

Investigative use of bronchoscopy, lavage and bronchial biopsies in asthma and other airways diseases

Workshop held in Columbia, Maryland on November 19-20, 1990

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While bronchoscopy is performed routinely in the clinical practice of pulmonary medicine, additional research applications have been developed using this procedure to obtain cellular and other components of the airway alveolar lining fluid from patients with a variety of lung diseases [1]. The results of these studies have enhanced our understanding of the pathogenesis and management of pulmonary disease.

Recently, extensive scientific data (over 200 published reports during the last five years) have accumulated, which document that investigative bronchoscopy can be safely performed to study the tracheobronchial tree of individuals with asthma and individuals with other obstructive pulmonary diseases. In subjects with asthma endoscopic procedures that include bronchoalveolar lavage [2, 3, 4], segmental airway lavage [5], mucosal biopsies, endobronchial brushing, and biopsies [6, 7, 8], placement of measurement devices [9] and direct assessment of airway physiology, can be performed when appropriate precautions are observed [10]. The research potential for investigative bronchoscopy and instrumentation of the airways is considerable and it is expected that these approaches will continue to provide information about the pathogenesis of airway diseases.

An earlier workshop, convened by the National Institutes of Health in 1984, developed guidelines [11] for ensuring the safety of subjects undergoing bronchoscopy and bronchoalveolar lavage. The purpose of the present workshop was to review the vast experience gained since then from the world-wide practice

of investigative bronchoscopy and, more importantly, to examine the possible need for developing revised guidelines and recommendations for its use.

While there is a more limited experience using investigative bronchoscopic techniques to study the pathophysiology of other forms of obstructive lung disease, the scientific indications and safety considerations should not differ from those for asthma. The present document, a summary of a workshop held on November 19-20, 1990, at Columbia, MD, USA, reviews the experiences with bronchoscopy, lavage, mucosal biopsy and airway instrumentation in asthma and the other obstructive pulmonary diseases and provides guidelines to ensure the safety of volunteer subjects participating in such procedures. These guidelines, for the most part, reflect the experiences gained using fiberoptic bronchoscopic techniques. However, studies have also been conducted in subjects with asthma using rigid tube bronchoscopy. Thus, the rigid tube techniques also appear safe when used by skilled investigators. Hopefully, these guidelines will aid in generating valid research data in an optimal fashion. These recommendations are limited to the research application of bronchoscopic procedures and will not address clinical indications which have been reviewed recently [12].

Past experience with airway lavage, bronchoalveolar lavage, mucosal biopsy and placement of measurement devices

The feasibility and utility of investigative bronchoscopy and airway instrumentation in asthma has been well documented. These techniques have been used to measure physiological function of the airways [10], evaluate inflammatory mediators and cells in lavage fluid [2-5], assess the effects of bronchial challenge, and study the morphology of accessible intrapulmonary airways [13, 14]. Recent studies have reported elevations of constrictor substances that include, but are not limited to, histamine, prostaglandins, leukotrienes and kinins in lavage fluid obtained from subjects with asthma [2-5, 13, 14]. Other investigators have reported

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elevated numbers of eosinophils, lymphocytes and mast cells in lavage fluid obtained from patients with asthma [15-19]. *In vivo* challenge with antigen and other provocative agents has been associated with release of inflammatory mediators into respiratory fluids and additional cellular events that include evidence of cell migration into the airway with increased numbers of eosinophils, as well as the accumulation of other inflammatory cellular elements [5, 13, 17, 20-26]. Mast cells and alveolar macrophages recovered from the airways of patients with asthma have been studied *in vitro* to evaluate cell function and immunopharmacological responsiveness [16, 27, 28]. These studies have demonstrated the potential interrelationships between different biochemical and cellular changes in asthma [28, 30]. Since most of these approaches have evaluated events on the airway surface, the need to understand morphological responses in the bronchial mucosa has led to the application of bronchial biopsy and brushing techniques in the study of airway disease. These studies have produced evidence of epithelial damage, eosinophilic infiltration and abnormal collagen deposition in the airways [6, 7, 30-32]. In addition, the effects of immunopharmacological interventions are being studied in lavage fluid and in biopsies from subjects with asthma [17, 33, 34]. The concomitant use of bronchial biopsy and lavage techniques will permit the investigation of the interrelationships between cellular and biochemical events both on the airway surface and in bronchial tissue.

