An International collaborative pathologic study of surgical lung biopsies from mustard gas-exposed patients

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Summary

Background: Recent studies have shown strong evidence that bronchiolitis obliterans is the major long-term sequelae of exposure to sulfur mustard (SM). This study is the first to examine the histopathologic spectrum of changes in a large number of surgical lung biopsies from patients exposed to SM.

Method: Fifteen patients with chronic respiratory disease from mustard gas exposure were divided into severe (6 cases) and mild exposure (9 cases). All had surgical (open or thoracoscopic) lung biopsy, pulmonary function tests (PFTs) and chest high-resolution computed tomography scan (HRCT).

Result: The mean age of the cases was 43.8 ± 9.6 (range 33–65). All patients had dyspnea and cough as the two main complaints. Only one patient was a smoker. Thirteen patients had normal PFTs, while one had obstruction and one had mild restriction. Six (66.6%) patients in the mild exposure and 3 (50%) in the severe exposure group showed evidence of more than 25% air trapping on chest HRCT. Among the mild group, 3 had features of constrictive bronchiolitis and another had features suggestive of this (bronchiolectasis and mucus stasis). The next most common finding was a mild-to-moderate chronic cellular bronchiolitis (3 patients). Two among the 6 in the severe group showed constrictive bronchiolitis and one showed features suggestive of constrictive bronchiolitis.

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Conclusion: We conclude that about half of patients had diagnostic constrictive bronchiolitis, or bronchiolectasis and mucus stasis consistent with more proximal luminal compromise. The fact that there were no differences between the low- and high-dose groups suggests that effects of SM are not solely dependent on the severity of exposure. The results also indicate that the diagnosis of chronic lung disease due to SM may be difficult. Surgical lung biopsy may be helpful in difficult cases, as constrictive (obliterative) bronchiolitis can be present in symptomatic patients with normal PFTs and chest HRCT.

Background and aims

Sulfur mustard (SM), a potent toxic fume, was used extensively against both military and civilian populations by Iraqi forces in the 1980–1988 Iran–Iraq war.[1] Acutely it produces severe mucosal injuries of the skin, eyes and respiratory system, but there is increasing evidence that it is also responsible for the ongoing respiratory injury that may not manifest for many years. Emad and Rezaian[2] reported the late clinical findings in 197 exposed patients and found evidence of chronic bronchitis (58.8%), bronchiectasis (8.62%) and fibrosis (12.8%). Other reports, however, have identified constrictive (obliterative) bronchiolitis as the main late complication of mustard gas exposure,[3–7] but no large comprehensive pathologic study based on surgical biopsies has yet been reported.

Therefore, we undertook an international collaborative study to better define the histopathologic features of chronic respiratory disease from mustard gas exposure and correlate them with clinical presentation, computed tomography appearance and pulmonary function tests (PFTs).

Design

Data were obtained from the medical records available at a major university hospital that provides tertiary medical care and maintains a large database of patients exposed to chemical warfare agents during the Iran–Iraq war. Standardized patient interviews utilizing a previously published questionnaire[8] were also performed.

Inclusion and exclusion criteria

For inclusion in the study, patients had to have some level of exposure to SM, have had a surgical (open or thoracoscopic) lung biopsy and PFTs. Patients were excluded if they had a history of significant occupational or other environmental exposures or a connective tissue disease. Smoking history was quantified by pack-years and did not constitute a reason for exclusion. Fifteen patients who had completed the consent form have been included in the study.

Data collection

All clinical records were reviewed and the following information were abstracted: age at presentation with symptoms, gender, time from exposure to presentation, PFTs including: FEV_1, FVC and FEV_1/FVC. A diagnosis of obstructive disease was rendered if FEV_1/FVC was <0.7, and the absolute FEV_1 $<80\%$ of the predicted. Restrictive disease was diagnosed when FVC was $<80\%$ of predicted, and the ratio of FEV_1/FVC was >0.7. The ratio of FEV_1/FVC >0.7 and FVC $>80\%$ was considered as normal.[9] Data was not available on lung volumes or diffusing capacity.

