DRUG EVALUATION

Future MICROBIOLOGY

Management of *Pseudomonas aeruginosa* infection in cystic fibrosis patients using inhaled antibiotics with a focus on nebulized liposomal amikacin

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Pseudomonas aeruginosa (*PsA*) is a highly prevalent bacterial organism recovered from the lungs of cystic fibrosis (CF) patients and chronic *PsA* infection is linked to progressive pulmonary function decline. The eradication and treatment of this organism from CF airways is particularly challenging to CF care providers. Aerosolized antibiotics that target *PsA* help to slow down growth, maintain lung function and reduce the frequency of pulmonary exacerbations. In this review, we discuss the currently available inhaled antibiotics for management of *PsA* lung infections in CF patients, with a focus on liposomal amikacin for inhalation (LAI). LAI is a unique formulation of amikacin under development that enhances drug delivery and retention in CF airways via drug incorporation into neutral liposomes. Factors such as once-daily dosing, mucus and biofilm penetration and potentially prolonged off-drug periods make LAI a potentially attractive option to manage chronic *PsA* lung infections in CF patients.

KEYWORDS
• amikacin • arikace
• arikayce • cystic fibrosis
• LAI • *Pseudomonas aeruginosa*

Cystic fibrosis (CF) is a life-limiting hereditary disorder occurring in virtually all ethnic groups with a prevalence of approximately 30,000 patients in the USA and over 70,000 worldwide. CF is caused by mutations in a chromosome 7 gene that encodes the CF transmembrane conductance regulator (CFTR) protein [1–4]. CFTR is a chloride and bicarbonate channel that regulates epithelial ion transport and hydration [5,6]. Mutations in CFTR affect the balance of epithelial ion and water transport, causing dehydrated and viscous mucus secretions in most CF-affected organs [7]. CF is a multisystem disease with prominent symptoms in the sinopulmonary tree, GI tract, pancreas, hepatobiliary system and male reproductive tract. Pulmonary infection and subsequent pulmonary decline is the leading cause of CF morbidity and mortality. A deficiency in mucociliary clearance makes the airways susceptible to bacterial and other infections that typically become chronic and lifelong [8]. Therefore, focusing efforts on treatment of lung infections in CF patients is a central component of CF care.

The burden of *Pseudomonas aeruginosa* pulmonary infection in CF patients

The microbial milieu of CF lungs changes with age. *Haemophilus influenzae* and *Staphylococcus aureus* (including both methicillin-sensitive and more recently methicillin-resistant strains) are more frequent pathogens in younger pediatric patients. With advancing age or disease, additional pathogens such as *Stenotrophomonas maltophila*, *Achromobacter xyloxidans*, *Burkholderia cepacia* complex, nontuberculous mycobacteria and other pathogens with inherent resistance to many antibiotics often predominate [9]. CF registry data confirm that *Pseudomonas aeruginosa* (*PsA*) typically becomes the predominant organism cultured from the respiratory tract by adulthood, with the prevalence rising

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from 20% in patients less than 5 years of age up to as high as 70% by 18 years of age [9]. It also is well established that chronic lung infection with *PsA* causes more-rapid pulmonary decline and contributes to early death in CF patients [10]. Thus, treatment of *PsA* infection has become an integral part of CF care. In addition to the 'traditional' CF pulmonary pathogens, recent research provides evidence for a highly complex microbiome in the CF airway. A better understanding of these organisms and the host:pathogen relationship in CF is believed to be critical to improved management and appropriate treatment of CF airway infections [11].

PsA isolates frequently possess features that contribute to their adaptation and persistence in the CF lung. They often possess a hypermutable genetic background that leads to the emergence of variants capable of long-term persistence, such as a mucoid phenotype, modified antigenic structures and antimicrobial resistance [12,13]. The mucoid phenotype is caused by extracellular alginate production, which can reduce antimicrobial effectiveness by limiting drug penetration. Evidence also suggests that *PsA* can grow as a biofilm in the lower respiratory tract of CF patients, rendering the organism resistant to many antimicrobials [14,15]. In-depth reviews of this topic have been published previously [16,17].

Although the definition of chronic *PsA* infection is still under debate [18,19], chronic *PsA* colonization is associated with a more-rapid decline in lung function [20]. The complex CF lower-airway environment (alginate production, neutrophilic inflammation and thick mucus) makes it particularly challenging to clear mucoid *PsA* from CF airways and thus management of chronic infection is a mainstay of current CF care.

