Public health nutrition intervention to delay the progression of HIV to AIDS among people living with HIV (PLWHIV) in Abuja, Nigeria

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PUBLIC HEALTH NUTRITION INTERVENTION TO DELAY THE PROGRESSION OF HIV TO AIDS AMONG PEOPLE LIVING WITH HIV (PLWHIV) IN ABUJA, NIGERIA

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ABSTRACT

Introduction: HIV/AIDS is a pandemic disease and its scourge has had a devastating impact on health, nutrition, food security and overall socioeconomic development in countries that have been greatly affected by the disease. The engagement of HIV/AIDS with under nutrition form a symbiotic relationship and one increases the prevalence and severity of the other.

Aim: The main goal of the study was to evaluate the effectiveness of public health-nutrition intervention programme designed to attenuate the progression of HIV to AIDS among people living with HIV in Abuja, Nigeria.

Methods: Local foods, which were known for their availability, accessibility, micro and macronutrient strengths were selected and optimised into a nutritional functional meal (*Amtewa*). 1000 PLWHIV were invited to participate in the research from all the HIV treatment centres in Abuja, Nigeria. Based on the sample size calculation, inclusion and exclusion criteria, 400 participants (adult, male and female from different religious background) were selected through simple randomisation. Out of these 400 participants, 100 were randomly selected for the pilot study. The participants in the pilot study overlapped to form part of the scale-up participants. The effect of daily consumption of Amtewa meal (354.92 kcal/d) for six and twelve months was ascertained through the nutritional status and biochemical indices of the study participants (n=100 pilot and n=400 scale-up interventions) who were/were not taking anti-retroviral drug therapy (ART).

Results: Mean CD4 cell count (cell/mm^3) for ART-Test group at baseline and sixth months increased by 12.12%. Mean mid upper arm circumference (MUAC) (cm) also increased by 2.52% within the same period (n=400). On the contrary, there were decreases in control groups of 14.9% CD4 count and 2.28% MUAC. Student's t-test analysis suggests a strong association between the intervention meal and mean CD4 count (It increased by 54.40 cells/mm^3 in the ART Test group ($p=0.05$)) on prolong use of Amtewa (up to 12 months).

Conclusion: These results ascertained the effectiveness of Amtewa meal on health status of HIV infected subjects and also underpinned its significant position within the National Health Services framework as innovative nutritional approach to delay the progression of HIV to AIDS in Nigeria.

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Special thanks to my lovely wife- Laura, my son- Abraham and my daughters – Nicole and Natalie, without whom this effort would have been worth nothing. Your love, support and constant patience have taught me so much about sacrifice, discipline and compromise. God bless you ALL!

……The race is not to the swift.

Nor the battle to the strong,

Nor bread to the wise,

Nor riches to men of understanding,

Nor favor to men of skill;

But time and chance happen to them all.

- Ecclesiastes 9:11 -

AUTHOR'S DECLARATION

I declare that all the materials contained in this thesis are my own work and was carried out in accordance with the Guidelines and Regulations of the University of Westminster. The work is original except where indicated by special reference in the text.

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LIST OF ABBREVIATIONS

Abbreviation	Full name
AIDS	Acquired Immune-Deficiency Disease Syndrome
ART	Anti-Retroviral Therapy
ARV	Anti- Retroviral
BMI	Body Mass Index
DRV	Dietary Reference Value
EARs	Estimated Average Requirements for Energy
FANTA	Food and Nutrition Technical Assistance Project Academy for Educational Development
FAO	Food and Agriculture Organization
FMM	Food Multi-Mix
FMOH	Federal Ministry of Health
GFATM	Global Fund to Fight AIDS, Tuberculosis and Malaria
GON	Government of Nigeria
HAART	Highly Active Anti –Retroviral Therapy
HCT	HIV Counselling and testing
HIV	Human Immune Deficiency Disease Syndrome
kcal	kilocalorie = 10^3 or 1000 calories
kJ	kilojoule = 10^3 1000 joules
MUAC	Mid–Upper Arm Circumference
NACA	National Agency for the Control of AIDS
PCV	Packed Cell Volume
PLWH	People Living with HIV
PLWHA	People Living with HIV/AIDS
PMTCT	Prevention of Mother to Child Transmission of HIV
RG	Random Glucose
RDI	Recommended Daily Intakes of Nutrients for the United Kingdom
RNIs	Reference Nutrient Intakes
SHMCA	State House Medical Centre, Abuja
SSA	Sub- Saharan Africa

USAID	United States Agency for International Development
UNAIDS	United Nations Joint Programme on AIDS
VCT	Voluntary Counselling and Testing
WHO	World Health Organization
SGOT	Serum Glutamic Oxaloacetic Transaminase

Chapter 1

Introduction

Contextualising the current PhD research

In 1986, the first case of HIV report in Nigeria was documented (FMOH, 2008). In line with guidelines from the World Health Organisation (WHO), the government adopted antenatal clinic (ANC) sentinel surveillance as the system for assessing the epidemic. The national HIV seroprevalaence level obtained from sentinel surveys of antenatal care attendees, increased from 1.8 percent in 1991 to 5.8 percent in 2001 and then declined to 5.0 percent in 2003 and further to 4.4 percent in 2005. This was followed by a rise to 4.6 percent in 2008 and then a recent decline to 4.1 percent in 2010 (NACA, 2012). Statistics obtained from attendance records at national antenatal clinics is used to gather information on HIV prevalence: although this is biased in as far as it does not capture information on men or non-child bearing women, it enables capture of comprehensive information on new cases of the disease. In addition, it allows the opportunity to strengthen seronegativity among pregnant women.

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 an estimated 1.6 million in 2005 and 2.1 million in 2012 (FMOH, 2007; NACA, 2012).

In 2005 the WHO reported complex interactions between nutrition and HIV/AIDS. Evidence highlights that undernutrition weakens the immune system, and in those living with HIV/AIDS in particular, this increases their susceptibility to the disease. Evidence from author groups including USAIDS (2004); Piwoz (2004); Drain et al. (2007) and Ivers (2014) indicates that the prevalence of macro and micronutrient (magnesium, selenium, zinc, vitamin C) deficiencies impacts negatively on optimal immune function, by a progressive depletion of the CD4 T-lymphocyte cells thus increasing susceptibility to morbidity and mortality among PLWH. Evidence also indicates that most HIV nutrition intervention programmes focus on the use of single or two synthetic micronutrients to delay the progression of the disease (Fawzi et al., 2005; Drain et al., 2007).

Given the economic instability evident in Nigeria, there is an inherent difficulty for drug manufacturing industries to purchase sufficient resources to manufacture and distribute synthetic micronutrients. More so, where synthetic micronutrients are available, they are not cost effective, less bioavailable and may not be

sustainable in the developing countries like Nigeria, compared to the natural micronutrients (Thiel, 2000).

It must be acknowledged that individual nutrients have been identified in the literature for their positive impacts on CD4 cell count in HIV infected individuals (Fawzi et al., 2004; Kaiser et al., 2006). Potential effects of nutrient supplement on the anthropometric profiles of HIV-positive patients and micronutrients supplementation in HIV- 1 disease progression are also documented in HIV management (Baum et al., 1995; Oguntibeju et al., 2008). However, there is a clear 'effect' of selected nutrients on outcome; we consume food, not nutrients.

Modifications to diet are required, and have variable effects (increasing intakes of fat may be offset reductions in sugar intakes to maintain energy balance) cannot be expected to have huge effects on outcome but additively given its acknowledged ability to impact various aspects of HIV pathology. These modifications have potential for positive effect for example: micronutrients attenuate HIV type 1 disease progression among adults and children (Fawzi, 2003); nutritional interventions for reducing morbidity and mortality in people with HIV (Mahlungulu, 2009). The concern is that in Nigeria, this cannot be financed unless the approach is cost effective and sustainable. For the purpose of this thesis, the concept of a naturally occurring, accessible and affordable nutrition intervention that demonstrates physiological and biochemical benefits in reducing the progression of HIV disease was investigated.

1.1 Background to HIV

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 (WHO, 2006; AIDS, 2009; WHO, 2013; CDC, 2014). Acquired immunodeficiency syndrome (AIDS) is a progressive deterioration of the immune status of the individual. It is characterised by the progressive depletion of the CD4 T-lymphocyte population (*cells that produces a specific immunity to a particular antigen*), which represents a major target of viral infection by the causative HIV (Table 1.1) (Vajpayee et al., 2005). In 2006, World Health Organization (WHO) provided a simplified HIV case definition designed for reporting and surveillance (Box 1.1).

Table 1. 1: Centre 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 generalised 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, extrapulmonary cryptococcosis, CMV, HIV-related encephalopathy, Kaposi's sarcoma, wasting syndrome due to HIV.

In Nigeria, CD4+ cell counts in healthy individuals have been found to range from 324 - 1160 cells/mm^3 of blood (FMOH, 2007). In Western countries, the mean value reported to be 1,000 - 1,100 cells/mm^3. Thus constant exposure to a number of other pathogens in sub-Saharan Africa may result in an overall less healthy immune system to fight HIV infection (FMOH, 2007).

Box 1.1: ***WHO case definition for HIV infection (WHO, 2006)***

Adult and children 18 months or older:

- HIV infection is diagnosed based on: Positive HIV antibody testing (rapid or laboratory based enzyme immunoassay). This is usually confirmed by a second HIV antibody test (rapid or laboratory – based enzyme immunoassay) relying on different operating characteristics

and/or;

- A positive virological test for HIV or its components (HIV – RNA or HIV – DNA or ultrasensitive HIV p24 antigen) confirmed by a second virological test obtained from a separate determination.

Children younger than 18 months

- HIV infection is diagnosed based on: A positive virological test for HIV or its components (HIV – RNA or HIV – DNA or ultrasensitive HIV p24 antigen) confirmed by a second virological test obtained from a separate determination taken more than four weeks after birth.

and/or;

- Positive antibody testing is not recommended for definitive or confirmatory diagnosis of HIV infection in children until 18 months of age.

1.2 Global epidemiology of HIV disease

At the end of 2010, an estimated 34 million people were living with HIV worldwide up by 17% from 2001(Figure 1.1) (UNAIDS, 2011). This reflects the continued large number of new HIV infections and a significant expansion of access to antiretroviral therapy, therefore declining mortality rates. Globally, there were 2.7 million new HIV infections in 2010, including an estimated 390,000 among children. However, sub-Saharan Africa (SAA) accounted for 70% of new HIV infections in 2010 (UNAIDS, 2011).

In many countries surveyed in SSA, more than half the people estimated to be living with HIV are not aware of their HIV status (WHO, 2013). Furthermore, in 2013, 67% (62-73%) of the estimated 1.4 (1.3-1.6) million pregnant women living with HIV in low and middle income countries received effective antiretroviral drugs to avoid transmission to their children while the remaining 33% had no access to ART (WHO, 2013). WHO (2013) also reported that close to half the people who test HIV-positive in SSA were lost between testing and being assessed for eligibility of ART and 32% of the people considered eligible were lost between being assessed for eligibility and initiating ART.

In WHO, UNAIDS and UNICEF (2011) report, AIDS had become one of the leading causes of adults dying in SSA and the full onslaught of the epidemic

could not be felt until 2006, when more than 2.2 million people died each year from AIDS-related causes such as undernutrition, tuberculosis and other opportunistic infections.

