Expert Opinion

Aerosol drug delivery in lung transplant recipients

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Background: Inhaled drug delivery after lung transplantation provides a unique opportunity for direct treatment of a solid organ transplant. At present, no inhaled therapies are approved for this population though several have received some development. Primary potential applications include inhaled immunosuppressive and anti-infective drugs. *Objectives*: The objective of this article is to review potential applications of inhaled medications for lung transplant recipients, the techniques used to develop inhaled drugs and the challenges of aerosol delivery in this specific population. *Methods*: The results of relevant studies are reviewed and two developmental examples are presented. *Results/conclusions*: Inhaled medications may provide significant advantages for lung transplant recipients. Past studies with inhaled cyclosporine and amphotericin-B provide useful guidance for clinical development of new preparations.

Keywords: aerosol drug delivery, inhaled amphotericin-B, inhaled cyclosporine, inhaled drug delivery, lung transplantation

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1. Introduction

Inhaled medications offer a potential opportunity to directly treat the lungs with therapeutic drug doses while sparing the patient the high systemic doses that may lead to unwanted side effects. In lung transplantation this concept may be particularly useful given the systemic side effect profiles associated with many of the immunosuppressive and anti-infective medications required by this population. However, the unique anatomy of lung transplant recipients and the pathophysiology of lung transplant rejection and post-transplant infection all require special consideration. Here we introduce lung transplantation and describe post-transplant complications and the unmet therapeutic needs of this population along with the potential advantages and disadvantages of inhaled therapies. We consider aerosol delivery in lung transplant recipients and detail the developmental path for therapies in this population through two examples: inhaled cyclosporine and inhaled amphotericin-B. We consider potential crossover therapies and look at future directions for therapeutic development.

2. Lung transplantation

2.1 Introduction

Lung transplantation provides a therapeutic alternative for end-stage lung diseases. The conditions most commonly treated through lung transplantation include chronic obstructive pulmonary disease (COPD), idiopathic pulmonary fibrosis, cystic fibrosis (CF), alpha-1 anti-trypsin deficient emphysema and primary pulmonary hypertension. In 2005, a total of 2,169 lung transplants were performed worldwide [1]. Depending on baseline condition, single and double lung as well as heart–lung transplants are performed. Double lung transplants

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have grown in proportion and now constitute the majority of procedures performed. Median survival at 5 years post-transplant is ~ 50% for all recipients [1].

2.2 Post-transplant complications

Graft failure and infectious complications are the most common causes of mortality during the first post-transplant year. Chronic rejection is the leading cause of mortality after post-transplant year 1 [1]. In the transplanted lung, chronic rejection manifests as obliterative bronchiolitis (OB), a fibroproliferative process that occludes the small airways, causing progressive and unrecoverable decreases in lung function. The loss of pulmonary function associated with chronic rejection is called bronchiolitis obliterans syndrome or BOS and is graded through a series of stages 0 – 3 [2]. This condition is, unfortunately, common post-transplant with 25% of lung recipients developing BOS within 2.5 post-transplant years, and 50% developing it within 5.6 years [1]. Chronic rejection can be diagnosed either through histology (on the basis of the results of *trans*-bronchial biopsies) or through spirometry (BOS).

Acute rejection of the allograft is a different process, specifically a vasculitis that occurs commonly after transplantation resulting in decreases in pulmonary function that are generally reversible with immunosuppressive therapy. Acute rejection events are very common in the early post-transplant course but mortality directly associated with acute rejection is not. It accounts for 4.7% of mortality within the first 30 post-transplant days and < 2% of post-transplant mortality after year 1 [1]. Repeated episodes of acute rejection are thought to lead to an increased incidence of chronic rejection [3]. Other factors described as potential risk factors for chronic rejection include lymphocytic bronchitis/bronchiolitis [3,4], cytomegalovirus pneumonitis [5], human leukocyte antigen mismatches [5], *Pseudomonas aeruginosa* colonization [6], gastro-esophageal reflux [7,8] and airway ischemia [9].

The immunosuppressive regimens associated with lung transplantation leave this population vulnerable to a host of opportunistic infections from bacterial, fungal and viral pathogens, with *P. aeruginosa*, Aspergillus species and cytomegalovirus being among the most common respectively [10]. Post–lung transplant anti-infective prophylaxis is typical [11-13], but infection is still the second leading cause of post-transplant mortality (after chronic rejection) from the end of the first transplant year forward [1].

2.3 Unmet needs and potential advantages/disadvantages of inhaled drugs

It is important to note that there are no medications specifically approved for post–lung transplant care at present, and that off-label use of drugs approved for other solid organ transplants is typical.

Induction therapy with different immunosuppressants is often used at the time of transplantation, followed by a strict regimen of maintenance immunosuppression through the entire post-transplant course [14-16]. Calcineurin inhibitors are a staple of post–lung transplant rejection prophylaxis with either cyclosporine or more commonly tacrolimus being used. An antimetabolite is also typically incorporated, with mycophenolate mofetil receiving the most use [1,17]. Oral steroids are also typically included as maintenance immunosuppression. Significant morbidity is associated with this immunosuppressive regimen. Nephrotoxicity is particularly common with the calcineurin inhibitors [18]. Other medication associated morbidities commonly reported in lung transplant recipients include hypertension, hyperlipidemia and diabetes mellitus [1]. Post-transplant care requires a constant need to balance the risks of rejection versus infection through titration of immunosuppressive medications.

Transplant recipients have a pressing unmet need for immunosuppressive therapies that offer maximum effect within the transplanted organ(s) with a minimum of systemic effects. In this regard, the development of inhaled therapies makes inherent sense for lung transplant recipients as they would allow for direct dosing of the transplanted organ, increasing the chance of efficient local treatment. Therapies for chronic lung transplant rejection are sadly lacking and represent a crucial unmet need. Pulsed IV steroids are often used, along with augmentation of maintenance immunosuppression. Azithromycin [19] and inhaled cyclosporine [20] have shown some promise in this role. Here an inhaled therapy might allow for high local dosing at the site of disease, especially if the drug could be targeted to the small airways.

The treatment of post–lung transplant infection involves the spectrum of antibiotic, antiviral and antifungal medications. Many of these medications also cause substantial systemic side effects. Consider, for example, the nephro- and ototoxicity associated with systemic aminoglycoside therapy [21]. Here again an inhaled route might offer locally effective treatment with minimal side effects. The development of antibiotic resistance would require careful study, however, especially given the potential for variable dosing in this population, which will be discussed in the next section. Inhaled antibiotics have been successfully developed to treat the infections associated with CF lung disease [22].

