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DEPARTMENT OF NUTRITON AND DIETETICS

ASSESSING NUTRITION KNOWLEDGE AND NUTRITIONAL STATUS OF SICKLE CELL DISEASE PATIENTS

BY

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DECLARATION

I, Faustina Ofosua Mintah, declare that this work is the report of the research I undertook at the Department of Nutrition and Dietetics, School of Biomedical and Allied Health Sciences, University of Ghana, under the supervision of Dr. Joana Ainuson-Quampah, Prof. Solomon Ofori-Acquah and Dr. Catherine Segbefia and neither the whole nor any part of it has been submitted for another degree at this or any other university. All references cited have been fully acknowledged.

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ABSTRACT

Background: Sickle cell disease (SCD) is a common haemoglobinopathy that is acquired by inheriting the defective gene from both parents. In recent years, it has become an important public health issue among people of African descent especially those in Sub-Saharan Africa. In Ghana, SCD occurs in almost 1 in 50 (2%) of all births annually. The disease is characterised by chronic haemolytic anaemia, vaso-occlusion and inflammation. These events increase the resting energy expenditure of individuals and contribute to the hypermetabolic state of a sickle cell patient, and consequently impact on the nutritional status. The poor nutritional status reported in individuals with SCD can also be attributed to reduced dietary intake due to low appetite leading to varied nutrient deficiencies including vitamin A, C, E and D deficiencies. In spite of these nutritional challenges faced by this group of people, frequent nutritional counselling is not part of the standard of care. However, recent studies have identified nutrition knowledge as one of the mechanisms which influence nutritional behaviour of individuals thereby improving their nutritional status.

Aim: To assess nutritional knowledge and nutritional status of people with sickle cell disease.

Methods: A cross-sectional study design was used. An estimated number of 173 adolescents and adults attending the Ghana Institute of Clinical Genetics, Korle Bu who were recruited for the SickleGenAfrica project were enrolled for this study. Nutrition knowledge of patients was assessed using a pre-tested questionnaire. Anthropometric, biochemical data and a one-week food frequency questionnaire were used to assess nutritional status. About 0.15ml of serum for the SickleGenAfrica project was sampled to assess serum vitamin E and IL-6 levels using Enzyme Linked Immunoassay technique. Stata/IC 14.0 was used for data analysis. Chi-squared was used to test for associations between nutrition knowledge and nutritional status. Linear regression was used to determine association between inflammation and nutritional

status. Multiple regression was used to test for the strength of the significant associations. A p-value < 0.05 was considered statistically significant.

Results: Malnutrition was recorded among 25% of the participants with prevalence of underweight, overweight and obesity as 15.2%, 7.6% and 1.8% respectively. Majority (84.8%) of the participants had high nutrition knowledge. There was no association between nutritional knowledge and nutritional status. The frequency of fruits and vegetables consumption was low. The mean serum vitamin E level of participants was 6.7±9.9µg/ml and there was no significant difference between mean levels of Hb SS and Hb SC individuals. Even in steady state, IL-6 levels were elevated with a mean of 18.6±33.7 pg/ml and this correlated negatively with body weight (r = -0.1698, p = 0.03). Body weight and height could predict 3% of the variability in IL-6 levels (F (2, 165) = 2.97; p – 0.05). There was a significant difference between the IL-6 levels of Hb SS and Hb SC individuals with means of 21.4±3.42 pg/ml and 11.2±2.70 pg/ml (p - 0.02) respectively.

Conclusion: Nutrition knowledge did not influence nutritional status. Results from this study show that participants are at risk of vitamin E deficiency. Hb SS individuals experienced higher levels of inflammation compared to Hb SC individuals.

DEDICATION

This work is dedicated to my parents, Mr. Samuel Ayim Mintah and Ms. Charlotte Adjei, for their immense support throughout my academic journey and my siblings for always motivating to be better and being my source of stress relief.

I also dedicate this work to all individuals with sickle cell disease.

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LIST OF ABBREVIATIONS

ACS –Acute Chest Syndrome

BAZ - BMI-for-age

BF - Body Fat

BMI - Body Mass Index

ELISA - Enzyme-Linked Immunosorbent Assay

FFM - Fat-Free Mass

FFQ - Food Frequency Questionnaire

Hb – Haemoglobin

HRP – Horseradish Peroxidase

IL-1 -Interleukin-1

IL-6 -Interleukin-6

IL-10 -Interleukin-10

MM - Muscle Mass

RBC - Red Blood Cell

REE - Resting Energy Expenditure

SCA - Sickle Cell Anaemia

SCD - Sickle Cell Disease

TNF – Tumour Necrosis Factor

TMB - 3, 3′, 5, 5′-Tetramethylbenzidine

WHO - World Health Organisation

CHAPTER ONE

1.0 INTRODUCTION

1.1 Background

Sickle cell disease (SCD) is a common genetic condition characterised by inheriting the defective haemoglobin gene from both parents (Makani et al., 2013). It is very common among people of Indian, Saudi Arabian, Mediterranean and sub-Saharan African descent (World Health Organisation (WHO), 2006). About 700 structural haemoglobin (Hb) variants have been identified. However only two, that is, Hb S and Hb C are very common in Africa with Hb C being exclusive to West Africa (Makani et al., 2013). Sickle cell disease is estimated to occur in about 300,000 births worldwide yearly (WHO, 2006). In Africa, about 10% to 40% of the population are carriers of the sickle cell gene leading to an estimated 2% prevalence of SCD in sub-Saharan Africa (WHO, 2006).

Patients with sickle cell disease usually suffer from anaemia, jaundice, infections and chronic organ damage. Vaso-occlusion is also very common and is identified as one of the contributors to the characteristic painful crises (Makani et al., 2013). Painful crises have been recognised as the cause of about 80-90% of hospitalisations (Lemanek et al., 2002). Frequent hospitalisations put financial stress on patients and make it difficult to work and raise money to support economic needs. Nutritional adaptations have been identified as one of the coping strategies that is adopted to help reduce the frequency of pain episodes and hospitalisations (Tanabe et al., 2010).

People with SCD are found to have a poor nutritional status with underweight being more prevalent among adolescents and adults compared to their healthy peers (Sobo et al., 2007). Some proposed mechanisms for the growth deficit include decreased intake, intestinal

malabsorption and increased catabolism. However, the most frequently reported mechanism is that sickle cell anaemia increases metabolic requirement due to recurrent haemolytic anaemia (Hyacinth, Gee, & Hibbert, 2010). This sub-optimal nutritional status has been linked to disease severity, number of hospitalisations, risk of infections just to name a few (Mandese et al., 2016). Common nutrient deficiencies in patients with SCD include vitamins A, C, E and D, zinc and magnesium (Hyacinth et al., 2010). As such, routine supplementations with folic acid and multivitamins are given to make up for some of these deficiencies. These findings have informed several studies on the role of nutrition in the management of SCD (Hundekar et al., 2011; Mandese et al., 2016; Mitchell, Kawchak, Stark, Zemel, Ohene-Frempong, et al., 2004; Nwaoguikpe & Braide, 2012; Tanabe et al., 2010). Therefore, there have been proposals to increase the recommended nutrient intake for sickle cell patients to make up for increased demand and ensure good nutritional status. Laboratory experiments have shown that supplementation of nutrients like protein, vitamins A, C and E, zinc and magnesium leads to an improvement in the clinical manifestations of the disease (Nwaoguikpe & Braide 2012; Ohnishi et al., 2000). This is suggestive of the need for nutritional care in the management of sickle cell disease.

Since the role of nutrition and the need for frequent nutrition counselling in the management of SCD has been emphasised by researchers and even some patients, it is necessary to assess nutrition knowledge and how this impact on their nutritional status. This is because nutrition knowledge is one of the factors that affect nutrition behaviour of individuals and subsequently overall health (Hakli et al., 2016).

1.2 Problem Statement

Sickle cell disease is not only a public health issue especially in Africa but also in Ghana. More than 75% of the people with SCD are in Africa (WHO, 2006). In Ghana, about 1.9% (1 in 50) of all new born babies annually have SCD (Ohene-Frempong et al., 2008). Ranked as the fifth commonest non-communicable disease in Ghana, the Ghana Health Service report for 2014 also showed an increase in the number of outpatient SCD cases from 2011- 2014 (from 29764 in 2011 to 43801 in 2014) (Ghana Health Service, 2015).

Sickle cell disease with its resultant increased energy and nutrient requirements coupled with inadequate food intake as a result of eating disorders such as poor appetite and pica, has adverse effects on body composition and disease severity (Hyacinth et al., 2010; Kawchak, Schall, Zemel, Ohene-Frempong, & Stallings, 2007; Sobo et al., 2007)

Therefore, improving nutritional status seems to be one of the techniques that can be used to improve disease outcome and overall quality of life. To achieve this, the nutritional knowledge of the patient is key since it has been mentioned as one of the mechanisms for improving nutritional status (Hakli et al., 2016). Subsequently, acquiring knowledge is influenced by education. However frequent nutrition counselling is still not part of the standard of care for the management of the disease.

There is also paucity of data on the nutritional knowledge and its association with the nutritional status of SCD patients. Thus, it is necessary to assess the nutrition knowledge of people with SCD and its association to their nutritional status so that the appropriate measures will be put in place to improve the quality of life of people with SCD.

1.3 Significance of Study

This study will provide baseline information on the nutrition knowledge and nutritional status of adolescent and adult sickle cell disease patients.

Information on the association between nutrition knowledge and nutritional status of patients with SCD will help the health team such as nurses, dietitians and doctors to come up with a comprehensive plan on how to improve care for the patient. It may also reinforce the need for nutrition education programmes for patients with SCD. Therefore, the appropriate stakeholders like Ministry of Health and Ghana Health Service can use findings from this study as a baseline to develop nutrition education programmes and policies that will help improve nutritional status and overall health of the SCD patient.

1.4 Hypothesis

H_0: There is no association between nutrition knowledge and nutritional status of people with SCD.

1.5 Aim

To assess nutritional knowledge and nutritional status of adolescents and adults with sickle cell disease.

1.6 Specific Objectives

1. To assess the nutritional knowledge of adolescents and adults with SCD.
2. To assess the nutritional status of adolescents and adults with SCD.
3. To assess the food consumption patterns of adolescents and adults with SCD.
4. To determine the association between nutritional knowledge and nutritional status of adolescents and adults with SCD.
5. To determine the association between inflammatory biomarker and nutritional status of people with SCD.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 History of Sickle Cell Disease

Sickle cell disease (SCD) refers to a group of haemoglobinopathies where at least one sickle β-globin gene is inherited together with another type of abnormal haemoglobin (Hyacinth et al., 2010). Haemoglobin (Hb) is a protein found in red blood cells (RBCs) and an oxygen transporter. In SCD, valine in the sixth codon is substituted with glutamic acid in the β-globin gene (Stuart & Nagel, 2004). Due to this abnormality, HbS can polymerise when deoxygenated. The polymer formed is an insoluble rope-like fibre that lines up with others to form a bundle, deforming the RBC into the characteristic sickle shape (Stuart & Nagel, 2004). Hence, the name sickle cell disease.

An intern of Dr. Irons named Dr. Herrick was the first to observe these elongated sickle-shaped RBCs during a routine blood test of a patient admitted for respiratory distress and leg ulcer (Herrick, 1910). An article about the disease was later published by Dr. Herrick with the caption "peculiar elongated and sickle-shaped red blood corpuscles in a case of severe anaemia" (Herrick, 1910). Although Dr. Herrick was the first to document a case report about the disease, it was until Dr. Mason observed the fourth reported case of the disease that he coined the name sickle cell anaemia (The Sickle Cell Association of Ontario, n.d.). He noticed the similarities between all the cases and observed that all the cases were black people. This gave rise to the idea that the disease originated from Africans (The Sickle Cell Association of Ontario, n.d.).

Since Dr. Mason made this remarkable observation, two schools of thoughts have emerged based on the origin of the disease. Whilst one school of thought theorise that the disease

originated from the same mutation occurring at five independent locations (multicentre theory), the other postulates that the disease originated from a single mutation occurring at a particular place (single centre theory). Pagnier et al. (1984) provided evidence for the multicentre theory in their study which examined different sickle β-globin haplotypes from four different regions in Africa. They concluded that the mutation occurred in 3 different locations in Africa namely, Benin, Bantu/ Central African Republic and Senegal. Other haplotypes that have been identified are Cameroon and Middle East and India (Arab-India) (Piel et al., 2017; Piel & Weatherall, 2015). Even though the hypothesis of five independent origins has been accepted almost globally over the years, recent reports have proven that the disease originated from just one mutation many years ago. A recent publication by Shriner and Rotimi concluded that the disease occurred due to a single mutation occurring at one geographic location. From their analysis, it was revealed that the mutation is of an African region and occurred once approximately 7,300 years ago either in West-Central Africa or the Sahara (Shriner & Rotimi, 2018).

2.1.1 Types of Sickle Cell Disease

Since the discovery of the disease, about 700 structural haemoglobin variants have been identified over the past decades (Makani et al., 2013). Some of the common subtypes which have been identified include haemoglobin (S, C, F and E) and thalassemia. Haemoglobin S (Hb S) is found to be common in areas of low altitude and high incidence of the *Plasmodium falciparum* malarial parasite (Piel et al., 2010). The other subtype haemoglobin C (Hb) is almost exclusive to West Africa with Burkina Faso and northern Ghana recording the highest frequencies (Grosse et al., 2011). This is reiterated by Makani et al. (2013) that Hb SS and Hb SC disease are the frequently observed SCD cases in Africa. Hyacinth et al. (2010)

mentions haemoglobin Sβ□thalassemia (Hb $S\beta^{thal}$) minor and major as other variants of SCD which are not common in Africa.

People with sickle cell anaemia are those who inherit two copies of the Hb S, that is, homozygous (Hb SS) (Hyacinth et al., 2010). Sickle Cell Anaemia (Hb SS) patients have the disease phenotype unlike the carriers (Hb AS) since haemoglobin A (Hb A) is the normal haemoglobin (Kanter & Kruse-Jarres, 2013). Alternative forms of the disease occur when Hb S is inherited together with other abnormal haemoglobin like Hb C and F. This gives rise to other forms of the disease like Hb SC, Hb SF, Hb $S\beta^{+}$ and other haemoglobin genotypes (Kanter & Kruse-Jarres, 2013). Hb SS and Hb $S\beta^{0}$ are known as the most severe forms of SCD (Chakravorty & Williams, 2015; Kanter & Kruse-Jarres, 2013). Aside Africa, SCD is very common among people of Indian, Mediterranean and Saudi Arabian descent (WHO, 2006). Migration has contributed to spreading of the disease to other parts of the world like America and Europe (Chakravorty & Williams, 2015; Moraes et al., 2017).

2.2 Prevalence of Sickle Cell Disease

The World Health Organisation (WHO) estimates that about 300,000 babies are born with major haemoglobin disorders annually (WHO, 2006). Majority of these newborns are in Nigeria, the Democratic Republic of Congo, and India (Piel et al., 2017). About 5% of the world's population are carriers of the genes responsible for haemoglobin disorders. In Africa, more than 200,000 newborns have sickle cell anaemia annually and an estimated 2% of all children born in Africa annually have SCD (WHO, 2006).

The WHO report also estimates that about 10% to 40% of people across equatorial Africa have sickle-cell trait and these decrease to < 2% in Northern and Southern Africa. It is

therefore not surprising that about two-thirds of the global population with SCD are in Sub-Saharan Africa (WHO, 2006). In West African countries like Nigeria and Ghana, the frequency of the trait ranges from 15% to 30%. However, in Uganda, there are significant variations among tribes with the Baamba tribe having a frequency as high at 45%. The variability in this distribution is linked to the survival advantage against malaria that the sickle-cell trait confers on individuals. Therefore, areas with a high malaria transmission have high frequencies of this mutated gene (WHO, 2006). A study conducted in Ghana has estimated about 1.9% of newborns having some form of SCD (Ohene-Frempong et al., 2008).

2.3 Complications of Sickle Cell Disease

People with SCD usually suffer from a lot of complications due to the disease. Makani et al., (2013) categorised the complications into four, namely, haemolysis and haematological complications, vaso-occlusion, organ dysfunction and infection.

2.3.1 Haemolysis and Haematological Complications

The haematological complications are marked by the chronic haemolytic anaemia that persists throughout life. Chronic haemolytic anaemia in SCD is due to the shortened lifespan of sickled RBCs. Unlike normal RBCs with a lifespan of 120 days, sickled RBCs have a lifespan between 10-20 days (Kanter & Kruse-Jarres, 2013). The rapid destruction of RBCs causes hyperbilirubinemia and subsequently leads to jaundice and gall bladder diseases. Aside chronic haemolytic anaemia, SCD patients sometimes have intermittent sessions of reduced haemoglobin known as anaemic crisis (Makani et al., 2013). This combined with chronic haemolytic anaemia is known as hyperhaemolysis crises and it is defined by a sudden fall in steady state haemoglobin coupled with increased reticulocytosis and exaggerated hyperbilirubinemia (Makani et al., 2013). Acute splenic sequestration is very common in

children and Makani et al. (2013) adds that it can also be a cause of anaemia because when red blood cells are trapped in the spleen there is a reduced number of circulating RBCs.

2.3.2 Vaso-occlusion

Vaso-occlusion which is thought to be the primary cause of the painful crises period in SCD occurs secondary to blockage of the blood vessels by the sickled RBCs (Chakravorty & Williams, 2015; Mulumba & Wilson, 2015; Serjeant, 2013). As mentioned earlier, polymerisation of Hb S when deoxygenated leads to the formation of strands of Hb that cause the sickled shape of the RBCs (Stuart & Nagel, 2004). The polymerisation also makes the cell rigid and increases its adhesion to the endothelium of the microvasculature. This can lead to blockage of the blood vessels which can progress to tissue ischaemia and necrosis (Piel et al., 2017). Vaso-occlusive crisis is named as the most common cause of frequent hospitalisations and pain episodes in SCD (Mulumba & Wilson, 2015; Serjeant, 2013; Stuart & Nagel, 2004). However, some patients manage this episode at home using hydration, anti-inflammatory drugs, opioid and non-opioid drugs (Stuart & Nagel, 2004).

2.3.3 End-organ Dysfunction

As the survival rate of SCD patients' increases, there is a rise in the destruction of major organs (Makani et al., 2013). End-organ dysfunction can be as a result of vaso-occlusion and the damage can affect any organ system. Given that, when blood vessels supplying an organ are occluded, there is decreased blood flow to the organ leading to ischaemia and finally tissue necrosis. Some organs that are mostly affected are the lungs and brain leading to acute chest syndrome (ACS) and stroke respectively (Kanter & Kruse-Jarres, 2013). Stroke and ACS are reported to be very common in SCD and are risk factors for mortality (Makani et al., 2013).

2.3.4 Infections

People with SCD are highly susceptible to infections from encapsulated bacteria such as *Streptococcus pneumonia, Neisseria meningitis* and *Haemophilus influenza* (Bender, 2003 & Makani et al., 2013). Reports prove that prophylactic penicillin and vaccination programmes significantly reduce mortality (Bender, 2003). According to Makani et al. (2013), the increased risk to infection is attributed to the low immunity and complications and/or treatment of the disease itself. For example, patients with splenic dysfunction have an increased risk to septicaemia and meningitis because the spleen is a very vital organ in the immune system. Iron overload due to frequent blood transfusions can also increase risk of infections due to *Yersinia Enterocolitica.* End-organ damage to liver, lung, skin and kidney leave these sites susceptible to infections (Makani et al., 2013). The reduced immunity is in part related to the poor nutritional status of people with SCD (Makani et al., 2013).

2.4 Nutritional Status of People with SCD

Numerous studies have documented poor nutritional status which is characterised by underweight, stunting and wasting in SCD patients. Anthropometric, dietary and biochemical data have been used in the assessment of nutritional status of people with SCD.

2.4.1 Anthropometry of Adolescents and Adults with SCD

Anthropometry entails taking body measurements of an individual and comparing them to reference standards (Litchford, 2017). This gives an indication of growth and development over a period time. Litchford (2017) mentions that anthropometric indices are good predictors of over-nutrition and undernutrition. Common anthropometric indices that have been used to assess nutritional status in adults include Body Mass Index (BMI) and waist-to-hip ratio as

predictors of overweight and risk of developing cardiovascular diseases respectively (Litchford, 2017).

People with SCD usually have a thinner outlook compared to their peers and available anthropometric data shows growth deficits in this group of people regardless of their geographic location. In a review by Al-Saqladi et al. (2008) on the growth and nutritional status of people with homozygous SCD, it was documented that most of the subjects in the 46 studies reviewed had growth deficits. Sobo et al., (2007) found that underweight was prevalent among SCD patients in Ogun State, Nigeria with the mean BMI being 17.66±2.41 kg/m^2. A comparison of BMI between people with SCD and non-SCD persons also showed that unlike the non-SCD group, overweight and obesity was not prevalent in the SCD group. Thus the BMI of the SCD group were mostly underweight or normal (Aminu et al., 2017). In contrast, another study conducted among SCD patients of African descent living in London reported patients having a normal BMI with a mean BMI of 24.56±4.69 kg/m^2 (Osuobeni et al., 2009). Although Helvaci and Kaya (2011) also reported significant variance between the mean BMI of non-SCD patients and SCD patients, with the SCD group recording a lower BMI than control, the mean BMI was 20.7 ± 2.9 which is within the normal range.

Cases of overweight and obesity have also been reported among people with SCD especially in developed countries. Nearly half of the subjects in a study carried out in United States of America were reported to be overweight with 20% being obese (Pells et al., 2005). This is reinforced by a current study conducted by Kaufman et al. (2018) which reported that 44% of their study participants were overweight, that is, BMI ≥25 kg/m^2 .

There is paucity of data regarding the nutritional status of adolescents and adults with SCD living in Ghana. Most data in Ghana seems to focus on the nutritional status of children with SCD and these studies have shown that wasting, stunting and underweight are very prevalent among SCD children in the country (Bonsu, 2017; Boadu, 2016 & Osei-Yeboah, Rodrigues, & Enweronu-Laryea, 2011). Drawing from these findings, it is likely that the story may be similar amongst adolescents and adults. For the adolescents, poor nutritional status may even be more prominent since puberty puts additional metabolic stress on the body.

2.4.1.1 Body Composition of SCD adolescents and adults using Bioelectric Impedance Analysis

The Bioelectric Impedance Analysis (BIA) scale can also be used to determine the body composition of an individual in terms of percentage body fat and body mass and has been widely used to assess nutritional status in different populations (VanderJagt, Okolo, Rabasa, & Glew, 2000). The principle of operation is that tissues contain electrolyte-containing fluids and conduct an electric current. Resistance (R, opposition to flow through intra- and extracellular ionic solutions) and reactance (Xc, extra opposition from the capacitant effect of cell membranes and tissue interfaces) affects current flow. Impedance (Z), is expressed as a composite of R and Xc. The association between Xc and R in circuits is given by phase angle (PA), arctangent of Xc/R. A lower PA is related with cell loss whilst a high PA indicates increased body cell mass (VanderJagt et al., 2000).

Using this technique, VanderJagt et al. (2000) conducted a study in Nigeria which compared the body composition of children and adolescents with SCD to healthy controls. Their study saw a significant difference in percentage body fat (%BF) and percentage fat free mass (%FFM) between subjects aged 10years and above in the two groups. The cases had a significantly lower and higher %FFM and %BF respectively compared to controls.

A similar study was conducted in the United States and contrary to the Nigerian study, there were significantly lower and higher %BF and %FFM respectively between SCD males and controls (VanderJagt, Harmatz, Scott-Emuakpor, Vichinsky, & Glew, 2002). Unlike the Nigerian study which recorded significant differences in the body composition of SCD females and controls, this study recorded no significant difference between cases and controls (VanderJagt et al., 2002).

2.4.2 Dietary Intake and Patterns of SCD Adolescents and Adults

Assessment of dietary intake and patterns can be done using numerous tools like the 24-hour recalls, food frequency questionnaire and weighed food record (Raymond & Couch, 2017). Poor nutritional status in SCD is often attributed to inadequate dietary intake. This has led researchers to investigate the dietary intakes of this group of people. Early studies have shown that male SCD patients consumed significantly less total calories, carbohydrates, fat and protein daily compared to controls however there was no significant difference the dietary intake of SCD females and their controls (Modebe & Ifenu, 1993). Kawchak et al., (2007) followed Hb SS children and adolescents for four years and assessed their dietary intake over these years by collecting 24-hour dietary recalls daily. Analysis of the dietary data showed suboptimal energy and nutrient intake with vitamin D, folate, vitamin E and calcium being the most deficient nutrients in the diets of participants. More importantly, the study showed a reduced dietary intake among adolescents with protein, vitamin C, riboflavin, vitamin B-12 and magnesium consumption being significantly low. In contrast, a study among Jamaican cohorts documented an increased consumption of fats and carbohydrates, however, protein intake was within normal ranges (Anglin et al., 2011). Carbohydrates and fats intake exceeded the Dietary Reference Intake by 51% and 48% respectively. Anglin et al.

(2011) further documented that except for vitamin D and E, intake of the other vitamins was higher than the Recommended Daily Allowance. Calcium intake was also higher than the Adequate Intakes.

Tanabe et al. (2010), documented that subjects consumed a healthy diet made of fruits, whole grain and vegetables with a limited intake of fast foods and refined foods. Changes in feeding behaviour have also been reported during painful episodes as a study showed that 87% of its subjected consumed less food when in pain (Pells et al., 2005). In this study, subjects reported a decrease in fat and protein intake but not sugar and salt.

Although there may be contrasting findings on the dietary intake of SCD adolescents and adults, most studies report suboptimal energy and nutrient intake compared to healthy controls. This is usually attributed to reduced appetites especially during painful episodes and frequent hospitalisations (Hyacinth et al., 2010; Mitchell, Kawchak, Stark, Zemel, & Ohene-Frempong, Kweku Stallings, 2004).

2.4.3 Biochemical Assessment of Nutritional Status in SCD

Dietary intake may not paint the true nutritional status picture due to reporting biases. Additionally, intestinal malabsorption can impede absorption leading to no correlation between dietary intake and serum/plasma levels of nutrients. Biomarkers are therefore used as better predictors of nutrient deficiencies or excesses. Although they present objective ways of assessing nutritional status, identifying the ideal biomarker is quite challenging because numerous factors may affect them. For instance, serum albumin which historically used to be a common biomarker for assessing nutritional status has been proven to be an unreliable marker for assessing protein-energy malnutrition (Bharadwaj et al., 2016). Other biomarkers

like prealbumin, retinol-binding protein and transferrin have also been shown to be weak predictors of malnutrition especially when the person has a chronic disease condition (Bharadwaj et al., 2016).

To make things easier, some studies use analysis of individual nutrients as indicators of specific nutrient deficiencies. In SCD, common nutrient deficiencies that have been identified are zinc, iron, magnesium, vitamin C, D and E (Mohanty et al. 2017; Hyacinth et al. 2010; Al-Saqladi et al. 2008; Mohanty et al. 2008).

Hyacinth et al. (2010) cites zinc as the most studied micronutrient in SCD. One of the earliest observations of zinc deficiency among adult SCD patients was made by Prasad and colleagues. in 1975 (Prasad et al., 1975). In this study, it was seen that plasma, erythrocyte, urinary and hair zinc levels were low in study subjects giving an indication of zinc deficiency. On the contrary, Abshire, English, Githens and Hambidge (1988) found that zinc deficiency was not prevalent amongst young adults assessed in their study.

Apart from zinc, the antioxidant vitamins have also been widely studied because of the role they play in vaso-occlusion and haemolysis. These studies have found deficiencies in people with SCD. Oyeyemi, Sonuga and Oyebanji (2017) found reduced serum vitamin C and E levels amongst Hb SS patients as compared to that of carriers (AS) and non-carriers (AA). An assessment of the vitamin E status of steady state Hb SS patients also showed a decrease in serum levels compared to healthy controls (AA) (Tukur et al., 2015). The reduction in these vitamins supports the hypothesis that SCD puts oxidative stress on the cells and causes a decrease in level of antioxidant vitamins (Oyeyemi et al., 2017). Furthermore, the deficiencies seem to occur at a very early age and runs through adulthood.

Quite recently, there has been a growing interest in the role of inflammation on nutritional status. Therefore, there are studies where inflammatory markers have been used as predictors of nutritional status in certain chronic inflammatory diseases. For example, elevated C-reactive protein (CRP) levels is considered as a good predictor of cancer cachexia and is sometimes used by some researchers in the definition cancer cachexia (McMillan, 2009). Another study that explored association between CRP markers and nutritional status of people with Chronic Obstructive Pulmonary Disease (COPD) found that CRP levels increased when subjects anthropometric measurements like skinfold thickness, mid upper arm circumference and BMI decreased (Arora et al., 2010).

A linkage has also been made between nutrient deficiencies and disease severity. This has led to the investigations on the effect of nutrition in the management of SCD.

2.5 Factors That Contribute to Poor Nutritional Status in SCD

Poor nutritional status refers to over or undernutrition. In SCD this usually presents as undernutrition and is characterised by growth deficits like wasting, stunting and underweight. Various factors have been linked to the poor nutritional status of people with SCD. Some of these include inadequate dietary intake, increased catabolism and intestinal malabsorption. Recently, the most common mechanism cited by researchers as the cause of the poor nutritional status in SCD is the hypermetabolic nature of the disease (Hyacinth et al., 2010). This has been shown to cause an increase in the energy and nutrient demands.

According to Hibbert et al. (2006), the rapid rate of erythropoiesis and increased cardiac output have been suggested as the driving force for the hypermetabolism. Inflammation is

also cited as a factor for the hypermetabolic nature of the disease (Bandeira et al., 2014 & Keikhaei et al., 2013).

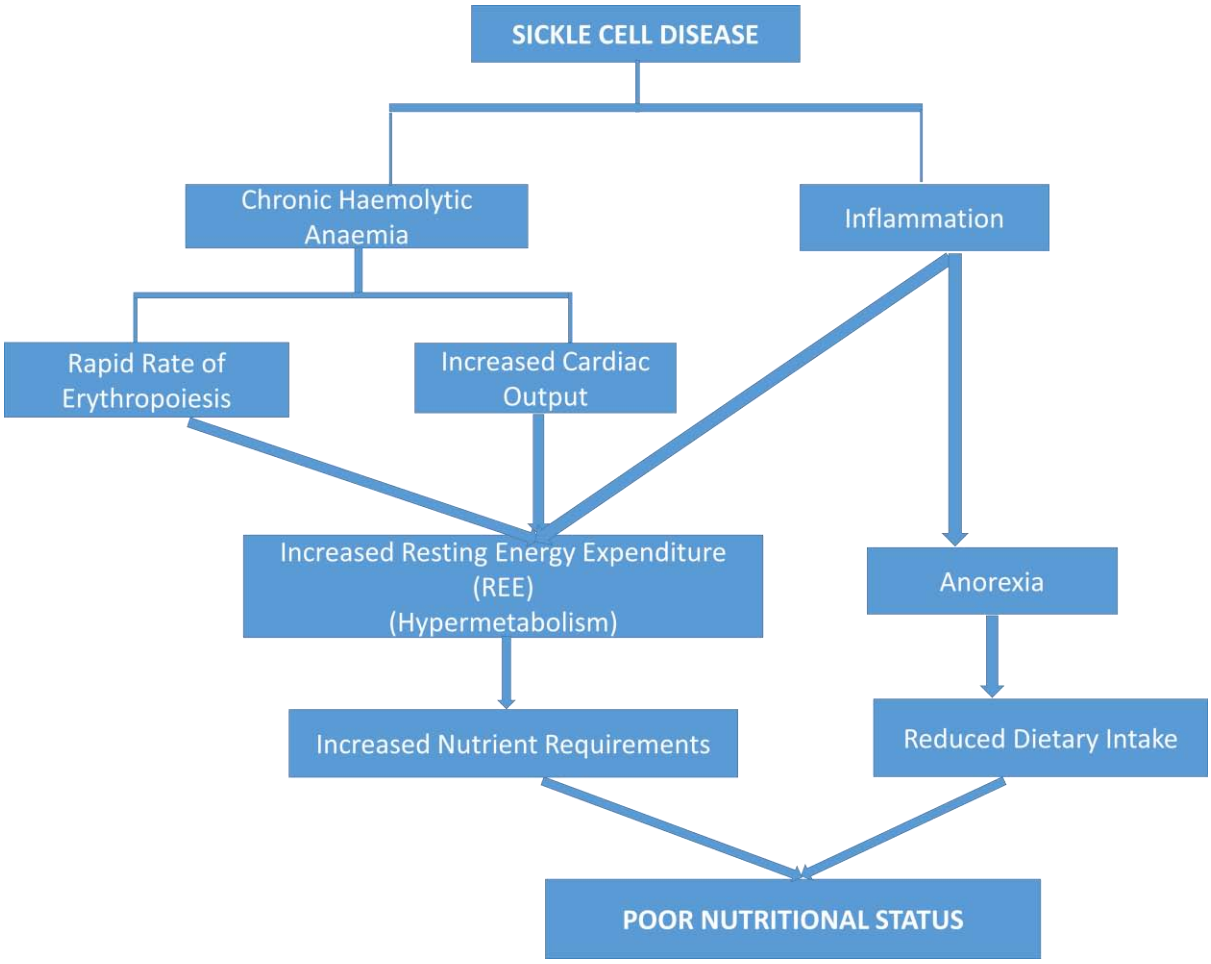


Figure 1: Factors that Contribute to Poor Nutritional Status in SCD (Author's own construct)

2.5.1 Chronic Haemolytic Anaemia and Nutritional Status

Unlike normal RBCs with a lifespan of about 100-120 days, sickled RBCs have a life span of 8-25 days (Hyacinth, Adekeye, & Yilgwan, 2013). The rapid destruction of these cells create a need for increased erythropoiesis; this leads to increased protein turnover and subsequently increased energy demand (Hibbert et al., 2006).

Hibbert et al. (2006) also observed that there is an increase in myocardial energy demand in Hb SS patients due to an increased heart rate. This is explained as a compensatory

mechanism to maintain constant tissue oxygenation since anaemia leads to low RBC count. The rise in myocardial energy requirement increases the resting energy expenditure (REE), which is the energy the body expends at rest, of SCD patients. This indicates that most of the energy is directed from building muscle for growth and other functions to meeting the increased myocardial energy demand and erythropoiesis (Hibbert et al., 2006).

2.5.2 Inflammation and Nutritional Status

Inflammation is a very common phenomenon in SCD as the disease is even described as a chronic inflammatory disease (Bandeira et al., 2014). This chronic inflammatory state is attributed to the nature of the RBCs in SCD and infections (Makani et al., 2013; Lee, 2006 & Stuart & Nagel, 2004). Sickled cells have an increased adhesiveness to the endothelium even during their steady state. The pathophysiology of vaso-occlusion, a popular complication in SCD is linked to this adhesive property of sickled cells which is a stimulator of inflammation (Lee, 2006). Increased adhesion of these cells to the endothelium leads to chronic activation and damage of the endothelium as well as increased production of inflammatory mediators like (IL-1, IL-6, IL-8, TNF-α) by endothelial cells (Keikhaei et al., 2013). Conversely, the presence of these cytokines also exacerbate inflammation as they activate the vascular endothelium, elevate sickle cells adhesion to the endothelium and ultimately lead to painful vaso-occlusive crisis (Keikhaei et al., 2013 & Pathare et al., 2004). According to Hibbert et al.(2005), CRP, a positive acute phase reactant, levels were raised in steady state Hb SS patients and this correlated strongly to resting energy expenditure (REE). This gives an indication that chronic inflammation leads to hypermetabolism which can consequently affect nutritional status. This is because a high REE confirms that the body is using a lot of energy and an inadequate supply of energy will lead to depletion of body stores.

In their study on pro-inflammatory cytokines and hypermetabolism in children with SCD, Hibbert et al. (2005) also found that steady state Hb SS patients had elevated levels of IL-6. Interleukin-6 is an inflammatory cytokine that has been reported as a strong predictor of malnutrition especially disease-induced malnutrition (Hyacinth et al., 2010). This cytokine is reported to cause anorexia, weight loss, changes in the lipid and protein metabolism, increased concentrations of catabolic hormones, reduction of anabolic hormones and metabolic impairment (Belizário, Oliveira, Borges, Kashiabara, & Vannier, 2016). Since the disease presentation is similar for all age groups, similar trends may also be observed among adolescents and adults. Tsujinaka et al. (1998) found that elevated levels of IL-6 in transgenic mice correlated positively with muscle wasting since IL-6 is considered as a proteolysis-inducing factor because of its modulation of muscle proteolytic systems.

Aside the nature of sickled RBCs contributing to inflammation, frequent infections is also a contributor to the chronic inflammatory nature of the disease. As stated earlier, SCD patients are particularly highly susceptible to infections from microorganisms such as *Streptococcus pneumonia, Neisseria meningitis* (Bender, 2003). Since inflammation is a natural response to any foreign agent in the human body, a state of recurrent infections will lead to frequent inflammation in attempts to remove the injurious agent.

2.6 The Role of Nutrition in SCD

Hyacinth et al. (2013) document that several studies report nutritional deficiencies, that is, macro and micronutrients deficiencies in SCD patients. Attempts have also been made by some researchers to examine the efficacy of nutritional interventions on improving clinical outcomes of the disease. Adequate nutrition has been indicated as a means of decreasing severity the disease and Mandese et al. (2016) documented that children with poor nutritional

status had an increase in the risk of disease severity. Some indicators of disease severity are the number of pain episodes, hospitalizations, total haemoglobin, foetal haemoglobin and lactose dehydrogenase (Mandese et al., 2016).

Antioxidant supplementation prevents the oxidative damage to erythrocytes and this leads to reduced frequency of haemolysis (Hundekar et al., 2011). Prophylactic folic acid is also given to patients to prevent the development of folate deficiency due to increased folate turnover (Hyacinth et al., 2010).

These interventional studies have recorded improvements in disease outcomes such as improved growth, decreased pain episodes and frequency of hospitalisation (Hundekar et al., 2011; Williams et al., 2004; Ohnishi et al., 2000). This gives a hint that some of the complications of the disease have nutritional underpinnings just like they have genetic underpinnings. Nutrition interventions also seem to be the most affordable and readily accessible treatment option especially considering that majority (about 75%) of the people with SCD are in Africa, a resource poor area (Hyacinth et al., 2013; WHO, 2006).

Evidence of the importance of nutrition in the management of the disease is not only from experimental studies. However, some of these nutritional modifications are being practiced by patients sometimes without the guidance of a healthcare provider. SCD patients have also mentioned that they use nutrition as a coping strategy in managing the disease (Tanabe et al. 2010). This is to say that they employ nutritional strategies and techniques like following a healthy diet consisting of fruits, vegetables, and whole grain fibre; and reducing intake of fast foods and refined foods to help improve their health and disease outcome (Tanabe et al., 2010).

Although nutrition has been identified as an important means of managing the disease, patients have challenges in making the right meal choices especially in an era where there is a lot of meal options due to food processing. This coupled with reduced dietary intake due to chronic inflammation and frequent hospitalisations poses a serious threat to the nutritional status of people with SCD. This calls for serious interventions to help manage the situation and improve overall quality of life of these people. Given the important influence of nutrition knowledge on dietary intake and nutritional status, it will be thought-provoking to assess nutrition knowledge and its possible association with nutritional status of people with SCD.

CHAPTER THREE

3.0 METHODOLOGY

3.1 Study Design

A cross-sectional study design was used.

3.2 Study Site

The study site was the Ghana Institute of Clinical Genetics (GICG) at Korle Bu. The GICG is located between the fevers unit of the Korle Bu Teaching Hospital and the Korle Bu Polyclinic. Established in 1947, the institute provides comprehensive outpatient care for people aged 13 years and above with sickle cell disease. As the first adult sickle cell clinic in Ghana, it has the largest number of registered sickle cell adolescents and adults. Patients from all over Ghana can attend the clinic however most patients are usually from the southern part of Ghana. The clinic operates every weekday except for holidays and has an average attendance of 50 patients per day.

3.3 Study Population

3.3.1 Inclusion Criteria

- All patients aged (16 years and above) diagnosed with SCD attending the clinic in steady state and recruited for the SickleGenAfrica project. The vision of the SickleGenAfrica project is to align the survival of individuals with the disease in Africa with national norms. Understanding and improving the nutritional knowledge and status of the people with SCD will help improve on the survival of these group of people.

3.3.2 Exclusion Criteria

- Patients with other chronic diseases that can affect growth like HIV, cancer, chronic kidney disease, congenital heart disease and cerebral palsy.

- Patients who were unwell or have recently been unwell (not less than a week). This is because levels of IL-6 would increase under this condition.
- Patients who were recently transfused (<3 months). This is because transfusion can lead to iron overload and subsequently depletion of vitamin E (Marwah et al., 2002). Thus, if these patients are used, their serum vitamin E may not be a true representation of their vitamin E status.
- Patients who are pregnant or breastfeeding because during these phases of life there is weight gain and this will not give a true representation of the patient's weight.

3.4 Sampling Technique

Convenience sampling technique was used. At the clinic, patients were approached individually and briefed about the study. Interested patients who met the inclusion criteria were enrolled for the study.

3.5 Sample Size Determination

In studies designed to measure a characteristic in terms of a proportion, the equation for calculating sample size is $N = \frac{Z^2 \times P(1-P)}{d^2}$ (Wayne, 1999)

Where N – sample size

Z_{crit} – 1.96 for a confidence interval of 95%

d – Allowable error = 0.075

P – Estimate of proportion of population with adequate nutritional knowledge and good nutritional status = 50 %

P was set at 50% because there is paucity of data on the nutrition knowledge of SCD patients in Ghana. Thus, P at 50% estimates that about half of the population SCD patients have adequate nutritional knowledge.

Therefore, $N = \frac{(1.96)^2 0.5\ (0.5)}{0.075^2} = \frac{3.84\ (0.25)}{0.0056} = 171.4 \approx 171$ people

3.6 Data Collection

An interviewer administered questionnaire was used to gather data on the socio-demographic details and nutritional knowledge of participants. Nutritional status of sickle cell patients was assessed using anthropometric and biochemical data. Food consumption pattern was assessed using a one-week food frequency questionnaire. The anthropometric and dietary data were collected by the researcher. The estimated time for data collection was 25-30 minutes per participant.

3.6.1 Socio-demographic Data

A structured questionnaire was used to obtain socio-demographic information such as age, gender, educational level and sickle cell disease type.

3.6.2 Nutritional Knowledge Data

Nutritional knowledge was assessed using a modified pre-tested questionnaire adapted from Tandoh (2015). The questionnaire contains only close-ended questions. The questionnaire is categorised in three sections: the first section assessed the knowledge of participants on food sources of some nutrients, the second section - knowledge on some nutrient deficiencies (focusing on the common nutrient deficiencies in SCD patients) and the last section - knowledge on expert opinion and healthy choices. A score of 1 was assigned to a correct answer and 0 to a wrong answer. A total score of more than half of the questions was categorised as high nutritional knowledge and below half as poor nutritional knowledge (Tandoh, 2015).

3.6.3 Nutritional Status Data

3.6.3.1 Anthropometric Data

All anthropometric measurements were taken using standard procedures.

Height

The Seca 786 mechanical column scale stadiometer (Hamburg, Germany) was used to measure the height of participants. Participants stood erect on a base plate without shoes with their heads in Frankfurt's plane position and back straight, feet together and heels touching the back of the plate. The head plate was lowered to touch the top of the head and height recorded. The heights were recorded to the nearest 0.1cm.

Weight

The Omron BF511 body composition scale was used to determine the weight as well as other body composition parameters like body fat, muscle mass and visceral fat. To do this, participants' personal data was entered into the unit. Immediately the display screen shows 0.0 kg, they were asked to stand on the scale barefooted with their knees and back straight and feet positioned on the electrodes whilst holding the display unit in their hands firmly. With the display unit in their hand, they were required to press their palms firmly on the grid electrode, raise their arms and extend their elbows straight to form a 90º angle to their bodies. Participants were asked to step off the scale after their weights have been displayed. Weight readings were recorded to the nearest 0.1kg.

3.6.3.2 Dietary data

A modified pretested Food Frequency Questionnaire (FFQ) by Asante, Pufulete, Thomas, Wiredu, & Intiful, (2015) was used to collect information on the dietary pattern of adults with sickle cell disease over a one-week period. The questionnaire contained a list of common Ghanaian foods, participants were required to recall the number of times they had eaten a particular food within the week prior to data collection.

3.6.3.3 Biochemical data

About 0.15 ml of serum obtained from the SickleGenAfrica project stored at -80ºC was used to assess biomarkers like serum vitamin E and interleukin-6 levels. Among the antioxidant vitamins, vitamin E deficiency has been identified in a lot of SCD patients. Also because it has antioxidant properties, deficiencies will contribute to haemolysis and subsequently poor growth due to the hypermetabolic burden of haemolysis (Hyacinth et al., 2010). A link has also been established between CRP and IL-6 and muscle wasting, higher levels correlates with increased muscle wasting (Belizário et al. 2016 ; Archer et al. 2008).

Procedure for analysing vitamin E levels

Enzyme-Linked Immunosorbent Assay (ELISA) kit from Kamiya Biomedical Company (vitamin E (VE) ELISA Cat. No. KT-32385) was used for this analysis. This kit employed the competitive inhibition enzyme immunoassay technique to determine the serum vitamin E levels of participants. Standards, controls and samples were pipetted into specific wells on a microplate pre-coated with monoclonal antibody specific to vitamin E. A biotin labelled vitamin E (Detection reagent A) was added to each well and a competitive inhibition reaction was launched between the pre-coated antibody specific to vitamin E and the Detection reagent A plus standard or control or sample. After 1hour of incubation at 37ºC, the microplate was aspirated and washed with an automatic washer. Excess water was blotted out against an absorbent paper to prevent dilution of solutions that will be added later. Next, avidin conjugated to Horseradish Perioxidase (HRP) was added to each well and also incubated for 30 minutes at 37ºC. The aspiration and wash procedure was repeated after incubation. 3, 3′, 5, 5′-Tetramethylbenzidine (TMB) substrate solution was added to each well and the plate was incubated for 13 minutes at 37ºC. A blue colour was observed in most of the wells. The intensity of the colour was inversely proportional to the concentration of

vitamin E in the sample. After incubation, a stop solution was added to each well and the blue colour changed to yellow, this was mixed thoroughly by gently tapping the sides of the plate.

The absorbance of each well was read using a spectrophotometer at a wavelength of 450nm. A standard curve was plotted and used to determine the concentration of vitamin E in the samples.

Procedure for analysing interleukin-6 (IL-6)

The Abcam IL-6 Human High Sensitivity ELISA kit which employed the sandwich ELISA technique was used for this assay. Standards, controls and samples were pipetted into specific wells on a microplate pre-coated with monoclonal antibody specific for interleukin-6. A Biotinylated anti-IL-6 was then added to each well, the plate was covered and incubated at 21ºC for 3 hours. After incubation, each well was aspirated and washed using an automatic washer. Excess water was blotted against an absorbent paper to prevent dilution of solutions that will be added. Next, Streptavidin-HRP solution was added to each well and incubated for 30 minutes at 21ºC. After incubation, the aspiration and wash procedure was repeated again. Chromogen TMB substrate solution was added to each well and incubated for 13 minutes at 21ºC in the dark by wrapping the plate in aluminium foil. There was a blue colour development of different intensities. After addition of the stop solution, the blue colour turned yellow.

The absorbance was read immediately using a spectrophotometer at a wavelength of 450nm. A standard curve was drawn and the concentrations of IL-6 calculated.

3.7 Data Handling

The raw data from this study was under restricted access by password protection. Answered questionnaires were stored in a locked cabinet which was accessible to only researchers and

also uploaded unto RedCap for safety. It was withheld from public access during the period of the research. Respondents were assigned random codes for the purpose of confidentiality.

3.8 Ethical Consideration

This project is under the SickleGenAfrica project and thus operated under its ethical clearance from the Ethical and Protocol Review Committee of the College of Health Sciences, University of Ghana. Additionally, informed consent was sought from all participants aged 18 years and above as well parents of participants below 18 years before they were recruited for the study. An assent was also sort from participants below 18 years.

This project benefitted from NIH/NHLBI grant for the SickleGenAfrica project, project number 1U54HL141011-01.

3.9 Data Analysis

Data was entered in Microsoft Excel and analysed using the Stata/IC 14.0.

Nutritional status was assessed using Body Mass Index (BMI) and participants were categorized into underweight, normal, overweight or obese based on WHO recommendations (World Health Organisation, 2009). For participants below 20 years BMI-for-age Z-scores were calculated using the WHO AnthroPlus software v1.0.4.

The biochemical tests, that is, serum IL-6 and vitamin E values were compared to reference values.

Descriptive analysis like means, percentages, standard deviations and frequencies were used to summarise continuous and categorical variables. Some continuous variables include age and height. Categorical variables include nutrition knowledge, nutritional status, sex and sickle cell type. Dietary data from the FFQ was presented as frequencies.

Chi-squared test was used to test the associations between the nutritional knowledge and nutritional status. A linear regression was used to test for the association between IL-6 and body weight. Multiple regression was used to test for the strength of this association. A p-value ($p < 0.05$) was considered as statistically significant.

CHAPTER FOUR

4.0 RESULTS

4.1 Socio-Demographic Characteristics of Participants

One hundred and seventy-three people were recruited for the study over a four month period. However, after confirmation of their Hb genotypes 2 people were excluded from the study. Table 1 shows the socio-demographic information of the study participants. The mean age was 26.7± 9.07 years with the male to female ratio being 1:1.5. Majority of the participants had sickle cell anaemia (Hb SS).

Almost all of the participants reside in the Greater Accra region (94.1%) and majority were Christians (88.3%). Majority were single (85.4%) and almost all (99.4%) of the participants had received some form of formal education. A little over half (58.5%) of the participants were either students or unemployed, they represented the group with no source of monthly income. However, for the working group, most of them (12.3%) earned between GH¢300-GH¢ 499 monthly.

Table 1: Socio-demographic characteristics of respondents (N = 171)

Variable	n (%)
Age groups	
<20	35 (20.5)
20-29	84 (49.1)
30-39	37 (21.6)
≥40	15 (8.8)
Gender	
Male	105 (61.4)
Female	66 (38.6)
Haemoglobin genotype	
SS	122 (71.4)
SC	49 (28.6)
Place of Residence	
Greater Accra	161 (94.1)
Central	8 (4.7)
Eastern	2 (1.2)
Religion	
Christian	151 (88.3)
Muslim	20 (11.7)
Marital Status	
Single	145 (84.8)
Married	25 (14.6)
Divorced/Widowed	1 (0.6)
Educational Level	
No formal education	1 (0.6)
Primary	11 (6.4)
JSS	23 (13.4)
SSS/SHS/Vocational	75 (43.9)
Tertiary	61 (35.7)
Occupation	
Formal	31 (18.1)
Informal	41 (24.0)
Unemployed	39 (22.8)
Students	60 (35.1)
Monthly Income	
No income	99 (57.9)
Below 300	18 (10.5)
GH¢ 700 - GH¢ 899	6 (3.5)
GH¢300- GH¢ 499	22 (12.9)
GH¢500 - GH¢ 699	14 (8.2)
≥GH¢ 900	12 (7.0)

4.2 Feeding Behaviour and Source of Nutrition Information

4.2.1 Feeding Behaviour

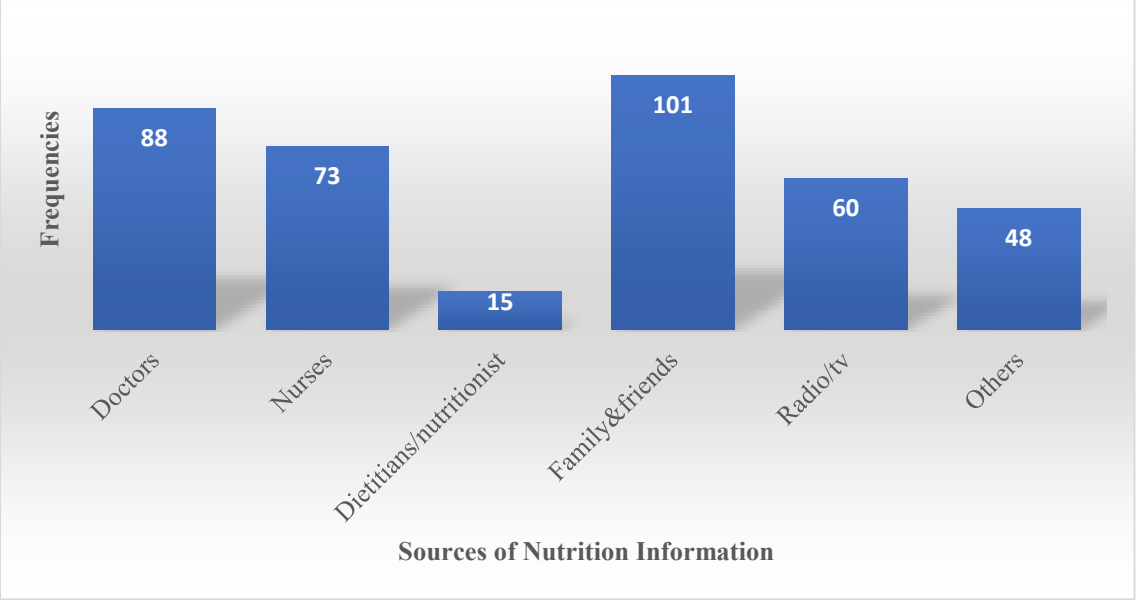
The participants were asked whether they had a good appetite for food or not for which they responded "yes" or "no". Majority (79.0%) responded that they had a good appetite for food. Except for one respondent who mostly ate once daily, all respondents ate more than once daily with about 49.1% of the participants eating twice daily.

Table 2: Feeding behaviour

Variable	n (%)
Appetite	
Yes	135 (79.0)
No	36 (21.0)
Meals per day	
Once	1 (0.6)
Twice	60 (35.1)
Thrice	84 (49.1)
>3x	26 (15.2)

4.2.2 Sources of Nutrition Information

Almost all participants (94%) had received some form of information about nutrition. Participants who had received some form of nutrition information were asked to identify sources of their information. Figure 3 shows the sources of nutrition information according to participants. The most frequent source of nutrition information was family and friends as illustrated in figure 4. This was followed by doctors and nurses. Dietitians and nutritionists are the least contacted for nutrition information. Some participants also got information from other sources like the internet, gym instructors and teachers.



*Others represents internet, gym instructors, teachers and magazines

Figure 2: Sources of nutrition information

4.3 Nutritional Knowledge of Participants

A 14-item questionnaire was used to assess the nutritional knowledge of participants. These questions are divided into three sub-topics as shown below. The first section (Table 3) assessed participants' knowledge on food sources of some nutrients. Most of the participants (80%) were able to identify food sources of the macronutrients, that is, carbohydrates, protein and fat. Less than half of the participants were able to identify the food source of iron and dietary fibre. Table 4 shows participants' knowledge on some nutrient deficiencies and for all questions, less than half of the participants were able to answer correctly. Majority of the participants knew what nutrition experts' recommend and what constituted a balanced diet as shown in table 5.

Table 3: Participants' knowledge on food sources of some nutrients (N = 171)

Variable	Proportion of participants who answered correctly n (%)
1. Cassava, yam and gari are rich sources of;	148 (86.6)
2. Chicken, eggs and fish are rich sources of;	148 (86.6)
3. Which of the following is a rich source of fat?	164 (95.9)
4. Milk and milk products are rich in;	98 (57.3)
5. Which of the following is a rich source of iron?	74 (43.3)
6. Which of the following is a rich source of dietary fiber/roughage;	62 (36.3)
7. Which of the following will give you blood?	167 (97.7)

Table 4: Participants' knowledge on some nutrient deficiencies (N = 171)

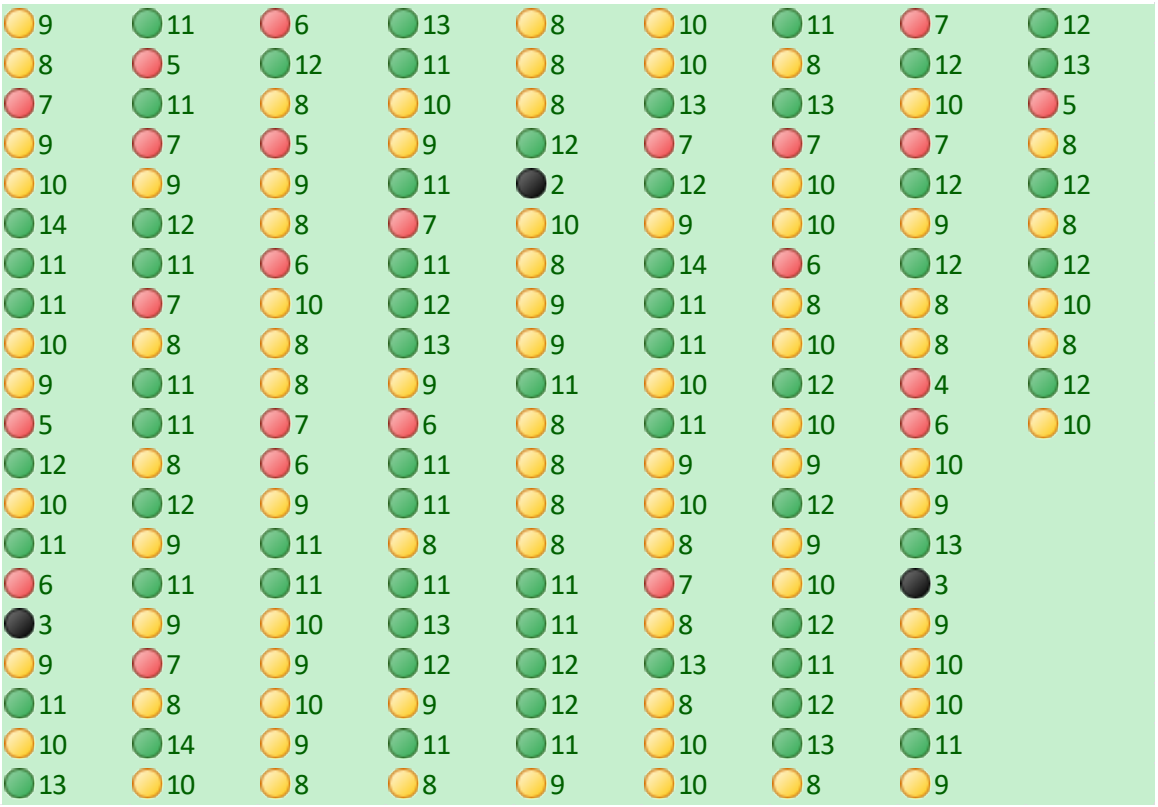
Variable	Proportion of participants who answered correctly n (%)
8. Inadequate intake of …….will make you look very thin;	78 (45.7)
9. Rickets/ bow legs are caused by lack of;	35 (20.5)
10. Anaemia is caused by lack of;	85 (49.7)
11. Bleeding of gums is caused by inadequate intake of;	77 (45.0)

Table 5: Participants' knowledge on expert opinion and making healthy meal choices (N=171)

Variable	Proportion of participants who answered correctly n (%)
12. Do you think fruits and vegetables should be eaten frequently?	168 (98.3)
13. Which of the following snacks is healthy?	161 (94.2)
14. Which of the following foods is balanced?	149 (87.1)

4.3.1 Nutrition knowledge scores and ranking

The nutrition knowledge questionnaire was scored; a correct answer was scored as 1 and a wrong answer, 0. Figure 3 shows a heat map of the nutrition knowledge scores of all participants. Based on the score, participants' nutrition knowledge was ranked as either low or high. A score from 1-7 was ranked as low and 8-14 was ranked as high. The mean score was 9.5±2.26, the minimum and maximum scores were 2 and 14 respectively. Majority (85%) of the respondents had high nutrition knowledge.



Keys: Black – below 4; red – 4 – 7; yellow – 8-10; green – 11 -14

Figure 3: Heat map of nutrition knowledge scores of participants

4.4 Nutritional Status of Participants

4.4.1 Body Fat Distribution among Males and Females

The percentage body fat and muscle mass of participants were estimated and ranked into low, normal, high and very high based on the standards of the manufacturers of the body composition monitor attached as appendix 4. The percentage body fat (%BF) of 85% of the participants was obtained. However, the %BF of 15% of the participants was not obtained due to technical errors that occurred during measurements. Out of the 85% that was obtained, females represented a greater majority (64%). The percentage body fat distribution among males and females is shown in tables 6 and 7 respectively. For both genders, a little over half of the participants had a normal percentage body fat. However, more males than females had a low body fat percentage. In contrast, more females than males had a high and very high

percentage body fat. The mean %BF for females and males are 30.6±7.04 and 11.9±5.82 respectively.

Table 6: Body fat levels of males

Age group (years)	Low n (%)	Normal n (%)	High n (%)	Very High n (%)	TOTAL N (%)
16	1 (50)	1(50)	-	-	2 (100)
18-39	15 (31.3)	28 (58.3)	3 (6.3)	2(4.1)	49(100)
40-59	-	2 (100)	-	-	2 (100)
TOTAL	16(30.8)	32(59.6)	3(5.8)	2(3.8)	52 (100)

Table 7: Body fat levels of females

Age group (years)	Low n (%)	Normal n (%)	High n (%)	Very High n (%)	TOTAL N (%)
16	-	3(100)	-	-	3 (100)
17	-	5(100)	-	-	5 (100)
18-39	3 (4.1)	39 (52.7)	25 (33.8)	7(9.5)	74 (100)
40-59	1 (9.1)	6 (54.6)	3 (27.3)	1 (9.1)	11 (100)
TOTAL	4 (4.3)	53(57.0)	28(30.1)	8 (8.6)	93 (100)

4.4.2 Muscle Mass Distribution among Males and Females

Although the percentage muscle mass (%MM) of 145 (84.8%) of the participants was obtained, only that of 135 participants could be ranked as low, normal, high or very high. This is because per the manufacturer's standards, the %MM of persons below 18years is not ranked and there were 10 participants below 18years.

Table 8 shows the %MM of males was skewed towards the high and very high end, this represented 86% of males. The mean %MM for males is 43.3±6.32. Majority (74.1%) of the females had a normal %MM as shown in table 9 with a mean of 28.1±3.17.

Table 8: Muscle mass levels of males

Age group (years)	Low n (%)	Normal n (%)	High n (%)	Very High n (%)	TOTAL N (%)
18-39	1 (2.1)	5 (10.4)	12 (25.0)	30(62.5)	48(100)
40-59	-	1 (50.0)	1 (50.0)	-	2 (100)
TOTAL	1(2.0)	6(12.0)	13(26.0)	30(60.0)	50 (100)

Table 9: Muscle mass levels of females

Age group (years)	Low n (%)	Normal n (%)	High n (%)	Very High n (%)	TOTAL N (%)
18-39	2 (2.7)	56 (75.7)	15 (20.3)	1(1.3)	74 (100)
40-59	1 (9.1)	7 (63.6)	3 (27.3)	-	11 (100)
TOTAL	3(3.5)	63(74.1)	18(21.2)	1(1.2)	85 (100)

4.4.3 BMI Categories of Participants

To determine overall nutritional status of participants, the WHO standard definition for underweight, normal, overweight and obese using Body Mass Index (BMI) was used. However, for participants below 20 years, BMI-for-age (BAZ) scores were used to classify them into underweight, normal, overweight and obese as recommended by WHO. Table 10 and 11 show the nutritional status of participants below 20 years based on BAZ scores and BMI categories respectively.

Table 10: Nutritional status of participants below 20years using BMI-for-age (BAZ) scores (N=35)

Nutritional status	n (%)
Underweight	1 (2.8)
Normal	33 (94.4)
Overweight	1 (2.8)

According to WHO, 2009; BAZ score: >2SD – Overweight; 1≥ x ≥ -2 SD – Normal; < -2 SD – Wasted / Underweight

Table 11: Nutritional status of participants 20years and above based on BMI categories (N =136)

Nutritional status	n (%)
Underweight	25 (18.4)
Normal	96 (70.6)
Overweight & Obese	12 (8.8)
Obese	3 (2.2)

BMI Interpretation: <18.5 kg/m^2 – Underweight; 18.5 – 24.9 kg/m^2 – Normal; 25.0 – 29.9 kg/m^2 – Overweight; ≥30 kg/m^2 – Obese

Combining the BAZ categories of participants below 20 years with the BMI categories of participants 20 years and above, the nutritional status of all participants was obtained and shown in figure 4. Majority of the participants were well-nourished with 25% being malnourished, that is, underweight, overweight or obese. Table 12 shows the nutritional status distribution per haemoglobin genotype, there was no significant difference between the nutritional status of participants in the two genotypes ($p = 0.09$).

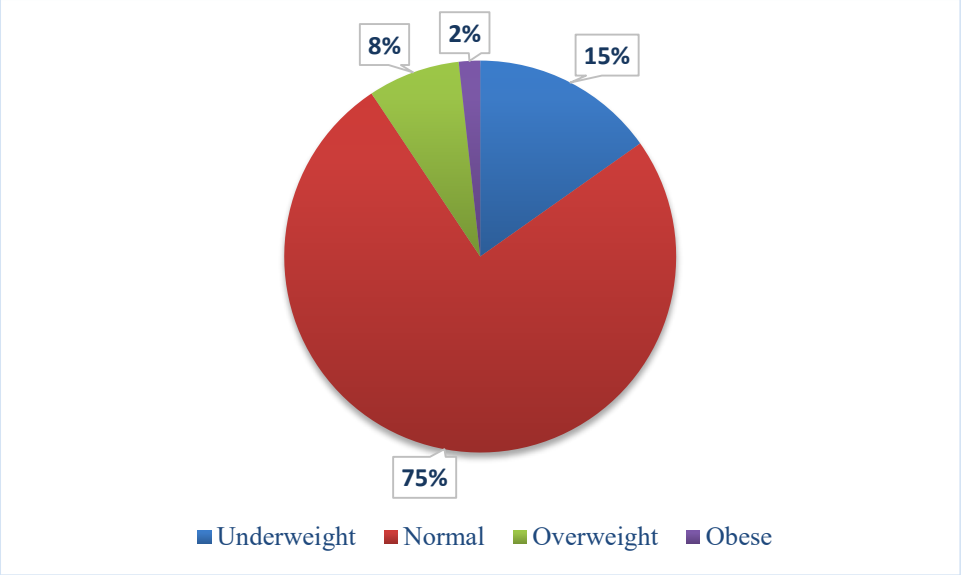


Figure 4: Nutritional status of participants

Table 12: Nutritional status per haemoglobin genotype

Haemoglobin genotype	Underweight n (%)	Normal n (%)	Overweight & Obese n (%)	Total N (%)	p-value
SC	5 (10.2)	36 (73.5)	8 (16.3)	49 (100)	0.09
SS	21 (17.2)	93 (76.2)	8 (6.6)	122 (100)	
Total	26 (15.2)	129 (75.4)	16 (9.4)	171 (100)	

4.4.4 Serum Interleukin-6 and Vitamin E levels of Participants

Out of the 171 participants, the serum IL-6 and vitamin E levels of 168 participants were analysed and the results presented in figures 5 and 6. Both figures show that the levels of these biomarkers in the participants were skewed to the right. The mean IL-6 and vitamin E level were 18.6±33.7 pg/ml and 6.7±9.9 µg/ml respectively. An independent t-test shows that IL-6 levels were significantly elevated in individuals with sickle cell anaemia compared to Hb SC individuals as shown in table 13.

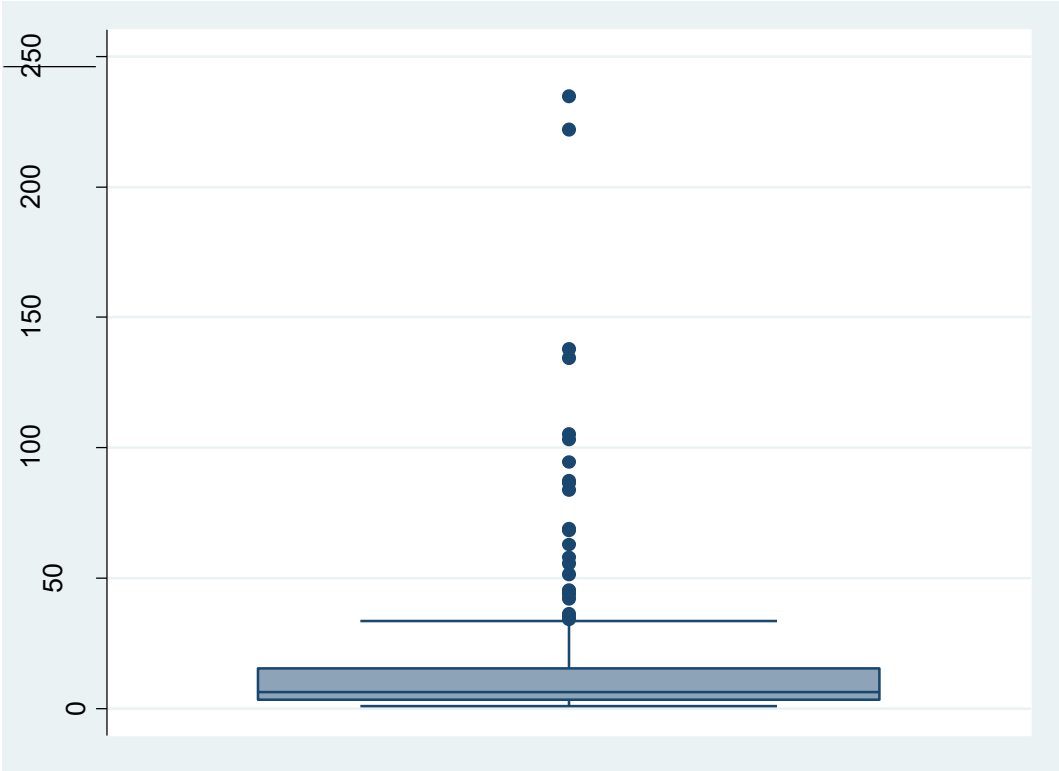


Figure 5: Serum interleukin-6 levels of participants

Table 13: Comparison of mean IL-6 levels between Hb genotypes

Hb genotype	Observations	Mean±SD	p-value
SS	121	11.19±2.7	0.02*
SC	47	21.49±3.4	

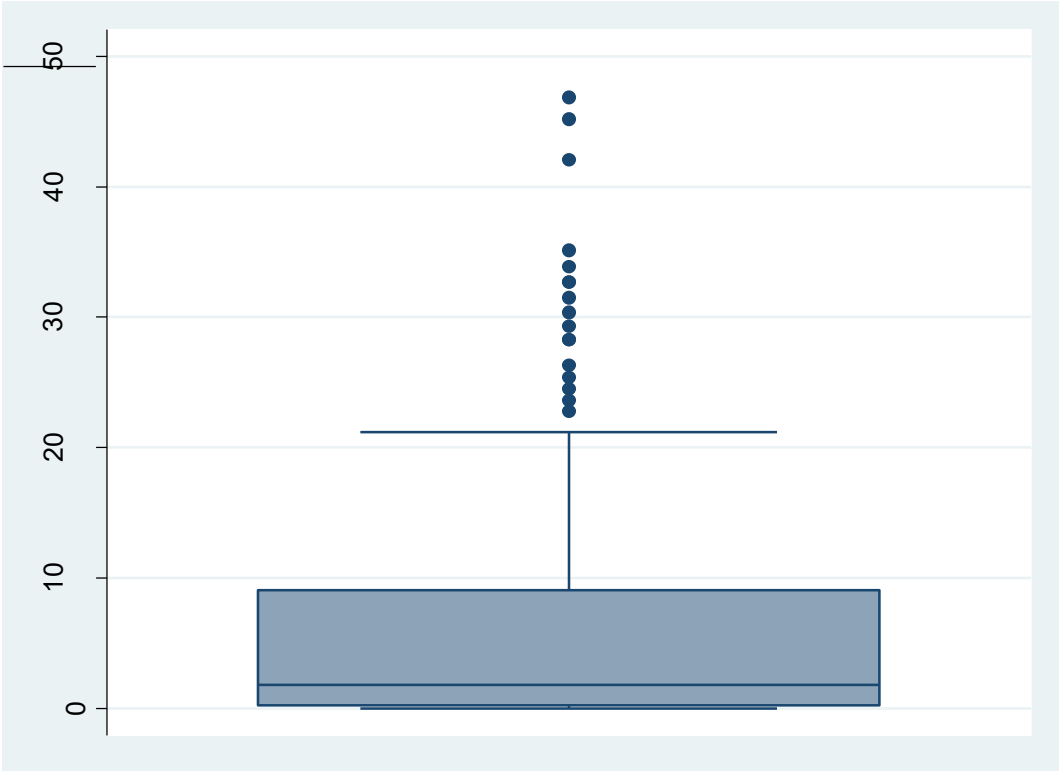


Figure 6: Serum vitamin E levels of participants

4.5 Food Consumption Pattern of Participants

Out of the 171 participants interviewed, the dietary pattern data of 168 was obtained. Figure 7-22 shows the dietary patterns of these participants over a one-week period. The food items are categorized into groups as shown in the figures below.

4.5.1 Pattern of fruit consumption

Figure 7 shows that fruit consumption was generally low amongst participants as more than half of them had not consumed the fruits mentioned except for banana which recorded almost 60% consumption rate. Grapes were the least consumed fruit and less than 10% of the participants consumed fruits almost daily. Most participants ate fruits once/twice in a week. Since the data was collected over a 5 month period and fruit availability is seasonal, the fruit consumption pattern was sub-divided into the various months and presented below. Banana recorded the highest frequency of consumption in all the months. Pawpaw, apples and citrus

recorded the second highest consumption rate depending on the month of assessment. Except for October, grapes were the least consumed fruit in all the other months.

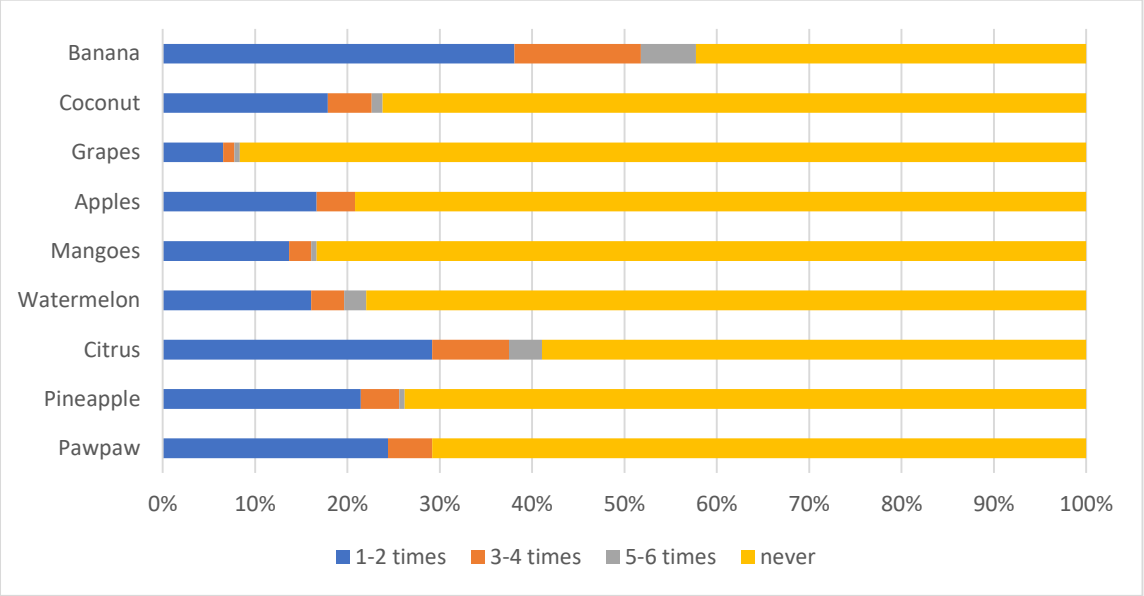


Figure 7: Daily fruit consumption over a one-week period

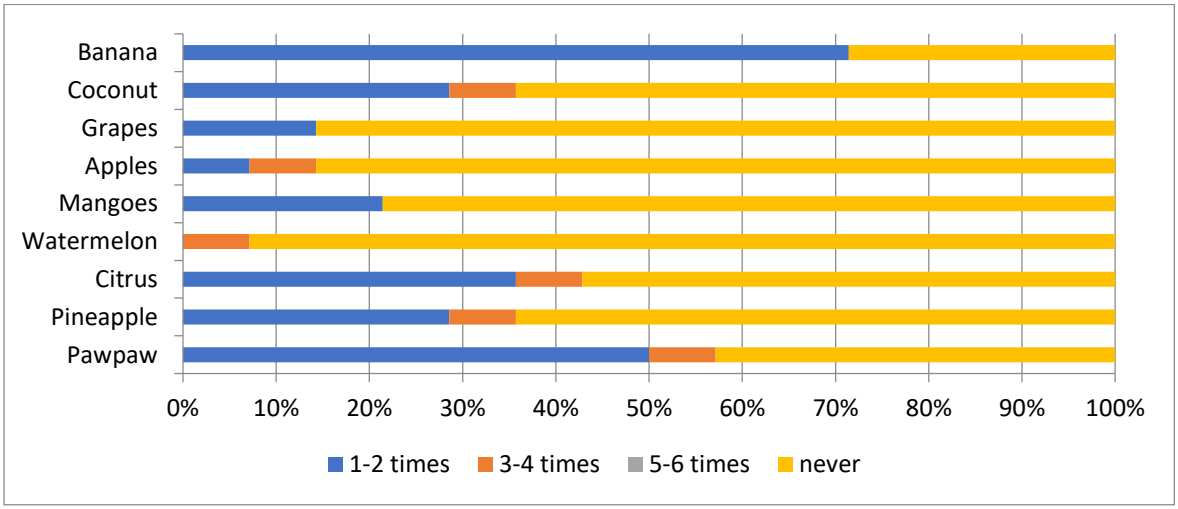


Figure 8: Daily fruit consumption in October over a one-week period

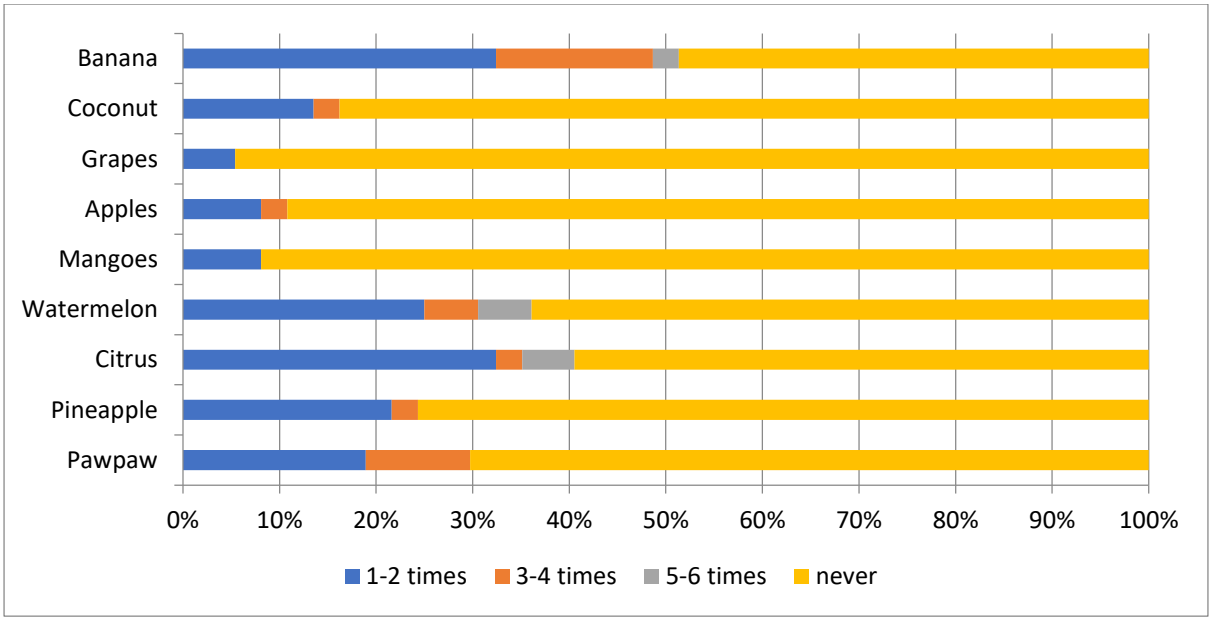


Figure 9: Daily fruit consumption in November over a one-week period

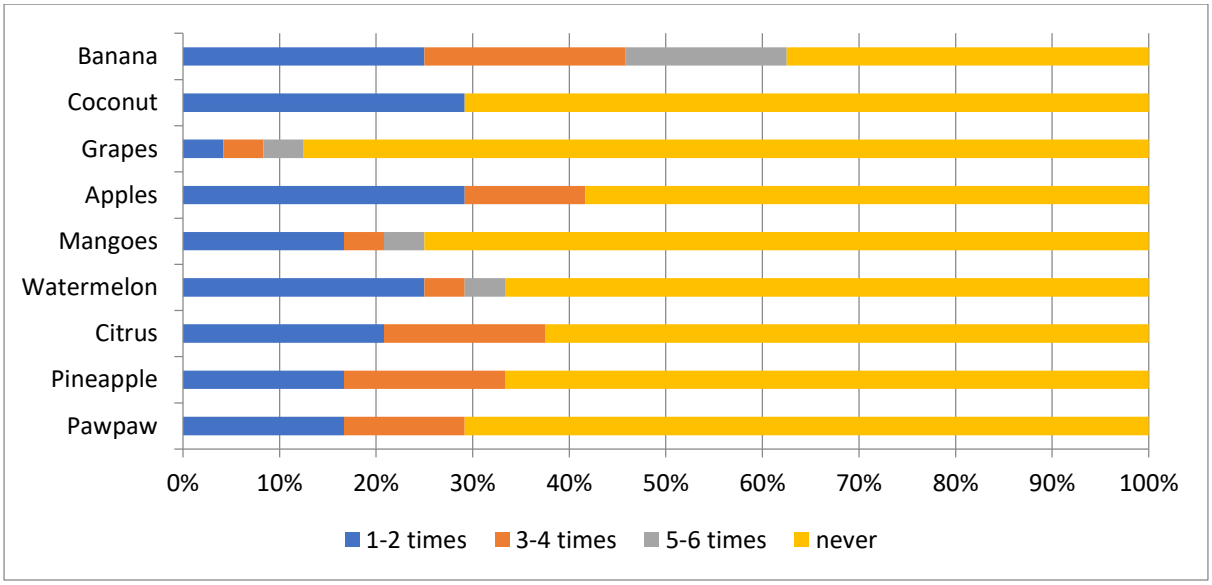


Figure 10: Daily fruit consumption in December over a one-week period

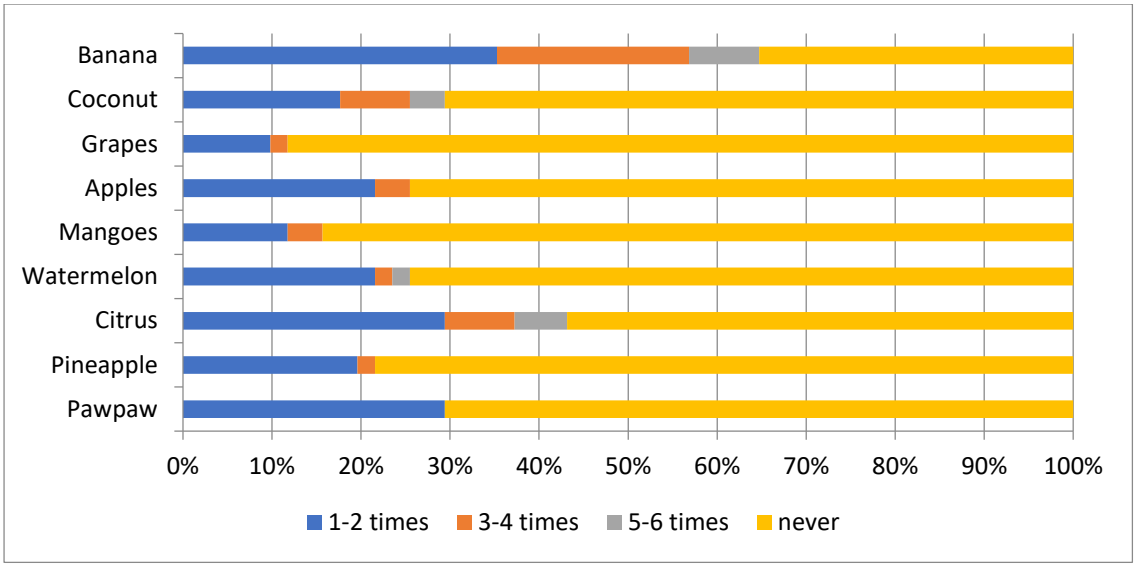


Figure 11: Daily fruit consumption in January over a one-week period

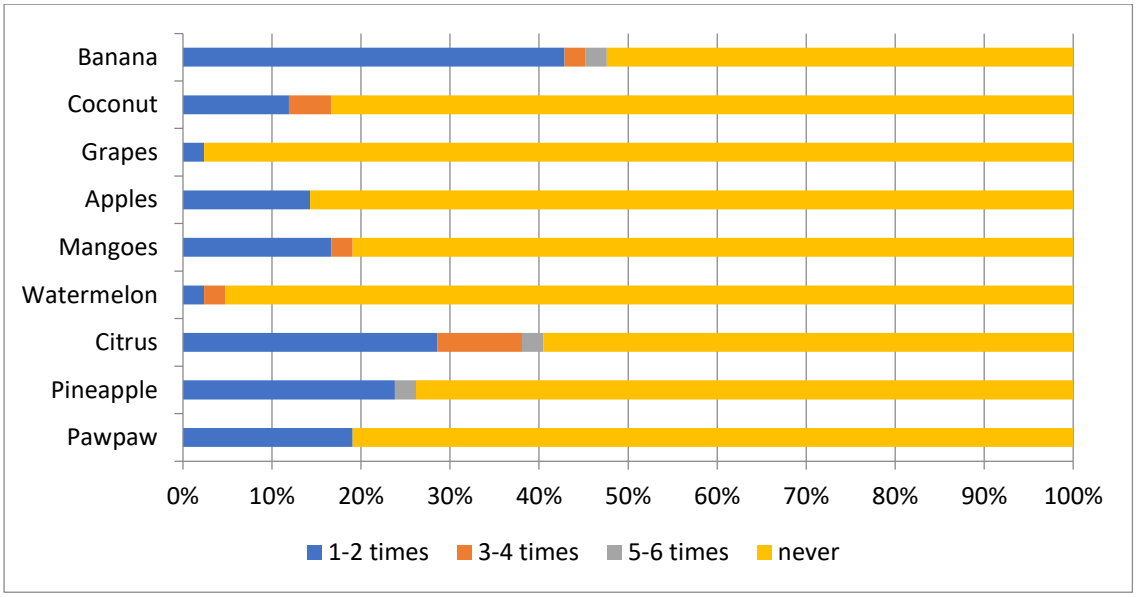


Figure 12: Daily fruit consumption in February over a one-week period

4.5.2 Pattern of vegetables, stews and soups consumption

Except for cucumber, more than half of the participants consumed cabbage/lettuce, green pepper and carrots weekly as presented in figure 13. However, most of the participants did not consume these vegetables on a daily basis rather once/twice weekly. Figure 14 shows that except for tomato stew, less than half of the participants had consumed the other stew types like okro, beans and kontomire stew. About 40% of the participants consumed tomato stew

about once/twice weekly. Consumption of kontomire with agushie recorded the lowest frequency.

More than half of the participants had not consumed all the soups assessed except for light soup as shown in figure 15. For all the soups, the highest rate of consumption was once/twice weekly although a few people consumed it almost every day.

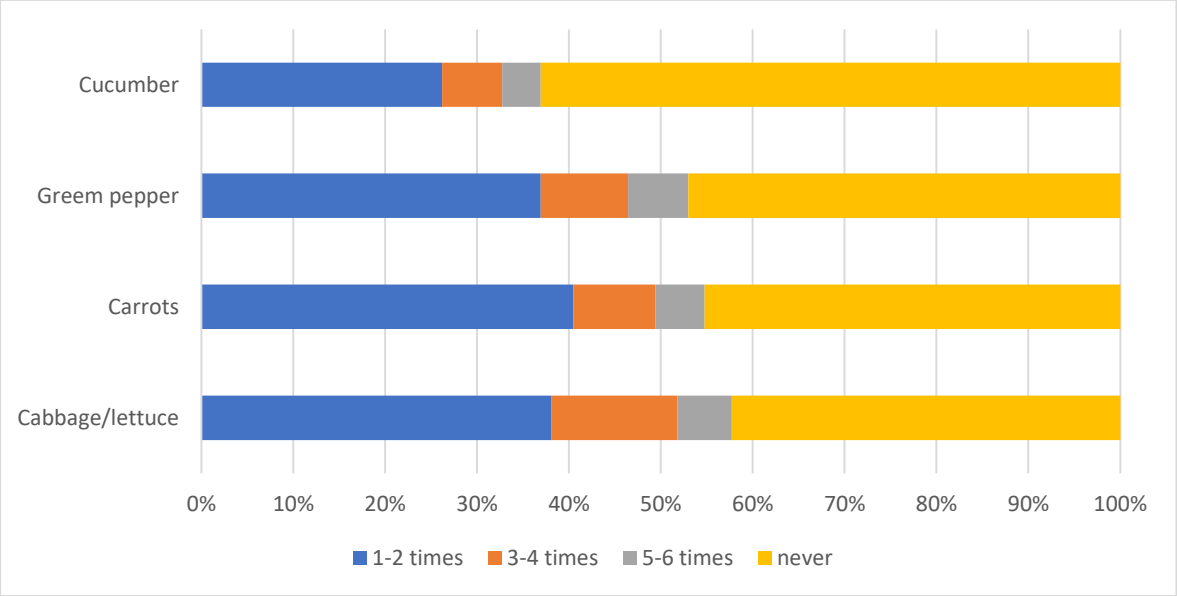


Figure 13: Daily vegetables consumption over a one-week period

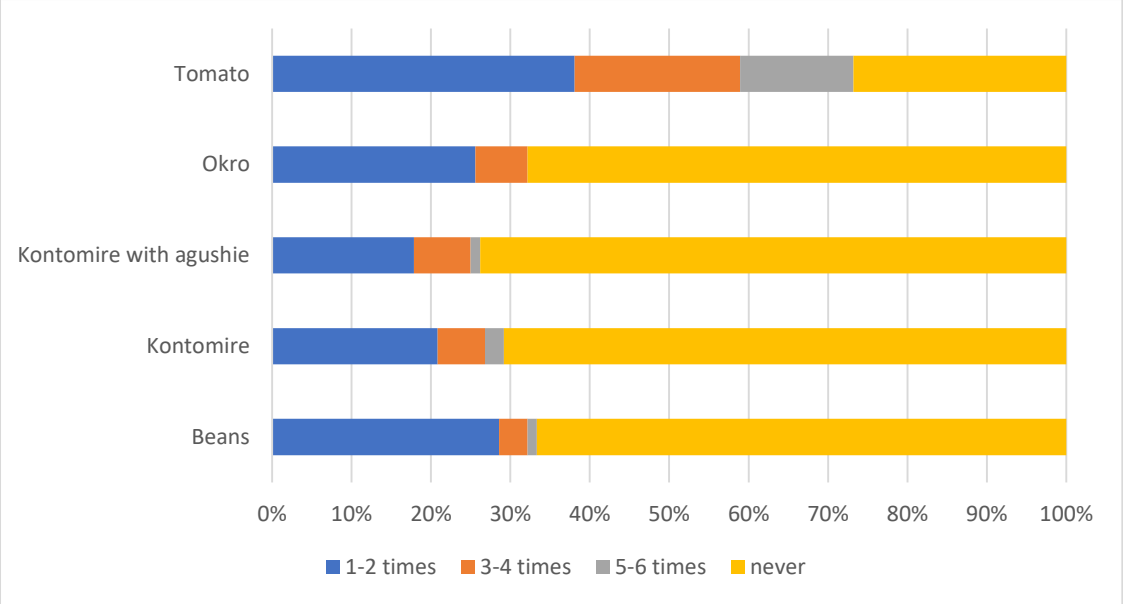


Figure 14: Daily stew consumption over a one-week period

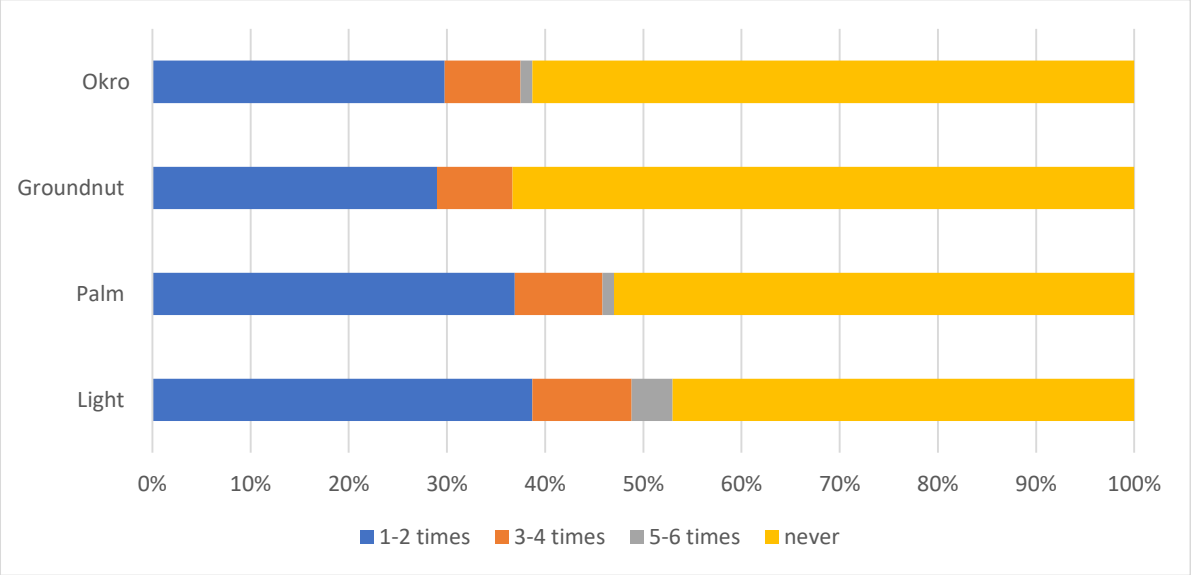


Figure 15: Daily soups consumption over a one-week period

4.5.3 Pattern of consumption of protein-rich foods

The most consumed fish was salmon (>50%), herring was the least consumed fish (<30%). A little over half of the participants had consumed meats like goat and cow meat and almost 60% had consumed chicken as illustrated in figure 16. Eggs were consumed by most of the participants at least once/twice in a week. Less than 20% of the participants had consumed

bush meat, offal, crabs and shrimps. Less than half of the participants consumed milk and milk products as shown in figure 17. Evaporated milk consumption was most frequent amongst the participants compared to powdered milk. Cheese was the least consumed milk product.

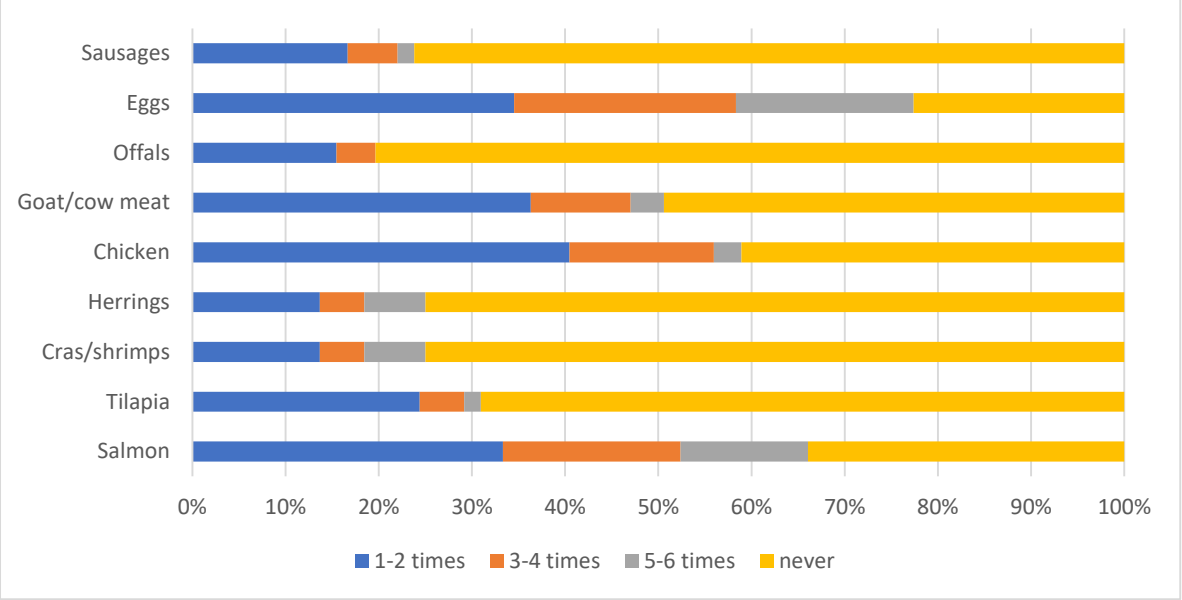


Figure 16: Daily meat, poultry, fish and egg consumption over a one-week period

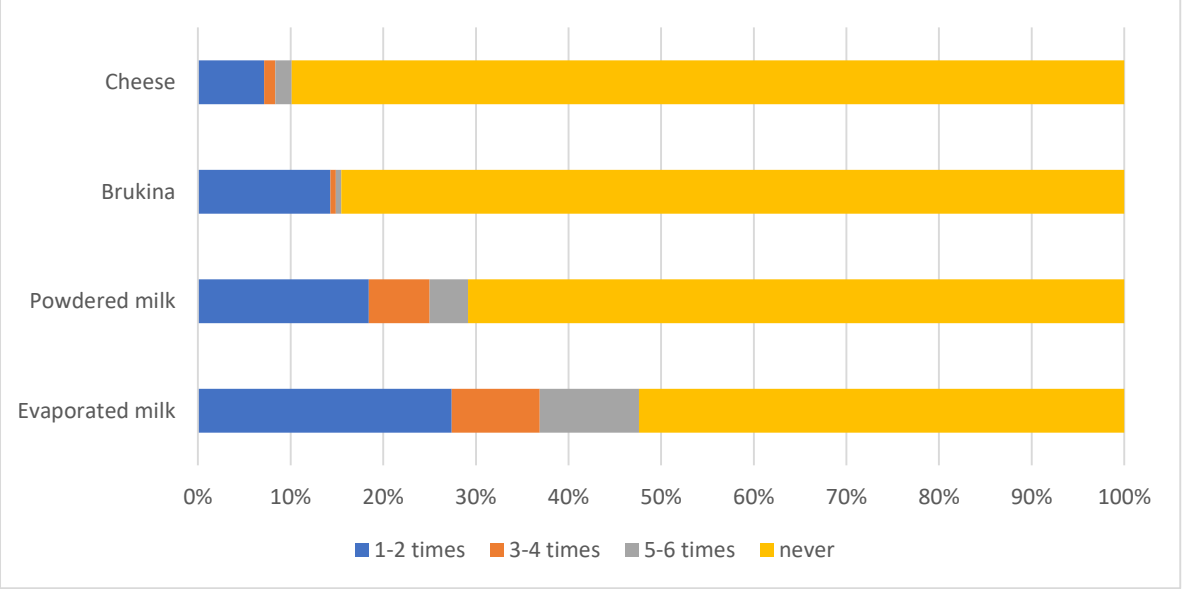


Figure 17: Daily milk and milk products consumption over a one-week period

4.5.4 Pattern of consumption of foods high in fats

Most of the participants had not consumed spreads during the period of data collection. Although margarine was most frequently consumed, just about a quarter of the participants had consumed it as shown in figure 18. The frequency of consumption of deep-fried foods was also low. Even though fried about 40% of participants consumed fried plantain, it was mostly consumed once/twice weekly. As illustrated in figure 19, more than 60% of participants had consumed white bread (cake bread, tea bread, sugar bread and butter bread) and most (about 40%) of them consumed it once/twice in the week. Biscuit was the second most consumed pastry with about half of the participants reporting that they had consumed it during the week of assessment. The most reported rate of consumption of pastries was once/twice in a week. Doughnuts were the least consumed pastry.

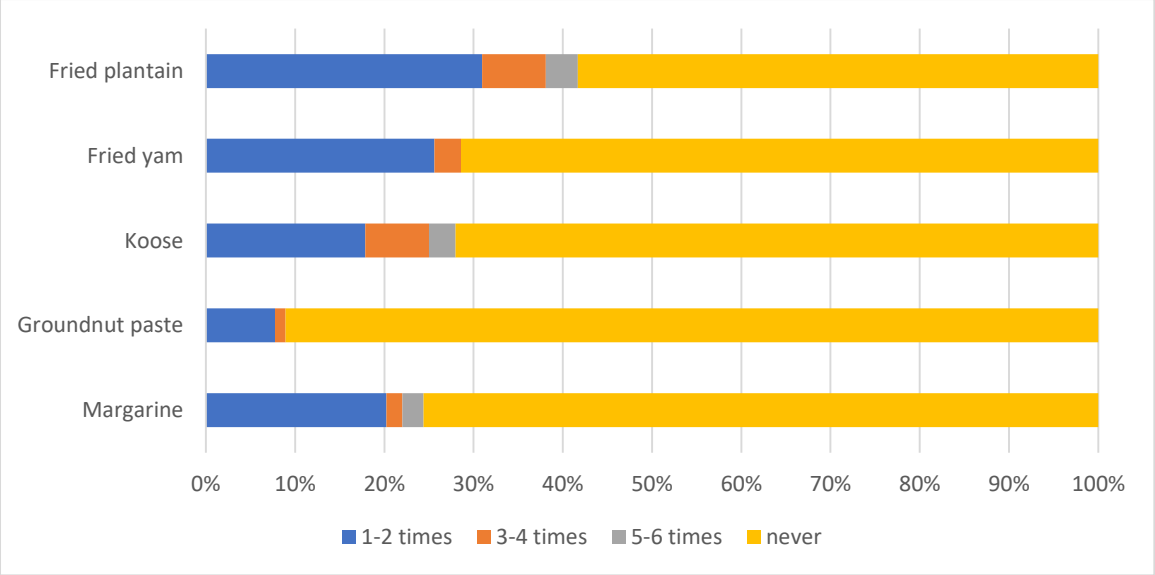


Figure 18: Daily deep-fried foods and spreads consumption over a one-week period

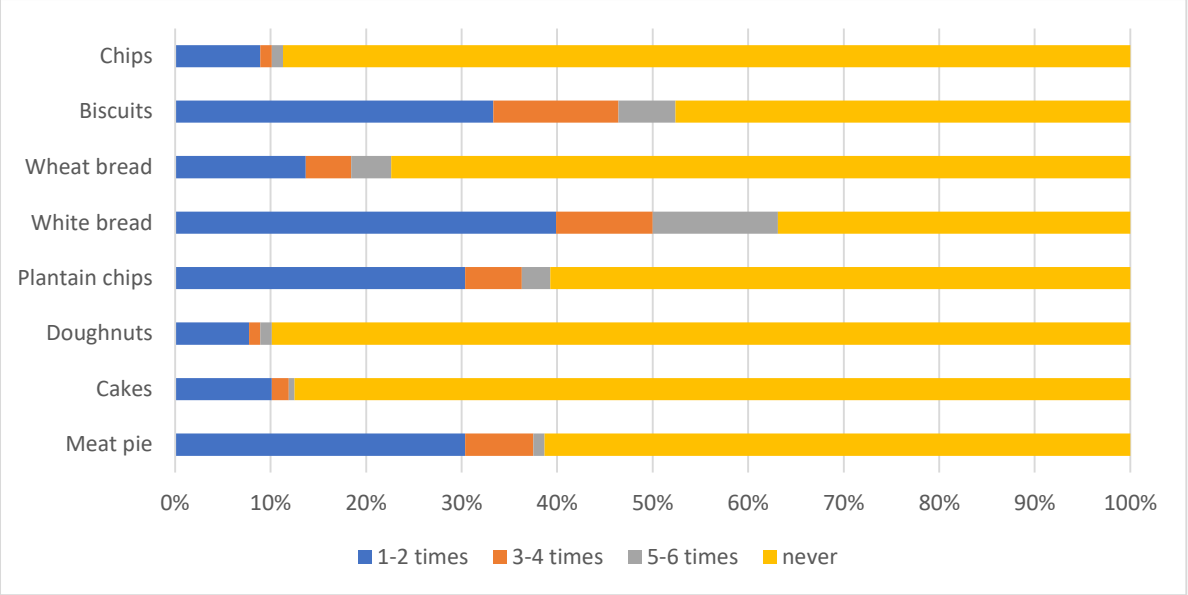


Figure 19: Daily pastries consumption over a one-week period

4.5.5 Pattern of consumption of carbohydrate-rich foods

As shown in figure 20, for the common breakfast meal options (beverages and porridges) assessed, more than half of the participants had not consumed them. However, the most consumed beverage and porridge was cocoa and millet porridge respectively where majority of participants had consumed these foods at least once/twice within a week. Rice, wheat and oats porridges were the least (<20%) consumed among participants.

Except for banku, plain rice and jollof, more than half of the participants had not consumed the major staples in the country. Gari was the least consumed starchy food followed by plantain. Although noodles and spaghetti are not traditional staples in Ghana, its consumption is almost at par with traditional staples like yam, fufu and kenkey. Almost 90% of the participants had eaten plain rice during the week of assessment with most of them eating it once/twice weekly followed by those who ate rice almost every day. Figure 22 shows that consumption of sugar-sweetened (soda) was very common among participants (60%) with a

consumption rate mostly once/twice weekly. This was followed by fruit juice consumption. Alcohol consumption beverages represented less than 10% of the participants.

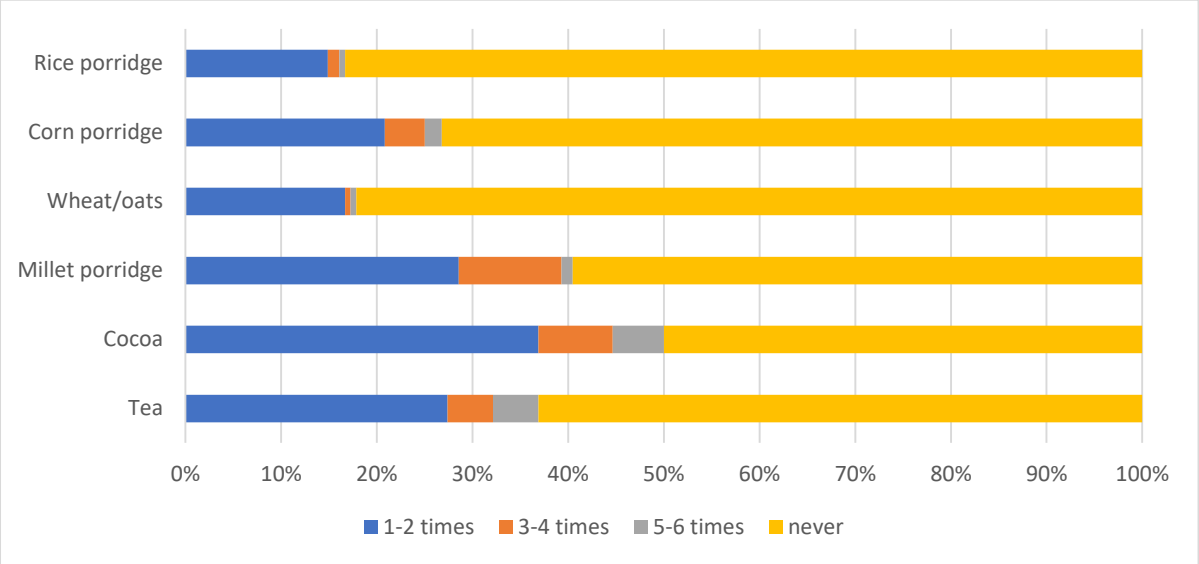


Figure 20: Daily beverages and porridges consumption over a one-week period

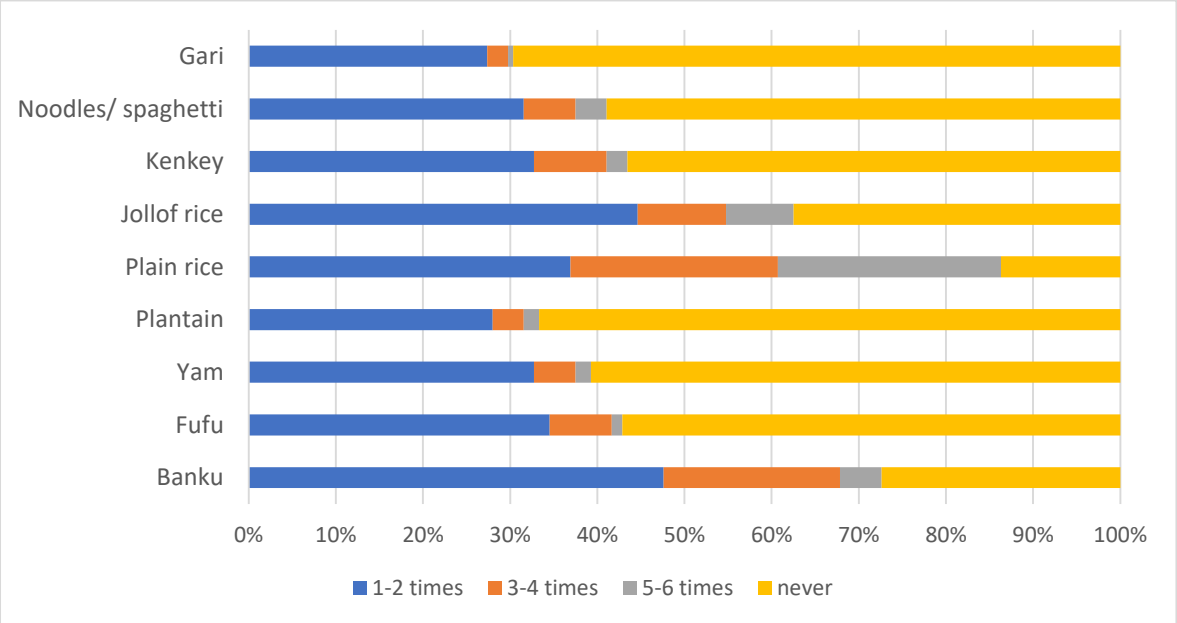


Figure 21: Daily starchy foods consumption over a one-week period

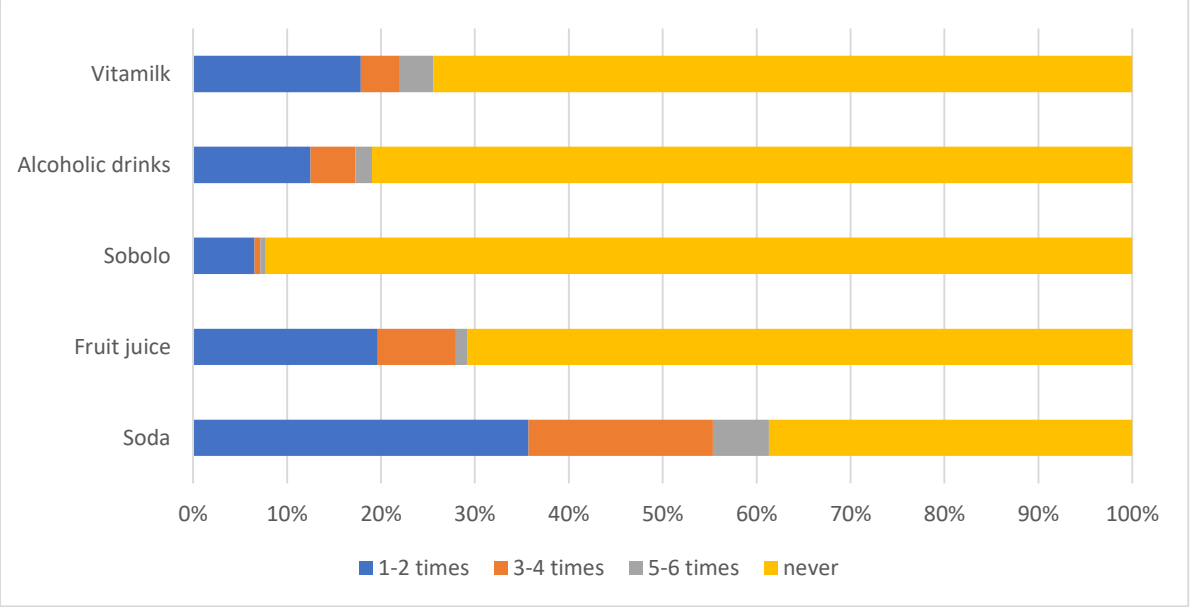


Figure 22: Daily soft drinks and alcoholic beverages consumption over a one-week period

4.6 Association between Nutrition Knowledge and Nutritional Status

A chi-squared test was conducted to determine the association between nutritional knowledge and nutritional status and the results presented in Table 14. A $p>0.05$ was obtained; therefore, we do not reject the null hypothesis.

Table 14: Association between nutritional status and nutritional knowledge

Overall nutritional status	Nutritional knowledge ranking			
	Low	**High**	**Total**	**p-value**
Malnourished	9	33	42	0.196
Well-nourished	17	112	129	
Total	26	145	171	

4.7 Association between Inflammatory Biomarker and Nutritional Status

A linear regression showed that weight was a predictor for IL-6 levels such that a unit change in weight will lead to a 0.66 decrease in IL-6 levels (F (1, 166) = 4.93 ; p = 0.03). After adjusting for height, the results are presented in the table below (F (2, 165) = 2.97; R^2- 0.03 p = 0.05).

Table 15: Strength of association between IL-6 levels and weight

Variable	Coefficient (confidence interval)	p-value
Weight	-0.589 (-1.194 – 0.016)	0.06
Height	-0.340 (-1.007 – 0.327)	0.32

CHAPTER FIVE

5.0 DISCUSSION

5.1 Socio-demographic Characteristics of Participants

Historically, people with SCD were known to have a shorter life expectancy compared to non-SCD individuals. However, due to improvements in care, the life expectancy of these individuals has increased. A recent report by Asare stated that most (86%) of the patients at the Ghana Institute of Clinical Genetics were between the ages of 13-39 years (Asare et al., 2018). Similarly, this study also had majority (91.2%) of participants within the age range of 16-39 years. More females than males (1.6:1) participated in this study which is also in agreement to the report by Asare et al. (2018). Generally, more females attended the clinic compared to males in a ratio of 1.6:1 and this was attributed to the better health seeking behaviour of females than males (Asare et al., 2018). In agreement to the current study, Nyante, Oppong and Bonney (2019) also found the prevalence of females (60%) with SCD to be higher than males (40%). Another explanation given by Asare et al. (2018) was that Ghanaian women had a higher life expectancy compared to males based on a WHO report.

Individuals with sickle cell anaemia represented a larger number of the participants. Aside sickle cell anaemia, the Hb SC subtype also represented 30% of the participants. A preliminary study conducted at a hospital in the Volta region of Ghana also reported a higher percentage of Hb SS to Hb SC patients in a ratio of 1:0.4. Sickle cell anaemia (Hb SS) and Hb SC were the reported genotypes probably because Hb S and Hb C are the commonest variants found in Africans with the Hb C variant being almost exclusive to West Africa (Makani et al., 2013). Even though individuals with SCD suffer from periodic crisis and may be absent during school hours, this did not deter them from pursuing formal education as this study shows that almost all participants had some level of formal education with most of

them attaining at least a Secondary or Vocational or Technical certificate. Opposing to this study, Nyante et al. (2019) reported that most (47.2%) of their participants had attained tertiary education. Most Ghanaians in secondary or vocational school are in their teenage years (13-18years) and this may explain why this study recorded a higher percentage for secondary or vocational level whereas Nyante and colleagues reported a higher percentage for tertiary education because their study did not include persons below 18 years.

5.2 Source of Nutrition Information

Due to the frequent reports on the nutritional challenges faced by individuals with SCD, there have been proposals to involve experts like dietitians and nutritionists in the standard care protocol (Hyacinth et al., 2013). Findings from this study suggest that this proposal has not fully been implemented since nutrition experts are the least consulted for nutrition information. This study revealed that the services of the nutritionists are not frequently sort after although there is a consulting nutritionist at the institute every Tuesday and Friday. A survey by a group of scientists showed many young adults could not afford the fee (GH₵ 10 - GH₵ 30) needed to consult a dietitian because of their limited income (Quaidoo et al., 2018). Given the mean age of this study's participants, it is likely that most of them faced similar challenges and hence could not schedule an appointment with a dietitian or nutritionist.

A study among healthy young adults in Accra reported that health professionals were the least consulted for nutrition information whereas the internet, radio, television, family and friends were the most consulted (Quaidoo et al., 2018). This trend was attributed to the preference for self-care practices by people in developing countries (Obasola & Agunbiade, 2016). A similar trend for self-care practices can also be seen in this study as family and friends were the most consulted source of nutrition information whereas radio, television and

internet were most consulted compared to dietitians. For individuals with SCD the quest for self-care may have negative consequences on them due to probability of misinformation.

Although Quaidoo et al., (2018), documented health professionals as the least consulted for nutrition information, this study reports that doctors are the most consulted health professional for nutrition counsel. The differences in the findings may be due to study participants. Whilst the study by Quaidoo et al. (2018) was among healthy individuals, this current study was among individuals with a chronic condition who visit the clinic and interact with doctors and nurses very often. It is therefore not surprising that doctors and nurses were the most consulted health professionals for nutrition information because they are the primary providers of health care for these individuals. People with SCD need a comprehensive nutrition management due to the nutritional challenges they face and doctors and nurses are not equipped to provide such care. Hence, doctors and nurses should refer cases that require nutrition attention to the appropriate professionals since they are well equipped to provide information and other strategies that can improve nutritional status.

5.3 Food Consumption Patterns of Individuals with Sickle Cell Disease

The food consumption patterns of the study participants were similar to that of healthy individuals in Ghana. For fruits, banana was the most consumed followed by citrus this is contrary to findings by Asante et al. (2015) who reported a citrus as the most frequently consumed fruit followed by banana. The monthly consumption also showed similar trends, banana was the most consumed fruit in each month followed by oranges except for October and December where pawpaw and apple consumption recorded the second highest frequencies respectively. Based on the article by Banson, Sun and Banson (2016), banana is available all year round, this may account for it being the most consumed fruit in all the

months. The period of data collection may explain why the frequency of orange consumption was the second highest in January and February because oranges are available from January-March and June-September (Banson et al., 2016). Although apples and grapes are exotic fruits, consumption of apples was more frequent than grapes and this agrees with the study conducted by Asante and colleagues (Asante et al., 2015).

In accordance with the study by Asante and colleagues this study also found that consumption of fresh vegetables was low. This is because Ghanaians prefer consuming their vegetables in the form of stews and soups. Fruits and vegetables are the main sources of vitamins and minerals in our diet, assessment of nutrition knowledge showed that participants did not know the importance of micronutrients to the body. This may also explain the low frequency of consumption of these foods.

For the starches, rice consumption was the most frequent probably due to the busy urban lifestyle and this is in accordance to a study conducted in the Ashanti region which also reported frequent consumption of rice compared to other staples like “banku” and “fufu”(Frimpong, 2013). Asante et al. (2015) also documented that preferred breakfast options for Ghanaians in Accra were corn and millet porridge but in this study cocoa beverages were the most preferred choice of breakfast followed by millet porridge. Eggs and salmon were the most frequently consumed protein-rich food compared to chicken and meat. Tanabe et al. (2010) mentioned that individuals with SCD do not consume fatty foods as a coping strategy to prevent crises. Since meats are often perceived to fatty, it is likely that participants preferred consuming salmon which is seen to be less fatty. Although participants knew sugar-sweetened beverages and pastries were not healthy snacks, they still consumed them in relatively high frequencies.

Fruits and vegetables are very important in the diets of individuals. This is because they contain vitamins and minerals which are very essential to human living. For example, vitamin A, C and E are antioxidant vitamins and this property has been shown to help reduce the frequency of crisis and disease severity in people with SCD (Hyacinth et al., 2010). Given the numerous nutrient deficiencies that people with SCD present with, it is necessary that they consume adequate amounts to help reduce incidence of these nutrient deficiencies. From the food consumption data gathered, it is seen that the consumption of these group of foods is very low among participants. It is therefore necessary, that in the nutritional management of people with SCD, dietitians and nutritionists emphasize on the need for these persons to consume adequate amounts of fresh fruits and vegetables since these foods contains the minerals and vitamins that most SCD patients that will help improve the numerous complications that they face.

5.4 Nutritional Status of Participants

The average body fat level of both males and females were within the normal range with the mean levels of females and males being 30.6±7.04 and 11.9±5.82 respectively. Two studies have also reported the mean body fat levels of males with SCD to be 8.9% and 10.8% (Eke, Chukwu, Ikefuna, Ezenwosu, & Emodi, 2015 & VanderJagt, Harmatz, Scott-Emuakpor, Vichinsky, & Glew, 2002). However, these studies report significantly lower levels of body fat in females with body fat levels of 25.7 % and 14.7% compared to the 30.6% reported in this study. For muscle mass, Eke et al. (2015) also reported higher levels (33.1%) among females compared to this study (28.1%) and vice versa for the males. A correlation analysis showed that body fat increased with age and muscle decreased with age this could explain the differences observed between the two studies since Eke and colleagues assessed the body composition of adolescent whereas this study was conducted among adolescents and adults.

In healthy populations, there is muscle loss with increasing age due to many factors like reduced activity, lower concentration of growth hormones and insulin-like growth factor (Seene & Kaasik, 2012). This age-related sarcopenia coupled with the hypermetabolic nature of the disease may contribute to the reduced %MM reported. In contrast, although VanderJagt et al. (2002) also conducted their study among adolescents, the percentage muscle mass of both genders (males - 91.1% and females – 72.1%) were significantly higher than the present study. It is worth noting that the study by VanderJagt and colleagues was conducted in the United States which is a developed country unlike Ghana and Nigeria which are still developing. Hence, individuals with SCD in the developed countries are more likely to receive better health care compared to Ghana. The body composition of females is quite worrying because a high percentage body fat has been shown to increase cardiovascular disease risk (Valentino et al., 2015). Almost 40% of the females had high and very high percentage body fat levels and this may put them at risk of cardiovascular diseases which will put another metabolic stress on the body in addition to the SCD.

The overall nutritional status based on BMI show that majority of the respondents were well-nourished with a few malnourished cases. Historically, individuals with SCD were mostly reported to be underweight; however, due to improvements in the standard of care, there is a shift from underweight to normal as reported in this study. This study also reports cases overweight and obesity almost close to prevalence underweight. Cox et al. (2011), did not report any case of overweight or obesity in their study among Hb SS individuals in Tanzania. Conversely, Akodu, Diaku-Akinwumi and Njokanma (2012) documented a 2.5% prevalence of obesity in their study conducted in Nigeria. A lower prevalence rate was reported compared to this study because Akodu et al. (2012) conducted their study among individuals with SCD aged 2-15 years old whilst the current study was conducted among individuals

aged 16 years and above. Unlike the study carried out in Nigeria, studies in Ghana conducted among children have not reported any case of overweight or obesity (Boadu, 2016; Bonsu, 2017; Osei-Yeboah et al., 2011). One of such studies, explained that dietary energy and nutrient intake of these Ghanaian children were sub-optimal (Boadu, 2016).

The prevalence of overweight and obesity among individuals with SCD in developed countries is higher than what has been reported in developing countries. An overweight and obesity prevalence rate of 44% was reported among African-Americans with SCD in the United States and the BMI was higher in Hb SC individuals compared to Hb SS (Kaufman et al., 2018). An equally higher prevalence (54%) of overweight and obesity was documented in another study conducted in the United States among SCD patients aged 16 years and above (Farooqui et al., 2014). Farooqui et al. (2014), states that these high rates are reflections of the rapid increase of obesity in the general population of developed countries. These findings on the prevalence of overweight and obesity suggests that whilst health professionals focus on ensuring almost all underweight patients attain a normal weight, care should be taken to prevent patients from gaining excessive weight.

5.5 Serum Vitamin E and Interleukin-6 levels of Participants

Low circulating levels of antioxidant vitamins has been reported in individuals with SCD. This study also reported lower levels of vitamin E as compared to the reference range. This shows participants are at risk of vitamin E deficiency since the reference range is 5.5-17µg/ml (Wallach, 1996). A similar study by Hasanato records mean vitamin E levels as low as 2 mg/L (Hasanato, 2006). This is worrying because participants are given multivitamins to supplement their dietary intake and help prevent these nutrient deficiencies. The low vitamin E level is also suggestive that even in steady state; there is an increased demand for nutrients

to compensate the hypermetabolic activities like subclinical inflammation taking place. Inflammation is likely to be more pronounced in persons with lower levels of vitamin E because they will not benefit from the ability of this vitamin to prevent oxidative stress and reduce rate of haemolysis.

Evidence of the hypermetabolism is the elevated IL-6 levels recorded even in steady state. Keikhaei et al. (2013) reported elevated IL-6 among SCD subjects compared to controls with a mean 12.1±1.98 pg/ml. Similarly, Pierrot-Gallo et al. (2015) also reported higher IL-6 levels among SCD subjects with a mean level of 15.9±9.1 pg/ml. These are not extremely different from the levels recorded in this study. Some studies have documented extremely high IL-6 levels in steady state SCD individuals. One of such studies reports mean levels of 71±40 pg/ml (Hibbert et al., 2005). A recent report by Elzubeir and colleagues also reported extremely high circulating IL-6 levels in steady state SCD individuals (Elzubeir et al., 2017). These findings suggest that even in study state, inflammation is ongoing in this group of people.

This chronic inflammatory response may be responsible for the low body weight recorded in some participants. An independent t-test showed that individuals with sickle cell anaemia had a significantly lower body weight and elevated IL-6 compared to those with Hb SC. This is because a negative correlation existed between IL-6 levels and body weight ($r = -0.1698$, $p = 0.03$). Interleukin-6 is a pro-inflammatory cytokine that has been implicated in cancer cachexia for causing weight loss and reducing appetite (Carson & Baltgalvis, 2010). Thus chronic elevated levels will lead to a reduction in body weight. A linear regression demonstrated that for every unit increase in weight, IL-6 levels will decrease by 0.66. After adjusting for height, weight was not a significant predictor of IL-6 levels.

5.6 Association between nutrition knowledge and nutritional status

Contrary to reports that nutrition knowledge was associated with nutritional status such that a high nutritional knowledge will lead to a good nutritional status and vice versa, this study reported no association between these two variables. This buttresses the statement by Hakli et al. (2016) that nutrition knowledge alone may not be enough to improve nutritional status if not translated into dietary behaviour and practices. In this study, although almost all the participants knew that sugar-sweetened beverages and pastries were not healthy snacks, the consumption rate was high. This lack of translation of knowledge to behaviour may explain the result obtained.

Unlike healthy individuals, persons with SCD are in a hypermetabolic state and this is reinforced by the elevated IL-6 levels recorded even in steady state. This increases the energy and nutrient requirements of affected persons. Therefore, having a high nutritional knowledge will not automatically translate into a good nutritional status if adequate energy and nutrients are not consumed to support the hypermetabolism and growth. Hence, nutrition experts should not only focus on providing nutrition information during counselling sessions but also have a means of assessing nutrition behaviour and practices. There is also a need for frequent monitoring to ensure that the nutrition counsel provided is being adhered. For individuals who may be struggling with underweight especially persons with Hb SS, laboratory tests can be requested to ascertain the intensity of inflammation and the appropriate dietary counsel that can meet metabolic demands and support growth given.

5.7 Limitations

Some limitations of this study include:

1. The dietary assessment tool used in this study may have some levels of under- or over-reporting.
2. It is also possible that eating habits were modified to impress the researcher and these may affect the results.
3. The results of the study cannot be generalized because of the sampling technique used.

CHAPTER SIX

6.0 CONCLUSION AND RECOMMENDATIONS

6.1 Conclusion

Although, almost all participants (90%) had not received counselling from a dietitian or nutritionist, majority had a high nutritional knowledge. However, nutritional knowledge did not influence nutritional status. Even though majority of the participants were well nourished (had a normal BMI), this study also found that the prevalence of overweight and obesity is almost equal to that of underweight. Haemoglobin SS participants were more likely to have a lower BMI compared to other SCD types.

Food consumption patterns of participants was similar to the healthy Ghanaian population with low frequency of consumption of fruits and vegetables was low and high consumption of sugar-sweetened beverages and pastries.

Serum interleukin 6 levels were significantly elevated in individuals with sickle cell anaemia compared to those with Hb SC. Also, IL-6 levels were slightly higher in participants with a normal nutritional status compared to those who were malnourished. Serum vitamin E levels of participants with a normal nutritional status were lower than those who were malnourished. However, the difference between the groups was not significant.

6.2 Recommendations

1. Since nutritional knowledge did not influence nutritional status, there is a need for dietitians to assess dietary practices and habits of individuals during counselling sessions.
2. There is a need to educate participants on the micronutrients and the importance of these nutrients to the body.

3. Patients at risk of being overweight or obese should be referred to dietitians for nutrition therapy to prevent these persons from developing non-communicable diseases like diabetes and dyslipidemia in addition to their present condition.
4. Future research should engage in interventional studies that will improve the dietary habits and intake of participants.

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APPENDICES

APPENDIX I

UNIVERSITY OF GHANA

SCHOOL OF BIOMEDICAL AND ALLIED HEALTH SCIENCES

INFORMED CONSENT

My name is **Faustina Ofosua Mintah** and I wish to conduct research on the topic **"Assessing Nutrition Knowledge and Nutritional Status of Sickle Cell Disease Patients"**. I am a Dietetic student of the School of Biomedical and Allied Health Sciences, College of Health Sciences, University of Ghana.

General Information about Research: The aim of the study is to assess nutrition knowledge and the nutritional status of sickle cell disease (SCD) patients. This study will help us know your nutritional status. It will also help us understand the relationship between your nutrition knowledge and nutritional status.

You will be required to fill a questionnaire that will provide information on your nutrition knowledge. You will also be required to give information on what you eat and how much you eat. Your body height and weight will be measured to determine your nutritional status. A small amount (about half of a teaspoon) of your blood that was taken for SickleGenAfrica will be taken for some blood tests. This will help us know if you have some nutritional problems or not. All these processes will take about 45 minutes.

Possible risks and Discomforts: There is no risk involved in taking part in this study.

Possible benefits: There is no direct benefit to you. However, the results will be used to plan nutrition and health intervention programmes to improve your care and SCD patients in general.

Confidentiality: The information you provide will be kept confidential by the researcher. You will not be identified by your name. You will be given codes as a form of identification.

Voluntary Participation: Participating in this study is voluntary without any costs. You are free to withdraw from the study at any point in time and your care at the clinic will not be affected.

Contacts for Additional Information: The researcher will however be available and willing to answer any further questions about the research, now or during the course of the project. You can contact me through this number should you have any questions or concerns, 0248568483.

VOLUNTARY AGREEMENT

I agree that the research project named above has been read and explained to me. I have also been given the opportunity to ask questions about the research and I have been answered to my satisfaction. I therefore agree to take part in this study. I understand that I am agreeing with my signature/thumbprint on this form to take part in this research project.

Name of participant / parent ……..

Signature /mark of participant / parent …….

Tel No: ……

Date ………

If volunteers cannot read the form themselves, a witness must sign here:

I was present while the benefits, risks and procedures were read to the patient. All questions were answered and the patient has agreed to take part in the research.

Name of witness ……..

Signature of witness ……... **Date** ……

I certify that the nature and purpose, the potential benefits, and possible risks associated with participating in this research have been explained to the above individual.

Name of Person Who Obtained Consent ….....

Signature of Person Who Obtained Consent ….....

Date ……..

APPENDIX II

CHILD ASSENT FORM

INFORMATION SHEET

My name is **Faustina Ofosua Mintah** and I wish to conduct a research on the topic **"Assessing Nutrition Knowledge and Nutritional Status of Sickle Cell Disease Patients"**. I am a Dietetic student of the School of Biomedical and Allied Health Sciences, College of Health Sciences, University of Ghana.

I am asking you to take part in this study because I am trying to learn more about the relationship between your nutrition knowledge and nutritional status. This will take about 45 minutes.

General information: If you agree to be part of this study, your height and weight will be measured. We will also ask you questions about what you eat and how much you eat. A little amount of the blood that was taken for SickleGenAfrica will be collected for some blood tests. Your participation in this study will give us important information to help plan a programme to improve your care and health.

Possible risk and discomforts: You will not experience any pain for taking part in this study.

Voluntary Participation and Right to Leave the Research: You can stop participating in this study at any point and nobody will get angry with you.

Confidentiality: The information you provide will be kept confidential. Your name will not be written against any response you give.

Contact for Additional Information: You can also call me on 0248568483 or contact me anytime you see me.

Please talk to your parents before you decide whether or not to be part of this study. I will also ask your parents for permission before you are chosen for the study. Even if your parents say "yes" you can still decide not to participate in the study.

VOLUNTARY AGREEMENT

By marking or thumb printing below, it means that you understand and know the issues concerning this study. If you do not want to be part of the study, please do not mark or thumbprint. A copy of this form will be given to you and your parents after you have marked/ thumb printed it.

This assent form which describes the risks, benefits and procedures of the research project named above has been explained to my satisfaction. I have been given the opportunity to have any questions about the research answered to my satisfaction. I agree to take part in this study.

NAME OF CHILD…….

DATE……

MARK/THUMBPRINT…….....

NAME OF RESEARCHER…….

DATE…….. SIGNATURE ……

TELEPHONE NUMBER ……

If child cannot read the form themselves, a witness must sign here:

I was present while the benefits, risks and procedures were read to the child. All questions were answered and the child has agreed to take part in the research.

Name of witness ……..

Signature of witness ……... **Date** ……

APPENDIX III

QUESTIONNAIRE

Participant's code ……..

SOCIO-DEMOGRAPHIC DATA

1. Age of participant ……
2. Gender: 0- Male ☐ 1- Female ☐
3. Body measurements

Height (cm)	
Weight (kg)	
BMI (kg/m^2)	
BF (%)	
MM (%)	
VF	
RM	

4. Sickle cell genotype: 0- SS ☐ 1- SC ☐ 2- SF ☐ Other……
5. Place of residence.....
6. Ethnicity: ……
7. Religion

   0- Christian ☐ 1- Muslim ☐ 2- Traditionalist ☐

   Other (specify) ……
8. Educational background

   0- No formal education ☐ 1- Primary ☐ 2- JSS ☐

   3- SSS ☐ 4- Tertiary ☐ 5- Vocational/Technical ☐
9. Occupation: 0- Public servant ☐ 1- Civil Servant ☐

   2- Trader ☐ 3- Seamstress/ Hairdresser ☐ 4- Unemployed ☐

   5 – Student ☐ 6 – Other …….
10. How much do you earn monthly?

    0-GH¢ 100 - GH¢ 299 ☐ 1- GH¢300- GH¢ 499 ☐ 2- GH¢500 - GH¢ 699 ☐

    3- GH¢ 700 - GH¢ 899 ☐ 4- ≥GH¢ 900 ☐ 5- No income ☐

11. What is your marital status?

0- Single ☐ 1- Married ☐ 2- Divorced/Widowed ☐

12. How long have you known your sickle cell status? ……year

13. Do you have any disease aside sickle cell? Yes ☐ No ☐

14. If yes, name them…….

FEEDING BEHAVIOUR AND GENERAL NUTRITION INFORMATION

1. Do you readily accept meals offered to you?

   0- Yes ☐ 1- No ☐

2. How many times do you eat in a day?

   0- Once ☐ 1- Twice ☐ 2- Thrice ☐ 3- > 3x ☐

3. Do you take dietary supplements? Yes ☐ No ☐
4. If yes, name them

   ……

   ……

5. What is your source of nutrition information? Tick as many that apply

   Doctors ☐ Nurses ☐ Dietitian / Nutritionist ☐

   Family and friends ☐ Radio/ Television programmes ☐

   Other ……

NUTRITION KNOWLEDGE QUESTIONNAIRE (Adapted from Tandoh, 2015)

1. Cassava, Yam and Gari are rich sources of:
   1- Protein 2- Carbohydrates 3- Fats 4- Iron 5- Don't know

2. Chicken, Eggs and Fish are rich sources of:
   1- Protein 2- Carbohydrates 3- Fats 4- Iron 5- Don't know

3. Which of the following is a rich source of fat?
   1- Orange 2- Margarine 3- Carrot 4- Rice 5- Don't know

4. Milk and milk products are rich in
   1- Sodium 2- Calcium 3- Iron 4- Zinc 5- Don't know

5. Which of the following is a rich source of iron?
   1- Liver 2- Tomatoes 3- Carrot 4- Rice 5- Don't know

6. Which of the following is a rich source of dietary fibre/roughage?

1- Beans 2- white rice 3- Groundnut 4- Beef 5- Don't know

7. Inadequate intake of …… will make you look very thin.
   1-Protein 2- Carbohydrates 3- Vitamin A 4- Vitamin K 5- Don't know

8. Rickets/ bow legs are caused by lack of
   1- Vitamin K 2-Vitamin D 3-Vitamin E 4-Vitamin A 5- Don't know

9. Anaemia is caused by lack of:
   1- Iodine 2- Iron 3- Calcium 4- Sodium 5-Don't know

10. Bleeding of gums is caused by inadequate intake of:
    1-Vitamin C 2- Vitamin K 3- Vitamin E 4- Vitamin A 5- Don't know

11. Which of these foods would give you blood?
    1- Kontomire 2- Oranges 3- Banku 4- Fish 5- Don't know

12. Do you think fruits and vegetables must be eaten frequently?
    1- Yes 2- No 3- Don't know

13. Which of the following snack options is healthy?

    1- Coca-cola 2- Ice-cream 3- Banana and groundnut 4- Biscuit

    5- Don't know

14. Which of these foods is balanced?

    1- Corn dough porridge with bread 2- Boiled yam with kontomire stew and fish

    3- Kenkey with pepper sauce 4 - Fufu with palm-nut soup

    5- Don't know

FOOD FREQUENCY QUESTIONNAIRE

For the past one week, how often has you have eaten the following foods? (Please tick one)

Food/Dish	Once a day	More than once daily	1-2x a week	3-4x a week	5-6x a week	Rarely	Never
BEVERAGES							
Tea							
Cocoa e.g. Milo							
PORRIDGES							
Corn porridge							
Millet porridge							
Oats/wheat							
Rice porridge							
Tombrown							
MILK AND MILK PRODUCTS							
Evaporated							
Powdered							
Brukina							
Cheese/ wagashie							
SPREADS							
Margarine							
Peanut butter							
BREAD							
Wheat bread							
White bread							
DEEP FRIED FOODS							
Fried yams							
Fried plantain							
Beans cake(koose)							
FISH AND SEAFOOD							
Salmon							
Herrings							
Tilapia							
Crab/Shrimps							
VEGETABLES							
Carrots							
Green pepper							
Cabbage/lettuce							
Cucumber							

Food/Dish	Once a day	More than once daily	1-2x a week	3-4x a week	5-6x a week	Rarely	Never
MEAT AND MEAT PRODUCTS							
Meat (beef, goat meat)							
Chicken							
Offals							
Sausages							
Cow's skin (wele)							
Game (Bush meat)							
Eggs							
STARCHES							
Banku							
Kenkey							
Fufu							
Plantain							
Yam							
Gari							
Cocoyam							
Plain Rice							
Jollof							
Indomie/ spaghetti							
LEGUMES & NUTS							
Black/ red-eyed beans							
Baked beans							
Soy Beans							
SOUPS							
Palm soup							
Groundnut soup							
Light soup							
Okro soup							
STEWS							
Kontomire stew (no agushie)							
Kontomire stew (plus agushie)							
Okro stew							
Ghanaian gravy							

Food/Dish	Once a day	More than once daily	1-2x a week	3-4x a week	5-6x a week	Rarely	Never
FRUITS							
Pawpaw							
Pineapple							
Mango							
Banana							
Citrus fruits (Orange, tangerine)							
Apples							
Watermelon							
Avocado pear							
Grapes							
Guava							
Coconut							
DRINKS							
Fanta, coke, sprite etc..							
Fruit juices							
Vitamilk							
Sobolo							
Beer							
Wine/spirits							
SNACKS							
Meat pie							
Cake							
Doughnuts							
Chips							
Plantain chips							
Biscuits							
Roasted groundnut							

APPENDIX IV

INTERPRETATION OF BODY FAT AND MUSCLE MASS PERCENTAGES

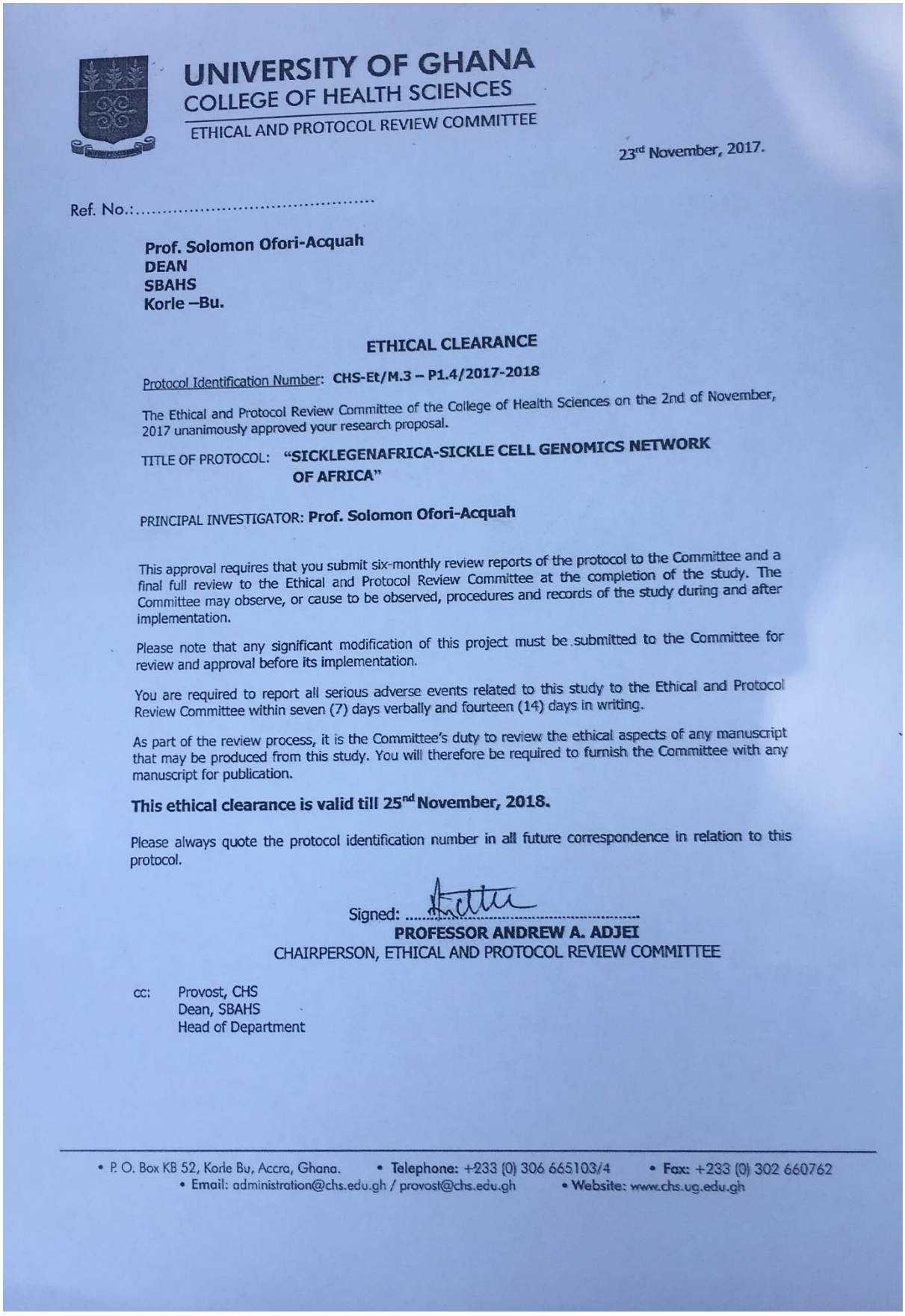
Body fat level interpretation for males				
	16years	**17years**	**18-39 years**	**40-59 years**
Low	< 10.4%	< 10.1%	< 8.0%	< 11.0%
Normal	10.4 - 24.7%	10.1 - 24.2%	8.0 - 19.9%	11.0 - 21.9%
High	24.8 - 27.7%	24.3 - 26.8%	20.0 - 24.9%	22.0 - 27.9%
Very high	≥27.8%	≥26.9%	≥25.0%	≥28.0%

Body fat level interpretation for females				
	16years	**17years**	**18-39 years**	**40-59 years**
Low	< 15.8%	< 15.4%	< 21.0%	< 23.%
Normal	15.8 - 34.5%	15.4 - 34.7%	21.0 - 32.9%	23.0 - 33.9%
High	34.6 - 37.1%	34.8 - 37.3%	33.0 - 38.9%	34.0 - 39.9%
Very high	≥ 37.2%	≥ 37.4%	≥39.0%	≥ 40.0%

Muscle mass level interpretation for males		
	18-39 years	**40-59 years**
Low	< 33.3%	< 33.1%
Normal	33.3 - 39.3%	33.1 - 39.1%
High	39.4 - 44.0%	39.2 - 43.8%
Very high	≥44.1%	≥43.9%

Muscle mass level interpretation for females		
	18-39 years	**40-59 years**
Low	< 24.3%	< 24.1%
Normal	24.3 - 30.3%	24.1 - 30.1%
High	30.4 - 35.3%	30.2 - 35.1%
Very high	≥35.4 %	≥35.2 %

APPENDIX V
ETHICAL CLEARANCE



UNIVERSITY OF GHANA
COLLEGE OF HEALTH SCIENCES
ETHICAL AND PROTOCOL REVIEW COMMITTEE

23rd November, 2017.

Ref. No.:.....

Prof. Solomon Ofori-Acquah
DEAN
SBAHS
Korle –Bu.

ETHICAL CLEARANCE

Protocol Identification Number: **CHS-Et/M.3 – P1.4/2017-2018**

The Ethical and Protocol Review Committee of the College of Health Sciences on the 2nd of November, 2017 unanimously approved your research proposal.

TITLE OF PROTOCOL: **"SICKLEGENAFRICA-SICKLE CELL GENOMICS NETWORK OF AFRICA"**

PRINCIPAL INVESTIGATOR: **Prof. Solomon Ofori-Acquah**

This approval requires that you submit six-monthly review reports of the protocol to the Committee and a final full review to the Ethical and Protocol Review Committee at the completion of the study. The Committee may observe, or cause to be observed, procedures and records of the study during and after implementation.

Please note that any significant modification of this project must be submitted to the Committee for review and approval before its implementation.

You are required to report all serious adverse events related to this study to the Ethical and Protocol Review Committee within seven (7) days verbally and fourteen (14) days in writing.

As part of the review process, it is the Committee's duty to review the ethical aspects of any manuscript that may be produced from this study. You will therefore be required to furnish the Committee with any manuscript for publication.

This ethical clearance is valid till 25nd November, 2018.

Please always quote the protocol identification number in all future correspondence in relation to this protocol.

Signed: .....
PROFESSOR ANDREW A. ADJEI
CHAIRPERSON, ETHICAL AND PROTOCOL REVIEW COMMITTEE

cc: Provost, CHS
Dean, SBAHS
Head of Department

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• Email: administration@chs.edu.gh / provost@chs.edu.gh • **Website:** www.chs.ug.edu.gh

STUDENT ID: 10382594
STUDENT NAME: FAUSTINA OFOSUA MINTAH

COMMENTS	STUDENT'S RESPONSE TO COMMENTS
EXAMINER 1	
Change "dietary patterns" to "food consumption pattern" (page 4)	Food consumption pattern has been used
Provide reference for classification of nutrition knowledge (page 24)	The reference (Tandoh, 2015) has been provided for the classification of nutrition knowledge.
Provide detailed information on methods for dietary data (page 25)	A modified pre-tested food frequency questionnaire from Asante et al., 2015 was used. The questionnaire was used once to gather data on the food consumption pattern of participants. Participants were required to recall the frequency of consumption of the listed foods within the past one-week prior to data collection.
Present text before figure (page 48)	The text has been presented before figure.
EXAMINER 2	
THEORITICAL FOUNDATION	
Spelling mistake on title	The word "nutrition" has been spelt correctly.
Use "adolescents and adult" instead of "people" in aim	"adolescents and adult" have been used instead of "people" in aim
Old statistics used in 1st paragraph (page 3)	Current statistics have been provided using the Ghana Health Service report for 2014.
Change wording in last sentence in 1.3 (page 4)	"to come up with a nutrition education program" has been changed to "develop a nutrition education program".
KNOWLEDGE ON PERTINENT LITERATURE	
The section on inflammation under biochemical assessment of nutritional status needs some more work.	An extensive review has been conducted in section 2.5.2 to address the case on inflammation and nutritional status.
DESIGN AND RELEVANCE OF METHODOLOGY USED	
Incorrect date and provide time frame for average attendance in section 3.2	The date has been changed from "194" to "1947". Average attendance is 50 people per day.
Provide aim for SickleGenAfrica project and link it to this study.	The link between the two studies has been stated in section 3.3.1.
Who collected the anthropometric and dietary data?	This was done by the researcher.

STUDENTS' COMMENTS MUST EXPLICITLY STATE HOW THE COMMENTS HAVE BEEN ADDRESSED

STUDENT ID: 10382594
STUDENT NAME: FAUSTINA OFOSUA MINTAH

Provide detailed description of nutrition knowledge questionnaire.	More details on the questionnaire has been provided in section 3.6.2
Declare source of funding	Source of funding declared in section 3.8
MAJOR FINDINGS	
Establish link between dietary data and nutritional management of SCD	The link has been stated in the last paragraph of section 5.3
What is the reason for combining daily beverages and porridges?	These two were combined because they are usually breakfast options for Ghanaians
Simple pie charts were not necessary	Simple pie charts have been removed and the figures stated in text.
Refrain from adding interpretation in results	All interpretations have been removed from the results section.
CLARITY AND JUSTIFICATION OF CONCLUSIONS AND RECOMMENDATIONS	
Is it necessary to conduct further studies to understand dietary practices?	Recommendation 4 has been changed.

STUDENTS' COMMENTS MUST EXPLICITLY STATE HOW THE COMMENTS HAVE BEEN ADDRESSED

STUDENT ID: 10382594
STUDENT NAME: FAUSTINA OFOSUA MINTAH

DECLARATION

I, Faustina Ofosua Mintah hereby declare that the above are my responses to the examiners comments and have been corrected in my thesis. This was done under the supervision of Dr. Joana Ainuson-Quampah, Prof. Solomon Ofori-Acquah and Dr. Catherine Segbefia.

.....

Faustina Ofosua Mintah
(Student)

04/02/2020
Date

.....

Dr. Joana Ainuson-Quampah
(Supervisor)

04/02/2020
Date

.....

Prof. Solomon Ofori-Acquah
(Supervisor)

11/03/2020
Date

.....

Dr. Catherine Segbefia
(Supervisor)

12/06/2020
Date

STUDENTS' COMMENTS MUST EXPLICITLY STATE HOW THE COMMENTS HAVE BEEN ADDRESSED