Bronchiolar airflow impairment after lung transplantation:
an early and common manifestation.

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Abstract:

**Background:** Bronchiolitis obliterans syndrome (BOS) is the major limitation to long term survival after lung transplantation (LTx). We studied extent and frequency of airflow limitation after LTx and its value for the diagnosis of BOS.

**Methods:** Flow-volume measurements were analysed retrospectively in 36 recipients of a bilateral LTx, with a median follow-up of 32.9 months. The prevalence and onset of a decline of FEV₁, FEF₂₅, FEF₅₀, FEF₇₅ and MMEF₇₅/₂₅ were evaluated and subsequently related to the occurrence of BOS grade 1.

**Results:** BOS grade 1 was diagnosed in 16 recipients at a median of 218 (range 88-1007) days after LTx. A persistent and significant decrease in FEV₁, FEF₂₅, FEF₅₀, FEF₇₅ and MMEF₇₅/₂₅ was observed in 23, 24, 30, 32 and 29 patients, respectively. In those patients developing BOS during follow-up this decrease was determined at 147 (55-657), 130 (78-932), 110 (21-573), 103 (32-657) and 121 (32-657) days after LTx (p<0.0005) respectively. The respective predictive values of these parameters for the occurrence of BOS grade 1 (within 120 days) were 88%, 60%, 50%, 35% and 41%.

**Conclusion:** Bronchiolar dysfunction is a common and early finding after LTx. The decrease of FEV₁ in BOS is significantly preceded by a decrease of bronchial airflow. Airflow markers may be used as an early warning sign for the development of BOS, although their predictive values are moderate.
Introduction:

Lung transplantation (LTx) is an accepted therapeutic option for patients with an end-stage lung disease\textsuperscript{1}. The main cause of late morbidity and mortality after LTx is bronchiolitis obliterans syndrome (BOS)\textsuperscript{2}. This syndrome is physiologically characterized by a progressive decline in lung function. Once started, BOS is usually an ongoing process leading to airflow limitation and later on to respiratory failure. The precise pathogenesis is unknown but experimental and clinical evidence suggests an immune mediated process\textsuperscript{3}. Histologically BOS is characterized by fibrous submucosal scarring of the terminal and respiratory bronchioles, resulting in complete or partial obstruction of the lumen (obliterative bronchiolitis (OB))\textsuperscript{4}. Decline of lung function is the most common sign of OB\textsuperscript{5}. The forced expiratory volume in one second (FEV\textsubscript{1}) is widely accepted as the indicator of pulmonary graft dysfunction\textsuperscript{6}. This parameter is a general measure of airway function comprising both small and large airways. More detailed evaluation of the lung function after LTx can be achieved by flow-volume measurements. Flow-volume curves in patients developing BOS after LTx usually show a concavity of the expiratory part, due to a decrease of the mid- and end-expiratory flow\textsuperscript{7}. Functional markers of the large, intermediate and small airways in flow-volume measurements are the forced expiratory flow after 25%, 50%, 75% of the forced vital capacity and the maximal mid-expiratory flow rate between 25% and 75%: FEF\textsubscript{25}, FEF\textsubscript{50}, FEF\textsubscript{75} and MMEF\textsubscript{75/25} respectively\textsuperscript{8,9}. Previous studies have suggested the usefulness of markers other than the FEV\textsubscript{1} as functional indicators of OB\textsuperscript{10,11}. An isolated decrease of the FEF\textsubscript{75} with normal FEV\textsubscript{1} is considered as a sensitive test for detecting small airway diseases like early abnormalities in asymptomatic smokers or in early emphysema. Also decline in small airway function as a precursor to BOS is widely acknowledged. However, most of this information arises from case studies\textsuperscript{7} and little quantitative information is available regarding earlier detection of BOS. In the present study the value of flow markers, representing the large, intermediate and small airways (FEF\textsubscript{25}, FEF\textsubscript{50}, FEF\textsubscript{75} and MMEF\textsubscript{75/25}) after lung transplantation were studied retrospectively in relation to FEV\textsubscript{1} and the development of BOS.

