Mustard lung secrets: Long term clinicopathological study following mustard gas exposure

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Received 18 December 2005; accepted 20 April 2006

Abstract

Considering the indefinite nature of lung pathology in patients exposed to sulfur mustard (SM) many years after exposure, we conducted this study to document and quantify lung disease in this setting.

In a cross-sectional study, we selected 23 patients exposed to SM gas approximately 14 years ago during the Iran–Iraq war (1980–1988). We studied their clinical history, physical examination, pulmonary function test (PFT), high-resolution computed tomography scan (HRCT) of the chest, bronchoscopy, and bronchoalveolar lavage (BAL) sampling, and transbronchial lung biopsies. Other potential causes of lung disease, including smoking of cigarettes, were excluded.

All 23 patients were symptomatic with cough, dyspnea, and/or felt tight in the chest. All of them had significant air trapping in HRCT and a marked increase of residual volume in PFT. The most common inflammatory cell in BAL fluid was neutrophil (88%). Of the 23 cases, there was sufficient tissue for detailed evaluation in 22. Histologically, 11 cases showed airway epithelial injury, and nine of the 14 lung biopsies with alveoli had histopathological changes diagnosable as organizing pneumonia (OP) or bronchiolitis obliterans OP (BOOP). Two out of 14 cases showed changes suggestive of OP.

Inhalation of SM can lead to persistent and clinically significant lung disease, including bronchial mucosal injury and OP, many years after exposure.

Keywords: Sulfur mustard; Chronic lung disease; Bronchiolitis obliterans organizing pneumonia; Organizing pneumonia; Chemical Warfare

Introduction

Sulfur mustard (SM) gas was a cause of casualties in the second half of World War I, and was used as a vesicant chemical warfare agent in the Iran–Iraq War (1981–1989) against Iranians [21]. This agent is capable of producing severe chemical injuries primarily in three major organs: skin, eyes, and lungs. Few studies have been done with regard to chronic respiratory effects, and there is controversy about their results. However, with the introduction of advantages in diagnosis criteria, some studies could benefit from these methods, and formerly erroneous diagnoses can be corrected in complementary investigations. They are useful in clinical
practice, leading to reasonable agreement between those using it.

One of these studies showed that patients developed a variety of chronic pulmonary diseases, including asthma, chronic bronchitis, bronchiectasis, and interstitial lung diseases (ILD, including pulmonary fibrosis [10,11]). Another study, based on high-resolution computed tomography (HRCT), reported 50 cases, suggesting that bronchial wall thickening, ILD, and emphysema are common sequelae of SM injury [3]. In a recent study using HRCT, air trapping was reported as common, which suggested bronchiolitis obliterans organizing pneumonia (BOOP) as an underlying disease process [15]. Two recent case reports also described BOOP following exposure to SM [8,23].

Due to lack of large-scale clinicopathological studies, we evaluated chronic effects of SM by a combination of chest HRCT, pulmonary function test (PFT), and transbronchial lung biopsy (TBLB). The patients in this study were exposed to SM about 14 years ago and are still suffering from chronic respiratory diseases.

Materials and methods

Thousands of Iranians were exposed to mustard gas during the Iran–Iraq war (1980–1988) [21]. A governmental association responsible for supporting the war victims and veterans’ affairs has gathered filed records of almost all the known victims who have regular biannual referrals to special clinics held by that association. About 14 years after exposure, in September 2002, the association called on all provinces to refer all suspect cases suffering from severe respiratory disorder for a complete checkup and management arranged for treatment of central airway stenosis. Between 2002 and 2003, among them, 23 patients were enrolled in our study. Examination of the patients, imaging, and PFTs were done at Research Center for Chemical Gas Injuries, Baqiyatallah Medical Sciences University, Tehran, Iran. The histopathologic study was performed and the slides reviewed in Tehran and at Massachusetts General Hospital, Harvard Medical School, Boston, Massachusetts.