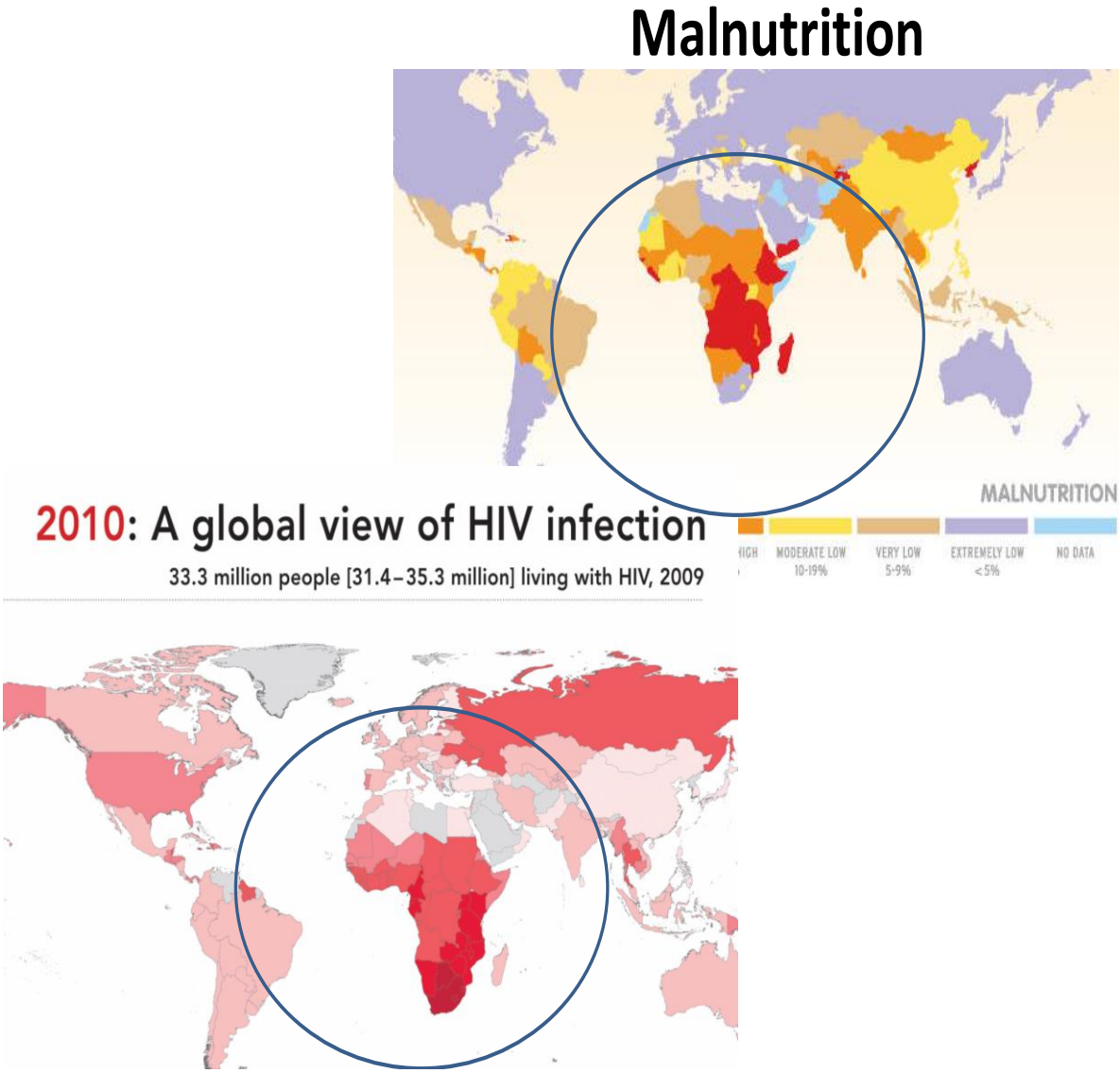


Figure 1. 1: A global view of HIV infection and malnutrition. (Source: UNAIDS, 2010; FAO, 2010)

Sixty one percent of people infected with HIV in SSA are women and several population based survey in Africa have found extremely high rates of infection amongst young women e.g.: The HIV infection rate among women between 25 and 29 years in rural South Africa was estimated at 5.1% (Pribram, 2011; NACA, 2012). HIV remains the single greatest cause of death in SSA and the disease is responsible for more than 20% of death in the region (Pribram, 2011). Globally, Nigeria ranks second most affected by HIV/AIDS after South Africa (Table 1.2) (CIA, 2012). Nigeria is among the 15-focus countries, which collectively

represent 50% of HIV infections worldwide. Although the HIV prevalence of approximately 3.1% appears relatively low compared with other countries in SSA, it nevertheless translates into over 3.4 million people infected with HIV in Nigeria (UNAIDS, 2009 and 2010; CIA, 2012).

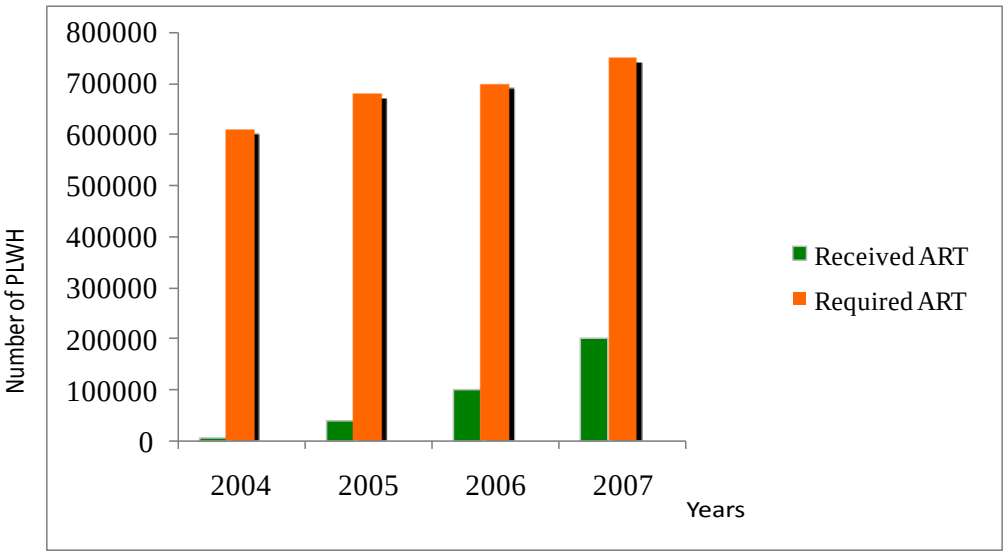
1.3 HIV disease progression in Nigeria

The first case of HIV in Nigeria report was in 1986. Since then the number of people living with HIV or AIDS (PLWHA) steadily increased and the epidemic moved into a 'generalised' state with an increase in seroprevalence from 1.8% in 1991 to 5.8% in 2001 (Figures 1.3 & 1.4) (FMOH, 2007, NACA, 2012). With initiation of some national intervention programmes (employing mainly ART: Lamivudine, Nevirapine, Zidovudine, Tenofovir and Efavirenz) there was slight drop by 0.8% which was recorded in 2003 that was sustained in 2005 with a seroprevalence rate of 4.4% (UNAIDS and WHO, 2009).

Table 1. 2: Estimate of all people (adults and children) alive at year end with HIV infection (CIA World Factbook 2012).

Rank	Country	HIV/AIDS People Living With HIV/AIDS
1	**South Africa**	6,070,800
2	**Nigeria**	3,426,600
3	**India**	2,085,000
4	**Kenya**	1,646,800
5	**Mozambique**	1,554,700
6	**Uganda**	1,549,200
7	**Tanzania**	1,472,400
8	**Zimbabwe**	1,368,100
9	**United States**	1,200,000
10	**Malawi**	1,129,800

In 2007, it was estimated that 200,000 people were receiving antiretroviral therapy out of the approximately 750,000 PLWH that required antiretroviral treatment (CD4 count is <350 cells/mm^3). This figure represented about 27% of all the people who need the treatment leaving 73% to suffer and die without medical intervention. This presents a serious challenge to the Government of Nigeria (GON) and its health programmes (Figure 1.2). Between 2005 and 2009, the AIDS related deaths fell from 220,000 to 170,000 while those orphaned as a result of the disease rose from 1.6 million in 2005 to 2.1 million in 2012. Estimates also show a cumulative death of 2.82 million people as at 2010 (FMOH, 2007; NACA, 2012).



PLWH = People living with HIV
ART = Anti Retroviral Treatment

Figure 1.2: Number of PLWH who received ART against those who needed it (Source: FMOH, 2007; UNAIDS, 2009).

Many factors are responsible for only 200,000 receiving ART in 2007, such as: quality of diagnosis, accurate prescribing, selection, distribution, and dispensing of ARVs. One of the most significant barriers to access is the price of ARVs. Currently, in most developing countries like Nigeria, high price of second line ARVs condemn people with AIDS to a premature death.

NACA (2012) also reported the trend of HIV across age groups in male and female. The prevalence rate of 5.7% (female) and 5.1% (male) between the age group of 30 – 39 years is an indication of the impact of the disease on Nigeria work force (Figure 1.4).

The impact of the political will in Nigeria have direct repercussions on the price of ARVs. Countries which imports ARVs (e.g. South Africa, Nigeria) have much higher prices than countries like Brazil and Senegal. Brazil settled a national protocol on HIV treatment while Senegal negotiated the Accelerated Access Initiative. The initiative by Brazil and Senegal proved that the price of ARVs can be brought at affordable level for the population.

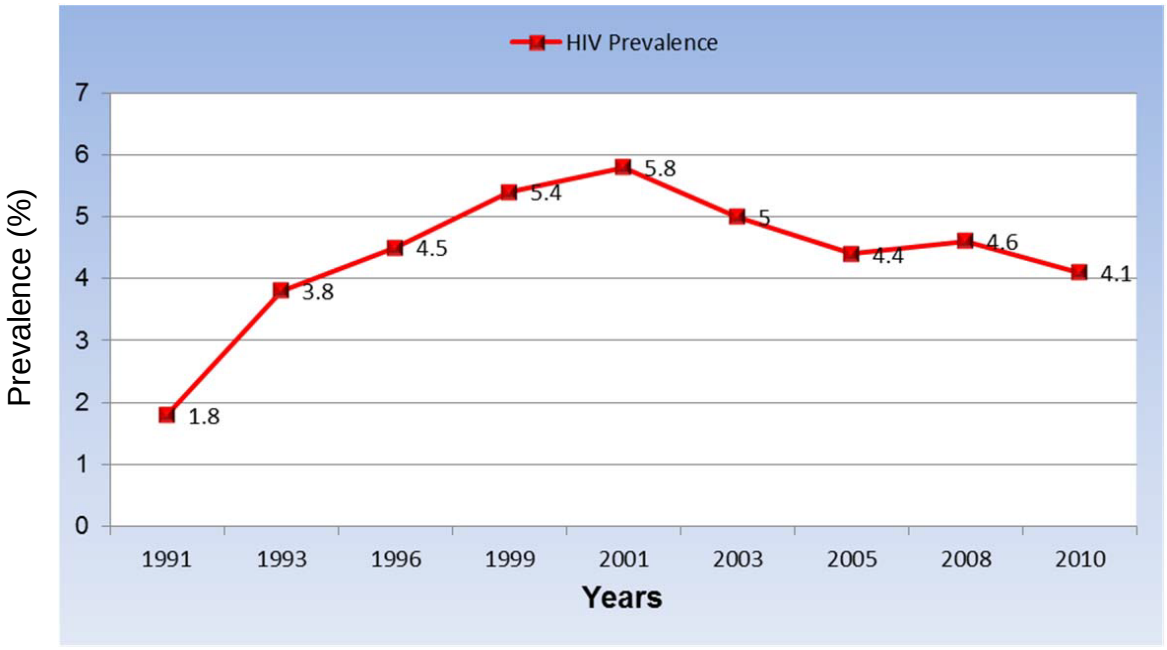


Figure 1. 3: National HIV Prevalence Trend (1991 - 2010). Source: National Agency for Control of AIDS, 2012.

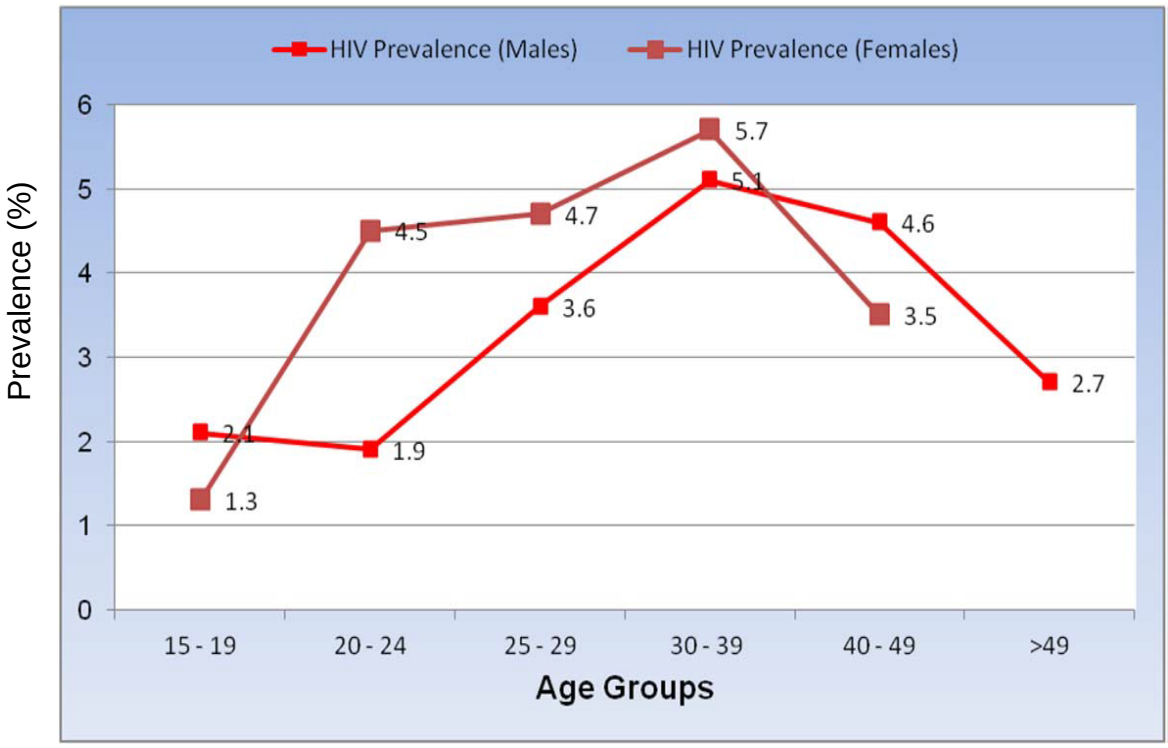


Figure 1. 4: National HIV Prevalence Trend across age groups (years) in male and female PLWHIV in Nigeria. Source: National Agency for Control of AIDS, 2012.