Materials and methods:

For the purpose of this study all flow-volume measurements of 36 patients were evaluated. From December 94 till May 98, 62 patients underwent a lung transplantation in the University Hospital Groningen. Twenty-six patients were excluded: 7 recipients of a single LTx, 4 patients with endoscopically significant airway pathology, 9 patients with a follow-up of less than 3 months after transplantation (e.g. due to early death), 5 patients with a still increasing FEV\textsubscript{1} after transplantation, and 1 patient with continuous infectious sequella. All patients received immunosuppression induction with usually 3 courses of rabbit-antithymocyte globulins (Thymoglobulin® (Merieux, Lyon, France), 3 mg/kg intravenously) and orally cyclosporine administered to reach whole blood trough levels of 400 µg tapered in 3 weeks after transplantation to 150 µg (measured by high pressure liquid chromatography), azathioprine (1-3 mg/kg) and prednisolone 15 mg daily. Herpes and pneumocystis carinii prophylaxis was given. The transplant function was monitored by flow-volume measurements: 2-3 times weekly during hospitalisation, in the outpatient setting weekly during the first 2-3 months after LTx, thereafter tapering to a minimum frequency of once every 3 months. These measurements were performed using a heated pneumotachograph (Masterscreen, E.Jaeger Würzburg,
Germany), according to standard guidelines. Subjects did not use bronchodilators within 12 hours before the measurements. Protocol bronchoscopies were performed once during the first month after LTx and every 6 months during the first 2 post-operative years and on clinical indication. Since lung transplantation patients were not expected to show the same variation in flow-volume values as in the normal population, the intra-individual variation of these parameters was determined in patients who did not develop BOS. The coefficient of variation ($V_{coef}$) of the FEV$_1$, FEF$_{25}$, FEF$_{50}$, FEF$_{75}$ and MMEF$_{75/25}$ was calculated from flow-volume values in a period of stable lung function without infection or acute rejection, using data from 20 patients based on 18 measurements (mean) during at least one year. Subsequently, a lower limit percentage for each of the parameters was calculated by the formula $(100 - 1.64*V_{coef})\%$, the factor 1.64 being the lower 5th percentile of the normal distribution. Therefore, in case a lung function value falls below this calculated lower limit, the probability that this decrease is by chance is less than 5%. A persistent decline in any parameter was defined as a decline below the lower limit for more than 3 successive measurements.

BOS was defined as a persisting decline of FEV$_1 < 80\%$ of baseline, with or without histologic changes (i.e. grade 1A or 1B according to ISHLT-criteria). The calculated baseline values for FEV$_1$, FEF$_{25}$, FEF$_{50}$, FEF$_{75}$ and MMEF$_{75/25}$ were similarly defined as the average of the highest two values three to six weeks apart after LTx.

FEF$_x$ values are dependent on lung volume. To ensure that the FEF$_x$ values were measured at the same lung volume, comparison of intra-individual FEF$_x$ values were only made when the forced vital capacity (FVC) did not differ more than 5%.

The lower limit of the usual variation of each pulmonary function parameter was determined by multiplying the above-mentioned lower limit percentage with the baseline values calculated for all individual patients. Onsets of a persistent decline of the respective lung function markers below the lower limit values were plotted for each patient, and compared with the onset of BOS grade I.

Differences in onset of a significant decline (i.e. below the lower limit) of the lung function values were assessed by the Kendall coefficient of concordance. In the case of a statistically significant difference, the Wilcoxon signed-rank test was used to compare 2 groups. P-values $< 0.05$ were considered significant.

In order to investigate their diagnostic power in relation to BOS grade I, positive predictive values of all evaluated flow markers were analysed.
Results:

The 36 patients studied were divided in patients with BOS (16) and patients without BOS (20) (table 1). The median follow-up of the total group was 32.9 months. The mean values of the $V_{coer}$ in the non-BOS group of FEV$_1$, FEF$_{25}$, FEF$_{50}$, FEF$_{75}$ and MMEF$_{75/25}$ were 4.0%, 7.4%, 9.8%, 14.9% and 10.7%, respectively. These values are comparable with the variation in the normal population. The lower limits of FEV$_1$, FEF$_{25}$, FEF$_{50}$, FEF$_{75}$ and MMEF$_{75/25}$ were 93.3%, 87.9%, 84.0%, 75.6% and 82.4%, respectively, of the baseline value. The median follow-up of the non-BOS group was 37.7 months.

