Growth and Nutritional Status of Children and Adolescents with Sickle Cell Anemia

Running Title: Nutritional status of Patients with Sickle Cell Anemia

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Abstract

Background

Sickle cell anemia (SCA) is a chronic condition that impacts the nutritional status and growth of patients through various mechanisms.

Objectives

This case-control study aimed to determine the growth patterns and nutritional status of pediatric patients with SCA and investigate the effects of selected clinical, hematological and biochemical parameters on the nutritional status.

Subjects and Methods

The study included 168 children and adolescents with SCA (in a steady state), and 181 healthy children as a control group.

Data (anthropometry, complete blood count, and levels of lactate dehydrogenase, C-reactive protein (CRP), zinc and copper) were evaluated for both groups. The WHO Child Growth Standards were used to assess the nutritional status of participants.

Results

Underweight, stunting and wasting were detected in 13.55%, 25.25% and 12.5% of patients with SCA compared with 2.6%, 2.75% and 2.20% of healthy children and adolescents, respectively ($P < 0.05$). In contrast, 8.33% of patients and 7.18% of individuals in the control group were overweight and obese, respectively.

The weight curve at the age of 6-7 years in females and 9-10 years in males was significantly lower among patients with SCA than the control group. While the height

curve started to decrease significantly at the age of 8-9 years in male patients and ≥ 11 years in female patients compared to the control group.

Age of patients and high CRP levels were independent risk factors for stunting. Meanwhile, number of hospital admissions/year were independent risk factors for wasting.

Conclusions

A high prevalence of poor growth was observed in children with SCA, and the weight curve started to decrease earlier in females, while the height curve decreased earlier in males. Stunting was the most common nutritional problem and was positively associated with age of patients.

Keywords: growth, nutritional status, sickle cell anemia

Introduction

Sickle cell anaemia (SCA) is a well-known inherited disorder with a disruptive metabolic status causing pain and a reduced life expectancy for the patient. Multiple deficiencies in some micronutrients, vitamins, antioxidants and certain lipid constituents have been shown to be prevalent in patients with SCA and associated with an increased severity of the disease [1].

Many complications associated with SCA, such as growth retardation, delayed sexual maturation, and low immunity, are likely partially due to nutritional deficiencies [2]. In addition, multiple external and internal factors are likely to interact with genetic, environmental and socio-economic factors to adversely affect the growth of patients with SCA. The main factors that have been reported to adversely affect growth of children with SCA are endocrine dysfunction, inadequate nutritional intake, micronutrient deficiencies and hypermetabolism [2,4]. Abnormalities in the levels of growth hormone (GH), insulin like growth factor-I (IGF-I), and IGF-binding protein 3 (IGF-BP3) also partially contribute to the growth failure experienced by children with SCA [5].

Children with SCA have a poorer nutrient intake than children matched for age and race. This poor nutrient intake correlates with the poor growth observed among children with SCA.[6] Ill health and frequent hospitalizations may also be associated with varying degrees of anorexia and reduced feeding times in children with SCA [3].

The circulating levels of interleukin-6 (IL-6), which acts within the brain to suppress appetite and subsequently decreases food intake and causes wasting, are elevated in individuals with SCA [7,8]. In addition, the metabolic demands of increased erythropoiesis

and cardiac energy consumption account for the excess protein and energy metabolism reported in these children [9].

In addition to the correction of micronutrient deficiencies and suboptimal nutritional intake among the paediatric SCA population, the use of hydroxyurea (HU) improves growth among children with SCA because HU decreases the resting energy expenditure (REE) and number of required blood transfusions [10,11].

Importantly, in recent studies, children and adolescents with SCA have also been reported to be overweight and obese [12,13].

Despite advances in our understanding of the molecular and genetic bases of SCA, little progress has been achieved in understanding the nutritional problems faced by children with SCA. A limited number of evaluations of a variety of nutritional interventions that may improve the nutritional status and growth of these children and have a favourable impact on their clinical course and prognosis have been performed [3]. In addition, the determinants of poor growth in patients with SCA are not well understood and are probably due to phenotypic polymorphisms caused by the haplotype, genetic factors, foetal haemoglobin levels, specific nutrient deficiencies, and environmental factors [14,15].

