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REVIEW

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The remaining barriers to normalcy in CF: Advances in assessment of CF lung disease

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Abstract

Despite early diagnosis of cystic fibrosis (CF) through newborn screening, a substantial proportion of infants and young children with CF still demonstrate physiologic and structural evidence of lung disease progression, such as obstructive airway disease and bronchiectasis. The growing availability of highly effective CF transmembrane conductance regulatory modulator therapy to the vast majority of people with CF has led to the potential to alter the natural history of CF lung disease, but to assess the full impact of these therapies on CF lung disease and to help guide treatment, sensitive measures of early and mild disease are needed. Chest imaging using computed tomography or magnetic resonance imaging is one approach, but technologic barriers and/or concern about exposure to ionizing radiation may limit its use. However, advances in physiologic measurement techniques and exhaled breath analysis offer another option for assessment of CF lung disease

KEYWORDS

biomarkers, cystic fibrosis, pulmonary function testing, pulmonary physiology

1 | INTRODUCTION

Despite early diagnosis of cystic fibrosis (CF) through newborn screening (NBS), a substantial proportion of infants and young children with CF still demonstrate physiologic and structural evidence of lung disease progression, such as obstructive airway disease and bronchiectasis.[1] The growing availability of highly effective CF transmembrane conductance regulatory (CFTR) modulator therapy to the vast majority of people with CF has led to the potential to alter the natural history of CF lung disease,[2] but to assess the full impact of these therapies on CF lung disease and to help guide treatment, sensitive measures of early and mild disease are needed. Chest imaging using computed tomography or magnetic resonance imaging is one approach, but technologic barriers and/or concern about exposure to ionizing radiation may limit its use. However, advances in physiologic measurement techniques and exhaled breath analysis offer another option for assessment of CF lung disease.

2 | INFANT PULMONARY FUNCTION TESTING

The lungs of infants with CF are normal at birth. However, within a few days chronic airway infection and inflammation develops.[3] Hence, the ability to monitor for CF lung disease in infancy is critical, and a variety of infant pulmonary function tests (IPFTs) have been used to study lung disease in this population.

2.1 | Raised volume rapid thoracoabdominal compression

The raised volume rapid thoracoabdominal compression technique (RVRTC) replicates adult-type spirometry in infants.[4] RVRTC requires sedation with chloral hydrate, and after adequate sedation is achieve, the lungs are inflated to 20 to 30 cm H_2O using a mask sealed with therapeutic putty. Forced expiratory flows are then

generated by rapidly inflating a jacket around the chest and abdomen, and the jacket pressure is gradually increased until flow limitation is achieved.

Early studies using RVRTC demonstrated that obstructive airway disease occurs in infants clinically diagnosed with CF.[5-7] Risk factors associated with lower lung function at 1 year of age include infection with *Pseudomonas aeruginosa* (Pa) or *Staphylococcus aureus*, a history of wheezing, and hospitalization for pulmonary exacerbation.[6,8] RVRTC has also shown that lung disease is present and progresses despite early diagnosis of CF through NBS.[1,9-11] However, lung function appears to be better than that of infants diagnosed clinically, and there is some evidence that it can improve over the first year of life.[11] These results suggest that while lung disease progression cannot be prevented through CF NBS, earlier diagnosis may help preserve lung function and growth.

In addition to detection of early lung disease, RVRTC has also been used to assess response to treatment, $FEV_{0.5}$ and FVC after antibiotic treatment, further supporting a degree of reversible lung disease.[12] In a randomized clinical trial, inhaled hypertonic saline significantly improved FEV0.5 compared to isotonic saline in infants with CF.[13] These studies suggest that RVRTC can be used as an objective outcome measure in clinical trials of therapies in infants with CF.

Although RVRTC has provided many important insights into early CF lung disease in infancy, it has multiple disadvantages that limit its use in clinical care and research. RVRTC requires sedation, and there are supply issues related to chloral hydrate.[14] Other medications have been studied, but they also have their drawbacks.[15] RVRTC is also very time and labor intensive, and a high degree of proficiency is required to consistently acquire useable data. Finally, manufacturing and support of commercial RVRTC devices is phasing out. While it is unlikely that there will be widespread use of RVRTC technique for clinical care or research, it may still play a role at specialized research laboratories with the necessary expertize and equipment.

