CURRENT THERAPEUTIC RESEARCH®
VOLUME 65, NO. 6, NOVEMBER/DECEMBER 2004

Efficacy of Concomitant Administration of Clarithromycin and Acetylcysteine in Bronchiolitis Obliterans in Seventeen Sulfur Mustard–Exposed Patients: An Open-Label Study

Mostafa Ghanei, MD, Kamran Abolmaali, MD, and Jafar Aslani, MD

Research Center of Chemical Injuries, Baqiyatallah Medical Sciences University, Tehran, Iran

ABSTRACT

Background: Victims of sulfur mustard (SM) gas exposure experience different types of chronic pulmonary disease, manifested as cough, sputum production, and dyspnea. Conventional therapies (eg, immunosuppressive drugs, corticosteroids) have not been effective in these patients.

Objective: This study was carried out to determine the efficacy of concomitant administration of the macrolide clarithromycin and the mucolytic agent acetylcysteine in the treatment of bronchiolitis obliterans in SM-exposed patients.

Methods: This open-label clinical study was conducted at the Research Center of Chemical Injuries, Baqiyatallah Medical Sciences University, Tehran, Iran. Clarithromycin and acetylcysteine were administered concomitantly for 6 months to male SM-exposed patients with chronic bronchitis and bronchiolitis obliterans who were nonresponsive to conventional treatments. Efficacy analysis included symptom assessment and pulmonary function tests (forced expiratory volume in 1 second [FEV_1], forced vital capacity [FVC], and FEV_1/FVC ratio) using spirometry, performed at baseline and after 2 and 6 months of treatment.

Results: Seventeen male patients (mean [SD] age, 38.3 [5.3] years [range, 31–50 years]; mean [SD] body weight, 77.9 [15.7] kg) were included in the study. Cough and sputum production were each found in 10 of 17 patients (58.8%) at baseline and were improved in all 10 patients after the administration of clarithromycin and acetylcysteine. FEV_1 and FVC also were improved, by mean (SD) 10.6% (9.7%) ($P < 0.001$ vs baseline) and 12.9% (13.6%) ($P = 0.001$ vs baseline). No significant change in FEV_1/FVC ratio was found.

Conclusions: In this study of concomitant administration of clarithromycin and acetylcysteine for the treatment of bronchiolitis obliterans in SM-exposed patients, symptoms and pulmonary function were improved. These results may have been related to the therapeutic effects of a macrolide antibiotic on chronic bronchitis and bronchiolitis obliterans in these patients. Based on the results of this study, we recommend this treatment for chemical warfare victims with

Accepted for publication October 5, 2004.
doi:10.1016/j.curtheres.2004.12.001
0011-393X/04/$19.00

recurrent exacerbation of bronchitis who do not respond to conventional treatment. (*Curr Ther Res Clin Exp.* 2004;65:495–504)

Key words: sulfur mustard, chemical warfare, chronic bronchitis, bronchiolitis, clarithromycin, acetylcysteine.

INTRODUCTION

Macrolides have been shown to have some anti-inflammatory effects that may contribute to their therapeutic efficacy. Newer macrolides (eg, tacrolimus), which have mainly immunosuppressive efficacy, are used in the management of bronchiolitis obliterans after lung transplantation.[1,2] The beneficial effects of the mucolytic agent acetylcysteine in the treatment of chronic bronchitis have also been noted.[3] Acetylcysteine reduces the elasticity of mucous secretions by reducing disulfide bonds.[3] In addition, acetylcysteine is a potent antioxidant that acts as a prodrug for cysteine and glutathione.[4]

During the Iran-Iraq War (1980–1988), thousands of Iranians were exposed to sulfur mustard (SM) gas.[5] Most survivors (~60%) have chronic pulmonary disease with different degrees of impairment.[6] Chronic bronchitis, bronchiectasis, and pulmonary fibrosis have been reported as late complications of SM exposure.[6] Two studies[7,8] have shown that bronchiolitis obliterans is another complication in this population. As in lung transplant recipients,[9,10] conventional drugs (eg, immunosuppressive drugs, corticosteroids) have been shown to be ineffective in the treatment of bronchiolitis obliterans in SM-exposed patients.[7]

In this study, we examined the efficacy of concomitant administration of the macrolide clarithromycin and acetylcysteine in the treatment of bronchiolitis obliterans in SM-exposed patients.

