Multiple-breath washout to detect lung disease in patients with inborn errors of immunity

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Abstract

Background Pulmonary manifestations are the major cause of morbidity and mortality in patients with inborn errors of immunity (IEI). New and more sensitive diagnostic methods can potentially lead to earlier recognition and treatment of IEI lung disease and improve outcome. The aim of this study was to compare multiple-breath washout (MBW) and spirometry in patients with IEI and cystic fibrosis (CF) as well as healthy controls (HC) and to evaluate the sensitivity of lung clearance index (LCI) to assess lung disease in IEI.

Methods IEI patients (n=114) were recruited from our paediatric and adult immunodeficiency outpatient clinics and compared to age-matched CF patients (n=114) and HC (n=114). MBW measurements and spirometry were performed in the study participants, and MBW testing was repeated after 63–707 days in IEI patients (n=70).

Results The LCI was significantly higher in IEI patients than in HC (p<0.001) and significantly lower than in CF patients (p<0.001). The forced expiratory volume in 1 s (FEV$_1$) z-score was significantly lower in IEI patients than in HC (p<0.01) and significantly higher than in CF patients (p<0.01). LCI and FEV$_1$ z-score correlated moderately negatively in the total cohort, the IEI group and the CF group. Nineteen (20.7%) of 92 IEI patients and 35 (33.3%) of 105 CF patients had an elevated LCI but a normal FEV$_1$ z-score. After a median of 364 days, the median LCI of 70 IEI patients increased significantly by 0.2.

Conclusion MBW is useful to detect lung disease in IEI and is more sensitive than spirometry.

Introduction

Pulmonary symptoms and complications are not only a frequent clinical manifestation of inborn errors of immunity (IEI), but also the major cause of morbidity and mortality in this patient group [1–3]. Despite improved treatment options for defects of the immune system including immunoglobulin replacement therapy, respiratory tract infections continue to occur, leading to chronic structural lung diseases [1, 4, 5]. In Europe, immunodeficiency is the fifth most frequent identified cause of non-cystic fibrosis (CF) bronchiectasis occurring in 4.1% of bronchiectasis patients [6]. Between 20% and >60% of patients with common variable immunodeficiency (CVID) develop bronchiectasis [7, 8]. Furthermore, non-infectious pulmonary complications caused by immune dysregulation, such as granulomatous-lymphocytic interstitial lung disease (GLILD), negatively impact the prognosis of IEI patients [1, 3]. GLILD was shown to negatively impact the outcome and to shorten the survival of patients with CVID [9, 10]. Additionally, IEI

patients have an increased risk to develop pulmonary lymphoma [11, 12]. Since the early recognition and treatment of respiratory manifestations in IEI patients can improve their outcome, sensitive diagnostic methods are urgently needed [1, 3]. At present, spirometry is commonly used for the assessment of lung function. Furthermore, in patients with antibody deficiencies, an annual body plethysmography with measurement of the diffusing capacity of the lung for carbon monoxide (D_{LCO}) is recommended, and D_{LCO} was shown to be sensitive to detect GLILD [3, 9, 13]. However, it was demonstrated that spirometry as well as body plethysmography and D_{LCO} do not detect mild structural lung disease in patients with IEI [8].

A promising method for the detection of early lung disease is the multiple-breath washout (MBW) test with the lung clearance index (LCI) as the most reported outcome parameter reflecting global ventilation inhomogeneities, gas mixing and physiological dead space [14]. MBW is performed noninvasively and has shown its feasibility in clinical settings even in early childhood in patients with chronic lung diseases [15–17]. In adult and paediatric patients with CF, studies demonstrated that LCI correlates with other parameters of pulmonary function but is more sensitive to detecting early lung disease than spirometry [18, 19]. In addition, MBW can detect progression of CF lung disease [20, 21]. Further, previous studies in CF patients demonstrated a good correlation of LCI with findings in chest magnetic resonance imaging (MRI) and computed tomography (CT) and that structural lung disease such as bronchiectasis is more readily detected by MBW than by spirometry [22–24]. Furthermore, CF patients with bacterial lung infections seem more likely to have an abnormal LCI than those without, and LCI detects treatment effects even in patients with mild CF lung disease [19, 25–27]. While the high sensitivity of LCI to detect lung disease has also been confirmed in patients with primary ciliary dyskinesia (PCD) and COPD, there is little data available on MBW for detection of different features of lung disease in IEI patients [15, 28–30].

Therefore, the aim of this study was to evaluate the ability of LCI to assess lung disease in comparison to spirometry in children and adults with IEI, CF patients and healthy controls (HC). Additionally, we evaluated longitudinal MBW measurements in IEI patients to assess progression of lung disease.

Methods

Study population and design

We performed a monocentric observational study over 2 years approved by the Ethics Committee of the Charité – Universitätsmedizin Berlin (EA2/003/21). Informed written consent was obtained from all participants and/or their parents or legal guardians. Patients with IEI attending the paediatric or adult outpatient immunodeficiency clinic were eligible for this study if they were able to perform a MBW measurement. Eight patients unable to perform MBW successfully were excluded from this study. Of those, three were below the age of 5 years, one was older than 60 years, two had ataxia telangiectasia and two had DiGeorge syndrome. Study visits took place during routine visits in clinically stable patients. IEI patients were classified according to the International Union of Immunological Societies Classification for Human IEI [31]. In addition, equal-sized and age-matched groups of CF patients as disease control as well as HC were recruited from other ongoing observational studies at our centre. An active lung disease and the use of respiratory medications were exclusion criteria for HC. Medical history and demographics were collected for all participants. Information on lung imaging in the previous 5 years, history of respiratory infections, previous lung surgery, diagnosis of bronchial asthma and lung-specific therapies were collected for IEI and CF patients. Data on previous allogeneic haematopoietic stem cell transplantation (allo-HSCT) and immunoglobulin replacement therapy were collected for IEI patients. For CF patients, data on CF transmembrane conductance regulator (CFTR) mutations, pancreatic insufficiency, chronic *Pseudomonas aeruginosa* infection according to the Leeds criteria [32] and the use of CFTR modulator therapy were collected. Bronchiectasis was confirmed by CT or MRI and defined as an increased airway diameter with an increased broncho-arterial ratio of >1–1.5 [33].

