Effect of formoterol, tiotropium, and their combination in patients with acute exacerbation of chronic obstructive pulmonary disease: A pilot study

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Summary The aim of our study was to evaluate the pharmacodynamic effects of 1-day treatment with formoterol, tiotropium and their combination in patients with acute exacerbation of chronic obstructive pulmonary disease (AECOPD). Twenty-one (19 males, mean age 72 ± 8 years, mean FEV\textsubscript{1} 38 ± 14% of predicted values) patients with mild to moderate AECOPD were enrolled. Patients received formoterol (12 \textmu g deliver via Modulite\textsuperscript{R} b.i.d.), tiotropium (18 \textmu g dry powder capsules delivered via HandiHaler\textsuperscript{R} once daily), and their combination, in randomized sequence. Serial measurements of FEV\textsubscript{1}, FVC, IC, SpO\textsubscript{2} and HR were performed over 24 h. Formoterol, tiotropium, and their combination significantly improved the area under curves (AUCs) for FEV\textsubscript{1}, FVC and IC over 12 and 24 h. The mean FEV\textsubscript{1}, FVC and IC AUC\textsubscript{0–12 h} and AUC\textsubscript{0–24 h} after formoterol and tiotropium combination were significantly higher than formoterol and tiotropium alone, whereas the differences between the two single drugs were not statistically significant. Formoterol, either alone or in combination with tiotropium, elicited a significantly faster onset of action, and combination elicited a greater maximum bronchodilation than both single drugs in terms of FEV\textsubscript{1} and FVC. After 24 h the bronchodilating effect of the three treatments disappeared, with the exception of the combination on FEV\textsubscript{1}. The results of this study have documented that, although the time course of the effects of evaluated drugs differs significantly from that in stable COPD, with a shorter bronchodilation...
Introduction

Acute exacerbations of chronic obstructive pulmonary disease (AECOPD) are associated with symptoms that include increased dyspnea, cough, and sputum volume, as well as changes in sputum colour. Symptoms are of fundamental importance and are the primary concern of the patient; it is generally a change in symptoms that prompts contact with healthcare professionals. Consequently, one of the most important goals of treatment of AECOPD is the fast resolution of symptoms.2

COPD exacerbations have also been associated with decrements in pulmonary function.1 Since in most patients with AECOPD, there appears to be a component of the worsened airflow obstruction that is particularly responsive to bronchodilators,3 which can lead to a symptoms reduction by affecting hyperinflation,4 bronchodilators are considered central in the symptomatic control of AECOPD.2,3 In effect, patients presenting with AECOPD should be given short-acting inhaled bronchodilators at high doses.5

In the acute exacerbation, there is little evidence conclusively establishing that any specific agent has superior efficacy as compared to others. However, there may be some advantages to the longer-acting agents in that there will be prolonged bronchodilation. In fact, the use of long-acting β2-agonists has recently been suggested as a potential option in the therapy of AECOPD,6–9 although these agents are currently not approved for use in this pathologic condition. In particular, formoterol has been proposed for the symptomatic treatment of AECOPD,10–11 since its onset of action is similar to that of salbutamol in patients with COPD.12,13

There is enough evidence in the literature that combination therapy with β2-adrenoceptor agonists and anticholinergics is effective and has a good safety profile.5 Unfortunately, to our best knowledge, no study has still investigated the impact of a combination therapy with formoterol and an anticholinergic agent in AECOPD, although there is documentation that in patients with stable COPD, treatment with a combination of formoterol and ipratropium bromide is more effective than a combination of salbutamol and ipratropium.14

The introduction into the market of tiotropium bromide, a long-acting anticholinergic bronchodilator that maintains bronchodilation for at least 24 h, allowing once-daily administration,15 and induces greater bronchodilator effect than ipratropium bromide,16 has offered a new drug able to improve dynamic lung hyperinflation and exertional dyspnea.17,18 The documentation that tiotropium alone or in combination with once-daily formoterol improved mean trough and average FEV1 and trough FVC values from baseline,19 and this even to a greater extent than twice-daily formoterol,20 suggests the need for also exploring the effect of a combination of formoterol and tiotropium in the therapy of AECOPD.

