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An oral selective M3 cholinergic receptor antagonist in COPD

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ABSTRACT: Cholinergic antagonists have been used since the early 1900s as bronchodilators for chronic obstructive pulmonary disease (COPD). The present study investigated whether an oral muscarinic M3-selective anticholinergic agent (OrM3) would provide an improved therapeutic advantage compared with an inhaled anticholinergic agent in patients with COPD.

A 6-week, multicentre, randomised, placebo- and active-controlled, parallel-group study was performed at 56 sites in the USA. In total, 412 male and female patients (aged 35–86 yrs) with a clinical history consistent with COPD were randomised to receive OrM3 0.5, 2, 3 or 4 mg orally once daily, ipratropium bromide 36 μg by inhalation four times daily or placebo.

OrM3 demonstrated a significant dose-related improvement in serial forced expiratory volume in one second and a trend for dose-related improvement in patient-reported symptoms compared with placebo. However, at a dose that provided efficacy less than that of ipratropium, the incidence of dose-related, mechanism-based side-effects for OrM3 exceeded those observed for ipratropium.

In patients with chronic obstructive pulmonary disease, the oral M3-selective agent did not offer a therapeutic advantage over inhaled ipratropium. These results do not support the hypothesis that high selectivity for muscarinic M3 receptors over airway neuronal M2 receptors will represent a more effective therapy than current inhaled anticholinergics in obstructive airway disease.

KEYWORDS: Anticholinergics, antimuscarinic agents, bronchodilators, chronic obstructive pulmonary disease, ipratropium bromide, muscarinic receptors

Chronic obstructive pulmonary disease (COPD) is the fourth leading cause of chronic morbidity and mortality in the USA [1]. A survey conducted in 2000 estimated that physician-diagnosed COPD affected ~10 million people in the USA, and 24 million adults had evidence of airflow limitation [2]. The incidence of COPD is rising worldwide, and the World Health Organization expects this disease to be the fifth most prevalent disease and the third most common cause of death by 2020 [3].

Cigarette smoking plays a key role in the development of COPD in the majority of patients. Smoking cessation is the only intervention that has been proven to modify the natural clinical course of COPD [4]. Although aggressive anti-smoking programmes, pharmacotherapy and counselling have improved patients' adherence to smoking abstinence [5], many individuals are either unable or unwilling to quit smoking, and many who do quit eventually relapse.

Current pharmacological treatments for COPD do not slow the rate of decline in lung function but can improve the health status of patients [4, 6]. Bronchodilators, including short- and long-acting β-adrenergic agonists and muscarinic cholinergic antagonists (anticholinergics), are the mainstays of therapy. With regard to the latter, three muscarinic cholinergic receptors (M1, M2 and M3) have relevant physiological roles in the human airways. The M3 subtype is expressed on airway smooth muscle and in salivary glands and is believed to mediate bronchoconstriction *via* parasympathetic nerve signal transduction [7, 8]. In contrast, prejunctional M2 receptors are expressed in nerves innervating the heart and lungs and function as negative-feedback regulators of parasympathetic signalling; inhibition of these receptors is likely to increase the risk of tachycardia and bronchoconstriction [9].

Anticholinergic agents, such as ipratropium bromide (Atrovent®; Boehringer Ingelheim, Ridgefield, CT, USA) and tiotropium bromide (Spiriva®; Boehringer Ingelheim/Pfizer, New York, NY, USA), administered by the inhalation route, have demonstrated efficacy as bronchodilators in COPD [10, 11]. Both agents are functionally selective for muscarinic M1 and M3 receptor

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subtypes and disassociate quickly from M2 receptors [11]. It has been hypothesised that use of an M3-selective antagonist may reduce the incidence of side-effects, thus allowing higher exposures, increased efficacy, and an improved therapeutic margin. However, no large study to date has tested this hypothesis.

Several 4-acetamidopiperidine derivatives have been studied to develop a novel bronchodilator with a high level of selectivity for M3 receptors and thus a reduction in side-effects [9]. One such agent, oral M3 (OrM3), demonstrated a high degree of selectivity (120-fold) for the M3 receptor (K_i=4.2 nM) over M2 receptors (K_i=490 nM) [9]. It was hypothesised that this compound would also be selective for M3 receptors in the airways. Unlike currently available inhaled anticholinergic bronchodilators, OrM3 was formulated as an oral tablet, a potentially more convenient formulation, particularly for less compliant patients and those who have difficulty using aerosol therapy. Dosed orally, pharmacokinetic data demonstrated that OrM3 has a long half-life ($t_{1/2}$=14–20 h), which would potentially allow for a once-daily dosing regimen.

The purpose of the current study was to determine whether an oral M3-selective anticholinergic agent would provide an improved therapeutic margin over currently available inhaled anticholinergics. The present authors, therefore, compared the safety and efficacy of oral OrM3 with inhaled ipratropium bromide in patients with COPD.

MATERIALS AND METHODS

Patient selection

Male and female patients aged ⩾35 yrs with ⩾1-yr history of symptoms consistent with COPD and a smoking history of ⩾10 pack-yrs who were otherwise healthy were eligible to participate. To qualify, a minimum grade of 2 (indicative of shortness of breath when hurrying on level ground or up a slight hill) on the 5-point Medical Research Council dyspnoea scale was required [12].