2.2 | Body plethysmography

Body plethysmography can be used to measure functional residual capacity (FRC) in a technique similar to that for older children and adults using the infant's tidal breaths to generate pressure swings rather than a panting maneuver.[16] In contrast to the relatively large number of studies using RVRTC, data on FRC during infancy in CF are more limited. A multicenter study of infants with clinically diagnosed CF reported a significantly increased mean FRC and higher FRC was associated with worse Wisconsin chest radiograph scores and *S. aureus* infection.[17] There have only been two studies of FRC in CF NBS infants. A study performed in the UK found that FRC was significantly elevated in CF NBS infants by 3 months and persisted at 12 months, suggesting that peripheral small airway disease is present early in life and may not be completely reversible.[9] A recent study performed in the USA also demonstrated hyperinflation in CF NBS infants.[18] In this study, 42% of infants had an FRC ≥ 2 Z scores, and 9% of these infants had an FEV0.5 within normal limits, suggesting that FRC is a more sensitive measure of early CF lung disease. Body plethysmography is easier to perform than RVRTC, and success rates in obtaining research quality data are higher for BP compared to RVRTC.[19]

2.3 | Tidal breathing measurements

Tidal breathing measurements require less technical skill when compared to RVRTC and can be performed during quiet sleep, making these tests more appealing during infancy.[20] The ratio of the time to peak expiratory flow over the total expiratory time (Tpef/Te) has been shown to be decreased in obstructive lung disease in adult and children.[21] Tpef/Te measured in CF infants at 3 months was not significantly different compared to healthy controls.[9] In another study, neither there was significant difference in Tpef/Te between CF and healthy controls at 7 months of age, nor there was an association between $FEV_{0.4}$ and Tpef/Te.[22] Tpef/Te has also not been associated with later spirometry.[23] The results of these studies suggest that Tpef/Te is not a sensitive measure of early CF lung disease.

Respiratory rate (RR) is easy to measure and does not require specialized equipment. Several studies have reported an elevated respiratory rate (RR) in CF infants when compared to healthy controls.[5,18,22] Elevated RR has been found to correlate with LCI and FRC.[18,24] These studies suggest that RR may serve as an easily obtained measure of early CF lung disease, but further work is needed to determine the baseline variability of RR and whether an objective measurement of RR using a monitoring device (eg, respiratory inductance plethysmography) is required.

3 | ASSESSMENT OF LUNG DISEASE IN PRESCHOOLERS

Preschool-aged children (3-5 years) are too old for IPFTs but are usually unable to reliably perform the maximal forced expiratory maneuver required for spirometry. Several alternatives to conventional spirometry have been developed to assess lung disease in preschoolers with CF.

3.1 | Forced oscillometry

Forced oscillometry (FO) measures respiratory system impedance by analyzing changes in pressure and flow in response to an oscillatory pressure signal introduced at the airway opening.[25] Impedance is the vector sum of respiratory system resistance (R) and reactance (X), and the latter is a reflection of the viscoelastic properties of the respiratory system. Impulse oscillometry (IOS) is a form of FO using pressure impulses as the input signal. Studies using IOS and FO in CF preschoolers have yielded conflicting results.[26-31] One study of IOS

studied children with CF across a broad age range (3-18 years) and found higher R in CF patients.[31] However, a different study evaluated IOS in CF children at 2 to 8 years old and failed to detect a significant difference in values between CF subjects and healthy controls.[29] Studies using non-IOS devices have found abnormalities in children with CF, but less so than with other pediatric respiratory diseases.[26,27] In a large multicenter study evaluating lung function in preschoolers with CF versus healthy controls, FO neither distinguished CF subjects from healthy controls, nor was it associated with risk factors such as Pa infection.[28,30] In older children with CF, a retrospective study noted an improvement in both the R and X after treatment of a pulmonary exacerbation, and changes in R and X correlated with improvements in FEV1.[32] Differences in baseline disease severity and FO equipment may account for some of the differences in results. However, overall the data suggest at FO does not reliably detect early or mild lung disease in the preschool CF population.

3.2 | Interrupter resistance

Resistance of the respiratory system can also be obtained using the interrupter technique (R_{int}), where there is a brief occlusion at the mouth during tidal breathing. R_{int} has been found to increase over time in a longitudinal study but is not significantly abnormal in CF patients, which may limit its clinical utility.[29] In a different study of preschool CF patients, R_{int} was found to be higher in CF children when compared to controls. When examining bronchodilator response, R_{int} decreased similarly with bronchodilator in CF patient and healthy controls.[33] There remain limited data on R_{int} in this age group and further studies are needed to determine its clinical usefulness.

3.3 | Specific airway resistance

Specific airway resistance (sRaw) was studied in a prospective longitudinal study of CF preschool and school-age children.[29] It was found to be abnormal in those with CF when compared to controls, correlated with spirometry and remained abnormal throughout the study. sRaw became more abnormal with time and may be able to detect milder CF lung disease, though further research is needed.[29] Differences in BTPS compensation algorithms and the effects of the local measurement conditions also limit the use of sRaw as a multicenter study outcome and comparison of sRaw values between different centers.