• The goals of treatment of PsA infection in CF patients

The goals of chronic PsA treatment can be broadly categorized into two aims: the treatment of acute pulmonary exacerbations and the management of chronic *PsA* infection during periods of relative clinical stability. Acute pulmonary exacerbations in CF typically include an increase in pulmonary and systemic symptoms (e.g., increased cough, chest congestion and sputum production accompanied by malaise and weight loss) coupled with physical findings (e.g., crackles), as well as a reduction in lung function as measured by the forced expiratory volume in 1 s (FEV_1) [21–25]. Treatment includes an increase in airway clearance and the addition of antibiotics that target known lower airway pathogens. In patients with chronic *PsA* infection, this usually includes the addition of anti-*PsA* antibiotics via oral, inhaled and/or intravenous routes. Although this approach is beneficial, a significant percentage of patients (~25%) fail to fully recover the lost lung function [26,27]. As this decline in lung function can be accumulative over the lifespan of CF patients, it is imperative to develop strategies to restore lung function in all affected patients.

Management of established *PsA* infections during clinical stability is a common point of CF care, and typically includes the cycling of antibiotics. Aerosolized antibiotics have the advantage of achieving high intrapulmonary concentrations with the potential for fewer associated systemic side effects. The use of inhaled antibiotics for chronic *PsA* infection has become a standard of care for CF. The goal of this therapy is the reduction of *PsA* density and host inflammation, maintenance of lung function and reduction in the frequency of acute pulmonary exacerbations [28,29].

PsA infections can manifest in different ways, with early *PsA* infections being mostly transient. Clearance of organisms can occur either spontaneously or after treatment with anti-*PsA* antibiotics [30,31]. These infections are typically caused by organisms that are sensitive to a variety of antibiotics, and evidence suggests that these transient infections are not associated with long-term decline in pulmonary function [10,32]. Several European CF care centers have adapted a strategy to aggressively treat early *PsA* infections, resulting in delay of the onset of chronic *PsA* colonization [33–35]. Two large controlled studies have examined similar strategies to eradicate early *PsA* infection (Early Inhaled Tobramycin for Eradication, and Early Pseudomonas Infection Control) [31,36–38]. These studies demonstrated that treatment of early *PsA* infection with tobramycin inhalational solution (TIS; TOBI®, Novartis Pharmaceuticals Corporation, USA) twice daily for 28 days (with or without oral ciprofloxacin) leads to successful eradication of *PsA* in up to 85% of CF patients, with >60% of patients remaining *PsA* negative at 6 months post-treatment.

As stated previously, chronic *PsA* infection (frequently with mucoid strains) is an important complication in CF, and is therefore the prime target of long-term inhaled antibiotic therapy

in these patients. The remainder of this review will focus on established therapies and therapies under development to manage chronic *PsA* infection in CF.

• Overview of the market: currently available therapies to manage chronic *PsA* infection

Tobramycin inhalational solution

Tobramycin is an aminoglycoside antibiotic produced by *Streptomyces tenebrarius* as a component of the nebramycin antibiotic complex [39]. It is bactericidal mechanism of action is disruption of bacterial protein synthesis. Tobramycin inhalational solution (TIS) is a sterile, clear aqueous solution with a pH and salinity specifically modified for administration by a reusable air-driven nebulizer (PARI LC Plus). TIS is formulated as single-use 5 ml ampules containing 300 mg tobramycin and 11.25 mg sodium chloride in sterile water. A landmark study evaluating TIS in CF patients chronically infected with *PsA* (FEV_1 of 25–75% of predicted) demonstrated that three 28-day TIS-treatment cycles (alternating with 28 days off treatment) over 6 months produced sustained improvements in lung function, reductions in *PsA* density and pulmonary exacerbations, and improvements in weight compared with placebo [40]. Based primarily on these results, current Cystic Fibrosis Foundation pulmonary treatment guidelines recommend TIS for the management of chronic *PsA* infections in CF patients 6 years of age and older. The cycled dosing is frequently used (particularly in the USA), and has been proposed to allow the airways to remain largely colonized with susceptible organisms, thereby maintaining tobramycin efficacy over time [41].