Lung function testing

MBW was performed in all and spirometry in study participants above 5 years of age at the initial study visit. In IEI patients, MBW measurements were repeated at subsequent outpatient visits after 3 to 24 months. MBW testing was performed with Exhalyzer D and (re-)analysed using spiroware version 3.3.1 (both Eco Medics, Duernten, Switzerland). 100% oxygen was used to wash out resident nitrogen (N_2) from the lungs as previously described [22, 34]. Measurements were performed in awake, upright sitting subjects using a mouthpiece as interface while wearing a nose clip. At least two acceptable trials were necessary [34]. In HC, LCI increased from the age of 50 years. Therefore, upper limits of normal (ULN) for LCI were determined separately for patients <50 and ⩾50 years of age. ULN were calculated as mean+1.96 × SD of the respective HC subgroup. Spirometry was performed according to the criteria of the American Thoracic Society and European Respiratory Society [34]. Forced expiratory volume in 1 s (FEV_1) was used as the primary spirometric outcome parameter and values were converted to z-scores

using the Global Lung Function Initiative reference equations [35]. A z-score <-1.96 was classified as abnormal for FEV_1.

Statistical analysis

SPSS Version 27 (SPSS Inc., Chicago, IL, USA) was used for the statistical analyses. Data are represented as n (%) or median (interquartile range (IQR)), if not indicated otherwise. Normal distribution of data was assessed by the Shapiro–Wilk test. The Mann–Whitney U-test or Wilcoxon signed rank test were used for comparison of continuous data and chi-square test or Fisher exact test for comparison of categorical data, as appropriate. Correlation analyses were performed by using the Spearman's rank correlation (r_s). A p-value <0.05 was considered statistically significant. In the case of multiple comparisons, individual p levels are indicated only if not rejected by Bonferroni correction.

Results

Clinical characteristics of the study population

In total, 114 HC with a median (range) age of 24.4 (3.7–73.8) years, 114 IEI patients with a median (range) age of 20.7 (3.8–76.9) years and 114 CF patients with a median (range) age of 20.4 (3.9–72.7) years were included in this study (table 1). More CF patients than IEI patients had a history of recurrent

TABLE 1 Characteristics of the study population			
	HC	IEI	CF
Participants n	114[#]	114	114
Age years	24.4 (3.7–73.8)	20.7 (3.8–76.9)	20.4 (3.9–72.7)
Patients ≥18 years	71 (62.3)	68 (59.6)	69 (60.5)
Female	67 (58.8)	39 (34.2)[¶]	44 (38.6)
BMI kg·m^{-2} for patients ≥18 years	23.6 (21.2–27.7)	23.6 (20.8–27.4)	22.2 (20.1–25.0)
BMI z-score for patients <18 years	0.2 (−0.5–1.0)	0.1 (−0.9–1.2)	−0.3 (−0.9–0.3)
MBW performed	114 (100.0)	114 (100.0)	114 (100.0)
Spirometry performed	111 (97.4)	92 (80.7)	105 (92.1)
Lung imaging (CT/MRI) performed	-	32 (28.1)	114 (100.0)
With signs of bronchiectasis	-	18 (15.8)	89 (78.1)
With signs of interstitial lung disease	-	4 (3.5)	0 (0.0)
History of recurrent lower airway infections	-	62 (54.4)	113 (99.1)
Lung surgery performed	-	3 (2.6)	0 (0.0)
Diagnosis of bronchial asthma	-	23 (20.2)	2 (1.8)
Post-allogeneic stem cell transplantation	-	11 (9.6)	0 (0.0)
Immunoglobulin replacement therapy	-	84 (73.7)	0 (0.0)
Pulmonary therapy	-	34 (29.8)	111 (97.4)
Inhaled hypertonic saline	-	13 (11.4)	108 (94.7)
Dornase α	-	1 (0.9)	72 (63.2)
Bronchodilators	-	24 (21.1)	77 (67.5)
Leukotriene receptor antagonists	-	1 (0.9)	0 (0.0)
Inhaled glucocorticosteroids	-	18 (15.8)	32 (28.1)
Inhaled antibiotics	-	1 (0.9)	39 (34.2)
Respiratory physiotherapy	-	8 (7.0)	67 (58.8)
CFTR genotype			
F508del/F508del	-	-	53 (46.5)
F508del/other	-	-	38 (33.3)
Other/other			23 (20.2)
Exocrine pancreatic insufficiency	-	-	102 (89.5)
Chronic *Pseudomonas aeruginosa* lung infection	-	-	31 (27.2)
CFTR modulator therapy	-	-	61 (53.5)
Ivacaftor	-	-	2 (1.8)
Lumacaftor/ivacaftor	-	-	25 (21.9)
Tezacaftor/ivacaftor	-	-	9 (7.9)
Ivacaftor/tezacaftor/elexacaftor	-	-	25 (21.9)

Age is given as median (range), BMI and BMI z-score are given as median (interquartile range), other results are given as n (%) unless stated otherwise. BMI z-scores were calculated using values from a German reference population [36]. HC: healthy controls; IEI: inborn errors of immunity; CF: cystic fibrosis; BMI: body mass index; MBW: multiple-breath washout; CT: computed tomography; MRI: magnetic resonance imaging; CFTR: cystic fibrosis transmembrane conductance regulator. [#]: 28 healthy controls were previously part of a study [22]; [¶]: lower percentage of female patients due to x-linked inheritance of several IEI.

lower airway infections, were diagnosed with bronchiectasis or received pulmonary therapies (table 1). The diagnosis of bronchial asthma was more prevalent in IEI patients than in CF patients (table 1). The majority of IEI patients received an immunoglobulin replacement therapy (table 1) as about two-thirds of the IEI patients had a predominant antibody deficiency (table 2). The next most common entities were combined immunodeficiencies with associated or syndromic features and congenital defects of phagocyte number, function or both (table 2). 11 IEI patients who underwent allo-HSCT were included (table 2). Five (45.5%) had lung involvement prior to allo-HSCT. The inclusion of these patients did not impact the results (supplementary table S1).

LCI and FEV1 z-score of IEI patients differ significantly from HC and CF patients

Spirometry was performed successfully in 111 (97.4%) HC, 92 (80.7%) IEI patients and 105 (92.1%) CF patients, while successful MBW was an inclusion criterion for this analysis. The median (IQR) LCI was 6.5 (6.2–6.9) in HC, 7.0 (6.5–7.9) in the IEI group and 8.9 (6.8–14.2) in the CF group (figure 1a). The LCI was significantly higher in the IEI patients than in the HC cohort ($p < 0.001$) and significantly lower in the IEI patients than in the CF patients ($p < 0.001$) (figure 1a). FEV_1 z-score had a median (IQR) of −0.1 (−0.8–0.5) in the HC group, −0.6 (−1.4–0.1) in the IEI group and −1.1 (−2.7–0.3) in the CF group (figure 1b). The FEV_1 z-score of the IEI patients was significantly lower than that of the HC cohort ($p < 0.01$) and significantly higher than that of the CF patients ($p < 0.01$) (figure 1b).

