

# Intravenous N-acetylcysteine in respiratory disease with abnormal mucus secretion

W. TANG<sup>1,2</sup>, D. ZHU<sup>3</sup>, F. WU<sup>4</sup>, J.-F. XU<sup>5</sup>, J.-P. YANG<sup>6</sup>, Z.-P. DENG<sup>7</sup>,  
X.-B. CHEN<sup>8</sup>, A. PAPI<sup>9</sup>, J.-M. OU<sup>1,2</sup>

<sup>1</sup>Department of Pulmonary and Critical Care Medicine, Ruijin Hospital, Shanghai Jiao Tong University School of Medicine, Shanghai, China

<sup>2</sup>Institute of Respiratory Diseases, Shanghai Jiao Tong University School of Medicine, Shanghai, China

<sup>3</sup>Respiratory Department, Jinhua Hospital Affiliated to Medical College of Zhejiang University, Zhejiang Province, China

<sup>4</sup>Respiratory Department, the Affiliated Hospital of Yangzhou University, Jiangsu Province, China

<sup>5</sup>Department of Respiratory and Critical Care Medicine, Shanghai Pulmonary Hospital, Tongji University School of Medicine, Shanghai, China

<sup>6</sup>Respiratory Department, Inner Mongolia Baogang Hospital, Inner Mongolia Autonomous Region, China

<sup>7</sup>Department of Pulmonary and Critical Care Medicine, The First People's Hospital of Zigong, Sichuan Province, China

<sup>8</sup>Infectious Disease Department, Deyang People's Hospital, Sichuan Province, China

<sup>9</sup>Department of Respiratory Medicine, University of Ferrara Medical School, Ferrara, Italy

**Abstract. – OBJECTIVE:** Evidence for the mucolytic and expectorant efficacy of intravenous (IV) N-acetylcysteine (NAC) is limited. This study aimed to evaluate in a large, multicenter, randomized, controlled, subject, and rater-blinded study whether IV NAC is superior to placebo and non-inferior to ambroxol in improving sputum viscosity and expectoration difficulty.

**PATIENTS AND METHODS:** A total of 333 hospitalized subjects from 28 centers in China with respiratory disease (such as acute bronchitis, chronic bronchitis and exacerbations, emphysema, mucoviscidosis, and bronchiectasis) and abnormal mucus secretion were randomly allocated in a 1:1:1 ratio to receive NAC 600 mg, ambroxol hydrochloride 30 mg, or placebo as an IV infusion twice daily for 7 days. Mucolytic and expectorant efficacy was assessed by ordinal categorical 4-point scales and analyzed by stratified and modified Mann-Whitney U statistics.

**RESULTS:** NAC showed consistent and statistically significant superiority to placebo and non-inferiority to ambroxol in change from baseline to day 7 in both sputum viscosity scores [mean (SD) difference 0.24 (0.763),  $p < 0.001$  vs. placebo] and expectoration difficulty score [mean (SD) difference 0.29 (0.783),  $p = 0.002$  vs. placebo]. Safety findings confirm the good tolerability profile of IV NAC reported from previous small studies, and no new safety concerns were identified.

**CONCLUSIONS:** This is the first large, robust study of the efficacy of IV NAC in respiratory

diseases with abnormal mucus secretion. It provides new evidence for IV NAC administration in this indication in clinical situations where the IV route is preferred.

#### Key Words:

Abnormal mucus secretion, Mucolytic, Expectorant, Sputum viscosity, Expectoration difficulty, N-acetylcysteine, Intravenous, Ambroxol.

## Introduction

N-acetylcysteine (NAC) was introduced in the 1960s and is well-established as a mucolytic agent for acute and chronic respiratory conditions<sup>1</sup>. In several European countries, NAC is currently licensed in this indication and is available in oral, inhaled, and parenteral formulations. Based on many years of use in clinical practice, NAC has shown an excellent safety and tolerability profile<sup>2</sup>.

The efficacy of oral NAC as a mucolytic has been demonstrated in several clinical studies<sup>3</sup>. A systematic review<sup>3</sup> of randomized placebo-controlled clinical studies on oral NAC in chronic bronchitis (CB) was published in 2011. It included 11 studies and over 2,000 patients and concluded that oral NAC reduces the risk of exacerbations

and improves symptoms compared to placebo<sup>3</sup>. A more recent meta-analysis<sup>4</sup> published in 2015 included 13 studies covering 4,155 patients with CB or chronic obstructive pulmonary disease (COPD; n=1,933 receiving NAC and n=2,222 placebo or controls) and concluded that NAC-treated patients had significantly fewer exacerbations of CB or COPD (relative risk 0.75, 95% CI: 0.66-0.84;  $p<0.01$ ).

