Original Research Article

FORMULATION AND EVALUATION OF READY-TO-USE THERAPEUTIC FOODS USING LOCALLY AVAILABLE INGREDIENTS IN BAUCHI, NIGERIA

Highlights:

- Globally, severe acute malnutrition (SAM) is reported to affect 19 million children 0-5 years of age, and is associated with 1 to 2 million preventable child deaths every year.
- 60-90% of children with SAM without medical complications can be treated without being admitted to health facilities using Ready-to-use Therapeutic Food (RUTF).
- RUTF formulated using locally available ingredients are acceptable, comparable to existing RUTF, and meet the WHO recommended minimum nutrient requirements for RUTF.

Running Title

FORMULATION AND EVALUATION OF READY-TO-USE THERAPEUTIC FOODS

Abstract

Globally, severe acute malnutrition (SAM) is reported to affect 19 million children 0-5 years of age, and is associated with 1 to 2 million preventable child deaths every year. 60-90% of children with SAM without medical complications can be treated without being admitted to health facilities using Ready-to-use Therapeutic Food (RUTF). Shipping costs, delays & donor fatigue lead to periodical unavailability of RUTF in Nigeria, undermining its effectiveness in combating malnutrition. The aim of this study was to produce RUTF from locally available ingredients, and to determine the proximate composition and evaluate the acceptability of the RUTF. The study produced and evaluated eight samples of RUTF from locally available ingredients such as soybean, acha, (fonio), guinea corn, crayfish, peanuts, cashew nut, milk, sugar, vegetable oil and date palm, but discarded five of the samples based on costs and acceptability. Sensory evaluation of the three selected samples of RUTF (AOB, BOC and PCO) was carried out. The energy content (523kcal) of PCO, AOB (555kcal) and BOC (573kcal) were comparable to the recommendation of 520-550 kcal by the WHO. The

fat contents (45.11g and 43.04g) of BOC and AOB respectively were higher, while that of PCO (32.14g) was within the recommendation of 45-60% for fat. The protein contents of AOB, BOC and PCO (22.7g, 24.11g and 21.70g respectively) were higher than the recommendation of 10-12% of energy. The ash contents (3.5g and 4.38g) of AOB and BOC were similar to that of Plumpy'Nut. BOC was the most acceptable in terms of flavor, color and consistency. There was no significant difference in flavor and color ($p>0.05$) but there were significant differences in consistency and taste ($p=0.025$ and 0.008 respectively) between the samples.

Introduction

Malnutrition is the most common nutritional disorder in developing countries and it remains one of the most common causes of morbidity and mortality among children worldwide[1]. Severe acute malnutrition is defined by a very low weight-for-height (below -3 z scores of the median WHO growth standards) and a mid-upper arm circumference less than 115 mm, with or without nutritional oedema[2-4]. Globally, 52 million children under five years of age – one in twelve children in this age group – suffer from acute malnutrition[5]. Globally, 19 million children 0-5 years of age are affected by severe acute malnutrition (SAM), which is associated with 1 to 2 million preventable child deaths every year[6,7].

In Nigeria, there has been a 97.7% increase in the prevalence of SAM over 10 years (from 4.4% in 2003 to 8.7% in 2013). The highest spikes in SAM prevalence within this period have been documented in the country's north-eastern and north-western regions respectively (1.2% to 9.3% - an increase of about 775%; and 3.8% to 15.3%, an increase of 402.6%)[8]. An acutely malnourished child under 5 years is 20 times at higher risk of dying than a well-nourished child[9]. Acute malnutrition inhibits children's physiological and mental development, has life-long implications for their health, and heavily mortgages the opportunities available to future generations[10].

Ready-to-use therapeutic foods (RUTF) are energy-dense, micronutrient enhanced pastes used in therapeutic feeding, which have greatly improved the recovery rate of children with severe acute malnutrition (SAM) in sub-Saharan Africa[11-12]. Ready-to-use Therapeutic Food (RUTF) can be used to treat 60-90% of children with SAM without medical complications without admitted them to health facilities[13]. Shipping costs, delays & donor fatigue lead to periodical unavailability of RUTF in Nigeria, undermining its effectiveness in combating malnutrition. Peanut milk-based ready-to-use therapeutic food (P-RUTF) which is used in community-based treatment of SAM is expensive[14]. Sustainable treatment of SAM can be challenging in the absence of locally produced RUTF.[15]. RUTFs can be made with local ingredients to fit local taste preferences[16]. Substituting soy for much of the milk in RUTF might reduce its cost and/or increase its availability[17].

For an ingredient to be described as local, a country has to have 500 metric tonnes or more of a given ingredient available, whether nationally produced or imported, in the locale of RUTF production[18]. Nigeria's current annual production of soya beans is about 500,000 to 600,000 metric tons (10 million to 12 million bags of 50kg)[19]. Nigeria's guinea corn production is about 6,550,000 metric tons[20]. In Nigeria, an annual output of 126,000 metric tonnes of acha (fonio) has been reported[21]. Nigeria is the fourth largest producer of cashew in Africa and the sixth in the world with an output of 160,000 metric tons per year[22]. Nigeria is the third highest producer of groundnut in the world, with a world share of 7.8% and production of 3,413,100 tons in 2014[23]. In Nigeria, Bauchi State (in the north-east where this work was carried out) is one of the leading producers of groundnuts[24]. This study was carried out in northern Nigeria which lies mostly in the Sudan Savanna and the arid Sahel zone, with a longer period of dry season and low rain fall. This region produces grains massively, which include millet, fonio, soybean, cowpeas, and sorghum (guinea corn)[25].

It is against this background that this study was designed to formulate and evaluate RUTF using locally available ingredients in different ratios, in order to meet the recommended nutrient composition for RUTFs while achieving products that are culturally acceptable at a lower cost.

MATERIALS AND METHODS

Eight cereal, legume and oils mixtures were formulated and evaluated. In particular, efforts were made to combine the various cereal, legume and oil seed mixtures to maximize the protein quality, attempting to offset any essential amino acid deficiencies in one ingredient by combining it with another ingredient that was high in that particular amino acid[26.] After evaluating the proximate composition, sensory qualities, cost of production (particularly the quantity of milk used) and overall acceptability of the 8 RUTF formulations, 5 were eliminated based on the parameters listed above, and only the most acceptable and cost-effective formulations which satisfied the nutrient recommendations were further evaluated.

The eight samples of RUTF (AOB, BOC and PCO) were produced using the following ingredients: powdered milk, sugar, peanut paste, vegetable oil, soybean flour, crayfish powder, guinea corn flour, rice flour, cashew nut paste, acha flour, dried date palm powder and a vitamin-mineral mix in different combinations as presented in tables 1A, B and C.

The grains, powdered milk, sugar, crayfish, date palm and vegetable oil were purchased from Muda Lawal market and Central Market, Bauchi.

The vitamin-mineral mix was purchased from Bio-Organics Nutrient Systems Ltd, Ogun State, Nigeria.

Production of the grain flours

Acha flour and guinea corn flour were produced using a modified method of processing of grains for "Tom Brown", a complementary food commonly prepared from a mixture of toasted cereals and legumes. The grains were sorted, washed, fermented, drained, dried and toasted until very crisp and golden brown, cooled and dry-milled into flour. Acha was steeped for 12 hours and dried at 60^0C, while guinea corn was steeped for 24 hours and dried at 70^0C[27-29]. Soybean flour was produced by sorting, washing, fermenting for 12 hours, decortication, boiling for 20 minutes, cooling, drying at 70^0C, toasting, cooling and dry-milling into flour[30-32]. All grains were toasted to enhance the flavour of the products.

Date palm powder was produced by sorting, washing, drying and dry-milling into powder. Peanut paste and cashew nut paste were produced by sorting, washing, air-drying, toasting and milling into a paste. Crayfish powder was produced by sorting, washing, drying, slight toasting and milling into powder.