Techniques have been developed to assess physiological function in the airways of subjects with asthma. These studies have employed fiberoptic bronchoscopic techniques to directly measure airway mechanics, and have shown that mild asthmatics have elevations in peripheral airway resistance [10]. Instrumentation to assess intra-airway thermal events and bronchial surface fluid balance has been developed. These studies have reported a relationship between thermal gradients and alterations in pulmonary mechanics and bronchial blood flow in exercise induced asthma [9, 35]. Other investigators have reported evidence of altered permeability and other inflammatory abnormalities in the airways of patients with asthma [29, 36, 37]. These multifaceted approaches will enable the investigation of the relationship between physiological function and inflammatory events in airways of subjects with asthma and other obstructive airways diseases.

Indications for investigative bronchoscopic procedures in asthma and other obstructive lung diseases

There are multiple scientific indications for investigative bronchoscopy and instrumentation of the tracheobronchial tree of subjects with airway diseases. The major areas of study are the following:

1. Fluids, cells and tissue samples in order to investigate the morphological, pathological, molecular

biological, immunological, biochemical, pharmacological, and neurobiological factors that are important in understanding the pathogenesis, natural history, pathophysiology, and clinical features of asthma, and other forms of obstructive and inflammatory airway disease.

2. Regional airway physiology and the factors that influence it in normal and diseased states.

3. Bronchial blood flow in normals and in subjects with airways diseases.

4. The effects of acute and chronic therapeutic and pathological interventions on physiological function, morphology, cellular interactions, and biochemical events in the airways.

5. Airway epithelial function *in situ*.

6. The pathogenesis of microbial and viral infections in airway disease.

This list is not inclusive, and other indications may exist for investigative bronchoscopy in asthma and obstructive airway disease. Additional indications may develop in the future that are based on the results of current research on the pathophysiology of these disorders.

Contraindications to investigative bronchoscopy

There are a few absolute contraindications to investigative bronchoscopy in asthma and other airway diseases. These include sensitivity to the local anaesthetic or other medication used during the procedure and an uncorrected bleeding diathesis, particularly if bronchial biopsy is planned. The major relative contraindications involve the activity of the subject's airway disease, level of bronchial responsiveness, the degree of airflow obstruction present at the time of study, and the presence of coexisting diseases particularly those involving the cardiovascular system. These major relative contraindications may affect the safety of the subject with airways disease during investigative bronchoscopy. Combinations of these factors may substantially increase the risk of untoward reactions during these procedures.

The extent to which the degree of airflow obstruction and bronchial hyperresponsiveness that exists prior to instrumentation, adversely influences the outcome is unknown. Bronchoscopy with lavage and/or biopsies has been successfully performed in subjects with asthma, chronic obstructive lung disease and cystic fibrosis, whose values for (FEV₁) are <60% of predicted [6, 8, 16, 26, 36, 37]. The general experience in asthma, however, indicates that the greater the degree of bronchial responsiveness and/or the more severe the airflow obstruction at the time of bronchoscopy, the greater the potential risk of precipitating acute severe airway obstruction. In a few subjects with asthma who have high levels of bronchial responsiveness and normal or near normal pulmonary function, acute bronchial narrowing may develop during the early phases of the bronchoscopic procedure, even if bronchodilator premedication has been administered. This acute airflow obstruction is rare and it improves after

bronchodilator therapy. Obviously, in individuals with significant preexisting bronchial obstruction such an adverse effect potentially could be dangerous. Thus, if bronchoscopy is to be performed in subjects with high levels of bronchial hyperresponsiveness, the bronchoscopist must be prepared to terminate the procedure and to treat the subject with bronchodilators, if bronchospasm occurs. In our current state of knowledge, the committee felt that fiberoptic bronchoscopy should not be performed in subjects with extreme bronchial hyperresponsiveness and severe airflow obstruction.