There are several potential disadvantages that must be considered with all inhaled medications. Local toxicity and the potential for airway irritation and bronchospasm must be considered with any new inhaled preparation in any population. It is important to note that a record of safe systemic delivery does not preclude this possibility when the medication is inhaled [23]. Inhalation toxicology studies provide the best means of assessing airway and lung toxicity in advance of clinical studies. Another potential disadvantage of inhaled medications is the unintentional generation of systemic doses. In many cases inhaled medications do provide lung dosing that is accompanied by

delayed and lower systemic doses; however, these kinetics can by no means be assumed. Absorption in the lung can vary substantially by medication [24] and by patient, and proper pharmacokinetic testing is necessary to ensure desired dosing. For example, systemic toxicity has been occasionally reported with inhaled tobramycin [25-27].

2.4 Aerosol delivery to lung transplant recipients

2.4.1 The physics of aerosol delivery

As previously described inhaled medications may provide some specific opportunities for lung transplant recipients. However, this population also provides some unique challenges in terms of aerosol delivery.

The dose of an inhaled medication that deposits in the lungs is affected by various patient factors (age, disease state, breathing pattern and flow rates, airway anatomy) and aerosol/device factors (aerosol size, output dose/rate) [28-33]. Aerosol deposition in the large airways, where flow velocities are high, is caused mostly by inertial impaction. Aerosol size is an important factor in this mechanism. Particles or droplets that are larger and heavier accumulate momentum causing them to move along straight-line paths rather than negotiating the complex flow pattern of the airways, ultimately causing them to deposit. The complex aerodynamics associated with obstructive lung diseases (sudden rapid changes in flow direction or constrictions/expansions of flow area) can increase the propensity for inertial impaction and cause regions of heavy local deposition in the airways, preventing aerosols from penetrating further into the lungs [34]. Aerosols that reach the slower flow regions of the peripheral lung deposit mostly owing to gravitational sedimentation, a process that occurs more quickly when aerosols are larger or heavier. For an aerosol to reach and deposit in the alveolar portion of the lung, it must be small enough to avoid the inertial filters of the upper airways and still be large enough to deposit quickly to avoid exhalation before deposition. In healthy lungs the ideal aerosol size for maximum delivery to the alveoli would be in the 2 – 3 μm range with some variation on the basis of breathing conditions [28]. As inhaled aerosols are dispersed exclusively by air flows, poorly ventilated regions of the lung may not be reached by aerosols, and many studies have demonstrated substantial variability in dosing with lung disease [35-39].

The most difficult lung zone to target with an aerosol is the small airways. Aerodynamic conditions in this zone vary substantially by airway generation. Flow areas increase and flow velocities slow down while moving peripherally, as airways become smaller in size and larger in number, lessening the impact of inertial deposition. There is no ideal aerosol size that independently targets this zone. Combinations of aerosol size and breathing technique to target the small airways have been described. For example, the inhalation of a 6 μm aerosol followed by a breath hold has been proposed [29].

2.4.2 The effects of post-transplant anatomy and pathophysiology

Specific aspects of post–lung transplant pathophysiology pose unique challenges to aerosol delivery. Elements of obstruction may be present in the large airways because of infection/inflammation, bronchial stenosis, bronchomalacia, the associated placement of airway stents [40] or surgical/anatomical factors unique to the transplant. Obstruction may be present in the small airways as a consequence of the OB associated with chronic rejection. These obstructions are particularly important to consider as they are a potential target for aerosol therapies. Obliterative bronchiolitis is a patchy process and may be present in the lung before the detection of the pulmonary function changes associated with BOS or changes in histology seen in samples obtained by means of transbronchial biopsy. Obliterative bronchiolitis is described as "a dense eosinophilic hyaline fibrosis in the sub-mucosa of membranous and respiratory bronchioles, resulting in partial or complete luminal occlusion" [41]. On the basis of Weibel's approximations, these bronchioles would be of the order of 0.5 mm in diameter and could number > 200,000 [42]. (A significant number of occlusions would be necessary to cause the decreases in pulmonary function associated with BOS.) Low air flow rates would be anticipated in these zones and the exact mechanisms causing aerosol deposition at the OB lesion are difficult to speculate on. Air trapping is commonly associated with OB, potentially causing initially decreased aerosol penetration followed by an increased residence period within the alveoli.

Aerosol delivery in single lung recipients poses a unique challenge. Quantitative studies of aerosol deposition in single lung transplant recipients typically demonstrate dominant deposition in the transplanted lung. This lung in most cases dominates ventilation and is subsequently exposed to more of the aerosol [43,44]. The interaction between the transplanted and native lungs is uniquely affected by the disease state of the native. Fibrotic native lungs have been shown to deposit smaller amounts of aerosol [44]. Hyperinflated native lungs may limit ventilation of the transplant. The lungs may have very different time constants in this setting, and aerosol flow from one lung to the other is possible. In general delivery to the native can be challenging, but is very necessary for some inhaled medications, particularly anti-infective drugs.

Overall pulmonary function will also vary in lung transplant recipients potentially affecting their ability to effectively inhale medications and endure long treatment periods.

2.5 Developmental examples

Two examples of the clinical development of inhaled medications for lung transplant recipients are illustrated here. A key element of this development is the early establishment of proper dosing. It is important to establish correct dosing with an inhaled medication, both in terms of

pulmonary and systemic dose during the early phases of clinical study, ahead of clinical trials to determine efficacy. The deposited dose of medication in the lungs can be measured using deposition scintigraphy techniques. Pharmacokinetic studies (which can and should if possible be performed simultaneously) provide an understanding of the corresponding systemic doses over time. The use of these techniques is further described later.

2.5.1 *Inhaled cyclosporine*

Inhaled cyclosporine was developed as an adjunct therapy to the usual post-transplant immunosuppressive regimen. It has been studied both as a rejection prophylaxis and as a treatment for refractory acute rejection and chronic rejection. It can be hypothesized that delivery of the drug directly to the small airways affected by OB might explain the efficacy demonstrated with the drug.

2.5.1.1 *Formulations*

Oral and i.v. formulations of cyclosporine are not suitable for inhalation. New formulations were developed specifically for inhaled delivery. Early development of these formulations was done in animal lung transplant models [45-47]. Formal inhalation toxicology studies were done during the later phases of development [48].

