CASE REPORT

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Distinction between mesothelioma and lung adenocarcinoma based on immunohistochemistry in a patient with asbestos bodies in bronchoalveolar fluid – case report

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Abstract

Asbestos is a mineral-mined form the rocks, consisting in amosite (brown asbestos), crocidolite (blue asbestos) and/or chrysotile (white asbestos) used in many industries. Researches about the exposure to asbestos dust and asbestosis related diseases started almost a century ago. The first case report of fatal asbestosis disease was published in 1906, in England, by Dr. Hubert Montague Murray. A decade after, asbestos "curious bodies" were firstly described in the lung tissue by Cooke (1926) and McDonald (1927). Occupational exposure to asbestos is now regulated in Romania, but past exposure is still a cause of asbestosis-related diseases (ARDs), including lung cancer. A peculiar association between a lung adenocarcinoma, a previously healed pulmonary tuberculosis (PTB) disease, is reported in a 61-year-old nonsmoker white man, a former factory worker with 29 years of occupational exposure history to cement and asbestos fibers. The positive diagnosis of asbestos exposure was facilitated by asbestos bodies determined in bronchoalveolar lavage fluid. The main purpose of this case report is to describe the development of a right pleural effusion which was not revelatory for a mesothelioma but for an adenocarcinoma of the lung. An accurate morphologic and immunohistochemistry assessment of a pleural biopsy sample excluded mesothelioma and was crucial in the positive diagnosis of adenocarcinoma. In conclusion, unilateral paraneoplastic pleural effusion in a nonsmoker male with occupational exposure to asbestosis fibers was suggestive for adenocarcinoma related asbestosis of the lung. Lung cancer and malignant pleural exudate developed after a long latency cumulative retention time of asbestos fibers.

Keywords: asbestos exposure, asbestosis bodies, bronchoalveolar lavage, lung adenocarcinoma, tuberculosis.

Introduction

In recent decades, lung cancer has become a worldwide leading cause of morbidity and mortality among neoplastic diseases, especially in males, according to *World Health Organization* (WHO) reports [1]. The exposure to carcinogens occupational factors, air pollution, the coexistence of other diseases lung, unhealthy diet, male gender and races differences, various viral infections, and not least the legacy of a genetic susceptibility are risk factors of malignant lung tumors, but the most important risk factor remains smoking [1].

In 2002, the *International Agency for Research on Cancer* (IARC) defined known and probable occupational exposure to more than 100 chemical or biological human carcinogens, including asbestos, as "any contact between the human body and a potentially harmful agent or environment in the workplace", and considered that the likelihood of lung cancer in exposed workers could be influenced by the dose and potency of the carcinogen, occupational turn-over, additive tobacco smoking exposure, and individual susceptibility [1]. The risk of developing both lung cancer and malignant mesothelioma among workers exposed both to asbestos fibers and tobacco smoke is high, and more than 100 000 deaths caused by asbestos-related diseases are WHO reported annually [2]. Pleural effusions caused by asbestos are particular forms, which may vary from an asymptomatic or mild pleurisy with completed resolution to massive and recurrent bloody pleural effusion if mesothelioma, a primary malignancy of pleura, appears. Secondary malignant pleural determination with pleural effusion is frequently diagnosed in patients with lung cancer. The interpretation of immunohistochemistry panel of antibodies may provide essential evidence for the differential diagnosis between mesothelioma and lung adenocarcinoma [3]. The newest discovered molecular targets of non-small cell lung cancer (NSCLC), as echinoderm microtubule-associated protein like protein 4 (EML4) and anaplastic lymphoma receptor tyrosine kinase (ALK) fusion oncogene, are very important in the positive diagnosis of adenocarcinoma subtype [4].