Intravenous (IV) administration of NAC as a mucolytic for respiratory conditions may be preferred in certain circumstances, e.g., severely ill hospitalized patients. As we are not aware of clinical trials evaluating the efficacy and safety of IV NAC as a mucolytic, it is important to establish the efficacy of IV NAC as a mucolytic and expectorant in a large, robust clinical trial.

The current study aimed to demonstrate in a large, multicenter, randomized, controlled, subject and rater-blinded study that NAC injection [600 mg twice daily (BID)] is superior to placebo in sputum viscosity score and expectoration difficulty score at the end of 1 week of treatment and non-inferior to ambroxol hydrochloride in adult subjects with respiratory diseases and abnormal mucus secretion.

## Patients and Methods

### Patients

Hospitalized adult subjects aged  $\geq 18$  were enrolled in the study from 28 study centers in China. All subjects had a respiratory disease, including acute bronchitis, chronic bronchitis and exacerbations, emphysema, mucoviscidosis and bronchiectasis, and abnormal mucus secretion (defined by sputum viscosity score  $\geq 2$  and expectoration difficult score  $\geq 2$ ). Subjects with active tuberculosis, lung cancer, pulmonary fibrosis, acute pulmonary thromboembolism, markedly abnormal hepatic enzymes, or markedly elevated creatinine were excluded from participation in the study.

### Study Design

This was a subject- and rater-blind, randomized, multicenter study. A double-blind study design was not possible because the NAC formulation releases a distinguishable smell and because the appearance of the NAC ampoules and ambroxol vials is different. Subjects and personnel involved with efficacy assessments or

care of patients were blinded to the identity of the study medication (subject- and rater-blind design). Study staff administering study medication were aware of the identity of treatment but were not involved in any efficacy assessments or the care of patients.

The study was powered to reach statistical significance for the superiority comparisons of NAC vs. placebo and the non-inferiority comparisons of NAC vs. ambroxol hydrochloride (key secondary study objective) in at least one of the two co-primary endpoints after 7 days of treatment. The sample size calculation assumed a standard deviation (SD) of 0.79 for sputum viscosity and 0.77 for expectoration difficulty scores and a correlation between the two endpoints equal to 0.31. For the comparison of NAC vs. placebo (primary study objective) and NAC vs. ambroxol hydrochloride (non-inferiority), a sample size of 100 subjects in each group was estimated. Assuming a drop-out rate of about 10% over 1 week of entry, a total of 333 subjects were randomized (111 subjects to the NAC group, 111 subjects to the placebo group, and 111 subjects to the ambroxol group).

### Ethics

The protocol and informed consent form were approved by the Ruijin Hospital Ethics Committee, Shanghai, China, on January 24<sup>th</sup> 2019, and the study was performed according to Good Clinical Practice and in accordance with the Declaration of Helsinki. The study was registered in the ClinicalTrials.gov registry (NCT03843541). The patients were included after releasing a signed informed consent to participate. Informed consent was obtained from each subject (or the subject's legally authorized representative) before the subject was admitted to the study. The investigator did not undertake any investigation specifically required for the clinical study until valid consent had been obtained.

### Treatments

Eligible subjects were randomized in a 1:1:1 ratio to one of three treatment arms (NAC, ambroxol or placebo). Study medication was administered in the hospital, morning and evening, by slow IV infusion over at least 5 minutes for 7 days as follows: NAC (Zambon) 600 mg (in 10 mL NaCl 0.9% saline); ambroxol hydrochloride (Boehringer Ingelheim) 30 mg (in 10 mL NaCl 0.9% saline); or placebo (10 mL NaCl 0.9% saline).

Concomitant expectorants, antitussive agents, sedatives, and Traditional Chinese Medicine (TCM) treatments were not permitted during the study.

### Assessments

Subjects were assessed at baseline, treatment day 3, and treatment day 7, in the hospital. At each control, sputum viscosity, expectoration difficulty, sputum color, and cough were recorded on an established 4-point ordinal categorical scale (Table I)<sup>5</sup>. Sputum volume was assessed based on sputum collected over 24 hours by the subject in a graduated container.