Table 1: Patient characteristics

<table>
<thead>
<tr>
<th></th>
<th>BOS group</th>
<th>non-BOS group</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of patients</td>
<td>16</td>
<td>20</td>
</tr>
<tr>
<td>Gender</td>
<td>m 8 / f 8</td>
<td>m 11 / f 9</td>
</tr>
<tr>
<td>Age (median, range)</td>
<td>41 (12-64)</td>
<td>43 (22-60)</td>
</tr>
<tr>
<td>FEV1 baseline (median, SD)</td>
<td>2.84 (0.62)</td>
<td>3.54 (1.24)</td>
</tr>
<tr>
<td>Diagnosis</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Chronic obstructive pulmonary disease</td>
<td>1</td>
<td>4</td>
</tr>
<tr>
<td>Emphysema due to alfa1-antitrypsin-deficiency</td>
<td>5</td>
<td>11</td>
</tr>
<tr>
<td>Bronchiectasies</td>
<td>3</td>
<td>1</td>
</tr>
<tr>
<td>Cystic fibrosis</td>
<td>5</td>
<td>2</td>
</tr>
<tr>
<td>Idiopathic pulmonary fibrosis</td>
<td>2</td>
<td>1</td>
</tr>
<tr>
<td>Eisenmenger syndrome</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>Maximal BOS stage during follow-up</td>
<td>1a: 3</td>
<td>N.A.</td>
</tr>
<tr>
<td></td>
<td>1b: 0</td>
<td></td>
</tr>
<tr>
<td></td>
<td>2a: 1</td>
<td></td>
</tr>
<tr>
<td></td>
<td>2b: 3</td>
<td></td>
</tr>
<tr>
<td></td>
<td>3a: 2</td>
<td></td>
</tr>
<tr>
<td></td>
<td>3b: 7</td>
<td></td>
</tr>
</tbody>
</table>

A persistent and significant decrease in FEV$_1$, FEF$_{25}$, FEF$_{50}$, FEF$_{75}$ and MMEF$_{75/25}$ was observed in 23, 24, 30, 32 and 29 patients, respectively. FEV$_1$ in 7 patients was below their individual lower limit but did not reach the 20% fall in FEV$_1$ required for the diagnosis of BOS grade 1. The proportion of patients maintaining a stable pulmonary function for the FEF$_{75}$ (i.e. not descending under the lower limits) and occurrence of BOS in relation to time after LTx is given in figure 1. In the BOS group, BOS grade 1 was diagnosed 218 (88-1007) days after LTx (median, range). A persistent decrease of FEV$_1$, FEF$_{25}$, FEF$_{50}$, FEF$_{75}$ and MMEF$_{75/25}$ below their lower limits however was observed at 147 (55-657), 130 (78-932), 110 (21-573), 103 (32-657) and 121 days (32-657) after transplantation, respectively. A significant difference in days after LTx between the groups was observed (Kendall’s W-test, p < 0.0005). Subsequent comparison between the onset of BOS grade 1 and onsets of a significant decrease of each of the flow-volume markers again showed significant differences (p < 0.001, Wilcoxon).
Figure 1: The proportion of patients maintaining a stable pulmonary function (i.e. not descending under the lower limits of the respective markers) over time after LTx.

The FEF\(_{75}\) was the first parameter with a decline below the lower limit in 10 out of the 16 patients (example in figure 2).

Finally, the value of a significant decline of the respective flow-volume markers to predict the occurrence of BOS grade 1 within a subsequent period of 120 days, was assessed (table 2). A significant decline of the FEF\(_{75}\) was available in 29 patients, of whom 10 developed BOS grade 1 within the subsequent 120 days (positive predictive value (PV+): 34.5%). Four patients had no decline below the lower limit value. Three patients were not available due to insufficient follow up (<120 days) in this data set. Patients without a decline below the lower limit did not develop BOS grade 1A or 1B within a subsequent period of 120 days. Decline below the lower limit of FEV\(_1\) showed the highest positive predictive value (88.2%). Intermediate values of the PV+ were found for the other markers.
Table 2: Persistent declines below the lower limit and positive predictive values for development of BOS grade 1 within 120 days after this decline.

<table>
<thead>
<tr>
<th>Functional marker</th>
<th>BOS grade 1</th>
<th>FEV₁</th>
<th>FEF₂₅</th>
<th>FEF₅₀</th>
<th>FEF₇₅</th>
<th>MMEF. 75/25</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lower limit</td>
<td>80*</td>
<td>93.3</td>
<td>87.9</td>
<td>84.0</td>
<td>75.6</td>
<td>82.4</td>
</tr>
<tr>
<td>Onset**</td>
<td>218</td>
<td>147</td>
<td>130</td>
<td>110</td>
<td>103</td>
<td>121</td>
</tr>
<tr>
<td>Positive predictive value</td>
<td>100%</td>
<td>88.2%</td>
<td>60.0%</td>
<td>50.0%</td>
<td>34.5%</td>
<td>40.7%</td>
</tr>
<tr>
<td>(evaluable patients***</td>
<td></td>
<td>(17/23)</td>
<td>(20/24)</td>
<td>(22/30)</td>
<td>(29/32)</td>
<td>(27/29)</td>
</tr>
</tbody>
</table>

*) by definition  
**) onset of BOS grade 1 and a persistent decline of flow values below the lower limit (median values, days after LTx)  
***) patients with a persistent decline below the coefficient of variation / total number of patients with a sufficient follow-up of at least 120 days in the present data set
Discussion:

The present study indicates 1) that the majority of LTx recipients will, in due time, develop pulmonary dysfunction, as defined by all markers tested here, and 2) that the diagnosis of BOS and a persistent decrease of FEV\textsubscript{1} is preceded by persistent decreases of expiratory flow. The data described here suggest that earlier detection of BOS is possible, using the decline of flow measurements.