LCI is abnormal in more patients than is the FEV_1 z-score

The ULN of the LCI derived from HC was 7.2 in participants <50 years of age and 8.7 in participants ⩾50 years of age. In total, 40 of 114 IEI patients (35.1%) and 79 of 114 CF patients (69.3%) had an elevated LCI. 19 of 92 IEI patients (20.7%) and 41 of 105 CF patients (39.0%) had a decreased FEV_1 z-score. Among participants with both measurements, 14 of 92 IEI patients (15.2%) and 39 of 105 CF patients (37.1%) had both an abnormal LCI and an abnormal FEV_1 z-score (figure 1c and d). 19 of 92 IEI patients (20.7%) and 35 of 105 CF patients (33.3%) had an elevated LCI but a normal FEV_1 z-score,

TABLE 2 Diagnosis and classification of the Inborn Errors of Immunity Group according to the International Union of Immunological Societies Classification for Inborn Errors of Immunity [33]

Group	Diagnosis	Total	Post allo-HSCT
All patients with IEI		114 (100.0)	11 (9.6)
Predominantly antibody deficiencies n=80 (70.2%)	Activated PI3 kinase delta syndrome	3 (2.6)	1 (0.9)
	CVID with no gene defect specified	38 (33.3)	0 (0.0)
	IgA with IgG subclass deficiency	2 (1.8)	0 (0.0)
	IKAROS deficiency	2 (1.8)	0 (0.0)
	Isolated IgG subclass deficiency	4 (3.5)	0 (0.0)
	NFKB1 deficiency	5 (4.4)	0 (0.0)
	NFKB2 deficiency	1 (0.9)	0 (0.0)
	UNG deficiency	1 (0.9)	0 (0.0)
	X-linked agammaglobulinaemia	16 (14.0)	0 (0.0)
	Antibody deficiency, unclassified	8 (7.0)	0 (0.0)
CID with associated or syndromic features n=11 (9.6%)	Ataxia telangiectasia	2 (1.8)	0 (0.0)
	AD-Hyper-IgE syndrome	5 (4.4)	0 (0.0)
	ICF3 syndrome	1 (0.9)	1 (0.9)
	NEMO deficiency	2 (1.8)	0 (0.0)
	Wiskott–Aldrich syndrome	1 (0.9)	0 (0.0)
Congenital defects of phagocyte number, function, or both n=9 (7.9%)	Chronic granulomatous disease	9 (7.9)	4 (3.5)
Immunodeficiencies affecting cellular and humoral immunity n=6 (5.3%)	CD40 ligand deficiency	2 (1.8)	1 (0.9)
	STK4 deficiency	1 (0.9)	1 (0.9)
	Combined immunodeficiency, unclassified	3 (2.6)	3 (2.6)
Others n=8 (7.0%)	ADA2 deficiency	1 (0.9)	0 (0.0)
	CD70 deficiency	1 (0.9)	0 (0.0)
	Good syndrome	2 (1.8)	0 (0.0)
	IRAK4 deficiency	1 (0.9)	0 (0.0)
	LRBA deficiency	2 (1.8)	0 (0.0)
	XIAP deficiency	1 (0.9)	0 (0.0)