3.4 | Spirometry

Although most preschoolers cannot meet standard acceptability criteria for spirometry, flow limitation can be achieved using modified criteria.[34] Several studies in CF preschoolers have demonstrated abnormal lung function when compared to healthy controls.[35] FEV0.5 is also significantly decreased from baseline during pulmonary exacerbations.[36] Brasfield chest radiograph scores are also associated with spirometry data in preschoolers with CF patients.[37] These results suggest that spirometry data correlate with clinical risk factors for lung function decline and can be used to monitor early lung disease. Forced expiratory flows and $FEV_{0.5}$ have been shown to be more abnormal in preschool age children when compared to healthy controls and may be more sensitive in detecting early lung disease when compared to FEV_1.[28,38] Success rates for preschool spirometry have ranged from 55% to 85%. This may limit spirometry as a research tool, but it can still be useful in a clinical setting.

4 | MULTIPLE BREATH WASHOUT

CF lung disease is characterized by heterogenous airway obstruction, which results in ventilation inhomogeneity. Multiple breath washout (MBW) quantifies the efficiency of gas mixing and is abnormal in the setting of ventilation inhomogeneity.[39] MBW requires much less cooperation and co-ordination compared to spirometry; an adequate mouthpiece/facemask seal and a regular tidal breathing pattern are required. MBW can either be performed by washing in a tracer gas (eg, sulfur hexafluoride [SF_6] or helium) followed by a washout with ambient room air or by washing out the resident N2 in the lungs by inhalation of 100% oxygen instead of room air. Because of differences in measurement and technique, MBW results from N2 washout cannot be directly compared to those from SF6 wash-in/washout. MBW test performance has been standardized in international guidelines.[34,40,41]

4.1 | The lung clearance index

The primary outcome measure of the MBW test is the lung clearance index (LCI), which is a global measure of ventilation inhomogeneity. The LCI is an index that represents the number of times the resting or end-tidal lung volume or FRC has to be "turned over" to clear the tracer gas from the lungs[39] so that the higher the LCI the greater the ventilation inhomogeneity. While other indices of ventilation inhomogeneity can be derived from the washout, LCI is the most robust and commonly used.[41]

LCI discriminates between CF subjects and healthy controls[42-44] and is abnormal in the majority of preschoolers[45,46] and school-age[45,47,48] CF patients, even when FEV_1 is in the normal range. An abnormal LCI in preschool years has been shown to predict abnormal spirometry at 6 years[49] and also predicts the onset of pulmonary exacerbations.[48] A prospective longitudinal study of preschool children with CF and healthy controls demonstrated that LCI deteriorated in the CF subjects but did not change in healthy controls.[46] Higher LCI values correlate with structural lung damage identified by high resolution computed tomography (CT), and both CT and LCI can be abnormal with FEV_1 in the normal range.[1,50-52]

LCI is a reliable, valid and responsive functional test and is now an established outcome measure in interventional trials.[53] The LCI has been shown to detect treatment effects in trials involving both

preschool[54,55] and school-age CF subjects[56-59] where a change in FEV_1 with treatment was not detected. The published treatment effects for LCI range from 0.6 to 2.2 units.[60]

Taken together, this suggests that LCI could help to identify patients with early lung disease and intervene before permanent lung damage occurs. However, exactly how the test should be used to guide clinical care and what defines a clinically meaningful change is still unknown.[61] MBW also takes longer to perform than other tests, such as spirometry and FO, which may limit its use in a busy clinical setting.

4.2 | Multiple breath washout test in infants

There are key differences between MBW testing in infants compared with older populations. Despite recent advances in the field of infant MBW testing, there are many obstacles related to the choice of device, gas and standardization across systems.[62] MBW in infants is performed in the supine position and usually requires chloral hydrate sedation, both of which can affect respiratory mechanics and distribution of ventilation. There presently is also a lack of commercially available infant MBW devices, limiting its use for multicenter studies. Another limitation is a result of the potential effects of 100% oxygen on the infant respiratory drive, which has led to SF6 being the preferred tracer gas.

Despite these limitations and challenges, studies of MBW in infants with CF have been performed, and approximately a third have an elevated LCI in comparison to healthy controls.[63] Furthermore, LCI can be normal in the presence of abnormal infant spirometry[9] or CT.[52] Although mean LCI z-score has been reported to be significantly higher in CF infants than controls at 2 years, it did not increase significantly during the second year of life.[64] In contrast to data in older children with CF, the relationship between structural lung changes on CT and LCI is not as strong in infants with CF.[51,52,64]

4.3 | Summary of MBW

In summary, the MBW test is a noninvasive outcome measure that is sensitive to early obstructive lung disease. LCI is an established primary endpoint in interventional trials and has an expanding evidence base supporting its use in clinical care.[40] Although MBW is feasible to perform, like any physiological tests, the MBW requires trained operators and technically acceptable tests to ensure accurate interpretation.[40] Future work should focus on streamlining the test for enhanced feasibility and efficiency and developing new software to automate quality control review to allow for rapid evaluation of test results.