Most clinical developmental efforts have involved the use of a 62.5 mg/ml propylene glycol based formulation developed at the University of Pittsburgh. The formulation was delivered using jet nebulizers (AeroTech II, *CIS*-US, Bedford, MA, USA) driven with heavy-duty compressors (8650D, DeVilbiss, Somerset, PA, USA) and tested through a series of clinical studies [20,49-51]. In these studies the drug was delivered at (nebulizer loaded) doses of 100 – 300 mg. This aerosol, when characterized, was notably small by comparison with other inhaled medications: mass median aerodynamic diameter was 1.6 µm with 90% of the aerosol mass being in droplet sizes < 5.0 µm [49]. This formulation is now being used in Phase III clinical trials in lung transplant recipients (APT Pharmaceuticals, Burlingame, CA, USA; National Clinical Trial #NCT00755781).

An early liposomal form of the medication was developed and tested in healthy volunteers [52]. Delivery was also performed using the AeroTech II nebulizer. More recently a different liposomal form has been developed for delivery using the EFlow vibrating mesh nebulizer by Pari Pharma (Munich, Germany). Dry powder forms of cyclosporine for inhalation are also under development (NCT00378677) [53].

2.5.1.2 *Deposition scintigraphy*

Developmental work with aerosol cyclosporine included the performance of deposition scintigraphy studies in lung transplant recipients [43,54]. This testing involved the addition of a radiopharmaceutical to the inhaled medication. The radiopharmaceutical acts as a drug analogue allowing the medication dose in the lungs, mouth, throat/esophagus and stomach to be estimated after delivery. In deposition scintigraphy the radiopharmaceutical can either be directly bound to the drug product or just put into solution along with the active drug. The latter approach was chosen for testing with inhaled cyclosporine. Technetium 99m bound to the small molecule DTPA (Tc-DTPA) was added in small volumes to the (propylene glycol based) cyclosporine solution in the nebulizer and delivered along with the drug.

Necessary preclinical testing demonstrated a proportional relationship between drug mass and radioactivity. The typical method for establishing this relationship involves aerodynamically separating the aerosol into its constituent size classes through cascade impaction, and then comparing drug content to radioactivity within each size range [44]. The use of Tc-DTPA for deposition scintigraphy does require some amount of correction during analysis to account for the absorption [55].

On average, single lung recipients deposited from 2.2 to 9.2% of the loaded nebulizer dose in their transplanted lung whereas double lung recipients deposited 3.3 – 7.1% per transplanted lung. A relationship was noted between deposited peripheral dose in the transplanted lung and improvement in pulmonary function during the trial period after scintigraphy testing (up to 2 years of total dosing) [43]. This led to the determination of a therapeutic dose for the treatment: 5 mg of drug deposited in the peripheral portion of the transplanted lung. Six of seven single lung recipients demonstrated preferential deposition in the transplanted lung [49].

2.5.1.3 *Pharmacokinetics*

The pharmacokinetics of inhaled cyclosporine were studied in eight lung transplant recipients and compared to i.v. pharmacokinetics. Minimum detection levels were ≤ 10 ng/ml. The average maximum whole blood concentration of cyclosporine (C_{max}) seen after aerosol delivery was 206 mg/ml. Average AUC (0 – 24) was 1,034 ng/ml [56]. C_{max} after oral cyclosporine delivery has been reported to be 1,710 ng/ml with trough levels of 346 mg/ml. AUC (0 – 12) in those same studies was 7,447 ng/ml-h [57]. Notably the inhaled preparation demonstrated a half-life of 40.7 h after delivery versus 6.5 for i.v. and 3 – 4 for oral cyclosporine. These studies taken together demonstrate the advantages of inhaled delivery in minimizing systemic dosing. Non-simultaneous deposition scintigraphy studies were performed in five subjects participating in the PK studies. A liner relationship was demonstrated between deposited dose and 24 h absorption as calculated on the basis of pharmacokinetic results.

2.5.1.4 *Clinical studies*

Many of the early clinical studies with inhaled cyclosporine involved open label rescue use for either chronic rejection

or refractory episodes of acute rejection [51,58,59]. Typical outcomes for these studies included changes in histology based on *trans*-bronchial biopsy or changes in pulmonary function (FEV1). Changes in cytokine levels (IL-6, and INF-γ) in bronchoalveolar lavage were also demonstrated in association with inhaled cyclosporine [60]. Retrospective analysis of survival after a histologic diagnosis of chronic rejection demonstrated a protective effect associated with inhaled cyclosporine [20].

Placebo controlled studies were done to determine whether inhaled cyclosporine provided effective prophylaxis of acute rejection. This study included regular use of the drug (3 days/week) over the first 2 post-transplant years. A propylene glycol placebo was used. Acute rejection rates were not significantly improved with the drug, but survival and chronic rejection free survival were both improved with inhaled cyclosporine. Survival of the placebo group was well matched with other multicenter control groups [50].

Upper airway irritation has been reported during clinical studies with inhaled cyclosporine. The bulk of these studies have used the propylene glycol form of the medication, and some debate has existed as to whether the carrier, the active drug or both are the source of this irritation. In the previously described placebo controlled studies symptoms of upper airway irritation were reported in both drug and (propylene glycol) placebo groups. These were reported more often in the drug group though the differences were not statistically significant [50]. Inhalation toxicology studies with the drug mixture and vehicle controls imply that cyclosporine is the more active substance [48]. Symptoms of upper airway irritation have also been reported with the older liposomal forms of the medication, which contain no propylene glycol [52].

2.5.2 Inhaled amphotericin-B

Several amphotericin-B formulations have been developed as inhalation treatments for lung transplant recipients with the goal of providing prophylaxis from invasive fungal infections, especially those associated with Aspergillus species. Invasive fungal infections cause significant morbidity and mortality in these recipients. The incidence of invasive aspergillosis in one population of lung transplant recipients was reported to be 6.2%, with mortality rates of 52% [61]. Inhaled formations are attractive for this medication as they may provide substantial and sustained dosing in the lungs while minimizing the side effects otherwise associated with systemic (i.v.) administration.

