

Induced sputum and occupational diseases other than asthma

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Purpose of review

Induced sputum noninvasively provides information on cellular and soluble material from airways. It has been successfully applied for assessing airway inflammation in asthma and chronic obstructive pulmonary disease, producing reliable results comparable to biopsy and bronchoalveolar lavage. Induced sputum research in the field of occupational medicine has mainly focused upon occupational asthma, and less to other types of occupational diseases.

Recent findings

Particulate size distribution in induced sputum samples points to accumulation over time, leading to the consideration that this measurement may serve as a time-dependent marker for biological monitoring. Qualitative analysis of chemical composition of induced sputum particles is well correlated to the chemical elements spectrum in bronchoalveolar lavage lung cells and in biopsy thin sections. T cell subsets in induced sputum can be used as a marker of granulomatosis in chronic beryllium disease. Cytokines retrieved from induced sputum samples in exposed workers show a differential pattern compared to nonexposed workers.

Summary

Induced sputum is a well tolerated, noninvasive technique that is opening a new window in the field of occupational diseases of the lung and can be integrated into the well established criteria for diagnosing and monitoring these diseases, especially when invasive techniques are clinically contraindicated or impractical.

Keywords

induced sputum, monitoring, occupational exposure

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Introduction

Lung diseases caused by exposure to particulate matter and toxic chemicals in the workplace have been studied mainly by the relatively invasive fiberoptic bronchoscopy and bronchoalveolar lavage (BAL) procedures [1[•],2,3]. Noninvasive alternatives include fractional exhaled nitric oxide [4^{••}] and exhaled breath condensate collection [5[•]]. Induced sputum is the only other noninvasive method for direct study of soluble mediators and cellular components of the lung [6^{••},7^{••}]. Induced sputum is mostly used for diagnosing and managing asthma, chronic obstructive pulmonary disease (COPD) and chronic bronchitis. This review presents the published results of sputum analysis in diagnosing, assessing and monitoring other occupation-associated pulmonary diseases.

Induced sputum in workers exposed to silica and metal particles

The results of an evaluation of silica-exposed and hard metal-exposed workers [8] revealed that BAL and induced sputum yielded similar quantitative and qual-

itative results in terms of the number of particles that were present in the samples and the chemical analysis of the particles. The qualitative analysis of chemical particles was performed by scanning electron microscope (SEM), whereas the quantitative analysis was done by the Cis-100 analyzer (Ankersmid, Yokneam, Israel) which allows a rapid analysis using minute quantities of biological material. Around 70% of particles internalized by macrophages or bronchial epithelial cells present in induced sputum as well as in BAL samples were smaller than 2.5 μ (<PM 2.5). These particles represent the largest fraction of the weight of the dust that enters the lower lung fields [8]. It should be noted that these tests were performed in optimal clinical laboratory conditions. The use of induced sputum for large-scale screening of workers would necessitate carrying out the sputum induction at the workplace in order not to cause unacceptable inconvenience in the work schedule. Our group therefore attempted to induce sputum in the foundry workers on-site during their routine periodic health examinations. We sought to determine whether the distribution of particle size in induced sputum specimens was different between exposed and nonexposed

workers, whether smoking was a confounder of the association, and whether there was a correlation between the distribution of particle size and the results of these workers' pulmonary function tests. We demonstrated significant differences in the proportion of particles with diameters greater than 2 and 5 μ between the exposed and nonexposed groups. There was a positive correlation between the proportion of particles greater than 5 μ and flow expiratory volume in one second (FEV1)/forced vital capacity (FVC) ratio for all workers ($n=54$, $r=0.32$, $P=0.02$), being stronger for nonsmokers ($r=0.47$, $P=0.02$) [9]. The high FEV1/FVC ratio would be consistent with restrictive disease. A semi-automated sputum cytometry method to detect atypical nuclei was found to have a sensitivity of 75% and specificity of 89.3%, compared to the sensitivity of 83% and specificity of 97% of conventional cytology for establishing the final diagnosis of lung cancer in slides of sputum samples in former uranium miners with silicosis and patients with asbestosis [10].