The aim of our study was, therefore, to evaluate the pharmacodynamic effects of 1 day treatment of formoterol, tiotropium and their combination in patients with AECOPD. Considering that conventional chlorofluorocarbon (CFC)-containing pressurized metered-dose inhalers (pMDIs) generate coarse, fast-moving clouds which can impede the optimal deposition of drug in the lung, a further problem in patients suffering from AECOPD, in this study we used a formoterol Modulite® solution formulation. In fact, the Modulite® inhaler, which has been developed to provide stable and uniform dose delivery using hydrofluoroalkane (HFA)—instead of CFC-based formulations, has been designed to improve the characteristics of the cloud emitted from the device to make the coordination of inspiration with drug delivery easier for the patient,21 and this is extremely convenient for patients with an AECOPD.

Material and methods

Subjects

Twenty-one (19 males, mean age 72 ± 8, range 58–86 years, mean smoke history 54 ± 26 pack-years, mean body mass index 27.0 ± 5.1 kg/m²) patients with mild to moderate AECOPD22 were enrolled (Table 1). Mean baseline FEV1 was 0.99 l (95% CI: 0.77–1.21, 38 ± 14% of predicted values) and mean forced vital capacity (FVC) was 1.83 l (95% CI: 1.48–2.18, 56 ± 19% of predicted values). All patients fulfilled the criteria proposed by GOLD guidelines for COPD5; they had cough, sputum production, or dyspnea, and/or a history of exposure to risk factors for the disease. The
Diagnosis was confirmed by spirometry (post-bronchodilator FEV1 < 80% of the predicted value, and FEV1/FVC < 70%). Patients with allergic rhinitis, atopy, positive skin test, or with a total blood eosinophil count over 400 /µL were excluded. Patients’ domiciliary treatment consisted in long acting β2-agonists (formoterol 12 µg b.i.d. and salmeterol 50 µg b.i.d. in 12 and 9 patients, respectively), anticholinergic (oxitropium bromide 200 µg b.i.d. in 18 patients) and inhalatory steroids (budesonide 400 µg b.i.d. and fluticasone 250–500 µg b.i.d. in 12 and 9 patients, respectively). No patient was in treatment with theophylline. Acute exacerbations of COPD, according to Anthonisen et al.,1 were defined by the presence of almost two of three of the following symptoms: increase dyspnoea, increased volume of sputum and purulence of sputum. The study was carried out according to the rules of the declaration of Helsinki. It was approved by the local ethical committee. All patients provided written informed consent to participate in the study.

Protocol

This was a double-blind, double-dummy, cross-over, randomized study. Patients received formoterol (12 µg deliver via Modulite® b.i.d.), tiotropium (18 µg dry powder capsules delivered via HandiHaler® once daily), and their combination, in randomized sequence on days 1, 3 and 5. During days 2 and 4 and patients were treated with salbutamol (last assumption almost 8 h before the study drugs inhalation). The washout period between the test days was at least 48 h in order to reduce the carry-over effect of tiotropium that has been described in patients with stable COPD.23 No other oral bronchodilators were allowed during the study. All patients were treated with systemic corticosteroids (40 mg of prednisolone or equivalent) and antibiotics (ciprofloxacin, levofloxacin, amoxicillin-clavulanate ± aminoglycosides) according to treating doctor. Consumption of cola drinks, coffee, tea, and smoking prior and during the investigation were avoided.

Physiological measurements

On test days, measurements of FEV1, FVC, inspiratory capacity (IC) (Elite DL, MedGraphics®, St Paul, MN, USA), oxygen saturation (SpO2) (by pulse oximetry Nonin Medical, Inc. Plymouth, MN, USA) and heart rate (HR) were recorded just before dosing and 30, 60, 120, 180, 240, 360, 480, 600, 720, and 1440 min after the study drug administration.
Statistical analysis

This was a pilot study and the first known comparison of formoterol, tiotropium, and their combination in patients with AECOPD. In view of the lack of previous experiences, no statistical hypotheses were drawn and consequently no formal sample size calculation was made. The use of pilot study such as the current study in clinical research is a well-established scientific procedure and only through the use of a pilot study can statisticians clarify data distributions and determine appropriate sample sizes for full-scale clinical trials.

According to the number of enrolled patients, a Kolmogorov–Smirnov test was performed before the data analysis in order to examine the data distribution. This test documented that the entire evaluated variable were distributed in a normal way, and then we used parametric tests for data management.

