

# Effect of nasal continuous positive airway pressure on methacholine-induced bronchoconstriction

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Bronchial hyper-responsiveness is a cardinal feature of asthma. To determine whether nasal continuous positive airway pressure (NCPAP) influences airway smooth muscle in response to exogenous stimuli, we examined the effect of NCPAP on aerosolized methacholine-induced bronchoconstriction in 16 stable asthmatic patients. The dose-response curve for each subject was measured by a log transformation and linear regression analysis as well as a formula fitted to the data points to obtain values for  $a$  (slope) and  $b$  (position). The  $PD_{20}FEV_1$  significantly increased in patients receiving 8 cmH<sub>2</sub>O of NCPAP by one doubling dose compared with that in patients using sham pressure. NCPAP shifted the dose-response curves to be flatter, deviated upwards and to the right. The coefficient  $a$ , indicating bronchial reactivity, was significantly lower in patients receiving NCPAP. The coefficient  $b$ , indicating the bronchial sensitive threshold, was higher after applying NCPAP. In contrast, coefficients  $a$  and  $b$  did not change in subjects with sham pressure. NCPAP also significantly enhanced the bronchodilator effect of inhaled salbutamol in response to methacholine-induced bronchoconstriction. In summary, we have shown that NCPAP therapy improves bronchial smooth reactivity with an increase in  $PD_{20}FEV_1$  and a reduction in the bronchial reactivity and bronchial sensitivity. Therefore, NCPAP may provide an adjuvant therapy in patients with acute bronchial asthma.

## Introduction

Continuous positive airway pressure (CPAP) is a form of spontaneous breathing where positive airway pressure is applied throughout the respiratory cycle. It has been established in the management of patients with acute diffuse lung injury resulting in adult respiratory distress syndrome (1), and when applied through the nares, it is also the most effective treatment for patients with obstructive sleep apnoea (2,3). In asthmatic subjects, the effectiveness of nasal continuous positive airway pressure (NCPAP) is still conflicting (4,5).

CPAP has been shown to reduce the load on the inspiratory muscles and, therefore, decrease the work of breathing (5) in histamine-induced bronchospasm. A recent report also suggests CPAP at either 5 or 7.5 cmH<sub>2</sub>O reduces dyspnoea without interference with expiratory airflow (4). However, the application of NCPAP in patients with nocturnal asthma was suggested to be effective only if sleep apnoea was

present, or it would worsen sleep patterns without improving pulmonary function (6).

Bronchial smooth muscle contraction, airway oedema, mucus secretion, and airway inflammation constitute the pathophysiological features in bronchial asthma and contribute to the development of a fundamental defect in asthma, bronchial hyper-responsiveness (7). It has been demonstrated that bronchial hyper-responsiveness may be related to the airway resting tone in patients with bronchial asthma (8). Therefore, it is possible that the application of continuous positive pressure in the airways may alter the resting tone and subsequently the response of smooth muscle to stimuli. In the present study, NCPAP was applied to stable asthmatic patients in whom bronchoconstriction was induced by methacholine to assess whether NCPAP may influence bronchial sensitivity and reactivity.

## Materials and Methods

### SUBJECTS

Sixteen adult asthmatic patients (10 men and six women), who consented to the study, were investigated in outpatient clinics. Their ages ranged from

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27 to 65 (a mean of  $46.7 \pm 11.8$ ) years. All subjects met the criteria for the definition of asthma by the American Thoracic Society (9). There were no other pulmonary or systemic disorders or any occupational exposure history in these patients. All of them were in a clinically steady state without exacerbation of asthma or a respiratory tract infection for 8 weeks before the study. Their usual maintenance therapy included inhaled  $\beta_2$ -agonists on a 'as required' basis and oral theophylline which was used at least 4 weeks before and throughout the study without changing the dosage. No subject was taking corticosteroids in any form 6 weeks before or during the study. They had not used inhaled bronchodilators for 6 h and theophylline for 24 h prior to the methacholine challenge test.

