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Local and systemic effects of inhaled AZD9164 compared with tiotropium in patients with COPD

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Summary
There is still a need for new agents which improve upon the therapeutic index of tiotropium, the current standard of care for many patients with chronic obstructive pulmonary disease (COPD). We examined in patients with COPD the efficacy of single doses of AZD9164, an M_3-selective muscarinic antagonist, to identify an appropriate dose-range for future studies.

COPD patients ($n = 28$) inhaled AZD9164 (100, 400 and 1200 μg), tiotropium (18 μg) and placebo at 5 study centre visits (Clinicaltrials.gov identifier NCT00939211). The effects of these test drugs on average (E_{av}), peak (E_{max}) and trough (E_{22-26}) forced expiratory volume in one second (FEV_1) were assessed, as were systemically-mediated effects and the safety and exposure of single doses of AZD9164.

AZD9164 100, 400 and 1200 μg caused increases in FEV_1 to peak effects of 12, 17 and 12% above baseline respectively, following an initial transient and dose-related fall in FEV_1 and associated increase in mild respiratory symptoms such as cough. Bronchodilation was maintained overnight, with minimal FEV_1 decline. AZD9164 400 and 1200 μg produced larger effects than tiotropium on E_{22-26} ($p < 0.05$; both doses) while AZD9164 400 μg also had larger effects on E_{max} ($p = 0.001$) and E_{av} ($p < 0.05$). There were no serious adverse events and statistically significant systemic effects were observed only with AZD9164 1200 μg.

AZD9164 may improve upon the therapeutic index of tiotropium, increasing the magnitude and duration of lung function improvements without increasing systemically-mediated adverse events. The initial bronchoconstrictor effect of AZD9164 requires further investigation.

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Introduction

Current guidelines for the treatment of chronic obstructive pulmonary disease (COPD) recommend the use of long-acting bronchodilators in patients who remain symptomatic with short-acting bronchodilators.[1] Tiotropium is the only inhaled long-acting muscarinic receptor antagonist currently available and has become the standard of care for many patients with COPD, being both well tolerated and efficacious at the recommended dose.[1,2]

Anticholinergic medications cause bronchodilation primarily by blocking muscarinic (M) 3 receptors. These receptors mediate bronchoconstriction via vagally derived acetylcholine on airway smooth muscle. M_1 receptors facilitate cholinergic transmission and thereby potentiate the effects of vagal bronchoconstriction. Antagonism of M_1 receptors should further increase the bronchodilator effect of M_3 antagonism.[2,3]

M_2 receptors at cholinergic nerve endings inhibit the release of acetylcholine and act as inhibitory feedback receptors.[2] Antagonism of prejunctional M_2 receptors therefore results in increased acetylcholine release in human airways which may overcome the blockade of M_3 receptors in the muscle.[2] However, M_2 receptors are also abundant on airway smooth muscle and contribute to muscle contraction by limiting adrenergic relaxation through inhibition of ß$_2$-adrenergic receptor-mediated increases in adenylate cyclase. Blocking of postjunctional M_2 receptors on airway smooth muscle may thus provide potential additional bronchodilation by removing the inhibitory effect they have on ß$_2$-agonist-induced relaxation.

The binding affinity of tiotropium and ipratropium has been reported to be similar for M_1, M_2, and M_3 receptors, but tiotropium dissociates >100 times more slowly than ipratropium from M_1 and M_3 receptors, whereas dissociation from M_2 receptors is more similar.[2,4] This suggests that tiotropium has a kinetic selectivity for M_1 and M_3 receptors over M_2 receptors. This kinetic selectivity is believed to be why tiotropium is a more effective and longer-acting bronchodilator than non-selective antagonists such as ipratropium.[2,4]

