Effects of Tiotropium or Combined Therapy with Salmeterol on Hyperinflation in COPD

YOHSUKE EGUCHI, YOSHITAKA TATEISHI, NOBUAKI UMEDA, TAKAHIRO YOSHIKAWA, HIROSHI KAMOI, HIROSHI KANAZAWA, SHINZOH KUDOH, KAZUTO HIRATA, and SHIGEO FUJIMOTO

YOHSUKE EGUCHI, YOSHITAKA TATEISHI, NOBUAKI UMEDA et al. Effects of Tiotropium or Combined Therapy with Salmeterol on Hyperinflation in COPD. Osaka City Medical Journal. 2007, 53, 25-34

Osaka City Med. J. Vol. 53, 25-34, 2007

Effects of Tiotropium or Combined Therapy with Salmeterol on Hyperinflation in COPD

YOHSUKE EGUCHI[1)], YOSHITAKA TATEISHI[2)], NOBUAKI UMEDA[1)], TAKAHIRO YOSHIKAWA[1)], HIROSHI KAMOI[2)], HIROSHI KANAZAWA[2)], SHINZOH KUDOH[2)], KAZUTO HIRATA[2)], and SHIGEO FUJIMOTO[1)]

Department of Sports Medicine[1)], and Department of Respiratory Medicine[2)], Osaka City University, Graduate School of Medicine

Abstract

Background

Hyperinflation is widely accepted as an abnormal state affecting clinical symptoms, activities of daily living and exercise tolerance in chronic obstructive pulmonary disease (COPD). Reducing hyperinflation is an essential theme in COPD treatment. In this study, we let patients with COPD hyperventilate to evoke hyperinflation, and evaluated the effects of tiotropium alone or in combination with salmeterol on hyperventilation-evoked hyperinflation.

Methods

Thirty-eight patients with COPD received pulmonary function tests including hyperventilation-evoked hyperinflation testing and the St. George's Respiratory Questionnaire (SGRQ) before treatment, after tiotropium administration for 8 weeks, and after combined therapy with salmeterol for 8 weeks.

Results

Before treatment, inspiratory capacity (IC) after hyperventilation decreased significantly in a breathing frequency-dependent manner. After tiotropium administration, forced expiratory volume in one second (FEV_1) increased significantly. IC after hyperventilation decreased significantly in a breathing frequency-dependent manner; however, IC was significantly greater than that before treatment (at rest, $p=0.001$; after hyperventilation at twice the resting respiratory rate, $p=0.0009$; and after hyperventilation at three times the resting respiratory rate, $p<0.0001$). The SGRQ score also improved significantly. After combined therapy with salmeterol, FEV_1 increased significantly compared with after tiotropium alone. However, there was no significant difference between the IC after tiotropium alone and that after combined therapy, at each stage. However, after combined therapy the SGRQ score significantly improved compared with that after tiotropium alone.

Received September 7, 2006; accepted November 28, 2006.

Correspondence to: Yohsuke Eguchi, MD.
Department of Sports Medicine, Osaka City University, Graduate School of Medicine,
1-4-3, Asahimachi, Abeno-ku, Osaka 545-8585, Japan
Tel: +81-6-6645-3790; Fax: +81-6-6646-6067
E-mail: m1159783@med.osaka-cu.ac.jp

Conclusions

Tiotropium improved airflow obstruction and hyperventilation-evoked hyperinflation. In combination with salmeterol, the improvement in airflow obstruction was greater, but hyperventilation-evoked hyperinflation was not further improved.

Key Words: Hyperinflation; Tiotropium; Salmeterol; Airflow obstruction; Inspiratory capacity