Tobramycin inhalational powder

Tobramycin inhalational powder (TIP) is delivered by a hand-held dry-powder inhaler device (T-326 and together known as a TOBI Podhaler™, Novartis Pharmaceuticals Corporation, USA). TIP (112 mg administered as four powder capsules twice daily) is licensed for use in 28-day cycles similar to TIS. A large multicenter study (EAGER trial) showed that the safety profile of TIP and TIS were similar and they had comparable effects on lung function and sputum *PsA* density in CF patients chronically infected with *PsA* [42]. The incidence of cough, dysphonia and dysgeusia was slightly higher in patients receiving TIP. The administration time for TIP was approximately 14 min faster than TIS (excluding time required to set up, clean and disinfect the TIS nebulizer), and patient-reported satisfaction was significantly higher for TIP compared with TIS [42].

Concentrated tobramycin solutions for inhalation

Concentrated solutions of tobramycin for inhalation (75 mg/ml formulated in 4 ml) enable delivery of a more highly concentrated solution with the same total dose of tobramycin compared with TIS (300 mg/5 ml of tobramycin). This allows for delivery in a somewhat shorter period of time (approximately 15 min), achieving a higher peak sputum concentration compared with TIS [43]. Concentrated tobramycin solution for inhalation is currently marketed in multiple countries (Bramitob®; Chiesi Farmaceutici, Parma, Italy [44] and Bethkis®; Chiesi USA, Inc., NC, USA [45]).

In the USA, the concentrated formulation of tobramycin (4 ml single-use ampules containing 300 mg of tobramycin) is administered by inhalation using a hand-held nebulizer (PARI LC PLUS® Reusable Nebulizer; PARI Respiratory Equipment, Inc., VA, USA) with an air compressor (PARI Vios®).

The European counterpart has a concentration, formulation and delivery time similar to the USA version. When compared with TIS, both concentrated tobramycin formulations lead to a reduction in the density of *PsA* in sputum after both short-term (4 weeks) and long-term (over 56 weeks) intermittent therapy in patients with CF [43]. FEV_1% predicted increased from baseline and non-inferiority between treatments was met and maintained throughout the extension phase of the study over 56 weeks [43]. No remarkable safety issues were identified throughout both study phases, with similar percentages of patients reporting adverse events (AEs) in the two treatment groups during the 8-week core phase (Bramitob®/Bethkis [31.4%]; TIS [28.0%]).

Aminoglycosides are an important antibiotic class to manage *PsA* lung infections in CF [10]. Inhaled tobramycin is well tolerated in CF patients and in a study by Ramsey *et al.* was shown to be more effective than bronchodilators or chest physiotherapy in improving lung function [4,5,40,46]. As recently summarized in a Cochrane review, inhaled antibiotics, particularly tobramycin, improve lung function and decrease pulmonary exacerbations when

compared with placebo or usual treatment. Aerosolized tobramycin at a 600 mg daily dose (300 mg twice daily) has been shown to be safe and without clear renal or audiologic complications associated with either short- or long-term use [4,5,47,48].

Nebulized concentrated tobramycin

Nebulized concentrated tobramycin (NCT; Vantobra™, PARI Pharma GmbH, Starnberg, Germany) was recently approved in Europe. NCT includes 170 mg tobramycin/1.7 ml delivered via a drug-specific nebulizer handset (TOLERO®, operated with eBase controller; PARI Pharma) that is more concentrated than TIS with 100 mg tobramycin per ml (T100). The chemical formula of NCT is identical to that of TIS ($C_{18}H_{37}N_5O_9$ for tobramycin). Pulmonary exposure to tobramycin is similar to TIS due to the delivery device used with the concentrated product. Since the composition of NCT is quite similar to TIS, bioequivalence to the TIS reference product has been the focus of NCT development for regulatory agencies.

Sands and colleagues compared the T100 product to the standard combination of TIS/PARI LC Plus in a randomized, two-period, multicenter, cross-over 14-week Phase II study. Fifty-eight patients ≥4 years of age with chronic *PsA* infection were enrolled.

Pharmacokinetics showed a nonsignificant trend toward higher sputum concentrations of T100 (potentially reflective of higher local pulmonary concentrations) and lower serum concentrations (consistent with a lower systemic exposure). There was a reduction in *PsA* CFU after treatment with both drug/device combinations, with no difference between combinations at the end of Treatment period 1, but a statistically significant difference in $\log_{10}$ CFU pre- versus post-treatment between T100 and TIS at the end of Treatment period 2 favoring T100 ($p = 0.0225$). Effect on $FEV_{1\%}$ predicted was similar for both groups for Treatment period 1; T100 (8.2 ± 9.49%; difference pre- vs post-treatment: $p < 0.0001$) and TIS (4.8 ± 9.58%; difference pre- vs post-treatment: $p = 0.0132$). The treatment effect similar for NCT and TIS after Treatment period 2 (2.4 ± 10.64% $p = 0.2436$ vs -0.44 ± 8.10% $p = 0.7862$).

