# ****Clinical characteristics and lung function in older children vertically infected with HIV in Malawi****

## ****Running title****

**Lung health in vertically acquired HIV**

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### Keywords

HIV

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## Abstract

### Rationale

Antiretroviral therapy has led to increased survival of children with vertically acquired HIV infection. Significant morbidity arises from respiratory symptoms, but aetiology and pulmonary function abnormalities have not been systematically studied.

### Method

HIV positive children aged 8-16 years were systematically recruited within clinics in Blantyre, Malawi. Clinical review, quality of life assessment, spirometry and chest radiography were performed.

### Results

160 participants had mean age 11.1 (range 8-16) years and 50.0% were female. Cough was present in 60 (37.5%), 55 (34.4%) had moderate or severe dyspnoea. 34 (22.1%) had digital clubbing. 33 (20.6%) were hypoxic at rest. 118 (73.8%) of children were receiving antiretroviral therapy (ART); median CD4 count was 698 cells/µl in these compared with 406 cells/µl in ART-naïve individuals (p<0.001). From 145 spirometry traces (90.6%), mean FEV1 and FVC were 1.06 and 0.89 standard deviations below predicted respectively. 21 (14.5%) traces demonstrated obstructive defects and 26 (17.9%) reduced FVC. Lung function abnormality was not associated with any clinical findings. Of the 51 individuals with abnormal lung function, the mean increase in FEV1 after salbutamol was 3.8% (95%CI 0.02 to 7.53). “Tramlines” and ring shadows were seen on chest radiographs in over half of cases.

### Conclusion

Symptoms of chronic lung disease were highly prevalent with two main clinical phenotypes: “cough” and “hypoxia”. Lung function abnormalities are common, poorly responsive to bronchodilators and apparent throughout the age range of our cohort. Pathological causes remain to be elucidated. “Cough” and “hypoxic” phenotypes could be a useful part of diagnostic algorithms if further validated.

***Abstract word count: 237***

## Introduction

An estimated 3.2 million children are living with HIV[1](#_ENREF_1), most of whom live in sub-Saharan Africa. 199,000 children were newly infected in 2013. Scale-up of antiretroviral (ART) provision is likely to contribute to improvements in survival amongst children with HIV[2](#_ENREF_2). However, in sub-Saharan Africa, only 22% of HIV positive children are receiving ART, lagging behind 39% coverage in adults[1](#_ENREF_1). Prevention of vertical infection through Prevention of Mother to Child Transmission (PMTCT) programmes is effective, but worldwide coverage requiring ART has been suboptimal. An estimated 1 to 3% of all 10-year-olds in Southern Africa are HIV infected long-term survivors, with 68% of eligible women received ART as PMTCT in 2013 compared with 33% five years previously[1](#_ENREF_1). In a hospital based study from Zimbabwe, advanced HIV infection was the single most common cause of admission and death in adolescents[3](#_ENREF_3).

With longer survival, lung effects of HIV become more prominent. HIV-infected children in sub-Saharan Africa are subject to frequent pulmonary infections[4](#_ENREF_4), and commonly develop chronic cough in older childhood. In a Zimbabwean study of 116 HIV infected adolescents receiving HIV care, dyspnoea was often disabling, and resting hypoxia or desaturation at submaximal exercise was present in 40%[5](#_ENREF_5). Chest radiographs were abnormal in two thirds of patients, characterised by ring and tramline opacities whose presence is unrelated to clinical symptoms. High resolution computerised tomography (HRCT) scans suggested small airways disease as the most common cause, however bronchodilator response was not assessed[6](#_ENREF_6). Despite this, data on symptoms of chronic lung disease and lung function testing, particularly in developing countries, are lacking.

We describe the burden and clinically useful phenotypes of chronic lung diseases, and assesses the bronchodilator response with inhaled beta-agonist therapy in HIV infected children aged 8 to 16 receiving HIV care in Blantyre, Malawi.