Data are presented as n (%). allo-HSCT: allogeneic haematopoietic stem cell transplantation; IEI: inborn errors of immunity; CVID: common variable immunodeficiency; Ig: immunoglobulin; CID: combined immunodeficiency.
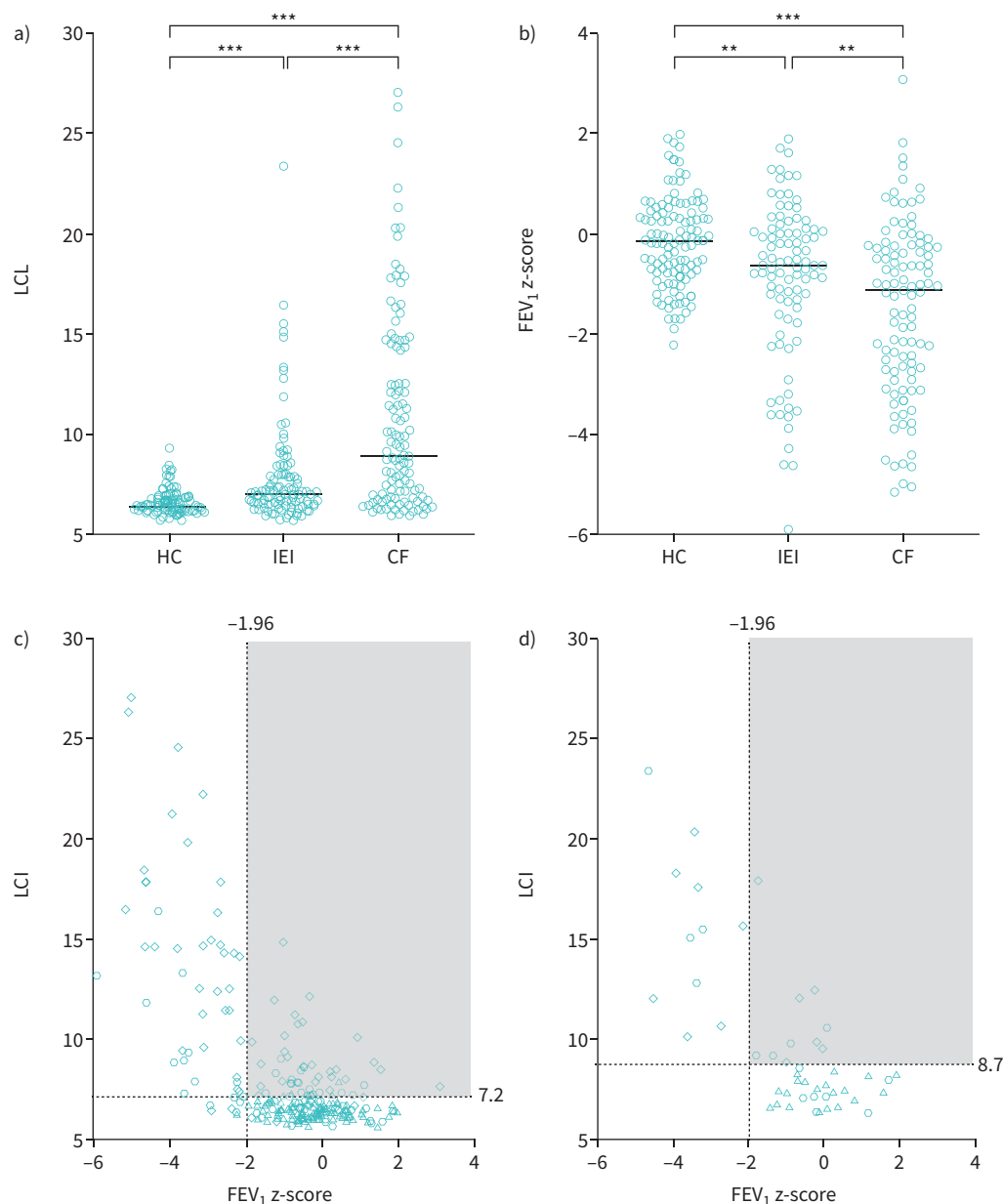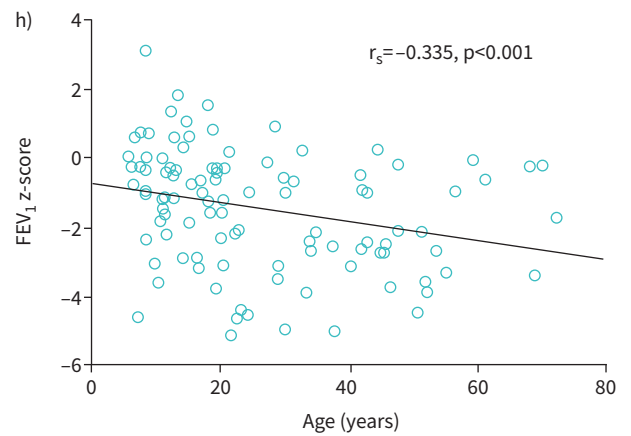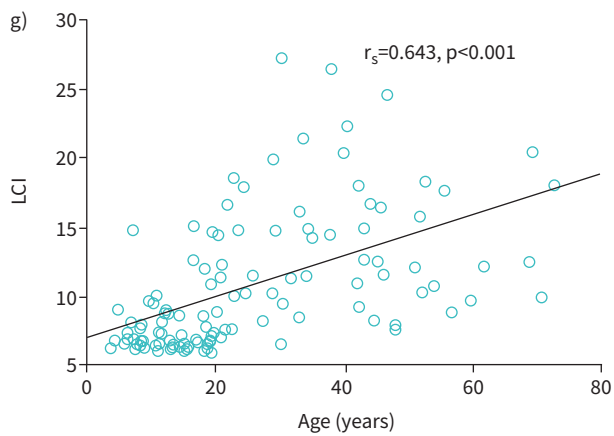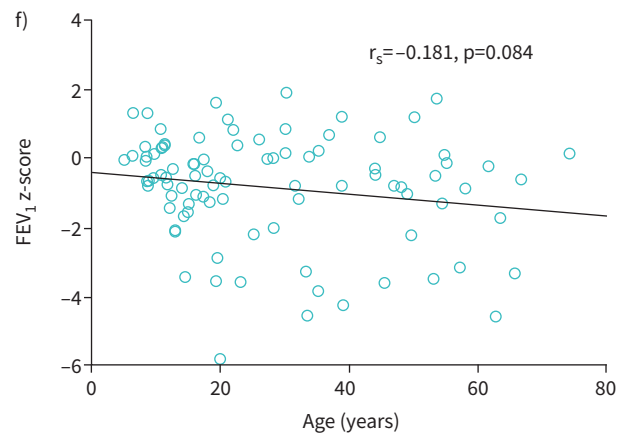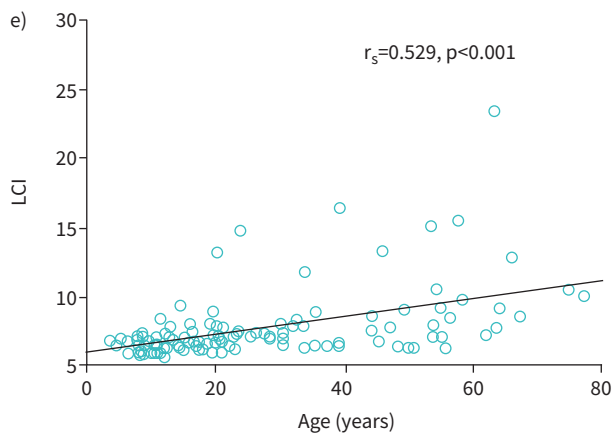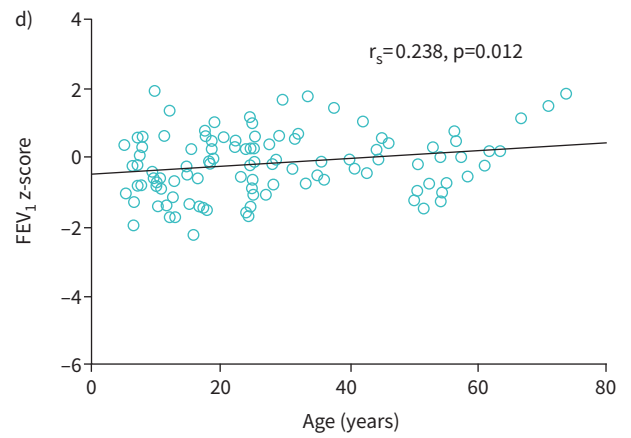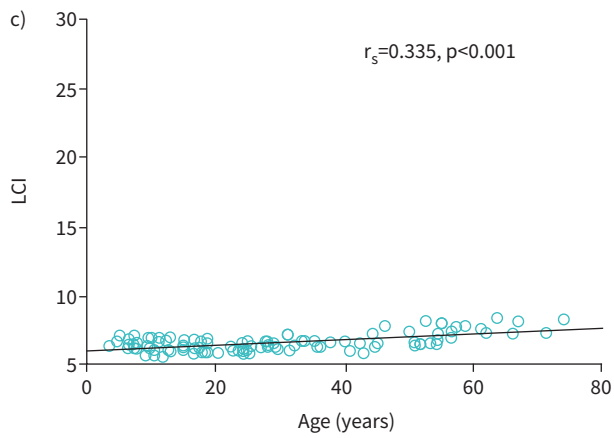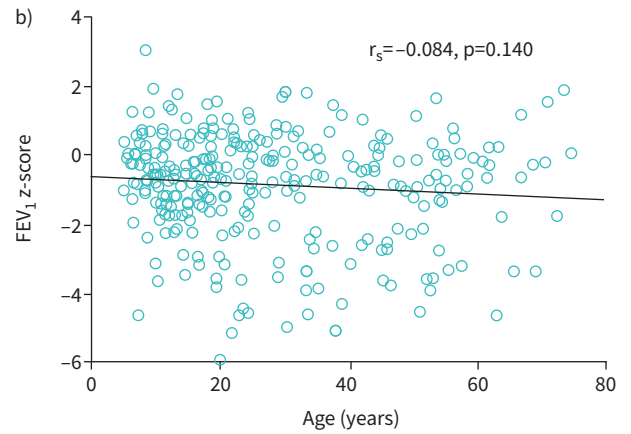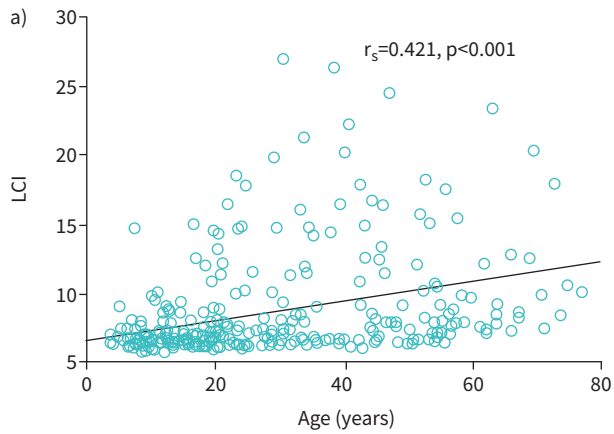
FIGURE 1 Lung clearance index (LCI) and forced expiratory volume in 1 s (FEV_1) z-score of healthy controls (HC), patients with inborn errors of immunity (IEI) and patients with cystic fibrosis (CF). Comparison of a) LCI and b) FEV_1 z-score between HC, patients with IEI and patients with CF. Relationship between LCI and FEV_1 in c) participants under 50 years of age and d) 50 years and older. p-values refer to the comparison of LCI and FEV_1 z-score between the groups. Triangles represent HC, diamonds represent patients with CF and hexagons represent patients with IEI. The horizontal lines indicate the median value. The dotted lines indicate the lower limit of normal of the FEV_1 z-score and the upper limit of normal of the LCI. The grey area highlights patients with elevated LCI values but normal FEV_1 z-scores. **p<0.01; ***p<0.001.