Comparison of baseline characteristics among the 3 days and the treatments were performed by ANOVA analysis for repeated measurements, while post-hoc analysis was performed by Bonferroni-test. As an expression of the total effect of each treatment, the areas under the FEV1, FVC and IC time responses curves (AUCs) were calculated for each patient by means of the trapezoidal rule. Comparison of AUCs and maximum response values among the three treatments were performed as before described. In the time course of bronchodilating effect, the comparisons vs. baseline values were performed by paired t-test. Data are presented as mean± standard deviations (sd) in the text with 95% confidence interval, while in figures bars represent standard error (se). A probability level <0.05 (P<0.05) was considered significant for all tests. Data were analysed by Statistical Package for the Social Science (version 13.0, SPSS, Chicago, USA).

Results

All the 21 patients completed the study. There were no significant differences among the baseline FEV1, FVC and IC values in the 3 days of treatment (P = 0.218, 0.325, and 0.570, respectively) and between the three treatments (P = 0.338, 0.265, and 0.680, respectively).

FEV1, FVC and IC areas under the curves

Formoterol, tiotropium, and their combination significantly (P<0.01) improved the AUCs for FEV1, FVC and IC over 12 and 24 h (Fig. 1). The mean FEV1, FVC and IC AUC0–12 h and AUC0–24 h after formoterol and tiotropium combination were significantly higher than formoterol and tiotropium alone (all, P<0.01), whereas the differences between the two single drugs were not statistically significant.

![Graphs showing FEV1, FVC, and IC AUCs](image-url)

**Figure 1** Mean changes (±se) from pre-dosing value on each of the treatment days in FEV1 (2a), FVC (2b) and IC (2c) area under curve (AUC) during the first 12 h (AUC 0–12 h) and 24 h (AUC 0–24 h). Formoterol (12 µg deliver via Modulite® b.i.d.), tiotropium (18 µg dry powder capsules delivered via HandiHaler® once daily), and combination (formoterol+tiotropium).
Time course of bronchodilating effect

The actual data of FEV$_1$, FVC and IC before and after the three treatments are shown in Fig. 2. The three therapies elicited a different bronchodilation profile, with a faster onset of action for formoterol alone or in combination with tiotropium. At 30 min, the combination of formoterol and tiotropium induced a significant higher improvement in FEV$_1$ and FVC than those obtained after single treatment administration ($P<0.01$ for all comparisons). The difference between formoterol and tiotropium resulted statistically significant ($P<0.01$). At the same time, the combination provided a higher IC than tiotropium ($P<0.05$) but not than formoterol. At 12 h, the mean increase in FEV$_1$ form pre-dosing value were 0.019 l (95% CI: -0.026–0.063; not significant) for formoterol, 0.097 l (95% CI: 0.033–0.161; $P<0.01$) for tiotropium and 0.118 l (95% CI: 0.055–0.182; $P<0.01$) for the combination. According to IC and FVC values, at 12 h the bronchodilation effect of the two single drugs disappeared, whereas the combination elicited a persistent bronchodilation ($P<0.05$). Twelve hours after the inhalation of formoterol associated to tiotropium the mean FVC increase was 0.150 l (95% CI: 0.003–0.296; $P<0.05$) and in IC was 0.135 l (95% CI: 0.009–0.261; $P<0.05$). At this time the differences among treatments were significant, with an higher mean FEV$_1$ for the tiotropium and the combination vs. formoterol ($P<0.01$), a significantly higher mean FVC for the combination vs. the single drugs ($P<0.01$) and an higher mean IC for the combination vs. formoterol ($P<0.05$) but not vs. tiotropium. After 24 h the bronchodilating effect of the three treatments disappeared (Fig. 2), with the exception of the combination on FEV$_1$ (increase of 0.039 l—95% CI 0.005–0.073; $P<0.05$). At this time, post-hoc analysis found a significantly higher mean FEV$_1$ after the inhalation of formoterol and tiotropium combination than after the two single drugs ($P<0.05$) and a higher mean FVC after the combination than after formoterol administration ($P<0.05$).