#### METHACHOLINE CHALLENGE

Forced expiratory volume in 1 s ( $FEV_1$ ) was measured by a Spiroanalyzer ST-350R (Fukuda Sangyo. Co Ltd.). The best of three reproducible values (with a difference within 200 ml or less than 5%) was used for the calculation. Methacholine in phosphate-buffered saline or phosphate-buffered saline alone as its buffer solution was delivered by a Rosenthal dosimeter (Model PF2A, Laboratory for Applied Immunology Inc.) using a 646 DeVilbiss nebulizer. Phosphate-buffered solution was inhaled first, followed by methacholine solution at increasing concentrations of 0.075, 0.15, 0.3, 0.6, 1.25, 2.50, 5.0, 10 and 25 mg  $ml^{-1}$  at 5 min intervals. Patients received five inhalations for each concentration. The  $FEV_1$  was measured 5 min later before inhalation of the next concentration. The log dose-response curves for methacholine were constructed as the percentage changes in  $FEV_1$  from the baseline (post-buffer) value. The cumulative dose of methacholine causing a 20% fall in  $FEV_1$  ( $PD_{20}FEV_1$ ) was measured by linear interpolation.

#### DETERMINATION OF THE LEVEL AND DURATION OF NCPAP

Patients were fitted with a suitable nasal mask and CPAP was delivered from a flow generator (VitalAire, Marietta, U.S.A.) that provided a continuous flow of room air throughout the respiratory cycle. The pressure was adjusted with threshold valves.

To select a universally acceptable pressure and duration of NCPAP for each subject, different levels of pressure with variable duration were applied to all our patients in a preliminary study 4 weeks earlier. A NCPAP level of 8  $cmH_2O$  for 10 min was chosen since it was tolerated well by all patients. Higher level

( $\geq 10 cmH_2O$ ) or longer duration of NCPAP was not tolerated in some patients due to irritation and dryness of nasal mucosa. Patients had been acclimatized to NCPAP before entering the study.

#### PROTOCOL

On the first visit, all patients received spirometry and methacholine challenge tests without NCPAP. On the second visit (1 week later), the baseline  $FEV_1$  was assessed first, then NCPAP was applied to each subject. The patients were selected randomly into a study group of nine patients (four men and five women) receiving NCPAP of 8  $cmH_2O$  and seven patients (six men and one woman) served as a control group wearing a NCPAP mask without applying positive pressure (sham pressure). After NCPAP or sham pressure for 10 min, methacholine-challenge began. NCPAP or sham pressure was only discontinued during inhalation of methacholine solutions and performance of forced expiratory manoeuvres to measure  $FEV_1$ . After completion, all patients were given two puffs of metered-dose salbutamol (100  $\mu g$ /puff) to relieve bronchoconstriction induced by methacholine. On both visits, the studies were carried out at similar times of the day for each subject (morning or afternoon). All measurements were made with the subjects seated in the upright position.

To determine whether the effect of NCPAP could remain constant following the removal of NCPAP to measure  $FEV_1$ , spirometry was performed 30 s, 1, 2, 3, 4 and 5 min after the removal of NCPAP (8  $cmH_2O$ ,  $n=6$ ) or sham pressure ( $n=5$ ), which was applied for 10 min after an induction of a similar magnitude of bronchoconstriction by methacholine (by decreasing  $FEV_1$  20%).

To further investigate the effect of NCPAP on the airway smooth muscle in response to bronchodilators, a decrease in  $FEV_1$  by 50% of the baseline value was induced by methacholine inhalation in six subjects (three men and three women) on two separate occasions (3–5 days apart). On each occasion, after completion of an induced bronchoconstriction by methacholine, nebulized salbutamol (1 mg in 1.5 ml of 0.45% saline) was given and immediately followed by the application of either NCPAP (8  $cmH_2O$ ) or sham pressure on each subject for 30 min, except when spirometry was measured at 5, 10, 20 and 30 min after inhalation of salbutamol.