Introduction

Chronic obstructive pulmonary disease (COPD), a common condition characterized by poorly reversible airflow obstruction[1,2], is widely recognized as a major cause of death[3]. As the disease progresses, loss of lung elastic recoil and development of airflow obstruction lead to progressive air trapping with an increase in end-expiratory lung volume and a decrease in inspiratory capacity (IC)[4]. Static hyperinflation and its increase during exercise (dynamic hyperinflation) have been associated with limitations in the functional capacity of patients with COPD[5,6]. There is some debate about the use of FEV_1 as a single parameter for evaluation of patients with COPD[7]. Measurement of FEV_1 has been widely accepted for the classification of the severity of COPD[1,2]. However, since FEV_1 may be a poor predictor of clinical symptoms, exercise tolerance, and response to bronchodilators[8], additional parameters have been sought. As an alternative, exercise testing with repeated measurement of IC has been used to detect dynamic hyperinflation and evaluate response to bronchodilators[9-11]. Previous studies have shown that measurements of IC and endurance time during submaximal cycle exercise testing are highly reproducible in COPD[12,13]. Moreover, progressive reduction of IC during exercise reflects dynamic hyperinflation, and is a good predictor of decreased exercise ability as well as of increased exertional dyspnea[11]. In addition, since there is a close relationship between IC and exercise performance in COPD, hyperinflation estimated by IC could be a predictor of value in the natural course of the disease among patients with COPD, independent of FEV_1. Thus, changes in IC reflect dynamic hyperinflation and are correlated with breathing frequency in patients with COPD[11]. We hypothesized that increase in breathing frequency would produce changes in IC similar to the changes in IC observed during exercise.

Current therapy for COPD has improved the management of the disease. GOLD guidelines[2] now recommend the use of long-acting bronchodilators as the mainstay of COPD management. The introduction of the long-acting β2 agonist salmeterol and the anticholinergic agent tiotropium bromide have made important progress in the management of COPD. Tiotropium is a specific muscarinic receptor antagonist with kinetically controlled selectivity for the M3 subtype. The agent has been shown to achieve sustained bronchodilation and to improve health-related quality of life in patients with COPD[14-16]. The improvement in health status is likely to be multifactorial[17], but enhanced symptom control and increased exercise capacity are potentially important factors[17]. Salmeterol has also been reported to be of therapeutic utility in patients with COPD. Tashkin et al[18] have recently suggested the efficacy of salmeterol in addition to tiotropium in patients suffering from COPD with more severe symptoms. Therefore, this study was designed to evaluate the effects of monotherapy with inhaled tiotropium bromide, and combined therapy with salmeterol on airflow obstruction and hyperventilation-evoked hyperinflation in patients with COPD.

Materials and Methods

Patients

Study subjects were required to have a clinical diagnosis of COPD according to GOLD guidelines[2]. All patients were clinically stable without exacerbations for at least 8 weeks prior to the present study. Patients with a recent history of myocardial infarction, heart failure, or cardiac arrhythmia requiring drug therapy were excluded from the study. In addition, patients with respiratory infection in the previous 6 weeks were excluded. Patients with known hypersensitivity to anticholinergic drugs, symptomatic prostatic hypertrophy, or narrow-angle glaucoma were also excluded. Demographic and baseline characteristics of patients are summarized in Table 1.

Table 1. Subject characteristics at study entry

Variables	Value
No of subjects	38*
Age (yrs)	70.2±8.3
Sex	
Male	34
Female	4
BMI (kg/m^2)	20.7±2.6
Outpatients/hospitalized	38/0
Respiratory rate (/min)	16.7±3.2
Smoking history	
Cigarette use (pack•yrs)	66±37
current smoker	6
ex-smoker	32
non-smoker	0
Severity †	
Mild, ≧80% of predicted FEV_1,%	10
Moderate, 50-79% of predicted FEV_1,%	10
Severe, 30-49% of predicted FEV_1,%	10
Very severe, <30% of predicted FEV_1,%	8
Prestudy medication for COPD	
Xanthines	9
oral corticosteroids	4
inhaled corticosteroids	10

Data are presented as n or mean±SD.

*: patients included in efficacy analysis; Two patients were excuded from the final analysis because of acute exacervation during tiotropium administration, and because of palpitation during conbined therapy with salmeterol.

†: according to the Global Initiative for Chronic Obstructive Lung Disease (GOLD) criteria.

COPD, chronic obstructive pulmonary disease; BMI, Body mass index.