Age alone should not be a factor for excluding any subject from investigative bronchoscopic procedures. Bronchoscopy with various kinds of instrumentation has been safely performed in infants and older adults without adverse sequelae. There are few data in subjects over the sixth decade and with increasing age there is an increased risk of concomitant disease. When bronchoscopy is considered in subjects over 60 yrs of age, additional precautions are indicated. Older individuals should be carefully evaluated for the presence and severity of co-existing diseases, especially those involving the cardiopulmonary system. However, it should be noted that many clinical bronchoscopies are performed in older individuals with significant obstructive lung disease and coexisting cardiovascular disease [1, 12]. With respect to the paediatric age group, while there is now a small, but significant, experience with investigative bronchoscopy in infants and children [38], specific recommendations cannot be made at this time. There are substantial issues related to ethics, informed consent, instrumentation and technique that make bronchoscopy in paediatric age groups more complex than in adults.

Safety and potential hazards

The safety of the subject is of paramount importance and cannot be jeopardized for the sake of completing a research protocol or obtaining scientific data. The available experience indicates that the risks of bronchoalveolar lavage and bronchial brushing or biopsies are similar and increase those of bronchoscopy alone. The potential hazards include, but are not limited to, acute airway obstruction, laryngospasm, hypoxaemia, and adverse reactions to premedications. Bleeding is an unusual complication that has been associated with bronchial biopsies or brushings. When it occurs, blood loss is insignificant. Airway perforation is a theoretical, but as yet unreported, possibility with bronchial biopsies or brushings. Fever and transient asymptomatic pulmonary infiltrates occasionally occur, especially following bronchoalveolar lavage [1]. Infection is a potential but rare complication after bronchoscopic procedures. It is important to recognize that the combination of two procedures such as biopsy and bronchoalveolar lavage tends to increase the duration of the procedure and the risk of hypoxaemia.

Subject evaluation

Because of the possibility of precipitating acute severe bronchial obstruction, the following evaluation is recommended prior to initiating bronchoalveolar lavage, bronchial brushings or biopsy, local airway challenge or other types of airway instrumentation. The investigator must obtain a complete medical history and physical examination. The history should include information about the severity and frequency of asthma attacks, current medications and specific allergies. The severity and activity of the subject's illness should be ascertained by some assessment of the duration and frequency of acute episodes of asthma, number and duration of hospitalizations, frequency of nocturnal awakenings, circadian variations in lung function, medication requirements, or measures of the level of responsiveness to standard bronchoprovocation with methacholine, histamine, exercise, or isocapnic hyperventilation. In addition, the history of other co-existing medical conditions should be recorded with specific emphasis on the cardiovascular system. Laboratory data, consisting of baseline pulmonary function studies that objectively measure the level of airflow obstruction and arterial oxygen saturation by oximetry, should be obtained prior to initiating the bronchoscopic procedures. Arterial oxygen saturation, cardiac rhythm, and blood pressure should always be monitored during these procedures. Prior to the study of patients with severe airflow obstruction, measurement of arterial blood gases may be indicated in addition to oximetry to exclude the presence of alveolar hypoventilation. In general, subjects with asthma who have a history of acute severe respiratory failure with severe episodes of airflow obstruction should not be selected for these studies.

Experience has demonstrated that endoscopy with biopsy, lavage, segmental airway challenge, and placement of measurement devices can be safely carried out in subjects with mild to moderate asthma who are asymptomatic at the time of study, but have reduced pulmonary function. The feeling was unanimous that in individuals with an $FEV_1 > 60\%$ of predicted, the endoscopy procedures could be safely performed. In subjects with chronic airflow obstruction and FEV_1 values $< 60\%$ predicted, investigative bronchoscopy may be safer than in subjects with asthma, because these subjects usually have lower levels of bronchial hyperresponsiveness. There are also reports that investigative bronchoscopic procedures have been safely carried out in subjects with asthma with FEV_1 values $< 60\%$ predicted [6, 8, 16, 36, 38]. If investigative fiberoptic bronchoscopy is to be performed in subjects with asthma with FEV_1 values $< 60\%$ of predicted, there should be valid scientific indications to study these subjects with more severe airflow obstruction, and additional care must be taken in their evaluation, monitoring and medical follow-up.

