Bronchodilator Response to Albuterol After Regular Formoterol and Effects of Acute Corticosteroid Administration*

Brian J. Lipworth, MD; and Imran Aziz, MD

Background: There is controversy about the development of bronchodilator subsensitivity after regular administration of long-acting β₂-agonists.

Objectives: The purpose of the study was to evaluate whether regular treatment with formoterol affects the bronchodilator response to repeated puffs of albuterol, and also to assess the effects of acute administration of a bolus dose of IV or inhaled corticosteroid.

Materials and methods: Twelve patients (mean [SD] age, 43 [15] years; FEV₁, 57 [17] % predicted) with stable, moderate to severe persistent asthma who were all taking inhaled corticosteroids were evaluated in a randomized, placebo-controlled, double-blind, double-dummy, crossover study. Patients received treatments each for 2 weeks followed by a bolus (IV/inhaled) of corticosteroid or placebo: (1) placebo inhaler bid followed by placebo; (2) formoterol Turbuhaler 24 μg metered dosage bid (delivered dosage 18 μg bid) followed by placebo; (3) formoterol 24 μg bid + bolus IV hydrocortisone, 200 mg; or (4) formoterol 24 μg bid + bolus inhaled budesonide, 1,600 μg. Bronchodilator response to repeated puffs of albuterol (200 to 1,600 μg) for >80 min was measured at 2 h after bolus administration of placebo or corticosteroid. The study was powered at the 80% level to detect a 20% difference in area under curve between 20 and 80 min (AUC) for FEV₁ response to albuterol as change from baseline (primary end point).

Results: There was significant subsensitivity (p = 0.01) of the mean albuterol FEV₁ response (as AUC, L × s) after formoterol alone (737) as compared to placebo (1,453) along with partial reversal by steroid administration: formoterol + hydrocortisone (1,050), and formoterol + budesonide (942). There was a similar pattern of subsensitivity (p = 0.03) for the mean albuterol forced expiratory flow rate between 25% and 75% of vital capacity response (as AUC, L): placebo (2,149), formoterol alone (1,002), formoterol + hydrocortisone (1,402), and formoterol + budesonide (1,271).

Conclusion: Regular treatment with formoterol produced significant bronchodilator subsensitivity to repeated puffs of albuterol, which was partially reversed by a bolus dose of systemic or inhaled corticosteroid.

Key words: β₂-adrenoceptor; albuterol; asthma; bronchodilator; corticosteroids; formoterol; genetic polymorphism; subsensitivity; tachyphylaxis

Abbreviations: Arg = arginine; AUC = area under curve between 20 and 80 min; Bmax = lymphocyte β₂-adrenoceptor binding density; CI = confidence interval; DRC = dose-response curve; FEF₂₅–₇₅% = forced expiratory flow rate between 25% and 75% of vital capacity (mid-expiratory flow); Gln = glutamine; Glu = glutamic acid; Gly = glycine; Kd = lymphocyte β₂-adrenoceptor binding affinity (dissociation constant)

Salmeterol and formoterol are long-acting β₂-agonists that are recommended for use as additional therapy to inhaled corticosteroids. There is much debate about the regular use of these drugs, particularly in regard to the development of tolerance to their bronchodilator and bronchoprotective effects. We have previously shown that 36 h after stopping regular treatment with twice daily salmeterol there is persistent bronchodilator subsensitivity to repeated puffs of albuterol along with down-regulation of lymphocyte β₂-adrenoceptors. In another study with a similar design, there was no bronchodilator subsensitivity to albuterol 36 h after stopping regular treatment with twice daily formoterol. There is, however, evidence of persistent bronchodilator subsensitivity to repeated puffs of formoterol 24 h after stopping regular treatment.
with twice daily formoterol. There was criticism of the latter study because in real life clinical practice inhaled formoterol would not normally be used as repeated puffs for acute relief therapy.