Maximum response

The mean maximum increase in FEV$_1$ from pre-dosing value on each of the dosing days were 0.222 l (95% CI: 0.139–0.305) for formoterol, 0.150 l (95% CI: 0.089–0.210) for tiotropium and 0.327 l (95% CI: 0.236–0.418) for the combination and occurred 2 h after formoterol and combination and 3 h after tiotropium inhalation. All comparisons between drugs resulted significant (combination vs. both

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**Figure 2** Mean changes in FEV$_1$ (1a), FVC (1b), IC (1c) and SpO$_2$ (1d) from pre-dosing value on each of the treatment days up to 24 h. Formoterol (12 µg deliver via Modulite® b.i.d.), tiotropium (18 µg dry powder capsules delivered via HandiHaler® once daily), and combination (formoterol+tiotropium). *$P<0.01$; †$P<0.05$. 

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The mean maximum increase in FVC from pre-dosing value on each dosing day were 0.272 l (95% CI: 0.157–0.387) for formoterol, 0.218 l (95% CI: 0.132–0.303) for tiotropium and 0.428 l (95% CI: 0.270–0.586) for the combination and occurred 1 h after formoterol and 2 h after inhalation of tiotropium and the combination. The differences between combination and both the two single drugs were significant (post-hoc \( P < 0.05 \)).

The mean maximum increase in IC from pre-dosing value on each of the dosing days were 0.243 l (95% CI: 0.103–0.384) for formoterol, 0.188 l (95% CI: 0.058–0.317) for tiotropium and 0.366 l (95% CI: 0.214–0.518) for the combination and occurred 2 h after formoterol and combination and 3 h after inhalation of tiotropium. The difference between treatments was border-line significant (ANOVA \( P = 0.053 \)).

**Pulse rate and oxygen saturation**

The actual data of \( \text{SpO}_2 \) before and after the three treatments are shown in Fig. 2. There were no significant differences between the baseline oxygen saturation (\( \text{SpO}_2 \)) in the 3 days (\( P = 0.128 \)) and in the three treatment groups (\( P = 0.124 \)). The mean \( \text{SpO}_2 \) AUC\(_{0–24\text{h}}\) was significantly higher after the inhalation of the combination (2275 h.l, CI 95%: 2251–2299) than after the inhalation of both formoterol (mean 2271 h.l, CI 95%: 2247–2296) and tiotropium (mean 2270 h.l, CI 95%: 2245–2295) alone (\( P < 0.05 \) for both the comparisons). No statistically significant modifications from baseline and between the three treatments in HR were found.

**Discussion**

The aim of the present study was to investigate the pharmacodynamic effects of 1 day treatments with formoterol delivered by Modulite\(^{16}\), tiotropium and their combination in patients with AECOPD. The results documented that the combination of formoterol and tiotropium induced a greater overall bronchodilation, as expressed by AUC both after 12 than 24 h, than the single drugs. As expected, formoterol elicited a fast onset of action that, intriguingly, was enhanced by the concomitant inhalation of tiotropium. In our opinion, this finding is important because one of the goals of AECOPD management is the prompt resolution of symptoms\(^2\) that can also be achieved by a rapid bronchodilation. Interestingly, the maximum bronchodilation obtained by the combination was higher than that recorded after the administration of the single drugs. On the other hand, the bronchodilator effect of formoterol disappeared after 12 h and that of tiotropium after 24 h. This trend was in accord with the documentation that the duration of the bronchodilator effect of short acting, inhaled \( \beta_2 \) agonists is decreased in COPD exacerbation.\(^{24}\) The speculative explanation of the shorter bronchodilation that we have found may be the increase of bronchial inflammation, which is present during exacerbation.\(^{25,26}\) It is a possibility that the inflammation may modify the pharmacokinetic and pharmacodynamic properties of inhalatory drugs, although we must emphasize that the amount of bronchodilation we observed, expressed as maximum response, did not differ significantly by that found in patients with stable COPD.\(^{19}\) In any case, the combination of formoterol and tiotropium allowed a sustained improvement in FEV\(_1\) up to 24 h. It must be highlighted that the effect of treatments on \( \text{SpO}_2 \), although statistically significant, is not clinically relevant.