**Methods**

### Participants

Participants were recruited from out-patient HIV clinics in Queen Elizabeth Central Hospital, Blantyre between July and December 2011. The first 3 eligible patients per day were included. Patients were not eligible for recruitment if they: resided outside urban Blantyre; were currently taking tuberculosis (TB) treatment; had Kaposi’s sarcoma; reported acute respiratory symptoms (≤1 week of any one or more of fever, purulent sputum, pleuritic chest pain); required emergent hospitalisation.

At baseline we assessed medical history, symptoms, quality of life and functional status by standardised verbal questionnaires administered in the local language. Examination included assessment of finger clubbing, growth and WHO clinical staging of HIV disease. CD4 count was performed, and TBsmear and culture done in all participants who could spontaneously expectorate. Participants performed a 200m sub-maximal walk test, unless contraindicated due to resting hypoxia (SpO2 <92%) or tachypnoea (>24/min). Within two weeks, participants had spirometry unless there was evidence of TB or acute respiratory illness[7](#_ENREF_7). Chest radiographs were reported by two independent clinicians using a standardised scoring system[6](#_ENREF_6), with discrepancies resolved by consensus.

### Quality of life assessment

In the absence of a disease-specific tool, quality of life was assessed using the Cystic Fibrosis Questionnaire-Revised (CFQ-R, for 6-13 year olds and their carers in parallel). These incorporate nine quality of life domains (physical, school, vitality, emotion, social, body image, eating, treatment burden, health perception) and 3 symptom domains (respiratory, digestion, weight). Translation was by two independent translatiors, collation by consensus, and back-translation for accuracy[8](#_ENREF_8).

### Spirometry and radiology

Spirometry was performed according to ATS/ERS guidelines[9](#_ENREF_9) by experienced nursing staff and a respiratory physician. Forced exhalation following maximal inspiration was recorded while seated using an EasyOne World spirometer (ndd, Switzerland). Up to eight trials were recorded, and assessed by two clinicians independently for quality. The best FEV1 and FVC values from 3 admissible traces were included for analysis. Primary reference values were taken from the Global Lung Initiative (GLI)[10](#_ENREF_10), using the 5th centile of the reference population as the lower limit of normal (LLN, 1.64SD below the mean). Results of FEV1, FVC, FEV1:FVC and FEF25-75% are reported as residual standard deviations (z-scores). Participants with FEV1 and FEV1:FVC ratio of less than the LLN were classified as having obstructive spirometry. Where FVC was reduced below LLN and FEV1:FVC was not, we recorded “reduced FVC” as we were unable to measure total lung capacities. In participants with abnormal spirometry, testing was repeated after nebulised salbutamol (2.5mg via face mask). Reversibility was defined as improvement of ≥12% in best FEV1 or FVC[9](#_ENREF_9). Participants meeting this criterion were prescribed inhaled salbutamol via metered dose inhaler with an Aerochamber device (GSK, UK), 200µg at least twice a day, and additionally when symptomatic. These participants returned for clinical reassessment after 4 weeks of treatment.

Secondary interpretation used a locally derived reference range[11](#_ENREF_11), with LLN of 80% predicted. Results are presented as “percentage of normal” given uncertain population standard errors.

### Laboratory methods

CD4 counts were determined by flow cytometry (BD FACSCount™, CA, USA). Participants with cough were asked to provide two sputum specimens. Concentrated decontaminated sputum specimens were examined with auramine, and cultured using Mycobacterial Growth Indicator Tubes (MGIT™, Becton Dickinson, Belgium). Positive mycobacterial cultures were confirmed by Ziehl-Neelsen staining and speciated using the Hain® assay (Hain LifeScience GmbH, Germany).

### Ethical approval

Ethical approval was obtained from the College of Medicine Research Ethics Committee, Malawi (P.02/11/1039) and the London School of Hygiene and Tropical Medicine Ethics Committee (5964). Informed written or witnessed thumbprint participant assent and parental or guardian consent were required for recruitment.