Patients were divided into those with severe exposure (defined by their having a documented hospitalization (frequently with other symptoms of exposure, such as skin blistering) at the time of initial exposure) and mild exposure (defined as those who reported having been exposed, but who were not actually hospitalized and lacked a history of blistering or other acute phase manifestations).

Chest high-resolution computed tomography scan (HRCT) was performed on all patients using the following protocol: collimation: 1–1.5 mm, interval scan: 10 and 2 mm, scan time: 1–2 s, matrix: 512×512, field of view as small as possible including both lungs, window level = −600 HU (range −500 to −900 HU), with level I, 500 HU (range I, 100–2000 HU).

Surgical procedures were performed under general anesthesia with concurrent bronchoscopy. If possible, biopsies were obtained from two different lobes of the lung at the interface between apparent normal and pathologic tissue. All specimens were sent fresh to pathology, paraffin embedded and stained with hematoxylin–eosin, Masson trichrome and elastic tissue stains according to standard procedures.

Data review

All cases (see below) were initially reviewed in a blinded fashion without the knowledge of clinical features by 6 pathologists, from Italy (Department of Pathology, University of Verona), USA (Department of Laboratory Medicine and Pathology, Mayo Clinic Arizona) and Iran (Research Center of Chemical Injuries, Baqiyatallah Medical Sciences University and Shahid Beheshti University, Tehran). Standardized scoring sheets were developed with special attention to bronchiolar pathology from a previously published system utilized in the assessment of lung pathology in idiopathic pulmonary fibrosis and nylon-flock workers.[8] After scoring all features individually, each pathologist provided a preliminary diagnosis for each case. The next session of the work group was held with the attending pulmonologists, radiologists and pathologists. All cases were reviewed with full clinical history, radiologic findings, collected histopathologic scoring and checklist. A final pathologic diagnosis was then determined for each patient.

Ethical issues

The study was conducted in accordance with the principles embodied in the declaration of Helsinki, and the project design was approved by an appropriate institutional ethics committee. All patients had signed an informed consent form before participating in the study.

Results

Between February 2004 and May 2005, 15 patients met our inclusion criteria and were entered into the study. The clinical characteristics of the patients are summarized in Table 1. All patients were exposed to SM approximately 20 years previously and are still alive at the time of this report. The mean age and standard deviation (mean $\pm$ SD) of the cases was 43.8 ± 9.6 (range 33–65). All patients were suffering from chronic respiratory disease for approximately 17 years. Dyspnea and cough were the main complaints. Other common complaints were, in decreasing order of frequency: sputum production, hemoptysis and chest pain. Only one patient was a smoker. Thirteen patients had normal PFTs, while one had obstruction and one had mild restriction (Table 1).

Chest HRCT findings

Six (66.6%) patients in the mild exposure group and 3 (50%) of the severe exposure group showed evidence of air trapping involving >25% of the lung.

Histologic findings

Among the mild group all had some evidence of pathology centered on the small airways, although one current cigarette smoker had very mild changes, which could be attributable to smoking or mustard gas exposure. Interestingly, this patient also had a normal CT scan and minimal evidence of obstruction. One patient had respiratory bronchiolitis, implying that he smoked, but he denied this history, a phenomenon that has been previously reported.[10] Three had features of obliterative (constrictive) bronchilitis. All these cases had partial luminal narrowing by the presence of plaque-like increases in submucosal collagen (Figure 1). In some cases this increase in collagen was circumferential, while in others it was partial. These airway walls frequently appeared somewhat rigid and lacked the normal mucosal convolutions (Figure 2). The process was sometimes accompanied by a mild-to-moderate lymphocytic infiltrate. In some of the cases, other bronchioles in the biopsy were dilated and showed mucus stasis indicating proximal obstruction. The next most common finding was the presence of a mild-to-moderate chronic cellular bronchiolitis. One of these patients (65-year-old man) also had goblet cell metaplasia of terminal bronchioles, mild bronchiolectasis and mucus stasis, the latter suggesting that he may have had airway stenosis proximally. The biopsy from the patient with follicular bronchiolitis as the main finding also had honeycomb change, but fibroblast foci or other features to suggest a diagnosis of usual interstitial pneumonia were absent. Two among the 6 severely exposed patients showed constrictive bronchiolitis and one showed bronchiolectasis (Figure 3), again suggesting that he likely had airway stenosis not represented on the slides.