while five of 92 IEI patients (5.4%) and two of 105 CF patients (1.9%) had a decreased FEV_1 z-score and a normal LCI (figure 1c and d).

LCI correlates with age and FEV_1 z-score in IEI patients

LCI and age correlated significantly in the total cohort (r_s=0.42, p<0.001) as well as in the HC group (r_s=0.34, p<0.001), the IEI group (r_s=0.53, p<0.001) and the CF group (r_s=0.64, p<0.001) (figure 2). FEV_1 z-score and age correlated significantly in the HC group (r_s=0.24, p<0.05) and in the CF group

FIGURE 2 Correlation of the lung clearance index (LCI) and the forced expiratory volume in 1 s (FEV$_1$) z-score with age. a and b) Correlation of the LCI and the FEV$_1$ z-score with age is shown for the total cohort, c and d) healthy controls, e and f) patients with inborn errors of immunity and g and h) patients with cystic fibrosis. Spearman's rank correlation coefficients (r_s) and p-values are provided for each correlation.

($r_s = -0.34$, p<0.001), but not in the IEI group ($r_s = -0.18$, p=0.084) (figure 2). The LCI and the FEV$_1$ z-score negatively correlated in the total cohort ($r_s = -0.49$, p<0.001), the IEI group ($r_s = -0.52$, p<0.001) and the CF group ($r_s = -0.65$, p<0.001) (figure 3).

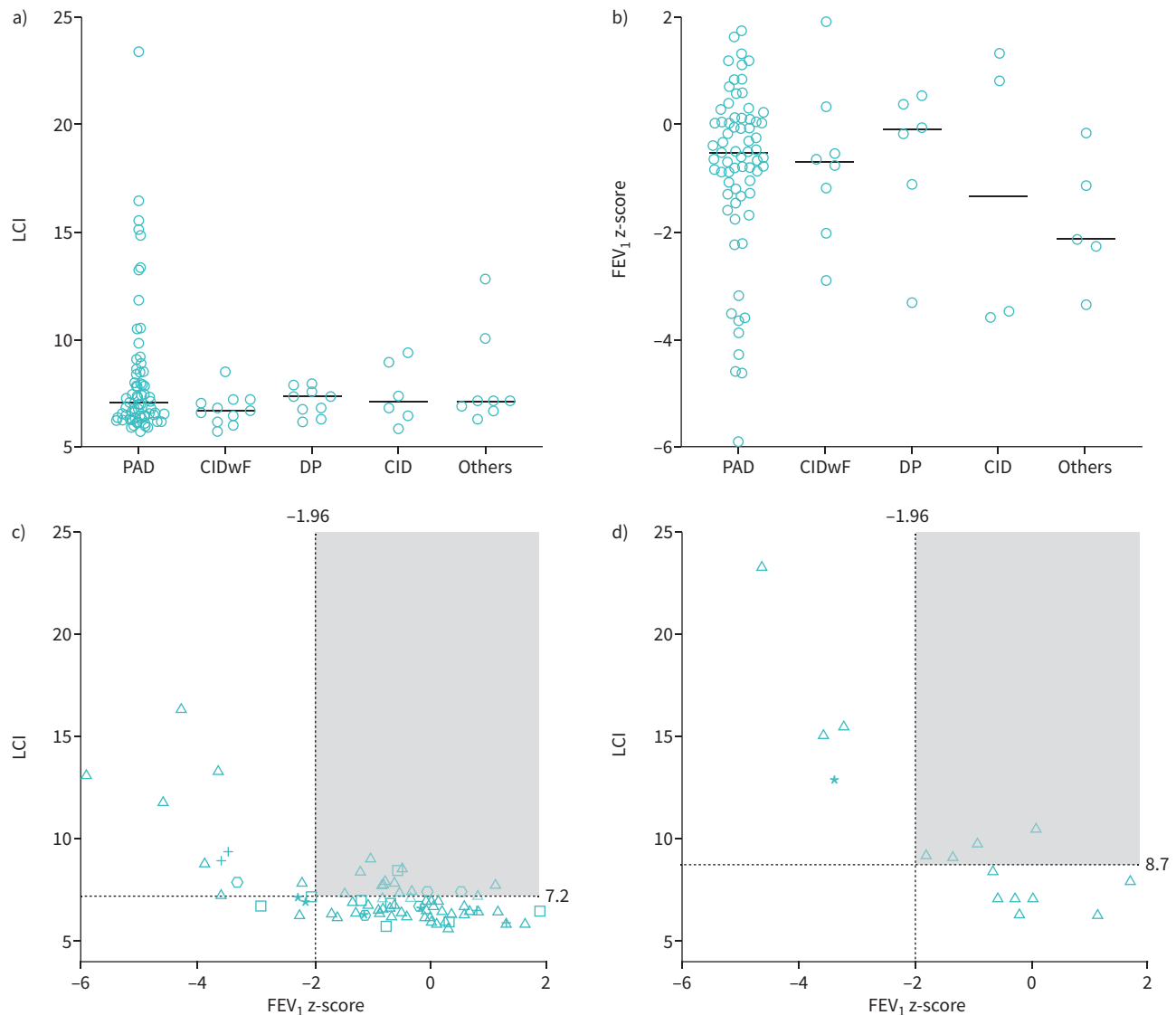### LCI and FEV$_1$ z-score in the different IEI categories

