**TITLE PAGE**

**Title:** Pulmonary function profile of children with sickle cell anaemia in Steady State in Lagos, Nigeria

**Running title:** Lung function status of children with sickle cell anaemia

**ABSTRACT**

**Background:** Respiratory disordersare responsible for considerable morbidity and mortality in children with sickle cell anaemia. Spirometry is a useful tool for detection and monitoring of respiratory disorders, but it is underused by healthcare workers treating children with sickle cell anaemia. Most of the studies assessing pulmonary function in sickle cell anaemia were done among adult patients.

**Objective:** To describe the lung function profile of children with sickle cell anaemia in steady state.

**Methodology:** In this study, spirometric indices of 100 HbSS aged 5 years to 12 years were compared with 100 matched HbAA control group.

**Results:** Irrespective of gender, meanPeak Expiratory Flow Rate (PEFR) values were significantly higher in HbAA controls than their HbSS counterparts. The mean Forced Expiratory Volume in one second **(**FEV1) values of males and all subjects irrespective of gender were also significantly higher among controls compared to sickle cell anaemia subjects. The mean Forced Vital Capacity (FVC) values were higher in HbSS subjects than their HbAA controls but the observed differences were not significant. The mean FEV1/FVC values were also not significantly different for Hb AA subjects and their counterparts with Hb SS. The overall prevalence of restrictive pulmonary abnormalities among AA controls and SS subjects 0.0% Vs 6.0% respectively.

**Conclusion:** Children with sickle cell anaemia irrespective of gender have significant lower PEFR and FEV1, implying relatively lower amount of airflow out of the large airways of the lungs. Restrictive lung abnormalities occur commonly among subjects with sickle cell anaemia while there is rarity of obstructive abnormalities.

**Key words:** spirometry; obstructive lung disease; restrictive lung disease; sickle cell anaemia

**INTRODUCTION**

Sickle cell disease is one of the commonest genetic disorders worldwide and is the most common inherited haematological disease affecting man1. This condition is inherited as an autosomal recessive disorder. The homozygous state otherwise known, as sickle cell anaemia is a lifelong disorder affecting about 2-3% of Newborns2 especially children, and this makes it a major health problem in Nigeria. With the population of Nigeria at about 140 million and of which about 50% are children,3 it is estimated that there are about 4.2 million Nigerians suffering from sickle cell anaemia.

One major mechanism in the pathophysiology of sickle cell anaemia is the sickling phenomenon in which there is obstruction of the microvasculature by abnormally shaped (sickle) red blood cells. This phenomenon affects all organs and tissues of the body including the lungs.4 Recurrence of these sickling phenomenon may lead to deterioration of lung function as a result of potential complications such as lung fibrosis, chronic hypoxia and pulmonary hypertension.5 In the lungs, sickle cell anaemia is associated with complications such as acute chest syndrome, pulmonary thromboembolism, pulmonary fat embolism and lung fibrosis. These abnormalities also referred to as sickle cell chronic lung diseases (SCCLD) are often heralded by recurrent acute chest syndrome in late infancy and early childhood. It is progressive in nature and continues into adulthood causing pulmonary hypertension, heart diseases and death. 6,7 Lung function abnormality in sickle cell anaemia can be obstructive, restrictive or both. The exact prevalence of SCCLD and method of diagnosis of SCCLD have not been established owing to lack of detailed epidemiological studies.

In Nigeria, there have been studies of lung function in apparently healthy children and in children with asthma. On the contrary, limited studies are available on lung function of children with sickle cell anaemia. In one of the few available studies, Vanderjagt et al8 studied lung function in children and young adults attending the sickle cell clinic at the Federal Medical Center, Gombe and the Jos University Teaching Hospital, Jos and concluded that abnormal lung function can occur in Nigerian children with sickle cell disease compared to controls.

With improved management, the life expectancy of SCA patients has increased significantly over the years. Consequently, complications of the disease associated with long-standing illness would expectedly increase in prevalence also. Pulmonary complications account for significant morbidity and mortality in patients with sickle cell disease. These complications can cause deterioration in lung function that can impact negatively on the quality of life of patients and may lead to mortality if not detected early and managed appropriately. A study of lung function in children with sickle cell anaemia is desirable for elucidating the pathophysiology of the disease. This will enable better understanding of progression of morbidity and clinical problems associated with this disease.

