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Dose-response study with high-dose inhaled salmeterol in healthy subjects

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A double-blind, placebo-controlled, cross-over study in 10 healthy male subjects has been carried out to investigate the non-pulmonary effects of single inhaled doses of salmeterol 100 μg, 200 μg and 400 μg and salbutamol 400 μg from a metered-dose inhaler. At all doses tested, salmeterol produced statistically significant changes in pulse rate, tremor, blood glucose and plasma potassium concentrations, compared with placebo. All changes were dosed related. A number of dose-related adverse events including tremor, awareness of heart beat and headache were reported after salmeterol administration.

Keywords salmeterol salbutamol placebo high dose inhaled

Introduction

Salmeterol has been shown in animals to be a selective β_2-adrenoceptor agonist (Ball *et al.*, 1987). Studies in asthmatic patients have shown the therapeutic dose to be 50 μg twice daily which can be increased to 100 μg twice daily in patients with more severe reversible airways obstruction. β_2-adrenoceptor agonists at high doses can produce a number of non-pulmonary effects including an increase in physiological tremor and pulse rate as well as biochemical changes (Lipworth *et al.*, 1990). This early study in healthy subjects was designed to investigate whether these effects occurred after high doses of inhaled salmeterol and to compare the effects with those observed after inhaled salbutamol. The doses chosen were 100 μg, 200 μg and 400 μg salmeterol, that is between two and eight times the therapeutic dose. The dose of salbutamol used for comparative purposes was 400 μg, that is two to four times the dose used clinically.

Methods

Ten healthy male subjects (three smokers and four atopics) participated in the study. Their mean age was 34.3 years (range 27–44 years) and mean weight 71.2 kg (range 47.7–100.0 kg). A full medical examination (including a 12-lead ECG) and laboratory safety screen were undertaken before and after the study. Informed written consent was obtained in each case. Approval was obtained from the Glaxo Group Research Ethics Review Committee.

A double-blind, cross-over study was carried out. Each subject received three doses of salmeterol (100 μg, 200 μg and 400 μg) as the xinafoate salt, one dose of salbutamol 400 μg (Ventolin©) and placebo on separate occasions at intervals of not less than 6 days. Salmeterol was given in order of increasing dose and the sequence of the placebo and salbutamol doses was randomised.

Subjects fasted from midnight on the evening before each study day and consumed no food or beverages containing caffeine on the day of the study. An intravenous cannula was inserted into a forearm vein for collection of blood samples and kept patent with heparinised saline. Subjects lay on a couch in a semi-recumbent position. Pulse rate and blood pressure were measured using an automated sphygmomanometer (Narco Scientific BP 203 NA). A 12-lead ECG and rhythm strip were recorded (Marquette MAC1 or 2). Tremor was recorded using an Endevco transducer and the signal was analysed using a Tremor Analyser (Maconochie *et al.*, 1989).

Blood pressure, pulse rate, ECG and tremor were monitored at 5 min intervals until stable. The baseline readings were established when the resting pulse rate did not vary by more than 10 beats min^{-1} over three consecutive readings. The prescribed treatment was administered and these measurements, together with plasma potassium and blood glucose, were repeated for up to 8 h after each treatment.

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For each parameter, a weighted mean response was calculated by dividing the area under the response-time profile (calculated by trapezoidal integration) by the time over which measurements were made. The weighted means for tremor were transformed to logarithms prior to analysis to improve the validity of the required normality and variance homogeneity assumptions. The weighted mean responses were subjected to analysis of covariance using the mean baseline response as a covariate. The analysis also took account of variation between subjects, study visits and treatments.

For each response, the difference between each dose of salmeterol and placebo was calculated. Significance probabilities for tests of no effect were based on two-sided *t*-tests based on the analysis of covariance and 95% confidence intervals were calculated.

Results

Adjusted mean responses for the ten subjects after treatment are shown in Figure 1 and Table 1.

The effect of salmeterol on mean pulse rate was dose-related. All doses of salmeterol produced significantly higher mean pulse rates compared with placebo. After administration of salmeterol 100 μg, 200 μg and 400 μg, the mean increase in pulse rate compared with placebo was 6, 7 and 16 beats min^{-1}, respectively. In two subjects, minor non-specific T-wave changes were observed after salmeterol 400 μg; QT_c was prolonged (> 440 ms) in one subject after salmeterol 400 μg and in two subjects after salbutamol 400 μg. Salmeterol had no effect on systolic or diastolic blood pressure.

Salmeterol produced a dose-related increase in mean tremor. Each of the three doses of salmeterol produced significantly higher mean responses compared with placebo. In comparison with placebo, mean tremor after 100 μg, 200 μg and 400 μg salmeterol was increased by factors of 1.5, 2.5 and 3.5, respectively.

There was a dose-related increase in blood glucose concentration after salmeterol. All doses of salmeterol gave significantly higher weighted mean responses than placebo. Compared with placebo, the mean increase in blood glucose after salmeterol 400 μg was 1.23 mmol l^{-1}. Salmeterol produced a dose-related decrease in plasma potassium concentration. All three doses of salmeterol produced significantly lower mean responses compared with placebo. Compared with placebo, the mean decrease in plasma potassium concentration after salmeterol 400 μg was 0.45 mmol l^{-1}.

Apart from the changes presented on blood glucose and plasma potassium concentrations, no significant changes occurred in any routine laboratory safety test as a result of any of the treatments.

A number of adverse events were noted after administration of salmeterol, salbutamol and placebo. Headache was the most commonly reported adverse event, 1/10 subjects after placebo, 1/10, 3/10, 5/10 subjects after salmeterol 100, 200 and 400 μg respectively and 3/10 after salbutamol. Tremor was reported in 5/10 subjects after salmeterol 400 μg and 2/10 subjects after salbutamol

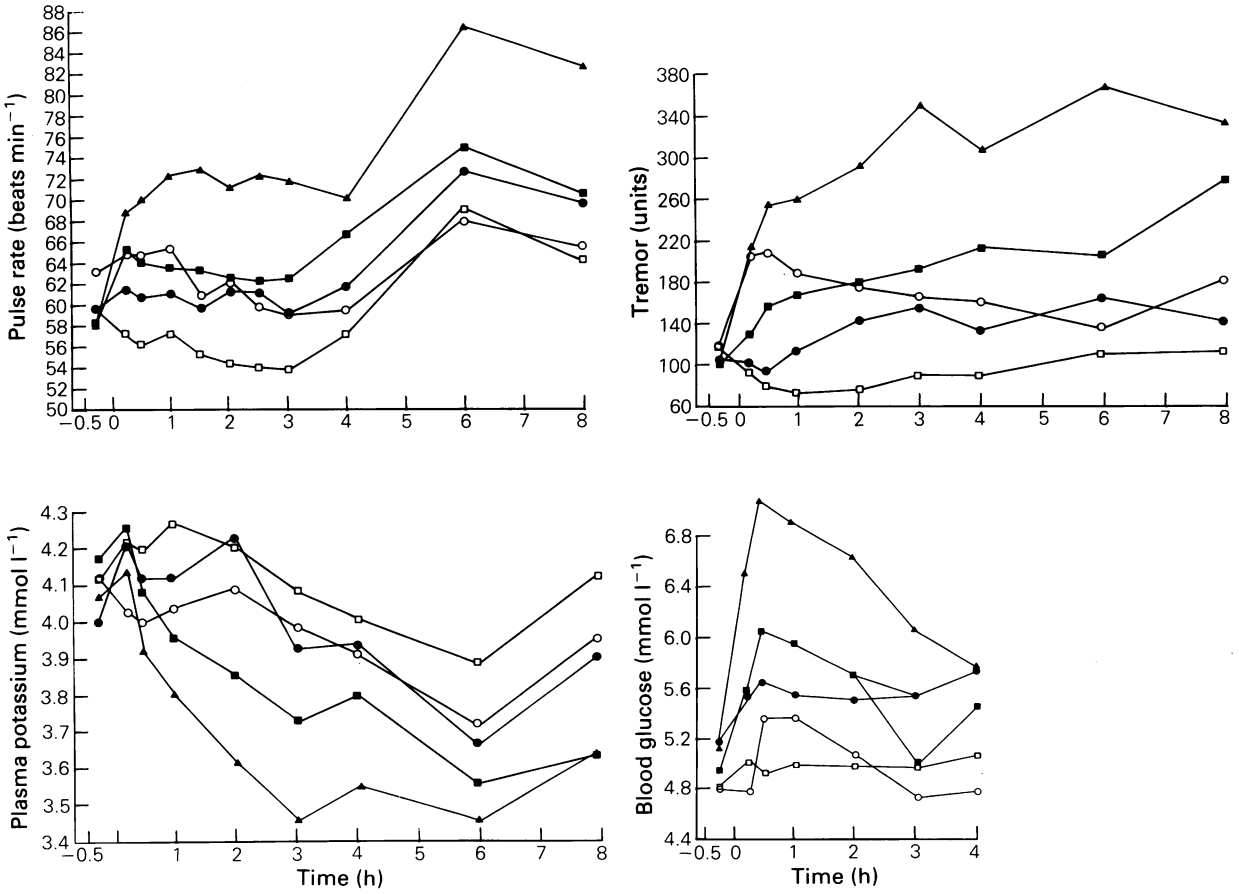

Figure 1 The mean effect of salmeterol, 100 μg (●), 200 μg (■), 400 μg (▲), salbutamol, 400 μg (○) and placebo (□) on pulse rate, tremor, plasma potassium and blood glucose in 10 healthy male subjects.

Table 1 Adjusted mean results in 10 subjects after salmeterol, salbutamol and placebo

Treatment	Dose (μg)	Pulse rate (beats min^{-1}) Mean	Pulse rate (beats min^{-1}) Difference from placebo (95% CI)	Systolic BP (mm Hg) Mean	Systolic BP (mm Hg) Difference from placebo (95% CI)	Diastolic BP (mm Hg) Mean	Diastolic BP (mm Hg) Difference from placebo (95% C I)
Salmeterol	100	66	6 (2,10)	117	−1 (−4,1)	73	−1 (−3,2)
	200	68	7 (4,11)	118	0 (−3,2)	72	−1 (−3,1)
	400	77	16 (12,20)	117	−2 (−4,1)	74	0 (−2,2)
Placebo	—	61		119		74	
Salbutamol	400	63		117		72	

Treatment	Dose (μg)	Tremor (units) Mean*	Tremor (units) Difference from placebo (95% CI)	Glucose (mmol l^{-1}) Mean	Glucose (mmol l^{-1}) Difference from placebo (95% CI)	Potassium (mmol l^{-1}) Mean	Potassium (mmol l^{-1}) Difference from placebo (95% C I)
Salmeterol	100	122	1.55+ (1.14,2.10)	5.40	0.61 (0.16,1.05)	3.94	−0.12 (−0.22,−0.02)
	200	198	2.49+ (1.88,3.31)	5.41	0.61 (0.21,1.02)	3.74	−0.32 (−0.41,−0.22)
	400	279	3.53+ (2.60,4.77)	6.03	1.23 (0.79,1.67)	3.61	−0.45 (−0.55,−0.35)
Placebo	—	79		4.80		4.06	
Salbutamol	400	147		4.99		3.91	

* Geometric mean; + Ratio.

400 μg and awareness of heart beating by 1/10 and 3/10 subjects after salmeterol 200 μg and 400 μg respectively.

Discussion

As anticipated, well-documented β_2-adrenoceptor agonist effects were observed after administration of salmeterol in terms of a dose-dependent increase in pulse rate, physiological tremor, blood glucose concentrations and decrease in plasma potassium concentration.

All doses of salmeterol produced a significantly higher mean pulse rate over the 8 h of the study than placebo. Similar effects have been shown with high doses of other β_2-adrenoceptor agonists (Crane *et al.*, 1989; Scheinin *et al.*, 1987; Whitsett *et al.*, 1981). This increase in pulse rate is probably due to the combination of an indirect effect through peripheral vasodilation and a direct chronotrophic effect through β_2-receptors in the heart (Hall *et al.*, 1989). Prolongation of QT_c has been observed by a number of investigators after high doses of β_2-adrenoceptor agonists (Crane *et al.*, 1989; Wong *et al.*, 1991). After administration of salmeterol the incidence was small and was less than after the same dose of inhaled salbutamol.

Salmeterol had no effect on blood pressure at the doses used.

Objective measures of tremor are very sensitive at picking up increases in tremor, but subjects are aware of tremor only when their objective reading has more than doubled (Watson & Richens, 1974). This was observed with salmeterol at a dose of 400 μg. After regular treatment with β_2-adrenoceptor agonists in asthmatic subjects, tachyphylaxis to the increase in physiological tremor usually occurs (Lipworth *et al.*, 1990; Minton *et al.*, 1991), so that after high doses of inhaled salmeterol in asthmatic patients, the effect on tremor may be expected to be less after regular treatment.

β_2-adrenoceptor agonists are known to increase blood glucose levels, probably through an effect on gluconeogenesis. These changes in blood glucose concentration observed after administration of salmeterol probably have little clinical significance in asthmatic subjects as tachyphylaxis occurs to these effects after regular treatment (Lipworth *et al.*, 1990).

The hypokalaemia observed after β-adrenergic stimulation is probably mainly due to increased intracellular uptake by both liver and skeletal muscle secondary to an activation of cell membrane sodium potassium ATPase (Brown, 1985; Vick *et al.*, 1972). As with other non-pulmonary effects of β_2-adrenoceptor agonists, tachyphylaxis has been observed to these falls in plasma potassium in the asthmatic subject (Lipworth *et al.*, 1990). β_2-adrenoceptor agonist-induced hypokalaemia has been accompanied by corresponding ECG alterations, most noticeably a decrease in T-wave amplitude (Struthers *et al.*, 1983) and prolongation of QT_c (Crane *et al.*, 1989).

We have shown that salmeterol at doses higher than those recommended for treatment in asthma is well tolerated in a group of healthy subjects. Clinically significant changes in a number of non-pulmonary effects occurred after 400 μg salmeterol, eight times the dose used therapeutically.

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