ELSEVIER

The Journal of
Heart and Lung
Transplantation

http://www.jhltonline.org

Bronchiolitis obliterans syndrome, hypogammaglobulinemia, and infectious complications of lung transplantation

Daniel C. Chambers, MBBS, MRCP, FRACP, MD,[a,b] Belinda Davies, BN,[b] Ann Mathews, BSc,[a] Stephanie T. Yerkovich, BSc(Hons) PhD,[a,b] and Peter M. Hopkins, MBBS, FRACP[a,b]

From the [a]School of Medicine, The University of Queensland; and the [b]Queensland Lung Transplant Service, The Prince Charles Hospital, Brisbane, Queensland, Australia.

KEY WORDS:
bronchiolitis obliterans syndrome;
hypogammaglobulinemia;
fungal infection;
community-acquired respiratory viral infection;
lung transplantation

BACKGROUND: Because infection has been associated with the development of bronchiolitis obliterans syndrome (BOS), we hypothesized that post-transplant hypogammaglobulinemia would be associated with infection and BOS.
METHODS: Cross-sectional levels of serum immunoglobulins were measured on 2 occasions in our transplant cohort and models developed to explain serum immunoglobulin levels and BOS-free survival.
RESULTS: A total of 139 patients (median age, 46.6 years) were evaluated at 47 months (range, 15–74 months) after transplant, and 87 were re-evaluated at 72 months (40–107 months). Of this cohort, 44% were immunoglobulin (Ig) G deficient and levels remained stable across the study period, and 27% were IgA deficient and levels fell slightly over time ($p = 0.003$). Both immunoglobulin classes were lower in patients with a history of invasive fungal infection, whereas IgA levels were lower in patients with a history of community-acquired respiratory viral infection. Low IgG was independently associated with shorter BOS-free survival (hazard ratio, 0.79; 95% confidence interval, 0.71–0.88; $p < 0.001$).
CONCLUSION: Serum immunoglobulin deficiency is common after lung transplantation and is associated with community-acquired respiratory viral infection, invasive fungal infection, and BOS.
J Heart Lung Transplant 2013;32:36–43

Lung transplantation is the only treatment option available for many patients with end-stage pulmonary disease, but long-term survival is compromised by the development of bronchiolitis obliterans syndrome (BOS). Although the strongest risk factor for the development of BOS remains a history of acute rejection,[1] infectious complications may also contribute. Cytomegaloviral and community-acquired respiratory viral infections (CARV)[2,3] as well as colonization with *Aspergillus* spp[4] and *Pseudomonas aeruginosa*[5] have all been implicated in BOS onset and progression.

Mucosal surfaces cover an area of more than 400 m^2 and harbor a vast population of potential pathogens[5] that are kept in check by the barrier function of these surfaces, secreted bactericidal peptides and proteins, and by the humoral arm of the immune system, in particular immunoglobulins (Ig) A and IgG. As the only commonly transplanted organ with a mucosal surface, the lung allograft may be particularly susceptible to the impaired mucosal defense associated with immunoglobulin deficiency.

Hypogammaglobulinemia is common after solid-organ transplantation and has been variously implicated as a causal factor in the infectious complications associated with these procedures. After lung transplantation, hypogammaglobulinemia (defined as IgG levels below age-specific ranges for pediatric patients[6] or < 6 g/liter[7] or 7 g/liter[8,9] for adult patients) is almost the norm, with a prevalence of between 48% and 73%.[6–9] Severe IgG deficiency (< 4 g/liter) is less common, with a prevalence between 8% and 15%, but has been associated with a higher risk of pneumonia, bacteriemia,

Reprint requests: Daniel C. Chambers, MBBS, MRCP, FRACP, MD, Queensland Lung Transplant Service, The Prince Charles Hospital, Brisbane, QLD 4032, Australia. Telephone: 61-7-3139-4000. Fax: 61-7-3139-5696.
E-mail address: daniel_chambers@health.qld.gov.au

1053-2498/$ - see front matter
http://dx.doi.org/10.1016/j.healun.2012.10.006

and invasive fungal infection.[8,9] Given the prevalence of hypogammaglobulinemia after solid-organ transplantation and the putative role of infection in BOS pathogenesis, we hypothesized that IgG and IgA deficiency would be common after lung transplantation and would be associated not only with infectious complications but also with BOS.

Methods

The study was approved by The Prince Charles Hospital Human Research and Ethics Committee.

Patients and study design

This study is a mixed retrospective review of pre-transplant immunoglobulin levels and a prospective cross-sectional assessment of post-transplant immunoglobulin levels. We assessed serum immunoglobulin levels in 139 lung transplant patients at our center in 2009 (97% of the entire cohort) and reassessed 87 patients (63%) 2.5 years later. All patients were censored in November 2011. In a sub-set of 75 patients, antibody levels were measured at the pre-transplant assessment. The study excluded patients receiving supplemental immunoglobulins.