Study design

The study protocol was approved by the hospital medical ethics committee, and all patients gave written informed consent. The protocol is shown in Figure 1. This was a single-center 20 weeks study consisting of a 4 weeks run-in period, followed by two 8 weeks treatment periods. Patients received baseline pulmonary function tests including hyperventilation testing and the

St. George's Respiratory Questionnaire (SGRQ)[19] at the end of run-in period. Patients then entered an 8 weeks treatment period with inhaled tiotropium powder 18 μg once daily in the morning in order to achieve a pharmacodynamic steady state. After tiotropium administration, patients received pulmonary function tests including hyperventilation testing and the SGRQ. Subsequently, inhaled salmeterol powder 50 μg twice daily was added and combined therapy was continued for another 8 weeks. The evening dose of salmeterol was administered approximately 12 hr after the morning dose of the study medication. After combined therapy with salmeterol, patients again received pulmonary function tests including hyperventilation testing and the SGRQ.

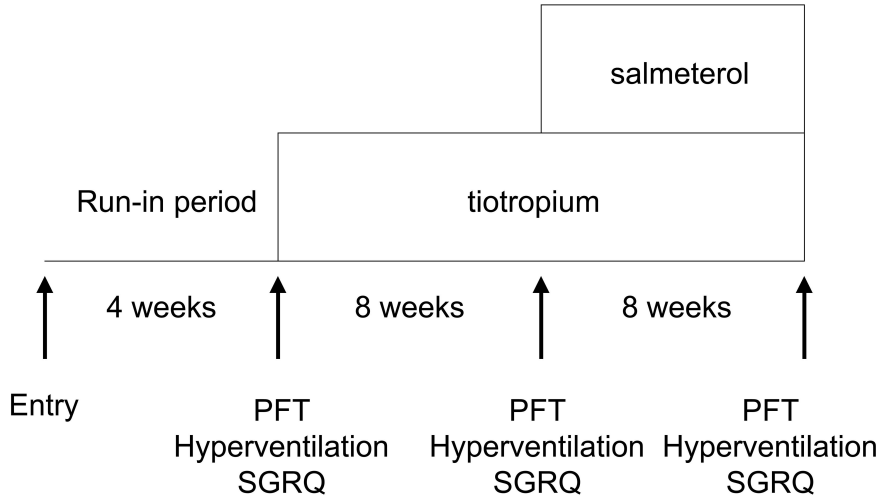

Figure 1. Study design. This study was a 20 weeks study consisting of a 4 weeks run-in period, followed by two 8 weeks treatment periods consisting of tiotropium administration followed by combined therapy with salmeterol. Patients received pulmonary function tests including hyperventilation testing, and the St. George's Respiratory Questionnaire (SGRQ) at the end of the run-in period and after each regimen. PFT, pulmonary function tests; and Hyperventilation, hyperventilation testing.

Throughout the whole study, patients were permitted to continue previously prescribed regular medications for their COPD at usual doses including inhaled steroids, oral steroids, theophylline preparations and mucolytics.

Pulmonary function tests

Pulmonary function tests were conducted in accordance with recognized standards[20]. Measurements were performed with a spirometer meeting American Thoracic Society criteria[21]. The highest FEV_1 and FVC values from three technically adequate measurements were retained. IC measurements were performed as described by O'Donnell et al[17] and were conducted before FEV_1 and FVC measurements. Residual volume was evaluated by body plethysmography using a pressure-compensated flow plethysmograph.

Hyperventilation testing

We evaluated hyperventilation-evoked hyperinflation using the method described by Gelb et al[8]. Patients were coached to maintain a respiratory rate synchronous with a metronome. Near-constant dynamic tidal volume during hyperventilation was achieved by having patients observe a graphical display of their breathing pattern. After hyperventilation at twice or three times the

baseline respiratory rate for 30 seconds, measurements of IC were performed.

Quality of Life (QOL)

QOL was evaluated at the end of the run-in-period and at the end of each treatment regimen using the St. George's Respiratory Questionnaire (SGRQ). The SGRQ consists of 50 disease-specific questions classified into three domains (symptoms, activity and impact). SGRQ total score was calculated as the sum of scores for each domain, with a decrease in score indicating an improvement: a change of more than 4 points was considered clinically significant.