This case-control study was performed to determine the frequency of nutritional problems (wasting and stunting) and the growth pattern of patients with SCA. We also investigated the potential risk factors associated with wasting and stunting in these patients.

Subjects and Methods

This case-control study was conducted on 168 children and adolescents with SCA who consulted the Center for Hereditary Blood Diseases (CHBD) at Basra Maternity and

Children's Hospital. The sample size was determined based on the expense of data collection and the need for sufficient statistical power.

Detailed histories, including sociodemographic and clinical data, were obtained and complete examinations, including growth parameters, were conducted for both patients and controls.

Drugs taken by patients, such as prophylactic antibiotics, folic acid and zinc supplements, non-steroidal anti-inflammatory drugs (NSAIDs), narcotics, HU and iron-chelating agents, were recorded.

Children with SCA were evaluated while in the steady state, which was defined as the absence of any painful crisis during the preceding 4 weeks, no recent decrease in haemoglobin levels and the absence of any symptoms or signs attributable to an acute illness [16].

The severity of SCA was assessed as the frequency of vaso-occlusive crises (VOCs), hospital admissions and blood transfusions [17,18].

Exclusion criteria were a Glucose -6- Phosphate Dehydrogenase (G6PD) deficiency, HU and zinc supplementation, a history of overt stroke, chronic medical or orthopaedic problems, fever or painful crises during the last 4 weeks, and blood transfusions within the last three months [16,19-21].

The control group included 181 apparently healthy children aged 1-15 years with normal haemoglobin patterns (Hb AA). These children were recruited from 3 primary health centres and were attending these centres for school registration, vaccinations or for minor health problems such as the common cold.

Z-scores for weight-for-age (WAZ), height-for-age (HAZ) and body mass index (BMI) for age (BMIZ) were calculated for all participants and compared with the World Health Organization (WHO) Multicentre Growth Reference Study (WHO MGRS 2006/2007) reference values using WHO Anthro software version 3.2.2 (Department of Nutrition, WHO, Geneva, Switzerland) for the global application of WHO child growth standard for children aged 1-5 years. WHO AnthroPlus software version 1.0.4 (Department of Nutrition, WHO, Geneva, Switzerland) was used for the global application of 2007 WHO reference standards for children and adolescents aged 5-19 years [22].

WAZ, HAZ and BMIZ scores were used to determine the severities of underweight, stunting and wasting conditions. Underweight, stunting and wasting were defined as WAZ, HAZ, and BMIZ scores < -2 standard deviations (SD) from the median WHO Child Growth Standards. Z-scores < -2 but ≥ -3 were considered moderate, and scores > -3 were considered severe. A BMIZ score ≥ 1 SD was considered "overweight" and > 2 SDs was considered "obese", according to the WHO [23].

SCA was diagnosed using high-performance liquid chromatography (HPLC), (VARIANT™, β-Thalassemia Short Programs; Bio-Rad Laboratories, Hercules, CA, USA) performed at the CHBD. A foetal haemoglobin (Hb F %) level < 10% was regarded as a low level, and ≥ 10% was regarded as a high level [24,25].

The complete blood count (CBC) was measured using the SYSMEX KX-21N automated haematological analyser (Wakinohama, Japan).

Lactate dehydrogenase (LDH) levels were measured using the colorimetric method from Audit Diagnostic kits. The LDH level was estimated with a spectrophotometer (Abbott Architect plus C4000, Japan), and the normal values ranged from 55 to 110 U/L.

C-reactive protein (CRP) levels were estimated with the rapid latex slide test using a Spectrum kit. A CRP level < 6 mg/L was deemed a normal value, and a value ≥ 6 was regarded as an elevated level.

Serum ferritin levels in patients were measured using the MINIVIDAS system immunoassay (BioMerieux, Italia). Serum copper and zinc levels were measured using a spectrophotometric method (Abbott Architect plus C4000, Japan). The normal serum zinc and copper levels in children are 63-110 and 80-190 µg/dL, respectively.

An informed consent was obtained from one of the parents before recruitment into the study. This study was approved by the Ethical Committee of Basra Medical College.

Statistical analyses were performed using SPSS program V.18, (IBM, Chicago, USA, SPSS Inc.). Data are presented as means ± standard deviations. A comparison of proportions was performed using the chi-square test when each value has an expected frequency of ≥ five, and Fisher's exact test was used when one or more of values have an expected frequency of < five. The independent t-test was used for a quantitative comparison between two means from different samples. A binary logistic regression analysis was also performed to analyse the potential independent risk factors associated with wasting and stunting. For all tests, a P value < 0.05 was considered significant.

Results

The total number of children and adolescents enrolled in this study was 349, and their ages ranged from 1- 15 years. Among these subjects, 168 were patients with SCA and 181 comprised the control group. The demographic data revealed that a significantly greater number of patients resided in rural areas and had parents with low educational levels compared to the control group; $P < 0.05$, Table 1.

One hundred forty-three (85.1%) patients had experienced one or more VOCs during the past year, and 93 (55.4%) required hospitalization; 65 (38.70%) required hospitalization fewer than 3 times last year. In addition, 94 (55.95%) of the patients did not require blood transfusions.

None of the patients had iron-deficient anaemia, as indicated by low serum ferritin levels, while 64 (38.1%) of the patients had a serum ferritin level > 1000 ng/mL. Male patients with SCA had significantly higher mean Hb F percentages than female patients; 20.87 ± 8.05 vs. 18.60 ± 0.82 respectively, $P < 0.05$.

The frequencies of underweight, stunting and wasting conditions were significantly higher among patients with SCA than in the healthy control group, $P < 0.05$. Overweight and obesity were reported in both groups; $P > 0.05$, Table 1.

The mean WAZ (-1.58 ± 1.11) and HAZ (-2.03 ± 1.21) scores among children with SCA and a Hb F level < 10% were significantly lower than in children with SCA whose Hb F was ≥ 10% (-0.79 ± 1.09 and $- 0.78 \pm 1.94$, respectively); $P < 0.05$. The BMIZ score was not significantly different between children with low and high Hb F levels (-1.22 ± 1.53 and -0.70 ± 1.27, respectively); $P = 0.075$.

The growth of patients of both sexes (both weight and height) in early age groups was comparable to the control group, $P > 0.05$. The weight curve at the age of 6-7 years in females and 9-10 years in males was significantly reduced among patients with SCA compared with the control group, $P < 0.05$, Fig. 1. The height curve started to decrease significantly at the age of 8-9 years in males and ≥ 11 years in females among patients with SCA compared to the control group, Fig. 2.

No differences in BMI were observed among patients or in control growth curves from early to late age groups among males or females, except for males aged 11-12 years and females aged 9-10 and 12-13 years, Fig. 3.

Significantly lower serum Hb, PCV, MCV, MCH, MCHC and Zn levels were observed among patients with SCA. Significantly higher WBC, platelets, reticulocyte counts, ESR, and serum copper, LDH and CRP levels were observed among patients with SCA ($P < 0.05$), Table 2.

A logistic regression analysis of potential risk factors for stunting and wasting revealed significant positive correlations between the number of hospitalisations with wasting and both age and CRP levels with stunting adversely affected the nutritional status of these patients, Table 3. Other sociodemographic, clinical and laboratory variables were not found to be associated with the nutritional status of SCA patients.

Discussion

The current study has revealed significantly higher frequencies of underweight, stunting and wasting conditions among patients with SCA than in the healthy control group, and indicators of a severe disease, such as frequent painful episodes and hospitalisations, and

low Hb F levels, adversely affected the nutritional status of these patients. Moreover, the growth of patients (both weight and height) in early age groups was comparable to the control group. However, the weight curve started to decrease at the age of 6-7 years in females and 9-10 years in males with SCA. Meanwhile, the height curve started to decrease significantly at the age of 8-9 years in male patients and ≥ 11 years in female patients with SCA compared to the control group.

Many factors affect the nutritional status and growth of patients with SCA. Importantly, the socioeconomic status of patients and their families may affect the patient's growth. Although this study did not indicate parental education and residence as risk factors for stunting and wasting. This finding differs from that of Rahimy et al., who reported that inadequate education, a limited number of health care facilities, limited parental understanding, poverty, and low compliance with routine health care visits, which are vital to the success of any comprehensive clinical care programme, exert a positive and sustained impact on the growth of patients with SCA, regardless of their age [26].

The negative impact of disease severity, mainly frequent hospitalizations, on the growth pattern of children with SCA is likely attributed to a reduced dietary intake prior to admission, increased resting energy expenditure during VOCs that leads to increased erythroid hyperactivity and red cell turnover, chronic anaemia associated with hypoxia and a hyperdynamic circulation that consequently increases the demand for energy and nutrients, chronic and acute consequences of vaso-occlusion, endocrine dysfunction, and a low socioeconomic status [4,27].

Although a higher frequency of malnutrition was reported among patients with SCA compared to the control group in other studies, the frequencies of stunting, wasting and underweight varied. de Souza et al. reported stunting in 15.5% and wasting in 5.7% of children and adolescents with SCA in Brazil [28]. Underweight, stunting, and wasting were observed in 47.7%, 10.5%, and 50.3% of children with SCA in Central Africa, respectively [29]. Low WAZ, HAZ and BMIZ scores (< -2 SD) were observed in 45%, 54% and 35% of children with SCD, respectively, in Yemen [30].

In the current study, 8.33% and 7.18% of patients and controls were obese or overweight, respectively, indicating that the incidence of overweight and obesity in children with SCD parallels the incidence of these conditions in the general paediatric population. The comparable frequencies of overweight and obesity between both groups are likely attributed to overprotection, a sedentary lifestyle, feeding habits and the presence of a milder disease in our patients, as indicated by a high mean Hb F level. The percentages of overweight and obesity are lower than the incidence of 22.4% reported by Chawla et al. in the USA [12], but higher than the incidence reported by Akodu et al. in Nigeria (2.5% and 3.8% among subjects with SCA and healthy controls, respectively) [31]. These findings lead to the conclusion that public health programmes aimed at preventing and controlling obesity must include children with SCA [31].

In the present study, the growth of patients was comparable to the control group. This finding was particularly apparent among early age groups, when the impact of the disease process on the growth of patients was limited. With aging, a significant difference appeared between the patient and control group, with weight specifically being affected at an earlier

age in females and height at an earlier age in males. This result may be attributed to the delay in pubertal attainment in children with SCA than in healthy controls [32].

Cox et al. reported that the greatest deficit in height among children with SCA occurs during adolescence, although an extended growth period and the potential for catch-up growth is more pronounced in females [33]. Many researchers have described the nutritional characteristics of children and adolescents with SCA and concluded that anthropometric indicators (weight and height) are often lower in children with SCA than in healthy groups or reference populations in all geographic areas [4,34], with good evidence linking growth failure to endocrine dysfunction, metabolic disorders, and specific nutrient deficiencies [4].

In the present study, the Hb level was not an independent risk factor for malnutrition. This finding may be attributed to the high Hb F level, which decreases hemolysis and the associated metabolic requirements to subsequently reduce growth retardation. However, the Hb level was an independent risk factor for decreased growth in studies by Cox et al. conducted in Tanzania [33], Singhal et al. in Jamaica [35], and Zemel et al. in the USA [36].

A high CRP level was identified as an independent risk factor for stunting among our patients. This finding is consistent with the results reported by Hibbert et al. [37], who attributed stunting to the strong association between CRP levels and REE, indicating a link between inflammation and the hypermetabolic state in patients with SCA.

This study has many limitations. One limitation is that it employed a cross-sectional design that lacks monitoring of the growth parameters in children with SCA over time. Other limitations are that dietary habits and physical activity were not considered in this study.

Conclusion

This study is the first to describe the nutritional status encountered in Iraqi children and adolescents with SCA. A high prevalence of poor growth was observed in children with SCA, and the weight curve started to decrease earlier in females with SCA, while the height curve decreased earlier in males with SCA. Stunting was the most common nutritional problem, indicating the chronicity of the condition, and was associated with increasing age of patients.

Nutritional interventions, regular health care visits and growth monitoring will likely improve the growth and general health status of children with SCA.

Author contributions

RS, MK and LM designed and planned the study. RS collected the data. RS, MK, and LM analysed the data. All authors contributed to the writing of the manuscript and read and approved the final manuscript.

