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Title: Quality control for Multiple Breath Washout tests in multicentre bronchiectasis studies: Experiences from the BRONCH-UK Clinimetrics study

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Abstract

Introduction: Multiple Breath Washout (MBW) to measure Lung Clearance Index (LCI) is increasingly being used as a secondary endpoint in multicentre bronchiectasis studies. LCI data quality control or "over-reading" is resource intensive and the impact is unclear.

Objectives: To assess the proportion of MBW tests deemed unacceptable with over-reading, and to assess the change in LCI (number of turnovers), LCI coefficient of variation (CV%) and tidal volume (VT) CV% results after over-reading.

Methods: Data were analysed from 250 MBW tests (from 98 adult bronchiectasis patients) collected as part of the Bronch-UK Clinimetrics study in 5 UK centres. Each MBW test was over-read centrally using pre-defined criteria. MBW tests with < 2 technically valid and repeatable trials were deemed unacceptable to include in analysis. In accepted tests, values for LCI, LCI CV% and VT CV% before and after over-reading, were compared.

Results: Insufficient data was collected in 10/250 tests. With over-reading, 30/240 (12%) were deemed unacceptable to include in analysis. In those accepted tests, overall the change in LCI, LCI CV% and VT CV% with over-reading was not statistically significant. When MBW new sites were compared to MBW expert sites, the change in LCI with over-reading was significantly greater in MBW new sites (p=0.047). Data suggests that over-reading could be important up to at least 12 months post initiation of MBW activity.

Conclusion: MBW over-reading was important in this study as 12% of tests were considered unacceptable. Over-reading improved test result accuracy in sites new to MBW.

Introduction

Lung Clearance index (LCI) is the most commonly reported multiple breath washout (MBW) parameter. It has good clinimetric properties as an outcome measure and has been adopted as a surrogate endpoint in cystic fibrosis (CF) clinical trials (1-4). There is an increasing body of evidence that forced expiratory volume in one second (FEV_1) is insensitive to early lung disease, and the majority of clinical trials in bronchiectasis have been unable to demonstrate a treatment effect using FEV_1 (5,6). This highlights the need for other, sensitive and responsive markers of lung function in bronchiectasis. LCI is being used in multicentre bronchiectasis studies as it has been shown to have good intravisit repeatability with better sensitivity in detecting lung disease on CT scan compared to FEV_1 (7-9). Much effort has been made to improve standardisation of MBW training, testing and analysis (10-14). Accurate estimation of LCI and other MBW parameters depends on correct operation of the device and appropriate analysis and interpretation of the collected data. Acquisition of good quality MBW data can be influenced by operator training, competence and experience in testing and reporting of the data.

MBW test results from different devices are not interchangeable and standardization of the device used is required in multicenter studies (15). However, even with standardization of the device, differences in software settings, patient interface dead space, breathing pattern protocols and operator technical expertise can all impact on results (14,16,17). Central co-ordination and data quality control or "over-reading" service could improve standardisation in testing and reporting of the data to a research quality standard in accordance with consensus statement guidance (10).

An over-reading protocol, to systematically evaluate MBW measurements for technical elements and stability of the breathing pattern, has been used in children with CF. The impact of the protocol on inter-observer agreement and reported MBW outcomes was assessed across 8 MBW operators from 4 institutions. Overall, use of the protocol resulted in improved inter-observer agreement but no change in reported MBW outcomes after over-reading. In 50 MBW tests (25 healthy children and 25 children with CF), application of an over reading protocol resulted in the rejection of 16.6% for technical reasons and a further 10.7% due to inappropriate breathing pattern (11). Over-reading in longitudinal studies was highlighted as important consideration, as the variability of the outcome within and between subjects will affect interpretation. In a multicentre study of 183 CF patients and 136 healthy volunteers from 8 centres, 24% of measurements in both groups were excluded due to quality issues (18). This study emphasised the importance of site training and a central over-reading process in multicentre studies. More recently, central training and assessment of MBW tests in CF pre-school children and infants, reported high rates test success (91.8%) (19).

These studies highlight the importance of central over-reading for the accuracy of MBW measurements in the multicentre setting in CF. However, over-reading of MBW data is resource intensive and significantly increases study costs. Currently there are no studies on the impact of central over-reading for MBW testing in the adult bronchiectasis population. In this study, we hypothesise that a central over-reading process in a multicentre bronchiectasis study will improve MBW result accuracy.

Aim

To determine the impact of central MBW over-reading in the bronchiectasis multicentre clinical study setting using data collected in the BRONCH-UK Clinimetrics study (ClinicalTrials.gov Identifier: NCT02468271). The impact of site MBW experience, length of sites current MBW testing activity and patient clinical status during MBW testing on the outcome of over-reading was also assessed. All sites completed a certification process before collecting MBW data in the study.

Objectives

- To assess the proportion of MBW tests deemed unacceptable after over-reading.
- To assess the change in LCI (number of turnovers), LCI variability (coefficient of variation [CV%]) and tidal volume (VT) variability (CV%) after over-reading.

- To assess the change in LCI (no. turnovers), LCI CV% and VT CV% in MBW expert sites versus MBW new sites.
- To assess the change in LCI (no. turnovers), LCI CV% and VT CV% in the first 12 months of site MBW study activity versus the remaining study period.
- To assess the change in LCI (no. turnovers), LCI CV% and VT CV% in clinically stable versus pulmonary exacerbation MBW tests.

Methods

The BRONCH-UK Clinimetrics study is a prospective cohort study to determine the utility of a range of outcome measures including LCI, in clinical trials in bronchiectasis (https://www.bronch.ac.uk/clinimetrics-study; ClinicalTrials.gov Identifier: NCT02468271). During the study, patients performed a MBW test in up to 6 study visits over a 24 month period (including 4 clinically stable and 2 exacerbation study visits). A pulmonary exacerbation was defined as an acute respiratory infection requiring oral or IV antibiotics in the presence of four abnormalities in the following nine categories: 1. increased sputum production; 2. increased dyspnoea; 3. increased cough; 4. fever >38^{o}C; 5. increased wheeze; 6. decreased exercise tolerance/malaise; 7. 10% decrease in FEV_1 from baseline; 8. new radiographic changes of a pulmonary process; 9. changes in chest sounds {{1426 O'Donnell,A.E. 1998}}.

The study is currently on-going and data collected from November 2015 to July 2017 were used in this study. The data collected were from five UK sites, trained and certified in MBW testing in the bronchiectasis patient population. Three of the sites were previously naive to MBW testing (sites 2, 3, 4) and 2 sites were MBW expert sites (sites 1, 5). During a scheduled research study visit, a Multiple Breath Nitrogen Washout (MBN_2W) test was performed using the Ecomedics Exhalyzer® D (Spiroware software version 3.1.6) and a published Standard Operating Procedure (SOP) developed for CF by Jensen and working group (http://lab.research.sickkids.ca/ratjen/mbw-centre/). MBW was carried out prior to spirometry and patients were also instructed to withhold bronchodilators prior to the study visit (SABA were withheld for at least 4 hours; LABA for > 12 hours). There was no specific instruction regarding withholding of physiotherapy airway clearance. Patients performed at least 3 trials during which they breathed 100% oxygen during tidal breathing until N_2 was washed out to <2.5% for at least 3 consecutive breaths. Sites followed quality control steps as detailed in the SOP (http://lab.research.sickkids.ca/ratjen/mbw-centre/) in addition to using quality control feedback provided by the Spiroware software (appendix 1). After the study visit, sites sent MBW data to the central over-reading facility in Belfast. Each test was assessed for validity and quality by a trained "over-reader" (KO'N, KF, DC) (appendix 2),

using pre-defined technical (signal misalignment, leak, did not meet end of test criteria, N_2 did not return to baseline between trials) and qualitative (repeatable testing session which reflects tidal breathing) criteria (11). Troubleshooting teleconferences between over-readers in Belfast and 1-2 independent over-reader(s) from the Royal Brompton London convened monthly to discuss and compare over-reading practice in accordance with criteria. Questionable tests were assessed by the group and inter-rater agreement sought.

To derive a LCI result, a minimum of 2 technically valid and repeatable trials which represented tidal breathing were required. Tests with ≥3 trials are required to calculate LCI CV% and VT CV% in accordance with analysis guidelines in the inert gas washout consensus statement (10). Values for LCI, LCI CV% and VT CV% before and after over-reading were recorded. Only those tests with a LCI value before and after over-reading (i.e. deemed to have a minimum of 2 technically valid and repeatable trials which represented tidal breathing) could be included in the comparison of LCI, LCI CV% and VT CV% before and after over-reading (i.e. those deemed not to meet this criteria either by the site or the over-reader did not have matching data). Only MBW tests with data before and after over-reading were included in the subsequent analysis. Before analysis, the data underwent a data cleaning process, where all entries were checked against source data (original spx. MBW data file) for accuracy.