Patients were excluded from participation if they had a history of asthma or glaucoma, a total peripheral blood eosinophil count >6% or >440·μL^{-1}, required on average <1 puff·day^{-1} of β-agonist, had a daytime room air oxygen saturation <90% or required oxygen therapy for use other than nocturnal use (maximum 2 L·min^{-1}), or had symptomatic prostatism. While withholding β-agonist for at least 6 h, patients were required to demonstrate a forced expiratory volume in one second (FEV_1) ⩾0.70 L and ⩽65% of predicted [13] and an FEV_1/forced vital capacity (FVC) ratio of ⩽70% on at least two occasions each during the pre-study visit and placebo run-in period. Patients were also required to demonstrate responsiveness to anticholinergic agents at least once during the pre-study and placebo run-in periods as evidenced by an increase in FEV_1 of ⩾10% 45–60 min after inhaled ipratropium bromide (36 μg).

Patients were allowed to take concomitant COPD therapy, including inhaled short-acting β-agonist on an ''as needed'' basis; inhaled or oral corticosteroids (inhaled beclomethasone ⩽2,000 μg·day^{-1}, inhaled fluticasone ⩽1,000 μg·day^{-1}, oral prednisone ⩽10 mg·day^{-1}, or equivalent) at stable doses beginning ⩾4 weeks before the pre-study visit; and oral short-acting theophylline (twice daily formulations only), at a stable dose, beginning ⩾5 days before the pre-study visit.

Study design

This was a randomised, multicentre, double-blind, parallel-group, dose-ranging study conducted at 56 outpatient centres in the USA. Written informed consent, approved by the respective institutional review boards, was obtained for each patient at or before the pre-study visit. Initially, 412 patients entered the study, beginning with a 2-week, single-blind, placebo run-in period (period 1).

Upon completion of the single-blind placebo run-in period, patients entered period 2 and were allocated to one of six double-blind treatments using a computer-generated random allocation schedule: OrM3 at 4.0 (n=67), 3.0 (n=69), 2.0 (n=73), or 0.5 mg (n=72) once daily in the morning; ipratropium bromide 36 μg four times daily (standard inhaled dose; n=63); or placebo (n=68; fig. 1).

Period 2 was followed by a 2-week, double-blind, placebo-controlled, treatment/washout period (period 3), during which each of the four OrM3 arms and the ipratropium arm were split in a 2:1 ratio according to the original allocation schedule. One third of each arm was placed on placebo for the duration of period 3, and two thirds continued on the treatment of period 2. These three periods were considered the base study. On completion of period 3, all remaining patients that provided informed consent entered period 4, a 16-week double-blind, safety extension study.

Patients were not aware that the study consisted of different periods and were not told when they were entering the treatment period. Exact-matching placebos for both the oral and inhaled anticholinergic agents were manufactured by the sponsor and distributed in a double-dummy fashion. Additionally, all patients were supplied with a salbutamol inhaler by the investigator to be used on an ''as needed'' basis.

Patients were scheduled to return to the clinic every 2 weeks during the study for assessment of pulmonary function and adverse experiences.

Pulmonary function testing

Pulmonary function testing was performed using a standard spirometer (Puritan Bennett PB100/PB110; Nellcor, Kansas City, KS, USA) according to the standards of the American Thoracic Society [13]. Spirometric manoeuvres were conducted in triplicate, and the results of the largest FEV_1 and FVC were recorded. Predicted normal values for patients were based on age, height and sex [13]. To ensure standardised conditions on all pulmonary function test days, patients were required to withhold theophylline and short-acting antihistamines ⩾24 h and β-agonist and oral/inhaled corticosteroids ⩾6 h before each visit.

Serial spirometry measurements were performed at the clinic before dosing and 1, 2, 4, 6 and 10 h after dosing 2 weeks after initiation of active treatment in period 2. Patients took their second dose of study inhaler after the 10-h post-dose measurement, followed by the third dose in the evening; no additional study drug (tablets or inhaler) was given before completion of the 24-h serial spirometry. If β-agonist rescues