2.5.2.1 Formulations

Three i.v. formulations of amphotericin-B are commonly available: amphotericin-B desoxycholate (ABD), lipid complex amphotericin-B (ABLC) and liposomal amphotericin-B (ABLS). The latter formulations were created to provide decreased side effect profiles versus ABD when delivered systemically [62,63]. All three forms have received some development for nebulized delivery including some fairly extensive studies in lung transplant recipients [44,64-68]. Differences in the tolerability have been noted [65,67]. Dry powder forms of amphotericin-B specifically for inhaled delivery have also been developed [69]. Nektar Therapeutics (San Carlos, CA, USA) now lists an amphotericin-B powder (NKTR-024) in its developmental pipeline.

2.5.2.2 Scintigraphy

Delivery of medication to all sites of potential infection is important with any inhaled anti-infective treatment, including inhaled preparations of amphotericin-B. For prophylaxis of Aspergillus infections, aerosol delivery to the airways and the peripheral lung is necessary as invasive pulmonary aspergillosis, tracheobronchitis and anastomotic infections all occur in significant proportion in lung transplant recipients (32, 37 and 20% of reported infections; remainder were disseminated aspergillosis) [61]. For single lung recipients, aerosol delivery to both the transplanted and native lungs is also needed as the native lung often serves as a nidus for fungal infections. Various reports have shown a higher incidence of invasive aspergillosis in single lung recipients with 40 – 90% of Aspergillus infections being noted first in the native lung [61,70].

Deposition studies with ABLC have been performed in lung transplant recipients [44]. As the ABLC formulation is a suspension, deposition scintigraphy testing required direct binding of Technetium 99m to the drug product. For these studies this was accomplished through a simple stannous reduction technique [44,71]. The dose of amphotericin-B loaded into the nebulizer was 35 mg (7 ml of ABLC). An AeroEclipse I nebulizer (Monaghan Medical, Plattsburgh, NY, USA) was used with an 8650D compressor (DeVilbiss, Somerset, PA, USA). Testing was performed with six single and six double lung recipients. The average total pulmonary dose measured in single lung recipients was 6.0 ± 0.6 mg. Double lung recipients averaged 6.8 ± 1.7 mg. Native lung doses were 50 ± 30% of transplanted lung doses on average in single lung recipients. Two tested single lung recipients with idiopathic pulmonary fibrosis had the highest transplanted lung doses and the lowest native lung doses. In these subjects, the peripheral native lung doses were 0.3 and 0.6 mg, respectively. These doses still seemed likely to provide the 90% MIC90 of amphotericin-B for *Aspergillus*: 2 μg/ml when measured *in vitro* [72], in lung regions that are accessible for aerosol delivery.

Deposition studies have also been performed with inhaled ABD. A System 22 Acorn nebulizer was used with a CR_{60} compressor (Alerbio, Madrid, Spain). These studies also reported preferential drug deposition in the transplanted lung of single lung recipients (72 versus 28% on average). They also reported preferential deposition in the lower lobes and a close correlation between drug deposition and perfusion [66].

2.5.2.3 Pharmacokinetics

Surveillance bronchoscopy with bronchoalveolar lavage (BAL) is frequently performed as a part of post–lung transplant care. The lavage procedure involves washing small volumes of saline into and out of the lung. Samples of these fluids are typically cultured to detect infection. They also provide an opportunity to quantify drug levels within the lung. There are techniques that allow for correction of the dilution associated with lavage so that a concentration of drug within pulmonary fluids can be estimated [73]. Amphotericin-B concentration was measured in a series of BAL samples collected from 4 – 48 h after the inhalation of 6 mg of ABD. Concentrations were 15.75, 13.66, 11.02 and 10.58 μg/ml at 4, 12, 24 and 48 h, respectively [66]. This gives an indication that inhaled ABD may provide therapeutic dosing that endures for an extended period of time after the treatment. This might allow for dosing at extended intervals (weekly versus daily).

Other studies have considered plasma levels of amphotericin-B after inhaled delivery to lung transplant recipients. Plasma samples were gathered from 20 lung transplant recipients 30 – 60 min after inhaled delivery of either 100 (ventilated patients) or 50 mg of ABLC. Treatments were delivered using an Updraft nebulizer (Hudson RCI, Temecula, CA, USA) driven by oxygen at 7 – 8 liters per minute (LPM). No samples demonstrated detectable levels of amphotericin-B (minimum detectable level 50 ng/ml) [67]. In another study amphotericin-B levels were measured in the plasma of 9 subjects (5 ABD, 4 ABLS), all of whom had received inhaled treatments for ≥ 1 week. Samples were collected 1 h after treatment. Minimum detection levels were < 0.2 μg/ml. In 5 subjects receiving ABD, plasma levels ranged from 0.2 to 0.9 μg/ml with 2 subjects having detectable levels. None of the 4 subjects receiving ABLS had detectable blood levels [68].

2.5.2.4 Clinical studies

The safety of inhaled ABLC was considered in a study that included 58 lung transplant recipients. The study included some subjects who received the treatments during mechanical ventilation (69 of 381 study treatments). These treatments were delivered using an Updraft nebulizer and oxygen at 7 – 8 LPM. Reported adverse effects were very limited with only two subjects reporting nausea and vomiting and no subjects reporting coughing or shortness of breath. Dynamic compliance was measured to determine safety in ventilated subjects. Only one treatment in one patient resulted in a ≥ 20% decrease in dynamic compliance [67]. A retrospective analysis was performed to determine the risk factors associated with Aspergillus infection that included 55 lung transplant recipients. Inhaled amphotericin-B (presumably ABD) was used by 44 of these subjects. Cytomegalovirus (CMV) disease was shown to be a significant risk factor. The use of inhaled amphotericin-B was shown to be protective (odds ratio: 0.13, 95% CI 0.02 – 0.69) [64]. The safety and efficacy of inhaled ABD and inhaled ABLC were compared in a study that included 100 lung transplant recipients. The medications were delivered using an Updraft nebulizer and oxygen at 7 – 8 LPM. Shortness of breath occurred in 19.9% of ABD subjects versus 2.1 of ABLC subjects. Overall, 42.9% of ABD subjects experienced some kind of adverse event versus 27.9 of ABLC subjects ($p = 0.02$ by generalized estimating equation (GEE) regression). This study also considered the incidence of invasive fungal infection in these subjects who inhaled the treatments once per day for 4 days and then once per week for 7 weeks. In the ABD group 7/49 subjects suffered invasive fungal infections versus 6/51 in the ABLC group. The meaning of these results is difficult to discern without the inclusion of a placebo group, however [65]. ABLS demonstrated similar and good tolerability when compared to ABD in another study [68].