A high lung cancer risk induced by asbestos exposure independent of smoking was reported by Karjalainen *et al.*, in 1994 [5]. The study of Segarra-Obiol *et al.* in asbestos exposed workers revealed a higher incidence of pulmonary tuberculosis (PTB) in people with asbestosis *versus* those without asbestosis disease [6]. The relationship between pulmonary tuberculosis and asbestosis disease,

chronic inflammation, fibrosis and scars of the pulmonary parenchyma induced by both diseases could be the initial condition for the debut of lung adenocarcinoma. Excess risk of death of asbestos could be attributable to lung cancer among workers with a history of PTB, as Tse *et al.* reported [7].

Aim

The main purpose of this case presentation is to describe that the development of a pleural effusion in a patient with asbestos exposure is not relevant for a pleural mesothelioma. The morphopathological and immunohistochemistry evaluation of the biopsy sample is trenchant for the positive diagnosis.

Case presentation

A 61-year-old white man was admitted in November 2014, in the Clinic of Occupational Diseases, "Colentina" Hospital of Bucharest, Romania, for an accurate evaluation of a chronic lung disease complicated with a unilateral mild pleural effusion. Patient's medical history revealed, two years before, in February 2012, a previous treated right PTB complicated with a massive right pleural effusion. A pulmonary homogenous solitary nodule, measuring 3 cm, relatively well defined, was observed in the right lower lobe by a routine standard chest radiography performed 10 months later, in December 2013. The patient refused further investigations at that time but he returned in June 2014, when right pleural effusion occurred again. In August 2014, patient was investigated in "Marius Nasta" Pneumophthisiology Institute, Bucharest. Several procedures were performed, such as chest X-ray, contrast enhanced computed tomography (CT) scan (Figure 1, a–d), thoracentesis with pleural fluid analysis, pleural biopsy, complete bronchoscopic examination including bronchoalveolar lavage, cytokeratin (Cyfra) 21-1 tumor marker, carcinoembryonic antigen (CEA) assays.

Bronchoalveolar lavage (BAL) was performed by a trained chest physician through bronchofibroscopy at the level of middle lobe of the right lung. The collected 60 mL of BAL fluid (BALF) were centrifuged and obtained samples of the sediment were spread on slides, stained and analyzed. May–Grünwald–Giemsa (MGG), Ziehl–Neelsen (ZN), and Berliner Blau iron stainings with optical microscopic examination were recommended for the detection and counting of asbestos bodies (ABs) by every mL of BALF. Ziehl–Neelsen staining and cultures for *Mycobacterium tuberculosis* were performed.

Right video thoracoscopy and pleural biopsy revealed small fragments of the pleura, which were histologically and immunohistochemically (IHC) investigated.

Retention time of asbestos fibers was defined as the time period from the first year of employment (1978) until "Colentina" Hospital admission year when asbestosis was finally diagnosed (2014).

For the recurrent episodes of bloody pleural effusion, which appeared before December 2014, chemical and mechanic pleurodesis was performed.

Occupational history revealed a former unqualified worker in an asbestos-cement industry, with a 29 years of exposure to cement, crocidolite (blue asbestos) and chrysotile (white asbestos) fibers (from 1978 to 2007).

Asbestos related disease was diagnosed based on positive history of asbestos fiber, long occupational exposure with a high retention time of 36 years (from 1978 to 2014).

Medical history revealed a white retired man, non-smoker, without a positive family history of tuberculosis or cancer. Previous secondary pulmonary tuberculosis disease, diagnosed in February 2012, associated with a mild unilateral right pleural effusion, was apparently cured after six months of standard regimen with anti-tuberculosis drugs as isoniazid, rifampicin, ethambutol and pyrazinamide. Smears and cultures from sputum, bronchial aspirate and pleural fluid were all negative for *M. tuberculosis*.

From December 2013, when a 3 cm solitary lung nodule was observed in the lower right lobe, until August 2014, the patient developed chronic productive cough, progressive dyspnea, fatigability, mild weight loss and painful right chest pain but he categorically refused the biopsy of the lung nodule.

