests.

R_{20} was expected to reflect the changes in central airway resistance. It seemed reasonable that an insignificant increase in R_{20} was evidenced, despite the presence of bronchospasm. From the physiological point of view, X_5 has relevance with regard to the reaction of peripheral airways. However, owing to a marked variation and outlier values following inhalation challenge, the change in X_5 was not useful when the baseline level was very small, and could even be zero from the mathematical background. It would have been more reasonable to argue that X_5 was not feasible to define the positive response of bronchospasm and should be excluded for further analysis.

The overall diagnostic power of IOS parameters was highest for Z_5, followed by resonance frequency, R_5, X_5 (R_5–R_{20}), and R_{20}. Again, this has led us to conclude that neither the change in (R_5–R_{20}) nor in R_{20} was reliable. Z_5, resonance frequency, and R_5 were characterized by a similar diagnostic power (area under the curve 0.82, 0.82, and 0.81, respectively). An increase of 57%, 43%, and 63% in Z_5, R_5, and resonance frequency could be regarded as the positive threshold, based on the cutoff points derived from the receiver operating characteristic curve.

Two studies[29,30] have reported a higher assay sensitivity of IOS parameters, compared with spirometry, which is inconsistent with our findings. This could have stemmed from the difference in study design, leading to varying conclusions. Importantly, the positive response, defined as when any of the individual parameters (resonance frequency, Z_5, or R_5) reached the threshold, was characterized by a higher sensitivity. The sensitivity of 0.45 for resonance frequency, Z_5, and R_5 was documented[25] in a study that also showed a higher sensitivity and specificity (0.70 and 0.88, respectively) when either individual parameter reached the positive threshold. This warranted the combination of, but not individual, parameters for assessment. There were 2 major factors that could have contributed to the distinction among the literatures. First, since the inclusion criteria differed, the results might have been affected by the different proportion of subjects with asthma and healthy subjects, or those with different control status. Second, the various positive criteria of IOS parameter and the threshold have rendered it impossible to compare the data directly.

The limitations of IOS technique (ie, high inter-individual variability in X_5, lower repeatability than spirometry, and the potential impact of posture) have been undeniably shown. Although most of these could not be temporarily resolved, further studies that explore the changes in IOS parameters in subjects with asthma under different control status and the comparison with miscellaneous measures are necessary.

Conclusions

IOS is characterized by acceptable diagnostic power, compared with spirometry, for bronchial provocation tests, given the study design, in which spirometry was adopted as the gold standard for inhalation challenge test. Z_5, resonance frequency, and R_5 are the IOS parameters that have the highest diagnostic power. Either a 57% increase in Z_5 or a 63% increase in resonance frequency may be regarded as a surrogate of FEV_1 decrease to determine airway hyper-responsiveness in asthma.

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REFERENCES

1. Drazen JM. Leukotrienes as mediators of airway obstruction. Am J Respir Crit Care Med 1998;158(5 Pt 3):S193-S200.
2. O'Byrne PM. Leukotrienes in the pathogenesis of asthma. Chest 1997;111(2 Suppl):27S-34S.
3. Lee TH, Woszczek G, Farooque SP. Leukotriene E_4: perspective on the forgotten mediator. J Allergy Clin Immunol 2009;124(3): 417-421.
4. Salvi SS, Krishna MT, Sampson AP, Holgate ST. The Anti-inflammatory effects of leukotriene-modifying drugs and their use in asthma. Chest 2001;119(5):1533-1546.
5. Baur X. Bronchial challenge tests. Pneumologie 2011;65(6): 340-346.
6. Mulder A, Gauvreau GM, Watson RM, O'byrne PM. Effect of inhaled leukotriene D4 on airway eosinophilia and airway hyperresponsiveness in asthmatic subjects. Am J Respir Crit Care Med. 1999;159(5 Pt 1):1562-1567.
7. Wood-Baker R, Town GI, Benning B, Holgate ST. The reproducibility and effect on non-specific airway responsiveness of inhaled prostaglandin D_2 and leukotriene D_4 in asthmatic subjects. Br J Clin Pharmacol 1995;39(2):119-123.
8. Schelfhout V, Van De Velde V, Pauwels R, Joos G. The effect of the leukotriene receptor antagonist zafirlukast on neurokinin A-induced bronchoconstriction in patients with asthma: a comparison with leukotriene D_4 induced broncoconstriction. Pulm Pharmacol Ther 2008; 21(2):276-284.