Follow-up

All patients have had slowly progressive symptoms, despite treatment with inhaled bronchodilators and corticosteroids, macrolides. In the severe group progression was initiated immediately after presentation and followed without any symptom-free period, but it was approximately after 7 years in the mild group. None, however, required lung transplantation.

Discussion

This study represents, to our knowledge, the largest series of patients with histologically documented bronchiolar disease due to SM exposure. Among our 14 patients with sufficient material for analysis, 50% had diagnostic obliterative bronchiolitis or features which strongly suggest that it was present, i.e. bronchioliectasis and mucus stasis. The next most common group had varying degrees of bronchiolar inflammation which could account for their symptoms; and interestingly 2 patients had disease likely not the result of exposure, but due to smoking and organic antigens, although the patient with histologic features of hypersensitivity pneumonitis had no obvious exposure.

This study is also the first to classify patients according to the degree of exposure. Our clinical and pathologic data show that there were no differences between the low- and high-dose groups. This suggests that effects of SM are not dependent on exposure severity.

In addition to airway pathology, one patient had honeycomb lung (7%). This figure is very close to the findings of Emad and Rezaian[2] who identified pulmonary fibrosis in 12% of patients, although their findings were not based on histologic analysis.

This study highlights several other interesting points about SM gas-exposed patients, which are relevant to the diagnosis of new patients. First, air trapping was identified in 60% of our group, similar to the reports of other authors.[4,6,7] Air trapping is recognized as the main imaging finding in patients with obliterative bronchiolitis although it is not specific and may also be seen in patients with various obstructive and airway diseases, including emphysema, asthma, Swyer–James syndrome, bronchiectasis, and cystic fibrosis. Also, 20% of our patients with air trapping seen on routine expiratory chest HRCT scans had inspiratory scans considered normal. Although a variety of diseases can show air trapping on expiratory chest HRCT scans, its presence in patients with normal findings on inspiratory scans are seen primarily in cases of bronchiolitis obliterans and asthma.[11–16] Interestingly, it was identified in 3/9 (33.3%) of our patients who did not have constrictive bronchiolitis histologically. Although air trapping does point to the presence of significant airway disease, it can be seen in healthy subjects with normal PFTs.[17–20] There was no specific pattern of chest HRCT for toxic fume-induced constrictive bronchiolitis in our series. In the post lung transplant setting, chest HRCT reportedly has 91% sensitivity

Table 1 Demographic, PFT, Chest HRCT data and pathologic diagnosis of patients.

	Low-dose exposure group							High-dose exposure group							
Age (years)	40	40	56	33	36	65	33	38	40	38	41	48	47	43	60
Sex	m	m	m	m	m	m	m	m	m	m	m	m	m	m	m
Years from exposure	17	21	18	19	23	21	20	20	18	20	20	21	20	20	21
Smoking (pack-years)	–	–	–	–	–	–	–	5	–	–	–	–	–	–	–
Signs and symptoms															
Cough	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Dyspnea	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Sputum	+	–	–	+	+	+	+	+	+	+	+	–	–	–	+
Chest pain	–	+	+	+	–	+	–	–	–	–	–	+	+	–	-
Hemoptysis	+	–	+	–	–	–	+	+	+	+	+	–	–	–	+
Spirometry															
FEV$_1$ (percent)	3.8 (95)	2.9 (81)	3.2 (90)	2.0 (52)	3.3 (86)	2.3 (85)	4.3 (88)	4.4 (104)	3.8 (92)	4.4 (99)	2.9 (76)	3.3 (96)	3.0 (83.2)	3.7 (84.3)	2.8 (82.5)
FVC (percent)	4.8 (91)	3.7 (88)	4.3 (96)	2.2 (47)	5.9 (88)	3.0 (86)	6.1 (102)	5.0 (102)	5.1 (101)	5.5 (104)	3.7 (75)	4.5 (101)	4.0 (87.3)	4.7 (90.3)	3.2 (76.6)
FEV$_1$/FVC	77	76	75	94	56	76	71	87	75	80	79	73	75	78	86
Pattern of disorder	Nl	Nl	Nl	Obstruction	Mild restriction	Nl	Nl	Nl	Nl	Nl	Nl	Nl	Nl	Nl	Nl
Chest HRCT															
Air trapping	+	+	+	+	+	+	N	N	N	+	+	N	N	N	+
Bronchiectasis	N	N	N	N	N	N	N	N	N	N	N	N	+	N	N
Bronchial wall thickening	+	N	+	+	N	N	N	N	+	N	N	+	+	N	+
Pathology diagnosis	Constrictive bronchiolitis	Constrictive bronchiolitis	Constrictive bronchiolitis	Chronic cellular bronchiolitis	Chronic cellular bronchiolitis	Chronic cellular bronchiolitis and bronchiolectasis	Respiratory bronchiolitis	Mild chronic peri-bronchiolitis	Follicular bronchiolitis	Constrictive bronchiolitis	Constrictive bronchiolitis	Chronic cellular bronchiolitis	Bronchiolectasis	Hypersensitive pneumonitis	Inadequate