AEs were noted in 29 out of 58 (50%) patients in both groups; three were severe and none were felt to be drug related (hospitalizations). Tinnitus was observed in two patients treated with T100 (3.4% of all patients) [49]. The nebulization times for the T100 drug/device combination was a mean of 4.4 min per treatment compared with the standard TIS mean of 24.3 min per treatment [49], with lowest reported duration of 14.6 min [50].

Comparable overall efficacy over the entire study supports therapeutic equivalence, and T100 had lower systemic exposure along with a similar safety profile compared with TIS.

Aztreonam solution for inhalation

Aztreonam solution for inhalation (AZLI; Cayston®, Gilead, CA, USA) is delivered via a nebulizer system (Altera® Nebulizer System; PARI Respiratory Equipment, Inc., VA, USA), utilizing a rapid delivery device (e-Flow®, PARI Pharma). It is a monobactam antibacterial dry mixture of aztreonam and arginine that is dosed three-times daily for 28-day treatment cycles. Delivery time is typically 2–3 min, and it is approved in the USA and many other countries for the management of *PsA* infection in CF patients. The AIR–CF1 trial published by McCoy and colleagues demonstrated that a 28-day course of AZLI given three-times daily led to delayed time to need inhaled or intravenous anti-*PsA* antibiotics, improved respiratory-symptom scores, increased FEV_1 and decreased sputum density of *PsA* [51]. The AIR-CF2 trial confirmed that 28 days of AZLI (with either twice or thrice daily dosing) immediately following a 28-day course of TIS delayed the time to need for additional inhaled or systemic anti-*PsA* antibiotics [52]. Subsequently, the AIR-CF3 trial showed that repeated cycled treatment with AZLI was well tolerated and demonstrated clinical benefits in respiratory-symptom scores, FEV_1 and weight gain [53]. A recently completed open-label trial in CF patients chronically treated with TIS showed that AZLI (three-times daily) had superior effects on lung function when compared with TIS over three 28-day courses [54]. The effects of both agents on FEV_1 were relatively small compared with earlier nebulized antibiotic trials. The smaller treatment effect and AZLI treatment advantage may reflect the impact of prior long-term nebulized TIS use in this population.

Inhaled colistimethate sodium

Colistin is an antibiotic belonging to the polymixin family that exerts its action through disruption of the cell-wall membrane of Gram-negative rod bacteria [55,56]. The narrow spectrum

of colistin activity targets several potential CF-related Gram-negative pathogens, such as *Pseudomonas*, *Enterobacter*, *Acinetobacter*, *Citrobacter* and *Klebsiella*. Given the low rates of resistance to colistin, it is an attractive choice to treat multidrug resistant isolates of *PsA*. Inhaled colistin (Colobreathe™, Forest Laboratories, Inc., NY, USA) is a capsule containing 1,662,500 IU of colistimethate sodium (approximately equal to 125 mg) that is administered with a portable inhaler (Turbospin®, PH&T, Milan, Italy) that relies on the patients' inspiratory flow to drive dry-powder delivery. It is dosed as one capsule twice a day and administration time is less than 60 s. The device requires little cleaning or maintenance and is disposable at the end of each 28-day pack. The Freedom study investigated the efficacy and safety of three 28-day cycles of inhaled dry-powder colistimethate sodium compared with TIS in patients with CF aged ≥6 years chronically infected with *PsA* and reported non-inferiority to TIS over 24 weeks of active cycled treatment [57]. Both study arms showed only modest effects on lung function over the course of the study. However, the study subjects rated inhaled colistin higher than TIS for ease of delivery. AEs were similar in both groups except for cough and dysgeusia, which were more frequently reported in the colistin group [57]. Inhaled colistin has gained regulatory approval in several countries to treat chronic *PsA* infection in CF, but it is not currently FDA approved in the USA.

It is worth noting that in a recent network meta-analysis, Littlewood and colleagues concluded that tobramycin inhalational preparations, AZLI and inhaled colistin had comparable efficacy in CF patients chronically infected with *PsA* when assessing improvement in FEV_1% predicted at 4 weeks [58].