5 | EXHALED BREATH ANALYSIS

Another approach to assessment of CF lung disease is analysis of exhaled breath. Exhaled breath contains numerous potential biomarkers of CF lung disease.[65] Breath collection is noninvasive, making breath biomarkers especially useful in the pediatric setting in which invasive procedures are particularly stressful. Breath is also available in virtually unlimited amounts from patients, unlike blood and other biofluids that are available in limited quantities.[65] These benefits have contributed to a dramatic increase in interest in breath biomarkers over the past two decades. Several breath tests are now clinically approved for use and others have been validated and standardized as clinical research tools.

5.1 | Components of exhaled breath

Exhaled breath can be divided into three fractions: gaseous breath, volatile breath, and breath condensate.[65] Most breath tests relevant to pediatric CF involve the volatile breath fraction or breath condensate. The volatile fraction of breath comprises less than 1% of its total volume and contains thousands of detectable compounds. These exhaled volatile organic compounds (VOCs) arise from three sources:

1. Exogenous: environmental VOCs are ubiquitous, originating from combustion, scented products, and other sources.
2. Endogenous: endogenous VOCs arise from host metabolism that results in the production, consumption and alteration of VOCs.
3. Biological/non-host: These VOCs are produced by bacteria that form the respiratory microbiome.

The method of breath collection can allow for preferential sampling of specific compartments of the respiratory tract.

Exhaled breath condensate (EBC) is a fluid that consists of the aerosolized droplets of airway lining fluid and the water vapor that is carried by humidified exhaled breath. The aerosolized droplets contain trapped particles and the water vapor contains dissolved components, both of which can be measured in EBC. Since the water vapor fraction of EBC is aqueous, biomarkers contained in it must be water-soluble and are, therefore, generally small and/or volatile, whereas the aerosol fraction can contain larger particles including proteins.

5.2 | Exhaled breath collection and analysis

VOCs can be analyzed online or offline, with specific methods for capture and analysis for each method. Online analysis allows for rapid, point-of-care sample analysis and negates the need for sample storage—a patient simply exhales into an analyzer or a bag of exhaled breath is injected into it. For offline analysis of VOCs, samples must be captured, stored and transported to the analyzer.[65] The gold standard for VOC identification, gas chromatography (GC-MS), requires offline analysis. It allows confident identification of VOCs present at very low concentrations in a sample. EBC collection simply involves directing exhalation onto a cold surface, such as a tube

stored in liquid nitrogen. Exhaled breath condenses onto the surface, where it can be collected and analyzed.

5.3 | Breath biomarkers in CF

Several breath biomarkers have been found to be altered in people with CF compared to the normal population. Several studies have shown that alterations in breath biomarkers correlate with measure of disease activity, such FEV1 and frequency of exacerbations.[65-68] These include exhaled breath pH, nitrite and other nitrogen oxides, 8-isoprostane, and glutathione (markers of oxidative stress), and interleukin-8.[65,66] Metabolomic analysis of exhaled breath is another approach that has been applied to the study of CF lung disease.[67] However, current limitations to their application to clinical care include lack of standardization in measurement, insufficient specificity, and poorly defined thresholds for what constitutes a clinically significant change.

5.4 | Summary of exhaled breath analysis

Breath biomarkers are a promising, minimally invasive, low-risk method for assessing CF lung disease. However, standardization of collection and analysis is necessary before exhaled breath analysis can be widely used by multiple clinical and research centers. More data on the sensitivity and specificity of exhaled breath to monitor early or mild lung disease or acquisition of new pathogens, such as Pa, are also needed. Efforts to address both these issues are underway and will provide greater clarity for the role of exhaled breath analysis in assessment and treatment of CF lung disease.

6 | SUMMARY

The need to accurately monitor early and mild CF lung disease will continue to grow in importance as highly effective CFTR modulator therapy becomes more widely available to a larger proportion and broader age range of people with CF. MBW has been shown to be a sensitive measure of early CF lung disease, and future work in this area should focus on optimizing it for routine clinical use. Infant PFTs can be useful in the assessment of lung disease in infants with CF, but issues with equipment, technical expertize, and sedation will limit the use of RVRTC to specialized research centers. Other methods, such as tidal breathing analysis and respiratory rate monitoring, require further investigation into their suitability for monitoring infant CF lung disease. Exhaled breath analysis is an emerging technology that has great potential as a noninvasive method to assess CF lung disease, but further work needs to be done to standardize methods and define the clinical relevance of test results. In summary, measurements of airway function and physiology will continue to play an important role in the treatment and study of CF lung disease.

CONFLICT OF INTERESTS

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