The objectives of the present study were twofold. First, we wanted to assess whether bronchodilator subsensitivity to repeated puffs of albuterol occurs after regular treatment with formoterol 24 h after stopping treatment. Second, we wanted to evaluate the effect of acute administration of an IV bolus or inhaled corticosteroids on the albuterol response. We have previously shown that acute administration of a bolus dose of IV hydrocortisone, 200 mg, along with oral prednisolone, 50 mg, rapidly reversed bronchodilator subsensitivity to repeated puffs of inhaled formoterol. In the present study, we have investigated the acute effects of a lower dose of systemic corticosteroid (IV hydrocortisone, 200 mg, alone) as well as a high dose of inhaled steroid (budesonide, 1.6 mg, via Turbuhaler; Astra Pharmaceuticals; Kings Langley, UK).

**Materials and Methods**

**Patients**

Twelve patients with stable, moderate to severe persistent asthma (7 women and 5 men; mean ± SD age, 43 ± 15 years; all taking inhaled corticosteroids) were recruited to take part in a randomized, placebo-controlled, double-blind, crossover study (Table 1). All were using inhaled short-acting β2-agonists (less than two puffs per day) for symptomatic relief purposes. Two subjects were taking inhaled salmeterol and one subject was taking inhaled formoterol twice daily. Three subjects were also taking oral theophylline twice daily. The subjects had stable asthma according to the American Thoracic Society criteria for at least 3 months before randomization, and no one had received oral steroids or antibiotics during this time. All subjects were nonsmokers and had demonstrated ≥ 15% reversibility in FEV1, or forced expiratory flow rate between 25% and 75% of vital capacity (FEF25–75%; mid-expiratory flow) to albuterol, 400 μg at recruitment. All subjects gave written informed consent before being randomized in the study, which was approved by the Tayside committee on medical research ethics.

On completion of the study, we also performed a post hoc genotype analysis of our study patients for β2-adrenoceptor polymorphisms. All patients who were homozygous for the glutamic acid (Glu)-27 were also homozygous for glycine (Gly)-16, which is a well-known linkage disequilibrium.

**Protocol**

A randomized, double-blind, double-dummy, crossover design was used. After an initial 1-week run-in period, subjects were randomized to receive four consecutive 2-week randomized treatment blocks with either inhaled placebo bid (in one limb) or inhaled formoterol 24 μg bid (in the three other limbs). Formoterol was delivered by dry powder Turbuhaler, 12 μg metered dose per actuation, 9 μg delivered dose (formoterol fumarate, Oxis Turbuhaler). Placebo was also delivered using Turbuhaler. The medication was taken between 7:00 PM and 9:00 PM for the evening dose and between 7:00 AM and 9:00 AM for the morning dose. The last dose of each treatment was taken in the morning 24 h before each laboratory visit.

From the start of the run-in period until the end of the study, all β2-agonist therapy was stopped and was substituted with inhaled ipratropium bromide (Atrovent Forte; Boehringer Ingelheim; Bracknell, UK) to be used as two puffs (80 μg) for symptomatic rescue relief. The subjects continued to use their inhaled corticosteroids unchanged throughout the study apart from stopping them for 24 h before each study visit (ie, the last 24 h before each study visit).

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dose of the study inhaler was taken with the last dose of inhaled corticosteroid). Similarly, the oral theophylline was omitted for 48 h before each study visit.

**Albuterol Dose-Response Curve**

Subjects attended the laboratory at 7:30 AM, approximately 24 h after the last dose of each of the four treatment periods, having withheld their reliever inhaler (ipratropium) for at least 12 h. An IV cannula was inserted and kept patent with a bolus of heparinized saline solution. After at least 30 min of supine rest, blood was withdrawn for 8:00 AM plasma cortisol after removing the cannula dead space of 1 mL, which was discarded.