• Therapies in development to manage chronic *PsA* infection

Fosfomycin/tobramycin for inhalation

Fosfomycin/tobramycin for inhalation (FTI) is a 4:1 combination of fosfomycin and tobramycin for nebulized delivery [59]. Fosfomycin is an inhibitor of bacterial cell-wall synthesis that is active against Gram-negative, Gram-positive and anaerobic bacteria [60]. Its potent activity against *S. aureus* (both methicillin sensitive [MSSA] and methicillin resistant [MRSA]) is of particular relevance to CF, since chronic MRSA infection is associated with higher mortality in CF patients [61]. Trapnell and colleagues reported that in a Phase II study of CF patients ≥8 years of age infected with *PsA*, FTI administered twice daily for 28 days was well tolerated and maintained the substantial improvements in FEV_1% predicted that were achieved following a run-in period with a different inhaled antibiotic (AZLI), when compared with placebo. In addition, there were fewer AE reported with the lower FTI 80/20 mg dose compared with the FTI 160/40 mg dose [59].

Levofloxacin

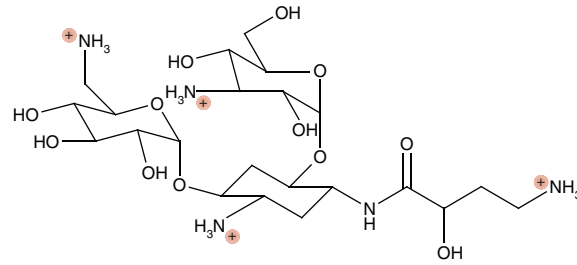
Levofloxacin is a fluoroquinolone used extensively in its oral and parenteral form for bronchopneumonia caused by *PsA* in patients with CF. MP-376 (Aeroquin™; Forest Pharmaceuticals, Inc, NJ, USA) [62] is a solution of levofloxacin (240 mg/2.4 ml) administered via a customized nebulizer (eFlow®; PARI Pharma) [63]. Results from a Phase II study of nebulized levofloxacin in CF patients chronically infected with *PsA* demonstrated reduced sputum *PsA* density at Day 28 and improvement in FEV_1, with the 240 mg twice daily dose demonstrating greater efficacy relative to other doses or placebo regimens [64]. Results of two Phase III studies were announced in a news brief by Aptalis Pharma in January 2013 [65]. In a randomized placebo-controlled single treatment-cycle study (Study 207) of 330 CF patients, comparing 240 mg of MP-376 nebulized twice daily failed to demonstrate statistical significance for time to first pulmonary exacerbation. However, MP-376 demonstrated efficacy in important secondary end points (e.g., lung function, *PsA* in sputum). Another Phase III study (Study 209) was a randomized open-label study comparing MP-376 with TIS for three 28-day on/off treatment cycles in 282 CF patients. Levofloxacin demonstrated noninferiority to TIS in relative change from baseline in FEV_1% predicted after the first treatment cycle of 28 days with a maintained effect on lung function over all three treatment cycles. Levofloxacin was also similar or superior to TIS in clinically significant secondary end points (quality-of-life measures and time to pulmonary exacerbation) [65]. Detailed results from these two trials have not yet been published.

• Liposomal amikacin for inhalation (LAI) for management of chronic PsA infection

Introduction to the compound

Amikacin is an aminoglycoside derived from kanamycin that is active against most

Table 1. Summary table of liposomal amikacin for inhalation.

Drug name	Liposomal amikacin, Arikayce™
Phase	Phase III
Indication	Chronic *Pseudomonas aeruginosa* infection in CF
Pharmacology description	Aminoglycoside
Route of administration	Inhaled
Chemical structure	 Pharmaprojects – copyright to Citeline Drug Intelligence (http://informa.citeline.com)
Pivotal trial(s)	[67,69,71,76]

CF-relevant gram-negative bacteria, including *PsA* [66]. LAI (Arikayce™, previously Arikace™; Insmed, NJ, USA) is a unique formulation that encapsulates aqueous amikacin in charge-neutral, highly biocompatible liposomes composed of dipalmitoyl phosphatidyl choline and cholesterol [67,68]. These lipids, which are found within the recycling pool of natural surfactant, are proposed to 'shield' positively charged amikacin from the polyanionic mucins that are characteristic of CF sputum [69]. A summary of LAI has been provided in **Table 1**.