1.4 HIV/AIDS as a public health problem in Nigeria

In Nigeria, the high burden of the disease with its associated morbidity and mortality continues to constitute a major public health concern for the country (Table 1.3 – 1.5) (FMOH, 2007).

Table 1. 3: Cumulative HIV deaths in Nigeria (FMOH, 2005; FMOH, 2007)

	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 HIV/AIDS epidemic has further weakened and threatened to overwhelm the Nigerian health care system. Additionally, it increased the number of orphans and increased the cost of achieving set developmental goals by decreasing the size of the workforce, affecting as it does, mainly adults in their most productive years of life (15-60 years). The high manpower-intensive sectors of the economy are most affected; in Nigeria this includes the agricultural, educational and health sectors as well as the rural economy (Table 1.4) (FMOH, 2007; FMOH, 2010).

Table 1. 4: HIV/AIDS Burden: Socio-economic implications in Nigeria (FMOH, 2007)

	2005	2010
Number of children orphaned due to AIDS	1,640,000	2,680,000
Number of orphans	7,820,000	9,130,000
Number of HIV positive pregnant women	227,900	223,300
Life expectancy	47 years	43.4 years

Interestingly, Stillwagon (2002), in her pioneering work, found falling calorie and protein consumption to be strongly correlated (R-squared is 0.545 or 0.517) with HIV prevalence in 44 SSA countries. The more severe the decrease in nutrition and the more unequal the distribution of income in a country, the higher is the rate of HIV. In Stillwagon's regression analysis, the prevalence of HIV is strongly correlated with income inequality, falling protein consumption, falling calorie consumption. Similarly, Fawzi et al. (2004) proposed that multivitamin supplements (Vitamins A, B, C, D and E) as low-cost immunomodulating interventions may slow the progression of HIV disease by significantly (P=0.02) increasing CD4+ and CD8+ cell counts and significantly decreasing (P=0.003) viral loads. Numerous studies also reported micronutrient deficiencies impair responses, weaken epithelial integrity, and are associated with HIV disease progression (Tang and Smit, 1998; Jiamto et al., 2003; Gillespie and Kadiyala, 2005; Kaiser et al., 2006 and Drain et al., 2007). Therefore, this research was intended to demonstrate the efficacy of macro and micro- nutrients derived from

food sources in Nigeria to delay the onset of advanced HIV disease and the need for antiretroviral therapy, saving antiretroviral drugs for when they may be most needed and reducing drug-related adverse events and costs.

In summary, the impact of HIV/AIDS on Nigeria's social fabric and on its economic development and well-being country to be pervasive and, unless controlled through multidimensional approaches to include nutritional intervention as proposed in this research, will continue to undermine the quality of life of Nigerians (Table 1.5) (FMOH, 2007; FMOH, 2010).

Table 1. 5: Contribution of HIV/AIDS to morbidity in Nigeria (FMOH, 2007)

S/N	Disease conditions	Contribution (%)
1	HIV/AIDS	16.0%
2	Respiratory diseases	14.0%
3	Malaria	11.0%
4	Cardiovascular diseases	10.0%
5	Childhood diseases	9.0%
6	Diarrhea diseases	7.0%
7	Injuries (Road accidents, drowning, violence)	7.0%
8	Prenatal conditions	4%
9	Others (cancer, urinary diseases, TB, etc)	22%

1.5 Research Aim and objectives

1.5.1 Aim

To design and evaluate a public health-nutrition intervention programme which aims to delay the progression of human immune-deficiency virus (HIV) to AIDS (*acquired immune-deficiency disease syndrome*) among people living with HIV in Abuja, Nigeria.

1.5.2 Objectives

The objectives of the study are:

1. To assess the quality and quantity of specific macro/micronutrients in local vegetables and grains consumed by People Living with HIV/AIDS and compare these values with age-specific recommended intakes
2. To design and optimise a nutritionally functional meal (Amtewa *meal*) that contains known concentrations of macro and micronutrients to be employed in the public health nutrition intervention.

3. To assess the nutritional intake and biomedical indices prior and post the pilot intervention (N=100) (baseline vs. end point; with follow-up at three and six months interval)
4. To appraise the outcomes of Amtewa meal on health status of HIV infected subjects (with CD4 values above 200/mm^3) at baseline and compare these at post-pilot and larger scale intervention (N= 400)
5. To evaluate the effectiveness of macro and micronutrient interventions as possible measure to attenuate the progression of HIV to AIDS.
6. Draw up a sustainability plan for outcome of discussion and conclusion

Chapter 2

Literature Review

2.0 Introduction

This chapter includes the current knowledge and substantive findings in the literature, as well as theoretical and methodological contributions to HIV/AIDS disease and the symbiotic relationship between HIV and nutrition. It explains and justifies how public health nutrition intervention may help answer some of the questions or gaps in this area of research in Nigeria.

2.1 Classification and Structure of HIV

The aetiology of AIDS has been identified as HIV-1 and HIV-2 (FMOH, 2007; AIDS, 2012). These viruses belong to the Lentivirus group and Retroviridae family (Figure 2.1). The Family Retroviridae of viruses includes three sub-families: Oncovirinae, Lentivirinae and Spurnavirinae. All the members of the family contain an enzyme called reverse transcriptase that is used for the synthesis of proviral DNA from the infecting viral RNA. The provirus (also called the complimentary or cDNA) integrates itself into the chromosome of the host cell with the aid of additional enzymes encoded by the viral *pol* gene.

The integration of the viral cDNA into the host cell genome serves as the basis for the continuous viral replication characteristic of retroviruses as well as the unconventional (reverse) method of transcription by these groups of viruses than the conventional flow (transcription) of the genetic information from DNA to RNA. This group of Retroviruses is associated with many diseases including rapid and long latency, malignancies, wasting disease, neurological disorders, immunodeficiency as well as long viraemia in the absence of any obvious clinical disease (FMOH, 2007).

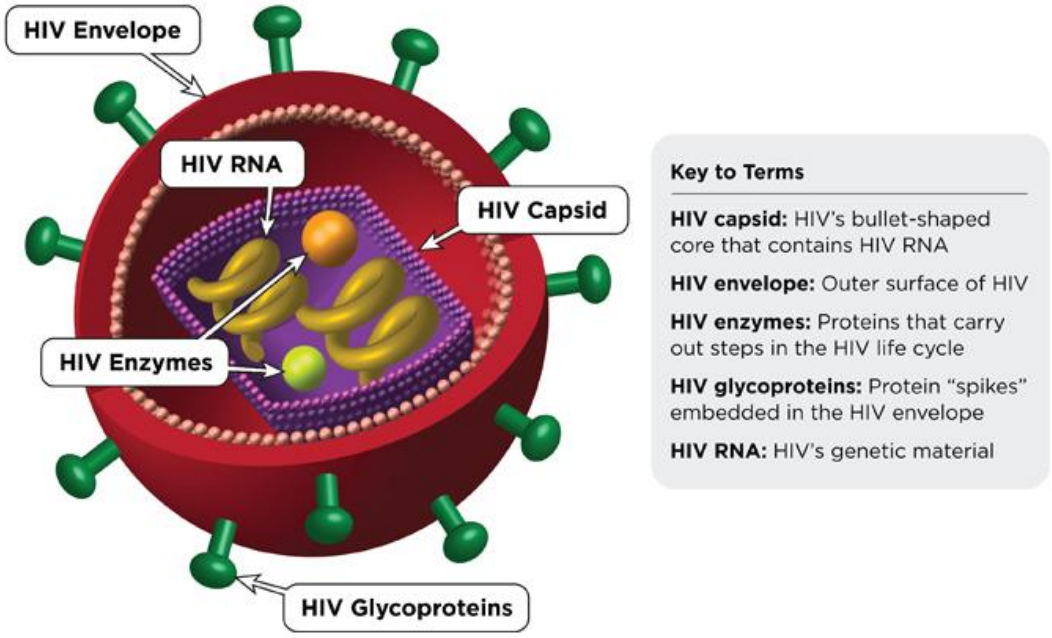


Figure 2. 1: Structure of HIV. (Source: AIDS info., 2014)

The HIV particle contains three components: the core, the surrounding protein matrix and the outer lipid envelop. The core contains genetic material, RNA, encapsulated by the capsid protein p24, which contains enzyme (reverse transcriptase, integrase and protease) involved in viral replication. The glycol proteins gp41 and gp120, which is attached to the envelop enable HIV to bind and fuse with target host cells (Pribram, 2011).

2.2 Cellular Receptors of HIV

The primary receptor for HIV is the CD4 molecule on the human T-helper cells. CD4 lymphocyte cells (also called T-cells or T-helper cells) are the primary targets of HIV. The CD4 count and the CD4 percentage mark the degree of immune compromise. The CD4 count is the number of CD4 cells per micro liter (µL) of blood. It is used to stage the patient's disease, determine the risk of opportunistic illnesses, assess prognosis, and guide decisions about when to start antiretroviral therapy (ART) (HRSA, 2011). Variation in CD4 receptor molecule on T-cell surface may influence the ability of HIV to bind and eventually penetrate the target cell (AIDS, 2013). In addition, attachment to and fusion with the target cells is determined not only by its binding with CD4 molecules, but also other secondary binding sites known as Beta chemokine such as CCR5 and CXCR4.There are several reports to show that individuals who are homozygous for a deletion in the CCR5 or CXCR4 gene are less frequently infected with HIV,

whereas individuals who are heterozygous for the same mutation become infected but can be protected against rapid progression to disease compared with infected individuals homozygous for the normal CCR5 or CXC gene (FMOH, 2007). Because HIV infects CD4 cells and uses them to produce more HIV copies, HIV infection is characterised by a progressive fall in the number of T-helper/inducer CD4 positive cells (Pribram, 2011). Helper T cells are clearly critical to the operation of the immune system. If they are destroyed because of an HIV infection, the whole system is crippled. The immune system is described as having two “arms”: the **cellular** arm, which depends on T cells to mediate attacks on virally infected or cancerous cells; and the **humoral** arm, which depends on antibodies to clear antigens circulating in blood and lymph. As an HIV infection progresses, destroying helper T cells, both arms of immunity are impaired.

2.3 Natural History of HIV infection

The course of HIV infection varies within a population (FMOH, 2007). Nonetheless, a typical infection can be divided into three stages: primary (acute) infection, asymptomatic (clinical latency) stage and symptomatic (AIDS) stage.

Only certain fluids—blood, semen (*cum*), pre-seminal fluid (*pre-cum*), rectal fluids, vaginal fluids, and breast milk—from an HIV-infected person can transmit HIV. These fluids must come in contact with a mucous membrane or damaged tissue or be directly injected into the bloodstream (from a needle or syringe) for transmission to possibly occur (Figure 2.2). Mucous membranes can be found inside the rectum, the vagina, the opening of the penis, and the mouth (AIDS, 2009; CDC, 2014).

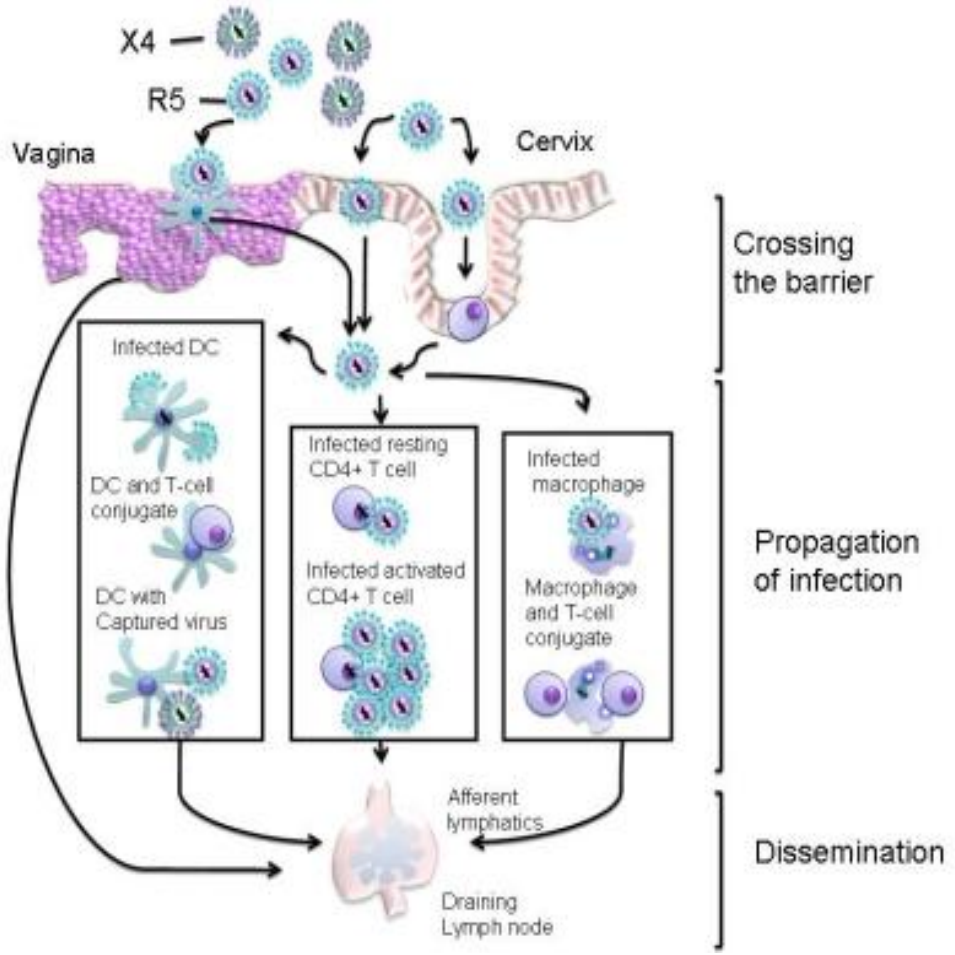


Figure 2. 2: HIV Transmission (Source: Pope & Haase, 2003)

During early infection, HIV remains concentrated in the lymph nodes, where it replicates in huge numbers and infects more CD4 T cells. Swollen lymph nodes are often the only clinical feature seen in a person with HIV infection for the first months or years of infection. The immune system gradually deteriorates to the point that the human body is unable to fight off other infections. The HIV viral load in the blood dramatically increases while the number of CD4+ T cells drops to dangerously low levels (Figure 2.3). An HIV-infected person is diagnosed with AIDS when he or she has one or more opportunistic infections, such as pneumonia or tuberculosis, and has fewer than 200 CD4+ T cells per cubic millimeter of blood (WHO, 2006; NIH, 2011).

