Microbiological Status as a Factor of Airway Complications After Lung Transplantation

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ABSTRACT

Background. Lung transplantation (LTx) is the only treatment for patients with end-stage lung disease. This procedure is associated with a risk of complications related to airway stenosis, which can be treated by means of bronchoscopic interventions (BI). Microbiological colonization may have an impact on airway complications. The aim of the study was to investigate the effect of presence of microbiological pathogens in graft among lung recipients and frequency of BI, considered as the indicator of severe complications.

Materials and Methods. The study design was single-center retrospective cohort research; cases of 116 patients with complete microbiological data who underwent LTx from April 2013 to June 2019 were reviewed (70.3% of transplanted patients). All statistical analyses were performed with SPSS version 25.0 and R 3.5.3. For analyses involving the number of bronchoscopy interventions, univariate and multivariate Poisson regression were used. Interaction effect of variables in multivariate Poisson regression was assessed with partial response plot.

Results. The mean number of pathogens colonizing each patient was approximately 4.66 (range, 0 to 19) with *Candida albicans* (n = 42, 36.2%), *Aspergillus spp.* (n = 33, 28.4%), *Pseudomonas aeruginosa* (n = 32, 27.59%), and methicillin-sensitive *Staphylococcus aureus* (MSSA) (n = 29, 25%) being the most prominent. Microbiological agents causing the greatest increase in the risk of intervention are as follows: *Proteus mirabilis* by 3.84 times, *Aspergillus spp.* by 3.53 times, and *Stenotrophomonas maltophilia* by 3.09 times. *Burkholderia multivorans*, *Enterococcus spp.*, and *Klebsiella spp.* do not have a statistically significant impact on the number of BI.

Conclusions. Some pathogens increase the frequency of complications, which are associated with deterioration of the general condition. Therefore, patients should be monitored for the presence of pathogens in the airways.

INFECTIONS serve as a major source of morbidity and mortality after lung transplantation (LTx). Lung transplant recipients undergo an intensive immunosuppressive regimen to prevent acute and chronic rejection as well as to maintain proper graft function. The downside of such treatment is that it contributes to the increased risk of opportunistic pathogens as well as any infections in transplanted patients. In addition, the lungs, unlike other solid organ transplants, are continuously exposed to contact with the external environment enabling the direct contact with pathogens. According to the International Heart and Lung

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Transplantation Registry, infections are the second leading cause of mortality within the first 30 days after transplantation (17.2%), and between 30 days and 1 year they are the leading cause of death among patients (33.1%) [1]. Regarding fungal infections, even up to 8.6% of patients during the first year after the procedure become infected [2–4]. The most common pathogens that cause airway infections are as follows: *Candida albicans, Aspergillus spp., Pseudomonas aeruginosa, Staphylococcus aureus, Enterobacteriaceae, Klebsiella spp., Escherichia coli, Acinetobacter, Enterococcus faecalis,* cytomegalovirus (CMV), herpes simplex virus (HSV), and more [5–9]. Despite *P aeruginosa* and *S aureus* infections being among the most prominent, they turned out to be quite manageable after LTx, in contrast to fungal infections. *Aspergillus spp.* especially remains a major source of therapeutic concern. Risk factors for fungal infections include single lung transplantation procedure, pretransplant *Aspergillus* colonization, cystic fibrosis (CF) as an underlying disease, aggressive immunosuppression, and airway stenting [6,8,10–14]. One of the most significant complications of LTx are airway complications (AC). Their prevalence is believed to be 10% to 15%, with bronchial stenosis being the most common. Such complication often requires bronchoscopic interventions (BI) such as balloon bronchoplasty, argon plasma coagulation, laser therapy, cryotherapy treatment, or even stent placement. The aim of the study was to examine the effect of presence of pathogens on the airway complications after LTx in a single center retrospective study.

MATERIALS AND METHODS

Patients

The study design was single-center retrospective cohort research. We analyzed data from 116 patients with complete microbiological data (70.3% of all transplanted patients) who underwent LTx from April 2013 to June 2019 in the Silesian Center for Heart Diseases (Zabrze, Poland).