Immunoglobulin measurements

Total IgA, IgG, and IgM were measured by nephelometry (Siemens Healthcare Diagnostics, Deerfield, IL), with normal reference range of >7 g/liter for IgG, >1 g/liter for IgA, and > 0.4 g/liter for IgM. Immunoglobulin deficiency was defined as a serum level below the lower limit of the predicted normal range.

Clinical information

Clinical parameters were collected including demographic data, immunosuppression, history of basiliximab or rituximab use, rejection burden (sum of A grades), history of proven (by polymerase chain reaction of nasopharyngeal swab/aspirate or bronchoalveolar lavage for respiratory syncytial virus, parainfluenza, influenza, human metapneumovirus, or adenovirus) CARV, cytomegalovirus infection, *P aeruginosa* colonization status (defined as isolation of *P aeruginosa* on at least 2 bronchoalveolar lavage samples taken at least 1 month apart), history of definite or probable invasive fungal infection[10] and BOS status.[11]

Immunosuppression protocols, consisting of a calcineurin inhibitor, mycophenolate mofetil, and prednisolone, have been uniform since the inception of the program in 1997. Induction therapy is not routinely used. Basiliximab is reserved for patients requiring a delay in calcineurin inhibitor exposure. Rituximab has been used rarely in patients with hyperacute or acute humoral rejection. Anti-microbial prophylaxis consists of targeted anti-bacterial drugs in the first 7 to 10 days after transplant, anti-fungal prophylaxis (voriconazole or itraconazole) for the first 3 months, and at times of augmented immunosuppression thereafter.

Statistical analysis

Results are presented as median and interquartile range (IQR). Statistical analysis was performed using STATA 11 software (StataCorp, College Station, TX) and $p < 0.05$ was considered statistically significant. Group differences were assessed by Kruskal-Wallis test, Mann-Whitney U test, Pearson chi square, or the Fisher exact test, as appropriate. Paired samples were compared by the Wilcoxon signed-rank test. Correlations between variables were determined using Spearman's ρ.

Simple and multivariate linear regressions were performed with IgG or IgA as the dependent variable. Simple linear regression analysis was initially used to evaluate the relationship between variables and immunoglobulin levels, and those variables with $p < 0.1$ were then subjected to multiple linear regression analysis. The time from transplant to BOS was modeled using Cox proportional hazard regression. Potential predictors included demographic factors, IgG, IgA, and IgM measured post-operatively, and previous infection, considered as a time-dependent covariate. The final model was obtained by forwards and backwards selection, retaining covariates where inclusion or exclusion changed the coefficients of other predictors by $> 10\%$ or where predictors were statistically significant at $\alpha = 0.05$.

Results

Full demographic data for the cohort are reported in Supplementary Table 1 (available on the JHLTonline.org Web site). Patients (48% women) were a median age of 46.6 years (IQR, 31.0–56.7 years) at transplantation. The diagnosis was cystic fibrosis in 34% and chronic obstructive pulmonary disease (COPD) in 37%. The transplant procedures were bilateral in 86%, single-lung in 7%, heart-lung in 5%, and heart-lung-liver in 2%. At the census, 51% of the cohort had a diagnosis of BOS grade ≥ 1.

Immunoglobulin levels fall after transplantation and remain low

At the first measurement at a median of 47 months after transplant, 44% of the patients were IgG-deficient (7.4 [IQR, 5.6–9.7] g/liter) and 27% were IgA-deficient (1.4 [IQR, 1.0–2.2] g/liter; Figure 1). IgG and IgA levels were only modestly correlated ($r = 0.349$, $p < 0.001$). Only 2 patients (identical twins with a history of cystic fibrosis and without associated IgG deficiency) were totally IgA-deficient (< 0.07 g/liter). IgM levels were better maintained after transplant, with a median IgM level of 0.9 g/liter (IQR, 0.6–1.5 g/liter) and with only 13% of the cohort deficient. Levels of IgG and IgA were below normal in 20 patients (14%), and all 3 immunoglobulin classes were deficient in 6 (4%).

Paired immunoglobulin levels before and after transplant were available for 47% (IgG), 52% (IgA), and 54% (IgM) of the cohort (Figure 2 and Supplementary Figure 1, available on the JHLTonline.org Web site). Age, sex, diagnosis, and transplant type were not different between those with and without pre-transplant immunoglobulin levels (data not shown). Pre-transplant, median (% deficient) IgG, IgA and IgM levels were 11.4 g/liter (9%), 2.5 g/liter (6%), and 1.1 g/liter (5%), respectively. The peri-transplant fall in IgG, IgA, and IgM levels was highly significant ($p < 0.001$ for each).