Review

Aminoglycoside therapy against *Pseudomonas aeruginosa* in cystic fibrosis: A review

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Abstract

In patients with cystic fibrosis (CF), respiratory infections with the opportunistic bacterial pathogen *Pseudomonas aeruginosa* have a major impact on morbidity and mortality. Aminoglycosides, especially tobramycin, have been used successfully to combat these infections. Aminoglycoside penetration of bronchial secretions is poor when the antibiotic is administered intravenously. Nebulization allows direct delivery of the drug to the sites of infection within the airways, while avoiding systemic exposure. Published clinical data show that inhaled tobramycin reduces the bacterial load, improves lung function and reduces the number of hospital admissions. Inhaled tobramycin has been used successfully to eradicate *P. aeruginosa* in patients with early infection. Maintaining clinical benefits requires chronic tobramycin treatment, and the concept of chronic intermittent inhaled treatment (typically, alternating drug and drug-free periods of 28 days) was introduced to minimize the emergence of aminoglycoside resistant *P. aeruginosa* strains. Other therapeutic advances include the development of different tobramycin formulations and nebulizers that reduce delivery time without compromising efficacy. An optimal treatment regimen for patients with CF with early or intermittent *P. aeruginosa* infections remains a high priority to maintain long-term lung health.

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Keywords: Cystic fibrosis; *Pseudomonas aeruginosa*; Aminoglycoside; Tobramycin

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1. Introduction

Approximately 70,000 patients worldwide have cystic fibrosis (CF), although in some developing countries the disease is not always diagnosed [1]. Since the early description of CF, pulmonary disease and accompanying infection have been recognized as playing the leading role in morbidity and mortality, resulting in premature death in the vast majority of patients [2]. Pulmonary disease is caused by mutations in the CF transmembrane conductance regulator (CFTR) gene [1], which encodes a membrane-bound cAMP-regulated chloride channel. Loss or misregulation of chloride and water secretion leads to viscous secretions in the airways of patients with CF [3–5]. This impairs mucociliary clearance [3,4], thereby facilitating chronic bacterial infections, which may start in infancy [2,6]. Pseudomonas aeruginosa, an opportunistic Gram-negative bacterium found in many natural and man-made water sources, is the most frequently identified pathogen in patients with CF [2,7]. Epidemiological data from the Cystic Fibrosis Foundation, USA, that includes more than 24,000 patients, revealed that approximately 27% of patients aged between 2 and 5 years, rising to approximately 80% of those between the ages of 25 and 34 years, have chronic P. aeruginosa infection [2].

Although P. aeruginosa infection does not appear to cause an immediate decline in lung function, it has been shown that developing P. aeruginosa infection at an early age affects long-term pulmonary disease and survival [8]. P. aeruginosa infections are difficult to treat and develop from an initial colonization into intermittent and, subsequently, chronic infections. This is mainly due to the survival of the pathogen in biofilm-like macrocolonies, which develop during chronic infections in the viscous, hypoxic mucus layers [9,10]. Most evidence would suggest that the initial infection occurs by invasion of P. aeruginosa into airway mucus and it is therefore the conductive airways that are considered to be the major target for treatment of the infection. The extent of alveolar involvement is still largely unclear, recent data from patients with end-stage disease would suggest both airways and lung tissue involvement in the absence of an aggressive antibiotic treatment able to control the infection [11]. Chronic pulmonary P. aeruginosa infection remains one of the major risk factors for lung function decline in patients with CF, particularly in children and adolescents [12]. In recent decades, however, various antibiotic treatment strategies have been developed that, in association with advances made in mucus clearance and management of the pancreatic disease, have contributed significantly to the improvement of the prognosis for patients with CF.

Here, we review published data on the impact of aminoglycoside therapy on the clinical status of patients with CF and on the development of resistance in P. aeruginosa, the use of aminoglycoside monotherapy versus use in combination with other classes of antibiotics, the pharmacokinetics of aminoglycosides, and intravenous and nebulized aminoglycoside therapeutic strategies in patients with CF.