Any adverse events (AEs) were recorded at each assessment, with an additional follow-up telephone call 2 weeks after the last administration of study medication for monitoring of any post-treatment AEs.

### Statistical Analysis

The primary efficacy endpoint was the change from baseline to day 7 in the mean sputum viscosity score or mean expectoration difficulty score (co-primary endpoints). The primary analysis was the comparison between NAC and placebo, with a secondary analysis of NAC *vs.* ambroxol, with a further analysis of ambroxol *vs.* placebo.

Non-key secondary endpoints included change from baseline to day 3 in mean sputum viscosity score or mean expectoration difficulty score, change from baseline to day 3 and day 7 in mean sputum color score, mean cough severity score, and mean sputum volume.

Safety endpoints included the frequency of AEs occurring from the first day of study medication administration (overall and considered related to study medication), clinical laboratory

evaluations, vital signs, ECG parameters, and physical examination findings.

Efficacy analyses were performed on the modified Intent-to-Treat (mITT) population (all randomized subjects who received at least one dose of study medication). The co-primary efficacy endpoints (change from baseline to day 7 in sputum viscosity score or expectoration difficulty score) were analyzed by means of stratified Mann-Whitney U statistics to test the hypotheses of the superiority of NAC *vs.* placebo. Additionally, a modified Mann-Whitney U statistics was used to test the hypotheses of non-inferiority of NAC *vs.* ambroxol assuming a margin (i.e.,  $\delta'$ ) of non-inferiority equal to 0.30. Non-inferiority was declared if the one-sided CI was within the interval  $(1/2-\delta', 1)$ , where " $\delta'$ " was the non-inferiority margin converted on a probability scale from a point scale  $\delta'$ . All tests were performed at the nominal significance level of  $\alpha=0.025$ .

The overall type I family-wise error rate was preserved at the one-tailed 0.025 nominal level (i.e.,  $\alpha=0.025$ ) by controlling multiplicity over the two co-primary endpoints and the two study targets (superiority and non-inferiority) using the multiple-sequence gatekeeping procedure.

The primary approach for handling missing data was the missing value treatment failure (MVTf) method, which represents a more conservative approach than expectationmaximization (EM) and last observation carried forward (LOCF) methods.

## Results

### Patients

A total of 333 subjects were randomized (IV NAC n=111; ambroxol n=111; placebo n=111), and 288 of them (86.5%) completed the treatment period (Table II and Figure 1).

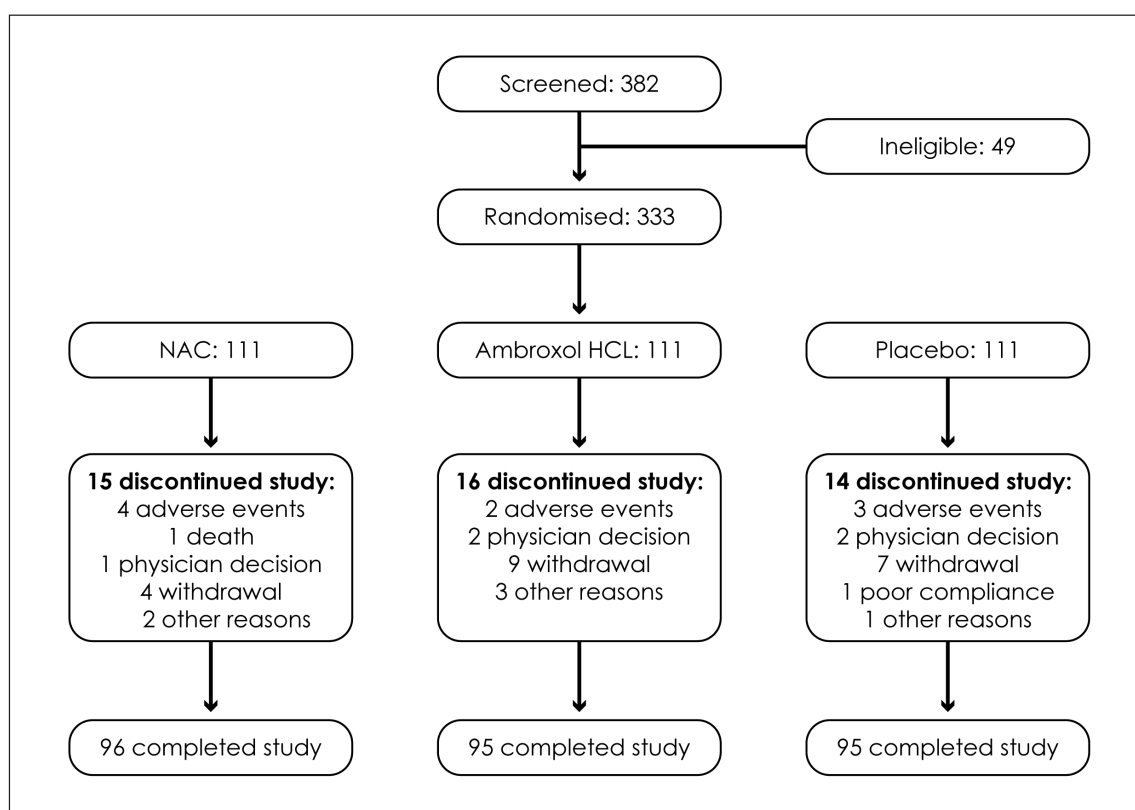
**Table I.** Scoring criteria for efficacy endpoints.