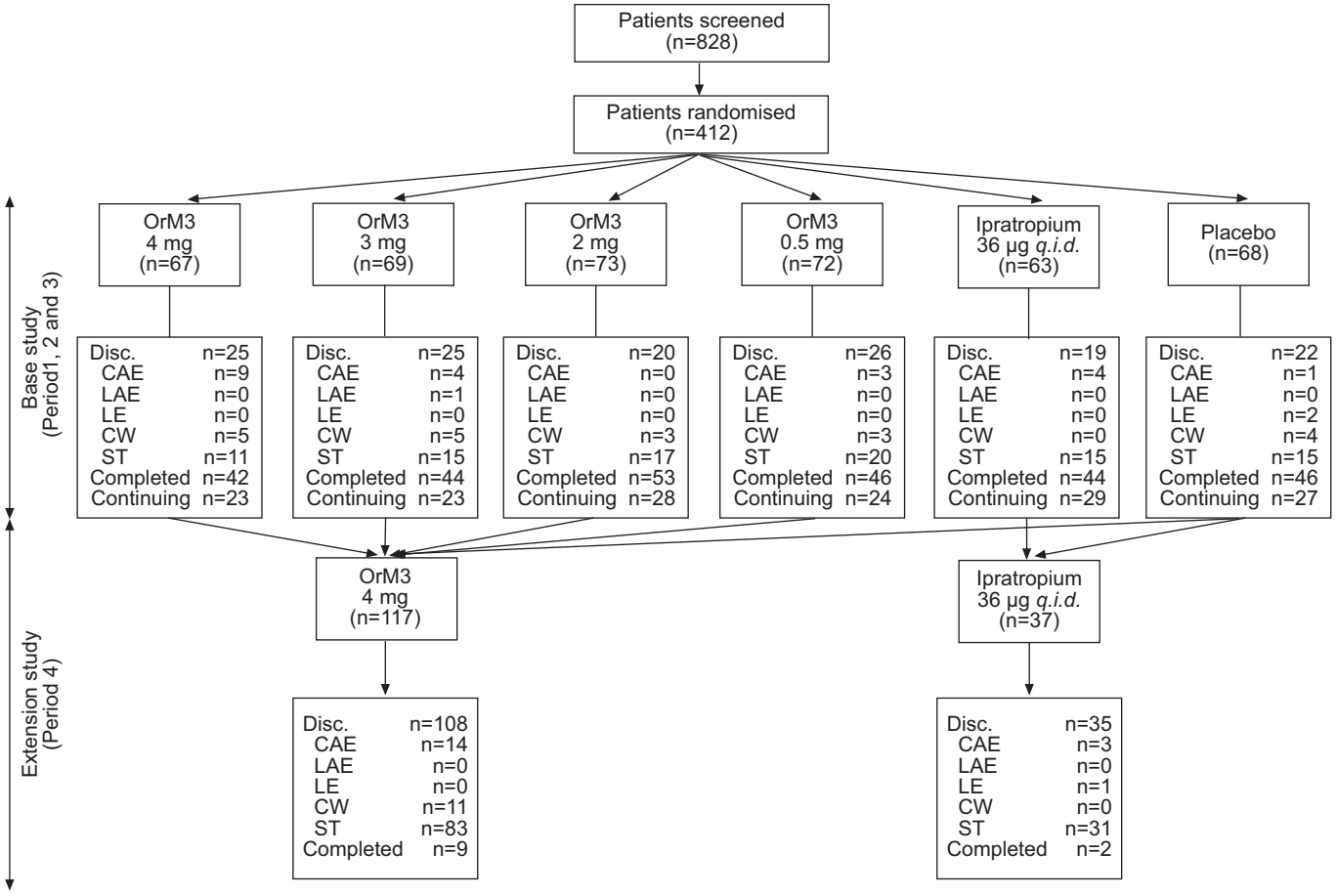

FIGURE 1. Study design for evaluation of an oral, M3-selective cholinergic receptor antagonist (OrM3) in patients with chronic obstructive pulmonary disease. Disc.: discontinues; CAE: clinical adverse experience; LAE: laboratory adverse experience; LE: lack of efficacy; CW: consent withdrawn; ST: site terminated.

were needed during the serial spirometric measurements, spirometry was attempted before the β-agonist rescue and again after 30 min. The spirometry data were electronically transmitted to a spirometry quality-control centre on a weekly basis for rigorous review of data quality and adherence to spirometry inclusion criteria [14].

In addition to the serial spirometry measurements above, baseline (trough) spirometry measurements were taken at visits two and three (period 1), and FEV_1 and FVC were measured between 06:00 h and 09:00 h at baseline and after 2, 4, 6 and 8 weeks of treatment.

Dyspnoea rating

Change in dyspnoea was assessed using the baseline dyspnoea index (BDI) completed at randomisation, and the transition dyspnoea index (TDI) completed after 2, 6 and 8 weeks treatment [15]. The focal score of the BDI was calculated as a sum of three domains: functional impairment, magnitude of task, and magnitude of effort. Total baseline score could range from 0 (severe dyspnoea) to 12 (no dyspnoea limitation). The TDI focal score was defined as a sum of the three domains using a scale of -9 (major deterioration) to +9 (major improvement).

Patient diary card

Patients recorded their COPD symptoms, morning and evening peak expiratory flow rates (PEFR), β-agonist use, and nocturnal awakenings due to COPD on a daily diary. The diary included six COPD symptom questions that focused on overall time having symptoms due to COPD, shortness of breath, cough, mucus production, difficulty in carring out routine activities (light), and difficulty in doing activities that require moderate-to-high physical movement. A 6-point scale was used to evaluate these patient-reported end-points (0=none of the time/none/no difficulty, to 5=all of the time/a very large or massive amount/so difficult couldn't do it at all).

Information on COPD exacerbations was also recorded in the patients' diaries. A COPD exacerbation was defined as worsening COPD symptoms requiring: a call to a doctor; visit to a doctor or an emergency room; hospital admission; or treatment with a corticosteroid and/or antibiotic.

Quality of life

The chronic respiratory questionnaire (CRQ), a COPD quality-of-life measure, is a 20-item questionnaire with four domains: dyspnoea, fatigue, emotional function, and the feeling of

mastery over the disease [16]. Questions in each domain were rated by the patients on a 7-point scale (1=poorest function to 7=optimal function). The CRQ was completed at the same visits as the BDI and TDI.

Global evaluations

Upon arriving at the clinic, all patients completed the global evaluation as the first procedure at the 6-week visit. Patients and physicians independently evaluated the change in the overall perception of the patient's COPD by selecting the most appropriate response using a 7-point Likert-type scale (7=very much better to 1=very much worse).