Inclusion criteria

1) Documentation of chemical exposure by military health services at the time of exposure. In this study, exposure is defined as a single high-dose exposure to SM. Patients had been transferred to local military hospitals. Chemical reaction kits had been used to confirm the presence of SM at the time of exposure. A previously validated questionnaire was used to identify the patients so exposed [14].

2) Onset of respiratory symptoms immediately after exposure to SM and continuation with no symptom-free period.

3) Radiographic evidence of expiratory air trapping >25% [16] and mosaic parenchymal attenuation. The criteria used for the presence of air trapping were alteration of normal anterior-posterior lobar attenuation gradients and/or lack of homogeneous increase in lung attenuation, resulting in persistent areas of decreased attenuation. The mosaic parenchymal pattern was defined as areas of heterogeneous lung attenuation in a lobular or multilobular distribution in expiratory phase [22].

4) Bronchoscopic performed and bronchial mucosa and lung parenchymal tissue obtained by bronchoscopic biopsy available to specifically look for diseases related to toxic exposure.

Exclusion criteria

Patients were excluded from the study and did not undergo lung biopsy if they had a positive history for the following:

1) History of smoking.

2) History of lung disease before exposure to SM.

3) Any occupational history of toxic fume exposure or occupational risk factors that could lead to ILD.

4) Any associated chronic disease (such as heart failure or connective tissue disease) with potential pulmonary involvement.

5) History of treatment with drugs that may cause acute pneumonitis or ILD as a side effect.

Pulmonary function tests

We evaluated the lung function of all patients by spirometry using the criteria of the American Thoracic Society [1]. We measured forced vital capacity (FVC) and forced expiratory volume in 1 second (FEV1) under the direction of physicians. TLC and RV were measured by the helium dilution method with a Master Screen apparatus (Jaeger, Hochberg, Germany), and diffusing capacity of the lung for carbon monoxide (DLco) by the single breathholding helium dilution method [7,19] using an infrared analyzer (SensorMedics Corporation, Anaheim, California, USA, Vmax software version 04-4), which utilizes methane as inert tracer gas.

The patients were seated with a nose clip in place and were asked to perform at least three forced expiratory manoeuvres. They were told to continue to blow maximally until they felt there was no air to expel. Both the patients and the technician received a visual feedback from a monitor during the test, which was repeated until three technically satisfactory curves with reproducible contour were obtained. All the indices used for the analysis were derived from the same manoeuver, which was the one with the largest FVC.
High-resolution computed tomography scan

HRCT images were performed on an electron beam CT machine. All 1.5 mm sections were obtained at full inspiration, with the patient in both supine and prone position. In all cases, additional expiratory films and prone sections were taken. Images were photographed at window settings appropriate for viewing the lung parenchyma (−500 HU center, 1500 HU width). The scans, together with plain chest radiographs, were assessed by a pulmonary radiologist. The presence or absence of patchy ground glass pattern, reticular pattern, emphysema, honeycombing, airways distortion, and bronchial wall thickening scans was recorded.

Transbronchial lung biopsy

All 23 patients underwent bronchoscopy followed by biopsies of the respiratory mucosa and lung parenchyma. Biopsy specimens were obtained from all three lobes on the right side or from the upper and lower lobes of the left lung. However, the site of biopsies could not be exactly matched with chest HRCT findings site. The biopsies were processed, sectioned, and stained with hematoxylin and eosin, Masson’s trichrome, and elastic-van Gieson stains. The slides were reviewed by pulmonary pathologists in Tehran, Iran, and at the Massachusetts General Hospital, Boston, Massachusetts.