#### EXPRESSION OF RESULTS

##### *Methacholine dose-response curve*

The methacholine concentration expressed on a log scale was related to the percentage change in

Table 1 Characteristics of patients and baseline pulmonary function on two visits

Subject no.	Age (yr)	Sex	Baseline FEV <sub>1</sub> (l)	
			1st visit	2nd visit
NCPAP (8 cmH <sub>2</sub> O) group				
1	55	M	1.09	1.38
2	34	F	2.50	2.13
3	49	F	1.59	1.50
4	31	F	2.65	2.81
5	35	M	3.57	3.51
6	57	M	2.12	2.04
7	39	F	1.92	1.58
8	65	M	1.43	1.67
9	54	F	2.04	2.01
Mean	46.5		2.10	2.07
SE	4.03		0.24	0.23
Control (0 cmH <sub>2</sub> O) group				
10	57	M	1.31	1.46
11	53	M	3.30	3.31
12	47	F	2.13	2.04
13	32	M	3.06	3.03
14	57	M	1.83	1.81
15	55	M	1.40	1.09
16	27			
M				
3.00	2.71			
Mean	46.8		2.29	2.20
SE	4.69		0.31	0.31

Abbreviations: NCPAP, nasal continuous positive airway pressure; FEV<sub>1</sub>, forced expiratory volume in 1 s; M, male; F, female.

FEV<sub>1</sub> using linear regression analysis. The log-concentration of methacholine was plotted on the abscissa and the decline of FEV<sub>1</sub> on the ordinate to make a log dose-response curve. The formula  $y = ax + b$  was derived by the linear regression method. The coefficient  $a$  represented the slope of the dose-response curve and indicated bronchial reactivity (10,13), and the coefficient  $b$  represented the position of the curve and indicated bronchial sensitivity (11). The values of  $\ln[\text{Delta Ratio}]$  were obtained from the natural-log transformation of the percentage changes in PD<sub>20</sub>FEV<sub>1</sub> before and after the application of NCPAP or sham pressure.

#### Data analysis

Data were expressed by mean  $\pm$  SEM. The responses obtained in the same subject before and after NCPAP or sham pressure were compared using a Student's paired  $t$ -test. Comparisons between groups (NCPAP to sham pressure) were made using an unpaired  $t$ -test. Mann-Whitney U test was used to compare differences in PD<sub>20</sub>FEV<sub>1</sub> between the two groups. The Bonferroni correction was applied to

the results of multiple comparisons. The level of statistical significance was chosen at  $P < 0.05$ .

#### Results

Table 1 gives the age, sex and baseline pulmonary function (FEV<sub>1</sub>) for the studied subjects. For each subject, the baseline FEV<sub>1</sub> was not significantly different between two visits. Neither was there a difference in the baseline FEV<sub>1</sub> nor in the mean age between the NCPAP group and the sham pressure group (Table 1). With the application of NCPAP (8 cmH<sub>2</sub>O), the PD<sub>20</sub>FEV<sub>1</sub> increased significantly from  $2.2 \pm 1.9 \text{ mg ml}^{-1}$  before to  $3.7 \pm 2.9 \text{ mg ml}^{-1}$  ( $n=9$ ,  $P < 0.01$ ) (Fig. 1). In contrast, PD<sub>20</sub>FEV<sub>1</sub> in the sham pressure control group decreased from  $4.4 \pm 6.0 \text{ mg ml}^{-1}$  before to  $2.4 \pm 2.3 \text{ mg ml}^{-1}$  ( $n=7$ ,  $P > 0.20$ ). The natural log of the delta ratio of PD<sub>20</sub>FEV<sub>1</sub> was significantly more pronounced in the NCPAP group ( $4.2 \pm 0.3 \text{ mg ml}^{-1}$ ,  $n=9$ ,  $P < 0.05$ ) compared with that in the sham pressure control group ( $-1.4 \pm 1.4 \text{ mg ml}^{-1}$ ,  $n=7$ ) (Table 2). There was no significant difference in the initial PD<sub>20</sub>FEV<sub>1</sub>