Scoring criteria	Score			
	0	1	2	3
Sputum viscosity	Liquid (normal viscosity)	Fluid (mildly increased viscosity)	Viscous (moderately increased viscosity)	Sticky (severely increased viscosity)
Expectoration difficulty	No difficulty	Mild difficulty	Moderate difficulty	Marked difficulty
Sputum color	Mostly white	Mostly pale yellow	Mostly dark yellow	Very dark yellow/green
Cough	No cough	Sporadic and mild cough	Moderate cough	Severe cough

**Table II.** Demographic and clinical characteristics of the intent to treat population at baseline.

Demographic and clinical characteristics	NAC, n (%)	Ambroxol, n (%)	Placebo, n (%)
Subjects	108	110	110
Male	72 (66.7)	79 (71.8)	78 (70.9)
Female	36 (33.3)	31 (28.2)	32 (29.1)
Age (years), mean ± SD:	64.7 ± 11.65	64.7 ± 12.76	64.5 ± 12.79
Smoking history			
Smokers	22 (20.4)	21 (19.1)	18 (16.4)
Ex-smokers	29 (26.9)	36 (32.7)	40 (36.4)
Never smokers	57 (52.8)	53 (48.2)	52 (47.3)
Weight (kg), mean ± SD:	57.60 ± 11.47	59.71 ± 12.31	59.02 ± 11.03
Height (cm), mean ± SD:	163.57 ± 8.59	165.30 ± 7.42	164.79 ± 8.18
Respiratory conditions*: COPD	61 (55.0)	36 (32.4)	51 (45.9)
Bronchiectasis	23 (20.7)	24 (21.6)	24 (21.6)
Emphysema	17 (15.3)	17 (15.3)	15 (13.5)
Respiratory failure	14 (12.6)	16 (14.4)	18 (16.2)
Asthma	15 (13.5)	10 (9.0)	13 (11.7)
Chronic bronchitis	11 (9.9)	12 (10.8)	14 (12.6)
Other	13 (11.7)	21 (18.9)	17 (15.3)
Sputum viscosity score (mean ± SD)	2.2 ± 0.40	2.3 ± 0.44	2.3 ± 0.46
Expectoration difficulty score (mean ± SD)	2.2 ± 0.43	2.2 ± 0.41	2.3 ± 0.44
Sputum color score (mean ± SD)	1.3 ± 0.82	1.4 ± 0.85	1.5 ± 0.81

NAC, N-acetylcysteine; SD, standard deviation; COPD, chronic obstructive pulmonary disease. \*Respiratory conditions were reported only for the ITT (not for the mITT).



**Figure 1.** Disposition of study subjects. 382 patients were screened at 28 centers in China and 333 patients were randomized to one of the three groups: NAC (600 mg BID), ambroxol hydrochloride (30 mg BID) and placebo.

Of the 45 (13.5%) who did not complete the treatment, 5 (1.5%) were randomized but did not receive any study medication, with 40 subjects discontinuing treatment before completing the planned 7-day treatment period. A total of 328 randomized subjects received at least one dose of study medication (mITT: IV NAC n=108; ambroxol n=110; placebo n=110).