The subjects then received from a second blinded investigator one of the following: (1) IV saline solution and inhaled placebo (after placebo pretreatment limb); (2) IV saline solution and inhaled placebo (after formoterol pretreatment); (3) IV hydrocortisone 200 mg (Solu-cortef, hydrocortisone sodium succinate; Pharmacia and Upjohn Ltd; Knowlhill, UK) and inhaled placebo (after placebo pretreatment limb); (2) IV saline solution and inhaled budesonide 1.6 mg Turbuhaler (after formoterol pretreatment); or (4) IV saline solution and inhaled budesonide 1.6 mg Turbuhaler (after formoterol pretreatment). Thus, the following treatment combinations were administered: (1) placebo alone, (2) formoterol alone, (3) formoterol and budesonide, (4) IV saline solution and inhaled budesonide 1.6 mg Turbuhaler (after formoterol pretreatment); or (4) IV saline solution and inhaled placebo (after placebo pretreatment limb); (2) IV saline solution and inhaled placebo (after formoterol pretreatment); or (4) IV saline solution and inhaled placebo (after placebo pretreatment limb). Thus, the following treatment combinations were administered: (1) placebo alone, (2) formoterol alone, (3) formoterol and budesonide, (4) IV saline solution and inhaled budesonide 1.6 mg Turbuhaler (after formoterol pretreatment). Plasma cortisol was collected at 8:00 AM before inhalation and injection, and again at 9:00 AM, 10:00 AM, 11:00 AM, and 12:00 noon. At 2 h after placebo or steroid administration, 40 mL of blood was withdrawn for lymphocyte β2-adrenoceptor analysis. A dose-response curve (DRC) to inhaled albuterol (Ventolin Accuhaler, albuterol sulfate, 200 µg per actuation; Allen and Hanburys; Uxbridge, UK) was then constructed, using consecutive doses of 200 µg, 200 µg, 400 µg, and 800 µg albuterol (ie, cumulative doubling doses of 200 µg, 400 µg, 800 µg, and 1,600 µg, respectively). The doses were separated by 20-min intervals with measurements made over an 80-min period from baseline. Spirometry was performed before the first dose at baseline and 15 min after each dose increment and before the next dose of albuterol.

**Domiciliary Peak Flow**

The patients were instructed to record their morning and evening peak expiratory flow rate using a peak flowmeter (Vitalograph Ltd; Buckingham, UK) immediately before taking their study medication. The subjects took three readings and recorded the highest reading in the diary card.

**Spirometry**

Spirometry was performed according to American Thoracic Society criteria using a compact spirometer (Vitalograph Ltd) with a pneumotachograph head and pressure transducer and on-line computer-assisted determination of FEV1 and FEF25–75%, using best test values.

**Lymphocyte β2-Adrenoceptor Binding Variables**

Lymphocyte β2-adrenoceptor binding variables were measured as previously described, using (−)125I-ioctoanopindolol. Assessable data for lymphocyte β2-adrenoceptor binding density (Bmax) and lymphocyte β2-adrenoceptor binding affinity (dissociation constant; Kd) were only available in 9 of 12 patients for all four treatments because of a malfunction in the binding assay.

**Plasma Cortisol Assay**

The assays were performed in duplicate using a commercial radioimmunoassay kit (Immunodiagnostic Systems Ltd; UK), which has no cross-reactivity with budesonide, but could evidently not be used with hydrocortisone. The coefficients of variation for analytical imprecision for intra-assay and inter-assay were 7.1% and 7.2%, respectively.

**Identification of β2-Adrenoceptor Polymorphisms**

β2-Adrenoceptor polymorphisms were identified as previously described. In brief, genomic DNA was extracted from whole blood, and a 234-base pair fragment generated by polymerase chain reaction spanned the regions of interest. Genotype was determined by allele-specific oligonucleotide hybridization using probes homologous for the arginine (Arg)-16, glycine (Gly)-16, glutamine (Gln)-27, or glutamic acid (Glu)-27 forms of the receptor.