Chronic lung transplant dysfunction or bronchiolitis obliterans syndrome is the most important impediment to long term survival after lung transplantation. The condition is often progressive in spite of the usual approach of augmentation or conversion of immunosuppression. Current efforts are directed at delineation of the pathophysiology of this heterogeneous syndrome and at the development of early diagnostic and prognostic markers. Target areas of research are immunohistology, broncho-alveolar lavage studies and studies of functional markers.

Patterson et al. suggested that a decline in MMEF\textsubscript{75/25} to <70% of predicted could be used to define the clinical onset of OB\textsuperscript{10}. This parameter appeared to be a more sensitive index than 20% decline in FEV\textsubscript{1}. Reynaud-Gaubert et al.\textsuperscript{13} found similar differences in days after LTx in MMEF\textsubscript{75/25} compared to the decline in FEV\textsubscript{1} as in the present study, although not statistically significant. In addition these authors state that the slope of the single breath nitrogen wash-out curve is a better indicator. However, the number of days after LTx at the time a significant change took place, is in the same order of magnitude as the results of the present study using FEF\textsubscript{75}.

In our study, first the V\textsubscript{coef} for the different lung function parameters was calculated from a group of LTx recipients without BOS, in order to avoid improper extrapolation from the normal population. Higher values for the V\textsubscript{coef} were expected because patients after LTx are more susceptible for infection due to the combination of immunosuppression, denervation of the allograft, decreased mucociliary clearance and a disrupted lymphatic drainage. The observed variations, however, appeared to correspond with those found in healthy volunteers.\textsuperscript{14,15,16,17}

The baselines of the small airway markers were calculated in the same way as the FEV\textsubscript{1} baseline values according to the ISHLT-criteria for the grading of BOS. It has been observed that these values are sometimes abnormally high in the early post-operative period in patients after bilateral lung transplantation, though not described in literature. A possible source of error may occur since decrease from this high value may not be as important as a decrease in FEV\textsubscript{1}.

As it is clear from our clinical experience, more than 40% of the LTx recipients will develop BOS grade I or higher within 4 year after LTx. An arbitrary, but clinically useful follow-up period of 120 days was chosen to calculate positive predictive values for the occurrence of BOS grade 1 after a significant decline of the respective flow markers.

Using the lower limit of the FEV\textsubscript{1}, in the present study calculated as 93.3% of baseline, an early diagnosis of BOS grade I seems possible by 71 days. A persistent decline below this lower limit showed a positive predictive value for developing BOS grade 1 within 120 days of almost 90%. These data suggest that the generally accepted 20% decline in FEV\textsubscript{1} for the diagnosis of BOS is probably a conservative criterion. From a clinical perspective, a significant decline of FEF\textsubscript{75} may be used as an early warning sign for the development of BOS. However, the clinician should be aware that the probability of developing BOS within 120 days is approximately 35%. Obviously, a gain in early diagnostic potential is lost in a diminished positive predictive value. In practice, the composite finding of declining values of FEF\textsubscript{75} and FEV\textsubscript{1} may urge additional diagnostic procedures including transbronchial biopsies to obtain insight in the cause of transplant dysfunction. The role of flow-volume
measurements should be of more importance in the search for early signs of BO. It is an easy, cheap and non-invasive method in contrast with obtaining histology or very sophisticated lung function tests. Our results show that absence of decrease in pulmonary function makes invasive diagnostic procedures to detect BO(S) unnecessary.

A diagnostic approach, using bronchoscopy based on declining values of FEF\textsubscript{75} and FEV\textsubscript{1} below the coefficient of variation will be subject to further study on early intervention of BOS and on survival and cost-effectiveness.

In conclusion, ongoing decline of pulmonary flow 1) is a common finding after LTx, 2) significantly precedes a decrease of FEV\textsubscript{1} or the onset of BOS, and 3) may be used as an indication for performing invasive diagnostic procedures.

References: