Global Lung Function Initiative reference values for multiple breath washout indices

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Abstract

Background Multiple breath washout is a lung function test based on tidal breathing that assesses lung volume and ventilation distribution. The aim of this analysis was to use the Global Lung Function Initiative methodology to develop all-age reference equations for the multiple breath washout indices lung clearance index (LCI) and functional residual capacity (FRC).

Methods Multiple breath washout data from healthy individuals were collated from sites. Data were re-analysed using the latest software versions. Reference equations were derived using the lambda-mu-sigma method using the generalised additive models of location shape and scale programme in R. The impact of equipment type, inert tracer gas and equipment dead space volume on the derived reference ranges were investigated.

Results Data from 23 sites (n=3647 test occasions) were submitted. Reference equations were derived from 1579 unique observations from participants between the ages of 2 and 81 years. Equipment type, inert tracer gas and equipment dead space volume did not significantly affect the prediction equations for either LCI or FRC. Reference equations for LCI include age as the only predictor, whereas sex-specific reference equations for FRC included height and age.

Conclusions Global Lung Function Initiative reference equations for multiple breath washout variables provide a standard for reporting and interpretation of LCI and FRC.

Introduction

Standard pulmonary function tests measure a range of indices that inform clinicians about different aspects of lung physiology [1]. Interpretation of pulmonary function tests requires an understanding of the range of

values that can be expected in otherwise healthy individuals. The Global Lung Function Initiative (GLI) Network has established a protocol to collate previously collected healthy pulmonary function data and to derive a single standard for the interpretation of measurements against a healthy population [2]. All-age GLI reference equations are now available for spirometry, static lung volumes and gas transfer [3–5].

Multiple breath washout (MBW) is a lung function test based on efficiency of washout of an inert tracer gas from the lungs during tidal breathing. It assesses the unevenness of gas mixing within the lung, also known as ventilation inhomogeneity. This is expressed by a number of MBW variables, the most commonly reported of which is the lung clearance index (LCI). A substantial body of literature on LCI, predominantly derived over the past two decades, has consistently shown it to be more sensitive at detecting early obstructive lung disease than spirometry variables in children with cystic fibrosis (CF) [6–8]. MBW requires less active patient cooperation than spirometry and can be reliably measured in young children and infants, including unsedated infants [9–11]. LCI is now being routinely measured in some CF centres to assist clinical decision-making and has been identified in CF guidelines as a clinical outcome of interest [12]. Results obtained for LCI in other obstructive disease conditions in research studies suggest that it may also have future important applications in asthma [13], bronchiectasis [14, 15], primary ciliary dyskinesia [16, 17], early COPD [18], lung disease following haematopoietic stem cell transplantation [19], occupational lung disease [20, 21] and lung transplantation [22, 23].

The other index consistently generated from multiple breath washout is functional residual capacity (FRC), a measure of resting end-expiratory lung volume. This also provides a quality control check to assess the technical acceptability of the trials and overall test session [24]. Over time, the focus of MBW testing has evolved from FRC being the main index of interest, to one focused on LCI for research and clinical use. Though infrequently used on its own in clinical practice, FRC may provide complimentary information about gas trapping (due to peripheral airways disease) [25] and can also be used to assess response to therapies [26].

Technical standards for MBW equipment and how to perform MBW testing, along with frameworks for training operators, and applying quality control have been published [24, 27–30]. Commercially available devices have evolved significantly over the past decade and have incorporated several advances with improved accuracy and precision of measurements. Differing inert tracer gas options exist, with differing intrinsic properties, such as nitrogen (N_2) and sulfur hexafluoride, which may impact on gas mixing and excretion [31, 32]. Importantly, the methodological approach chosen for indirect N_2 concentration measurement has also been shown to directly influence results [33–35].

Differences in equipment, tracer gases and algorithms to calculate LCI have limited previous attempts to collate healthy reference equations for LCI and FRC derived from MBW [18, 36, 37]. Consequently, and in line with consensus recommendations [24], individual centres have developed their own normal ranges for LCI and FRC. This often restricts individual datasets to narrow age ranges, single devices and limited sample sizes [38–42]. Centre- and device-specific reference equations make it challenging to interpret measurements collected elsewhere, or to understand how much of the differences are due to sampling variability and technique compared to fundamental differences between sites, equipment and protocols. The lack of all-ages data also makes it difficult to accurately interpret how ventilation inhomogeneity changes with growth and ageing.

A single standard for the interpretation of MBW indices across all ages is an important resource for the use of MBW more widely in research and clinical practice. The aim of this study was to collate MBW data from healthy individuals and to derive GLI reference equations for LCI and FRC across all ages. In addition, we aimed to investigate how differences in equipment, tracer gas and equipment dead space volume impacted outcomes.

Methods

A European Respiratory Society (ERS)-approved task force was formed in May 2020 to develop all-age reference equations for LCI and FRC measured by MBW. The task force comprised researchers and healthcare professionals with expertise in developing international standards, lung physiology, pulmonary function testing and biostatistics.

Data sources

A pragmatic review of the literature was conducted to identify published studies that included measurement of LCI and FRC in healthy children and adults. Task force members searched for articles published between 2005 and 2023 in Embase, Ovid MEDLINE, Web of Science, PubMed and Scopus databases.

Publications were then screened to remove duplicates, articles without MBW data, articles without healthy individuals and reviews/editorials without original data (details provided in the supplementary material). The authors of studies with ⩾50 healthy participants were contacted and invited to share their data with the task force (this restriction was applied to ensure that data were collected only from centres with significant experience conducting LCI assessments). Invitations were also circulated through international and local respiratory societies (American Thoracic Society (ATS), Asia Pacific Society of Respirology, ERS, Latin American Thoracic Association, Pan African Thoracic Society, Thoracic Society of Australia and New Zealand) to solicit unpublished data.

Contributors were required to obtain approval from the local ethics committee to contribute data to the GLI Network. All data were pseudo-anonymised before submission and entered to a standard data template. Study data were collected and managed using Research Electronic Data Capture (REDCap) hosted at Dalhousie University (Halifax, NS, Canada) [43, 44].

Data were collated for LCI and FRC as well as demographic variables: sex, age, height and weight. The mean LCI and FRC from all acceptable trials were included in the analysis from test occasions with a minimum of two acceptable trials. If a centre contributed more than one data point per individual participant, one measurement only was randomly selected and included. Metadata included equipment type (and any modification of commercial equipment), tracer gas and equipment dead space volume. Due to the published impact of different software versions on MBW outcomes [33, 37], we only accepted data analysed using specific software versions (Eco Medics Exhalyzer D: Spiroware 3.3.1; ndd EasyOne Pro: Easy One Pro Lab/Easy One Connect V03.08.00.11 or higher until the date of data submission) [45, 46]. Sites were given detailed instructions and supported to reanalyse data from previously collected washout tests to maximise the inclusion of available data. Quality control assessments were performed by the individual sites according to international guidelines [24, 27, 28, 30, 47]. All submitted data were reviewed to identify missing data and implausible outliers; contributors were contacted directly to clarify discrepancies.

Healthy participants were defined as a person who was not a current smoker, had never smoked and was not obese. Participants aged <12 years with missing cigarette smoking status were assumed to be never-smokers. Incorporating any missing observations on smoking status in those aged >12 years, we created two definitions of healthy: a healthy participant was defined as a person with no reported currently smoking (not smoking now) and no reported smoking in the past (has not smoked >100 cigarettes). This definition includes individuals with a missing currently smoking or ever-smoked status (*i.e.* they were assumed to be healthy). A second definition "strictly healthy" was defined as an individual who was reported to be a never-smoker. This definition excluded participants who had missing data with respect to currently smoking or ever-smoked status (*i.e.* no assumptions were made on this subset). Body mass index (BMI) measurements were used to categorise subjects as overweight or obese. The Centers for Disease Control and Prevention growth charts were used for children aged 2–18 years, with overweight defined as BMI >85th percentile and obesity defined as BMI >95th percentile. For adults, we used measures of BMI >25 $kg \cdot m^{-2}$ to categorise an individual as overweight and those with a BMI >30 $kg \cdot m^{-2}$ to define obesity.

Statistical analysis

The generalised additive models of location shape and scale (GAMLSS) programme, previously used for other GLI projects, was used to develop the reference equations of LCI and FRC measured by multiple breath washout to 2.5% of starting inert tracer end-tidal concentration. Briefly, the GAMLSS technique allows the median value to be summarised as a function of multiple explanatory variables (*e.g.* height, age, sex), the spread of values around the median value to be constant or vary by a function of explanatory variables, and any departure from a normal distribution (skewness, kurtosis) to be transformed to normal using a Box–Cox transformation. Thus, the resulting model residuals are normally distributed. The BCCG (or Box–Cox Cole and Green family) distribution and a log transformation of the response variable was applied. All analyses were performed using the GAMLSS package in the statistical programme R (The R Project for Statistical Computing; www.r-project.org, version 4.3.1).

Age, sex, height, weight and total equipment dead space volume were treated as explanatory variables and were evaluated independently and in a multivariable model. Total equipment dead space constitutes both the pre-capillary dead space (volume between the airway opening and inert gas sampling point) and post-capillary dead space (volume between the gas sampling point and bias flow). These variables have previously been shown to influence LCI and/or FRC. We did not investigate ethnicity as a predictor as race and ethnicity are social constructs without a consistent definition globally, and recent statements endorsed by both the ATS and ERS have recommended against its continued use in reference equations [48, 49]. Variables that improved overall model fit were retained in the final model. The goodness of fit of each

model was assessed by Schwarz Information Criteria (SBC), residual Q–Q plots and residual plots. Differences in LCI and FRC between equipment types and sites were investigated after the final models were derived. The upper and lower limits of normal for LCI and FRC from the final equations were compared to previously published equations. Sensitivity analyses were performed to determine the impact of the definitions of healthy and strictly healthy.

Results

Study population

Data from 3647 individuals were submitted by 23 studies from nine countries (figure 1). Since infant MBW is particularly challenging, and it became clear early in the analysis that approaches to infant MBW varied widely (*e.g.* positioning of infant, technological approaches, sedated *versus* unsedated), infants (defined as those aged <2 years) were excluded from analysis. After exclusions of ineligible studies, data from 1579 healthy individuals aged between 2 and 81 years from 17 studies and eight countries were available (table 1, figure 2). A large number of observations (n=474, 30%) were from children aged between 5 and 10 years (figure 2a).