Table 2 PD<sub>20</sub>FEV<sub>1</sub> and values from dose-response curve before and after nasal continuous positive airways pressure (NCPAP)

Subject no.	PD <sub>20</sub> FEV <sub>1</sub> (mg ml <sup>-1</sup> ) doubling dose ln[Delta Ratio (%)]	a		b	
		Pre-NCPAP	Post-NCPAP	Pre-NCPAP	Post-NCPAP
NCPAP (8 cmH <sub>2</sub> O)					
1	4.39	22.17	22.60	20.07	8.11
2	3.85	32.70	28.50	37.61	32.71
3	3.26	23.63	12.54	3.45	5.12
4	4.45	22.53	9.51	16.53	11.57
5	4.34	9.72	9.60	16.05	6.86
6	5.99	29.42	22.12	52.46	28.84
7	5.46	34.09	17.46	38.38	18.48
8	3.56	29.42	18.97	34.84	24.66
9	2.77	11.07	12.72	17.27	14.25
Mean (SE)	4.23 (0.34)	23.86 (2.92)	16.57 (2.19)*	26.30 (5.10)	16.73 (3.35)†
Control (0 cmH <sub>2</sub> O)					
10	- 3.30	16.25	35.58	18.55	34.07
11	- 2.94	18.74	29.62	26.97	35.13
12	- 3.50	13.80	12.70	16.94	13.90
13	3.18	9.44	8.22	2.55	1.18
14	- 4.29	8.77	11.66	9.39	12.01
15	- 3.83	21.24	32.85	30.45	47.21
16	4.65	18.25	13.82	27.60	20.48
Mean (SE)	- 1.43 (1.39)	15.21 (1.79)	20.64 (4.35)	18.92 (3.89)	23.43 (6.06)
NCPAP vs control					
P<0.05					

\*P&lt;0.05; †P&lt;0.01; comparison between data before NCPAP and after NCPAP.

Abbreviations: a, slope of dose-response curve; b, position of dose-response curve; PD<sub>20</sub>, provoking concentration for 20% fall in FEV<sub>1</sub>.