On readmission in August 2014, the patient was afebrile, with dullness in the lower middle part of the right side of the chest, without crackles at this side. Contrast enhanced computed tomography (CT) scan performed on August 21, 2014 showed a large lung mass measuring 10.2×8.4 cm localized in lower right lobe, consistent with malignancy, with mediastinal lymph node involvement, metastatic coin lesions, with diameter between 11 to 19 mm, disseminated in both lungs, right-sided pleural effusion and uniform thickening of the posterior-basal right pleura, probably of a tuberculosis origin (Figure 1, a–d).

Cyfra 21-1 tumor marker and CEA assays were in normal range of values.

Bronchoscopic examination revealed bronchial stenosis of right main bronchia, rare macrophages, eosinophils, grouped and isolated neoplastic cells in bronchial fluid, suggestive for adenocarcinoma (ADK). No culture of sputum or bronchial fluid was positive for *M. tuberculosis*.

Asbestosis bodies were identified through MGG, ZN and Berliner Blau iron stainings and counted more than 1/mL of BALF (Figure 2, a–d).

For malignant right pleural effusion (PE) initially considered mesothelioma, patient benefited of thoracentesis, which revealed hemorrhagic exudate with increased proteins (4.7 g/dL), glucose (107 mg/dL) and low adenosine deaminase (26.6 U/L). Standard PE cytology provided frequent erythrocytes and lymphocytes counts above 85% (89%), rare eosinophils (2%), neutrophils (7%) and frequent ADK cells.

After prolonged occupational exposure to asbestos fibers and asbestosis disease identified, according to the presence of asbestos bodies in the collected bronchoalveolar lavage, the patient was transferred from "Marius Nasta" Pneumophthisiology Institute, Bucharest, to the Clinic of Occupational Diseases, "Colentina" Hospital, Bucharest, with a presumptive diagnosis of mesothelioma and histological exam of pleural samples on going. After 10 days, histological exam of pleural samples collected through video-assisted thoracoscopic surgery (VATS) revealed mucinous adenocarcinoma of the lung. This type of lung cancer was histopathologically considered as the primary malignant tumor, which cause the right pleural effusion.

Additional IHC investigation of pleural samples revealed positive cytokeratin (CK) 7, CEA and thyroid transcription factor 1 (TTF1), negative CK20 and CDX and presumptive diagnosis of mesothelioma failed.