Statistical Analysis

Comparisons of SGRQ data were made using two-tailed paired *t*-tests. Pearson correlations were performed in order to examine the strength of the relationship between SGRQ and IC at rest and after hyperventilation. Comparisons of other data were performed using analysis of variance (ANOVA) followed by Bonferroni's test. Values are presented as mean±SD unless otherwise specified. Statistical significance was set at p-values of <0.05.

Results

In this study, we used the hyperventilation testing to induce hyperinflation. Before treatment, IC after hyperventilation decreased significantly in a breathing frequency-dependent manner (IC at rest, 1.87±0.56 L; IC2, 1.36±0.55 L, $p<0.0001$; IC3, 1.15±0.57 L, $p<0.0001$; where IC2 and IC3 are inspiratory capacity after hyperventilation at twice and three times the resting respiratory rate, respectively; Fig. 2). Moreover, SGRQ symptom scores correlated significantly with IC at rest ($p=0.002$, $r=0.48$), IC2 ($p=0.007$, $r=0.43$), and IC3 ($p=0.006$, $r=0.44$) (Fig. 3).

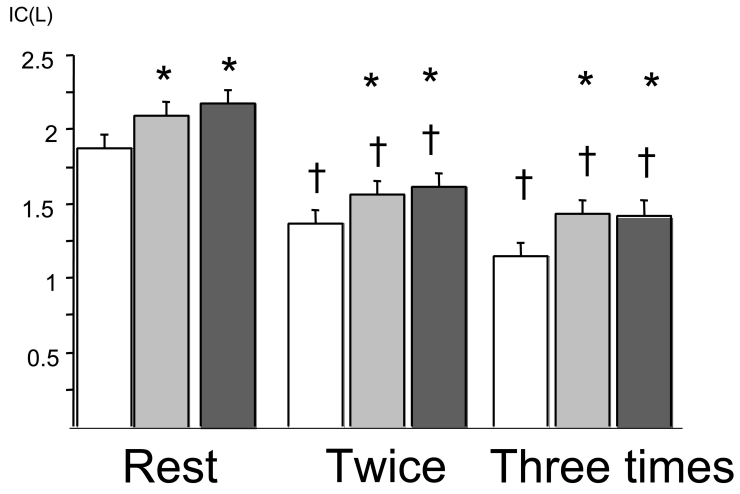

Figure 2. Inspiratory capacity (IC) in 38 patients with COPD at rest and after hyperventilation, before treatment (white bars), after tiotropium administration (gray bars), and after combined therapy with salmeterol (black bars). After tiotropium administration, IC after hyperventilation decreased significantly in a breathing frequency-dependent manner. However, IC was significantly greater than that before tiotropium administration, at each stage of the protocol. On the other hand, there were no significant differences between IC after tiotropium administration and that after combined therapy. Rest, at rest; Twice, after hyperventilation at twice the resting respiratory rate; and Three times, after hyperventilation at three times the resting respiratory rate. †: significant difference compared with IC at rest. *: significant difference compared with IC before tiotropium administration. Comparisons were performed using analysis of variance (ANOVA) followed by Bonferroni's test. Values are adjusted means±SE.

