Enhanced synergy between fluticasone propionate and salmeterol inhaled from a single inhaler versus separate inhalers

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Background: The coadministration of long-acting inhaled β_2-agonists and inhaled corticosteroids is the most effective treatment for persistent asthma.
Objective: This meta-analysis aimed to determine the efficacy of fluticasone propionate and salmeterol inhaled from a single inhaler (combination therapy) or from separate inhalers (concurrent therapy).
Methods: Four similarly designed double-blind studies individually confirmed equivalence between combination and concurrent therapy on the basis of the primary efficacy measure (morning peak expiratory flow [PEF]). Each study showed a consistent trend in favor of combination therapy. Individual patient data from these studies were combined to provide overall estimates of treatment effect for morning PEF and other efficacy measures.
Results: Fixed-effects meta-analysis showed a significant advantage for combination therapy compared with concurrent therapy in morning PEF (mean difference between groups in change from baseline over 12 weeks of 5.4 L/min; *P* = .006; 95% CI = 1.5-9.2). Logistic regression analysis showed that the odds of achieving a greater than 15 or greater than 30 L/min improvement with combination therapy were increased by approximately 40% compared with those after concurrent therapy (15 L/min: odds ratio = 1.42, *P* = .008, 95% CI = 1.1-1.8; 30 L/min: odds ratio = 1.40, *P* = .006, 95% CI = 1.1-1.8), representing an additional 7% to 9% and 5% to 14% more patients, respectively, on combination therapy responding compared with those on concurrent therapy.
Conclusion: The meta-analysis indicates that the fluticasone propionate plus salmeterol combination offers the potential for increased clinical efficacy over concurrent use of the same doses of the same 2 drugs. After administration from a single inhaler, fluticasone propionate and salmeterol might codeposit in the airways. It is hypothesized that this codeposition offers an increased opportunity for synergistic interaction to occur.
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Key words: *Fluticasone propionate, salmeterol, synergy, asthma, Advair/Seretide, combination therapy*

Abbreviations used
ICS: Inhaled corticosteroid
ITT: Intent to treat
LABA: Long-acting β_2-agonist
PEF: Peak expiratory flow

Asthma is a disease characterized by airway inflammation and smooth muscle dysfunction. Therefore to achieve optimum asthma control, therapy should be targeted against these underlying components. The combination of a long-acting β_2-agonist (LABA) and inhaled corticosteroid (ICS) has been shown to improve lung function and to control symptoms and exacerbations more effectively than double the dose of ICSs in patients with varying degrees of asthma severity who are symptomatic while taking ICSs.[1-5] Indeed, the addition of an LABA to a low-to-moderate dose of ICS is a recommended treatment in the National Heart, Lung, and Blood Institute and Global Initiative for Asthma guidelines.[6,7] Inhalers delivering fixed combinations of ICSs and LABAs are now available. One particular combination, fluticasone propionate and salmeterol (Seretide/Advair), has shown superior efficacy compared with either agent given alone,[8,9] an increased dose of ICS alone,[10,11] or the leukotriene receptor antagonist montelukast either with or without an ICS.[12,13]

Preclinical work has shown that fluticasone propionate and salmeterol have complementary mechanisms of action and, in addition, interact in a synergistic manner at the receptor, molecular, and cellular levels.[14] However, for optimal interaction, the 2 drugs must reach the same target cell together in adequate concentrations. Therefore their coadministration, resulting in a greater amount of drug being delivered to the same site, favors this synergistic activity.

Four pivotal clinical studies[15-18] compared combination therapy with fluticasone propionate and salmeterol with the concurrent use of the individual components administered separately at the same doses. Although these studies met predetermined criteria for clinical equivalence, the results of the individual studies showed that there was a consistent and sustained trend in favor of combination therapy. A meta-analysis of these clinical studies was therefore conducted to assess further the effect of fluticasone propionate and salmeterol when delivered in combination from a single inhaler in comparison with delivery from separate inhalers.