The groups were comparable with respect to baseline characteristics, including baseline sputum viscosity and expectoration difficulty scores (Table II). Most subjects received at least one prior medication (started and stopped prior to the first dose of study medication) (81.1%, ambroxol 82.9%, placebo 83.8%), and almost all subjects received at least one concomitant medication (including those medications stopped on or after the date of the first dose of study medication). The proportions of subjects who had at least one concomitant medication were similar among the treatment groups (100.0% subjects in the NAC group, 99.1% subjects in the ambroxol group, and 99.1% subjects in the placebo group). Permitted concomitant medications included any appropriate treatment for any respiratory tract disease that, in the investigator's opinion, did not interfere with the measurements contributing to the efficacy outcomes. The most commonly reported concomitant medications were glucocorticoids, followed by fluoroquinolones and xanthines (52.3%, 51.1%, and 46.8% of subjects in total, respectively). Concomitant expectorants, antitussive agents, sedatives, and Traditional Chinese Medicine treatments were not permitted during the study.

### Efficacy

Both NAC and placebo groups showed decreasing sputum viscosity scores and decreasing expectoration difficulty scores over time and reached the nadir on day 7 for both primary endpoints. NAC resulted in statistically significant improvements from baseline to day 7 in both sputum viscosity score and expectoration difficulty score vs. placebo (Table III).

There was a statistically significant difference in favor of NAC for mean change from baseline to day 7 in sputum viscosity score [-1.2 in the NAC group and 1.0 in the placebo group, mean (SD) difference between groups of 0.24 (0.763),  $p < 0.001$ ], and in expectoration difficulty score [-1.4 in the NAC group and -1.1 in the placebo group, mean (SD) difference between groups 0.29 (0.783),  $p = 0.002$ ].

A significant difference favoring NAC for both primary endpoints was observed using the more conservative MVTf method for handling data. A significant difference in favor of NAC was also observed for both endpoints using the EM and LOCF methods (sensitivity analysis, data not shown).

NAC showed non-inferiority vs. ambroxol in both mean changes from baseline to day 7 in sputum viscosity score and expectoration difficulty scores.

The mean change from baseline to day 7 in sputum viscosity score was -1.2 in both the NAC and ambroxol groups (Table III), with a one-sided 97.5% CI of the difference between the two groups of 0.43, 1.00, within the interval for non-inferiority (0.39-1).

The mean change from baseline to day 7 in expectoration difficulty score was -1.4 in the

**Table III.** Change in sputum viscosity score or expectoration difficulty score from baseline to day 7 in the intent to treat population.

	NAC (n = 108)	Ambroxol (n = 110)	Placebo (n = 110)	p-value
Sputum viscosity score	-1.2 (0.74)	-1.2 (0.78)	-1.0 (0.78)	NAC vs. ambroxol, $p = 0.002$ NAC vs. placebo, $p < 0.01$ Ambroxol vs. placebo, $p = 0.007$
Expectoration difficulty score	-1.4 (0.78)	-1.3 (0.75)	-1.1 (0.78)	NAC vs. ambroxol, $p < 0.001$ NAC vs. placebo, $p = 0.002$ Ambroxol vs. placebo, $p = 0.018$

Data are mean (standard deviation).

NAC group and -1.3 in the ambroxol group (Table III), with a one-sided 98.75% CI of the difference between the two groups of 0.45, 1.00, within the interval for assessing non-inferiority (0.40-1).

The mean change from baseline to day 7 in both sputum viscosity scores ( $p=0.007$ ) and expectoration difficulty scores ( $p=0.018$ ) was significantly higher for ambroxol than for placebo-treated patients (Table III). No significant differences between ambroxol and placebo were observed for mean change from baseline to day 3 in sputum viscosity score or mean expectoration difficulty score.

Sputum color scores, cough severity scores, and sputum volumes showed a decreasing trend over time. They reached the nadir on day 7 in all groups, with no statistically significant differences observed for these parameters, with the exception of a difference in change from baseline to day 7 in sputum volume in favor of ambroxol vs. placebo ( $p<0.025$ ).

### Safety

A total of 191 (58.2%) subjects experienced at least one AE (Table IV). No difference in the frequency of AEs across treatment groups was observed. Most AEs [138 (42.1%) subjects] were mild in severity [moderate 37 (11.3%) subjects, severe 16 (4.9%) subjects], with a similar proportion of mild, moderate, or severe across groups.

Ten (3.0%) subjects experienced treatment emergent adverse events leading to discontinuation of study medication [NAC  $n=5$  (4.6%); ambroxol  $n=2$  (1.8%); placebo  $n=3$  (2.7%)].