Varying proportions of these ingredients were combined experimentally, with the aim of arriving at the formulations with the least content of milk, yet meeting the recommended nutrient compositions for RUTF. The ingredients were processed in such a way as to approximate the flavour of "dakuwa", a delectable indigenous snack made from toasted peanuts, sugar and toasted cereals, which is commonly consumed in northern Nigeria.

Tables 1A, B and C show the ingredient composition of the eight RUTF formulations

Table 1A: Ingredient composition of the RUTF formulations (AOB, BOC, PCO)

Sample AOB		**Sample BOC**		**Sample PCO**	
Ingredient	%	Ingredient	%	Ingredient	%
Rice flour	18	Acha flour	18	Guinea corn flour	18
Peanut paste	27	Cashew nut paste	25	Peanut paste	27
Soya bean flour	18	Soya bean flour	18	Soybean flour	18
Date powder	14	Sugar	14	Sugar	15
Milk	12	Milk	14	Milk	10
Vegetable oil	10	Vegetable oil	10	Vegetable oil	12
Multimix	0.07	Multimix	0.07	Multimix	0.07
		Crayfish powder	1		

AOB – Peanut, soybean, date
BOC – Acha, Cashewnut, Soybean, Crayfish
PCO - Guinea corn, peanut, soybean in varying proportions

Table 1B: Ingredient composition of the RUTF formulations (CBM, TCG, PGS)

Sample CBM		**Sample TCG**		**Sample PGS**	
Ingredient	%	Ingredient	%	Ingredient	%
Milk	15	Milk	10	Milk	20
Sugar	25	Sugar	30	Sugar	25
Guinea corn	15	Guinea corn	15	Guinea corn	10
Peanut paste	25	Peanut paste	25	Peanut paste	25
Vegetable Oil	10	Vegetable Oil	10	Vegetable Oil	10
Soybean	10	Soybean	10	Soybean	15
Multimix	0.07	Multimix	0.07	Multimix	0.07

CBM, TCG, PGS – Guinea corn, peanut, soybean in varying proportions

Table 1C: Ingredient composition of the RUTF formulations (CMP, MBC)

Sample CMP		**Sample MBC**	
Ingredient	%	Ingredient	%
Milk	30	Milk	30
Sugar	20	Sugar	20
Peanut paste	25	Peanut paste	20
Vegetable Oil	10	Vegetable Oil	10
Soybean	15	Soybean	20
Multimix	0.07	Multimix	0.07

CMP, MBC - Guinea corn, peanut, soybean in varying proportions

An electric blender was used for mixing the different products. They were continuously mixed until a fine consistency was achieved. The products did not contain lumps and water was not added during mixing. The locally produced RUTF was in a paste form.

Determination of the proximate composition of the RUTF

The protein content of the three samples AOB, BOC PCO was determined using microkjeldahl method. Also, the fat content of the samples was also analysed using Soxhlet method. The crude fiber, ash, and moisture content of the samples were determinedas described below.The determination of each nutrient was done in triplicate.

Procedure for moisture determination

a) The samples were mixed thoroughly.

d) The water content was determined by weighing 2.5g of each sample into a silica dish, which had been previously weighed.

c) The dish containing the sample was placed inside a hot air oven (due to unavailability of a vacuum oven) for 24 hours at 70-80^{0}C. Drying at high temperature may result in losses of heat liable or volatile components)[33]. Lipid oxidation and a resulting sample weight gain can occur at high temperatures in an air oven, hence samples were dried at a lower temperature[34].

d) It was finally dried at to a constant weight and allowed to cool for ten minutes in a desiccator before weighing.

$$\% \text{ moisture} = \frac{W1-W2}{W1} \times 100$$

W_1 = Weight of biological material before drying

W_2 = Weight of biological material after drying

Nitrogen determination by micro kjeldahl method (crude protein)

The nitrogen of protein and other compounds were converted to ammonium sulphate by acid digestion with boiling sulphuric acid.