The study group consisted of 47 women (40.52%) and 69 men (49.58%). The median age at referral for LTx was 41 years old (range, 15-65 years old). Heart-lung transplantation and retransplantation recipients were excluded from the study. The number of double lung transplantation patients was 87 (75.0%). The most common diagnoses were as follows: CF (n = 37), chronic obstructive pulmonary disease (COPD) (n = 30); idiopathic pulmonary arterial hypertension (IPAH) (n = 16); interstitial lung diseases (ILD) (n = 27); and other (n = 6). ILD included idiopathic pulmonary fibrosis (IPF), sarcoidosis, histiocytosis, hypersensitivity pneumonitis, and lymphangioleiomyomatosis. Other diseases observed in patients were Osler-Weber-Rendu syndrome, Williams-Campbell syndrome, pulmonary veno-occlusive disease, bronchiectasis, and status post severe pulmonary embolism. Microbiological status was assessed from the material obtained by means of mini-bronchoalveolar lavage during bronchoscopy.

Airway Complications

Despite the vastness of the term "airway complication," this article describes interventions required in case of any airway stenosis (at the bronchial anastomosis or lower, eg, bronchus intermedius), granulation, or the presence of necrotic tissue. BI were as follows: balloon bronchoplasty, stent placement, argon plasma coagulation, laser therapy, or cryotherapy. None of the studied patients presented with partial or total anastomotic dehiscence; therefore, such complication is not described in this study.

Statistical Analysis

All statistical analyses were performed with SPSS version 25.0 (IBM, Armonk, NY, United States) and R 3.5.3 (The R Foundation for Statistical Computing, Vienna, Austria). *P* levels lower than .05 were deemed statistically significant. For analyses involving the number of BI, tests adequate for Poisson distribution were used, because our data consisted of counts in a given period of time. Specifically, univariate and multivariate Poisson regression were used as well as Poisson test. The interaction effect of variables in multivariate Poisson regression was assessed with partial response plot. In a series of univariate Poisson regression analyses Holm correction was applied to preserve overall alpha level equal to 0.05. For dichotomic discreet variables (occurrence or lack of intervention) multivariate logistic regression analysis was performed.

RESULTS

In our study group BI was required in 38.55% of recipients (n = 65).

The mean number of pathogens colonizing each patient was approximately 4.66 (range, 0 to 19) with *C albicans* (n = 42, 36.2%), *Aspergillus spp.* (n = 33, 28.4%), *P aeruginosa* (n = 32, 27.59%), and methicillin-sensitive *Staphylococcus aureus* (MSSA) (n = 29, 25%) being the most prominent. Regarding the average number of pathogens per patient in a given diagnosis, the data are distributed as follows: CF, 5.59; IPAH, 5.375; IPF, 5.18; and COPD, 4.55. Data of percentage distribution of pathogens in terms of underlying disease are as follows: CF, 37%; COPD, 23%; IPAH, 16%; ILD, 14%; and other, 10%.

According to univariate analysis presented in Fig 1, the presence of each pathogen causes a statistically significant increase in the number of interventions. Microbiological agents causing the greatest increase in the risk of intervention are *Proteus mirabilis* by 3.84 times, *Aspergillus spp.* by 3.53 times, and *Stenotrophomonas maltophilia* by 3.09 times. The significance of the impact of methicillin-resistant *Staphylococcus aureus* (MRSA) on the risk of performing an intervention is doubtful due to the small number of patients and the wide confidence intervals.

Nevertheless, considering the multivariate analysis presented in Fig 2, MRSA, *Burkholderia multivorans, Enterococcus spp.*, and *Klebsiella spp.* do not have a statistically significant impact on the number of BI. The chances of developing AC with each pathogen present are multiplied as follows: *Aspergillus spp.*, 3.35; *P mirabilis*, 2.87; MSSA, 2.33; *S pneumoniae*, 2.16; and *B cepacia*, 2.1. Additional presence of *P aeruginosa* or *Achromobacter xylosoxidans* is a factor causing a statistically significant decrease in the number of BI.