2.6 Potential for cross-development

Inhaled antibiotic medications have been used to treat CF lung disease for some time. *Pseudomonas aeruginosa* is the primary bacterial pathogen affecting CF patients and the target of several inhaled antibiotic therapies. Inhaled tobramycin is approved for use in this population [74]. Inhaled colistin has received substantial off-label use [75] as has inhaled ceftazidime [76]. Inhaled amikacin has received substantial development [77] as has inhaled aztreonam [78]. Inhaled forms of ciprofloxacin, levofloxacin and a fosfomycin/tobramycin mixture are also listed on the Cystic Fibrosis Foundation's therapeutics pipeline [79]. Pseudomonas is the most common lung pathogen in lung transplant recipients comprising nearly 58% of all pathogens reported in a recent survey [10]. However, the inhaled antibiotics developed for CF have received relatively little formal study in lung transplant recipients. As with the other inhaled preparations described, inhaled antibiotics could potentially provide high local dosing with minimal side effects in lung transplant recipients. However, antibiotic resistance must be considered as a potential drawback. Sufficient delivery of the medication to all sites of infection would decrease the chance of such resistance, though as previously discussed this may be difficult in some lung transplant recipients. There is also a report of systemic toxicity in a lung transplant recipient using inhaled tobramycin [27].

Several studies have considered the use of inhaled steroids in lung transplant recipients without demonstrating substantial effect [80-82]. One study did demonstrate decreases in exhaled nitric oxide (a marker of inflammation) and increases in pulmonary function after inhaled budesonide was used to treat lymphocytic bronchiolitis [83]. Inhaled beta-agonists have been shown to improve mucociliary clearance after lung transplant [84]. Otherwise inhaled beta-agonists and anticholinergics have not received substantial study in lung transplant recipients.

3. Conclusions

Inhaled medications for lung transplant recipients provide a unique opportunity to directly treat a solid organ transplant. At present no medications, inhaled or otherwise, are specifically approved for post–lung transplant care. One area of potential opportunity is the development of inhaled immunosuppressants that could provide local treatment in the lungs while minimizing systemic dosing and associated side effects. Inhaled cyclosporine has demonstrated efficacy in both the prevention and treatment of chronic rejection [50], the leading cause of mortality after the first post-transplant year [1]. It is now being tested in Phase III trials (NCT00755781) and being developed in several different formulations.

Another potential opportunity lies in the development of inhaled anti-infective medications for this population. Inhaled forms of the antifungal medication amphotericin-B have been researched in lung transplant recipients, and have demonstrated substantial periods of residence within the lung with very minimal systemic concentrations [65-68]. Inhaled antibiotic medications being developed for use in CF may provide an opportunity for cross-development in lung transplant. The potential generation of bacterial resistance must be considered and proper dosing to reach all sites of infection must be pursued if these treatments are developed.

Lung transplant recipients offer unique challenges in terms of aerosol delivery. The unusual anatomy associated with lung transplant and the pathophysiology of post-transplant infection and rejection may make it difficult to deliver therapies consistently to the sites of disease. Inter-subject dosing variability should be anticipated. Substantial early clinical testing using deposition scintigraphy and pharmacokinetic techniques is suggested to establish proper dosing before studies evaluating efficacy. Other potential drawbacks to inhaled delivery include the possibility of upper airway irritation or bronchospasm and the development of unintended systemic doses. Inhaled medications have been successfully used in many lung diseases and their limited use in lung transplant recipients has so far been very successful.

4. Expert opinion

Lung transplant recipients are a growing population with substantial unmet therapeutic needs. The total number of lung transplant procedures performed in the US has increased substantially from 959 in 2000 to 1,468 in 2007 [85]. At present, this population depends on the off-label use of medications developed for other solid organ transplants, many of which have not been thoroughly studied in lung recipients.

Along with the further development of the medication classes previously described, lung transplant recipients would benefit substantially from the development of improved aerosol drug delivery technologies. Technologies to increase delivery to underventilated regions of the lungs could provide benefits to them and many other populations affected by lung disease. These technologies would allow for more consistent aerosol delivery to regions obstructed by infection or the fibroproliferative obstructions associated with OB. Increased delivery of aerosols beyond points of obstruction has been accomplished in asthma using low-density gases such as helium–oxygen mixtures, which flow with less resistance through the airways [86-88]. Aerosol size, shape and density could also be potentially manipulated to provide more efficient delivery of aerosols to low-ventilation regions. Also, extra dispersion mechanisms beyond those associated with inhaled air flows could be incorporated. For example, surfactant carriers have been shown to improve drug dispersion over the surface of airway models after aerosol deposition [89]. Technologies to improve the precision of dosing would be of use to this population as well. Much of the technology has been or is in the process of being developed for new inhaled medications that require more precise dosing (insulin and opiates, e.g.,). Flow-rate sensing is being incorporated into many newer aerosol delivery devices. Precise control of inhalation flow-rate and aerosol size could be used to control aerosol deposition through well-established mechanisms. For lung transplant recipients this precision delivery technology would allow for even more targeted delivery of inhaled therapies, providing efficacy while even further limiting systemic dosing. Finally technologies designed to improve the convenience of inhaled drug delivery would increase compliance and quality of life for lung transplant recipients. Some nebulizer treatments can require 30+ min of dosing or several doses per day. Improvements would allow for easier incorporation of inhaled therapies into current regimens.

The measurement of delivered dose is a vital part of the development of any inhaled medication. This is particularly important for lung transplant recipients because of the potential for dose variability caused by anatomic and disease factors. Deposition scintigraphy is the best-available method for measuring delivered dose. If possible, this should be performed simultaneously with pharmacokinetic testing so that pulmonary and systemic doses can be directly related. Establishing controlled and sufficient dosing is vital before the performance of clinical trials designed to demonstrate efficacy.

Increasing the number of clinical trials performed with lung transplant recipients would contribute to the infrastructure needed for further therapeutic development. Clinical trials in this population can be somewhat involved based on small numbers and the normal complications anticipated after lung transplantation. However, recent studies have fostered the development of expertise on clinical trials at many transplant centers and allowed for the formation of center networks. Exploring new study

endpoints specific to lung transplantation would also be of great benefit in accelerating therapeutic development. Recent studies of noninvasive methods for detecting chronic allograft rejection may be particularly useful [90-93].

Declaration of interest

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Bibliography

Papers of special note have been highlighted as either of interest (•) or of considerable interest (••) to readers.

