Responsiveness to leukotriene D₄ and methacholine for predicting efficacy of montelukast in asthma

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ABSTRACT
A lower responsiveness to leukotriene D₄ (LTD₄) or higher LTD₄/[methacholine (MCh)] potency ratio might suggest preferable outcomes of short-term montelukast monotherapy in terms of airway inflammation and lung function in asthmatic patients.

KEY WORDS
Asthma; leukotriene D₄ (LTD₄); leukotriene-responsive; leukotriene-unresponsive; methacholine (MCh); montelukast


Introduction
Leukotrienes are pivotal inflammatory mediators of asthma (1) and potently elicit bronchoconstriction by combination with CysLT₁, the major leukotriene receptor (2). Despite that leukotriene receptor antagonists (LTRA) have been clinically applied for years, a considerable number of asthmatic patients did not respond preferentially. Although increased in vivo leukotriene release (3) has been linked to preferable response to montelukast, this appeared clinically complicated. Urinary LTE₄/exhaled nitric oxide ratio (4) reflected the efficacy in childhood asthma yet remained elusive in adults. We hypothesized that airway responsiveness to leukotriene D₄ (LTD₄) [cumulative dose of leukotriene D₄ causing a 20% fall in FEV₁ (PD₂₀FEV₁-LTD₄)] and LTD₄/[methacholine (MCh)] potency ratio could predict the efficacy of LTRA, based on the available methodology of LTD₄ bronchial provocation test (5,6) and the difference in responsiveness to LTD₄ and MCh (6).

Methods
Recruitment ran from March 2010 to August 2010. The inclusion and exclusion criteria have been described previously (6). Asthmatic patients who tested positively to LTD₄ inhalation challenge were, in this open-label pilot trial, allocated to receive 28-day montelukast monotherapy (10 mg, once daily) followed by reassessment 3 to 5 days after montelukast withdrawal. Salbutamol was allowed as needed. Subjects were instructed to record the peak expiratory flow (PEF) thrice daily and as-needed use of salbutamol (puffs). Measurement of fractional exhaled nitric oxide (FENO) (7), airway responsiveness to LTD₄ and MCh, Asthma Quality of Life Questionnaire (AQLQ) score in the symptom dimension and asthma control test (ACT) score were performed prior to and after the treatment. Approval was obtained from Ethics Committee of First Affiliated Hospital of Guangzhou Medical University. All subjects gave informed consent prior to the study.

The methodology of LTD₄ and MCh inhalation challenge has been introduced previously (6). Both tests were performed at a 2- to 14-day interval. Measurement of FeNO was conducted by using NIOX MINO (Aerocrine Co, Sweden). All maneuvers met the guideline established by American Thoracic Society (7).

The geometric means of PD₂₀FEV₁-LTD₄ (0.533 nmol) and LTD₄/MCh potency ratio (3647), as determined by a previously conducted cross-sectional study, were adopted to identify leukotriene-responsive or leukotriene-unresponsive subjects (6). Subjects having a PD₂₀FEV₁-LTD₄ ≤0.533 nmol and LTD₄/MCh potency ratio ≥3,647 were deemed leukotriene-responsive, whilst those with PD₂₀FEV₁-LTD₄ >0.533 nmol and LTD₄/MCh potency ratio <3,647 were allocated to leukotriene-unresponsive group. The remaining were assigned to unclassified group. Data were expressed as mean ± standard deviation (χ±s) for normal distribution, while median (interquartile range) [M(QR)] was otherwise applied. Analysis of variance (ANOVA) was conducted for among-group comparison on data with normal distribution.

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distribution, whilst Kruskal-Wallis test was otherwise employed. Statistical analyses were performed using SPSS 16.0.

**Results**

Of 32 asthmatic patients allocated, 23 completed end-point reassessment. The 9 subjects dropped out owing to poor compliance (n=5, diary recording <50%), asthma exacerbation (n=3) and respiratory tract infection needing systemic therapy (n=1). All subgroups of subjects, mostly comprised of those with uncontrolled asthma, did not differ statistically (all P>0.05) in demography nor spirometry or FeNO (Table 1). Between-subgroup difference in pre- and post-treatment spirometry, quality of life, daily reliever use or airway hyperresponsiveness did not reach statistical significance (all P>0.05), except for that in PD$_{20}$FEV$_1$-LTD$_4$ and cumulative dose of methacholine causing a 20% reduction in FEV$_1$ (PD$_{20}$FEV$_1$-MCh) (both P<0.05). There was a trend towards a favorable improvement in FeNO, FEV$_1$, and AQLQ scores in those with PD$_{20}$FEV$_1$-LTD$_4$ <0.533 nmol or LTD$_4$/MCh potency >3,647 (Table 2). Further comparison was conducted in leukotriene-responsive/-unresponsive asthmatic patients. As compared with leukotriene-unresponsive group, a similar trend towards preferentially improved FeNO and FEV$_1$ in leukotriene-responsive group was noted though the difference did not reach statistical significance (Table 3).