Safety and tolerability evaluations

Adverse experiences were recorded and monitored throughout all periods of the study. Patients underwent clinical evaluations, including vital signs, physical examinations, ophthalmic examinations, electrocardiograms, adverse experience monitoring and laboratory safety testing (complete blood count, serum chemistries and urinalysis), prior to randomisation and at designated visits throughout all four periods of the study. Final safety evaluations were conducted at the final scheduled visit for period 3 or 4 (extension) or at the discontinuation visit.

Statistical analyses

The primary efficacy analyses were based on an intention-to-treat approach, defined as a population of patients who had a baseline value and at least one treatment period measurement. Missing values were not imputed for any end-points. For FEV_1, the end-point for every patient was defined as the average of all measurements. For example, if a patient had only week 2 trough FEV_1 and then discontinued from the study, the average trough was set to be the week 2 value. Statistical analyses were based on two-tailed tests conducted at the 0.05 significance level; data are presented as least square means (95% confidence intervals), unless otherwise stated.

The primary efficacy end-point was the between-group comparison of mean serial FEV_1 assessed as the average of FEV_1 values measured over 24 h after 2 weeks of treatment, which was analysed using an ANCOVA model with treatment and study site as factors and baseline FEV_1 and ipratropium reversibility as covariates. A stepwise linear contrast test based on the ANCOVA model was used to examine the dose-response relationship for the 0.5-, 2-, 3- and 4-mg doses of OrM3 and provided for a more effective comparison of the doses *versus* placebo.

Specific between-group comparisons (*i.e.* among OrM3 doses, each OrM3 dose *versus* ipratropium, ipratropium *versus* placebo) were based on specific pairwise contrasts from the ANCOVA model above. Other efficacy end-points were analysed in a similar way, using the ANCOVA model and including treatment and study site as factors, and baseline (where applicable) as a covariate. In addition, global evaluations were separated into three categories (better, no change and worse) and analysed with a Cochran–Mantel–Haenszel test [17]. A *post hoc* analysis of the percentage of patients with at least one COPD exacerbation was performed. An interim analysis was performed to obtain preliminary safety and efficacy information on OrM3.

A sample size of 85 patients per group was estimated to provide 80% power to detect (α=0.05, two-sided) a 0.094-L between-group difference in average FEV_1 values measured over 24 h after 2 weeks of treatment. This sample size also had 80% power to detect (at α=0.05, two-sided) a 7.8% between-group difference in the per cent change from baseline of pre-dose (trough) FEV_1.

RESULTS

Patients

A total of 828 patients were screened for the trial, with 412 randomised into the active treatment period (fig. 1). The most common reasons patients were not randomised included inability to meet pulmonary function criteria, use of excluded medications and history of symptomatic prostatism. It was aimed to have 85 patients in each treatment group. However, an interim analysis was conducted after 412 patients had been randomised, and the study was terminated earlier than planned due to the incidence of side-effects and a lack of clear superior efficacy for OrM3 compared with standard treatment (ipratropium).

Of the 412 patients who were randomised, 275 completed the base study (fig. 1). In total, 21 patients discontinued due to a clinical adverse experience (OrM3 4 mg, n=9; 3 mg, n=4; 0.5 mg, n=3; ipratropium, n=4; placebo, n=1) and one patient in the 3 mg OrM3 group discontinued due to a laboratory adverse experience (AE). In total, 22 patients withdrew consent (n=20) or discontinued due to lack of efficacy (n=2) and 93 patients discontinued when the study was terminated at various sites based on results from the interim analysis.

Of the 275 patients who completed the active treatment period, 154 continued into the extension period (OrM3 4 mg, n=117; ipratropium, n=37; fig. 1). The primary reasons for discontinuation prior to initiation of the extension period were termination of the site and withdrawal of consent by the patient. Of the 154 patients that continued into the extension study, 143 discontinued during that period, primarily due to termination of the study by the sponsor, withdrawal of consent, or clinical AEs; 11 patients completed all 16 weeks of the extension period (fig. 1).

Of the 412 patients randomised, 387 patients who completed $\geqslant$2 weeks of study therapy and had valid serial spirometry performed at week 2 were included in the primary end-point analyses. All other analyses, including the safety analyses, include data from all randomised patients up to the point at which enrolment was terminated.

Most allocated patients had severe-to-very severe COPD (Global Initiative for Chronic Lung Disease Stage III–IV) [18]; the mean$\pm$SD per cent predicted FEV_1 value at baseline was 40.8$\pm$14.2. Ipratropium reversibility was similar in all groups, with a mean$\pm$SD change in FEV_1 of 21$\pm$13% after 36 μg of ipratropium bromide. Based on the mean focal BDI scores, all groups were similar at baseline with moderate impairment due to their dyspnoea (table 1). All six treatment groups were similar with regard to demographics and other baseline characteristics (table 1).