2. Impact of aminoglycoside therapy on the clinical status of patients with CF and the development of resistance in P. aeruginosa

The inability to eradicate chronic P. aeruginosa infections in patients with CF, even with high-dose antibiotics, necessitates multiple courses of antibiotics (i.e., maintenance therapy) in order to control the bacterial load in the airways [13]. Several studies, including Phase 3 placebo-controlled, randomized clinical trials, performed in patients with CF with chronic P. aeruginosa infection, have demonstrated the improvement in pulmonary function after aminoglycoside therapy. Ramsey and colleagues in 1999 [14], reported the benefit of tobramycin in patients with CF through increased forced expiratory volume in 1 s (FEV1), expressed as a percentage of the predicted value. FEV1 was used as a primary outcome parameter in this study as it has been shown to be the best predictor of survival in patients with CF [15]. The observation that antibiotic therapy was more effective than bronchodilators or chest physiotherapy alone strongly suggests that the antibiotic therapy is directly responsible for the improvement in lung function [14]. Furthermore, aminoglycoside therapy has a positive impact on morbidity, expressed as the numbers of days of hospitalization [14]. While improvements in FEV1 could result in improved mortality, there are currently no long-term studies to confirm this [16,17].

A key feature of P. aeruginosa is its capacity to develop resistance to virtually all antipseudomonal agents, due to elevated mutation frequencies in naturally occurring strains [18]. Clinical trials have shown that repeated courses of treatment may increase the resistance rates of the pathogen to the applied antibiotics, although resistance rates against some antibiotics such as colistin remain low [13,14,19]. Continuous use of inhaled tobramycin 600 mg three times daily for 3 months led to an increase in the proportion of patients with a minimum inhibitory concentration (MIC) > 8 µg/ml, used as a measure of resistance, from 29% to 73% [19]. The increase in resistance rates was lower when inhaled tobramycin was administered intermittently [13,14], as tobramycin-resistant P. aeruginosa clones appear to grow more slowly than susceptible clones. Hence, once the antibiotic selective pressure is removed, susceptible clones overgrow resistant clones [20]. In a 5-year (1998–2002) cohort study of 4293 individuals with P. aeruginosa infection followed in the Cystic Fibrosis...
Foundation Registry, long-term inhaled tobramycin usage was identified as an independent risk factor for multiple antibiotic-resistant *P. aeruginosa* (HR, 2.08; 95% CI, 1.56 to 2.77), among other parameters (e.g. frequent courses of intravenous antibiotics and hospitalizations) [21]. However, it has been reported that the emergence of resistant *P. aeruginosa* isolates with repeated treatment is not associated with any clinical deterioration [19,22]. Furthermore, it has been shown in a 2-year study that patients infected with resistant strains (defined by parenteral MIC breakpoints), may benefit in terms of lung function improvement [23].

In addition to antibiotic exposure, impairment of the endobronchial environment stimulates hypermutable (or mutator) *P. aeruginosa* strains that are deficient in the DNA mismatch repair system [24]. These strains display a large variety of genotypic and phenotypic traits, including resistance to multiple antimicrobial agents in 30–60% of patients [24,25]. The most consistently reported mutational mechanism of aminoglycoside resistance in *P. aeruginosa* is hyperexpression of the chromosomally encoded multidrug efflux pump MexXY-OprM. The majority of isolates from patients with CF exhibiting significant resistance to aminoglycosides overproduce MexXY-OprM as a result of mutations occurring in the repressor gene *mexZ*, the product of which down-regulates the expression of the MexXY operon [26]. However, in an animal model of chronic *P. aeruginosa* infection, tobramycin treatment did not lead to the occurrence of MexXY-OprM up-regulation, unlike ciprofloxacin [27]. Overall, these results also indicate that both sensitive and highly resistant colonies of a given strain of *P. aeruginosa* may be present simultaneously in a single sputum specimen from a patient with CF.

