HHS Public Access

Author manuscript

Expert Rev Respir Med. Author manuscript; available in PMC 2016 September 22.

Published in final edited form as:

Expert Rev Respir Med. 2013 June ; 7(3): 231–243. doi:10.1586/ers.13.25.

Novel endpoints for clinical trials in young children with cystic fibrosis

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Introduction

Cystic fibrosis (CF) is a disease caused by mutation in the gene encoding the cystic fibrosis transmembrane conductance regulator (CFTR) protein. Although CF is a multi-system disorder, progressive lung disease is the most common cause of mortality. Decline in lung function is largely a result of structural lung damage associated with neutrophil-dominated inflammation and chronic pulmonary infections (1). Evidence of structural lung disease on chest computed tomography (CT), pulmonary inflammation and endobronchial infection (2, 3) (4) have all been reported in the first months of life in infants diagnosed with CF following newborn screening (NBS), and often in the absence of respiratory symptoms (5, 6). However, lung function is reported to be within normal limits for the majority of young infants diagnosed by (7–9), indicating a critical window of opportunity for intervention. While many new and exciting CF disease modifying agents are undergoing clinical trial evaluation (see (10) for review), including CFTR potentiators and correctors, clinical trials are difficult in patients younger than 5 years due a lack of age-appropriate clinical trial endpoints (ref 1. Davis SD, Brody AS, Emond MJ, Brumback LC, Rosenfeld M. Endpoints for clinical trials in young children with cystic fibrosis. Proc Am Thorac Soc. 2007 Aug 1;4(4):418–30). This population, with less severe lung damage, are most likely to benefit from intervention with disease-modifying therapies and implementation of such therapies in infancy has the potential to change the prognosis for CF patients. As such, methods to assess early lung disease with the potential to serve as outcome measures for clinical trials are urgently needed. The increased emphasis on the detection and treatment of early CF lung disease over recent years (11) has driven the need for the development of sensitive, reproducible, and feasible outcome measures for quantifying mild lung disease in young children with CF, including advancements in infant and preschool lung function testing,

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imaging of the chest, and the development of biomarkers; some of which will be reviewed herein as potential objective endpoints.

Lung function testing in infants and young children

As CF lung disease begins in the peripheral airways (see (12) for review), lung function tests that are sensitive to small airway dysfunction are required. While spirometry is the standard lung function measure in CF related clinical trials, it is generally considered insensitive to changes in small airway function (13–15) and cannot be reliably performed in young children. Techniques to measure lung function during infancy and the preschool years have developed over recent decades (16). In infancy their routine utilisation remains limited due to the requirement for sedation and the technical nature of infant lung function testing. In the preschool years lung function measures requiring tidal breathing are generally more successful than spirometry (17) as young children often lack the coordination for the forced exhalation, especially once low lung volumes are reached. We have reviewed the currently available or emerging lung function tests which have the potential to be applied in young children with CF.

Inert tracer gas washout

Inert tracer gas washout tests provide a measure of ventilation inhomogeneity (VI), over single or multiple breaths (SBW or MBW). The MBW involves tidal breathing of an inert tracer gas, generally helium (He) or sulfur hexafluoride (SF_6), which is first "washed in," then "washed out." Alternatively, 100% oxygen (O_2) can be used to "wash out" the resident nitrogen from the lungs, though this technique is not standard practice in infants due to the effects of 100% O_2 on breathing pattern during infancy (18). A range of outcomes can be calculated, including the lung clearance index (LCI) and moment ratios, as well as indices representing the convection dependent inhomogeneity of the conducting airways (S_{cond}) and the diffusion convection dependent inhomogeneity at the entry to the acinus (S_{acin}) derived from phase III slope analysis (See (19) for review). However, the most sensitive measure for detection of CF lung disease is not yet known.