**Discussion**

A 3- to 5-day withdrawal period following 28-day montelukast therapy was designed inasmuch that we aimed to determine the gross improvement in clinical indices and that all subjects might otherwise test negatively to LTD$_4$ challenge (unpublished data). This might, conceivably and inevitably, obscure the treatment effects of montelukast and lead to a relapse in some asthmatic patients. Interestingly, that subjects with PD$_{20}$FEV$_1$-LTD$_4$ <0.533 nmol or LTD$_4$/MCh potency ratio >3,647 showed significantly favorable improvement in FeNO and lung function indices seemed to suggest the roles that leukotrienes play in asthma. Our results were partly comparable to those of Athavale et al. (8), who reported markedly increased FEV$_1$, PEF and asthma symptom score and reduced salbutamol use in 148 clinically stable asthmatic patients receiving a 4-week open-labeled montelukast trial. The difference between these the two studies could have stemmed from the inclusion criteria, standards for allocation, concomitant use of medication and ethnicity.

Noticeably, several limitations must be addressed. Firstly, the subject enrollment might not be sufficient to warrant a clear-cut discrimination for between-group comparison, and hence, a sound conclusion. Secondly, the compelling results might be attributable to the lack of a placebo group. Thirdly, the comparatively short course of montelukast therapy appeared insufficient to clinical practice, particularly to those with uncontrolled asthma, who were expected to benefit more following a prolonged treatment. Finally, the different proportion of asthmatic patients with various levels of asthma control might have inevitably biased the results. However, subgroup analysis

**Table 1. Baseline levels.**

<table>
<thead>
<tr>
<th>Indices</th>
<th>Subgroup 1</th>
<th>PD$_{20}$FEV$_1$-LTD$_4$ $\leq$0.533 nmol</th>
<th>PD$_{20}$FEV$_1$-LTD$_4$ $&gt;$ 0.533 nmol</th>
<th>LTD$_4$/MCh potency ratio $&lt;$3,647</th>
<th>LTD$_4$/MCh potency ratio $\geq$3,647</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. of patients</td>
<td>18</td>
<td>5</td>
<td>13</td>
<td>10</td>
<td></td>
</tr>
<tr>
<td>Age (years)</td>
<td>38.11±11.71</td>
<td>40.00±8.12</td>
<td>39.69±11.92</td>
<td>37.00±10.53</td>
<td></td>
</tr>
<tr>
<td>Height (cm)</td>
<td>159.00 (11.50)</td>
<td>164.20±7.26</td>
<td>161.42±6.80</td>
<td>160.65±7.66</td>
<td></td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>61.33±12.25</td>
<td>66.80±8.58</td>
<td>61.23±11.92</td>
<td>64.20±11.59</td>
<td></td>
</tr>
<tr>
<td>Male/female ratio</td>
<td>8/10</td>
<td>3/2</td>
<td>6/7</td>
<td>5/5</td>
<td></td>
</tr>
<tr>
<td>Partly controlled (No.)</td>
<td>3</td>
<td>0</td>
<td>2</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>Uncontrolled (No.)</td>
<td>10</td>
<td>4</td>
<td>8</td>
<td>6</td>
<td></td>
</tr>
<tr>
<td>Pre-challenge FEV$_1$ (pred%)</td>
<td>91.22±14.60</td>
<td>101.12 (15.97)</td>
<td>95.65±15.99</td>
<td>88.17±10.53</td>
<td></td>
</tr>
<tr>
<td>FeNO (ppb)</td>
<td>45.50 (47.00)</td>
<td>29.40±13.83</td>
<td>40.00 (52.00)</td>
<td>56.70±40.70</td>
<td></td>
</tr>
<tr>
<td>PD$_{20}$FEV$_1$-LTD$_4$ (nmol)</td>
<td>0.25±0.12**</td>
<td>0.68 (0.86)</td>
<td>0.40 (0.29)</td>
<td>0.20 (0.26)</td>
<td></td>
</tr>
<tr>
<td>PD$_{20}$FEV$_1$-MCh (μmol)</td>
<td>1.12±0.79**</td>
<td>4.39±4.84</td>
<td>0.95±0.64**</td>
<td>1.71 (3.00)</td>
<td></td>
</tr>
</tbody>
</table>

*P<0.05, **P<0.01 for between-group comparison; FeNO, fractional exhaled nitric oxide; FEV$_1$, forced expiratory volume in one second; PD$_{20}$FEV$_1$-LTD$_4$, provocative dose of LTD$_4$ causing a 20% reduction in FEV$_1$; PD$_{20}$FEV$_1$-MCh, provocative dose of methacholine causing a 20% reduction in FEV$_1$. 
on the basis of asthma control level would be necessary when an adequate enrollment has been secured. Our hypothesis therefore urgently needs to be further tested in successive studies with a prolonged treatment course.

Conclusions

Lower PD\textsubscript{20}F\textsubscript{E}V\textsubscript{1}-LTD\textsubscript{4} or higher LTD\textsubscript{4}/MCh potency ratio might suggest preferable outcomes of montelukast therapy in
terms of airway inflammation and spirometry, but not airway hyperresponsiveness or quality of life in asthmatic patients.

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**References**


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