**METHODS**

The study was hospital-based and the primary subjects were sickle cell anaemia patients (aged five to 12 years) diagnosed by cellulose acetate electrophoresis, attending the Paediatric sickle cell clinic at Lagos State University Teaching Hospital (LASUTH). Apparently healthy (children who have no symptoms or sign attributable to an acute illness in the preceding four weeks), age and sex-matched children that met the inclusion criteria and have haemoglobin genotype AA, attending routine follow-up Paediatrics clinics were recruited as controls.

**Inclusion criteria for sickle cell anaemia subjects**

1. Confirmed HbSS by electrophoresis.
2. Age five to 12years
3. Subjects who were in a steady state - steady state is defined as absence of any crisis in the preceding four weeks, no recent drop in the haemoglobin level and absence of any symptoms or sign attributable to an acute illness9
4. No acute illness such as coryza and pneumonia for at least four weeks prior to recruitment
5. No medical history suggestive of bronchial asthma

**Exclusion criteria for sickle cell anaemia subjects**

1. Structural abnormality of the rib cage
2. Denial of consent
3. Patients on drugs that can affect lung function indices such as steroids.
4. Patients with overt mental subnormality; HIV; Oedema; Heart failure

**Inclusion criteria for controls (genotype AA) subjects**

1. Age five to 12years
2. Confirmed Hb AA by Hb electrophoresis
3. Subjects who had no symptoms or sign attributable to an acute illness in the preceding four weeks.

**Exclusion criteria for controls (genotype AA) subjects**

1. Structural abnormality of the rib cage
2. Denial of consent
3. Patients on drugs that can affect lung function indices such as steroids.
4. Patients with overt mental sub-normality; HIV; Oedema; Heart failure
5. Children with acute illness

The minimum sample size to measure the proportion of sickle cell anaemia children with lung function abnormality as against children without sickle cell anaemia was calculated using the formula:10 N = ( Zα/2 + Z1 - β )2 (σ12+σ22) /µ2

Where,

N = estimated sample size

Z1 - β = One sided percentage point of the normal distribution corresponding to 100% minus power, (1.96 for 95% power)

1 – β = power = 95%

Zα/2 = percentage point of the normal distribution corresponding to the (two sided) significance level = 1.96 (95% level of significance)

σ1 =standard deviation for cases which is 0.194

σ2 = standard deviation for controls which is 0.241

µ = the difference to be detected between the means of the two samples = 0.15

The average monthly clinic attendance of sickle cell anaemia patient was 110 out of which there were about 60 children aged 5 - 12 years. Further stratification of the 60 patients showed that about 18 were 5 - 6 years; about 18 were 7 - 8 years; 12 were 9 - 10 years while 12 were11 - 12 years. This gives a distribution ratio of 3: 3: 2:2. The average male to female ratio in these distributions was about 1.1: 1

In order to avoid lopsided clustering of subjects around a particular age or sex, the calculated sample size was stratified according to the distribution pattern of patients in the follow up clinic.

All patients with sickle cell anaemia attending routine Sickle Cell Anaemia follow up clinic within the study period who satisfied the study criteria were recruited. Healthy controls that satisfied the study criteria were recruited from the Dermatology and other follow up clinics. They were matched in respect of, age and sex with the sickle cell anaemia study group.

Ethical approval was obtained from the Ethics Committee of LASUTH prior to the commencement of research. Subjects were recruited following detailed explanation about the research. All recruitments were strictly voluntary and backed by written informed consent.

Data were taken from both the caregiver and patient. The questionnaire was administered principally in English language and translated to the local languages including Pidgin English as each case demanded. However, the consistency of all the content was maintained following translation. Thereafter, patients were physically examined and the findings entered into the study proforma. The physical examination was to:

1. Exclude respiratory or cardiovascular disorders like pneumonia or congenital heart disease
2. Exclude structural rib and spinal deformities
3. Take relevant anthropometric measurement

Socio-economic status of each subject was determined using the Oyedeji11 method of classification. Thereafter they were classified into upper (social-economic class 1 and II), middle (social-economic class III) and lower (socio economic class IV and V) socioeconomic strata.

**Measurement of Forced Expiratory Volume in one second (FEV1).** F**orced Vital Capacity (FVC) and Peak Expiratory Flow Rate (PEFR):** This was done with One Flow Spirometer (Clement Clerk International, England). The author did the spirometry after receiving training at the chest clinic where spirometry is regularly done under the supervision of chest physician. Hygiene was strictly observed by using one mouthpiece per patient. A Universal Sterilizable Mouthpiece with mouth end diameter of 22 millimeter was used. At the end of each day the mouth pieces were rinsed with cetrimide and thereafter autoclaved for the next day’s use. Measurement of FEV1. FVC and PEFR were with the One Flow Spirometer.

**Spirometric tests:** were performed in the Consultant Outpatient Clinics for subjects and controls using the One-flow spirometer (Clement Clerk International, England). One flow spirometer is a digital light weight hand held spirometer that can be held comfortably by a child of 5 years and older. It has an inlet through which air can blow through into the spirometer. It also has a small digital screen from where the values obtained can be read. The control of the spirometer is through the buttons on the side of the screen. The spirometer is battery-powered with an advantage that patients will not be delayed during public power outage. A spare battery was kept handy in case of low battery.

Detachable Universal Sterilizable Mouthpieces were included in the spirometer pack. The spirometer can also be connected to a personal computer to store values and to print values obtained. The spirometer is quick to operate, and does not scare children. Displaying of the value on a digital screen also encouraged the children to do better in subsequent manoeuvre.

Respiratory function tests were performed using spirometry system to measure FEV1, PEFR, FVC, and FEV1/FVC. Three completed attempts were recorded. The highest of the best three readings was taken as the result for the parameter.12 Predicted FEV1, PEFR, FVC, and FEV1/FVC values, using formulae obtained for normal Nigerian Children13, were compared to the observed values for the purpose of assessing pulmonary function test.

The pattern of ventilation which spirometry results show may be normal, obstructive, restrictive, or mixed. The pulmonary function of each subject and control was classified into four categories based on American Thoracic Society criteria14: –

* If FVC and FEV1 are not less than 80% of predicted value and FEV1/FVC ratio less than 70% below the predicted ratio, it is normal.
* A reduction in FEV1 less than 80% of predicted value for age, sex, height and FEV1/FVC ratio less than 70% below the predicted ratio is diagnostic of airway obstruction.
* A reduction in FVC less than 80% of predicted value for age, sex, height and race in the presence of FEV1/FVC ratio not less than 70% is suggestive of restrictive disease.
* Mixed obstructive/restrictive lung disease is suspected when there is a reduction in FVC less than 80% of predicted value for age, sex, height and race in the presence of FEV1/FVC ratio less than 70%.

All data collected were entered into a standard proforma. The statistical analysis was done using the SPSS software version 17.0 Continuous variables were expressed as mean +/- standard deviation (SD) and categorical variables as percentages. Differences in categorical variables were assessed by chi-square analysis while the Student t-test was used for the comparison of continuous variable. Level of significance was set at p = < 0.05.

**RESULTS**

**Characteristics of the study populations**

A total of 200 children, 100 each withgenotype SS and AA respectively, who met the study criteria, were recruitedover a study period of four months. The demographic characteristics of the studypatients are given in Table I. The age of the subjects ranged from five to twelve years. Age groups five to six years and seven to eight years accounted for 30% each of the total subjects, while age groups nine to ten years and eleven to twelve years each accounted for 20% of the study group. Approximately a third (34.0%) of the subjects belonged to the upper socioeconomic strata (Socioeconomic class I and II), while 45% and 21% belonged to the middle (Socioeconomic class III) and lower (Socioeconomic class IV and V) socioeconomic strata respectively.

**Pulmonary function tests of subjects**

Table II shows pulmonary function test values of subjects. In both males and females, the mean PEFR values were higher among haemoglobin genotype AA controls than their SS counterparts but the observed difference was not significant (p = 0.066 and 0.370 for males and females, respectively). The mean FVC and FEV1/FVC values of males and females were also not significantly different for Hb AA subjects and their counterparts with Hb SS (p > 0.05).

Males with haemoglobin genotype AA had significantly higher FEV1 values when compared with their haemoglobin genotype SS counterparts (p = 0.001). Irrespective of gender, the mean PEFR and FEV1 valueswere significantly higher in subjects with haemoglobin genotype AA when compared with their haemoglobin genotype SS counterparts (p = 0.048 and 0.000 respectively). The lung function indices were comparable in males and females subject with sickle cell anaemia. A similar pattern was also observed among the controls.

**Distribution of individual criteria for diagnosing pulmonary abnormalities among subjects**

Comparison of some selected spirometry parameters which are used as criteria for defining pulmonary abnormalities among subjects according to some sociodemographic characteristics are shown in Table III. The useful variables for spirometry interpretation are FEV1, FVC and FEV1/FVC ratio. Six of the subjects with sickle cell anaemia have abnormal FEV1 and FVC values while the only HbAA controls with abnormal spirometry results had FEV1 values below 80% predicted value. However, it was in the FVC measurement that the observed difference between subjects with sickle cell anaemia and controls was significant (p = 0.029).

