eISSN 2255-0569

ORIGINAL

Assessment of pubertal delay in adolescents with sickle cell anemia in two health facilities in Cameroon

Evaluación del retraso puberal en adolescentes con anemia falciforme en dos centros de salud de Camerún

Andreas Chiabi[1], Ladwena Shuymuh[1], Cecilia Fomenky[1], Kate Kan[1], Denis Nsame[2]

1. Department of Clinical Sciences, Faculty of Health Sciences, University of Bamenda.
2. Department of Pediatrics, Regional Hospital Bamenda, Cameroon

Corresponding author
Andreas Chiabi
E-mail: andy_chiabi@yahoo.co.uk

Received: 8 - I - 2023
Accepted: 10 - II - 2023

doi: 10.3306/AJHS.2023.38.03.94

Abstract

Objectives: Onset and progression of puberty is reported to be delayed in children with sickle cell anemia. This study sought to determine the prevalence, assess pubertal delay and evaluate the factors associated with pubertal delay in adolescents with SCA.
Methods: We conducted a cross-sectional analytic study on 72 sickle cell adolescents, at the sickle cell clinics of the Regional Hospital Bamenda and Nkwen Baptist Hospital in Cameroon, from 15th February 2022 to 20th June 2022. We assessed sociodemographic variables, clinical and Tanner stages of the participants. Chi square test was used to test for associations, and statistical significance was set at $p<0.05$.
Results: Pubertal delay was found in 48.5% of participants with a male predominance of 75.0%. The mean age at onset of puberty (B2) in girls with delay was 13.3±1.4 years compared to 15±1.4 years in those without delay, with observe difference statistically significant ($p=$ ***0.001***). The mean age of menarche was delayed by 2.3 years in sicklers with delay. Amongst males, the mean age at onset of puberty (G2) was 14.6±1.5 in sicklers with delay compared to 16.9±1.9 in sicklers with no delay and was statistically significant (***p*=0.001**). Amongst the factors evaluated receiving three or more blood transfusions was significantly associated with pubertal delay. (AOR= 5.7(1.7-19.2) and $p=$**0.01**).
Conclusion: A high prevalence of pubertal delay (48.5%) was recorded amongst children with sickle cell anemia. The mean age of menarche was delayed by 2.3 years in sicklers with pubertal delay. Three or more transfusions were significantly associated with pubertal delay.

Key words: Pubertal delay, Adolescents, Sickle cell anemia, Assessment.

Resumen

Objetivos: Se informa que el inicio y la progresión de la pubertad se retrasan en niños con anemia de células falciformes. Este estudio buscó determinar la prevalencia, evaluar el retraso puberal y evaluar los factores asociados al retraso puberal en adolescentes con SCA.
Métodos: Realizamos un estudio analítico transversal en 72 adolescentes con anemia falciforme, en las clínicas de anemia falciforme del Hospital Regional Bamenda y el Hospital Bautista Nkwen en Camerún, del 15 de febrero de 2022 al 20 de junio de 2022. Evaluamos variables sociodemográficas, etapas clínicas y de Tanner de los participantes Se usó la prueba de chi cuadrado para probar las asociaciones y la significación estadística se fijó en p<0,05.
Resultados: Se encontró retraso puberal en el 48,5% de los participantes con un predominio masculino del 75,0%. La edad media de inicio de la pubertad (B2) en las niñas con retraso fue de 13,3±1,4 años en comparación con 15±1,4 años en las que no tenían retraso, observándose una diferencia estadísticamente significativa (p=0,001). La edad media de la menarquia se retrasó 2,3 años en las falciformes con retraso. Entre los hombres, la edad media de inicio de la pubertad (G2) fue de 14,6 ± 1,5 en las falciformes con retraso en comparación con 16,9 ± 1,9 en las falciformes sin retraso y fue estadísticamente significativa (p=0,001). Entre los factores evaluados, recibir tres o más transfusiones de sangre se asoció significativamente con el retraso puberal. (ORA= 5,7(1,7-19,2) y p=0,01).
Conclusión: Se registró una alta prevalencia de retraso puberal (48,5%) entre los niños con anemia de células falciformes. La edad media de la menarquia se retrasó 2,3 años en las drepanocíticas con retraso puberal. Tres o más transfusiones se asociaron significativamente con retraso puberal.

Palabras clave: Retraso puberal, Adolescentes, Anemia de células falciformes, Evaluación.

Introduction

Sickle cell anaemia (SCA) is an autosomal recessive disease characterized by crescent- or sickled shape red blood cells due to a point mutation in the beta globin chain of haemoglobin (Hb) in chromosome 11 causing the hydrophilic glutamic acid to be replaced with the hydrophobic valine at the 6th position[1,2]. According to World Health Organisation(WHO) about 5% of the world's population carry genes for haemoglobinopathies and 2.9% carries mutation for SCA[3,4].In a state of low oxygen, decreased pH, dehydration and infection, erythrocytes alter their shape invivo and undergo repeated cycle of sickling and unsickling until they become irreversibly sickled. This in turn reduce blood supply to tissues leading to organ malfunction as well as complications involving many systems, which in the long run affects development[5-7].

The clinical manifestations of SCA most often begins as from 6 months, with painful swelling of hands and or feet, severe painful crises, recurrent infections, bone and joint necrosis and priapism[2]. Moreover, deleterious consequences such as vaso-occlusion, haemolysis, infection and organ dysfunction occur in these set of population and delayed puberty happens to be one of the prominent complications SCA patients experienced. Pubertal delay is the absence of breast development in girls by 13 years of age and absence of testicular growth to at least 4mL in volume or 2.5 cm in length in boys by 14 years of age[8]. Pubertal changes occur at different ages depending on various genetic and environmental influence. Amongst children with sickle cell anaemia the onset of puberty is affected mainly due to chronic hypoxia from recurrent vaso-occlusive events which together with chronic anaemia causes hypoplasia of pituitary gland and gonads. Consequently, altering the process that initiate complex neuroendocrine machinery for the pulsatile secretion of gonadotropin releasing hormone(GnRH) in the hypothalamic pituitary-gonadal (HPG) axis[8,9]. In addition, the toxicity of iron and increase basal metabolism secondary to haemolysis and multiple transfusions also affects these axis hence delayed pubertal development[9,10].

In South America, Gomes et al, carried-out a study on Growth and puberty in a prospective cohort of patients with SCA and found that, patients with SCA showed growth impairment and pubertal delay of 46.66% compared with 6.66% of healthy controls[11]. Uchendu et al in Nigeria in a study to evaluate sexual maturity among adolescent males, SCA patients showed delayed onset and completion of sexual maturation compared to others[12]. A similar study by Betoko et al in Yaounde on puberty during sickle cell anemia noted that 27.3% of girls and 10% of boys with SCA had delayed puberty with recuurent infections and low Hb being the leading associated factors during the study[9].

So far no study on puberty in sickle cell anemia has been done in the North West Region of Cameroon.Therefore this study will focus in determining the prevalence, assessing pubertal delay and evaluating the factors associated with pubertal delay in adolescents with SCA in the Regional Hospital Bamenda (RHB) and Nkwen Baptist Hospital (NBH).

Materials and methods

Study Setting

This study was conducted in the general paediatric units of 2 hospitals in Bamenda (the Regional Hospital Bamenda and Nkwen Baptist Hospital) found in the North West Region of Cameroon. These two hospitals have specialised units for the follow up of sickle cell patients.

Study design, population and selection criteria

Cross-sectional analytic study, at the general pediatric ward of the Regional Hospital Bamenda and Nkwen Baptist hospital specifically in the sickle cell units. We assessed pubertal delay in adolescents with sickle cell anemia. This study was carried out over a period of 4 months,15th February to 20th June 2022. The study population comprised all SCA patients being followed up at the sickle cell clinic of the aforementioned hospitals during the study period. A consecutive sampling method was used in recruiting the study population.

Included in our study, were children in the age group of 8-19 years whose genotypes have been confirmed by Hb electrophoresis and are regularly followed up in the RHB and NBH. The age group of 8-9 years was chosen because in Cameroon, puberty usually begins as from the age of 8 and 9 years in girls and boys respectively, and progresses to adolescence at 19 years. We excluded chronic pathologies (heart failure, HIV, renal pathologies), and those who did not give in their consent.

Data collection

For each participant that was eligible for the study, a verbal interview was done using an adequately pre-designed questionnaire to interview participants. The information gotten from the participants and /or caregivers and parameters measured were then filled by the investigator into the questionnaire. A total of 85 participants were approached. After elimination using both the inclusion and exclusion criteria, 72 participants were included in the study.

Data analysis

Data were entered into SPSS for windows version 23.0, Microsoft excel sheet 2021, Epi info version 7 and WHO Anthro version 3.2.2 statistical software for analysis and analysis was done. Chi-square test was used to compare categorical variables while the student-t test

was use to compare the mean values. A p-value < 0.05 was considered as statistically significant.

Ethical considerations

Ethical clearance was obtained from the institutional Review board of Nkwen Baptist hospital and of the Faculty of Health Sciences, University of Bamenda, while administrative authorization was obtained from the North West Regional Delegation of Public Health, as well as directors of the aforementioned hospitals. Before obtaining information from the participants, their assent and consent were also obtained. Participant anonymity and confidentiality were maintained throughout the period of study.

Results

Majority of the 72 children followed up during the study period were between the age group 10-15years (43.1%), and of male predominance (58.3%), however about 65.3% were underweight (**Table I**). Forty were males, and 30 females giving a sex ration of 1.3. In addition, majority of children were diagnosed as early as between the ages 1-5years with about 37.5% presenting with more than 2 crises and more than half already being transfused at least once per year.

Table I: Sociodemographic characteristics of the children.

Variable	SCAa(N=72)	
	Number	%
Age Range (in years)		
[8-10]	25	34.7
]10-15[	31	43.1
≥15	16	22.2
Sex distribution		
Male	42	58.3
Female	30	41.7
Level of education		
None	3	4.2
Primary	41	56.9
Secondary/High	26	36.1
Higher	2	2.8
BMIb		
[<18.5[	47	65.3
[18.5-24.9]	23	31.9
≥25	2	2.8

aSicke Cell Anemia b Body Mass Index

Table II: Mean age at Different Tanner Stages for Breast Development in Girls.

Variable	Number	Percentage (%)	P-Value	Mean (±SDa)	Age	P-Value
	Delay	No Delay		Delay	No delay	
B1	3 (75)	0	***0.04***	14±1.4	0	
B2	1 (25)	2 (25)		13.0±1.4	15±1.4	***0.001***
B3	00	3 (37.5)		00	16.3±1.5	
B4	00	2 (25)		00	17±2.8	
B5	00	1 (12.5)		00	19±0.0	
Total	4 (100)	8 (100)				
Menarche	1 (16.7)	5 (83.3)		14.6±1.5	16.9±1.9	***0.001***

a standard deviation

Girls without delay were older compared to those with delay at the onset of puberty (B2). The mean age at onset of breast development in sicklers with delay was 13.3±1.4 compared to 15.0±1.4 in those with no delay and was statistically significant (P=0.001). The proportions between the overall breast development in delay and no delay was statistically significant(P=0.04) (**Table II**).

In boys, the mean age at Tanner stage 2 of testicular development (G2) in male sicklers with delay was lower (14.8±1.8 years) against 16.0±2.0 years for those without delay with statistically significant value (P=0.001). The proportions between the overall genital development in delay and no delay was statistically significant (P=0.005) (**Table III**). The mean age of Tanner stage 2 pubic hair growth (P2) in sicklers without delay was higher compared to those with delay with a statistically significant (P= 0.001). The proportions between the overall pubic hair development in delay and no delay was statistically significant (P=<0.000) (**Table IV**). About 16.7% of sicklers with delay had their first menstruation compared to 83.3% of sicklers with no delay.

The mean age of menarche was 16.9±1.9 in sicklers with pubertal delay compared to 14.6±1.5 in no delay with a statistical significant of (P = <0.001) (**Table III**). Pubertal delay was noted in 48.5% of sicklers with a high rate recorded in males (75.0%). The mean age (±SD) of all sicklers with pubertal delay was 14.7(±1.53), and it was higher in males (15.0±1.9) compared to females (13.8±1.0).Amongst the factors evaluated, only number of blood transfusion over three statistically influence puberty delay (AOR= 5.7(1.7-19.2) and P=0.01) and were 32 times more prone to PD (**Table V**). None of the sociodemographic factors were associated with pubertal delay (**Table VI**).

Table III: Mean age at Different Stages for Testicular Development.

Variable	Number	Percentage (%)	P-Value	Mean (±SDa)	Age	P-Value
	Delay	No Delay		Delay	No delay	
G1	3 (25)	0	***0.005***	15±1.4	0	
G2	9 (75)	3 (33.3)		14.8±1.8	16.0±2.0	***0.001***
G3	00	1 (11.1)		00	16.0±0	
G4	00	2 (22.2)		00	18.0±1.4	
G5	00	3 (33.3)		00	18.3±1.2	
Total	**12 (100)**	**9 (100)**				

Table IV: Mean age at Different Tanner Stages for Pubic Hair Development.

Variable	Number	Percentage (%)	P-Value	Mean (±SDa)	Age	P-Value
	Delay	No Delay		Delay	No delay	
P1	15(93.8)	0	***0.000***	14.6±1.5	0	-
P2	1(6.2)	8(47.1)		15±0.0	15.6±1.3	***0.001***
P3	00	3(17.6)		00	17.3±2.1	
P4	00	2(11.8)		00	18.0±1.4	
P5	00	4(23.5)		00	18.5±1.0	
	16(100)	17(100)				

Table V: Factors Associated with Delayed Puberty from the Disease.

Variable	Delay N=16[a]	No delay N=17[b]	Binary regression		Multivariate regression	
Variable	**Frequency (%)**	**Frequency (%)**	**OR [95%CI]**	**P-value**	**AOR [95%CI]**	**P-value**
Gender						
Male	12 (60.0)	8 (40.0)	3.4 [0.8-14.8]	0.11	2.7 (0.5-13.1)	0.2
Female	4 (30.8)	9 (69.2)	Reference	Reference		
BMI[c]						
<18.5	13 (65.0)	7 (35.0)	6.2 [1.3-30.2]	***0.02***	5.7 (1.7-19.2)	0.05
≥18.5	3 (23.1)	10(76.9)	Reference	Reference		
Number of transfusions ever received						
≥3	15 (65.2)	8 (34.8)	16.9 (1.8-158.1)	***0.01***	32 (2.2-482.1)	***0.01***
<3	1 (10.0)	9 (90.0)	Reference	Reference		
Number of transfusions per year						
≥3	8 (57.1)	6 (42.9)	1.8 [0.5-7.4]	0.4	1.6 (0.4-6.8)	0.5
<3	8 (42.1)	11 (57.9)	Reference	Reference		
Number of crisis per year						
≥3	8 (53.3)	7 (46.7)	1.4 (0.4-5.7)	0.6	1.5 (0.4-6.0)	0.6
<3	10 (55.6)	8 (44.4)	Reference	Reference		
Hospitalizations per year						
≥3	8 (57.1)	6 (42.9)	1.8 [0.5-7.4]	0.4	1.5 (0.4-6.6)	0.6
<3	8 (42.1)	11 (57.9)	Reference	Reference		
Baseline haemoglobin g/dl						
<7	7 (46.7)	8 (53.3)	0.9 (0.2-3.5)	0.9	1 (0.1-6.0)	0.8
≥7	9 (50.0)	9 (50.0)	Reference	Reference		

Table VI: Sociodemographic Factors Associated with Delayed Puberty.

Variable	Delay N=16*	No delay N=17**	Binary regression		Multivariate regression	
Variable	**Frequency (%)**	**Frequency (%)**	**OR [95%CI]**	**P-value**	**AOR [95%CI]**	**P-value**
Age of PCG						
≤40	9 (40.9)	13 (59.1)	0.4 (0.1-1.8)	0.2	0.2 (0.03-1.5)	0.1
>40	7 (63.6)	4 (36.4)	Reference	Reference		
Level of education						
<Secondary	8 (44.4)	10 (55.6)	0.7 (0.2-2.8)	0.6	0.8 (0.2-3.2)	0.7
≥ Secondary	8 (53.3)	7 (46.7)	Reference	Reference		
Occupation						
Liberal	14 (51.9)	13 (48.1)	2.2 (0.3-13.8)	0.4	1.8 (0.3-12.3)	0.5
Non-liberal	29 (33.3)	4 (66.7)	Reference	Reference		
Marital status						
Single	8 (53.3)	7 (46.7)	1.4 (0.4-5.7)	0.6	1.3 (0.3-5.33)	0.7
Living as couple	8 (44.4)	10 (55.6)	Reference	Reference		
Socioeconomic status						
< Middle	9 (3)	3 (17.6)	5.3 (1.1-26.6)	0.04	5.3 (1.0-29.4)	0.06
≥Middle	7 (57.6)	14 (82.4)	Reference	Reference		

OR=Odds ratio, C. I=Confidence Interval,N=16* and17**: Number of sicklers with and without delay, AOR=Adjusted odds ratio, PCG=Primary care-giver

Discussion

For pubertal assessment, the onset and completion of sexual maturation among sicklers was noted to be delayed. Below the age of 13 years, only about 38.5% of the study population had attained stage 2 for breast and pubic hair. However, there was a relative delay in children with SCA below 13years and 14 years in girls and boys respectively.

The results obtained in our study reported that, girls without delay (13-19 years) at each level of pubertal stages were older than those with delay with a gap of 2.0 years at B2.The mean age of female sicklers with delay at the onset of puberty B2 was 13.0±1.4 years this is lower than the reports by Uchendu et al in Nigeria with a mean age of 14.7±1.5 years[13]. The delay in the onset of puberty amongst girls observed in this study may be due to the deleterious effects of hypoxia and chronic hemolysis on the hypothalamic-pituitary-gonadal axis[8].

The mean age of menarche in sicklers with delay was 14.6±1.5 years. These results were lower than the findings of M'Pemba et al (15.2 years ± 1.6). In addition, sergeant et al in Jamaica girls demonstrated that the mean age at menarche in sicklers was 15.4 years and they noted that greater weight was associated with earlier age at menarche and weight status was a predictive

factor for the age of menarche in their cohort[14].This delay in the onset of menses could be explained by low weight and low BMI identified in SCA patients with delay compared to others in our study. The notion of target weight is described in literature as the determining factor for menarche in girls[15].

Amongst boys, sickle cell anemic patients showed delay in their ages of attainment of corresponding genital stages of development. Boys without delay were older than boys with delay at the onset of puberty (G2) with a gap of 2.8 years. The mean age of male sicklers with delay at the onset of puberty (stage G2) was 14.8±1.8 (G2) and this results were slightly higher than the findings of Uchendu et al in Nigeria who found that onset of puberty in male sicklers started at 14.7±1.5 years[13]. Our results reflect delayed onset of puberty among boys and can be explained by recurrent vaso-occlusive crises which lead to hemolysis and chronic anemia. However multiple blood transfusions and chronic hemolysis may induce iron overload which is toxic for the pituitary gland and gonads[16]. M'Pemba et al found hemoglobin level of less than 7 g / dl to be associated with delayed testicular development in contrast to our study. This may suggest that anemia is a deleterious factor for testicular development. However, a low hemoglobin level exposes the patient to high transfusion requirements which could lead to an iron overload, toxic for gonadal cells[16].

Globally in our study, the overall prevalence of pubertal delay amongst sicklers was reported to be 48.5%. with a male predominance (75%). A previous study in Cameroon in 2019 reported lower findings: 27.3% in girls and 10% in boys[9]. This difference could be explained by a difference in sampling methods, sample size and the age groupings in our study. Our results were similar to the proportions reported in previous studies around the world which varies from 37% to 50% for girls and 28.57-73 % for boys[17,18]. These could be explained by the similar size of their study populations with ours though their study groups included adolescents and young adults who were older than our subjects. This fact may have increased the probability to have a greater proportion of delayed puberty with respect to age groups.

Amongst the factors evaluated, multiple blood transfusions, low BMI and low socioeconomic class were found to be significantly associated with delayed puberty on bivariate analysis and on multivariate analysis we noted that, the number of blood transfusions received remained the only factor associated with pubertal delay. This was similar to the findings of Betoko et al who also found multiple blood transfusions, to be associated with pubertal delay[9]. This could be explained by the fact that high transfusion can cause iron overload, which is toxic for gonadal cells[16] but this was in contrast to a study by Zemel et al in 2007, who found that blood transfusion status had no effect on pubertal development as it rather improves their health and decrease the number of crises[19].

According to literature, many mechanisms are described for delayed puberty in SCA. Chronic hypoxia related to recurrent vaso-occlusive events and chronic anemia lead to hypoplasia of pituitary gland and gonads[8]. On the other hand, iron overload secondary to hemolysis and multiple transfusions also negatively affects the gonadotropic axis[10,16]. Micronutrient deficiency is also mentioned by some authors as part of pathogenesis of delayed puberty. Zemel et al. in 2002 found an improvement in weight and height in pre-pubertal children after zinc supplementation[20].

In conclusion this study indicates a high prevalence of pubertal delay in adolescents with sickle cell anemia. The mean age of menarche was delayed by 2.3 years in sicklers with pubertal delay. Three or more transfusions were significantly associated with pubertal delay. Therefore, psychological counselling on pubertal development during the follow-up of these children is necessary.

Acknowledgements

We are very grateful to the staff of the pediatric ward of the RHB and NBH for their collaboration during the realization of this work.

Declaration of Competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have influenced the work reported in this paper.

Funding

The authors declare that this study has received no financial support.

References

1. Creary M, Williamson D, Kulkarni R. Sickle Cell Disease: Current Activities, Public Health Implications, and Future Directions. J Womens Health. 2007;16(5):575-82.

2. kliegman RM, Stanton BF, St Geme III JW, Schor NF. Nelson Textbook of Pediatrics. 20th ed. Behrman RE, editor. Vol. 2. Philadelphia: ELSEVIER; 2016. 1-5315 p.

3. World Health Organisation WHO. Sickle Cell Anemia. 2006 Apr p. 1–5. Report No.: 59TH.

4. Chemegni CB, Kenmegne AB, Ngo SF, Ngouadjeu E, Megningue NE, Bengondo C, et al. Epidemiological, Clinical and Biological Characteristics of Cameroonian Children and Adolescents with Sickle-Cell Anemia. Arch Med. 2018 Feb 14;10(1:6):1-3.

