AN INTERNATIONAL ISHLT/ATS/ERS CLINICAL PRACTICE GUIDELINE: DIAGNOSIS AND MANAGEMENT OF BRONCHIOLITIS OBLITERANS SYNDROME

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ABSTRACT

Background: Bronchiolitis obliterans syndrome (BOS) is a major complication of lung transplantation that is associated with poor survival.

Purpose: The American Thoracic Society (ATS), International Society for Heart and Lung Transplantation (ISHLT), and European Respiratory Society (ERS) convened a committee of international experts to describe and/or provide recommendations for (1) the definition of BOS, (2) the risk factors for developing BOS, (3) the diagnosis of BOS, and (4) the management and prevention of BOS.

Methods: A pragmatic evidence synthesis was performed to identify all unique citations related to BOS published from 1980 through March, 2013. The expert committee discussed the available research evidence upon which the updated definition of BOS, identified risk factors and recommendations are based. The committee followed the GRADE approach to develop specific clinical recommendations.

Results: The term BOS should be used to describe a delayed allograft dysfunction with persistent decline in FEV1 that is not caused by other known and potentially reversible causes of post-transplant loss of lung function. The committee formulated specific recommendations about the use of systemic corticosteroids, cyclosporine, tacrolimus, azithromycin and about re-transplantation in patients with suspected and confirmed BOS.

Conclusions: The diagnosis of BOS requires the careful exclusion of other post-transplant complications that can cause delayed lung allograft dysfunction, and several risk factors have been identified that have a significant association with the onset of BOS. Currently available therapies have not been proven to result in significant benefit in the prevention or treatment of BOS. Adequately designed and executed randomized controlled trials that properly measure and report all patient-important outcomes are needed to identify optimal therapies for established BOS and effective strategies for its prevention.

TABLE OF CONTENTS

EXECUTIVE SUMMARY

Many lung transplant recipients develop delayed allograft dysfunction that has been traditionally referred to as bronchiolitis obliterans syndrome (BOS), which is thought to be caused by inflammation, destruction, and fibrosis of small airways in the lung allograft that leads to obliterative bronchiolitis (OB). Because a definitive diagnosis of OB is difficult to make without a surgical lung biopsy, a decrease in the FEV1 has been used as a surrogate marker to identify patients who develop a syndrome of significant and persistent loss of lung allograft function with onset three or more months following transplantation. However, it is now recognized that numerous factors apart from OB can lead to a delayed onset, significant decline in lung function, and these causes of delayed onset graft dysfunction must be carefully excluded when a diagnosis of BOS is made. In general, BOS responds poorly to therapeutic interventions but may stabilize, and some patients may have a significant improvement in FEV1 with certain therapies.

A comprehensive review of the literature on lung transplantation and bronchiolitis obliterans syndrome allowed the committee to reach a number of conclusions, which are given in Table 1. Additionally, committee members used a systematic approach to formulate a number of specific evidence-based recommendations for the prevention and management of BOS, which are given in Table 2. Evidence tables are provided in the online supplement. It is our hope that this guideline will promote an understanding of the current approach to the evaluation and management of lung transplant recipients who develop delayed allograft dysfunction, as well as stimulate additional research that will provide higher quality evidence upon which future guidelines may be based.

SCOPE AND PURPOSE

The purpose of this document is to revise the definition of BOS, discuss the risk factors for the development of BOS, and provide guidance about the management of patients with suspected or confirmed BOS. The target audience of these guidelines is specialists in respiratory medicine managing adults and children who have received lung transplants. Other specialists in respiratory medicine may also benefit from these guidelines.

INTRODUCTION

Obliterative bronchiolitis, first described in recipients of heart-lung transplants in 1984 (1), is recognized as a major cause of lung allograft dysfunction following lung transplantation (2-5). Post-transplant OB is characterized by progressive obliteration of small airways (Figure 1) that is typically accompanied by a

persistent decline in spirometric measures of lung function, a spirometric pattern that is usually obstructive, and an essentially clear chest radiograph (4). However, OB is difficult to detect via transbronchial lung biopsy and cannot be confidently diagnosed via non-invasive testing (6-10). Therefore, previously published consensus statements have designated a persistent decline in FEV1 to ≤80% of baseline post-transplant FEV1 that is present for a minimum of 3 weeks (in the absence of confounding conditions) as a surrogate marker of probable OB, and such FEV1 decline has been termed bronchiolitis obliterans syndrome (BOS) (4,5).

The BOS classification scheme adopted in 1993 (4) provided a staging system based on the severity of lung function decline after transplant and has been used for clinical decision-making and research purposes. This grading system was most recently modified in 2002 (Table 3) (5). Baseline values for FEV1 and FEF_{25-75} are defined as the average of the two highest values for each measurement that were obtained at least 3 weeks apart post-transplant without the administration of a bronchodilator. To help distinguish BOS from acute and/or subacute complications of lung transplantation and taking into account the time needed to establish both a baseline FEV1 and a decline in FEV1 ascertained by two FEV1 measurements performed 3 weeks apart, by definition, 3 or more months are required to have elapsed from the time of transplantation in order for the diagnosis of BOS to be made (4,5). Additionally, it has become clear that lung function decline consistent with a diagnosis of BOS can stabilize in some patients and not lead to sustained, progressive deterioration in allograft function and graft loss. Because of concern that setting the cutoff value for FEV1 at 80% of the best post-transplant value may be insensitive to early decline in allograft function due to early OB, stage BOS-0p (≥10% but less than 20% decline in FEV1 and/or ≥25% decline in FEF_{25-75}) was added to the staging system to signify "potential BOS" (5).

BOS affects 50% or more of recipients who survive beyond 5 years and accounts for a considerable proportion of cases of lung allograft loss and recipient death beyond 3 months post-transplant. It is the leading cause of death for recipients who survive beyond one year post-transplant (2,3), and it is widely perceived as the physiological surrogate of immunologically-mediated phenomena due to many observations that include its association with acute cellular rejection (11), the tendency of recipients who develop BOS to have greater degrees of HLA mismatch (12), and accumulating evidence of the involvement of autoimmune pathways (13). Furthermore, there are striking similarities to OB that can occur in allogeneic bone marrow or stem cell transplant recipients as well as patients with connective tissue diseases, which are also perceived as alloimmune or autoimmune disorders respectively. Therefore, BOS is frequently equated with the term chronic rejection. However, various interventions including intensified immunosuppression may have little or no effect on progressive loss of allograft function in patients with BOS. Additionally, many non-immune mechanisms have also been implicated or

suggested as playing a role in BOS pathogenesis. These include airway injury due to primary graft dysfunction (PGD), gastroesophageal reflux (GER), various infections, and airway ischemia due to disruption of the bronchial circulation (14-16). These "non-immune" factors may promote tissue damage and inflammation that in turn initiates and intensifies an alloimmune recipient response. Established OB displays variable evidence of inflammation, alloimmune reactions, autoimmunity, and fibroproliferation with airway obliteration that leads to allograft airway remodeling and loss of function (14-16). OB may represent a final common end-point for a variety of forms of allograft injury.

Because BOS is clinically defined by a persistent decline in lung function, post-transplant decline in FEV1 may be incorrectly perceived as exclusively due to OB. It has been increasingly recognized that other allograft disorders can occur in the chronic post-transplant setting (Table 4), and some of these entities may cause allograft dysfunction that may not be reversible yet meet spirometric criteria for the diagnosis of BOS, as many of these entities may also lead to a sustained decline in FEV_1. The situation can be further complicated by the simultaneous existence of other pathophysiological entities (infection, various forms of rejection, diffuse alveolar damage) when OB is also present in the allograft.

This International Society for Heart and Lung Transplantation (ISHLT)/American Thoracic Society (ATS)/European Respiratory Society (ERS) clinical practice guideline provides a comprehensive, conceptually balanced, and evidence-based perspective that examines the concepts pertaining to the diagnosis and management of BOS that have appeared in the medical literature since this syndrome was first described. It is intended to provide guidance in management whenever possible and to identify gaps in knowledge and issues that need to be addressed via additional basic and clinical research. It should be recognized that the vast majority of patients described in the published literature have been adults, and some recommendations may not have as firm a basis when applied to pediatric lung transplant recipients.

HOW TO USE THESE GUIDELINES

The ATS/ISHLT/ERS guidelines about the management of BOS are not intended to impose a standard of care. They provide the basis for rational decisions in the management of patients with suspected or confirmed BOS. Clinicians, patients, third-party payers, institutional review committees, other stakeholders, or the courts should never view these recommendations as dictates. No guidelines and recommendations can take into account all of the often-compelling unique individual clinical circumstances. Therefore, no one charged with evaluating clinicians' actions should attempt to apply the recommendations from these guidelines by rote or in a blanket fashion.

Statements about the underlying values and preferences as well as qualifying remarks accompanying each recommendation are its integral parts and serve to facilitate more accurate interpretation. They should never be omitted when quoting or translating recommendations from these guidelines.

METHODS

An ISHLT, ATS, and ERS-sponsored Ad Hoc Committee held preliminary meetings in April and May of 2008 to begin the process of identifying and prioritizing topics to be covered in this guideline. The chairs were approved by the three societies. Panel members were identified as leaders in the field of lung transplantation and were selected from established transplant centers worldwide by the chairs to review the existing literature and to answer clinical questions based upon the published evidence or, when such evidence was lacking, provide guidance based upon the observations in their clinical practice. All members of the committee disclosed potential conflicts of interest, which were vetted according to the policies of the ISHLT, ATS, and ERS. Each member of the committee was involved in developing the conclusions and recommendations provided by this document.