Statistical analyses were conducted using SPSS version 22 (IBM Corporation, Somers, New York, USA). As the database contained multiple entries from individual patients, the mean ratio of LCI, LCI CV% and VT CV% values before and after over-reading was calculated for analysis. Mean ratios were log transformed to facilitate analysis and simple t-tests were performed on the log ratios to test the null hypothesis of no effect i.e. no change in LCI, LCI CV% and VT CV% with over-reading. Sub group analysis of logged mean ratios of LCI, LCI CV% and VT CV% were used to assess for differences based on expert MBW site vs. new MBW site; stable vs. exacerbation patient visit; length of site MBW testing activity (first 12 vs. second 12 months), using independent samples t-test. A p value <0.05 was considered significant. Results are presented in raw values of LCI (number of turnovers), LCI CV% and VT CV%.

Results:

Patient data

Data from 250 MBW tests collected from 98 patients over 5 UK sites were analysed in July 2017. Mean (SD) age was 65.4 (10.8) years, 63 (63%) were female. For all patients, the aetiology of bronchiectasis was idiopathic or post infectious. The database contained between 1-6 visits per patient during clinical stability and at the start and end of the protocol

defined pulmonary exacerbation, totalling 250 tests. Figure 1 presents steps of the over-reading process and identifies the MBW tests included in analysis.