In this study, we examined the bronchodilation profile of the single treatments by evaluating not only FEV\(_1\) and FVC, but also IC because dyspnea better correlates with changes in hyperinflation parameters, such as IC\(^27\) and functional residual capacity.\(^8\) Although all the explored spirometric parameters had a similar trend over the time (Fig. 2), the differences between the three treatments were less evident when changes in IC were considered. It is likely that, although a fast improvement in IC can be observed after the administration of tiotropium,\(^{28}\) a maintenance treatment with this agent is required to achieve maximal reduction in hyperinflation.\(^{16}\)

Adverse cardiovascular effects such as heart-rate changes, and ECG tracings may occur during treatment with \( \beta_2 \)-agonists, and the use of a long-acting molecule may be less favourable than the short-acting ones.\(^{29}\) However, several studies on patients with AECOPD and often older than 65 years have been unable to demonstrate a significant impact of a higher that customary dose of formoterol and salmeterol both on HR and \( \text{SpO}_2. \)\(^{10}\) According to our previous studies, we did not find significant changes in heart rate and \( \text{SpO}_2, \) although recently we have documented a little but significant reduction of PaO\(_2\) after formoterol administration,\(^{30}\) but it is well know that \( \text{SpO}_2 \) is a less sensitive parameter than PaO\(_2\).

In the present study, all patients suffering from mild to moderate AECOPD were treated with systemic corticosteroids and antibiotics, as...
suggested by international guidelines. Short courses of systemic corticosteroids in AECOPD have been shown to improve spirometric outcomes. In any case, we cannot exclude that a synergistic interaction between corticosteroids and long-acting bronchodilators, with the resulting synergetic effect being greater than the sum of responses achieved from each drug alone, might justify the findings of the present study. This type of synergetic effect has only been documented with corticosteroids and long-acting $\beta_2$-agonists. and, in effect, there is documentation that the addition of a corticosteroid influences the fast onset of action of formoterol. However, in addition to the up-regulation of $\beta_2$-adrenergic receptors and anti-inflammatory effects by corticosteroids, part of the beneficial effect of corticosteroids in AECOPD therapy may include a reduction in muscarinic receptor expression in airway smooth muscle, allowing for easier muscle relaxation by $\beta_1$-adrenergic agonists. In fact, it has been shown that, at least in dogs, a treatment with methylprednisolone led to a decreased expression of both $M_2$ and $M_3$ muscarinic receptors in airway smooth muscle. More recently, it has also been documented that dexamethasone decreases airway responsiveness to vagal stimulation via two mechanisms: increased $M_2$ receptor function that results in decreased acetylcholine release, and increased degradation of acetylcholine by cholinesterases.

It must be emphasized that our study has a number of limitations. First of all, we used a crossover protocol which can be controversial in patients during AECOPD. In effect, spontaneous changes of the bronchial obstruction level due to the systemic therapy might have introduced a bias in the comparison of day 1 and 5. We chose a crossover design because it allows paired comparisons of the results which are characterized by an higher statistical power than unpaired comparisons. This statistical approach is convenient for a pilot study with a low number of enrolled patients. Nonetheless, we believe that drugs randomization faced this possible bias and, moreover, our data analysis did not show basal differences among the three test days and among the three drugs. In any case, previous studies demonstrated a very slow recovery of pulmonary function after exacerbation, with a considerable number of patients that had not recovered their peak-expiratory flow (PEF) to baseline after 35 days. However, for a large scale study, a parallel groups design appears to be more appropriate. Secondly, the failure to show a statistically significant difference between treatments was likely associated with an insufficient statistical power in the study. We believe that there was a possibility of a type II error, which supported the lack of significance that we have repeatedly observed. This is unfortunately, one limit of the pilot studies. Nonetheless, researchers may start with "qualitative data collection and analysis on a relatively unexplored topic, using the results to design a subsequent quantitative phase of the study". Thus, pilot studies are conducted for a range of different reasons such as establishing whether the sampling frame and technique are effective, identifying logistical problems which might occur using proposed methods, estimating variability in outcomes to help determining sample size, and collecting preliminary data. Our study has clearly documented that the use of a combined therapy with formoterol and tiotropium is a possibility that is worthy of further investigation.

In conclusion, the results of this study have documented that, although the time course of the effects of evaluated drugs differs significantly from that in stable COPD, with a shorter bronchodilation both for tiotropium and formoterol, these two long-acting bronchodilators appear to also be complementary in mild to moderate AECOPD. In fact, whereas on the one hand formoterol ensures a fast onset of action, which is improved by the concomitant inhalation of tiotropium, on the other hand tiotropium adds prolonged and sustained bronchodilation that is enhanced by the association of formoterol. This finding let us to suggest a larger evaluation of this combination in the broncholytic treatment of AECOPD.

References

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