PATIENTS AND METHODS

This open-label clinical study was conducted at the Research Center of Chemical Injuries, Baqiyatallah Medical Sciences University, Tehran, Iran, between March 2001 and September 2002. The study was approved by the ethics committee at the university.

Inclusion and Exclusion Criteria

Men with chronic bronchitis and bronchiolitis obliterans who were exposed to SM between 1985 and 1988 (during the Iran-Iraq War) and were receiving long-term follow-up care were enrolled. Additional inclusion criteria were as follows:

1. Documented chemical exposure based on official certification from the Bonyad Mostazafan & Janbazan Foundation, Tehran
2. Military/medical records showing that the patient was transferred to a local military hospital after attack, where, based on signs and symp-

toms and the use of special kits by military services, the types of chemical agents to which the patient was exposed were determined, after which the patient received care according to standard protocols determined by military health services

3. Pathologic findings compatible with bronchiolitis on transbronchial lung biopsy
4. Radiologic evidence of bronchiolitis obliterans on high-resolution computed tomography (HRCT) of the chest (described later) (ie, the presence of air trapping in ≥25% of the cross-sectional areas of an affected lung on ≥1 scan level and with ≥3 months free of immunosuppressive drug therapy)
5. Nonresponse to high-dose bronchodilator therapy (salmeterol, 25 µg, 4 puffs BID) and inhaled corticosteroid therapy (fluticasone propionate, 125 mg, 2 puffs BID) ≥3 months before the study.

Each HRCT examination was conducted using the Hi Speed Advantage Scanner (General Electric Medical Systems, Milwaukee, Wisconsin) and consisted of five 1.0-mm collimation images obtained during deep inspiration and full expiration, with the patient supine. Images were obtained at the levels of the aortic arch, midway between the aortic arch and the tracheal carina, midway between the tracheal carina and the right hemidiagram, and 1 cm superior to the right hemidiagram. IV contrast material was not administered. All images were reconstructed using a high spatial resolution algorithm and displayed at standard (level, –700 Hansfield units [HU]; width, 1500 HU) and narrow (level, 700 HU; width, 1000 HU) lung window settings. The HRCT scans were reviewed by a radiologist and a pulmonologist, who knew only the age, sex, and the SM exposure history of the patients. The interobserver agreement for air trapping and mosaic parenchymal attenuation was registered. The expiratory images were assessed for the presence and lobar distribution of air trapping, defined as alteration of normal anteroposterior lobar attenuation gradients and/or lack of homogeneous increase in pulmonary attenuation resulting in persistent areas of decreased attenuation. The extent of air trapping was qualified and classified using the same system as defined for hyperlucent regions on inspiratory images. The presence of air trapping was considered to be an indication of bronchiolitis obliterans only if it exceeded 25% of the cross-sectional area of an affected lung on ≥1 scan level. Expiratory images displayed at standard and narrow window settings were directly compared to determine differences in the conspicuity of air trapping.

Patients with the following characteristics were excluded from the study:

1. Radiographic evidence of pneumonia, active tuberculosis, lung cancer, or an infection that necessitated the use of a concomitant antibiotic
2. Cigarette smoking or occupational exposure to toxic agents in the previous 10 years

3. History of hypersensitivity to beta-lactam or macrolide antibiotics
4. Treatment with a systemic antibiotic, any investigational medicine, or a long-acting injectable antibiotic within 7 days, 4 weeks, and 6 weeks, respectively, before the study, or the use of concomitant theophylline or carbamazepine (unless the serum concentration was regularly monitored)
5. Exacerbation phase of respiratory complications.

Written informed consent was obtained from all eligible patients before the study.

Study Drug Administration and Efficacy Assessments

The treatment regimen consisted of clarithromycin* (500-mg tablet QD) and acetylcysteine effervescent† (600-mg tablet QD), both given for 6 months. Measurements for the therapeutic efficacy of this regimen were evaluated before, during, and after the treatment period. Clinical and paraclinical parameters monitored were the severity of cough and sputum production, and changes in pulmonary function. Bronchodilator and inhaled corticosteroid therapy were continued throughout the study.