The challenge facing both microbiologists and treating physicians is to provide *in vitro* microbiological results with clinical meaning. Traditionally, bacterial resistance is assessed using minimal inhibitory concentrations (MIC) [28]. MIC is defined as the lowest concentration of an antibiotic that is needed to inhibit growth of a bacterial isolate *in vitro* — for example, MIC$_{90}$ is the lowest concentration required to inhibit the growth of 90% of isolates tested. Breakpoints are discriminatory antibiotic concentrations that are used to define the resistance or susceptibility of bacterial isolates. The breakpoint is, therefore, a function of the MIC for the pathogen and the achievable, non-toxic serum levels of an antibiotic. For *P. aeruginosa*, the parenteral tobramycin breakpoint is 8 μg/ml; an MIC of ≤4 μg/ml indicates that the isolate is susceptible, an MIC of ≥16 μg/ml indicates a resistant isolate, and isolates with an MIC of 8 μg/ml are categorized as intermediate. To ensure consistency across laboratories, antibiotic breakpoints for specific organisms are established by the National Committee for Clinical Laboratory Standards. It is important to note that the concept of a breakpoint is based on several assumptions: the site of infection is in the bloodstream (or a pharmacokinetically comparable compartment), the bacterial population is homogeneous and there is a clear clinical endpoint. Although breakpoints have been adopted for systemic treatments of systemic infections, establishing breakpoints for inhaled therapy in treating airway infection becomes problematic. A single-compartment pharmacokinetic assumption no longer applies, as the site of infection is a complex compartment (the inflamed lung and associated biofilms [9,10]). In addition, the bacterial population is not homogeneous. In chronically infected patients with CF, more than one strain or clone of *P. aeruginosa* with diverse phenotypes are usually found, and these different strains may have different susceptibilities. The main difference to classical breakpoints is that with inhalation therapy very high local concentrations can be achieved in the airways while avoiding side effects associated with systemic exposure. Therefore, *in vitro* sensitivity testing will have limited value for predicting in vivo activity in this setting.

One of the most frequently used options to prevent rapid development of antibiotic resistance in *P. aeruginosa* in patients with CF is combination therapy with *P. aeruginosa*-specific antibiotics that display different modes of action. In a mouse model, a combination of tobramycin and ciprofloxacin had a synergistic effect in preventing resistance [27,29] and reducing bacterial load [27], although in a clinical study in which tobramycin and a β-lactam antibiotic (azlocillin) were combined, development of resistant strains was not prevented [30]. Based on these assumptions, antibiotic combinations are frequently used in patients with CF, especially during exacerbations of pulmonary symptoms [31].

### 3. Intravenous antipseudomonal antibiotic therapy in CF

Traditionally, during acute exacerbations, intravenous administration of aminoglycosides has been preferred and continues to be used, often in combination with other classes of antibiotics [31]. The mechanistic justification and the clinical evidence for the efficacy of this route of administration remain to be confirmed. To date, no clear correlation has been established between intravenous aminoglycoside therapy and improved clinical outcome. However, the potential for severe systemic side effects is certainly greater with this route of administration, compared with topical (inhaled) treatment. A recent publication has challenged the view that effective concentrations at the site of infection can be achieved using intravenous treatment during an acute exacerbation in CF, neither with single nor with combination antibiotic treatments [32].