The LCI is elevated in the majority of preschool and school-aged children with CF (13, 15, 20) and is more sensitive than spirometry in this age group (13, 21). In addition, the LCI has been reported to increase with *Pseudomonas aeruginosa* infection in pre-school children (15) and to decrease following intravenous antibiotic therapy for pulmonary exacerbation (22), indicating that LCI may be a suit(23)able clinical tool for examining the response to therapeutic intervention in this age group (11, 24, 25). While LCI has recently been shown to be increased in the presence of Pseudomonas infection and airway inflammation (26), the LCI is less often abnormal in infants with CF (26, 27) and does not correlate with the presence of bronchiectasis on chest CT (23)

The SBW is not routinely used in the infant / preschool population due to the requirement of a vital capacity manoeuvre. However, recently, a SBW of a double tracer gas mixture (SF_6 and He) during tidal breathing (approximately 30 seconds) was shown to be feasible in healthy adults (28). In short, the gases are thought to distribute unequally in the lung periphery due to their marked differences in molecular mass resulting in a non-linear

washout relationship which may be altered in small airway disease (28–30). If proven to be feasible and a sensitive marker of lung disease in the paediatric CF population, such a technique may eliminate the requirement of vital capacity manoeuvres and long washout times; though much groundwork remains regarding the sensitivity, potential confounders and appropriate modelling of the outcomes obtained from this test (28).

The inert gas washouts are potentially advantageous in that they can be performed during infancy without sedation (31) though their sensitivity for detecting small airways disease is unknown in this age group. In addition, differences in reference ranges between the N_2 and SF_6 washout methods will require specific reference equations for both methods from infancy to the school years for this technique to become a valuable clinical trial endpoint.

Raised volume rapid thoraco-abdominal compression technique (RVRTC)

RVRTC allows the measurement of spirometry in sedated infants. Briefly, the lungs are inflated to a pressure of 30 cm H_2O after which a jacket fitted around the chest is rapidly inflated causing the lungs to empty to residual volume (32). The majority of young infants diagnosed with CF by NBS demonstrate measures of forced expiratory volume ($FEV_{0.5}$), forced vital capacity (FVC) and forced expiratory flows (FEF) within the normal range (7–9), with lung function deteriorating later in infancy (7, 8), particularly following infection with *Staphylococcus aureus* or *Pseudomonas aeruginosa* (8). Diminished lung function (FEF_{50}) by RVRTC has been shown to track into school age in a small group of infants (33), though further longitudinal studies examining the long term impact of reduced RVRTC measures during infancy are required. In addition RVRTC has been used as an outcome measure in a study assessing the tolerability of inhaled hypertonic saline in infants (34–36) indicating that it may be suitable for monitoring lung function in response to intervention in infants with CF. In addition to the requirement of further work aimed at elucidating the relationships between RVRTC outcomes and spirometry outcomes at school age, routine use of RVRTC is limited by the requirement of sedation.

Forced Oscillation Technique (FOT)

The FOT measures input impedance of the respiratory system (Zrs) by applying pressure oscillations to the airway opening. The measured Zrs can be separated into respiratory system resistance (Rrs) and reactance (Xrs), which includes both elastic and inertive properties of the respiratory system. Measurement of Zrs at low frequencies (LFOT) allows the partitioning of lung function into components representing airway and parenchymal tissue mechanics (37) and as such offers significant potential for detection and monitoring of early (peripheral) lung disease in infants with CF. Despite this only one small study, demonstrating that several markers of inflammation correlate with LFOT outcomes, has been published in infants with CF (38). The lack of commercially available equipment, sufficient expertise, reference equations and the need for sedation significantly limit the ability of the LFOT to be used in large scale clinical trials.