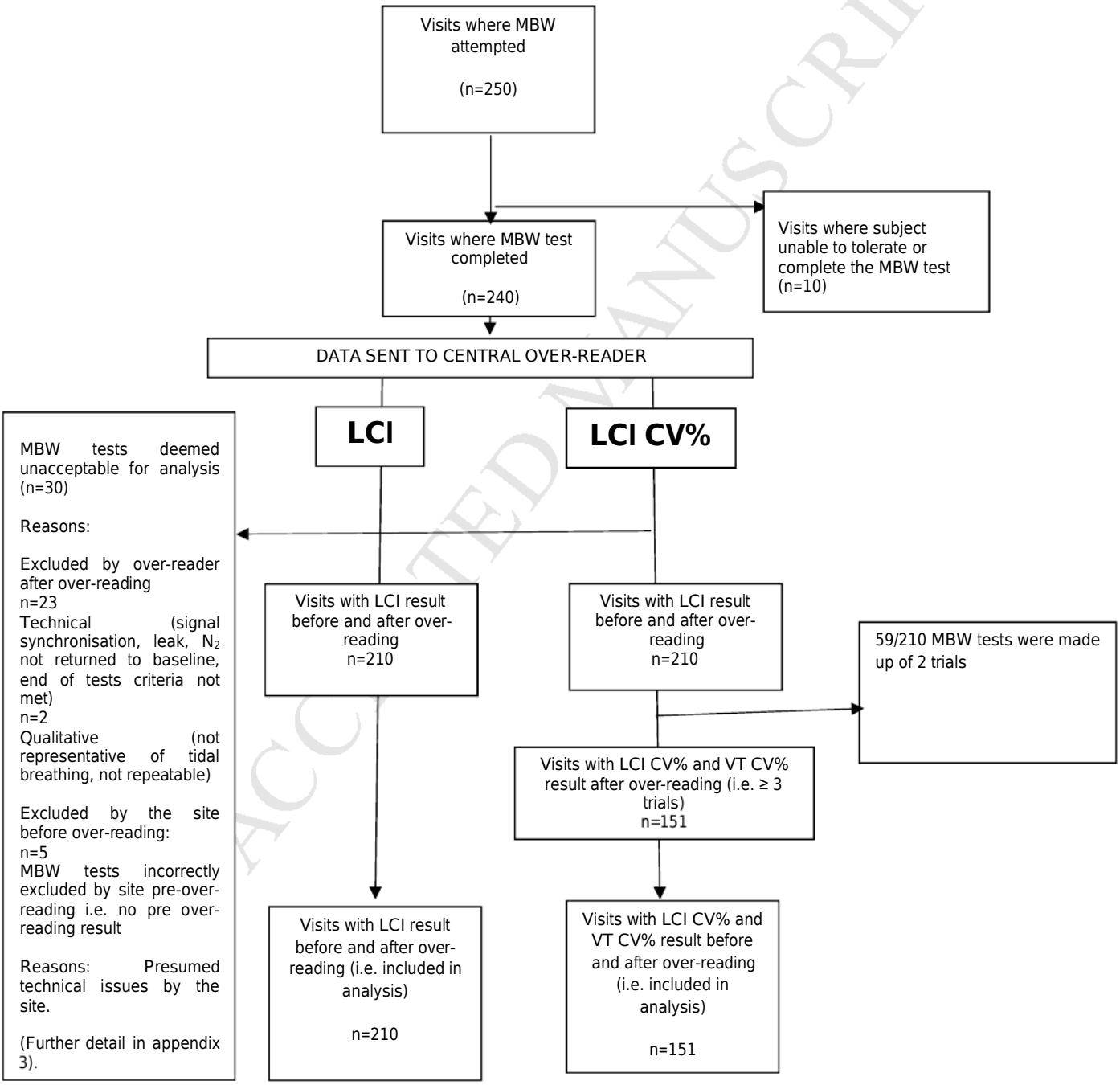

Figure 1: Flowchart identifying MBW tests across 5 sites to be included in analysis

During 10/250 visits, the patient was unable to tolerate the test long enough to collect enough data. Of those 240 visits with data, 30/240 (12%) were deemed unacceptable for analysis. 25/30 were excluded by the over-reader after over-reading and 5/30 were excluded by the site before over-reading. Without over-reading, these data would have otherwise been incorrectly included or excluded in the database analysis (Table 1). The mean (SD) [range] LCI no. turnovers of the data included in the analysis was 13.1 (3.7) [7.4-23.3]. Mean (SD) [range] LCI CV% and VT CV% of the data included in the analysis was 3.7 (2.2) [0.4-10.0] and 5.8 (4.8) [0.7-27.4]. FEV_1 data was available for 243/250 visits. Mean (SD) FEV_1 % predicted was 70.2 (20.6%), range 25.6 to 126.0. Considering the range of severity, FEV_1>80% in 79/243 (32%), FEV_1=50-80% in 120/243 (49%), FEV_1=30-50% in 38/243 (16%) and FEV_1<30% in 6/243 (2%).

The n (%) of tests excluded or included post over reading across the categories of disease severity according to FEV_1 were 8/40 (23) [FEV_1>80%], 23/40 (58) [FEV_1=50-80%], 4/40 (10) [FEV_1=30-50%]. Five excluded tests did not have FEV_1 data. Thirty-two patients accounted for the 40 tests excluded (25 patients had 1 test excluded, 6 patients had 2 tests excluded, 1 patient had 3 tests excluded). Therefore, there was no indication that test exclusions were higher in patients with more severe disease or that a core group of patients were solely responsible for the tests excluded.

The LCI CV% of those deemed unacceptable for analysis (30/240) was significantly larger than the LCI CV% in the 210/240 tests suitable for analysis (mean [SD] CV% 7.9 [2.3] vs. 2.7 [0.2] p=0.004). In total, 210 tests had LCI results which were suitable for subsequent analysis (matched data before and after over-reading). One hundred and fifty one tests had 3 or more trials and therefore had a LCI CV% and a VT CV% available for analysis.

The change in LCI, LCI CV% and VT CV% with over-reading in these tests was not statistically significant (Table 1).

The mean (SD) LCI CV% across the categories of disease severity according to FEV_1 were 3.5 (2.0) [FEV_1 >80%], 4.0 (2.3) [FEV_1=50-80%], 3.5 (2.2) [FEV_1=30-50%] and 3.6 (1.7) [FEV_1<30]. The mean (SD) VT CV% across the categories of disease severity according to FEV_1 were 6.2 (5.2) [FEV_1>80%], 5.7 (4.4) [FEV_1=50-80%], 6.2 (6.0) [FEV_1=30-50%] and 2.4 (0.7) [FEV_1<30]. Therefore there was no pattern between the variability of LCI or tidal breathing and disease severity.

