# MANAGEMENT OF MODERATE ACUTE MALNOURISHED UNDER-FIVE CHILDREN IN EKITI SOUTH SENATORIAL DISTRICT OF EKITI STATE USING MAIZE-PEANUT AND MAIZE-SOYBEAN BLENDS

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# TITLE PAGE

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#### **DEDICATION**

This work is dedicated to God, the Lord Almighty in appreciation of His mercy, protection,

ry, g His name.

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#### ABSTRACT

Moderate Acute Malnutrition (MAM) has been threatening the life and health of majority of global <5 years old children in sub-Saharan Africa, which is reduced by promoting affordable, readily available but locally grown supplementary food. Therefore, this study aimed to formulate and evaluate the effect of locally grown maize-peanut and -soybean supplementary food on the anthropometric parameters of <5 MAM children in Ekiti South Senatorial District of Ekiti State. Maize-Peanut {MPN} (149.4 g) and Maize-Soybean {MSB} (147.4 g) composite flours were produced and re-constituted to slurry. Proximate composition, minerals, vitamins, anti-nutritional and phytochemical composition, aflatoxin and microbiological assays and sensory properties were determined using standard analytical methods. The descriptive study involved four-stage sampling methods where children were randomly divided into four treatment groups comprising 18 each including Plumpy sup (control), maize-peanut blend (MPN), maize-soybean blend (MSB) and Nutrition Education (NE). Feeding trials (1 sachet per day, 149.4g for MPN and 147. 4g for MSB) and nutrition education were implemented for 12 weeks. Anthropometric parameters including body weight, height and mid upper arm circumference (MUAC) were assessed fortnightly. The proximate composition showed that the crude protein and carbohydrate contents of MSB (7.59 and 88.31%) were significantly (p < 0.05) higher than the MPN (7.19 and 87.63%) but with lower fat (3.81; 4.79%), respectively. The amino acid profile of both gruels was rich in hydrophobic and essential ones such as, valine, isoleucine, leucine and phenylalanine. The mineral contents (calcium, iron, zinc, iodine) for MSB and MPN gruels are 14.15±0.00, 11.81±0.00 mg; 5.54±0.00, 4.84±0.00 mg; 3.98±0.00, 3.13±0.00 mg; and 297.33±1.16, 293.00±2.00 µg, respectively. The vitamin B<sub>1</sub>, B<sub>3</sub>, B<sub>9</sub>, A and K contents of MSB and MPN gruels are 2.30±0.02, 2.22±0.03 mg; 0.08 ±0.02, 0.06 ±0.03 mg; 300.00±26.46, 150.00±17.32 mg;  $1.24\pm0.05$ ,  $1.13\pm0.04$  RE; and  $0.86\pm0.02$ ,  $1.98\pm0.02$  µg, respectively. The MSB and MPN gruels contained very low antinutritional (tannin, saponin, oxalate and alkaloid) contents (<1 mg/100 g) as well as low amount (0.04-0.05 mg/100 g) of Aflatoxin (AFB1 AFB2, AFG1 and AFG<sub>2</sub>) contents. The gruels showed a low population of yeast  $(1.4-4.2 \times 10^8 \text{ cfu/g})$  and moulds  $(1.2-2.3 \times 10^8 \text{ cfu/g})$  with the MSB gruel having higher consumer acceptability than the MPN and plumpy sup, respectively. The age, MUAC and weight-height z-score at enlistment and end line of the children fed with plumpy sup, MSB, MPN and NE were 14, 19 months; 11.21, 18.90 cm; 13.41, 15.67 and 18.53, 15.52, respectively. A 100% recovery from MAM was recorded for the children from plumpy sup, MSB, and MPN groups but 0.0% for NE group. Following complete MAM relief with the use of locally produced supplementary foods from both MSB and MPN, it is recommended that management of moderate acute malnutrition should be given necessary priority in primary health care in Nigeria and government should support mass production of local supplemntary foods.

#### **CHAPTER ONE**

#### **INTRODUCTION**

# **1.1 Background of the Study**

Childhood malnutrition, a public health problem of decades, played significant roles in the escalation of childhood infections, impaired physical, social and intellectual development as well as causing high rates pf death and sicknesses most especially in the developing countries, like Nigeria. It is a common phenomenon that undernutrition in early childhood predominantly evolves as from six months and two years when children transit from exclusive breast feeding to complementary feeding with rapid linear growth and intellectual development characteristic of the first 2 years of life. This is obviously as a result of intake of diets with poor nutrient and energy quality produced majorly from carbohydrate-based staples which are given to complement breast milk. Growth failure and developmental retardation are some of the consequences for the consumption of complementary food with poor nutrient and energy quality with low bioavailable vitamins and minerals by young children who are also not exclusively breastfed (WHO & UNICEF, 2003).

Childhood malnutrition has been classified into severe and moderate acute malnutrition. The World Health Organization (2012) defined moderate acute malnutrition (MAM) as moderate wasting with weight-for-height of -3 and -2 (international standard z-scores) or the mid-upper arm circumference (MUAC) between  $\geq$ 11.5 and <12.5 cm. Globally, over 150 million children are malnourished, Asia contributed about 55% of the entire stunted children while Africa had about 39%, 68% of children who are moderately malnourished were in Asia and about 28% were in Africa (UNICEF, WHO & World Bank, 2019). Studies revealed that several millions of children globally are suffering from moderate acute malnutrition (wasting), which has constituted a major worldwide health concern and threatens the life of more than 500,000 under five children yearly (Bari, Nazar, Iftikar & Mehreen, 2019; WHO, 2013; Bain, et al., 2013; ACC/SCN, 2000). As reported by Marzia, Laura and Paola

(2013), the annual prevalence of wasting (MAM) in West Africa is 15% of children less than five years of age.

Multiple Indicator Cluster Survey (MICS, 2017) carried out by National Bureau of Statistics indicated that 31.5% of under-five children in Nigeria were underweight, 43.6% were stunted and 10.8% were wasted, while in Ekiti State, 11.6% were underweight, 21.9% were stunted and 6.8% were wasted. This high prevalence at both the national and state levels calls for effective management programs. Unfortunately, strategies for the management of MAM in the vulnerable is yet to receive the necessary attention in the past 30 years, contrary to severe acute malnutrition (SAM), despite the fact that MAM affects many children in the developing countries (WHO, 2012). Lack of MAM programmes may be responsible for approximately 45% of death of preschool children (Black et al., 2013). Malnutrition and inadequate diet has been adjudged to be the largest determinant accountable for the global burden of disease and death most especially for the vulnerable among who are mostly children and adolescents (Forouzanfar et al., 2015).

Two broad approaches employed in managing children with moderate malnutrition, are supplementary feeding and dietary counseling. In most situations especially in the communities, dietary counseling is given to caregivers during contacts (ante-natal clinic, child welfare clinic, sick children clinic, food demonstration clinic, home visit) because health workers believe that caregivers are deficient in the skill to process complementary foods that are high in nutrients and energy which are important for proper growth and healthy living of their children. In cases of inadequate access to nutritiously adequate foods and food supplements, conventional ready-to-use supplementary (RUSF) or therapeutic foods are supposed to be provided but this has been in short supply and instead, fortified blended (supplementary) foods are usually given. This study was designed to develop supplementary foods formulated from ingredients sought locally for managing moderately malnourished children who are often not catered for in nutrition programming.

#### **1.2 Statement of the Problem**

Hands on experience showed that development partners are getting fatigued and supplies (such as lipid based/conventional RUSFs) for nutrition programmes are gradually prioritized, hence focus are being shifted from the production and supply of supplementary foods for the management of MAM to where cases of SAM are high. Meanwhile, attentions are being focused on children with SAM alone, while children with MAM are, therefore, left at the mercy of dietary counseling/nutrition education of the caregivers, and this usually does not lead to improvement in their nutritional status. Thus, the child may eventually present with SAM, which is more complicated and expensive to manage and may lead to death if adequate care is not provided on time.

It has been observed that MAM and SAM affected approximately 52 million children under five globally, out of which approximately 33 million are affected with MAM (Lenters, Wazny, Webb, Ahmed & Bhutta, 2013). It was reported through National Nutrition and Health Survey (NNHS) in 2018, that 5.5% of under- five in Nigeria has MAM and 1.5% has SAM while in Ekiti State, 3.5% of under-five children suffered MAM and 0.3% are severely acutely malnourished (NBS, 2018). Although, there is a nutrition intervention/ programme by the federal government to address the problem of SAM most especially in the Northern states and some states in the south but the contrary is for MAM. Children with wasting have different nutritional requirement from well-nourished and those that are severely malnourished.

Reports from Ekiti South Senatorial District, most especially during routine Child Welfare Clinic and Maternal, New-born and Child Week, shows that nutrition screening are done for under-five children using Mid Upper Arm Circumference (MUAC) tape, while the senatorial district recorded the highest number of children (11,616) with MAM over a period of three years (2015–2017) (Ekiti State Primary Health Care Development Agency, 2017 EKSPHCDA). There is also an inadequate supply of RUSF, thereby necessitating prioritizing usage for SAM while children with MAM benefit only from nutrition health talk given to caregivers.

The non-availability of necessary intervention for MAM contributed to child mortality, hence, the need to promote the use of readily available and locally produced RUSF. Standard RUSF (such as energy biscuit and plumpy sup) include different types of foods suitable for feeding severely malnourished children. These are often supplied by agencies such as UNICEF, but in recent times, these type of supports to states have stopped hence the need to produce one from commonly available food products. The RUSF proposed in this study will be developed using maize, soya beans and peanut, which are often used by the caregivers but with improper processing. The proposed formulated supplementary food is expected to address the gap occasioned by the stoppage of the standard RUSF usually supplied by donor agencies for the management of MAM. These food items are locally available, relatively affordable, acceptable and easily processed.

#### **1.3 Objectives of the Study**

# 1.3.1 General Objective

This research aimed at evaluating the management of moderate acute malnourished underfive children in Ekiti South Senatorial district of Ekiti State using maize-peanut and maizesoybean flour blends.

## **1.3.2 Specific Objectives**

The specific objectives of the study were to:

1. determine the proximate (moisture, protein, fat, carbohydrate, fiber and ash) composition of the formulated composite flours based on maize, peanut and soybean;

- assess the mineral (calcium, iodine, phosphorus, manganese, potassium, sodium, magnesium, zinc, copper, and selenium) composition of the formulated composite flours;
- 3. investigate the vitamins, (A, D, B<sub>1</sub>, B<sub>2</sub> and B<sub>6</sub>, B<sub>12</sub>, K, C, E, biotin, and folic acid) composition of the formulated composite flours;
- 4. examine the phytochemical and anti-nutrients (oxalate, alkaloids, saponin and tannin) composition of the formulated composite flours;
- 5. study the amino acid profile of the maize based formulated composite flour;
- 6. investigate the acceptability of the formulated composite flours;
- 7. assess the microbiological and aflatoxin content of the formulated composite flours and
- evaluate the effect of the formulated blended foods on the anthropometric parameters of moderate acute malnourished children in the Southern Senatorial District of Ekiti State.

# 1.4 Significance of the Study

The outcome from the research will be disseminated through different media by submitting to the School of Postgraduate Studies, Department of Nutrition and Dietetics and the library of the University of Nigeria Nsukka, respectively. The findings of this study will be presented at various global scientific conferences. The research will also be published in a peer-reviewed journal for the benefit of the scientific community, researchers and the global public.

This study is significant in the sense that by production of the supplementary food for children under-five years to prevent malnutrition, the caregivers whose children have MAM will have access to the locally produced and available supplementary food if found effective in the management of their malnourished children, thereby saving time, money and reduce under-five mortality and morbidity. The packaged products will be available at the nutrition unit of the LGAs for sale at affordable price to caregivers who are unable to produce the products themselves for lack of time.

It is hoped that there will be equally more production of the food items (raw materials) to be used for the production of this supplementary food because market would be created for the farmers and this will empower them economically. Large scale production of the proposed supplementary food will create more employment and both the wholesalers and the retailers will also be economically empowered. The supplementary food being locally produced, will cause government to spend less on importation of the standard ones, due to their affordability and easy accessibility. Availability of the supplementary food will afford the nutritionists and dietitians the opportunity of better management of moderately malnourished children they would encounter in the course of carrying out their duties.

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#### **CHAPTER TWO**

### LITERATURE REVIEW

#### 2.1 Management of Moderate Acute Malnutrition

Under-nutrition has always been recognized as a major determinant in child mortality and has been reported to cause about 28% of deaths of under-five children (Black et al., 2008). Wasting (MAM) and severe acute malnutrition (SAM) are the two types of undernutrition that affected 13% of children less than five years globally (Navarro-Colorado et al. 2008; UNICEF, 2010). The number of children affected by MAM (about 33 million) worldwide is unacceptably high and this has become a great global health challenge which has been contributing to the escalated rates diseases and deaths and also intellectual disabilities (Black et al., 2008; Black et al., 2008; Black et al., 2013). About eleven percent (10.8%) of children under-five years in Nigeria and 6.8% in Ekiti State have MAM (NBS & UNICEF, 2017). Supplementary feeding programme is employed to manage MAM in order to prevent the progression to SAM (WFP, 1999) and hence reduce child mortality and morbidity through malnutrition. Therapeutic foods typically employed in supplementary feeding program in managing malnourished children most especially those with either severe acute malnutrition or wasting. However, it has been observed that there are no evidence-based and scientifically valid guidelines for specially formulated supplementary diets in managing/treating children with MAM. Several types of specially formulated foods with different nutrient compositions are employed in managing malnourished children with the intention of improving their nutritional status and enhance their recovery but evidences have shown that their efficacy and effectiveness have not been satisfactory (WHO, 2012).

The resultant outcome of using therapeutic foods to manage children with MAM has been reported to be conspicuous because the children were observed to regain their appetite and consume adequate food for their weight to increase tremendously (Golden, 2009). Several negative outcomes would result if emphasis is placed on energy density, for example, the children would not recover their physiological or immunological normality; thus, hypersensitivity is delayed (Golden, Golden, Harland & Jackson, 1978), and sodium pump function (Patrick, Golden & Golden, 1980), glucose tolerance (Hadden, 1967), renal concentrating ability (Klahr & Alleyne, 1981) and muscle size (Hansen-Smith, Picou & Golden, 1979) remained anomalous, post treatment. Even though the children given these foods recovers speedily and attained the required weight-for-height, they often have shortfalls in functional tissue and have surplus of fat tissue and ended up being relatively obese (Golden & Golden, 1985) as a result of imbalance nutrients leading to the synthesis of inappropriate amounts of lean tissue.

Prior to the publication of the expert report on supplementary foods recommended to manage infants and children 6-49 children with MAM, WHO collaborated with other United Nations bodies (UNICEF, WFP & UNHCR, 2008) hosted a consultative meeting in 2008, the meeting was aimed at reviewing the authentication of the techniques and programmatic approaches to the management of moderate wasting. The technical note which is a comprehensive review that highlighted the proposed recommended nutrients requirement for children who malnourished moderately and choice of food and ingredients to be used for the formulation therapeutic foods (Shoham & Duffield, 2009; Golden, 2009; Michaelsen et al., 2009) was issued. No consensus on dietary composition of supplementary food towards managing MAM was reached at the meeting. The importance of developing specific guidelines and suggestions for requisite diet formulation and production of foods for different categories of malnourished children, most especially those with MAM (WHO/UNICEF/WFP/UNHCR, 2009) was however noted.

The consultation meeting also identified following areas; 1) dietary management of acute malnutrition and the nutritional needs; 2) Knowledge gap created by modalities for

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dietary provision and management which is expected to be addressed by research. Because of the uncertainties in the derivation of the recommended nutrient intake (RNI) for normal, healthy children in respect of some nutrients as a result of insufficient extant data to set RNI (Golden, 2009), absence of clear guidelines and recommendations for the formulation and production of supplementary diets from the locally available foods, this gave room for different types of local formulations whose efficacy and effectiveness remain doubtful. In view of this, World Health Organization in 2012, came up with nutrient requirement to be met by the formulated supplementary foods employed in managing children with MAM and the principles for their nutritional management, although it is still a proposal. According to Golden (2009), the recommendation, most especially the demand for most nutrients, occurred using the requirements between a well-nourished child living in an environment devoid of danger, clean and a malnourished child recuperating in inhospitable environment.

There have been several gatherings of international experts thereafter aimed at fine turning the guidelines earlier proposed and to ascertain their practicability as specifications for manufacturers of therapeutic foods (i.e. their capability to explain such suggestions into low-cost, pleasant tasting and effectual products), and look out for the clinical apposite of extreme precision in the context of compounded and confused operational realities. This has led to standardization of micronutrient premix formulation for Fortified Blended Foods (FBF) and RUSF. Beyond food and supplement composition, the gathering also suggested that general dietary sufficiency of MAM children could be used in peaceful environment for counseling approach that depends on household and family sources of food (Briend & Prinzo, 2009). Ashworth and Ferguson (2009) reported that the current practices that encourage the use of locally processed and produced foods for the treatment malnutrition by some agencies (and national programmes where such exist) revealed that very few counseling approaches adopts the recommended and the appropriate practices. It was discovered that specified

messages centered on the frequency, adequacy, density, utilization and safety of foods including those from animal origin were usually not conveyed. Several studies have appraised the outcomes of dietary/nutrition counseling for the caregivers of malnourished children who were taught about the rehabilitation of their children at home using the family foods. According to Annan, Webb & Brown (2014), nutrition counseling alone as a strategy for the management of MAM has not been effective, this may not be due to non- effectuality of nutrition education and counseling but may rather be due to non-adherence to instructions and guidelines.

Moderate malnourished children receive treatments and as well managed from home in supplementary food programmes where supplementary foods, such as Lipid-based RUSF or FBF, such as blended corn-soy plus milk and oil (CSB++) are distributed as take-home ration. Research have revealed that standard ready-to-use- supplementary foods has increased convalescence rates compared to other locally formulated foods most especially blended cornsoy food (De Pee & Bloem, 2009; Nackers, et al., 2010). The efficacy of corn soya blend fortified with milk and oil has also been revealed (LaGrone et al., 2012). Chang et al (2013) revealed that some Southern Malawian foods products (such as, CSB++, soy RUSF and soy/whey RUSF) were compared in order to observe their effectiveness and test the supposition that the clinical results obtained 12 months' post recovery from MAM would be preferable in children managed with standard ready-to-use- supplementary food. The team concluded that children who recuperated from MAM using standard RUSF should be seen as precarious and also as concern of interventions to minimize this probability of malnutrition in subsequent evaluations because the supplies of the standard RUSF may not be sustainable. This implied that it is better to manage children with MAM with local supplementary food that could be available throughout the year than depending on supplies from government or development partners that may not always be available.

Studies have shown that RUSF is effective but could not be considered as a permanent solution as possibility of relapse is high, if nutritious foods are not given to the children after recovery. For several years, supplementary feeding programs have always been used in economically developing nations as one of the strategies to manage MAM but there have been insufficient evidences about the efficacy of the locally formulated foods supplied in the course these programmes (Beaton & Ghassemi, 1982; Navarro-Colorado, Mason & Shoham, 2008.; Pe´rez-Expo´sito & Klein, 2009). Fortified blended supplementary food, such as maize-soya (CSB) and maize-peanut (CPB) blends made into slurry, are the traditionally used diets in supplementary feeding programs (Dewey & Adu-Afarwuah, 2008).

Specially formulated foods such as enriched blended foods (e.g. blended corn-soy (CSB) and the conventional ready-to-use-foods are said to be the cornerstone of treatment or community management of moderate acute malnutrition (Lenters et al., 2013). However, disquiet concerning the nutritional appropriateness of these formulated foods in connection with questions that surrounds adequate knowledge about home preparation and the evidence-based accomplishment of therapeutic diets in managing SAM have resulted to the evolution of surrogate foods for MAM management (Duffield, Reid, Walker & Shoham, 2004; Bhutta et al., 2008; De Pee & Bloem, 2009). Lenters et al., (2013) reported a well- organized review of community focused treatment of SAM and MAM in under-five children which is an approach used in economically developing nations that involves RUTF was carried out to identify the deficiencies in literature and to come up with the conclusions necessary for the inclusion of supplementary feeding programme in Lives Saved Tool (LIST) was carried out. They reported that there is a fundamental change towards community-focused treatment of malnutrition, which modified the perspective to the treatment of acute malnutrition and that community-based management is supported by absolute programmatic evidences.

Integrated Child Development Scheme (ICDS) and FAO have suggested the uses of locally accessible foods such as cereals and legumes in the formulation and production of supplementary diets to fight against malnourishment among the vulnerable in the low- and medium-income nations, because of they are easy to access, reconstitute and prepare (Baskaran, Mahadevamma, Malleshi, Shakara & Lokesh, 1999). The blend of cereals and legumes that bear a resemblance to the primordial diets and prepared in a manner that is comparable to the predominant food consumed at the family level have been suggested for the management of children that are moderately malnourished (Dijkhuisen, 2000). The nutrient quality and appropriateness of foods from cereals and legumes sources used for the formulation of the locally produced supplementary/complementary foods can be improved on by different processing and local fortification method (Natarajan, Shakar, Srihari, Rao & Rao, 1979; Malleshi & Desikachar, 1982).

## 2.2 Causes and Consequences of moderate acute malnutrition

The consequential effect of MAM on child diseased state and death in economically developing nations can be very high and devastating. Chandra (2002) revealed that aside the exposure of children to infections, malnutrition weakens their immune function and make them susceptible to acute ailments and hence they face a higher fatality threat. Findings have revealed that moderately malnourished children are subjected to an increased threat of comprehensive mortality approximately three- to four-fold compared to their healthy peers (Caulfield, De Onis, Blössner & Black, 2004; Black et al., 2008). Threat associated with mortality of children with MAM in the economically developing countries have been reported to have increase through common diseases such as pneumonia, measles and malaria which have been implicated as the specific causes (Black et al., 2008). Moreover, children with MAM with their compromised immune system, if not quickly attended to and treatment or management not initiated early enough, can proceed rapidly and degenerate towards severe

acute malnutrition (SAM), which is presumed to be deadly, fatal, difficult and expensive to manage (Garenne et al., 2009).

Contrary to the perception and believe that mortality increases only among severely malnourished, findings have revealed that mortality as a result of malnourishment (about 83%) transpires in children with mild or moderate malnutrition, which are sizeable number of the total malnourished children, instead of those who are severely undernourished (Pelletier et al., 1995; Black et al., 2008). In agreement with this assertion, World Bank (2006) implicated acute malnutrition been responsible for about 14.6% of annual child mortality in developing countries, severe acute malnutrition contributes about 4.4% while 10.2% was as a result of moderate acute malnutrition. The report went further to state that global acute malnutrition (presence of MAM and SAM in a population) is accountable for 14.8% of the entire disease adjusted life years (DALYs) in <5 years' children. Acute undernutrition has been indicted as one of the major precarious situations accompanying the occurrence of diseased conditions such HIV and tuberculosis which in turn increases the fatality. Hence programmes aimed at reducing undernutrition should also have an influence on these specifies diseased conditions (World Bank, 2006). Food insecurity has also been reported to compromise malnourished children's ability to learn thereby reduces their educational performance and over all development as a result of their poor attention, low interest and poor cognitive performances (World Bank, 2006). Therefore, undernutrition is said to have an undesirable effect or negative impact on the intellectual and societal aspects of child health.

#### 2.3 Nutritional Management of MAM Children

World Health Organization in 2012 made the following suggestions in managing moderate malnutrition through diets:

1. Cost effective nutrition interventions that includes but not limited to breastfeeding promotion, adequate supplementary feeding practices and support, nutrition education

and diet counseling for families most especially the caregivers, food security and other actions that recognizes and intercepts the root causes of under-nutrition should be included in the treatment of children 6-59 months that are moderately malnourished. However, cash transfer (conditional or non-conditional) should be provided and agriculture should be supported through strategies such as crop diversification to improve food security.

- 2. Diets are adequate in terms of quality and quantity should be consumed by all children, particularly those that are undernourished, this is important for standard growth and development as described by WHO (WHO, 2006).
- 3. Consumption of diets that are high in nutrient content supplied through either the conventional or locally formulated RUSF in addition to the family foods by children below five years that are moderately malnourished is important to enhance their additional needs required for the early functional recovery, weight and height gain, hence should be encouraged.
- 4. Diets with adequate nutrient contributes to the chances of children's ability to increase assimilation of nutrients necessary to accomplish calorie prerequisite and the indispensable nutrients need and so should be consumed. Amino acid and the essential nutrients needed by children recovering from malnutrition are expected to be met by the consumption of foods from animal sources, there is the need therefore to combine such foods with those from plant sources to complement each other. Adequate protein can also be obtained in foods from plant sources, most especially legumes or diets prepared when cereals and legumes are combined. Foods from plant sources contain some anti-nutrients and phytochemicals which oftentimes inhibits digestive enzymes and limits the absorption of some micronutrients but different processing methods has been reported to eliminate them.

- 5. Fermentation, germination, malting, soaking e.t.c. are some of the processing methods capable of reducing most of the compounds that interferes with the assimilation of nutrients and other toxins that occurs naturally, cyanogens or other prospective deadly or harmful ingredients found in foods from plant sources. Such should be part of the process employed in producing supplementary foods.
- 6. Foods used in managing or treating wasting (MAM) must supply all the necessary nutrients at the magnitude that will not be detrimental to children with MAM when ingested for a long period of time most especially when such foods are the main sources of calorie.
- 7. Availability, accessibility and adequacy of nutrient of the family food, possibility of sharing the take home ration of the supplementary foods and whether the children are still on breastfeeding are some of the factors to be considered when the quantity of supplementary foods to be given to a malnourished child is to be determined.
- 8. Safety and effectiveness of supplementary foods during formulation and production should be taken seriously most especially when they are the only source of calorie for the moderately malnourished children.
- 9. Easily digestible and water-soluble salts should be used in the formulation of supplementary foods because of the compromised digestive competency of the malnourished children, there is the need for regulatory agencies to authorize and monitor the mineral contents of the food and issue guidelines for the locally produced ones.
- 10. Hygiene standards set by Codex Alimentarius for the production of both supplementary/complementary foods should be complied with.

#### 2.4 Nutritional Requirement of Children with Moderate Acute Malnutrition.

Golden (2009) reported that statistics colleceted from well nourished person, in good health that lives in healthy and secured environs, who are also thriving was used to generate experimental data obtained at both national and international recommended nutrient intake (RNIs). He was of the opinion that the existing RNIs should not be applicable to most of the world population most especially those in the low and medium income countries where there are endangered children whose environment is deleterious; whose growth and development are retarded, who usually have repetitive infections, where there is absence of portable water and instead drink polluted water, who are vulnerable to fumes from cooking fires, consume foods contaminated with noxious substances from fungi and bacteria and lives on a meagre variety of plant products cultivated in the surroundings of their residences.

As the result of the foregoing, it was suggested that the requirement for nutrient for this category of individuals should be higher because their diets are much more poorer, they lose appetite when they get infection, they experience acute loss of weight compared with the children living without stress, and stays in safe and secured environments whose foods are diversified (Golden, 2009).

Studies have shown that children in impoverished households, who cannot experience steady growth during recuperation from sickness, whose diets are not nutrient dense enough to substitute for the nutrients expended in the course of ailment and to permit their coming back to the standard nutritional status and who are definitely malnourished has no internationally agreed RNIs. The published recommendations and the levels of RNI chosen has for this group of children is yet to be justified (Golden et al.,1995; Golden, 2001; IoM, 2002). However, the need to have RNIs for children in this category was addressed by various United Nation Organization (UN) and the Institute of Medicine at different times in their various reports but there was no proposal for changes of RNIs for vulnerable children (WHO, 1996; FAO & WHO, 2001; IoM, 2002).

FAO/WHO RNIs and AIs standard for well- nourished was compared with the nutrient

.h .a about

Nutrient (Absolute amount)		FAO/WHO RNIs		MAM (Food based)				MAM		
									(Comple	ment based)
Age range	Unit	7-12mnt	1-2yrs	3-5yrs	7-12mnt	1-2yrs	3-5yrs	7-12mnt	1-2yrs	3-5yrs
Energy used as divisor										
	kcal	673	956	1,242	673	956	1,242	673	956	1,242
Protein	G	10.1	11.1	14.5	16	23	30	17	25	32
Nitrogen	G	1.6	1.8	2.3	2.6	3.7	4.8	2.8	4.0	5.2
Minerals							$\rightarrow$			
Sodium	Mg	-	-	-	370	530	680	370	530	680
Potassium	Mg	-	-	-	950	1,350	1,750	1,050	1,550	2,000
Magnesium	Mg	53	60	73	135	190	250	200	290	370
Phosphorus	Mg	300	430	560	400	570	750	600	860	1,120
Sulphur	Mg	0	0	0	0	0	0	135	190	250
Zinc (high)	Mg	2.5	2.4	3.1	-	2 -	-	-	-	-
Zinc (moderate)	Mg	4.1	4.1	5.1		-	-	-	-	-
Zinc (low)	Mg	8.3	8.4	10.3	9	12	16	13	19	25
Calcium	Mg	400	500	600	400	570	740	560	800	1,050
Copper	μ	-	-	-	450	650	850	600	850	1,100
Iron (15%)	Mg	6	4	4	-	-	-	-	-	-
Iron (12%)	Mg	8	5	5	-	-	-	-	-	-
Iron (10%)	Mg	9	6	6	-	-	-	-	-	-
Iron (5%)	Mg	19	12	13	6	9	11	12	17	22
Iodine	μ	135	75	110	135	190	250	135	190	250
Selenium	μ	10	17	21	20	30	35	35	55	70
Manganese	Mg		-	-	0.8	1.1	1.5	0.8	1.1	1.5
Chromium	μ	-	-	-	7	11	14	7	11	14
Molybdenum	μ		-	-	10	15	20	10	15	20
Source: Golden (2009).										

Table 2.1a: Recommended nutrient intakes for the moderately malnourished expressed in absolute units.
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Nutrient (Absolute amount)		FAO/WHO RNIs			MAM (Food based)			1	MAM (Complement based)	
Age range	Unit	7-12mnt	1-2yrs	3-5yrs	7-12mnt	1-2yrs	3-5yrs	7-12mnt	1-2yrs	3-5yrs
Vitamins, water soluble										
Thiamine (vitamin B <sub>1</sub> )	μ	300	500	600	400	575	750	670	950	1,250
Riboflavin (vitamin B <sub>2</sub> )	μ	400	500	600	540	770	990	1,200	1,700	2,250
Pyridoxine (vitamin B <sub>6</sub> )	μ	300	500	600	540	770	990	1,200	1,700	2,250
Cobalamin (vitamin B <sub>12</sub> )	Mg	500	900	1,200	675	960	1,240	1,750	2,500	3,200
Folate	μ	80	160	200	150	210	270	240	330	430
Niacin	Mg	4	6	8	6	8	11	12	17	22
Ascorbate (vitamin c)	Mg	30	30	30	50	70	90	60	90	120
Pantothenic acid	Mg	1.8	2	3	2.0	3.0	3.5	2.0	3.0	3.5
Biotin	μ	6	8	12	6.5	9.5	12.5	8.5	12.5	16.0
Vitamins, fat soluble										
Retinol (vitamin A)	μ	400	400	450	650	920	1,190	1,280	1,820	2,360
Cholecalciferol (vitamin	μ	5	5	5	5	7	9	7	11	15
D)										
Tocopherol (vitamin E)	Mg	2.7	5	5	8	11	14	15	20	25
Phytomenadione (vitamin	μ	10	15	20	13	20	25	25	40	50
<u>K</u> )				Y						
Source: Golden (2009).		VER								

Table 2.1b: Recommended nutrient intakes for the moderately	malnourished exp	pressed in absolute units.
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There is the need for children who are malnourished to restore their lost and wasted tissues resulting from their current nutritional status and the requirement to have improved growth during recovery from illness, their requisite for nourishment of the tissues is often higher when compared with their peers' who has normal nutritional status. Malnourished children usually acquire weight and height slowly and the increase in their nutrient uptake is needed for their development which is more than what is required by well- nourished children for maintenance. Malnourished children require high proportion of nutrients changes for their growth and a richer and more nutrient-dense diet for rapid synthesis of functional tissue (Golden, 2009). There is the need to adjust the RNIs to accommodate some of the factors that are peculiar to moderately malnourished children.

#### 2.5 Recommended Nutrient Composition of Diets to manage and treat MAM Children

In order to prevent the conditions of children with MAM from degenerating to SAM, World Health Organization (2012) suggested that the supplementary diets for their treatment and management should be formulated and produced using foods available locally which should be accessible for the improvement of their nutritional status. Moreso, where there is food shortage as common in economically developing countries or where there are inadequate nutrients through the family foods, supplementary foods most especially from the combination of cereals and legumes should be use to treat or manage children with MAM. With the current climatic change, nutrition security is at risk and the situation would definitely aggravate during emergency situations such as during droughts and/or displacement as it has been observed in some countries in savannah Africa, some part of Asia and south America. De Pee and Bloem (2009) revealed that regular and the family diets cannot supply all the needed nutrients and calories by malnourished children and to bridge this gap there is the need to provide and make available specially formulated supplementary foods. Effectiveness of supplementary foods with diverse nutrient make-up has been observed to be suboptimal despite been recommended to ease the rates of recovery of moderate acute malnourished cchilden. Having this in mind, WHO (2012) suggested different strategies and approaches such diversification of diets, vitamin and mineral fortification of certain staple foods and different processing methods to improve the family foods that are usually deficient in different nutrients.

Before the nutrient requirement of supplementary foods for the management of malnourished was proposed, it was presumed that their chief diets commonly comprised basically of cereals and pulses, therefore the composition of the foods was arrived at, using different food composition databases (WHO, 2012). It was reported that the achievement of the recommended nutrient intake is possible by either providing supplementary foods or through the regular foods intake to compensate for the shortfall and to avoid any potential toxic effects, the quantity of nutrient and energy density necessary in attaining the recommended daily energy and nutrient requirement as well as the safety of the ingredients to be used in as al supplementary foods formulation was also considered (WHO, 2012).
Nutrient per 1000kcal	Unit	Minimum	Maximum
Protein	g	20	43
Fat	g	25	65
Mineral			
Sodium (Na)	mg	-	500
Potassium (K)	mg	1500	2200
Magnesium (Mg)	mg	280	420
Phosphorus (P)	mg	850	1400
Zinc (Zn)	mg	20	35
Calcium (Ca)	mg	1000	1400
Copper (Cu)	mg	1	35
Iron (Fe)	mg	18	30
Iodine (I)	μg	150	350
Selenium (Se)	μg	35	90
Manganese (Mn)	mg	1	2
Vitamins (Water soluble)			
Thiamin (B1	mg	>1	-
Riboflavin (B2)	mg	>4	-
Pyridoxine (B6)	mg	>2	-
Cobalamine (B12)	μg	>4	-
Folate (dietary folate equivalent	μg	>400	-
Niacin	mg	>25	-
Ascorbate (vitamin C)	mg	>150	-
Pantothenic acid	mg	>5	-
Biotin	μg	>20	-
Vitamins (Fat Soluble)			
Retinol (vitamin A)	μg	2000	3000
Cholecalciferol (vitamin D)	μg	20	60
Vitamin E (dl-α tocopherol acetate)	μg	>30	-
Phytomenadione (vitamin K)	μg	>50	-
Fatty acids			
ω-6 fatty acid	% energy	>4.5	<10
ω-3 fatty acid	% energy	>0.5	<3
Trans-fatty acids	% total fat	-	3
Source: WHO (2012)			

 Table 2.2: Proposed recommended nutrient composition of supplementary foods for use

 in the management of moderate acute malnutrition in children

 Table 2.3 Nutrient content per 100g powder of complementary food, from natural ingredients and premix

Nutrient content per 100g	Min	Target	Max	References			
Powder	(RNI/100g)	(RNI/100g)	(RNI/100g)				
Energy (kcal)	400	420	440	RNI	RNI	DRI	DRI
Protein (g)	8	16	16.5	7-	1-	6-	1-3yr
Protein from dairy sources (g)	3.6	7.2	-	12m	3yr	12m	
Fat (g)	9	10	-				
Linoleic acid (g)	1.5	2.5	-				
Sugar added (g)	0	5	10				
Vitamin A (µg RE)	300	800	1250	400	400	500	300
Thiamin B1 (mg)	0.3	0.6	-	0.3	0.5	0.3	0.5
Riboflavin B2 (mg)	0.4	0.8	-	0.4	0.5	0.4	0.5
Niacin B3 (mg NE)	4	8	10	4	4	6	4
6Pantothenic acid (mg)	1.8	3.6	-	1.8	2	1.8	2
2Pyridoxine B6 (mg)	0.3	0.6	-	0.3	0.5	0.3	0.5
Biotin B7 (µg)	6	12	-	6	8	6	8
Folate B9 (µg) DFE	80	160	300	80	150	80	150
Vitamin B12 (µg)	0.5	1.4	-	0.7	0.9	0.5	0.9
Vitamin C (mg)	15	60	-	30	30	50	15
Vitamin D (mg)	5	10	24	5	5	10	15
Vitamin E (mg)	2.7	5.4	-	2.7	5	5	6
Vitamin K (µg)	2.5	20	-	10	15	25	30
Calcium (mg)	260	800	-	400	500	260	700
Copper (mg)	0.22	0.44	1.0	-	-	0.22	0.34
Iodine (µg)	90	90	200	90	90	130	90
Iron (mg)	11.6	23	40	18.6	11.6	11	7
Magnesium (mg)	54	108	168	54	60	75	80
Manganese (mg)	0.6	1.2	2	-	-	0.6	1.2
Phosphorus (mg)	180	550	-	-	-	275	460
Potassium (mg)	700	773	-	-	-	700	3000
Selenium (µg)	10	15	36	10	17	20	20
Sodium (g)	-		0.4	-	-	0.37	1.0
Zinc (mg)	4.2	8.4	14	8.4	8.3	3	3

Source: WFP, 2018

#### 2.6 Supplementary Feeding Programme

Supplementary feeding has been described as the programme that provides additional food or supplementary ration to children and families beyond the standard portion of their household dietary regime to ameliorate or prevent under-nutrition (Beaton & Ghassemi, 1982; Kristjansson, et al., 2016). Visser et al., (2018) reported that supplementary feeding as a strategy, was adopted to proffer solution to temporary hunger and deep-rooted food shortage for the purpose of improving the nutritional status in both emergency and non-emergency situations to prevent nutritional deterioration of specific populations. Sometimes government through a programmatic approach at the state and national levels provide supplementary foods to increase health facility and services utilization, compliance to treatment procedures or attendance (and performance) at school. Like complementary feeding, the focus of supplementary feeding is to bridge the gaps in daily energy and nutrient requirements for malnourished children (Motuma, Azeb & Bekesho, 2016). Through supplementary feeding programme, supplementary foods are provided at different settings such as in kindergarten, crèche or community or given to children as take-home ration, it can also be delivered directly to homes and at times additional nutrition education are provided to parents and caregivers (Kristjansson, et al., 2016). Supplementary feeding is of two different types which includes: targeted supplementary feeding and blanket supplementary feeding.