The medium frequency FOT (see (39) for review) has been applied to young children with CF (≥2 years), with several authors concluding that this technique fails to adequately identify airway obstruction in CF (40–42). Gangell et al. reported higher Rrs and lower Xrs

in young children with CF than in healthy children; however, the majority of young children with CF had lung function within the normal range (43). Interestingly, children with current symptoms exhibited worse lung function when compared with asymptomatic children (43). As early CF lung disease is known to affect the peripheral lung, measures of Xrs or the area under the reactance curve (AX) may be more sensitive to changes in disease status than the traditionally reported Rrs. The technique is easy to perform and commercial equipment is available, as are comprehensive reference ranges (44). Further studies examining the relationship between FOT outcomes and structural lung damage, the presence of pulmonary infection and inflammation, and during interventions would be advantageous to determine this technique's ability to be utilised as a clinical trial endpoint in preschool children.

Interrupter technique

The interrupter technique is based on the theory that mouth pressure and alveolar pressure will equilibrate when expiratory flow is interrupted, allowing the calculation of the interrupter resistance (Rint) (45). While some studies of preschool children with CF have demonstrated marginally higher Rint than their healthy counterparts (46), most studies have shown that Rint measurements do not distinguish health from disease (40, 47–50). In addition, no deterioration in Rint was noted in a longitudinal study despite radiographic worsening in preschool children with CF (49). Application of the interrupter technique is limited in CF as the mouth pressure and alveolar pressure are unlikely to have reached equilibration during the short occlusion time (typically 100 ms) in patients with airflow obstruction, leading to an underestimation of airway resistance (47). Therefore, this technique is unlikely to provide the required sensitivity for a clinical trial endpoint.

Plethysmography and plethysmography derived airway resistance

Plethysmography uses Boyles Law to measure intrathoracic gas volume during breathing efforts against a closed shutter. Since gas trapping is an early marker of respiratory dysfunction in infants with CF (5, 23) the use of this may provide valuable information about early lung disease. Previous studies of infants with CF have reported varying incidences of increased FRC measured by plethysmography (9, 51, 52) and demonstrated associations with pathogen density in lower airways at the time of clinically-indicated bronchoscopy (53). However, neither measurements of thoracic gas volume or airway conductance were able to detect differences in lung function between infants on continuous prophylactic antibiotics and those who received antibiotics when clinically indicated, with the mean values for both groups falling within normal limits (54). The use of plethysmography as a clinical trial endpoint is potentially limited by the variability of the end-expiratory volume defining FRC during infancy, particularly in the setting of obstructive lung disease. In addition, normal reference data using commercially available equipment is currently scarce.

Specific airway resistance (sRaw), corrected for total lung volume, is determined by simultaneously measuring airflow and the changes in whole body plethysmograph box volume with the shutter open, avoiding the need for the panting manoeuvre (55). Nielson *et al.*, showed increased sRaw in young children with CF (40) however, algorithms applied to measures of sRaw vary in their thermal correction factors (56) and therefore are not

comparable between systems. Indeed, centre differences are apparent even with the same equipment (57) thus severely limiting the usefulness in multi-centre clinical trials.

Summary of lung function

Much progress in standardising lung function tests, which is essential for multi-centre collaborations, has been made in this age group over recent years (58, 59). However, measures of lung function in infants and preschool children continue to be limited by numerous factors including the need for sedation, lack of appropriate reference data, time and resource intensity as well as the apparent limited sensitivity of these tests in very young children with generally mild lung disease.

Imaging of the chest

In the absence of pathological specimens, imaging endpoints provide the most direct assessment of lung structure (60) and detect regional heterogeneity of disease (61). The pathological changes detected by imaging include bronchiectasis, or the pathological dilatation of airways, bronchial wall thickening, mucous plugging of airways by thickened secretions and air trapping, which is an indirect assessment of small airways disease, as well as parenchymal disease including atelectasis and consolidation (61). The three imaging modalities that have the greatest potential as endpoints for CF clinical trials are traditional chest radiography, chest computed tomography (CT) and lung magnetic resonance imaging (MRI) (61).