A total of 14 (4.3%) subjects experienced at least one serious AE (SAE) during the study

[NAC  $n=2$  (1.9%); ambroxol  $n=5$  (4.5%); placebo  $n=7$  (6.4%)]. None were considered related to the study medication. One death occurred in the NAC group due to the progression of an underlying disease (cardiac failure), considered to be unrelated to the study medication.

AEs considered by the investigator to be at least possibly related to study medication occurred in 11 (10.2%) subjects in the NAC group, 11 (10.0%) in the ambroxol group, and none reported in the placebo group. The only treatment-related AEs occurring in more than one subject per treatment group were in the ambroxol group (ALT increased in two subjects and dry mouth in three subjects).

No trends were observed over time, and no notable differences among treatment groups in clinical laboratory evaluations, vital signs, ECG parameters, and physical examination findings.

### Discussion

This multicenter randomized study found that NAC 600 mg BID administered IV was significantly superior to placebo and non-inferior to IV ambroxol 30 mg BID in improving sputum viscosity and expectoration difficulty after 7 days of treatment.

Although NAC is widely used as an adjunctive treatment for respiratory diseases, evidence for the efficacy of IV NAC in such diseases is limited<sup>1,2,6</sup>. The use of the intravenous route may be suitable for older, disabled, or seriously ill patients when oral administration is not efficient. Based on a literature search, we identified

**Table IV.** Adverse events recorded during the study. N represents the number (%) experiencing at least one of the events; E represents the Number of events.

	NAC (n = 108), n (%) E	Ambroxol (n = 110), n (%) E	Placebo (n = 110), n (%) E
Subjects with*			
AE	67 (62.0) 157	66 (60.0) 170	58 (52.7) 147
Treatment-related AE	11 (10.2) 14	11 (10.0) 13	0
AE leading to drug discontinuation	5 (4.6) 6	2 (1.8) 2	3 (2.7) 4
Severe AE	5 (4.6) 8	4 (3.6) 4	7 (6.4) 9
SAE	2 (1.9) 2	5 (4.5) 5	7 (6.4) 7
SAE leading to drug discontinuation	1 (0.9) 1	1 (0.9) 1	1 (0.9) 1
Subjects			
Discontinued study			
Due to AE	5 (4.6) 6	2 (1.8) 2	3 (2.7) 4
Due to SAE	1 (0.9) 1	1 (0.9) 1	1 (0.9) 1

AE, adverse event; SAE, serious adverse event. \*Some subjects were in more than one route of the administration group but were analyzed independently.

seven clinical studies to date evaluating the mucolytic and/or expectorant efficacy of IV NAC in patients with respiratory conditions<sup>5,7-12</sup>. Of these, three<sup>5,7,8</sup> were randomized controlled trials (including 111 patients overall), and four<sup>9-12</sup> were uncontrolled studies (with 74 patients overall) (Table V). To our knowledge, the current study represents the first large, randomized, controlled subject- and rater-blinded study to evaluate the mucolytic and expectorant efficacy of IV NAC in patients with acute and chronic respiratory tract disease. Indeed, the results from the current study confirm previous reports from small studies of IV NAC, which showed improvements in sputum viscosity and expectoration difficulty<sup>5,7-12</sup>.

We observed the maximum effect of IV NAC on day 7, while no statistically significant differences between treatment groups were observed on day 3, suggesting that NAC may activate supportive mechanisms different from those of ambroxol, including anti-inflammatory and antioxidant activities, which could be responsible for prolonged protective effects<sup>13</sup>.

The study was a large, randomized, placebo- and active-controlled clinical trial and used an established consistent scoring system for the co-primary endpoints.

### Limitations

However, the study has some limitations. Firstly, it was not double-blind due to the medication

**Table V.** Adverse events recorded during the study. N represents the number (%) experiencing at least one of the events; E represents the Number of events.