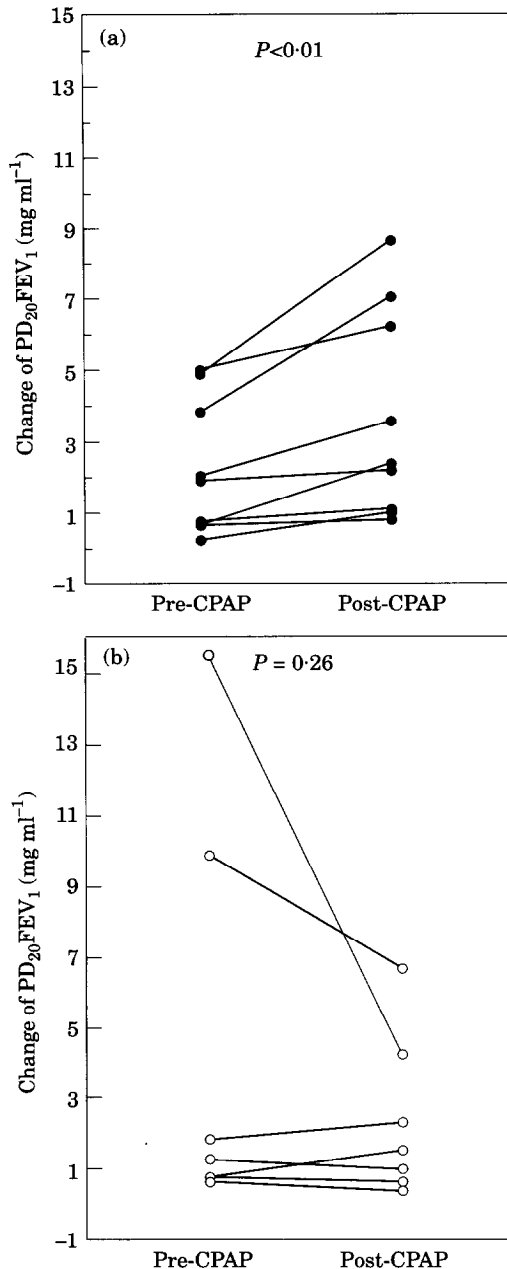


Fig. 1 The changes in  $PD_{20}FEV_1$  in patients receiving NCPAP of (a) 8 cmH<sub>2</sub>O (●,  $n=9$ ) or (b) sham pressure (○,  $n=7$ ).  $P < 0.01$  compared with baseline.

between the two groups. Neither did NCPAP or sham pressure influence the baseline  $FEV_1$  before methacholine challenge.

The typical dose-response curves of methacholine in the NCPAP group (subject 4) and the sham

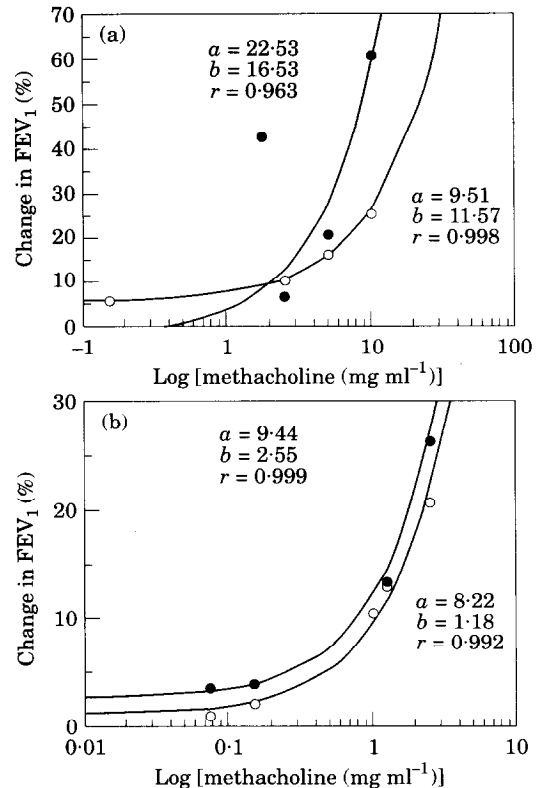


Fig. 2 Dose-response curves of methacholine for (a) a subject with NCPAP (8 cmH<sub>2</sub>O) (subject 4) and (b) a subject with sham pressure (subject 13) before (pre-NCPAP, ●) or after (post-NCPAP, ○) the application of pressure. For definition of abbreviations for  $a$  and  $b$ , see Table 2.

pressure control group (subject 13) are demonstrated in Fig. 2. With NCPAP, the curves were flatter and shifted to the right compared with baseline curves. Sham pressure failed to modify the dose-response curves. The values for  $a$ , indicating slope and bronchial reactivity, in NCPAP subjects were significantly lower ( $16.6 \pm 2.2$ ) than those before NCPAP ( $23.9 \pm 2.9$ ) ( $n=9$ ,  $P < 0.05$ ). The coefficient  $a$  did not show any significant difference after sham pressure (Table 2). The values for  $b$ , indicating position and bronchial sensitivity, were also significantly ( $n=9$ ,  $P < 0.01$ ) lower with NCPAP ( $16.7 \pm 3.4$ ) than those before ( $26.3 \pm 5.1$ ). Sham pressure failed to alter the coefficient  $b$  (Table 2).