Studies have shown that there are important lung function benefits associated with tiotropium therapy that are maintained over periods up to 4 years, along with improvements in quality of life and a reduced risk of exacerbations and hospitalisations.[5–7] However, current licenced use of tiotropium, like ipratropium, is limited by potential adverse effects (e.g. dry mouth, cough) to dose levels that are below the maximal bronchodilating dose in man.[8] Early studies of tiotropium via the Handihaler® suggested potential for greater bronchodilator efficacy at doses higher than those currently approved and well tolerated. Recent data with the Respimat® device have also highlighted potential increased cardiovascular safety concerns that may be due to higher systemic exposure with the new device and formulation.[9]

The desire to improve upon the therapeutic index of tiotropium and ipratropium has led to the development of new anticholinergic agents with differing pharmacokinetic and pharmacological characteristics, with the aim of maximising efficacy and minimising potential adverse effects. AZD9164 is the result of a collaboration between AstraZeneca Discovery and Pulmagen Therapeutics Limited (formerly Argenta Discovery Limited), and is a selective, competitive antagonist at the human M_3 receptor. AZD9164 has potential as an inhaled, once-daily, long-acting anti-muscarinic bronchodilator.

The aim of the study was to examine the efficacy of single doses of AZD9164 in COPD patients and to identify an appropriate dose-range for future clinical studies. The doses of AZD9164 in the present study were chosen based on safety data from a single ascending dose (SAD) study in healthy volunteers, and the potency of AZD9164 was examined in terms of lung function and systemic effects in relation to tiotropium.[10,11]

Methods

A total of 60 patients were enrolled in the study, of whom 28 were randomised to treatment after a rigorous screening process to ensure both their safety and eligibility to participate in the study. To be eligible for the study, patients were required to have had a diagnosis of COPD with a post-bronchodilator FEV_1 between 40% and 80% of predicted normal and a FEV_1/FVC ratio less than 70%. Patients were further required to demonstrate reversible airway obstruction (i.e. >10% increase in FEV_1, after 3 × 40 μg ipratropium [Atrovent®]) on two separate occasions. The study comprised nine clinic visits, Visit 1 was for enrolment and to obtain informed consent, Visits 2 and 3 assessed ipratropium reversibility and COPD severity, Visits 4–8 were study drug treatment visits with Visit 9 for follow-up (Fig 1). Informed consent was needed prior to Visits 2 and 3 in order to be able to withdraw bronchodilators prior to these lung function tests. Long-acting β_2-agonists and muscarinic antagonists were not allowed before Visit 2 and throughout the study. Short-acting β_2-agonists were to be withheld 6 h and muscarinic antagonists 8 h before study drug intake, but were allowed between visits.

Treatments and study design

The trial was a double-blind, double-dummy, placebo-controlled, randomised, multi-centre, 5-way cross-over, single dose study carried out at four centres in Sweden. Patients inhaled three different single doses of AZD9164 (100 μg, 400 μg and 1200 μg estimated lung deposited doses) or saline placebo via a Spira Electro 2 Dosimeter jet nebuliser, 1 dose of tiotropium (18 μg) or lactose placebo from a Handihaler® dry-powder inhaler at five separate overnight visits. Irrespective of treatment, patients were required to take 19 inhalations from the nebuliser. Based on times of the PK sample taken immediately after end of inhalation, the inhalation procedure could be estimated to be 6–9 min long. In the morning of each treatment visit, the patient inhaled in a double-dummy fashion; first from the nebuliser and then from the HandiHaler®. The placebo and active capsules were loaded into the Handihaler® by a neutral observer to maintain double-blind status. Training in use of nebuliser and Handihaler® was given prior to treatment at Visit 3. The single dose administrations were

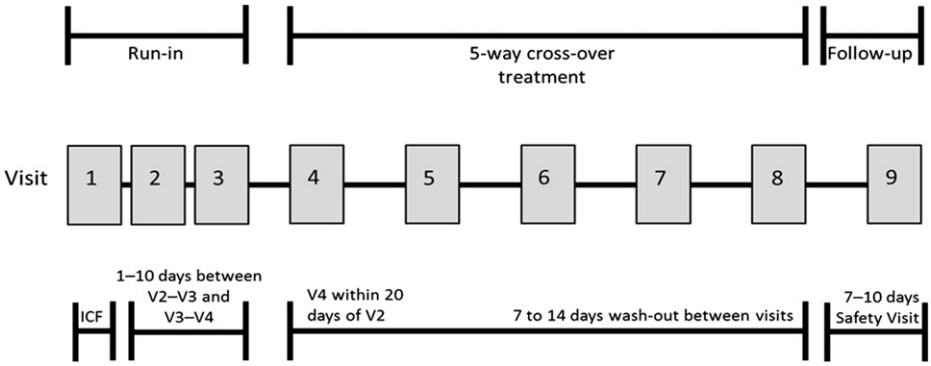

Figure 1 Study flow chart. ICF, Informed Consent Form.