Preparation

The procedure should be performed by an experienced bronchoscopist with facilities available for the management of medical emergencies and cardiopulmonary resuscitation. A route for injection of intravenous medication must be secured. Arterial oxygen saturation should be monitored continuously, and oxygen administered in sufficient quantities to maintain oxygen saturation within normal limits. Premedication with atropine, bronchodilators and sedatives can be either administered or omitted, as required by the specific research protocol and the comfort and safety of the subject. A history of bleeding disorders and the use of medications that can affect clotting should be sought and recorded. Prior to bronchial biopsy or brushings, coagulation studies should be obtained to exclude abnormal bleeding states. Topical anaesthesia of the upper and lower airways should be achieved using the lowest doses of topical anaesthetic possible. Additional mild sedation may help to make the procedure more comfortable for the research subject.

Procedures

Bronchoscopy with instrumentation, airway lavage, bronchoalveolar lavage, biopsy and brushing have been performed before and after a number of acute bronchoprovocations including local instillation of antigen, nonisotonic solutions, air pollutants, and thermal stimuli such as exercise and hyperventilation [5, 17, 20, 21, 24, 25, 26, 39, 40]. They have also been undertaken to evaluate the mechanisms by which therapeutic interventions affect physiological, biochemical and cellular function in the lung [10, 33, 34, 40]. The techniques used to obtain fluid and tissue have varied widely, so that no detailed specific methodological recommendation can be made. This experience, although establishing the feasibility and safety of these approaches to provide meaningful scientific data, has also demonstrated that extreme care needs to be exercised during any challenge procedure. When a provocation is to be performed in a subject in association with airway instrumentation, the magnitude of the provocative stimuli should be adjusted to produce the minimum functional decrements commensurate with obtaining the desired experimental results. Administration of allergen directly to airway segments carries the additional potential risk of producing systemic allergic reactions and late phase pulmonary responses [21, 25].

The quantity of fluid used in lavages has varied with the needs of the protocol. Lavages have been performed with as little as 5 ml of fluid, and with as much as 400 ml. Data on the use of larger amounts of lavage fluid than this amount is not available and is not recommended. While lavage has been performed in all lung segments, the return of fluid from the right middle lobe, lingula, or lower lobes is higher than from upper lobes and these areas are the usual regions for

lung washings. Lavage fluid should be aspirated with gentle suction. Periodic deep breathing by the subject may aid in recovery of the effluent, although with asthma and other obstructive airways diseases, fluid recovery is lower than in normal volunteers.

With respect to mucosal biopsies, three to six 2 mm tissue samples from a combination of the main carinae, and one or more of the segmental or subsegmental carinae have been obtained during a single procedure without sequelae. Acquisition of more biopsies than this during a single bronchoscopy will increase the duration of the procedure, and information on the safety of additional biopsies is not available. Bronchial biopsies should always be performed under direct vision. Brushings of two to four areas have been performed without difficulty in subjects with asthma. There are no data on either the indications or safety of transbronchial lung biopsies in the investigation of obstructive airway disease but this procedure is likely to be associated with a higher level of morbidity.

The safety of repeat lavages and airway biopsies from different segments of the lung has been established. Two such procedures in a 24 h period have been performed and appear to be well-tolerated. More than this number has not been attempted and with the present state of knowledge is not recommended. Serial biopsies with lavages have also been performed in the same subject over longer periods of time, the minimum reported interval between procedures has been six weeks. The shortest interval over which serial bronchial biopsies can be safely performed has not yet been determined.

It should be realized that obtaining valid scientific data from these bronchoscopic procedures is difficult and complicated. They should be undertaken only after the investigator has developed the skills and resources to make appropriate measurements and evaluate the specimens accurately. The assessment of biochemical mediators in biological fluids requires careful measurement and standardization. Technical skill and laboratory resources are especially important in processing the small biopsy specimens obtained during fibreoptic bronchoscopy. They require careful histological preparation and processing. The biochemical and morphological analysis of tissues and fluids obtained during investigative bronchoscopy are not routine clinical procedures. Careful review of the techniques employed and of findings from previous studies are important in planning future research endeavours.