The bronchodilator effect of NCPAP on methacholine-induced bronchoconstriction persisted for at least 5 min after the removal of NCPAP. The improvements in  $FEV_1$  in NCPAP (8 cmH<sub>2</sub>O) group, compared with that in the sham pressure group, were significant at 30 s and 1 min, and persisted until 5 min

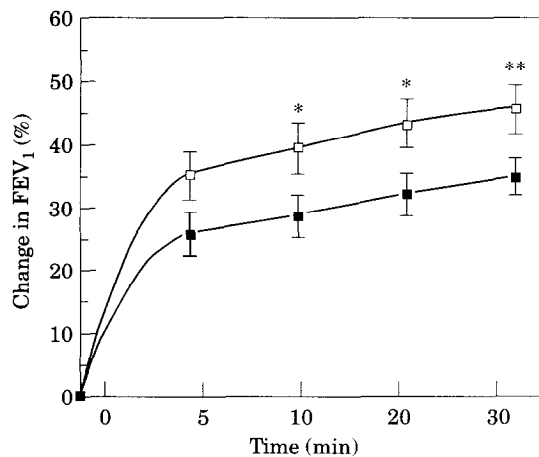


Fig. 3 The time-course for the bronchodilator effect of inhaled salbutamol in asthmatic patients with NCPAP (8 cmH<sub>2</sub>O, □,  $n=6$ ) or sham pressure (■,  $n=6$ ) after induction of a 50% decrease in FEV<sub>1</sub> by methacholine. Data are mean  $\pm$  SEM. \* $P<0.05$ ; \*\* $P=0.05$  compared with corresponding sham pressure controls.

after removal of NCPAP (by  $21.0 \pm 2.7\%$  vs.  $2.3 \pm 4.2\%$ ,  $24.2 \pm 4.3\%$  vs.  $3.8 \pm 3.1\%$ ,  $28.5 \pm 3.3\%$  vs.  $3.1 \pm 5.0\%$ ;  $P<0.05$ ,  $n=6$ ,  $n=5$ , respectively).

NCPAP also significantly enhanced the bronchodilator effect of salbutamol compared with sham pressure, significant at 10 and 20 min after inhalation of salbutamol (by  $39.8 \pm 4.1\%$  vs.  $29.0 \pm 3.4\%$ ,  $43.8 \pm 3.8\%$  vs.  $32.5 \pm 3.4\%$ ;  $P<0.05$ ,  $n=6$ ,  $n=6$ , respectively) (Fig. 3).

## Discussion

In the past, CPAP was often avoided in patients with obstructive airways disease to prevent the risk of barotrauma and adverse haemodynamic effects. However, recent studies showed CPAP might be beneficial in acute asthma. Shivaram *et al.* suggested CPAP reduced both respiratory rate and dyspnoea, and assisted inspiratory muscles in patients with acute asthma (4,10). Martin *et al.* also concluded that in induced asthma, CPAP reduced the pulmonary resistance and the load on inspiratory muscles to improve ventilation (5). Little attention has been given to the effect of CPAP on the airway functions. In the present study, the application of NCPAP in patients with methacholine-induced bronchoconstriction both significantly lowered the PD<sub>20</sub>FEV<sub>1</sub>, and altered the dose-response regression lines. Patients with a medium level of NCPAP (8 cmH<sub>2</sub>O) showed improvements in airway sensitivity and airway reactivity compared to those with sham pressure.

Since methacholine-induced bronchoconstriction may regress either spontaneously or by transient deep inspiration (11), it is possible that NCPAP-induced reversibility may be due to spontaneous regression or an effect of deep breathing in repetitive spirometry measurement. However, the negative response to sham pressure does not support this possibility. The bronchodilator effect of NCPAP or sham pressure was found to remain constant for more than 5 min after removal of applied pressure, suggesting that the change in the methacholine-induced constrictive responses, determined by measuring the changes in FEV<sub>1</sub> within 3 min of removal of applied pressure in our subjects, was adequate to represent the effect of NCPAP.