Table 1: Change in LCI, LCI CV% and VT CV% with over-reading in each site and overall

Site	1	2	3	4	5	Overall
N tests attempted N patients unable to tolerate test therefore no data	74 2	33 0	25 2	59 5	59 1	250 10
N with data N (%) test excluded after over-reading N (%) test included after over-reading	72/74 4 2	33/33 6 0	23/25 4 0	54/59 9 1	58/59 2 2	240/250 25/240 (10%) 5/240 (2%)
Total excluded post over reading	6	6	4	10	4	30/240 (12%)
N tests with data before and after over reading	66	27	19	44	54	210
Mean (SD) LCI [range] (no. turnovers) change with over-reading	0.07 (0.18)	0.19 (0.28)	0.25 (0.73)	0.10 (0.21)	0.03 (0.09)	N=210 0.10 (0.28) [0- 3.21] p=0.07
Mean (SD) LCI CV% change with over-reading	0.37 (1.46)	0.96 (2.03)	0.05 (0.11)	0.38 (1.60)	0.21 (0.80)	N=151 0.35 (1.33) p=0.78
Mean (SD) VT CV% change with over-reading	0.14 (0.57)	1.66 (4.57)	0.59 (1.83)	0.09 (0.46)	0 (0)	N=151 0.25 (1.50) p=0.79

CV%= coefficient of variation; LCI= lung clearance index; N=number; SD=standard deviation; VT= tidal volume

In those tests analysed (n=210) the change in LCI with over reading was compared in MBW new sites vs. MBW expert sites. The mean change in LCI in sites who were new to MBW (mean [SD] {range} change=0.16 [0.39] {0 - 3.21} lung turnovers) was significantly larger compared with MBW expert sites (mean [SD] {range} change= 0.05 [0.15] {0 – 1.02} lung turnovers) (p=0.047). The change in LCI CV% (p=0.74) or VT CV% (p=0.46) was not significant, comparing sites new to MBW vs. MBW expert sites.

The over-reading outcome from MBW tests from the first 12 months of site MBW test activity compared to the remaining time period of MBW test activity (9 months), showed no difference LCI (p=0.23), LCI CV% (p=0.33) or VT CV% (p=0.51). The impact of the duration of current MBW activity on the change in LCI after over-reading, in new vs. expert sites was

also explored. In MBW new sites, change in LCI after over-reading were reduced to a level similar to that of MBW expert sites by 6 months (mean [SD] change=0.07 [0.14] vs. 0.02 [0.05] lung turnovers), but increased again at 12 months (mean [SD] change=0.23 [0.60] vs. 0.06 [0.18] lung turnovers), indicating that over-reading may be of benefit to MBW new sites up to at least 12 months post initiation of MBW activity.

There was no difference in the change in LCI (p=0.85), LCI CV% (p=0.41) or VT CV% (p=0.75) after over-reading between clinically stable and pulmonary exacerbation MBW tests.

Discussion

This is the first study to assess the impact of central over-reading on MBN_2W variables in a bronchiectasis multicentre clinical trial setting, including patients across the range of the disease severity spectrum. We found that 12% of MBW tests were considered unacceptable after over-reading due to technical or quality issues. Without over-reading, these tests which had significantly greater LCI variability (CV%), would have incorrectly been included in the database for analysis. This demonstrates the value of over-reading to both avoid the inclusion of invalid data, and avoid the loss of valid MBW data. For those MBW tests included in the dataset, over-reading did not result in any change in LCI, LCI variability or VT variability values. However, sub-group analysis found that there was a significant difference in the change in LCI after over-reading in sites new to MBW testing compared with those sites with MBW experience.

Our data in BE is similar to those reported in CF (11), showing no group level change in the LCI value but a substantial proportion of tests excluded due to technical or qualitative issues. Our study had a lower proportion of tests excluded compared with Jensen et al (12% versus 27%). The larger proportion of tests excluded in Jensen et al may be explained by the greater incidence of quality issues relating to patient non-cooperation and irregular breathing pattern seen in the paediatric age group during testing. In this study there was a strong emphasis on training and ongoing mentorship to the sites involved, with the support of an interactive e-learning tool (www.mbwtraining.com); given that the majority of sites were new to MBW. In addition, over-readers from the Belfast site were available via phone or email to answer MBW test queries in real time and provide feedback including picture snapshots and a training point summary to resolve/avoid the quality issue. This may have helped to minimise the overall proportion of tests excluded. In addition to these findings, our study highlighted the importance of over-reading in sites new to MBW testing, demonstrating that over-reading improves accuracy of LCI results in these sites. Results from the study by Fuchs et al also found that operator experience influenced MBW test success rate and indicated that it may improve with increasing study duration and experience with the test