1. Trulock EP, Christie JD, Edwards LB, et al. Registry of the International Society for Heart and Lung Transplantation: twenty-fourth official adult lung and heart-lung transplantation report-2007. J Heart Lung Transplant 2007;26:782-95

•• **Yearly summary of lung transplant data presented by the International Society for Heart and Lung Transplantation.**

2. Estenne M, Maurer JR, Boehler A, et al. Bronchiolitis obliterans syndrome 2001: an update of the diagnostic criteria. J Heart Lung Transplant 2002;21:297-310

• **Defines criteria for bronchiolitis obliterans syndrome (BOS).**

3. Sharples LD, Mcneil K, Stewart S, Wallwork J. Risk factors for bronchiolitis obliterans: a systematic review of recent publications. J Heart Lung Transplant 2002;21:271-81

4. Burton CM, Iversen M, Scheike T, et al. Is lymphocytic bronchiolitis a marker of acute rejection? An analysis of 2,697 transbronchial biopsies after lung transplantation. J Heart Lung Transplant 2008;27:1128-34

5. Kroshus TJ, Kshettry VR, Savik K, et al. Risk factors for the development of bronchiolitis obliterans syndrome after lung transplantation. J Thorac Cardiovasc Surg 1997;114:195-202

6. Botha P, Archer L, Anderson RL, et al. Pseudomonas aeruginosa colonization of the allograft after lung transplantation and the risk of bronchiolitis obliterans syndrome. Transplantation 2008;85:771-4

7. D'ovidio F, Mura M, Tsang M, et al. Bile acid aspiration and the development of bronchiolitis obliterans after lung transplantation. J Thorac Cardiovasc Surg 2005;129:1144-52

8. Stovold R, Forrest IA, Corris PA, et al. Pepsin, a biomarker of gastric aspiration in lung allografts: a putative association with rejection. Am J Respir Crit Care Med 2007;175:1298-303

9. Babu AN, Murakawa T, Thurman JM, et al. Microvascular destruction identifies murine allografts that cannot be rescued from airway fibrosis. J Clin Invest 2007;117:3774-85

10. Valentine VG, Bonvillain RW, Gupta MR, et al. Infections in lung allograft recipients: ganciclovir era. J Heart Lung Transplant 2008;27:528-35

11. Chmiel C, Speich R, Hofer M, et al. Ganciclovir/valganciclovir prophylaxis decreases cytomegalovirus-related events and bronchiolitis obliterans syndrome after lung transplantation. Clin Infect Dis 2008;46:831-9

12. Husain S, Paterson DL, Studer S, et al. Voriconazole prophylaxis in lung transplant recipients. Am J Transplant 2006;6:3008-16

13. Fishman JA. Prevention of infection caused by Pneumocystis carinii in transplant recipients. Clin Infect Dis 2001;33:1397-405

14. Ailawadi G, Smith PW, Oka T, et al. Effects of induction immunosuppression regimen on acute rejection, bronchiolitis obliterans, and survival after lung transplantation. J Thorac Cardiovasc Surg 2008;135:594-602

15. Hartwig MG, Snyder LD, Appel JZ, et al. Rabbit anti-thymocyte globulin induction therapy does not prolong survival after lung transplantation. J Heart Lung Transplant 2008;27:547-53

16. McCurry KR, Iacono A, Zeevi A, et al. Early outcomes in human lung transplantation with Thymoglobulin or Campath-1H for recipient pretreatment followed by posttransplant tacrolimus near-monotherapy. J Thorac Cardiovasc Surg 2005;130:528-37

17. Snell GI, Westall GP. Immunosuppression for lung transplantation: evidence to date. Drugs 2007;67:1531-9

18. Kahan BD. Cyclosporine. N Engl J Med 1989;321:1725-38

19. Gottlieb J, Szangolies J, Koehnlein T, et al. Long-term azithromycin for bronchiolitis obliterans syndrome after lung transplantation. Transplantation 2008;85:36-41

20. Iacono AT, Corcoran TE, Griffith BP, et al. Aerosol cyclosporin therapy in lung transplant recipients with bronchiolitis obliterans. Eur Respir J 2004;23:384-90

• **Retrospective study of inhaled cyclosporine for treatment of chronic lung transplant rejection.**

21. Label for Nebcin® (Tobramycin for Injection, USP)

22. Ramsey BW, Pepe MS, Quan JM, et al. Intermittent administration of inhaled tobramycin in patients with cystic fibrosis. Cystic Fibrosis Inhaled Tobramycin Study Group. N Engl J Med 1999;340:23-30

23. Alothman GA, Alsaadi MM, Ho BL, et al. Evaluation of bronchial constriction in children with cystic fibrosis after inhaling two different preparations of tobramycin. Chest 2002;122:930-4

24. Patton JS, Fishburn CS, Weers JG. The lungs as a portal of entry for systemic drug delivery. Proc Am Thorac Soc 2004;1:338-44

• **Describes factors determining absorption rates of drug from the lungs.**

25. Abdulhamid I, Wise TL, Andrews S, et al. Elevated serum tobramycin concentrations after treatment with tobramycin inhalation in a preterm infant. Pharmacotherapy 2008;28:939-44

26. Laporta R, Ussetti P, Carreno MC. Renal toxicity due to inhaled tobramycin in lung transplant recipients. J Heart Lung Transplant 2006;25:608

27. Ahya VN, Doyle AM, Mendez JD, et al. Renal and vestibular toxicity due to inhaled tobramycin in a lung transplant recipient. J Heart Lung Transplant 2005;24:932-5

28. Heyder J, Gebhardt J, Rudolf, G. et al. Deposition of Particles in the Human Respiratory Tract in the Size Range of 0.005-15 micron. J Aerosol Science 1986;17:811-25

•• **Classic report detailing the relationship between aerosol size and deposition location.**

29. Heyder J. Deposition of inhaled particles in the human respiratory tract and consequences for regional targeting

in respiratory drug delivery. Proc Am Thorac Soc 2004;1:315-20

• **Review of the basics of aerosol deposition in the lungs.**

30. Chua HL, Collis GG, Newbury AM, et al. The influence of age on aerosol deposition in children with cystic fibrosis. Eur Respir J 1994;7:2185-91

31. Usmani OS, Biddiscombe MF, Barnes PJ. Regional lung deposition and bronchodilator response as a function of beta2-agonist particle size. Am J Respir Crit Care Med 2005;172:1497-504

