CURRENT STATUS OF LUNG TRANSPLANTATION

The current status of the field over the past several years was summarized in the 2005 Official Report of the Registry of the International Society for Heart and Lung Transplantation (ISHLT) (1). The overall level of activity reported in 2003 was close to that of 2002, including 931 bilateral- and 772 single-lung transplantations, and 74 heart–lung transplantations. The number of bilateral procedures surpassed the number of single procedures by a larger amount in 2003 than in 2002, and the number of heart–lung procedures continued to decrease. The primary indications were chronic obstructive pulmonary disease (COPD)/α1-antitrypsin deficiency for 61.2% of all single procedures and 32% of all bilateral procedures, idiopathic pulmonary fibrosis (IPF) for 24% of all single procedures, and cystic fibrosis (CF) for 32% of all bilateral procedures. Survival rates at 3 yr have improved from 55.7% for patients undergoing transplantation surgery between 1988 and 1994 to 63.3% for patients undergoing transplantation between 2000 and June 2003.

DONOR ISSUES

Two studies examined the use of donors with marginal oxygenation. Luckraz and colleagues retrospectively analyzed outcomes of 362 consecutive heart–lung and lung transplant recipients according to whether they had received organs from “low PaO2” donors (PaO2, 225–300 mm Hg on 100% FiO2) or “normal” PaO2 donors (PaO2 > 300 mm Hg) (2). There was a trend toward lower 30-d survival (p = 0.08) in the low PaO2 cohort but no difference in 1- or 5-yr survival rates. In a more elaborate multicenter study involving 785 patients, Thabut and colleagues used multivariate analysis to define the relationship between donor characteristics and outcomes (3). Among donor-related variables, only the PaO2/FiO2 ratio was found to be predictive of early post-transplant graft function, as assessed by recipient PaO2/FiO2 ratio within the initial 6 h and by duration of mechanical ventilation. In contrast to the findings of the Luckraz and colleagues’ study, a low donor PaO2/FiO2 ratio (< 350) was associated with a significantly increased risk of death after transplantation (hazard ratio [HR], 1.43; 95% confidence interval, 1.10–1.85). Fischer and colleagues found no difference in early gas exchange, duration of mechanical ventilation, length of intensive care unit (ICU) stay, or short- and long-term survival rates among recipients of organs from donors 50 yr and older versus donors who were younger than 50 yr (4).

Hartwig and colleagues analyzed outcomes of 29 patients who received allografts from donors who tested positive for hepatitis B core antibody (5). All recipients had been immunized with hepatitis B vaccine before transplantation and none developed evidence of viral transmission post-transplantation, suggesting that this practice is safe. In contrast, all three recipients of lungs from hepatitis C antibody–positive donors tested positive for this virus post-transplantation, and one of these patients died of accelerated viral hepatitis.

Gamez and colleagues reported on their initial experience with lung transplantation using non–heart-beating donors (6). Five procedures were performed, with total ischemic times ranging from 9 to over 11 h. All patients were successfully discharged from the hospital and were “alive and well” 2 to 13 mo after the procedure.

LUNG PRESERVATION

Using a swine model, Roe and colleagues examined the impact of various pulmonary vascular flush techniques on bronchial mucosal temperature and regional lymph node production of inflammatory cytokines (7). They found that antegrade pulmonary artery flush combined with bronchial artery flush resulted in a greater degree of cooling of the bronchial mucosa than did the more conventional techniques of antegrade pulmonary artery flush with or without retrograde pulmonary vein flush. In addition, the antegrade pulmonary artery/bronchial artery route resulted in down-regulation of IFN-γ expression in regional lymph nodes.

Sommer and coworkers assessed the impact of adding the free-radical scavenger glutathione to low potassium dextran flush preservation solution in a swine lung transplant model (8). After 24 h of cold storage, transplantation of lungs flushed with the glutathione-containing solution resulted in lower pulmonary vascular resistance and pulmonary artery pressures, and higher PaO2/FiO2 ratios. This improvement was associated with a decrease in neutrophil sequestration and lung water accumulation within the allograft, suggesting that glutathione attenuated the magnitude of ischemia-reperfusion injury.

Wittwer and colleagues evaluated the effect of donor pre-treatment with inhaled iloprost before harvest and protracted cold ischemic storage in a pig model (9). Early post-transplant measurements of lung compliance and vascular resistance were significantly better in the iloprost-treated group compared with control animals, and the treated allografts were less edematous. Similar results were achieved when inhaled iloprost was used in non–heart-beating animal donors (10), suggesting that this technique may be valuable in enhancing the function of allografts harvested from this emerging lung donor pool.