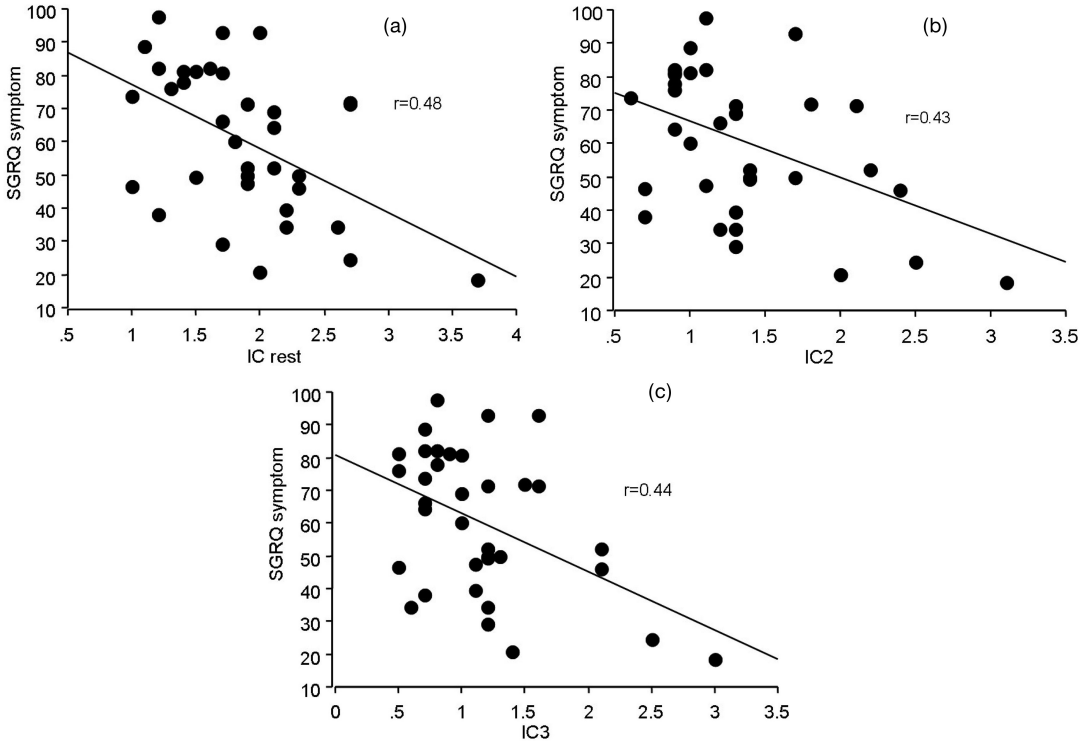

Figure 3. Relationship between SGRQ symptom score (vertical axis) and inspiratory capacity (IC) measured at rest (a), after hyperventilation at twice the resting respiratory rate (b) and after hyperventilation at three times the resting respiratory rate (c), before tiotropium administration. SGRQ symptom score correlated significantly with IC at rest, IC2 and IC3. SGRQ symptom, SGRQ symptom score before tiotropium administration; IC rest, IC at rest before tiotropium administration; IC2, IC after hyperventilation at twice the resting respiratory rate before tiotropium administration; and IC3, IC after hyperventilation at three times the resting respiratory rate before tiotropium administration. Pearson correlations were performed in order to examine the strength of the relationship between SGRQ and IC at rest or after hyperventilation.

After tiotropium administration, values for FEV_1, forced vital capacity (FVC), specific airways conductance (sGaw), and inspiratory to total lung capacity ratio (IC/TLC) increased significantly ($p<0.0001$, $p=0.0004$, $p=0.0049$, and $p<0.0001$ respectively; Table 2). IC after hyperventilation decreased significantly in a breathing frequency-dependent manner (IC at rest, 2.08±0.56 L; IC2, 1.55±0.58 L, $p<0.0001$; IC3, 1.42±0.53 L, $p<0.0001$, respectively; Fig. 2). However, IC at each stage was significantly greater than that before treatment (at rest $p=0.001$; IC2, $p=0.0009$; IC3, $p<0.0001$, respectively; Fig. 2). The improvement in SGRQ total score (13 points reduction, change >4U) and in each SGRQ domain score after tiotropium administration was statistically significant (Fig. 4).

After combined therapy with salmeterol, FEV_1 and sGaw increased significantly compared with tiotropium alone ($p=0.005$, $p=0.02$, respectively; Table 2). However, FVC and IC/TLC did not increase significantly compared with tiotropium alone ($p=0.28$, $p=0.34$, respectively; Table 2). IC after hyperventilation decreased significantly in a breathing frequency-dependent manner (IC at rest, 2.15±0.60 L; IC2, 1.60±0.53 L, $p<0.0001$; IC3, 1.42±0.63 L, $p<0.0001$, respectively; Fig. 2). However, there was no significant difference between IC after tiotropium administration and IC after combined therapy either at rest or after hyperventilation (IC at rest, $p=0.24$; IC2, $p=0.31$; IC3, $p=0.96$, respectively; Fig. 2). The improvement in SGRQ total score with combined

Table 2. Pulmonary function data before treatment, after tiotropium administration and after combined therapy with salmeterol