Previous reports have suggested there is a significant relationship between PD<sub>20</sub>FEV<sub>1</sub> and the baseline FEV<sub>1</sub> (8), suggesting that airway hyper-reactivity is dependent on the initial airway tone. Therefore, it is possible that the improvement in PD<sub>20</sub>FEV<sub>1</sub> by NCPAP in the present study is merely due to a non-specific bronchodilator effect of NCPAP on baseline airway resistance. However, a trivial effect of NCPAP on baseline FEV<sub>1</sub> discounts this possibility and indicates that NCPAP provides a protective action in airway smooth muscle against spasmogen challenge. Another possible mechanism postulated by Smith and Marini (12) is that NCPAP reduces expiratory airway resistance and avoids airway collapse by increasing the intraluminal pressure, therefore, pneumatically splinting the airways. This could explain in part the improvement in PD<sub>20</sub>FEV<sub>1</sub> in our study.

Orehek *et al.* (13,14) have suggested that inhalation dose-response curves should be studied to evaluate the airway reactivity. The coefficient  $a$  indicates the slope of the dose-response curve. The lower its value, the flatter the slope. The coefficient  $b$  is the position of the curve. The higher the value, the more the curve is displaced to the right (15). Orehek *et al.* supposed that the bronchial sensitivity (position) would indicate the intrinsic properties of the smooth muscle and the extrinsic stimuli acting on it. The bronchial reactivity (slope) represents levels of bronchoconstriction due to different drug doses (13). Similar suggestions were also reported by Woolcock *et al.* (15–17) and Beaupre and Mato (18). In the present study, NCPAP, but not sham pressure, elevated both the threshold of bronchial reactivity and sensitivity, indicating NCPAP not only offers a pneumatic splitting effect on airway smooth muscle but also affects the intrinsic properties of smooth muscle contraction, at least in response to methacholine.

It has been demonstrated that stimuli on the bronchopulmonary stretch receptors create a vagal positive feedback loop for increasing the contractile response, defined as 'reflex bronchoconstriction'. This has been documented in human airways (19–21) and is likely to be enhanced in asthmatic subjects by increasing the cholinergic tonic discharge. The increase in reflex results in greater bronchial reactivity (13). Hoffstein and Slutsky proposed that CPAP could stimulate pharyngeal mechanoreceptors to stiffen the upper airway (22). Some investigators also suggested that the use of CPAP may stabilize the upper airway and remove the chronic irritation to the oropharyngeal area, with subsequent elimination of the reflex bronchoconstriction (23). Thereby, NCPAP may intervene in the reflex responses in the upper airways and decrease bronchial reactivity. Stimulation of inhibitory neural pathways in airways, such as the non-adrenergic non-cholinergic (NANC) pathway has been suggested to be contributing to the response to CPAP in the upper airways (24).

This study also showed that NCPAP combined with  $\beta_2$ -agonist inhalation had a more profound bronchodilator effect than the  $\beta_2$ -agonist alone in pharmacologically-induced bronchoconstriction. Our study provided no clue for an enhancement of NCPAP effect on the  $\beta$ -adrenoceptor but suggests NCPAP can be used as an adjuvant therapy in relief of acute bronchoconstriction. Aside from the effect on airway function, NCPAP may reduce the load on the inspiratory muscles and decrease breathing work (5), resulting in improved ventilation function which may also contribute to the effects of NCPAP on airway function. Whatever the exact mechanism is, NCPAP provides a potent inhibitory action against increased airway resistance induced by smooth muscle constrictors.

Taken together, we have demonstrated that NCPAP may improve airways hyper-responsiveness to exogenous spasmogens and increase the efficacy of  $\beta_2$ -agonists in relief of bronchoconstriction. Our results indicate a potential therapeutic direction for the treatment of acute asthmatic attack.

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