Pulmonary function tests (PFTs) were performed at baseline and after 2 and 6 months of treatment, using spirometry (MasterScope PC, Jaegger, Würzburg, Germany), based on American Thoracic Society criteria.[11] Briefly, this testing involved measuring forced expiratory volume in 1 second (FEV_1) and forced vital capacity (FVC) and calculating the FEV_1/FVC ratio. The tests were performed by experienced respiratory technicians under the direction of licensed physicians, using a standard spirometer. The technicians were blinded to the study. All of the patients were experienced with PFTs.

While seated with a nose clip in place, patients were asked to perform ≥3 forced expiratory maneuvers, with verbal encouragement to blow maximally throughout expiration until they felt there was no air to expel. Both the patient and the technician received visual feedback from a monitor during the test, which was repeated until 3 technically satisfactory curves with reproducible contours were obtained. All of the indices used for the analysis were derived from the maneuver with the largest FVC.

Statistical Analysis

Data are expressed as mean (SD). The Student *t* test and SPSS version 11.5 (SPSS Inc., Chicago, Illinois) was used to calculate the differences between baseline and end-of-study results. Differences of $P < 0.05$ were considered statistically significant.

*Trademark: Klacid® (Abbott GmbH, Wiesbaden, Germany).
†Trademark: Flumacil® (Zambon Group, Vicenza, Italy).

RESULTS

Seventeen male patients (mean [SD] age, 38.3 [5.3] years [range, 31–50 years]; mean [SD] body weight, 77.9 [15.7] kg) were included in the study. All patients were treated with the 2 study drugs and completed the study. In all of the patients, the chemical insult occurred between 1985 and 1988. None of the patients were active smokers at the time of the study; however, 4 patients (23.5%) had a history of smoking >10 years before the study. In all patients, findings on HRCT were consistent with a diagnosis of bronchiolitis obliterans (air trapping on expiration, 13 patients [76.5%]; bronchiectasis, 12 patients [70.6%]; and inspiratory mosaic parenchymal attenuation, 10 patients [58.8%]) (**Table I**).

Although the precise amount of exposure to chemical agents could not be determined on the battlefield, according to each patient's medical records, the patients were symptomatic at the time of the chemical attack, but symptoms of the acute phase were transient, resolving after the patients were transported for medical care.

At baseline, cough, sputum production, and dyspnea were the most common symptoms, each occurring in 10 patients (58.8%). However, because dyspnea is a highly subjective symptom and is difficult to quantify, cough and sputum production were used to assess efficacy. The prevalence of cough and sputum production before and after treatment are shown in **Table II**.

At baseline, mean (SD) FEV_1 was 2.57 (0.98) L and mean (SD) FVC was 3.31 (0.92) L. FEV_1/FVC ratio was 75.8%. After 2 months of therapy, FEV_1, FVC, and FEV_1/FVC ratio were calculated as 2.91 (0.96), 3.90 (0.90), and 74.1%, respectively. FEV_1 improved by mean (SD) 10.6% (9.7%) ($P < 0.001$ vs baseline) and FVC improved by mean (SD) 12.9% (13.6%) ($P = 0.001$ vs baseline). The distributions of percentage improvement from baseline in FEV_1 and FVC after 6 months of treatment are shown in **Figures 1** and **2**, respectively. No significant change in FEV_1/FVC ratio was found after treatment.

DISCUSSION

Chronic bronchitis affects a significant proportion (~60%) of chemical warfare victims.[6] This condition is characterized by frequent, recurrent, acute

Table I. Baseline findings on high-resolution computed tomography (HRCT) of the chest in sulfur mustard gas–exposed patients (N = 17).

HRCT Finding	Prevalence, No. (%)
Air trapping on expiration	13 (76.5)
Bronchiectasis	12 (70.6)
Inspiratory mosaic parenchymal attenuation	10 (58.8)
Interlobular septal thickening	5 (29.4)

Table II. Prevalence (no. [%]) of cough and sputum production before and after 6 months of concomitant administration of clarithromycin and acetylcysteine in sulfur mustard gas–exposed patients with bronchiolitis obliterans who were symptomatic at baseline (n = 10/17).