Chest Radiography

Several CF specific chest radiograph scoring systems have been developed and validated, and these correlate with clinical status and lung function (62, 63). Chest radiograph scores have been used as surrogates for lung disease in studies following infants from diagnosis after detection by newborn screening (64, 65), with radiographic progression evident in 85% of children by age 5 years (64). In preschool children, chest radiograph scores worsen over 3 years despite stable lung function measured by the interrupter technique (49). However, mild structural changes that typify early CF lung disease in infancy are generally subtle on traditional chest radiography, which provides a two-dimensional representation of a complex three-dimensional structure. Further, early or mild bronchiectasis may not be identified on chest radiographs (60). The advantages of chest radiography lie in its ease of use and ready availability in most centres without a requirement for sedation, however its poor sensitivity for early bronchiectasis is likely to translate into a requirement for large sample sizes in clinical trials, and the focus therefore shifted to cross-sectional imaging endpoints with increased sensitivity to detect bronchiectasis.

Lung MRI

Lung MRI is a more recent imaging technique that does not involve ionising radiation exposure, and may therefore be appropriate for more frequent assessments of lung structure, particularly in older children and adults where sedation is not required (61). However, its use in early CF lung disease is in its infancy, and it remains to be validated as an appropriate endpoint for clinical trials in early childhood. Lung MRI lacks the spatial resolution of chest

CT (66), particularly in its ability to identify segmental and subsegmental healthy and diseased bronchi (67). Dilated subsegmental bronchi with minimal bronchial wall thickening or mucous plugging remain poorly visualised with standard techniques (68). As CF lung disease begins in the peripheral airways (12), it is critical that small airways disease is accurately assessed in studies examining early structural changes in infants and young children. In addition to difficulties directly visualising peripheral airways, standard lung MRI acquisition techniques poorly visualise air trapping (66, 69), an indirect measure of small airways obstruction, and while specialised sequences can improve detection rates, the sensitivity remains, at best, moderate (70). At this stage there are no studies linking structural changes detected by lung MRI to CF pathobiology and clinical endpoints. Further research demonstrating the ability of lung MRI to track disease progression in early childhood and predict clinical outcomes is required before lung MRI can be considered a valid endpoint for CF clinical trials commencing in infancy.

Chest CT

Chest CT has received the most attention as a potential endpoint for clinical trials in CF and is recognised for its ability to detect disease in asymptomatic children with normal chest radiographs and pulmonary function tests (61). The biggest advantage of chest CT as an endpoint to monitor disease progression in CF is its ability to objectively and directly assess changes in lung structure in patients of all ages, from infants soon after diagnosis (5) to patients with severe advanced lung disease awaiting lung transplant (71). Semi-quantitative CF specific CT scoring systems have consistently high within and between observer agreement (reviewed by de Jong et al (72)). Importantly, studies in younger (73) and older (74) children have demonstrated sensitivity to detect disease progression, with improvements seen in young children treated for pulmonary exacerbations (75). From early childhood to adolescence and adulthood, chest CT correlates with lung function (76–79), and in older children, chest CT is more sensitive to disease progression than traditional spirometric assessments of lung function (74). Chest CT is predictive of mortality (80), correlates with clinical endpoints including quality of life (81) and pulmonary exacerbations (82), and correlates with underlying CF pathobiology, including measures of inflammation and infection (4, 5, 73, 75).

Standardisation of lung volume during CT acquisition will facilitate comparisons of airways between infants or in the same child over time. Long *et al.* demonstrated that controlled ventilation limited slice chest CT scans in infants result in end-inspiratory images that are more sensitive to detect bronchiectasis than images acquired at end-expiration or during quiet breathing (83). Further studies are required to determine whether motion-free images obtained during quiet breathing in unsedated infants using modern ultrafast CT scanners provide images that are comparable over time for longitudinal studies.