Pharmacokinetics & pharmacodynamics

Okusanya and colleagues evaluated the pharmacokinetic and pharmacodynamic (PK–PD) properties of LAI in a cohort of 105 CF patients chronically infected with *PsA*. PK–PD relationships between efficacy end point measures (change in lung function from baseline) and *PsA* burden (change in $\log_{10}$ CFU from baseline) and amikacin exposure (dose, Day 1 AUC, dose/MIC ratio and Day 1 AUC/MIC ratio) were observed. An LAI dose of 560 mg once daily for a total of 14 days was associated with statistically significant improvements in FEV_1 and FEV_1% predicted and a reduction in $\log_{10}$ CFU of *PsA* from baseline. Doses >560 mg per day were not studied, and thus extrapolations of drug effects beyond this dose level are not available. These data serve to support the possible efficacy of a once-daily dose of 560 mg LAI for the treatment of CF patients with chronic infections due to *PsA* [67].

Clinical efficacy

LAI is delivered using an optimized investigational nebulizer (eFlow® Nebulizer System, PARI Pharma, Munich, Germany) [70] that rapidly delivers drug throughout the respiratory tree. Due to their uniform size, LAI (~300 nm) particles have been demonstrated to penetrate the meshwork of CF sputum where they are retained and subsequently lyzed by local *PsA*-derived products (rhamnolipids) [71,72]. Thus, LAI has the capacity to deliver drug deeply into CF sputum, where active drug is released at the site of bacteria. LAI has been shown to have a prolonged lung half-life (several hours) relative to liposome-free antibiotics [71–73], which allows for once-daily dosing and drug accumulation in the lower airway of CF sputum. In a preclinical rat model of *PsA* lung infection, once-daily nebulized LAI compared favorably with liposome-free amikacin and tobramycin at more frequent dosing (twice daily) to reduce *PsA* density [49].

Extensive preclinical studies also demonstrated that while LAI can concentrate in alveolar macrophages (thought to be part of the natural clearance response to inhaled lipids), this was not associated with macrophage defects in cytokine signaling or pathogen-killing function [74]. These studies point toward the potential use of LAI to treat intracellular pathogens such as nontuberculous mycobacteria (NTM) infection in patients with and without CF (ClinicalTrials.gov identifier: NCT01315236). Indeed, treatment of pulmonary NTM infection is one of the greatest current challenges

in CF care and therapeutic regimens typically include prolonged (months to years) treatment with intravenous amikacin combined with other antibiotics [32,75].

Recently, the results of a multinational Phase II, randomized, double-blind placebo-controlled trial of LAI in CF patients chronically infected with *PsA* was published [76]. The inclusion criteria were as follows: CF patients ≥6 years of age, FEV1 ≥40% predicted, chronic *PsA* infection, at least 28 days of clinical stability off of inhaled or intravenous antibiotics prior to enrollment and no known allergy to amikacin. Subjects harboring *Burkholderia cepacia* or nontuberculous mycobacteria, active allergic bronchopulmonary aspergillosis or significant comorbid medical conditions placing the subject at potential risk from enrollment in the study were excluded. A total of 105 subjects were enrolled and dosed across 32 international sites (19 in the USA and 13 in Europe), and cohorts were treated with different doses of LAI (70, 140, 280 and 560 mg) once daily for 28 days compared with placebo. Mucoid *PsA* infection rates were >80% at screening, and chronic TIS use was somewhat less common in the European versus USA subjects (19 and 35%, respectively). The European CF subjects were also younger than those enrolled in the USA (median age of 16.5 vs 30.5 years for the US subjects), but with similar lung function (median FEV_1% predicted of the European subjects was 65.7% [±20%, SD] vs 65.3% [±19%, SD] for the USA subjects). Due to the large number of sites enrolling a relatively small number of subjects, and the disparity in age of subjects enrolled from Europe versus the USA, the potential for bias and/or confounders cannot be excluded.

Patients treated with 280 mg (n = 21) or 560 mg of LAI (n = 36) had significant improvements in lung function relative to the placebo controls (n = 36) after 28 days of treatment ($p < 0.05$ for both dose cohorts). In the 560 mg dose group, FEV_1 improvements at Day 28 compared with placebo were 0.081 l ± 0.161 versus 0.011 l ± 0.101; p = 0.033. Consistent with a prolonged and efficacious depot effect of study drug, a prolonged treatment effect (relative difference in FEV_1 of 12.5%) was observed 1 month after discontinuing LAI compared with the placebo controls (day 56, $p < 0.01$). This was accompanied by a modest increase in systemic exposure and renal clearance, further supporting prolonged deposition of LAI in the lung following nebulized delivery. There was a significant improvement in patient-reported respiratory symptoms associated with LAI treatment; 67% of subjects in the high-dose LAI group (560 mg) reported improved respiratory symptoms compared with 36% of subjects treated with placebo ($p < 0.01$). These improvements in respiratory symptoms correlated with FEV_1 improvements at the end of treatment (Day 28, $p < 0.001$). There was a rapid and sustained reduction in *PsA* sputum density of 1–2 logs in the higher dose groups, including mucoid *PsA* strains of enrolled subjects.