32. Laube BL, Jashnani R, Dalby RN, Zeitlin PL. Targeting aerosol deposition in patients with cystic fibrosis: effects of alterations in particle size and inspiratory flow rate. Chest 2000;118:1069-76

33. Svartengren M, Philipson K, Camner P. Individual differences in regional deposition of 6-micron particles in humans with induced bronchoconstriction. Exp Lung Res 1989;15:139-49

34. Brown JS, Zeman KL, Bennett WD. Regional deposition of coarse particles and ventilation distribution in healthy subjects and patients with cystic fibrosis. J Aerosol Med 2001;14:443-54

35. Ilowite JS, Gorvoy JD, Smaldone GC. Quantitative deposition of aerosolized gentamicin in cystic fibrosis. Am Rev Respir Dis 1987;136:1445-9

36. Smaldone GC, Fuhrer J, Steigbigel RT, McPeck M. Factors determining pulmonary deposition of aerosolized pentamidine in patients with human immunodeficiency virus infection. Am Rev Respir Dis 1991;143:727-37

37. Diot P, Palmer LB, Smaldone A, et al. RhDNase I aerosol deposition and related factors in cystic fibrosis. Am J Respir Crit Care Med 1997;156:1662-8

38. Mukhopadhyay S, Staddon GE, Eastman C, et al. The quantitative distribution of nebulized antibiotic in the lung in cystic fibrosis. Respir Med 1994;88:203-11

39. Condos R, Hull FP, Schluger NW, et al. Regional deposition of aerosolized interferon-gamma in pulmonary tuberculosis. Chest 2004;125:2146-55

40. Burns KE, Orons PD, Dauber JH, et al. Endobronchial metallic stent placement for airway complications after lung transplantation: longitudinal results. Ann Thorac Surg 2002;74:1934-41

41. Stewart S, Fishbein MC, Snell GI, et al. Revision of the 1996 working formulation for the standardization of nomenclature in the diagnosis of lung rejection. J Heart Lung Transplant 2007;26:1229-42

42. Weibel ER. Morphometry of the human lung. Berlin, Springer; 1963

43. Corcoran TE, Smaldone GC, Dauber JH, et al. Preservation of post-transplant lung function with aerosol cyclosporin. Eur Respir J 2004;23:378-83

• **Includes deposition studies of inhaled cyclosporine.**

44. Corcoran TE, Venkataramanan R, Mihelc KM, et al. Aerosol deposition of lipid complex amphotericin-B (Abelcet) in lung transplant recipients. Am J Transplant 2006;6:2765-73

• **Aerosol deposition studies with inhaled lipid-complex amphotericin-B.**

45. Mitruka SN, Won A, Mccurry KR, et al. In the lung aerosol cyclosporine provides a regional concentration advantage over intramuscular cyclosporine. J Heart Lung Transplant 2000;19:969-75

46. Mitruka SN, Pham SM, Zeevi A, et al. Aerosol cyclosporine prevents acute allograft rejection in experimental lung transplantation. J Thorac Cardiovasc Surg 1998;115:28-36; discussion 36-27

47. Zenati M, Duncan AJ, Burckart GJ, et al. Immunosuppression with aerosolized cyclosporine for prevention of lung rejection in a rat model. Eur J Cardiothorac Surg 1991;5:266-71; discussion 272

48. Wang T, Noonberg S, Steigerwalt R, et al. Preclinical safety evaluation of inhaled cyclosporine in propylene glycol. J Aerosol Med 2007;20:417-28

49. Corcoran TE. Inhaled delivery of aerosolized cyclosporine. Adv Drug Deliv Rev 2006

50. Iacono AT, Johnson BA, Grgurich WF, et al. A randomized trial of inhaled cyclosporine in lung-transplant recipients. N Engl J Med 2006;354:141-50

• **Placebo-controlled study of inhaled cyclosporine for prophylaxis of lung transplant rejection.**

51. Iacono AT, Keenan RJ, Duncan SR, et al. Aerosolized cyclosporine in lung recipients with refractory chronic rejection. Am J Respir Crit Care Med 1996;153:1451-5

52. Gilbert BE, Knight C, Alvarez FG, et al. Tolerance of volunteers to cyclosporine A-dilauroylphosphatidylcholine liposome aerosol. Am J Respir Crit Care Med 1997;156:1789-93

53. Zijlstra GS, Rijkeboer M, Jan van Drooge D, et al. Characterization of a cyclosporine solid dispersion for inhalation. Aaps J 2007;9:E190-199

54. O'riordan TG, Iacono A, Keenan RJ, et al. Delivery and distribution of aerosolized cyclosporine in lung allograft recipients. Am J Respir Crit Care Med 1995;151:516-21

55. Coates AL, Green M, Leung K, et al. The challenges of quantitative measurement of lung deposition using 99mTc-DTPA from delivery systems with very different delivery times. J Aerosol Med 2007;20:320-30

•• **Describes the limitations of aerosol deposition testing with Technetium 99m DTPA and suggests correction factors.**

56. Burckart G, Smaldone GC, Eldon M, et al. Lung Deposition and Pharmacokinetics of Cyclosporine after Aerosolization in Lung Transplant Patients. Pharm Res 2003;20:252-6

• **Includes pharmacokinetic studies of inhaled cyclosporine.**

57. Knoop C, Vervier I, Thiry P, et al. Cyclosporine pharmacokinetics and dose monitoring after lung transplantation: comparison between cystic fibrosis and other conditions. Transplantation 2003;76:683-8

58. Iacono AT, Smaldone GC, Keenan RJ, et al. Dose-related reversal of acute lung rejection by aerosolized cyclosporine. Am J Respir Crit Care Med 1997;155:1690-8

59. Keenan RJ, Iacono A, Dauber JH, et al. Treatment of refractory acute allograft rejection with aerosolized cyclosporine in lung transplant recipients. J Thorac Cardiovasc Surg 1997;113:335-40; discussion 340-331

60. Iacono A, Dauber J, Keenan R, et al. Interleukin 6 and interferon-gamma gene expression in lung transplant recipients with refractory acute cellular rejection: implications for monitoring and inhibition by treatment with aerosolized cyclosporine. Transplantation 1997;64:263-9

61. Singh N, Husain S. Aspergillus infections after lung transplantation: clinical differences in type of transplant and implications for management. J Heart Lung Transplant 2003;22:258-66