The requirement for anaesthesia for lung volume control results in an increased incidence of atelectasis (84), although significant proportion of preschool children with CF undergoing unsedated chest CT also have some atelectasis (85). Anaesthesia related atelectasis can be reduced by a recruitment manoeuvre following induction of anaesthesia prior to acquisition of images, involving 10 deep inspirations to 30 cm H_2O airway opening pressure (Regli and

Powers, unpublished observations). It is unclear whether a similar procedure is required for infants undergoing sedation rather than anaesthesia.

One of the argued limitations to chest CT as an endpoint is the risks associated with exposure to ionising radiation. One method to reduce exposure is to perform limited slice high resolution scans.. In school age children, limited slice approaches have poor sensitivity to detect disease extent for both airways disease (86) and air trapping (87, 88).

Modern chest CT scanners can perform full volumetric inspiratory and expiratory acquisitions in young children at relatively low doses equivalent to 2 or 3 PA/lateral chest xray series. A recent retrospective study supports arguments that scanning protocols currently suggested for imaging the lung in children with CF have a low risk of exposure related cancer. Using these data, the lifetime excess cancer risk for children scanned annually for the first 10 years of life is approximately 0.3%. However, we can expect that about 50% of children born now will die before the age of 50 years due to progressive lung disease.

Whether volumetric assessments of structural disease are more closely related to clinical features and underlying pathophysiology in young children is not known. From a clinical trials perspective, a volumetric approach is likely to reduce required sample sizes through improvements in sensitivity and precision, and should allow development of (semi-) automated analysis techniques to quantify disease. However, the standardisation of CT settings to achieve comparable images in terms of image quality and associated radiation exposures (92) will be important for multicentre studies involving chest CT.

To date, most studies using chest CT have employed a semi-quantitative CF specific CT scoring system. Tools to quantify structural lung disease in CF are in various stages of development (93–96), and (semi-) automated quantitative assessments of lung structure are likely to be preferred assessment tool in the future. Further research is required to determine the most appropriate tools to quantify early structural lung disease (gas trapping, bronchial wall thickening, bronchiectasis) and mucus plugging in early childhood, particularly assessing airway dimensions and air trapping, including formulation of appropriate reference ranges from corresponding normative data.

Summary of imaging

Chest CT is currently the imaging modality with the most promise as a robust clinical trials endpoint, particularly for the detection of airways disease including bronchiectasis, bronchial wall thickening and mucous plugging, and identification of air trapping as a surrogate measure of small airways obstruction. However, this technique is not without its limitations including the requirement for sedation to achieve lung volume control and concerns regarding radiation exposure. While there is sufficient data validating chest CT as an endpoint for clinical trials in CF, including studies from infancy, further research regarding acquisition techniques, assessment tools and standardisation for multicentre studies are required

Biomarkers

Biochemical biomarkers are a mainstay for monitoring clinical endpoints in many diseases. In contrast to lung function or imaging studies, biomarkers can assess the specific aspects of pathophysiology targeted by a potential therapeutic agent. However, identifying and measuring biomarkers of CF lung disease has proven challenging, particularly in infants and young children.

Blood

Measuring biomarkers in serum or plasma is perhaps the most common strategy for assessing endpoints in clinical trials, reflecting the relative ease with which blood based biomarkers can be obtained and evaluated. A large number of blood inflammatory markers are elevated relative in CF, including C-reactive protein (97–100), immunoglobulin G (98, 101, 102), cytokines (99), tumor necrosis factor (103), and transforming growth factor β (104). Most studies of blood biomarkers in CF have demonstrated differences from healthy controls, cross-sectional relationships to other disease measures (97, 98) or changes with exacerbation (99, 100, 104, 105). However, an ideal clinical trial endpoint would be able to assess longitudinal changes from baseline lung disease and reflect both acute and chronic disease severity. A few studies suggest that some blood based biomarkers may meet these criteria. Wolter *et al.* demonstrated that median C-reactive protein decreased in patients treated with azithromycin and correlated with lung function (106), and Proesmans *et al.* found that serum IgG concentrations are predictive of lung function over time (101). Similarly, Oliero *et al.* found lipid signatures predictive of longitudinal changes in lung function (107). While promising, these markers require further study to assess their reliability and validity as well as their ability to detect meaningful changes in response to therapy. One significant concern for any blood based inflammatory biomarker of the fact that non-pulmonary inflammation could impact biomarker concentrations and reduce the specificity of the biomarker for lung disease. Furthermore, because most studies have focused on older children and adults, it is unclear whether these inflammatory markers would be effective in young children who have correspondingly less disease.

Urine

Urine is attractive as a source of biomarkers since it can be collected non-invasively, though collection can be more challenging in younger children. There is some evidence that urinary biomarkers may be useful in CF lung disease. For example, urinary concentrations of the breakdown products of elastin, desmosine and isodesmosine, correspond to neutrophil elastase activity and reflect neutrophilic airway inflammation (108). Although there is evidence that changes in these compounds may reflect therapeutic effects (109, 110), there is currently insufficient evidence to recommend using such markers as outcome measures in young children.

Airway

In theory, the best way to track disease processes occurring in the lung is to obtain samples directory from the airway. This potential has been well demonstrated through analyses of sputum, and several sputum biomarkers appear to track lung inflammation and have

potential as clinical trial endpoints (111). However, sputum biomarkers are of limited utility in the youngest children with CF, who cannot generally produce sputum. As sputum markers have been reviewed extensively elsewhere (112) and are more relevant to an older population, they are not considered further in this review.

In younger children, flexible bronchoscopy with bronchoalveolar lavage (BAL) represents the primary means to obtain airway secretions for analysis (113). Lower airway microbiology can be assessed through analysis of BAL fluid, as can numerous inflammatory markers including neutrophil counts, neutrophil elastase, interleukin-8 and other cytokines, and others (3, 4, 114–117). Many of these inflammatory markers correlate with other aspects of disease severity including respiratory pathogens (118), radiologic findings (4, 73, 75), and outcomes from infant lung function testing (3, 117). However, bronchoscopy has several significant limitations. CF lung disease is heterogeneous, and the location of lavage can significantly influence measures of disease markers (75, 119, 120). Furthermore, bronchoscopy requires considerable time, expertise, and expense and entails risks of sedation as well as possible pyrexia, or bronchoconstriction. As a result of these limitations, most studies of BAL fluid biomarkers have relied on clinically indicated procedures, and there is a paucity of data on longitudinal validity, reliability, and sensitivity to treatment effects. Some of these issues are being addressed through the AREST CF protocol, which includes annual bronchoscopy with BAL at clinical baseline in the first six years of life (4). These studies are providing critical data on the longitudinal variation in common BAL fluid biomarkers including cell counts, neutrophil elastase, and IL-8 (4, 73)

Although the utility of BAL fluid biomarkers to longitudinally assess CF lung disease remains unproven, bronchoscopy has proven useful for assessing other clinically relevant endpoints. For example, Gibson *et al.* utilized eradication of *Pseudomonas* infection in BAL fluid cultures as a primary outcome measure in their assessment of inhaled tobramycin (121), and others have used BAL to assess concentrations of inhaled antimicrobials (122) and other drugs (123) in the lower airway. Thus, bronchoscopy with BAL does currently have a role in clinical trials, though mainly in early phase studies for targeted endpoints.

Exhaled Breath

Exhaled breath contains a number of volatile and non-volatile compounds, including many that are effective biomarkers in sputum or bronchoalveolar lavage (124). Non-volatile markers can be collected as exhaled breath condensate (EBC), and since collection only requires the subject to exhale through a chilled tube, obtaining EBC can be simple and completely non-invasive even in young children. However, EBC is mostly condensed water vapour with a small and highly variable fraction of airway secretions, complicating analysis of non-volatile EBC biomarkers. Therefore, extremely sensitive methods are needed to assess the low concentrations of biomarkers found in EBC, ideally coupled with some method to control for variable dilution (125, 126).