Author	Study design	Treatment groups (duration)	Study population	Key findings
<b>Controlled</b>				
Porsio et al <sup>7</sup>	Open-label, crossover, randomized	NAC IV 500-1,000 mg/day Placebo (7 days)	34 patients with pulmonary tuberculosis	Improvement in sputum variables, particularly viscosity and in cough, dyspnea, and nocturnal rest
Grassi et al <sup>5</sup>	Open-label, parallel, randomized*	NAC oral 200 mg TID NAV IM 300 mg BID NAV IV 500 mg OD (6 days)	27 patients with chronic bronchitis/bronchiectasis	Improvement in sputum viscosity and expectoration with significantly better effect and faster onset with IV route
Henneghien et al <sup>8</sup>	Open-label, parallel, randomized	NAC oral 200 mg TID NAC IV 300 mg TID (3-10 days)	50 patients with acute exacerbation of chronic bronchitis	Improvement in sputum variables
<b>Uncontrolled</b>				
Gambini et al <sup>9</sup>	Open-label, single-arm safety evaluation	NAC IV 500 mg BID (12 days)	6 patients with chronic bronchitis	Improvement in expectoration, no significant changes in blood tests or ECGs
Domenichini et al <sup>10</sup>	Open-label, single-arm	NAC IV 500 mg BID w (8 days)	30 patients with acute illness requiring mechanical ventilation	Improvement in sputum viscosity and decrease in airway resistance
Gunella et al <sup>11</sup>	Open-label, single arm	NAC IV 500 mg BID [5-41 days (median 13.5 days)]	28 patients with chronic respiratory impairment	Improvement in sputum variables, pulmonary function tests and arterial blood gases
Balestra et al <sup>12</sup>	Open-label, single-arm	NAC IV 500 mg BID (8-10 days)	10 patients with tracheotomy due to acute respiratory disorders	Confirmation of mucolytic effect on sputum histochemistry

NAC, N-acetylcysteine; TID, three times daily; BID, twice daily; OD, once daily; IV, intravenous, IM, intramuscular.

being identifiable. However, the subject- and rater-blinding should be adequate to avoid bias in the results. Secondly, the study used sputum-related endpoints rather than endpoints directly related to patient outcomes. However, as the study aimed to evaluate the mucolytic and expectorant efficacy of IV NAC, sputum-related endpoints would appear to be appropriate. Lastly, the study included only subjects from China. Although genetic or environmental factors might impact the pathophysiology of respiratory diseases, our results demonstrated the efficacy of NAC in a large sample, suggesting a possible generalization to other populations.

## Conclusions

In conclusion, the study provides new evidence to support the efficacy and safety of IV NAC as a mucolytic and expectorant treatment for patients with respiratory tract disease and abnormal mucus secretion in clinical situations where the IV route is preferred.

## Conflict of Interest

WT, DZ, FW, JFX, JPY, ZPD, XBC, JMQ were investigators of this study. AP received fees or honoraria from Chiesi, AstraZeneca, GSK, Menarini, MSD, Mundipharma, Novartis, Sanofi, Zambon, IQVIA, Avillon, Elpen, Edmon Pharma.

## Acknowledgements

Zhang Xinri (The First Hospital of Shanxi Medical University, Shanxi, Taiyuan, China), AN Dongshan (Jilin Province 'People's Hospital, Changchun, Jilin, China), Xiang PingChao, (Peking University Shougang, Beijing, China), Liang Zong-An (Sichuan University, West China Hospital, Sichuan, Chengdu, China), Guo Shuliang (The First Affiliated Hospital of Chongqing Medical University, Chongqing, China), Gu Wei (Nanjing First Hospital, Nanjing, China), Xiong Shengdao (Tongji Medical College of Huazhong University of Science & Technology, Hubei, Wuhan, China), Yan Chunsong (Nanchang University, The Second Affiliated Hospital, Nanchang, China), Li Dong Ming (Affiliated Hospital of Guangdong Medical University, South Guangdong, Zhanjiang, China), Liu Xiaofang (Beijing Tongren Hospital, Beijing, China), Zhang Min (Shanghai General Hospital, Shanghai, China), He Zhengguang (Suining Central Hospital, Suining, Sichuan, China), Xu Maoye (Inner Mongolia 'People's Hospital, Saihan District, Hohhot, China), Zheng Xiwei (General Hospital of Ningxia Medical University, Ningxia, Yinchuan, China), Yan Zhi Jun (Minhang District Central Hospital, Shanghai, China), Zhao Yinying (Wuxi Peoples' Hospital, Jiangsu, Wuxi, China), Dong Limin (Jiangxi Pingxiang 'People's Hospital, Pingxiang

City, China), Chen Mei (Chengdu Fifth 'People's Hospital, Sichuan, Chengdu, China), Hua Shucheng (The First Hospital of Jilin University, Changchun, Jilin, China), Han Wei (Qingdao Municipal Hospital, Qingdao, China), Xu Xiaomao (Beijing Tongren Hospital, Beijing, China).