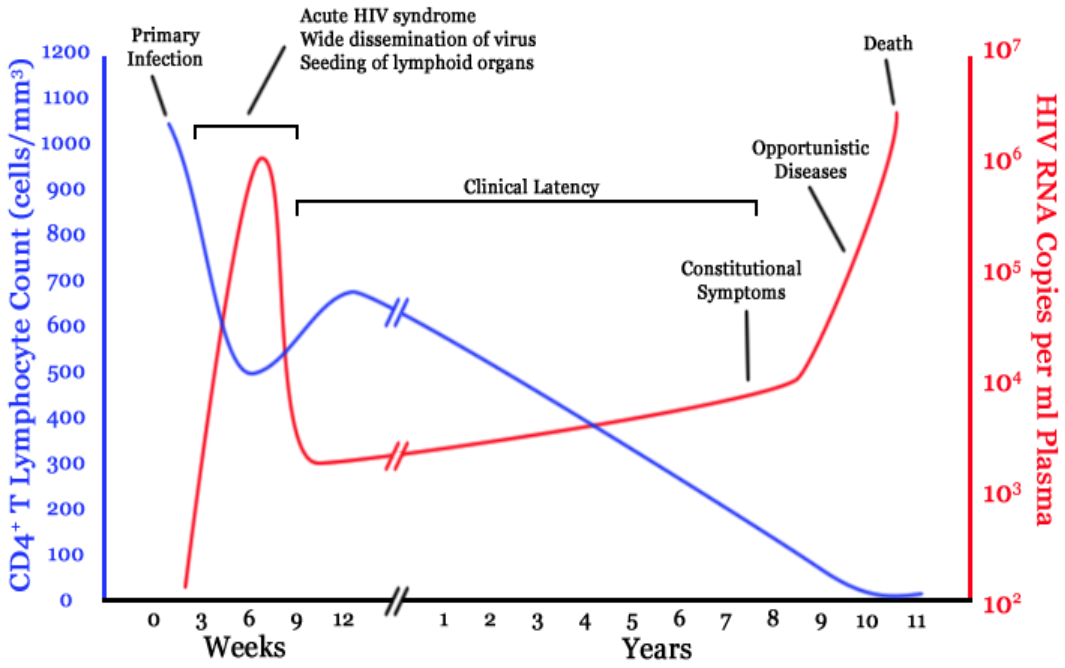


Figure 2. 3: Clinical progression of HIV to AIDS (Source: The naked scientist, 2007).

2.3.1 Acute infection stage

Within *2-4 weeks* after HIV infection, many, but not all, patient develops flu-like symptoms, often described as "the worst flu ever." Symptoms can include fever, swollen glands, sore throat, rash, muscle and joint aches and pains, fatigue, and headache. This is called "acute retroviral syndrome" (ARS) or "primary HIV infection," and it's the body's natural response to the HIV infection. During this early period of infection, large amounts of virus are being produced in the body. The virus uses CD4 cells to replicate and destroys them in the process. Because of this, theCD4 count can fall rapidly. Eventually the immune response will begin to bring the level of virus in the body back down to a level called a *viral set point*, which is a relatively stable level of virus in the body. At this point, the CD4 count begins to increase, but it may not return to pre-infection levels (WHO, 2006; AIDS, 2013).

2.3.2 Clinical latency stage

After the acute stage of HIV infection, the disease moves into a stage called the "clinical latency" stage. "Latency" means a period where a virus is living or developing in a person without producing symptoms. During the clinical latency stage, people who are infected with HIV experience no HIV-related symptoms,

or only mild ones. (This stage is sometimes called “asymptomatic HIV infection” or “chronic HIV infection”). During the clinical latency stage, the HIV virus continues to reproduce at very low levels, although it is still active. Patients on ART may live with clinical latency for several decades because treatment helps keep the virus in check. For those who are not on ART, the clinical latency stage lasts an average of 10 years, but some people may progress through this stage faster. Therefore intervention means to delay /attenuate the progressions of latency stage through adequate nutritional assistance warrant research investigation by health authorities (WHO, 2006; Pribram, 2011). It is important to remember that patients in this symptom-free stage are still able to transmit HIV to others, even if they are on ART, although ART greatly reduces the risk of transmission.

2.3.3 AIDS Stage

This is the stage of HIV infection that occurs when the immune system is badly damaged and become vulnerable to infections and infection-related cancers called *opportunistic infections*. When the number of your CD4 cells falls below *200 cells per cubic millimeter of blood (200cells/mm^3)*, patients are considered to have progressed to AIDS. In someone with a healthy immune system, CD4 counts are between 500 and 1,600 cells/mm^3. A patient is also considered to have progressed to AIDS if he/she develops one or more opportunistic illnesses, regardless of the CD4 count (WHO, 2006). Without treatment, patients who progress to AIDS typically survive about three years. Once you have a dangerous opportunistic illness, life-expectancy without treatment falls to about one year. However, if a patient is taking ART and maintains a low viral load, then he/she may enjoy a near normal life span and will most likely never progress to AIDS (WHO, 2006).

Despite repeated exposure, some individuals never become infected with HIV. These individuals often have unusual helper T cells with a less-efficient variant of the co-receptor CCR5, which is necessary for viral entry into helper T cells (FMOH, 2007). There are also individuals who become infected, but do not progress to AIDS. These long-term survivors, or long-term non-progressors (LTNP), include individuals who have been AIDS-free as long as eighteen years after infection (FMOH, 2007; Praveen, 2013). Five percent of total HIV

populations are LTNP (Praveen, 2013). A variety of factors may be responsible; for example, infection with less-virulent viruses. Some long-term non-progressors seem to have CD8 cells, which are particularly adept at curtailing HIV infection (FMOH, 2007; Praveen, 2013).

2.4 Replication of HIV

Replication of the virus particle begins with attachment of gp 120 to the CD4 on the surface of a target cell (Figure 2.4). Following the gp 120-CD4 binding, a structural change allows for the interaction of the V3 loop region in the gp 120 with a chemokine receptor, including CCR5 and CXCR4 (WHO, 2006; FMOH, 2007).

The reaction with the co-receptor results in another conformational change in the viral surface glycoprotein, which exposes a fusion domain contained within the envelop trans-membrane glycoprotein. Exposure of the fusion domain results in the insertion of the gp41 into the cellular membrane. Subsequent to the fusion event, the viral core is released into the cytoplasm of the host cell. Once in the cytoplasm, the viral RNA genome is uncoated and reverse transcribed by the virally encoded Reverse Transcriptase (RT) enzyme to generate a double-stranded viral DNA pre integration complex (FMOH, 2007).

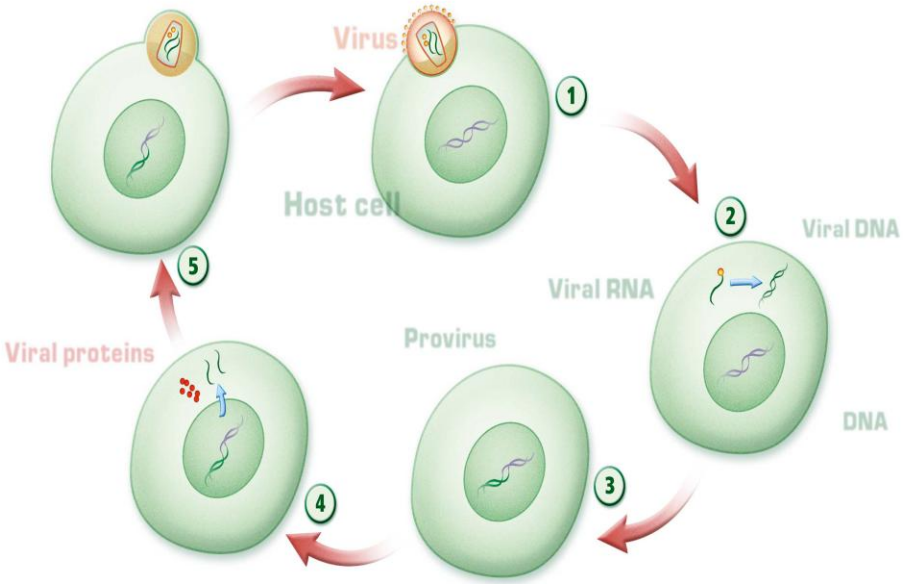


Figure 2. 4: HIV Replication cycle

The double stranded DNA is then transported into the host cell nucleus and via catalysis by integrase, becomes integrated into the host cell chromosome, where it resides as provirus. Once the viral genome has been integrated into the host cell genome, it can remain in the latent state for many years or can begin the production of new viral RNA. If the host cell is activated, the host cell enzyme RNA polymerase II will transcribe the proviral DNA into messenger RNA (mRNA). The mRNA is then translated into viral proteins that undergo extensive post-translational modifications. The viral RNA becomes the genetic material for the next generation of viruses. Viral RNA and viral proteins assemble at the cell membrane. After proper assembly and processing, new infectious virus particles are released by budding from the cell membrane (FMOH, 2007; FMOH, 2010).

2.5 Variability of HIV isolates

The human immunodeficiency virus exhibits marked genetic diversity among different isolates. This heterogeneity is distributed throughout the viral genome and most of it is located in the *env* gene. Based on this variability, HIV has been classified into types 1 and 2. HIV 1 has a global distribution while HIV -2 is limited to West Africa. Nevertheless, HIV-1 is still the predominant type in this sub-region. The 2 sub-types of the virus also vary in their biological characteristics. The rate of transmission (sexually and MTCT) as well as progression to disease of HIV-1 is faster than that of HIV-2.

HIV-1 isolates are classified into subtypes using the nucleotide sequence of the *gag, pol* and *env* genes. Subtypes of HIV-1 isolates using the env gene has been based mainly on the third variable region (V3 loop) of the Gp 120, known to be important in viral cell type tropism, virus cell fusion and cytopathology. Based on the nucleotide of the V3 loop of the Gp 120, HIV-1 has been classified into three major groups, the group M (major), N (non-M, non-O) and group O (Outlier). The group M virus has been further classified into at least 11 different subtypes (A to K). In addition, recombination of the virus subtypes is a well known phenomenon and many recombinant forms including the A/G which is the most predominant subtype in West Africa have been identified. In Nigeria, the A/G and the G subtypes are predominant. HIV-2 is classified into 5 subtypes, A-E and recombinant forms have not been identified (FMOH, 2007).

Co-infection or super-infection of an individual by HIV-1 and HIV-2 has been well documented (FMOH, 2007). Infection of an individual with different HIV-1 and

HIV-2 subtypes has also been identified. This large number of variants makes the virus more difficult to treat and hinders vaccine development.
In addition, because of its rapid rate of evolution, even within a single individual, HIV can quickly evolve resistance to the drugs the individual is taking to combat the virus.

2.6 Laboratory diagnosis and HIV Counselling and testing

Laboratory diagnosis of HIV infection is based on the demonstration of antibody in plasma or serum, and a virus in the blood. The virus can be demonstrated in the blood with nucleic acid-based test (PCR for proviral DNA and RT-PCR for plasma viral RNA), culture and p24 antigen assay. HIV antibodies are detectable within four to six weeks of infection, and within 24 weeks in virtually all infected individuals. However, virus can be detected in plasma at least one week earlier. The period of absent antibody in the presence of virus in the plasma is called the "window period".