#### 3.1. Pharmacokinetics

The pharmacokinetics of aminoglycosides in patients with CF has been evaluated extensively [33–44]. The volume of distribution per kg body weight is often increased, and the elimination half-life is decreased [40]. The higher body clearance in patients with CF, due to increased renal and non-renal elimination, demands that aminoglycosides are administered at higher doses and more frequently than in other patients [39]. Aminoglycosides diffuse poorly across lipid membranes and into bronchial secretions, and their distribution is restricted mainly to the extracellular fluid [45]. Therefore, relatively high parenteral aminoglycoside doses must be administered [16,17]. For aminoglycosides, which generally display concentration-dependent activity, the free drug maximum concentration...
The initial doses of aminoglycosides may be calculated based on body surface area [36] or, more conveniently, on body weight [30,39]. Intravenously administered aminoglycosides have traditionally been given three times daily, and initial tobramycin doses of approximately 10 mg/kg/day, or higher, have been recommended [33]. An adjustment of dosing to achieve high serum levels has a significant effect on pulmonary function [39]. If aminoglycosides are given at higher doses, the dosing frequency may be reduced due to the sub-inhibitory MIC effect, the bactericidal concentration-dependent killing and dosing frequency may be reduced due to the sub-inhibitory MIC effect, the bactericidal concentration-dependent killing and adaptive resistance [41]. Indeed, high aminoglycoside doses given at once-daily intervals have been shown to be less toxic and equally effective when data from four studies comparing once- and three-times-daily aminoglycoside regimens were analysed [37]. No significant differences in several lung function and weight parameters, or otoxicity, were found between the once-daily and three-times-daily treatments [37]. The percentage change in serum creatinine significantly favoured once-daily treatment in children, whereas in adults no difference could be found. Dosing recommendation for once daily aminoglycoside therapy have usually been adapted from regimens using bid or tid regimens. In a study to determine the optimal tobramycin dose for a once-daily regimen to treat acute pulmonary exacerbations in children with CF, it was shown that the optimal initial dosing regimen for female patients aged $\geq 14$ years should be 7 mg/kg/day tobramycin given intravenously once daily, whereas for all other patients with CF an initial dose of 9 mg/kg/day given intravenously once daily was calculated as appropriate [47]. In addition, there is evidence of less nephrotoxicity in children with once-daily treatment [36].

3.2. Adverse effects

Patients with CF are treated with antibiotics throughout their lives: for the eradication of a new infection, during pulmonary exacerbation, or for control of a chronic infection. Thus, any new or unusual symptom must be pursued rigorously and considered as a possible side effect. The potential toxicities of different aminoglycosides are similar. Overall, only a few side effects have been reported using high-dose aminoglycoside therapy, which are largely confined to the vestibular auditory and renal systems [38,48–53].

Typical manifestations of ototoxicity reported in patients with CF following parenteral aminoglycoside administration include loss of hair cells and degeneration of ganglion cells resulting in cochlear damage [54,55], which may lead to high-tone deafness [55]. Acute vestibular toxicity may be seen if the drug is given rapidly [56]. A slow infusion of the drug is, therefore, preferable. Due to the toxicity of aminoglycosides, serum trough and peak levels are normally determined [16]. Potentiating risk factors for aminoglycoside-induced vestibular toxicity include renal dysfunction, hyperthermia, concomitant use of other ototoxic agents (e.g., furosemide, cisplatin and vancomycin) and prior exposure to aminoglycosides [57]. Unfortunately, there is little evidence to support any of these claims. Of note, some cases of vestibular toxicity have been reported in patients with low serum aminoglycoside concentrations [57]. Therefore, therapeutic monitoring should not simply focus on maintaining target-range serum concentrations, but also on the development of symptoms — for example, special attention is warranted in patients who develop dizziness, as this is a potential sign for vestibular toxicity.

Some studies suggest that tobramycin may be less nephrotoxic than gentamicin [16]. Acute renal failure has been very rarely reported in patients with CF. Smyth et al [58] recently published a case-control study of 24 patients with CF from 20 UK centres. They found that use of intravenous aminoglycoside is a risk factor for renal impairment, with gentamicin being more nephrotoxic than tobramycin. Other known risk factors for renal impairment (prior renal disease, acute dehydration, or long-term treatment with a nephrotoxic drug) were also present in most of the cases in that study [58]. Therefore, for patients at risk of developing acute renal failure, the authors recommended either withholding aminoglycosides or monitoring their use closely [58].

To avoid aminoglycoside toxicity, liposome-encapsulated aminoglycosides have been developed and shown in vitro to be more effective than the corresponding free drug against P. aeruginosa [59]. An alternative strategy is to administer aminoglycosides directly to the infected airways in patients with CF (see below).

Another potential side effect of tobramycin is the promotion of co-infections with other pathogens. Exposure to tobramycin has been demonstrated in vitro to protect Staphylococcus aureus from eradication when co-cultured with a virulence factor secreted by P. aeruginosa [60]. Furthermore, prolonged incubation of the S. aureus with either P. aeruginosa or the virulence factor, selected for small-colony variants that were resistant to aminoglycosides [60].