ISCHEMIA-REPERFUSION INJURY AND PRIMARY GRAFT DYSFUNCTION

Epidemiology

Ischemia-reperfusion injury, which manifests clinically as primary graft dysfunction (PGD), remains a major complication that contributes significantly to early post-transplantation mortality, increases costs, and leads to protracted and often compromised recovery among survivors (11, 12). Christie and colleagues provided a comprehensive view of the epidemiology of PGD through analysis of data on 5,262 patients in the United Network
for Organ Sharing (UNOS)/ISHLT registry (13). They found an overall incidence of PGD of 10.2%. The 30-d mortality rate for patients with PGD was 42% compared with only 6% for those without this complication. The risk of death remained excessive even beyond the first year, suggesting that PGD has lingering adverse consequences well after resolution of the acute event.

**Pathogenesis**

Using oligonucleotide microarrays, Yamane and colleagues examined changes in gene expression in a rat model of ischemia-reperfusion injury (14). Among the genes demonstrated to undergo up-regulation were those encoding for proinflammatory cytokines, adhesion molecules, chemokines, and procoagulants. This technique may ultimately facilitate identification of new targets for therapeutic intervention.

Two studies focused on the role of factors involved in the regulation of apoptosis, specifically the transcription factor nuclear factor-κB (15) and caspase (16). The studies demonstrated that these factors were up-regulated after experimental transplant-related ischemia-reperfusion injury, and that inhibition of these factors attenuated the severity of lung injury and the extent of apoptosis.

**Risk Factors**

Using a database encompassing 752 patients from seven centers in France, Thabut and colleagues performed Cox regression analysis and found an independent, inverse relationship between graft ischemic time and recipient PaO2/FiO2 ratio within the first 6 h post-transplantation (17). In addition, a direct relationship between graft ischemic time and mortality was demonstrated. The risk of death was greatest within the first year and increased sharply in association with ischemic times of greater than 330 min.

Pilcher and coworkers explored donor, recipient, and procedure-related parameters predictive of early graft dysfunction (18). Higher donor age, lower donor PaO2/FiO2 ratio, and recipient need for higher doses of inotropes were found to be independently associated with lower graft PaO2/FiO2 ratios within the initial 24 h.

**Prognostic Factors**

In a retrospective review of 68 bilateral lung transplant recipients, Oto and colleagues found that the PaO2/FiO2 ratio at 6 h was a useful prognostic parameter that correlated with duration of intubation, length of ICU stay, and 30-d mortality (19). Pilcher and colleagues corroborated the association between low PaO2/FiO2 ratio and inferior outcomes (18). In addition, they found that an elevated central venous pressure measured within the first 3 d was independently predictive of a prolonged need for mechanical ventilation and ICU care, and increased early mortality.

**Treatment**

In a prospective placebo-controlled trial involving 59 patients, Keshevjee and colleagues examined the ability of an inhibitor of complement activation to attenuate ischemia-reperfusion injury (20). The rate of extubation by 24 h was significantly higher in the treatment arm (50 vs. 19%), though the reason for this was not readily apparent because neither the initial PaO2/FiO2 ratio nor the extent of opacities on chest radiography differed between the two groups. There was no significant difference in length of hospital stay or short-term mortality.

**RECIPIENT SELECTION**

Two studies addressed the issue of selection of appropriate candidates with CF. Using the large CF Foundation Patient Registry and the UNOS database, Liou and colleagues used Cox proportional hazards models to identify variables predictive of an increased risk of post-transplantation mortality and to assess the impact of these variables on survival benefit (21). No survival benefit was provided by transplantation in patients younger than 18 yr, patients with CF-related arthropathy or pretransplant colonization with *Burkholderia cepacia*, and patients with a predicted 5-yr “natural history” survival of greater than 50% (as determined by a previously validated survival model of CF). Ellaffi and coworkers found that nearly half of the patients with CF admitted to their ICU for severe pulmonary exacerbations died during the hospitalization or within 8 mo of discharge, suggesting that these patients should be considered for urgent listing if they are otherwise suitable candidates (22).

**IMMUNOSUPPRESSION**

In a phase I–II study, McCurry and colleagues treated 48 patients with either Campath-1H (alemtuzumab) or Thymoglobulin (antilymphoid antibody preparation) immediately before transplantation, with maintenance immunosuppression consisting of tacrolimus only, or tacrolimus and low-dose prednisone (23). Patients treated with Campath-1H had fewer and less severe acute rejection episodes than patients treated with Thymoglobulin, and both groups tended to have a better survival than historical control subjects treated with daclizumab induction and triple immunosuppression.