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Table 1. Sociodemographic Characteristics of Patients with SCA and Controls.

Variable		**SCA group** (Total 168) N. (%)	**Control group** (Total 181) N. (%)	**P value**
Age in Months (Mean ± SD)		89.67 ± 48.21	96.56 ± 49.62	0.189**
Age groups (Years)	< 2	13 (7.74)	13 (7.18)	0.738*†
	2- 4	43 (25.60)	39 (21.55)	
	5- 9	59 (35.12)	63 (34.81)	
	10-15	53 (31.54)	66 (36.46)	
Sex	Female	82 (48.81)	88 (48.61)	0.972§
	Male	86 (51.19)	93 (51.38)	
Residence	Urban	78 (46.42)	117 (64.64)	0.001§
	Rural	90 (53.57)	64 (35.35)	
Mother's Education				
Illiterate		36 (21.42)	21 (11.60)	< 0.001§
Primary		76 (45.23)	40 (22.09)	
Secondary		49 (29.16)	55 (30.38)	
Higher Education		7 (4.16)	65 (35.91)	
Father's Education				
Illiterate		2 (15.47)	29 (16.02)	0.012§
Primary		73 (43.45)	52 (28.72)	
Secondary		43 (25.59)	51 (28.17)	
Higher Education		26 (15.47)	49 (27.07)	
Nutritional Status				
Underweight*	Moderate	11 (9.32)	2 (1.72)	0.009†
	Severe	5 (4.23)	1 (0.86)	
	Total	16 (13.55)	3 (2.58)	
Stunting	Moderate	32 (19)	3 (1.65)	< 0.001†
	Severe	11 (6.55)	2 (1.10)	
	Total	43 (25.55)	5 (2.75)	
Wasting	Moderate	14 (8.33)	3 (1.65)	0.001†
	Severe	7 (4.17)	1 (0.55)	
	Total	21 (12.5)	4 (2.20)	
Overweight		11 (6.55)	12 (6.63)	0.452§
Obese		3 (1.78)	1 (0.55)	0.121†

*Until 10 years of age, according to the WHO growth chart. SCA (No. 118), control (No. 116).
**Independent t-tests were used to calculate P value,
§ the chi square test was used the P values, † Fisher's exact test was used to calculate the P values.

Table 2. Selected Haematological and Biochemical Variables among Patients with SCA and Controls.

Variable		**SCA (Mean ± SD)**	**Control (Mean ± SD)**	**P value***
Hb (g/L)		81.51 ± 13.5	120.80 ± 19.61	< 0.001
PCV (g/L)		25.42 ± 4.11	36.11 ± 5.92	< 0.001
MCV (fL)		78.44 ± 10.82	82.03 ± 3.86	< 0.001
MCH (pg)		25.45 ± 3.14	30.69 ± 2.60	< 0.001
MCHC (g/L)		31.69 ± 1.91	34.15 ± 0.69	< 0.001
Total WBC ($\times 10^9$/L)		11.04 ± 5.20	6.31 ± 1.15	< 0.001
Platelets ($\times 10^9$/L)		325.92 ± 144.59	291 ± 53.85	0.003
Reticulocytes (%)		7.57 ± 6.70	1.6 ± 0.46	< 0.001
ESR (mm/hr)		20.42 ± 5.56	11.11 ± 5.12	< 0.001
S. Zn (µg/dL)		61.57 ± 12.85	91.54 ± 14.18	< 0.001
S. Cu (µg/dL)		121.98 ± 30.49	90.49 ± 14.16	< 0.001
LDH (U/L)		136.30 ± 70.98	104.23 ± 37.36	< 0.001
CRP N. (%)	High	49 (29.2)	19 (10.5)	< 0.001
	Normal/ Low	119 (70.8)	162 (89.5)	< 0.001

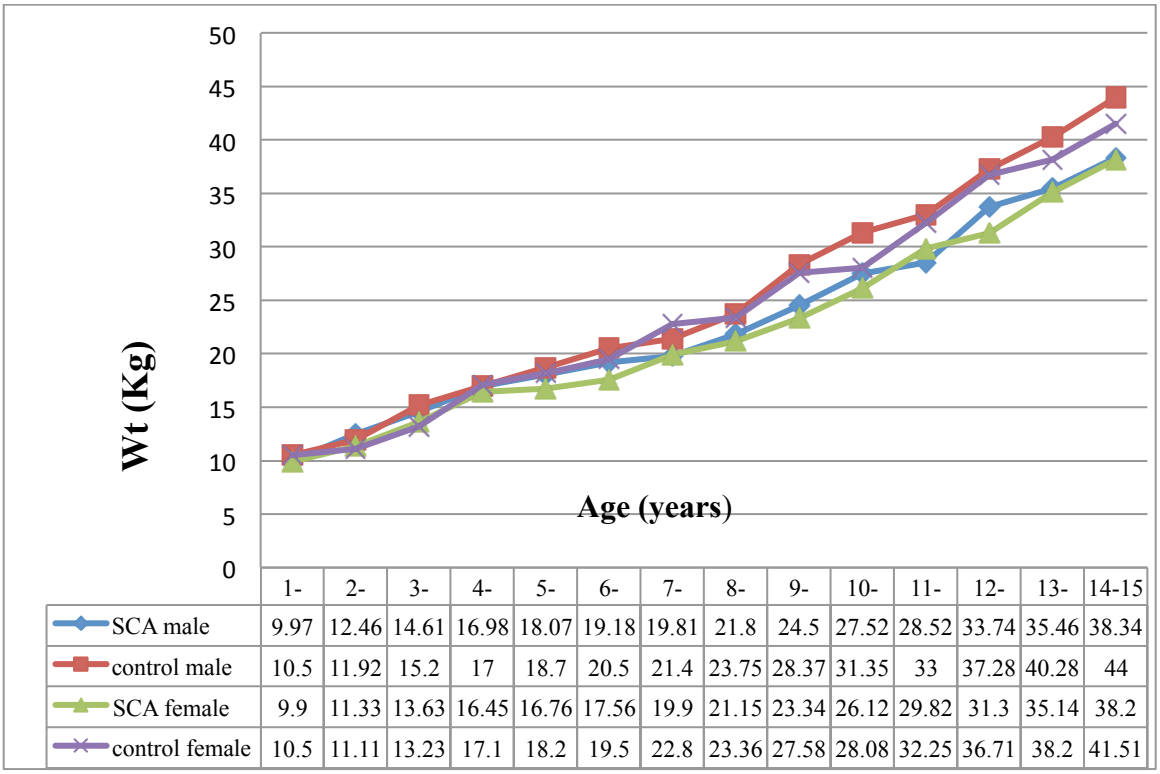
* Independent t-tests were used to calculate P values for all variables except CRP levels (chi square was used).

Hb: hemoglobin; PCV: packed cell volume; MCV: mean corpuscular volume; MCH: mean corpuscular hemoglobin; MCHC: mean corpuscular hemoglobin concentration; ESR: erythrocyte sedimentation rate; WBC: White blood cells; S. Zn: serum zinc; S. Cu: serum copper; LDH: lactate dehydrogenase, CRP: C-Reactive Protein.

Table 3. Independent Risk Factors Associated with Stunting and Wasting among Patients with SCA

Variables	β coefficient	OR	95% CI	P value
Stunting				
Age	0.017	1.017	1.008- 1.026	<0.001
CRP	1.932	6.900	3.010- 15.820	< 0.001
Wasting				
N. of Hospital admissions	0.262	0.092	1.069- 1.580	0.009

CRP: C-reactive protein; OR: Odd Ratio; CI: Confidence Interval.

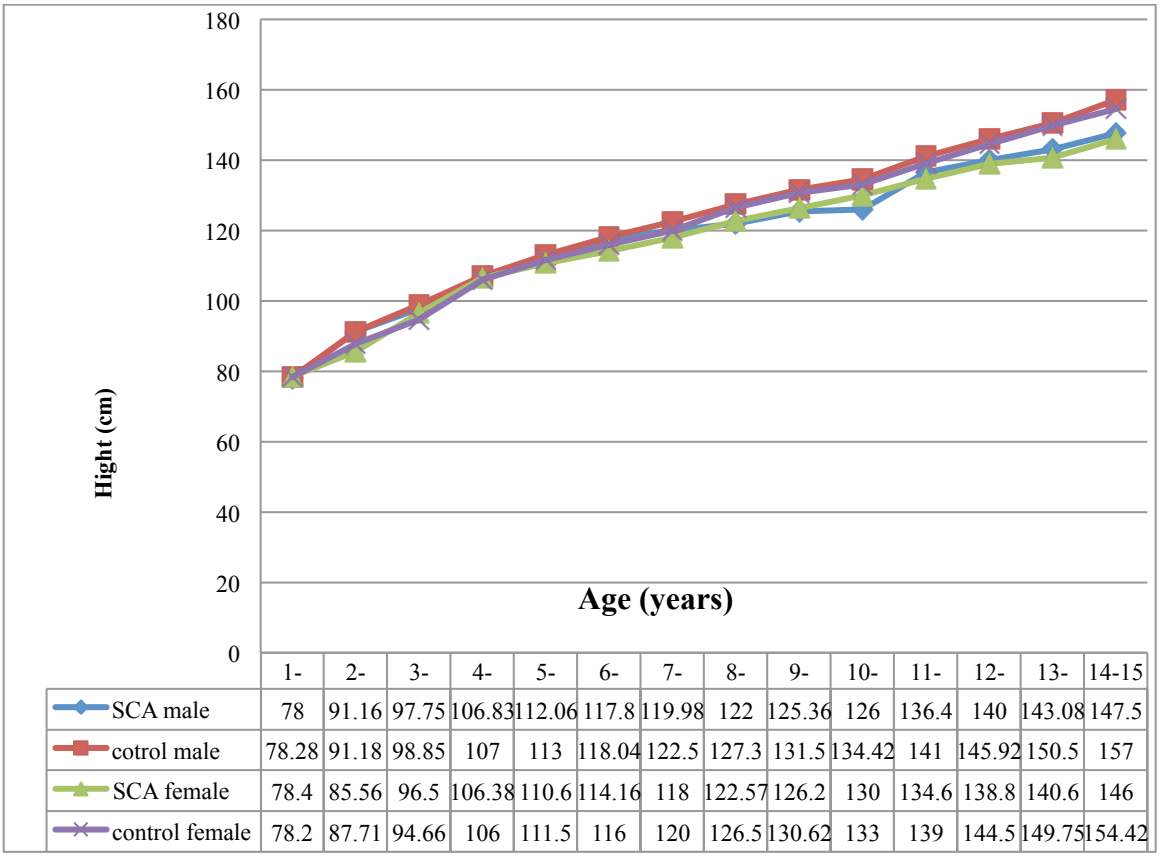


	1-	2-	3-	4-	5-	6-	7-	8-	9-	10-	11-	12-	13-	14-15
SCA male	9.97	12.46	14.61	16.98	18.07	19.18	19.81	21.8	24.5	27.52	28.52	33.74	35.46	38.34
control male	10.5	11.92	15.2	17	18.7	20.5	21.4	23.75	28.37	31.35	33	37.28	40.28	44
SCA female	9.9	11.33	13.63	16.45	16.76	17.56	19.9	21.15	23.34	26.12	29.82	31.3	35.14	38.2
control female	10.5	11.11	13.23	17.1	18.2	19.5	22.8	23.36	27.58	28.08	32.25	36.71	38.2	41.51

Figure 1. Weight for Age Chart for Males and Females with SCA and the Control Group

Independent t-tests were used to calculate P values.

P value < 0.05; males aged 9-10, 11-12 and 13-14 years and females aged 6-7, 9-10 and 12-13 years only.

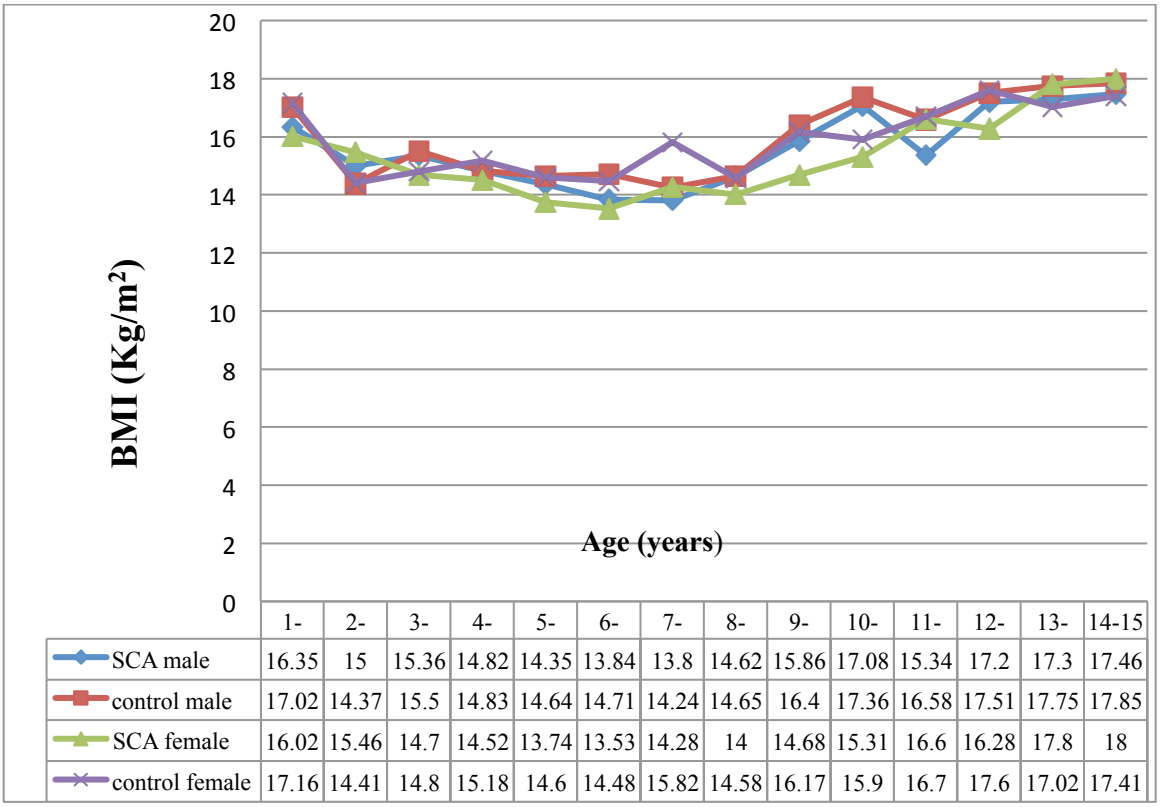


	1-	2-	3-	4-	5-	6-	7-	8-	9-	10-	11-	12-	13-	14-15
SCA male	78	91.16	97.75	106.83	112.06	117.8	119.98	122	125.36	126	136.4	140	143.08	147.5
cotrol male	78.28	91.18	98.85	107	113	118.04	122.5	127.3	131.5	134.42	141	145.92	150.5	157
SCA female	78.4	85.56	96.5	106.38	110.6	114.16	118	122.57	126.2	130	134.6	138.8	140.6	146
control female	78.2	87.71	94.66	106	111.5	116	120	126.5	130.62	133	139	144.5	149.75	154.42

Figure 2. Height for Age Chart for Males and Females with SCA and the Control Group.

Independent t-tests were used to calculate P values.

P value < 0.05; males aged 8-9, 10-11, 11-12, 13-14 and 14-15 years, females aged 11-15 years.



	1-	2-	3-	4-	5-	6-	7-	8-	9-	10-	11-	12-	13-	14-15
SCA male	16.35	15	15.36	14.82	14.35	13.84	13.8	14.62	15.86	17.08	15.34	17.2	17.3	17.46
control male	17.02	14.37	15.5	14.83	14.64	14.71	14.24	14.65	16.4	17.36	16.58	17.51	17.75	17.85
SCA female	16.02	15.46	14.7	14.52	13.74	13.53	14.28	14	14.68	15.31	16.6	16.28	17.8	18
control female	17.16	14.41	14.8	15.18	14.6	14.48	15.82	14.58	16.17	15.9	16.7	17.6	17.02	17.41

Figure 3. Body Mass Index for Age Chart for Males and Females with SCA and the Control Group.

Independent t-tests were used to calculate P values.

P value < 0.05; males aged 11-12 years and females aged 9-10 and 12-13 years only.