Nl: normal.
N: negative.
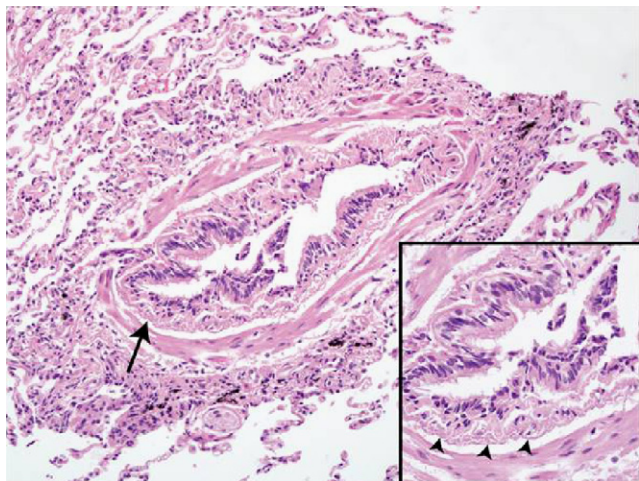+ Air trapping: air trapping involving >25% of the lung.

Figure 1 Constrictive (obliterative) bronchiolitis in a 40-year-old patient with mild exposure to SM 17 years previously with circumferential increase in collagen, highlighted by arrowheads (inset).
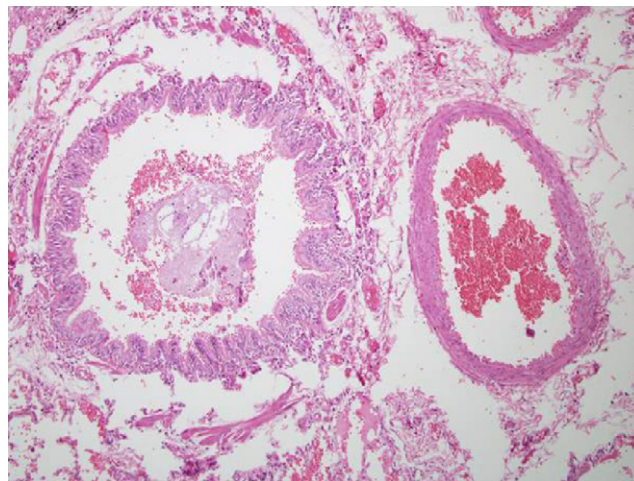

Figure 2 Constrictive (obliterative) bronchiolitis in a 56-year-old man patient with mild exposure to SM 18 years previously. A, low power, shows the increase in connective tissue bands cut longitudinally (arrows) with flattening of the overlying epithelium, while B shows connective tissue bands in cross-section (arrowheads).