A comprehensive literature search was performed by a medical librarian. PubMed interface was used to search Medline for relevant publications (original articles and systematic reviews) in the English language from 1980 through 2009. The search was updated twice in 2012 and in March 2013. The search terms included “lung transplantation”, “bronchiolitis obliterans syndrome” and terms specific to management options considered in the clinical questions. A total of 10,031 manuscripts were identified using the electronic searches. Relevant publications were selected by committee members using pre-specified inclusion criteria, and the bibliographies of selected articles were reviewed to identify additional articles. The pragmatic evidence synthesis was primarily qualitative, rather than quantitative (i.e., few data could be pooled via meta-analysis). The methods used for this guideline are summarized in Table 5.

Members of the committee were provided with the entire collection of compiled documents and subcommittees were formed to address specific topics. Each subcommittee reviewed, appraised, and summarized the relevant evidence. The Grading of Recommendations, Assessment, Development, and Evaluation (GRADE) approach was used to appraise the quality of the body of evidence supporting each recommendation. Clinical questions related to treatment versus no treatment, one treatment versus an alternative treatment, or which populations to treat were answered with recommendations that were formulated and graded using the GRADE approach (Table 6) (17). Disagreements were resolved by discussion and consensus. The final recommendations and grades were reviewed by the entire committee

and approved in September 2013. In contrast to the systematically developed recommendations, other committee's conclusions were based upon the literature appraisal and committee deliberations.

A strong recommendation was made if the committee felt confident of the balance between desirable and undesirable consequences. A conditional recommendation was made if the committee felt less confident of the balance between desirable and undesirable consequences. Factors that influence the strength of recommendations include the estimates of effect for desirable and undesirable outcomes of interest, confidence in these estimates of effects, estimates of values and preferences, and resource use. In any case the appropriate course of action depends upon the clinical context. The committees' judgments about the underlying values and preferences of well-informed patients were based upon the committee members' clinical experience. Evidence tables summarizing the relevant literature for each recommendation are provided in the online supplement.

The committee identified very few experimental studies of the management of BOS. Available data are very limited owing to the small number of subjects. Thus, most of the recommendations are based upon observational studies with or without a control group and the clinical experience of the committee members (i.e., unsystematic clinical observations from their clinical practices).

TERMINOLOGY USED FOR BOS

Several confounding conditions that are potentially reversible may cause delayed decline in allograft function (Table 4). When such entities are excluded and a significant decline in FEV1 meets criteria for BOS, a diagnosis of BOS may be made. However, BOS with obstructive physiology may be distinguished from the recently described entity of restrictive allograft syndrome (RAS), which is characterized by restrictive physiology with evidence of allograft parenchymal fibrosis (18). Therefore, it should be recognized that not all patients in whom a decline in FEV1 and/or airflow obstruction develops necessarily have BOS. Additionally, occult OB may be present in allografts that do not display a significant pattern of FEV1 decline that meets currently accepted criteria for the diagnosis of BOS (19,20).

The term chronic lung allograft dysfunction (CLAD) has been used in reference to BOS and chronic rejection, and these three terms have been used interchangeably in a number of published manuscripts. However, CLAD is a term that needs to have a rigorous and widely accepted definition. The indiscriminate, interchangeable use of these terms may be perceived as indicating that a decline in FEV1

always indicates the presence of OB due to chronic rejection, but FEV1 decline may occur for a variety of reasons as stated above.

BOS PHENOTYPES

The identification of patient groups with specific attributes or patterns of disease may allow the recognition of specific risk factors, pathogenetic disease mechanisms, and/or strategies for treatment and prevention that pertain to an identifiable subset (phenotype) of patients with BOS. Patients with a pattern of early decline in FEV1 that meets BOS criteria may represent a BOS phenotype that has more severe and aggressive OB that is characterized by rapid progression and poor prognosis (19,21-23). However, some patients with rapidly declining lung function may stabilize despite an initial rapid onset and loss of lung function (24). Another potential BOS phenotype suggested in recent literature consists of recipients with significant bronchoalveolar lavage (BAL) neutrophilia who respond to azithromycin therapy (25,26); FEV1 may improve such that the recipient no longer meets spirometric criteria for BOS. These patients appear to have a reversible, BOS-like syndrome associated with BAL neutrophilia, and the recently published, randomized prospective clinical trial conducted by Vos et al. (27) suggested that prophylactic administration of azithromycin initiated shortly after transplantation can suppress the development of this syndrome. Patients who meet BOS criteria but do not respond to azithromycin may represent a phenotype with fibroproliferative OB (25). Nonetheless, distinct phenotypes of BOS that are based upon specific risk factors (Table 7) or other parameters have yet to be definitively established.

RISK FACTORS ASSOCIATED WITH THE DEVELOPMENT OF BOS

Non-minimal (Grade ≥A2) acute cellular rejection and lymphocytic bronchiolitis

Grade A2 or greater acute cellular rejection (AR) on lung biopsy (28) has been linked to subsequent development of BOS (6,29-36). Late AR (30,31,33) and both increasing frequency and severity of AR (6,30,33) have been found to be risk factors for BOS. Most such investigations have found that AR is a major risk factor for BOS even after other clinical events are accounted for by time-dependent Cox regression models and multivariate analyses. Grade B rejection (lymphocytic bronchiolitis, LB) has also been identified as a risk factor for the development of BOS (30,33,34,37,38). See table 3, as well as the online supplement, for a description of the grading of acute cellular rejection.

Our literature search identified no studies that compared augmented immunosuppression with no augmented immunosuppression in patients with non-minimal (Grade ≥A2) AR or LB. Such studies will probably never be done because augmented immunosuppression for non-minimal AR or LB is so widely

accepted that it is unlikely that a control group is possible (i.e., patients are unlikely to accept the chance of being placed in the no augmented immunosuppression group). However, we identified two relevant studies that support the notion that augmented immunosuppression may decrease the risk of BOS among patients with non-minimal AR or LB (33, 39).

The first study was a case-control study that found that patients who developed BOS were more likely to have had inadequate maintenance immunosuppression (cyclosporine, azathioprine, and prednisone with cyclosporine levels <200 ng/mL) than patients without BOS ($p<0.0001$) (33). The second study was a case series that revealed that augmented immunosuppression was associated with improved or eliminated cellular rejection in 54% of patients with Grade A2 AR, 48% with Grade A3 AR, 83% with Grade A4 AR, and 43% with LB; patients whose AR or LB neither improved nor resolved usually remained stable (9). AR and LB are markers (i.e., surrogate measures) of risk for BOS. The series did not measure how many patients developed BOS, nor did it specify which regimen(s) were used to augment immunosuppression. See Table 2a in the online supplement.

When deciding whether or not augmented immunosuppression is warranted, the likelihood of preventing BOS described above must be balanced against the harms of the increased immunosuppression. This balance will vary depending upon the regimen chosen; however, a short course of systemic steroids is the most common regimen selected (40). The best evidence regarding the potential adverse effects of a short course of systemic steroids is indirect, extrapolated from randomized trials conducted in patients having an exacerbation of chronic obstructive pulmonary disease. Such trials have found that short courses of systemic steroids increase the incidence of adverse effects, particularly hyperglycemia and weight gain. See Table 2a in the online supplement.

Our confidence in the accuracy of the reported effects of augmented immunosuppression on the development of BOS in patients with non-minimal AR or LB (i.e, the quality of evidence) is very low because the estimates are derived from an observational study and a case series, which were limited by risk for bias, small sample sizes, indirectness of the population (included all post-transplant patients rather than specifically patients with non-minimal AR or LB (33)), and indirectness of the outcome (measured the change in AR or LB, rather than the development of BOS (39)). Our confidence in the accuracy of the reported adverse effects of systemic steroids is moderate to high because the estimates derive from randomized trials, some of which were limited by a small sample size. See Table 2b in the online supplement.

The committee suggests augmented immunosuppression for patients with non-minimal AR or LB in order to prevent BOS. This is based upon our assessment that the potential benefits of the most common regimen used for augmented immunosuppression (a short course of systemic steroids) outweigh the risks, both in terms of importance (i.e., preventing a life threatening complication versus hyperglycemia and weight gain) and duration (i.e., the potential benefits are long-term, whereas the risks are only short-term and reversible upon discontinuation of therapy). Moreover, a short course of systemic steroids is not overly costly or burdensome and the committee's collective clinical observations suggest that there is a beneficial effect from such therapy in this population of patients.

The recommendation is conditional because the very low quality of evidence provides little certainty that the desirable consequences of augmented immunosuppression exceed the undesirable consequences. Although the initial approach to augmenting immunosuppression in this setting that is generally employed by transplant centers worldwide is to give high-dose corticosteroids intravenously (e.g. methylprednisolone at 1,000 mg daily for 3 days), other therapies may be required (e.g., lymphodepletion-inducing agents) for a rejection episode. Additionally, various changes in the maintenance immunosuppression drug regimen may also be appropriate (see Recommendation 4 below).

Recommendation 1. *For lung transplant recipients who have non-minimal acute cellular rejection (Grade ≥A2) or lymphocytic bronchiolitis on transbronchial lung biopsy specimens, we suggest augmented immunosuppression with a course of systemic steroids to prevent the development of Bronchiolitis Obliterans Syndrome (conditional recommendation, very low quality evidence).*

Values and preferences: This recommendation places a high value on preventing a life-threatening complication of lung transplantation and a lower value on avoiding short-term adverse effects.