In a recent work, our group recruited asymptomatic welders with either short-term exposure to aluminum-iron or long-term exposure to cadmium chromium-iron-lead-nickel and compared them to a control group of nonexposed workers in order to identify the effect of inhaled particulate matter inhaled during welding on subclinical inflammatory status. This was the first demonstration of the crucial factor of the duration of exposure over time: there was a higher percentage of particles greater than 2 μ in the airways of welders in correlation to the years of exposure to the noxious air-borne substances [11^{••}]. We then hypothesized that the quantitative analysis of particles recovered by induced sputum, as was shown in those studies, can be added to the periodic health examinations of healthy workers exposed to hazardous dusts and serve as a tool for biological monitoring of their pulmonary status together with the traditional occupational parameters of history and environmental measurements. This screening would correspond to the currently employed biological monitoring of workers exposed to toxic agents, such as some metals (e.g. lead, cadmium) and solvents (e.g. toluene, trichloromethane) that can be estimated by measuring them or their metabolites in blood, urine or exhaled air [12].

Mineral analysis of tissue specimens has provided very useful information in pathological conditions associated with occupational exposures to mineral particles, particularly when combined with epidemiological information. In this context, our group compared the first qualitative SEM analysis of particles in induced sputum vs. BAL and found a very similar spectrum of mineral particles in both samples [8]. We also conducted a comparative SEM analysis of particles in induced sputum vs. lung biopsy samples in silica and hard metal-exposed workers and were able to demonstrate that SEM analysis of induced

sputum samples was as efficient for detecting hazardous particles as was SEM analysis of lung tissue samples. Silica was detected even better in sputum, whereas exposure to silica was the main chemical element associated with a high likelihood to show 'pathological' findings in induced sputum [odds ratio (OR) 19.41, 95% confidence interval (CI) 0.270–1398.33] [13^{••}]. The sample was considered 'pathological' if SEM analysis identified silica or hard metals compatible with the exposure background as self-reported in the questionnaire.

Induced sputum in workers exposed to asbestos and glass wool fibers

Several investigators have shown the relevance of asbestos bodies in spontaneous sputum compared to BAL. Sputum analysis for asbestos bodies was, however, much less sensitive than BAL fluid analysis and useful only in workers with high levels of exposure [14]. Although another group of investigators found agreement of asbestos body detection in only 56.8% of their cases, they concluded that the two methods (BAL and induced sputum) were complementary since both are useful in increasing the probability of detection [15]. In another study, the induced sputum samples of ex-workers with asbestosis on high-resolution computerized tomography (HRCT) showed reduced sputum cellularity but higher macrophage/neutrophil ratio and percentage macrophage than the induced sputum samples of nonexposed controls [16^{••}]. Sputum cytology was related to lung function impairment [16^{••}].

There are two reported cases in which fibers could be visualized in induced sputum and in which induced sputum assisted in evaluating and diagnosing suspected occupational lung diseases. The first was a case of a miner who worked under Nazi captivity in Silesia. He was referred for induced sputum analysis due to progressive shortness of breath. His chest computed tomography showed diffuse interstitial fibrosis, and pulmonary function testing revealed a restrictive pattern with decrease in diffusion capacity. Analysis of his induced sputum sample demonstrated clay minerals, quartz, aluminum, and asbestos fibers [17]. The second case was a bus driver who had used the same vehicle for 11 years. He had undergone bronchoscopy 4 years earlier and the results showed thickening of interstitium, bronchial wall with epithelial shedding, and thickening of the basal membrane, bronchial wall inflammatory infiltration. Additionally, some multinuclear giant cells, resembling those of foreign body cells, were found. He was referred for a more thorough occupational exposure evaluation because of worsening of clinical parameters. The SEM analysis of his induced sputum sample revealed the presence of fibers with a chemical composition compatible with that of glass wool. The results were confirmed by an SEM analysis of a lung tissue biopsy. Investigation of the

working environment of this patient revealed that the roof of his bus was insulated with glass wool and that the covering over the driver's seat was full of holes [18].