Despite these complications, studies have identified several EBC biomarkers elevated in CF including multiple cytokines (127–130), 8-isoprostane (127, 131), nitrates (127, 132), leukotrienes (133), and purines (117, 134) EBC pH has also been shown to be decreased in subjects with CF (135–137). EBC pH, leukotrienes and purines have been shown to track

changes in treatment of CF exacerbations (133–137). However, application of EBC biomarkers as clinical endpoints will require further efforts to explore the longitudinal variability and sensitivity to treatment effects.

Applying EBC methodology to the youngest children poses additional challenges, since infants and young preschoolers cannot voluntarily exhale through the EBC collection tube. Several studies have demonstrated that EBC can be collected from infants sedated for lung function testing (138, 139), and a recent investigation demonstrated that EBC purines correlate with infant lung function measures (140). Further study will be required to determine whether the EBC approach can be successfully applied to infants and young children.

While EBC is ideal for assessing traditional, non-volatile biomarkers, exhaled breath also contains a number of volatile compounds that could potentially serve as disease markers (141, 142). Such biomarkers have appeal, since they can potentially be measured at the point of care using devices such as an ‘electronic nose’ (143). For now, exhaled volatile markers remain at a fairly early stage of development and require further study before determination of clinical applicability.

Summary of biomarkers

Applying biochemical biomarkers of CF lung disease in young children holds great promise but poses considerable challenges. At this time, there are no biomarkers proven as a endpoints for clinical trials in young children with CF. Ongoing and future studies assessing the validity, reliability, and sensitivity to treatment effects are necessary to develop such biomarkers into clinically useful tools. Because biochemical biomarkers can assess unique aspects of disease pathophysiology, they are likely to serve as a complementary role to lung function and imaging studies in clinical trials.

Conclusions

The ideal endpoint in any clinical trial needs to be accurate, precise and reliable, with minimal risk and across all ages, easy to perform, and be inexpensive (144). Over the past 10 years, much progress has been made in developing infant and preschool lung function tests, chest imaging modalities and biomarkers as potential clinical trial endpoints for the youngest patients with CF. However, a common limitation across all of these potential targets for clinical trial endpoints is the requirement for sedation or anaesthesia in our youngest patients. While paediatric anaesthesia is considered to be fairly safe (145, 146), there are some concerns about potential neurotoxicity of sedative and anaesthetic agents (147). A reasonable approach is to minimise unnecessary exposure to procedures requiring sedation where possible, however the theoretical risks of neurotoxicity must be weighted against the potential advantages of sedation for lung function measures, lung volume control for chest CT and bronchoscopic collection of BAL fluid in a life limiting disease.

CT scans are particularly promising because of their ability to detect early structural lung changes. In fact, this year, the first study to determine whether an early intervention can prevent structural lung disease (COMBAT CF: clinicaltrials.gov identifier NCT01270074) in

a newborn-screened population will commence. This pioneering study will use chest CT as a trial endpoint to investigate whether azithromycin use from diagnosis reduces the prevalence of CT-detected bronchiectasis at age 3 years. There is no doubt that this trial will further our knowledge on the challenges of standardised operating procedures for multicentre trials, agreement on the most appropriate scoring systems for young children, and a better understanding of the sensitivity and specificity of chest CT to treatment effects.

Newborn screening for CF affords the opportunity to intervene before lung function declines; however, a distinct lack of appropriate outcome measures has severely limited the availability of clinical trials in this age group. While currently in various stages of development, the potential candidates reviewed herein may come to play a role in infant and/or preschool clinical trial endpoints. As none of these techniques is without limitation, and it remains unknown what treatment effects are achievable in this population with mild lung disease, it is likely that future trials in the youngest CF patients will require a combination of structural, functional and bronchoscopic outcomes to successfully detect and monitor CF lung disease over the course of an intervention.

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