Remarks: A typical course of systemic corticosteroids used to augment immunosuppression in adult recipients is intravenous methylprednisolone 1,000 mg daily for three days (many centers use 10-15 mg/kg/day for smaller patients).

Minimal (Grade A1) acute cellular rejection

The significance of minimal AR (Grade A1) rejection is controversial. Hopkins et al. (41) have reported that grade A1 AR remains relatively prevalent on surveillance transbronchial biopsies up to 2 years post-transplant. Available studies suggest that patients with multiple episodes of Grade A1 rejection have an earlier onset of BOS, and a single episode of A1 rejection was found to be independently associated with

progression to BOS (42). See table 3, as well as the online supplement, for a description of the grading of acute cellular rejection.

Our search identified two observational studies that suggest that augmented immunosuppression for minimal (Grade A1) AR may decrease the risk of developing BOS (41,43). In one study, intravenous steroids followed by a tapering course of oral steroids was not associated with development of BOS (p=0.48), whereas lack of treatment with systemic steroids was associated with development of BOS (p=0.01) (43). In the other study, a course of oral steroids reduced progression to higher grades of AR and LB (markers of risk for BOS) by 16.7% and 15.6%, respectively (41). Neither of the studies reported adverse effects from the systemic steroids. See Table 3a in the online supplement.

When deciding whether or not augmented immunosuppression is warranted, the likelihood of preventing BOS described above must be balanced against the harms of the increased immunosuppression. The best evidence regarding the adverse effects of short courses of systemic steroids is indirect, extrapolated from randomized trials conducted in patients having an exacerbation of chronic obstructive pulmonary disease. Such trials have found that short courses of systemic steroids increase the frequency of adverse events, particularly hyperglycemia and weight gain. See Table 3b in the online supplement.

Our confidence in the accuracy of the reported effects of augmented immunosuppression on the development of BOS in patients with minimal AR (i.e., the quality of evidence) is very low because the estimates derive from observational studies that are limited by a risk for bias indirectness of either the outcome (measured progression to higher grades of AR and LB rather than development of BOS (41)) or use of an indirect comparator (looked at associations in treated and untreated patients separately, rather than directly comparing treatment with no treatment (43)). Our confidence in the accuracy of the reported adverse effects of systemic steroids is moderate to high due because the estimates derive from randomized trials, some of which were limited by a small sample size. See Table 3b in the online supplement.

The committee suggests that patients with clinically significant minimal AR be treated with a course of systemic steroids. This reflects the committee's judgment that the possible benefits (i.e., preventing BOS) of therapy exceed the risks (i.e., hyperglycemia, weight gain), cost, and burden in such patients. The rationale for treating minimal AR with systemic steroids is similar to that provided above for treating non-minimal AR and LB with systemic steroids. The recommendation is conditional because the balance of desirable versus undesirable consequences is uncertain due to the very low quality of the evidence, and the uncertainty is reinforced by the committee's clinical experience. In contrast, for clinically stable

patients with grade A1 AR on a surveillance biopsy, the decision about whether to immediately treat the patient with augmented immunosuppression or observe and repeat the biopsy before reaching the decision to augment immunosuppression should be made on a case-by-case basis.

Recommendation 2. *For lung transplant recipients who have clinically significant minimal acute cellular rejection (Grade A1) on transbronchial lung biopsy specimens, we suggest augmented immunosuppression with a course of systemic steroids to prevent the development of Bronchiolitis Obliterans (conditional recommendation, very low quality evidence).*

Values and preferences: This recommendation places a high value on preventing a life-threatening complication of lung transplantation and a lower value on avoiding short-term side effects.

Remarks: We consider Grade A1 acute cellular rejection to be clinically significant if it is associated with clinical findings such as symptoms (e.g. dyspnea, fatigue, new-onset cough) or objective measurements (e.g. decline in FEV1, oxyhemoglobin desaturation with ambulation) that suggest the presence of allograft dysfunction. A typical course of systemic steroids used to augment immunosuppression in adult recipients is intravenous methylprednisolone 1,000 mg daily for three days (many centers use 10-15 mg/kg/day for smaller patients).

Other risk factors

Other risks factors for the development of BOS are discussed in the on-line supplement and include the presence of anti-HLA antibodies (44-48), primary graft dysfunction (49-52), and the presence of significant (abnormal in degree) gastroesophageal reflux (GER) (66-68). Additional associated risk factors include viral infection (29,31,32,57-65), bacterial infection (53-56), and fungal infection (69-71). Cytomegalovirus infection engages both the innate and adaptive components of immunity and causes upregulation of HLA class I and class II antigens on epithelial cells (72,73), and it stimulates and augments the generation of allogeneic immune responses and pro-inflammatory cytokines (72,74). Transient bacterial airway colonization can significantly increase BAL neutrophils and other indicators of lung inflammation (75). Botha et al. (66) examined 155 consecutive lung transplants and reported that *de novo* allograft colonization with *Pseudomonas aeruginosa* was strongly associated with developing BOS within 2 years of transplant, and Vos et al. (67) reported that persistent Pseudomonas colonization was an even greater risk for BOS than *de novo* colonization. Additionally, Gottlieb et al. (68) found that persistent allograft colonization with Pseudomonas in recipients with cystic fibrosis (CF) significantly increased the prevalence of BOS. Valentine et al. (69) identified fungal pneumonia or pneumonitis as an

independent predictor of subsequent BOS, and Weigt et al. (70) reported that *Aspergillus* colonization was independently associated with the subsequent development of BOS.

A prospective study that monitored peripheral blood mononuclear cell responses in 54 lung transplant recipients over a 7-yr period showed a strong association of collagen V-specific responses with the incidence (HR 5.4 for BOS-1, HR 9.8 for BOS-2) and severity of BOS (13), and induction of collagen V reactivity has been associated with abnormal GER and the development of BOS (76). Additionally, Saini et al. (77) found a strong association of the appearance of donor-specific anti-HLA antibodies with the detection of antibodies directed against self-antigens (collagen V and K-α1 tubulin) in a retrospective analysis of 42 lung transplant recipients with BOS.

Numerous studies have shown evidence of neutrophil recruitment and activation when BAL was performed in recipients with acute rejection, infection, and/or BOS (78-81), and Neurohr et al. (82) found that BAL neutrophilia was predictive of subsequent BOS. Schloma et al. (83) also reported that increased BAL neutrophils were associated with early onset BOS, and subsequent investigations by Gottlieb et al. (26) and Vos et al. (25) have also linked BAL neutrophilia to BOS.

Recommendations for mitigating risk factors for BOS are beyond the scope of this guideline, but may be addressed in future guidelines.

DIAGNOSIS OF BOS

An FEV1 decline should trigger concern that graft dysfunction and possibly BOS is evolving, and considerable allograft damage from evolving BOS may have already occurred by the time FEV1 has declined by 20% from its baseline value. When clinically stable patients develop symptoms (e.g. dyspnea, cough, fatigue, fever) and/or signs (decline in FEV1 on home spirometry or at clinic visit follow-up evaluation) that may indicate allograft dysfunction, a comprehensive evaluation to determine cause is typically initiated (Figure 2). This usually includes a routine evaluation in the clinic that is followed by specific testing (imaging, confirmatory spirometry, and bronchoscopy as indicated) to identify a specific cause or causes of lung function decline. If BOS appears to be the cause of lung function decline, treatment approaches discussed in the next section can be considered.

Lama et al. (84) found that the probability of testing positive for BOS-0p by the FEV1 criterion was 71% at two years before the onset of BOS and the specificity of the FEV1 criterion was 93% in single lung transplant (SLT) recipients. Hachem and colleagues (85) reported a positive predictive value of 79% and

negative predictive value of 82% for stage 0-p by FEV1 criteria in 203 adult bilateral lung transplant recipients, but the FEF_{25-75} 0-p criterion had poor predictive value. The prevalence of BOS in the study was 41 to 63% depending upon the criteria used to define BOS. In contrast, Nathan et al. (86) found a 80% sensitivity and 82.6% specificity of the FEF_{25-75} 0-p criterion in a cohort of 43 single lung transplant recipients. Differences between these studies may be related to different statistical techniques, sample size, and follow up time.

HRCT may detect diagnostically useful pleuroparenchymal changes and/or air trapping to which routine CXR is insensitive (87-94). Bronchoscopy with TBLB and BAL is useful to detect infection or other entities that may be the cause of functional decline. Although changes consistent with OB may be obtained via TBLB, non-surgical lung biopsy is insensitive, and a lack of changes of OB on TBLB has poor predictive value. Although many potential biomarkers of BOS have been reported in the literature, none have been validated as having adequate sensitivity and specificity.

Evaluation and/or screening of children for changes in lung function are particularly challenging, and children under 4 years of age may be unable to perform spirometry, necessitating specialized approaches (95,96). Pediatric centers are likely to use alternative lung function measurements or imaging modalities such as ventilation/perfusion scanning and inspiratory/expiratory HRCT scanning to enhance the ability to detect the presence of airflow obstruction. Because TBLB is difficult to perform in infants and small children, many pediatric centers use surgical lung biopsy to confirm a diagnosis of suspected OB (97).

Diagnostic recommendations for BOS are beyond the scope of this guideline, but may be addressed in future guidelines.

TREATMENT AND PREVENTION OF BOS

Intensified pharmacologic immunosuppression has little effect on established BOS in the absence of confounders such as AR, AMR, or lack of BAL neutrophilia.