DISCUSSION

Many articles pertaining to the topic of the presence of pathogens in the bronchial tree after LTx point to the

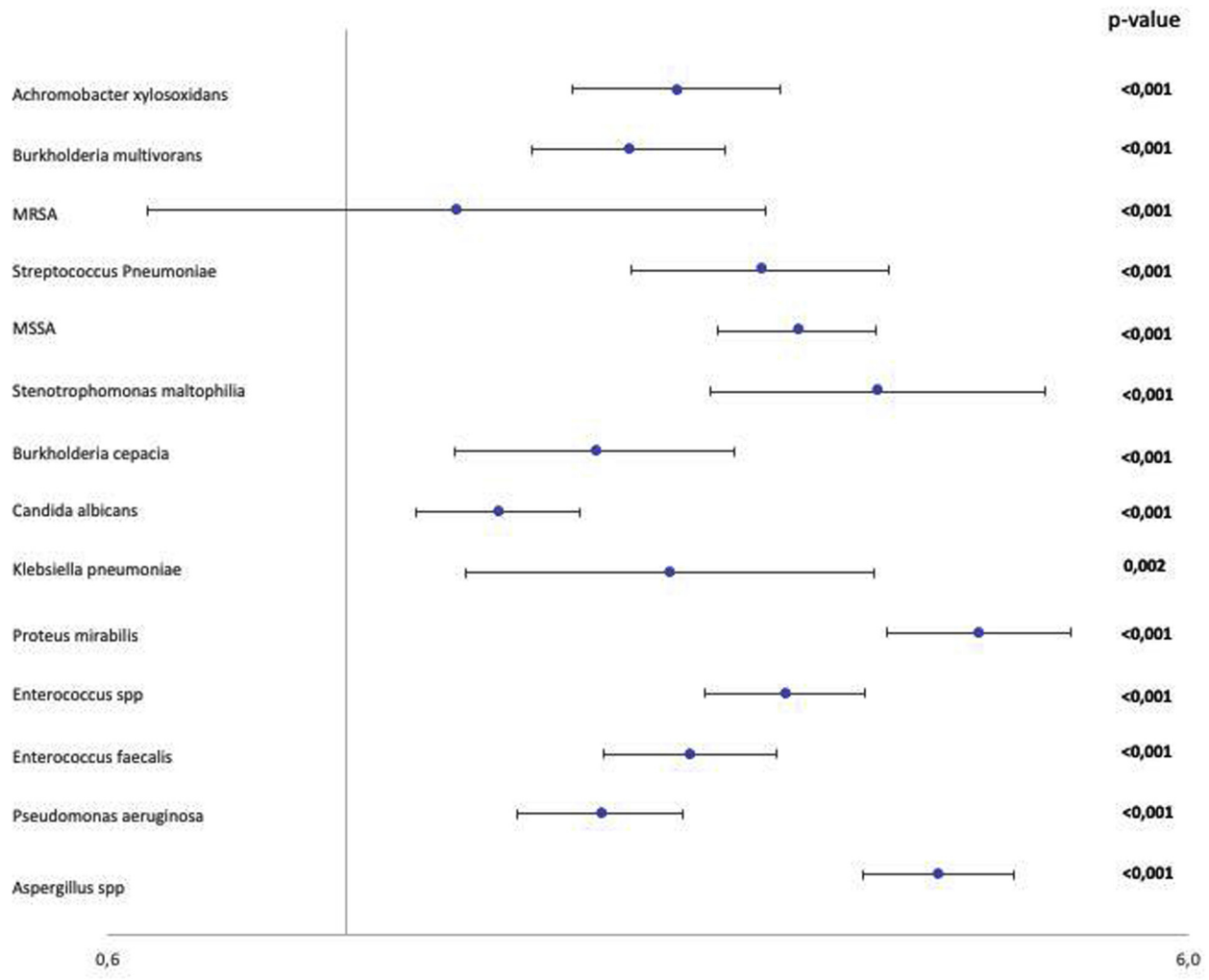

Fig 1. Univariate logistic regression analysis of the influence of microbiological presence on bronchoscopic interventions.