2.6.1 Antibody Assays

The antibody assays that are used for HIV diagnosis consist of screening tests: rapid tests or ELISA, and confirmatory tests: Western blot and indirect immune fluorescent assay. Routine antibody testing is performed with the serial or parallel testing algorithm using rapid or ELISA test kits

Rapid Rests

Rapid tests are suitable for use in laboratories that have limited facilities and process few samples. They are technically simple to perform, do not require any major equipment, but have a sensitivity and specificity comparable to ELISA. This commonly used rapid antibody tests in laboratories are based on the principles of dot immunoassay, or particle agglutination (e.g. gelatin or latex).

Enzyme linked immunosorbent Assay (ELISA)

The ELISA procedure is carried out to screen for HIV IgG antibodies in plasma or serum. The principles of ELISA are classified as direct, competitive and sandwich test. The competitive principle is not popular because of the low sensitivity. Antigens derived from HIV grown in human T-lymphocytes or recombinant proteins or synthetic peptides are used to coat beads or microtitre plates. To perform an ELISA, the patient's serum is incubated with the antigens

on the beads or in the microtitre plates. A conjugate, i.e., enzyme-labelled antibody specific for human immunoglobulin is then added. Detection of the enzyme-labelled antibody is carried out by the addition of a substrate that produces a colour reaction. ELISA requires a plate washer and ELISA Reader and printer. It is a suitable test for use in the laboratories where large numbers of samples are tested each time (FMOH, 2007).

Western Blot

Western blot is the standard confirmatory test for HIV antibody assays. The test utilizes HIV antigens from purified viruses that have been electrophoretically separated and “blotted” (transferred) onto a nitrocellulose paper. The paper is then cut into strips, each containing all the separated HIV antigens. To carry out a Western blot assay, a strip is incubated with the patient’s serum. Antibody in the serum binds to the HIV antigens on the strip, and is detected with the aid of a conjugate consisting of labeled anti-human immunoglobulin. Studies have shown that combinations of ELISAs or rapid assays can provide results as reliable as the Western blot at a much lower cost (FMOH, 2007).

Indirect Immunofluorescent Antibody Assay (IFA)

IFA employs HIV-infected cells (lymphocytes) fixed to the microscope slide. The patient serum is added and reacts (if antibody is present) with the intracellular HIV antigen. After washing the slide, a conjugate consisting of anti-human immunoglobulin labeled with FITC added, and the reaction is visualised under a fluorescent microscope. IFA has been used to confirm diagnosis in sera producing indeterminate results in Western blot (FMOH, 2007).

Serial testing

Algorithm refers to the use of 2 screening tests employed sequentially to test for HIV antibody. If the initial screening is negative, no further testing is required. If the initial test is positive, it is followed by one more test. The first test should be the most sensitive test and the second test should be very specific, and be based on an antigen source different from that of the first test. Samples that produce discordant results in the two tests are subjected to further testing.

Parallel testing

Involves two screening tests performed simultaneously. Samples reactive to both tests are regarded as positive. However, those with discordant results require further testing. Parallel testing is performed to minimize the chances of false negative results and to guard against technical errors. It is often used when a very sensitive test is not available for the initial screening, and when the concordance of two tests is to be evaluated.

2.6.2 Nucleic Acid-based Tests

These consist of DNA Polymerase Chain Reaction (DNA PCR) and reverse transcriptase Polymerase Chain Reaction (RT-PCR). These tests are not routinely used for laboratory diagnosis of HIV infection in adults and adolescents in Nigeria (FMOH, 2007).

HIV DNA Polymerase Chain Reaction

The DNA PCR involves the amplification of specific DNA sequences in the proviral DNA that has been integrated in the host cell. This test is the preferred procedure for diagnosing HIV infection in infants less than 18 months of age. Due to the high sensitivity of the test, false positive results may occur as a result of contamination by minute quantities of extraneous DNA.

RT – PCR

RT-PCR is used to detect and quantify the amount of HIV RNA in plasma. The assay requires the conversion of viral RNA to DNA and amplification of specific sequences in the DNA produced by a process known as reverse transcriptase polymerase chain reaction (RT-PCR).

2.6.3 Other Tests

These tests are not routinely used for laboratory diagnosis of HIV infection.

Antigen detection

Detection of p24 antigen is an ELISA-based test. The reliability of the test is in doubt because of specificity and sensitivity problems.

Virus isolation

HIV is usually isolated in PBMCs. The procedure involves co-cultivating the PBMCs from a patient with those obtained from a healthy donor. HIV isolation in PBMC is quite sensitive and is comparable to DNAPCR in sensitivity.

2.7 Antiretroviral Therapy

Standard antiretroviral therapy (ART) consists of the combination of at least three antiretroviral (ARV) drugs to maximally suppress the HIV virus and stop the progression of HIV disease (WHO, 2014). Huge reductions have been seen in rates of death and suffering when use is made of a potent ARV regimen, particularly in early stages of the disease.

2.7.1 Pre-treatment evaluation

This includes:

- Complete history and physical examination
- Clinical and Immunological classification of the patient
- Check Laboratory results (FBC with differentials, ALT, Creatinine, CD4+ cell count, pregnancy test
- Evaluation of nutritional and psychosocial status
- Assessment of readiness for therapy
- Development of patient-specific adherence strategy

2.7.2 Criteria for initiation of ART

Initiation of therapy depends on CD4+ cell count and WHO clinical staging (Table 2.1)

- WHO Stage IV disease irrespective of CD4+ cell count
- WHO Stage III disease with CD4+ cell counts <350/mm^3
- WHO Stage I or II disease with CD4+ cell count =200/mm^3
- Any WHO Stage with CD4+ cell count 200 – 350 /mm^3, consider therapy.

Table 2. 1: WHO Adult HIV Clinical Staging

	Stage 1 Asymptomatic	Stage 2 Mild Disease	Stage 3 Moderate Disease	Stage 4 Severe Disease (AIDS)
Symptoms Treat common and opportunistic infections according to Adult Care and/ or guidelines.	No symptoms or only persistent generalised lymphadeno-pathy.	Weight loss 5-10% Sores or cracks around lips, Herpes zoster, Recurrent upper respiratory infections such as sinusitis or otitis, Recurrent mouth ulcers	Weight loss >10%. Oral thrush, More than 1 month diarrhea or unexplained fever, Severe bacterial infections, Pulmonary TB, TB lympadenopathy Acute necrotizing ulcerative gingivitis/ Periodontitis	HIV wasting syndrome, Oesophageal thrush, More than 1 month Herpes simplex ulceration, Lymphoma, Kaposi sarcoma, Invasive cervical cancer, CMV retinitis, Pneumocystic pneumonia, Extrapulmonary TB, Toxoplama brain abscess, Cryptococcal meningitis, Visceral leishmaniasis, HIV encephalopathy.
Prophylaxis		Cotrimoxazole Prophylaxis	Cotrimoxazole prophylaxis Other prophylaxis on treatment plan	Cotrimoxazole prophylaxis Other prophylaxis on treatment plan
ARV therapy	Only if CD4 <200	Only if CD4 <200 or Total lymphocyte <1200/mm^3	Treat if more than one sign or repeated/chronic stage 3 problems or CD4 <200 Evaluate for ART Prepare for adherence	All in stage 4 are medically Eligible for ART Evaluate for ART Prepare for adherence
Suggested action	Nutrition support ‡	Nutrition support ‡	Nutrition support ‡	Nutrition support ‡

‡ Piwoz and Preble, 2000; WHO, 2003; USAID, 2004; WHO, 2005; FANTA/USAID 2007; WHO, 2009; Pribram, 2011

2.8 Nutrition role in HIV

Nutrition has always been an important aspect of HIV care (Pribram, 2011). Similarly, HIV as a pandemic disease its impact is worsened by the presence of other conditions such as under-nutrition and opportunistic infections (Anabwani and Nazario, 2005). HIV/AIDS scourge has had a devastating impact on health, nutrition, food security and overall socioeconomic development in countries that have been greatly affected by the disease. There is a need for renewed focus on and use of resources for nutrition as a fundamental part of the comprehensive package of care at the country level (Anabwani and Nazario, 2005; FMOH, 2007; WHO, 2010; Ivers, 2014).

In human beings, HIV/AIDS and undernutrition form a symbiotic relationship and one increases the prevalence and severity of the other (Bijlsma, 2000; Yale University, 2007; Pribram, 2011; Ivers, 2014). Moreover, despite the effectiveness of highly active antiretroviral therapy (HAART), there is evidence that HIV-related wasting remains an important co-morbidity factor in many patients (FAO/WHO, 2002; Tang et al., 2002; Yale University, 2007; Pribram, 2011; Ivers, 2014).

Micronutrient deficiencies significantly contribute to HIV progression to AIDS; deficiencies of essential vitamins (A, B-complex, C and E) and minerals (selenium and zinc), are common in People Living with HIV (PLWH) and these micronutrients are required by the immune system to combat infection (WHO, 2005; Hurwitz et al., 2007; Duggal, 2012). 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 (USAIDS, 2004; Piwoz, 2004; Drain et al., 2007 and Ivers, 2014).

The effects of under-nutrition on the immune system are well documented and include decreases in CD4 T cells, suppression of delayed hypersensitivity and abnormal B-cell responses (USAIDS, 2004; Drain et al., 2007). Interestingly, the immune suppression caused by protein-energy malnutrition (PEM) mechanism is similar in many ways to the effects of HIV infection in PLWH as illustrated in Figure 2.5 (RCQHC and FANTA, 2003; FANTA, 2004; USAIDS, 2004; Pribram, 2011).

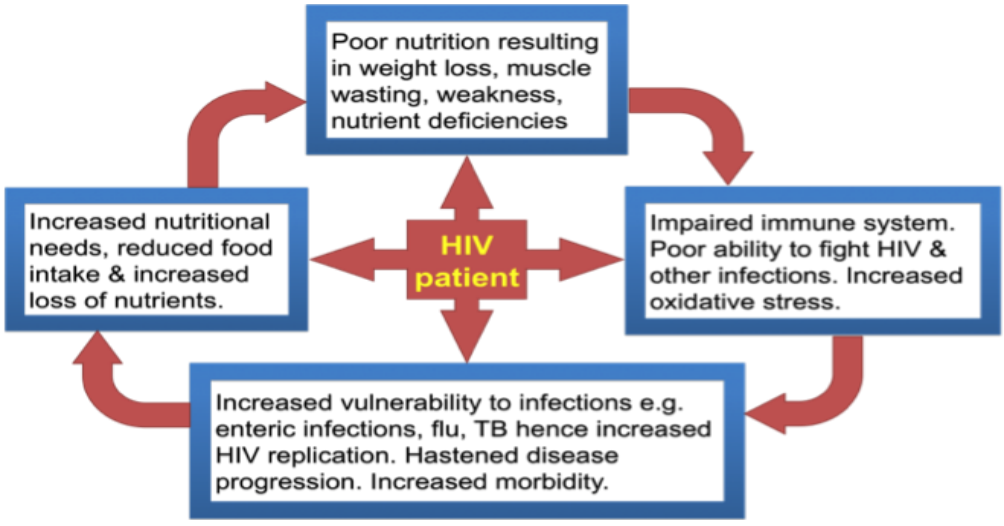


Figure 2. 5: The vicious cycle of malnutrition in the HIV patient (Source: RCQHC and FANTA 2003)

Action and investment to improve the nutritional status of People Living with HIV/AIDS (PLWHA) should be based on sound scientific evidence, local resources, and programmatic and clinical experience with the prevention, treatment, and management of the disease and related infections (USAID, 2004; UNAIDS, 2008; Tewfik et al., 2010; UNAID, 2010; WHO 2010). Although there are gaps in scientific knowledge, much can and should be done to improve the health, nutrition and quality of care for PLWHA and their families and communities (WHO, 2005; Hurwitz, et al., 2007).

An earlier review by Piwoz and Preble (2000) examined preliminary evidence that improving nutrition status may improve some HIV-related outcomes. HIV infection increases energy requirements through increases in resting energy expenditure (REE 12% higher), while reduced food intake, nutrient mal-absorption, negative nitrogen balance and metabolic alterations exacerbate weight loss and wasting, perpetuating the cycle (Melchior et al., 1991; FAO/WHO, 2002; Piwoz, 2004; WHO, 2009). Increase in REE may be due to the production of tumor necrosis factor- alpha and /or interleukin-l in patients with AIDS. However, potential mechanisms of wasting invoked in AIDS patients and their impact on energy intakes (e.g., severe oral and /or esophageal candidiasis, viral esophagitis or extensive oral kaposis sarcoma) are perhaps more relevant. Anorexia can occur in response to systemic infection or mental depression, and intestinal mal-absorption, increased loss of nutrients; muscle wasting and weakness were identified in AIDS (Melchior et al., 1991).

2.9 Current intervention programmes at international level

The number of people dying of AIDS-related causes fell to 1.8 million in 2010, down from a peak of 2.2 million in the mid-2000s. A total of 2.5 million deaths have been averted in low- and middle-income countries (e.g. Nigeria, South Africa) since 1995 due to progresses that resulted from expanded access to antiretroviral therapy, antiretroviral prophylaxis and safe infant-feeding interventions (WHO, 2010; UNAIDS, 2011).