Bronchoscopy and bronchoalveolar lavage sampling

Bronchoalveolar lavage (BAL) was performed in all subjects using a flexible fiber-optic bronchoscope (Olympus BF1T, Tokyo, Japan). The upper respiratory tract was anesthetized with 2% lidocaine. Atropine (0.75 mg intramuscularly) was administered before the procedure. Supplemental oxygen was given throughout the procedure, and the oxygen saturation was monitored by continuous pulse oxymeter. The bronchoscope was wedged for lavage in the middle lobe segmental bronchus, and four 60-ml aliquots of sterile physiological saline solution warmed to 37 °C were infused. The fluid was immediately recovered by gentle suction after each instillation.

Results

All the victims were men, with a mean age of 39.4 years (range 35–52 years).

Pulmonary function tests

The PFT results of the cases can be seen in Table 1. The residual volume (RV) is markedly increased in all cases. The result from DLCO was normal in all cases (mean ± SD was 98 ± 3.8).

Chest HRCT

For controlling hyperventilation-induced parenchymal attenuation, chest HRCTs were reviewed, and none of the cases was in hyperventilation state during the taking of chest HRCTs. The chest HRCT results of the cases are illustrated in Table 2. Air trapping is the most common finding in these results. A typical HRCT image of one of the patients showing significant air trapping can be seen in Fig. 1.

Bronchoscopy

The bronchoscopic appearance of airway mucosa was that of a combination of erythema, chronic inflammatory changes, and mucosal thickening in all of the cases.

Histopathology

The percentages of inflammatory cell count in BAL fluid were 88, 16, 62, and 0% for neutrophils, lymphocytes, macrophages, and eosinophils, respectively. Of the 23 cases, there was sufficient tissue for detailed evaluation in 22. Eleven had pulmonary alveolar parenchyma, three had alveolar parenchyma and respiratory mucosa, and eight had only respiratory mucosa. Nine of the 14 lung biopsies with alveoli had histopathological changes diagnosable as organizing pneumonia (OP) or BOOP. The parenchymal involvement

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<th>Table 1. Pulmonary function data of 23 patients 14 years after exposure to sulfur mustard</th>
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<th>Table 2. Chest HRCT findings of 23 patients 14 years after exposure to sulfur mustard</th>
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<td>Chest HRCT Findings</td>
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</tr>
<tr>
<td>Air trapping</td>
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<tr>
<td>Bronchiectasis</td>
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<td>Mosaic parenchymal attenuation</td>
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<td>Interlobular septal thickening</td>
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<td>Bronchial wall thickening</td>
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was that of myxoid tufts of organizing connective tissue within the alveolar ducts and alveoli with incorporation into alveolar walls (Figs. 2 and 3). Also present was alveolar wall expansion by a mild mononuclear cell infiltrate, edema, and delicate interstitial thickening. Tichrome stains emphasized the nature of fibrosis to be young and myxoid, with only thin delicate strands of collagen fibers (Fig. 4). Slight intra-alveolar fibrin mixed with mononuclear cells and was present in most cases, but was most noticeable at the periphery of the foci of OP (Fig. 2). The OP was patchy, with areas of normal alveoli around and in between the areas involved (Figs. 2 and 5). Two of the 14 cases had mild disease suggestive of OP. Two of the 14 cases had anthracotic pigment and no other condition. One of the 14 cases showed no diagnostic abnormality.

The histopathologic findings in the eight specimens of respiratory mucosa and three specimens of respiratory mucosa with alveoli showed a combination of basement membrane thickening, edema and mononuclear cell infiltration of the lamina propria, fibrosis of lamina propria, and hyperplasia of muscularis mucosa. Each of these findings was seen in mild to marked degrees in different specimens. One specimen showed marked hyperplasia of goblet cells.

**Discussion**

SM inhalation can produce a variety of lung injury patterns. The least specific of the reported findings is
pulmonary fibrosis [10]. It is not clear whether it has an interstitial pneumonitis pattern similar to other ILDs, or a somewhat less specific pattern. We found lung parenchymal injury and airway mucosal injury.