**Statistical Analysis**

The study was powered at the 80% level with 12 subjects to detect a 20% difference in the primary end point as ΔFEV1 response (area under curve between 20 and 80 min [AUC]) between placebo and active treatments. All DRC variables were analyzed as change in response from baseline. Comparisons for Δ responses from the DRC were made as AUC and as peak response. The lymphocyte β2-adrenoceptor binding parameters were logarithmically transformed prior to analysis. Comparisons were made by multivariate analysis of variance, using subject, study treatment, albuterol dose, and period as factors for the analysis. This was followed by applying Bonferroni multiple-range testing (set at 95% confidence interval [CI]) to obviate multiple pairwise comparisons. The domiciliary morning and evening peak flow data were calculated as the mean of 7 days prior to the last dose of each treatment period. Data analysis was performed using a Statgraphics statistical analysis software package (STSC Software Publishing Group, Rockville, MD). The genotype data were not subjected to statistical analysis because of the small number of subjects within a given genotype for each codon.

**RESULTS**

Thirteen subjects were recruited in total; 1 subject dropped out because of an exacerbation of asthma during the third formoterol limb (the last limb of the trial) caused by a chest infection. The data for this patient were excluded from the analysis, and one more subject was randomized to completion. All of these 12 subjects completed the study, and no other adverse events were reported. Treatment with formoterol produced significant improvements in morning (p = 0.004) and evening (p = 0.0004) domiciliary peak flow recordings as compared with placebo during all three active treatment limbs (Fig 1).

There were no significant (p = 0.3) differences in mean values for baseline FEV1 (ie, before the DRC) between the four treatment periods (Table 2). For the primary end point (ΔFEV1 AUC), treatment with formoterol produced significant (p = 0.01) bronchodilator subsensitivity compared with placebo for all measured variables, as evidenced by a rightward shift in the albuterol DRC (Fig 2 and Table 3).
Mean values for ΔFEV₁ (as AUC, L × s), comparing placebo vs formoterol, were 1,453 vs 736 (95% CI for difference, 132 to 1,302); and for peak ΔFEV₁, placebo vs formoterol, 0.39 vs 0.24 L (95% CI for difference, 0.001 to 0.28 L); Fig 2 and Table 3. Similarly, there was significant (p = 0.03) subsensitivity for ΔFEF₂₅–₇₅% (as AUC), comparing placebo vs formoterol: 2,149 vs 1,002 L (95% CI for difference, 163 to 2,130 L); and for peak ΔFEF₂₅–₇₅% response, placebo vs formoterol: 0.57 vs 0.31 L/s (95% CI for difference, 0.03 to 0.50 L/s); Fig 2 and Table 3.

Bmax (as geometric mean) showed significant down-regulation (p = 0.02) with formoterol as compared to placebo: 2.56 vs 3.00 fmol/10⁶ cells, a 1.2-fold difference (95% CI, 1.02-fold to 1.35-fold; p = 0.03). Values for Bmax with IV steroid (2.63 fmol/10⁶ cells) or inhaled steroid (2.75 fmol/10⁶ cells) showed no difference from placebo or formoterol alone. Values for Kd (as geometric mean) showed no difference (p = 0.2) between any of the treatments: placebo (21 pmol) vs formoterol (23 pmol) vs formoterol and hydrocortisone (17 pmol) vs formoterol and budesonide (24 pmol).