The revised recommendation for antiretroviral therapy (ART) included an earlier start to treatment for all HIV-infected individuals with a CD4-cell count of 350 cells/mm^3 or chronic hepatitis B irrespective of CD4-cell count. They are based on evidence of both individual and public health benefits of starting treatment earlier (WHO, 2010). However, WHO recommends that nutritional care and

support with macro/micronutrients must be started at the early stages of the infection prior to the initiation of ART in order to prevent weight loss and undernutrition (Piwoz and Preble, 2000; WHO, 2009; Pribram, 2011).

2.10 Current intervention programmes in Nigeria

There are four major HIV interventions programmes in Nigeria: HIV counselling and testing (HCT), HIV and AIDS treatment and care in adolescents and adults, paediatric HIV and AIDS treatment and care, prevention of mother-to child transmission of HIV (PMTCT). Current antiretroviral drug treatments in these programmes control HIV infection and prevent severe wasting, as well as other AIDS-related conditions (FMOH, 2007 and 2010). Emaciated people tend to regain weight once they begin treatment. Nevertheless, the drugs do not eliminate wasting.

In addition, some antiretroviral drugs have been linked to lipodystrophy. Whereas HIV-related wasting tends to deplete lean tissue, lipodystrophy involves changes in fat distribution. Nutrition intervention programme may improve strategies to ameliorate these limitations associated with the current HIV intervention programmes in Nigeria.

2.10.1 HIV Counselling and Testing (HCT)

During the past 15 years, the introduction of effective ART and its demonstrated medical benefits has shown the usefulness and importance of expanding HCT services to facilitate early diagnosis and treatment of HIV- infected persons (FMOH, 2007; NACA, 2012). It has also been shown that early knowledge of HIV infection can result in tremendous public health benefits through decreasing risk behaviors that could transmit HIV to uninfected persons. Furthermore, uninfected persons may benefit from HIV testing if knowing their HIV status assists them in modifying or reducing risk behaviours.

All patients undergoing HIV testing must receive pre- and post- test counselling, and give their consent before the test is performed on their specimens. Post- test counselling should be done irrespective of the test result (FMOH, 2007). Testing may be performed without consent if the patient is unable to give his/her consent and the test result is needed in an emergency to provide medical care.

A high level of confidentiality must be maintained during testing (for ethical reason). Careful record keeping is essential to ensure confidentiality. HCT

includes counselling and testing in a variety of settings. Traditional Voluntary Counselling and Testing (VCT), provider- initiated counselling and testing, opt-out counselling and testing are examples of HCT methods. VCT is a client-initiated approach by an individual to find out his HIV status and in the process receive Counselling. Provider-initiated Counselling and testing is done based on the recommendation of the care provider to the client. The opt-out approach occurs where HCT is routinely offered with the patient having the option to decline testing and is utilised in the health care setting to capture all patients presenting for other health care services. HCT should be considered whenever there is care provider/patient contact (FMOH, 2007; NACA, 2012).

Benefits of HCT for individual include; Improved health through educational and nutritional advice; early access to care (including ART) and prevention of HIV-related illnesses; emotional support and better ability to cope with HIV-related anxiety; awareness of safer options for reproduction and infant-feeding; motivation and initiate or maintain reduced risk behaviors (FMOH, 2007).

Benefits of HCT for the public health of the nation include: Reduced transmission following increased knowledge; reduced stigmatization as a result of widespread Counselling services; improved health and productivity of PLWHA as a result of utilisation of care, support and ART services (FMOH, 2007).

HCT is being rapidly scaled up using innovative, ethical and practical approaches. Services can be provided at free-standing, mobile, primary, secondary and tertiary facilities. These facilities must meet national minimum standards for HIV testing applicable to their various levels. For instance, category one laboratories will carry out voluntary counselling and testing using rapid kits. In the case of free- standing, mobile, and primary facilities, referral of seropositive individuals to secondary or tertiary facilities for pre-assessment for ART should be done (FMOH, 2007).

HCT long-term impact on other outcomes is mixed. These include: Increased sexual risk following receipt of a negative result may be a serious unintended consequence of HCT intervention. The new strategy for HIV prevention, “Universal Test and Treat”, whereby everyone is tested for HIV once a year and treated immediately with antiretroviral therapy (ART) if they are infected should be enforced. This strategy could eliminate the epidemic and reduce ART costs in

the long-term as substantial proportion of HIV-infected individuals do not present for HIV testing until late in infection; these individuals are often ill, have a high mortality risk, and are less likely to respond to treatment when initiated. Stigmatised attitudes among health care workers, family and friends of PLWH influence the decision-making process of PLWH and stop them from accessing HCT care, support, and treatment services (FMOH, 2007; NACA, 2012)..

2.10.2 HIV and AIDS Treatment and Care in Adolescents and Adults

An increasing challenge today is the need to standardize HIV treatment to ensure the highest quality of care. With current advances in technology and better understanding of the infection, case management of HIV and AIDS will continue to improve and guidelines on HIV and AIDS care and treatment will continue to be subject to regular review as indications emerge from scientific research and advancement. The National ART Guidelines (FMOH, 2007) has been updated based on literature review and relevant local experiences and is meant for the treatment of adults and adolescents. It is hoped that it will provide relevant, simplified but adequate information required for the effective management of PLWHA in Nigeria.

It is also expected that the Nigeria ART Guidelines will assist building capacity among clinicians who have the primary responsibility of managing the patients.

2.10.3 Paediatric HIV and AIDS treatment and care

The World Health Organization (WHO) in 2008 estimated that 2.1 million children <15 years were living with HIV infection largely acquired through mother to child transmission (FMOH, 2010). It is estimated that over 1150 children <15 years are infected with HIV every day all over the world (FMOH, 2010). In 2008, 390,000 children were newly infected. Recent data from WHO and Joint United Nations Programme on HIV and AIDS (UNAIDS) indicate that only 15% HIV positive African children needing Antiretroviral therapy (ART) in West and Central Africa were receiving it in 2008 (FMOH, 2010).

In Nigeria, seroprevalence Sentinel Surveys showed HIV prevalence of 1.8% in 1992, 5.8% in 2001, and 4.4% in 2005, a slight rise to 4.6% in 2008 and currently in 2010, the prevalence is 4.1% (FMOH, 2010; NACA, 2012). Estimates from the Survey also showed that 52,000 new infections occurred in children <15 years. The number of HIV positive children requiring ART in 2009

was estimated to be 92,000 of which <15% actually received it. According to the 2009 Universal Access Report, Nigeria accounts for 30% of the global burden of mother-to-child transmission of (MTCT) of HIV and 10% of Paediatric HIV and AIDS. As at the end of 2009, the human immunodeficiency virus accounts for 3% of deaths in children younger than five years in Nigeria.

In 2007, the first Paediatric Guidelines was developed. With the WHO, (2010) recommendations on antiretroviral therapy for infants and children, it becomes necessary for the National Guidelines for Paediatric HIV and AIDS Treatment and Care to be updated accordingly.

Objectives of the Paediatric National Guidelines

The overall objectives of the National Guidelines on Paediatric HIV and AIDS Treatment, Care and Support are:

1. To provide standardised management protocols based on current evidence for children infected with or exposed to HIV infection.
2. To provide guidelines for monitoring and evaluation of comprehensive Paediatric HIV and AIDS services
3. To serve as a reference document to advisory boards, programme managers and other policy makers involved in paediatric HIV/AIDS programming.

2.10.4 Prevention of Mother-to-Child Transmission of HIV (PMTCT)

Most children less than 15 years living with HIV acquire the infection through mother-to-child transmission (MTCT). This can occur during pregnancy, labour and delivery or during breast-feeding. In the absence of interventions, the risk of such transmission is 30 – 45% (FMOH, 2010).

The high burden (2.1 million children orphaned by AIDS) of MTCT (Figure 2.6) in sub-Saharan Africa (compared to the rest of the world) is due to higher (336,379 infection in 2008) rates of heterosexual transmission, higher (4.6% National ANC Median HIV prevalence) prevalence of HIV in women of reproductive age, high (56,681 annual HIV positive births) total fertility rate, characteristically prolonged breast feeding culture, as well as poor access to PMTCT interventions. The risk of MTCT can be reduced to less than 2% by interventions that include the use of antiretroviral (ARV) as either prophylaxis or therapy given to women in pregnancy, labour and during breastfeeding (FMOH, 2010). In situations where a

mother is not receiving ARVs during the breastfeeding period, the breast-fed infant should receive ARV prophylaxis until one week after cessation of all breast- feeding. Where breastfeeding is not possible however, it should be noted that the use of commercial infant formula is an alternative (FMOH, 2010).

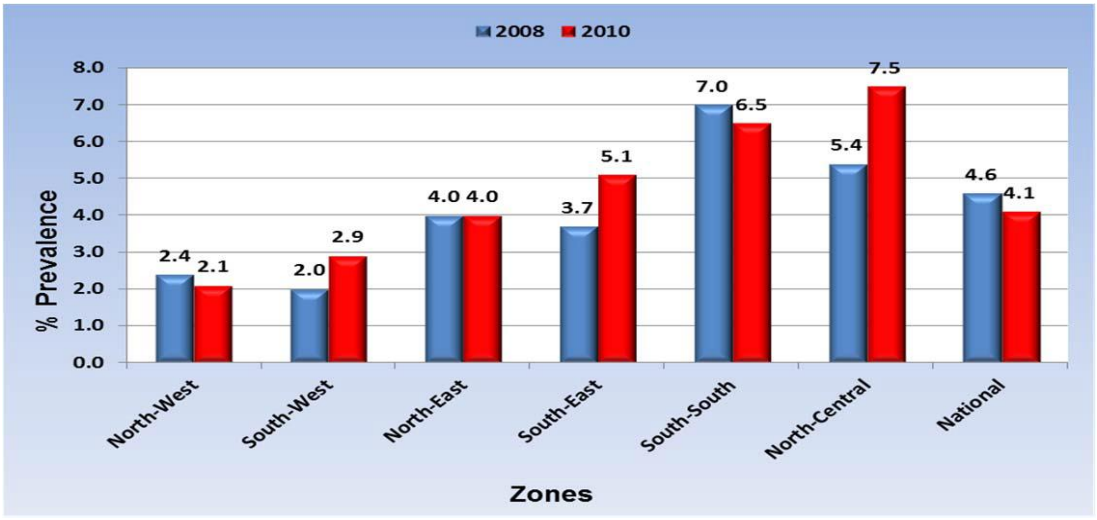


Figure 2. 6: HIV among pregnant women by geopolitical zone in Nigeria (Source: NACA, 2012)

The Nigerian National goal for PMTCT as contained in the 2003 AIDS Policy is to reduce the transmission of the HIV through MTCT by 50% by the year 2010 and to increase access to quality HIV Counselling and testing services by 50% by the same year. To achieve this goal, a comprehensive four-pronged strategy to prevent HIV infection among infants and young children is being implemented in Nigeria since 2001. The strategy includes the following elements:

- Primary prevention of HIV infection in women of reproductive age group and their partners
- Prevention of unintended pregnancies among HIV positive women
- Prevention of HIV transmission from HIV infected mothers to their infants
- Care and support for HIV infected mothers, their infants and family members.

2.11 Nutrition problems in Nigeria

Undernutrition is a problem that aggravates the spread of HIV in Nigeria (FMOH, 2011). HIV prevalence in the country can be said to be stabilising but undernutrition is on the increase (FMOH, 2011). In 2008, the Nigeria Demographic and Health Survey (NDHS) showed that 41% of children aged less

than five years were stunted, 23% were underweight and 14% were wasted. The Nigeria Food Consumption and Nutrition Survey (2003) also reported that 11.6% of women of childbearing age were suffering from chronic undernutrition. Prevalence of micronutrient deficiencies was also high, for instance: Among children under five years old, 29.5% suffered from vitamin A deficiency. 27.5% were iron deficient and 27.5% suffered various degrees of iodine deficiency. Among women of childbearing age, 13.1% were vitamin A deficient (19.1% of pregnant women), 12.7% iron deficient and 15.3% iodine deficient (FMOH, 2011).

Research in HIV-positive children demonstrated that multiple large doses of vitamin A (50,000 IU at 1 and 3 months, 100,000 IU at 6 and 9 months, 200,000 IU at 12 and 15 months) reduced diarrhea episodes (OR=0.51; 95% CI=0.27,0.99), increased CD4 cell counts, and reduced all-cause morbidity (OR=0.69; 95% CI=0.48, 0.99) (Coutsoudis et al., 1995). Anaemia is a common problem affecting PLWH and may result from cytokine-induced suppression of red cells production; chronic inflammation; and/or reduction in dietary intake, absorption and retention of iron (Piwoz and Preble, 2000).

Similar data on the prevalence of other micronutrient deficiencies like magnesium, selenium, and vitamin C are not available, but these nutrients, which are important for optimum immune functions, are also deficient among large sections of the population (Piwoz and Preble, 2000). It has long been established that undernutrition impacts negatively on optimal immune function (Figure 2.7), thus increasing susceptibility to morbidity and mortality among HIV positive patients. It is therefore important to include nutritional care and support in the provision of quality care and support for People Living with HIV (PLWH) (FMOH, 2011).