problem of distinguishing colonization from infection and lack of proper definition [15,16]. The presence of microbiological agents in the lungs of transplanted patients may be a source of AC, which still remains a major source of morbidity and mortality after LTx. Anastomotic infections appear to predispose to airway complications such as dehiscence, bronchomalacia, bronchial stenosis, and fistula formation [17,18]. This is the reason why bronchoalveolar lavage (BAL) cultures should be performed as well as proper antibiotic therapy applied to prevent aforementioned complications. BAL is considered to be a very thorough diagnostic tool for assessing and detecting inflammation of the lower respiratory tract [19–23]. In various publications considering the impact of pathogens on post-LTx outcomes, *Aspergillus* colonization is proven by numerous studies as being a risk factor for the development of AC [11,12,17]. According to the literature, the incidence of *Aspergillus spp.* ranges between 31% and 50% [24–26], which is similar to the frequency in our center (28.4%). In multivariate logistic regression analysis, *Aspergillus* has been shown to significantly increase the risk of AC, as has been previously described in several studies [17,27]. In addition, our result indicates a specific (3.35 times) increase in AC risk requiring BI in the presence of this fungus. Felton et al demonstrated that the presence of *Aspergillus* also affects mortality [11,28].

Regarding bacteria colonization, the most prominent cases vary in different studies, but usually include *P aeruginosa, S aureus, S maltophilia, K pneumoniae, S pneumoniae, Acinetobacter baumannii, Serratia marcescens,* and *Enterobacteriaceae* [5–9,16] According to the literature, *P aeruginosa, S aureus, Enterobacter spp., Klebsiella spp.*, and *B cepacia* are the most common bacteria causing complications after LTx [29,30]. In our center *C albicans, P aeruginosa, Aspergillus spp.*, MSSA, and *Enterococcus spp.* are the most prominent pathogens (listed in order of occurrence). Moreover, *Aspergillus* and MSSA are the first and third pathogen, respectively, according to multifactorial analysis,