# **Targeted Supplementary Feeding**

Targeted supplementary feeding is aimed to forestall the decline of nutritional status of MAM children to severe malnutrition and to rehabilitate them. Targeted supplementary feeding usually provided supplementary ration or food to specified members of endangered group or children who may unable to access the main portion. UNICEF (1997) reported that mild and moderately malnourished individuals and other vulnerable groups may not have access to the general ration in targeted supplementary feeding. Apart from following up from the therapeutic

feeding, the overall objective of targeted supplementary is the reduction in the prevalence mortality arising from acute and severe malnutrition. This is achieved through rehabilitation of children with MAM and other endangered groups with the provision of food supplements to them. Apart from the provision of treatment for malnourished children, it also makes available take home ration of locally formulated supplementary foods while micronutrient powder may or may not be added (WFP, 2016).

#### **Blanket Supplementary Feeding**

This type of feeding program provides supplementary diets for a specified period to the general population of recognized endangered groups which includes under- five children, elderly persons, pregnant women and lactating mothers. This is done to hamper the decrease in their nutritional status and thereby reduce death and disease conditions. Blanket Supplementary Feeding is adopted during emergencies when food insecurity is pronounced (WFP, 2016). This type of feeding program is aimed at providing either food or and micronutrient supplements for all members of specified groups irrespective of their nutritional status. Blanket supplementary feeding is peculiar to situation of extraordinarily high Global Acute Malnutrition (WFP, 2011).

# 2.7 Supplementary Food

Programmatic roles and ingredients used in the formulation are used as criterion for the classification of specialized food products or other food commodities employed to manage moderate acute malnutrition. The quality attributes of supplementary foods are; high energy and nutrients density, must be safe and without anti-nutritional components, must retain the qualities of palatability and must be acceptable (Motuma, Azeb & Bekesho, 2016). Because supplementary food and other food products used in MAM treatment are formulated using different types of ingredients, they are grouped based on whether they are used for treatment or prevention and whether they are lipid-based and non-lipid-based (WFP, 2014). To confront

undernutrion among the vulnerables in economically developing nations, Integrated Child Development Scheme (ICDS) and Food and Agriculture Organization (FAO) recommended that supplementary foods should be formulated and produced from locally available cereals and legumes (Natrajan et al. 1979; Malleshi & Desikachar, 1982; Anjum ChikkeGowda & Bhagya, 2013).

#### **Types of Supplementary Foods**

There are three forms of supplementary food which are Lipid-based nutrient supplement (LNS) e.g. Ready-To-Use Supplementary Food (RUSF), locally prepared foods with micronutrient powder and blended cereals with modified high nutrient density and micronutrient makeup which is required for the nutritional needs of moderately malnourished children to be met (WFP, 2016).

# Lipid-based Nutrient Supplement (LNS)

Annan, Webb and Brown (2014) reported that LNS are reffered to as RUSF. RUSF are foods which requires no preparation before consumption, their storage without refrigeration is safe, the risk of contamination is low because they have moisture content that is low and no dilution or cooking is required. World Food Program (2012) affirmed that RUFs are considered "lipid-based" because they have a large content of the energy which is provided by fats, also present are vitamins, minerals, protein and essential fatty acids (EFA). Varieties of products fall under this category. These ready to use foods are commercially produced, and are prepared from varying amounts of vitamins, minerals, sugar, peanut, vegetable oil and milk powder (James, 2011). According to Defourny, Seroux, Abdelkader and Harczi (2007), LNS has been successfully employed for the management of children with MAM. Energy biscuit, plumpy nut and plumpy sup are examples of LNS.

#### **Locally Prepared Foods**

The foods used in the management moderately malnourished children 6-59 months are prepared from locally available sources in the absence of lipid based nutrient supplement. Home fortification using micronutrient powder (MNP) is usually done by adding it to the prepared family foods. If locally formulated supplementary foods are produced with ingredients high in energy and macronutrient (protein, fat and carbohydrate) composition they could be comparable to the conventional RUSF.

### **Fortified Blended Foods (FBF)**

Fortified blended foods (FBF) are produced using the mixture of ingredients that includes but not limited to cereal, legumes, dried skimmed milk (DSM), sugar and oil, oftentimes, the blends are fortified with vitamins and minerals and are prepared in form of slurry, gruel or solids products. World Health Assembly (WHO, 1981; WHO, 2008) through relevant resolutions and recommendations that FBFs should be formulated as such that there would be no competition between such food and breastfeeding in compliance with local and international statute, most especially the International Code on Marketing of Breast-Milk. FBFs are developed in line with relevant Codex Alimentarius standards and technical specification for ready-to-use supplementary food, which include quality and safety, hygienic preparation and addition of additives aimed to improve the qualities of vitamins, minerals and protein (WFP, 2016; WFP, 2018; WFP; 2020). As expected, some of the locally formulated FBF would not meet all technical specifications for supplementary foods, their nutrient and energy content could be higher or lower than specified. The FBFs are used to augment the habitual diet of the child or the family foods in the course of treating or managing children with moderate malnutrition. The formulation of some of the fortified blended foods are not limited to cereals, legumes and sugar or vegetable oil alone, however there are others in which milk and additional fat sources are added to provide the required essential fatty acids, high-quality protein and

macro-minerals, like calcium and phosphorus (WFP, 2006). De pee and Bloem (2009) reported that the energy density, protein, fat and micronutrient content of blended foods are specially formulated and modified to meet the standard required for children that are moderately malnourished. The blended foods are distributed to children either as ready to eat or as dry take home portion for preparation and consumption (WFP, 2016).

#### 2.8 Effectiveness of targeted Supplementary Food programme.

The World Bank (2006) carried out a review on the effectiveness of targeted supplementary food programmes (SFPs) and was published in 2008 by Navarro-Colorado et al. The review evaluated the effect and effeciency of targeted SFPs in severe exigencies managed by different participating organizations. The report stated that from the raw data pooled together from all programmes reviewed, out of the 376,179 beneficiaries, 260,034 children equivalent to 69% recovered, 67,366 children (17.9%) defaulted while 1,763 children (0.46%) died, 47,016 (12.5%) did not respond to treatment. Forty-one percent of the programmes were up to date in all indicators in respect of recovery, defaulting and death. It was revealed from the report that eighty-two SFPs that had 376,179 beneficiaries were reviewed and that almost sixty-four percent of SFPs had recovery rates which is equal to or above seventy-five percent. The analysis went further to state that when those who defaulted (i.e. children who left the programme before their recuperation from moderate acute malnutrition) were added in this analysis, it was discovered that less than forty percent (39.3%) showed SFP is cost effective. Conclusively, Navarro-Colorado et al (2008), posited that many SFPs were ineffective because of some contextual factors and challenges that affected the implementation, however the recovery rate of 69% and death rate as low as 0.46% is enough to prove the effectiveness of supplementary diet in MAM management.

#### 2.9 Efficacy of Specially Formulated Foods in the treatment of MAM

Previous works by (Delchevalerie et al. 2010 and Ackatia-Armah et al. 2012 reviewed how safe and efficient different types of exclusively prepared food products for the management of moderately malnourished children in economically developing nations. In the systemic review carried out by Delchevalerie et al (2010), one of the studies enrolled 10,037 children with MAM and the compliance with peculiar nutritional composition such as WHO technical specification regarding safety and effectiveness of the formulated foods were assessed. Two of the studies draw an analogy between the provisions of foods with counseling for the management of MAM, Lipid-based Nutrient supplement (LNS) and blended foods were compared by seven studies. Also blended foods were compared to complementary LNS by two studies while three of the studies compared other LNS to specially formulated foods such as CSB++ or CSB premix were compared to other blended foods by another study. No significant difference was observed in death rates, advancement to chronic under-nutrition and the number of children that defaulted when lipid based nutrient supplement and dry food mixtures, with low lipid content were compared as showed in the findings of the review. Also from the review, it was revealed that there was an improvement in the number of children that recovered in the five trials as a result of the significance of LNS. Improved weight gain, WFH, and MUAC associated with LNS was also observed in the review, the recorded improvements were considered to be moderate. It was noticed in one of the trials that more children vomited in LNS group when compared with blended food group. The CSB++ showed less effectiveness when compared with locally formulated food as pointed out by one of the trials. In conclusion, findings from the reviews showed both the LNS and the blended foods were effective in treating and managing moderately malnourished children. Studies however revealed that where MAM is prevalent, most especially in Asian countries, there has been no such studies ever published (Matilsky et al., 2009; Hossain et al., 2011; Chang et al., 2013).

The efficacy of RUSF compared with corn-soy blend in managing and treatment of under-five malnourished children was investigated through five studies by Lenters et al (2013). Cluster Randomized Controlled Trials (CRCT) was used for two of the studies and the remaining three were RCTs designed. Finding revealed that the mortality rates of children fed with RUSF was not significantly different from those that received CSB when the outcomes were compared. Non-response rates showed a lower level of significance in RUSF compared to other groups, however the children fed with RUSF recorded greater mid upper arm circumference measurement of 0.04 mm/day while the weight increased by 0.61 g/kg/day than the group that were fed with CSB. The researchers observed the existence of considerable differences amidst the program appraised and they posited that the effect of all-inclusive management perspective for moderate malnutrition was indecisive and as such, more research with extensive scope and higher standard program assessment was recommended (Nackers et al., 2010; Ackatia-Armah, McDonald, Doumbia & Brown, 2012; Karakochuk, Van Den Briel, Stephens & Zlotkin, 2012; LaGrone, 2012).

Suri, Moorthy and Rosenberg (2016) reviewed the management of under- five moderately malnourished children with different dietary and non-dietary interventions (including fortified blended food and lipid based nutrient supplement) in twenty-two studies. It was revealed through the report that LNS had greater rate of recuperation and fast weight gain but not height gain, when compared with children fed with FBF. The researchers pointed out a wide variation in food supplements used in the formulation of foods (including foods that belong to the same category, such as lipid based nutrient supplement). The importance of extended research on SFPs to improve its efficacy was highlighted by Webb et al., (2014) like in other studies.

#### 2.10 Challenges in Community Management of Moderate Acute Malnutrition

Past study (Annan et al. 2014) identified the following challenges in the community management of MAM *viz*:

I. Low attention and lack of consensus: Despite the global caseload and associated mortality risk, MAM has not received the required attention which has been identified as a key constraint when compared with SAM. Some reasons have been adduced to this heedlessness and indifference managing MAM with products with high unit cost as it is being currently practiced, low coverage usually experienced in the programme, high percentage and frequent defaulting and instead of focusing on season-specific incidence rates rather emphasis is often placed on generalized prevalence rates. Appropriate definitions, acceptable admission and discharge criteria, types of supplementary foods to be use, treatment protocol that is defined and proofs from different studies are issues considered to lack consensus around programmatic guidance for MAM and this may be part of the reasons why attention given to MAM is less compared to SAM. De Onis et al (2013) however observed that there may be some improvement with the introduction of a target for moderate acute malnutrition among the six nutrition goals for 2025 set by the World Health Assembly because the goals put wasting and SAM at par alongside obesity and stunting including other nutritional challenges to be addressed globally. According to Bhutta et al (2013), there exist an evidence-based intervention from Lancet series highlighting adequately demonstrated efficiency to permit action that will scale up MAM and SAM management. United Nation (2013), also reported a proposal targeted at global improvement of nutrition and food security that is specifically aimed at wasting on the Post-2015 development agenda recommended by a panel consisting of high-level eminent persons. All the aforementioned initiatives are pointers to the continuous consciousness of the necessity

proffer solutions to moderate acute malnutrition simultaneously with other key consequences of undernutrition through the life process. Studies have shown that there is still the need to have an improved management and prevention strategies, ways of carrying out appropriate monitoring through feasible targets that would lead to appreciable annual rates of reduction of MAM whose progress recorded is adequately reported (Annan, Webb & Brown, 2014; Korenromp & Wustefeld, 2015; Deconinck, Bahwere, Diene, De- Bernardo & Adou, 2011).

II. Focus on products: Despite their known limitations, targeted and blanket supplementary feeding are predominantly the approaches focused on to address MAM. Part of the limitations of these forms of supplementary feeding is that they focus on product and without considering the underlying causes of malnutrition. Also, the approaches do not take into cognizance the appropriate consciousness needed to build a corporate and administrative capacity or improve on the training and educational demand of the professionals and also improve the skills and awareness of MAM children's caregivers. According to Jackson, Ashworth, Mohktar and Uauy (2014), before supplementary feeding which is seen as both community and facility-based intervention, can be adjudged to be a success, it is important to improve on human resource development, professionalize programming, invest more in behavioural change and awareness creation. The aforementioned strategies are necessary because beneficiaries of these supplementary feeding programs are often at risk of relapse (Matilsky, Maleta, Castleman & Manary, 2009). Confusion has been generated from the wide choice of specialized food items and their uses, such confusion surrounds treatment against prevention, SAM against MAM and supplement against diet replacement etc, for example, where there is existence of general food distribution or where there is an assurance of adequate access to specific ready to use supplementary food such as Plumpy'Doz. This type of food aside from been suitable only in emergency conditions it is also outlined to be distributed in a small daily portion to deliver key nutrients under specific conditions for specified number of children most especially those around 0.5-3 years old (WFP, 2012). There is therefore the need for mote guide and clarity on the varities of food to be used, the target of the food should be used for and what time and purpose such should be used.

- III. Inconsistent definitions in MAM Management programmes: There has been no consensus definition for MAM management programmme. While some supplementary feeding programmes include parameters such as underweight which is otherwise known as low weight-for-age (WFA) or stunting referred to as low height-for-age (HFA), children enlisted to participate in MAM programmes while mid-upper-arm-circumference measurement is used by some others. Studies have shown that the absence of agreed definition usually results in uncertainties over what criteria to be used for children that will be admitted and excluded during programme planning and this makes evaluation of the efficacy of MAM programs and associated products very difficult (Action Center, 2011; De Pee & Bloem, 2009). Annan, Webb & Brown (2014) therefore, suggested the need for an agreed term, approaches for reporting, how to monitor the actual programs by using different admission and discharge criteria, and the methods to be adopted for the assessment of prevalence, coverage and efficacy.
- IV. Confirmation gaps: According to Annan, Webb and Brown (2014), there are scarcity of empirical evidence in several key areas of MAM management and treatment. It has been reported that most often caregiver of children with MAM are given nutrition counseling and there has been no enough evidence on the effectiveness of nutrition counseling, whether when the counselling is combined with supplementary foods and medical interventions or done separately either at the facility or community levels

(Annan, Webb & Brown, 2014). The nutrition counseling received by the caregivers at all points of contact with the health workers requires high quality and intensive behavior change communication which is predicated on empowering them as part of the intervention in the process of treating/managing the children, this is what community MAM entails (USAID/SPRING., 2014). The cost-effectiveness of supplementary feeding program intervention and its context has been poorly documented, thereby making the acceptance very low (UNHCR/WFP., 1999). Thirdly, there has been confusion about appropriate comparison of products, rather than using research plan that makes promotion and programming a rule, compares calorie and nutrient densities and also compare packaging remains an exception. Care has to be taken in drawing conclusions from various studies because it has been observed in several studies that claimed to have compared different foods that the mode of delivery, dosage or portion size and Behaviour Change Communication (BCC) strategies are all different (Annan, Webb & Brown, 2014). Fourthly, the empirical valid studies on MAM requires expansion and a wider variety of settings and context because most of the large-scale studies done were derived from only a few countries especially Africa. Fifthly, there is the need for documented evidences on the usage of product meant to be used for community interventions and the outcomes need to be measured as well because evidence on sharing of products remains on hand (Annan, Webb & Brown, 2014).

V. Limited understanding of Nutrient Requirements for MAM: Based on WHO guidelines proposed in 2012, it is unfortunate there are still confusion about the clear definition of nutrient necessary for children who are moderately malnourished who are habitating in environment that is harmful, unhygienic and vulnerable. It is also clear that guidelines on formulation of food/diets, portion size and substitute to nutrient supplies are yet to be determined. Also, research should not be limited to nutritional

effectiveness and cost-effectiveness of individual products, such should be extended to varieties of actions and programs that have clearly-designated intents of MAM management with regards to nutrients supplied or provided and anticipated end results (Annan, Webb and Brown, 2014). The need to have a well-defined perception about the essence of weight increase in relation to the management of moderate acute malnutrition with different products cannot be overemphasized, there is the need to create better understanding while further research is expected on the gold standard for the content of the types of fatty acids most especially those that contained >1 more double bond (polyunsaturated fatty acids) in supplementary diets/food (Yameogo et al., 2017). Digestion and proper absorption of nutrients are important during the management or treatment of malnourished children, hence there is the need to study the efficacy of different levels of fiber consumption in children in this category (Michaelsen et al., 2009).

- **VI. Gaps in consensus around MAM management protocols:** Consensus on MAM management protocols is still lacking in some areas and the skepticism surrounding the management of MAM in specific or special situations is yet to be cleared. As reported by WHO/UNICEF/WFP/UNHCR (2010), the situation includes:
  - ✓ Management of MAM with Diarrhea: Recognition and treatment of diarrhea pathogens on admission contributes to the improvement of treatment of moderate acute malnutrition through rates of recuperation which is usually high when compared with weight increase etc.
  - ✓ MUAC for Monitoring: Abnormal fat distribution during Antiretroviral Therapy may make MUAC measurement an inappropriate strategy for monitoring progress in the program.

- ✓ Anthropometric Measurements: There are different responses to the newly developed MAM treatment and management by the anthropometric indicators such as weight-forheight, mid-upper-arm-circumference, weight-for-age and height-for age measurement and this has resulted in difficulties being encountered in taking decisions on suitability of the specific and combined indicators to be adopted. It is not yet clear as a result of continuing debate whether MUAC alone or in combination with weight–for-height should be for admission is enough as anthropometric indicator for moderate undernutrition in view of the dispute bothering on body shape (Cormic index) and its effect on weight-for-height z scores.
- ✓ Integration of Guidelines: The inconsistent in guidelines between and within nations necessitate the need for the integration of nutrition and HIV guidelines, this is important because of the discovery that there will be increase in decentralization and reduction in defaulting if community management of acute malnutrition and HIV-nutrition programs are rolled out at the same time. Important terms such as single entry, exit and referral criteria need to be clearly defined.
- MAM Admission and Discharge Criteria: There are some issues yet to be understood and such includes: appropriate criteria to be used to admit and discharge children who measures less than 67 cm and above 6 months of age, appropriate functionality to monitor treatment progress within the programme and finally how to appraise prevalence in programme situation. A key knowledge gap between the definition and identification of MAM and how treatment should commence for children below 6 months of age remain unaddressed.
- VII. Absence of basic medical treatment: Rather than citing treatment centers at the primary health care facilities where there are basic medical treatments, MAM programmes usually done at the community level where basic medical treatment are

not provided, children with MAM are therefore referred to the facilities where these medical services exist. Such children are often lost to follow-up because of the long distance they would have to travel long before treatment can be accessed (Annan, Webb & Brown, 2014). The long-run efficacy of MAM management has been observed to have been compromised because essential attendant care is absent. There is the need therefore for capacity building for institutions as well as professionals and it is very important to integrate MAM into national systems and protocols.

VIII. Monitoring and reporting of MAM management data constraints: Aggregation and analysis at a central level has been a major challenge in MAM treatment information. Information relating to MAM admission and management is usually not collected, collated and analyzed were program decisions are made and policies are formulated because MAM management is often implemented by NGOs and development partners (Annan, Webb & Brown, 2014).

# 2.11 Type of Foods and constituents for Moderately Malnourished Under – Five Children

Michaelsen et al., (2009) reported that populations that are prone to high prevalence of malnutrition usually consume diets that consist less or no fruits and vegetable, sparse quantity of plant protein from pulses and legumes with little no animal source food but feed principally on carbohydrate-based staple that include cereal (maize, rice, millet, sorghum) and tubers (cassava, yam etc). Such poor diets lead to situation that increases the risk of contagious sicknesses among the vulnerable most especially among children, because the diets are bulky with low nutrient and energy density as well as low digestibility of minerals which eventually results in weakened immunity, poor development and impeded growth. The vulnerability of children to malnutrition is as a result of their high growth velocity as well as energy and nutrient needs that are also high. They are equally exposed more to the consequences of synthetic

compounds that obstruct assimilation of nutrients, such as phytate, whose high level of presence impede several growth-limiting minerals absorption (Michaelsen et al., 2009).

Global figures on nutritional status as pointed out by Shrimpton et al. (2001) revealed that under-nutrition among under- five children commence from 6 to 18 months. Their brain develops to adult size at nearly 90% and growth velocity are high during this period up to the age of 2 years. Continued breastfeeding is considered to be very important (whenever possible) for moderate malnourished children most especially when they are less than two years of age and care must be taking in ensuring that breast milk is not replaced with supplementary foods/diets during MAM treatment/management. Study have shown that the time of recovery from MAM, SAM and stunting differ considerably (Michaelsen et al., 2009). According to the study, moderate wasting can be reversed within 2 to 4 weeks with appropriate management and treatment, whereas reversing moderate stunting (if it is possible at all) may take months or years. It may be easier to reverse stunting if the management and treatment started early most importantly before the first 2 years which has been globally acclaimed as the only opportune moment to address issues of under-nutrition malnutrition in children (Michaelsen, et al., 2009). Diets that are in high energy density has been recommended to play important roles in MAM prevention, most especially where there is high rate of malnutrition. Such diets are beneficial to children within 6-24-month age group, whether or not they have moderate wasting or they have moderate stunting (Michaelsen, et al., 2009). Children with moderate wasting may have the same requirement with those that are stunted or much difference may not exist when their requirement for more nutrients is demonstrated regarding calorific content are compared (Golden, 2009; WHO, 2007). But this requirement differs when malnutrition results from gastrointestinal challenges (which impairs assimilation of nutrients, or it is as a result of occurring contagious illnesses) compare to where insufficient diet or food insecurity results in malnutrition (Michaelsen, et al., 2009).

#### 2.12 Basic Ingredients used in Supplementary Food formulation

Diets meant for the treatment and management of MAM are formulated with the intention to be used for the rehabilitation of children with malnutrition or by meeting the additional needs of those most at risk by preventing the deterioration of their nutritional condition which focuses mainly on children between 6-59 month. These foods are meant to treat or manage moderate malnourished children from 6 months and above (WFP, 2019). Table 3 show the characteristics and recommendations of FBF for children whose age is between 6-23 months.

FAIGHREAM

	Characteristics	Recommendations			
_	Recommended number of servings	One serving per day			
	Kcal/kilojoules (kJ) per serving:	100-150 kcal / 418-628 kJ			
	Kcal/g of product as served	~0.8 kcal/g (3.3 kJ/g) but higher energy density is			
		preferable			
	A volume served from prepared/ready to	< 175 ml			
	eat product):				
	Viscosity	Not easily fed in a bottle			
	Consistency of product or when eaten.	Semi-solid (not liquid)			
	Protein:	10 -15% of energy as protein			
	PDCAAS	> 70%			
	Fat:	~20% of energy as fat and preferable			
	Essential fatty acids:	R			
	Alpha-linolenic acid content:	Ratio linoleic to alpha-linolenic acid 5 to 10:1			
	The hydrogenated fats with trans-fatty	Contain at least 130 mg, more preferable			
	acids is not allowed				
Added amount of sugar: Milk:		Preferably <10% of energy			
		Should contain ~5 g of milk solids non-fat (MSNF);			
		sources of MSNF include dried skim milk, whey, full-			
	C Y	fat milk, and semi-skimmed milk solids			
	Aflatoxins:	Maximum <5 ppb			
	Anti-nutrients:	If feasible, low extraction flours should be used to			
		reduce phytate levels, and dehulled legumes should			
		be used to lower fiber and polyphenols if colored			
	$\mathbf{N}^{\mathbf{Y}}$	varieties are used			
	Source: GAIN (2014)				

# Table 2.4: Recommendation of Fortified Blended Foods for <2 years old Children</th> Characteristics Recommendations

### Cereal

Globally, cereals constitute the major staple foods for mankind, most especially in the developing world. Cereals are the predominant foods in most countries and are considered as chief sources of nutrients. Serna-Saldivar (2010) pointed out that cereals are rich sources of both macro and micro nutrients most especially carbohydrates, fats, oils, and protein, anti-nutrients and phytochemicals. Cereals are the major ingredients that are used for the formulation and production of different types of foods. Food Standards Programme Codex Alimentarious Commission recommended that all grinded cereals are acceptable for human consumption can be used to formulate supplementary food, but they should be processed properly and adequately to reduce the fiber content and to eliminate anti-nutrient content as well as other phenolic materials which lowers the protein digestibility and mineral absorption (FAO/WHO, 1995).

# Legumes

Legumes are Leguminosae, characterized by the production of seed with a pod (Staniak, Księżak & Bojarszczuk, 2014; Kouris-Blazos & Belski, 2016). Well-known legumes consumed by human are peas, peanut, green bean, broad beans, soybean, lentils, lupins, mung bean, lotus and sprouts, are also called grain legumes or food legumes (Anonymous, 2013; Yorgancilar & Bilgicli, 2014). Oil seed and pulses are the two groups into which food legumes are divided into and their high oil content distinguished oil seeds such as soy bean and peanuts while pulses are dry seeds which are used as traditional food (Maphosa & Jideani, 2017). Legumes are good sources of essentially-needed amino acids and proteins, minerals, vitamins, complex carbohydrates, unsaturated fats and dietary fiber and are therefore nutritionally valuable (Bouchenak & Lamri-Senhadji, 2013; Rebello, Greenway & Finly, 2014; Annor et al., 2014). Apart from being nutritionally exceptional, legumes are also playing economic, cultural and medicinal roles because they contain beneficial bioactive substances (Philips,

1993). Messina (2016) and Ndidi et al (2014) reported many of the useful health attributes of legumes to include hypocholesterolemic, antiatherogenic, anticarcinogenic and hypoglycemic properties when consumed.

Maphosa and Jideani (2017) reported that because of the health and nutritional benefits of legumes, their demand is increasing worldwide. It was also revealed that people are replacing animal protein with plant protein which subsequently increased the request for legumes as their principal source of plant proteins. Before the increased clamour can be met, attention should be directed at the nutritional outline of different legumes, utilization of under-utilized legumes must be increased and production should be cheap. Also, there should be innovation on the upgraded products from legumes, consumers should be educated on the nutritional value of legumes and also new ways of inspiring the use of the subsisting ones should be sought (Maphosa & Jideani, 2017). Although, legumes was reported as an outstanding source of quality protein (as high as 20–45%) and high in lysine (Philips, 1993) but comparatively, their protein content varies, for example peas and beans have lower content that ranges between 17 and 20%, while lupins and soybeans has higher range of between 38 and 45% (Kouris-Blazos & Belski, 2016; Mlyneková, Chrenková & Formelová, 2014). FAO on the 7th of March 2016 through its Latin America and the Caribbean Regional Representative stated that legumes are very high in protein content and about twice that of cereals and this corroborated with what Kouris-Blazos and Belski (2016) and Leonard (2012) reported earlier. Protein from legume (except soy protein) are incomplete source of protein because they are very low in methionine, cystine and cysteine as well as in tryptophan (Kouris-Blazos & Belski, 2016).

Legumes contain remarkable quantity of different types of dietary fibers (both soluble and insoluble) and hence are important source of dietary fiber (5–37%) (Philips, 1993; Leonard, 2012; Kouris-Blazos & Belski, 2016). Non communicable diseases that are diet related including diabetes, cardiovascular complications and cancer are some of the emanating health

challenges confronting global population in recent times but high dietary fiber food plays important health roles in their prevention and treatment and also in the management of conditions like constipation, obesity, diabetes and piles (Anonymous, 2013; Karner, 2016; Maphosa & Jideani, 2017). Dietary fibers, most especially the soluble one through the formation of a protective gel lining along the intestinal walls reduces glucose and cholesterol assimilation into the bloodstream through the reduction of the cholesterol in the blood and glycemic index as well as the improvement of glucose tolerance (Maphosa & Jideani, 2017; Karner, 2016). Studies also show that legumes are invaluable part of human diet because they contain insoluble dietary fibers which are low in densities, increase fecal bulk and promote normal laxation because they are porous (Myriam, et al., 2016; Bliss, et al., 2014; Bliss, Weimer, Jung & Savik, 2013).

Messina (2016) reported that about 5% of energy in legumes are from fat and generally have no cholesterol because they are low (in fat), however peanuts and soybeans derives their energy, 45% and 47% respectively, from fat. Kouris-Blazos & Belski (2016) pointed out that legumes contain virtually no saturated fat they however contain remarkable quantity of monoand polyunsaturated fatty acids (PUFA). Legumes are inexpensive protein source, unsaturated fats, contain important bioactive phytochemicals as well as dietary fiber, different micronutrients and complex carbohydrates, eating them therefore contribute to a healthier lifestyle. To explore the nutritional and health benefits of legumes and also to reduce malnutrition, legumes are recommended to be included or added to children and infants' habitual diets and also through school feeding programmes, especially in low- and mediumincome nations. According to Maphosa and Jideani (2017) many functional foods could be formulated and produced from legumes because of the aforementioned nutritional attributes.

#### **Flours and Oil Seed Protein Products**

Codex Alimentarius (CAC/GL 8 -1991) recommended protein products and flours from oil seed as acceptable constituents of supplementary food because of their rich source of protein (50–95%). They can be used as the principal source of proteins for the manufacturing and formulation of complementary foods for older and younger infants as well as supplementary diets for malnourished children. The FAO/WHO (1995) report also recommended that flours, isolate and protein concentrates from the following that sources; sunflower (defatted flour), sesame seed (whole blended and defatted flour), groundnut (defatted and isolate), cotton seed (defatted flour) and soy bean flour (full fat, defatted, isolates and concentrates) if appropriately and processed to specifications.

#### **Fats and Oils**

Fats and oils are recommended to be added in quantities to supplementary foods to increase their energy density. However, caution must be applied not to add oxidized fat which could be detrimental to the wholesomeness and taste as well as the life-span of the final product (FAO/WHO, 1995).

#### **Essential Fatty Acids**

Studies have shown that growth retardation results from low intake of essential fatty acid and hence their presence in complementary/supplementary foods has been found to be beneficial (Uauy & Castillo, 2003; Hoffman, et al., 2004) therefore, taking diets that have high linolenic acid is be expected (MIYCN, 2009). The linoleic and alpha-linolenic acid contents of soybean and canola oil makes them more preferable to corn, peanut and palm oil that contains less omega-3 fatty acids (GAIN, 2014).

# Fiber and anti-nutrients

At the informal consultation meeting of some United Nation Agencies held in October 2008, emphasis was placed on the significance of minimizing anti-nutrients and fibrous

materials in complementary foods (WHO, 2010). Michaelsen et al., (2009) reported that substances like phytates, tannins and saponins when present in various food ingredients are capable of inhibiting bioavailability of nutrients, also inhibitors such alpha amylase, trypsin and haemagglutenins interfere with digestion. However, processing methods such as milling which is often used in traditional complementary food processing for whole grains are capable of reducing the high levels of dietary fiber and anti-nutritional factors they contain. De-hulling which is another processing method that can remove phytate and polyphenols (in coloured legumes or certain sorghums and millet), reduce tannin (found in chickpeas) and fiber (present in some legumes). Thus, it is not advisable to use un-milled grain for both complementary and supplementary foods (Michaelsen, Hope, Lauritzen & Molgaard, 2007).

## **Other Ingredients**

Some other ingredients recommended to be included in the formulation and production of complementary/supplementary foods includes dairy products and digestible carbohydrates including sugar and flavours (FAO/WHO, 1995), they could enhance the quality of nutrients and/or acceptance of the food, however they must be readily accessible.

#### 2.13. Food processing methods

Floros et al (2010) defined food processing as the modification of foods from their original state for better preservation status and improved nutrient quality. International Food Information Council pointed out that food processing is seen as an intentional change in foods which is brought about before their accessibility for consumption. Food processing as a chain of different methods is an important part of food production that linked agricultural production which leads to provision of food in an acceptable state and necessity time by consumers (Floros et al., 2010). There could be so many reasons why food are preserved, food can be preserved for use in times of scarcity; to increase life span; detoxify; anti-nutrients removal, improvement of digestion and making nutrients available; improvement of pleasant tasting and functional

properties. Processing most often led to changes of the constituents of food and some of these changes can bring about improvement and enhancement in the quality of the food, it could also be detrimental, depending on the type of processing method employed (Weaver et al., 2014). Heat treatment has a negative effect (as a processing method) on the quality of foods, for instance, causing trans fats in foods, acrylamide formation, elevated sugar level in supplementary foods and nutritional degradation, have been reported, but to balance this view, it is important to look at the beneficial effect as well (Van Boekel et al., 2010). The effects of food processing procedures on their nutritional qualities differs depending on the specific methods desired. Mensah and Tomkins (2003) comprehensively reviewed how household processing methods can be engaged to impact positively on the nutritional value and safety of complementary/supplementary diets and provided a primitive classification of the importance of these technologies and their beneficial influence.

# Soaking or steeping

Soaking or steeping is a pretreatment of grain aimed at the removal of the husk or skin. Soaking process has some effects but not limited to increase moisture content, total carbohydrate, starch, hydration coefficient, total protein, seed weight, ash, fat, stachyose, fiber, raffinose, non-protein nitrogen, reducing sugars and minerals content of cereals and legumes. Soaking the seeds in water and processing effectively removed the anti-nutrients which includes tannin, phytic acid, trypsin inhibitor and also heme-agglutinin activities were decreases when 0.5% sodium bicarbonate is added (El-Adawy, Rahma, El-Bedwy & Sobibah, 2007). Time and period for steeping however depends on temperature and degree of aeration of the steep water. For instance, a temperature of 10-12°C is recommended with steeping times of 40-60 h for cereals, but when the steeping temperature increases, the steeping tome reduces. However, a temperature of 20-25 °C is recommended with steeping times of 16-20 h for legumes (Kent & Evers, 1994).

# Drying

Sontakke and Salve (2015) defined drying or dehydration as the transfer of heat under controlled condition or simply as application of heat, purposely to reduce the water content of foods through evaporation resulting in solid products resulting from concurrent heat and mass transfer. Reduction of water activity in foods is necessary to extend their shelf-life, reduces their weight and as well reduce transportation and storage cost. Also, drying transforms foods to different forms which makes storage, packaging, transportation and use more convenient. For example, when liquids such as milk or coffee extract are dried, they are transforms to a dry powder which can be reconstituted with water to the original form (instant products) when the need arises (Kumar, Tiwari, & Belorka, 2015). Drying techniques is applicable in the production of many products ranging from dairy products (milk, whey, and creamers), starch derivatives, sugar beet pulp, beverages (coffee, powdered drinks), fruits, processed foods, vegetables, spices and potato tubers. According to Lewicki (1998), absence of enough water most especially during storage prevent the activities of micro-organisms that promotes unpleasant chemical changes in food composition, causes food spoilage, decay and many enzymes reactions (Lewicki, 1998). Dehydrated products, in most cases, are further processed or consumed after rehydration and drying often produce a crumbly, readily milled stable product that may be stored for long (Mercer, 2014).

# Roasting

Roasting is responsible for flavor enhancement through some reactions known as caramelization and maillard browning reaction that show on the surface of the food and thereafter give the foods their toasty brown color and sweet, caramelized flavors (Schulze, 2019). Dry roasting is used in the processing of foods such as nuts and seeds, foods processed through this method are stirred periodically, as they are roasted to ensure even heating (Gahlawat & Sehgal, 1994) and prevent burning. This heat processing method, reduces the moisture content, thereby concentrating the food value. Roasting imparts a nutty flavor in foods and thereby enhances the acceptability. Elimination of most anti-nutrients in legumes (such as, trypsin inhibitor, hemagglutinin, goitrogens, etc.) could be done through roasting process (Ndidi, et al., 2014). Similarly, the *in vitro* protein, starch digestibility and *in vitro* iron availability of complementary foods is elevated by 15-21, 16-19 and 12-19%, respectively after roasting process (Gahlawat & Sehgal, 1994).

# Milling

Milling or size reduction are some sequential operations that involve breaking, grinding and separation and through this the bran and germ are reduced into a uniform particle size (flour) (Hassoon, Daziki, Mis, & Biernacka, 2021). The outer bran in coarse grains is fibrous, bitter, astringent, or colored. Milling of the coarse grains is therefore desirable to confer adequate acceptance by the consumers (FAO., 1994). Size reduction of cereals change the whole grain into flour before different products can be prepared for human consumption. The milling quality of cereals plays a significant role in further processing and diversified food uses of the crops. Generally, the cereals are expected to produce high quality flour, which is achieved by separation of endosperm from the bran and germ (Khatkar, Panghal & Singh, 2009).

#### Blending

Blending is a process of combining substances, which may not necessarily be homogenous but becomes inseparable in a comparatively gentle process. Blending is a unit operation and one of the important phases in a manufacturing process. It is the homogenous mixing of the entire ingredient (Baumber, 2021). Two or more ingredients are combined together during blending so that they lose their individual characteristics and become smooth and uniform. The main objective of blending is to combine or mix so that the constitute parts are indistinguishable from one another resulting into the lipid-based paste product (Amagloh et al., 2012).

# Sieving

Sieving or screening is a process of segregation of solid particles or a laboratory procedure in size analysis and it remains of the oldest operation yet an important technique in food processing (Liu, 2009). Sieving is understood to be that process that involves separation of smaller particles from bigger ones by passing them through a sieve (Jillavenkatesa, Dapkunas & Lum, 2001). The process of sieving separates particles according to their sizes by passing the material across a number of sieves of non-identical mesh sizes and determines the particle size distribution of a material. Sieving is applicable in a unit operation that contain mixtures whose components are of different sizes. Sieving process prevents contamination of products from foreign bodies which is an important role in food industries and this is achieved using sieves (which often vibrating). Sieves are of different types and sizes, the types to be used depend on the types of particles to be separated. It is used in flour mill or other food processing companies, where impurities like husks and stones are removed from wheat.

# Packaging

Packaging is a process that involves the technology of putting products in different types of materials aimed protecting the content from environment during the period of storage, distribution, sales, and use. Beside protection of contents from different types of contamination, packaging is also engaged in designing, evaluating and preserving food products. Packaging materials plays important roles in transportation of goods/products, warehousing, logistics, sale, and end use. Soroka (2002) reveals that packaging is important because it encloses, offers protection, preserves, makes transport easy, educates and sells the products. No doubt, packaging in the production chain is an indispensable part of processing and distribution of

foods. Packaging has different functions among of which preservation plays a major role and this should be clearly understood by food manufacturers (Coles, 2003).

#### Food packaging materials

Apart from offering protection, packaging aids for food demonstration, tampering resistance, and special physical, chemical, or biological needs, and also allows customers to make an informed choice. Packaging materials usually carries labels that has nutrition facts and useful information about the product (Paine & Paine, 1992) and they are categorized into two main classes which are shipping containers and retail containers.

Retail containers protects and advertise products for retail sales and domestic storage. Examples of retail containers are sachet, paper bags, overwraps, metal cans, flexible plastics, collapsible tubes, glass or plastic bottles and jars, pots and trays, plastic tubs and paper cartons.

# Special feature required for RUSF packaging

The RUSF are lipid-based paste products. The fat content is generally high so, it is very susceptible to oxidative rancidity. To prevent leakage and protect the product against contamination or any environmental factors that may negatively affect RUSF, they are packaged in food-grade sachets that are hermetically. World Food Program (2016) emphasized that when it is opened and put in contact with the mouth sachet material should not represent a hazard for infants and young children.

# High Density Polyethylene (HDPE)

High density polythene is one of the largest globally malleable material classified after polyvinyl chloride. High-density polyethylene (HDPE) was reported by Kumar and Singh (2013) to be a thermoplastic material comprising of carbon and hydrogen which are atoms bonded to form a high-molecular weight product. The HDPE is produced at lower temperatures and atmospheric pressure as a liquid phase process. It softens at 120-130 °C and so it can be used for hot filling, steam sterilizing or cook in the bag applications. Due to its greater rigidity,

it can be used in thinner gauges thereby saving money. It has excellent retention of essential oils such as aromas. In general, the polyethylene are soft and flexible in film form with good impact resistance. However, they can be hard to open. They are very resistant to water and water vapor; the higher the density, the greater the resistance, i.e. the lower the value of water vapour transmission rate (WVTR), but the oxygen transmission rate is high (Coles, 2003). The main advantages of HDPE are:

- a) Water proofness, low gas and water vapor permeability.
- b) Good aroma retention.
- c) It is heat sealable, can be oriented and made into bags.
- d) It is useful in wrapping meat, fish and dried foods (Marsh & Bugusu, 2007).

# 2.14 Shelf life

Shelf life is the active time for storing the fresh (harvested) produces without becoming unwholesome or spoilt before final consumption. In food processing, a product's shelf life is described as the period between when the food is produced or packaged and the time it becomes unwholesome and becomes a risk to customers when consumed. Shelf life of food depend on the following factors formulation, processing, packaging and storage and can be impacted by various features, such as; contamination by microorganisms, exposure to light, mechanical stress, heat, transmission of gases and moisture. Concentration of chemical compound, moisture content or microbiological index are some of the parameters around which product quality is often mathematically modeled (Azanha & Faria, 2005).

# 2.15 Relevant foods and ingredients and their nutritive value

#### Maize

Maize (*Zea mays*) also known as corn is an important food that is eaten routinely and constitutes a dominant size of a standard in Sub Saharan Africa. Both human and livestock uses maize as their main energy and protein sources as well as other nutrients. Maize is one, if not the most important and widely distributed cereals crops of the world. The chief proteins of

maize are glutelin and prolamine (zein). The zein fraction was shown to be very low in lysine content and lacking in tryptophan. Whole maize is a good source of thiamine, pyridoxine, pantothenic acid, fair sources of riboflavin but poor sources of niacin (Iken & Amusa, 2004). Maize accounted for up to 60% of the daily human protein supply (Sofi, Wani, Rather, & Wani, 2009). It contained 4-10% protein, 2-4% fat and some of the important vitamins and minerals (Envisi, Umoh, Whong, Abdullahi & Alabi, 2014). Lysine and tryptophan are the two essential amino acids lacking in varieties of maize (Serna-Saldivar et al. 2008), which are nutritionally essential for humans and animals. Despite this limitation, maize is widely playing important roles as the major ingredient in complementary foods for children and also could be fortified with legumes to provide the missing nutrients. Supplementing grains with key amino acid during fermentation has provided the needed solutions to the aforementioned problems/challenges (Prasanna, Vasal, Kassahun & Singh, 2001), although amino acids are often lost from foods through different processing methods most especially in maize. Prasanna et al., (2001) stated that tangible achievement can be realized through conventional breeding to increase the nutritional quality of food crop but the attainment of this requires continuous administrative, financial and scientific effort, long-term investment, sustained research efforts and patience. This is the context through which the story of quality protein maize assume significance, therefore the need to genetically ameliorate the poor nutritive value of cereal grains such as maize has been recognized over time.

Corn has been adjudged as one of the leading cereals globally followed by rice and wheat respectively because of their high starch content (Prasanthi, Naveena, Rao & Bhaskarachary, 2017). It has been playing prominent roles in formulation of blended locally produced complementary and supplementary foods to forestall malnutrition most especially in economically developing nations apart from being used as a major source of food for human nutrition after different processing methods (Kent & Evers, 1994). Maize is prominent and accepted not only because it is a source of food for both humans and animals (Nuss & Tanumihardjo, 2010) but also for industrial products including industrial alcohol, fuel, ethanol starch, sweeteners, corn oil and beverages. Brown et al., (1988) stated that 15% of world's protein as well as 20% calories is provided by maize, and more than 200 million people derives their dietary staple from it. The main structural composition of maize is the endosperm that make about 71% of the total content, it is a complex carbohydrate and good source of pure calorie (Dias et al. 2010). Nuss & Tanumihardjo (2010) reported that the three distinct compartments of the edible kernels make up about forty-two percent of the maize plant dry weight (DW) and the 3 distinct compartments are the endosperm which is about eighty-three percent, embryo (germ) also about eleven percent and the pericarp. According to USDA (2009) maize supplies about 365kcal/100g energy density, similar to wheat (340 kcal/100g) and rice (360 kcal/100g) apart from containing about seventy-two percent starch, ten percent protein, and four percent lipid. Corn is processed industrially to flour, corn meals and its products globally, employing different food processing methods, the products obtained are oftentimes precooked, refined, dehydrated and fermented (Prasanthi et al., 2017). Corn, apart from the provision of energy as a food source, has many health benefits, for example study revealed that it is a reliable source of carotenoids well as polyphenols which associated with lowering the risk of various worsening illnesses when consumed (Messias, Galli, Silva, Schirmer & Rombaldi, 2013). Processing methods plays important roles in vitamin and mineral content of various products from maize because they go along divergent pathways of production running through the raw material to end products during processing which eventually leads to the changes in their nutrient constitution.

Maize is high in three major minerals namely; potassium phosphorus and magnesium, however, it is low in calcium, zinc, copper, sodium, iron and manganese (Asiedu, Nilsen, Lie & Einar, 1993; Prasanthi et al. 2017). Difference in mineral composition of products developed or formulated from maize may has been reported to result from genetic make- up and environmental factors, soil composition, fertilizer used and the processing methods adopted (Food Standard Australia New Zealand, 2006; Ikram, Mohammed & Arifa, 2010). The significant physiological role of maize is as a result of the considerable amounts of dietary fiber it contributes by the grain, the kernel seed coat functions well because of the crude fiber (eighty-seven percent of the seed coat) which is also exist in minute quantity in the endosperm and germ walls (Nuss & Tanumihardjo, 2010). The importance of dietary fiber to our digestive health and regular bowel movements has been reported, it can improve cholesterol and sugar level, can helps to feel fuller for longer period and this can assist in preventing ailments such as diabetes, heart related disease and cancer most especially bowel cancer (Nutrition Australia, 2014). Fiber has been reported to helps normalize intestinal obstipation (severe constipation) most especially the insoluble part, it equally accelerates and increases the feacal bulk and also contributes to low availability of iron in maize (Nuss & Tanumihardjo, 2010).

#### Soya beans

Soya bean contain high quality and cheap source of protein and oil; it is a member of the family Leguminosae. It has 30% protein content which is the highest among the food crops and ranked after peanut regarding oil content (18%) within food legumes. Soybean is the cheapest of protein-rich foods when compared to others such as meat, fish, and eggs. WHO (1988) revealed that other plant protein sources have inferior amino-acid profile compared to soybean. Soybean, contains trypsin and growth inhibitor and hemagglutinin which can be inactivated by autoclaving soybean at pressure of 14 lb for 30 min. or roasting in 100-110° C for some time (Khokhar & Richard, 2003). When soybean protein is heat processed, the PER ranges from 1.9-2.2 and when it is supplemented with methionine, it increases markedly to 2.8-3.0. The chief protein of soyabean is a globulin known as glycinin; other proteins present in small amount are phaseotin and legumelin. It is a rich source of lysine and threonine. Although

deficient in methionine and cysteine but this can be overcome by either the addition of 0.15% of methionine which results in better protein energy ratio than casein or by blending with other protein to provide a good balance of amino acids (Islam, Chowdhury, Islam & Islam, 2007). Soybean occurs in different colours ranging from yellow, black, blue, green and brown. It provides high content of oil and protein; hence it is an important crop. Oil (22%) and protein (37%) content of soybean flour is responsible for about 60% of its dry weight. The presence of all essential amino acids is responsible for the high standard of the protein content, also high quantity of lysine present differentiates soybeans from other legumes and cereals (Nwokolo, 1996; Liu, 2004) and as a result its protein digestibility corrected amino acid scores (PDCAAS) is very high. Soybean as an excellent source of protein could be recommended for those who perpetually consume diets that are low in animal-source foods because of the aforementioned reasons.

The presence of elevated content of n-3  $\alpha$ -linolenic fatty acid, a proportion of unsaturated fatty acid and -6/-3 Polyunsaturated Fatty Acid ratio of about ~7, makes the oil a high quality. Dried soybean is made up of about 35% carbohydrates which includes pectin, hemicellulose, cellulose and a trace amount of starch which are otherwise known as insoluble carbohydrate. Soybeans contain both water-soluble (which are folic acid, thiamine, vitamin C, niacin, pantothenic acid and riboflavin) and fat-soluble vitamins. Vitamin C content of soybean is insignificant in mature beans but the quantity is measurable in both unripe and sprouted seeds (Liu, 2004). Soybean contain no vitamins D and K essentially, but A and E are the fat-soluble vitamins present, like other plant sources, with provitamin  $\beta$ -carotene as the vitamin A form and very negligible content in matured seeds. Soybean is a very good sources of phosphorus and potassium but very low in sodium as in most legumes, however the phosphorus content has only partial biological availability because a remarkable quantity is present in phytic acid form. Apart from been an excellent source of calcium and magnesium, soybean also contain

important protease inhibitors, lectin and phytase which are anti-nutritional factors, it is also poor in iron and contain minimal quantity of zinc and iodine (Liu, 2004).

Lusas and Riaz (1995) highlighted whole soybeans or grits, defatted soy flour and fullfat soy flour as some products from soybean that are used to formulate diets in managing vulnerable and undernourished populations. Formulated foods such as corn–soy blend and concentrated products like soy protein concentrates and soy protein isolates are produced from non-dehulled and dehulled soybeans and also from defatted and toasted soy flour (Rackis, Honig, Sessa & Steggerda, 1970; De Pee & Bloem, 2009). Studies have showed that despite the fact that various soy flour contain fiber and reduced quantity of anti-nutrients but defatted soy flour (32%), soy concentrates (21%), and soy isolates (3%) also contain different proportion of carbohydrate content (Rackis et al. 1970; Sandstrom, Kivisto & Cederblad, 1987; Davidsson et al. 1994). Conclusively, the protein quality of soybean is high because of its balanced amino acid composition and good quality of fat content, although the presence of antinutrients is prominent, especially phytate and phytoestrogens, however the probable dismissive consequence of the anti-nutrients in soybeans in malnourished infants and young children is yet to be investigated or researched.

# Peanut

Peanut is globally recognized as one of the most popular legumes or pulses and also one of the most nutritious seeds known as groundnut. There are many ways of preparing groundnut but it is prepared as fresh boiled or roasted in Africa. Nwokolo (1996) reported that peanut is available in the market as ingredient for the preparation of peanut sauce when grounded or milled. When compare with other legumes, peanut is an excellent source of fat (it is made up of more than 40% fat). Although, it is deficient in methionine and lysine but the protein content is about 25% and has carbohydrate content that is relatively low (about 28%). Apart from having a very strong nutritional profile, peanut is said to be an excellent source of Vitamins  $B_3$ ,  $B_6$  and E but very low or has no carotene (Sanders, 2003). Being an adequate source of healthy fat, peanut has been revealed to contains many key vitamins and minerals, an outstanding source of plant-based protein and fiber. Peanut can be processed into various products including, peanut butter, roasted, chocolate-coated and salted foods with different nutritional profiles and various health advantages. As reported by the world healthiest food (2019), a periodic journal published by George Mateijan Foundation, peanut and peanut butter has the capability of reduction in risks associated with cardiovascular disease by an estimated 21% as revealed in a randomized study using double-blind and cross-over study involving 22 subjects.

The periodic journal (George Mateijan Foundation) in April 2019 reported that apart from containing monounsaturated fat, peanuts has good quantity of other nutrients that promotes heart health as shown in numerous studies. According to the journal, when compared with the U. S, people in France have a lower risk of heart related illnesses because they consume diets that is low in fat, although peanuts contain resveratrol which is the phenolic antioxidant that is also present in red grapes and red wine which is said to be responsible for the French paradox (GMF, 2019).

# Vegetable Oil

Vegetable oils are triglyceride produced from seeds, fruits, or plant seedlings. According to Orsavov, Misurcova, Ambrozova, Vicha & Micek, (2015) vegetable oil is the end product of the combination of higher saturated triglycerides and unsaturated fatty acids. Oils are very concentrated source of energy and they improve the palatability and consistency of a food without increasing its bulk (Drewnowski & Almiron-Roig, 2010). The amounts eaten each day vary widely according to cooking patterns, availability and cost. It is difficult for a small child to eat enough food to cover his energy requirements unless his diet contains some fat or oil or fat rich food (Burgess & Glasauer, 2004). Before fat soluble vitamins could be
absorbed, fats and oil are necessary, they are also sources of essential fatty acids (National Research Council (US) Committee on Diet and Health RCCDH., 1989).

# Sucrose (Table Sugar)

Many plants and plant parts produce a common, naturally occurring carbohydrate known as sucrose. Sucrose is a disaccharide (consisting of glucose and fructose having a general formula of  $C_{12}H_{22}O_{11}$ ) which is easily hydrolyzed by enzyme sucrase present in intestinal juice. For human consumption, sucrose is often extracted and refined from either cane or beet sugar. It plays important functions as flavor enhancer and consumed all over the world to provide energy as well as palatability (Dekker, 1996).

# 2.16 Standard Ready to Use Supplementary Food (Plumpy sup corn formular)

Plumpy-Sup corn formula is a RUSF made from corn, milk and soy. It is specially designed and formulated for treatment and management of moderate acute malnutrition. Plumpy sup corn formular is developed or formulated to reinforce ready-to-use supplementary food's acceptability and programme adherence with specific focus on inhabiting where corn is a staple food and part of a daily diet. This increases availability of RUSF and enables scale up of programs and increase vulnerable population coverage and also to provide humanitarian organizations and programs with a new reference as an alternative within Nutriset product range.

The adoption of Plumpy-Sup corn formula is to prevent relapse into severe acute malnutrition of children who finish therapeutic nutrition programs. Because of the ingredients or constituents, it is recommended not to be given people who have allergic to soy or dairy products. However, it can be used in targeted Supplementary Feeding Programs ("targeted SFPs") or general supplementation ("blanket SFPs") (ACFI., 2011).

## Benefits

Using plumpy sup corn formular helps children with MAM to regain their normal nutritional status. The formulated diet contain elevated nutrient and energy required to complement the habitual diet for the management and treatment of MAM. It is high in vitamin and mineral content and also a lipid-based nutritional supplement that is habituated to the needs and requirement of moderate acute malnourished children. As a ready to use supplementary food, it is eaten without prior preparation which means, it can be used directly (Nutrivita., 2016).

#### Uses

Plumpy-Sup corn formula was designed to be directly eaten from the packaging material with no initial cooking or reconstitution. The supplementary food is packaged is such a way that it can be easily unlatched by gently tearing off one corner of the sachet. Children eats the food on their own without any assistance from their caregivers or anyone else. Suitable drinking water is recommended to always be available when a child is consuming Plumpy-Sup corn formula. The sachet can be kept throughout the day after opening. Pumply sup corn formular is meant to supplement the usual diet, hence should not replace the family food nor used in place of breast milk (Nutrivita., 2016).

It is employed in managing children with wasting from the age of 6 months and above. A sachet supplies a minimum of 75 kcal/kg body weight /day and should be continuously consumed until the target weight is attained. A sachet per day is recommended for the management of moderately acutely malnourished child (Nutrivita., 2016). It contains skimmed powder, vegetable oils (rapeseed, palm, soy in varying proportions), vitamin and mineral complex, sugar, defatted soy flour, stabilizer (fully hydrogenated vegetable fat, mono and diglycerides) and extruded corn flour (Nutrivita., 2016). Plumpy-Sup corn formula does contain no ingredients of animal origin besides dairy products and there is no presence any Genetically Modified Organism (GMO) (Nutrivita., 2016). It contains no ingredients of animal

origin besides dairy products.

Proximate			
Parameters	Minimum	Maximum	Target
Energy	520 kcal	550 kcal	537 kcal
Proteins	10.8 g	14.2 g	12.5 g
Carbohydrates	40.3 g	45.5 g	43 g
Dry skimmed milk protein	3.6 g	-	3.6 g
Lipids	34.1 g	35.9 g	35 g
-3 fatty acids	0.4 g (0.5% total	0.6 g (3% total	0.5 g (about 0.8%
	energy)	energy)	total energy)
-6 fatty acids	2.6 g (4.5% total	5 g (10% total	4.3 g (about 7.2%
	energy)	energy)	total energy)
Minerals			
Calcium	510 mg	770 mg	610 mg
Phosphorus	435 mg	580 mg	490 mg
of which free phosphorus*	400mg	560mg	425 mg
Potassium	765 mg	1200mg	945 mg
Magnesium	145mg	205 mg	170 mg
Zinc	10.2 mg	13.2 mg	11 mg
Copper	0.5 mg	0.85mg	0.7mg
Iron	9.4mg	15mg	12 mg
Iodine	78 µg	150 μg	116 µg
Selenium	18 µg	40 µg	20 µg
Manganese	0.5mg	0.85mg	0.65 mg
Sodium	-	270 mg	<180mg
Vitamins	1020	1(70	1000
Vitamin A	1020µg	1670 μg	1020 μg
Vitamin D	10.2 μg	33.6µg	10.2 μg
Vitamin E	15.3 mg	21.6mg	15.3 mg
Vitamin C	76.5mg	200mg	76.5mg
Vitamin BI	0.5 mg	0.9mg	0.5mg
Vitamin B2	2mg	2.4mg	2mg
Vitamin B6	Img	1.4mg	Img
Vitamin B12	2.6 μg	3.6 µg	2.6 μg
Vitamin K	26 μg	42 μg	26 μg
Biotin	10.2 μg	16 μg	10.2 μg
Folic acid	340 μg DFE	415 μg DFE	340 μg DFE
Pantothenic acid	2.6mg	5mg	2.6 mg
Niacin	12.8 mg	15 μg	12.8 mg

 Table 2.5: Nutritional Content for 100g of Plumpy Sup Corn Formular

Source: Nutrivita (2016)

### 2.17 Review of previous research on supplementary feeding

Karakochuk et al. (2012) managed moderate malnourished children with prepared RUSF. The effectiveness of locally formulated corn-soyabean based food was compared with the conventional RUSF in a randomised-cluster trial that was included in a conventional supplementary feeding program. The methodology of the study involved random allocation of two districts using blinded draw from a non-transparent bag and they received either corn-soybean based or RUSF. The study ran for 6 months and at the end of the research appraisal, it was discovered that the recovery rate of the conventional RUSF was superior compared to the locally formulated but that the cost and availability of the formulated corn-soya blend have a direct effect on the efficacy of supplementary feeding using locally available foods.

Lazzerini, Rubert and Pani (2013) evaluated how safe and efficacious were the diverse specially formulated foods use in managing moderately malnourished children in economically developing countries. The research was aimed at assessing the compliance of supplementary diets used in managing/treating MAM with specific nutrition composition as contained in the World Health organization technical specifications. Out of the eight randomised control trials that enrolled 10,037 children, seven was conducted in Africa and at the end of the trial, evidences that ranged from moderate to exceptional quality revealed the effecacy of both the LNS and blended foods in managing moderately malnourished children. And also in comparing the children that recovered using blended foods with that of lipid-based nutrient supplements (LNS), it was assumed that there was a pragmatically remarkable benefit in the later , this does not translate to reduction in mortality neither does it reduces the possibility of default or degeneration to SAM.

Rice, lentil and chickpea was previously used in Bangladesh as ingredients to develop a supplementary food (Ahmed et al., 2014). The planting and utilization of these ingredients produced and widely used in Bangladesh and other South Asian countries would make them available, accessible and affordable. Two different types of RUSF were locally formulated and developed, the first one was formulated from rice and lentil while the second one was chickpea based, other ingredients such as vitamin and mineral premix, sugar, dried skim milk powder and soybean oil were added to the two RUSFs. Different food processing techniques such as roasting, particle size reduction, homogenous blending and packaging were employed in their production and fed to children. However, their caregivers accepted the two formulated RUST despite their production from locally available food ingredients which is seen as new way of ensuring that children's diets are nutritional adequate. The acceptability was well pronounced most especially among those living in food insecure context, the study went further to show that the two diets were more adequate nutritionally when compared with Pushti packet (a standard supplementary food).

In South Africa, Steenkampa, Lateganb and Raubenheime (2015) studied the effectiveness of supplementation of 12-60 months MAM children with RUSF to measure their catch-up growth. Although the study was done in functional health system with sufficient human resources but it was a government program, so standard supplementary foods were used and it was targeted at a specific group of children suffering from MAM. It was observed and concluded that the time frame for the targeted supplementary feeding program did not benefit the children beyond six weeks, the period which was seen as been too short to achieve total recovery. As a result of this, there is no justification for governments provision of expensive supplement to MAM children resulting from food insecurity in households through food supplementation program. Sustainability of costly standard supplementary food attempted to be used for catch-up growth in children residing in food in-secured household has been impossible, the appropriated fund for such exercise should be channel to support community-based interventions such as local production of enriched supplementary food and increase household food production which would grant malnourished children more access.

Systemic review on the efficacy of supplementary feeding on the health of disadvantaged children and infants was carried by Kristjansson et al in 2016. From their findings there was small but statistically significant positive effects on weight and height of young children in economically developing countries when supplementary food was provided, there was as little as 0.12 kg weight gain and the height increased by 0.32 cm. Other physical outcomes such as hemoglobin levels, height- and weight-for-age z scores showed there was positive effects. Their report also revealed some evidences on the improvement in psychomotor development of children fed with supplementary food in addition to the family food when compared to those who were fed on their habitual diets alone and not given additional food. Through the review, the supplementary food showed more efficacy in <2 years old children of poor nutritiona or malnourishment status. Findings from the review also revealed that some of the supplementary foods supplied the better part of the recommended daily allowance for energy and because the feeding was supervised, it ensured the children ate the food as intended and greater effectiveness was recorded. An average of 36% of energy in the supplementary food were accessed by the children when the food was home-delivered or is in the form of takehome rations compared to 80% when the food was given at school to preschool children at school or day care centers, this was revealed through supervision and hence showed how important supervision could be in supplementary feeding program.

Medoua, Ntsama, Ndzana, Essa'a, Tsafack, & Dimodi, (2016) carried out a randomised control trial with the objective of testing the hypothesis that a satisfactory rates of recovery is possible when 50% of childs energy requirement is supplied by either CSB ++ or RUSF and the remaining 50% came from the family or household food. The randomised trial tend to study the rates of recovery of moderate acute malnourished children managed with RUSF or improved corn-soya blend (CSB+). The study enlisted eighty-one malnourished children whose ages ranges between 25 and 29 months and had their weight-for-height z score of -3 and

-2. The children were fed for 56 days, each enlisted child was given daily ration that supplied 40 kcal/kg body weight, home monitoring and follow-up visits was carried out every 14th day during which the caregiver received nutrition education and dietary counselling. Findings at the end of the study revealed that the children that received CSB had 73 % recovery from moderate acute malnutrition while RUSF recorded 85 % with no significant differences in the two treatments. RUSF group required mean treatment duration of 44 days while CSB++ acieved recovery within 51 days. They concluded the study by emphasizing that treating moderate acute malnutrition with the two different supplementary foods (CSB++ and RUSF) was successful relatively.

A past study (Isanak et al., 2019) from Mali, through cluster-randomised MAM treatment study, assessed the effficacy of cost of community-focused screening and MAM management, the study revealed the outcomes of four dietary supplements used in managing MAM during the trial. The formulated blended food (CSB++) was produced using micronutrient pre-mix, dehusked soybean flour, dried skimmed milk, maize flour, and soy oil, Plumpy sup (a conventional ready-to-use supplementary food), Misola, produced locally from micronutrient-fortified cereal-legume blend and locally milled flour was formulated using micronutrirnt powder mixed with millet, beans, oil and sugar, all the four dupplementary food were employed in the trial. Decision tree model was use to calculate the incremental costeffectiveness ratios (ICERs) of the supplementary foods by comparing their health and economic outcomes in estimating the long term outcomes. The outcome revealed that the risk of death in CSB ++, RUSF, MI and LMF was reduced by by 12.7%, 15.4%, 11.9% and 10.3%, respectively when the four treatment were compared with no MAM treatment. The result further showed that when RUSF treatment was compared with no MAM treatment, the incremental cost-effectiveness ratios (ICERs) of US \$9821 per death and US \$347 per day were averted.

In 2020, Das, Salam, Saeed, Kazmi & Bhuta (2020) reported that the existing data on RUSF suggested that there could be an improved recuperation and weight gain when compared with corn-soya blend (a locally formulated supplementary food) for MAM management. This was revealed from a study on the efficacy of the interventions for MAM management in <5 years in economically disadvantaged nations using systemic and meta-analysis method.

Cliffer et al., (2020) revealed that locally formulated supplementary foods are cost effective compared to the stadard supplementary food and considering these gaps, it is imperative that a supplementary diet from locally accessible ingredients be developed. This research is an attempt to come up with such quality nutritional supplementary food. It is designed to use cereal and legumes in the formulation of composite blends which is expected to provide the needed nutrients required in managing children with MAM. And also make the diet readily accessible and inexpensive to caregiver in both rural and urban centers and it can be used when supplies from government and development partners stops or delayed to prevent mortalities among under-five children as a result of malnutrition.

MUERSI

#### CHAPTER THREE

# MATERIALS AND METHODS

#### **3.1 Procurement of materials**

The yellow matured maize, pest-free soya bean and disease-free peanuts were procured from King's market in Ado Ekiti, Ekiti State, Nigeria while polyethylene was obtained from Agbeni/Ogunpa Market in Ibadan, Nigeria. The standard RUSF was procured from Nutriset, Malaunay, Northwestern, France.

### **3.2 Product preparation**

**Production procedure for soybean flour:** This is done according to the previously described method of Ihekonroye and Nogoddy (1985). After sorting and removal of the extraneous materials, the soybean was washed and soaked in clean water for 12 h at room temperature. After draining, it was blanched for 10min at 80°C for 10min, allowed to cool and dehusked, dried for 2 hr at 100°C in a hot air oven. Using hammer mill, the bean was dry-milled, and the flour packed in a polyethylene after sieving and then stored at room temperature prior to formulation.

**Production procedure for fermented maize flour:** Stones and other physical contaminants were removed from the maize grains and thereafter washed, using water. Fermentation was done by steeping in water (48 h), while the fermented maize was cleaned followed by wet milling using attrition mill. The slurry was sieved to remove the husk, allowed to settle overnight and thereafter decanted, packed in a clean jute bag and was dewatered by pressing, dried in an oven for 48-hr at 60<sup>o</sup>C, dried milled and packaged prior to formulation according to the previously described method of Awoyale et al. (2016).

**Processing method for peanut grit:** The disease-free peanuts was sorted for extraneous materials removal, washed and dried at room temperature (12 h). It was roasted winnowed and

blended into grits. It was thereafter packed and refrigerated before the formulation according to the previously described method of Ikeze et al. (2016).

Flow chart diagram of different ingredients used in preparing the locally formulated supplementary food is shown in Fig 3.1. The quantity of ingredients was calculated based on dry weight basis, homogenized using platinum mixer (HM990-SR7198111) (Platinum Machines, Sr. No. 34/1, Dagde Wasti, Maharashtra, India). The mixed products were then .90. products were kept a. weighed with Camry electronic kitchen scale model: EK5055, packaged (nylon) and sealed with QASA polyethylene sealing machine (QNS-3200H) (Qlink Corp, Dongguan City, Guangdong Province, China). The packaged products were kept at room temperature for the



Fig 3. 1: Flow chart for the production of soybean flour Ihekoronye & Ngoddy (1985)



# Fig 3.2: Flow chart for the production of fermented maize (ogi) flour.

# Awoyale, Maziya-Dixon & Menkir, 2016



# Fig 3.3: Flow chart for the production of groundnut grit

Modified method of Ikeze et al, 2016



Fig 3.4: Packaged MPN Flour

MINERS,



Fig 3.5: Packaged MSB Flour



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# Fig 3.6: Plumpy Sup corn formular

Source: www.nutriset.fr/product

# **3.3 Diet Formulation**

The technical specification for RUSF by World Food Program (WFP, 2016; 2018; 2020) stipulated that the protein content of supplementary food to manage MAM children is expected at 11 - 16 g/ 100 g of the finished product, hence this specification was followed in the formulation of maize –peanut grits flour (MPN) and maize-soybean flour.

# **Diet 1 = Plumpy Sup maize formula**

Plumpy sup maize formula is the conventional ready-to-use-therapeutic food used in supplementary feeding program for the management of children with MAM, it was used as a control for the feeding aspect of the research. It is packed in 100g per sachet and 12g protein, 35g fat and supplies 537 kcal of energy.

# **Diet 2 = Maize Flour + Peanut grits (MPN)**

The technical specification for the formulation ready-to-use supplementary food using the World Food Program (2016, 2018; 2020) stipulated a 16g protein from natural components and premix in the 100 g powder formulation of the foods. Initial protein contents of the peanut grits before the diet formulation from the peanut grit and maize are 35.15 and 3.8%, respectively. For instance, the peanut contributed 65% protein in the basal 16% protein of the composite flour (100 g) whereas the remaining 35% was from maize (i.e. 10.4 and 5.8% of the final 16% protein are from peanut grit and maize flour, respectively):

Peanut grits =  $\underline{16} \times 65 = 10.4\%$  (contributed protein from peanut grits) 100

Maize =  $\frac{16}{100}$  x 35 = 5.8% (contributed protein from maize flour) 100

In order to calculate the weight of each component making up the Maize-peanut composite flour

## **Peanut grits**

100g = 35.15g protein (protein content of the peanut grits)

Xg = 10.4g of protein (contributed protein from peanut grits)

 $Xg = 100 \times 10.4$  = 29.6 g (peanut grit in each sachet of MPN composite flour) 35.15

# Maize flour

100g = 3.8g protein (protein content of the maize flour)

Xg = 5.8g protein (contributed protein from maize flour)

 $Xg = \frac{100 \times 5.8}{3.8} = 152.6g$  (maize flour in each sachet of the MPN composite flour)

Total weight of MPN sachet = 182g (i.e., 29.6 g peanut grits + 152.6 g maize flour)

# **Diet 3 = Maize Flour + Soy-bean flour (MSB)**

The technical specification for the formulation RUSF using the World Food Program (2016, 2018; 2020) stipulated a 16g protein from natural components and premix in the 100 g powder formulation of the foods. Initial protein contents of the soybean grits before the diet formulation from the soybean grit and maize are 43.8 and 3.8% protein, respectively. For instance, the soybean contributed 65% of protein in the basal 16% protein content of the 100g composite flour while the remaining 35% was from maize (i.e. 10.4 and 5.8% of the final 16% protein are from soybean grit and maize flour, respectively):

In order to calculate the weight of each component making up the Maize-soybean composite flour

# **Soybean Flour**

 $\underline{65} \times \underline{16} = 10.4\%$  (contributed protein from soybean flour) 100 1

# Maize flour

 $\frac{35 \times 16}{100}$  = 5.8% (contributed protein from maize flour) 100

## Soy bean flour

100g = 43.8g protein

Xg = 10.4g of protein

 $Xg = 100 \times 10.4$  = 23.7g (soybean flour in each sachet of the MSB composite flour) 43.8

# Maize

100g = 3.8g protein

Xg = 5.6g

 $Xg = \frac{100 \times 5.6}{3.8} = 147.4g$  (maize flour in each sachet of the MSB composite flour)

Total weight of MSB sachet = 171.2g (i.e., 23.7g soybean flour + 147.4g maize flour)

### **3.4 Chemical Analysis**

## **Proximate composition**

### **Moisture determination**

The determination of the moisture content of the homogenized samples was done using the hot air oven method described by Association of Official Analytical Chemists (AOAC) procedure for hot air oven method of moisture determination (AOAC, 2010). This method employed drying of a food sample under a controlled temperature and pressure until when a constant weight was achieved. The nutrient content per weight basis of the samples are expressed through moisture content. The stainless steel oven dishes for placing the samples were washed, cleaned thoroughly and dried at 100 °C for 1 h in hot air oven until a constant weight is achieve. The stainless dishes were cooled in a desiccator and then weighed. Two grams of each of the samples was weighed into each dish and dried at 100 °C in the oven until constant weight was achieved. The dishes together with their contents were cooled in a desiccator and weighed. The cooling process in the desiccators was repeated before taking the final weight.

% Moisture Content = 
$$\frac{W_2 - W_3}{W_2 - W_3}$$
 x 100

Where

W1 = weight of dish

W2 = weight of dish + sample before drying

W3= weight of dish + sample after drying

### **Crude protein determination**

Crude protein content was determined by using the Kjeldahl method (AOAC, 2010). Two gram of each of the samples was weighed into a micro-Kjedahl flask. Five grams of anhydrous sodium sulphate and one gram of copper sulphate, a speck of selenium and 25 ml of concentrated sulphuric acid was added. The solution was heated in the fume chamber and shaken occasionally until the solution changes to green, the heating continue until the green colour disappears became clear. The digest (solution) was allowed to cool to room temperature and transferred into a 250 ml volumetric flask and made up of 100 ml with distilled water. During distillation process, the distillation unit was cleaned and the apparatus set up. Five ml of 2% boric acid with a few drops of methyl red indicator was introduced into a distillate collector (100 ml conical flask). The conical flask was placed under the condenser. Then 5ml of the sample digest was pipetted into the apparatus and washed down with distilled water. 5ml of 60% sodium hydroxide solution was added to the digest. The solution was heated until 100 ml of ammonium sulphate (distillate) was collected in the receiving flask. The receiving flask was removed and the content was titrated with 0.049 M H<sub>2</sub>SO<sub>4</sub> to a pink coloured end point. A blank with filter paper was subjected to the same procedure.

Calculation:

% Total Nitrogen =  $(titre-blank) \times Normality of acid \times N_2$ Weight of sample.

Nitrogen factor = 6.25

Crude protein = % total N x 6.25

#### **Determination crude fat**

The fat content of the samples was determined using Soxhlet extraction method (AOAC, 2010). A 500 ml capacity round bottom flask, which has been thoroughly washed and dried in oven for about 30 min at 105-110 °C and cooled in a desiccator. The flask was filled with 300 ml petroleum ether and fixed to the soxhlet extractor. Two gram of the sample was weighed into a labeled thimble. The extractor thimble was sealed with cotton wool. The soxhlet apparatus was set and allowed to reflux for about 6 hours. The thimble was removed with care and the petroleum ether on top of the container was collected until the flask is free of petroleum ether for reuse. The extract in the flask was removed and oven dried at 105 °C for 1 h, cooled in a desiccator and weighed.

Calculation:

% Fat =  $\frac{\text{Weight of } E1xtract}{\text{Weight of Fat Sample}}$  x  $\frac{100}{1}$ 

#### Ash content determination

Ash was determined according to AOAC (2000) procedure. Crucibles were washed thoroughly, dried in hot-air oven at 100 °C, cooled in a desiccator and weighed. Two gram of the sample was weighed into the crucible of known weight and put in a muffle furnace, heat was applied gradually until the temperature rise to 550 °C and the temperature was maintained for about 3 h until a white or grey ash was obtained. The crucible with its content was removed, cooled in a desiccator and weighed.

% ash = 
$$\frac{W_3 - W_1}{W_2 - W_1} \frac{100}{1}$$

Where: W<sub>1</sub> = Weight of dish W<sub>2</sub> = Weight of dish + sample before ashing W<sub>3</sub> = Weight of dish + sample after ashing

### Crude fibre determination

The determination of crude fibre was done using AOAC procedure (AOAC, 2010). Three gram of the sample was weighed into a 50 ml beaker and fat was extracted with petroleum ether by stirring, settling and decanting three times. The extracted sample was air dried and transferred to a 600 ml dried beaker. Then 200 ml of 1.25% dilute H<sub>2</sub>SO<sub>4</sub> and few drops of anti-foaming agent were added to the beaker. The beaker was placed on digestion apparatus with pre-adjusted hot plate and boiled for 30 min, rotating the beaker periodically to keep the solids from adhering on the sides of the beaker. At the end of 30 min, the mixture was allowed to stand for 1 min and then filtered using a Buckner flask covered with a white calico cloth connected to vacuum pump. Without breaking suction, the filtered residue was washed with boiling water several times until the washing is no longer acidic and was returned into the same beaker using a wash bottle containing 200 ml of 1.25% NaOH solution. This was again boiled briskly for 30 min with similar precautions as before. After boiling for 30 min, it was allowed to stand for 1 min and then filtered immediately under suction. The residue was transferred into ash dish and dried at 100 °C to a constant weight. Incineration to ash was done at 600 °C for about 30 min, cooled in a desiccator and weighed. The difference to weight between oven dry weight and the weight after incineration was taken as the fibre content of the sample. This was expressed as a percentage weight of the original sample taken for analysis: % Crude fibre =  $\underline{\text{Oven dried sample} - \text{weight of sample incineration}} \times 100$ Weight of sample taken

### **Carbohydrate estimation**

The carbohydrate was obtained by difference as follows:

% Carbohydrate = 100 – (% Moisture +% Ash + % Protein +% Fat + % crude fibre)

## **Energy calculated**

Energy was calculated by the "Atwater factor". The value obtained for carbohydrate, fat and protein was used to calculate the calorific content value of the samples as expressed below: 17:37:17 (kJ/g) or 4:9:4 (kcal/g) the "Atwater factors" respectively (FAO, 2003).

Where,

Protein = Crude Protein x 17 KJ/g or 4 kcal/g

Fat = Fat x 37 KJ/g or 9 kcal/g

Carbohydrate = CHO x 17 KJ/g or 4 kcal/g

# **Mineral analysis**

# Determination of calcium (Ca) and magnesium (Mg)

Calcium and magnesium contents of the samples extract were carried out by versanate EDTA complexiometric titration used by James (1995) and described by AOAC (2010). Twenty millimeter of each extract was measured and dispersed into a conical flask and pinch doses of the masking agents (potassium cyanide, potassium ferocyanide, hydroxylamine hydrochloride) were added to it. Ammonia buffer (20 ml) was added to adjust the pH to 10.0. A pinch of the indicator Erichrome blank T was added and then the mixture was shook very well. It was titrated against 0.02 N EDTA solution, until the color changes from mauve to a permanent deep blue color. This titration gave a reading for combined concentration of Ca and Mg ions. This was as a result of  $Ca^{2+}$  and  $Mg^{2+}$  forming complexes at pH 10.0 with EDTA. A second titration was conducted to determine Ca alone. This was a repeat of the previous one with a slight change, in that 10% NaOH solution was used to raise the pH of the digest to 12.0 and then was titrated with 0.02N EDTA using selechrome dark blue as indicator in place of Erichrome blank T. At pH 12.0, the  $Ca^{2+}$  complexed with EDTA. A reagent blank was titrated to serve as control. The experiment was repeated two more times. The calcium and magnesium contents were calculated separately using the formula;

% Calcium or Magnesium =  $\frac{100}{W}$  x EW x N x  $\frac{V_f}{V_a}$  x T - B

Where:

W = weight of sample analyzedEW = Equivalent weightN = Normality of EDTA $V_{f} = Total volume of extract$  $V_{a} = Volume of extract titrated$ T = Titre value of sampleB = Titre value of blank

## Determination of potassium (K) and sodium (Na)

Flame photometry was used to determine the concentrations of potassium and sodium as described by James (1995). The instrument (photometer) was set up according to the manufactures instructions. The equipment was switched on and allowed to stay for about 10 min. The gas and air inlets were opened and the start knob was turned on, the equipment being self-igniting. After ignition, the flame was adjusted to a non-luminous (blue) flame. Standard potassium and sodium solutions were prepared separately and each was diluted to a concentration of 2, 4, 6, 8 and 10 ppm. The appropriate filter was selected i.e. for potassium and sodium. The highest concentration standard solution (10 ppm) was aspirated and its emission intensity adjusted to 100 units. Thereafter, starting with the least concentrated (2 ppm), each standard solution was aspirated and caused to spray over the nonluminous butane gas flame. The emission intensity was read directly on the instrument and readings were recorded. The sample digest was also aspirated and their readings recorded. The emission intensities of the standards were plotted against their concentrations to obtain a standard curve (calibration graph) for each element. Subsequently, the optical density emissions recorded from each of the samples was matched against those in the curve. Using the curve to extrapolate the quantity of each potassium and sodium ions in the sample, the experiment was repeated two more times to get a mean concentration. The concentration of the test mineral was calculated as follows;

K or Na mg/100g = 
$$\frac{100}{W} x \frac{1}{1000} x X x \frac{V_f x}{Va}$$
  
Where:

W = Weight of sample used X = Concentration (in ppm) from curve  $V_f$  = Total volume of extract  $V_a$  = Volume of the extract (digest) flame D = Dilution factor where applicable

### Determination of manganese (Mn), selenium (Se), iron (Fe), zinc (Zn), copper (Cu)

An atomic absorption spectrophotometer (AAS) was used to determine the concentration of the trace elements/metals as described by James (1995). The solution (digest) from the ash was used. A solution containing metal ions was aspirated into a flame in which they were converted to a free atom vapour. A monochromatic light source was directed through the flame and the amount of radiation of a specific energy absorbed by the solution was recorded. A calibration graph was then prepared for the element and from this, the amount of the element present in each sample was read. The final computation was based on their relationship

$$X = \frac{C \times V}{Weight of sample used}$$

Where:

X =content of minerals in the sample

C = Concentration of the metal from the sample detected by the AAS.

V= Volume of which the solution was diluted.

# **Phosphorus determination**

Phosphorus was determined by the Molybdovanadate method. Two milligram of dry ash digest of sample was dispersed into a 50 ml volumetric flask. The same volume of distilled water and standard P solution were measure into different flask to serve as reagent blank and standard respectively. Two millimeter of phosphorus colour reagent (Molybdo vanadate solution) was added to each of the flasks and allowed to stand at room temperature for 15 min. The content of the flask was diluted to the 50 ml mark with distilled water and is absorbance was measured in spectrophotometer at a wavelength of 540 nm with the reagent blank at zero. The phosphorus content was calculated using the formula:

$$X = \frac{100}{W} x \frac{A_u}{A_s} x C x \frac{V_t}{V_a}$$

Where:

X= Content of phosphorus in the sample, mg/100 g.
W= Weight of ashed sample
Au = Absorbance of standard phosphorus solution.
As= Absorbance of standard phosphorus solution
C = Concentration of standard phosphorus solution.
Vt = Total volume of extract

Va = Volume of extract analyzed.

# **Iodine determination**

Iodine in the samples was determined trimetrically using the method described by Kirk and Sawyer, 1991. The iodine in the samples was oxidized under acidic condition and the free iodine is titrated with thiosulphate. About 5 gram of the sample was mixed with 100ml distilled water in a flask and shacked vigorously. It was made faintly acidic to methyl orange colouration and 1ml of saturated bromine water was added to it. A few glass beads were added and it was boiled for 5 min. It was cooked and 2 ml of molar HCl solution was added to it followed by 0.2 g potassium iodide. It was mixed well and titrated against 0.005m sodium thiosulphate solution using 1% starch solution as indicator. The iodine solution was calculated using the relationship that:

1ml of 0.005m Na<sub>2</sub>S<sub>2</sub>O<sub>3</sub>

# Vitamin analysis

# Determination of thiamine (vitamin B1)

Thiamine was determined using the method described by Okwu & Josiah (2006) and adopted by Nwaoguikpe et al. (2012). Five grams of each sample was homogenized with 50 ml of 2 M Ethanolic Sodium hydroxide (EtNaOH). This was filtered into a 100 ml flask. 10 ml of the filtrate was precipitated and color developed by the addition of 10 ml of 5% Potassium dichromate ( $K_2Cr_2O_7$ ) solution in a flask and absorbance of the sample was measured at 360 nm using Spectrum lab 752s spectrophotometer (Shanghai Xunda Medical Instrument Co., Ltd. Shanghai, China). A blank sample was prepared and the color also developed to calibrate the instrument to zero and read at the same wavelength. The thiamine content was calculated using the formula:

Thiamine (mg/100 g) =  $\frac{100}{W} x$  Au  $x \frac{C}{x} x$  Vf x  $\frac{D}{Va}$ 

Where:

W = Weight of sample analyzed

Au = Absorbance of standard thiamine solution

C = Concentration of standard thiamine solution

Vf = Total volume of filtrate

Va = Volume of filtrate analyzed

D = Dilution factor where applicable

# Determination of riboflavin (vitamin B<sub>2</sub>)

Riboflavin was determined using the method previously described and adopted methods (Okwu & Josiah, 2006; Nwaoguikpe et al., 2012), respectively. Five gram of each of the samples were extracted with 100 ml of 50% ethanol solution and shaken for 1 h. This was filtered into a 100 ml flask. Ten mills of the extract was pipetted into 50 ml volumetric flask. Ten mills of 5% KMnO<sub>4</sub> solution and 10 ml of 0.5% of H<sub>2</sub>O<sub>2</sub> were added and allowed to stand over a hot water bath for about 30 mins. 2 ml of 4% Sodium sulphate (NaSO<sub>4</sub>) solution was added. This was made up to 50 ml mark and the absorbance measured at 510 nm using Spectrum Lab 752s (Shanghai Xunda Medical Instrument Co., Ltd. Shanghai, China) and calculated as:

Riboflavin (mg/100 g) =  $\frac{100}{W}$  x Au x  $\frac{C}{As}$  x Vf x  $\frac{D}{Va}$ 

Where:

W = Weight of sample analyzed

Au = Absorbance of standard thiamine solution

C = Concentration of standard thiamine solution

Vf = Total volume of filtrate

Va = Volume of filtrate analyzed

D = Dilution factor where applicable

# **Determination of niacin (vitamin B3)**

Niacin was determined using the method described by Okwu and Josiah (2006). Five gram of the sample was treated with 50 ml of 1 N sulphuric acid (H<sub>2</sub>SO<sub>4</sub>) and shaken for 30 min. Three drops of ammonia solution were added to the sample and filtered. Ten mills of the filtrate was pipette into a 50 ml volumetric flask and 5 ml potassium cyanide was added. This was acidified with 5 ml of 0.02 N H<sub>2</sub>SO4 and absorbance measured in the spectrophotometer at 470 nm wavelengths. The reagent blank was used to calibrate the instrument at zero. The niacin content was calculated using the formula:

Niacin mg/100g = 
$$\frac{100}{W}$$
 x Au x  $\frac{C}{As}$  x Vf x  $\frac{D}{Va}$ 

Where:

W = Weight of sample analyzed

Au = Absorbance of standard thiamine solution

C = Concentration of standard thiamine solution

Vf = Total volume of filtrate

Va = Volume of filtrate analyzed

D = Dilution factor where applicable

# Determination of pyridoxine (vitamin B<sub>6</sub>)

This was determined using the colorimetric method described by Ashok et al (2011). Five gram of each sample was mixed with 50 ml of distilled water in a conical flask and agitated to effect solubilization of the vitamin. After 30 min, the mixture was filtered and the filtrate was used for the analysis. An aliquot of the extract (2 ml), was mixed with 1 ml of ammonia buffer solution and 1 ml of 20% sodium acetate solution was added to it and mixed well. Then 1 ml of dye solution (2, 6–dichloroquine chloride) was added to it. It was mixed very well. A standard solution of vit,  $B_6$  (Pyridoxine hydrochloride) was prepared and diluted to contain 0.005 mg/ml. 1ml of the standard solution was treated as described for the sample extract with 2 ml distilled water and was used as reagent blank. After mixing very well, the absorbance of the sample extract and the standard Vit  $B_6$  solution was read in a UV-visible spectrophometer at a wavelength of 650 nm. The reagent blank was used to set the instrument at zero. The formula below was used for the calculation.

Vitamin B6 ( $\mu g/100 g$ ) =  $\underline{100} x \underline{Au} x C x \underline{Vf} x D$ As Va

Where:

W = Weight of sample analyzed

 $Au = Absorbance of standard Vitamin B_6 solution$ 

 $C = Concentration of standard Vitamin B_6 solution$ 

Vf = Total volume of filtrate

Va = Volume of filtrate analyzed

D = Dilution factor where applicable

# **Determination of biotin (vitamin B7)**

Biotin was determined using the method described by Gill et al. (2018). Two grams (2g) of the sample was weighed into a 50 ml disposal centrifuge tube, was made to 25ml with water and vortexed for 30 s. Three mills of ascorbate solution, 500  $\mu$ l of stable isotope-labeled internal standard and 20 ml of acetate buffer was added to the sample and vortexed for 30s. The centrifuge tube was placed in a horizontal shaker for 5 min and then autoclaved at 121 °C for 30 min. The centrifuging tube was removed from the autoclave and was cooled to room temperature in a cold water bath, it was vortex mixed and placed in a centrifuge at 2700  $\times$  g for 5 min and the extract will be filtered through a syringe filter into an HPLC vial. A linear calibration plot was constructed by plotting the ratio of peak areas against ratio of

concentrations. The concentrations of biotin  $\mu g/dL$  was calculated using the following equations:

Vitamin ( $\mu g/dL$ ) =  $\underline{A_{NV}}_{A_{IS}} \times \underline{1} \times \underline{(CIS \times VIS)}_{M_S} \times \underline{100}_{1000}$ 

Vitaminconcentration of biotinor folic acid in sample (µg/dL);

A<sub>NV</sub> peak area of biotinor folic acid in sample;

A<sub>IS</sub> peak area of 2H4-biotinor 13C5-folic acidin sample;

L slope of the calibration curve

C<sub>IS</sub> concentration of 2H4-biotinor 13C5-folic acidin internal standard (ng/mL)

V<sub>IS</sub> volume of internal standard spiked to sample (mL);

M<sub>S</sub> mass of sample (g);

V<sub>S</sub> volume of sample (g);

1000 concentration conversion factor (ng/g to  $\mu$ g/g);

100 mass conversion factor ( $\mu g/g$  to  $\mu g/hg$ ).

# Folic acid determination (Vitamin B<sub>9</sub>)

The folic acid content of the samples was determined using spectrophotometric method described by Kumar et al (2011). Briefly, 1 g of the sample solution was prepared dissolving it in 100 ml of distilled water. 2 ml of the standard and sample solution was taken in marked test tubes. In each test tube 2 ml of 0.02% potassium permanganate solution, 2 ml of 2% sodium nitrate solution, 2 ml 4 N HCl solution, 1 ml of 5% ammonium sulphamate solution and 1 ml dye solution (0.1% N, N diethyl aniline solution in isopropyl alcohol) were and mixed well and kept at room temperature. The absorbance reading was taken at 535 mn against the blank Calculation:

Vitamin B<sub>9</sub> (mg/100 g) = <u>Absorbance</u> x C Slope

Where C is the concentration of the standard.

# Determination of cobalamine (vitamin B<sub>12</sub>)

The colorimetric method described by Ashok et al (2011) was employed. Five grams of the samples was mixed with 50 ml of distilled water and shaken well. It was allowed to stand

for 30 min at room temperature with thorough agitation before it was filtered through whatman No 1 filter paper. The resulting filtrate (extract) was used for the assay. The extract was treated with 1.25 g of diabasic sodium phosphate crystals and shaken to dissolve. This was followed by the addition of 1.1 g of anhydrous citric acid and 1.0 g of sodium matabisulphite and the volume was made up to 100 ml with distilled water. The mixture was heated under pressure at 121  $^{O}$ C for 10 min, filtered through Whitman No 1 filter paper and the absorbance was measured at 530 nm wavelength. Meanwhile, a standard solution of the vitamin B<sub>12</sub>was prepared and diluted to contain 0.05 µg/ml. 1ml of it was treated with the reagent blank was set up. The absorbance of the sample and standard solution was read at 530 nm and the reagent blank was used to set the instrument at zero. The formula below was used to calculate the Vitamin B<sub>12</sub> content.

Vitamin B<sub>12</sub> (mg/100 g) =  $\frac{100}{W}$  x  $\frac{Au}{As}$  x C x  $\frac{Vf}{Va}$  x D

Where:

W = Weight of sample analyzed

Au = Absorbance of standard Vitamin B12 solution

C = Concentration of standard Vitamin B12 solution

Vf = Total volume of filtrate

Va = Volume of filtrate analyzed

D = Dilution factor where applicable

## **Determination of Vitamin A (Carotenoid)**

The Vitamin A (Carotenoid) was done using the method described by Rodriguez-Amaya and Kimura (2004). The extraction was done with acetone using a mortal and pestle. Ten grams (10 g) of sample was weighed and crushed in mortar and 2 g of Hyflosupercel (celite) was added. Thirty mills of cold acetone (acetone refrigerated for about 2 h) to extract the carotenoid. The mixture was filtered with suction through a sintered glass funnel. The mortar, pestle, funnel, and residue were washed with 50 ml of acetone into the suction flask through the funnel. **Partition to Petroleum Ether**: Two hundred (200) ml of petroleum ether (PE) was poured into 500 ml separatory funnel with teflon stop-cock and acetone was added to each of the filterate. 300 ml of distilled water was slowly introduced to the walls of the separating funnel using wash bottles. The two phases were allowed to separate and the lower aqueous phase was discarded. The washing was done 4 times using 200 ml of distilled water each time to remove residual acetone and the Petroleum Ether phase was collected in a 50 ml volumetric flask which was made to pass through a small funnel containing 15 g anhydrous sodium sulfate to remove residual water (a glass wool plug was put to hold the sodium sulfate). The volume was made up to mark with petroleum ether and the absorbance was read at 450 nm using spectrophotometer. The total carotenoid content was calculated using the following formula:

Total carotenoid content ( $\mu g/g$ ) =  $\frac{A \times \text{volume (ml)} \times 10^4 \times \text{Df}}{A^{1\%}_{1\text{cm } X} \text{ sample weight (g)}}$ 

Where:

A= absorbance.

Volume = total volume of extract (50 or 25 mL).

 $A^{1\%}_{1cm}$  = absorption coefficient of  $\beta$ -carotene in PE (2592).

Df = Dilution factor.

Multiply by 100 to give the carotenoid content in  $\mu g/100$  g.

# **Determination of vitamin D**

The spectrophotometric method of the Association of Vitamin Chemists described by Kirk and Sawyer (1991) was employed. Five grams of each sample was saponified by boiling in alcoholic potassium hydroxide and reflux. The unsaponified portion was separated in a separating funnel and used for the determination. Accordingly, 10 ml of the unsaponified matter was treated with 9 ml of 20% (v/v) Antimony Chloride solution in chloroform containing 4 ml of acetyl chloride. Main while a standing Vitamin D solution (Cholecalciferol) was prepared and diluted to specific concentration. 1 ml of the standard Vitamin solution was treated as described for the sample. The absorbance of the standard solution and the samples

were read at 500nm while a reagent blank was used to set the instrument at zero. The formula below was used for the calculation.

Vitamin D (Cholecalciferol)  $(\mu g/100 \text{ g}) = \frac{100}{W} x \text{ Au } x \frac{C}{As} x \text{ Vf } x \frac{D}{Va}$ Where:

W = Weight of sample analyzed

Au = Absorbance of standard thiamine solution

C = Concentration of standard thiamine solution

Vf = Total volume of filtrate

Va = Volume of filtrate analyzsed

D = Dilution factor where applicable

### **Determination of vitamin E**

The spectrophotometric method of the Association of Vitamin Chemists described by Kirk and Sawyer (1991) was used. One gram of each test sample was mixed with q0ml of absolute alcoholic suphuric acid solution. The mixture was boiled under reflux, under reduced light (aluminum wrapped containers) for 45 min. 50 ml of distilled water was added and the mixture was transferred to a separation funnel using additional 50 ml of distilled water to washout. The unsaponified matter was extracted with 5 portions of 30 ml diethyl ether. The combined extract was washed free from acid by using several portions of distilled water and then dried over sodium sulphate in desiccator. Thereafter, the extract was evaporated until the solvent was gone. The extract was then re-dissolved in 10 ml of absolute ethanol. Standard vitamin E solution was prepared and diluted. Two mills of the extract solution and 2 ml of the standard solution as well as 2 ml of distilled water were dispensed into 3 different test tubes to serve as sample standard and blank respectively. Five mills of absolute alcohol was added to each test tube followed by 1ml of concentrated HNO<sub>3</sub> which was added with great caution. The test tubes were placed in a water bath at 90  $^{\circ}$ C until the alcohol boiled and boiling was allowed

for 3 min only. The volume of the content was measured at 470 nm with the blank at zero. The Vitamin E content was calculated as shown below:

Vitamin E (mg/100 g) =  $\frac{100}{W} x \frac{Au}{As} x C x \frac{Vf}{Va} x D$ 

Where:

W = Weight of sample analyzed

Au = Absorbance of standard Vitamin E solution

C = Concentration of standard Vitamin E solution

Vf = Total volume of filtrate

Va = Volume of filtrate analysed

D = Dilution factor where applicable

### Vitamin K determination

The Vitamin K content was determined using spectrophometric method described by Ashok and Kumar (2011). About 0.5 g of each sample was dissolved in 5 ml of distilled water and then extracted with 4x10 ml chloroform. The water layer was discarded while the chloroform layer was transferred to a clean dried conical flask passed through anhydrous sodium sulphate for the removal of water residue. It was made up to 50 ml in the flask. This extract was used for the assay. An aliquot (5ml) of the extract and 1 ml of standard Vitamin K solution were put in separate test tubes. A reagent blank was set up in a separate test tube as well. The content of each test tube was treated with 2 ml of 0.2% solution of 2, 4 di-nitrophenyl hydrazine in HCl and alcohol at ratio 5:1 (v/v). After mixing very well, the tubes were heated in a water bath until almost completely dry and were allowed to cool to room temperature. Then, 15 ml of alcoholic ammonia (1:1 v/v) solution was added to each and mixed well and their respective absorbance was measured at wavelength of 635 nm. The reagent blank was used to set the instrument to zero while the vitamin was obtained according from the equation as follows:

Vitamin K ( $\mu g/100 g$ ) =  $\frac{100}{W} x Au x \frac{C}{As} x Vf x \frac{D}{Va}$ 

Where:

W = Weight of sample analysed

Au = Absorbance of standard thiamine solution

C = Concentration of standard thiamine solution

Vf = Total volume of filtrate

Va = Volume of filtrate analysed

D = Dilution factor where applicable

# Vitamin C determination.

The Vitamin C content was determined using the method described by Okwu and Josiah, 2006. Ten grams (10 g) of the sample was weighed into an extraction tube and 50 ml of EDTA/TCA (2:1) extracting solution were mixed and the mixture shaken for 1hr. This was transferred into a centrifuge tube and centrifuged at 3000 rpm for about 20 min. It was transferred into a 50 ml volumetric flask and made up to 100 ml mark with the extracting solution. Twenty mills of the extract was pipette into a 250ml conical flask and 10ml of 30% starch indicator (Kl) and also 50ml of distilled water was added. This was followed by 2ml of 1% starch indicator. This was titrated against 0.01ml CuSO4 solution to get a dark end point.

Vitamin C (mg/100g) = 0.88 x  $\frac{100}{10}$  x  $\frac{V_F}{20}$  x  $\frac{T}{1}$ 

## Where:

 $V_{\rm f}$  = Volume of extract

T = Sample titre - blank tire

# Determination of amino acid profile in MSB and MPN Flours

The Amino Acid profile of the samples was determined using methods described by Benitez (1989). The known sample was dried to constant weight, defatted, hydrolyzed, evaporated in a rotary evaporator and loaded into the Applied Biosystems PTH Amino Acid Analyzer (Applied Biosystems, Inc. Foster City, California, USA). **Defatting Sample:** The sample was defatted using chloroform/methanol mixture of ratio 2:1. About 500 mg of the sample was put in extraction thimble and extracted for 15 h in soxhlet extraction apparatus (AOAC, 2006).

**Nitrogen Determination:** A small amount (1150 mg) of ground sample was weighed, wrapped in whatman filter paper (No.1) and put in the Kjelcihal digestion flask. Concentrated sulphuric acid (10 ml) was added. Catalyst mixture (0.5 g) containing sodium sulphate (Na<sub>2</sub>SO<sub>4</sub>), copper sulphate (CuSO<sub>4</sub>) and selenium oxide (SeO<sub>2</sub>) in the ratio of 10:5:1 was added into the flask to facilitate digestion. Six pieces of anti-bumping granules were added. The flask was then put in Kjeldhal digestion apparatus for 3 h until the liquid turned light green. The digested sample was cooled and diluted with distilled water to 100 ml in standard volumetric flask. Aliquot (10 m1) of the diluted solution with 10 ml of 45% sodium hydroxide was put into the Markham distillation apparatus and distilled into 10 ml of 2% boric acid containing 4 drops of bromocresol green/methyl red indicator until about 70 ml of distillate was collected. The distillate was then titrated with standardize 0.01 N hydrochloric acid to grey coloured end point. Percentage Nitrogen = (a-b) x 0.01 x 14 x V x 100

witrogen =  $(a-b) \ge 0.01 \ge 14 \ge 0.01$ W x C

Where:



**Sample hydrolysis**: A known weight (0.8264 g for MSB and 0.9804 g for MPN) of the defatted sample was weighed into glass ampoule. Seven mills of 6 N HCL was added and oxygen was expelled by passing nitrogen into the ampoule (this is to avoid possible oxidation of some amino acids during hydrolysis e.g methionine and cystine). The glass ampoule was then sealed

with Bunsen burner flame and put in an oven preset at  $105\pm5$  <sup>o</sup>C for 22 h. The ampoule was allowed to cool before broken open at the tip and the content was filtered to remove the humins. It should be noted that tryptophan is destroyed by 6 N HCL during hydrolysis. The filtrate was then evaporated to dryness using rotary evaporator. The residue was dissolved with 5 ml to acetate buffer (pH 2.0) and stored in plastic specimen bottles, which were kept in the freezer.

**Loading of the hydrolysate into analyzer**: The amount loaded was 60 ul. This was dispensed into the cartridge of the analyzer. The analyzer is designed to separate and analyze free acidic, neutral and basic amino acids of the hydrolysate.

**Method of Calculating Amino Acid Values:** An integrator attached to the analyzer calculated the peak area proportional to the concentration of each of the amino acids.

# Determination of Amino Acid Profile in the gruel Sample

Quantification of amino acids in the pap samples (MSB and MPN) were carried out using the method outlined by Wang & Cavins (1989) as described by AOAC (2006), with little modifications where necessary. Precise volume of 50cm3 was measured into 250ml quick fit round bottom flask and dried using rotary evaporator. A clean spatula was used to remove the dried sample and defatted in soxhlet extraction apparatus using chloroform methanol mixture (2:1). Known weight (1.000g for MSB and 1.234g for MPN) was hydrolyzed, filtered using non- absorbent cotton wool or glass wool.

**Nitrogen determination:** A small amount (200 mg) of grounded sample was weighed, wrapped in whatman filter paper (No.1) and put in the Kjeldhal digestion flask. Concentrated sulphuric acid (10 ml) was added. Catalyst mixture (0.5g) containing sodium sulphate (Na<sub>2</sub>SO<sub>4</sub>), copper sulphate (CuSO<sub>4</sub>) and selenium oxide (SeO<sub>2</sub>) in the ratio of 10:5:1 was added into the flask to facilitate digestion. Six pieces of anti-bumping granules were added while the flask was then put in Kjeldhal digestion apparatus for 3 h until the liquid turned light green. The digested sample was cooled and diluted with distilled water to 100ml in standard volumetric flask. Aliquot (10 m1) of the diluted solution with 10 ml of 45% sodium hydroxide
was put into the Markham distillation apparatus and distilled into 10ml of 2% boric acid containing 4 drops of bromocresol green/methyl red indicator until about 70 ml of distillate was collected. The distillate was then titrated with standardize 0.01 N hydrochloric acid to grey coloured end point.

Percentage Nitrogen =  $(a-b) \ge 0.01 \ge 14 \ge V \ge 100$ W \x C

Where:

a.	=	Titre value of the digested sample
b.	=	Titre value of blank sample
v.	=	Volume after dilution (100 ml)
W.	=	Weight of dried sample (mg)
C.	=	Aliquot of the sample used (5 ml)
14.	=	Nitrogen constant in mg.

**Hydrolysis of the sample**: A known weight (1.000 g for MSB and 1.234 g for MPN) of the defatted sample was weighed into glass ampoule. 7 ml of 6 N HCL was added and oxygen was expelled by passing nitrogen into the ampoule (this is to avoid possible oxidation of some amino acids during hydrolysis e.g methionine and cystine). The glass ampoule was then sealed with Bunsen burner flame and put in an oven preset at  $105\pm5$  °C for 22 h. The ampoule was allowed to cool before broken open at the tip and the content was filtered to remove the humins. It should be noted that tryptophan is destroyed by 6 N HCL during hydrolysis. The filtrate was then evaporated to dryness using rotary evaporator. The residue was dissolved with 5 ml to acetate buffer (pH 2.0) and stored in eppendorf plastic specimen bottles, which were kept in the freezer.

**Loading of the hydrolysate into analyzer**: The amount loaded was 60 uL. This was dispensed into the cartridge of the analyzer. The analyzer is designed to separate and analyze free acidic, neutral and basic amino acids of the hydrolysate.

**Method of Calculating Amino Acid Values:** An integrator attached to the analyzer calculated the peak area proportional to the concentration of each of the amino acids.

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#### **Determination of Tryptophan in the Liquid Sample**

Tryptophan is a difficult amino acid to determine in proteins and peptides because it chemically decomposes during acid hydrolysis, hence it was determined separately. It should be noted that tryptophan is destroyed by 6N HCL during hydrolysis. Antioxidants such as thioglycolic acid or dodecanethiol have been used to preserve tryptophan. Alkaline hydrolysis has also been studied and was shown to produce higher tryptophan recovery than acid hydrolysis. The addition of phenol has also been reported. Alkaline hydrolysis was improved by using sodium hydroxide (NaOH) instead of barium hydroxide to prevent problems with both precipitation and adsorption of tryptophan.

**Determination:** The tryptophan in the known sample was hydrolyzed with 4.2 M Sodium hydroxide (Maria et al. 2004). The known sample was dried to constant weight, defatted, hydrolyzed, evaporated in a rotary evaporator and loaded into the Applied Biosystems PTH Amino Acid Analyzer. Quantification of amino acids of the pap samples were carried out using the method outlined by Wang and Cavins (1989), with little modifications where necessary. Precise volume of 50 cm<sup>3</sup> was measured into 250 ml quick fit round bottom flask and dried using rotary evaporator. A clean spatula was used to remove the dried sample and defatted in soxhlet extraction apparatus using chloroform methanol mixture (2:1). The known weights of gruelsta (1.000 g for MSB and 1.234 g for MPN) and flours (0.8264 g for MSB and 0.9804 g for MPN) were hydrolyzed, filtered through non-absorbent cotton wool or glass wool.

**Nitrogen Determination**: Nitrogen content was determined using kjeldahl method. A small amount (200 mg) of ground sample was weighed, wrapped in whatman filter paper (No.1) and put in the Kjeldhal digestion flask. Concentrated suphuric acid (10 ml) was added. Catalyst mixture (0.5 g) containing sodium sulphate (Na<sub>2</sub>SO<sub>4</sub>), copper sulphate (CuSO<sub>4</sub>) and selenium oxide (SeO<sub>2</sub>) in the ration of 10:5:1 was added into the flask to facilitate digestion. Four pieces of anti-bumping granules were added. The flask was then put in Kjeldhal digestion apparatus for 3 h until the liquid turned light green. The digested sample was cooled and diluted with

distilled water to 100 ml in standard volumetric flask. Aliquot (10 ml) of the diluted solution with 10ml of 45% sodium hydroxide was put into the Markham distillation apparatus and distilled into 10 ml of 2% boric acid containing 4 drops of bromocresol green/methyl red indicator until about 70ml of distillate was collected. The distillate was then titrated with standardize 0.01 N hydrochloric acid to grey coloured end point, while the percentage nitrogen in the original sample was calculated using the formula:

Percentage Nitrogen =  $(\underline{a} - \underline{b}) \ge 0.01 \ge 14 \ge 0.01 =$ 

Where:

a = Titre value of the digested sample

b = Titre value of blank sample

v = Volume after dilution (100ml)

w = Weight of dried sample (mg)

c = Aliquot of the sample used (10ml)

14 = Nitrogen constant in mg

# Anti-nutritional Analysis Saponin determination

The saponin content of the sample was determined by modified double extraction gravimetric method described by Obadoni & Ochuko (2002) and Onwuka (2005). Briefly, five grams (5 g) of the dried sample was mixed with 50ml of 20% aqeous ethanol soultion in a conical flask. The mixture was heated with periodic agitation in water bath for 4hours at 55  $^{\circ}$ C, it was then filtered through whatman filter paper (No 42) and the residue was further etracted with 50 ml of 20% ethanol. The combined extract was evaporated on a hot water bath at about 90  $^{\circ}$ C until 40ml volume was achieved. Diethyl ether (40 ml) was added to the concentrate in 250 ml separator funnel followed by vigorous shaking. The aqueous layer was recovered while the ether layer was discarded. The purification process was repeated. N-butanol (60 ml) was

added and washed twice with 10 mL of 5% aqueous sodium chloride. The remaining solution was heated in a water bath after which the sample was dried in oven at 60 <sup>o</sup>C, weighed and saponin content was calculated as percentage of original sample thus:.

% Saponin = 
$$\frac{W_2 - W_1}{Wt \text{ of sample}}$$
 x  $\frac{100}{1}$ 

Where:

 $W_1$  = weight of evaporating dish

 $W_2$  = weight of dish + sample

#### **Tannin Determination**

Tannin content was determined by the Folis-Denis colorimetric method described by Kirk and Sawyer (1991) and Onwuka (2005). Sample (5 g) was dispersed in 50 mL of distilled water and shaken. The mixture was allowed to stand for 30 min at 28 °C, filtered through Whatman No 42 grade of filter paper. The extract (2 ml) was dispersed into a 50 mL volumetric flask. Also 2 ml standard tannin solution (tannic acid) and distilled water (2 ml) were added in separate volumetric flasks followed by addition of reagent to each flask to serve as standard. Saturated Na<sub>2</sub>CO<sub>3</sub> solution (2.5 ml) was added. The content of each flask was made up to 50 ml with distilled water and allowed to incubate at 28°C for 90 min. Their respective absorbance was measured in a spectrophotometer at 260 nm using the reagent blank to calibrate the instrument at zero.

## **Determination of Alkaloids**

Alkaloid content was determined gravimetrically by the method described by Harborne (1973) and Onwuka (2005). Five grams of the sample was dispersed in 20 ml of 10% acetic acid solution in ethanol to form a ratio of 1:10 (10%). The mixture was allowed to stand for 4 h at 28 °C. It was later filtered via Whatman no 42 grade of filter paper. The filtrate was concentrated to one quarter of its original volume by evaporation and treated with drop wise addition of concentrated aqueous NH<sub>4</sub>OH until the alkaloid was precipitated. The alkaloid

precipitated was received in a weighed filter paper, washed with 9% ammonia solution dried in the oven at 80 °C for 30 min cooled in a desiccator and reweighed. The Alkaloid content was calculated and expressed as a percentage of the weight of sample analyzed as shown below:

% Alkaloid =  $\underline{W_2 - W_1}$  x  $\underline{100}$ Weight of sample 1

Where:

 $W_1$  = weight of filter paper

W<sub>2</sub> = weight of filter paper + alkaloid precipitate

#### **Determination of Oxalate**

Oxalate content of the sample was determined by the method described by Onwuka (2005). Two grams of the sample was suspended in 190 ml distilled water in a 250 ml volumetric flask, followed by the addition of 10 mL of 6M HCl at 100 °C for 1 h. The digested sample was cooled and made up to 250 ml mark before filtration. Triplicate portion of 125 ml of the filtrate was measured into beakers and three drops of methyl red indicator was added. This was followed by the addition of conc. NH<sub>4</sub>OH solution until the test solution changes from salmon pink colour to a faint yellow colour. Each portion was heated again to 90 °C and 10 ml of 5% CaCl<sub>2</sub> solution was added while being stirred continuously. It was cooled and left overnight at 5 °C. The solution was centrifuged at 2500 rpm for 5 min. The supernatant was decanted and the precipitate completely dissolved in 10 ml of 20% H<sub>2</sub>SO<sub>4</sub> solution. The filtrate resulting from precipitation was made up to 300 ml. Filtrate (25 ml) was heated until near boiling point and then titrated against 0.05 M standardized KMnO<sub>4</sub> solutions to faint pink colour which persisted for 30 s. The oxalate content was calculated using the formula.

$$\frac{\% \text{ Alkaloid } = \text{T x (Vme) (DF) x 10^5}}{(\text{ME}) \text{ x Mf}} \qquad \text{mg/100 g}$$

Where:

T = Titre of KMnO4

Vme = the volume - mass equivalent (i.e 1cm3 of 0.05 MKMnO4 solution is equivalent to 0.00225g anhydrous oxalic acid).

DF = the dilution factor Vr/A (2.4 where Vr is the total volume of titrate (300ml) A = is the aliquot used (125ml) ME = the molar equivalent of KMnO4 in oxalate (KMnO4 redox reaction Mf = the mass of flour used

# Analysis of aflatoxin

The determination of aflatoxin was done using the ELISA method described by Leszczynska, Maslowska, Owczarek, and Kucharska (2001). Two grams of samples were weighed and was extracted and homogenised simultaneously for 10 min in a homogeniser with 6ml of methanol–water mixture (7:3 v/v) to separate aflatoxins and centrifuged (10 min at 3500 rpm) at room temperature. About 0.1 ml of the supernatant was diluted with 0.6 ml of phosphate buffer solution (pH 7.2) and the resultant solution was used for determinations. 0.1 ml of the examined sample solution and 0.05 ml aflatoxin-peroxidase conjugate and 0.05 ml of the mouse antibody solution against aflatoxin were added to each well of the plate used. The determination was repeated three times. The samples were incubated for 30 min at room temperature in the darkness. The free and peroxidase-combined aflatoxins compete for the combining site with antibodies to mouse antibodies immobilized on the plate. Each plate was emptied and washed five times with 5 ml phosphate buffer at pH = 7.2. Then, 0.05 ml of tetramethylbenzidine and 0.05 ml of urea peroxide were added to each tube, mixed and incubated again for 30 min in darkness. The reaction was terminated and their respective absorbance was measured at a wavelength of 450 nm, using an ELISA reading apparatus. The content of aflatoxins was calculated using the calculation below:

$$AFT (\mu g/kg) = \frac{1000}{W} x \qquad \frac{Au}{As} x C x \frac{Vf}{Va} x D$$

Where:

W = weight of sample

Au = Absorbance of sample extract

As = Absorbance of standard aflatoxin solution

 $C = Concentration (\mu g/ml)$  of standard aflatoxin solution

Vf = Total value of extract

Va = Volume of extract analyzed

D = Dilution factor where applicable.

#### Microbiological Analysis.

#### Coliforms, feacal coliforms and E. coli

This was examined microbiologically using the procedure described by Feng, Weagant, Grant and Burkhardt (2002).

**Presumptive Test**: Fifty grams (50 g) of each of the sample was weighed into sterile highspeed blender jar. Four hundred and fifty mililiters of butterfield's phosphate-buffered water was dded and blended for 2 min. The total volume in the blender jar should completely cover the blades. Decimal dilutions were prepared with sterile Butterfield's Phosphate diluent or equivalent. The number of dilutions that were prepared depended on anticipated coliform density. All the suspensions were shaken for 25 times in 30 cm arc. At least 3 consecutive dilutions were used and 1ml aliquots from each dilution was inoculated into 3 Lauryl tryptose broth tubes for a 3 tube Most Probable Number analysis. For better accuracy, a 1 ml or 5 ml pipet was used for inoculation. Pipet was held at an angle so that its lower edge rested against the tube. Care was taken so that not more than 15 minutes would elapse from the time the sample was blended and dilutions are inoculated in the media. Lauryl tryptose broth tubes was inoculated at 35±0.5 °C. The tubes was examined and reactions was recorded at 24±2 h for gas. Gas-negative tubes was re-incubated for an additional 24 h and examine and record reactions again at 48±3 h. Confirmatory test was carried out on all presumptive positive (gas) tubes.

**Confirmed test for coliforms**: From each gassing lactose broth tube, a loopful of suspension was transfer to a tube of Brillant Green Lactose Bile broth. The Brillant Green Lactose Bile tubes was incubated at  $35\pm0.5$  °C and was examined for gas production at  $48\pm3$  h. Most probable number (MPN) of coliforms based was calculated based on proportion of confirmed gassing Lauryl tryptose broth tubes for 3 consecutive dilutions.

**Confirmed test for faecal coliforms and** *E. coli*.: From each gassing Lauryl tryptose broth tube from the Presumptive test, a loopful of each suspension was transferred to a tube of *E. coli*, broth. EC tubes was incubated for  $24\pm 2$  h at 44.5 °C and will be examine for gas production. If the result is negative, the tubes was reincubate and re-examine again at  $48\pm 2$  h. The results of this test was used to calculate fecal coliform most probable number.

**Completed test for** *E. coli*.: From the confirmatory test for *E. coli*, each of the gassing EC tube was gently agitated and a loopful of broth was removed and streak for isolation on a Levine's eosin-methylene blue (L-EMB) agar plate and was incubated for 18-24 h at  $35\pm0.5$  °C. Plates was exaamined for suspicious *E. coli* colonies. Five suspicious colonies was gently transferred from each L-EMB plate to Plate Count Agar (PCA) slants and was incubated for 18-24 h at  $35\pm0.5$  °C and was used for further testing.

# Isolation and enumeration of Staphylococcus aureus

Isolation and enumeration of *Staphylococcus aureus* was done using using the procedure described by Bennett & Lancette (2001). For each dilution to be plated, 1ml sample suspension was aseptically transfer to 3 plates of Baird-Parker agar and 1ml of inoculum was equitably distributed to 3 plates. The inoculum was spread over the surface of the agar plate using sterile bent glass streaking rod. The plates was in an upright position until inoculum is absorbed by agar (about 10 min on properly dried plates). The plates was inverted and was incubated at between 45-48 h at 35-37 °C. Plates containing 20-200 colonies was selected, unless only plates at lower dilutions (>200 colonies) have colonies with typical appearance of *S. aureus*. Colonies of *S. aureus* was identified with the following features; circular, smooth, convex, moist, 2-3 mm in diameter on uncrowded plates, gray to jet-black, frequently with light-colored (off-white) margin, surrounded by opaque zone and frequently with an outer clear zone; colonies have buttery to gummy consistency when touched with inoculating needle.

**Count and record colonies**: Colonies which appeared to be *S. aureus* on selected plates was identified and number of colonies of each type was counted and recorded separately. The plates containing more than 200 colonies with the typical appearance of *S. aureus*, these plates was used for the enumeration of *S. aureus and* nontypical colonies was not counted. Colonies more than one were selected, counted and test for coagulase production. Colonies were added on triplicate plates represented by colonies giving positive coagulase test and multiply by the sample dilution factor. The number were reported as the number of *S. aureus*/g of food tested.

## Salmonella Determination

Salmonella was cultured and detected by using the method described by Andrew (1992).

**Sample Preparation**: Each sample was mixed to ensure homogeneity before withdrawing a 25 g analytical unit. Aseptically 25 g of the two samples was weighed into sterile 250 ml beaker. Using sterile glass funnel 25 g analytical unit was poured gently and slowly over surface of 225 ml lactose broth contained in sterile 500 ml Erlenmeyer flask. The container was allowed to stand undisturbed for  $60\pm5$  min and was incubated while keeping the cap loosely. The cap of the incubated samples was tightened and was gently shaken. One mill of the mixture was be transfer to 10ml selenite cystine (SC) broth and another 1 ml mixture to 10 ml tetrathionate (TT) broth. SC and TT broths will be incubated for  $24\pm2$  h at 35 °C. The incubated TT broth was mixed and 3 mm loopful were streaked on bismuth sulfite (BS) agar. Bismuth sulfite plates were prepared the day before streaking and store in dark at room temperature until streaked. The same process was repeated with 3 mm loopful of SC broth and was incubate plates  $24\pm2$  h at 35 °C.

**Examination for the Presence of Salmonella**: The plates of Bismuth Sulfite Agar were examined for the presence of colonies suspected to be Salmonella, typical Salmonella colonies may appear brown, gray, or black; sometimes they have a metallic sheen. Surrounding medium is usually brown at first, but may turn black in time with increased incubation, producing the

so-called halo effect. Some strains may produce green colonies with little or no darkening of surrounding medium.

**Confirmation using Rapid Agglutination Assay:** A suspect colony was taken from the Bismuth Sulfite Agar plate and was streaked on to a fresh blood agar plate and incubates to obtain a pure culture. One colony from the plate was taking and mixed in a test circle with a drop of test latex. Using a fresh loop, a second colony was mixed in a second test circle with a drop of the control latex. The mixing continued for 10–15 s. The cards were gently rocked in a circular motion for up to 2 min and observe for agglutination.

**Reading and Interpretation:** A result is positive when agglutination of the test latex occurred within 2 min, while a result is negative if no agglutination of either the test latex or the control latex occurred within 2 min.

# Yeast and Molds isolation and enumeration

Yeasts and molds were cultured and detected by using the method previously described by Tournas, Stack, Mislivec, Koch, and Bandler (2001).

**Sample Preparation:** Under aseptic condition, 25 g of the samples was weighed into a sterilized conical flask with glass stopper and 225 ml of 0.1 peptone water was added to achieve  $10^{-1}$  dilution, then mixture is transferred into a sterile blender jar, it was covered and blended at low speed for 1min. 1:10 (1+9) dilutions in 0.1% peptone water was obtained. A sterile pipette was used to transfer 1ml of the  $10^{-1}$  dilution to 99ml of sterile Phosphate buffered saline, the cap was replaced and was mixed by shaking it for 3min resulting in  $10^{-3}$ . Dilutions of  $10^{-6}$  should suffice. 1ml of the  $10^{-3}$  dilution was transferred to another sterile bottle of 99ml of sterile water and mixed to result in  $10^{-6}$  dilution.

**Plating and incubation of sample:** Aseptically 0.1 ml of each dilution was pipetted on prepoured, solidified Dichloran rose bengal chloramphenicol agar plates and the inoculum was spread with a sterile, bent glass rod. Each dilution was plated in triplicate, inverted and incubated at 27 °C for 5 days.

**Counting of plates:** After 5 days of incubation each plate was counted. Results were reported in colony forming units (CFU)/g or CFU/ml based on average count of triplicate set. Counts were round off to two significant figures.

# 3.5 Ethical clearance and informed consent

The consent for the research was secured from Ekiti State Ministry of Health, Ado – Ekiti. The caregivers of the children also gave their informed consent through the completion and signing of the consent form before the enlistment of their children in the study (annexure 9).

## 3.6 Preliminary visit

Permission was sought from the authorities of the study sites, Ekiti State Primary Health Care Development Agency and the Primary Health Care Departments of the six LGAs, with approvals EK/PHCDA/ADM/288/184 and EK/PHCDA/ADM/288/185 to all the six Local Government Areas before the commencement of the study. Most of the Primary Health Care facilities in Ekiti State have designated Nutrition Liaison Officers. This group of health workers was extremely useful and important in this study. There were preliminary visits to each of the LGAs with the approval letters from the State Primary Health Care Development Agency to the Coordinating Directors of Medical Services (CDMS), discussion was held with them (CDMS), Local Government Nutrition Focal Persons (LGNFPs) and the Facility Nutrition Liaison Officers (FNLOs) on the scope and methodology that were to be used in the research. The caregivers were met during one of their child welfare clinics and they were informed about the research and specific date was picked for the caregivers to bring their under-five children to the clinic for screening and thereafter children that fell within the scope of the research were enlistment and their consent and they all signed the consent form after the information in the form was carefully interpreted to them in Yoruba and Ekiti dialect.

### 3.7 Recruitment and training of Research Assistants.

Six Nutrition Officers from Ekiti State Primary Health Care Development Agency, Six Local Government Nutrition Focal Persons of the LGAs (LGNFPs) and 18 Facility Nutrition Liaison Officers (FNLOs), (one in each health facility per political ward, and in three political ward per Local Government Area in each of the six LGAs of Ekiti South Senatorial District) were recruited as research assistants for the study. There was 1-day training at the State and each LGA for the State Officers, LGNFPs and FNLOs respectively.

#### **3.8 Inclusion and Exclusion Criteria**

Children who were engaged in similar study concurrently or supplementary feeding program, have an incapacitating ailment (not including HIV or tuberculosis), or had a report of peanut or soybean sensitivity were excluded in the research. In addition, MAM children who have received treatment or management in one-month prior the presentation were excluded in order to concentrate the research essentially on the initial MAM treatment. However, children between 6 to 59 months, whose MUAC measurement were between  $\geq 11.5$ cm and  $\leq 12.5$  cm without bilateral pitting edema were included to the study.

# **3.9** Sample size determination

The study sample size was done *via* the following:

$$\mathbf{N} = \frac{\mathbf{4P} (\mathbf{1} - \mathbf{P})}{\mathbf{W}^2}$$

Where N = total number of MAM children required in the study.

 $\mathbf{P}$  = proportion of Children that have MAM in Ekiti State.

W = required precision level or probability level taken for this study (0.05 or 5%).

$$N = \frac{4 \times 3.5\% (1-3.5\%)}{0.05^2}$$

 $N = \frac{4 \times 3.5}{100} \times \frac{1-3.5}{100}$  $\frac{1-3.5}{100}$ 

$$N = \frac{0.14 \text{ x} (1-0.035)}{0.0025}$$
$$N = \frac{0.14 \text{ x} 0.965}{0.0025}$$
$$N = \frac{0.1351}{0.0025}$$

= 54.04

In order to make room for dropouts, the number was made up to 72 (30% increase). This percentage increase was purposively chosen to avert the danger of refusal by the caregivers to engage in the study. The number of MAM children per Local Government Area was calculated by simple proportion (16%), which were 12. This number was divided across the four treatments and in 3 political wards per LGA. The study sample size made use of 18 children for each treatment. There were four treatments namely: maize-soybean, maize-peanut, Plumpy Sup and Nutrition Education, using 72 children in total. One primary health care facility in each of the three political wards was used in each of the 6 LGAs.

#### 3.9 Selection of Subject

Experimental and cross-sectional sampling design were employed in this study. Experimental design involves a deliberate change in more than one operation variables (or factors) for the purpose of observing the effect, the switch has on more than one feedback variables (Crawford, 1997). The (statistical) design of experiments (*DOE*) is a systematic approach for planning experiments for data obtained to be analyzed in order to yield logical and unprejudiced conclusions (Setia, 2016). A cross-sectional study requires examining data generated from a population at a specified point in time. The participants involved in this kind of research are picked based on specific variables of interest (Kesmodel, 2018). Cross-sectional studies are empirical in nature and are descriptive research type, i.e. not informal or affinitive or comparative, which means that they can be used to determine the source of an ailment, for example, investigators documented the information that occurred in a population without manoeuvrering the variables (Kesmodel, 2018).

Descriptive study is a type of study that describes a community, circumstances, or occurrence that is being researched (Loeb, et al., 2007). It center attention on answering the how, what, when, and where questions of a study problem, rather than the why. Descriptive study is grouped into different types with respect to the type of approach that is used in administering such study (Mack, Woodsong, MacQueen, Guest & Namey, 2005). Its design involved four multistage sampling methods. Multistage sampling method involves the division of the population into groups or clusters for conducting research in order to make primary data collection more manageable (McCombes, 2019).

# **Stage 1: Selection of the Senatorial District**

The purposive sampling method was used in selecting the senatorial district where the study was carried out. South Senatorial district of Ekiti State was purposively selected because it was the senatorial district where the highest number of children with MAM was recorded during the Maternal Newborn and Child Health Week implemented in Ekiti State between 2015 and 2017. A purposive sample also referred to as a reproachful, deprecatory or expert sample, is a type of non-probability sample (Palinkas, et al., 2016). The core objective of a purposive sample is to generate a kind that can be reasonably assumed to be the population's representation (Dolores, 2007). This is often achieved by putting in expert knowledge of the population to pick randomly, a sample of elements that is a representative of a cross-section of the population.

### Stage 2: Selection of political wards/communities.

The research was executed in all the six local government areas (LGAs) that made up the senatorial district. All political wards in each LGA of the 6 LGAs were identified and the names of the political wards where the study took place were randomly selected from the poll of all the political wards in each of the 6 LGAs using a simple random sampling method. A community/health facility that is central to each of the selected political wards were picked as the venue for the screening.

#### Stage 3: Screening of Children with MAM

The children of 6-59 months in each of the 18 selected political wards, across the six LGAs in the senatorial districts of the State, were screened for MAM with Mid Upper Arm Circumference (MUAC) tape and anthropometric measurement.

MUAC is a rapid and easy way to establish whether a child is undernourished using a simple three - colored plastic strip. MUAC is appropriate to use on children from 1 year up to the age of 5 years. However, it is equally used for children who are over six months provided the length is above 65 cm. The MUAC of the children was determined by bending the left arm, (the left upper arm (a non-dominant arm that is assumed as the left one) is being used for MUAC measurement because its triceps and biceps muscle development is asymmetric unlike the right and presumed dominant arm with greater triceps and biceps muscles (Blackwell et al., 2015). This is done by using the tape to measure between the shoulder and elbow with marked mid-point while the arm was relaxed and allowed to hang down by the side of the body. Care was taking to ensure the tape did not pinch the arm nor left loosen up while taking the MUAC measurement. The measurement was thereafter read through the window of the tape and recorded to the nearest 0.1 cm. The measurement taken in the yellow section and 11.5 - 12.5 cm meant that the child is at risk of malnutrition. Children with MUAC measurement of 11.5 - 12.5 cm were thereafter recruited.

The anthropometric measurements of the children were taking using the United State Centre for Disease Control and Prevention's previously described method (CDC, 2007). The weights of participants were measured using electronic weighing scale with participants standing erect and where the children cannot stand, their weights were taking while sitting on the scale. Participants were put on light clothing with no shoes and socks before standing or sitting on the scale. Weights were taking in duplicate to  $\sim 0.1$  kg and recorded.

The height/length of participants was also measured at the time of enrolment in duplicate to the nearest 0.1 cm with a portable stadiometer or measuring board. Children under 24 months, 2 -3 years who could not stand and/or measure <30 inches were measured in recumbent position (lying down).

The board was covered with a clean paper drape/cloth and children's shoes, outer clothing, hair ornaments were removed while thick braids hair that could interfere with the measurement were loosened. The children were laid on their back in the center of the board with the head touching the fixed headboard, the body straight in line with the board and the shoulders and buttocks flat against the surface. Their heads were held gently against headboard with their eyes looking straight up. Gently, but firmly, the children's knees were pressed down with left hand to fully extend both legs, while using the right hand to slide the moveable foot board firmly against the sole of the feet ensuring that the toes point straight up. The measurements were read to the nearest 0.32 cm inch and the figures were recorded. The procedure was repeated. Where the children were uncooperative, the parents were allowed to try to calm the children before trying to re-measure. Two measurements were usually compared. Where they agreed within 0.61cm inch, the second reading was recorded. Where they did not agree within 0.61cm inch, the procedures were repeated until two measurements agree and the second measurements were recorded.

Height measurement was done for children who are 24 months and older who were able/willing to stand. As usual, the headboard surface was disinfected and thereafter hands were washed. Children who had thick socks, shoes, jackets or bulky clothing and any hair ornaments or braids on top of head were removed. The children stood with their back against the measuring surface of the standiometer, with their feet together flat on the floor, arms at side and knees and back straight. Their heads, heels, buttocks and shoulder blades touched the measuring surface. The headboard was gently slid down to the head, compressing the hair while the children looked straight ahead, with the headboard level and at right angles to the tape and the heels still flat against the floor. The height was measured to the ~0.32 cm inch, while their eyes were on the same level with the indicator, and the readings were recorded. A footstool was used to read the tape at eye level, where necessary. The children stepped away, and then the procedure was repeated completely. The two measurements were compared. Where they agree within 0.61cm inch, the second reading was recorded. Where they did not agree within 0.61cm inch, the procedure was repeated until two measurements agreed and then the second measurement was recorded. Length gain (in cm) was recorded over the entire duration (12 weeks) of the study. Using the WHO growth chart, children whose weight-for-height z score was between <-2 and  $\geq$ -3 with no bipedal edema were recruited for the study. Based on the MUAC measurement ( $\geq$  11.5 and  $\leq$  12.5cm) and weight – for – height z score (between <-2 and  $\geq$ -3), four (4) children with MAM were randomly selected from each of the eighteen (18) political wards across the six (6) LGAs in Ekiti South Senatorial District.

# Stage 4: Allocation of Children to groups

The 72 MAM children selected in stage 3 above were assigned to control and treatment groups by investigators who performed the anthropometric assessments, using investigatorblinded sampling method/technique. There are four treatment groups with the control (Plumpy sup which is the control, MPN, MSB and NE). A blinded-experiment is an evaluation that concealed the details concerning the test from the participant, in order to prevent bias, until after an experimental result is known (Day & Altman, 2000). Blind testing is employed wherever objects are to be compared without influences from testers' preferences or expectations (Day & Altman, 2000). Eighteen MAM children were eventually assigned into each of the four treatment groups including the control.

### 3.10 Preparation procedure of the formulated diets

Food demonstration session was organized for the caregivers of the selected children. According to World Food Program (2018), 50 g of flour is expected to be reconstituted with 200ml of water and cooked with 250 ml since there will be no evaporation. Each sachet of both MPN (182.2 g) and MSB (171.2 g) was a little less than 200 g, 700ml of clean water at room temperature was used to reconstitute a sachet or packed of the formulated supplementary food to form slurry. To ensure that the slurry does not have lumps, it was thoroughly mixed with spoon while 850ml of clean and boiled water was thereafter added slowly and continuously stirred on fire to be cooked for about 5 min. Eight gram (two tea spoons) of sugar and 8.4 g of vegetable oil, respectively were added for the energy content increase, fullness as well as taste improvement. The subsequent preparation was done at individual caregivers' household. The gruel was thereafter served with spoon.

# 3.11 Feeding and nutrition education of the selected moderate acute malnourished children

Three groups were fed with each of the three samples. Group one was the children managed with nutrition education/dietary counseling without feeding, groups two, three and four were fed with MPN, MSB and Plumpy sup respectively. The MAM children were managed under the four treatments for 90 days. Fourteen sachets of each of MPN, MSB and plumpy sup were given to each child under each treatment on bi-monthly basis and every child consumed one sachet per day for 90 days. The children were fed with the supplementary foods one sachet a day while supplementing with the family foods. Nutrition education on the frequency, adequacy, density, utilization and safety were given before the commencement of research and during the bi-monthly follow-up visits. Anthropometric measurements and MUAC of all the enlisted children were taken bi-monthly (every two weeks) throughout the duration of the research (90 days) to measure the effect of locally formulated supplementary

foods (MPN and MSB), the standard ready-to-use-supplementary food (plumpy sup) and nutrition education (NE) on the anthropometric indices and MUAC of the children.

# 3.12 Sensory evaluation

The formulated supplementary diets were subjected to sensory evaluation. The coded reconstituted samples (MPN and MSB) were cooked and allowed to cool to about 45 °C while the control (Plumpy Sup) was ready to eat. The gruel and the control (paste) were assessed for organoleptic properties and overall acceptability using ten (10) panelists selected among the caregivers (Nursing mothers) attending Child Welfare Clinic at Okeyinmi Comprehensive Health Centre (CHC), Ado-Ekiti. Prior to each evaluation, the caregivers were intimated with the task of the test. About 30 g each of the three samples (MSB, MPN and Plumpy Sup) were portioned in a coded plate for each caregiver. The caregivers were asked to rinse their mouths with clean water prior proceeding to the next food testing (Mekuria, Kinyuru, Mokua & Tenegashaw, 2021). The same caregivers were engaged in all the steps of the sensory evaluation, in order to obtain accurate data. Each caregiver was handed a questionnaire with the sample code, which they are expected to complete after tasting the samples and the identity of each sample was not revealed out to them. Each panelist was asked to evaluate for the sensory attributes (such as, appearance, aroma, taste, consistency and overall acceptability) to obtain information on their genuine opinion of the supplementary diets. Tasting of the gruels was carried out in the food demonstration shed of the CHC under broad daylight. The assessment was done using the 9-point Hedonic scale (1= disliked extremely, 2= disliked very much, 3= disliked moderately, 4= disliked lightly, 5= neither liked nor disliked, 6= liked slightly, 7=liked moderately, 8=liked very much, 9= liked extremely) as previously described (Stone & Sidel, 2004).

## **3.13 Data and statistical analysis**

Data from the treatment groups were screened and respondents who were absent on any visit were taken out. Anthropometric indices were calculated using WHO (2006) standards (ANTHRO version 3.2.2 January 2011) and recorded as weight-for-height (WHZ), weight-forage (WAZ) and height-for-age (HAZ) z-scores. All the data from the study were coded into the computer for descriptive and statistical analysis using Statistical Package for Social Science (SPSS) software (version 22 for windows). Data from chemical composition analysis were ле. ле meas compared using Analysis of Variance (ANOVA) while means were separated by turkey test at

#### **CHAPTER FOUR**

### RESULTS

#### 4.1 Proximate compositions of supplementary foods (flour)

Table 4.1 showed the proximate compositions of the formulated food (flour). The result revealed that protein (14.68%), ash (1.88%) and carbohydrate (70.69%) contents of maize-soybean (MSB) flour were significantly (p<0.05) higher than those of maize-peanut (MPN) flour (protein 10.80%, ash 1.68% and carbohydrate 65.03%). Meanwhile, the MPN flour has significant (p<0.05) higher moisture (7.21%), fat (8.89%), fiber (2.42%) and energy (384.14kcal) than MSB flour moisture, fat, fiber and energy (6.50%, 7.81%, 2.33 and 411.77kcal), respectively.

# 4.2 Proximate compositions of supplementary foods (gruel as consumed)

Table 4.2 showed the proximate composition and energy values of consumable grules supplementary diets. The moisture, crude protein, ash and carbohydrate (74.62, 7.59, 0.11 and 88.31%) contents of MSB gruel were significantly (p<0.05) higher than 69.11%, 7.19%, 0.10% and 87.63% of MPN gruel, respectively. However, the energy value, crude fibre, and crude fat obtained for MPN (422.39 Kcal, 0.28% and 4.79%) are significantly higher when compared to MSB (417.89 kcal, 0.17% and 3.81%), respectively.

	MSR FLOUR	MPN FLOUR			
-	MBD FLOOK	MINTLOOK			
Parameter	Mean ± SD	Mean ± SD	*Codex Std	t-value	p-value
Moisture (%)	$6.50\pm0.02$	$7.21\pm0.12$	<5	63.62	0.000***
Crude Protein (%)	$14.68 \pm 0.04$	$10.80\pm0.02$	15	166.43	0.000***
Crude Fat (%)	$7.81\pm0.12$	$8.98 \pm 0.10$	10-25	19.35	0.000***
Crude Fibre (%)	$2.33\pm0.04$	$2.42\pm0.02$	<5	3.78	0.019*
Ash (%)	$1.88 \pm 0.02$	$1.68 \pm 0.04$	<3	8.34	0.001**
Carbohydrate (%)	$70.69\pm0.07$	$65.03\pm0.12$	60-70	70.71	0.000***
Energy (Kcal)	411.77 ±0.15	$384.14\pm0.51$	400-425	11.28	0.000***

Table 4.1: Proximate composition (%) and energy values of supplementary foods (flour) per100g

Values are means  $\pm$  standard deviation of triplicates determination of samples. P < 0.05 is significant.

MSB = Maize – Soybean, MPN = Maize-peanut. \* CODEX CAC/GL08, \*P<0.05, \*\*P<0.01 \*\*\*P<0.001,

Table 4.2: Proximate composition (%)	) and energy values of formulated supplementary
foods (gruel as consumed) per 100g	

MSB GRUEL MPN GRUEL						
Parameter	Mean ± SD	Mean ± SD	t-value	p-value		
Moisture (%)	$74.62\pm0.02$	69.11 ± 0.01	523.0	0.000***		
Crude Protein (%)	$7.59\pm0.02$	$7.19\pm0.02$	22.3	0.000***		
Crude Fat (%)	$3.81\pm0.02$	$4.79\pm0.04$	-38.3	0.000***		
Crude Fibre (%)	$0.17\pm0.02$	$0.28\pm0.03$	-5.3	0.006**		
Ash (%)	$0.11\pm0.02$	$0.10\pm0.02$	0.4	0.692		
Carbohydrate (%)	$88.31\pm0.02$	$87.63\pm0.06$	-123.8	0.000***		
Energy (Kcal)	$417.89{\pm}0.08$	$422.05\pm0.22$	-194.6	0.000***		

Values are means  $\pm$  standard deviation of triplicates. determination of samples p < 0.05 is significant.

MSB = Maize – Soybean, MPN = Maize-peanut. \*P < 0.05 \*\* P < 0.01 \*\*\* P < 0.001

#### 4.3 Mineral contents of formulated supplementary foods (flour)

The mineral compositions of the formulated supplementary diets (flour sample) are presented in Table 4.3. The sodium (14.06 mg/100 g), phosphorus (111.95 mg/100 g), manganese (14. 06 mg/100 g) and iodine (508.33  $\mu$ g/100 g) of MSB flour were higher than the sodium, phosphorus, manganese and iodine (13.74, 107.74, 13.74 mg/100g and 303.0  $\mu$ g/100g, respectively) of MPN flour. Contrarily, the magnesium (10.06 mg/100g), iron (3.58 mg/100g, potassium (2.76 mg/100g) and zinc (4.22 mg/100g) contents of MPN flour are slightly higher compared to those of MSB flours (9.94, 3.56, 2.72 and 4.01 mg/100 g, respectively). MPN had higher Ca/K ratio (8.09 mg/100g) than MSB (6.07 mg/100g). The selenium contents of both flours (3.33 and 2.33 mg) were not significantly different after comparison.

# 4.4 Minerals contents of formulated supplementary foods (gruel as consumed)

The mineral compositions of the formulated supplementary foods (gruel as consumed) are presented in Table 4.4. The copper content of both gruels are <0.05 mg/100 g as well as the same potassium contents (2.7 mg/100 g). The iodine, calcium, phosphorus, zinc, iron and sodium contents of gruel from MSB (297.33  $\mu$ g/100 g, 14.15, 97.16, 3.98, 5.54 and 12.58 mg/100 g) were comparatively higher than MPN gruel (293.00  $\mu$ g/100 g, 11.81, 87.48, 3.13, 4.84 and10.87 mg/100 g), respectively. However, the manganese and magnesium contents (10.87 and 14.30 mg/100 g) of MPN gruel were significantly (p<0.05) higher than 9.99 and 11.10 mg/100 g obtained for MSB, respectively. Interestingly, the Ca/K ratio of 4.95 and 4.13 mg/100g was obtained for MSB and MPN gruels, respectively.

Minerals	MSB FLOUR	MPN FLOUR	*WHO	t-value	p-value
Sodium (mg)	14.06±0.00	13.74±0.00	500	166.7	0.000
Calcium (mg)	$16.51 \pm 0.00$	$22.34 \pm 0.00$	1000	-4843.1	0.000
Magnesium (mg)	$9.94 \pm 0.00$	$10.06 \pm 0.00$	280	-87.7	0.000
Iron (mg)	$3.56{\pm}0.00$	$3.58 \pm 0.00$	18	-10.2	0.001
Potassium (mg)	$2.72 \pm 0.00$	$2.76\pm0.00$	1500	-23.0	0.000
Zinc (mg)	$4.01{\pm}0.00$	$4.22 \pm 0.00$	20	-213.9	0.000
Phosphorus (mg)	$111.95{\pm}0.00$	$107.74 \pm 0.00$	850	2976.2	0.000
Iodine (µg)	508.33±1.16	$303.00{\pm}1.00$	150	232.8	0.000
Selenium (mg)	3.33±1.16	2.33±1.16	35	1.3	0.251
Copper (mg)	$0.04 \pm 0.00$	$0.03 \pm 0.00$	1	12.9	0.000
Manganese (mg)	$14.06 \pm 0.00$	$13.74 \pm 0.00$	1	215.9	0.000
Ca/K	$6.07 \pm 0.04$	8.09±0.03			

 Table 4.3: Mineral content (mg/100 g) of formulated supplementary foods (flour)

Values are means  $\pm$  standard deviation of triplicate determination of samples. P < 0.05 is significant.

MSB = Maize – Soybean, MPN = Maize-peanut, WHO, 2012

	MSB GRUEL	MPN GRUEL	*WFP		
Minerals	Mean ± SD	Mean ± SD		t-value	p-value
Sodium (mg)	12.58±0.00	$10.87 \pm 0.00$	270	1209.9	0.000
Calcium (mg)	$14.15 \pm 0.00$	$11.81 \pm 0.00$	535	1403.8	0.000
Magnesium (mg)	$11.10 \pm 0.00$	$14.30 \pm 0.00$	150	-2259.2	0.000
Iron (mg)	$5.54 \pm 0.00$	4.84-±0.00	10	479.5	0.000
Potassium (mg)	$2.86{\pm}0.00$	$2.86\pm0.00$	900	0.0	1.000
Zinc (mg)	$3.98{\pm}0.00$	3.13±0.00	11	526.0	0.000
Phosphorus (mg)	$97.16{\pm0.00}$	$87.48 \pm 0.02$	450	828.7	0.000
Iodine (µg)	297.33±1.16	293.00±2.00	100	3.2	0.031
Copper (mg)	$0.02\pm0.00$	$0.04 \pm 0.00$	1.4	-14.5	0.000
Manganese (mg)	9.99±0.00	$10.87 \pm 0.00$	1.2	-658.0	0.000
Ca/K	4.95±0.02	4.13±0.02			

# Table 4.4 Mineral contents (mg/100 g) of supplementary foods (gruel as consumed)

 $Values \ are \ means \pm standard \ deviation \ of \ triplicate \ determination \ of \ samples \ P < 0.05 \ is \ significant. \ MSB = Maize - 1000 \ model{eq:standard}$ 

Soybean, MPN = Maize-peanut, \*WFP, 2016

#### 4.5 Vitamin compositions (/100g) of formulated supplementary foods (flour)

Table 4.5 revealed the vitamin compositions of the formulated supplementary diets (flour). Vitamins  $B_1$  (2.87 mg),  $B_2$  (0.07 mg),  $B_3$  (0.15 mg),  $B_6$  (0.26 mg) and A (2.25 RE) of MSB flour were reportedly higher than those (2.49, 0.06, 0.10, 0.18 and 1.67 RE) of MPN flour, respectively. Meanwhile, the MPN flour had higher vitamins  $B_{12}$  (500.00 µg), E (1.72 mg) and C (62.05 mg) than those (393.3 µg, 0.62 and 56.02 mg) of MSB flour, respectively. Notably, both MSB and MPN flours had similarly comparable Vitamins  $B_2$ ,  $B_3$  and A.

# 4.6 Vitamin compositions of formulated supplementary foods (gruel as consumed)

Table 4.6 revealed the vitamin contents of the complementary foods as consumed. The MSB gruel had highest vitamins K (0.04  $\mu$ g) B<sub>9</sub> (300.00  $\mu$ g), B<sub>7</sub> (2.95 mg) and B<sub>1</sub> (2.30 mg), contents but lowest vitamins B<sub>3</sub> (0.08 mg) and B<sub>2</sub> (0.05 mg) when compared to MPN gruel (0.03 $\mu$ g, 150.00 $\mu$ g, 2.16mg, 2.22mg, 0.06 mg and 0.03 mg, respectively). However, vitamins C (45.37 mg), B<sub>12</sub> and E (190.00 and 1.98  $\mu$ g of MPN gruel are significantly (p<0.05) higher than 41.00 mg, 120.00  $\mu$ g and 0.86  $\mu$ g, respectively in MSB gruel.

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Vitamins	Mean ± SD	Mean ± SD	WHO	t-value	p-value
Vitamin B <sub>1</sub> (mg)	2.87±0.02	2.49±0.02	>1	21.7	0.000
Vitamin B <sub>2</sub> (mg)	$0.07 \pm 0.02$	0.06±0.03	>4	0.5	0.624
Vitamin B <sub>3</sub> (mg)	0.15±0.03	0.10±0.01	>25	3.3	0.028
Vitamin B <sub>6</sub> (mg)	0.26±0.03	0.18±0.02	>2	4.7	0.009
Vitamin B <sub>7</sub> (mg)	18.09±16.76	9.25±55.08	>20	118.5	0.000
Vitamin B <sub>9</sub> (µg)	396.67±25.17	263.00±50.86	>400	4.1	0.015
Vitamin $B_{12}(\mu g)$	393.33±11.54	500.00±36.06	>4	-4.9	0.008
Vitamin A (RE)	2.25±0.05	1.67±0.04	2000	16.2	0.000
Vitamin D(µg)	$0.52 \pm 0.02$	0.56±0.00	20	-17.2	0.000
Vitamin E(µg)	$0.62 \pm 0.02$	1.72±0.02	>30	-73.8	0.000
Vitamin K(µg)	3.20±2.31	1.89±3.06	>50	593.8	0.000
Vitamin C(Mg)	56.02±0.08	62.05±0.16	>150	-60.2	0.000

Table 4.5: Vitamin composition of formulated supplementary foods (flour)MSB FLOURMPN FLOUR

Values are means  $\pm$  standard deviation of triplicate determinations of samples P < 0.05 is significant. MSB = Maize – Soybean, MPN = Maize-peanut. WHO, 2012.

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	MSB GRUEL	MPN GRUEL	*WFP		
Vitamins	Mean ± SD	Mean ± SD		t-value	p-value
Vitamin B <sub>1</sub> (mg)	$2.30\pm0.02$	2.22±0.03	1.0	5.1	0.007
Vitamin B <sub>2</sub> (mg)	0.05±0.03	0.03 ±0.01	2.1	1.1	0.346
Vitamin B <sub>3</sub> (mg)	$0.08 \pm 0.02$	$0.06 \pm 0.03$	13	1.0	0.368
Vitamin B <sub>6</sub> (mg)	0.18 ±0.02	0.20±0.01	1.8	-1.6	0.184
Vitamin B7(mg)	2.95 ±0.08	2.16±0.04	60	20.9	0.000
Vitamin B <sub>9</sub> (µg)	300.00±26.46	150.00±17.32	330	8.2	0.001
Vitamin B <sub>12</sub> (µg)	120.00±20.00	190.00±30.00	2.7	-3.4	0.028
Vitamin A (RE)	1.24 ±0.05	$1.13 \pm 0.04$	550	3.4	0.031
Vitamin D(µg)	0.18±0.00	0.06 ±0,00	15	59.4	0.000
Vitamin E(µg)	$0.86 \pm 0.02$	1.98 ±0.02	16	-63.3	0.000
Vitamin K(µg)	0.04 ±2.52	0.03 ±2.31	900	9.5	0.001
Vitamin C(Mg)	41.17 ±0.28	45.37±0.12	60	-34.0	0.000

 Table 4.6: Vitamin contents (/100g) of supplementary foods (gruel as consumed)

Values are means  $\pm$  standard deviation of triplicate determinations of samples P < 0.05 is significant. MSB = Maize – Soybean, MPN = Maize-peanut, WFP, 2016

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### 4.7 Anti-nutrient and phytochemical (%) content of formulated supplementary flours

Table 4.7 showed the anti-nutrient contents of formulated supplementary foods (flour). There exists insignificant difference (p>0.05) in the oxalates found in MPN (1.53 mg) and MSB (1.48 mg) flour, respectively. The tannin and saponin contents (1.23 and 0.83 %, respectively) of MPN flour were significantly (p<0.05) higher than 1.12 and 0.74%, respectively obtained for MSB flour. However, the alkaloid content (0.55 mg) of the MSB flour was significantly higher (P<0.05) than 0.51 mg obtained for MPN flour.

# 4.8 Anti-nutrient and phytochemical content of the formulated supplementary foods

# (gruel as consumed)

Table 4.8 showed the anti-nutrient and phytochemical contents of the formulated supplementary foods in gruel form. There exists insignificant difference (p>0.05) in the tannin and saponin contents found in MPN (0.93 and 0.34%) and MSB (0.90 and 0.30%) gruels, respectively. In contrast, the alkaloid and oxalate contents of MSB gruel (0.12 and 0.24 mg) were higher than 0.10 and 0.11 mg reported for MPN gruel, respectively.

NERP

# Table 4.7: Anti-nutrient and phytochemical content of formulated supplementary foods (flour)

ANTI-NUTRIENTS	MSB FLOUR	MPN FLOUR		
PHYTOCHEMICAL	Mean ± SD	Mean ± SD	t-value	p-value
Tannin	1.12±0.02	1.23±0.02	-7.2	0.002
Saponin	0.74±0.02	0.83±0.03	-5.3	0.006
Oxalate	$1.48 \pm 0.57$	1.53±0.03	-02	0.880
Alkaloid	0.55±0.02	0.51±0.0	3.6	0.023

Values are means  $\pm$  standard deviation of triplicate determinations of samples P < 0.05 is significant.

MSB = Maize – Soybean, MPN = Maize-peanut.

# Table 4.8 Anti-nutrient and phytochemical content of the formulated supplementary

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# foods (gruel as consumed)

ANTI-NUTRIENTS/	MSB	MPN		
PHYTOCHEMICAL	Mean ± SD	Mean ± SD	t- value	p-value
Tannin (%)	0.90±0.03	0.93±0.02	-1.4	0.238
Saponin (%)	0.30±0.04	0.34±0.03	-1.4	0.241
Oxalate (mg)	0.24±0.02	0.11±0.01	10.5	0.000
Alkaloid (mg)	0.12±0.02	0.10±0.02	1.6	0.184

Values are means  $\pm$  standard deviation of triplicate determinations of samples P < 0.05 is significant.

MSB = Maize - Soybean, MPN = Maize-peanut.

#### 4.9 Amino acid (/100g protein) profile of the formulated supplementary foods (flour)

Table 4.9 showed the amino acid profile of the formulated supplementary diets (flour sample). Among the non-essential amino acids, the MSB flour has higher proline and tyrosine (4.98 and 3.10 mg, respectively) contents than those (4.87 and 2.24 mg, respectively) of MPN flour. Contrarily, the glycine (5.03mg), alanine (4.02 mg), serine (4.21 mg), aspartate (9.96), glutamate (15.29 mg), cysteine (1.21 mg) and arginine (8.26 mg) respectively present in MSB flour were lower than those (glycine (7.12 mg), alanine (4.55 mg), serine (4.59 mg), aspartate (10.30 mg), glutamate (16.80 mg), cysteine (1.82 mg) and arginine (10.32 mg) respectively) of MPN flour.

The mean total essential amino acid content of MSB flour (37.49mg) was significantly higher than that of MPN flour (28.42 mg). The valine (6.02 mg), histidine (3.00 mg), isoleucine (3.99 mg), leucine (7.18 mg), lysine (5.46 mg), methionine (2.99 mg), phenylalanine (4.43 mg) and tryptophan (1.42 mg) contents of MSB flour were higher than the contents in MPN flour (valine- 3.8 mg, histindine - 2.49 mg, isoleucine - 2.49 mg, leucine - 6.48 mg, lysine - 3.02 mg, methionine - 2.19 mg, phrnylalanine - 3.55 mg and tryptophan - 1.13 mg respectively).

# 4.10 Amino acid profile of the formulated supplementary foods (gruel as consumed)

The amino acid profiles of the formulated supplementary diets (gruel as consumed) are presented in Table 4.10. The mean total non-essential amino acid contents of MPN gruel (39.75mg/100g) was than MSB gruel (39.09mg/100g). The results of the non-essential amino acids of the gruel form further revealed that MSB had higher glycine, alanine, serine, proline, aspartate and tyrosine (3.61, 3.00, 3.00, 3.04, 7.19 and 2.06 mg/100g, respectively) content than gruel from MPN (3.21, 2.84, 2.43, 2.94, 6.21 and 1.55 mg/100g, respectively). In contrast, the gruel from MPN has higher glutamate, cysteine, and arginine (10.45, 0.97, and 7.23 mg/100g, respectively) than the gruel from MSB (9.31, 0.72, and 5.16mg/100g, respectively).

The mean total essential amino acid content was comparatively higher in MSB gruel (21.95mg/100g) than MPN gruel (16.11 mg/100 g). All the essential amino acid contents of MSB gruel; valine (4.21 mg), histidine (2.01 mg), threonine (2.05 mg), isoleucine (2.10 mg), leucine (4.73 mg), lysine (3.24 mg), methionine (1.92 mg), phenylalanine (2.75 mg) and .e.1. .jaanin 1.3 tryptophan (0.95 mg) respectively were higher than Valine 2.96, histidine 1.92, threonine 1.83, isoleucine 1.21, leucine 4.00, lysine 2.01, methionine 1.60, phenylalanine 1.69, and tryptophan

	MSB	MPN	*RDA
Amino acids	Mean ± SD	Mean ± SD	
Non-essential amino acids (NEAA)			
Alanine	$4.02 \pm 0.00$	4.55±0.00	-
Aspartate	9.96±0.00	10.30±0.00	-
Serine	4.21±0.00	4.59±0.00	-
Glutamate	$15.29 \pm 0.00$	16.80±0.00	-
Conditional essential amino acids (TCEA)			
Proline	4.98±0.00	4.870±0.00	-
Glycine	$5.03 \pm 0.00$	7.12±0.00	-
Arginine	8.26±0.00	$10.32 \pm 0.00$	
Cystine	1.21±0.00	1.82 ±0.00	2
Tyrosine	3.10±0.00	2.24±0.00	-
Essential Amino Acids (EAA)	RY		
Valine	$6.02 \pm 0.00$	3.80±0.00	3.5
Histidine	3.00±0.00	2.49±0.00	1.9
Threonine	3.00±0.00	3.27±0.00	3.4
Isoleucine	3.99±0.00	2.49±0.00	2.8
Leucine	$7.18 \pm 0.00$	$6.48 \pm 0.00$	6.60
Lysine	5.46±0.00	3.02±0.00	5.8
Methionine	$2.99 \pm 0.00$	2.19±0.00	2.2
Phenylalanine	4.43±0.00	3.55±0.00	2.8
Tryptophan	$1.42\pm0.00$	1.13±0.00	1.1
TEAA	37.49	28.42	33.9
TSAA (Meth + Cyst)	4.2	4.0	2.5
TArAA (Phenyl+Tyro)	7.53	5.79	6.3
ТАА	93.55	91.03	
ΣΤΕΑΑ / ΣΤΑΑ (%)	40.07	31.22	

 Table 4.9: Amino acid profile of the formulated supplementary foods (flour)

MSB = Maize – Soybean, MPN = Maize-peanut. TNEAA = Total Non-Essential Amino Acid, TEAA = Total Essential amino acid, TSAA = Total sulphur Amino Acid, TArAA = Total Aromatic Amino Acid, Source of RDA: FAO/WHO. 1991

	MSB	MPN	*RDA
Amino acids	Mean ± SD	Mean ± SD	
Non-essential amino acid			
Alanine	3.00±0.00	2.84±0.00	-
Aspartate	7.19±0.00	6.21±0.00	-
Serine	3.00±0.00	2.43±0.00	-
Glutamate	9.31±0.00	10.45±0.00	-1
Conditional essential amino acid			
Proline	3.04±0.00	2.94±0.00	
Glycine	3.61±0.00	3.21±0.00	-
Arginine	5.16±0.00	7.23±0.00	-
Tyrosine	2.06±0.00	1.55±0.00	-
Cystine	0.72±0.00	0.97±0.00	2.70
Essential amino acid			
Valine	4.21±0.00	2.96±0.00	4.30
Histidine	2.01±0.00	1.92±0.00	2.00
Threonine	2.05±0.00	1.83±0.00	3.10
Isoleucine	2.10±0.00	1.21±0.00	3.20
Leucine	4.73±0.00	4.00±0.00	6.60
Lysine	3.24±0.00	2.01±0.00	5.70
Methionine	1.92±0.00	1.60±0.00	2.70
Phenylalanine	2.75±0.00	1.69±0.00	5.20
Tryptophan	0.95±0.00	0.81±0.00	0.85
TEAA	23.96±0.00	18.03±0.00	
TSAA (Meth + Cyst)	2.64	2.57	2.5
TArAA (Phenyl+Tyro)	4.81	3.24	6.3
TAA	61.05	55.86	
ΣΤΕΑΑ / ΣΤΑΑ (%)	39.25	32.28	

Table 4.10: Amino acid profile of supplementary foods (gruel as consumed
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MSB = Maize – Soybean, MPN = Maize-peanut. TNEAA = Total Non-Essential Amino Acid, TEAA = Total Essential amino acid. TSAA = Total Sulphur Amino Acid, TArAA = Total Aromatic Amino Acid, Source of RDA: FAO/WHO. 1991

# **4.11:** Sensory attributes of the formulated supplementary foods (gruel as consumed) and the control (plumpy sup)

The sensory attributes of gruel samples from the formulated complementary food diets are shown in Table 4.11. The result revealed that plumpy sup was ranked the highest in all the sensory attributes with mean scores of colour ( $8.6\pm0.51$ ), flavor ( $8.5\pm0.52$ ), taste ( $8.5\pm0.52$ ), texture/mouth feel ( $8.5\pm0.52$ ), and general acceptance ( $8.5\pm0.52$ ). The MSB gruel was ranked highest and next to plumpy sup in flavour ( $6.7\pm1.76$ ), taste ( $7.2\pm0.92$ ), texture/mouth feel ( $8.2\pm0.78$ ) and general acceptance ( $7.4\pm1.71$ ) when compared to MPN gruel that had  $5.2\pm1.93$ for flavour,  $6.7\pm1.88$  for taste,  $6.3\pm2.00$  for texture/mouth feel and  $5.3\pm2.11$  for general acceptance, the differences were significant (p>0.05). However, the colour ( $7.7\pm0.94$ ) of MPN gruel was ranked better next to plumpy sup when compared with  $6.9\pm2.50$  for MSB gruel.

# 4.12 Microbial counts of the formulated supplementary foods (flours)

Table 4.12 showed the microbial counts in the supplementary foods (flour). The total count of yeast was higher in MSB flour ( $3.2 \times 10^8$  cfu/g) than MPN flour ( $2.1\times10^8$  cfu/g). While the total counts of *S. aureus* in MSB flour was 2.4  $\times10^8$ cfu/g while that of MPN was 2.3  $\times 10^8$  cfu/g. There was no observable growth of mould and coliform in both MSB and MPN flour samples. Although, no growth of salmonella was observed in MPN flour but it was isolated ( $1.0\times10^8$  cfu/g) in MSB flour. Likewise, no growth of *E. coli* was noticed in MSB flour but it was isolated ( $1.2\times10^8$  cfu/g) in MPN flour.

SENSORY	MSB Mean ±SD	MPN Mean ± SD	PLUMPY SUP Mean ± SD	p-value
ATTRIBUTES				
Colour	6.9±2.50	7.7±0.94	8.6±0.51	0.70
Flavour	6.7±1.76	5.2±1.93	8.5±0.52	0.68
Taste	7.2±0.92	6.7±1.88	8.5±0.52	0.56
Texture/ Mouth Feel	8.2±0.78	6.3±2.00	8.5±0.52	0.57
General Acceptance	7.4±1.71	5.3±2.11	8.5±0.52	0.71

 Table 4.11: Sensory evaluation of the formulated supplementary foods (gruel)

Values are means  $\pm$  standard deviation of triplicate determinations of samples. (P < 0.05 is significant. MSB = Maize – Soybean, MPN = Maize-peanut, Plumpy sup = control.

Table 4.12. Whet obtai counts of the formulated supplementary roous nour	Table 4.12: Microbial	counts of the	formulated	supplement	ary foods flou
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MICRO-	MSB	MPN	
ORGANISMS	Cfu/g	Cfu/g	
Yeast	$3.2 \times 10^8$	$2.1 \text{ x} 10^8$	
Mould	NG	NG	
E. coli	NG	$1.2 \ge 10^8$	
Salmonella	$1.0 \ge 10^8$	NG	
Coliform	NG	NG	
S. aureus	$2.4 \times 10^8$	$2.3 \times 10^8$	

 $\checkmark$ 

MSB = Maize – Soybean, MPN = Maize-peanut. NG = No growth

#### 4.13 Microbial counts of the formulated supplementary foods (gruel as consumed)

The results presented in Table 4.13 showed that yeast was isolated in MSB  $(1.4 \times 10^8 \text{ cfu/g})$  and MPN  $(4.2 \times 10^8 \text{ cfu/g})$  gruels, respectively. Similarly, mould was isolated in MSB  $(1.2 \times 10^8 \text{ cfu/g})$  and MPN  $(2.3 \times 10^8 \text{ cfu/g})$  gruels as well. However, no growths of *E. coli*, salmonella, coliform and *S. aureus* were detected in both MSB and MPN gruel samples.

#### 4.14 Aflatoxins content of supplementary flours

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Table 4.14 showed the aflatoxin contents of the formulated supplementary food diets from both flour samples. The result showed the mean scores of aflatoxin B<sub>1</sub> (AFB<sub>1</sub>) (1.52 µg/kg), aflatoxin B<sub>2</sub> (AFB<sub>2</sub>) (1.49 µg/kg), aflatoxin G<sub>1</sub> (AFG<sub>1</sub>) (1.48 µg/kg) and aflatoxin G<sub>2</sub> (AFG<sub>2</sub>) (1.50 µg/kg) respectively in MPN flour, were significantly higher than the mean scores in MSB flour (AFB<sub>1=</sub>0.87 µg/kg, AFB<sub>2=</sub> 0.88 µg/kg, AFG<sub>1=</sub> 0.93 µg/kg and AFG<sub>2 =</sub> 0.88 µg/kg respectively). The total aflatoxin content (5.99 µg/kg) of MPN flour was equally significantly (p<0.05) comparable to the total aflatoxin content (3.56 µg/kg) of MSB flour.

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MICRO-	MSB	MPN	
ORGANISMS	Cfu/g	Cfu/g	
Yeast	1.4 x 10 <sup>8</sup>	4.2 x 10 <sup>8</sup>	
Mold	$1.2 \ge 10^8$	2.3 x 10 <sup>8</sup>	
E. coli	NG	NG	
Salmonella	NG	NG	
Coliform	NG	NG	
S. aureus	NG	NG	AP

#### Table 4.13 Microbial counts of the supplementary foods (as consumed)

MSB = Maize – Soybean, MPN = Maize-peanut. NG = No growth.

Table 4.14: Aflatoxins content of supple	ementary foods (flour)

	MSB	MPN	*MP		
Aflatoxins	Mean ± SD	Mean ± SD		t-value	p-value
AFB <sub>1</sub>	0.87±0.00	$1.52 \pm 0.33$	<4	-22.5	0.000
AFB <sub>2</sub>	$0.88 \pm 0.00$	$1.49\pm0.00$	<4	-90.1	0.000
AFG1	$0.93\pm0.05$	$1.48 \pm 0.04$	<4	-15.3	0.000
AFG <sub>2</sub>	$0.88 \pm 0.02$	$1.50\pm0.00$	<4	-51.5	0.000
Total Aflatoxin	$3.56 \pm 0.54$	5.99 ±0.00	<4		

Values are means  $\pm$  standard deviations of triplicate determinations of samples, P < 0.05 is significant. MSB = Maize – Soybean, MPN = Maize-peanut, AFB<sub>1</sub> = Aflatoxin B<sub>1</sub>, AFB<sub>2</sub> = Aflatoxine B<sub>2</sub>, AFG<sub>1</sub> = Aflatoxin G<sub>1</sub>, AFG<sub>2</sub> = Aflatoxin G<sub>2</sub>. \* Maximum permitted aflatoxins B and G contents in food products ( $\mu g/kg$ ) according to Regulations (EC) No 1881/2006 and (EU) 165/2010

#### 4.15 Aflatoxin contents of supplementary foods (gruel as consumed)

The aflatoxin contents of the formulated complementary food diets from both gruels are presented in Table 4.15. The result showed no significant difference in the aflatoxin contents of both samples. However, MPN gruel had the same quantity  $(0.05\mu g/kg)$  of AFB<sub>2</sub>, AFG<sub>1</sub> and AFG<sub>2</sub> except for the quantity of AFB<sub>1</sub> (0.18 $\mu$ g/kg). Interestingly, the same quantity (0.04  $\mu$ g/kg) of aflatoxin AFB<sub>1</sub>, AFB<sub>2</sub>, and AFG<sub>1</sub> were found in MBS gruel except for the quantity of AFG<sub>2</sub> (0.05  $\mu$ g/kg).

# 4.16 Effect of the formulated supplementary foods on the anthropometric indices of the children

Table 4.16 shows the effect of the formulated supplementary diets on the anthropometric indices of the children in the study area. The result showed that the weight increased significantly ( $P \le 0.05$ ) by 22.9%, 16.15%, 15.36% and 0.90 with PST (Plumpy sup treatment), MSB, MPN and NE respectively. Height increased by 0.46% with PST, 0.06 with MPN, 0.51% with MSB and 0.10% with N E. There was an increase of 11.76% in MUAC with PST, 10.55% with MSB, 10.06% with MPN and 0.88 with NE.

Aflataring	MSB MPN		*MP		
Anatoxins	Mean ± SD	± SD Mean ± SD		t-value	p-value
AFB <sub>1</sub>	$0.04\pm0.00$	$0.18\pm0.23$	<4	-1.0	0.354
AFB <sub>2</sub>	$0.04\pm0.00$	$0.05\pm0.00$	<4	-4.2	0.013
AFG1	$0.04\pm0.00$	$0.05\pm0.00$	<4	-2.4	0.076
AFG <sub>2</sub>	$0.05\pm0.00$	$0.05\pm0.00$	<4	-0.4	0.741
<b>TOTAL</b> Aflatoxins	$0.05\pm0.00$	$0.05\pm0.00$	<4	-0.9	0.001

<b>Table 4.15:</b>	Aflatoxins content	t of supplementary	y foods (Gruel	as consumed)
1 abic 4.15.	matoring content	i or supprementary	Toous (Or ucr	as consumeu)

Values are means  $\pm$  standard deviations of triplicate determinations of samples, P < 0.05 is significant. MSB = Maize – Soybean, MPN = Maize-peanut AFB<sub>1</sub> = Aflatoxin B<sub>1</sub>, AFB<sub>2</sub> = Aflatoxine B<sub>2</sub>, AFG<sub>1</sub> = Aflatoxin G<sub>1</sub>, AFG<sub>2</sub> = Aflatoxin G<sub>2</sub>, \* Maximum permitted aflatoxins B and G contents in food products (µg/kg) according to Regulations (EC) No 1881/2006 and (EU) 165/2010

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#### of the children

		After			Percentage
<b>Treatment Group</b>	Baseline	Treatment	t-value	p-value	difference
PST-WEIGHT (kg)	6.58	8.09	-9.98	*0.000***	↑22.9
MSB-WEIGHT (kg)	6.21	7.29	-17.99	*0.000***	<b>↑16.15</b>
MPN-WEIGHT (kg)	6.49	7.57	-11.37	*0.000***	15.36
NE-WEIGHT (kg)	6.59	6.71	-3.99	*0.005**	↑0.90
PST-HEIGHT (cm)	69.87	70.19	-5.05	*0.000***	↑0.46
MSB-HEIGHT (cm)	68.27	68.62	-10.25	*0.000***	↑0.51
MPN-HEIGHT (cm)	69.71	69.95	-5.95	*0.000***	↑0.06
NE-HEIGHT (cm)	70.27	70.41	-2.31	0.054	↑0.10
PST-MUAC (cm)	11.78	13.25	-20.65	*0.000***	↑11.76
MSB-MUAC (cm)	11.77	13.07	-20.34	*0.000***	<b>↑10.55</b>
MPN-MUAC (cm)	11.79	13.04	-17.66	*0.000***	10.06
NE-MUAC (cm)	11.76	11.96	-2.62	*0.034*	↑0.88

Values are means ± standard deviation of triplicates. \*p<0.05, \*\*p<0.01, \*\*\*p<0.001 PST = Plumpy sup treatment, MSB = Maize – Soybean, MPN = Maize-peanut, NE = Nutrition Education. MUAC = Mid Upper Arm Circumference

## 4.17. Categorized baseline and after treatment anthropometric status of the children in different treatment groups

Table 4.17 showed both the categorized baseline and after treatment anthropometric status of the children in different treatment groups. For the baseline, the mean wasting z score for plumpy sup treatment group was -2.70. None (0%) of the children had the normal weight-for-height (-2 to 2) Z score, all (100%) the children were moderately wasted and none were severely wasted. Fifty percent (50.0%) had normal height – for – age, 22.2 % were moderately stunted while 27.8 % were severely stunted. The percentage of children with normal weight – for-age was 5.6 %, 50.0 % were moderately underweight while 4.4% were severely underweight. The MUAC of all the children (100%) in this group were between 11.5cm and 12.5cm which indicated that they had moderate acute malnutrition (MAM).

For MSB treatment group, the mean wasting was -2.71. Majority (94.7 %) was moderately wasted, 63.2 % had normal height-for –age, moderately and severely underweight children were 42.1% respectively. The MUAC of all the children (100 %) in this group were between 11.5 cm and 12.5 cm which indicated that they had moderate acute malnutrition (MAM). For MPN treatment group, the mean wasting z score was -2.72. Ninety-four percent (94.1 %) were moderately wasted, 23.5% were moderately stunted, and 58.8% were severely underweight. The MUAC of all the children (100%) in this group were between 11.5cm and 12.5cm which indicated that they had moderate acute malnutrition (MAM).

For nutrition education treatment group, the mean wasting was -2.65. A little above ninetyfour percentage (94.4%) of the children were moderately wasted, 27.8% were severely stunted, and 44.4% were severely underweight. The MUAC of all the children (100%) in this group were between 11.5cm and 12.5cm which indicated that they had moderate acute malnutrition (MAM). For the after-treatment anthropometric status of the children in different treatment groups. There was reduction in the number of respondents across the treatment groups as a result of skipping at least one visit. For all the treatments (plumpy sup, MSB and MPN) except NE, all the children (100 %) attained normal weight- for- height while the mean wasting was - 0.24 for plumpy sup, - 0.83 for MSB, - 0.93 for MPN and - 2.46 for NE respectively.

Across the four treatments, 15.4 % of the children were nurtured to their normal height – for – age Z score with plumpy sup, 26.7 % with MSB, 10 % with MPN and 28.6 % with NE. However, 30.8 % remain moderately stunted with plumpy sup, 26.7 % with MSB, 20 % with MPN and 42.9 % NE while 53. 8 % had severe stunting with plumpy sup, 46.7 % with MSB, 70 % with MPN and 28.6 % with NE.

About 53.8 % of children managed with plumpy sup attained the normal weight- for – age, 40 %, and 10 % was achieved with MSB and MPN respectively while none managed with NE had normal weight-for-age. While 23.1 %, 33.3 % 30 % and 42 % of children managed with plumpy sup, MSB, MPN and NE respectively were moderately underweight, also 23.1 % of children under plumpy sup, 26.7% under MSB, 40 % under MPN and 57.1% under NE were severely underweight.

All the children managed with the four treatments except NE recorded normal MUAC (> 12.5 cm) while only NE recorded 100 % MUAC of  $\geq$ 11.5 to  $\leq$ 12.5 cm.

	Plumpy Sup				
	Base	Baseline After treatm			
Maximum (n)	1	8	М	13	
Age at enlistment (months)	Mean 13.83	SD 11.98	Mean 18.69	SD 13.87	
Weight (kg)	6.51	1.76	8.02	2.36	
Length/height (cm)	69.43	9.10	69.97	10.63	
WHZ	-2.70	0.22	-0.24	0.56	
HAZ	-2.29	2.08	-3.85	2.25	
WAZ	-3.30	1.37	-2.54	1.56	
MUAC	11.76	0.17	13.25	0.18	
NUTRITION INDICATORS					
Number (n)	1	8	N	13	
Normal (WHZ -2 to 2)	N 0	% 0.0	N N	% %	
Moderate wasting (WHZ -3 to < -2)	18	100	13	100.0	
Severe wasting (WHZ < -3)	0	0.0	0	0.0	
Normal (HAZ -2 to 2)	9	50.0	0	0.0	
Moderate stunting (HAZ $-3$ to $< -2$ )	4	22.2	2	15.4	
Severe stunting (HAZ < -3)	5	27.8	4	30.8	
Normal (WAZ -2 to 2)	1	5.6	Ν	%	
Moderate underweight (WAZ -3 to < - 2)	9	50.0	7	53.8	
Severe underweight (WAZ < -3)	8	44.4	3	23.1	
Normal (MUAC > 12.5cm)	0	0.0	3	23.1	
MAM (MUAC 11.5 to 12.5cm)	18	100.0	13	100.0	

## **4.17A** Categorized baseline and after treatment anthropometric status of the children in Plumpy Sup treatment groups

n = number of subject, WHZ = weight for height z-score, HAZ = height for age z-score, WAZ = weight for age z-score, MUAC = midupper arm circumference. MSB = Maize –Soybean, MPN = Maize-peanut, NE = Nutrition Education, MAM = moderate acute malnutrition, SAM = severe acute malnutrition.

		MSB			
	Bas	eline	After	treatment	
Maximum (n)	1	9		15	
	Mean	SD	Mean	SD	
Age at enlistment (months)	11.21	5.17	13.90	5.38	
Weight (kg)	6.32	1.19	7.19	1.23	
Length/height (cm)	68.87	6.81	68.16	6.83	
WHZ	-2.71	0.21	-0.83	0.58	
HAZ	-1.82	1.65	-3.23	1.76	
WAZ	-3.01	1.01	-2.50	1.02	
	0.01		2.00	1.02	
MUAC	11.80	0.23	13.07	0.14	
	11.00	0.20	12.07	0.11	
NUTRITION INDICATORS					
Number (n)		9		15	
	N	%	Ν	%	
Normal (WHZ $_{-2}$ to 2)	0	0.0	15	100.0	
$\operatorname{Horman}(\operatorname{WHZ}^{-2} \operatorname{to} 2)$	U III	0.0	15	100.0	
Moderate wasting (WHZ $3$ to $< 2$ )	18	047	0	0.0	
which have a string (which have $-2$ )	10	JH./	0	0.0	
Source westing (WHZ $< 2$ )	1	53	0	0.0	
Severe washing (WHZ < -3)	1	5.5	0	0.0	
	10	(2)	4	$\mathcal{D}(\mathcal{T})$	
Normal (HAZ -2 to 2)	12	03.2	4	20.7	
Madamata structing (IIA7 2 to < 2)	2	150	1	$\mathcal{D}(\mathcal{T})$	
Moderate stunting (HAZ -3 to $< -2$ )	3	15.8	4	26.7	
	4	01.1	7		
Severe stunting $(HAZ < -3)$	4	21.1	/	46./	
	2	1 7 0		10.0	
Normal (WAZ -2 to 2)	3	15.8	6	40.0	
			_		
Moderate underweight (WAZ -3 to $< -2$ )	8	42.1	5	33.3	
× Y					
Severe underweight (WAZ $<$ -3)	8	42.1	4	26.7	
Normal (MUAC > 12.5cm)	0	0.0	15	100.0	
MAM (MUAC 11.5 to 12.5cm)	19	100.0	0	0.0	

### **4.17B** Categorized baseline and after treatment anthropometric status of the children in MSB treatment groups

n = number of subject, WHZ = weight for height z-score, HAZ = height for age z-score,

WAZ = weight for age z-score, MUAC = mid-upper arm circumference. MSB = Maize –Soybean, MPN = Maize-peanut, NE = Nutrition Education, MAM = moderate acute malnutrition, SAM = severe acute malnutrition.

			MPN			
	Basel	ine	Af	ter treatment		
Maximum (n)	17			10		
	Mean	SD	Mean	SD		
Age at enlistment (months)	13.41	7.92	18.53	9.62		
Weight (kg)	6 48	0.91	7 59	1 16		
(in origin (ing)	0.10	0.91	1.05	1.10		
Length/height (cm)	69.68	5.11	69.93	5.97		
N / 1 / 7	0.70	0.00	0.02	0.41		
WHZ	-2.72	0.22	-0.93	0.41		
HAZ	-2.07	2.21	-3.77	1.35		
			-			
WAZ	-3.09	1.21	-2.79	1.05		
MUAC	11.85	0.22	13.04	0.13		
Mone	11.05	0.22	15.04	0.15		
NUTRITION INDICATORS						
Number (n)	17			10		
	N	%	N	%		
Normal (WHZ -2 to 2)	0	0.0	10	100.0		
	Y					
Moderate wasting (WHZ -3 to $< -2$ )	16	94.1	0	0.0		
Severe wasting (WHZ < -3)	1	5.9	0	0.0		
Normal (HAZ $-2$ to 2)	7	41.2	1	10.0		
	,	11.2	1	10.0		
Moderate stunting (HAZ -3 to $<$ -2)	4	23.5	2	20.0		
Severe stunting (HAZ < -3)	5	29.4	7	70.0		
Normal (WAZ -2 to 2)	3	17.6	1	10.0		
Moderate underweight (WAZ -3 to $<$ -2)	4	23.5	5	50.0		
Severe underweight (WAZ < -3)	10	58.8	4	40.0		
Normal (MUAC $> 12.5$ cm)	0	0.0	10	100.0		
$\mathbf{WOAC} > 12.5 \mathbf{CIII}$	U	0.0	10	100.0		
MAM (MUAC 11.5 to 12.5cm)	17	100.0	0	0.0		

4.17C Categorized baseline and after treatment anthropometric status of the children in MPN treatment groups

n = number of subject, WHZ = weight for height z-score, HAZ = height for age z-score, WAZ = weight for age z-score, MUAC = mid-upper arm circumference. MSB = Maize -Soybean, MPN = Maize-peanut, NE = Nutrition Education, MAM = moderate acute malnutrition, SAM = severe acute malnutrition.

			NE			
	Base	line	Afte	r treatment		
Maximum (n)	1	8		7		
	Mean	SD	Mean	SD		
Age at enlistment (months)	15.67	12.05	15.52	5.68		
6						
Weight (kg)	6 80	1 57	6.86	0.78		
() orgine (Kg)	0.00	1.57	0.00	0.70		
Langth/haight (am)	71.06	<u> 8 02</u>	71 17	1 52		
	/1.00	0.03	/ 1.1 /	4.55		
N 71 1/7	0.65	0.17	0.16	0.24		
WHZ	-2.65	0.17	-2.46	0.24		
HAZ	-1.99	2.22	-2.51	0.98		
WAZ	-3.00	1.23	-3.09	0.58		
MUAC	11.84	0.21	12.03	0.14		
NUTRITION INDICATORS						
Number (n)		8		7		
	N	0%	N	0%		
Normal (WHZ $2 \pm 2$ )		<i>/</i> 0	0	<i>/</i> 0 0.0		
Notifial ( $W \Pi Z = 2 \text{ to } 2$ )	0	0.0	0	0.0		
	17	04.4	7	100.0		
Moderate wasting (WHZ -3 to $< -2$ )	1/	94.4	/	100.0		
Severe wasting (WHZ $< -3$ )	1	5.6	0	0.0		
Normal (HAZ -2 to 2)	9	50.0	2	28.6		
Moderate stunting (HAZ -3 to $< -2$ )	4	22.2	3	42.9		
Severe stunting (HAZ < $-3$ )	5	27.8	2	28.6		
Normal ( $WAZ = 2$ to 2)	3	167	0	0.0		
Normal (WAZ -2 to 2)	5	10.7	0	0.0		
	7	29.0	2	42.0		
Moderate underweight (wAZ -3 to $< -2$ )	1	38.9	3	42.9		
	0					
Severe underweight (WAZ $< -3$ )	8	44.4	4	57.1		
Normal (MUAC > 12.5cm)	0	0.0	0	0.0		
MAM (MUAC 11.5 to 12.5cm)	18	100.0	7	100.0		

4.17D Categorized baseline and after treatment anthropometric status of the children in NE treatment groups

MAM (MUAC 11.5 to 12.5cm)18100.0710n = number of subject, WHZ = weight for height z-score, HAZ = height for age z-score, WAZ = weight for age z-score,WAZ = weight for age z-score,WAZ = weight for age z-score,MUAC = mid-upper arm circumference.MSB = Maize – Soybean,MPN = Maize-peanut,NE = Nutrition Education,

= moderate acute malnutrition, SAM = severe acute malnutrition.

#### 4.18: Summary of each treatment group mean anthropometry at each visit

Table 4.19 showed the summary of each treatment group mean anthropometry at each visit. Firstly, there was reduction in the number of respondents across the treatment groups in the resultant visits due to skipping of a visit. For plumpy sup treatment group, the children's weight and height (6.51kg and 69.43cm) at enlistment visit and gradually increased at each visit till the last visit (8.02kg and 69.97cm). The mean weight-for-height z score (WHZ) at enlistment (-2.70) gradually improved at each visit and the children ended with mean WHZ of -0.24. The mean MUAC of the children at enlistment was 11.76cm and it increased to 13.51cm at the last visit.

For MSB treatment group, the children's weight and height (6.32kg and 68.87cm) at enlistment visit gradually increased at each visit till the last visit (7.19kg and 68.16cm). The mean weight-for-height z score (WHZ) at enlistment (-2.71) gradually improved at each visit and the children ended with mean WHZ of -0.83. The mean MUAC of the children at enlistment was 11.80cm and it increased to 13.07cm at the last visit.

For MPN treatment group, the children's mean weight and height (6.48kg and 69.68cm) at enlistment and gradually increased at each visit till the last visit (7.59kg and 69.93cm). The mean weight-for-height z score (WHZ) at enlistment (-2.72) gradually improved at each visit and the children ended with mean WHZ of -0.93. The mean MUAC of the children at enlistment was 11.85cm and it increased to 13.04cm at the last visit.

For Nutrition education treatment group, the children's weight and height (6.80kg and 71.06cm) at enlistment visit and gradually increased at each visit till the last visit (6.86kg and 71.17cm). The mean weight-for-height z score (WHZ) at enlistment (-2.65) gradually improved at each visit and the children ended with mean WHZ of -2.46. The mean MUAC of the children at enlistment was 11.84cm and it increased to 12.03cm at the last visit.

Visits	Plumpy sup MSB MPN					]	NE	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Enlistment visit (n =18, 19, 18, 18)								
Age (m)	13.83	11.98	11.21	5.17	13.41	7.92	15.67	12.05
Weight (kg)	6.51	1.76	6.32	1.19	6.48	0.91	6.80	1.57
Length (cm) 0-24 month ( $n = 15$ ,	66.93	6.86	68.87	6.81	68.59	4.23	68.80	5.63
19.15.15)								
Length (cm) >24 month (n = 3, 0, 2, 3)	81.93	9.68		_	77.00	5.66	82.33	9.87
Length/Height (cm)	69.43	9.10	68.87	6.81	66.68	5.11	71.06	8.03
WHZ	-2.70	0.22	-2.71	0.21	-2.72	0.22	-2.65	0.17
HAZ	-2.29	2.08	-1.82	1.65	-2.07	2.21	-1.99	2.22
WAZ	-3.30	1.37	-3.01	1.01	-3.09	1.21	-3.00	1.23
MUAC	11 76	0.17	11.80	0.23	11.85	0.22	11 84	0.21
	111/0	0.17	11.00	0.20	11.05	0.22	11.01	0.21
First visit (n=15, 18, 16, 14)								
Age (m)	15.06	13.05	11.46	5.24	13.90	8.18	16.53	12.33
Weight (kg)	6.63	1 98	6 37	1.20	6.65	0.92	7.02	1 69
Length (cm) 0-24 months ( $n = 12, 18$	66 28	7 50	68 53	6.84	68 90	4 31	69.48	6.01
14 12)	00.20	1.00	00.22	0.01	00.70		07.10	0.01
Height (cm) > 24 months (n = $3, 0, 2, 2$ )	81 93	9 68	_		77.00	5 66	88.00	1 41
I ength/Height (cm) $I = 3, 0, 2, 2$	69.41	9.88	68 53	6 84	69.92	5.00	72.12	8 71
WHZ	-2.43	0.25	-2.47	0.01	-2.49	0.19	-2.59	0.14
HAZ	-2.19	2 20	-2.06	1 70	-2.08	2.06	-2.08	1.89
WA7	-3.37	1.20	_2.00	1.70	-2.00	$\frac{2.00}{1.14}$	-3.02	1.07
MUAC	12 01	0.25	12.02	0.26	12.00	0.25	11 84	0.23
MUAC	12.01	0.25	12.02	0.20	12.00	0.25	11.04	0.25
Second visit (n=15, 17, 14, 13)								
Age (m)	15.80	12.96	11.59	5.09	15 14	8 4 8	14 31	6 98
Weight (kg)	6.91	1 97	641	1 17	677	0.99	678	1 45
Length (cm) 0-24 months ( $n = 12, 16$	66 91	7 50	67.27	6.03	68 33	4 35	69 48	6.01
12 12	00.71	7.50	07.27	0.05	00.55	4.55	07.40	0.01
Height (cm) 24 months $(n - 3, 1, 2, 1)$	82.00	9.62	80.60	_	77.05	5 73	87.00	_
I ength/Height (cm) $I = 3, 1, 2, 1$	69.93	10.21	68.15	6 70	69.96	5.75	70.43	4 39
WH7	-2.08	0.24	-2 21	0.70	-2.18	0.17	-2 58	0.17
HAZ	-2.00	2.24	-2.21	1 75	-2.10	1.56	-2.50	1.85
WA7	-2.02	1.51	-2.31	1.75	-2.70	0.96	-2.10	1.05
MUAC	12.36	0.17	12.30	0.23	12.10	0.25	-5.00	0.22
MORE	12.30	0.17	12.30	0.25	12.20	0.25	11.01	0.22
Third visit $(n - 14, 17, 12, 9)$								
$\Delta ge(m)$	16 53	13/11	12.05	5 09	16 21	9.05	13/16	5.05
Weight (kg)	7 18	2 00	6 59	1 10	6.98	1.05	6.68	0.81
Length (cm) 0.24 months $(n - 11, 16)$	66 7 <i>1</i>	2.07	67.36	6.04	68 53	1.05	0.00 70.43	1 30
10, 9	00.74	7.82	07.30	0.04	08.55	4.00	70.43	4.39
Height (cm) > 24 months (n = 3, 1, 2, 0)	82.10	9.70	80.80	-	77.10	5.66	-	-
Length/Height (cm)	70.03	10.21	68.15	6.70	69.96	5.64	70.43	4.39
WHZ	-1.65	0.23	-1.91	0.30	-1.97	0.19	-2.60	0.18
HAZ	-3.07	2.26	-2.49	1.77	-3.01	1.47	-2.06	1.06
WAZ	-3.05	1.58	-2.86	1.13	-3.09	0.96	-2.97	0.57
MUAC	12.64	0.17	12.52	0.17	12.49	0.12	11.79	0.16

Table 4.18A: Summary of each treatment	group mean anthropometry at each visi
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WHZ = weight for height z-score, HAZ = height for age z-score, WAZ = weight for age z-score, MUAC = mid-upper arm circumference. MSB = Maize – Soybean, MPN = Maize-peanut, NE = Nutrition Education.

Visits	Plumpy sup		MSB		MPN		NE	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Third visit (n=14, 17, 12, 9)								
Age (m)	16.53	13.41	12.05	5.09	16.21	9.05	13.46	5.05
Weight (kg)	7.18	2.09	6.59	1.19	6.98	1.05	6.68	0.81
Length (cm) $0-24$ months (n = 11, 16, 10, 9)	66.74	7.82	67.36	6.04	68.53	4.66	70.43	4.39
Height (cm) > 24 months (n = 3, 1, 2, 0)	82.10	9.70	80.80	-	77.10	5.66	-	-
Length/Height (cm)	70.03	10.21	68.15	6.70	69.96	5.64	70.43	4.39
WHZ	-1.65	0.23	-1.91	0.30	-1.97	0.19	-2.60	0.18
HAZ	-3.07	2.26	-2.49	1.77	-3.01	1.47	-2.06	1.06
WAZ	-3.05	1.58	-2.86	1.13	-3.09	0.96	-2.97	0.57
MUAC	12.64	0.17	12.52	0.17	12.49	0.12	11.79	0.16
Fourth visit (n=13, 16, 10, 7)				ć				
Age (m)	17.46	13.87	12.50	5.24	17.30	9.62	14.29	5.68
Weight (kg)	7.44	2.24	6.71	1.20	7.12	1.11	6.77	0.84
Length (cm) 0-24 months $(n = 10, 15, 8, 7)$	66.20	7.96	66.89	5.85	68.04	4.76	71.09	4.52
Height (cm) > 24 months (n = 3, 1, 2, 0)	82.23	9.59	80.90	-	77.10	5.66	-	-
Length/height (cm)	69.90	11.00	67.76	6.65	69.85	5.98	71.09	4.52
WHZ	-1.15	0.42	-1.56	0.37	-1.67	0.16	-2.60	0.15
HAZ	-3.41	2.25	-2.85	1.72	-3.38	1.42	-2.06	1.08
WAZ	-2.92	1.60	-2.82	1.08	-3.11	0.95	-2.96	0.60
MUAC	12.88	0.13	12.72	0.14	12.71	0.10	11.87	0.16
Fifth visit (n=13, 16, 10, 7)								
Age (m)	17.92	13.87	12.96	5.24	17.76	9.62	14.75	5.68
Weight (kg)	7.78	2.34	6.94	1.17	7.40	1.13	6.81	0.78
Length (cm) 0-24 months (10, 15, 8, 7)	66.22	7.97	66.93	5.88	68.08	4.79	71.09	-
Height (cm).> 24 months (3, 1, 2, 0)	82.23	9.59	80.90	-	77.15	5.59	-	-
Length/height (cm)	69.92	10.59	67.81	6.66	69.89	6.00	71.09	4.52
WHZ	-0.62	0.51	-1.16	0.55	-1.21	0.36	-2.51	0.19
HAZ	-3.67	2.24	-3.13	1.73	-3.61	1.38	-2.34	1.02
WAZ	-2.70	1.56	-2.70	0.99	-2.91	1.05	-3.04	0.60
MUAC	13.08	0.15	12.89	0.14	12.90	0.11	11.94	0.14
Sixth visit (n=13, 15, 10, 7)								
Age (m)	18.46	13.87	13.67	5.38	18.30	9.62	15.29	5.68
Weight (kg)	8.02	2.36	7.19	1.23	7.59	1.16	6.86	0.78
Length (cm) $0-24$ months (n = 10, 14, 8, 7)	66.26	8.02	67.24	6.06	68.13	4.76	71.17	4.53
Height (cm) > 24 months (n = 3, 1, 2, 0)	82.33	9.50	81.00	-	77.15	5.59	-	-
Length/height (cm)	69.97	10.63	68.16	6.83	69.93	5.97	71.17	4.53
WHZ	-0.24	0.56	-0.83	0.58	-0.93	0.41	-2.46	0.24
HAZ	-3.85	2.25	-3.23	1.76	-3.77	1.35	-2.51	0.98
WAZ	-2.54	1.56	-2.50	1.02	-2.79	1.05	-3.09	0.58
MUAC	13.25	0.18	13.07	0.14	13.04	0.13	12.03	0.14

 Table 4.18B: Summary of each treatment group mean anthropometry at each visit

WHZ = weight for height z-score, HAZ = height for age z-score, WAZ = weight for age z-score, MUAC = mid-upper arm circumference. MSB = Maize – Soybean, MPN = Maize-peanut, NE = Nutrition Education.

#### **CHAPTER FIVE**

#### DISCUSSION, CONCLUSION AND RECOMMENDATIONS

#### **5.1 DISCUSSION**

The present research focused at developing, formulating and determining the efficacy of locally produced supplementary food (maize-peanut {MPN} and maize-soybean {MSB}) on the anthropometric indices of under-five children with moderate acute malnutrition in Ekiti South Senatorial District, Ekiti State

#### Proximate compositions of supplementary flours and gruels

The moisture content (7.21 and 6.50%) for MPN and MSB composite flours were significantly different and within the limit (<10%) recommended by FAO/WHO (1992). Makinde and Ladipo (2012) reported the implication of moisture in terms of the microbiology quality of food and consistency/texture. Prevention of microbial contaminations that could cause children to be diarrhea prone after consumption is better achieved through low moisture content of supplementary foods which also has a positive effect on its shelf life (Gebrezgi & Yildiz, 2019). Finding is comparable to past result (5.47–6.87 %) for fermented maize, rice, soybean and fishmeal complementary foods (Amankwah, Barimah, Acheampong, Addai & Nnaji, 2009). The present result is higher than the range (2.29 % - 5.53%) reported for the Ethiopian cereal/legumes-based complementary diets (Yohannes, Makokah, Kanensi, & Tenagashaw, 2020). However, findings were comparative low to values reported for dried foods by past studies (Ndife, Abdulraheem, Zakari, 2011; Bolarinwa, Olajide, Oke, Olaniyan & Grace, 2016) and in African yam bean, sorghum and crayfish supplementary foods (Egbujie & Okoye, 2019). The low moisture content reported for the different flours could suggest that MSB and MPN when stored at room temperature would have little or no adverse effect in terms of quality (nutrient retention), although the present research does not include the shelf life of the flours. The low moisture content is also a useful property for convenient packaging and transportation of products (Oduro, Ellis, Sulemana & Oti-Boateng, 2007).

Comparing the value obtained (69.11–74.62 %) in the present study, the previous ones, (Ponka et al, 2015; Ukegbu & Ayika, 2012) reported 82.10-86.85 % and 74.50-77.29 % for five pap varieties and supplemented maize pap from Maroua, Far-North Cameroun and Ngor-Okpala, Nigeria, respectively. Previous findings (Okeke & Eze, 2006; Nwamarah & Amadi, 2009) showed similar high moisture content for complementary/supplementary foods and gruel. Contrarily, the result was higher than 63.26 and 64-52% obtained for fermented maize/millet and soybean/sorghum pap (Omenna, Olanipekun & Ogunwale, 2018), respectively. Past works (Ukegbu & Anyika, 2012; Ponka et al., 2015) have previously reported a foreseable decline in the energy density and nutrient of the complementary foods with elevated moisture content. FAO report presented a high starch and heavy densities in some food crops (such as millet, maize and sorghum) due to absorbption of lot of water during cooking, thus leading to consumption of huge quantity of such foods for adequate energy and nutrients (FAO, 2001). Meanwhile, children are endowed with small stomach structure, which presented a challenge of consuming huge amount of food at a sitting and thus, limiting the intakes of adequate energy and nutrients. Therefore the frequency of their feeding is expectedly to be increasedly done by small quantity but bulk foods (FAO, 2001).

The crude protein content presents in the flour formulated supplementary foods are between 10.80% and 14.68%. The protein contents were more than the benchmark of 6% recommended for complementary foods by the Codex Alimentarius (FAO/WHO, 2013). The protein content of MSB flour was within the values (14.52-37.70 %) specified by Codex Alimentarius, for maximum complementation of amino acids in foods and growth (FAO/WHO Codex Alimentarius Standards, 1995). As one of the most important nutrients, protein is a prerequisite required for the formulation of supplementary foods (GAIN, 2014). The high protein content in MSB is a result of presence of soya bean in the supplementary food. This confirms the study by Ogbemudia, Nnadozie & Anuge (2017) who reported that soya bean is of high quality

protein. Furthermore, Wakil and Onilude (2009) reported that a cereal-based combination produces better protein content when compared to the protein content from a single plant-based food (cereal) because of the complementing effects of the amino acids. Protein is necessary for growth and substitution of worn-out tissues. Findings of this study are similar to 6.84-13.44 % reported for complementary food from the blends of sorghum-african yam bean (Okoye & Ojobor, 2016), 10.6-15.5% in maize-plantain-soybean (Noah, 2017) and 8.70-15.05 % in maize, crayfish with black bean (Okoye and Ene, 2018) flour blends, respectively. However, protein contents of current finding are lower than 23.85–28.84 % and 13.56-23.88 % reported for complementary foods produced using germinated and fermented popcorn (Ijarotimi & Keshinro, 2012) and from sorghum, African yam bean and crayfish flours (Egbujie & Okoye, 2019).

The protein content of both gruels are significantly obtained as 7.19-7.59 %, which corresponds to 6.76-7.88 and 4.3-8.44 % previously reported by Anigo, Ameh, Ibrahim and Danbauchi (2010) and Omenna et al., (2018) for North-western Nigeria maize-millet and millet-co-fermented maize-soybean-sorghum-pap, respectively. Interestingly, the result is substantially better than 2.22, 4.48 and 4.67-5.23 % previously reported by Makanju (2012) and Anigo et al., (2009) for yellow pap, sorghum pap consumed in Nasarawa State and those from North Western of Nigeria, respectively.

The fat content of these two composite flours (7.81-8.98 %) is in line with the recommendation (<10 %) of Protein Advisory Group (1972) but did not meet the 10-25 % WHO least fat requirements for infant food (WHO/ FAO, 2004; WHO, 2012). The fat content is lower than 11.5–24.8 % reported for composite flour from maize and other crops (Solomon, 2005) but higher than 1.36 and 1.85-3.64 % for wheat-green gram (Ghavidel & Davoodi, 2011) and sorghum African-yam bean, crayfish flours (Egbujie & Okoye, 2019), consecutively.

A concrete comparative with the previous values (0.47-0.72 %, 0.91-3.50%, 2.94 %) from other studies (Omenna et al., 2018; Anigo et al., 2009; Okeke & Eze, 2006) respectively, showed a higher content of the present gruels (3.81-4.79 %). The importance of vegetable oil inclusion in infants and children foods as done in this study accroding to the recommendations of FAO and WHO (1998), helped to increase the energy density, enhance greater assimilation of fat-soluble vitamins and for the provision of essential fatty acids needed for habitual cognitive development (Aranceta & Pérez-Rodrigo, 2012). However, spoilage occurs as a result of oxidative deterioration when the fat content of food is high (Tiencheu et al, 2016).

The importance of fiber as an important dietary component cannot be over emphasized since its roles include but not limited to, aiding digestion and absorption, prevention of overweight, constipation control, management of cardiovascular disease, diabetes as well as colon cancer (Mosha, Laswai, & Tetens, 2000). The crude fiber content of the supplementary food (composite flour) is obtained as 2.33-2.42 % and is lower than 5 % recommended daily fiber allowance in complementary foods (FAO/WHO, 1985). This could be attributed to different processing methods (milling and sieving) involved during the production of the formulated diets. Although, the values obtained are within the range reported for maize-soy bean-carrot flours (Arawande & Borokini, 2010; Butt & Batool, 2010) but lower than the 2.35-4.05 % found in maize, crayfish and black bean comparative flours (Okoye & Ene, 2018). Furthermore, in the current research, the crude fiber content of the supplementary food was comparable lower to 4.69 - 8.36 % and 3.1-4.1 % reported in complementary food blended from maize (Yohannes et al., 2020) and maize, roasted pea, and malted barley (Fikiru, Bultosa, Forsido & Temesgen, 2016).

A similar value (0.17-0.28 %) was reported by Ponka (2015) and Ukegbu & Anyika (2012), who gave values of 0.14-0.21 % and 0.33-2.57 %, respectively. However, low fiber level was recommended in infant formulation for the purpose of improving digestion and assimilation

(Adepeju et al., 2016). Findings have revealed that polyphenols and non-starch polysaccharides which are fiber related fractions participate in binding minerals such as calcium, iron and zinc, hinder their presence for human (Fairweather-Tait & Hurrel, 1996). Hence, to prevent elevated water assimilation, displacement of energy and nutrient needed for children growth, complementary foods should then be of low fiber (Michaelsen et al., 2009; Klim, Isaac & Joseph, 2001). Another study (Annan et al., 2014) asserted dietary fibers to reduce energy and digestion of nutrient, which could be injurious to malnourished children and at the same time increases the bulk and satiety of food, therefore supplementary food with low fiber would be best food to manage children with MAM.

The quantity of minerals a food product possess is an indication of the ash content of such food material (Ishiwu & Onyeji, 2004; Fusuan, Fawale, Enwerem, Uche & Ayodele, 2017). Building of bones and teeth are part of the primary functions of minerals in addition to their roles in muscles and nerves, blood clotting, synthesis of hemoglobin, hence their importance in supplementary/complementary food cannot be overemphasized as they are essential in early childhood. Whitney, Hamilton and Rolfes (1990) revealed the usefulness of minerals as enzymes/coenzymes (employed for various metabolic path-ways), according to them their function includes enhancement of the body's immune system which in turn lead to reduction in diseases and advance proper operation of other organs of the body. The ash content obtained for the two supplementary composite flours (1.68-1.88 %) far less to the acceptable to 5 % standard level recommended by the Protein Advisory Group (1972). However, ash content standard has not been specified for supplementary/complementary food by the Codex Alimentarius Standards towards management of MAM (Annan, Webb, & Brown, 2014; FAO/WHO, 1995). The present content is comparatively lower compared than 0.56-2.00 and 1.88-3.17 % of supplementary food prepared from fermented carrot-soybean-maize (Barber et al., 2017) and sorghum-groundnut-crayfish (Nwaejike & Nzeagwu, 2008) composite flours.

This similar to those (0.83–1.35 and 0.75–4.82%) reported for pap from Maroua (Ponka et al., 2015) and Ngor-Okpala LGA, Nigeria (Ukegbu & Anyika, 2012), respectively.

The growth and physical activity in children is an indicator of carbohydrate being a good source of energy is needed in good quantities. The carbohydrate content of the MPN and MSB composite flour blends was high (65.03-70.69 %). The MPN flour sample falls within the range (67 -78 %) suggested for most complementary foods (FAO/WHO, 1995). The current findings are in line with the finding on sorghum composite flour blends (Yusufu, Egbunu, Egwujeh, Opega & Adikwu, 2013) but lower than findings on maize-soybean-sweet potato flour (71.49-78.82 %) reported by Okoye & Egbujie (2018). A significantly high (87.63-88.31 %) carbohydrate content was obtained for both formulated diets. Ukegbu & Anyika (2012) and Ponka et al. (2015) reported a corresponding result for pap consumed in Marou, Far-North Cameroun (79.49-85.29%) and Nigeria (77.29%), respectively. The high carbohydrate content of the complementary diets could be ascribed to the composition of maize (a cereal), which formed the larger components of most carbohydrate-based diets. This could also be due to microflora enzyme hydrolytic fermentation that caused the synthesis of complex carbohydrates from other nutrients made up of carbon skeletons (Onoja et al., 2014). The result is in the ranges of WHO recommended values of 55-75 % for all ages excluding the <2 years old children (WHO, 2003). However, carbohydrate content of the formulated foods, which is relatively high, is an indication of effectiveness of the products to manage the children with MAM (Ibironke et al., 2012).

The energy content for the composite flours in this study (384.14-411.77 kcal) is higher than 257-337 kcal reported for spices-soy-maize complementary blends (Folorunso, Ayetigbo & Afolabi, 2018). Contrarily, the Ethiopian selected cereals and legumes formulated blends (in Gondar province) showed higher energy content (394-560 kcal) (Yohannes et al., 2020). Nevertheless, the gruels had calorie content (417.96-424.05 kcal) lower than the range

(409.16–490.17 kcal) reported for pap from Ngor-Okpala, Nigeria (Ukegbu & Anyika, 2012). Notably, the present calculated total energy values did not meet the codex recommended energy value (400-425 kcal/100g). However, the sources of energy of the formulated diets (flour and gruel) might be from its carbohydrates, fat and protein. As a result of high energy yield of fat of these products (Folorunso et al., 2018), they could hence, meet the energy needs of the MAM.

#### Minerals content of formulated supplementary flours and gruels

The results of the mineral compositions revealed iodine and phosphorus as the most abundant minerals present in the formulated supplementary diets. The calcium, potassium, iron, magnesium and zinc contents in the present study were lower than the WHO (2012) recommended standards for complementary foods to manage children with MAM, due to ingredients (plant sources) used in the food samples formulation. Besides, However, the foods, when complemented with family diets, are okay in providing the nutrition yearnings of children with MAM.

The formulated diets had low sodium (13.74-14.06 mg/100 g) when compared to those 32.7–42.00 (mg/100 g) reported by Bello, Akpaoko and Ntukidem (2020) from maize-carrot-pigeon pea complementary foods but similar to 11.1–21.1 mg/100g previously reported by Solomon (2005) for three potential cereals and legumes complementary foods. However, the result is comparatively high with those (7.12 -8.56 mg/100 g) reported by Akinsola, Idowu, Oke, Idowu and Laniran (2017) for maize-millet-soybean complementary foods. Moreso, various sodium contents, such as, 1.11-2.75 (Ukegbu & Anyika, 2012), 0.78-3.39 (Ponka et al., 2015) and 73.78-150 (Anigo et al., 2010) mg/100 g have been previouly obtained for some complementary foods, respectively. Elevated blood pressure from childhood up till adulthood has been associated with high sodium intake (Gilbert & Heiser, 2005), hence the prevention and reduction in the risk of hypertension could be achieved through decreased sodium intake

in children (Naima et al, 2018). The result obtained agrees with the low sodium suggested by the Scientific Advisory Committee on Nutrition (SACN, 2003). Moreso, children with severe malnutrition usually have untypically elevated sodium and imparment potassium levels (Golden, 2009), therefore their supplementary foods is expected to have adequante potassium for renal and fecal excretion sustainability (Amegovu et al, 2013). The present supplementary food with low sodium content would thus, pose no harmful and adverse effects on their kidney, when used in managing and treating the MAM children (Derbyshire & Davies, 2007).

Calcium is an important element in teeth and bone development and also plays prominent roles in body composition and clotting of blood. The insufficiency of calcium in the diet leads to formation of rickets and osteomalacia in early childhood and adults, respectively. As expected, calcium content of both composite flours (16.51 - 22.34 mg/100 g) decreased when made in gruel (14.15 - 11.81mg/100 g). The finding from this research indicated substantial low calcium of the formulated diets when compared with the recommended allowance for children. However, this is higher than the value reported Okoye & Ene (2018) (11.10-13.15 mg/100g) and Onoja, Akubor, Gernar & Chinmma (2014) (6.44-12.14 mg/100g) for nutritional contents of fortified local staple composite flours flours. Different studies (Bolarinwa et al., 2016; Anigo et al., 2010; Ponka et al., 2015) have indicated higher calcium contents of ; 18.01-25.10, 27.68-47.95 and 30.2-89.96 mg/100 g in complementary/supplementary foods, respectively. The current result was comparatively low to WHO (2012) standards (170-400 mg/100 g) recommended for locally produced and formulated complemetary diets. Besides the use of calcium for teeth formation and bone maintenance, the present formulated diets may not meet these functions and then could be fortified or combined with family food. This could assist in the significant reduction of osteoporosis among MAM children (Adams & Berridge, 1969). The importance of calcium for vasodilation, vascular contraction, nerve transmission, muscle

functions, besides building strong bones and teeth, hormonal secretion and intracellular signaling has been reported (Beto, 2015).

The current magnesium content (11.10–14. 30 mg/100g) of both gruels is substantial low compared to 420 mg/100 g recommended by WHO (2012) to manage MAM children. The 0.64-1.26 and 3.57-5.78 mg/100g for the fortified complementary foods by Nwakalor & Obi (2014) and Okoye & Ene (2018), respectively were to those obtained in this findings. Meawhile, higher values (49.35–80.56 and 107.56-146.91 mg/100g) than the present values are reported for five Cameroonian commonly consumed pap varieties (Ponka *et al.*, 2015) and malted North-Western Nigerian cereal-soyabeans-groundnut complementary diets (Anigo et al., 2010), respectively. Moreso, current values was comparatively low to 40-76 mg/100g recommended by the Codex Alimentarius Commission for formulated complementary foods. Some food crops are known for their low magnessium levels, such as corn flour, cassava and polished rice (FAO and WHO, 2001). Essentially, maize being the significant part of the formulated diets could contribute to the low magnesium content. Magnesium also served as enzyme activators and when included with calcium and phosphorus assisted in bone formation (Agu and Aluya, 2004).

The iron obtained in this current study (4.48 - 5.54 mg/100g) is lower but higher than and similar to the values  $(18 - 30, 1.65 \cdot 2.08 \text{ and } 3.35 \cdot 6.25 \text{ mg}/100\text{g})$  in WHO (2012) recommendations to manage MAM children, complementary foods from cereal and legume (Solomon, 2005) five Cameroonian pap varieties (Ponka et al., 2015), respectively. Iron is bonded to oxygen to produce oxy-myoglobin for human muscles development and adequate metabolisms (Okaka, Akobundu, Okaka, 2006). To prevent anemia during childhood, an iron-rich food should be regularly consumed for blood cells formation.

The potassium content of the composite flours are 2.72-2.76 mg/100 g. This is low when compared to 293.11-346.20 mg/100g reportedly found in sorghum-African yam bean and-

crayfish complementary foods (Egbujie & Okoye, 2019) but similar to the potassium contents (4.55-6.32 and 4.26-5.28 mg/100 g) for soyabean and sweet potato complemetary diets (Okoye & Egbujie, 2018) and malted millet-plantain-soyabean flour (Bolarinwa et al., 2016). The gruels had same potassium content (2.86 mg/100 g), which is comparatively low to 414.8-648.8, 198.20-322.22 and 57.36 mg/100g reported for millet-maize-soybean (Akinsola et al., 2017), five Cameroonian varieties (Ponka et al., 2015) and Jos maize-based (Solomon et al., 2000) pap, respectively. The WHO (2012) recommended contents of 1500 – 2200 mg/100 g to manage children with MAM, hence the low contents in the complementary diets, which could be due to maize fermentation (Tautau et al., 2009) is expected to be supplemented with family foods. Previous study (Maguy et al., 2018) reported the proper operation of nerve and muscle cells, muscle contraction, blood clotting and essential protein synthesis by potassium due to its intracellular cation status (Moochhala & Unwin, 2015).

Zinc is an important factor in metabolic processes such as division of cells, synthesis of protein, regulation of appetite and regulation of food consumed (Mittal & Dipti, 2016). A rapid increase in weight of malnourished children with malnutrition when given zinc supplements (FAO and WHO, 1998). The MSB and MPN composite flours had zinc content of 4.01-4.22 mg/100 g, which is similar to 3.90-5.36 mg/100 g present in complementary foods from maize-(Okoye & Ene, 2018) but higher than 0.02- 3.20 mg/100g in fortified complementary foods from local staples (Onoja et al., 2014). Notably, comparing the gruels showed that zinc for MSB and MPN is high (3.98 mg/100g) and low (3.13 mg/100g), respectively. Soybean is well known as a good zinc source (Planhar, Okezie & Gyato, 2003). The current result was similar to 1.5- 4.9 mg/100 g and higher than 0.75-1.83 mg/100 g present in Nigerian maize-based pap (Ukegbu & Anyika, 2012) and Maroua, Cameroun (Ponka *et al.*, 2015), respectively. Legumes, wholegrain cereals and pulses contained excellent amounts of zinc but, which could be reduced by milling process (FAO & WHO, 1998), as being revealed in the low quantity in this study. Apart from playing

the roles mentioned above, zinc has been adjudged as an important enzymes co-factors (Maret, 2013; Roohani et al, 2013). The consequential effect of low or lack of zinc could be linked with liver and spleen enlargement, anemia, skeletal development impairment and growth failure as previously reported by Okoye & Egbujie (2018).

The present phosphorus (87.48 – 97.16 mg/100 g) was significantly high when compared with past results (1.42–2.47 and 28.90–31.13 mg/100 g) by Bello et al. (2020) and Akinsola (2017), respectively. However, phosphorus contents of 105.35-162, 92.03-171.32 and 141.8–231 mg/100g were obtained for millet-based and malted cereals-soybeans-groundnut ogi, respectively (Shalem, Ayasi & Onah, 2019; Anigo, Ameh, Ibrahim & Danbauchi, 2010). Several foods (like vegetables, grains, poultry, nuts, eggs, fish and legumes) are reportedly described as good means for phosphorus (McClure et al., 2017). The high phosphorus content obtained in this study is ascribed to the raw materials compositions and processing methods employed. For instance, their high phosphorus content could be due to presence of peanuts and soyabeans in the formulated diet (Hother et al., 2016) as well as fermentation process employed during production, which helped to release phytate-bound phosphorus. Every living cells has phosphorus is an important constituent. Apart from playing significant roles in the body's ATP formation (Okaka et al., 2006), phosphorus is very unique for bone formation (Berdanier & Zempleni, 2009).

The iodine contents of both composite flours are  $303.00 - 508.00 \ \mu g/100g$ . This is higher than 350  $\mu g$  WHO (2012) recommendations to manage the MAM and the range (0.15 -0.51  $\mu g/100g$ ) found in fortified local staples complementary food (Onoja et al., 2014). Although, there exists no difference in the iodine content of the MSB and MPN (297.33 and 293.0  $\mu g/100$  g) gruel samples but were comparatively lower than 350  $\mu g/100$  g (WHO, 2012) obtained for WHO recommendations. Iodine in the thyroid gland, produced homones useful to control body metabolic and growth rates (Zelman, 2019). Therefore, the iodine content present in the

complementary diet would aid the development and growth of the children with MAM and prevent goiter and/or other irreversible mental reterdation.

The selenium content (2.33-3.33  $\mu$ g/100g) obtained in the flour samples of the formulated diet was within the range  $(2.64 - 4.07 \mu g/100g)$  reported by Akinsola et al. (2017) in the evaluation of the nutritional qualities of maize-millet-soybean complementary foods but lower than the WHO (2012) recommended value (90 µg/100 g) for the management of MAM children. However, selenium was absent in the slurry samples of the diets. Studies have related the interdependence of selenium contents of soil, plants and its products (Sunde, 2010). Hence, selenium content in food is dependent on the its accumulation capability of selenium from the soil source (Kieliszek & Blazjak, 2016). Besides low selenium content in cereal products (0.01-0.55  $\mu$ g/g) as reported by Sunde (2012), the processing methods (fermentation soaking and heating) employed aided the reduction of the element in complementary diets (Khanam & Platel, 2016). Selenium, an antioxidant-rich element is equally important in enzyme glutathione peroxidase (GPx) content as well as in important seleno-proteins (Rayman, 2000). Expanded threat of some of the childhood illnesses such as asthma has been attributed to low selenium levels (Rubin, Navon & Cassano, 2004). Adequate selenium intake is therefore recommended for malnourished children through the consumption of food from sea foods, animal protein, fish and egg (Chun, et al., 2010) because of their exposure to increased infections-related oxidative stress (Golden & Ramdath, 1987).

The copper content of MSB composite flour is comparatively high (0.04 mg/100 g) to MPN (0.03 mg/100 g). The values were lower than the range (1.22 - 2.68, 0.19-0.23 mg/100 g) reported by Akinsola et al. (2017) and Yohannes et al. (2020) in maize-millet based complementary foods fortified with soybean and in cereal-legume based complementary foods, respectively. Although, there exists similarity in the result for MSB (0.02 mg/100 g) and MPN (0.04 mg/100 g) gruel samples but were comparatively lower than 0.17-0.40 and 1-35 mg/100

g obtained for five common varieties of pap in Maroua, Cameroon (Ponka et al., 2015) and WHO (2012) recommendations to manage the MAM children.

#### Vitamin composition of formulated supplementary flours and gruels

The formulated supplementary diets had thiamin (vitamin  $B_1$ ) contents of 2.49-2.87 mg/100 g, which is comparatively low with those (10.27-15.09 and 6.75-11.02 mg/100 g) by Okoye, Umerah and Ani (2019) and Okoye & Ene (2018) for supplementary foods from pigeon peamillet-crayfish and maize-black bean-crayfish flour blends, respectively. It has been reported that children below 6 months,  $\leq 3$  and 4-8 years needed a daily recommended allowance of 0.2, 0.3 and 0.6 mg thiamin (Hiffler et al., 2016). Vitamin  $B_1$  is a vital vitamin with metabolic functions in fats, protein and carbohydrates conversion into energy apposite and healthy functions of heart and nerve cells (IoM & FNB, 1998). The body weight, age individual metabolism and physiological conditions with respect to the overall energy content in the diet are the important parameters that determines daily requirement of thiamin, which is essentially dependent on diatery supply (i.e not synthesised endogenously (Hiffler et al., 2016). Insufficient thiamin has been shown to be responsible for the overall reduction in carbohydrate metabolism and despite the fact that malnourished children needed adequate energy, however, the inter-connectivity between thiamine and of amino acids (through a-keto acids) results in continual grave consequencial effect on the reduction of the acetylcholine formation during neural function (FAO & WHO, 1998). It functioned prominently in the maintenance of healthy mental attitude of children with MAM as well as beriberi treatment (Okoye et al., 2019). The current thiamin is within the WHO (2012) recommendations (>1 mg/100 g) for the complementary diet to manage children with MAM. Moreso, its unmarkedly untainted safety outline without higher dose level (Hiffler et al., 2016), makes the formulated diet a good source of thiamin with no negative health effect.

Riboflavin is another micronutrient whose actions are essential components of flavin adenine dinucleotide (FAD) and flavin mononucleotide (FMN) and its presence is critical for metabolism of energy and fat, normal vision, cell function, as well as healthy skin, hair and nail growth (McCormick, 2010; Okoye & Ene, 2018; Okoye et al., 2019) in young people. Golden (2009) revealed the heat stability of riboflavin but with high rate of destruction during light exposure or free radical reactions. The present riboflavin (0.06-0.07 mg/100g) of both complementary diets are similar to but lower than 0.06-0.13 and 8.10-14.02 mg/100 g present in plantain-crayfish-millet ogi (Shalem et al., 2019) and maize-crayfish-black beans complementary foods (Okoye & Ene, 2018). A shortage of riboflavin caused low FAD and FMN levels and thereafter causing inefficient metabolism of energy and subsequent reduction in energy. Past works reported its antioxidative roles to reduce oxidative stress in the malnourished children tissues (Perampalli et al., 2010; Ashoori & Saedisomeolia, 2014), which could be supplemented through additional sources.

Niacin in the MSB flour (0.15 mg/100g) was higher than those (0.10 mg/100g) in MPN flour and similar to but lower than 0.07- 0.12 and 3.17-8.72 mg/100g reported for malted milletplantain-soyabean (Shalem et al., 2019) and maize blends (Bolarinwa et al., 2016) complementary food, respectively. Humans being blood cholesterol can be reduced through the help of niacin. Niacin is another vitamin that is soluble in water and important for transfer reactions of energy in the assimilation of macromolecules (Darnton-Hill, 2019) as a result of its coenenzymes status as nicotinamide adenine dinucleotide (NAD) and adenine dinucleotide phosphate (NADP). Its absence in diets caused night blindness, beriberi and pellagra (Okoye *et al.*, 2019). Besides dehydration, diarrhea caused nutritional imbalance, food intake and nutrient assimilation reduction as well as increased nutrient reserves dissilation in malnourished children (USNRC., 1985). Hence, niacin-rich foods such as, poultry, fish, eggs, red meat, are recommended for consumption towards rehabilitation of MAM children (Golden, 2009).

The pyridoxine (vitamin B<sub>6</sub>) contents of MSB (0.26 mg/100g) and 0.18 mg/100g in MPN flour are lower than 2.43-3.84 mg/100g for Ethiopian supplementary foods from some selected cereals and legumes (Yohannes *et al.*, 2020). Pyridoxine are in three forms namely; pyridoxal, pyridoxine and pyridoxamine that are absorbed in the body as pyridoxal phosphate for coenzyme functions in glycogen, amino acids and sphingoid bases metabolism. Pyridoxine determines the nervous and enzymes system regulation during neurotransmitters formation in the brain as well as in the red blood cells transportation (Mackey et al., 2005). Its deficiency caused children seizure, non specific pruritic rash, normocytic anaemia, cheilitis with scaly lip of the mouth and tongue swelling (Brown et al., 2020). If malnourished children will be eating diets prepared from maize, beans, and oil; it is important their pyridoxine stores is good because the net of foods used in supplementing their diets may negatively affects bioavailability of pyridoxine (Golden, 2009). This is because these foods have poor pyridoxine (Mackey et al., 2005) and adequate consumption of pyridoxine-enriched diets is encouraged for effective correction of blood abnormalities caused by sideroblastic anaemia (Fishman et al., 2000).

The composite flours had vitamin A content (1.67-2.25 mg/100g) that corresponds to and lower than 0.31-2.25 and 3.01-14.66 mg/100g previously obtained for Ethiopian cereal and legume-based complementary food (Yohannes et al., 2020) and maize- crayfish-black bean complementary foods (Okoye & Ene, 2018), respectively. Although, it is importantly known for good vision maintenance and immune system booster but when indiscriminately increased could casue 25% reduction in childhood morbidity arising from malaria, measles, pneumonia and diarrhea (UNICEF, 2017). Lower risk of lung cancer are associated with higher comsuption of carotenoids, fruits and vegetables has been revealed by studies (Johnson & Russell, 2010). Good sources of vitamin A include food crops such as, orange and yellow

vegetables, green leafy vegetables, tomato products, egg, fish oil ,fruits, some vegetable oils, liver and milk (Ross, 2010).

An elevated Vitamin A level in the diet has been recommended for malnourished children because of the effect of its defiency on them as a result of thrit need to restock their decreased liver stores, and as well complement their diets that is usually low in fat content. For instance, a low vitamin A capsule dose of 5000 IU combined with food based approach has been suggested for children with MAM (Golden, 2009).

The vitamin E content of the both flours are 0.62-1.72  $\mu$ g/100g with highest and least values in MPN and MSB flours, respectively. The high content in MPN could be due to peanut (an excellent vitamin E source) in its composition (Sanders, 2003). Meanwhile, the present result is lower than 2840-12450  $\mu$ g/100g reported (Okoye & Ene, 2018) for maize-black beancrayfish complemetary foods. According to Okoye and Ene (2018), the vitamin E is principally needed for proper functioning of body organs. Berdanier and Zempleni (2009) reported that vitamin E, as an antioxidant, protected the body system against free radicals damages, inhibition of platelet aggregation as well as enhancement of immune system. The children with MAM are exposed to oxidative stress and usually have low vitamin E, plasma selenium, cholesterol, zinc and albumin; hence, vitamin E supplementation and dietary diversification is a possible solution in most MAM children who may not get adequate quantity from their diet (Tatli et al., 2000; Manary et al., 2000; Kwena & Nyandieta, 2003).

The ascorbic acid play important roles in the elimination of scurvy, infants healthy immune system and L-carnitine, collagen, certain neurotransmitters (Carr & Frei, 1999) as well as protein metabolism biosynthesis (Li & Schellhorn, 2007). Vitamin C, also played important roles in the development of bone and soft tissue as well as improving the absorption of non-heamoglobin indirectly (Atanassova & Tzatchev, 2008). The present result (56.02- 62.05 mg/100g) is higher than 2.34 - 50.41, 38.03 -50.13 and 8.32 - 13.23 mg/100g reported for

soybean-maize-banana (Ezeokeke & Onuoha, 2016), sprouted paddy sprouted African yam bean-rice-pawpaw (Obasi et al., 2018), black bean-maize-crayfish (Okoye & Ene, 2018) complementary foods, respectively. Interestingly, the current result is higher than 40 and 13.34 mg/100g recommended for blended-fortified diets (IoM, 1997) and as Codex standard for ascorbates in most complementary diets.

# Anti-nutrient and phytochemical content of the formulated complementary flours and gruels

Past study reported the significance of antinutritional factors present in complementary diets to saliently obstruct the effective nutrients utilization and assimilation in humans (Mbofung et al., 1990). The tannin composition of the formulated flour blends varied from 1.12-1.23 %. MPN had the highest tannin content which differed significantly from MSB flour. The tannin content of this study is within the range (0.0-2.41 %) of tannin content in maize-soybean-banana flour blends (Ezeokeke & Onuoha, 2016). The present (0.90 - 0.93 %) result is lower than but similar to 15.57-37.42 and 0.12 - 0.22 % found in malted cereals, soybeans and groundnut (Anigo et al., 2010) and millet-co-fermented maize-soybean-sorghum (Omenna et al., 2018) gruels, respectively. Tannin impede the assimilation of iron and hinders trypsin, chymotrypsin, amylase and lipase after forming insoluble compounds with proteins thereby decreases the digestion and utilization of protenious food (Omenna et al., 2018). However, scientific studies affirmed the combination of fermentation, cooking and germination processing methods to improve and reduce the food products nutritional quality and some anti-nutritional factors to a safe limit, respectively (Syed et al., 2011; Ibrahim et al., 2002; Mbithi-Mwikya et al., 2001).

The saponin content (0.74 - 0.83 %) in both flours significantly lower than 1.41 - 3.13 % obtained for plantain, malted millet and soybean flours (Bolarinwa, Olajide, Oke, Olaniyan &

Faromiki, 2016). Although, saponin is richly present in soybeans but the low saponin in MSB could arise from the blanching and soaking process the current bean is subjected to.

Irrespective of harmless status of low oxalate, yet past studies (Ladeji et al., 2004 and Chai & Liebman, 2004) have reported a structural with calcium, thereby forming the insoluble calcium oxalate, when present in large quantities, and this cannot easily be absorbed in the GIT, but rather causing kidney stones. Oxalate was the highest phytochemical found in both MSB and MPN formulated flour diets. The insignificant or low amount (1.48-1.53%) is higher than 0.04-0.24 % reported for our commonly used for local complementary foods (Adepoju & Etukumoh, 2014).

The alkaloid contents (0.51 - 0.55%) of the formulated diet flours (MPN and MSB) significantly lower than 9.26 - 14.16% reported for defatted soybean-malted red sorghum supplementry food (Agbaje, Oloye, Olatunji & Olawale-Olakunle, 2017). The saponin, tannin, alkaloid and oxalate contents of both gruel samples cannot prevent the nutrients bioavailablity due to their insignificantly amount, which have been reduced by several processing methods.

#### The Amino acid profile of the formulated supplementary foods (flour and gruel).

The result showed that glutamate; a non-essential amino acid, was the most prominent in MSB and MPN gruels samples while leucine was the most substantial essential amino acid. The amino acid profile of the formulated supplementary diets (gruel samples) showed the dominance of glutamic acid, which has close similarity to the previous findings (Olaofe, Adeyemi, & Adeniran, 1994; Oshodi, Esuoso & Akintayo, 1998; Adeyeye, 2004; Aremu et al., 2006; Ponka et al., 2015). All the essential amino acids were present in the formulated supplementary diets. Essential amino acids are not produced by human body; it is therefore important to consume foods that contains them. Nuss and Tanumihardjo (2011) reported that diets of children with limited tryptophan and lysine (essential amino acids) may expose them to high risk of stunting. When malnourished children are defficient in the essential amino acids, the chances of their exposure to infectious diseases is high (Peng, Yin, Li, Kim & Wu, 2007).

However, the burden of infectious disease on the body may use the little essential amino acids in supporting the immune system activation and growth (Kampman-van de Hoek et al., 2016). MSB gruel met FAO (1991) recommended value for most of the essential amino acids while MPN gruel did not meet the FAO (1991) reference standard for all essential amino acids. The supplementary diets are plant based and plant proteins when compared to animal proteins are generally incomplete because they do not have all the essential amino acids in their right amounts (Uloma, Afolami & Adepoju, 2014). However, soybeans have been reported to have all the essential amino acids, although lacking in methionine. This could have given the MSB gruel the room to meet the FAO (1991) reference standards. Except for MPN gruel, the essential amino acids constitute ~33 % of total amino acids in MSB gruel, this is an indication of a good balance in the amino profiles (Blankership and Alford, 1983). Total amino acid (TAA) above average is an indication that a food sample is nutritionally rich (Ijarotimi & Keshinro, 2013), these formulated diets can therefore serve as good diets to manage children with MAM.

#### Sensory evaluation of the formulated supplementary foods (flour and gruel).

Acceptability and the feasibility of introducing foods among a specified population is a prefiction of sensory evaluation of a food product (Paul, Dickin, Ali, Monterrosa & Stoltzfus, 2008). There was no significant difference in all the organoleptic properties (p>0.05) when the sensory evaluation scores of the formulated diets (MSB and MPN gruels) and the control (plumpy sup) were compared. Addition of sugar and oil to the diet could be attributed to this, similar results were reported by Martin, Laswai and Kulwa (2010) in study assessing nutrient content and acceptability of soybean based complementary foods. The result showed that plumpy sup was the most preffered when it comes to color, flavor, taste, texture and general acceptance between MSB gruel and plumpy sup. The MSB gruel ranked second with regards to flavor, taste, texture and general acceptance in comparison with MPN gruel, cooking and

mixing cereals with legumes increases the gruel texture (Shim & Lim, 2013). The two gruel compete favourably with plumpy sup, for instance maillard and caramelization (non-enzymatic) reactions that leads to the realease of flavour compounds (Muhimbula et al. 2011) during roasting of graoundnut. The MPN scored higher than MSB gruel in terms of color, but it was the least generally accepted gruel, colour acceptance of MPN gruel could be attributed roasted grandnut, Pelembe, Erasmusw and Taylor (2000) reported similar results. Muhimbula et al. (2011) reported that beside making home-based complementary food formulations energy dense, the sensory qualities should not be neglected because it gives rise to food predisposition for infants and young children leading to increased consumption. Giving infants improved complementary diets such as that formulated food used in this study will improve the growth of children (Lartey, Manu, Brown, Peerson & Dewey, 1999).

#### Microbial counts (cfu/g) of the formulated supplementary foods (flour and gruel)

Microbiological analyses have been the useful way for the evaluation of safety and quality of foods (Anigo *et al.*, 2007). For instance, the existence of *E. coli* is commonly used as proxy pointer to fecal contamination directly or indirectly in foods. Considerable number of *E. coli* in food is a sign suggesting a general unhygienical handling and poor storage (International Commission on Microbiology Specification for Food {ICMSF}, 1996). Ideally, *E. coli* should be absent and as such, a level of < 3 g-1 (most probable number test limit) has been defined as the satisfactory criteria for this organism (ICMSF, 1996). Detection of salmonella in food indicates unhygienic preparation and poor handling practices, hence such food should be adequately cooked prior to consumption (ICMSF, 1996). On the other hand, the presence of fungi, a spoilage microorganisms, in food is highly unacceptable (Adegoke, 2004) and have been incriminated in food poisoning illnesses (Yusuf, Egbunu, Egwujeh, Opega & Adikwu, 1992). The present result showed no growth for *E. coli, Salmonella, Coliform* and *S. aureus* in both MSB and MPN gruels, which could be due to cooking treatment. However, there was

presence of yeast and mould on the gruels which could have resulted from the source of raw materials used in formulation despite the fact that the food was prepared under laboratory conditions (Anigo, Ameh, Ibrahim & Danbauchi, 2010).

#### Aflatoxins content of supplementary foods (flour and gruel)

The MSB gruel had AFB<sub>1</sub> within the limits (0.10  $\mu$ g/kg) established by European Commission (EC) for AFB<sub>1</sub> in complementary foods and other processed cereal-based foods for Infant and Young Child (European Commission, 2006), MPN gruel had mean AFB<sub>1</sub> that exceeded it. The AFB<sub>1</sub> in this study was lower than 2.4  $\mu$ g/kg, 83.4  $\mu$ g/kg and 4.8  $\mu$ g/kg found in *Tom bran* and *ogi* respectively as previously reported (Ojuri et al., 2018). AFB<sub>1</sub> is seen as a potent cancer-causing microorganism which induces hepatocellular carcinoma (International Agency for Research on C cancer {IARC}, 1993). Reports have further shown that AFB<sub>1</sub> contributes to stunting in children (1ARC, 2015).

The total aflatoxins contents in both slurries (0.05  $\mu$ g/kg) are within the limit (4  $\mu$ g/kg) set by Nigeria regulation on aflatoxins in most foods, including baby foods. The total aflatoxins of this finding is lower than the total aflatoxins content found in *Tom bran* and *ogi* was 3.3  $\mu$ g/kg, 104  $\mu$ g/kg and 5.5  $\mu$ g/kg (Ojuri *et al.*, 2018) respectively. Findings were also less than the aflatoxin contents of maize nshima and maize porridge consumed by children in Chipata (2.2 mg/100g and 5.8 mg/100g) and in Monze (3.8 mg/100g and 2.0 mg/100g) districts of Zambia (Alamu, Godwe, Eyinla, & Maziya-Dixon, 2019). These gruels total aflatoxins could be considered low since they are lower than the tolerance limits for total aflatoxins in the groundnut and soybean set by European Union/National Agency for Food and Drug Administration and Control (4  $\mu$ g/kg) and American Food and Drug Administration (20  $\mu$ g/kg) respectively (FAO, 2004). Aflatoxins B<sub>1</sub> and B<sub>2</sub> as well as G<sub>1</sub> and G<sub>2</sub> are products of special strains of *A. flavus* and *A. parasiticus*, respectively, with mere production of aflatoxin B by *A. flavus*, while both species produced both aflatoxins (Aiko & Mehta, 2015; Fakruddin Chowdhury, Hossain & Ahmed, 2015; Ehrlich, Kobbeman, Montalbano & Cotty, 2007). Study has shown that contaminations resulting from aflatoxin in groundnuts, soybeans and their products could result to severe problems and many health hazards such as aflatoxicosis, a condition caused by aflatoxins in humans (Murshed, Bacha, & Alharaz, 2019). Epidemic acute hepatic injury has been reported to be the consequential effect of dietary exposure to B<sub>1</sub> (Sudakin, 2003). It has been reported that complete termination of this toxin is impossible, hence its reduction to levels < 20 ppb (Michaelsen, 2009; Golden, 2009) is highly recommended. Therefore, the very low contents of aflatoxin of the formulated diets make it suitable for consumption, hence recommended to manage children with MAM.

#### Effect of the supplementary foods on the anthropometric indices of the children.

Study (Kristjansson, et al., 2016) had shown that weight gain and linear height are the two common measures used to determine the outcome or success of supplementary feeding programme due to their adjudged simplicity, rapid and inexpensive data generation. It was observed that the standard (plumpy sup) had the highest percentage increase in median weight gain, the two formulated diets (MSB and MPN) had increased height higher than plumpy sup, which implied that the formulated diets can compete favourably with the coventional RUSF in the management of MAM children. This is in agreement with past findings (Kristjansson et al., 2016) on supplementary feeding that has small but statistically significance effect on weight and height (0.12 kg and 0.32 cm, respectively) of children who had supplementary food than their counterparts who did not have supplementary food within a period of 6 months. The increase in height observed in this study is in conformity with past finding (Kajjura, Veldman & Kassier, 2019) on the supplementation of locally produced food to manage infant and young children with MAM for a period of ninety days, thereby leading to a better weight-for-height z- score. From the MUAC measurement, an increase was observed in children treated with Plumpy Sup (11.76 cm), MSB (10.55 cm) and MPN (10.06 cm), this conforms with Huybregts

et al, (2012) that inclusion of RUSF to the family diet rations will not lead to cumulative reduction of wasting incidence /MAM children by having a significant gain in height-for-age ( $\sim 0.09$  cm/month) when compared to the control group.

In Nigeria, Global Nutrition Report (2016) showed that more than 50% of children under age 5 are either stunted or wasted as a consequence of poor diet and disease. About 31.5 %, 43.6 % and 10.8 % of Nigerian under-five children are underweight, stunted and wasted respectively (NBS, 2017). Among children aged 6-59months in developing countries, nutrient dense dietary intercession has been exemplified to be an effective process to reduce under-nutrition and iron-deficient anemia (Allen, Peerson & Olney, 2009; Nga et al., 2009). The supplementary food intervention across the treatment groups showed that the mean WHZ scores improved from between - 2.65 and - 2.70 (at enlistment) to between -0.24 and -2.46 (after treatment). The impact was more noticeable in children treated with plumpy sup but less improvement was observed in children managed with only nutrition education. The poor response observed in WHZ of the children treated with NE could be attributed to the nutrient content of the family food they received for period of the study. After the treatment, children fed with MSB and MPN gruels had mean WHZ scores that is within the normal range of weight-for-height of <5 years old children. However, MSB treatment group had higher WHZ score and this could be attributed to energy and nutrient contents of the gruel (MSB gruel had higher carbohydrate, protein, Ca, Fe, Zn, phosphorus, vitamin B<sub>1</sub>, B<sub>2</sub>, B<sub>3</sub>, B<sub>7</sub>, B<sub>9</sub>, A & K). Different studies have reported that providing energy rich, nutrient dense complementary foods increases recuperation of acute malnourished children as indicated by their weight-for-length (Lassi, Das, Zahid, Imadad & Bhutta, 2013; Dewey & Adu-Afarwuah, 2008; Phuka, Gladstone & Maleta, 2012; Lazzerini, Rubert & Pani, 2013; Kajjura, Veldman & Kassier, 2019). Finding of current study is also similar to the sharp increase in WHZ score of malnourised Vietnamese children fed with locally available RUTF for 4 weeks (Nga et al., 2013) and among 6-59 months children in Uganda whose MAM was treated using sorghum peanut blend (Amegovu et al., 2014). The result after treatment, showed 100 % recovery in children managed with plumpy sup, MSB and MPN respectively from both moderate and severe wasting, there was a bit of improvement from severe to moderate wasting in children managed with NE. The prevalence of MAM in children treated with nutrition education experienced very little reduction and this is not in line with the report that nutrition counseling alone can improve weight and height of children (Imdad, Yakoob & Bhutta, 2011).

Across the treatment groups, the mean HAZ score at enlistment ranged from -1.82 to -2.29 and further dropped to between -2.51 and -3.85 after treatment. This could be attributed to drop out of participants (at enlistment there were 73 participants while only 45 stayed till the end of the research), who could be the tallest which definitely affected the over all HAZ score, this is line with Dewey and Adu-Afarwuah (2008) who reported that supplementary feeding intervention programmes on linear growth has been inconsistent. Also, there could be no significant increase in HAZ because of duration of feeding (90 days), it takes some time before signifcant improvement can be observed in HAZ during supplementary feeding, this is in agreement with Isanaka et al (2009) that three months intervention may be too short to demostrate an important effect on linear growth and Michaelsen et al. (2009) that reversing moderate stunting (if it is possible at all) may take months or years. However, it does not mean that there was no growth at all, but the standard growth rate expected at the ages of the children could not be met and the gap keeps increasing as their age increases. Their age differences make this is more difficult, howerver the finding conformed with the report of Michaelsen et al (2009) who reported that reversing stunting may be easier if the management and treatment started early especially within the first 1000 days which is presumed to be a window of opportunity. The formulated diets are low in type two micronutrients (zinc, magnessium and potassium) and according to Golden (1995), linear growth requires micronutrients most especially type 2, and
since the presence of these micronutrients is possibly responsible for the quality and density of these diets and may have consequential effect on the linear growth. The quality and quantity of supplementary foods has equally been reported as reason for different linear catch-up in children with malnutrition (Kangas et al., 2019). Finding of this study however does not support the report of Nga et al. (2013) who found a reduced HAZ score after the intervention of 4 weeks and contrary to the findings on the effecacy of complementary food supplements used towards improving the nutritional status of children in the areas affected by Wenchaun earthquake over a period of one and half year (Dong *et al.*, 2013).

At the end of the treatment, there was a sharp enhancement in the mean WAZ of the children across 3 of the treatment groups and for nutrition education only slight improvement was observed. The poor effect of NE treatment on the WAZ of children is not in line with the report that educating food insecure population such as Nigeria would lead to improvement in linear growth and weight gain as seen in significant increase in HAZ and WAZ scores (Lassi *et al.*, 2013). However, the reduction of WAZ in the 3 treatment groups observed still have between 23.1 to 40% of the children severely underweight. Although, none of the children was obese, but because they experience rapid weight gain and attained their normal weight-for-height, they may have a shortage of effective tissue since the nutrients left would not be enough to permit suitable amount of lean tissue to be synthesized (Maclean & Graham, 1980; Golden & Golden, 1985). However, the reduced prevalence of WAZ observed in this research agrees with the findings of Dong et al. (2013).

The mean MUAC score of children at enlistment ranged from 11.76 to 11.84 cm which is <12.5 cm cut off point for a well nourished child, hence all the (100 %) children enlisted had moderate acute malnutrition (MAM). The result of treatment across the four groups showed that, all the children in Plumpy Sup, MSP and MPN, respectively except those managed with Nutrition education recovered from MAM (they all had their MUAC >12.cm) this is in conformity with

the previous findings that gain in MUAC is an indication of rapid catch-up growth in response to therapeutic diets (Dale, Myatt, Prudhon & Briend, 2013; Goossens, Bekele, Yun, Harci, Quannes & Shepherd, 2012).

# **5.2 CONCLUSION**

The formulation of supplementary diets from the locally available foods bridged the created vacuum through the inavailability and cost of coventional ready to use therapeutic food used in the management of MAM in children under – five years of age whose neglect in nutrition programme often increase the cases of SAM and malnutrition-related cases of under-five mortality. The two formulated (maize-soybeans and maize peanut) diets were energy dense, high in protein and carbohydrate. They also contain vitamins and essential minerals required for growth and development, had low antinutrients and insignificant total aflatoxin contents. The formulated diets had low fibre content, thus showcasing it as best food to manage children with MAM. The low anti-nutrient and phytochemicals levels of the formulated diets affirmed that they cannot constitute or be an hinderance to bioavailability of nutrients when used in the management of MAM children. The amino acid components of the gruels especially the essential amino acid constituents of MSB formulated diet met the FAO reference standards. The gruel prepared from MSB flour was the second best accepted formulated supplementary food. Indeed, the complementary diets were not only nutrient dense, but generally accepted by the panelists. There were significant improvement in the weight-for-height z score and weightfor-age z score of the MAM children managed with formulated diets but the children couldn't catch up with height – for-age z score standard. The study, however established that MAM can be managed with locally formulated and produced supplementary diets whose ingredients or constituents are readily available, cost-effective, energy and nutrient dense and culturally acceptable.

#### **5.3 RECOMMENDATIONS**

All the materials used for the formulation and production of the supplementary diets were from plant sources, addition of some ingredients from animal sources is recommended to augument the nutrient density of the products.

The nutrient requirement for children with moderate acute malnutriton most especially when managed with locally formulated food remain undefined, there has been challenges in meeting the dietary need of children with MAM in line with the proposed requirement prepared in the 2012 WHO technical note on food for the MAM management and treatment, there should be a simple and adaptable dietary requirement protocol that could be use in the formulation and local production of specific diets for MAM children.

The formulated diets have been used to successfully manage MAM children in the course of this research, efforts should be geared toward their mass production and making them available for children with MAM at the point of discovery and could be used as complementary food to prevent the occurrence of severe malnutriton in under- five children.

Government at the state level should encourage nutrition units of the relevant ministries, agencies and parastatals for the production these foods and give to mothers whose children are discovered to have MAM during nutrition screening at every contacts at health facility level. The foods can also be made available at a very cheap and affordable cost for the caregivers. Most of the children above one year do not come to health facilities after their immunization schedule, this makes nutrition screening at the facility difficult for children above one year. Community mobilization and sensitization was done during this research to gather under five children, this makes identification and management of malnourished children difficult. It is recommended that child welfare and food demonstration clinics should be encouraged at the facility level, this will afford early detection of malnourished children and management will commence early and there will be reduction of rates of child's death rates resulting from malnutrition.

The supplementary food products developed in this study from locally available food materials have high prospect in improving the nutritional status of children from the economically developing nations who may be malnourished. Behaviour change intervention ang edis indementaria indeme and counselling on MAM management and also improved infant and young children feeding

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#### ANNEXURE I

# UNIVERSITY OF NIGERIA, NSUKKA, ENUGU STATE, DEPARTMENT OF NUTRITION AND DIETETICS

## Informed consent form for caregiver to participate in the research:

# MANAGEMENT OF MODERATELY ACUTE MALNOURISHED UNDER-FIVE CHILDREN IN EKITI SOUTH SENATORIAL DISTRICT OF EKITI STATE USING MAIZE-PEANUT AND MAIZE-SOYBEAN BLENDED FOODS

# Ph.D RESEARCH

I requested your authorization, so that your child: \_\_\_\_\_

can participated in a research project whose objective is to evaluate the effect of maize-peanut and maize-soybean locally produced supplementary food on the anthropometric parameters of under five children with moderate acute malnutrition in Ekiti South Senatorial District of Ekiti State. It is expected to improve the nutrition status of children with moderate acute malnutrition and nurture them from their current nutritional state (MAM) to full recovery

In case of agreeing to participate in the study, the child will be given the following:

a) **Nutritional Screening:** in this section anthropometric measurement and MUAC of your will be taking to determine the nutritional status. b) **Supplementation:** the child can be supplemented or not according to the study group. c) **Follow-up visit:** There will be a weekly follow up visit for 12 week, during which there will be anthropometric measurement and MUAC screening of your child as well as nutrition education/dietary counseling.

The duration of the study will be 12 weeks. In case the proposed research is beneficial, I will provide the supplement and nutrition education/dietary counseling for 12 weeks. Before starting the study, an investigator-blinded sampling method/technique will be used to assign your child MSB group, MPN group, Plumpy Sup group or N E group (can be in any of four). I hope that the nutritional support will bring benefits to health of your child. Previous studies by other researchers have shown that children with moderate acute malnutrition tends to be prone to diarrhea and infections. It is known that a malnourished child has poor results in their rehabilitation and development. There may be some allergic effect or intolerance to any of the diets or food supplements that is unpredictable until they are consumed, if this is the case, the intake should be suspended until it is assessed in the weekly follow-up visit. There could be unpredictable risks that are beyond the investigator's knowledge. If you present an adverse effect or require other type of attention with respect to the study, it will be provided. The results obtained from this study are confidential and your participation has no cost and you will not

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receive payment for your participation. In addition, you and your child can leave the study at the time you decide without affecting the nutritional support you receive in the course of this study.

The study is supervised of Ade Adesanmi, Ekiti State Primary Health Care Development Agency, Ado - Ekiti (Mobile: 08034649765).

I, \_\_\_\_\_\_\_\_ have listened to the interpretation of the interpretation in yoruba/Ekiti dialect and understood the above information and my questions have been answered in a satisfactory manner. I have been informed and I understand that the data obtained in the study can be published or disseminated for scientific purposes. I agree that my child participate in this research study. I will receive a signed and dated copy of this consent form.

Signature of participant's caregiver or guardian

Date

This part must be completed by the Investigator (or his representative):

I have explained to Mr. (Mrs.) \_\_\_\_\_\_\_ the purposes of the investigation; I have explained to him (her) about the risks and benefits in this study. I have answered the questions and asked if they have any questions. I accept that I have read and know the corresponding regulations to carry out research with human beings and I am attached to it. Once the question and answer session were concluded, the present document was signed.

Investigator's signature

Date

# **Annexure II**

## Trend of mean weight-for height Z scores for each treatment group at different visits

The figure below showed the trend of the mean WHZ for each treatment group from enlistment to last visit. This result showed that children fed with plumpy sup for the period of the study improved more in their WHZ (-2.70 to -0.24 respectively) at enlistment and last visit, than those fed MSB ( -2.71 to -0.83), MPN ( -2.72 to -0.93). However, children that their caregivers were given nutrition education (NE) improved poorly within the study period as seen in their WHZ (-2.65 to -2.46, respectively) both at enlistment and last visit.



# Annex figure I: Trend of mean weight-for height Z scores for each treatment group at different visits

#### **Annexure III**

#### Trend of mean height-for-age Z scores for each treatment group at different visits

Figure 4.2 showed the trend of the mean HAZ for each treatment group from enlistment to last visit. This result showed that HAZ dropped from -2.70 to -3.85 for children fed with plumpy sup but the standard deviation (SD) increased from 2.08 - 2.25 between enlistment and the last visit. However, there was decrease in MSB (-2.49 to -2.50 HAZ and 1.77 to 1.76 SD) and NE (-2.06 to -2.61 for HAZ and 1.06 to 0.98) while for MPN the HAZ dropped from -3.01 to -3.77 and the SD increased from 1.47 to 1.55 within the study period.



Annex Figure 2: Trend of mean height-for-age Z scores for each treatment group at different visits
## **Annexure IV**

## Trend of mean weight-for-age Z scores for each treatment group at different visits

Figure 4.3 showed the trend of the mean WAZ for each treatment group from enlistment to last visit. There were a bit of improvement in WAZ and standard deviation respectively in children fed with plumpy sup (-3.30 to -2.54; 1.37 to 1.56), MSB (-3.01 to -2.50; 1.01 to 1.02), there was a drop in MPN standard deviation from 1.21 to 1.05 but the WAZ improved from -3.09 to -2.79, however both the WAZ and SD for NE decreased from -3.00 to -3.09 and 1.23 to 0.58 respectively within the study period.



Annex Figure 3: Trend of mean weight-for-age Z scores for each treatment group at different visits

## Annexure V

## Trend of mean MUAC values for each treatment group at different visits

Figure 4.4 showed the trend of mean MUAC values for each treatment group at different visits. Children at plumpy sup group had the highest improvement in their mean MUAC (from 11.76cm at enlistment to 13.25 at last visit). This was seconded by the mean MUAC of MSB treatment group (from 11.80cm at enlistment to13.07cm at last visit). MPN treatment group was the third though the pattern was similar to that of MSB treatment group (from 11.85cm at enlistment to 13.04cm at last visit). Lastly by NE treatment group, their mean MUAC increased from 11.84cm at enlistment to 12.03cm at last visit).



Annex figure 4: Trend of mean MUAC values for each treatment group at different

visits