procedure (18). Inclusion of a central over-reading process is resource intensive, with review of a single testing session requiring up to 1 hour including the completion of quality control logs and inter-rater agreement. Our study results may support a pragmatic approach to central over-reading i.e. sites new to MBW testing should complete over-reading for at least a 12-month duration. The degree of change in LCI with over-reading was small (mean change in MBW new sites = 0.16 lung turnovers) and much less than what could be considered a clinically meaningful change, as indicated in the CF literature (1.3 LCI lung turnovers; % change [95% limits] = 1.27 [-25 to 27] (20,21). However, the range of LCI change at an operator level (range of LCI change in MBW new sites= 0 – 3.21) suggests that impact of test quality could be larger within longitudinal studies or in studies measuring LCI treatment effect.

This study included a wide range of disease severity but we found no pattern between disease severity and MBW test quality (number of tests excluded, LCI CV% or VT CV%). Whilst the proportion of patients in the severe disease category were small (2%) and further study of MBW outcomes in this category is required, this result also demonstrates the robust nature and clinical utility of LCI as an outcome measure across a wide range of bronchiectasis disease severity.

Our results suggest that the level of MBW experience influences result accuracy but the definition of an "expert" MBW site is subjective. In this study, the "expert" sites had > 5 years' experience on site with MBW testing and were currently active as central over-reading sites. This included >2 staff who were routinely involved in MBW testing and over-reading, with regular troubleshooting discussions and interrater agreement. Clinical Research Organisation consideration of site MBW experience could reduce set-up time and result in better quality data for clinical trials. Site MBW experience is determined by individual operators and can be affected by staff turnover. Where a site is impacted by staff turnover it will take additional time re-accumulate experience, therefore consideration of this issue is important in assessing the feasibility of MBW at individual sites.

The most common reason for test exclusion was for technical reasons (leak, N_2 not returned to baseline, end of test criteria not met, signal misalignment). This highlights the key areas for ongoing training and mentorship in order to minimise the number of tests excluded due to these reasons (Table 2). In this study, fewer tests were excluded due to quality issues (reflective of tidal breathing pattern) however, this aspect has the potential to significantly alter results as demonstrated by Jensen et al. Currently, determination of tidal breathing is a subjective assessment taking into account the total duration of the washout. Further work to establish quantitative limits for breathing pattern may enable a more objective and standardised assessment. A limitation of this study is that findings are not directly applicable to other MBW devices and software.

Conclusions

As MBW testing is being increasingly used in bronchiectasis research, including sites new to the testing method, there is a need to determine what quality control measures are required to ensure that the data collected is research quality. The results from this study emphasise that central over-reading is required in bronchiectasis studies, to avoid inclusion or exclusion of invalid tests in analysis. In this study, over-reading improved result accuracy in MBW new sites and data suggests that over-reading could be important up to at least 12 months post initiation of MBW activity.

Table 2: Technical issues that result in test exclusion and key training points to reduce occurrence (10,11)

Technical issues	Key training points
Leak	Patient: • Fitting of mouthpiece and nose clip comfortably with a tight seal. • Distraction with TV to promote relaxed breathing. Recognition of leak: • Sudden spike in N_2 signal. • Deviation in N_2, O_2 or CO_2 signals inconsistent with phase of breath. • Sudden step change in volume trace. • Rise in N_2 signal early in expirogram. Action on recognition of leak: • Stop trial. • Check patient positioning, mouthpiece and nose clip. • Check fitting of patient interface components. • Repeat trial after patient rest and when N_2 returned to baseline.
Did not meet end of test criteria	Recognition of successful end of test: • ≥ 3 breaths < 2.5% N_2. • All 3 breaths reflective of tidal breathing.
N_2 not returned to baseline	Recognition of N_2 not returned to baseline: • End tidal N_2 ≥77% at start of first trial and within 1.5% of baseline on subsequent trials. Action on recognition of N_2 not returned to baseline: • Stop trial. • Allow sufficient time between trials.
Signal misalignment	Recognition of signal misalignment: • Spikes, deviation in N_2 inconsistent with phase of breath.

	Action on recognition of signal misalignment: • Stop trial. • Perform flow, gas calibration and flow gas signal synchronization calibration. • Seek advice on re-run of tests.