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TABLE I. Overview of studies comparing combination therapy (fluticasone propionate/salmeterol) with concurrent therapy with salmeterol and fluticasone propionate

Study	Patient age group	Duration (wk)	Baseline asthma therapy (total daily dose)	Treatments (μg) twice daily	N	Baseline morning PEF (L/min)	Baseline FEV_1 (% predicted)
Bateman et al[15]	≥12 y	12	400-500 μg of BDP/BUD or 200-250 μg of FP	FP/Sal 100/50; FP 100 + Sal 50	121; 123	368; 365	75; 76
Chapman et al[16]	≥12 y	12*	800-1200 μg of BDP/BUD or 400-600 μg of FP	FP/Sal 250/50; FP 250 + Sal 50	180; 191	398; 391	75; 77
Aubier et al[17]	≥12 y	12*	1500-2000 μg of BDP/BUD or 750-1000 μg of FP	FP/Sal 500/50; FP 500 + Sal 50; FP 500	167; 171; 165	359; 345; 351	73; 73; 73
van den Berg et al[18]	4-11 y	12	400-500 μg of BDP/BUD or 200-250 μg of FP	FP/Sal 100/50; FP 100 + Sal 50	125; 132	241; 243	86; 84

BDP, Beclomethasone dipropionate; *BUD,* budesonide; *FP,* fluticasone propionate; *Sal,* salmeterol.
*Twelve-week efficacy analysis; 28-week safety analysis.

METHODS

Clinical studies

The 4 pivotal clinical studies[15-18] that compared combination therapy and concurrent therapy with fluticasone propionate and salmeterol administered at the same doses are presented in Table I. All 3 strengths of the combination (100/50, 250/50, and 500/50) were used in these studies, with one study also including an additional arm of fluticasone propionate monotherapy.[17] Three studies were conducted in adult and adolescent patients aged 12 years or older with asthma across a range of severities enrolled in 142 centers in 12 countries, and one trial was conducted in pediatric patients (ie, 4 to 11 years of age) in 35 centers in 9 countries. All 4 studies were conducted simultaneously and had different entry criteria, ensuring that no patient was enrolled in more than one study. All studies were of similar design: randomized, double-blind, double-dummy studies with equivalence assessed over 3 months. The method of collecting and analyzing data was also similar across all trials. Patients attended the clinic and were assessed at weeks –2 (run-in), 0 (randomization), 2, 4, 8, and 12 of treatment. Patients kept a diary record card of their peak expiratory flow (PEF) values, symptoms, and use of a reliever inhaler.

Primary efficacy variable

The primary outcome measure for all 4 studies was the change from baseline in mean morning PEF over 12 weeks. Baseline was taken as the mean value over the 2-week run-in period, and the post-treatment value was the mean over the 12 weeks. The threshold for determining clinical equivalence in morning PEF was prespecified as ±15 L/min. The mean difference in morning PEF (using individual patient data) and the difference in the percentage of patients responding to treatment were determined by using analysis of covariance (morning PEF) and logistic regression (increase from baseline in morning PEF), adjusting for the following covariates: baseline morning PEF, age, sex, and geographic region. Individual study results were combined to provide a weighted average, with weights proportional to the inverse variance of the study-wise effect estimators (fixed-effect analysis). Evidence of between-study heterogeneity was assessed through estimation of the between-study component of variance and construction of the corresponding random-effects CIs.[19] The generally wider CIs associated with the random-effects approach reflect the reduced precision for estimating the expected response in a future trial. The between-study component of variance was estimated by using both the methods of moments[19] and restricted maximum likelihood.[20] In all cases, both methods provided identical estimates.

An analysis of the proportion of patients who achieved a clinically meaningful improvement (responders) was conducted to assess whether combination treatment is clinically more effective than concurrent therapy. Because a change in morning PEF of ±15 L/min was considered the threshold for representing a clinically meaningful change in the protocols, this value was used to assess the proportion of responders for each treatment group. A threshold of 30 L/min was also used to explore further the nature of the treatment group difference.

The meta-analysis was conducted by using both the intent-to-treat (ITT) and per-protocol populations, as defined in the original protocols. The ITT population included all those randomized to treatment who received at least one dose of study drug. The per-protocol population included that subset of the ITT population that strictly met the study entry criteria.

Secondary efficacy variables

Secondary efficacy variables included the mean change in evening PEF and clinic FEV_1 plus the median percentage of days, nights, or both free of symptoms and with no reliever inhaler. These variables were analyzed by using the same methodology as the primary variable.