### Table 2—Spirometry Prior to DRC*

<table>
<thead>
<tr>
<th>Treatment</th>
<th>FEV₁, L</th>
<th>FEV₁, % Predicted</th>
<th>FEF₂₅–₇₅%, L/s</th>
<th>FEF₂₅–₇₅%, % Predicted</th>
</tr>
</thead>
<tbody>
<tr>
<td>Placebo</td>
<td>1.67 (1.54–1.79)</td>
<td>54 (49–58)</td>
<td>1.30 (1.15–1.44)</td>
<td>32 (29–36)</td>
</tr>
<tr>
<td>Formoterol</td>
<td>1.79 (1.67–1.92)</td>
<td>57 (53–62)</td>
<td>1.43 (1.29–1.58)</td>
<td>36 (32–39)</td>
</tr>
<tr>
<td>Formoterol + hydrocortisone</td>
<td>1.67 (1.54–1.79)</td>
<td>54 (50–58)</td>
<td>1.35 (1.21–1.50)</td>
<td>34 (30–38)</td>
</tr>
<tr>
<td>Formoterol + budesonide</td>
<td>1.78 (1.65–1.90)</td>
<td>57 (53–61)</td>
<td>1.49 (1.34–1.64)</td>
<td>38 (34–42)</td>
</tr>
</tbody>
</table>

*There were no significant differences between the four treatments for FEV₁ (p = 0.3) and FEF₂₅–₇₅% (p = 0.4). Values are shown as mean (95% CI). Measurements were made 26 h after the last dose of prior treatment with placebo or formoterol, and 2 h after bolus administration of placebo, hydrocortisone, or budesonide.
There was partial reversal of the subsensitivity when formoterol was given in conjunction with inhaled or IV steroid in that the responses were not significant compared with placebo pretreatment, but at the same time were also not significant compared with pretreatment with formoterol alone. Mean values for peak delta response and AUC showed a similar trend in this respect for both FEV1 and FEF25–75% (Fig 2 and Table 3).

Individual data comparing the albuterol response between placebo and formoterol alone showed that for the ΔFEV1 (AUC) response in 8 of 12 subjects had >20% attenuation, whereas for ΔFEF25–75% (AUC), 9 of 12 subjects had >20% attenuation. Concerning patients who had the Gly-16 allele (ie, homozygous or heterozygous at codon 16), there were 2 of 11 who did not show subsensitivity for ΔFEV1 response and 1 of 11 did not show subsensitivity for ΔFEF25–75% response.

The 8:00 AM plasma cortisol (before steroid administration) was not significantly different (p = 0.8) among the three treatments (ie, placebo vs formoterol vs formoterol + budesonide; Table 4). The 4-h plasma cortisol profile (8:00 AM to 12:00 noon) showed significant (p = 0.003) suppression after budesonide administration compared to placebo (Table 4). The plasma cortisol profile after hydrocortisone could not be described as being caused by lack of separation between endogenous and exogenous glucocorticoids.

**Discussion**

Our results showed that regular twice-daily formoterol produced subsensitivity of the bronchodilator response to repeated puffs of albuterol for effects on both FEV1 and FEF25–75% response (as AUC). We found that an approximately eightfold higher dose of albuterol would be required to elicit the

![Figure 2. Mean DRC for ΔFEV1 and ΔFEF25–75% response to cumulative puffs of albuterol for the four treatments. *p < 0.05, response compared with placebo pretreatment. Error bars (± SEM) are only shown for the maximum albuterol dose. See Figure 1 for abbreviations.](http://publications.chestnet.org/pdfaccess.ashx?url=/data/journals/chest/21938/)

<table>
<thead>
<tr>
<th>Variables</th>
<th>Placebo</th>
<th>Formoterol</th>
<th>Formoterol + Hydrocortisone</th>
<th>Formoterol + Budesonide</th>
</tr>
</thead>
<tbody>
<tr>
<td>AUC FEV1, L × s</td>
<td>1453 (1,153–1,753)</td>
<td>736* (436–1,036)</td>
<td>1050 (750–1,350)</td>
<td>942 (642–1,242)</td>
</tr>
<tr>
<td>AUC FEF25–75%, L</td>
<td>2149 (1,644–2,652)</td>
<td>1002* (497–1,506)</td>
<td>1402 (898–1,907)</td>
<td>1271 (766–1,775)</td>
</tr>
<tr>
<td>Peak FEV1, L</td>
<td>0.39 (0.31–0.47)</td>
<td>0.24* (0.16–0.31)</td>
<td>0.31 (0.23–0.39)</td>
<td>0.29 (0.20–0.37)</td>
</tr>
<tr>
<td>Peak FEF25–75%, L/s</td>
<td>0.57 (0.45–0.69)</td>
<td>0.31* (0.18–0.42)</td>
<td>0.43 (0.31–0.55)</td>
<td>0.38 (0.26–0.50)</td>
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</table>