separated by wash-out periods of 7–14 days (Fig. 1). The first dose (Visit 4) was to be given between 7.00 and 9.00 in the morning. The dosing at the following visits (Visits 5–8) was to take place within ±1 h of the time of the dosing at Visit 4. Time of first inhalation was used to represent when the dose was administered.

If required, Bricanyl® Turbuhaler® could be administered to patients as rescue medication at 0.25 mg per inhalation. All use was recorded on study case report forms with time of administration and the number of doses given on each occasion.

The primary objective of the study was to investigate the pharmacodynamics of inhaled AZD9164 in comparison to tiotropium and placebo, with FEV_1 as the primary variable for local pulmonary effects. FEV_1 was assessed as the average effect over 24 h (E_{av} = AUC_{0-24}/24 h), the peak (maximum) effect over 24 h (E_{max}) and the trough effect (E_{22-26}), here assessed as the average effect over 22–26 h. Variables for systemically mediated effects included heart rate, heart rate corrected QT interval (QTc), pulse and blood pressure. The secondary objectives of the study were to examine the safety of single doses of AZD9164 and to investigate drug exposure to AZD9164.

Blood samples to determine the plasma concentration of AZD9164 were taken prior to the administration of each drug dose, and then repeated up to 24 h after administration. The lower limit of AZD9164 quantification in plasma was 0.01 nmol/l. The first blood sample for PK determinations after inhalation was drawn as soon as possible after the patient had inhaled the investigational product. Further samples were taken at 15, 30, 60 and 90 min and 2, 4, 6, 8, 12 and 24 h post-dose.

Statistical methods

Computed pharmacodynamic parameters based on the peak, trough and average effects of variables (FEV_1, pulse, blood pressure, heart rate and QTc) were compared between the five treatments using analysis of variance (ANOVA) models with fixed factors for treatment, period and patient, and using the baseline of the day (the pre-dose value; for FEV_1 the geometric mean of the two pre-dose values) as a covariate. Additive models were used for measures of systemic effects, while FEV_1 was analysed using multiplicative models. Pharmacokinetic parameters were calculated using standard non-compartmental methods. These included area under the plasma concentration–time curve from time 0 to 24 h post-dose (AUC_{0-24}), the maximum plasma concentration measured (C_{max}) and the time taken to reach C_{max} (t_{max}).

Results

Baseline characteristics

The baseline characteristics of patients are outlined in Table 1. All participants allocated to treatment were white with 13 (46%) male patients. The average patient age was

Table 1 Demographic and disease-related data at entry.

Patient characteristic	n = 28 (all)
Gender	Male n = 13, female n = 15
Age (years)	64.2 (51–71)
Race	White
BMI (kg/m^2)	25.7 (20–34)
Time since COPD diagnosis (years)[a]	5.5 (1–20)
Time since COPD symptoms (years)[a]	9 (2–20)
Smoking status	Former n = 15, current n = 13
Pack years (years)	35.1 (14–56)
Inhaled GCS	No n = 7, yes n = 21
Daily dose of inhaled GCS (μg)	588.6 (320–1000)
Long-acting β_2-agonist	No n = 10, yes n = 18
Long-acting muscarinic antagonist	No n = 14, yes n = 14
Short-acting β_2-agonist	No n = 17, yes n = 11
Short-acting muscarinic antagonist	No n = 22, yes n = 6
Post-bronchodilator FEV_1, % pred (Visit 2)	61.1 (41–80)
Post-bronchodilator reversibility % (Visit 2)	20.9 (11–61)
Post-bronchodilator reversibility % (Visit 3)	22.1 (10–50)

GCS, Glucocorticosteroids.
[a] Median.

64.2 years (range: 51—71), with an average body mass index (BMI) of 25.7 kg/m^2 (range 20—34 kg/m^2). Participants were either former (15 [54%]) or current smokers with a smoking history of at least 10 pack years, and a median time since COPD diagnosis of 6 years (range: 1—20). Inhaled corticosteroids were used by 21 (75%) patients prior to the start of the study, with 18 (64%) patients using long-acting β_2-agonists and 14 (50%) patients using long-acting anticholinergics. The average post-bronchodilator FEV_1 at Visit 2 was 61.1% predicted normal (range: 41—80). The average reversibility (after 120 μg ipratropium) was 20.9% (range 11—61%) and 22.1% (range: 10—50%) at Visits 2 and 3, respectively.

Pharmacokinetics

Due to the long half-life of AZD9164 (on average 123 h, as determined by the SAD study in healthy volunteers), many pre-dose plasma samples contained quantifiable concentrations of AZD9164 (all below levels considered therapeutic). This was not corrected for when computing pharmacokinetic parameters, thus exposure after low dose may have been slightly overestimated. The absorption rate of inhaled single doses of AZD9164 was fast, with a median t_{max} of between 15 and 60 min. Following the peak in plasma concentration of AZD9164, levels of the study drug declined in a multi-phasic manner over the 24-h test period (Fig. 2). The systemic exposure of AZD9164 increased slightly more than dose-proportionally in the range 100—1200 μg as nebulised solution.

Pharmacodynamics

Shortly after inhalation, patients given AZD9164 exhibited a rapid, dose-dependent decrease in FEV_1. At the 15-min post-dose time point, mean decreases in FEV_1 of 3, 11 and 15% were observed for AZD9164 100 μg, 400 μg and 1200 μg respectively. This is in contrast to patients' response to administration of placebo and tiotropium 18 μg, which resulted in mean increases of 2% and 7% in FEV_1 respectively (Table 2, Fig. 3).

A diurnal variation in FEV_1 was seen after placebo administration, with a 3—5% increase during the day time, followed by a decrease to baseline overnight before an increase in FEV_1 the following morning (Fig. 4). After tiotropium 18 μg, FEV_1 peaked after 1 h, remained stable during the day time (approximately 1—14 h post-dose), then declined during the night (approximately 14—22 h post-dose). The initial decrease in FEV_1 with AZD9164 100 μg and AZD9164 400 μg was followed by a rapid increase to reach peak effects of 12% and 17% respectively at 2—6 h post-dose; effects which were maintained for 22—26 h post-dose. After the decrease in FEV_1 following administration of AZD9164 1200 μg, it took approximately 2 h for FEV_1 to reach levels comparable with the placebo group. FEV_1 then increased by approximately 12% above baseline, with a minimal decline in FEV_1 overnight.

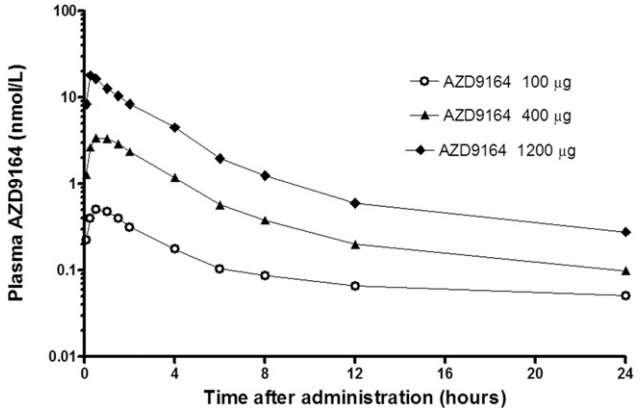

Figure 2 Mean plasma concentration curves on semi-logarithmic scale.

At each of the three study doses, AZD9164 was associated with statistically significant increases relative to placebo for E_{av}, E_{max} and E_{22-26}. These improvements were not dose-dependent, with AZD9164 400 μg consistently producing the largest effect. This was most likely due to the large initial decrease and slow recovery seen after AZD9164 1200 μg administration.

Tiotropium 18 μg produced statistically significant differences to placebo for E_{av} and E_{max}, but not on E_{22-26}. The two higher doses of AZD9164 (400 μg and 1200 μg) produced statistically significantly larger effects than tiotropium 18 μg on E_{22-26} while AZD9164 400 μg, but not 1200 μg, also had significantly larger effects on E_{max} and E_{av} (Fig. 5).

Rescue medication at clinical visits

In total, 10 patients were given 59 doses of rescue medication at 18 study treatment visits (Table 3). The use of rescue medication was most frequently reported during AZD9164 1200 μg treatment (8 out of 28 patients), with medication required shortly after inhalation of the study drug (within 30 min). In contrast, rescue treatment following tiotropium 18 μg was not initiated until during the night. The frequency of rescue treatment use was lowest during placebo days (1 patient).

ECG variables, pulse and blood pressure

Dose-dependent effects in heart rate were seen after AZD9164, with 1200 μg producing a statistically significant increase (mean increase 4 bpm) for both E_{max} and E_{av} in comparison to placebo, which caused a decrease. No other statistically significant effects were seen for heart rate or for QTc. The outcome for pulse following AZD9164 1200 μg closely followed the results for heart rate, with a statistically significant increase in pulse relative to placebo for both E_{max} and E_{av}. This was concomitant with a statistically significant increase in systolic blood pressure (as determined by E_{max}), but with no significant change in diastolic blood pressure. Tiotropium 18 μg had no significant effect on any of these parameters.

Safety evaluations

Extent of exposure and adverse events

Of the 28 patients randomised to treatment, there were no fatal adverse events, no serious adverse events, no

Table 2 Summary of individual changes in FEV_1 at 15 min.

Treatment	Decrease (no. of patients)	Increase (no. of patients)	Missing (no. of patients)	Mean change in FEV_1 (%)
AZD9164 100 μg	15	13	0	−3
AZD9164 400 μg	22	6	0	−11
AZD9164 1200 μg	24	2	2	−15
Tiotropium 18 μg	7	20	1	+7
Placebo	14	14	0	+2

discontinuation of investigational product due to an adverse event and no patient withdrawals. The highest frequency of adverse events was reported following AZD9164 1200 μg, both in terms of the number of patients with an event and the total number of events. The most frequently reported adverse effects were cough, throat irritation and dyspnoea (Table 4), appearing shortly after inhalation.

Clinical laboratory evaluation

No safety concerns were identified based on haematology or clinical chemistry, with no clinically significant changes in parameters from pre-drug administration to 24-h post-drug administration with investigational products in comparison to placebo. Measurement of urinalysis variables (protein, glucose and haemoglobin) pre- and post-drug administration demonstrated no clinically relevant changes.

Discussion

The present study demonstrated an increase in lung function (as measured by FEV_1) with all doses of AZD9164 (100 μg, 400 μg and 1200 μg) in comparison with placebo, following an initial bronchoconstrictor effect. Furthermore, the effect of AZD9164 400 μg and 1200 μg at 24 h was superior to that seen with tiotropium 18 μg — the current gold standard long-acting bronchodilator in COPD treatment. A low dose of AZD9164 (100 μg) appeared to give approximately the same 24-h bronchodilatory effect as tiotropium 18 μg, as determined by peak, trough and average FEV_1.

An unexpected finding of the study was an immediate but transient dose-related fall in FEV_1 with AZD9164 that was not observed following tiotropium or placebo treatments. This was accompanied by a dose-related increase in adverse effects, including cough, throat irritation and dyspnoea, and occasionally rescue medication needed to be given shortly after inhalation (30 min). The necessity of rescue medication was highest following inhalation of AZD9164 1200 μg (8 out of 28 patients). However, on an individual basis, there was no clear association between the degree of fall in FEV_1, the occurrence of adverse effects or the need for rescue medication. The high incidence of respiratory effects following AZD9164 1200 μg did not appear to influence absorption from the lungs into the systemic circulation.