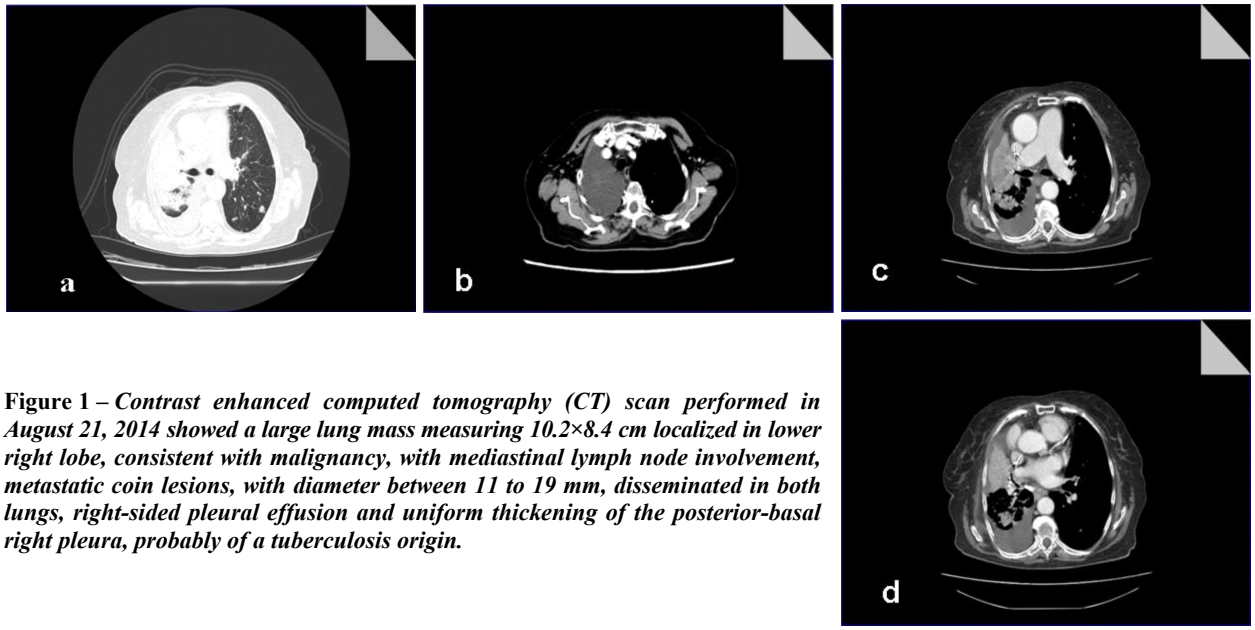

Figure 1 – ***Contrast enhanced computed tomography (CT) scan performed in August 21, 2014 showed a large lung mass measuring 10.2×8.4 cm localized in lower right lobe, consistent with malignancy, with mediastinal lymph node involvement, metastatic coin lesions, with diameter between 11 to 19 mm, disseminated in both lungs, right-sided pleural effusion and uniform thickening of the posterior-basal right pleura, probably of a tuberculosis origin.***

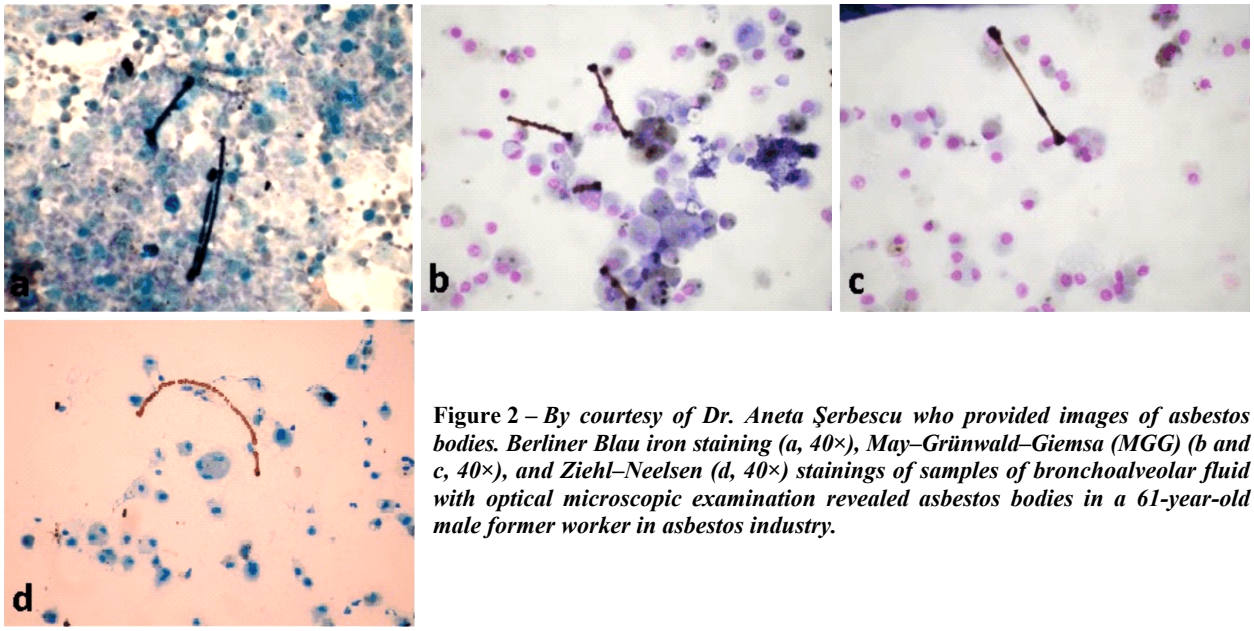

Figure 2 – ***By courtesy of Dr. Aneta Şerbescu who provided images of asbestos bodies. Berliner Blau iron staining (a, 40×), May–Grünwald–Giemsa (MGG) (b and c, 40×), and Ziehl–Neelsen (d, 40×) stainings of samples of bronchoalveolar fluid with optical microscopic examination revealed asbestos bodies in a 61-year-old male former worker in asbestos industry.***