*Significant difference in response between placebo and formoterol by Bonferroni multiple-range testing. Values given as mean (95% CI).
same increase in FEV₁ after formoterol compared to placebo. The present study only looked at airway β₂-adrenoceptor responses, although we have previously reported subsensitivity of systemic β₂-adrenoceptor responses to repeated puffs of albuterol after treatment with regular formoterol.¹²

There is, however, controversy about the development of bronchodilator desensitization with long-acting β₂-agonists. Ullman et al,¹³ Wilding et al,¹⁴ and Nelson et al¹⁵ with salmeterol, and Arvidsson et al¹⁶ with formoterol reported that bronchodilator subsensitivity to albuterol did not occur. In all four of these studies, the baseline spirometry (before albuterol) was performed within 12 h of dosing with active treatment, and consequently, values were higher as compared to placebo, which would result in confounding of the subsequent albuterol response. Also, β₂-agonist rescue medication was permitted instead of ipratropium (except for Nelson et al¹⁵), and there was no β₂-agonist washout period before the study. In our study, the baseline spirometry during active treatments was performed at 24 h and was not statistically different from placebo, which may explain why it was possible to demonstrate a rightward shift in albuterol response. Furthermore, the patients in our study had more severe asthma, and hence the ceiling of the FEV₁ DRC was probably not attained. It is important to point out that we decided before the study to analyze the albuterol FEV₁ response as change from baseline in keeping with analysis of DRCs from all of our previous studies.¹⁴,⁷,¹²,¹⁷ Inasmuch as the ΔFEV₁ response was used to power the study, we therefore feel justified in making valid conclusions on the basis of our primary end point.

We did not show, as in our previous study, that a bolus of systemic steroid completely reverses bronchodilator subsensitivity after regular treatment with formoterol 24 µg bid.⁷ Our results suggested only a partial reversal of subsensitivity with both IV hydrocortisone and inhaled budesonide, in that the albuterol responses were no longer significantly different from placebo, but at the same time they were not significantly different from formoterol alone. What are the possible explanations to account for this finding? We used a smaller dose of systemic steroid (hydrocortisone, 200 mg) in the current study, as compared with hydrocortisone, 200 mg, plus prednisolone, 50 mg (combined equivalent dose of hydrocortisone 400 mg) in our previous study. We chose to use the maximum recommended daily dose (1.6 mg) of inhaled budesonide (Astra Draco; Lund, Sweden) as well as a standard recommended dose of hydrocortisone (200 mg) for the treatment of acute asthma.¹⁸ It is conceivable that the acute effects of inhaled and IV corticosteroids on airway β₂-adrenoceptors may be dose related.

Another factor that may have influenced the results was that in our previous study we had used repeated puffs of formoterol to construct the DRC; consequently, we had evaluated the time profile after the last puff. We did not evaluate the time profile after the last puff in the present study when we used albuterol, because it has a much shorter duration of action. The degree of corticosteroid reversibility may also relate to the intrinsic activity of the agonist in terms of albuterol exhibiting a lower agonist activity compared with formoterol, a full agonist.¹⁸ We had also found no significant increase in Bmax at 1 h after the corticosteroid bolus dose in the previous study,⁷ as opposed to evaluating effects at 2 h in the current study. The choice of making measurements 2 h after corticosteroid administration was based on in vitro data showing that dexamethasone increases gene transcription of β₂-adrenoceptor messenger RNA, with a peak effect occurring within 2 h.¹⁹ Further studies using different corticosteroid doses and time profiles are indicated to further evaluate these effects.