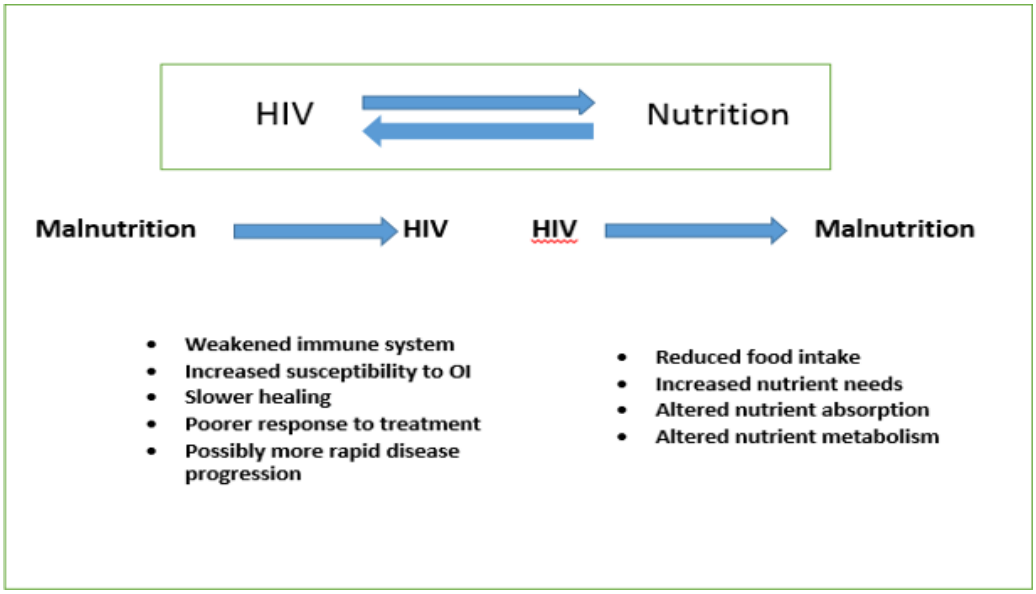


Figure 2. 7: HIV/AIDS and under-nutrition form a symbiotic relationship (USAIDS, 2004)

2.12 Potential role of Nutrition as modifiable risk factors to progression of HIV to AIDS

Nutrition complications of HIV infection, including wasting syndrome, nutrient deficiencies, and metabolic complications, have been well documented over the last 25 years (Coyne-Meyers, 2004). Several studies observed that the level of HIV replication is closely associated with both the rate of growth and the quantity of lean tissue stores (Apardi, 2005). Thus, dietary intake varies inversely with level of virus, suggesting that viral replication directly or indirectly suppresses appetite. In many regions of sub-Saharan Africa, diarrheal diseases represent an important and potentially modifiable factor involved in growth disturbances in PLWHA. The benefit of providing adequate amounts of calories, protein, and micronutrients for persons with HIV is well accepted (Coyne-Meyers, 2004). For example, low serum carotene concentration is common in AIDS patients and predicts death but supplementation with micronutrients and natural mixed carotenoids improved survival by correction of a micronutrient deficiency (Austin et al., 2006). Therefore, low weight gain and growth failure, which are both severe manifestations of AIDS and related to high viral load and low CD4 count, are risk factors that raise the mortality rate among PLWHA.

The correlation between clinical category and under-nutrition reinforces the concept of natural evolution that underlies the CDC classification of CD4 cell counts (Centeville, 2005). Thus the severity of AIDS manifestations is associated with nutritional status and with the age at onset of HIV disease symptoms.

2.13 Nutrient intakes of People Living With HIV/AIDS in Nigeria

Dietary assessment for PLWH in Nigeria and the National Guideline shows that the mean daily energy intake for PLWHIV in Nigeria is 1725 kcal (male) and 1885.85 kcal (female) (Table 2.2). The evidence on the low social economic status of PLWHA in Nigeria and the dietary assessment investigated by the FMOH on PLWH in Nigeria did not accentuate the need to examine the dietary assessment of the same group of patients in this research. Similar dietary assessment was investigated by Temitope et al. (2011) on PLWH (n=200).

Hunger and undernutrition remain the most devastating problems that dominate the health of underprivileged Nigerians (Temitope et al., 2011). At the national level, there was evidence of high expenditure by the citizens on staples with cereals attracting the highest expenditure of about N643.97/week (approximately $4) compared to an average expenditure of N347.73 on roots and tubers.

Results further revealed that some Nigerians spent more on sorghum (N228.3) than on either rice (N215.6) or maize (N127.2) (IITA, 2003). Nutrient intake is specific to a study population and depends on a variety of factors that are difficult to control. These factors include cultural, socioeconomic, environmental, and geographical determinants (Temitope et al., 2011). Carbonnel et al. 1997 illustrated that body weight loss result mainly from decreased energy intake in HIV infected patients. They also found that HIV infected patients had lower levels of energy intake (1,725 kcal for male and 1,885.85 kcal for female) than the RDA (2,800 kcal for male and 2,500 kcal for female). This probably contributed to the lower body weight observed in HIV infected patients, since body weight and energy intake were correlated. HIV infection compromises the nutritional status of the infected individuals and, in turn, poor nutritional status which affects the progression of HIV infection. In Nigeria, the mean energy, and protein in PLWH are below the RDA (Table 2.2). This result was based on 24 hours diet recall and dietary history, It showed that the mean daily energy intake of PLWHIV in Nigeria were below the recommended values by 38.4 per cent for male and 24.6 per cent for female (Temitope et al., 2011; FMOH, 2011). Although the data from the Federal Ministry of Health (2011) and Temitope et al. (2011) were generated from 24 hours diet recall and diet history; however Food Frequency Questionnaire method of dietary assessment would have been more appropriate because it is suitable for large scale surveys such as this intervention research.

Table 2. 2: Energy and Nutrient intake for adult PLWH in Nigeria. (Source: Temitope et al., 2011).

Gender	Energy (kcal)	Mean ± SD	Protein (g)	Mean ± SD	Iron (mg)	Mean ± SD	Vitamin A, g (RE)	Mean ± SD
Male								
Mean intake		1,725 ± 440.5		15.58 ± 3.36		5.55 ± 0.95		562.70 ± 122.45
RDA	2,800		68		6		750	
Intake percentage of RDA	61.6		22.9		92.5		75.0	
Female								
Mean intake		1,885.85 ± 447.4		14.08 ± 3.87		16.48 ± 2.43		495.19 ± 100.08
RDA	2,500		56		19		400	
Intake percentage of RDA	75.4		25.1		86.7		123.80	

The National Guidelines on Nutritional Care and Support for People Living with HIV in Nigeria specified 10% daily energy intake increase during asymptomatic phase of HIV infection and 20 – 30% daily energy intake increase during symptomatic phase. The meal should also provide multiple micronutrients. The most important nutrients are selenium, zinc, beta carotenes, and vitamins A, B, C and E. Selenium play an important role in metabolising reactive oxygen species and reducing oxidative stress. Selenium deficiency impairs the immune system and has been associated with faster HIV disease progression and reduced survival in adults (Piwoz and Preble, 2000; Kupka et al., 2004). Zinc is an essential component of the immune system and it is important for the development of non-specific and cell mediated immunity (particularly CD4 cells) (Piwoz and Preble, 2000; Kupka and Fawzi, 2002). Zinc supplementation (45.5mg/d orally) for one month plus AZT (Zidovudine) reduced the incidence of opportunistic infection, stabilised weight and improved CD4 count ($p < 0.01$) among adult with AIDS (Mocchegiani et al., 1995). Severe Zinc deficiency results in severe depression, frequent diarrhea and other infections (e.g. malaria) as well as mental disturbances (Piwoz and Preble, 2000; Kupka and Fawzi, 2002). Low beta-carotene concentration (>10mg/l) primarily reflect more active HIV-1 infection (Baeten et al., 2007). Vitamin A supplementation (50,000

IU at 1 and 3 months to 200,000 IU at 12 and 15 months) of HIV –infected children reduced diarrhea morbidity by 50 percent (Coutsoudis et.al., 1995; Piwoz and Preble, 2000; Mehta and Fawzi, 2007). Low serum B_{12} level (>180 ng/L) is associated with neurological abnormalities (e.g peripheral neuropathy, myelopathy); impaired cognition (e.g information processing, problem solving); reduced CD4 T-cell count (Tang et al., 1997; Piwoz and Preble, 2000). Vitamin C (80-500mg) significantly ($p < 0.05$) reduced oxidative stress and HIV viral load (Piwoz and Preble, 2000; Isanaka et al., 2012). Vitamin E (15-30mg) increases humoral and cell-mediated immune response, including antibody production, phargocytic and lymphocytic responses, and resistance to viral and infectious diseases (Piwoz and Preble, 2000; Isanaka et al., 2012).

The additional energy requirements as recommended by WHO (10 - 30% in addition to the daily energy intake) will provide more nutrients to compensate for nutrient losses and/or increased nutrient requirements, maintain body weight, manage specific symptoms (e.g. nausea, constipation and diarrhea and reduce the severity of symptoms by providing specific nutrient needs (Piwoz, 2004; FANTA, 2004; WHO, 2007).

2.14 Additional energy requirements (Asymptomatic vs. symptomatic)

Asymptomatic HIV-positive adults need 10% additional energy (per day) than HIV-negative individuals of the same sex. The additional energy requirement for symptomatic HIV positive adults is 20 to 30% (per day) than HIV-negative individual of the same sex while children with growth faltering require 50 to 100% additional energy (Table 2.3) (Piwoz, 2004; FANTA, 2004; WHO, 2007). Therefore, macro and micronutrients from naturally occurring (as opposed to synthetic with a less bioavailability) components tailored to meet the additional energy requirements of PLWHA will enhance well-being and health and/or reduce the risk of disease or provide health benefit so as to improve their quality of life (Roberfroid, 2002).

Table 2. 3: PLWHIV Nutrient Requirement (WHO, 2003; FANTA/USAID 2007).

•Energy **10% increase for asymptomatic** **20 – 30% increase for symptomatic** **50 – 100% increase for children with growth faltering** **•Protein** **12 – 15% of energy intake to maintain and/or recover lean body mass** **•Micronutrients** **Essential micronutrients at RDA** **High energy, nutrient dense food is required to meet needs – not just more of the same!**

2.15 The European Perspectives of Functional Foods and the concept of a Tailored Food Recipe

Because of increasing interest in the concept of "Functional Foods" and "Health Claims", the European Union set up a European Commission Concerted Action on Functional Food Science in Europe (FUFOSE). The report takes the position that functional foods should be in the form of normal foods and they must demonstrate their effects in amounts that can normally be expected to be consumed in the diet (EUFIC, 2006).

Currently, health concerns of communicable and non-communicable diseases have necessitated investigating into options for dietary interventions including the role of tailored food recipes *(e.g. Amtewa meal)* in HIV/AIDS management. The outcome of the research will have direct effect on 90% of HIV infected subjects in West Africa *vis-à-vis* slowing down /eliminating the progression of HIV to AIDS (EUFIC, 2006).

2.16 Justification for HIV nutrition intervention programme in Nigeria

Antiretroviral (ARV) drugs have been shown to reverse under-nutrition in HIV/AIDS but are usually used at the later stages of the disease when the patients are moribund (Kumar and Clark, 2005; Boon and Walker, 2006). Thus, presently, 75% of Nigerians infected with HIV as recommended by WHO do not require ART (CD4 ≥ 350cells/mm^3), but should receive nutritional assistance to maintain the immune system, sustain healthy levels of physical activity and for

optimal quality of life (WHO, 2010). Incidentally, in Nigeria all the HIV/AIDS programmes and interventions at the moment focus on the remaining 25% of HIV infected subjects that require ART (CD4<350cells/mm^3) (UNAIDS, 2009). The implication of the reality on the ground is that all the interventions at the moment are grossly unable to cope with the treatment of those who require ARVs urgently. In essence, this study proposed a nutrition intervention programme which was designed to circumvent undernutrition of the 75% of PLWH who do not require ART (and sustain their CD4 count level ≥350 cells/mm^3 i.e. not require initiation of ART) and equally supporting and improving the anthropometric and biochemical indices of the remaining 25% who are receiving ART treatment (CD4 count ≥200 cells/mm^3). See Figure 2.8.

Therefore, the focus of this intervention was to develop an optimised tailored functional recipe – TFR (as defined in box 1.2) that is readily available and accessible at low cost to Nigerians. TFR is containing macro and micronutrients from natural food sources in Nigeria. Then the efficacy of the optimised TFR to strengthen the immune system of PLWH has been evaluated. The domestic production of the constituents of the optimised meal by household, individual and PLWH in sufficient quantities and quality will ensure access to the intervention meal. Availability and access dimension of food security will also guarantee the stability of this intervention programme.