Parameter	Baseline	Tio	Tio+Sal
FVC (L)	2.80±0.85	2.99±0.78*	3.05±0.75*
FEV_1 (L)	1.21±0.65	1.32±0.65*	1.38±0.65*†
FEV_1 (% prediceted)	64.9±32.5	71.3±32.3*	74.8±32.4*†
FEV_1/FVC (%)	41.9±13.2	42.9±12.6	44.4±13.6*
MV (L)	11.7±2.3	12.3±2.8	12.9±4.2
MMF (L)	0.46±0.41	0.51±0.48	0.58±0.42*
RV (L)	2.51±0.65	2.34±0.57*	2.27±0.52*
RV/TLC (%)	45.3±8.7	42.6±8.1*	41.6±7.6*
%DLco (%)	73.6±31.6	75.1±30.1	75.2±38.3
DLco/VA (mL/min/mm Hg/L)	2.4±1.0	2.4±1.0	2.3±1.0
FRCbox (L)	4.07±0.88	3.85±0.88	3.82±0.84*
Raw (cmH_2O/L/sec)	5.0±3.7	3.7±1.4*	3.5±1.5*
sGaw (sec^{-1}•cmH_2O^{-1})	0.07±0.04	0.09±0.06*	0.10±0.06*†
IC/TLCbox	0.31±0.08	0.33±0.07*	0.34±0.07*

*: compared with that before treatment (significant difference; comparisons with analysis of variance (ANOVA) followed by Bonferroni's test).

†: compared with that after tiotropium administration (significant difference; comparisons with analysis of variance (ANOVA) followed by Bonferroni's test).

Data are presented as mean±SD unless otherwise indicated.

Baseline, before treatment; Tio, after tiotropium administration; Tio+Sal, combined therapy with salmeterol; FVC, forced vital capacity; FEV_1, forced expiratory volume in one second; and IC/TLC, inspiratory to total lung capacity ratio measured by body plethysmography.

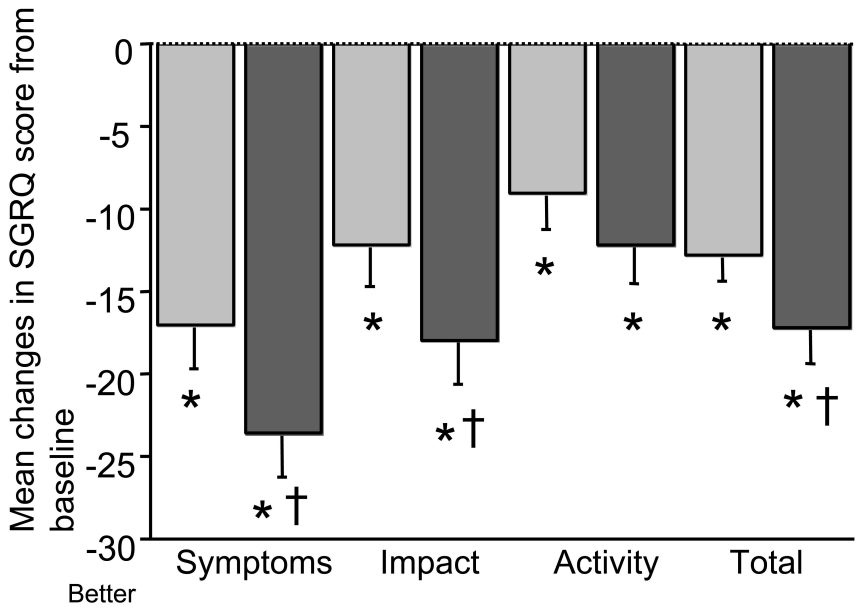

Figure 4. Mean changes in St. George's Respiratory Questionnaire (SGRQ) score after tiotropium administration (gray bars) and combined therapy with salmeterol (black bars) in patients with COPD. SGRQ symptoms scores (Symptoms), impact scores (Impact), activity scores (Activity) and total scores (Total) were measured before tiotropium administration, after tiotropium administration and after combined therapy with salmeterol: the changes from those before tiotropium administration are shown. A reduction in SGRQ score of 4 points was considered clinically meaningful. Values are adjusted means±SE. *: $p<0.05$ vs before tiotropium administration. †: $p<0.05$ vs after tiotropium administration. Comparisons were performed using two-tailed paired *t*-tests.

therapy (5 points reduction, change >4U) was significantly greater than with tiotropium alone (Fig. 4).