Long-term high-dose corticosteroids

Sustained treatment with high-dose corticosteroids (≥30 mg/day prednisone or an equivalent) has not been shown to improve BOS, and such therapy is associated with numerous and frequently severe side effects (98). Our search identified a case series of ten patients with lung function decline consistent with BOS (99). All ten patients exhibited progressive lung function decline despite receiving high-dose methylprednisolone. Adverse effects of the sustained high-dose methylprednisolone were not reported in

the case series; however, there is indirect evidence from patients with chronic lung diseases that sustained high-dose corticosteroids are harmful to patients. Specifically, sustained high-dose corticosteroids increase the incidence of osteoporotic fractures, cataracts, and dyspepsia. The findings that sustained high dose corticosteroids induce no beneficial effects on lung function, but cause numerous serious adverse effects, are supported by the collective clinical observations of the committee members. See Table 4a in the online supplement.

Our confidence in the accuracy of the reported effects of sustained high dose corticosteroids on the lung function of patients with a decline in FEV1 consistent with BOS (i.e., the quality of evidence) is very low because the estimates derive from one small case series. Our confidence in the accuracy of the reported adverse effects of systemic steroids varies from low to high depending upon the outcome because the estimates derive from observational studies with one outcome (osteoporotic fractures) upgraded because there was a dose-response gradient and an effect was seen even though confounders would tend to underestimate the effect. See Table 4b in the online supplement.

We suggest not using sustained high dose systemic corticosteroids for patients who have a decline in FEV1 consistent with BOS given the lack of proven benefit and the potential for serious adverse effects. The recommendation is conditional because the evidence provides very low confidence in the effect of sustained high dose systemic corticosteroids.

Recommendation 3. *For lung transplant recipients who develop a decline in FEV1 consistent with the onset of BOS, we suggest that clinicians do NOT use long-term, high-dose corticosteroids (conditional recommendation, very low quality evidence).*

Values and preferences: This recommendation places a high value on avoiding harmful effects due to ineffective therapies.

Remarks: We define sustained administration of high-dose corticosteroid as ≥30 mg/day of prednisone or an equivalent formulation.

Converting cyclosporine to tacrolimus

If patients are receiving CSA-based immunosuppression, switching from CSA to tacrolimus has been reported to slow lung function loss by a number of case series (99-108). However, no randomized trials have been performed to support this switch.

Our search identified ten case series that described the effects of converting cyclosporine to tacrolimus in lung transplant patients with BOS. Six of the series reported mitigation of lung function decline following conversion, while the remaining four series reported improvement of lung function following conversion. Most of the case series did not mention adverse effects; however, those that evaluated nephrotoxicity or hyperglycemia reported a frequent rise in the serum creatinine and glucose levels. None of the case series described infections or malignancy. See table 5a in the online supplement.

There is indirect evidence from randomized trials of patients who have undergone renal transplantation that indicates that tacrolimus does not increase the risk of infection, malignancy, nephrotoxicity, or hyperglycemia when compared with cyclosporine (109). We have no reason to believe that the adverse effects of tacrolimus and cyclosporine are different in lung transplant patients compared with renal transplant patients.

Our confidence in the accuracy of the reported effects of converting cyclosporine to tacrolimus on lung function, nephrotoxicity, and hyperglycemia (i.e., the quality of evidence) is very low because the estimates derive from small case series. See Table 5b in the online supplement.

In lung transplant recipients who develop BOS while receiving a maintenance immunosuppression regimen that includes cyclosporine, we suggest that the cyclosporine be converted to tacrolimus. This reflects the committee's opinion that the likely benefits of mitigation or reversal of lung function decline outweighs the risks of an increase in the serum creatinine and/or glucose levels. The recommendation is conditional because the balance of desirable and undesirable consequences is very uncertain due to the very low quality of the available evidence.

Recommendation 4. *For lung transplant recipients who develop BOS while receiving chronic immunosuppression with a regimen that includes cyclosporine, we suggest switching the cyclosporine to tacrolimus (conditional recommendation, very low quality evidence).*

Values and preferences: This recommendation places a higher value on mitigation of lung function decline and a lower value on avoiding nephrotoxicity and hyperglycemia.

Remarks: The conversion of cyclosporine to tacrolimus is generally performed by stopping cyclosporine and initiating tacrolimus while transiently increasing maintenance corticosteroid dosing until tacrolimus blood levels are ascertained to have reached the desired target range. The target range for therapeutic

trough blood levels of tacrolimus is generally considered to range from 5 to 15 ng/mL for patients who are 18 years of age or older once a steady state has been attained.

Azithromycin

Beneficial effects have been reported for approximately 35-40% of lung transplant recipients treated with azithromycin (25,26,110-116). Complete reversal of FEV1 decline may occur in some patients and patients with BAL neutrophilia appear to represent a subset of patients that are particularly likely to respond to azithromycin therapy (25,26,114).

Our literature search identified ten studies (one observational study and nine case series) that described the effects of azithromycin on the lung function of ~~in~~ lung transplant patients with BOS. The studies found that 30 to 83% of patients had improvement of lung function (defined as an increase in the FEV1 of ≥10%) after receiving azithromycin, even though the mean FEV1 did not increase in some studies because non-responders continued to have lung function decline. In addition, two observational studies ~~found that~~ were identified that described the effects of azithromycin on mortality in lung transplant patients with BOS. Both studies found that early treatment was associated with decreased mortality in some patients. In one study, lung transplant patients with BOS Stage 1 who received azithromycin had lower mortality than those who did not receive azithromycin (HR 0.29, 95% CI 0.11-0.82). The mortality decrease was not seen among patients with BOS Stage 2 (116). In the other study, 40% of patients responded to azithromycin and those patients had a reduction in mortality (HR 0.96, 95% CI 0.95-0.98). Responders tended to receive azithromycin earlier post-transplantation (25). Most of the studies did not mention adverse effects; however, the most common adverse effects reported were nausea, diarrhea, dyspepsia, and colitis, occurring in fewer than 5% of patients. See table 6a in the online supplement.

There is indirect evidence from randomized trials in other conditions that probably better estimates the incidence of adverse effects from azithromycin. A meta-analysis of 12 randomized trials with 1406 patients who received azithromycin to treat an acute lower respiratory tract infection found that 244 out of 1363 patients (17.9%) developed an adverse event (117). Most of the adverse events were minor nausea and diarrhea. Neither the studies identified by our systematic review, nor the meta-analysis of azithromycin for lower respiratory infection, reported fatal cardiac arrhythmias. However, there is other evidence that azithromycin is associated with fatal cardiac arrhythmias. In an observational study that looked at more than one million instances of taking azithromycin, patients who took azithromycin were more likely to suffer a fatal cardiac arrhythmia than those who did not take an antibiotic (RR 2.85, 95% CI 1.13-7.24) (118). The absolute risk of a fatal cardiac arrhythmia during azithromycin therapy was small (1.1 cases per 1000 person-years) and the risk was not increased compared with patients who took

an alternative antibiotic (RR 0.93, 95% CI 0.56-1.55) (118). These findings were supported by another study conducted by the maker of azithromycin (119). There is also evidence from a randomized trial that patients with COPD who are treated with chronic azithromycin therapy are more likely to experience a decrement in hearing and colonization with azithromycin-resistant organisms (120).

Our confidence in the accuracy of the reported effects of a trial of azithromycin on lung function, survival, gastrointestinal distress, and allergic reactions is very low because the estimates derive primarily from case series and a few small observational studies. See Table 6b in the online supplement. Similarly, our confidence in the accuracy of the reported effects of a trial of azithromycin on fatal cardiac arrhythmias is very low because it derives from observational studies in a different patient population. The relevance of the reported fatal cardiac arrhythmias to lung transplant patients is uncertain because lung transplant recipients are uniformly screened to rule out the presence of significant coronary disease or cardiac dysfunction prior to being listed for transplantation. Finally, our confidence in the accuracy of the reported effects of a trial of azithromycin on hearing loss and the acquisition of azithromycin-resistant colonization is moderate because it derives from a single randomized trial with a different patient population.

We suggest a trial of azithromycin in lung transplant recipients who develop BOS. This reflects the committee's judgment that the importance of improved lung function and decreased mortality exceed the risk of minor gastrointestinal distress, decreased hearing, colonization with azithromycin-resistant organisms, rare fatal cardiac arrhythmias, and rare allergic reactions. The recommendation is conditional because our very low to moderate confidence in the reported effects provides limited certainty that the benefits (improved lung function, decreased mortality) exceed the potential adverse events (nausea, diarrhea, fatal cardiac arrhythmias, decreased hearing, colonization with azithromycin-resistant organisms, and allergic reactions,).

Recommendation 5. *For lung transplant recipients who develop a decline in FEV1 consistent with the onset of BOS, we suggest a trial of azithromycin (conditional recommendation, very low quality evidence).*

Values and preferences: This recommendation places a high value on preventing lung function deterioration and possibly reducing mortality, and a lower value on avoiding adverse effects.

Remarks: Azithromycin is generally administered orally at 250 mg per day for five days and then 250 mg three times per week. We define a trial of azithromycin as treating continuously with azithromycin for a

minimum of 3 months. Additionally, it is unclear whether (2) azithromycin should be continued long-term if a beneficial response is observed or (2) whether it should be discontinued if lung function does not show improvement during followup clinical evaluation.