Two studies assessed tacrolimus pharmacokinetics in stable lung transplant recipients with and without CF, at least 3 mo after surgery (24, 25). The trough concentration (C0) was poorly correlated with tacrolimus exposure (as assessed by the area under the concentration curve) in both patient groups, but concentrations at 3 and 4 h (C3 and C4) postdose performed much better. The best predictive performance was obtained with 2 (e.g., C0, C4) or more sampling points. In a study of 48 de novo recipients, Akhlaghi and colleagues showed that cyclosporine C2 concentration correlated better with the incidence of acute rejection than did C0; an average C2 between 1,000 and 1,500 μg/L within the first postoperative month was associated with better graft outcomes (26).

In a prospective randomized trial, Snell and colleagues compared cellular and cytokine profiles in endobronchial biopsies and/or bronchoalveolar lavage fluid obtained in 13 patients treated with everolimus versus 10 patients treated with azathioprine (27). Patients receiving everolimus showed a significant reduction in the percentage or number of CD4 lymphocytes in the lavage, and a decrease in CD4 lymphocytes, CD4/8 ratio, and number of neutrophils in the biopsies. This immunologic profile might be associated with a lower incidence of bronchiolitis obliterans syndrome (BOS).

**MEDICAL COMPLICATIONS**

In a large, retrospective, single-center study, Rocha and colleagues reported an incidence of acute renal failure of 56% within 2 wk after transplantation, with 8% of the patients requiring dialysis (28). Patients requiring dialysis were more likely to have pulmonary hypertension and IPF as preoperative diagnoses; other independent predictors of the need for dialysis were baseline glomerular filtration rate, mechanical ventilation for more than 1 d, and parenteral amphotericin B use. One-year survival for this subgroup was only 22%, with a median time to death of 33 d.

Mughal and colleagues reported their experience with uncovered self-expanding metallic stents in seven patients with life-threatening bronchial dehiscence that could not be treated by open surgical repair (29). Stent placement resulted in complete
bronchial healing in all patients and was followed by successful weaning in the three patients who required mechanical ventilation.

BOS AND ACUTE REJECTION

Risk Factors

In a prospective analysis of 272 bronchoalveolar lavage samples, D’Ovidio and colleagues found that levels of bile acids were increased above the normal serum upper limit in 20 of 120 stable lung transplant recipients (17%), consistent with reflux from the duodenum (30). Bile acid levels were greater in patients with than without BOS, in particular if BOS had developed during the first postoperative year. Patients with high levels of bile acids also had increased alveolar neutrophilia and interleukin-8 levels, and early BOS development. Ward and colleagues measured pepsin levels in the bronchoalveolar lavage fluid of four normal subjects and 13 lung transplant recipients without BOS (31). Pepsin was absent in the normal subjects, but was increased in all patients, suggesting that gastric aspiration is common after lung transplantation and may be detected by the presence of pepsin in bronchoalveolar lavage fluid.

In a retrospective study in 228 patients monitored for 7 yr, Khalifah and colleagues assessed minimal (A1) rejection as an independent risk factor for BOS (32). Building on the previous study by Hopkins and colleagues (33), they found that patients with A1 rejection were two to four times more likely to develop BOS at stages 1 through 3 than recipients without BOS, and that the risk associated with minimal rejection was comparable to that associated with more severe acute rejection.

Infections with community-acquired respiratory viruses (34), Chlamydia pneumoniae (35, 36), and human herpesvirus-6 (37) may all increase the risk of developing acute rejection and/or BOS. Intravenous ribavirin for respiratory syncytial virus infection may help prevent BOS development (38).

Pathogenesis

Girnita and colleagues prospectively studied 51 patients during 4 yr to determine the direct impact of HLA-specific antibodies on BOS (39). Twelve patients developed de novo HLA antibodies after transplantation, in general during the first postoperative year, with donor specificity in seven patients. HLA antibodies were associated with persistent-recurrent acute rejection, lymphocytic bronchiolitis, and BOS. On average, antibodies were detected 1 yr before BOS onset, and they had a cumulative effect on the risk of BOS when associated with persistent-recurrent acute rejection or lymphocytic bronchiolitis. These findings are in keeping with the observation made by Maruyama and colleagues in murine heterotopic tracheal allografts that anti-HLA class I antibodies play an important role in the pathogenesis of obliterative airway disease by inducing growth factor production, apoptosis, and chemotaxis of inflammatory cells (40).

