Aspiration, Localized Pulmonary Inflammation, and Predictors of Early-Onset Bronchiolitis Obliterans Syndrome after Lung Transplantation

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BACKGROUND: We hypothesized that immune mediator concentrations in the bronchoalveolar fluid (BALF) are predictive of bronchiolitis obliterans syndrome (BOS) and demonstrate specific patterns of dysregulation, depending on the presence of acute cellular rejection, BOS, aspiration, and timing of lung transplantation.

STUDY DESIGN: We prospectively collected 257 BALF samples from 105 lung transplant recipients. The BALF samples were assessed for absolute and differential white blood cell counts and 34 proteins implicated in pulmonary immunity, inflammation, fibrosis, and aspiration.

RESULTS: There were elevated BALF concentrations of interleukin (IL)-15, IL-17, basic fibroblast growth factor, tumor necrosis factor−α, and myeloperoxidase, and reduced concentrations of α_1-antitrypsin, which were predictive of early-onset BOS. Patients with BOS had an increased percentage of BALF lymphocytes and neutrophils, with a reduced percentage of macrophages ($p < 0.05$). The BALF concentrations of IL-1β; IL-8; interferon-γ−induced protein 10; regulated upon activation, normal T-cell expressed and secreted; neutrophil elastase; and pepsin were higher in patients with BOS ($p < 0.05$). Among those with BOS, BALF concentrations of IL-1RA; IL-8; eotaxin; interferon-γ−induced protein 10; regulated upon activation, normal T-cell expressed and secreted; myeloperoxidase; and neutrophil elastase were positively correlated with time since transplantation ($p < 0.01$). Those with worse grades of acute cellular rejection had an increased percentage of lymphocytes in their BALF ($p < 0.0001$) and reduced BALF concentrations of IL-1β, IL-7, IL-9, IL-12, granulocyte colony-stimulating factor, granulocyte-macrophage colony-stimulating factor, interferon-γ, and vascular endothelial growth factor ($p \leq 0.001$). Patients with aspiration based on detectable pepsin had increased percentage of neutrophils ($p < 0.001$) and reduced BALF concentrations of IL-12 ($p < 0.001$).

CONCLUSIONS: The BALF levels of IL-15, IL-17, basic fibroblast growth factor, tumor necrosis factor−α, myeloperoxidase, and α_1-antitrypsin at 6 to 12 months after lung transplantation are predictive of early-onset BOS, and those with BOS and aspiration have an augmented chemotactic and inflammatory balance of pulmonary leukocytes and immune mediators. These data justify the surgical prevention of aspiration and argue for the refinement of antirejection regimens. (J Am Coll Surg 2013;217:90−101. © 2013 by the American College of Surgeons)

Lung transplantation patients continue to have the worst survival of all solid organ transplant recipients, despite attempts at refining surgical technique and antirejection regimens.[1] The reduced survivability after lung transplantation is multifactorial and involves donor-related factors, primary graft dysfunction, allorecognition, and bronchiolitis obliterans syndrome (BOS), which is characterized by progressive fibrous obliteration of the small

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Abbreviations and Acronyms

ACR = acute cellular rejection
A1AT = α_1-antitrypsin
BALF = bronchoalveolar fluid
BOS = bronchiolitis obliterans syndrome
GERD = gastroesophageal reflux disease
GM-CSF = granulocyte-macrophage colony-stimulating factor
IPO-10 = interferon-γ–induced protein 10
IQR = interquartile range
LARS = laparoscopic antireflux surgery
MPO = myeloperoxidase
NE = neutrophil elastase
OR = odds ratio

airways.[2,3] Affecting half of lung transplant recipients by 5 years,[1] BOS is also a multidimensional process that appears to involve both alloimmune and non-alloimmune factors, such as ischemia/reperfusion, infection, and gastroesophageal reflux disease (GERD)–related aspiration.[4]

Our work and that of others has identified GERD as exceedingly common among lung transplant recipients.[5-9] In addition, we have affirmed that the surgical correction of GERD is not only safe after lung transplantation,[10-12] but that it can also stabilize, if not prolong, pulmonary function.[7,8,13,14] Our most recent findings have demonstrated prevention of aspiration by way of reduced pepsin levels in the bronchoalveolar lavage fluid (BALF) after laparoscopic antireflux surgery (LARS),[14] which appears to parallel a less proinflammatory and fibrogenic environment within the pulmonary allograft.[15,16]

The aim of our current study was to characterize the biologic changes that occur with BOS, acute cellular rejection (ACR), and aspiration. In addition, we hoped to identify a unique pattern of immune mediators within the BALF that would be predictive of early-onset BOS when measured within the first year after lung transplantation. We hypothesized that a proinflammatory and fibrogenic pulmonary microenvironment is characteristic of ACR, aspiration, development of BOS, and timing of lung transplantation.

METHODS

Patients and parameters

From September 2009 to January 2012, there were 105 lung transplantation patients prospectively enrolled, on whom transbronchial biopsy and bronchoalveolar lavage were performed during routine surveillance or when clinically indicated by reduced pulmonary function on spirometry. At our institution, surveillance bronchoscopy is performed 1, 3, 6, 9, and 12 months after transplantation. Clinical variables and outcomes of interest were recorded, including age, sex, indication for transplantation, time since transplantation, identification of ACR by transbronchial biopsy, diagnosis of BOS, presence of GERD, and evidence of aspiration as determined by measureable pepsin in the BALF.

All study subjects provided informed consent. Participants were excluded for the following: age younger than 18 years, combined heart and lung transplantation, malignancy, current smoking, and pregnancy. This study was approved by the Loyola University Medical Center Institutional Review Board (LU202400).

Pulmonary function testing

All lung transplantation patients underwent serial pulmonary function testing according to institutional protocol, with spirometry and flow volume assessments performed at each clinic appointment and with any substantial change in respiratory symptoms. This generates a schedule of post-transplantation documentation of the forced expiratory volume in 1 second once per week for the first month, twice monthly for the next 2 months, then every third month, or more frequently depending on clinical indication. Additionally, full pulmonary function testing with and without bronchodilators is performed 6 months post transplantation, and annually thereafter. All forced expiratory volume in 1 second data consist of pulmonary function assessment without bronchodilators.

Immunosuppression

The standard maintenance immunosuppression regimen at our institution includes a calcineurin inhibitor (tacrolimus), an anti-metabolite (azathioprine or mycophenolate mofetil), and steroids. Patients routinely received induction immunosuppression with either basiliximab or daclizumab, with the exception of those patients seronegative for cytomegalovirus receiving an allograft from a cytomegalovirus-seropositive donor.

Esophageal function testing

Esophageal function testing was undertaken as we have described previously.[5] Briefly, proton pump inhibitors were stopped for 14 days and histamine H_2-receptor antagonists were stopped for 3 days before pH monitoring. A pH catheter (Sleuth system with BioVIEW software; Sandhill Scientific Inc.) was placed with the distal pH sensor positioned 5 cm superior to the manometrically determined upper border of the lower esophageal sphincter. The DeMeester score was calculated for the distal pH recordings, and a score >14.7 was considered diagnostic of GERD.