

A comparative study: long and short term effect of a nutrition sensitive approach to delay the progression of HIV to AIDS among people living with HIV (PLWH) in Nigeria

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Submission Date: November 8, 2015, Acceptance date: February 2, 2016: Publication date: February 4, 2016

ABSTRACT

Background: Malnutrition has a negative impact on optimal immune function, thus increasing susceptibility to morbidity and mortality among HIV positive patients. Evidence indicates that the prevalence of macro and micronutrient deficiencies (particularly magnesium, selenium, zinc, and vitamin C) has a negative impact on optimal immune function, through the progressive depletion of CD4 T-lymphocyte cells, which thereby increases susceptibility to morbidity and mortality among PLWH.

Objective: To assess the short and long term effects of a nutrition sensitive intervention to delay the progression of human immune-deficiency virus (HIV) to AIDS among people living with HIV in Abuja, Nigeria.

Methods: A randomized control trial was carried out on 400 PLWH (adult, male and female of different religious background) in Nigeria between January and December 2012. Out of these 400 participants, 100 were randomly selected for the pilot study, which took place over six months (January to June, 2012). The participants in the pilot study overlapped to form part of the scale-up participants (*n* 400) monitored from June to December 2012. The comparative effect of daily 354.92 kcal/d optimized meals consumed for six and twelve months was ascertained through the nutritional status and biochemical indices of the study participants (*n*=100 pilot interventions), who were and were not taking the intervention meal. The meal consisted of: Glycine max 50g (Soya bean); Pennisetum americanum 20g (Millet); Moringa oleifera 15g (Moringa); Daucus carota spp. sativa 15g (Carrot).

Results: At the end of sixth month intervention, mean CD4 cell count (cell/mm³) for Pre-ART and ART Test groups increased by 6.31% and 12.12% respectively. Mean mid upper arm circumference (MUAC) for Pre-ART and ART Test groups increased by 2.72% and 2.52% within the same period (*n* 400). Comparatively, participants who overlapped from pilot to scale-up intervention (long term use, *n* 100) were assessed for 12 months. Mean CD4 cell count (cell/mm³) for Pre-ART and ART test groups increased by 2.21% and 12.14%. Mean MUAC for Pre-ART and ART test groups increased by 2.08% and 3.95% respectively. Moreover, student's t-test analysis suggests a strong association between the intervention meal, MUAC, and CD4 count on long term use of optimized meal in the group of participants being treated with antiretroviral therapy (ART) (*P*<0.05).

Conclusion: Although the achieved results take the form of specific technology, it suggests that a prolong consumption of the intervention meal will be suitable to sustain the gained improvements in the anthropometric and biochemical indices of PLWHIV in Nigeria.

Keywords: HIV; AIDS; Nutrition-sensitive approach; CD4 cell count; Macro and Micronutrients; ART; Tailored Functional Recipe-TFR.

INTRODUCTION:

Human Immunodeficiency Virus (HIV) is a severely infectious and fast replicating retro-virus, genetically made up of a single stranded RNA molecule, which impairs and deteriorates the functioning of the immune system's cells [1,2]. The World Health Organization reported complex interactions between nutrition and HIV/AIDS [3]. Evidence highlights that malnutrition weakens the immune system; as a result, those living with HIV/AIDS are extremely affected, which thereby increases their susceptibility to the disease (Table 1.1) [4,5].

Table 1.1: Center for Disease Control and Prevention classification system for HIV infection

	Clinical categories		
	A	B	C
CD4+ T-cell count (cells/mm ³) (CD4%)	Asymptomatic, acute (primary) HIV or PGL*	Symptomatic, not A or C conditions [†]	AIDS-indicator conditions [‡]
> 500 (28%)	A1	B1	C1
200–499 (15–28%)	A2	B2	C2
< 200 (14%)	A3	B3	C3

* Category A: asymptomatic HIV infection, persistent generalized lymphadenopathy (PGL).

[†]Category B: oropharyngeal and vulvovaginal candidiasis, constitutional symptoms such as fever (38.5°C) or diarrhea lasting >1 month, herpes zoster (shingles).