Safety & tolerability

In the Phase II study of LAI, the LAI-treated cohorts demonstrated a similar safety and AE profile relative to controls over the course of the study, and AEs were consistent with underlying CF lung disease. There were no clear dose-dependent effects of LAI on a variety of pulmonary AEs (e.g., cough, dyspnea, dysphonia, pulmonary congestion, wheezing, hemoptysis) in the placebo-controlled trial. The median sputum concentration of amikacin prior to and at 1 h after dosing on days 1, 14 and 28 indicated that the airway clearance of amikacin was consistent over the 28-day dosing cycle.

During an open-label extension study completed in Europe (n = 49 subjects), LAI 560 mg administered once daily for 28 days followed by 56 days off study drug for six cycles (18 months total extension) was well tolerated, with an AE profile similar to that observed during the placebo-controlled study [76]. A total of 48:49 subjects reported an AE during the open label extension, with approximately one third experiencing an SAE (none were categorized as related to study drug). Of the 351 AEs reported, 33 were felt to be possibly or probably related to study drug Importantly, FEV_1 at the end of the sixth treatment cycle (end of treatment and 56 days off of study drug) remained above baseline values. This prolonged off-drug period has not typically been examined in CF studies of inhaled antibiotics, but it is provocative to consider whether prolonged off-treatment periods would translate into improved drug safety and/or efficacy. Together, the results of this study provided the necessary support for pivotal Phase III trials of LAI in CF patients chronically infected with *PsA*. Preliminary results from a randomized open-label Phase III study of once

Table 2. Comparison of dosing characteristics of liposomal amikacin for inhalation with other inhaled antibiotic drugs for treatment of *Pseudomonas aeruginosa* in cystic fibrosis.

Antibiotic	Drug class	Dosing and delivery
Liposomal amikacin for inhalation (LAI; Arikayce™)	Aminoglycoside	560 mg QD by nebulization with eFlow®
Tobramycin inhalation solution (TIS)	Aminoglycoside	300 mg/5 ml BID by nebulization with PARI LC Plus nebulizer
Tobramycin inhalation powder (TIP)	Aminoglycoside	112 mg BID by dry-powder inhaler with T-326 device
Nebulized concentrated tobramycin (NCT; T100; Vantobra)	Aminoglycoside	170 mg/1.7 ml BID by nebulization with TOLERO operated with eBase controller
Aztreonam solution for inhalation (AZLI; Cayston®)	Beta-lactam	75 mg TID by nebulization with eFlow
Colistimethate sodium (Colobreathe™)	Polymixin	1,662,500 IU inhalation powder, hard capsules. One capsule inhaled BID
Fosfomycin/tobramycin for inhalation (FTI)	Phosphoenolpyruvate synthetase inhibitor	80 mg/20 mg TID by nebulization with eFlow
Levofloxacin (Aeroquin™)	Fluoroquinolone	240 mg BID by nebulization with eFlow

BID: Twice daily; QD: Once a day; TID: Three-times daily; IU: International units.

daily LAI compared with twice daily TIS have been reported. Key enrollment criteria included age >6 years, FEV_1 >25% predicted, confirmed chronic *PsA* infection and no acute use of antibiotics for 28 days prior to initiating study drug. The preliminary results of this 6-month, three-cycle trial (28 day on/off LAI vs TIS), suggest that LAI is noninferior to TIS to maintain lung function in CF patients chronically infected with *PsA* [77]. In data presented in abstract and poster form, 371 patients were screened and 302 were randomized to LAI (n = 148) or TIS (146) arms. Safety data indicated that 84.5% of LAI and 78.8% of TIS subjects reported treatment emergent AEs (TEAEs). A total of 38.5% were categorized as drug related in the LAI group compared with 14.4% in the TIS group. A total of 95% of TEAEs were graded as mild or moderate in the two groups, and 10.1% of subjects in the LAI arm experienced an AE that led to discontinuation of study drug compared with 4.8% in the TIS group. The relative change in FEV_1 percent predicted from study baseline ranged from 4 to 8% (end of cycle) across the three cycles for both study groups. Both LAI and TIS reduced *PsA* sputum concentrations by 1.0–1.5 logs (end of cycle) across the three cycles, and trends in improved patient-reported respiratory symptoms in the LAI group were observed. Interim analysis from a subsequent rollover study reported that cycled LAI treatment was associated with sustained improvement in pulmonary function and a favorable tolerability profile with exposure up to 1 year [78]. **Table 2** details the characteristics comparing LAI to other inhaled antibiotics for treatment of chronic *PsA* in CF patients.