TABLE 1 Patient baseline characteristics

Characteristics	OrM3				Placebo	Ipratropium
	4 mg	3 mg	2 mg	0.5 mg		
Subjects n	67	69	73	72	68	63
Age yrs	67 (46–86)	65 (46–81)	67 (43–81)	66 (37–83)	64 (43–80)	66 (39–85)
Male	52	50	60	51	57	60
Ethnic origin						
White	94	87	95	99	93	96
Black	3	9	4	1	7	2
Other	3	4	1	0	0	2
Duration of COPD yrs	6.9±5.8	7.5±7.2	6.5±5.6	7.1±6.8	5.9±5.1	6.7±6.3
Smoking history pack-yrs	63.9±31.6	62.9±41.2	60.5±30.5	59.3±38.9	63.8±40.6	56.2±27.5
FEV_1 L	1.1±0.5	1.1±0.4	1.2±0.5	1.3±0.5	1.2±0.5	1.4±0.6
FEV_1 % pred	40.2±15.8	37.8±12.9	38.9±13.4	44.2±13.4	40.4±14.4	43.3±15.2
FEV_1 % pred ipratropium reversibility	23.3±15.6	21.2±10.5	22.0±12.9	20.2±15.2	20.0±11.5	21.3±10.1
FEV_1/FVC	51±17	50±17	51±17	53±17	53±22	52±16
Morning PEFR $L \cdot min^{-1}$	250.7±86.7	231.3±71.9	263.4±97.8	276.4±56.2	254.3±85.9	273.3±105.6
Evening PEFR $L \cdot min^{-1}$	241.6±84.6	217.4±73.7	254.4±96.6	260.1±86.0	244.4±91.5	260.3±104.7
β-agonist use $puffs \cdot day^{-1}$	5.1±4.0	5.5±3.8	5.4±4.3	4.6±3.5	5.3±3.4	5.3±4.4
MRC score	3.3±0.9	3.3±0.8	3.0±0.9	3.0±0.9	2.9±0.9	2.9±1.0
BDI focal score	5.6±2.1	5.3±1.6	5.6±1.9	5.8±1.9	5.7±2.1	5.2±2.0
CRQ average of 4 domains	4.3±0.9	4.1±0.8	4.4±0.9	4.3±0.9	4.3±1.0	4.2±1.0
Daytime overall COPD symptoms score	2.1±0.8	2.2±0.8	2.0±0.8	2.1±0.8	2.1±0.8	2.2±0.9
Night-time awakenings	0.6±0.5	0.5±0.4	0.7±0.7	0.6±0.5	0.6±0.6	0.9±0.7

Data are presented as median (range), % and mean±SD, unless otherwise stated. OrM3: oral M3-selective cholinergic receptor antagonist; COPD: chronic obstructive pulmonary disease; FEV_1: forced expiratory volume in one second; % pred: per cent predicted; FVC: forced vital capacity; PEFR: peak expiratory flow rate; MRC: Medical Research Council; BDI: baseline dyspnoea index; CRQ: chronic respiratory questionnaire.

Pulmonary function

OrM3 demonstrated a dose-related improvement in the primary end-point of serial FEV_1 over 24 h after 2 weeks of treatment (table 2). Improvement in the average serial FEV_1 measurements for the 3- and 4-mg doses were statistically significant compared with placebo ($p=0.01$ and 0.018, respectively), whereas the 0.5- and 2-mg doses were not different from placebo (table 2). The peak mean change from baseline (peak effect within 2 h of treatment) in the 4 mg group was about two thirds of the effect observed in the ipratropium group. The effect of the 4 mg dose was still apparent 24 h post-dose (fig. 2); in contrast, for patients receiving ipratropium, FEV_1 had returned to baseline within 10 h post-dose. An exploratory analysis of FEV_1 area under the curve was consistent with the results of the average serial FEV_1 analyses (data not shown).

The average percentage change in trough (pre-dose) FEV_1 from baseline over the 6 weeks of treatment demonstrated a modest, albeit statistically significant, improvement in the 4 mg group compared with placebo (table 2). During the washout period, there was no evidence of rebound worsening following withdrawal of either drug (data not shown).

Other efficacy measurements

For dyspnoea assessment, there was no significant difference in TDI focal score in any treatment group compared with placebo over the 6 weeks of treatment (table 2; fig. 3) or for any of the functional domains of the TDI (functional impairment, magnitude of task, and magnitude of effort; data not shown). However, a dose-related trend for improvement with OrM3 was observed in the mean TDI scores (table 2).

There was a statistically significant increase in mean (95% confidence interval) morning PEFR in the 2 mg (11.07 (5.93–16.22), $p=0.017$), 3 mg (10.52 (4.93–16.11), $p=0.029$), and 4 mg (14.14 (8.66–19.62), $p=0.002$) OrM3 groups over 6 weeks of treatment compared with placebo (2.18 (-3.34–7.71)). Morning PEFR was not different between the placebo and ipratropium groups (fig. 4). Evening PEFR responses were significantly improved for the 4 mg OrM3 group only (10.29 (4.42–16.16), $p=0.041$) compared with placebo (1.89 (-4.04–7.81), fig. 4).

There was no significant difference in total daily β-agonist use for any of the treatment groups compared with placebo, although OrM3 at 2, 3 and 4 mg and ipratropium showed numerically less daily β-agonist use (table 2).