Discussion

The novel finding of this investigation is that tiotropium improved airflow obstruction and static hyperinflation. Of further importance is that tiotropium reduced hyperventilation-evoked hyperinflation in patients with COPD. Moreover, combined therapy with the addition of salmeterol to tiotropium resulted in additional improvement of airflow obstruction. However, salmeterol administration did not yield additional improvement of hyperventilation-evoked hyperinflation. We also showed a significant correlation between SGRQ symptom scores and hyperventilation-evoked hyperinflation. Thus hyperventilation-evoked hyperinflation was strongly associated with quality of life for patients with COPD.

The hyperventilation testing is a reliable surrogate for exercise-induced hyperinflation. Although we did not compare hyperventilation-evoked hyperinflation with exercise-induced hyperinflation, Gelb showed a significant correlation between them[8]. Thus, it is appropriate to use the hyperventilation testing to detect dynamic hyperinflation. Our finding that tiotropium improved hyperventilation-evoked hyperinflation may have been due to a reduction in static hyperinflation. This data agrees with the previous results of O'Donnell showing that tiotropium improved static hyperinflation and dynamic hyperinflation in parallel[17].

The clinical efficacy of long-acting β2 agonists including salmeterol is also apparent in patients with COPD. Noord recently reported additional effects from the combination of formoterol with tiotropium on airflow obstruction and on static hyperinflation in patients with COPD[22]. On the other hand, in our study, there was no significant difference between IC after tiotropium alone and IC after combined therapy with salmeterol. However, the difference between his results and ours may be attributable to population differences. Moreover, in this study, combined therapy with salmeterol and tiotropium did not demonstrate additional improvement of hyperventilation-evoked hyperinflation. Pharmacological differences, including differences in sites of receptor activation, between anticholinergic agents and β2 agonists may explain our findings that combined therapy with salmeterol and tiotropium improved airflow obstruction, but not hyperinflation.

We showed in this study that IC at rest and IC after hyperventilation correlated with the SGRQ symptom score. Recent studies have shown that IC correlates strongly with dyspnea intensity[17]. We also showed that combined therapy with tiotropium and salmeterol yielded an additional improvement in SGRQ. This decrease in SGRQ may be attributable to the improvement of airflow obstruction. SGRQ is widely accepted as an index that reflects various factors including pulmonary function, activities of daily living and mental state. We propose that SGRQ can be used as a clinical surrogate for dyspnea intensity in COPD.

This study was not a randomized, double blind trial. Therefore the question of additional improvement of hyperinflation by combined therapy with salmeterol requires more study.

In summary, tiotropium improved airflow obstruction and hyperventilation-evoked hyperinflation. Tiotropium plus salmeterol further improved airflow obstruction but did not produce an additional reduction in hyperventilation-evoked hyperinflation.

Acknowledgements

The authors wish to thank Dr Keisaku Fujimoto, of the First Department of Internal Medicine, Shinshu University School of Medicine, for his kind assistance with the hyperventilation technique.