	Cough		Sputum Production	
Severity	Before Treatment	After Treatment	Before Treatment	After Treatment
A lot every day	9 (90.0)	3 (30.0)	8 (80.0)	4 (40.0)
A little every day	1 (10.0)	6 (60.0)	0 (0.0)	4 (40.0)
A little on most days	0 (0.0)	1 (10.0)	0 (0.0)	0 (0.0)
A little occasionally	0 (0.0)	0 (0.0)	1 (10.0)	0 (0.0)
Rarely	0 (0.0)	0 (0.0)	1 (10.0)	2 (20.0)

bacterial infection exacerbations, which are an important cause of morbidity in this patient population. Infectious exacerbations of chronic bronchitis are frequently induced by pathogens that reside in the respiratory tract. Risk factors for chronic bronchitis include factors that alter normal host defense mechanisms in these patients (eg, mucociliary dysfunction, accumulation of secretions, bacterial colonization). Clarithromycin is an

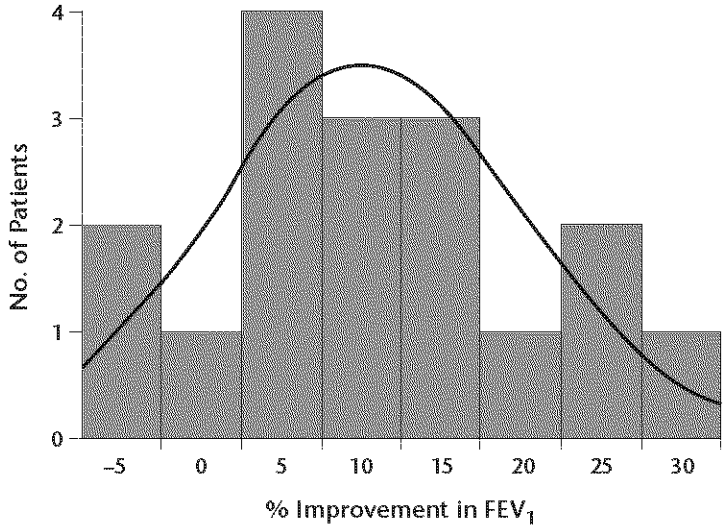

Figure 1. Distribution of patients according to percentage improvement in forced expiratory volume in 1 second (FEV_1) after 6 months of concomitant administration of clarithromycin and acetylcysteine in sulfur mustard gas–exposed patients with bronchiolitis obliterans (N = 17). Mean (SD) percentage improvement was 10.6% (9.7%) ($P < 0.001$ vs baseline). Curve = normal distribution.

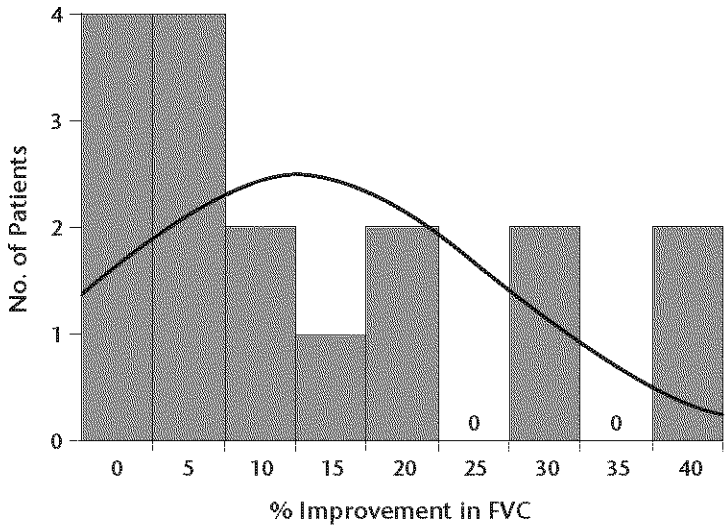

Figure 2. Distribution of patients according to percentage improvement in forced vital capacity (FVC) after 6 months of concomitant administration of clarithromycin and acetylcysteine in sulfur mustard gas–exposed patients with bronchiolitis obliterans (N = 17). Mean (SD) percentage improvement was 12.9% (13.6%) ($P = 0.001$ vs baseline).

advanced-generation macrolide with potent antimicrobial activity against the common gram-negative and gram-positive microorganisms that exacerbate chronic bronchitis.

The mechanism of macrolide antibiotic action is based on inhibition of bacterial protein synthesis by interaction with 23S rRNA in the central loop of the peptidyltransferase center, as well as with specific ribosomal proteins found in the same region of the ribosome. Macrolides are best known as anti-infective agents, but they also exert other important pharmacologic effects, such as immunosuppression and immunomodulation.

In this study, the combination of clarithromycin and acetylcysteine decreased the prevalence of cough and sputum production, thereby effectively treating exacerbations of SM–induced bronchiolitis. These findings are compatible with previous reports regarding the efficacy of combination therapy with clarithromycin and acetylcysteine in chronic bronchitis.

Several double-blind trials have demonstrated that symptoms of bronchitis and chronic obstructive pulmonary disease were improved by the use of acetylcysteine and that exacerbations of these diseases were prevented.[12–15] An additional study showed that pulmonary tissue may be protected by the antioxidant activity of acetylcysteine.[16]

A number of randomized, parallel-group studies[17–21] have used clarithromycin for the treatment of acute bacterial exacerbations of chronic bronchitis.

Macrolide antibiotics have been shown to decrease sputum volume and increase sputum elasticity in patients with diffuse panbronchiolitis, chronic bronchitis, and bronchiectasis.[22] Numerous studies have shown that macrolides decrease interleukin (IL)-8–induced neutrophil accumulation; suppress IL-6 expression; inhibit expression of intercellular adhesion molecule 1; decrease superoxide formation in neutrophils; decrease lipopolysaccharide-induced levels of tumor necrosis factor; and decrease lipopolysaccharide-induced neutrophil migration into bronchial tissues, neutrophil adhesion, and vascular leakage.[18,19,23,24] Two case-report studies[7,25] have shown that bronchiolitis obliterans is the main pulmonary abnormality after exposure to SM and that pulmonary function abnormalities were not reversed by treatment with corticosteroids or bronchodilators.

Fifteen years after exposure to SM as a chemical warfare agent, the patients in our study were experiencing the chronic and often disabling respiratory symptoms of dyspnea, cough, and sputum production. Most of these patients had unremarkable plain chest radiographs; however, the findings of bronchiectasis, air trapping on expiration, and inspiratory mosaic parenchymal attenuation on HRCT pointed to the diagnosis of bronchiolitis obliterans in this patient population.

CONCLUSIONS

In this study of concomitant administration of clarithromycin and acetylcysteine for the treatment of bronchiolitis obliterans in SM-exposed patients, symptoms and pulmonary function were improved. These results may have been related to the therapeutic effects of a macrolide antibiotic on chronic bronchitis and bronchiolitis in these patients. Based on the results of this study, we recommend this treatment for chemical warfare victims with recurrent exacerbation of bronchitis who do not respond to conventional treatment. However, this study must be regarded as a first step in examining the efficacy of these drugs for the treatment of individuals exposed to chemical warfare agents. To further assess our conclusions, larger, double-blind, case-control studies are required.

REFERENCES

1. Cairn J, Yek T, Banner NR, et al. Time-related changes in pulmonary function after conversion to tacrolimus in bronchiolitis obliterans syndrome. *J Heart Lung Transplant.* 2003;22:50–57.
2. Fieguth HG, Krueger S, Wiedenmann DE, et al. Tacrolimus for treatment of bronchiolitis obliterans syndrome after unilateral and bilateral lung transplantation. *Transplant Proc.* 2002;34:1884.
3. Fuloria M, Rubin BK. Evaluating the efficacy of mucoactive aerosol therapy. *Respir Care.* 2000;45:868–873.
4. Ruffmann R, Wendel A. GSH rescue by *N*-acetylcysteine. *Klin Wochenschr.* 1991;69:857–862.
5. New York Security Council of United Nations (NYSCUN). Report of specialists appointed by the secretary general to investigate allegations by the Islamic Republic

of Iran concerning the use of chemical weapons. New York: NYSCUN; 1986. Document no. S/16433/1986.

6. Emad A, Rezaian GR. The diversity of the effects of sulfur mustard gas inhalation on respiratory system 10 years after a single, heavy exposure: Analysis of 197 cases. *Chest.* 1997;112:734–738.
7. Thomason JW, Rice TW, Milstone AP. Bronchiolitis obliterans in a survivor of a chemical weapons attack. *JAMA.* 2003;290:598–599.
8. Ghanei M, Mokhtari M, Mir Mohammad M, Aslani J. Bronchiolitis obliterans following exposure to sulfur mustard: Chest high resolution computed tomography. *Eur J Radiol.* 2004;52:164–169.
9. Laohaburanakit P, Chan A, Allen RP. Bronchiolitis obliterans. *Clin Rev Allergy Immunol.* 2003;25:259–274.
10. Chan A, Allen R. Bronchiolitis obliterans: An update. *Curr Opin Pulm Med.* 2004;10:133–141.
11. American Thoracic Society. Standardization of spirometry, 1994 update. *Am J Respir Crit Care Med.* 1995;152:1107–1136.
12. Boman G, Bäcker U, Larsson S, et al. Oral acetylcysteine reduces exacerbation rate in chronic bronchitis: Report of a trial organized by the Swedish Society for Pulmonary Diseases. *Eur J Respir Dis.* 1983;64:405–415.
13. Long-term oral acetylcysteine in chronic bronchitis. A double-blind controlled study. *Eur J Respir Dis.* 1980;111(Suppl):93–108.
14. Grandjean EM, Berthet P, Ruffmann R, Leuenberger P. Efficacy of oral long-term *N*-acetylcysteine in chronic bronchopulmonary disease: A meta-analysis of published double-blind, placebo-controlled clinical trials. *Clin Ther.* 2000;22:209–221.
15. van Schayck CP, Dekhuijzen PN, Gorgels WJ, et al. Are anti-oxidant and anti-inflammatory treatments effective in different subgroups of COPD? A hypothesis [published correction appears in *Respir Med.* 1999;93:224]. *Respir Med.* 1998;92: 1259–1264.
16. Spiritue E, Devcich K, Dmec J, Hallwae R. Clarithromycin vs. cefaclor for acute bacterial exacerbations of bronchitis. In: *Proceedings of the First International Conference on the Macrolides, Azalides, and Streptogramine.* January 1992; Santa Fe, New Mexico. Abstract 239.
17. Aldons PM. A comparison of clarithromycin with ampicillin in the treatment of outpatients with acute bacterial exacerbation of chronic bronchitis. *J Antimicrob Chemother.* 1991;27(Suppl A):101–108.
18. Anzueto A, Jenkinson SG, for the Clarithromycin Chronic Bronchitis Study Group. Safety and efficacy of clarithromycin vs cefuroxime axetil in the treatment of acute bacterial exacerbation of chronic bronchitis. *Immunol Infect Dis (Oxford).* 1993;3:231–237.
19. Bachand RT Jr. Comparative study of clarithromycin and ampicillin in the treatment of patients with acute bacterial exacerbations of chronic bronchitis. *J Antimicrob Chemother.* 1991;27(Suppl A):91–100.
20. Guay DR, Craft JC. Comparative safety and efficacy of clarithromycin and ampicillin in the treatment of out-patients with acute bacterial exacerbation of chronic bronchitis. *J Intern Med.* 1992;231:295–301.
21. Neu HC, Chick TW. Efficacy and safety of clarithromycin compared to cefixime as outpatient treatment of lower respiratory tract infections. *Chest.* 1993;104:1393–1399.
22. Tamaoki J, Takeyama K, Tagaya E, Konno K. Effect of clarithromycin on sputum production and its rheological properties in chronic respiratory tract infections. *Antimicrob Agents Chemother.* 1995;39:1688–1690.

23. Tamaoki J, Sakai N, Tagaya E, Konno K. Macrolide antibiotics protect against endotoxin-induced vascular leakage and neutrophil accumulation in rat trachea. *Antimicrob Agents Chemother.* 1994;38:1641–1643.
24. Ras GJ, Anderson R, Taylor GW, et al. Clindamycin, erythromycin, and roxithromycin inhibit the proinflammatory interactions of *Pseudomonas aeruginosa* pigments with human neutrophils in vitro. *Antimicrob Agents Chemother.* 1992;36:1236–1240.
25. Dompeling E, Jobsis Q, Vandevijver NM, et al. Chronic bronchiolitis in a 5-year-old child after exposure to sulphur mustard gas. *Eur Respir J.* 2004;23:343–346.

Address correspondence to: Dr. Mostafa Ghanei, Research Center of Chemical Injuries, Baqiyatallah Medical Sciences University, PO Box 19945-546, Mollasadra Street, Tehran, Iran. E-mail: m.ghanei@bmsu.ac.ir