It is also worth comparing the current data with another study in which we observed rapid reversal of formoterol-induced subsensitivity to adenosine monophosphate bronchoprotection and lymphocyte β₂-adrenoceptor down-regulation, at 2 h after inhalation of a 1,600-µg bolus dose of budesonide.²⁰ This may reflect differential effects of corticosteroids on β₂-adrenoceptors of airway mast cells compared with smooth muscle. It is known that β₂-agonist–induced bronchoprotective subsensitivity is more pronounced for effects on mast cells than smooth muscle.²¹ The degree of formoterol-induced down-regulation of lymphocyte β₂-adrenoceptors was greater in our previous study,²⁰ probably reflecting the measurement at 2 h after the last dose of formoterol, in contrast to 24 h in the current study.

It is known that β₂-adrenoceptor polymorphism at

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**Table 4—Cortisol Before and After Steroid Administration**

<table>
<thead>
<tr>
<th>Variables</th>
<th>Pretreatment</th>
<th>Posttreatment Average</th>
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<tbody>
<tr>
<td></td>
<td>S:00 AM</td>
<td>to 12:00 Noon</td>
</tr>
<tr>
<td>Plasma Cortisol, nmol/L</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Placebo + placebo</td>
<td>476 (416–536)</td>
<td>321 (281–362)</td>
</tr>
<tr>
<td>Formoterol + placebo</td>
<td>471 (412–531)</td>
<td>324 (294–365)</td>
</tr>
<tr>
<td>Formoterol + budesonide</td>
<td>450 (391–510)</td>
<td>231† (191–272)</td>
</tr>
</tbody>
</table>

*Plasma cortisol values are shown as means (95% CI) at baseline prior to injected/inhaled corticosteroid or placebo administration and for 4 hours after corticosteroid administration, after pretreatment with placebo or formoterol.

†Significant suppression of plasma cortisol (8:00 AM to 12:00 noon) after administration of budesonide compared to placebo.
codon 16 determines the propensity for bronchodilator subsensitivity and β2-adrenoceptor down-regulation, in particular with the homozygous Gly-16 genotype.5,22 In our study, almost all of the patients who were homozygous Gly-16 or heterozygous Gly/Arg-16 exhibited bronchodilator desensitization. Indeed, the only subject who did not show subsensitivity for FEV1 response was homozygous for arginine at codon 16 of the β2-adrenoceptor. A population-based study has shown that only 11% of subjects have the homozygous Arg-16 genotype,11 and so the majority of patients who have the Gly-16 allele will be predisposed to down-regulation.

We accept the limitations of our study in that we have studied stable, moderate-to-severe asthmatic subjects in a controlled laboratory environment. In an acute exacerbation, there would be increased bronchomotor tone, which might influence the bronchodilator response to albuterol as a consequence of altered airway geometry.18 Our study had limited power (80%) because of the small sample size. Thus, we are unable to exclude the possibility that a difference between treatments in ΔFEV1 response (as AUC) of < 20% between treatments may have occurred, resulting in a false-negative result. Nevertheless, we were able to show a significant rightward shift in the albuterol dose response comparing placebo and formoterol pretreatment.

In conclusion, bronchodilator subsensitivity to repeated puffs of albuterol developed after regular treatment with formoterol, for effects on FEV1 and FEF25-75% response. This desensitization was only partially reversible 2 h after administration of a bolus dose of systemic hydrocortisone or inhaled budesonide. These findings highlight the potential problems associated with the regular use of long-acting β2-agonist therapy in asthma. Further studies are required to evaluate the effects of regular treatment with long-acting β2-agonists on the albuterol response in the setting of increased bronchomotor tone in acute severe asthma and the influence of acute corticosteroid administration. Such studies should also address acute effects in patients who are particularly susceptible to β2-adrenoceptor down-regulation because of genetic polymorphism.

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