a) A known weight of sample was placed in Kjeldahl flask and about 200mg of catalyst mixture was added.

b) 10.0mL of concentrated sulphuric acid was added to the content of the flask. It was heated gently for few minutes until frothing ceased. The heat was increased to digest for 3 hours. It was allowed to cool and made to a known volume with distilled water (100mL).

c) 10.0mL aliquot of the dilute solution of the digest was distilled by pipetting the volume into distillation chamber of micro Kjeldhal distillation apparatus. 10.0mL of 40% sodium hydroxide solution and steam distillate was added into 10.0mL of 2% boric acid containing mixed indicator (note colour from red-green). It was titrated with standard 0.2N hydrochloric acid to grey end point.

$$\% N = \frac{(a-b) \times 0.01 \times 14.0057 \times c \times 100}{d \times e}$$

a = titre value for the sample

b = titre value for the blank

c = Volume to which digest is made up with distilled water

d = Aliquot taken for distillation

e = Weight of dried sample (mg)

To convert to % crude protein, multiply by necessary conversion factor (6.25)

Ash determination

The residue was charred from the moisture determination in a muffle furnace between 500^0-600^0C until the ash turned grey or nearly white. It was cooled and weighed after 12 hours[35].

Fat determination (ether-extract) was done by the Soxhlet method.

Crude fibre determination

a) 500ml glacial acetic acid, 450ml water and 50ml concentrated Nitric acid were mixed.

b) 20gtrichoracetic acid was dissolved in this mixture

c) 1g of deffated material was weighed into a 250ml conical flask. 100ml of the TCA mixture was added into the flask. It was refluxed for exactly 40 minutes, counting from the time heating commenced.

d) A 3 feet long air condenser or a water-jacketed condenser was used to prevent loss of liquid.

e) The flask was disconnected and allowed to cool, it was filtered through a 15cmNo.4Wattman filter paper previously dried and weighed.

f) It was washed 10 times with hot distilled water and once with industrial methylated spirit. The filter paper containing the residue was dried in an oven at 105^0C overnight.

g) It was transferred to a desiccator and weighed after cooling. An ashing crucible was weighed and the weight of the crucible plus the filter paper containing the fiber was taken.

h) Ashing was done overnight at 500^0C, it was cooled and weighed. The percentage crude fibre was calculated (AOAC, 2006).

Sensory evaluation

The three products were evaluated using a 5 point hedonic scale based on colour, flavour, taste, consistency, and general acceptability. Plumpy' Nut was not available for evaluation because its use is strictly regulated for the management of CMAM cases in CMAM sites, and the researchers did not have access to it.

Fifty panellists (mother-child pairs) were chosen from Federal Polytechnic Bauchi out of which 25 were mothers and 25 were children. The panelists were shared into sub groups, to assess the products that were served to them. The mothers were asked to test one product at a time and express their degree of preference in relation to the sensory attributes listed above. The samples were presented to the children and their degrees of preference for each sample were interpreted by their mothers. The degree of preference was converted into numerical scores ranging from 1 to 5, whereby 1 was strongly disliked and 5 was strongly liked. After testing a product, panelists rinsed their palate before testing the next product.

The results were analysed using ANOVA.

RESULTS

Table 2: NUTRIENT COMPOSITION OF THE RUTFSAMPLES

Nutrients	Samples								
	CBM	**TCG**	**PGS**	**CMP**	**MBC**	**AOB**	**BOC**	**PCO**	**Plumpy’ Nut**
Energy (Kcal)	554.2	517.3	525.4	516.9	555.52	555.0	573.0	523.0	530.0
Protein (g)	19.44	17.84	21.54	23.74	24.31	22.7	24.11	21.70	14.5
Carbohydrate (g)	9.48	46.22	36.54	34.52	31.41	19.67	17.83	36.73	43.0
Fat (g)	35.39	29.01	32.56	31.54	36.96	43.04	45.11	32.14	33.5
Ash (g)	2.91	2.79	3.74	3.00	3.06	3.50	4.38	2.92	4.0
Moisture (g)	0.59	0.60	0.68	0.66	1.33	2.73	0.63	0.59	<5.0

AOB – Peanut, soybean, date
BOC – Acha, Cashew nut, Soybean, Crayfish
PCO, CBM, TCG, PGS, CMP, MBC – Guinea corn, peanut, soybeanin varying proportions

Table 2 shows the nutrient content of the eight samples.Sample BOC had the highest energy content of 573kcal while sample CMP had the lowest energy value of 516.9kcal. Sample MBC had the highest protein content of 24.31g while Plumpy’ Nut had the lowest protein content of 14.5g. Fat was highest in sample BOC (45.11g) and lowest in sample TCG (29.01g). Ash (4.39g) was highest in sample BOC and lowest in sample PCO (2.92). Sample AOB had the highest moisture content of 2.73% while moisture was lowest in samples PCO and CBM (0.59%).

Five samples (CBM, TCG, PGS, CMP and MBC) were eliminated from further evaluation either because the proportion of milk used in these combinations was high,(ranging from 15-30%), or because their energy profiles were poor (<520Kcal/100g). Therefore only samples AOB, BOC and PCO were subjected to further evaluation.

Table 3: PERCENTAGE CONTRIBUTION TO ENERGY OF THE MACRONUTRIENTS

Nutrients	Fat (%)	CHO (%)	Protein (%)
AOB	69.69	14.1	16
BOC	70.55	12.45	17
PCO	55.4	28.1	16.6

AOB – Peanut, soybean, date
BOC – Acha, Cashewnut, Soybean, Crayfish
PCO – Guinea corn, peanut, soybean

Table 3 shows the percentage contribution to energy of the macro nutrients.Sample BOC had the highest percentage contribution to energy from fat, while sample PCO had the lowest contribution from fat. Sample PCO had the highest contribution from carbohydrate, while BOC had the lowest.

Table 4: SENSORY EVALUATION SCORES OF RUTF

Sample	Flavour	colour	consistency	Taste
AOB	3.50a±1.09	3.56a±1.01	3.28a±1.01	3.48ab±1.03
BOC	3.88a±0.91	3.76a±1.20	3.84b±1.01	3.88ac±0.92
PCO	3.68a±1.02	3.50a±1.02	3.52ab±1.03	3.24bc±1.12

AOB – Peanut, soybean, date
BOC – Acha, Cashewnut, Soybean, Crayfish
PCO – Guinea corn, peanut, soybean

Any two means not followed by the same letter on the same column are significantly different ($p<0.005$) using multiple comparison of 50 panelists.

Table 4 shows the sensory evaluation scores of RUTF. Sample BOC was the most generally accepted sample and had the highest acceptability in terms of flavor, colour, consistency and taste when compared with samples AOB and PCO. There were no significant differences in flavor and colour, but significant difference in consistency and taste were observed.

DISCUSSION

In the current study, the energy values of the three samples of locally produced RUTF were found to be comparable to the imported RUTF (Plumpy' nut),and in conformity with the recommendations, which indicate that the energy content of RUTF should not be below 500kcal per 100g. The energy values of RUTF produced in the current study (555, 573 and 523 Kcals respectively) are also comparable to those of alternative RUTF formulations produced in Malawi, having energy contents of 551, 567 and 512 Kcal respectively[26]. However, the energy contents of RUTF in the present study (including sample PCO with 10% milk) are higher than those indicated by Oakley et al (2010), of 2000KJ (478.0 Kcal) for RUTF containing 10% milk. The lower energy content indicated by Oakley et al may be a possible explanation for their RUTF being less effective in the treatment of SAM[17]. The three samples (AOB, BOC, PCO) in the current study are therefore energy dense and suitable for feeding to children 0-5 years and other vulnerable individuals.

According to the recommendations, Protein should contribute 10- 12% of the energy value of RUTF[36]. Compared with the standard, the imported RUTF and the locally produced RUTF were similar in protein content. Samples AOB, BOC and PCO had protein contents of 16%, 17%, 16.6% respectively, which were higher than that of Plumpy'Nut (10.9%). The legumes used in producing the RUTF were roasted prior to milling into flour, to reduce anti-nutritional factors such as phytate.This is because the content of phytate in foods has a strong negative effect on bioavailability of important minerals, and food processing methods that reduce the phytate content of foods should be promoted, especially for children with SAM[37].

In addition, the legumes used in the formulation of the locally produced RUTF contributed to most of the protein content of the product. For instance, the average crude protein (CP) content of soybean is 38% with a rich and balanced amino acid profile, (rich in the amino acids lysine, tryptophan, threonine, isoleucine, and valine which are deficient in cereal grains[38].

The fat contents of the three locally produced RUTF (69.9%, 70.55%, and 55.4% for sample AOB, BOC and PCO respectively), are comparable to that of Plumpy'Nut (56.6%). According to WHO standard, fat should contribute 40-60% to the energy value. Poly unsaturated oils are used in the production of RUTF to provide essential fatty acids. Fat is very essential in the formulation of RUTF, this is so because Severe Acute Malnutrition

(SAM) leads to severe wasting and loss of subcutaneous fat[39]. Fat in addition to protein helps in tissue regeneration, protection, and normal functioning of the immune cells which prevent children from suffering from childhood diseases.

The moisture content of the three samples waslower than that of Plumpy' Nut. This will allow locally produced RUTF to be safely stored at ambient tropical conditions for 3-4 months.

Another major concern is the economics and sustainability of any wide use of RUTF to prevent malnutrition or even to treat mild malnutrition. The products are expensive. At around 35 US cents a package, a full two-month treatment with the imported RUTF costs around between USD 45 -53per carton(without shipping costs)[40]. Many parents of children with severe acute malnutrition cannot afford this. The costs so far have been borne primarily by United Nations agencies and by international non-governmental organizations such as Medecins Sans Frontiers (MSF). The findings from this study indicate that the cost of production of locally formulated RUTF is relatively cheaper when compared to the commercially produced RUTF. The cost of production of sample AOB was ₦79.19per packet (24.6cents per packet, USD36.9 per carton of 150 packets of 100g). The cost of sample BOC was ₦103.86 per packet (35 US cents per packet, USD 48.4per carton) sample PCO was the cheapest to produce ₦54.78 per packet (17 cents per packet, USD 25.5 per carton). The cost of sample BOC was higher than the two other samples because cashew nut (which is more expensive) was used to substitute groundnuts in order to provide an alternative with a lower risk of aflatoxin contamination. These costs were calculated based on the costs of ingredients, equipment, labour and utilities for production.

RUTF contains 25% milk powder, an expensive ingredient that is not readily available worldwide. Soybean has a high protein content and it is cheap. Therefore substituting milk powder with locally produced soybean can reduce the cost of Fortified Spreads without significantly changing the macronutrient content[41]. Recent studies demonstrate the potential of new RUTF, produced from locally available grains and legumes[14]. Other ingredients such as rice, acha, soya bean, guinea corn, crayfish, groundnuts, cashew nuts are locally grown in Nigeria especially in the Northern region where the prevalence of malnutrition is very high. Even with the importation of RUTF by UNICEF and other international agencies, the number of malnourished children is still very high in Africa and Nigeria in particular. Local production of RUTF in the regions with high burden of malnutrition will contribute immensely to the reduction of severe acute malnutrition. Some of the ingredients used in this study constitute the staple foods of the people living in northern Nigeria.

CONCLUSION

The three RUTF formulations made from locally available ingredients were found to be acceptable. These formulations are cheaper than the imported products, and they meet the WHO recommended minimum nutrient requirements for RUTF. This study has concluded that milk can be substituted with soya beans without any adverse effect on the macronutrient content of RUTF. Local production of RUTF is critical for the sustainable management of Severe Acute Malnutrition.

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