### Analysis

Univariable associations of abnormal lung function were assessed using logistic regression, with predictors at p<0.10 taken forward to a multivariable model in addition to age and sex.

Exploratory analysis was used to compare different prototype definitions of CLD, including two-way associations between individual variables, aiming for a definition that could be applied in outpatient clinics where investigations are limited to oximetry and symptom screening. We assessed univariable relationships, looking for a clinically useful phenotype. Multivariable logistic regression was then used to identify independent variables independently associated with these phenotypes. A hierarchical approach to modelling (two levels: distal and proximal) was used to account for factors that affect the lungs indirectly and directly, respectively[12](#_ENREF_12). Distal (indirect) factors were: stunting, orphanhoodandvariablesselected *a priori* for inclusion including sex, age, antiretroviral therapy and CD4 count. Proximal (direct) factors included symptoms, physiological observations and radiographic and spirometric abnormality. Participants with positive *Mycobacterium tuberculosis* culture were excluded from case definition analyses.

Statistical analysis usedStata v12 (STATA Corporation, TX).

## Results

The flow and baseline characteristics of the 160 participants are shown in Figure 1 and Table 1 respectively. All children in the study were black African, with a mean age of 11.1 years (SD 2.1). 114 (71.7%) were established on antiretroviral therapy, and 46 (28.3%) not yet meeting criteria for treatment. Median CD4 counts in these groups were 698 cells/µl and 406 cells/µl respectively (p<0.001, Wilcoxon rank sum test). Perinatal acquisition of HIV was assumed after a systematic review of participants’ risk factors: 16 (10.3%) had received blood transfusion; 11 had previous surgery (6.9%); 53 (33.1%) had injections outside the healthcare setting, including escarification; 3 (1.9%) reported sexual abuse; none reported other sexual activity. 89 (56%) had no risk factors other than maternal orphanhood or known HIV infection, although reported factors for other transmission routes were higher than in the Zimbabwe study[13](#_ENREF_13) Previous respiratory complaints were common: 30 (18.8%) treated for tuberculosis; 20 (12.5%) for asthma; 13 (8%) for chest infection requiring hospitalisation. Household air pollution was common. 76 (47.5%) and 117 (73.1%) of households used biomass fuel as the predominant energy source for lighting and cooking respectively. Passive smoking was reported in 23 (14.5%).

Notably, 91 (56.9%) participants had one or more of: cough; moderate or severe dyspnoea New York Heart Association (NYHA grade III and IV); wheeze. Of those with previous pulmonary tuberculosis 6 (3.8%) had received more than two courses. In the preceding year, 34 (21.3%) had used antibiotics for a lower respiratory tract infection: 17 (10.6%) had received multiple courses.

### Quality of life

Participants reported high quality of life in six domains (median scores 88.9-100.0 for emotional, eating, body image, treatment burden, respiratory and digestion), where 100 is the maximum score. Social and physical activity domains had the lowest median scores of 57.1 (IQR 47.6-57.1) and 83.3 (IQR 55.6-100.0), respectively.

### Developing a case definition and phenotypes of chronic lung disease

An association matrix was used to investigate potential case definitions (Supplementary Table 2). Two patients with active tuberculosis were excluded from this. Two candidate phenotypes were postulated: one characterised by cough (37.5%, 95%CI 30.0-45.1%) and the other by hypoxia or desaturation at submaximal exercise (38.8%, 95%CI 31.1-46.4%). While these might be expected to commonly co-exist, only 22 (13.8%, 95%CI 8.4%-19.1%) participants had both. There was lack of agreement between these two variables over that expected by chance alone (expected agreement 52.8%, observed agreement 51.3%, kappa = -3.3%). We therefore further characterised those two proposed phenotypes (“CLD-cough” and “CLD-hypoxia”) - see Figure 3. Table 2 summarises univariable and multivariable analysis of risk factors for chronic cough and hypoxia or desaturation individually.

**CLD-cough** was not associated with any indirect factors, but was significantly associated with wheeze (OR 11.47, 95%CI 2.44-53.83), abnormal chest radiograph (OR 3.00, 95%CI 1.15-7.85), and abnormal spirometry (OR 2.61, 95%CI 1.29-5.28). Participants with higher levels of exercise tolerance as measured by CFQ-R physical domain had reduced odds of CLD-cough (OR 0.98, 95%CI 0.97-0.99). After multivariable analysis, wheeze (OR 6.94, 95%CI 1.38-34.95, p=0.019), CFR-Q physical domain (OR 0.98, 95%CI 0.97-1.00, p=0.017) and abnormal chest radiograph (OR 3.43, 95%CI 1.01-7.85, p=0.048) remained significant.

**CLD-hypoxia** had more limited univariate predictors: only finger clubbing (OR 2.28, 95%CI 1.04-4.99) and respiratory rate (OR 3.00, 95%CI 1.52-5.92). Only resting tachypnoea remained significant in multivariable modelling adjusted for *a priori* variables. There was weak evidence for the association of CD4 <100cell/µl and CLD-hypoxia (OR 3.87, 95% 0.99-16.39, p=0.051) compared with children having a CD4 count of 350 or more.

### Spirometry

Spirometry results for 145 participants are summarised in Table 3 and Figure 2A. Median FEV1 and FVC were reduced compared with international reference ranges (1.31 SD and 0.89 SD below expected respectively). Categorically, 90 (62.1%) had normal spirometry, 26 (17.9%) obstructive defects and 29 (20.0%) reduced FVC. Fewer individuals were classified as having abnormalities using local compared with international reference range (43 vs 55). Within our cohort FEV1 z-score did not significantly decline with age (Figure 2, panel B, r2 = 0.026, p=0.054).

55 participants had abnormal spirometry of whom 47 adequately completed post-bronchodilator testing. Median change in FEV1 was 2.9% (IQR -4.3-9.2). Reversibility threshold of 12% increase was met in seven (33.3%) of those with obstructive abnormalities and eight (30.8%) with reduced FVC. When reviewed four weeks afterwards, despite being given salbutamol, only two participants continued to use their inhaler, and none reported symptomatic improvement.

### Chest radiograph abnormalities

The majority of radiographs had at least one abnormality (n=110, 68.8%). Upper or lower zone preponderance were uncommon (n=12, 10.9% and n=13, 11.8% respectively) compared with mid zone abnormality. The most frequently abnormality was ring or tramlining pattern (n=90, 56.3%). Two abnormalities (airspace shadowing and volume loss) were discriminatory for both CLD phenotypes (Table 2) and spirometric abnormality (Supplementary table 3). Air space shadowing (n=10, 6.3%), and loss of volume (n=3, 1.9%) were associated with reduced FEV1 (p=0.0032 and p=0.010 respectively). Other radiographic findings were not significantly associated with differences in FEV1 or FVC z-score.

### Clinical associations of lung function

Potential associates of abnormal spirometry were investigated (Supplementary data, table 2). Only one strong association emerged: individuals reporting cough for more than one month were 2.9 times more likely to have abnormal spirometry (95%CI 1.21–7.10).

### Microbiological findings

Sputum was obtained from 32/60 participants with cough, with the remainder unable to expectorate. There were 6 positive mycobacterial cultures; 2 M. *tuberculosis* and4 non-tuberculous mycobacteria.

## Discussion

This study demonstrates a high burden of symptoms in children aged 8 to 16 with vertically acquired HIV, consistent with a similar study from Zimbabwe[14](#_ENREF_14). Over half of our participants were coughing, wheezy or breathless. Within our cohort, there are two definable, common and independent phenotypes: children who cough (CLD-cough), and those who have hypoxia at rest or desaturate with submaximal exercise (CLD-hypoxia). Neither phenotype was associated with antiretroviral treatment.

For the CLD-cough phenotype**,** cough, wheeze and functional breathlessness were commonly associated with each other and also with radiological abnormalities of airspace shadowing and volume loss where parenchymal lung disease was likely. Previous treatment for tuberculosis was not a significant risk factor for this phenotype and symptoms were mostly chronic (individuals with symptoms of acute infection were excluded). Abnormal spirometry was associated with this phenotype but there was no preponderance of obstructive or restrictive types.

The CLD-hypoxia phenotype was predictably associated with tachypnoea. There was a suggestion that very low CD4 counts (<100) predicted hypoxia. Low numbers of individuals in this group limited our power to detect a difference. While not independently associated, there was a higher than expected rate of finger clubbing in these individuals.

Chronic lung disease in these children is likely to be multifactorial, and therefore difficult to clearly define[15](#_ENREF_15). Frequent bacterial, mycobacterial and viral respiratory infections were reported in this population (18.8% of our cohort had received treatment for chest infection in the preceding year), can also contribute to bronchiectasis. Consistent with underlying bronchiectasis there was a high rate of finger clubbing, reduced lung function and radiological abnormalities consistent with bronchiectasis. These features are insensitive and non-specific for its diagnosis in isolation[16](#_ENREF_16). A direct effect of HIV and chronic inflammation of the airways might give rise to reduced lung capacities and chronic chest X-ray findings, including lymphadenopathy. Findings that the pulmonary microbiota can be altered in adult HIV, notably for *Tropheryma whipplei* bacteria[17](#_ENREF_17), raise the possibility that these changes may reflect or drive long term disease in the airways, including chronic inflammation. HRCT findings from similar patients in Zimbabwe which are suggestive of airways disease[18](#_ENREF_18) can represent the final common pathway of many diseases, including post-infective change, although is uncommon outside of allogeneic transplantation[19](#_ENREF_19). In our study, the clinical syndrome of CLD-cough including non-reversible spirometry findings would be consistent with such pathology[20](#_ENREF_20).

Toro et al[21](#_ENREF_21) demonstrated a high burden of pulmonary lymphoid hyperplasia and lymphoid interstitial pneumonitis (LIP) in early life associated with a wide variety of radiological changes of which reticular infiltrates are most typical[22](#_ENREF_22). Early Western cohorts including the (P2C2 study) noted high rates of LIP and reported chest radiograph with LIP suggestive changes[23](#_ENREF_23), but this condition has been almost eliminated with effective ART provision[24](#_ENREF_24). Our participants were considerably older than the usual age of LIP presentation[25](#_ENREF_25), and started ART later.. In this case, the 2013 WHO guidelines to start all HIV positive children under 5 years on ART may improve rates of chronic lung disease in future.

A Zimbabwean study has shown a high frequency of cardiac abnormalities and cor pulmonale in adolescents with vertically transmitted HIV infection[14](#_ENREF_14). This raises the possibility that CLD-hypoxia, might represent pulmonary vascular disease or interstitial lung disease with secondary cardiac involvement.

The degree of impairment of lung function is marked when measured against both internationally used and locally derived reference ranges. Adult HIV patients in the US have higher rates of asthma than the general population[26](#_ENREF_26), but the generalizability to our age group and geography is uncertain. The ISAAC study[27](#_ENREF_27) did not cover Malawi, but prevalence of wheeze was 15.9% in English speaking African countries. In our study, bronchodilator reversibility was minimal and rates of wheeze were similar to regional rates in the general population suggesting asthma was unlikely to be a predominant pathology. FEV1 improvement after inhaled bronchodilator was disappointing, and on average was indistinguishable from zero. It is therefore possible that even those with >12% increase in FEV1 may represent bias related to regression to the mean. No participants found salbutamol helpful at four weeks: effective treatment options are urgently needed.