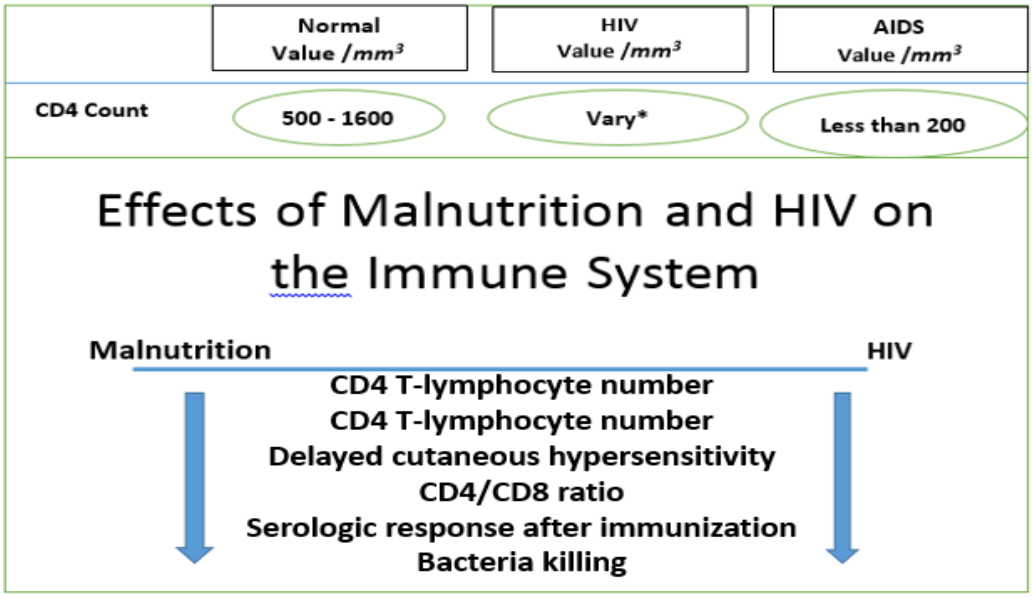


Figure 2. 8: Effect of malnutrition and HIV on the immune system

Box 1.2: Definition of TFR

*TFR: Food that is naturally occurring, accessible, affordable and perhaps consumed in unnatural concentrations as part of the usual diet and has demonstrated physiological and or biomedical benefits in reducing the risk of chronic disease beyond basic nutritional functions.. .

2.17 Originality of the research

Development of 'Amtewa meal'

Under-nutrition and micronutrient deficiency remain significant contributors to morbidity and mortality in developing countries (FAO/WHO, 2002; Amuna et al., 2004) and in economic terms, remain a major challenge. Food-based approaches need to be innovative, culturally acceptable, accessible, affordable, reliable and requiring low-tech approaches in order to assure compliance, sustainability and cost-effectiveness (Jiamto et al., 2003; Zotor et al., 2006). It is possible to improve the nutritive value of local foods through simple but scientific combinations of food ingredient in form of food multimixes (FMM) to develop Amtewa meal (Amuna et al., 2004).

Nutrient deficiencies associated with HIV are: total calories, proteins, vitamin A, vitamin B_6, vitaminB_{12}, vitamin C, vitamin E, magnesium, selenium and zinc (Kotler, 1992; Beach et al., 1992; Baum et al., 1995; Cimoch, 1997; Van Staden et al., 1998; Kotler et al., 1999; Periquet et al., 1995; Vilaseca, 2003 and Oguntibeju, 2008). Development of a biochemical deficiency of vitamins A, B_6 and B_{12} is associated with faster disease progression. Normalisation of plasma vitamin A, B_{12} and zinc levels is linked to slower disease progression (Baum et al., 1995). Tang et al. (1997) confirmed that low serum (>180ng/L) vitamin B_{12} precedes disease progression. Patients often have multiple nutrient deficiencies at once, and many of the nutrients likely to be deficient are directly or indirectly involved in maintaining normal immune system function (Tang and Smit, 1998). Amtewa meal is a combination of these macro and micro-nutrients carefully selected from locally available food in Nigeria (Table 2.4), analysed, optimised and formulated into a 100g pack for daily consumption and can contribute between 15 to 20% additional energy requirement by PLWH as recommended by WHO. This meal is a natural product that requires low-technology approaches in its development, has a greater bioavailability (The bioavailability of macronutrients – carbohydrates, proteins, fats – is usually very high at more than 90% of the amount ingested (EUFIC, 2010)) than synthetic nutritional

supplements. The nutritional content of the meal is tailored to: decrease functional impairment from under nutrition, improve immune function, preserve or increase fat-free mass, limit disease specific complications, improve tolerance to antiretroviral therapy (ART), provide relief from/prevent symptoms of HIV and improve quality of life of PLWH in Nigeria.(Kotler et al., 1999; Mahlungulu et al., 2009; Ukibe et al., 2013).

Bioavailability of natural food ingredients versus synthetic micronutrients

Studies suggest that the bioavailability of natural food complex vitamins is better than that of most isolated United States Pharmacopeial vitamins that they may have better effects on maintaining aspects of human health beyond traditional vitamin deficiency syndromes and at least some seem to be preferentially retained by the human body (Thiel, 2000). The first step in making nutrients in Amtewa meal bioavailable is to liberate it from the food matrix and turn it into a chemical form that can bind to and enter the gut cells or pass between them. Collectively this is referred to as bioaccessibility (EUFIC, 2010). The Nutrients are rendered bioaccessible by the processes of chewing (mastication) and initial enzymatic digestion of the food in the mouth, mixing with acid and further enzymes in the gastric juice upon swallowing, and finally release into the small intestine, the major site of nutrient absorption. In addition to the bodily means of mastication and enzyme action, the digestibility of Amtewa meal is aided by cooking or pureeing the food. For example, whereas raw carrots, spinach and moringa are good sources of dietary fibre, cooking them allows the human body to also extract a much larger fraction of the carotenoids contained.

Table 2. 4: Justification for the inclusion of some selected micronutrients in the optimised meal (Amtewa meal) and the sources of the nutrients from Nigerian foods (Opoku et al., 1981; Adeyeye and Ajewole, 1992; Bijlsma, 2000; Zotor et al., 2000).

Nutrient/100g	Deficiency	Sources	Literature support
Beta- carotene 5.04mg	Faster disease progression	Dried moringa leaves, dried carrot roots	Bijlsma, 2000; Zotor et al., 2000; Piwoz and Preble, 2000; Omale and Ugwu, 2011; Monica, 2013
Vitamin B_1 0.4mg B_2 3.08mg B_3 1.23mg	Psychoneurological symptoms ranging from peripheral neuropathies to spinal cord degradation and cognitive impairment	Dried moringa leaves	
Vitamin C 10.5mg	Decreases resistance to infection	Dried soya bean seeds, dried moringa leaves, dried carrot roots	
Phosphorus 442mg	Reduced utilization of energy and metabolism by cells	Dried soya bean seeds, dried moringa leaves, dried millet seeds	
Zinc 3.88mg	Faster disease progression	Dried soya bean seeds, dried moringa leaves, dried carrot roots, dried millet seeds	
Copper 0.33mg	Increased incidence of infection and cell-mediated immunity	Dried moringa leaves, dried carrot roots, dried millet seeds	
Iron 18.4mg	Anaemia	Dried soya bean seeds, dried moringa leaves, dried carrot roots, dried millet seeds	
Manganese 0.6mg	Decreased mitochondria ability to reduce level of oxidative stress	Dried carrot roots, dried millet seeds	
Sodium 6.57mg	Hyponatremia, muscular weakness	Dried soya bean seeds, dried carrot root, dried millet seeds	
Potassium 1188.4mg	Hypokalemia, impaired cellular processes	Dried soya bean seeds, dried moringa leaves, dried carrot roots, dried millet seeds	
Magnesium 218.4mg	Decreased protein synthesis,decreased transmission of nerve impulse	Dried soya bean seeds, dried moringa leaves, dried carrot roots, dried millet seeds	
Calcium 451.4mg	Decreased normal heart and muscle functions, blood clothing and pressure, and immune defence	Dried soya bean seeds, dried moringa leaves, dried carrot roots, dried millet seeds	
Selenium 0.008mg	Faster disease progression from oxidative damage	Moringa leaves, whole grain	

The above Table 2.4 highlighted the richness of some Nigerian foods with micronutrients that may have therapeutic effect once optimised in an intervention meal. This warrants an investigation to assess their effect to attenuate the progression of HIV to AIDS.

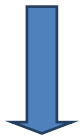
Chapter 3

Subjects, Materials and Methods

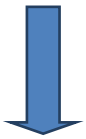
3.1 Subjects, Materials and Methods

This chapter elucidates the various stages taken in the laboratory and the study site to establish the effect of the intervention meal. The chapter also set up steps to provide reliable evidence to assess the effectiveness of the intervention meal. These stages were summarised as follows:

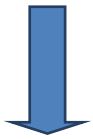
A. An overview of HIV in Nigeria



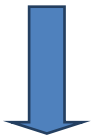
B. Sampling method and randomisation of study participants



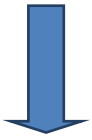
C. Scientific approach in the development of Tailored Food Recipe



D. Intervention setting, assessment tools and ethical consideration



E. Intervention strategy: Planning, implementation and evaluation



F. Sustainability plan

3.1.1 Overview of HIV/AIDS in Nigeria

In Nigeria, AIDS is one of the most challenging health problems of this era (FMOH, 2008; NACA, 2012). Since the first case report in 1986, the number of persons infected with the HIV has risen markedly, so that at present there are over 3.4 million Nigerians living with the virus, of which an estimated 750,000 require medical care (FMOH, 2007). Youth and young adults (18 – 30 years) from the three main religions (Christianity, Islam and Traditional religion) in Nigeria are particularly vulnerable to HIV, with young women at higher risk than young men (UNAIDS, 2008).

The care and support of people with HIV and AIDS presents special challenges for Nigerian health institutions unlike most other community encountered disease entities. HIV disease is multifaceted, and as such, the care provider is confronted with multiple manifestations of HIV associated disorders such as tuberculosis, undernutrition, depression etc (WHO, 2006; Pribram, 2011).

The complex nature of HIV infection and disease management in Nigeria calls for a multidisciplinary approach to the care of PLWHA. HIV presents as a complex mix of many disease states. There is the physical illness requiring physical – medical care and there are sundry manifestations of psychosocial disorders. The later often the direct consequence of stigma, fear and neglect that is associated with a diagnosis of HIV infection in Nigeria (FMOH, 2007).

To ensure that all persons living with HIV and AIDS have access to a comprehensive package of care and support, the Ministry of Health in Nigeria developed treatment guideline that anticipates and addresses the four components of HIV care in Nigeria.

HIV Counselling and Testing, PMTCT, Adult and Paediatric ART Care are vital components of the strategies to expand access to comprehensive care for Nigerian population of PLWHA and they are universally acknowledged.

Despite the care and support programmes adopted by the Ministry of Health in Nigeria, the high burden of HIV and AIDS calls for dedicated and sustained acceleration of prevention strategies against the transmission of the virus as well as affordable nutrition intervention to slow the progression of the disease in Nigeria. This study reviewed the existing epidemiologic evidences on the relation between nutrition and HIV disease and the basic principles of ethical practice in a nutrition intervention (see chapter 1 and 2). The study also has the potential to

provide a level of assurance about the role of locally formulated meal in HIV disease progression that is difficult to achieve with any observational design.

This chapter (3) will illustrate the major steps of the planning phase and these include:

Sampling methods

Sample size calculation

Inclusion,exclusion criteria and confounding factors

Scientific approach to develop Tailored Food Recipe (Amtewa meal), etc.

3.1.2 Sampling methods

Randomised Control Trials (RCTs) was used to test the efficacy or effectiveness of Amtewa meal nutrition intervention.

All patients who assess antiretroviral treatment at ARV treatment centres in Abuja, Nigeria between February 2002 and December 2010 were given opportunity to participate in the research. Demographic characteristics such as age, gender, occupation, marital status, number of spouses, past history of STIs, and blood transfusion were obtained from the patients' hospital record files.

1000 participants were invited to participate in the research from all the HIV treatment centres in Abuja, Nigeria. These PLWH from the HIV treatment centres in Abuja, were assessed for eligibility (PLWH with a CD4 count of ≥200cells/mm^3, above 18years, not pregnant and without HIV/AIDS complications – for details see section 3.1.2.3 inclusion/exclusion criteria). The Study participants were recruited based on the most recent CD4 count records obtained from the medical records department of the treatment centres.

Based on the sample size calculation, inclusion and exclusion criteria, participants (Pilot N=100; Scale-up N=400), (adult, male and female from different religious background) were selected through simple randomisation (Figure 3.1). The selected participants were randomly allocated into one of four groups as illustrated in the study design (Figure 3.2 and 3.3) and given the right to decline participation without jeopardizing receipt of care at the State House Medical Centre, or other treatment centres in Abuja. They were also subjected to comprehensive assessment (appendix 3.8), which includes demographic, physical assessment (anthropometric measurements) and selected biomedical indices. The study participants continued standard treatment for PLWH by the SHMCA (nutritional Counselling, vitamin supplements for Pre-ART group and

HAART, nutritional Counselling, vitamin supplements for the ART group). Half of each group (Pre-ART and ART) were dispensed Amtewa meal for daily consumption for six months. Prior intervention, project's information sheet and consent forms (appendix 3.3) were read /interpreted and signed by all the study participants. