A novel inhaled phosphodiesterase 4 inhibitor (CHF6001) reduces the allergen challenge response in asthmatic patients

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Abstract
CHF6001 is an inhaled phosphodiesterase 4 (PDE4) inhibitor in development for the treatment of obstructive lung diseases. The efficacy and safety of CHF6001 were investigated in a double blind, placebo controlled, 3-way cross-over study using the allergen challenge model. Thirty-six atopic asthmatics who were not taking inhaled corticosteroids and who demonstrated a late asthmatic response (LAR) to inhaled allergen at screening were randomised to receive CHF6001 400 mg or 1200 mg or placebo administered once a day using a dry powder inhaler. The three treatment periods were 9 days; allergen challenges were performed on day 9 and induced sputum was obtained after 10 h from challenge. Washout periods between treatments were up to 5 weeks.
Both CHF6001 doses significantly attenuated the LAR; the primary endpoint analysis showed that CHF6001 400 µg and 1200 µg caused reductions of 19.7% (p = 0.015) and 28.2% (p < 0.001) respectively of the weighted FEV1 AUC4-10h compared with placebo. The difference between the CHF6001 doses was not statistically significant (p = 0.223). Compared with placebo, CHF6001 caused greater reduction in sputum eosinophil counts, although these changes were not statistically significant. CHF6001 was well tolerated, with similar numbers of adverse events in each treatment period.
This inhaled PDE4 inhibitor has the potential to provide clinical benefits in patients with atopic asthma.

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1. Introduction

Cyclic 3’5’-adenosine monophosphate (cAMP) and cyclic guanosine monophosphate (cGMP) are intracellular signalling molecules that regulate immune cell function [1]. Phosphodiesterase (PDE) enzymes hydrolyse these second messengers thereby up-regulating the activity of immune cells [2]. The PDE4 subtype is selectively expressed in immune cells [2]; pharmacological targeting of PDE4 increases cAMP and cGMP levels and thereby reduces immune cell activity. Orally administered PDE4 inhibitors such as roflumilast have anti-inflammatory effects and clinical benefits in asthma and COPD, but also cause side effects such as gastro-intestinal disturbance and weight loss due to systemic exposure [3–6]. CHF6001 is a potent and selective PDE4 inhibitor that is in development for the treatment of obstructive lung diseases [7]. The compound is formulated as dry powder for delivery by inhalation with low aqueous solubility and oral bioavailability in order to reduce the potential for systemic side effects. Intratracheal delivery of CHF6001 has anti-inflammatory effects in different animal models, including allergic inflammation in rats [7].

While there are more clinical data showing the clinical effects of PDE4 inhibitors in COPD [3,4], there are a number of studies that have shown that roflumilast improves lung function in patients with asthma [6]. However, the side effect profile of roflumilast has prevented further development of this drug for asthma [5]. Inhaled
PDE4 inhibitors may be able to overcome this limitation, and provide an extra option for the treatment of asthma.

The inhaled allergen challenge model is commonly used to evaluate the anti-inflammatory effects of novel drugs at an early stage in clinical development [8–11]. Patients with allergic asthma can develop an early asthmatic response (EAR) due to mast cell mediator-induced bronchoconstriction and a late asthmatic response (LAR) due to airway inflammation in response to inhaled allergen. The efficacy and safety of CHF6001 were investigated in a double-blind, placebo-controlled, cross-over study using the allergen challenge model.

2. Methods

2.1. Subjects

Thirty-six (36) patients with physician-diagnosed asthma who had not been treated with inhaled corticosteroids for > 6 months were recruited. Subjects were required to be aged 18–60 years and non-smokers for at least 1 year with < 5 pack-year history. At screening patients were required to have an FEV1 > 70% predicted, have a positive skin test to either house dust mite, grass pollen or cat allergen (ALK-Abelló, Denmark), and to demonstrate both an EAR and LAR to one of these allergens when inhaled. Exclusion criteria are listed in the data supplement. All patients provided written informed consent. The study was approved by the local research ethics committee.

2.2. Study design

This was a three-centre, double-blind, randomised, placebo-controlled, three way cross-over study. Patients who were sensitive to grass pollen were enrolled outside the hay-fever season. Eligible subjects were randomised to receive CHF6001 400 μg (one 400 μg capsule plus two matching placebo capsules) or 1200 μg (three 400 μg capsules) or placebo (three placebo capsules) administered once a day in the morning using a dry powder inhaler (Aerolizer™) for 9 days. The washout period was 4–5 weeks between treatment periods. A balanced block randomization scheme was used. The treatment sequences were arranged according to a complete set of 3 × 3 Latin Square design (6 sequences). Patients were sequentially assigned to the lowest available randomization number.

At screening, the following measurements were performed: vital signs, ECG, forced expiratory volume in 1 s (FEV1), biochemistry and haematology blood tests, skin prick tests and inhaled allergen challenge with induced sputum at 10 h post-allergen challenge. At least 20 subjects able to produce adequate induced sputum at 10 h post-allergen challenge at screening had to be enrolled to the study. At least 20 subjects able to produce adequate induced sputum samples after allergen challenge at screening were required, in order to increase the chance of a subgroup of patients providing adequate sputum samples for analysis. Patients were trained in inhaler technique at screening using the In-check Dial device. The following measurements were performed at pre-dose on day 1, 4 and 9 of each treatment period: FEV1, vital signs and ECG. After dosing on day 9, an inhaled allergen challenge was performed with collection of induced sputum 10 h after challenge. Methacholine challenge was then performed 24 h post-allergen challenge. Adverse events were collected throughout the study using diary cards.

2.3. Allergen and methacholine challenges

Bronchial challenges were performed as previously described [8,9], with full details in the on-line supplement. For allergen challenge at screening, ascending concentrations from 500 SQ-U/ml to 32,000 SQ-U/ml were administered using a Mefar Dosimeter (Mefar-Bologna) until an early asthmatic response (EAR) was observed. The EAR was defined as a fall in FEV1 of ≥ 20% from the post-saline value, on at least one occasion, between 5 and 30 min after the final concentration of allergen. The late asthmatic response (LAR) at screening was defined as a fall in FEV1 of ≥ 15% from the post-saline value, on at least three occasions, two of which must be consecutive, between 4 and 10 h after the final allergen concentration. After randomization, allergen challenges administered a single dose of allergen which was the cumulative dose that caused the LAR at screening. The EAR was measured from 0 to 2 h post allergen, and the LAR from 4 to 10 h post-allergen.

2.4. Induced sputum

Induced sputum was obtained as previously described [9,12]. Sputum plugs were selected to separate sputum from saliva, and then processed using dithiothreitol (DTT, Sigma Aldrich, Poole, UK). Cell counts and viability were determined by the Trypan blue exclusion method utilising a Neubauer haemocytometer. Cytospins were prepared and stained with Rapi-diff (Triangle, Skelmersdale, UK) for differential cell counts. Four hundred leukocytes were counted and the results expressed as percentage of the total leukocyte count and the total cell count (TCC). An adequate sputum sample was defined as > 75 mg with a cell viability > 40%. Sputum supernatants were analysed for eosinophil cationic protein (ECP) using MESACUP ECP kit (MBL international, Aichi, Japan; lower limit of detection 0.125 ng/ml) and neutrophil elastase (NE) by Luminex assay (Merck-Millipore, UK; lower limit of detection 20 pg/ml) on a Magpix analyser (Luminex, USA).

2.5. Pharmacokinetics

Blood samples were collected by venipuncture on day 9 in order to obtain plasma for pharmacokinetic analysis. Plasma CHF6001 (the parent compound) and CHF5956 (a metabolite) concentrations were determined at Quotient Bioresearch Ltd. (UK) using a validated LC-MS/MS method, with a lower quantification limit (LQL) of 10 pg/ml for both analytes (validated range: 10–1,000 pg/ml). The precision and accuracy were within ± 20% at the LQL and within ± 15% at all other concentrations.

2.6. Statistics

The primary efficacy endpoint was the inhibition of the LAR on Day 9, expressed as the area under curve (AUC) of the FEV1 percent changes from post-diluent normalised by time (weighted AUC). The post-diluent FEV1 measurement was used as the pre-challenge reference value to calculate the change in FEV1 at each time point assessment. These changes were expressed as follows:

1) absolute change: \( \text{FEV1 at time } t - \text{ FEV1 post-diluent} \)
2) percent change: \( \frac{\text{FEV1 at time } t - \text{ FEV1 post-diluent}}{\text{FEV1 post-diluent}} \times 100 \)

The AUC of these changes was calculated using the linear trapezoidal rule as follows:

\[
\mathrm{AUC}(t) = \sum \left[ \frac{t_{i} + t_{i+1}}{2} \times (C(t) - C(t_{i})) \right],
\]

\[
C(t) = \text{cumulative AUC up to time } t = \text{AUC}(t-1) + 0.5 \times (\text{FEV1 % change from post-diluent} (t-1) + \text{FEV1 % change from post-diluent} (t)) \times (\text{time}(t)-\text{time}(t-1)).
\]

The AUC normalised by time (or weighted AUC) was obtained by dividing the AUC by time interval considered as follows: \( \text{AUC/actual time span by AUC} \).

A sample size of 24 evaluable patients ensures 85% power to detect a difference of at least 30% in attenuating weighted LAR with CHF6001 compared with placebo, with a 0.025 two-sided
3. Results

Thirty-six subjects were randomised (Table 1), with the following allergens administered: house dust mite n = 22, cat n = 5, grass pollen n = 9. Thirty-three subjects completed the study. One subject withdrew the consent and two others were withdrawn because of adverse events during the first treatment period. These adverse events were an asthma exacerbation after 4 days of treatment with CHF6001 400 µg, and a severe allergic reaction to inhaled allergen after placebo treatment.

3.1. Allergen challenge

The time profile of the FEV1 response to allergen challenge is shown in Fig. 1. Both doses of CHF6001 significantly attenuated the LAR compared to placebo. The primary endpoint analysis showed that CHF6001 400 µg and 1200 µg caused reductions of 19.7% (p = 0.015) and 28.2% (p < 0.001) respectively of the weighted FEV1 AUC4–10h (% change) compared with placebo (Table 2). The effect on AUC4–10h (% change) did not differ between the two CHF6001 doses (p = 0.223). Similar results were obtained using the absolute FEV1 change from baseline: attenuation of 20.3% with CHF6001 400 µg (p = 0.014) and 29.9% with CHF6001 1200 µg (p < 0.001) compared with placebo were observed, with no difference between CHF6001 doses. The maximum FEV1% fall was attenuated by CHF6001 400 µg and 1200 µg (15.6%; p = 0.028 and 22.9%; p = 0.001 respectively). There were similar results for the maximum absolute FEV1 fall (L) (Table 2). CHF6001 400 µg and 1200 µg had no significant effect on the EAR AUC0–2h (% change) or maximum fall compared to placebo (Table 2).

3.2. Pulmonary function before allergen challenge

FEV1 measured pre-dose on days 4 and 9 were similar to baseline values for CHF6001 and placebo (see on-line Table 1).

3.3. Methacholine challenge

The methacholine PC20 FEV1 geometric means (95% CI) 24 h post-allergen challenge were 0.652 (0.499; 0.851), 0.826 (0.627; 1.088) and 0.505 (0.378; 0.674) mg/mL after treatment with CHF6001 1200 µg, CHF6001 400 µg and placebo, respectively. There was a statistically significant difference in MCh PC20 FEV1 of 0.7 doubling doses between CHF6001 400 µg and placebo (p = 0.028). The doubling dose difference between CHF6001 1200 µg and placebo was 0.4 (p = 0.331).

3.4. Induced sputum

The numbers of viable sputum samples obtained after treatment with CHF6001 1200 µg, CHF6001 400 µg and placebo were 14, 14 and 17 respectively. Eosinophil counts are shown in Table 3. Compared with placebo, CHF 6001 caused greater reductions in eosinophil absolute counts and percentages, although these changes did not attain statistical significance. There were no differences for other cell types, or ECP or NE concentrations, as shown in on-line Table 2.

3.5. Pharmacokinetics

Plasma concentrations of CHF6001 and the metabolite CHF5956 on day 9 are shown in Fig. 2. The systemic exposure to CHF6001 was proportional to the dose administered, with AUC0–10h geometric means of 4,960 h pg/mL and 14,746 h pg/mL for CHF6001 400 µg and 1200 µg, respectively, indicating an approximately three times higher systemic exposure with a 3-fold increase in the dose. The Cmax geometric means followed a similar pattern; CHF6001 346 pg/mL and 1,025 pg/mL for 400 µg and 1200 µg, respectively. CHF5956 pharmacokinetics also showed a relationship to the dose administered, with AUC0–10h geometric means of 166 h pg/mL and 813 h pg/mL for CHF6001 400 µg and 1200 µg, respectively. The Cmax geometric means followed a similar pattern; 34.6 pg/mL and 95.8 pg/mL for CHF6001 400 µg and 1200 µg, respectively.

3.6. Adverse events

Adverse events were reported by similar numbers of patients within each treatment period; 38.2% and 30.3% for CHF6001 400 µg and 1200 µg, respectively, and 37.1% for placebo. Adverse events were mostly mild in nature, the most common being headache (see on-line Table 3). Gastrointestinal adverse events were uncommon, with one episode of dyspepsia during CHF6001 400 µg treatment and one during placebo treatment.

4. Discussion

This clinical trial was designed to evaluate the efficacy of the
Fig. 1. Time course of FEV<sub>1</sub> changes after allergen challenge conducted after 9 days dosing. Adjusted mean % change in FEV<sub>1</sub> (from post diluent value) is shown on y-axis. Error bars represent SEM.

Table 2
Late and early asthmatic response (FEV<sub>1</sub> weighted mean AUC and maximum fall) after 9 days treatment.

<table>
<thead>
<tr>
<th>Treatment effect</th>
<th>Difference versus placebo</th>
</tr>
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<tbody>
<tr>
<td><strong>LAR</strong></td>
<td></td>
</tr>
<tr>
<td>AUC 4–10 h (%)</td>
<td></td>
</tr>
<tr>
<td>400mcg</td>
<td>-16.70 (18.73; -14.68)</td>
</tr>
<tr>
<td>1200mcg</td>
<td>-14.94 (16.96; -12.92)</td>
</tr>
<tr>
<td>Placebo</td>
<td>-20.80 (22.98; -18.62)</td>
</tr>
<tr>
<td>AUC 4–10 h (L)</td>
<td></td>
</tr>
<tr>
<td>400mcg</td>
<td>-0.552 (-0.621; -0.483)</td>
</tr>
<tr>
<td>1200mcg</td>
<td>-0.486 (-0.555; -0.417)</td>
</tr>
<tr>
<td>Placebo</td>
<td>-0.693 (-0.767; -0.618)</td>
</tr>
<tr>
<td>Max Fall (%)</td>
<td></td>
</tr>
<tr>
<td>400mcg</td>
<td>-26.07 (-28.70; -23.44)</td>
</tr>
<tr>
<td>1200mcg</td>
<td>-23.84 (-26.47; -21.21)</td>
</tr>
<tr>
<td>Placebo</td>
<td>-30.91 (-33.74; -28.07)</td>
</tr>
<tr>
<td>Max fall (L)</td>
<td></td>
</tr>
<tr>
<td>400mcg</td>
<td>-0.865 (-0.956; -0.775)</td>
</tr>
<tr>
<td>1200mcg</td>
<td>-0.782 (-0.873; -0.691)</td>
</tr>
<tr>
<td>Placebo</td>
<td>-1.033 (-1.131; -0.915)</td>
</tr>
<tr>
<td><strong>EAR</strong></td>
<td></td>
</tr>
<tr>
<td>AUC0-2 h (%)</td>
<td></td>
</tr>
<tr>
<td>400mcg</td>
<td>-15.417 (-17.766; -13.074)</td>
</tr>
<tr>
<td>1200mcg</td>
<td>-14.901 (-17.244; -12.558)</td>
</tr>
<tr>
<td>Placebo</td>
<td>-16.115 (-18.511; -13.720)</td>
</tr>
<tr>
<td>AUC0-2 h (L)</td>
<td></td>
</tr>
<tr>
<td>400mcg</td>
<td>-0.511 (-0.583; -0.440)</td>
</tr>
<tr>
<td>1200mcg</td>
<td>-0.487 (-0.558; -0.415)</td>
</tr>
<tr>
<td>Placebo</td>
<td>-0.547 (-0.620; -0.473)</td>
</tr>
<tr>
<td>Max Fall (%)</td>
<td></td>
</tr>
<tr>
<td>400mcg</td>
<td>-27.710 (-30.646; -24.774)</td>
</tr>
<tr>
<td>1200mcg</td>
<td>-26.755 (-29.691; -23.818)</td>
</tr>
<tr>
<td>Placebo</td>
<td>-30.100 (-33.103; -27.098)</td>
</tr>
<tr>
<td>Max fall (L)</td>
<td></td>
</tr>
<tr>
<td>400mcg</td>
<td>-0.916 (-1.008; -0.825)</td>
</tr>
<tr>
<td>1200mcg</td>
<td>-0.889 (-0.981; -0.798)</td>
</tr>
<tr>
<td>Placebo</td>
<td>-1.008 (-1.102; -0.915)</td>
</tr>
</tbody>
</table>

<sup>a</sup> PE = Point Estimate.
<sup>b</sup> Dunnett’s adjustment.

inhaled PDE4 inhibitor CHF6001 on the response to inhaled allergen in patients with mild atopc asthma. CHF6001 significantly reduced the LAR, and was also well tolerated. Since orally administered PDE4 inhibitors cause side effects due to systemic exposure [3–5], CHF6001 has the potential for a better therapeutic index due to targeted lung delivery thereby markedly reducing systemic exposure.

There has been interest recently in the re-evaluation of the use of PDE4 inhibitors for the treatment of asthma [5,6,13]. A review of placebo controlled clinical trials in asthma using rufomilast reported a benefit in terms of lung function, including when administered in addition to inhaled corticosteroids [6]. However, further evidence for the symptomatic benefits of PDE4 inhibitors in asthma are needed. This study shows inhibition of allergic inflammation by CHF6001. Inhibition of the LAR has become a standard method for investigating the potential of novel drugs to suppress the allergic component of inflammation in asthma, as a lack of effect on the LAR indicates that such drugs will not be useful clinical treatments for allergic asthma [8,14]. This study confirms the potential for CHF6001 as a treatment for asthma, but further studies to investigate long term effects on lung function and symptoms are needed.

Table 3
Sputum percent differential cell count.

<table>
<thead>
<tr>
<th></th>
<th>Placebo N = 17</th>
<th>CHF 400mcg N = 14</th>
<th>CHF 1200mcg N = 14</th>
</tr>
</thead>
<tbody>
<tr>
<td>Eosinophil %</td>
<td>7.1 (4.6;10.8)</td>
<td>5.4 (3.4;8.8)</td>
<td>4.4 (2.7;7.1)</td>
</tr>
<tr>
<td>Neutrophil %</td>
<td>65.5 (58.4;73.5)</td>
<td>71.8 (62.9;81.8)</td>
<td>69.2 (60.6;78.9)</td>
</tr>
<tr>
<td>Macrophage %</td>
<td>12.2 (8.8;16.8)</td>
<td>6.9 (4.8;10.1)</td>
<td>10.5 (7.2;15.3)</td>
</tr>
<tr>
<td>Lymphocyte %</td>
<td>0.0 (0.0;0.0)</td>
<td>0.0 (0.0;0.0)</td>
<td>0.0 (0.0;0.0)</td>
</tr>
</tbody>
</table>

Data are adjusted geometric means and 95% confidence interval.
Previous studies have shown that the orally administered PDE4 inhibitors roflumilast [11,15] and MEM1414 [16], and the inhaled PDE4 inhibitor GSK256066 [17] inhibited the LAR, with effect sizes ranging from approximately 20–43% depending on the endpoint measured (AUC or maximum fall). In the current study, the level of inhibition of the LAR AUC of approximately 20–30% (varying according to the CHF6001 dose) was similar to these previous results. Comparisons between different allergen challenge studies are to be taken with caution due to important methodological differences, including the period of measurement of the LAR.

The lack of effect of CHF6001 on the EAR may indicate that this PDE4 inhibitor does not influence mast cell degranulation, as is the case for inhaled corticosteroids [18,19]. Previous allergen challenge studies using PDE4 inhibitors have shown variable EAR results, with both inhibition [11,15,17] and no inhibition [16,20] observed. This study was specifically powered for the primary endpoint of the LAR FEV₁ AUC, and we observed approximately 20–30% reduction in this parameter. The 1200 µg dose caused a numerically greater effect than 400 µg on the LAR (approximately 10% difference), but these differences were not statistically significant. It should be noted that the study was powered on the basis of a 30% difference between active treatment and placebo, and so the lack of statistical significance between the active treatments with a 10% treatment difference is likely to be related to insufficient statistical power.

The pharmacokinetic analysis showed a linear relationship between systemic exposure and the dose of CHF6001, as AUC₀–τ (geometric mean) was approximately 3 times higher for the 1200 µg dose compared with 400 µg. The increased exposure with 1200 µg appears to be associated with greater efficacy on the LAR (albeit not statistically significant) for the reasons already explained, but no increase in side effects. Indeed, the incidence and nature of side effects with inhaled CHF6001 were similar to placebo; this is encouraging for the further clinical development of this drug, as side effects of oral PDE4 inhibitors such as gastrointestinal disturbance, headache and weight loss are a concern in clinical practice. Although this was a short term study, gastrointestinal disturbance caused by PDE4 inhibitors tends to occur early after starting therapy [3–5,21], and so it is promising that there was an absence of this signal in the current study.