Discussion

Lung cancer is a most commonly asbestosis related disease in smokers and nonsmokers as well. Adenocarcinoma is also considered a scar cancer and it seems to be old tuberculosis (TB) related [8]. Because the patient initially refused pleural biopsy, TB etiology remains uncertain.

The relationship between asbestos workplace exposure, concentration of asbestos bodies (ABs) and the risk of lung cancer and mesothelioma was observed and reported in Finland [9]. The intensity of asbestos exposure is related with the density of ABs in BALF. It could be compared with the level of air pollution in a small monitored work area, which can be estimated by measuring specific pollutant concentration [10]. Therefore, counting 1 AB/mL with optical microscopy is considered to be predictive for a high density of ABs per every gram of lung tissue (from 100 to 10 000) [11]. The detection of ABs in the BAL sampling sustained the asbestos exposure.

The accumulation of fluid in pleural space is confusing in patients with PTB, asbestos exposure and/or lung cancer because a pleural effusion is a common manifestation of all these diseases. Chest pain is more suggestive for malignancy in a patient with previously treated PTB. Pleural effusion requires a great variety of investigations. Tissue specimen, including pleura, are very useful for genomic and IHC analysis [3, 4]. Mucinous adenocarcinoma of the lung was histopathologically determined as the primary malignant tumor, which caused the right pleural effusion. No IHC marker reactivity suggestive for an epithelial mesothelioma (HBME1, D2-40, CK 5/6 and calretinin) was identified. There are studies considering none of the lung carcinoma could be positive for D2-40

immunostaining [3]. Thus, in the lack of D2-40 staining, the patterns of tumor-type cytology and diffuse positive TTF1 and CEA were suggestive only for a mucinous adenocarcinoma. In the absence of IHC specific markers, mesothelioma remains an inconclusive supposition.

Synchronous epithelial mesothelioma with lung adenocarcinoma in exposed asbestos workers is rarely reported and described in literature. In 1993, Cagle *et al.* reported a rare case of a simultaneous occurrence of a mesothelioma and an adenocarcinoma of the lung in a patient with asbestosis [12] and, 10 years after, Attanoos *et al.* reported nine more cases of synchronous malignant mesothelioma and lung carcinoma in patients with history of asbestos exposure [13]. Pleural samples collected in our case report revealed no ovoid mesothelial cells with clear cytoplasm arranged eccentric-looking "signet ring" but very suggestive histopathological and IHC pattern of lung adenocarcinoma. Therefore, a synchronous mesothelioma involvement of pleura in a patient with an adenocarcinoma of the lung remains no more a matter of debate.

Conclusions

The etiology of pleural tuberculosis disease is sometimes difficult to sustain in the lack of a positive bacteriologic exam for *M. tuberculosis* and/or pleural biopsy. Misconception of asbestosis occupational exposure history was related with a prolonged misunderstanding of pleural effusion etiology. Primary lung adenocarcinoma was diagnosed almost two years after healed tuberculosis disease, and seemed to be more related with the prolonged asbestosis exposure than previous pulmonary tuberculosis disease. Malignant mesothelioma development was initially suspected but immunohistochemistry examination of pleural samples established the positive diagnosis a lung adenocarcinoma with secondary malignant pleural effusion. Occupational cancer must be suspected in all cases with workplace asbestosis exposure. Screening method for detecting asbestosis bodies in BALF is very useful for positive diagnosis of asbestosis. In the same time, malignancies must be systematically investigated.

Conflict of interests

The authors declare no conflict of interests.

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