Anti-reflux surgery

Abnormal GER (identified by esophageal pH probe in the majority of studies) is highly prevalent in patients with advanced lung disease and in lung transplant recipients (53,54,56,121-127), and it has been implicated as a risk factor for BOS (53,54,56,128,129). For this reason, committee members routinely test patients with new onset BOS for GER. Proximal gastrointestinal tract motility studies and pH/impedance testing can be used to diagnose motility abnormalities and abnormal acid and/or non-acid GER (130), and examination of BAL for markers of aspiration (e.g. oil red O staining and determination of a lipid index, BAL fluid pepsin, BAL fluid bile acids) has been reported as useful for the detection of microaspiration of refluxed gastroesophageal material (54,131,132). However, additional studies correlating BAL markers of aspiration with GER and BOS are needed to facilitate selection of recipients with BOS who may benefit from interventions such as laparoscopic fundoplication.

Anti-reflux surgery (e.g., Nissen fundoplication, Toupet fundoplication) can be performed safely on lung transplant candidates with advanced lung disease or lung transplant recipients with documented abnormal GER (124,125,128-130,133-139), thereby preventing reflux, aspiration of gastric secretions, and related sequelae.

Our literature search identified three observational studies and five case series that reported the effects of anti-reflux surgery on lung function and mortality in lung transplant recipients. Seven out of eight studies found that the FEV1 improved following anti-reflux surgery (including the two studies that looked specifically at lung transplant recipients with BOS) and the two studies that described long-term survival both reported improved survival after anti-reflux surgery. See table 7a in the online supplement.

With respect to the safety of anti-reflux surgery, one observational study and three case series reported a complication rate of less than 5% when pooled. Similarly, one observational study and six case series reported a peri-operative mortality rate of less than 1% when pooled. Three case series reported that 6 to 14% of patients develop post-operative dysphagia. See table 7a in the online supplement. There is also indirect evidence about the safety of anti-reflux surgery that can be extrapolated from non-transplant patients with GER. Consider the following examples. In one systematic review, a meta-analysis of three randomized trials with 111 non-transplant patients undergoing Nissen fundoplication found that intra-operative complications (e.g., bleeding, liver and spleen capsule tears, and stomach perforation) occurred

in 18% of patients and dysphagia developed in 14% (140). There were no peri-operative deaths. In a similar systematic review, a meta-analysis of five randomized trials with 388 non-transplant patients with GER found that peri-operative morbidity occurred in 14% of patients and dysphagia developed in 17% of patients (141). There were no peri-operative deaths.

There are conflicting data regarding whether lung transplant patients undergoing anti-reflux surgery have a higher incidence of non-fatal peri-operative complications than non-transplant patients undergoing anti-reflux surgery. One retrospective cohort study of 52 patients found no differences in estimated blood loss, duration of surgery, length of hospital stay, complications, or readmission rate (136). In contrast, another retrospective cohort study of 28 patients found a longer post-operative hospital stay (2.9 versus 0.7 days) and higher 30-day readmission rate (25% versus 3.2%) among lung transplant patients than non-transplant patients (23).

Our confidence in the accuracy of the reported effects of anti-reflux surgery on lung function, mortality, and peri-operative complications is very low because they derive from observational studies and cases series limited by indirectness of the population (most studies looked at lung transplant patients with GER in general rather than lung transplant patients with GER and probable BOS) and imprecision of the reported effects owing to few observed events. See table 7b in the online supplement.

We suggest that lung transplant patients with GER who develop a decline in FEV1 consistent with the onset of BOS be referred for potential fundoplication of the gastroesophageal junction. This is based upon the committee's observation that fundoplication may improve lung function and decrease mortality with low risk for peri-operative complications. Our recommendation is conditional because the very low quality of the available evidence provides little certainty that the desirable consequences outweigh the undesirable ones.

Recommendation 6. *For lung transplant recipients who develop a decline in FEV1 consistent with the onset of BOS and have confirmed GER, we suggest referral to an experienced surgeon to be evaluated for potential fundoplication of the gastroesophageal junction (conditional recommendation, very low quality evidence).*

Values and preferences: This recommendation places a high value on reducing the risk of lung function deterioration, and possibly mortality, and a lower value on avoiding surgical complications.

Remarks: Nissen fundoplication has been more extensively studied than Toupet fundoplication; however, we have no reason to believe that one is superior to the other and feel that the choice of the surgical technique should remain at the surgeon's discretion.

Re-transplantation

A number of single-center observational studies and case series have evaluated the outcomes of re-transplantation (142-148). Survival rates have improved significantly in the modern era (146,147), and outcomes following re-transplantation for carefully selected patients with BOS (ambulatory patients selected via the same process used for first-time transplantation) may approach those of first-time lung transplants if performed by experienced centers (143,149).

Our literature search identified four observational studies and three case series that reported the effects of re-transplantation on lung function and survival (142-148). With respect to lung function, freedom from BOS following re-transplantation surgery for BOS was reported to be 85-90% at 1 year, 70-77% at 2-3 years, and 50-77% at 4-5 years. Patients who underwent re-transplantation due to BOS had a higher risk of recurrent BOS than patients who underwent re-transplantation for other reasons, but it is uncertain whether they also had higher risk for BOS than patients who underwent first-time lung transplantation because the results were conflicting. Survival following re-transplantation for BOS was reported to be 60-78% at 1 year, 53-64% at 2 years, and 44-61% at 5 years. Survival was higher among patients who underwent re-transplantation for BOS than among patients who underwent re-transplantation for other reasons, but it was lower than that of patients undergoing primary lung transplantation. See table 8a in the online supplement.

The safety of re-transplantation has not been well studied. We identified no studies that reported peri-operative morbidity and only one observational study that reported peri-operative mortality. In that study (148), there were 39 deaths within 180 days among the 389 patients (10 percent) who underwent re-transplantation. The causes of death were infection, respiratory failure, and multi-organ system failure. Patients undergoing re-transplantation had an increased risk for death after the procedure compared with patients who underwent primary transplantation. See table 8a in the online supplement. Our confidence in the reports of freedom from BOS, survival, and peri-operative mortality (i.e., quality of evidence) is very low because the estimates derive from observational studies and case studies with limitations, usually indirectness of the population. See table 8b in the online supplement.

We suggest referring patients who develop refractory end-stage BOS to a transplant surgeon to be evaluated for re-transplantation for two reasons. First, re-transplantation is usually the only hope for

survival since such patients have already failed alternative interventions. Second, re-transplantation probably improves survival. The survival of those who undergo re-transplantation has been reported to be 45-78% at 1 year and 40-67% at 2 years (142-148). In contrast, survival without re-transplantation has been reported to be 51% at 3 years for all BOS stages (32) and is certainly much lower among those whose BOS is severe enough to require re-transplantation (i.e., patients are usually classified as Stage 3 when referred for re-transplantation), as suggested by the observed 2 to 3-fold increased risk of death with progression of each grade of BOS to a higher grade (e.g. stage 1 to 2, then stage 2 to 3) (23,32). Of note, survival has recently been reported to differ for early versus late onset BOS with a progressive reduction in mortality risk if BOS onset occurs later (e.g. <1, 1-2, 2-3, or >3 years) after transplantation versus earlier onset (150), suggesting that recipients with early onset BOS are at especially high risk for a poor survival outcome. Our recommendation is conditional because the very low quality of the available evidence provides little certainty that the desirable consequences (i.e., potential mortality reduction) outweigh the undesirable consequences (i.e., increased risk of recurrent BOS, peri-operative mortality, and resource utilization); therefore, the appropriate course of action likely depends upon the clinical context.

Recommendation 7. *For lung transplant recipients who have developed end-stage BOS refractory to other therapies, we suggest referral to a transplant surgeon to be evaluated for re-transplantation (conditional recommendation, very low quality evidence).*

Values and preferences: This recommendation places a high value on avoiding surgical complications (e.g., mortality), recurrent BOS, and resource utilization.

Remarks: The selection process for re-transplantation is the same as that used for first-time lung transplantation.

KEY UNANSWERED QUESTIONS AND SPECIFIC RESEARCH NEEDS

Key unanswered questions and research needs are listed in Table 8. Despite the identification of numerous risk factors that that are associated with the onset of BOS, the specific mechanisms by which BOS is initiated in the lung allograft remain unknown, and key mediators of airway injury that can be targeted by specific therapies need to be identified. Additionally, lung function decline characterized as BOS by the FEV1 criterion may be caused by a number of different mechanisms and additional research is needed to understand and characterize complex histopathologic changes that may be present in dysfunctional lung allografts and to identify and characterize BOS phenotypes that can be distinguished

on the basis of clinical, histopathologic, and/or pathogenic mechanisms. As treatments become available that may have a therapeutic effect on the course of BOS, reliable biomarkers of early disease need to be identified to optimize the impact of therapeutic interventions. Guidelines addressing how to detect abnormal GER, select patients for antireflux surgery, and select the appropriate type of antireflux surgery to prevent or treat BOS have yet to be established. In addition, optimal approaches to allograft surveillance (e.g. the role of bronchoscopy with transbronchial biopsies in clinically stable LTX recipients, screening for de novo anti-HLA antibodies and the presence of humoral rejection) have yet to be determined.

A number of complex issues need to be resolved by additional research. These include how to deal with categorization and management of recipients whose spirometry values fluctuate considerably over time, classification of patients who meet criteria for the diagnosis of BOS but subsequently experience a significant improvement in response to therapy (e.g. azithromycin, fundoplication) that leads to clinical and functional (FEV1) improvement such that criteria for BOS are no longer met, and the issue of FEV1 decline that meets BOS criteria when evidence of allograft infection is present. Indeed, infection (e.g. chronic bacterial infection) and OB may coexist and a diagnosis of BOS may only become apparent after a period of time has elapsed (e.g. 3-6 months) when infection has cleared or adequately suppressed and allograft function still does not significantly improve. Additionally, the role of inhaled antibiotics used to prevent or suppress bacterial infection in the prevention or management of BOS needs to be determined.