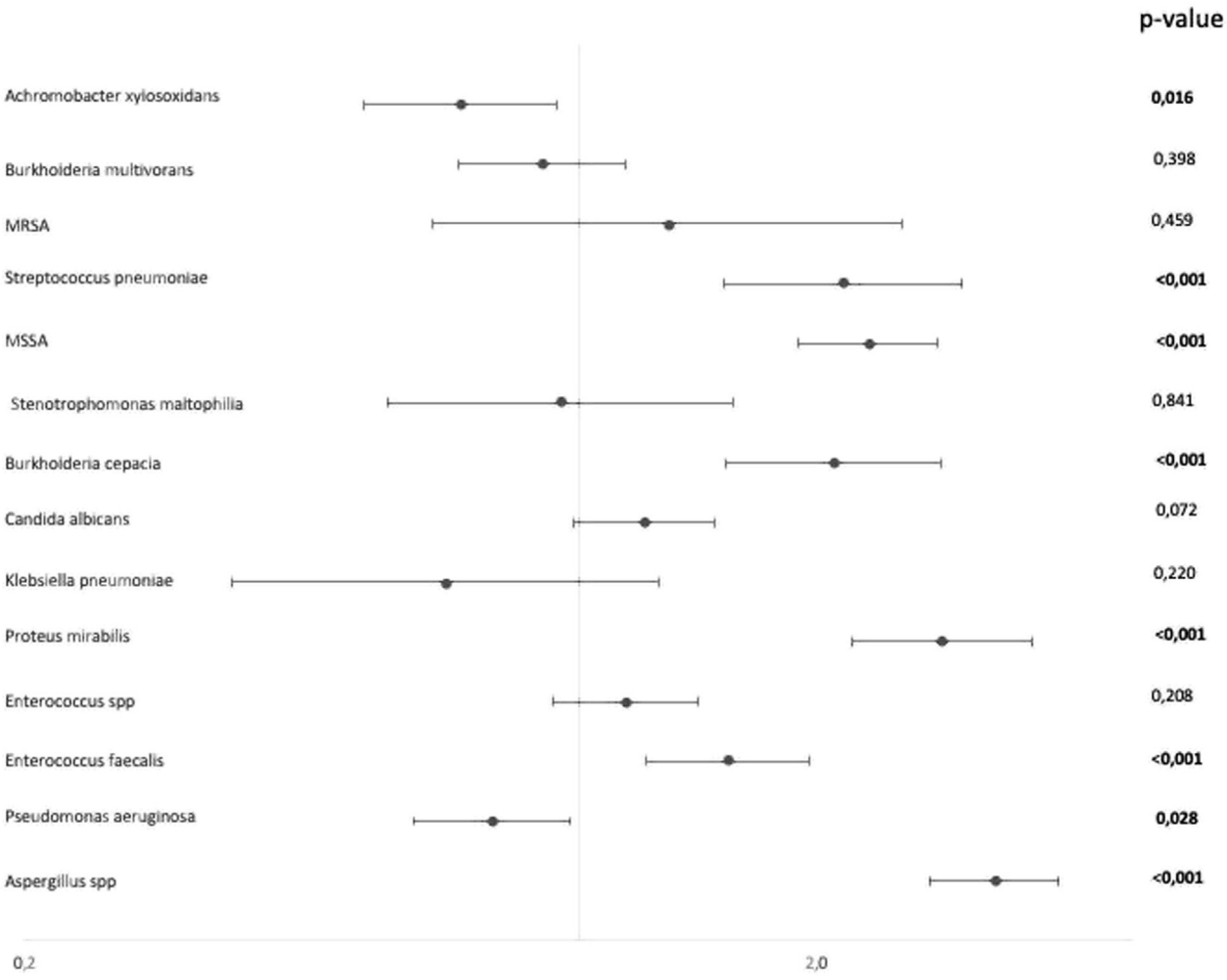

Fig 2. Multivariate logistic regression analysis of the influence of microbiological presence on bronchoscopic interventions.

causing the greatest risk of increasing the frequency of complications. For this reason performing BAL cultures is crucial to introduce treatment in a timely manner and to prevent the development of possible AC.

According to our results, the presence of *P mirabilis* is a serious problem in the risk of complications. This bacterium according to univariate analysis is the most important infectious factor in the occurrence of complications, and according to multivariate analysis it is the second most important factor after the presence of *Aspergillus*. To our knowledge, the risk posed by the presence of this microbe in the respiratory tract of patients after LTx has not yet been described.

Due to *B cepacia* colonization being a contraindication for qualifying patients for LTx, patients with positive BAL cultures were infected post-LTx. According to the literature, a vast majority of patients colonized with *B cepacia* are CF patients [31,32]. Nevertheless, in our study group 1 of 6 CF patients was colonized with this bacterium. Regarding *B multivorans* colonization, it is not considered to be a contraindication for LTx in our center. In addition, in contrast to the literature, fewer than 50% of patients were diagnosed with CF. Choong et al in a multivariate analysis found that *B cepacia* was a significant risk factor for the development of AC in pre-LTx-infected patients [30]. According to our knowledge, the impact of *B cepacia* on the risk of AC has been purely evaluated. Our results revealed significant influence of *B cepacia* on the occurrence of AC as one of the main agents. It should be emphasized that, unlike in the mentioned study, BAL cultures were positive only after LTx.

P aeruginosa remains 1 of the most prominent pathogens, especially in CF patients. Luong et al suggest the potentially negative impact of pretransplantation pseudomonal colonization on LTx outcomes [33]. Another study analyzing this matter was performed by Vos et al. Their work found that *P aeruginosa* colonization was an independent risk factor only in univariate analysis; however, the multivariate analysis

result remains insignificant [15]. Unlike the previously mentioned studies, in the study by Yserbyt et al, colonization by *P aeruginosa* did not have an impact on the frequency of AC [34]. We found that the presence of *Pseudomonas* not only is not a risk factor for developing AC, but it also resulted in a decrease of AC (OR, 0.777; 95% CI: 0.620-0.973; $P = 0.$). Suspicion was made that it may probably be associated with the fact that its presence prevents the extensive growth of other types of bacteria as they compete for resources. This finding is particularly important and interesting for us. Further studies examining this issue were in motion on the day we submitted this article. We are aware of limitations associated with the retrospective design of this study, and this issue requires further investigation in a larger group of patients in a prospective study.

CONCLUSIONS

Some pathogens increase the frequency of complications, which are associated with deterioration of the general condition of patients and more frequent hospitalizations.

Therefore, patients should be monitored for the presence of these pathogens in the airways. However, there are pathogens that have been shown to have a beneficial effect after statistical analysis, but we are aware of the controversy of this result; therefore, further research is required in a larger group of patients. This article also emphasizes the need and rationale for the thorough screening of fungal presence in the LTx recipients' bronchial trees.

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