This is the first large scale report of OP in patients exposed to SM. However, the discrepancy between these recent and previous reports can be confusing. Lung fibrosis, reported by Emad and Rezaian [10], was concluded based on the HRCT findings. This supine position can lead to a shift of blood to a lower level in chest cavity. It is an error to draw conclusions from biased reports of clinicians. Also, the patients did not undergo chest HRCT in the expiratory phase of the breathing cycle, but we performed all chest HRCTs during the expiratory phase. This is necessary for the detection of bronchiolitis because material filling the bronchioles causes their obstruction, resulting in the presence of air-trapping, visible on expiratory sections [17]. Reliability can be increased by providing a clear definition for each diagnosis, and by specifying discriminating signs. Other findings of his study, based on histopathological investigations, failed to disclose the exact nature of pathology because of the UIP-like pattern and a related misdiagnosis that can mimic interstitial fibrosis [9]. These conflicts are caused by some previous disadvantages in diagnosis and diagnosis criteria, as well as by the recent histopathological classification with different features consistent with an ILD. Our understanding of the various types and patterns of diffuse lung disease that might result in fibrosis has evolved considerably over the last 50 years [6].

A pulmonary air trapping percentage of more than 25% in expiratory HRCT is highly indicative of bronchiolitis obliterans [4,12]. In bronchiolitis obliterans, the mosaic pattern of lung attenuation is caused by hypoventilation of alveoli distal to bronchiolar obstruction. This leads to secondary vasoconstriction, seen on CT scans as areas of decreased attenuation. Uninvolved segments of lung show normal or increased perfusion, with normal to increased attenuation as a result. The patients were not in a hyperventilation state, and spirometry values of all patients were mildly obstructive and restrictive or near normal. Consequently, mosaic parenchymal attenuation was present, while none of the cases was in a hyperventilation state.

Causes of OP include pulmonary infections (particularly viral and mycoplasmal), toxic gas inhalation, graft-versus-host disease following bone marrow transplantation, chronic rejection following lung or heart–lung transplantation, and connective tissue disorders such as rheumatoid arthritis and polymyositis [5,22,24]. OP could also be idiopathic, which is then referred to as cryptogenic OP (COP) or idiopathic BOOP [13].

The terms BOOP, cryptogenic COP (COP), and OP have been used in different manners. The ATS/ERS international Multidisciplinary consensus panel for the classification of idiopathic interstitial pneumonias preferred the use of COP to BOOP for idiopathic cases [2]. Although BOOP is still used for cases secondary to a well-defined clinical disease (such as infection or collagen vascular disease), the more generic term OP is gaining more acceptance. It has been used as a pattern of reaction to a variety of known or suspected causes (viral, collagen vascular disease, aspiration pneumonia, eosinophilic pneumonia, hypersensitivity pneumonia, etc) [20] in distinction to COP as an idiopathic condition. Histopathologically, OP and BOOP are identical by these standards. We used the term OP for the described pattern of reaction to inhalation of SM.

Based on the size of biopsy samples, the term OP could generally be suggested as a uniform pathology in most of our cases, but for distinguishing different types of bronchiolitis and for differentiating between proliferative obliterative bronchiolitis and constrictive obliterative bronchiolitis, it is necessary to obtain larger biopsy samples that could be achieved by video-assisted thoracoscopy or open lung biopsy. Nevertheless, bronchoscopic biopsy is an accepted technique for the diagnosis and follow-up of patients with OP, as for example in the management of chronic rejection in lung transplant patients [18].

This study also showed airway injury in tracheobronchial tree. Biopsies of bronchial mucosa showed basement membrane thickening, edema and mononuclear cell infiltration of the lamina propria, fibrosis of lamina propria, and hyperplasia of muscularis mucosa in our cases. These can represent a chronic inflammatory response to the inhaled chemical agent.

Other studies have described pulmonary fibrosis in patients exposed to SM. Our study refines the long-term histological reaction pattern in conjunction with clinical and radiological findings, which all together indicate
that most cases show persistent airway mucosal injury, and many cases show OP many years after the insult.

References