References

1. Siafakas NM, Vermeire P, Pride NB, Paoletti P, Gibson J, Howard P, et al. Optimal assessment and management of chronic obstructive pulmonary disease (COPD). The European Respiratory Society Task Force. Eur Respir J 1995;8:1398-1420.
2. Pauwels RA, Buist AS, Calverley PM, Jenkins CR, Hurd SS. Global strategy for the diagnosis, management, and prevention of chronic obstructive pulmonary disease. NHLBI/WHO Global Initiative for Chronic Obstructive Lung Disease (GOLD) Workshop summary. Am J Respir Crit Care Med 2001;163: 1256-1276.
3. Murray CJ, Lopez AD. Mortality by cause for eight regions of the world: Global Burden of Disease Study. Lancet 1997;349:1269-1276.
4. Pride NB. The Respiratory System. In: Macklem PT, Mead J, editors. Handbook of Physiology: Section 3, Vol **Ⅲ**, Part 2. 2nd ed. Bethesda: American Physiological Society Book, 1986. pp. 659-692.
5. Diaz O, Villafranca C, Ghezzo H, Borzone G, Leiva A, Milic-Emil J, et al. Role of inspiratory capacity on exercise tolerance in COPD patients with and without tidal expiratory flow limitation at rest. Eur Respir J 2000;16:269-275.
6. O'Donnell DE, Revill SM, Webb KA. Dynamic hyperinflation and exercise intolerance in chronic obstructive pulmonary disease. Am J Respir Crit Care Med 2001;164:770-777.
7. Celli BR. The importance of spirometry in COPD and asthma: effect on approach to management. Chest 2000;117:15S-9S.
8. Gelb AF, Gutierrez CA, Weisman IM, Newsom R, Taylor CF, Zamel N. Simplified detection of dynamic hyperinflation. Chest 2004;126:1855-1860.
9. Belman MJ, Botnick WC, Shin JW. Inhaled bronchodilators reduce dynamic hyperinflation during exercise in patients with chronic obstructive pulmonary disease. Am J Respir Crit Care Med 1996;153:967-975.
10. Dodd DS, Brancatisano T, Engel LA. Chest wall mechanics during exercise in patients with severe chronic air-flow obstruction. American Review of Respiratory Disease 1984;129:33-38.
11. O'Donnell DE, Webb KA. Exertional breathlessness in patients with chronic airflow limitation. The role of lung hyperinflation. American Review of Respiratory Disease 1993;148:1351-1357.
12. O'Donnell DE, Lam M, Webb KA. Measurement of symptoms, lung hyperinflation, and endurance during exercise in chronic obstructive pulmonary disease. Am J Respir Crit Care Med 1998;158:1557-1565.
13. O'Donnell DE, Lam M, Webb KA. Spirometric correlates of improvement in exercise performance after anticholinergic therapy in chronic obstructive pulmonary disease. Am J Respir Crit Care Med 1999;160: 542-549.
14. Vincken W, van Noord JA, Greefhorst AP, Bantje TA, Kesten S, Korducki L, et al. Improved health outcomes in patients with COPD during 1 yr's treatment with tiotropium. Eur Respir J 2002;19:209-216.
15. Casaburi R, Mahler DA, Jones PW, Wanner A, San PG, ZuWallack RL, et al. A long-term evaluation of once-daily inhaled tiotropium in chronic obstructive pulmonary disease. Eur Respir J 2002;19:217-224.
16. Littner MR, Ilowite JS, Tashkin DP, Friedman M, Serby CW, Menjoge SS, et al. Long-acting bronchodilation with once-daily dosing of tiotropium (Spiriva) in stable chronic obstructive pulmonary disease. Am J Respir Crit Care Med 2000;161:1136-1142.
17. O'Donnell DE, Fluge T, Gerken F, Hamilton A, Webb K, Aguilaniu B, et al. Effects of tiotropium on lung hyperinflation, dyspnoea and exercise tolerance in COPD. Eur Respir J 2004;23:832-840.
18. Tashkin DP. Is a long-acting inhaled bronchodilator the first agent to use in stable chronic obstructive pulmonary disease? Curr Opin Pulm Med 2005;11:121-128.
19. Jones PW, Quirk FH, Baveystock CM, Littlejohns P. A self-complete measure of health status for chronic airflow limitation. American Review of Respiratory Disease 1992;145:1321-1327.
20. Cotes JE, Chinn DJ, Quanjer PH, Roca J, Yernault JC. Standardization of the measurement of transfer factor (diffusing capacity). Report Working Party Standardization of Lung Function Tests, European Community for Steel and Coal. Official Statement of the European Respiratory Society. Eur Respir J S**uppl** 1993;16:41-52.
21. Society AT. Standization of Spirometry 1994 update. Am J Respir Crit Care Med 1995;152:1107-1137.

22. van Noord JA, Aumann JL, Janssens E, Verhaert J, Smeets JJ, Mueller A, et al. Effects of tiotropium with and without formoterol on airflow obstruction and resting hyperinflation in patients with COPD. Chest 2006;129:509-517.