The LCI and the FEV$_1$ z-score of the IEI patients were evaluated according to the different IEI categories (figure 4). The median (IQR) LCI and FEV$_1$ z-score were 7.0 (6.4–8.0) and −0.6 (−1.3–0.1) for patients with predominantly antibody deficiencies, 6.7 (6.2–7.2) and −0.7 (−1.8–0.1) for patients with combined immunodeficiencies with associated or syndromic features, 7.3 (6.5–7.7) and −0.1 (−1.7–0.4) for patients with congenital defects of phagocytes number, function or both, 7.1 (6.3–9.1) and −1.3 (−3.6–1.2) for patients with immunodeficiencies affecting cellular and humoral immunity, and 7.1 (6.7–9.3) and −2.1 (−2.8–0.7) for those with other entities (figure 4a and b, and supplementary table S2).

LCI and FEV$_1$ z-scores are worse in IEI patients with bronchiectasis than in those without bronchiectasis

32 of 114 (28.1%) IEI patients received a chest CT or MRI within 5 years before the initial MBW measurement. Of those, 14 (43.8%) did not have bronchiectasis, while 18 (56.3%) were diagnosed with bronchiectasis. IEI patients with bronchiectasis had significantly lower body mass index values, more often
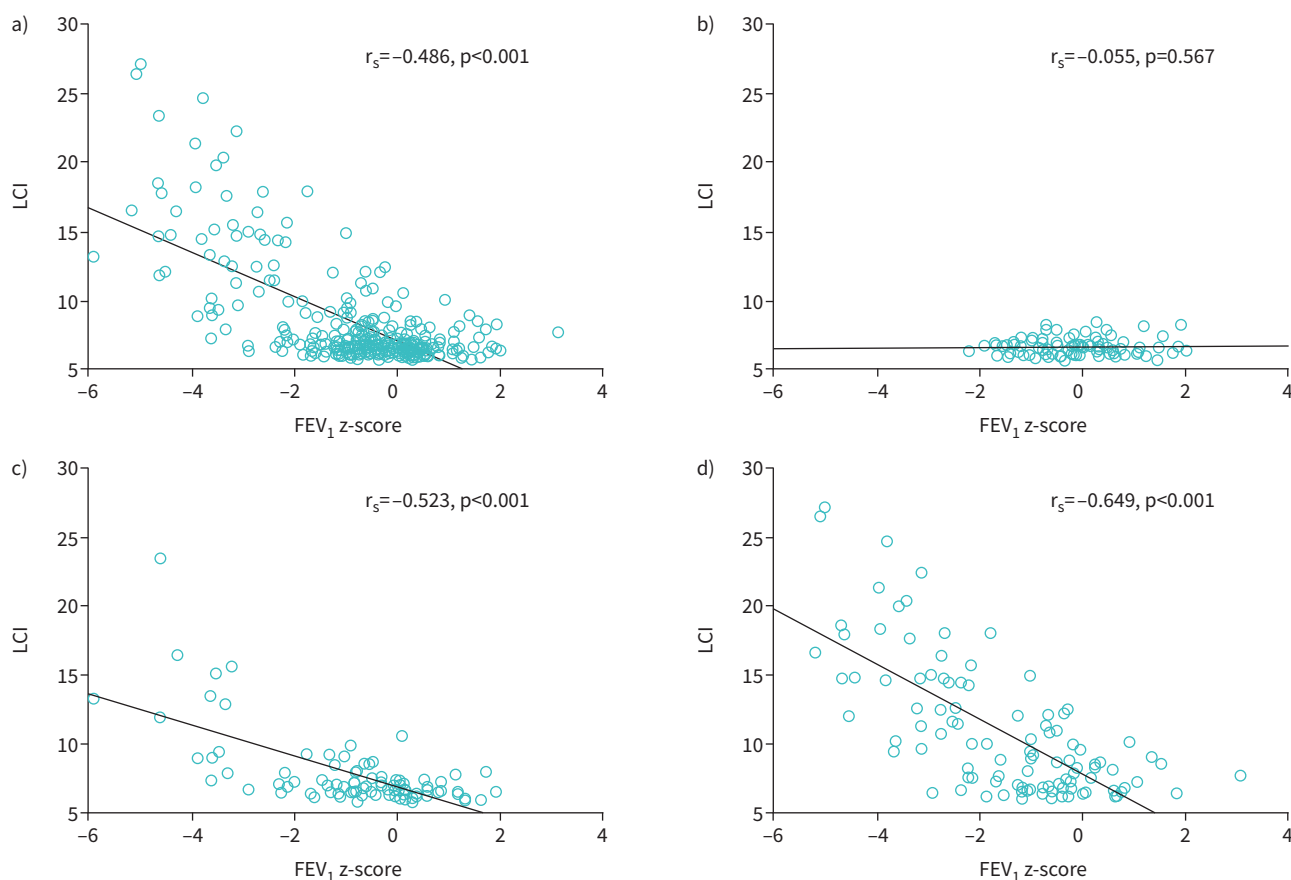
FIGURE 3 Correlation between the lung clearance index (LCI) and the forced expiratory volume in 1 s (FEV$_1$) z-score. Correlation between the LCI and the FEV$_1$ z-score is shown for a) the total cohort, b) the healthy controls, c) the patients with inborn errors of immunity and d) the patients with cystic fibrosis. Spearman's rank correlation coefficients (r_s) and p-values are provided for each correlation.