Editorial and graphical assistance were provided by Diana Barkley (SEEDHOPPER LLC) and by Laura Brogelli, PhD, Massimiliano Pianta, Valentina Attanasio, and Aashni Shah (Polistudium SRL, Milan, Italy). This assistance was supported by Zambon SpA.

## Authors' Contribution

Study conception: JMQ, data collection: WT, DZ, FW, JFX, JPY, ZPD, XBC, JMQ; manuscript drafting: JMQ; final manuscript revision and approval: WT, DZ, FW, JFX, JPY, ZPD, XBC, AP, JMQ.

## Ethics Approval

At each study site, the protocol and informed consent form were approved by the Ruijin Hospital Ethics Committee, Shanghai, China, on January 24th 2019, and the study was performed according to Good Clinical Practice and in accordance with the Declaration of Helsinki. The study was registered in the ClinicalTrials.gov registry (NCT03843541).

## Informed Consent

All patients released informed consent to participate.

## References

- 1) Micheletto C, Izquierdo JL, Avdeev SN, Rada Escobar RA, Pacheco Gallego MC. N-acetylcysteine as a therapeutic approach to post-COVID-19 pulmonary fibrosis adjunctive treatment. *Eur Rev Med Pharmacol Sci* 2022; 26: 4872-4880.
- 2) Di Marco F, Foti G, Corsico AG. Where are we with the use of N-acetylcysteine as a preventive and adjuvant treatment for COVID-19? *Eur Rev Med Pharmacol Sci* 2022; 26: 715-721.
- 3) Stey C, Steurer J, Bachmann S, Medici TC, Tramèr MR. The effect of oral N-acetylcysteine in chronic bronchitis: a quantitative systematic review. *Eur Respir J* 2000; 16: 253-262.
- 4) Cazzola M, Calzetta L, Page C, Jardim J, Chuchalin AG, Rogliani P, Matera MG. Influence of N-acetylcysteine on chronic bronchitis or COPD exacerbations: a meta-analysis. *Eur Respir Rev* 2015; 24: 451-461.
- 5) Grassi C, Morandini GC, Frigerio G. Clinical evaluation of systemic acetylcysteine by different routes of administration. *Curr Ther Res Clin Exp* 1973; 15: 165-179.
- 6) Santus P, Danzo F, Zuffi A, Pini S, Saad M, Visconti A, Radovanovic D. Oxidative stress and vi-



- ral Infections: rationale, experiences, and perspectives on N-acetylcysteine. *Eur Rev Med Pharmacol Sci* 2022; 26: 8582-8590.
- 7) Porsio A, Borgia M. Sull'impiego dell'acetilcisteina per via endovenosa: esperienze cliniche di tollerabilità e di attività [Intravenous administration of acetylcysteine: clinical experience with its tolerance and action]. *Clin Ter* 1970; 55: 123-132.
  - 8) Henneghien C, Remacle P, Bruart J. Study of the N-acetylcysteine (NAC) by oral or intravenous administration in chronic bronchitis. *Curr Ther Res* 1983; 34: 750-756.
  - 9) Gambini A, Gazzaniga A, Lodola E. Valutazione biochimica ed elettrocardiografica dell'acetilcisteina per via endovenosa [Biochemical and electrocardiographic evaluation of acetylcysteine in intravenous administration]. *Clin Ter* 1971; 57: 357-368.
  - 10) Domenichini E, Carbonera D, Bobbio-Pallavicini F. Effetto mucolitico e terapeutico dell'acetilcisteina par via endovenosa. Esperienze in rianimazione [Mucolytic and therapeutic effects of acetylcysteine administered by the intravenous route. Experiences in resuscitation]. *Minerva Anesthesiol* 1973; 39: 7-12.
  - 11) Gunella G, Melica A, Fabbri M, Brignani F. Effetti dell'acetilcisteina per via endovenosa, associata ad altre terapie, in broncopneumopatici cronici portatori di insufficienza ventilatoria globale o parziale. *Minerva Pneum* 1973; 12: 142-149.
  - 12) Balestra L, Mereto GC, Aliotta P. L'acetilcisteina per via venosa in rianimazione [Intravenous acetylcysteine resuscitation]. *Minerva Anesthesiol* 1971; 37: 394-398.
  - 13) Žuža O, Minić R, Kotur-Stevuljević J, Žujović D, Đorđević B, Ilić A. A combination of N-acetyl cysteine and propolis attenuates oxidative-inflammatory parameters during COPD exacerbation. *Eur Rev Med Pharmacol Sci* 2022; 26: 2467-2477.