9. Smith LJ, Hanby LA, Lavins BJ, Simonson SG. A single dose of zafirlukast reduces LTD_4-induced bronchoconstriction in patients on maintenance inhaled corticosteroid therapy. Ann Allergy Asthma Immunol 1998;81(1):43-49.
10. Christie PE, Tagari P, Ford-Hutchinson AW, Black C, Markendorf A, Schnitz-Schuamann M, Lee TH. Increased urinary LTE_4 excretion following inhalation of LTC_4 and LTE_4 in asthmatic subjects. Eur Respir J 1994;7(5):907-913.
11. Deykin A, Belostotsky O, Hong C, Massaro AF, Lilly CM, Israel E. Exhaled nitric oxide following leukotriene E_4 and methacholine inhalation in patients with asthma. Am J Respir Crit Care Med 2000; 162(5):1685-1689.
12. O'Shaughnessy TC, Georgiou P, Howland K, Dennis M, Compton CH, Barnes NC. Effect of pranlukast, an oral leukotriene receptor antagonist, on leukotriene D_4 (LTD_4) challenge in normal volunteers. Thorax 1997;52(6):519-522.
13. Casas A, Gómez FP, Dahlén B, Roca J, Barbera JA, Dahlén SE, Rodriguez-Roison R. Leukotriene D_4-induced hypoxaemia in asthma is mediated by the cys-leukotriene 1 receptor. Eur Respir J 2005; 26(3):442-448.
14. Gyllfors P, Dahlén SE, Kumlin M, Larsson K, Dahlén B. Bronchial responsiveness to leukotriene D_4 is resistant to inhaled fluticasone propionate. J Allergy Clin Immunol 2006;118(1):78-83.
15. Gyllfors P, Kumlin M, Dahlén SE, Gaber F, Ehrs PO, Dahlén B. Relation between bronchial responsiveness to inhaled leukotriene D_4 and markers of leukotriene biosynthesis. Thorax 2005; 60(11):902-908.
16. Bohadana AB, Peslin R, Megherbi SE, Teculescu D, Sauleau EA, Wild P, Pham QT. Dose-response slope of forced oscillation and forced expiratory parameters in bronchial challenge testing. Eur Respir J 1999;13(2):295-300.
17. Oostveen E, MacLeod D, Lorino H. The forced oscillation technique in clinical practice: methodology, recommendations and future developments. Eur Respir J 2003;22(6):1026-1041.
18. Kim HY, Shin YH, Jung DW. Resistance and reactance in oscillation lung function reflect basal lung function and bronchial hyperresponsiveness respectively. Respirology 2009;14(7):1035-1041.
19. O'Byrne P, Bateman ED, Bousquet JD, Clark T, Ohta K, Paggiaro P, et al. Global Initiative for Asthma (GINA). Global strategy for asthma management and prevention. Updated 2006. http://www.ginasthma.org/Guidelines/guidelines-resources.html. *Accessed September 20, 2013.*
20. Smith HJ, Reinhold P, Goldman. Forced oscillation technique and impulse oscillometry. Eur Respir J 2005;31(Suppl):72-105.
21. Miller MR, Hankinson J, Brusasco V, Burgos F, Casaburi R, Coates A, et al; ATS/ERS task force. Standardization of spirometry. Eur Respir J 2005;26(2):319-338.
22. Zheng JP, Zhong NS. Normative values of pulmonary function testing in Chinese adults. Chin Med J 2002;115(1):50-54.
23. Guan W, Zheng J, Gao Y, Jiang C, Xie Y, An J, et al. Leukotriene D_4 and methacholine bronchial provocation test for identifying leukotriene-responsiveness subtypes. J Allergy Clin Immunol 2013; 131(2):332-338.
24. Guan W, Zheng J, Gao Y, Jiang C, An J, Yu X, Liu W. Leukotriene D_4 bronchial provocation test: methodology and diagnostic value. Curr Med Res Opin 2012;28(5):797-803.
25. Wang HY, An JY, Luo DF, Zheng JP. Implication of impulse oscillometry in the measurement of bronchial hyperresponsiveness. Chin J Postgrad Med 2006;29(1):39-41.
26. Pairon JC, Iwatsubo Y, Hubert C, Lorino H, Nouaigui H, Gharbi R, Brochard P. Measurement of bronchial responsiveness by forced oscillation technique in occupational epidemiology. Eur Respir J 1994;7(3):484-489.
27. Snashall PD, Parker S, Ten Haave P, Simmons D, Noble MI. Use of an impedance meter for measuring airways responsiveness to histamine. Chest 1991;99(5):1183-1185.
28. Jee HM, Kwak JH, Jung W, Han MY. Useful parameters of bronchial hyperresponsiveness measured with an impulse oscillometry in preschool children. J Asthma 2010;47(3):227-32.
29. Almosawi AI, Keung EJ, Gauvreau GM, Killian KJ. assessing the sensitivity of IOS (Impulse Oscillation) compared to spirometry and plethysmography in detecting methacholine induced bronchoconstriction (abstract). Am J Respir Crit Care Med 2010;181: A4993.
30. Hira HS, Singh A. The site of airway obstruction among patients of bronchial asthma: role of impulse oscillometry (abstract). Chest 2008; 134:5004.