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Appendix 1:
Quality control messages provided by Ecomedics Exhalyzer® D using Spiroware software version 3.1.6.

QC MESSAGE	TECHNICAL	QUALITATIVE (BREATHING PATTERN)	MISCELLANEOUS (SETUP, CALIBRATION, TROUBLESHOOTING)
At least two trials needed			
BTPS Correction Flow Inspiration out of valid Range			
BTPS Correction Flow Expiration out of valid Range			
Channel Calibration skipped			
Flow Calibration skipped			
Inspiratory Flow too high			
LCI Coefficient of Variation too high			
LCI Target not reached			
N_2 Inspiration Mean out of valid Range			
O_2 Drift Correction out of valid Range			
O_2 End Expiration too high			
Sample Flow out of valid Range			
Standard versus CO_2 Cet out of valid Range			
Standard RQ out of valid Range Standard Deviation			
Wrong DSR used			
X2 Transit Time Error			

KEY: QC – Quality control; BTPS – Barometric temperature, ambient pressure; LCI – Lung Clearance Index; RQ – Respiratory Quotient; DSR – Dead space reducer

Appendix 2

Summary of over-reader training

Pre-requisites for training:

- Certificate in MBW testing for CF and/or Bronchiectasis patients.
- >2 years' experience in MBW testing.

Training:

1 day training with trained over-reader covering:

- Orientation and familiarisation with Spiroware.
- Orientation and familiarisation with pre-defined criteria for trial validity and quality.
- Orientation and familiarisation with quality control and results excel sheets for recording over-reading activity.

Key references and contacts:

- Jensen, R., Stanojevic, S., Klingel, M., Pizarro, M.E., Hall, G.L., Ramsey, K., Foong, R., Saunders, C., Robinson, P.D., Webster, H., Hardaker, K., Kane, M. & Ratjen, F. 2016, "A Systematic Approach to Multiple Breath Nitrogen Washout Test Quality", *PLoS ONE,* vol. 11, no. 6, pp. e0157523.
- Key reference documents from the LCI over-reading centre at the Royal Brompton London and Imperial College London.
- Key reference documents from the North American MBW centre: http://lab.research.sickkids.ca/ratjen/mbw-centre/#1476992018777-85ea347c-b8d3.

Certification:

- Review, analysis and submission of 20 MBW tests (provided by the North American MBW centre).
- Analyses and certification (≥80% agreement on over-reading outcome of test) issued by the North American MBW centre.

Appendix 3

Reasons for test exclusion

MBW tests deemed unacceptable for analysis (n=30)	
Reason	N (%)
Excluded by over-reader after over-reading for technical reasons including: • Patient related leak • Signal misalignment • Test did not meet end of test criteria • Equipment related leak • Other: Problems with equipment	 12/30 (40) 5/30 (17) 3/30 (10) 2/30 (7) 1/30 (3)
Excluded by over-reader after over-reading for qualitative reasons including: • Irregular breathing pattern	 2/30 (7)
Excluded by site pre-over-reading i.e. no pre over-reading result due to presumed qualitative and technical issues including: • Irregular breathing pattern • Patient related leak	 4/30 (13) 1/30 (3)

References

(1) O'Neill K, Tunney MM, Johnston E, Rowan S, Downey DG, Rendall J, et al. Lung Clearance Index in Adults and Children With Cystic Fibrosis. Chest 2016 Dec;150(6):1323-1332.

(2) Kent L, Reix P, Innes JA, Zielen S, Le Bourgeois M, Braggion C, et al. Lung clearance index: Evidence for use in clinical trials in cystic fibrosis. J Cyst Fibros 2014 Mar;13(2):123-138.

(3) Stanojevic S, Ratjen F. Physiologic endpoints for clinical studies for cystic fibrosis. Journal of Cystic Fibrosis 2016 7;15(4):416-423.

(4) Davies J, Sheridan H, Bell N, Cunningham S, Davis SD, Elborn JS, et al. Assessment of clinical response to ivacaftor with lung clearance index in cystic fibrosis patients with a G551D-CFTR mutation and preserved spirometry: a randomised controlled trial. The Lancet Respiratory Medicine 2013 10;1(8):630-638.