Within our cohort, there is no strong evidence for clinically significant decline in lung function with age. However, this could be confounded by age of ART initiation, and a longitudinal study to specifically examine this is in progress. Declining FEV1 is reported in chronic lung diseases such as cystic fibrosis and COPD. In other cohorts (chronic coughers with bronchiectasis which presented in childhood), FEV1 declined with age, but this was apparent only after many years[28](#_ENREF_28). Some decline may be artefactual relating to growth and maturationdelay, although the significant baseline abnormality suggests that earlier life events have already strongly affected the lung architecture. In any case, the lack of clinical predictors of lung function abnormality suggests that considerable lung abnormalities, through intercurrent disease or other effects on lung growth, may not be identified unless spirometry is performed. Rates of reported household biomass fuel use were typical for many sub-Saharan countries. This important public health problem may have contributed to reduced lung function in our population[29](#_ENREF_29).

Our data are limited by the cross-sectional nature of the study and the lack of total lung volume and transfer factor measurements, and the absence of non-infected controls. Reversibility studies might be more easily interpreted with either universal reversibility testing, or a control arm but this was not possible within our study. We did not have access to HRCT imaging, echocardiography or post-mortem tissue biopsies which would define the pathologies more clearly. and TB screening is limited by suboptimal diagnostics.

Prospective studies should examine our definition of the two phenotypes in relation to pathophysiology in a cohort in which intensive investigation is possible, for example with high resolution CT scanning, echocardiography and possibly, autopsy studies. If the phenotypes correlate with disease (we hypothesise “cough” with bronchiectasis or bronchiolitis obliterans, and “hypoxia” with interstitial lung disease), this could be useful to clinicians where such investigations are not available. Longitudinal cohort studies should assess longterm change in symptoms and lung function in CLD, and would facilitate therapeutic trials of immunomodulation (for example prednisolone in obliterative bronchiolitis) or antimicrobials (azithromycin in bronchiectasis).

Widespread evidence of pulmonary disease presented here adds to the case for treatment of all HIV infected children with antiretrovirals irrespective of CD4 count. At the least, as a WHO HIV Stage 3 criterion, there should be a strong emphasis on identifying children with chronic lung disease and establishing early antiretroviral therapy in those individuals. For this purpose, simple clinical definitions of CLD-cough (in the absence of TB) and CLD-hypoxia could be useful to clinicians in healthcare settings with few resources.

### Conflicts of interest

The authors declare no conflict of interest.

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## Table Demographic and clinical characteristics

|  |  |
| --- | --- |
|  | **n (%) unless stated** |
| Age, *median years (IQR)* | 11.1 (9.5-12.4) |
| Sex, female *(%)* | 80 (50.0%) |
| Age at HIV diagnosis, *median years (IQR)* | 7.9 (5.8-9.8) |
| Child aware of HIV diagnosisa, n (%) | 65 (41.1) |
| CD4 countc, *median cells/µL (IQR)* | 572 (370-876) |
| Taking co-trimoxazole prophylaxis, n (%) | 159 (99.4%) |
| On ARTa, n (%) | 114 (71.7%) |
| Duration of ARTb, *median years (IQR)* | 3.5 (1.3-4.6) |
| Chest infection in preceding year, *n (%)* | 30 (18.8%) |
| **Symptoms** |  |
| Cough, *n (%)* | 60 (37.5) |
| Sputum produced, *n (%)* | 32 (20.0) |
| Wheezing in last 12 months, *n (%)* | 13 (8.1) |
| Breathlessness (NYHA class), *n (%)* |  |
| 0 | 85 (53.1) |
| 1 | 9 (5.6) |
| 2 | 11 (6.9) |
| 3 | 9 (5.6) |
| 4 | 46 (28.8) |
| **Examination** |  |
| Stunted [HFA z<-2], *n (%)* | 89 (55.6%) |
| WFH z-score, *mean (SD)* | -0.82 (±1.09) |
| Finger clubbing, *n (%)* | 34 (22.1) |
| Resting pulse rate*, median min-1 (IQR)* | 87.0 (76.0-98.5) |
| Resting tachypnoea[>24/min], *n (%)* | 57 (35.6) |
| Resting hypoxia [SpO2<92%], *n (%)* | 33 (20.6) |
| Normoxemia but desaturates >4%, *n (%)* | 29 (18.1) |

Continuous data are represented as median (IQR). a missing data n=1; b unknown n=15; c data unavailable n=3. WFH = weight for height

### Table Risk factors for CLD defined by presence of cough and hypoxia

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  | **Presence of cough** | | | | **Presence of hypoxia or desaturation** | | | |
|  | Univariate  OR (95% CI) | p | Multivariate a  OR (95% CI) | p | Univariate  OR (95% CI) | p | Multivariate a  OR (95% CI) | p |
| **Distal (indirect) factors** |  |  |  |  |  |  |  |  |
| Sex, female | 0.72 (0.38-1.38) | 0.322 |  |  | 1.24 (0.65-2.35) | 0.514 |  |  |
| Age, years | 0.99 (0.84-1.16) | 0.883 | 1.00 (0.81-1.23) | 0.98 | 0.97 (0.82-1.14) | 0.68 | 0.95 (0.79-1.13) | 0.54 |
| Orphaned (1+ parent died) | 0.65 (0.34-1.24) | 0.192 |  |  | 1.36 (0.71-2.6) | 0.348 |  |  |
| Height-for-Age (HFA), z-score | 0.85 (0.64-1.12) | 0.25 |  |  | 0.90 (0.68-1.18) | 0.451 |  |  |
| Weight-for-Height (WFH), z-score | 0.96 (0.71-1.30) | 0.798 |  |  | 0.91 (0.67-1.22) | 0.515 |  |  |
| ART prescribed | 0.91 (0.45-1.84) | 0.788 | 0.79 (0.34-1.83) | 0.59 | 0.64 (0.32-1.28) | 0.207 | 0.83 (0.35-1.96) | 0.67 |
| Age at which ART started, years | 0.94 (0.82-1.08) | 0.372 |  |  | 0.92 (0.8-1.06) | 0.227 |  |  |
| CD4 c <100 | 1.93 (0.53 – 7.04) | 0.32 |  |  | 4.03 (0.99-16.39) | 0.051 | 3.87 (0.91-16.42) | 0.066 |
| 100-199 | 1.93 (0.53 – 7.04) | 0.32 |  |  | 1.72 (0.47-6.30) | 0.41 |  |  |
| 200-349 | 1.50 (0.52-4.31) | 0.45 |  |  | 0.79 (0.26-2.41) | 0.67 |  |  |
| >349 | 1.00 | - |  |  | 1.00 | - |  |  |
| **Proximal (direct) factors** |  |  |  |  |  |  |  |  |
| Previous TB | 1.49 (0.69-3.22) | 0.316 |  |  | 1.51 (0.7-3.24) | 0.295 |  |  |
| Smoker in household | 0.44 (0.15-1.25) | 0.104 |  |  | 0.49 (0.18-1.33) | 0.145 |  |  |
| NYHA grade 3 or 4 | 1.65 (0.84-3.24) | 0.148 |  |  | 0.77 (0.39-1.52) | 0.45 |  |  |
| Wheeze | 11.47 (2.44-53.83) | <0.001 | 6.94 (1.38-34.95) | 0.019 | 0.44 (0.12-1.66) | 0.198 |  |  |
| CFQR Physical domain | 0.98 (0.97-0.99) | <0.001 | 0.98 (0.97-1.00) | 0.017 | 1.00 (0.99-1.01) | 0.623 |  |  |
| Clubbing | 1.19 (0.54-2.63) | 0.666 |  |  | 2.28 (1.04-4.99) | 0.038 | 1.28 (0.50-3.25) | 0.60 |
| Respiratory rate, >25/min at rest | 0.98 (0.50-1.93) | 0.948 |  |  | 3.00 (1.52-5.92) | 0.001 | 2.39 (1.06-5.38) | 0.032 |
| Pulse rate, beats/min | 1.01 (0.99-1.03) | 0.185 |  |  | 1.00 (0.98-1.01) | 0.585 |  |  |
| CXR abnormality b | 3.00 (1.15-7.85) | 0.023 | 3.43 (1.01-11.65) | 0.048 | 1.04 (0.4-2.7) | 0.941 |  |  |
| Abnormal spirometry | 2.61 (1.29-5.28) | 0.007 | 2.09 (0.94-4.66) | 0.072 | 1.65 (0.83-3.28) | 0.153 |  |  |