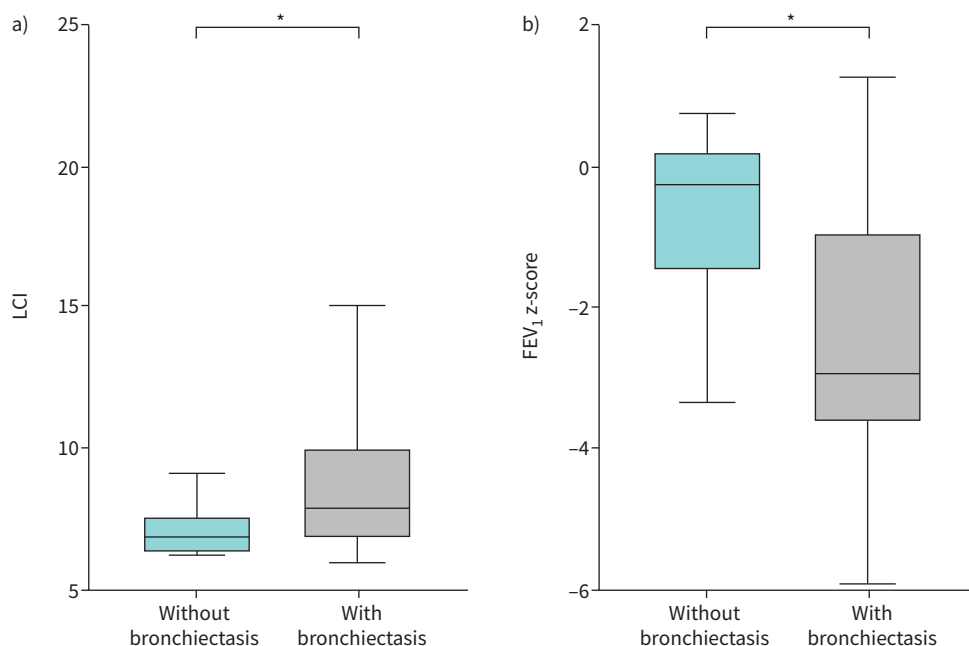FIGURE 4 Lung clearance index (LCI) and forced expiratory volume in 1 s (FEV_1) z-score according to entity of inborn errors of immunity (IEI). a) The LCI and b) FEV_1 z-score are given for the different IEI groups. Horizontal lines indicate the median value. The relationship between the LCI and the FEV_1 z-score in the different IEI groups is shown for c) patients with both measurements under 50 years of age (n=77) and d) 50 years and older (n=15). Symbols reflect the IEI groups: triangle: predominantly antibody deficiencies (PAD) (n=69); squares: combined immunodeficiencies with associated or syndromic features (CIDwF) (n=8); hexagon: congenital defects of phagocyte number, function or both (DP) (n=6); +: immunodeficiencies affecting cellular and humoral immunity (CID) (n=4); *: others (n=5), including ADA2 deficiency (n=1), Good syndrome (n=1), IRAK4 deficiency (n=1), LRBA deficiency (n=1) and XIAP deficiency (n=1). The dotted lines indicate the lower limit of normal of FEV_1 z-score and the upper limit of normal of LCI. The grey area highlights patients with elevated LCI values but normal FEV_1 z-scores.

a history of recurrent lower airway infections and received more frequently pulmonary therapies compared to IEI patients without bronchiectasis (supplementary tables S3 and S4). The median (IQR) LCI and median FEV_1 z-score for patients without *versus* patients with bronchiectasis were 6.9 (6.4–7.6) *versus* 7.9 (6.8–10.0; p<0.05) and −0.2 (−1.4–0.2) *versus* −2.9 (−3.6–0.9; p<0.05) (figure 5).

LCI in IEI patients increases over time

70 of 114 (61.4%) IEI patients received a second MBW measurement after a median (range) of 364 (63–707) days (supplementary tables S5 and S6). The LCI showed a significant median (IQR) increase of 0.2 (−0.2–0.5; 2.2% (−2.1–7.3)) at the second measurement (p<0.01) (figure 6a). The median (IQR) change in LCI per patient year was 0.1 (−0.2–0.5; 2.2% (−2.6–7.9)) (p<0.01) (figure 6b and c). 13 (18.6%) of 70 patients showed an increase of >10% after 1 patient year.

FIGURE 5 Comparison of the lung clearance index (LCI) and the forced expiratory volume in 1 s (FEV_1) z-score between patients with inborn errors of immunity (IEI) without and with bronchiectasis. Comparison between IEI patients without and with bronchiectasis is shown for a) the LCI and b) the FEV_1 z-score. Boxes extend from the 25th to the 75th percentile and horizontal lines indicate the median value. Whiskers mark the minimum and maximum values. p-values refer to the comparison of LCI and FEV_1 z-score between the groups. *$p < 0.05$.
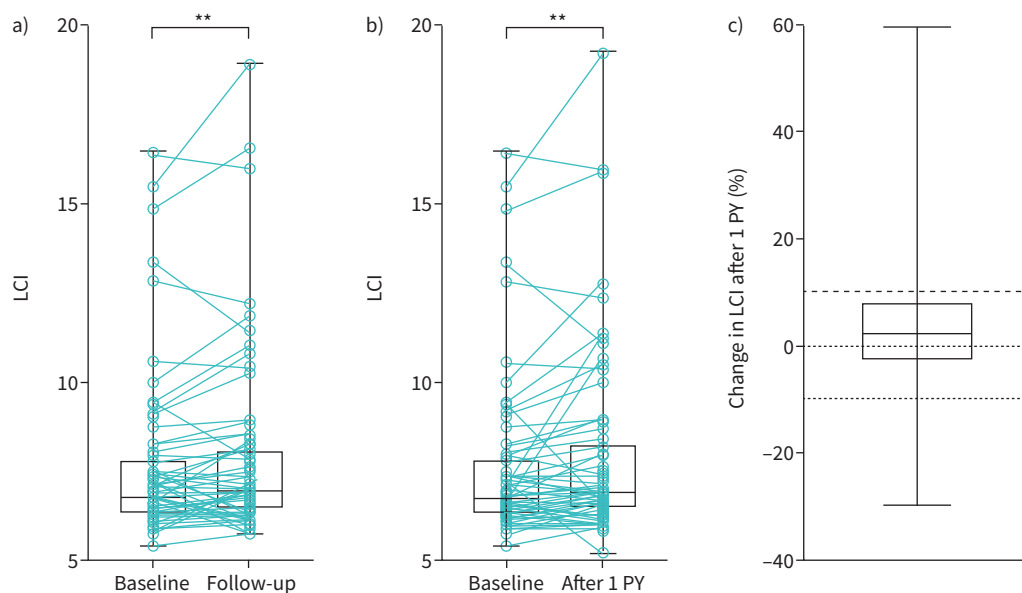
FIGURE 6 Repeated lung clearance index (LCI) measurements in patients with inborn errors of immunity (IEI). a) Change of LCI in clinically stable IEI patients from baseline to a second measurement (follow-up) after a median (range) of 364 (63–707) days. b) Change of baseline LCI after 1 patient year (PY). c) Change in LCI after 1 PY in per cent (%). p-values refer to the longitudinal change of LCI. Boxes extend from the 25th to the 75th percentile and horizontal lines indicate the median value. Whiskers mark the minimum and maximum values. The dashed lines indicate a clinically relevant change of 10%. **$p < 0.01$.

Discussion

This is the largest study to date comparing MBW and spirometry for IEI patients, CF patients and HC, demonstrating that MBW is feasible and sensitive to detect lung disease in IEI patients and that the LCI of IEI patients differs from CF patients and HC. In addition, we show that more patients are identified with abnormal results for LCI than for FEV_1. Therefore, LCI is more sensitive than FEV_1 to detect lung disease. Furthermore, we found that IEI patients with bronchiectasis have higher LCI values than those without and that LCI increases with time in IEI patients.

Previous studies showed that MBW can assess even mild lung disease in patients with various entities such as CF, PCD and COPD [15–30, 37]. The present study extends the use of MBW to IEI patients of a wide age range and with different underlying disease entities demonstrating that IEI lung disease can be assessed by MBW. As expected, the LCI of patients with lung diseases (both IEI and CF) is significantly higher than in HC [17–19, 22, 28, 30]. However, the level of lung disease detected by the LCI seems to be milder in IEI patients compared to age-matched patients with CF (figure 1). Comparable to previous findings in different lung diseases, we found a moderate negative correlation between LCI and FEV_1 in IEI patients (figure 3). In addition, our investigations in both patients with IEI and CF are in line with previous reports on a higher sensitivity of the LCI to identify patients with lung diseases than the FEV_1 [18, 19, 24, 30] (figure 1).

Previously, studies in CF and PCD found a correlation between LCI and radiological findings of lung pathology in CT and MRI [22, 24, 30]. In the present study, chest CT or MRI were available in 32 IEI patients at baseline. In this subgroup, IEI patients with bronchiectasis had significantly higher LCI values than those without (figure 5). This suggests a correlation between LCI and bronchial changes in IEI patients. Previous studies also demonstrated that the LCI correlates more strongly with structural lung changes assessed by radiological imaging than FEV_1 [24, 30]. However, our radiological data are limited, partially due to limited access to imaging during the COVID-19 pandemic. This resulted in imaging and pulmonary function testing not taking place at the same time. Therefore, structural lung changes could have developed by the time of MBW testing that were not present when lung imaging was performed. To validate our findings on LCI and to evaluate correlations with additional lung function parameters, the use of consensus diagnostic criteria of bronchial and parenchymal pathologies for CT/MRI lung imaging in IEI is warranted for further studies [8].

IEI lung disease was shown to progress with age regardless of immunoglobulin replacement therapy [1, 4, 5]. Therefore, regular assessment of pulmonary function is required which should provide information on even small changes in lung function to adequately adjust therapies to the individual level of lung disease. LCI was found to be the earliest and strongest lung function parameter to predict disease progression in a cohort of CF patients [38]. In the present study, LCI was used to assess lung function in clinically stable IEI patients of which 70 underwent a second MBW measurement after a median of 1 year. The LCI showed a median increase of 0.2 during this period (figure 6), leading to a comparable median increase per patient year. Almost a fifth of the IEI patients had an increase of LCI above 10% after 1 patient year, which is considered the cut-off for a clinically meaningful change [20]. This indicates that the lung disease of IEI patients progresses even in clinically stable patients within a short period of time and that LCI can detect this progression. However, not all IEI patients received a second measurement, and the follow-up period differed between the patients. Therefore, further investigations including lung imaging are needed for verification.

IEI patients who underwent allo-HSCT for IEI were also included in this study. During allo-HSCT, the mutated haematopoietic stem cells are replaced by healthy donor cells and the immunodeficiency is potentially cured in the long-term. However, lung disease can develop already prior to allo-HSCT (without complete reversion after transplant) or afterwards in the form of infectious or non-infectious pulmonary complications caused by graft-*versus*-host disease [39]. Since we aimed to monitor any respiratory manifestations, we included IEI patients post allo-HSCT. Of note, inclusion of transplanted IEI patients did not impact our results (supplementary table S1).

A limitation of this study is the heterogeneity of diagnoses in the IEI group. While there are a substantial number of patients with antibody deficiency, the numbers of patients for other IEI categories are small. Multicentre studies are required for a more detailed investigation of the LCI in different IEI categories, as well as for a comparison between those groups. However, a strength of our study is that MBW measurements were performed in a clinical setting, supporting the feasibility of lung function monitoring in IEI patients.

In summary, this is the largest study to date comparing LCI and FEV_1 z-score in patients with IEI. We demonstrated that the LCI is sufficiently sensitive to detect lung disease in IEI patients. Although our study shows a moderate negative correlation between LCI and FEV_1 z-score for IEI and CF patients, we demonstrated that more patients have abnormal LCI values than FEV_1 z-scores. In addition, we found that the LCI differs between IEI patients with bronchiectasis and those without. MBW may help to identify patients that should undergo CT or MRI lung imaging for early detection of lung disease facilitating early treatment initiation. We provide supportive evidence for the feasibility and benefit of LCI as an additional outcome parameter for the monitoring of IEI lung disease.

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