(5) Bilton D, Daviskas E, Anderson SD, Kolbe J, King G, Stirling RG, et al. Phase 3 randomized study of the efficacy and safety of inhaled dry powder mannitol for the symptomatic treatment of non-cystic fibrosis bronchiectasis. Chest 2013 Jul;144(1):215-225.

(6) Murray MP, Govan JR, Doherty CJ, Simpson AJ, Wilkinson TS, Chalmers JD, et al. A randomized controlled trial of nebulized gentamicin in non-cystic fibrosis bronchiectasis. Am J Respir Crit Care Med 2011 Feb 15;183(4):491-499.

(7) Rowan SA, Bradley JM, Bradbury I, Lawson J, Lynch T, Gustafsson P, et al. Lung Clearance Index Is a Repeatable and Sensitive Indicator of Radiological Changes in Bronchiectasis. Am J Respir Crit Care Med 2014 03/01; 2014/03;189(5):586-592.

(8) Gonem S, Scadding A, Soares M, Singapuri A, Gustafsson P, Ohri C, et al. Lung clearance index in adults with non-cystic fibrosis bronchiectasis. Respir Res 2014 May 18;15:59-9921-15-59.

(9) Grillo L, Irving S, Hansell DM, Nair A, Annan B, Ward S, et al. The reproducibility and responsiveness of the lung clearance index in bronchiectasis. Eur Respir J 2015 Sep 4.

(10) Robinson PD, Latzin P, Verbanck S, Hall GL, Horsley A, Gappa M, et al. Consensus statement for inert gas washout measurement using multiple- and single- breath tests. European Respiratory Journal 2013 March 01;41(3):507-522.

(11) Jensen R, Stanojevic S, Klingel M, Pizarro ME, Hall GL, Ramsey K, et al. A Systematic Approach to Multiple Breath Nitrogen Washout Test Quality. PLoS ONE 2016 06/15;11(6):e0157523.

(12) O'Neill K, Tunney MM, Elborn JS, Bradley JM. Training in Multiple Breath Washout (MBW) testing for bronchiectasis (BE) clinical trials. Eur Respir J 2017 European Respiratory Society;50(suppl 61).

(13) O'Neill K, Elborn J S, Tunney MM, O'Neill P, Rowan S, Martin S, et al. Training in multiple breath washout testing for respiratory physiotherapists. Physiotherapy 2017 Available online 25 April 2017.

(14) Saunders C, Bayfield KJ , Short C, Davies J C. 134 Training and qualifying international teams in standardised procedures: steps on the learning curve from the CTN LCI Core Facility. Journal of Cystic Fibrosis 2016 2018/02;15:S85.

(15) Poncin W, Singer F, Aubriot AS, Lebecque P. Agreement between multiple-breath nitrogen washout systems in children and adults. J Cyst Fibros 2017 Mar;16(2):258-266.

(16) Summermatter S, Singer F, Latzin P, Yammine S. Impact of Software Settings on Multiple-Breath Washout Outcomes. PLOS ONE 2015 07/13;10(7):e0132250.

(17) Yammine S, Singer F, Gustafsson P, Latzin P. Impact of different breathing protocols on multiple-breath washout outcomes in children. Journal of Cystic Fibrosis 2014 March 2014;13(2):190-197.

(18) Fuchs SI, Ellemunter H, Eder J , Mellies U, Grosse-Onnebrink J , Tümmler B, et al. Feasibility and variability of measuring the lung clearance index in a multi-center setting. Pediatr Pulmonol 2012;47(7):649-657.

(19) Stahl M, Graeber SY, Joachim C, Barth S, Ricklefs I, Diekmann G, et al. Three-center feasibility of lung clearance index in infants and preschool children with cystic fibrosis and other lung diseases. Journal of Cystic Fibrosis 2017 Available online 12 August 2017.

(20) O'Neill K, Elborn J S, Tunney MM, Bradley J M. Response. Chest 2016 Dec;150(6):1413-1414.

(21) Oude Engberink E, Ratjen F, Davis SD, Retsch-Bogart G, Amin R, Stanojevic S. Inter-test reproducibility of the lung clearance index measured by multiple breath washout. Eur Respir J 2017 Oct 5;50(4):10.1183/13993003.00433-2017. Print 2017 Oct.

Highlights

- Lung Clearance Index is emerging as a potential endpoint in bronchiectasis
- Data quality control or “over-reading” avoids the inclusion of invalid data
- MBW experience influences result accuracy