4. Sub-inhibitory concentrations of aminoglycosides in the airways of patients with CF

Poor penetration of intravenously administered aminoglycosides into bronchial secretions of patients with CF may impair aminoglycoside efficacy [48]. Mucous plugs in patients with CF are mainly composed of negatively charged glycoproteins, arising from hypersecretion of mucins, and DNA, due to massive necrosis of infiltrating neutrophils from the blood [64]. Positively charged aminoglycosides, such as tobramycin, may be bound to these compounds [61–63]. Consequently, bactericidal concentrations of aminoglycosides in mucous plugs may be difficult to attain, and it has to be assumed that when parenterally administrated often only sub-inhibitory aminoglycoside concentrations are present at the site of infection. The same is true for poorly ventilated areas. The biofilm mode of growth of P. aeruginosa in chronic lung infections poses additional problems, and investigators have suggested that antibiotic susceptibilities should be determined in vitro in
biofilm cultures rather than using simple bacterial isolates [64–66]. Interestingly, sub-inhibitory tobramycin concentrations induce P. aeruginosa biofilm formation in vitro [67], but its relevance in clinical practice is not fully understood.

5. Nebulized aminoglycosides in CF

5.1. Rationale for inhaled tobramycin

The bioactivity of tobramycin in sputum is low when administered intravenously [68]. In a study of intravenous tobramycin treatment in patients with CF, tobramycin sputum concentrations of at least 25 times the apparent MIC were required to produce a measurable decrease in P. aeruginosa density in sputum [68]. However, administering tobramycin as an aerosol improves the delivery of the antibiotic to the site of infection, namely in the airways (Fig. 1) [68,69]. Data from different studies comparing peak tobramycin levels in serum and sputum show that administration of aerosolized tobramycin results in low serum levels, whereas sputum levels more than 1000-fold greater than those in serum were achievable with aerosolized tobramycin (Table 1) [68–72].

5.2. Clinical efficacy

To raise concentrations in the infected respiratory tract while avoiding toxicity, aminoglycosides have been nebulized for direct delivery into the airways of patients. Aerosolized gentamicin [73], tobramycin [13,14,19,22,74,75] and amikacin [76], or a combination of an aminoglycoside with a β-lactam antibiotic, have been evaluated [33,80,81]. Most of these studies showed improved lung function or a slowing down of the deterioration in lung function in the active treatment group [13,14,19,73–78]. Additionally, studies reported a reduction in the number of hospital admissions related to respiratory events [25,78,82,83] and reduced use of parenteral antibiotic treatment [14]. Reduction in P. aeruginosa density has been reported in several studies [13,14,19,74–79]. A clinical pharmacology study showed that peak sputum concentrations using tobramycin for nebulization were higher than the threshold for a significant antibacterial effect against P. aeruginosa (>400 µg/g) for two different tobramycin nebulizer formulations [79]. A review of randomized, controlled trials showed the benefit/efficacy of nebulized tobramycin, with no demonstrable adverse effects [80]. Two independent multi-centre studies including a total of 520 patients with CF demonstrated conclusively that chronic intermittent therapy with aerosolized tobramycin solution for inhalation (300 mg/5 ml preservative-free (TOBI®, Novartis) to be administered with PARI LC Plus) improved lung function, reduced hospitalization and reduced concomitant intravenous antibiotic therapy [14]. More recently, safety and efficacy data have been published on another tobramycin solution for inhalation (300 mg/4 mL, Bramitob®, Chiesi) [74,75].