Induced sputum in the assessment of acute exposures

After the disaster at the World Trade Center (WTC) in New York, our group performed a study to determine whether induced sputum from highly exposed firefighters who had been caught in the dust cloud demonstrated a unique pattern of inflammation and particulate matter deposition compatible with WTC dust, 10 months post-collapse. Their induced sputum differential cell counts differed from those of healthcare worker controls, but not from those of firefighters in Tel Aviv [19]. Neutrophil and eosinophil levels increased with greater intensity of WTC exposure, and metalloproteinase-9 (MMP-9) levels positively correlated with neutrophil counts. The particles were larger and more irregularly shaped in the exposed New York group (1–50 μ ; Zn, Hg, Au, Sn, Ag) than in the Tel Aviv group (1–10 μ ; silica, clays).

The analyses of WTC dust and particle size demonstrated that a portion of the dust was composed of particles small enough to penetrate deep into the lungs, reaching distal airways, and alveoli. These particles could well have been the cause of the diffuse parenchymal lung disease that was diagnosed after exposure to the dust at WTC disaster site [20*].

Induced sputum in granulomatous lung diseases

Our group and others [21,22] demonstrated that the results of induced sputum and BAL are comparable with regard to high percentages of lymphocytes and predominance of CD4 in sarcoidosis.

Beryllium is a classic example of a metal-mimicking sarcoidosis, and chronic beryllium disease (CBD) has been and continues to be misdiagnosed as sarcoidosis [23,24]. A study on a cohort of dental technicians involved in casting, sprue cutting, grinding, polishing, and finishing of dental alloys containing beryllium discovered that half of them were suffering from CBD [23]. Induced sputum was performed also in the 12 patients who had CBD and tissue granulomatosis was correlated with high ratio CD4/CD8 (a high ratio of CD4/CD8 T cell subsets, range 2.9–12.8).

Soluble factors retrieved by induced sputum in occupational diseases

Metalloproteinase-9 was measured in the above-cited study on the firefighters exposed during WTC collapse

[19] because it plays an important role in remodeling and the inflammatory process in the lung [25] and also because it is detectable in induced sputum with reliable and reproducible results [26]. There was a trend for higher levels of MMP-9 in the induced sputum samples from the New York firefighters than in those from the Tel Aviv firefighters, and the levels of both groups were significantly higher than nonfirefighter controls. There was also a positive correlation between the accumulation of neutrophils and MMP-9 levels. In these nonsmoking firefighters, the increase in MMP-9 levels provided biochemical evidence for exposure-related immune activation in the lung, complementing the evidence from induced sputum differential cell counts. In a later study, our group demonstrated that the combination of MMP-9 values in induced sputum and the results of pulmonary function tests can serve as markers of damage and intensity of exposure to hazardous dust in a cohort of workers exposed to metal particles and supposedly nonexposed office workers. A linear regression model for MMP-9 based on exposure, smoking habits, and proportion of particles less than 5 μ revealed a positive correlation between each of the explanatory variables and MMP-9 values [27].

A correlation of proinflammatory cytokines and exposure to silica and asbestos was reported in two recent publications [16••,28]. In the first study, occupationally exposed patients with asbestosis on HRCT had cytologic abnormalities in their induced sputum samples and increased local and systemic proinflammatory status as expressed in high levels of interleukin-8 (IL-8) which were correlated to functional impairment [27]. In the second (pilot) study that evaluated a group of workers with asbestosis and another group of workers with silicosis, the levels of interleukin-1 β were higher in the sputum of both groups than in unexposed controls. Importantly, eosinophil, neutrophil, and IL-1 β levels were significantly correlated with the rate of decline in pulmonary function [28].

Conclusion

The convincing evidence that has accumulated over the past decade has validated the consideration of the sputum induction technique as a reliable tool in the diagnostic work-up of individuals whose workplace is known or suspected as being hazardous to pulmonary function. Unlike traditional and more invasive procedures, induced sputum can be performed on-site and as often as necessary to detect exposures and to assist in monitoring disease.

References and recommended reading

Papers of particular interest, published within the annual period of review, have been highlighted as:

- of special interest
- of outstanding interest

Additional references related to this topic can also be found in the Current World Literature section in this issue (pp. 178–179).

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