To assess whether innate immunity contributes to the development of acute rejection beyond the first 6 postoperative mo and to BOS, Palmer and colleagues correlated clinical outcomes in 170 lung transplant recipients with either of two loss-of-function polymorphisms in Toll-like receptor 4: Asp299Gly or Thr399Ile (41). Of the patients, 11% were heterozygous for one or both polymorphisms; this subgroup showed a reduced frequency, severity, and onset of acute rejection, and a trend toward reduced BOS 2 and 3. The authors concluded that activation of innate immune signaling contributes to the development of acute rejection and possibly higher BOS grades.

Leukotriene B

is a lipid mediator with potent chemotactic activity for effector T lymphocytes through its receptor BLT1. Using a novel transgenic murine model of antigen-specific T-cell–induced acute rejection (bronchiolitis), two murine hetero-
topic tracheal transplantation models, and studies of bronchoalveolar lavage fluid from patients with and without BOS, Medoff and colleagues convincingly showed that BLT1 contributes to the development of acute rejection and BOS by mediating CD8+ cell trafficking into the airways. BLT1 might thus be a new therapeutic target after lung transplantation (42).

Ward and colleagues performed a study to assess whether epithelial–mesenchymal transition (EMT) may be the source of the activated fibroblasts responsible for airway fibrosis after lung transplantation (43). Using staining of endobronchial biopsy specimens and cultures of primary bronchial epithelial cells obtained by airway brushings in patients without BOS, they showed the presence of both early (e.g., the human homolog of fibroblast specific protein 1, S100A4) and late (matrix metalloproteinases [MMPs]) markers of EMT. In a cross-sectional study, Taghavi and colleagues found that levels of MMP-2, MMP-8, and MMP-9 and their tissue inhibitors (TIMPs) in the bronchoalveolar lavage fluid increased markedly on going from BOS stage 0 to BOS 0p and BOS 1 (44). It is not yet clear, however, whether EMT and MMP production are merely a homeostatic reparative, or a deleterious pathologic, mechanism.

Murakawa and colleagues used a murine orthotopic tracheal transplantation model to assess the role of leukocyte function associated antigen-1 (LFA-1) and CD40L pathways in the pathogenesis of alloimmune-mediated airway injury (45). They found that early administration of anti–LFA-1/anti-CD40L antibodies altered antidonor reactivity and prevented epithelial lesions and subepithelial fibrosis, in part through dampening of CXC chemokine responses and by preventing the replacement of donor-derived epithelium by that derived from recipients. Additional experiments showed that the development of epithelial chimerism after relining of donor airways with recipient-dervied epithelium was strongly associated with airway remodeling.

Belpiero and colleagues found evidence of marked vascular remodeling in the airways of patients with BOS (46). In very detailed studies using both the analysis of bronchoalveolar lavage fluid from patients with and without BOS and the murine heterotopic tracheal transplantation model, they found that levels of multiple CXC chemokines were increased in BOS. Interaction of these chemokines with their receptor (CXCR2) induced a neovascularization that contributed to airway fibroproliferation. This angiogenic activity was independent of vascular endothelial growth factor (VEGF). In contrast, using a similar animal model, Krebs and colleagues found up-regulation of VEGF expression on tracheal smooth muscle cells and graft-infiltrating mononuclear inflammatory cells (47); VEGF promoted microvascular remodeling and fibroproliferation through increased platelet-derived growth factor signaling. Langenbach and colleagues reported an increase in large airway vascularity in 27 lung transplant recipients, but VEGF was not increased in bronchoalveolar lavage fluid and airway vascularity did not differ according to the BOS stage (48). Altogether, these studies emphasize the key role of angiogenesis-mediated airway fibroproliferation in the pathogenesis of BOS (49), though the complex mechanisms promoting angiogenesis still need to be clarified.