5. Chinenye AH. Physical Growth and Sexual Maturation of Female Adolescent in Sickle Cell Anemia Patients Seen in Lagos, Nigeria. Lagos University Teaching Hospital, Idi-Araba and Sickle Cell Foundation, Nigeria; 2014.

6. Sundd P, Gladwin MT, Novelli EM. Pathophysiology of Sickle Cell Disease. Annu Rev Pathol. 2019 Jan 24; 14:263-92.

7. Lichtin AE. Hematology and Oncology. In: Porter RS, editor. The Merck Manual of Diagnosis and Therapy. 19th ed. Merck Sharp and Dohme Corp; 2011. p. 942-5.

8. Klein DA, Emerick JE, Sylvester J, Vogt KS. Disorders of Puberty: An Approach to Diagnosis and Management. Am Fam Physician. 2017 Nov 1;96(9):590-9.

9. Hauschild M, Theintz G. Severe chronic anemia and endocrine disorders in children. Rev Med Suisse. 2007; 3:988-91.

10. Betoko RM, Um SSN, Alima AY, Chelo D, Nengom JT, Simon D, et al. Puberty During Sickle Cell Anemia in Cameroonian Children: A Case Control Study. [cited 2022 Sep 17]. Available from: https://abstracts.eurospe.org/hrp/0086/hrp0086p1-p739.

11. Soliman AT, Yasin M, El-Awwa A, Abdelrahman MO, De Sanctis V. Does blood transfusion affect pituitary gonadal axis and sperm parameters in young males with sickle cell disease? Indian J Endocrinol Metab. 2013;17(6):962-8.

12. Gomes ICP, Melo HN, Melo SIA, Menezes NV de, Dantas TVP, Cipolotti R. Growth and puberty in a prospective cohort of patients with sickle-cell anaemia: an assessment over ten years. J Hum Growth Dev. 2017;27(1):91-8.

13. Uchendu UO, Nwokocha AR, Ikefuna AN, Emodi IJ, Onwasigwe CN. Evaluation of Sexual Maturity among Adolescent Male Sickle Cell Anaemia Patients: The Usefulness of Testicular Volume Estimation. South Afr J Child Health. 2010 Mar 31;4(1):11.

14. Uchendu UO, Ikefuna AN, Nwokocha ARC, Emodi IJ. Impact of socioeconomic status on sexual maturation of Nigerian boys living with sickle cell anaemia. Hematology. 2010;15(6):414-21.

15. Serjeant GR, Singhal A, Hambleton IR. Sickle cell disease and age at menarche in Jamaican girls: observations from a cohort study. Arch Dis Child. 2001 Nov 1;85(5):375-8.

16. Kaplowit P. Link Between Body Fat and the Timing of Puberty. Am Acad OfPediatrics. 2008; 121:208-17.

17. Fung EB, Harmatz PR, Lee PDK, Milet M, Bellevue R, Jeng MR, et al. Increased prevalence of iron-overload associated endocrinopathy in thalassaemia versus sickle-cell disease. Br J Haematol. 2006;135(4):574-82.

18. M'Pemba-Loufoua AB, Nzingoula Z, Moubouh-Akouala F, Oba A. Développement pubertaire chez la fille drépanocytaire homozygote. A propos de 72 cas. Bull Soc PatholExot. 2001;326-9.

19. Zemel BS, Kawchak DA, Ohene-Frempong K, Schall JI, Stallings VA. Effects of Delayed Pubertal Development, Nutritional Status, and Disease Severity on Longitudinal Patterns of Growth Failure in Children with Sickle Cell Disease. Pediatr Res. 2007 May;61(5, Part 1):607-13.

20. Zemel BS, Kawchak DA, Fung EB, Ohene-Frempong K, Stallings VA. Effect of zinc supplementation on growth and body composition in children with sickle cell disease. Am J Clin Nutr. 2002 Feb 1;75(2):300-7.