Figure 3 Mild bronchiolectasis (in comparison to the adjacent artery) with mild mucus stasis in a 41-year-old man with sever exposure to SM 20 years previously. Other bronchioles showed constrictive (obliterative) bronchiolitis.
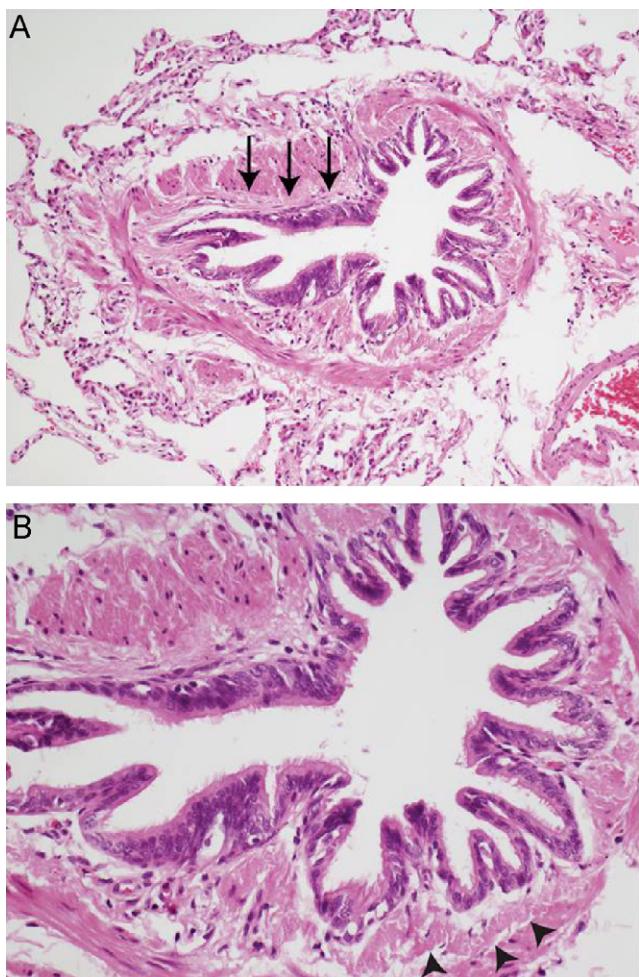
and it is a valuable tool for establishing the diagnosis of obliterative bronchiolitis.[21] But in our patients, it neither excluded nor confirmed the diagnosis of bronchiolitis. Perhaps this is because the degree of obliteration present in our patients was somewhat less than that seen in many lung transplant recipients.

Second, PFTs have been proven helpful in diagnosing idiopathic constrictive bronchiolitis and bronchiolitis obliterans syndrome (BOS) at an early stage.[22] However, all but two of our patients had normal PFTs. This is in keeping with other studies which have shown that patients with obliterative bronchiolitis may have a variety of PFTs patterns, including normal.[23] This emphasizes that the clinical work up may not always reflect the pathology as expected, and the type of PFT abnormality does not predict the mode of presentation, histology or chest HRCT results. Clinicians should have a high degree of suspicion for the diagnosis of SM-gas-induced airway disease if patients have an appropriate history of exposure and symptoms, despite atypical PFT findings.

Finally, the clinical presentation in these patients may not always suggest that airway disease is the main diagnostic problem for these patients. Although shortness of breath and chronic cough even with normal or near normal chest HRCT and PFTs would strongly suggest the presence of bronchiolitis to most clinicians, some of our patients presented with sputum production (66%) and hemoptysis (53%). The results of this study, therefore, suggest that hemoptysis alone or in combination with air trapping in a non-smoker may be more common in patients with bronchiolitis than previously appreciated and in SM-exposed individuals might be a clue to SM-induced airway disease.

In conclusion, significant airway disease including constrictive (obliterative) bronchiolitis may be present in symptomatic patients exposed to SM even when chest HRCT and PFTs are normal. Thus, open or thoracoscopic lung biopsy plays a critical role in the diagnosis of SM-exposed patients. Clinicians who encounter exposed patients with unexplained cough, progressive exertional dyspnea, and

fixed airflow limitation, with or without restriction, should consider constrictive (obliterative) bronchiolitis in the differential diagnosis.

Author contributions

Dr. Ghanei had full access to all of the data in the study, had the final responsibility for the decision to submit for publication and takes responsibility for the integrity of the data and the accuracy of the data analysis.

Study concept and design: Ghanei, Tazelaar, Chilosi.

Study supervision: Ghanei, Tazelaar, Chilpsi, Bahadori.

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Conflict of interest statement

All authors of this work disclose that they had no conflicts of interest.

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