The initial bronchoconstrictor effects of AZD9164 were unexpected as no similar effects were observed when single doses even higher than in present study (up to 1940 μg) were administered to healthy volunteers in the previous SAD safety study. However, it should be noted that FEV_1 measurements were not taken until 1.5 h after drug inhalation in the safety study. As the effects of muscarinic antagonism vary with age and severity of inflammation,[3] it is possible that patients with COPD may respond differently to inhalation of AZD9164 compared to healthy volunteers. An additional, non-pharmacological possibility is that local irritancy of AZD9164 may be greater in patients with COPD than in healthy patients.

Another unexpected finding was the lack of a clear dose—response to AZD9164 on FEV_1, with the 400 μg dose producing the largest effects. The dose—response and the extent of bronchodilation at later time points may have been affected by the magnitude of the initial drop in FEV_1,

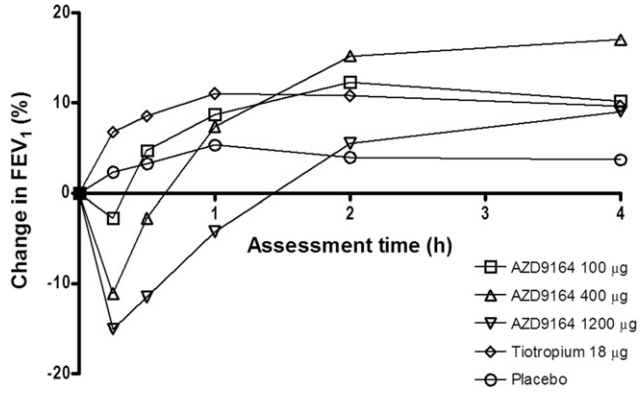

Figure 3 Mean value graph of change in FEV_1 during first 4 h, by treatment.

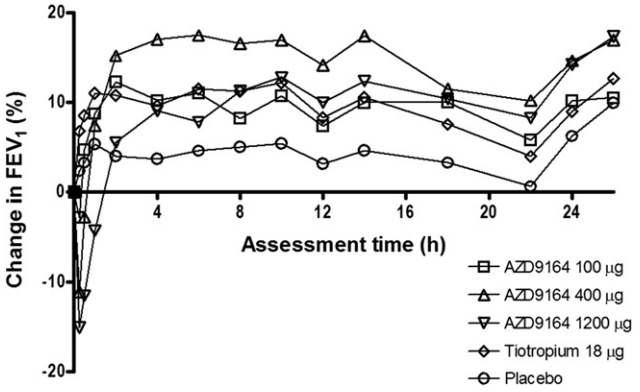

Figure 4 Mean value graphs of change from baseline in FEV_1 by treatment over 24—26 h.

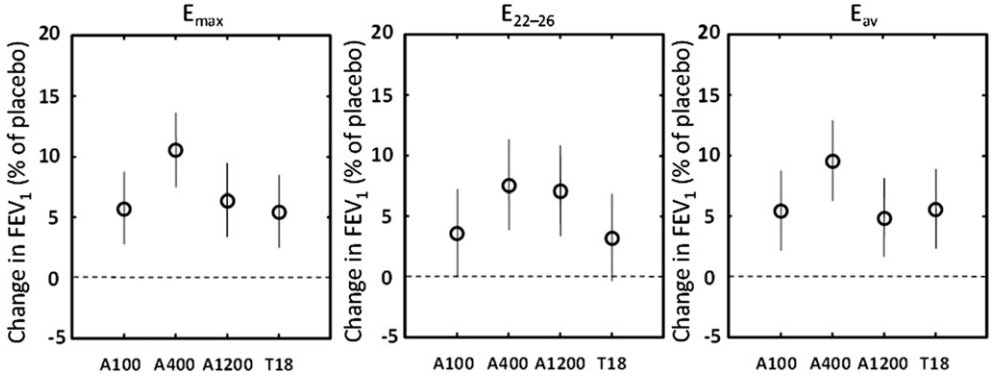

Figure 5 Adjusted mean FEV_1 ratios versus placebo with 95% confidence intervals. Treatment effects (differences versus placebo) are expressed as (baseline-adjusted) ratios of active treatment over placebo.

Table 3 Patients using rescue medication at clinic visits.

Treatment	*n*	Users	Doses
AZD9164 100 μg	28	3	8
AZD9164 400 μg	28	2	8
AZD9164 1200 μg	28	8	34
Tiotropium 18 μg	28	4	7
Placebo	28	1	2

particularly with the highest dose. It is not possible, therefore, to draw conclusions over the full dose-range at present.

Although an initial dip in FEV_1 following AZD9164 was observed, the subsequent bronchodilation was both profound and prolonged. Bronchodilation at 22—26 h was found to be significantly greater with AZD9164 than with tiotropium 18 μg. This increased bronchodilation at 22—26 h could be beneficial in combating the circadian drop in lung function overnight if replicated in other patients.

Table 4 Most frequent (> 5%) adverse events by preferred term.

Preferred term	A 100 μg	A 400 μg	A 1200 μg	T 18 μg	Placebo
Cough	0	6 (21%)	18 (64%)	1 (4%)	0
Throat irritation	0	5 (18%)	8 (29%)	0	0
Dyspnoea	1 (4%)	2 (7%)	7 (25%)	0	0
Headache	1 (4%)	3 (11%)	2 (7%)	1 (4%)	1 (4%)
Nasopharyngitis	2 (7%)	3 (11%)	0	0	2 (7%)
Chest discomfort	0	2 (7%)	4 (14%)	0	0
Thrombophlebitis	0	2 (7%)	1 (4%)	0	0
Chronic obstructive pulmonary disease	0	0	2 (7%)	0	0
Tremor	0	0	2 (7%)	0	0

n = 28 for all treatment groups. A, AZD9164; T, tiotropium.

If administration of AZD9164 resulted in either a brief initial partial M_1 agonist effect or an initial M_2 antagonist effect like that of ipratropium, before the expected prolonged blockade of M_3 receptors, this would provide a possible explanation for the brief, transient bronchoconstriction followed by prolonged bronchodilation observed in this study. A further possibility is a brief partial agonist effect on the M_3 receptor. However, none of these effects has been demonstrated in the laboratory or in preclinical studies with AZD9164. Further research will be required to investigate the dual drug response.

Even though a small initial bronchoconstriction was observed immediately after inhalation of AZD9164, no serious adverse events or systemic side effects (increases in heart rate, pulse and systolic blood pressure) were observed at the moderate doses, and could be detected only at the highest dose. Indeed, it is possible that different formulations of the AZD9164 molecule for inhalation could result in even better safety profiles.

Conclusions

This study demonstrates that it is possible to increase both the magnitude and duration of the improvements in lung function resulting from the administration of an inhaled muscarinic antagonist in patients with COPD. Tiotropium, still the only long-acting muscarinic antagonist currently available, is only approved at a dose of 18 μg via the Handihaler® and the potential to improve its bronchodilatory effects by administering higher doses may be restricted by an accompanying increase in anticholinergic effects, such as dry mouth. The present study with AZD9164 shows that it may be possible to improve on the bronchodilatory effects of tiotropium without an inevitable increase in such effects. However, the observed initial bronchoconstriction of AZD9164 would be deemed unacceptable, and needs further investigation prior to any larger clinical studies.

Conflicts of interest

LB has received honoraria for speaking and consulting and/or financial support for attending meetings from Almirall,

AstraZeneca, Airsonette, Andre Pharma, Boehringer, GlaxoSmithKlein, Merck, Mundipharma, Niigard, Novartis, Nycomed/Takeda and Orion Pharma. TB and CJ are employees of AstraZeneca. JL has received honoraria for speaking and consulting and/or financial support for research from AstraZeneca, Airsonette, GlaxoSmithKline, Merck/MSD and Novartis.

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