**Pulmonary function abnormalities of subjects**

The prevalence of pulmonary function abnormalities seen in spirometry results of subjects is shown in Table IV. Six subjects with HbSS have spirometry results suggestive of restrictive lung defect giving a prevalence of six percent among subjects while none was found among controls. This finding was significant ( p = 0.029). As regard obstructive lung defect, there were none HbSS subjects and controls with spirometry results that are diagnostic. This observed difference was also not significant. There were no subjects with spirometry results diagnostic of mixed obstructive/restrictive lung defect.

**DISCUSSION**

The present data were from children with sickle cell anaemia in steady condition. The mean FVC was lower in SS subjects than AA controls, although the difference was not significant. This finding is consistent with previous studies.15, 16, 17, 18, 19 Endothelial hypoxic injury due to hypoventilation during painful crisis affecting the lungs and the chest wall can eventually result in fibrotic lung injury, and this can reduce FVC in HbSS patients.20

The mean FEV1 among HbSS subjects irrespective of gender in the present study of 1.30L is lower than 1.70L reported by Onigbinde17 among children with sickle cell anaemia at Oshogbo and Ile Ife in 2006. The disparity in the age range of the study subjects may account for the observed difference in mean values; the upper age limit for the present study is 12 years while it was 15 years in the study by Onigbinde18. It is plausible that the lower mean FEV1 observed in present study was because younger children were studied than was the case in the study by Onigbinde18. Since FEV1 is directly related to height, weight, age and lung size, it is therefore expected that older children should have higher values. Additionally, the observed difference could possibly be an effect of the smaller sample size of 74 subjects used in the earlier study as against 100 subjects used in current study. Small sample size is known to give exaggerated sample mean value.21

FEV1, a timed subdivision of FVC in the first second is a measure of air flow rate in the air ways. The mean FEV1 was higher in controls than HbSS subjects. The lower values in SS subjects may be explained by ongoing subclinical inflammatory process within the respiratory system of children with sickle cell anaemia.22, 23 The ongoing inflammation may result in airway hyper-reactivity thereby causing spasm of bronchial smooth muscle and consequently reducing the diameter of the bronchioles. The reduced bronchial diameter result in reduced airflow rate which translate to reduced FEV1.

The mean PEFR was lower among HbSS subjects than HbAA controls. This corroborates reports from earlier studies.7, 8, 16, 18, 19, 24 This is because PEFR is dependent on lung size, lung compliance, rib cage size and mobility. Reduced values therefore, could be due to repeated pulmonary hypoxic injury from pneumonia, acute chest syndrome and pulmonary infarction associated with sickle cell anaemia.19

The study aimed to determine the pattern of lung function abnormalities among children with SCA. The two types of abnormalities identified were obstructive and restrictive. The basis for obstructive disease in HbSS patients is in chronic hypoxia and ongoing subclinical inflammation in the respiratory tract of children with SCA. Obstructive diseases affect the rate at which air can move through the lungs.

The zero prevalence of obstructive defect herein reported agrees with an earlier study conducted in southwestern Nigeria, which also reported zero prevalence of obstructive lung defect.18 On the other hand, Sylvester et al17 reported a prevalence of obstructive defect of 4.5% among black Americans. In an another study done among African–Americans,24 a much higher prevalence rate of 35% was reported. It is noteworthy that the upper age limit in the African–Americans study24 was 15 years (three years older than the subjects of the current study). It is however doubtful if that factor would be enough to explain the large difference in prevalence rates. Also, a much smaller sample size of 63 was used in the African–Americans study, which has the risk of generating high prevalence rates.21 Another explanation for this may be racial differences. Race is an important determinant of lung function. Previous studies have demonstrated that, on average, equations for predicted values of pulmonary function test data based on data generated in white subjects overestimate the FEV1, and FVC by 12%.25 Although all our data were corrected for race before analysis, there may still be a differential pattern present.

With respect to restrictive abnormalities, six subjects with sickle cell anaemia were identified with the disorder in the study. Restrictive lung defect means that there is interference with the ability of the lungs to expand Restrictive lung dysfunction probably reflects the effect of recurrent vaso-occlusive crisis affecting the lung and the rib cage. The resulting lung fibrosis and rib infarction restrict lung expansibility thereby reducing its volumes and capacities. The six percent prevalence of restrictive lung abnormality in current study is in line with the findings of the study conducted by Sylvester et al17 in which 6.3% of children studied had restrictive lung abnormality. This pattern of lung dysfunction in children with sickle cell anaemia is still a matter of continuing investigation. While some workers have reported high prevalence in children and adolescents, 18, 20, 22 others have emphasized its rarity.18

In conclusion, the PEFR and FEV1 are significantly lower among HbSS subjects compared to their HbAA counterparts. The prevalence of restrictive defect results was 6.0% among subjects with sickle cell anaemia. This work has shown that the most common pulmonary function abnormality among subjects with sickle cell anaemia was restrictive lung defect. There is a need for routine evaluation of pulmonary functions during follow-up visit of children with sickle cell anaemia to ensure early detection of lung volume abnormalities and subsequent intervention to correct such abnormalities.

**LIMITATIONS**

Plethysmography, a vital tool in the diagnosis of restrictive lung abnormality was not used because it is not available at the study centre. Spirometry, while excellent for the diagnosis of obstructive disease is at best, a screening tool for restrictive disease.

Predicted standards of lung function indices for children in the sub-region are yet to be generated. Consequently, the observed values among subjects in current study were compared with predicted values derived using the values of the Nigerian school children derived by Oduwole et al which are similar to the controls.13

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**TABLES**

**Table I: Demographic characteristics of study population**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Characteristic** |  | **AA**  **N (%)** | **SS**  **N (%)** | **p - value** |
| Gender  Age in years  Male  Female  Socioeconomic strata | Male  Female  5 - 6  7 – 8  9 - 10  11 - 12  5 - 6  7 – 8  9 - 10  11 - 12  Upper  Middle  Lower | 50 (50.0)  50 (50.0)  15 (15.0)  15 (15.0)  10 (10.0)  10 (10.0)  15 (15.0)  15 (15.0)  10 (10.0)  10 (10.0)  44 (44.0)  39 (39.0)  17 (17.0) | 50 (50.0)  50 (50.0)  15 (15.0)  15 (15.0)  10 (10.0)  10 (10.0)  15 (15.0)  15 (15.0)  10 (10.0)  10 (10.0)  24 (24.0)  51 (51.0)  25 (25.0) | 1.000  1.000  1.000  0.011 |

**Table II: Pulmonary function test values of study subjects**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Pulmonary Function Test** | **SS**  **Mean (SD)** | **AA**  **Mean (SD)** | **t-value** | **p - value** |
| PEFR ( L/m)  Males  Females  Males and Females    FEV1 (L)  Males  Females  Males and Females    FVC (L)  Males  Females  Males and Females  FEV1 (%)  FVC  Males  Females  Males and Females | 213.70(44.40)  216.20(56.58)  214.95(50.62)  1.28(0.24)  1.33(0.36)  1.30(0.31)  4.03(17.89)  4.12(18.17)  4.07(17.94)  85.88(5.12)  85.62(4.81)  85.75(4.95) | 234.30(60.64)  226.30(55.61)  230.30(58.03)  1.48(0.34)  1.48(0.44)  1.48(0.39)  1.70(0.43)  1.71(0.50)  1.71(0.47)  87.60(4.78)  86.28(4.59)  86.94(4.71) | 1.938  0.900  1.994  3.416  1.879  3.532  0.920  0.937  1.320  1.736  0.702  1.742 | 0.066  0.370  0.048\*  0.001\*  0.063  0.000\*  0.360  0.351  0.188  0.086  0.485  0.083 |

SD standard deviation, \* statistically significant; n= number in group

**Table III: Distribution of individual criteria for diagnosing pulmonary abnormalities among study subjects**

|  |  |  |  |
| --- | --- | --- | --- |
| **Pulmonary Function Test** | **SS** | **AA** | **p-value** |
| FEV1 (%)  <80% of predicted value  ≥80% of predicted value    FVC (%)  <80% of predicted value  ≥80% of predicted value    FEV1 (%)  FVC <70% of predicted value  ≥70% of predicted value | 6 (6.0)  94 (94.0)  6 (6.0)  94 (94.0)  0 (0.0)  100 (100.0) | 1 (1.0)  99 (99.0)  0 (0.0)  100 (100.0)  0 (0.0)  100 (100.0) | 0.118  0.029\*  1.000 |

\* Statistically significant

**Table IV – Prevalence of Pulmonary Function Abnormalities among study subjects**

|  |  |  |  |
| --- | --- | --- | --- |
| **Abnormal lung function** | **SS** | **AA** | **p-value** |
| Obstructive Lung Defect  Affected    Not affected    Restrictive Lung Defect    Affected    Not affected | 0 (0.0)  100 (100.0)  6 (6.0)  94 (94.0) | 0 (0.0)  100 (100.0)  0 (0.0)  100 (100.0) | 1.000  0.029 |

NB: Values in parenthesis are % of number in group