Based on daily diary scores, patients' overall COPD symptoms score decreased numerically in all active treatment groups, with the largest improvement occurring in the 4 mg OrM3 group (mean -0.15) compared with placebo (mean -0.01; $p=0.018$; table 2). For individual symptoms, daily dyspnoea

TABLE 2 End-points: primary study comparisons for patients with chronic obstructive pulmonary disease (COPD) treated with an oral selective M3 cholinergic receptor antagonist (OrM3)

End-point	OrM3				Placebo	Ipratropium
	4 mg	3 mg	2 mg	0.5 mg		
Primary						
FEV1 L over 24 h after 2 weeks of treatment	1.29 (1.25–1.33)*	1.29 (1.25–1.33)*	1.27 (1.23–1.30)	1.24 (1.20–1.28)	1.22 (1.18–1.26)	1.26 (1.22–1.30)
Secondary						
% change in trough from baseline FEV1#	5.11 (1.96–8.27)	3.58 (0.60–6.57)	1.51 (-1.35–4.37)	1.65 (-1.24–4.54)	-1.18 (-4.19–1.84)	-16.1 (-4.67–1.46)
TDI focal score#	1.51 (0.95–2.06)	1.46 (0.90–2.01)	1.22 (0.70–1.74)	0.98 (0.44–1.53)	1.04 (0.48–1.61)	1.09 (0.51–1.68)
β-agonist use puffs·day^{-1}#	-0.70 (1.24– -0.16)	-0.77 (-1.32– -0.22)	-0.56 (-1.08– -0.05)	-0.25 (-0.78–0.27)	-0.26 (-0.80–0.29)	-0.72 (-1.28– -0.16)
Exploratory						
Daytime overall COPD symptoms score#	-0.15 (-0.26– -0.05)	-0.09 (-0.2–0.01)	-0.12 (-0.22– -0.02)	-0.09 (-0.19–0.01)	-0.01 (-0.12–0.09)	-0.06 (-0.16–0.05)
Night-time awakenings#	-0.20 (-0.34– -0.07)	-0.09 (-0.22–0.05)	-0.05 (-0.18–0.07)	-0.21 (-0.33– -0.09)	-0.14 (-0.26– -0.02)	0.01 (-0.13–0.15)
COPD exacerbations¶	17.4	14.5	18.8	8.7	20.2	20.2
Quality of life score#						
Average of 4 domains	0.31 (0.16–0.46)	0.28 (0.14–0.43)	0.35 (0.22–0.49)	0.36 (0.21–0.50)	0.37 (0.22–0.52)	0.32 (0.17–0.47)
Dyspnoea	0.54 (0.32–0.77)	0.53 (0.39–0.75)	0.64 (0.43–0.85)	0.58 (0.37–0.80)	0.47 (0.25–0.70)	0.64 (0.41–0.87)
Fatigue	0.29 (0.10–0.47)	0.25 (0.07–0.44)	0.24 (0.06–0.41)	0.30 (0.12–0.48)	0.44 (0.25–0.62)	0.29 (0.10–0.47)
Emotional function	0.15 (-0.01–0.32)	0.10 (-0.06–0.27)	0.26 (0.10–0.42)	0.23 (0.07–0.40)	0.34 (0.17–0.51)	0.19 (0.01–0.36)
Mastery	0.19 (0.00–0.39)	0.22 (0.03–0.42)	0.29 (0.11–0.47)	0.26 (0.07–0.45)	0.20 (0.00–0.39)	0.14 (-0.06–0.34)

Data are presented as least square mean (95% confidence intervals) based on ANCOVA, or %. FEV1: forced expiratory volume in one second; TDI: transition dyspnoea index. #: results of treatment over 6 weeks; ¶: percentage of patients with more than or equal to one exacerbation during 8 weeks of treatment; *:$p<0.05$, significantly different from placebo.

scores significantly improved in the 4 mg OrM3 group only compared with placebo ($p=0.033$). There were no significant improvements in cough, mucus production and difficulty performing routine (light) or moderate-to-high physical activity in the OrM3 or ipratropium groups compared with placebo.

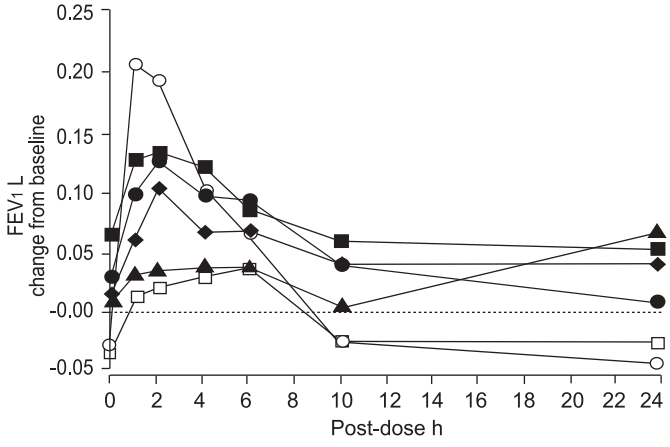

FIGURE 2. Average change in forced expiratory volume in one second (FEV1) from pre-randomisation baseline values, measured over 24 h following 2 weeks of treatment. The morning dose of study medication was taken after the first serial spirometric measurement (hour 0, pre-dose). The 3 and 4 mg oral M3-selective agent (OrM3) groups were significantly different from placebo ($p<0.05$). ■: OrM3 4 mg; ●: OrM3 3 mg; ♦: OrM3 2 mg; ▲: OrM3 0.5 mg; ○: ipratropium; □: placebo.

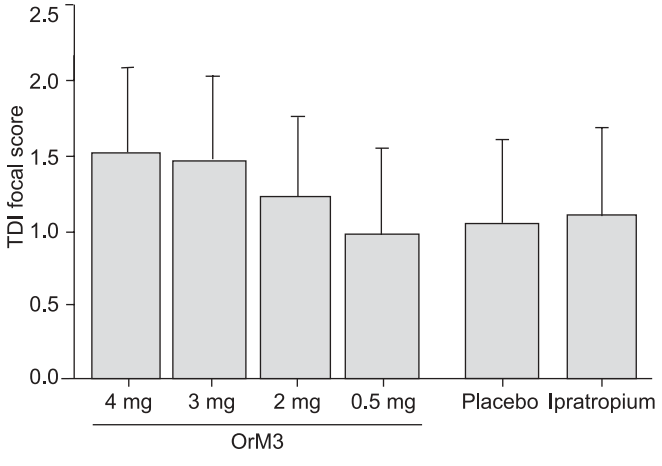

FIGURE 3. Average transition dyspnoea index (TDI) focal score over 6 weeks of treatment. Data are presented as mean and 95% confidence limits. There was no significant difference from placebo in any of the active treatment groups, although a dose-related trend, consistent with forced expiratory volume in one second results, can be seen. OrM3: oral M3-selective cholinergic receptor antagonist.

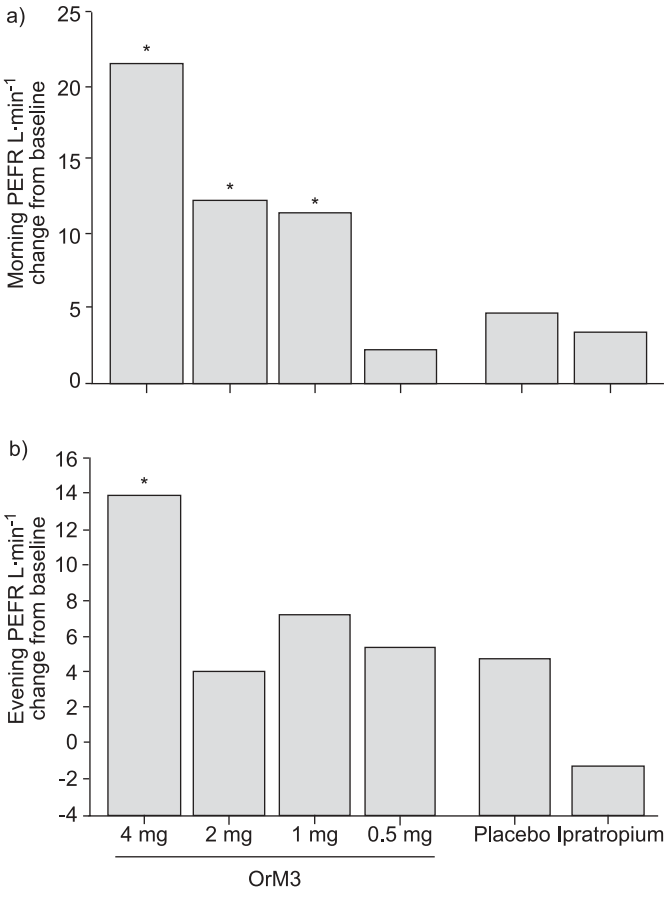

FIGURE 4. Mean change in a) morning and b) evening peak expiratory flow rate (PEFR) over 6 weeks of treatment. The 2, 3 and 4 mg oral M3-selective cholinergic receptor antagonist (OrM3) groups demonstrated a significant improvement compared with placebo in morning PEFR. The 4 mg OrM3 group also demonstrated a significant improvement compared with placebo in evening PEFR. *: $p \leqslant 0.05$.

Over the 6-week treatment period, there were no significant differences between the groups in nocturnal awakenings.

Approximately 17% of the patients experienced at least one COPD exacerbation over 8 weeks of treatment (table 2). Overall, treatment with OrM3 was associated with a slight reduction in the incidence of COPD exacerbations compared with either placebo or ipratropium, but a dose-related effect was not observed. Only two patients (both in the 4 mg OrM3 group) were hospitalised during the treatment period due to worsened COPD symptoms.

Neither active drug demonstrated any effect on overall quality of life (CRQ), individual domains of dyspnoea, fatigue, or mastery of the disease over the 6-week active treatment period compared with placebo (table 2). The emotional function domain significantly improved in the 3 mg OrM3 group compared with placebo ($p=0.043$), but was not significantly different for any other treatment group compared with placebo.

Patients' global evaluations at the end of the 6-week treatment period were significantly improved in the 4 mg OrM3 group (mean (95% confidence interval) difference *versus* placebo -0.49 (-0.93– -0.05), $p=0.029$). No differences from placebo were observed on the physicians' global evaluations for either OrM3 (mean difference for 4 mg *versus* placebo -0.17 (-0.54–0.20), $p=0.374$), or ipratropium (mean difference *versus* placebo -0.02 (-0.35–0.39), $p=0.921$).

Safety and tolerability

There were no serious drug-related adverse experiences in any treatment group in either the base or the extension study. Cardiovascular adverse events occurred in <5% of patients and no single cardiovascular adverse event occurred in more than one patient in any treatment group. Dose-related incidences of nonserious adverse experiences consistent with anticholinergic activity (*e.g.* dry mouth, dry eyes and throat, blurred vision, and constipation) in patients taking OrM3 were frequent during the base study (fig. 5). Dry mouth was the most commonly reported adverse experience occurring in patients taking OrM3, including 31 (46.3%) patients at 4 mg, 29 (42.0%) at 3 mg, 22 (30.1%) at 2 mg, and five (6.9%) at 0.5 mg. Dry mouth was reported for six patients taking ipratropium (9.5%). There was one incidence (1.5%) of dry mouth in the placebo group (fig. 5).

DISCUSSION

The primary goal of the present study was to test the hypothesis that a highly M3-selective anticholinergic agent administered orally would provide a superior therapeutic margin over that currently observed for inhaled anticholinergic therapies, either by improved efficacy and/or by improved safety and tolerability. Although inhaled therapies have demonstrated acceptable efficacy and safety/tolerability in COPD, numerous studies have demonstrated improved patient satisfaction and compliance with oral *versus* inhaled

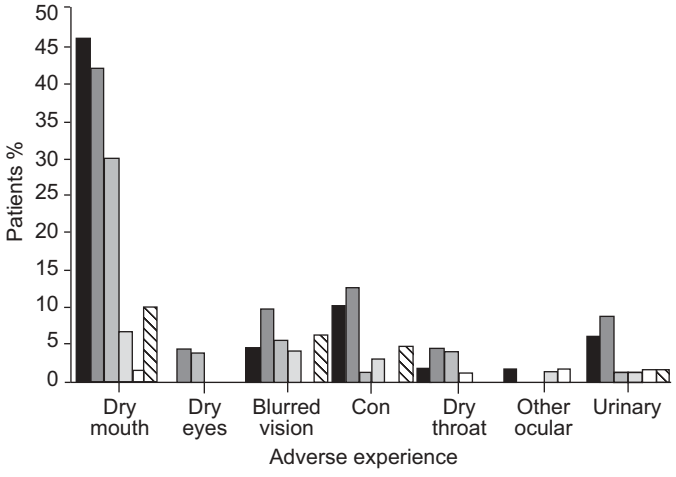

FIGURE 5. Percentage of patients with major anticholinergic adverse experiences (AEs) occurring during the base study (6-week treatment period plus 2-week washout period). Other ocular AEs included visual acuity decreased and visual disturbance. Urinary AEs included oliguria, urinary retention, urinary stream slowed and urination disorder (difficulty voiding). ■: oral M3-selective cholinergic receptor antagonist (OrM3) 4 mg (n=67); ■: OrM3 3 mg (n=69); ■: OrM3 2 mg (n=73); ■: OrM3 0.5 mg (n=72); □: placebo (n=68); ▨: ipratropium (n=63). Con: constipation.

medications [19]. Mechanism-based side-effects, such as dry mouth, tachycardia and visual disturbances, have limited the effectiveness of anticholinergic agents, and as a result, these drugs are currently delivered predominantly by the inhalational route to reduce systemic exposure. Since smooth muscle contraction is primarily mediated by M3 receptors expressed on the smooth muscle, it was speculated that an M3-selective antagonist might avoid some of the mechanism-based side-effects associated with less-selective antagonists, and thus achieve higher systemic exposures and potentially greater efficacy without worsening of side-effects compared with a nonselective antagonist. OrM3, an orally bioavailable, once-daily highly selective M3 anticholinergic agent, was ideally positioned to test this hypothesis.

Data from the current study suggest that OrM3 was efficacious in the treatment of COPD, with improvements noted for serial FEV_1, trough FEV_1, PEFR and patient global evaluations. However, oral OrM3 was inferior to inhaled ipratropium as a bronchodilator at the highest OrM3 dose tested (4 mg); the improvement in mean change from baseline in serial FEV_1 was less than that observed for ipratropium at 2 h post-dose (0.13 L 4 mg OrM3 *versus* 0.19 L ipratropium). The magnitude of the ipratropium response (peak FEV_1 change from pre-randomisation baseline ~208 mL after a single 36 μg dose) was consistent with that reported elsewhere [20], and thus, the failure to observe comparable efficacy with OrM3 in the current trial was unlikely due to patient selection or study design.

Mechanism-based side-effects, most prominently dry mouth, were higher in the 4 mg OrM3 treatment group than in the group treated with ipratropium. Thus, the improved M3-selectivity of OrM3 did not confer an improved therapeutic margin with regard to bronchodilator effects in patients with COPD. It is possible that administration of OrM3 by the inhaled route could have produced better efficacy and/or fewer side-effects; however, this was not the hypothesis of the study, and as such additional investigation would be needed to evaluate this possibility.

Whereas OrM3 was inferior to ipratropium as a bronchodilator, the positive efficacy data support the notion that the M3-cholinergic receptor is indeed the primary receptor mediating airway effects in humans. Conversely, the data also confirm that M3-receptor blockade is also primarily responsible for side-effects, such as dry mouth. This finding suggests that it will be quite difficult to identify systemically administered anticholinergic agents that are efficacious yet avoid significant dose-limiting, mechanism-based toxicities. For example, darifenacin, an oral M3-selective antagonist approved for treatment of urinary incontinence, has been reported to have dose-related incidences of dry mouth (13.2–31.3% of patients) in a clinical trial [21].

In conclusion, this proof-of-concept study demonstrates that selective antagonism of the M3 receptor causes an improvement in patients' airway function without the occurrence of M2 receptor-based side-effects, such as tachycardia. However, dose-limiting side-effects, such as dry mouth, presumably due to antagonism of M3 receptors in salivary glands, resulted in a reduced therapeutic margin relative to an inhaled anticholinergic agent. Thus, increased selectivity for the M3 cholinergic receptor is unlikely to allow development of oral anticholinergic drugs with improved therapeutic margins in chronic obstructive pulmonary disease.

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