[‡]Category C: Mycobacterium tuberculosis (pulmonary and disseminated), Pneumocystis carinii pneumonia, candidiasis of bronchi; trachea or lungs, extra pulmonary cryptococcosis, CMV, HIV-related encephalopathy, Kaposi's sarcoma, wasting syndrome due to HIV.

Globally, there were 2.7 million new HIV infections in 2010, including an estimated 390,000 among children. Significantly, sub-Saharan Africa (SAA) accounted for 70% of new HIV infections in 2010 [6]. Furthermore, the first case of HIV reported was documented in Nigeria in 1986. CD4+ cell counts in healthy individuals have been found to range from 324-1160 cells/mm³ of blood [7].

The impact of the HIV/AIDS pandemic on the children, families, communities, Nigerian work forces, and the national GDP scores is considerable. For example, the number of children orphaned by AIDS in Nigeria was estimated to be 1.6 million in 2005 and 2.1 million in 2012 (Table 1.2) [7,8].

Table 1. 2: Cumulative HIV deaths in Nigeria [7]

	2005	2006	2010
No. of people infected	2.86 million	2.99 million	3.4 million
No. of new HIV infections:			
• Adults	296,320	305,080	346,150
• Children (<15 years old)	73,550	74,520	75,780
No. requiring ART:			
• Adults	412,450	456,790	538,970
• Children (<15 years old)	94,990	98,040	106,840
Annual HIV (+ve) births	73,550	74,520	75,780
Cumulative deaths	1.45 million	1.70 million	2.82 million

The effects of malnutrition on the immune system are well documented and include decreases in CD4 T-cells, suppression of delayed hypersensitivity, and abnormal B-cell responses [9,10]. Interestingly, the immune suppression caused by protein-energy malnutrition (PEM) mechanism is similar in many ways to the effects of HIV infection in PLWH [11]. Numerous studies reported micronutrient deficiencies impair responses, weaken epithelial integrity, and are associated with HIV disease progression [12,13,14]. Deficiencies of essential vitamins (A, B-complex, C and E) and minerals (selenium and zinc), are common in PLWH and these micronutrients are required by the immune system to combat infection [15, 16]. Furthermore, deficiencies of antioxidants, vitamins and minerals contribute to oxidative stress (a condition that may accelerate immune cell damage), increase risk of diarrhea and therefore associated mortality in HIV positive children [17,18].

Nutrient deficiencies associated with HIV are: total calories, proteins, vitamin A, vitamin B₆, vitamin B₁₂, vitamin C, vitamin E, magnesium, selenium, and zinc [18, 19]. Development of a biochemical deficiency of vitamins A, B₆ and B₁₂ is associated with faster disease progression. In contrast, normalization of plasma vitamin A, B₁₂ and zinc levels is linked to slower disease progression [19]. Low serum (>180ng/L) vitamin B₁₂ precedes disease progression [20]. Patients often have multiple nutrient deficiencies at once, and many of the nutrients which are likely to be deficient are directly or indirectly involved in maintaining normal immune system function.

Modifications to one's diet are required, and have variable effects (increasing intakes of fat may offset reductions in sugar intakes to maintain energy balance), which cannot be expected to

have huge effects. However, there is acknowledgement of this change in diet's ability to impact various aspects of HIV pathology. For example, these modifications have potential for positive effect: micronutrients attenuate HIV type 1 disease progression among adults and children [21]; nutritional interventions for reducing morbidity and mortality in people with HIV [22]. However, there is a concern that within Nigeria, these diet modifications cannot be financed, unless the approach is both cost effective and sustainable. Therefore, this study investigated the long and short term impact of locally produced tailored functional recipes to attenuate the progression of HIV to AIDS among People Living with HIV (PLWH) in Abuja, Nigeria.

MATERIALS AND METHODS:

Participants: 400 participants assessed for eligibility (PLWH with a CD4 count of ≥ 200 cells/mm³, above 18 years old, not pregnant, and without HIV/AIDS complications) from all the HIV treatment centers in Abuja, Nigeria were recruited for a six-month nutrition intervention study. 100 participants (N=100; adult, male and female from different religious background) were selected through simple randomization out of the 400 participants and were monitored for one year in a follow up study to compare the short and long term efficacy of the intervention meal.

Design: The selected participants (n=100) were randomly allocated into one of four groups and given the right to decline participation without jeopardizing receipt of care as specified in the National HIV treatment guidelines. They were also subjected to comprehensive assessment which includes demographic, physical assessment (anthropometric measurements), and CD4 count measurement using flow cytometric analysis method. The study participants continued standard treatment for PLWH (nutritional counseling, vitamin supplements for Pre-ART group and HAART (Zidovudine or Tenofovir plus Lamivudine or Emtricitabine plus Efavirenz or Nevirapine), nutritional counseling, and vitamin supplements for the ART group. Half of each group (Pre-ART and ART) were dispensed the optimized meal for daily consumption (once daily or shared as twice daily meal) for six months. Prior intervention, project's information sheet and consent forms were read, interpreted, and signed by all the study participants. Pre and post intervention assessments were carried out as specified in the study design.

On completion of the pilot intervention, the participants (n=100) progressed to form part of the scale-up intervention. This group of participants who overlapped from pilot to scale-up phase was monitored and evaluated for one year (six month pilot and six month scale-up) to ascertain the effectiveness of Amtewa meal on prolong use (12 months).

Collected assessment tools were revised for completeness and data analyzed using SPSS Statistical Software. Simple frequencies were used for data checking. Additionally, simple descriptive statistics were used for summary of quantitative data and frequencies for qualitative data. Bivariate relationships were displayed in cross tabulations, i.e. the association of nutritional intervention on CD4 count and MUAC. Univariate group comparisons included Student's t- tests for continuous variables.

Ethical consideration: Research ethics review forms were submitted at the University of Westminster, London and State House Medical Center, Abuja, Nigeria. The ethical approvals

from the two centers were received before commencement of the research, in order to ensure that participants were fully aware of the nature of the research before they consent to take part in the study. Precautions were in place to deal adequately with the effect of participation.

RESULTS:

Optimization of macro and micronutrients: Table 1.3 shows the weight of carbohydrate (CHO), protein and fat in a known weight of each food sample. It also shows the percentage total solid and the weight of crude fiber in the samples. Of the four samples analyzed, 50g of soya bean seeds contain the highest amount of CHO (17.5g), protein (20g) and fat (10g) while 20g of the millet seeds contain 14.98g of CHO, 1.58g of protein and 0.64g of fat. These two samples formed 70% of Amtewa meal.

Table 1.3: Macronutrients compositions in g/100g of optimize Amtewa meal

SAMPLE	WT(g)	CHO(g)	Kcal	Prot(g)	Kcal	Fat(g)	%Total solid	Crude fibre
Soya bean seeds	50	17.5	70	20	80	10	92.47	4.65
Moringa leaves	15	5.73	22.9	4.07	16.26	0.35	29.26	2.88
Carrot roots	15	0	0	0.16	0.63	0	8.9	
Millet seeds	20	14.98	59.9	1.58	6.32	0.64	94.36	0.22
TOTAL	100	38.21	153	25.80	103.21	10.99	224.99	7.75
DRV¹	100	50		15		35		
(g/100g)								
DRV¹	100	50		15		35		
(Percent)								

¹Dietary Reference Value for Food Energy and Nutrients for the United Kingdom [23]

Table 1.4 shows the total weight of micronutrients in each of the analyzed samples. The four samples contain micronutrients (vitamin A from carotene, vitamin B, vitamin C, Ca, Mg, K, Na, Mn, Fe, Cu, Zn and phosphorus) which are essential for immune boosting. Soya bean seed and moringa leaves contain higher amounts of each micronutrient than carrot and millet. However, carrot contains 2.2mg of carotene in 100g Amtewa meal and Moringa contains 2.84mg of carotene in 100g of Amtewa meal which were the natural sources of vitamin A in the formulation. The total weight of each sample was compared to the DRV and RNI for the United Kingdom (UK) for the macro and micronutrients [23] to ensure that the summation of each micronutrient does not exceed the DRV and RNI. UK values were used because the DRV for the Nigeria population has not yet been determined. According to DRV for UK, Table 1.5 shows the EARs energy per day for male is 2500 kcal/d and for female is 2000 kcal/d (average 2250 kcal/d). The optimized 354.92 kcal/d of Amtewa meal which is 10% to 20% higher than the

average daily energy requirements for healthy male and female adults, was packaged and dispensed to PLWH to be consumed daily in addition to their normal daily nutritional intake.

Table 1. 4: Micronutrients compositions in mg (see details of Table below)

SAMPLES	Ca	Mg	K	Na	Mn	Fe	Cu	Zn	Se	P	Car	B1	B2	B3	Vit. C
SOYA	138.5	140	898.5	1	0	7.85	0	25	0.008	352	0	0	0	0	3
MORINGA	300.4	55.2	198.6	0	0	4.23	0.09	05		30.6	2.84	0.4	3.08	1.23	2.60
CARROT	0.48	0.12	1.34	0.17	03	0.13	0.12	05		0	2.2	0	0	0	4.9
MILLET	12	23.1	90	5.4	03	6.18	0.12	05		59.4	0	0	0	0	0
TOTAL	451.4	218.4	1188.4	6.57	06	184	0.33	3.88	.008	442	5.04	0.4	3.08	1.23	10.50
RNI (Males) ^{1,2}	700	300	3500	1600		8.7	1.2	9.5	0.075	550	700	1.0	1.3	17	40
RNI (Females) ^{1,2}	700	270	3500	1600		14.8 ³	1.2	7.0	0.06	550	600	0.8	1.1	13	40

¹Dietary Reference Value for Food Energy and Nutrients for the United Kingdom. ²19 years and above.

³Insufficient for women with high menstrual losses where the most practical way of meeting iron requirements is to take iron supplements.

Table 1. 5: Percentage composition of Carbohydrate, Protein and Fat contained in the Amtewa meal (100g)

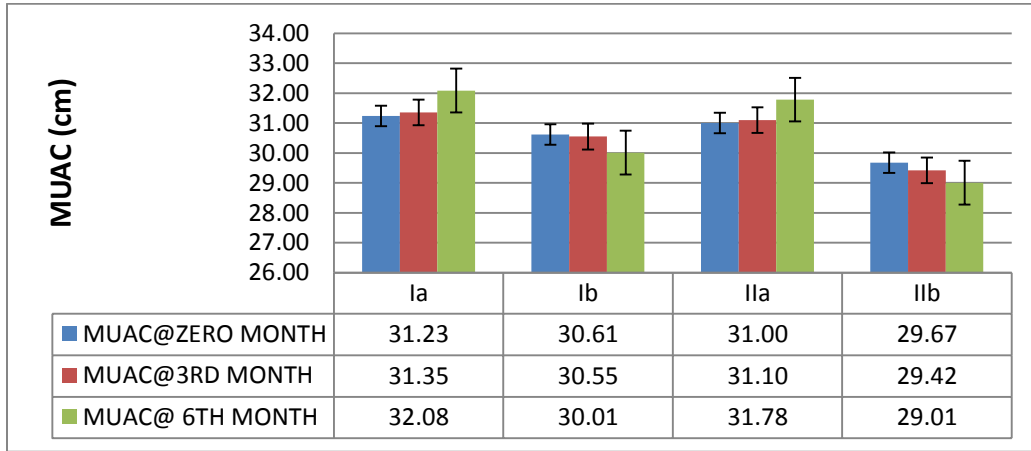
Total energy(kcal/d)	% CHO	% Protein	% Fat	EARs energy (males) kcal/d	EARs energy (females) kcal/d
354.92	38.21	25.80	10.99	2500 ¹	2000 ¹

¹Dietary Reference Value for Food Energy and Nutrients for the United Kingdom

Demographic characteristics of Participants: One hundred (100) PLWH were monitored for 12 months in an intervention program. All the research participants were above 18 years of age. 61.2% of the participants were female while 38.8% were male. Additionally, 0.26% divorced 36.98% single, 58.85% married, 3.39% widows, and 0.52% were widowers. The percentage of married participants may be a factor to explain the degree of adherence, or why most of the participants completed the six months or one-year intervention program. 10.1% of the participants were less than 30 years, 65.9% of the participants were between ages 31 to 50 years and 24% were greater than 50 years. Mean age (n=100) was 43.32 years, median age (n=100) was 43 years and the standard deviation (SD) was 9.721. The cumulative frequency was 100%. Therefore, intervention programs to alleviate stigmatization in PLWH/AIDS is more likely to impact more on PLWHIV/AIDS that are less than 30 years of age than those above 30 years of age.

Anthropometric indicator MUAC: Figure 1.1 shows significant increase in the MUAC from inclusion into the study (0 month) to the sixth month of the intervention. Mean MUAC in the

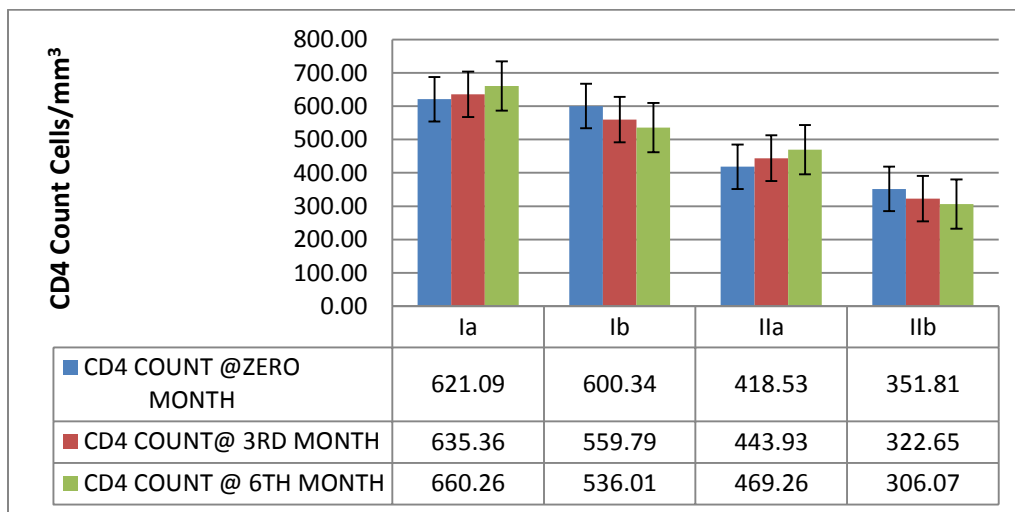
Pre-ART Test group increased by 0.38% at the third month and 2.72% at the sixth month. Similarly, mean MUAC in the ART Test group increased by 0.33% at the third month and 2.52% at the sixth month. Conversely, in the control group, mean MUAC in the Pre-ART Control decreased by 2% while a similar decrease of 2.28% was recorded for the mean ART Control group at the sixth month of the study. This is statistically significant (p=0.05).



Key: Ia = Pre-ART Test, Ib = Pre-ART Control, IIa = ART Test, IIb = ART Control

Figure 1.1: The impact of Amtewa meal on Mean MUAC (cm) at zero, three and six month intervals of the scale-up intervention (n=100).

Biochemical indicator CD4 cell count: Mean CD4 cell count of the Test groups (Amtewa meal intervention groups) of participants in the 12 month intervention increased. For example, in the Pre-ART group, the percentage increase in the mean CD4 counts at three and six months were 2.3% and 6.31% respectively. Similarly, in the ART test group, the percentage increases were 6.07% and 12.12% at three and six months respectively. In the control groups, mean CD4 count decreased by 64.33 cells/mm³ in the Pre-ART group and 45.74 cells/mm³ in the ART group as illustrated in Figure 1.2.



Key: Ia = Pre-ART Test, Ib = Pre-ART Control, IIa = ART Test, IIb = ART Control

Figure 1.2: The impact of Amtewa meal on Mean CD4 counts (cells/mm³) at zero, three and six month intervals of the scale-up intervention (n=100).

DISCUSSION:

The Amtewa meal nutrition intervention increased CD4 cell counts and MUAC of PLWH. This supports other trials which have demonstrated the positive effect of vitamins B, C, and E supplements on the immune status of HIV-infected persons, in addition to the relationships between micronutrients status and HIV disease progression among adults and children [21]. The human diet contains an array of different compounds which possess antioxidant activities, or have been suggested to scavenge reactive oxygen species (ROS) [24]. This ROS is responsible for the oxidative damage of biological macromolecules such as DNA, carbohydrates, and proteins. These processes are discussed as pathobiochemical mechanism involved in the initiation or progression phase of various diseases, including HIV/AIDS. Antioxidants such as selenium, zinc, and vitamin C in the Amtewa meal may be responsible for scavenging this ROS. There are various sources of specific ROS in the human organism. However, the superoxide radical anion appears to play a central role, since other reactive intermediates are formed in reaction sequences starting with O_2^- [24]. It is generated by enzymic one-electron reduction of oxygen from xanthine oxidase (EC 1.2.3.2), NADPH oxidase, or by leakage of the respiratory chain. It has been estimated that 1-3% of the O_2 we utilize is converted to O_2^- . To counteract the prooxidant load, a diversity of antioxidant defense systems is operative in biological systems, including enzymic and non-enzymic antioxidants [24]. These antioxidants (present in Amtewa meal) are substances present in low concentrations, compared to that of an oxidizable substrate which significantly delays or inhibits the oxidation of that substrate [25].

Carotenoids (present in Amtewa meal) are lipophilic antioxidants present in lipoproteins such as LDL and HDL. Some of the major sources are carrots (alpha carotene and beta carotene), tomatoes, citrus fruits, and spinach. Flavonoids are considered polyphenolic antioxidants that occur in several fruits, vegetables, and beverages. Enzymes such as glutathione peroxidase and superoxide dismutase, which require dietary supply selenium, copper, and zinc respectively, contributed to the overall oxidative defense mechanism achieved by the intervention meal [24].

Furthermore, micronutrients (zinc, copper, selenium) and vitamins (A, C, E, B6 and B12) contained in Amtewa meal are essential for maintaining proper immunological function, performing very important roles in the body's physiological and psychological (fear of death, anger, anxiety etc) processes and the regulation of energy metabolism [10,12]. Deficiencies of these micronutrients in HIV disease have been associated with higher risks of HIV disease progression and mortality. Studies have shown low or deficient serum concentration of several micronutrients, including thiamine, selenium, zinc, and vitamins A, B3, B6, B12, C, D and E to be individually associated with either low CD4 cell counts or advanced HIV-related diseases. Furthermore, faster disease progression or HIV-related mortality among HIV-positive persons not receiving ART was observed [10,12].

Some morbidity characteristics, such as diarrhea, nausea or vomiting, lower respiratory tract infections, oral ulcer, thrush, severe anemia, and low CD4+ are related to a higher risk of wasting. Moderate to severe malnutrition was present among 16% of patients at the time of starting ART, and was a significant predictor of death [26]. Vitamin B, in addition to vitamins C and E (contained in Amtewa meal) are essential vitamins that can reduce the risk of wasting [26]. Increased serum zinc levels (as optimized in Amtewa meal) are associated with improved virology control. In another investigation, an absolute CD4 count increased by an average of

24% in the micronutrient group versus no change in the control group ($p=.01$); serum parameters were not different among both groups, but the micronutrients supply, as proposed in the study, improved CD4 cells count reconstitution in HIV-infected person taking HAART [19]. Similarly, a synergistic effect in Amtewa meal nutrition intervention with PLWH already on HAART was documented (12.12% increase in the CD4 count of the ART test group (group on HAART) compared to the 6.31% increase in the Pre-ART test group (not on HAART)).

The choice of MUAC in the assessment of the nutritional status in this intervention was based on the fact that MUAC is simple and minimally invasive, and is rarely affected by edema than other anthropometric measurements [11]. The classification of mean MUAC (cm) according to age groups confirmed that the mean MUAC for men within the age group 18-74 years is 31.8cm, while that of women within the same age group (18-74 years) is 29.4cm [27]. Although the MUAC results (Test versus control) in the scale-up intervention indicated that both groups are within the normal range, there was still a consistent decrease in MUAC in the control (Pre-ART and ART) groups.

In support of our findings, an estimated relative risk of death was increased fivefold in HIV-infected children who had the lowest quantities of fat-free mass [28]. This implies that there is a strong association between poor growth and mortality in HIV, which may further be investigated in future work with the Amtewa meal. The possibility that poor nutritional status accelerates the progression of asymptomatic HIV infection to AIDS has already been suggested by various authors. Likewise, the link between nutritional status and clinical progression of HIV/AIDS has been re-emphasized in recent studies. Multivitamins alone significantly reduced the risk of a first episode of a low mid-upper arm circumference. Therefore, the Amtewa meal, which is a natural source of macro and micronutrients, may result in better outcomes in future comparative studies, with studies done with synthetic multivitamins.

The Malnutrition Universal Screening Tool (MUST) was developed by the British Association of Parenteral and Enteral Nutrition (BAPEN) and is supported by many professional bodies, including the British Dietetic Association, Royal College of Physicians and National Institute for Health and Clinical Excellence (NICE) classified MUAC values using the following reference tool [11]:

- If MUAC < 23.5cm, BMI < 20 kg/m² (underweight)
- If MUAC > 32cm, BMI > 30 kg/m² (obese)

From the above reference tool, the MUAC intervention results reveals that the participants monitored over a period of 6 months were not underweight, but the control participants consistent weight lost calls for an urgent intervention (Amtewa meal intervention), as studies have shown that among HIV-positive persons, 5% weight loss in 6 months is markedly associated with an increased risk of death [11, 12,13]

Given the economic instability currently evident in Nigeria, there is an inherent difficulty for drug manufacturing industries to purchase sufficient resources to manufacture and distribute synthetic micronutrients. Moreover, in the places where synthetic micronutrients are available, these micronutrients are not cost effective, are less bioavailable, and may not be sustainable in developing countries like Nigeria, compared to the Amtewa meal, which only costs an average of \$1 to produce a pack of 100g per day.

CONCLUSION:

Although the achieved results take the form of specific technology, it suggests that a prolonged consumption of the intervention meal (Amtewa) will be cost effective and suitable to sustain the gained improvements in the anthropometric and biochemical indices.

Abbreviations: Dietary Reference Value (DRV); Reference Nutrition Intake (RNI)

Competing interests: The authors declare that they have no competing interests

Author's contributions

Abraham M. Amlogu, Pharm.D, FPCPharm, PhD is the principal investigator for this study. He developed the conceptual framework, study design and writing the manuscript.

Sundus Tewfik, BSc, MSc, PhD is a Pharmaceutical scientist. She contributed to the conceptualization of the research and the analysis of the micro and macro nutrients of the intervention meal.

Charles Wambebe, B.Pharm, PhD is a Professor of Pharmacology. He contributed in the development of the research protocol, monitoring and management of research field work.

Ihab Tewfik, BSc, MPH, DrPH is a Fellow of the Royal Society of Public Health. He coordinated the initiatives to accelerate the development and subsequent production of the intervention meal. He also contributed in the study design and assisted in writing the manuscript.

Acknowledgements

This study was supported by State House Medical Center (SHMC) Abuja, Nigeria. We gratefully acknowledge the invaluable contributions of SHMC HIV/AIDS support group. We also thank the laboratory staff of both University of Westminster London and London Metropolitan University.

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