RESULTS

Clinical studies

In all studies, both combination therapy and concurrent therapy were associated with clinically significant improvements from baseline in mean morning PEF over the 12-week treatment period. There was a clear and consistent trend in favor of combination therapy over concurrent therapy, with the 90% CI for the difference falling outside the ±15 L/min limits for equivalence in the study by Bateman et al[15] (morning PEF: –17 to 0 L/min) and in the study by Chapman et al[16] (evening PEF: –16 to –4 L/min). Mean change in morning PEF during the 12-week treatment period is shown in Table II.

In absolute terms, the analysis showed that an additional 5% to 14% more patients treated with combination therapy achieved a greater than 30 L/min improvement in morning PEF compared with those treated with concurrent therapy in the 4 studies (Table III). Similarly, a greater than 15 L/min improvement was achieved in an extra 7% to 9% more patients receiving combination

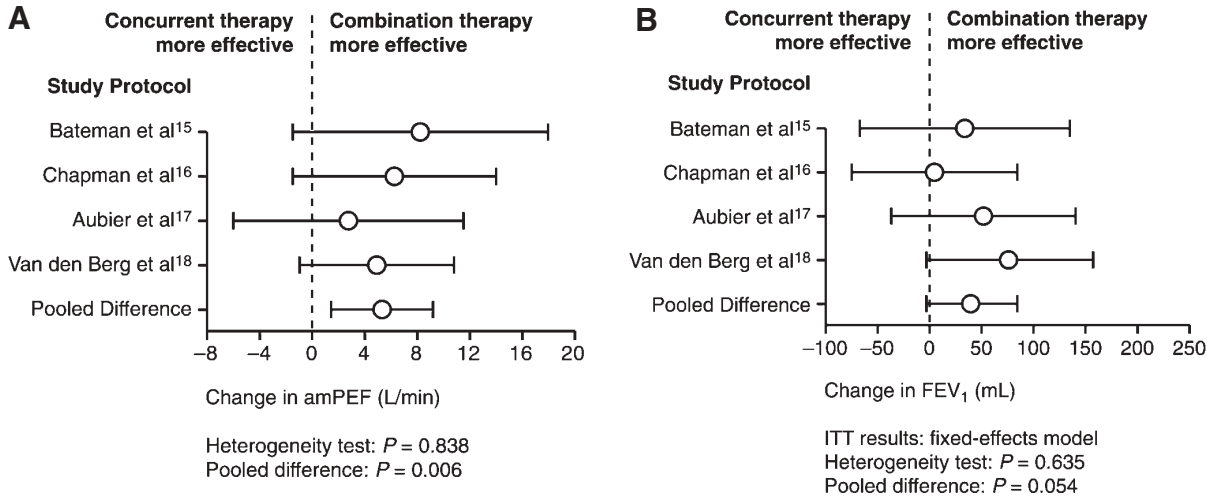

FIG 1. A, Difference in change in mean morning PEF *(amPEF)* over weeks 1 to 12 between ITT results (fixed-effects model). **B,** Mean change in clinic FEV_1 at week 12 (ITT results).

TABLE II. Adjusted mean change from baseline in mean morning PEF over weeks 1 to 12 for ITT and per-protocol populations

		Concurrent (FP + Sal)		Combination (FP/Sal)		Combination-concurrent	
Population	Study	N	Mean change (L/min)	N	Mean change (L/min)	Difference	95% CI
ITT	Bateman et al[15]	121*	33	121	42	8.6	–2 to 19
	Chapman et al[16]	191	36	180	43	6.4	–2 to 14
	Aubier et al[17]	170	33	167	35	2.7	–6 to 11
	Van den Berg et al[18]	132	28	125	33	4.9	–1 to 11
Per protocol	Bateman et al[15]	74	42	82	51	9.5	–3 to 22
	Chapman et al[16]	160	36	150	43	7.4	–1 to 16
	Aubier et al[17]	124	36	115	40	4.0	–7 to 14
	Van den Berg et al[18]	87	33	79	34	1.3	–6 to 9

*Two patients from the ITT population had missing data and could not be included in the analysis.

therapy versus those receiving concurrent therapy. Results in the ITT population were mirrored in the per-protocol population.