Advances in understanding the effects of allograft cellular senescence (accelerated aging) on allograft function and BOS risk are needed, and a better understanding of the role of IL-17 (151-157), autoimmune pathways, regulatory lymphocyte populations (158-164), and neutrophil responses as well as mechanisms by which the hypothetical phenomenon of epithelial-mesenchymal transition (which remains hypothetical and has not been well validated in humans) (165-168) leads to airway fibrosis may lead to novel therapies to prevent and treat BOS. Improved animal models of OB are needed and likely to be useful in improving our understanding of the role of these and other phenomena in the initiation, progression, prevention and treatment of OB following lung transplantation.

New methods of allograft conditioning, such as *ex vivo* lung perfusion (EVLP) (169-173) may lead to improved early allograft dysfunction, diminish the risk of PGD, and decrease both the incidence and severity of BOS, but data to determine the impact of EVLP on BOS risk are not yet available. Intensified immunosuppression with total lymphoid irradiation (TLI) (174,175) or extracorporeal photopheresis (ECPP) (176,177) suggest that these interventions may have a significant, beneficial impact on lung function decline due to BOS, but these interventions can have significant adverse effects and may be

associated with significant economic issues, and additional clinical research is required to establish the efficacy of these interventions. Single-center studies have suggested that other immunosuppressive therapies such as sirolimus (178), alemtuzumab (179,180), or anti-thymocyte globulin (181) may play a role in the prevention or management of BOS, but additional research is required to evaluate the utility of TLI, ECPP, or other changes in chronic maintenance immunosuppression by using alternative immunosuppressive agents (e.g. the mTOR inhibitor, everolimus) to prevent or manage BOS. Finally, multicenter trials are needed to better establish optimal regimens for the induction and maintenance of immunosuppression that can adequately prevent both acute and chronic allograft dysfunction yet not lead to excessive risk for infection or other untoward consequences, and collaboration among lung transplant centers to provide adequately powered clinical trials will greatly facilitate the identification of specific risk factors and interventions to both treat and prevent BOS.

CONCLUSIONS

This guideline is intended to enhance the understanding of the diagnosis and management of BOS by transplant physicians and other clinicians and to assist them in the making appropriate clinical decisions when evaluating patients in whom a diagnosis of BOS is suspected. The recommendations in this guideline were informed by a combination of published literature and the clinical observations of experts in the field of lung transplantation. Therefore, they can be used worldwide to help standardize the management of BOS. It is hoped that this guideline will provoke and facilitate future clinical studies in lung transplant recipients who develop delayed loss of allograft function.

Glossary of terms:

AMR – antibody-mediated rejection
OB – obliterative bronchiolitis
BOS – bronchiolitis obliterans syndrome
CARV – community-acquired respiratory virus
CF – cystic fibrosis
CMV – cytomegalovirus
CSA – cyclosporin A
CXR – routine postero-anterior and lateral chest x-ray
FEV1 – forced expiratory volume in one second
FEF25-75 – forced expiratory flow (average from 25-75% vital capacity)
VC – vital capacity
GER – gastroesophageal reflux

HLA – human leukocyte antigen

HR – hazard ratio

HRCT – high-resolution thoracic computed tomographic scan

ISHLT – International Society for Heart and Lung Transplantation

CLAD – chronic lung allograft dysfunction

BAL – bronchoalveolar lavage

LB – lymphocytic bronchiolitis

TBLB – transbronchial lung biopsy

MHC – major histocompatibility complex

MRI – magnetic resonance imaging

RSV – respiratory syncytitial virus

6-MWT – 6-minute walk test

RR – risk ratio

TLI – total lymphoid irradiation

ECPP – extra-corporeal photopheresis

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Table 1. Conclusions

1. The terms BOS and chronic lung allograft dysfunction (CLAD) should not be considered interchangeable or synonymous. Both are clinical terms that describe clinical syndromes. CLAD needs a precise definition, which has not yet been determined.
2. The term BOS should be retained to denote allograft dysfunction with delayed onset and persistent decline in FEV1 (which is usually accompanied by evidence of airflow obstruction) that is not caused by other causes (some of which may be potentially reversible) of post-transplant loss of function.
3. The timing of BOS onset and its subsequent course provide prognostic information and may be linked to different pathophysiological mechanisms.
4. The identification and detailed definition of BOS phenotypes that correlate with prognosis and response to therapy may be useful in understanding the natural course of BOS and the development of more targeted treatment modalities.
5. The following potential risk factors are associated with BOS:
    a. Primary graft dysfunction (PGD)
    b. Acute cellular rejection (AR) including Minimal Grade A1 and higher AR grades
    c. Lymphocytic bronchiolitis (LB) or Grade B rejection
    d. Antibody-mediated rejection (AMR)
    e. Gastroesophageal reflux (GER) (acid and non-acid)
    f. Cytomegalovirus (CMV) pneumonitis
    g. Symptomatic community-acquired respiratory virus (CARV) infection
    h. Colonization and infection of the lung by *Pseudomonas aeruginosa*
    i. *Aspergillus* colonization or fungal pneumonitis
    j. Autoimmune sensitization to collagen V
    k. Increased bronchoalveolar lavage (BAL) neutrophils on BAL differential cell count
6. BOS is generally suspected at an early stage when theFEV1 is ≤90% of baseline (i.e., BOS 0p) and/or the $FEF_{25-75\%}$ is ≤75% of baseline in both bilateral and single lung transplant recipients.
7. In most transplant centers, lung transplant recipients (including asymptomatic patients) receive sustained follow-up including routine clinical evaluation, spirometry (both in the clinic and in remote in-home settings), and other methods for monitoring allograft status (such as fiberoptic bronchoscopy as appropriate). Such monitoring is generally sustained beyond the first 6-12 months following transplantation.
8. When lung transplant recipients who have been clinically stable develop a decline in lung function, prompt clinical evaluation is usually performed to identify the likely cause.

9. Routine postero-anterior and lateral chest x-rays are neither sensitive nor specific for diagnosing BOS.
10. The findings of air trapping with expiratory views and/or mosaic attenuation patterns on HRCT imaging support the presence of BOS, but lack sensitivity and specificity.
11. Thoracic imaging assists in making a diagnosis of BOS by ruling out other causes of allograft function decline.
12. Surveillance bronchoscopy can safely evaluate the lung allograft for occult abnormalities, although a beneficial effect on recipient survival and prevention of BOS has not been clearly demonstrated. In most transplant centers, surveillance bronchoscopy is routinely offered to lung recipients to potentially allow early detection of occult chronic lung allograft dysfunction and/or the presence of occult infection.
13. Although bronchoscopy has poor sensitivity for the diagnosis of OB, bronchoscopy is frequently used to evaluate the lung allograft when evidence of clinical dysfunction is identified.
14. The presence of BAL neutrophilia suggests that OB may be occurring in the lung allograft and that the allograft is at increased risk for progression to BOS; infection is a confounder and may be the cause of BAL neutrophilia, although infection and OB/BOS may coexist in the allograft.
15. The presence of donor-specific antibody (DSA) suggests AMR when detected in context of a delayed allograft functional decline.
16. For lung transplant recipients who develop BOS and have evidence of allograft infection, aggressive measures to control and eradicate infection are routine.
17. Within the various classes of commonly used immunosuppressive agents in lung transplant recipients, there is no definitive evidence of superiority of one drug or drug combination for prevention of BOS.
18. Single-center studies suggest that some less commonly used immunosuppressive agents (i.e., sirolimus, alemtuzumab, and anti-thymocyte globulin) may improve outcomes in patients with BOS.
19. Extracorporeal photopheresis (ECPP) and total lymphoid irradiation (TLI) are therapies that some institutions consider for selected patients with progressive BOS.

Abbreviations: AMR = antibody-mediated rejection; AR = acute cellular rejection; BAL = bronchoalveolar lavage; BOS = bronchiolitis obliterans syndrome; CARV = community-acquired respiratory virus; CLAD = chronic lung allograft dysfunction; CMV = cytomegalovirus; DSA = donor-specific antibody; ECPP = extracorporeal photophoresis; FEV1 = forced expiratory volume in 1 second; FEF25-75 = forced expiratory flow from 25 to 75 % of vital capacity; GER = gastroesophageal reflux; HRCT = high-resolution computed tomography of the thorax; LB = lymphocytic bronchiolitis; OB = obliterative bronchiolitis; PGD = primary graft dysfunction; TLI = total lymphoid irradiation

Table 2. Recommendations

1. For lung transplant recipients who have non-minimal acute cellular rejection (Grade ≥2) or lymphocytic bronchitis on transbronchial lung biopsy specimens, we suggest augmented immunosuppression with a course of systemic steroids to prevent the development of Bronchiolitis Obliterans Syndrome (***conditional recommendation, very low quality of evidence***).
   Values and preferences: This recommendation places a high value on preventing a life-threatening complication of lung transplantation and a lower value on avoiding short-term adverse effects.
   Remarks: A typical course of systemic corticosteroids used to augment immunosuppression in adult recipients is intravenous methylprednisolone 1,000 mg daily for three days (many centers use 10-15 mg/kg/day for smaller patients).
2. For lung transplant recipients who have clinically significant minimal acute cellular rejection (Grade A1) on transbronchial lung biopsy specimens, we suggest augmented immunosuppression with a course of systemic steroids to prevent the development of Bronchiolitis Obliterans Syndrome if the finding of grade A1 acute cellular rejection is perceived to be clinically significant ***(conditional recommendation, very low quality of evidence).***
   Values and preferences: This recommendation places a high value on preventing a life-threatening complication of lung transplantation and a lower value on avoiding short-term side effects.
   Remarks: We consider Grade A1 acute cellular rejection to be clinically significant if it is associated with clinical findings such as symptoms (e.g. dyspnea, fatigue, new-onset cough) or objective measurements (e.g. decline in FEV1, oxyhemoglobin desaturation with ambulation) that suggest the presence of allograft dysfunction. A typical course of systemic steroids used to augment immunosuppression in adult recipients is intravenous methylprednisolone 1,000 mg daily for three days (many centers use 10-15 mg/kg/day for smaller patients).
3. For lung transplant recipients who develop a decline in FEV1 consistent with the onset of BOS, we suggest that clinicians do NOT use long-term, high-dose corticosteroids ***(conditional recommendation, very low quality of evidence).***
   Values and preferences: This recommendation places a high value on avoiding harmful effects due to ineffective therapies. Remarks: We define sustained administration of high-dose corticosteroid as ≥30 mg/day of prednisone or an equivalent formulation.
4. For lung transplant recipients who develop BOS while receiving chronic immunosuppression with a regimen that includes cyclosporine, we suggest switching the cyclosporine to tacrolimus ***(conditional recommendation, very low quality of evidence).***

Values and preferences: This recommendation places a higher value on mitigation of lung function decline and a lower value on avoiding nephrotoxicity and hyperglycemia. Remarks: The conversion of cyclosporine to tacrolimus is generally performed by stopping cyclosporine and initiating tacrolimus while transiently increasing maintenance corticosteroid dosing until tacrolimus blood levels are ascertained to have reached the desired target range. The target range for therapeutic trough blood levels of tacrolimus is generally considered to range from 5 to 15 ng/mL for patients who are 18 years of age or older once a steady state has been attained.

5. For lung transplant recipients who develop a decline in FEV1 consistent with the onset of BOS, we suggest a trial of azithromycin ***(conditional recommendation, very low quality of evidence).***

   Values and preferences: This recommendation places a high value on preventing lung function deterioration and possibly reducing mortality, and a lower value on avoiding adverse effects.

   Remarks: Azithromycin is generally administered orally at 250 mg per day for five days and then 250 mg three times per week. We define a trial of azithromycin as treating continuously with azithromycin for a minimum of 3 months. Additionally, it is unclear whether (2) azithromycin should be continued long-term if a beneficial response is observed or (2) whether it should be discontinued if lung function does not show improvement during followup clinical evaluation.

6. For lung transplant recipients who develop a decline in FEV1 consistent with the onset of BOS and have confirmed GER, we suggest referral to an experienced surgeon to be evaluated for potential fundoplication of the gastroesophageal junction ***(conditional recommendation, very low quality of evidence).***

   Values and preferences: This recommendation places a high value on reducing the risk of lung function deterioration, and possibly mortality, and a lower value on avoiding surgical complications.

   Remarks: Nissen fundoplication has been more extensively studied than Toupet fundoplication; however, we have no reason to believe that one is superior to the other and feel that the choice of the surgical technique should remain at the surgeon's discretion.

7. For lung transplant recipients who have developed end-stage BOS refractory to other therapies, we recommend referral to a transplant surgeon to be evaluated for re-transplantation ***(conditional recommendation, very low quality of evidence).***

   Values and preferences: This recommendation places a high value on avoiding surgical complications (e.g., mortality), recurrent BOS, and resource utilization.

   Remarks: The selection process for re-transplantation is the same as that used for first-time lung transplantation.

Table 3. Grading of bronchiolitis obliterans syndrome (BOS).

BOS Grade	Spirometry (% of baseline)	
	1993 Classification	**2002 Classification**
0	FEV_1 ≥80% of baseline*	FEV_1 >90% of baseline* and FEF_{25-75} >75% of baseline
0-p	Not included	FEV_1 81-90% of baseline and/or FEF_{25-75} ≤75% of baseline
1	FEV_1 66-80% of baseline	FEV_1 66-80% of baseline
2	FEV_1 51-65% of baseline	FEV_1 51-65% of baseline
3	FEV_1 ≤50% of baseline	FEV_1 ≤50% of baseline

*Baseline defined as the average of the two best FEV_1 (or $FEF_{25-75)}$ values (≥3 weeks apart) following functional recovery and stabilization post-LTX

- Other causes of lung function decline must be excluded (e.g. acute rejection, infection, native lung problems for single lung recipients, excessive recipient weight gain, anastomotic dysfunction, respiratory muscle dysfunction, effusion, or technical problems such as erroneous measurements due to device dysfunction)

Table 4. Differential Diagnosis of Delayed Post-Transplant Lung Function Decline

1. Bronchiolitis obliterans syndrome (BOS)
2. Non-BOS alloinflammatory processes
    a. Acute cellular rejection
    b. Lymphocytic bronchiolitis
    c. Antibody-mediated rejection (humoral, vascular)
3. Restrictive allograft syndrome
4. Inflammatory complications of the lung allograft
    a. Pleuro-parenchymal inflammation
        i. Bronchiolitis obliterans organizing pneumonia (BOOP)
        ii. Fibrinoid and organizing pneumonia (FOP)
    b. Chronic inflammation of airways
        i. Large airways (bronchiectasis*, bronchomalacia)
        ii. Bronchioles (follicular or exudative bronchiolitis)
    c. Chronic pleural inflammation
    d. Chronic vascular rejection
5. Infection
6. Surgical removal of lung tissue
7. Mechanical abnormality
    a. Airway dysfunction
        i. Anastomotic stricture/stenosis
        ii. Bronchomalacia (allograft, native airway in SLT)
    b. Allograft compression
        i. Weight gain
        ii. Abdominal distention
        iii. Hyperinflation of native lung in SLT for emphysema
        iv. Pleural complications
            1. Pneumothorax
            2. Pleural effusion
            3. Pleural fibrosis
            4. Bronchopleural fistula
    c. Impaired graft inflation
        i. Pain (vertebral fracture, fracture of ribs and/or sternum)
        ii. Ventilatory compromise
            1. Diaphragmatic dysfunction or paralysis
            2. Chest wall myopathy
        iii. Other (cerebrovascular accident, Parkinson's disease, etc.)
    d. Drug reaction (e.g. sirolimus, everolimus, amiodarone)
    e. Pulmonary edema
    f. Malignancy (PTLD, other)
8. Vascular obstruction
    a. Allograft anastomotic large vessel strictures
    b. Thromboembolic disease
    c. Tumor emboli
9. Allograft parenchymal abnormalities
    a. Transplant indication disease recurrence
        i. Interstitial diseases (e.g. sarcoidosis, PLCH, LAM)
        ii. Other (e.g. veno-occlusive disease, connective tissue disorders)
    b. Diffuse alveolar damage
    c. Organizing pneumonia
10. Aging

Abbreviations: LAM = lymphangioleiomyomatosis; PLCH = pulmonary Langerhans cell histiocytosis; PTLD = post-transplant lymphproliferative disease; SLT = single lung transplant;
*Bronchiectasis may be a manifestation of OB/BOS

Table 5. Methods

Category	Checklist Item	YES	NO
Panel Assembly	Included experts from relevant clinical and non-clinical disciplines	X	
	Included individual who represents views of patients and society at large		X
	Included methodologist with appropriate expertise (documented expertise in development of conducting systematic reviews to identify the evidence base and development of evidence-based recommendations	X	
Literature Review	Performed in collaboration with librarian	X	
	Searched multiple electronic databases	X	
	Reviewed reference lists of retrieved articles	X	
Evidence Synthesis	Applied pre-specified inclusion and exclusion criteria	X	
	Evaluated studies for sources of bias	X	
	Explicitly summarized benefits and harms	X	
	Used PRISMA1 to report systematic review		X
	Used GRADE to describe quality of evidence	X	
Generation of Recommendations	Used GRADE to rate the strength of recommendations	X	

Table 6. Quality of Evidence and Strength of Recommendations.

Quality of Evidence	High	Evidence includes well-designed, well-conducted randomized trials or meta-analyses of randomized trials, without risk of bias, indirectness, imprecision, inconsistency, or publication bias. Alternatively, the evidence may include well-designed, well-conducted observational studies with either a very large effect or at least two of the following: a large effect, dose-response gradient, and/or reverse confounding.
	Moderate	Evidence includes randomized trials or meta-analyses of randomized trials downgraded because of a serious risk of bias, indirectness, imprecision, inconsistency, or publication bias. Alternatively, the evidence may include well-designed, well-conducted observational studies upgraded because of a large effect, dose-response gradient, or reverse confounding.
	Low	Evidence includes well-designed, well-conducted observational studies, or randomized trials or meta-analyses of randomized trials downgraded two levels because of very serious risk of bias, indirectness, imprecision, inconsistency, or publication bias.
	Very low	Evidence consists of case reports, case series, or unsystematic clinical observations (i.e., clinical experience, expert opinion).
Strength of Recommendations	Strong	The committee feels certain that the benefits of the intervention substantially outweigh its risks, burdens, and costs.
	Weak	The committee believes, but is uncertain, that the benefits of the intervention substantially outweigh its risks, burdens, and costs.

Table 7. Risk Factors Associated with Bronchiolitis Obliterans Syndrome.

- Primary graft dysfunction (PGD)
- Acute cellular rejection
- Lymphocytic bronchiolitis
- Humoral rejection (e.g. *de novo* anti-HLA antibodies)
- Gastroesophageal reflux and microaspiration
- Infection
    - Viral
    - Bacterial
    - Fungal
- Persistent neutrophil influx and sequestration (BAL neutrophilia)
- Autoimmunity (collagen V sensitization)

Table 8. **Key Unanswered Questions and Research Needs.**

Unanswered Questions:

1. What are the roles and mechanisms of alloimmune and autoimmune responses in BOS pathogenesis?
2. Does antibody-mediated rejection play a role in BOS onset and progression?
3. What is the significance of the appearance of *de novo* anti-HLA antibodies in BOS pathogenesis, and when and how should screening and treatment for anti-HLA antibodies be performed?
4. Can specific biomarkers identify and reliably predict increased risk for the development of BOS, and can such biomarkers be used to detect the early (subclinical) onset of BOS?
5. Can specific BOS phenotypes be identified that are useful for predicting prognosis and response to therapy?
6. What specific agent or combinations of post-transplant immunosuppressive agents are most likely to prevent BOS and improve allograft and patient survival?
7. Does any early, specific therapy significantly alter the natural history of BOS?
8. When lung retransplantation is performed for end-stage BOS, is the retransplanted lung at increased risk for the development of rejection and/or OB?
9. Can patients who are more tolerant to their grafts and, therefore, require less intense immunosuppression be identified?
10. Can induction of tolerance to self-antigens (e.g. collagen V) or strategies to augment regulatory T or B cells to promote and maintain tolerance diminish risk for BOS?
11. Will the use of *ex vivo* lung perfusion (EVLP) techniques to condition the lung allograft diminish the risk of developing BOS?
12. What is the optimal frequency for obtaining spirometry to assist in the early detection of evolving BOS?

Research Needs:

1. Multi-center clinical investigations are needed to identify and assess risk factors for BOS.
2. Multi-center clinical trials are needed to evaluate potentially therapeutic interventions to treat BOS as well as strategies to prevent its onset.
3. Additional studies of mechanisms and phenotypes (animal models and lung allograft recipients) are needed.
4. Guidelines for optimal testing for abnormal GER and the selection of patients (and procedure) for antireflux surgery to prevent or treat BOS.
5. Identification of optimal approaches to allograft surveillance (e.g. the role of bronchoscopy with transbronchial biopsies in clinically stable LTX recipients, screening for de novo anti-HLA antibodies and the presence of humoral rejection).
6. Improved animal and other laboratory models of OB to better understand its pathogenesis and identify key mediators of airway inflammation and fibrosis.

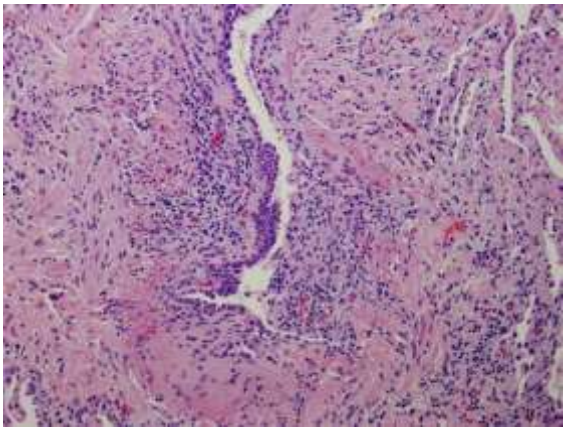

Figure 1a

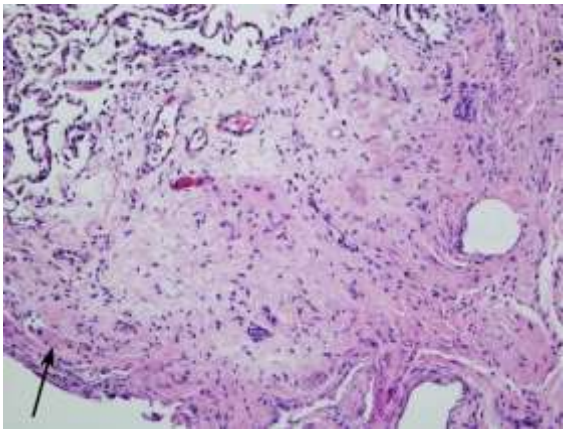

Figure 1b

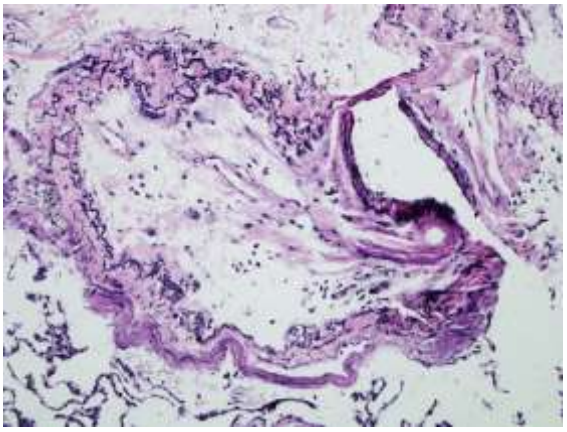

Figure 1c

Figure 1a: Bronchiolitis obliterans in a surgical lung biopsy with partial luminal compromise accompanied by mild chronic inflammation in the wall and focal ulceration of the mucosa.

Figure 1b: Bronchiolitis obliterans on a transbronchial biopsy with complete luminal obliteration. Scant bronchiolar muscle (arrow) helps to identify the scarred structure as residual airway (hematoxylin and eosin).

Figure 1c: An elastic tissue stain from a slightly deeper section of the same bronchiole (1b) highlights the residual elastica present. In contrast to the accompanying artery on the right, there is only one elastic lamella in the bronchiolar wall.

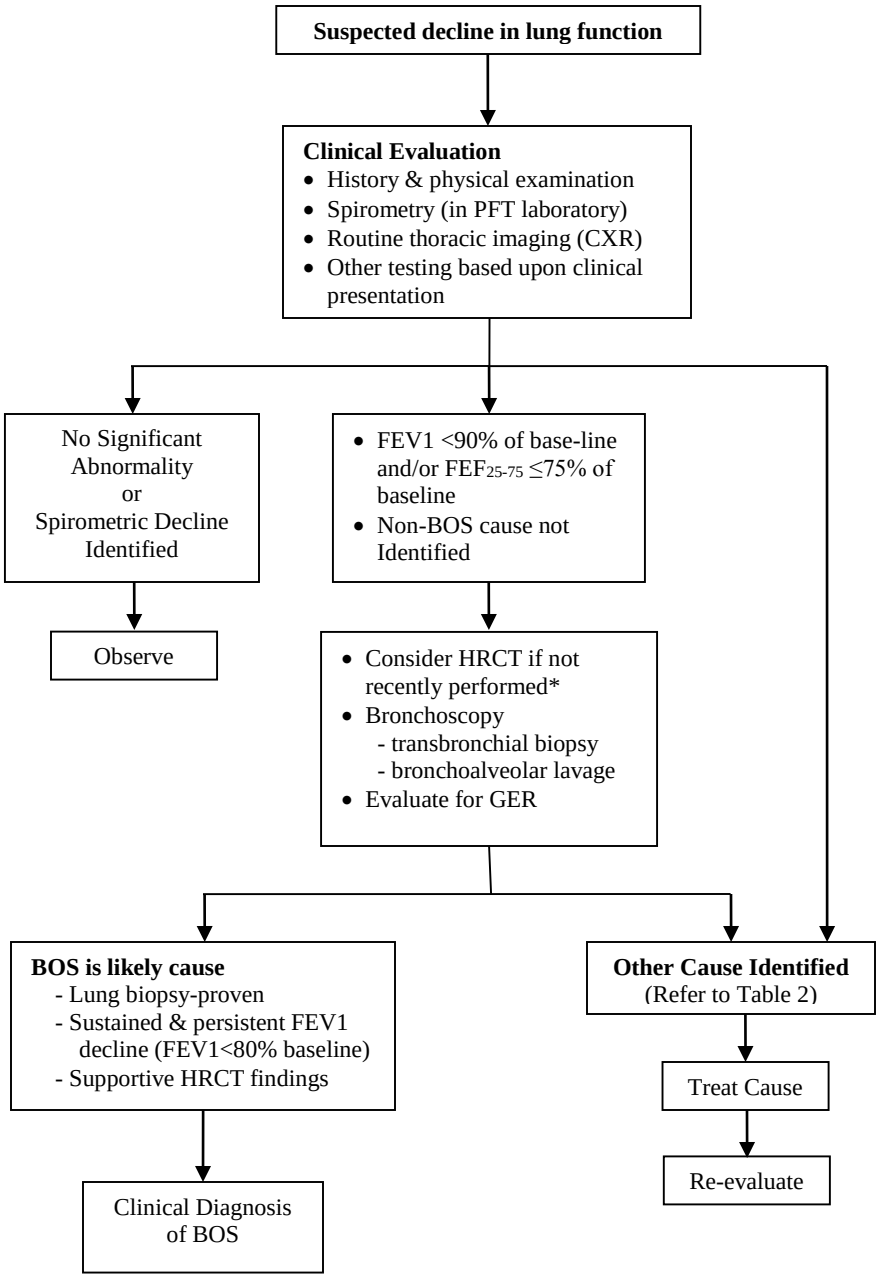

Figure 2. Algorithm for clinical evaluation of suspected BOS. This algorithm is a description of the collective clinical practices of the committee members. It is not based upon systematically-developed evidence-based diagnostic recommendations.
*Obtain both inspiratory and expiratory views to evaluate for air trapping