5.3. Adverse effects

When given as an aerosol, the resultant low serum aminoglycoside levels may avoid systemic toxicity, although this depends on the dose administered. Administration of aerosolized

![Fig. 1. The rationale of using inhaled tobramycin: mean sputum tobramycin concentrations after intravenous tobramycin 80 mg/m² three times daily (approximately 6–10.8 mg/kg/day) for 18–21 days in four patients with CF [71] and after inhaled tobramycin 300 mg/5 ml twice daily with PARI LC Plus for three 28-day on/28-day off treatment cycles in 247 patients with CF [72].](image-url)
tobramycin 600 mg daily dose, during tidal volume breathing manoeuvres did not lead to audiometric abnormalities or renal toxicity in patients with CF [19,81]. Similarly, no signs of renal or ototoxicity were detected after long-term aerosolized tobramycin therapy [19,22]. However, a patient with CF was reported to have developed recurrent eosinophilia and severe persistent bronchospasm following repeated administration of preservative-free tobramycin by inhalation [82]. In a European consensus on antibiotic therapy against *P. aeruginosa* in patients with CF, inhaled tobramycin has been recommended at doses between 80 and 300 mg given twice daily [16,17]. Long-term safety studies, using more sensitive methods for the investigation of renal function, may be of interest, in particular in patients with other risk factors for impaired renal function [16,17]. Tobramycin levels in serum samples from patients with CF may vary considerably after aerosol treatment [81], but concentrations are rarely high; therefore, monitoring serum concentrations is not considered to be a part of routine clinical care for patients using inhaled tobramycin exclusively. Patients receiving concomitant parenteral aminoglycoside therapy should be monitored as clinically appropriate, taking into account the potential risk of cumulative toxicity. Caution is also warranted in patients with known or suspected renal dysfunction.

5.4. Development of aminoglycoside resistance

Emergence of aminoglycoside resistance after aerosol therapy has been noted in several studies [13,14,19,78], although *P. aeruginosa* susceptibility was regained after drug-free periods [19]. This led to the concept of intermittent dosing [13,14]. Usually, inhaled tobramycin is given in cycles, consisting of 28 days on therapy followed by a drug-free interval of 28 days; the impact of antibiotic resistance on the efficacy of more prolonged aerosolized therapy is unknown. Breakpoints for inhaled tobramycin for *P. aeruginosa* isolates from patients with CF have been recommended (susceptible, ≤64 µg/ml; resistant, ≥128 µg/ml) [83]. Applying these breakpoints, 95.1% of *P. aeruginosa* isolates from patients with CF were susceptible to tobramycin using agar dilution or Etest high-range strips [83], but their clinical relevance has not been established.

5.5. Impact of the inhalation device and clinical status on aminoglycoside efficacy

Several factors concerning the delivery system and the patient can influence the efficacy of nebulized aminoglycosides in CF. Aerosol devices may deliver approximately 15–20% of the medication to the airways; the remainder is either impacted on the oropharynx and swallowed, or is exhaled [84,85]. There is a negative relationship between the degree of lung damage and distribution of the drug in the airways [86]; patients with mild CF and low sputum production benefit most from aerosolized antibiotic therapy [22]. A mathematical airway-surface liquid model combined with a lung-deposition model has been used to calculate tobramycin doses for inhalation [87]. Given the multiplicity of parameters interfering with optimal delivery to the site of infection, newly developed nebulizers need to be tested adequately in patients with CF to ensure the efficacy and safety of the inhaled aminoglycoside.

Aerosol treatment is often time-consuming and inconvenient for the patient. New strategies, such as a dry-powder formulation (tobramycin inhalation powder, TIP) with the excipient distearylphosphatidylcholine (DSPC), are in development to reduce the administration time. *In vitro* studies have shown that tobramycin encapsulated in DSPC more effectively retarded siderophore production by *P. aeruginosa* than did free tobramycin [88]. In a multicentre study, the delivery time with TIP was reduced to one-third of that with TOBI, and systemic exposure was comparable [89]. The expected benefits from TIP over existing treatments lie in its shortened administration time (about 3–7 min, instead of the average 15–20 min required for TOBI [89]), better portability (small inhaler that does not require a compressor or power supply), storage at room temperature, easy maintenance (disposable device that does not need burdensome cleaning and disinfection procedures) and, most importantly, convenience.

6. *P. aeruginosa* eradication in clinical trials

It is now generally recognized that aggressive antibiotic treatment can postpone chronic infection and even eradicate *P. aeruginosa* from the airways of a patient with CF when the treatment is started early, immediately after the first detection of the non-mucoid pathogen in the patient’s airway. In a placebo-controlled, double-blind, randomized trial, inhaled tobramycin significantly reduced the onset of chronic *P. aeruginosa* infection, compared with placebo, when given immediately after airway colonization by *P. aeruginosa* [90]. This is consistent with the effects of other antibiotics, such as aerosolized colistin monotherapy [91] or in combination with oral ciprofloxacin [92] and data comparing different regimens are still lacking. Data from subsequently reported small-scale studies have also demonstrated that early treatment of *P. aeruginosa* lung colonization effectively eradicated the pathogen [93–95]. These findings were based mainly on negative cultures and/or decreasing serum levels of *P. aeruginosa* antigens, using enzyme-linked immunosorbent assays [93,95,96], as well as genotyping of *P. aeruginosa* in isolates from patients with CF [97]. Antibiotic strategies for eradicating *P. aeruginosa* have recently been reviewed [98].

The initial *P. aeruginosa* infection often occurs in patients with CF under the age of 6 years [2,6]. Despite the medical need and the use in clinical practice of different treatment regimens in younger children, only limited reliable scientific data are available in this patient population. A large-scale multicentre trial of *P. aeruginosa* eradication therapy using tobramycin that included young patients (<6 years old) has recently been completed in Europe [99,100]. An additional multicentre trial in the USA that includes a similar patient population is ongoing.

A prerequisite for early eradication therapy is the reliable diagnosis of *P. aeruginosa* colonization/infection. This is particularly difficult in infants and small children unable to expectorate sputum [101,102]. Consequently, other methods...
(e.g., cough or oropharyngeal swabs, sputum induction and bronchoalveolar lavage) are needed.

Other techniques, such as sensitive serological tests for *P. aeruginosa* antigens, would be useful in this context [93,96,103], although their value in monitoring disease progression is not yet fully understood. Similarly, the possibility of using molecular methods (e.g., polymerase chain reaction) to screen for early *P. aeruginosa* infection would be valuable, but the lack of standardization may hamper the use of this technology for diagnostic purpose.

7. Aminoglycosides for improving the function of CFTR in patients with CF carrying stop mutations

Gentamicin has been shown to induce read-through of premature stop codons in class I CFTR mutations *in vitro*, and this leads to improvements in CFTR-dependent ion transport and protein localization on the apical surface of respiratory epithelial cells in patients with CF [104,105]. However, in a recent multicentre study [106] conducted in two cohorts of patients with CF, neither gentamicin nor tobramycin delivered nasally produced detectable changes in nasal ion transport or CFTR localization in cells obtained by brush sampling from either study group. These findings suggest the need for either improved drug delivery methods or more potent drugs. A compound without antibiotic activity, PTC 124, appears to be superior to aminoglycosides and has shown promise in an early clinical trial in patients with cystic fibrosis [107].

8. Summary

Aminoglycosides, and especially tobramycin, are clinically effective against *P. aeruginosa* when administered intravenously or by nebulization in patients with CF. Tobramycin increases pulmonary function and lowers *P. aeruginosa* colony counts in patients’ sputum samples. The use of intravenous aminoglycosides reduces the frequency of acute exacerbations. Nebulization and inhalation of tobramycin into the airways of patients with CF results in higher local drug concentrations in the lung/airways and diminishes toxicity, compared with intravenous administration. Early treatment of intermittent colonization by *P. aeruginosa* delays the onset of chronic *P. aeruginosa* infection. Aminoglycosides may also be effective in suppressing stop mutations in CFTR.

Conflict of interest

Florian Brockhaus and Gerhild Angyalosi are employees of Novartis.

Dr Felix Ratjen is the principal investigator of the Early Intervention TOBI Eradication (ELITE) trial, a study assessing the efficacy of inhaled tobramycin in early *P. aeruginosa* infection, which is sponsored by Novartis. Dr Ratjen is serving as an advisor to Novartis and has received support for lectures sponsored by Novartis.

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