Reference equations

There was a nonlinear relationship between predicted LCI and age, and age also contributed to the between-participant variability (figure 3). In other words, LCI was on average higher in younger children and older adults, but relatively stable during childhood and early adulthood. Furthermore, there was more variability in LCI in younger children and older adults, resulting in wider limits of normal. FRC reference equations included age and height as predictors, as well as sex-specific equations. The coefficients for each prediction equation are presented in table 2. Look-up tables and a worked example are available in the supplementary material.

Limits of normal and comparison with existing reference values

The upper limit of normal for LCI (as both +1.64 and +1.96 z-scores) using the derived equations and previously published equations is presented in table 3. The upper and lower limits of normal for FRC

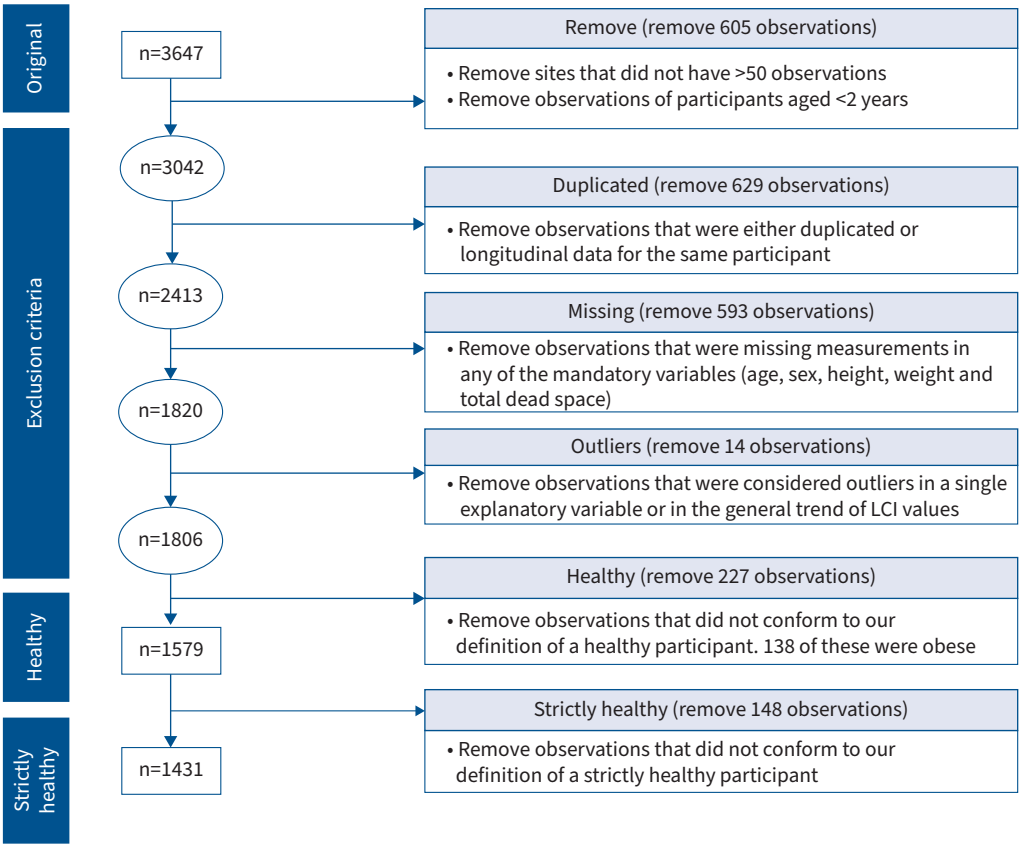

FIGURE 1 Flow diagram of exclusions used to reach the final datasets.

TABLE 1 Summary of healthy participants by site

Site	Participants n	Age years	Female	Overweight[#]	LCI	FRC L	Country	Equipment	Tracer gas	Was the equipment modified?	Breathing pattern
5	140	5–59	50.7	12.1	6.1 (5.8–6.4)	1.9 (1.4–2.7)	United Kingdom	Innocor closed circuit	SF_6 up to 0.4%	Yes	Tidal breathing
10	22	5–16	50.0	4.6	6.4 (6.3–6.6)	1.7 (1.1–2.2)	United Kingdom	Innocor open circuit	SF_6 up to 0.4%	Yes	Tidal breathing
12	27	2–11	55.6	0.0	6.6 (6.2–6.9)	0.6 (0.5–0.8)	United Kingdom	Mass-spectrometer	SF_6 4%	NA	Tidal breathing
13	73	20–77	50.7	32.9	5.9 (5.5–6.3)	3.4 (2.8–4.1)	United States	Eco Medics Exhalyzer D	N_2 (washout 100% oxygen)	No	Tidal breathing
15	75	3–67	52.0	10.7	6.5 (6.2–6.8)	2.0 (1.2–3.2)	Germany	Eco Medics Exhalyzer D	N_2 (washout 100% oxygen)	No	Tidal breathing
19	73	19–65	60.3	32.9	6.3 (6.0–6.7)	2.7 (2.2–3.2)	Australia	Eco Medics Exhalyzer D	N_2 (washout 100% oxygen)	No	Fixed breathing protocol
20	54	5–59	44.4	5.6	6.2 (5.9–6.5)	2.7 (1.7–3.3)	Belgium	Eco Medics Exhalyzer D	N_2 (washout 100% oxygen)	No	Tidal breathing
20	41	5–32	61.0	0.0	6.3 (5.9–6.7)	1.7 (1.1–2.3)	Belgium	Ndd Easy One Pro	N_2 (washout 100% oxygen)	No	Tidal breathing
24	275	5–20	49.5	1.5	6.2 (5.9–6.5)	1.0 (0.8–2.4)	Switzerland	Eco Medics Exhalyzer D	N_2 (washout 100% oxygen)	No	Tidal breathing
26	153	3–66	59.5	13.7	6.3 (6.0–6.6)	2.0 (1.0–2.7)	France	Eco Medics Exhalyzer D	N_2 (washout 100% oxygen)	No	Tidal breathing
33	238	20–81	50.0	28.6	6.4 (6.0–6.8)	3.2 (2.7–3.8)	Belgium	N_2 analyzer	N_2 (washout 100% oxygen)	No	Fixed breathing protocol
34	37	21–77	67.6	35.1	6.3 (6.0–7.2)	3.2 (2.6–3.5)	United Kingdom	Eco Medics Exhalyzer D	N_2 (washout 100% oxygen)	No	Tidal breathing
36	44	6–44	50.0	11.4	6.2 (6.0–6.6)	1.6 (1.2–2.8)	United Kingdom	Innocor open circuit	SF_6 up to 0.4%	Yes	Tidal breathing
37	95	6–59	71.6	20.0	6.4 (6.2–6.7)	2.8 (2.5–3)	Australia	Eco Medics Exhalyzer D	N_2 (washout 100% oxygen)	Yes	Tidal breathing
42	34	5–63	52.9	17.6	6.4 (6.0–6.9)	2.0 (1.5–2.7)	United Kingdom	Innocor open circuit	SF_6 up to 0.4%	No	Tidal breathing
47	41	5–74	65.9	14.6	6.4 (6.2–6.9)	3.0 (2.4–3.5)	Germany	Eco Medics Exhalyzer D	N_2 (washout 100% oxygen)	No	Tidal breathing
52	62	3–11	58.1	0.0	6.4 (6.1–6.7)	1.0 (0.8–1.2)	Australia	Eco Medics Exhalyzer D	N_2 (washout 100% oxygen)	No	Tidal breathing
59	95	6–7	47.4	0.0	6.8 (6.5–7.2)	0.6 (0.6–0.7)	South Africa	Eco Medics Exhalyzer D	N_2 (washout 100% oxygen)	No	Tidal breathing

Data are presented as range, % or median (interquartile range), unless otherwise stated. Each row represents a different site or measurement system. If the same site contributed data from studies with different equipment or protocols these were reported as separate sites. If the same participant was measured using two different protocols/equipment, only one measurement from each participant was randomly included. In some sites, there were <50 observations after data exclusions were applied, but all sites had to contribute data for a minimum of 50 participants. LCI: lung clearance index; FRC: functional residual capacity; SF_6: sulfur hexafluoride; NA: not applicable; N_2: nitrogen. [#]: based on a body mass index >85th percentile in children aged 2–18 years and >25 $kg \cdot m^{-2}$ in adults.

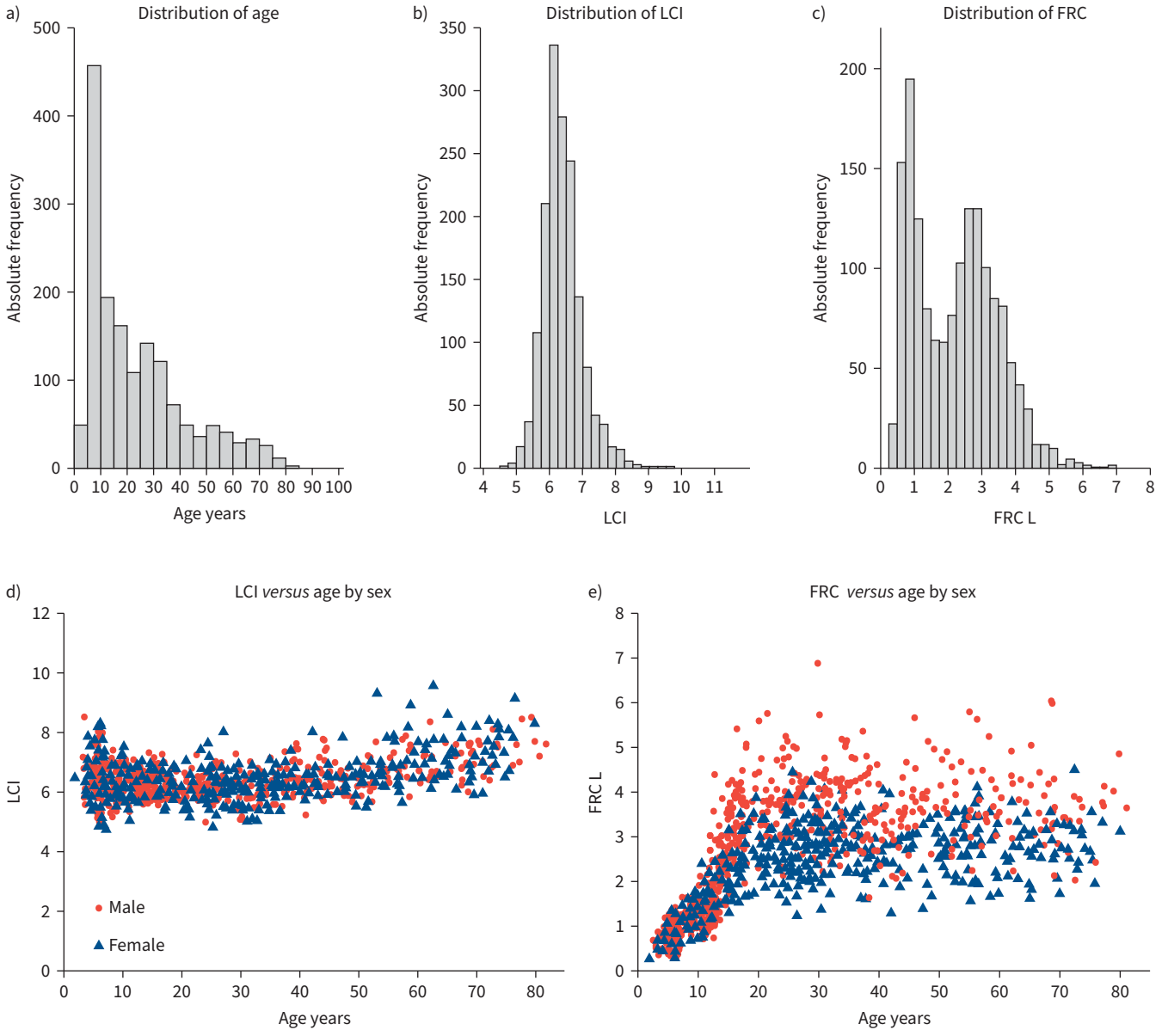

FIGURE 2 a) Age distribution of individuals considered to be healthy; b) histogram showing lung clearance index (LCI) measurements of healthy individuals; c) histogram showing functional residual capacity (FRC) measurements of healthy individuals; d) scatter plot of LCI against age stratified by sex of healthy individuals; e) scatter plot of FRC against age stratified by sex of healthy individuals.

(+1.64 and +1.96 z-scores) are presented in supplementary tables S1 and S2. The GLI equations were generally similar to previously published equations for FRC and LCI. The upper limits of normal for LCI from the derived GLI equations were higher in children aged <10 years compared to existing static reference values. The GLI upper limits of normal for FRC were lower in females, and higher in adult males compared to existing reference values. In the final models there remained small residual differences between sites and equipment types (figure 4).

Sensitivity analyses

We performed sensitivity analyses to determine the impact of including tracer gas or equipment device in the model. There were no significant discrepancies between the final model and the sensitivity models adjusted for either tracer gas (supplementary figure S1) or equipment (supplementary figure S2). These sensitivity analyses were limited by the fact that not all tracer gases or equipment devices spanned the entire age range. Based on this information, we did not include these variables in the final model.

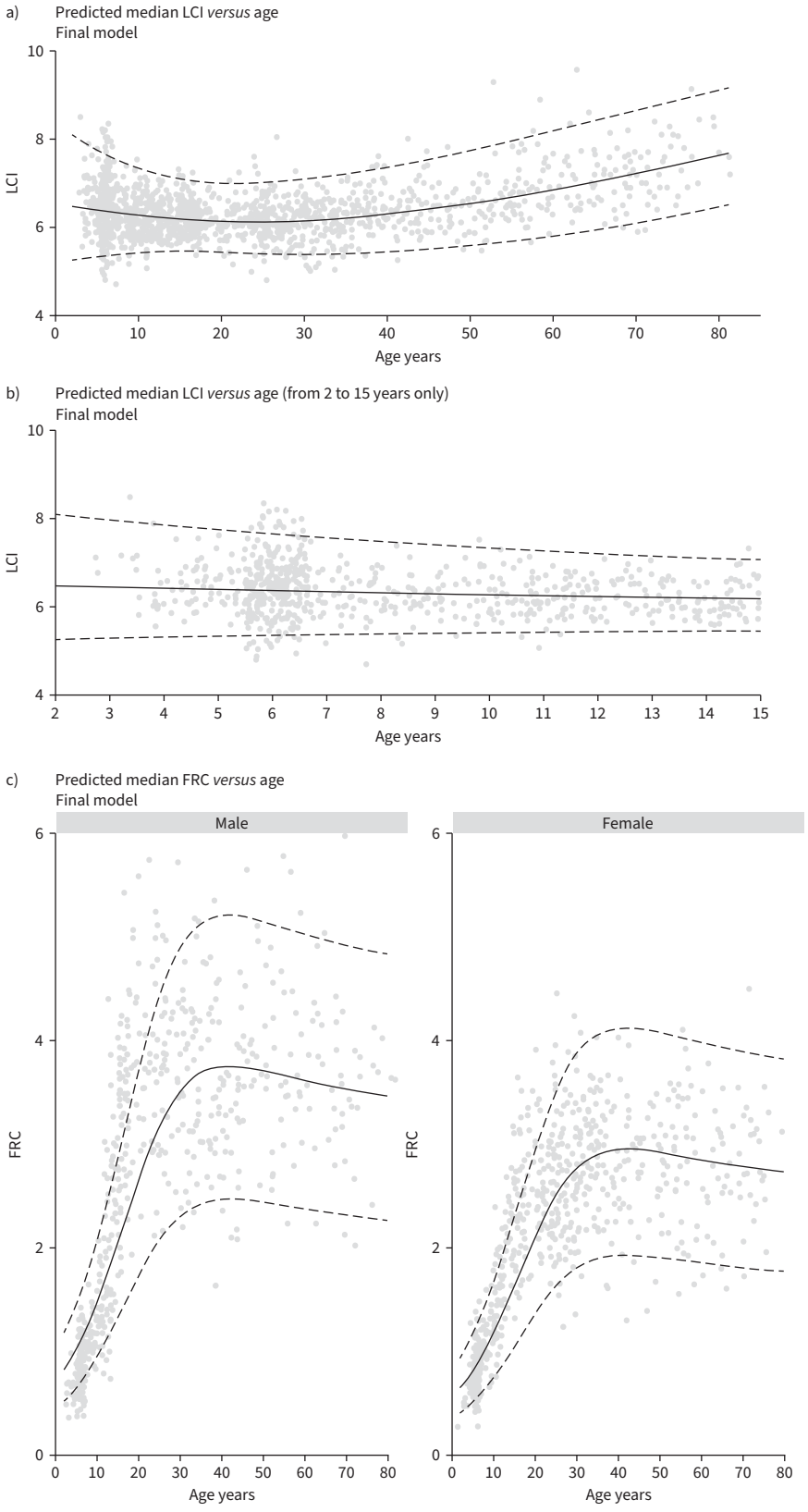

FIGURE 3 a) Predicted lung clearance index (LCI); b) graph in a) expanded to show only ages from 2 to 15 years; c) predicted functional residual capacity (FRC). Data are presented as the predicted values for age and 95% confidence limits. Predicted equations are overlaid on observed values.

TABLE 2 Coefficients and model fit for the Global Lung Function Initiative multiple breath washout equations

LCI	M (median)	$\exp\{1.817 + 0.001308 \cdot age + Mspline\}$
	S (variability around the median)	$\exp\{-2.540 - 0.0002242 \cdot age + Sspline\}$
	L (skewness)	−0.3964
FRC male	M (median)	$\exp\{-2.468 + 0.006038 \cdot age + 0.01957 \cdot height + Mspline\}$
	S (variability around the median)	$\exp\{-1.392 - 0.001590 \cdot height\}$
	L (skewness)	0.6471
FRC female	M (median)	$\exp\{-2.512 + 0.006038 \cdot age + 0.01957 \cdot height + Mspline\}$
	S (variability around the median)	$\exp\{-1.392 - 0.001590 \cdot height\}$
	L (skewness)	0.6471

Age is in years and height in centimetres. LCI: lung clearance index; FRC: functional residual capacity.

The GLI repository is set up to collect lung function data in healthy individuals. The assumption is that any submitted data are from healthy, nonsmoking individuals. Some individuals had missing smoking status which required us to create two definitions of healthy: healthy (assumed to be nonsmoker) and strictly healthy (reported nonsmoker) (supplementary figure S3). The mean difference in z-scores calculated using both definitions of nonsmoker was −0.052 (95% CI −0.056–−0.048) for LCI z-scores and 0.0042 (95% CI 0.0028–0.0055) for FRC z-scores. The lack of differences between the two definitions of smoking status allowed us to include those with missing data.

We performed a sensitivity analysis to test the impact of including obese individuals in our analysis (supplementary figure S4). The number of obese individuals was only 138, which represents 8.7% of the total population. The mean difference in z-scores calculated between the final model and model with obese individuals was 0.0007 (95% CI −0.002–0.0004) for LCI z-scores and 0.0018 (95% CI 0.00285–0.00545) for FRC z-scores. Due to the low sample of obese individuals, we did not feel that we could confidently conclude that obesity had no impact on LCI and FRC, and therefore made the decision to exclude obese individuals from the final model.

Discussion

We report the largest collation of MBW data from healthy participants across a wide age range, and a single reference equation for each of LCI and FRC. Data from healthy individuals collected using different inert tracer gases and a wide range of equipment showed clear overlap. Therefore, the reference equations derived from combined data are applicable across currently used protocols and equipment.

Methodological differences in measurement technique, tracer gases and equipment have previously limited efforts to collate healthy MBW data and made it challenging to agree on a consensus of what healthy LCI

TABLE 3 Upper limit of normal for lung clearance index (LCI) using the derived equations and previously published equations

Age years	GLI MBW			Horsley 2020 (Innocor closed circuit) ULN (1.64 z-scores) [38]		Verbanck 2016 (N_2 analyser) ULN (1.64 z-scores) [42]		Kentgens 2022 (Exhalyzer D) ULN (1.96 z-scores) [41]	
	Predicted	ULN (1.64 z-scores)	ULN (1.96 z-scores)	Predicted	ULN (1.64 z-scores)	Predicted	ULN (1.64 z-scores)	Predicted	ULN (1.96 z-scores)
3	6.5	7.7	8.0						
5	6.4	7.5	7.8	6.1	6.8				
7	6.4	7.4	7.6	6.1	6.8			6.3	7.1
10	6.3	7.2	7.3	6.1	6.8			6.3	7.1
15	6.2	6.9	7.1	6.1	6.8			6.3	7.1
20	6.1	6.9	7.0	6.1	6.8	6.1	6.7		
25	6.1	6.9	7.0	6.1	6.8	6.1	6.7		
30	6.1	6.9	7.1	6.1	6.8	6.1	6.7		
40	6.3	7.2	7.4			6.2	6.9		
50	6.5	7.5	7.7			6.4	7.2		
60	6.9	8.0	8.2			6.7	7.7		

GLI: Global Lung Function Initiative; ULN: upper limit of normal; N_2: nitrogen.

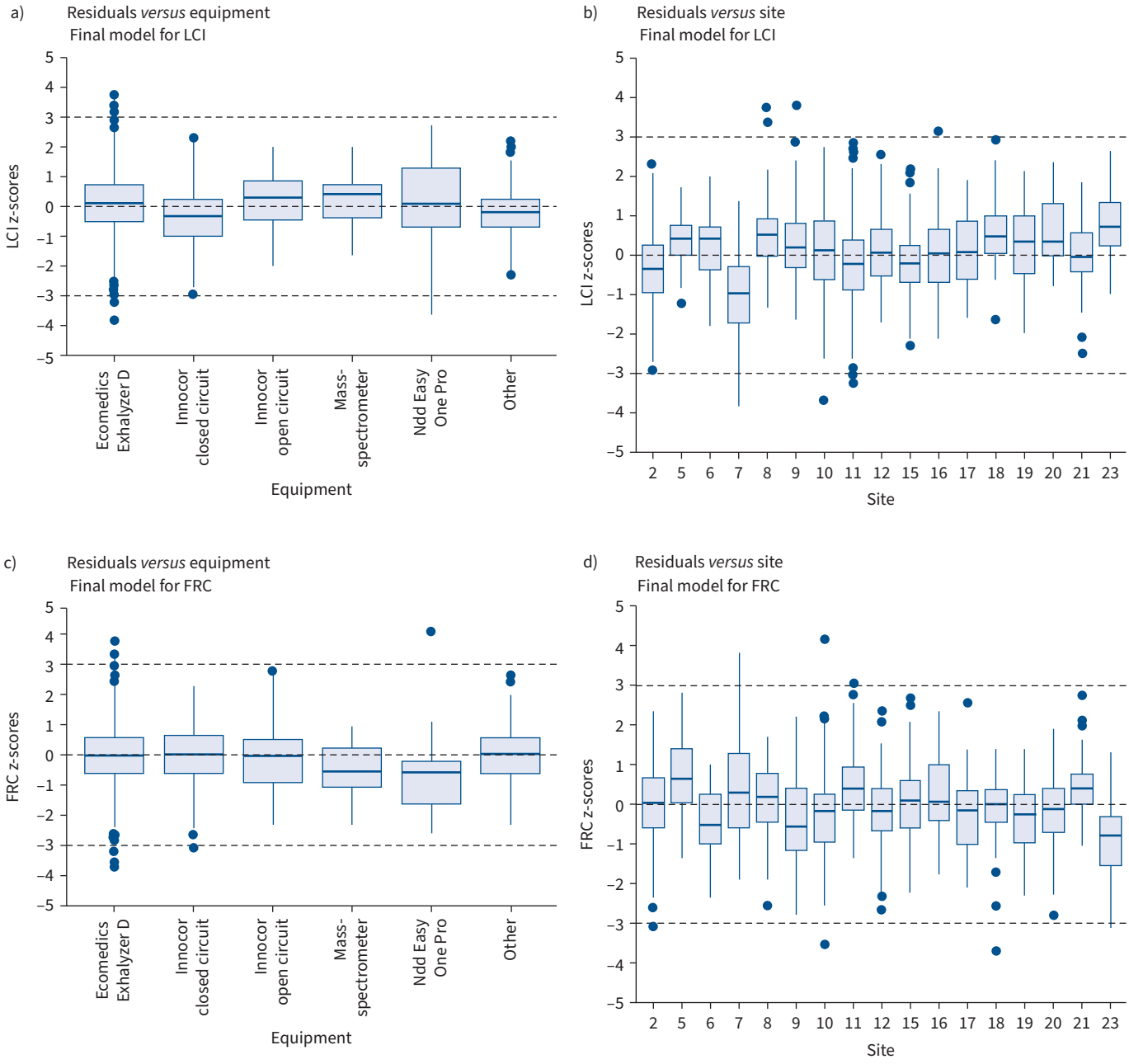

FIGURE 4 Box-and-whisker plots for a) lung clearance index (LCI) z-scores (residuals) by the different equipment in the final model; b) LCI z-scores (residuals) by the different sites in the final model; c) functional residual capacity (FRC) z-scores (residuals) by the different equipment in the final model; and d) FRC z-scores (residuals) by the different sites in the final model.

should look like [18, 36, 37]. Here we observed smaller equipment differences than previously shown between equipment type (including inert tracer gas and equipment dead space volume). Importantly, these different systems incorporated different tracer gases, different measurement techniques (including fixed volume breaths and free tidal breathing), and devices with in-house modifications. Despite this heterogeneity of approach there was good overlap in MBW indices between devices, adding confidence that our LCI equations are robust. All data collected using the Exhalyzer D device were reanalysed using a revised software version (Spiroware 3.3.1) prior to submission [46]. This version of the software has improved cross-sensitivity corrections for the oxygen and carbon dioxide sensors used in the Exhalyzer D system, which resulted in significant reductions in LCI and FRC compared to previous software versions [33]. The minimal equipment differences do not mean that different equipment devices and tracer gases

should be used interchangeably in the same individual over time, but rather that the variability of data from healthy individuals is not sufficiently explained by these variables.

The normal ranges presented here provide, for the first time, a consensus indication of where normal LCI should lie in healthy individuals. Different groups have previously used different thresholds to identify the upper limit of normal LCI, setting the level as either the top 2.5% (1.96 z-scores) or the top 5% (1.64 z-scores) of healthy LCI. In ordinary clinical practice, LCI is unlikely to be used on its own to diagnose a condition, and more likely to be used in longitudinal monitoring for those with a known diagnosis. In this context, the task force recommends using the cut-off that excludes the top 5% of healthy values (*i.e.* 1.64 z-scores), as highlighted in table 3. In different contexts, for example if LCI were to be used to diagnose illness in a population with low prior likelihood of lung disease, then a higher ULN might be appropriate (also provided in table 3 for comparison with previous normal ranges).

Reference equations for LCI were related to age, but not sex or height. LCI in healthy individuals was relatively age-independent between the ages of 10 and 25 years. In addition, there was greater variability in the residuals in the younger and older age ranges. These observations are consistent with smaller studies of narrower age ranges [38–41]. Technical and physiological factors may have influenced the relationship between LCI and age, and the distribution of the residuals with age. Equipment dead space has the potential to overestimate LCI in young children who have small tidal volumes, and differences in equipment, interface, measurement protocol and breathing pattern between sites covering different age ranges may also influence results [40, 41, 50–52].

Reference equations for FRC were sex-, age- and height-dependent. There were small deviations between the derived FRC equations and existing reference equations [3, 39, 42, 53]. The greatest deviations occurred in adult males aged >30 years, whereby the upper limits of normal for FRC from the GLI MBW equations were higher than the GLI static lung volume equations (which included FRC data from nitrogen washout, helium dilution and plethysmography). This could be explained by the greater variability in the residuals in the older age range in our model. However, overall, these differences were relatively small and provide validation that the FRC measurements are accurate. Choice of reference equation depends on the population under study and the purpose of the measurement. When measuring LCI, FRC is generally used to check that washout outcomes appear to be reasonable, and FRC z-scores or percentage predicted are rarely used to determine treatment. However, where FRC is the primary measurement of interest, GLI lung volumes equations are the recommended reference values.

Limitations

MBW remains a specialised pulmonary function test limited to experienced laboratories, and consequently the number of contributing sites and total sample size was small. All contributing sites, except one site, were from high-income countries with predominantly white European populations. The final models demonstrated residual variability in both LCI and FRC between sites. It is unclear whether this is due to differences in protocols and equipment *versus* population differences in lung health. Further work will be needed to understand whether differences in LCI between sites could also reflect the sensitivity of LCI to population level differences in respiratory exposures such as lower respiratory tract infections and air quality [54].

There are inevitable gaps in our data collection. The task force made multiple attempts to contact all authors with published MBW data, and the data collection phase was extended by 18 months. Furthermore, differences in data governance laws and interpretation of the definition of pseudo-anonymised data between jurisdictions prohibited some sites from contributing data. An important learning point for researchers is that provision to share data needs to be included in patient consent forms at the outset. We focused our analysis on FRC and LCI. Other MBW parameters exist, such as moment ratios and phase 3 slope analysis end-points, but data collection for these parameters was incomplete, analysis methods are nonstandardised, and their clinical application less robust. Additionally, different equipment systems were not equally represented in our dataset.

We were unable to include MBW data collected in healthy infants aged <2 years. Infants present unique technical challenges which resulted in wide discrepancies in data collected within and between sites. There were also limited data available on individuals aged >50 years.

The operational definition of healthy was robust to sensitivity analysis. It was possible to confirm the smoking exposure status for most subjects, and when this was not available, making the assumption that sites contributed data from healthy individuals did not influence the final results. We did not have a

sufficient sample size to determine the impact of obesity on reference equations for FRC or LCI in this study.

Implementation

The prediction equations and look-up tables and online calculator (www.lungfunction.org) are provided and a worked example is provided in the supplementary material. We have contacted manufacturers of MBW systems to help them incorporate these latest equations as an option in their devices. Changing to global reference equations will support implementation of LCI into clinical practice.

Conclusions

We have generated all-ages GLI reference equations for LCI and FRC that are independent of equipment type, inert tracer gas and measurement method. Our findings support the wide applicability of these equations to guide interpretation.

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