Regulatory affairs

Of note, the studies described in this section were performed predominantly in Europe, but may provide support for approval of LAI use in relevant countries. The Phase III trial achieved the primary end point of a noninferiority margin of 5% for LAI compared with TIS. A follow-up open label study in patients who completed the Phase III randomized trial is currently ongoing [79]. In addition, LAI has been granted Orphan Drug status in the US and Europe for Pseudomonas lung infections in CF patients [80].

Conclusion

There is a remarkable amount of active clinical research focused on developing new inhaled antibiotics to manage *PsA* lung infections in patients with CF. Several tobramycin-based preparations are currently available, including various liquid and dry powder formulations that have potential product-specific advantages as discussed in this review. In addition, AZLI has been approved for approximately five years in the USA and Europe, and is an established option to manage chronic *PsA* infection in CF. Colistimethate dry power has recently gained regulatory approval in Europe and is another convenient option. Efforts to develop quinolone- and tobramycin/fosfomycin-based inhaled antibiotics are ongoing, with antimicrobial targets that may extend beyond *PsA*. Compared with drugs that are approved or in development, LAI is a unique potential therapy for use in patients chronically infected with *PsA* for a number of reasons. First, it has a tolerable safety profile coupled with clear evidence of efficacy across a number of end points in Phase II clinical trials

(including lung function, sputum bacterial density and patient-reported outcomes). Second, it has been developed with an interest in reducing the treatment burden in CF patients, including the use of a rapid-delivery nebulizer and a once-daily dosing regimen. It is well known that adherence to CF-care regimens is suboptimal, and the establishment of chronic *PsA* infection typically adds another level of complexity to a CF patients' short-term and long-term care plans. Furthermore, sputum microbiology is an important aspect of care that is negatively influenced by factors such as poor adherence to prescribed antimicrobials. Thus, reducing the time and frequency of drug delivery offers the potential to optimize inhaled antibiotic adherence by simplifying chronic treatment regimens.

The fundamental difference between LAI and all other inhaled antibiotics that are approved or in development is the incorporation of liposomal technology. This novel concept also has several potential advantages, including shielding of aminoglycoside from charged sputum, delivery of antibiotic cargo deep into CF sputum, release of active drug at the site of desired action, potential activity in complex CF lung infections (e.g., nontuberculous mycobacteria) and prolonged deposition in the lung. This final attribute raises the potential for prolonged off-treatment regimens.

The availability of different inhaled-antibiotic options raises a number of questions for CF care providers, including how to best match patients to optimal treatment choices, whether to incorporate multiple inhaled antibiotics into individualized patient-treatment plans and whether continuously cycled or cotherapies are appropriate for select patients. Addressing these questions will be a significant challenge in the coming years, but will be facilitated tremendously by robust patient registries coupled with comparative effectiveness research across the CF community.

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EXECUTIVE SUMMARY

Mechanisms of action

- The active ingredient,amkacin, is an aminoglycoside antibiotic. It acts by irreversibly binding to the 30S subunit of bacterial ribosomes; blocking a recognition step in protein synthesis and therefore causing growth inhibition.
- Arikayce™ is a sustained-release lipid formulation of amikacin for inhalation.
- Charge neutral biocompatible liposomes (~0.3 μm) packed with amikacin enable penetration of the drug into biofilm and release of drug at the site of bacteria.

Pharmacokinetic properties

- High lung C_{max}, AUC and half- life results in an improved AUC:MIC ratio.
- *PsA* virulence factors can facilitate release of amikacin from Arikayce™ lipsome.
- Primarily excreted by the kidneys.

Clinical efficacy

- Prolonged deposition in the lung following treatment course resulting in maintained effect on lung function when off treatment.
- Improvement in respiratory symptoms, forced expiratory volume in 1 s and *Pseudomonas aeruginosa* burden.

Safety & tolerability

- Side-effect profile consistent with underlying cystic fibrosis lung disease and similar to placebo.
- Dysphonia observed with higher doses of drug.

Drug interactions

- Drug interactions for the inhaled formulation of amikacin have not been reported.

Dosage & administration

- Five hundered and ninety milligrams once daily delivered by eFlow® Nebulizer System.

Financial & competing interests disclosure

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