Post procedure observation

The subjects should be carefully monitored at regular intervals until their condition is stable. The time required to achieve this will vary from subject to subject and with the type of procedures performed. Suggested end-points include, but are not limited to, return of the gag reflex and the absence of any clinical

evidence of symptomatic airflow obstruction at the end of the protocol. This should be documented prior to discharge of the subject by objective measurement of forced expiration with spirometry.

The subjects should be given the name and telephone number of a physician to contact, should untoward symptoms such as fever, dyspnoea, chest pain, or airway obstruction develop. Many investigators feel it is prudent to contact their subjects the day following the procedure to document their state of health.

Other potential uses

Bronchoscopy provides a great potential for the *in situ* investigation of airway physiology, biochemistry and immunology in diseases such as asthma, cystic fibrosis, chronic bronchitis, bronchiectasis, and other chronic obstructive lung diseases. For example, this procedure permits the insertion or implantation of surface sensors for the study of epithelial biochemistry, thermodynamics and biophysics; studies of microbial adhesion to the epithelium; and the definition and monitoring of biochemical and/or physiological markers of disease. These approaches will further our understanding of complex pathophysiological processes and interrelationships that differentiate asthma and other forms of airway disease.

Limitations

The major limitation of investigative bronchoscopy is that this procedure is invasive. Endoscopy allows one to study the disease process only at one point in its natural history. Therefore, repeated studies are required to ascertain how events change over time. Further, it is difficult to examine changes in airway function or morphometry in acutely ill patients. The findings from bronchoalveolar or airway lavages reflect surface airway or alveolar events that may not accurately reflect the condition in the airway mucosa *per se*. Also, although the findings from studies of the lavage fluid may reflect the events within the airway wall, this relationship needs to be determined experimentally. Bronchial biopsies are limited to larger airways and may not reflect the findings in smaller peripheral airways. Finally, there is no standard way of normalizing information obtained from biopsies and lavages.

Future needs

The airway disease of each subject should be carefully characterized prior to performing endoscopic procedures. Improved techniques are needed for the preparation and standardization of samples and for morphological analyses. All specimens and biopsies should be processed for maximum utilization. Data are required to establish whether biopsies from larger

airways reflect the events throughout the tracheobronchial tree. In addition, longitudinal data are needed to ascertain how changes in disease status are reflected in tissue and cellular events. Finally, better techniques and instruments that permit the study of airway events *in situ* need to be developed.

The ability to use bronchoscopic techniques while adhering to carefully designed guidelines (table 1), to sample lower respiratory tract secretions and cells, measure physiological function, and to correlate these findings with bronchial morphology in carefully characterized subjects, provides a unique opportunity to investigate the pathophysiology of airway diseases in man. The mechanisms of action of different therapeutic interventions can be studied by combining clinical and physiological assessments with cellular and biochemical measurements. The multifaceted approach should lead to a better understanding of the pathogenesis of these diseases and should result in optimal regimens for their prevention and treatment.

Table 1. — Summary of recommendations and guidelines for investigative bronchoscopy in subjects with asthma and other obstructive airway diseases

Bronchoalveolar lavage, bronchial brushing and biopsy, and airway instrumentation can be safely performed in subjects with mild to moderate asthma.	
Subjects considered unsuitable	Those sensitive to local anaesthetics/other medications; individuals exhibiting extreme bronchial hyperresponsiveness and severe airflow obstruction; and those with uncorrected bleeding diathesis (biopsy).
Subjects at high risk from the procedures	Those with FEV ₁ <60% predicted and/or coexisting cardiopulmonary diseases
Procedural limitations	Instillation of no more than 400 ml fluid; Three to six 2 mm biopsies from a combination of the main carina and one or more segmental or sub-segmental carinae in a single procedure; and brushing limited to two to four areas
Potential hazards	Acute airflow obstruction; laryngospasm; hypoxaemia; apnoea; bleeding; drug reactions; airway perforation (biopsy); fever; pulmonary infiltrates; infections
Pre-procedure evaluation	Complete medical history; physical examination; electrocardiogram; blood pressure; oximetry
Post-procedure monitoring	Gag reflex; pulmonary function; stable clinical status; post-discharge follow-up

FEV₁: forced expiratory volume in one second.

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