Meta-analysis

Fixed-effects meta-analysis (morning PEF, ITT) showed a significant 5.4 L/min advantage for combination therapy over the 12-week treatment period ($P = .006$; 95% CI, 1.5-9.2; Table IV and Fig 1, *A*). In all cases the estimated between-study variance was zero so that the fixed-effects and random-effects estimates and CIs were identical. The odds of achieving a greater than 15 or greater than 30 L/min improvement in morning PEF with combination therapy were also increased by approximately 40% compared with the odds for patients receiving concurrent therapy (Fig 2).

The mean difference in FEV_1 was 40 mL in favor of combination therapy and approached statistical significance ($P = .054$, Fig 1, *B*). The difference in mean evening PEF was 6.11 L/min ($P < .001$), but there were no significant differences seen in the percentage of symptom-free and reliever-free days and nights (Table IV). For all analyses, there was no evidence of between-study heterogeneity. Furthermore, an analysis of the study drug dose and treatment effect for morning PEF was nonsignificant ($P = .06$). An analysis of the 3 studies in adult and adolescent subjects found that the average difference between combined versus concurrent treatment was 5.7 L/min compared with 5.4 L/min in all subjects, suggesting that the benefit of combination therapy is also insensitive to age.

DISCUSSION

Clinical studies have confirmed that combination therapy with fluticasone propionate and salmeterol delivered from the same inhaler is superior to monotherapy with the individual components[8,9] and at least equivalent to that of the components administered concurrently.[15-18] This meta-analysis has shown that the improvement in morning PEF is significantly greater with combination than with concurrent therapy, with a greater proportion of patients achieving a clinically significant improvement. Similar modest clinical benefits for evening PEF and perhaps also FEV_1 are apparent, with evidence that the combination therapy also provides a similar level of efficacy

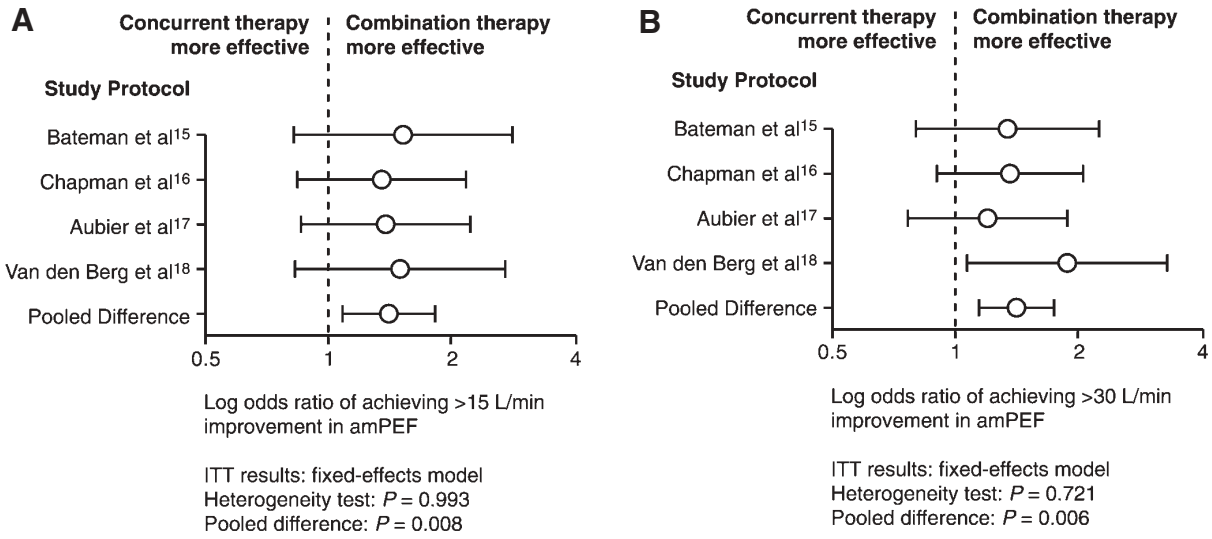

FIG 2. A, Odds of achieving a greater than 15 L/min improvement in mean morning PEF *(amPEF)* with combination therapy compared with concurrent therapy (ITT results). **B,** Odds of achieving a greater than 30 L/min improvement in morning PEF with combination therapy compared with concurrent therapy (ITT results).

TABLE III. Percentage of patients with an increase in mean morning PEF from baseline of greater than 15 or greater than 30 L/min (ITT population)

	Concurrent		Combination		
Study	n	Subjects with morning PEF increase, n (%)	n	Subjects with morning PEF increase, n (%)	Difference (%)
>15 L/min					
Bateman et al[15]	121*	87 (72)	121	96 (79)	7
Chapman et al[16]	191	130 (68)	180	135 (75)	7
Aubier et al[17]	170	106 (62)	167	115 (69)	7
Van den Berg et al[18]	132	81 (61)	125	87 (70)	9
>30 L/min					
Bateman et al[15]	121	66 (55)	121	75 (62)	7
Chapman et al[16]	191	89 (47)	180	100 (56)	9
Aubier et al[17]	170	75 (44)	167	82 (49)	5
Van den Berg et al[18]	132	48 (36)	125	63 (50)	14

*Two patients from the ITT population had missing data and could not be included in the analysis.

for other measures. It is suggested that these results are due to enhanced synergy of fluticasone propionate and salmeterol caused by codeposition in the airways after administration from a single inhaler.

Complementary and synergistic actions

There is growing evidence to show that LABAs and ICSs have complementary and synergistic efficacy, interacting usefully at the molecular, receptor, and cellular levels. With respect to complementary effects, the 2 classes of compound have very different modes of action, thereby targeting different aspects of the disease process: airway inflammation and smooth muscle dysfunction. LABAs have long-lasting effects on airway smooth muscle, inhibit mast cell mediator release, and reduce mucosa edema. ICSs have potent anti-inflammatory effects and reduce bronchial hyperreactivity. LABAs and ICSs in combination are therefore at least additive by virtue of having complementary modes of action and targeting different aspects of the underlying disease pathophysiology.[14]

LABAs and corticosteroids have, however, also been shown to interact at a receptor level. Corticosteroids increase β_2-adrenergic receptor transcription in the human lung[21] and increase the synthesis of respiratory mucosal β_2-receptors at clinical doses.[22] In turn, LABAs, such as salmeterol, have been shown to prime the inactive glucocorticoid receptor through a phosphorylation mechanism, rendering the receptor more sensitive to steroid-dependent activation.[23] Recent in vivo data showing increased glucocorticoid receptor nuclear translocation when fluticasone propionate is administered in combination with salmeterol further support a synergistic mechanism of action of fluticasone propionate and salmeterol.[24]

TABLE IV. Pooled difference between combined and concurrent therapy in mean morning PEF over weeks 1 to 12 and secondary efficacy measures

	Pooled estimate combination-concurrent				Heterogeneity		
	Difference	SE	*P* value	95% CI	Q	*df*	*P* value
Mean morning PEF (L/min)							
ITT: All studies	5.35	1.95	.006	1.52 to 9.17	0.847	3	.888
Per-protocol: All studies	4.67	2.33	.044	0.12 to 9.22	1.826	3	.609
Secondary variables (ITT population)							
FEV_1 (mL)	40	20	.054	0.00 to 0.08	1.708	3	.635
Mean evening PEF (L/min)	6.11	1.86	.001	2.48 to 9.75	1.580	3	.664
Median % days symptom free	0.00	0.68	.999	–1.32 to 1.32	0.757	3	.860
Median % nights symptom free	–1.15	1.01	.257	–3.14 to 0.84	1.018	3	.797
Median % days reliever free	–0.36	0.90	.685	–2.13 to 1.40	2.580	3	.461
Median % nights reliever free	–0.11	0.23	.645	–0.56 to 0.35	5.093	3	.165

Results shown are from the fixed-effects model. The estimate of the between-study component variance was zero by using either the method of moments or restricted maximum likelihood.

The positive interactions between corticosteroids and LABAs have been illustrated at the cellular level. For example, in steroid-sensitive systems, such as airway smooth muscle chemokine synthesis[25] and T-cell and eosinophil apoptosis,[26] salmeterol increases the potency of the steroid. In systems responsive to both ICSs and LABAs (eg, epithelial cell cytokine and chemokine release and cytoprotection of the respiratory mucosa against the damaging effects of microorganisms), the combination is more active than either drug alone.[25,27,28] A greater than additive anti-inflammatory effect on $CD3^+$ and $CD4^+$ T cells was demonstrated after 3 months' therapy with fluticasone plus salmeterol compared with therapy with low-dose fluticasone alone.[29] A recent 3-month biopsy study found that low-dose ICSs and salmeterol significantly decreased the number of blood vessels in the airway lamina propria compared with a higher dose of steroid. This effect on angiogenesis suggests a possible benefit of ICS plus LABA combination therapy in airway remodeling.[30]

These data suggest that there are more than complementary mechanisms involved in the simultaneous use of fluticasone propionate and salmeterol. This effect seems to occur when the 2 drugs reach the same cell together in adequate concentrations. Therefore their coadministration offers greater potential for this synergistic activity to occur provided that the 2 drugs are delivered to the same site in the lung at the same time.

Lung deposition pattern

Previous studies have demonstrated that there is no systemic pharmacokinetic or pharmacodynamic interaction between inhaled fluticasone propionate or salmeterol when administered in combination.[31] In addition, the amount of in vitro drug delivery of the fluticasone plus salmeterol combination from the Diskus inhaler demonstrates comparable performance with devices containing the individual drug products with respect to the fine particle mass of both fluticasone and salmeterol and to the particle size distribution of the emitted dose,[32,33] and hence greater total drug delivery with the combination product is not responsible for the greater clinical benefits. Although the differences between combined and separate inhalation treatment is not explained by differences in the total amount of drug deposited in the lung, these differences could be explained by breath-to-breath variability in deposition pattern. That is, delivery of 2 drugs simultaneously through a single inhaler avoids deposition variation caused by natural variation in inspiratory maneuvers on successive inhalations. This aids the colocation of the 2 compounds, which in turn increases the opportunity for synergistic interaction resulting from codeposition of salmeterol and fluticasone propionate at β_2 and glucocorticoid receptors in the same cell. When 2 drugs are administered in combination, regions of the lungs are exposed to a single aerosol cloud containing both active drugs. This is in contrast to the pattern of deposition when the 2 drugs are administered from separate devices, when the region of deposition will overlap but not coincide exactly. Indeed, given the degree of branching in the airways, the likelihood of drug codeposition by chance at the same cell is very low after successive inhalations. Although diffusion of drug molecules after dissolution in lung fluid will ensure that a proportion of cells will receive both drugs within the appropriate time window, the positive interaction between the 2 drugs at the cellular level is likely to be increased when the drugs are delivered as an aerosol cloud from a single device.

For fluticasone and salmeterol delivered from a single inhaler, the potential for airway codeposition to occur might further be enhanced by the tendency of the 2 drugs to form particle agglomerations within the inhaler device. It has been demonstrated that within an aerosol propellant system, there was an interaction between the drug particles.[34] Data are emerging on the nature of this agglomeration through the use of Raman laser spectroscopy, a technique capable of identifying individual drug particles and presenting an image in which the particles are tagged with a false color.[35] This technique has been used to analyze fluticasone plus salmeterol combination samples taken from stage 4 of an Anderson Cascade Impactor, the stage considered to represent the dose

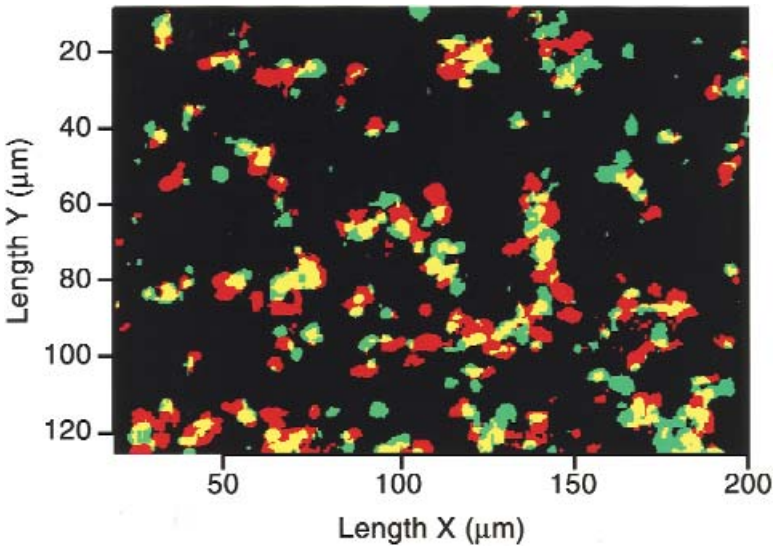

FIG 3. Raman laser analysis of Seretide metered-dose inhaler formulation on stage 4 of an Anderson Cascade Impactor showing particles of fluticasone propionate *(green)*, salmeterol *(red)*, and coassociation of both fluticasone propionate and salmeterol *(yellow)*.

delivered to the central airways. This analysis suggests that fluticasone plus salmeterol particle agglomeration persists after delivery from either the dry powder inhaler (as used in these clinical studies) or from the metered-dose inhaler (Fig 3). Work is ongoing to characterize further the nature of any possible physicochemical interaction of salmeterol and fluticasone propionate and whether it occurs with other drug and delivery device combinations.

Clinical data

The approach of adding an inhaled LABA to an ICS is supported by sound scientific and clinical evidence and supported by guideline recommendations.[6,14] There is now convincing clinical evidence showing that patients treated with the combination of fluticasone propionate and salmeterol gain significant benefit from the complementary effects of these drugs on inflammation and bronchoconstriction,[8,9,11] with associated improvements in quality of life.[36-38] However, this meta-analysis shows that the delivery of fluticasone propionate and salmeterol through a single inhaler provides an additional 5 L/min increase in morning PEF over the use of the same medications at the same doses but delivered through 2 separate inhalers. This difference is the same as the degree of improvement that would be seen if the dose of ICS had been doubled.[39] However, it is important to consider the effect on the individual patient to ascertain the clinical significance of this difference in mean morning PEF. This meta-analysis also shows a significant increase in the odds of achieving a clinically relevant improvement in morning PEF (of either >15 or >30 L/min) with combination therapy over and above that achieved with concurrent therapy. There was a trend for an improvement in FEV_1 across the 4 studies that supports the findings of the primary efficacy variable. The lack of any observed additional benefit of combination over concurrent therapy in the secondary outcome measures is of interest. This might reflect the relative insensitivity of these more subjective end points, especially given the large improvements from baseline seen with both groups across the 4 studies.

Similar clinical results have not been shown for an alternative LABA plus ICS combination, formoterol and budesonide in a single dry powder inhaler, although only data from one study have been published on which to base this comparison. Zetterström et al[40] showed that with the formoterol plus budesonide combination inhaler, there was a nonsignificant trend toward superior results seen only over the first 30 days of treatment when compared with those with concurrent budesonide plus formoterol. After 30 days, any differences between concurrent or combination therapy were negated or reversed. Each of the 4 individual studies as well as our meta-analysis of combined therapy with fluticasone propionate and salmeterol represents the treatment differences over the entire 12-week trial period, and the meta-analysis showed significant differences in morning PEF throughout this period. The phenomenon of enhanced synergistic potential through codeposition might be dependent on the specific drugs used.

Although the effects of this synergy are beneficial in terms of efficacy, they do not appear to be detrimental to safety. In the 4 pivotal studies adverse effects (including effects on serum cortisol levels) were low and similar in the concurrent and combination groups[15-18] and similar to those of fluticasone propionate alone.[17] It seems that there is a concentration response for the interaction between salmeterol and fluticasone propionate. Concentrations reached in the lung after inhalation are sufficient for the interaction to take place, but concentrations of either drug in the blood are not. Furthermore, the pharmacokinetic

time course is rather different for the 2 drugs, with salmeterol reaching a peak plasma concentration at around 5 minutes, whereas for fluticasone propionate, it is not until 1 hour that the peak concentration is reached.[31] The result is that there is an interactive effect in the airways with no evidence of interactive effect on the systemic effects of fluticasone propionate or salmeterol.

The results of the meta-analysis suggest that administration of a combination of fluticasone propionate and salmeterol through a single inhaler can provide significant improvement in lung function over and above that already seen with both agents administered concurrently through separate inhalers. This translates into an additional 7% to 9% of patients treated with combination therapy achieving a greater than 15 L/min improvement and an additional 5% to 14% with a greater than 30 L/min improvement in morning PEF. Recent Global Initiative for Asthma guidelines[7] and National Heart, Lung, and Blood Institute guidelines[6] recommend the use of an LABA with an ICS as highly effective asthma treatment and that the delivery of these agents in combination through fixed-dose inhalers aids compliance and convenience. Raman laser spectroscopy suggests that these additional benefits gained from delivery through a single inhaler are due to particle agglomeration of fluticasone propionate and salmeterol and subsequent codeposition within the airways.

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