62. Martino R. Efficacy, safety and cost-effectiveness of Amphotericin B Lipid Complex (ABLC): a review of

the literature. Curr Med Res Opin 2004;20:485-504

63. Olson JA, Adler-moore JP, Jensen GM, et al. Comparison of the physicochemical, antifungal, and toxic properties of two liposomal amphotericin B products. Antimicrob Agents Chemother 2008;52:259-68
64. Monforte V, Roman A, Gavalda J, et al. Nebulized amphotericin B prophylaxis for Aspergillus infection in lung transplantation: study of risk factors. J Heart Lung Transplant 2001;20:1274-81
65. Drew RH, Dodds Ashley E, Benjamin DK Jr, et al. Comparative safety of amphotericin B lipid complex and amphotericin B deoxycholate as aerosolized antifungal prophylaxis in lung-transplant recipients. Transplantation 2004;77:232-7

• **Safety results regarding inhaled amphotericin-B preparations.**

66. Monforte V, Roman A, Gavalda J, et al. Nebulized amphotericin B concentration and distribution in the respiratory tract of lung-transplanted patients. Transplantation 2003;75:1571-4
67. Palmer SM, Drew RH, Whitehouse JD, et al. Safety of aerosolized amphotericin B lipid complex in lung transplant recipients. Transplantation 2001;72:545-8
68. Lowry CM, Marty FM, Vargas SO, et al. Safety of aerosolized liposomal versus deoxycholate amphotericin B formulations for prevention of invasive fungal infections following lung transplantation: a retrospective study. Transpl Infect Dis 2007;9:121-5
69. Shah SP, Misra A. Development of liposomal amphotericin B dry powder inhaler formulation. Drug Deliv 2004;11:247-53
70. Westney GE, Kesten S, De Hoyos A, et al. Aspergillus infection in single and double lung transplant recipients. Transplantation 1996;61:915-9
71. Saha G. Fundamentals of Nuclear Pharmacy, Fifth Addition: Springer; 2004
72. Diekema DJ, Messer SA, Hollis RJ, et al. Activities of caspofungin, itraconazole, posaconazole, ravuconazole, voriconazole, and amphotericin B against 448 recent clinical isolates of filamentous fungi. J Clin Microbiol 2003;41:3623-6
73. Bayat S, Louchahi K, Verdiere B, et al. Comparison of 99mTc-DTPA and urea for measuring cefepime concentrations in epithelial lining fluid. Eur Respir J 2004;24:150-6
74. Ramsey BW, Dorkin HL, Eisenberg JD, et al. Efficacy of aerosolized tobramycin in patients with cystic fibrosis. N Engl J Med 1993;328:1740-6

• **Clinical study demonstrating the efficacy of inhaled tobramycin in cystic fibrosis patients.**

75. Diot P, Gagnadoux F, Martin C, et al. Nebulization and anti-Pseudomonas aeruginosa activity of colistin. Eur Respir J 1997;10:1995-8
76. Stead RJ, Hodson ME, Batten JC. Inhaled ceftazidime compared with gentamicin and carbenicillin in older patients with cystic fibrosis infected with Pseudomonas aeruginosa. Br J Dis Chest 1987;81:272-9
77. Nicholson. Development and Studies of Nebulized SLIT Amikacin for the Treatment of Pseudomonas Aeruginosa Airway Infection in CF (S12.3 Session Summary). Pediatr Pulmonol 2005;40:144
78. McCoy KS, Quittner AL, Oermann CM, et al. Inhaled Aztreonam Lysine for Chronic Airway Pseudomonas aeruginosa in Cystic Fibrosis. Am J Respir Crit Care Med 2008;178:921-8
79. Available from: http://www.cff.org/treatments/Pipeline/
80. Ward C, De Soyza A, Fisher AJ, et al. Reticular basement membrane thickening in airways of lung transplant recipients is not affected by inhaled corticosteroids. Clin Exp Allergy 2004;34:1905-9
81. Whitford H, Walters EH, Levvey B, et al. Addition of inhaled corticosteroids to systemic immunosuppression after lung transplantation: a double-blind, placebo-controlled trial. Transplantation 2002;73:1793-9
82. Zheng L, Walters EH, Wang N, et al. Effect of inhaled fluticasone propionate on BAL TGF-beta(1) and bFGF concentrations in clinically stable lung transplant recipients. J Heart Lung Transplant 2004;23:446-55
83. De Soyza A, Fisher AJ, Small T, Corris PA. Inhaled corticosteroids and the treatment of lymphocytic bronchiolitis following lung transplantation. Am J Respir Crit Care Med 2001;164:1209-12
84. Laube BL, Karmazyn YJ, Orens JB, Mogayzel PJ. Albuterol improves impaired mucociliary clearance after lung transplantation. J Heart Lung Transplant 2007;26:138-44
85. Available from: www.optn.org
86. Kim IK, Phrampus E, Venkataraman S, et al. Helium/oxygen-driven albuterol nebulization in the treatment of children with moderate to severe asthma exacerbations: a randomized, controlled trial. Pediatrics 2005;116:1127-33
87. Papamoschou D. Theoretical validation of the respiratory benefits of helium-oxygen mixtures. Respir Physiol 1995;99:183-90
88. Piva JP, Menna Barreto, SS Zelmanovitz, et al. Heliox versus oxygen for nebulized aerosol therapy in children with lower airway obstruction. Pediatr Crit Care Med 2002;3:6-10
89. Marcinkowski AL, Garoff S, Tilton RD, et al. Post-deposition dispersion of aerosol medications using surfactant carriers. J Aerosol Med and Pulm Drug Del 2008;21
90. Studer SM, George MP, Zhu X, et al. CD28 Downregulation on CD4 T-Cells is a Marker for Graft Dysfunction in Lung Transplant Recipients. Am J Respir Crit Care Med 2008
91. Golocheikine AS, Saini D, Ramachandran S, et al. Soluble CD30 levels as a diagnostic marker for bronchiolitis obliterans syndrome following human lung transplantation. Transpl Immunol 2008;18:260-3
92. Gast KK, Biedermann A, Herweling A, et al. Oxygen-sensitive 3He-MRI in bronchiolitis obliterans after lung transplantation. Eur Radiol 2008;18:530-7
93. Van Muylem A, Knoop C, Estenne M. Early detection of chronic pulmonary allograft dysfunction by exhaled biomarkers. Am J Respir Crit Care Med 2007;175:731-6

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