Lung Function

To assess the ability of stage BOS 0p to predict the development of BOS after single-lung transplantation, Lama and colleagues retrospectively analyzed spirometric data in 197 recipients (50). The predictive value of the FEV1 criterion for BOS stage 0p was superior to that of the FEF25–75 criterion, and it was higher in patients with restrictive than with obstructive underlying diseases; the probability to develop BOS stage 1 or greater or to die within 3 yr after reaching the FEV1 criterion was 81%, indicating that this criterion provides useful predictive information.
Treatment

To examine the effect of low-dose macrolides (250 mg azithromycin on alternate days) on lung function in patients with BOS, Yates and colleagues retrospectively evaluated 20 patients, of whom 2 were in BOS stage 0p, 6 in BOS 1, 2 in BOS 2, and 10 in BOS 3 at the start of study (51). After 3 mo, there was a mean increase in FEV1 of 110 ml (range,70–730 ml) or 14%. This improvement was sustained beyond 3 mo in 12 of the 17 patients who initially responded. An editorial commentary by Williams and Verleden accompanies this article (52). The same group of investigators reported their experience with total lymphoid irradiation in BOS. In this 12-yr study, Fisher and colleagues treated 37 recipients, of whom 27 (73%) completed the treatment (0.8 Gy twice weekly for 5 wk) (53). In these patients, the decline in FEV1 decreased from 123 ml/mo before treatment to 25.1 ml/mo after treatment.

OUTCOMES

To determine the optimal procedure for patients with pulmonary fibrosis, Meyer and colleagues compared the survival of patients in the UNOS national database who underwent single (n = 636) versus bilateral (n = 185) lung transplantation from 1994 to 2000 (54). For patients younger than 60 yr, performance of single-lung transplantation was associated with superior survival. This was attributable to excessive early mortality in the bilateral group, as no survival difference was observed between the two groups when the analysis was restricted to patients surviving to 3 mo. Among patients older than 60, the two procedures yielded equivalent outcomes, but this analysis was limited by a paucity of older patients undergoing bilateral-lung transplantation. The adverse effect of bilateral transplantation on early mortality among recipients with pulmonary fibrosis was corroborated in a second study (55).

The impact of secondary pulmonary hypertension on outcomes was the subject of two studies, with somewhat conflicting results. In a single-center retrospective cohort study, Fitch and colleagues found that severe secondary pulmonary hypertension (mean pressure \( \geq 40 \) mm Hg) was associated with lower \( P_{\text{aO}_2}/F_{\text{O}_2} \) ratios within the first 24 h after transplantation but this did not result in increased time on the ventilator, length of stay in the ICU, or overall mortality (56). Using the UNOS registry and focusing exclusively on patients with IFP, Whelan and colleagues reported that increased mean pulmonary artery pressure was an independent risk factor for early mortality (55).

Rutherford and colleagues reported on the health status of long-term survivors (57). They found that approximately one-third of the recipients at their center survived at least 10 yr. Only 18% of these long-term survivors were free of BOS; nearly half had BOS stage 1, 18% had BOS 2, and 18% had BOS 3. Median serum creatinine had doubled compared with pretransplant levels and 14% of patients required hemodialysis and/or kidney transplantation. Long-term survivors reported a significantly lower level of quality of life in multiple domains assessed by the standard Short-Form 36 questionnaire, including general health and physical function.

Bowdish and colleagues compared outcomes of 59 living-donor bilateral lobar transplant recipients with 43 recipients of conventional bilateral cadaveric grafts (58). Mortality at 3 mo was higher among the lobar recipients (25 vs. 6.5%). This finding likely reflects the increased acuity of illness of the lobar recipients, 73% of whom were hospitalized at the time of transplantation (vs. 2% in the cadaveric group). Long-term survival among patients living more than 3 mo was similar between the two recipient groups as was pulmonary function and maximum exercise capacity.

FUTURE DIRECTIONS IN LUNG TRANSPLANTATION

Wilkes and colleagues summarized the content of a workshop on lung transplantation sponsored by the National Heart, Lung, and Blood Institute (59). Key priorities for advancement of the field identified by workshop participants included expansion of the donor pool, accurate prediction and effective treatment of PGD and BOS, and development of strategies to facilitate induction of immune tolerance. Establishment of a multicenter collaborative network to promote clinical and basic science investigation was recommended.

HEMATOPOIETIC STEM CELL TRANSPLANTATION

In a retrospective analysis of 2,852 patients who received hematopoietic stem cell transplantation at a single center, Parimon and colleagues showed that compromised lung function before transplantation increases the risk of early post-transplant respiratory failure and death (60). The authors developed a lung function score based on pretransplant FEV1 and \( D_{\text{LCO}} \) that, by multivariate analyses, proved to be predictive of the risk of post-transplant respiratory complications and mortality.

Conflict of Interest Statement: Neither author has a financial relationship with a commercial entity that has an interest in the subject matter of this manuscript.

References

12. Meyers BF, de la Morena M, Sweet SC, Trulock EP, Guthrie TJ, Mendeloff EN, Huddleston C, Cooper JD, Patterson GA. Primary