OR: odds ratio, CI: confidence interval, NYHA: New York Heart Association breathlessness scale, ART: Antiretroviral Therapy, a =adjusted for priori variables: age, sex and being on ART and significant distal and proximal variables. b=defined here as consolidation, volume loss or lymphadenopathy as other findings were non-discriminatory. c=comparator population is those with CD4>349. Participants diagnosed with pulmonary tuberculosis are not included in this analysis.

### Table Spirometric indices

|  |  |  |
| --- | --- | --- |
| **Baseline spirometry (n=145)** | **GLI reference**[**10**](#_ENREF_10) | **Local reference** |
| FEV1 | -1.31 (-2.10 to -0.27) \* | 92.2 (79.5 to 104.6) † |
| FVC | -0.89 (-1.91 to -0.18) \* | 93.9 (81.8 to 104.2) † |
| FEV1/FVC | -0.27 (-1.21 to 0.35) \* | 87.9 (82.1 to 91.6) ‡ |
| FEF25-75% | -0.69 (-1.63 to 0.38) \* | not available |
| No abnormality, n (%) | 90 (62.1%) | 102 (70.3%) |
| Obstruction, n (%) | 26 (17.9%) | 18 (12.4%) |
| Reduced FVC, n (%) | 29 (20.0%) | 25 (17.2%) |
| **Reversibility testing** | **FEV % change** | **Reversible, *n (%)*** |
| Reduced FVC pattern (n=26) | 2.6 (-3.6 to 9.5) | 8 (30.8%) |
| Obstructive pattern (n=21) | 3.3 (-4.3 to 12.1) | 7 (33.3%) |
| All (n=47) | 2.7 (-4.3 to 10.1) | 15 (31.9%) |

Continuous data presented as median (IQR) due to skewed distributions. Includes all traces meeting ATS criteria (grades A and B), n=145 at baseline, n=47 for reversibility testing.

\* median z-score (IQR).† median percentage of predicted (IQR). ‡ median percentage (IQR)

### Figure Study flowchart

Flow diagram illustrates participant retention and quality of spirometry throughout the study.

### Figure Spirometry results overview

Graphs illustrating the degree and distribution of spirometric abnormality. **Panel A:** Boxes represent 25 and 75th centiles, whiskers represent 10 and 90th centiles, with outliers shown as individual dots. ULN and LLN are upper and lower limits of normal respectively, drawn by dashed line at +1.64SD and -1.64SD from the mean. **Panel B:** FEV z-score for all participants as a function of age. Linear regression model is shown as a solid line. There is a non-significant tendency to reducing FEV1 with increasing age in this cohort (r2 = 0.026, p=0.054). Similar results for FVC obtained (results not shown).

### Figure Proposed CLD phenotypes

Proportional areas diagram illustrating the proposed CLD phenotypes, “Cough” and “Hypoxia”, and their overlap with individuals with abnormal spirometry. Percentages indicate the proportion of the entire study population for which spirometry data were available.