The LAR involves an influx of inflammatory cells into the airways, including eosinophils [22]. CHF6001 caused a reduction in sputum eosinophil counts after allergen challenge, although not statistically significant. A subset of 20 patients who were able to produce adequate sputum samples at screening was specifically recruited, in order to enrich the study population with individuals who were more likely to produce good quality samples after allergen challenge. During the treatment periods, the number of sputum samples ranged from 14 to 17. The study was not powered for sputum eosinophils, and we saw a numerical suppression of these cells by CHF6001 that was not statistically significant. A larger sample size is probably needed to robustly evaluate this endpoint.

Despite the positive effect of CHF6001 on the primary endpoint (LAR), no consistent effect on secondary endpoints, including induced sputum and methacholine challenge conducted after allergen challenge was observed. However, the study was not powered for these secondary endpoints, and many other previous allergen challenge studies of novel therapies have also shown positive efficacy on the primary LAR endpoints but failed to show efficacy on secondary endpoints such as methacholine challenge [8,10,17,18,23].

The FEV₁ measurements on days 4 and 9 (before allergen challenge) showed no change compared to baseline. This might be due to the very short term treatment with CHF6001 and the very mild airway obstruction of enrolled patients.

Inhaled PDE4 inhibitors, such as CHF6001, are likely to be used as add on to inhaled corticosteroid plus long acting beta agonist. Bateman et al. recently reported that roflumilast added to inhaled corticosteroid plus long acting beta agonist plus leukotriene antagonist treatment resulted in an FEV₁ improvement of approximately 100 ml [24]. This demonstrates the potential for PDE4 inhibitors as add on treatment to other standard asthma therapies.

The current study was performed in asthma patients who were not taking ICS, in order to avoid any confounding effect of ICS treatment on the response to inhaled allergen challenge. This is the population used for most allergen challenge studies of novel drugs for asthma [8–11], and the demonstration of a positive effect using this study design usually encourages further studies of longer duration in moderate to severe asthma patients. The effects of CHF6001 in mild asthma patients not taking ICS observed here also suggests that longer clinical trials could be warranted in this patient group. However, the firmly established place of ICS as a first line maintenance treatment for asthma makes it difficult for any novel anti-inflammatory drug to be a recognised treatment before ICS.

The patients in this study all demonstrated a positive LAR. Clinical trials in asthma patients often require the demonstration of either bronchodilator reversibility or methacholine reactivity at screening to confirm the diagnosis of asthma. We used the presence of an LAR as confirmation, and so did not perform other confirmatory tests.

In conclusion, the inhaled PDE4 inhibitor CHF6001 significantly reduced the LAR in patients with atopic asthma. Further studies of the long-term efficacy and safety of CHF6001 are now needed, as inhaled delivery of this drug has the potential to provide clinical benefits with an acceptable safety profile.

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**Fig. 2.** Pharmacokinetic profiles of CHF6001 and CHF5956 after 9 days dosing. Mean values are shown.
Conflict of interest

D. Santoro is a full time employee of Chiesi Farmaceutici SpA.
M.A. Nandeul is a full time employee of Chiesi Farmaceutici SpA.
F. Mariotti is a full time employee at Chiesi Farmaceutici SpA.
P.J. Barnes has served on Scientific Advisory Boards of AstraZeneca, Boehringer-Ingelheim, Chiesi, Daiichi-Sankyo, GlaxoSmithKline, Glenmark, Johnson & Johnson, Merck, Novartis, Takeda, Pfizer, Prosonix, RespiVert, Sun Pharmaceuticals, Teva and Zambon and has received research funding from Aquinox Pharmaceuticals, AstraZeneca, Boehringer-Ingelheim, Chiesi, Daiichi-Sankyo, GSK, Novartis, Takeda, Pfizer, Sun Pharmaceuticals. He is also a cofounder of RespiVert (now part of Johnson & Johnson), which has discovered novel inhaled anti-inflammatory treatments for asthma and COPD.
S. Collarini is a full time employee at Chiesi Farmaceutici SpA.
B. Leaker has received research funding from AstraZeneca, Chiesi and Pfizer.
D. Singh has received sponsorship to attend international meetings, honoraria for lecturing or attending advisory boards and research grants from various pharmaceutical companies including Almirall, AstraZeneca, Boehringer-Ingelheim, Chiesi, Genentech, GlaxoSmithKline, Glenmark, Merck, NAPP, Novartis, Pfizer, RespiVert, Skypharma, Takeda, Teva, Therevance and Verona.
M. Boyce has no conflicts of interest.

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Appendix A. Supplementary data

Supplementary data related to this article can be found at http://dx.doi.org/10.1016/j.pupt.2016.06.011.

References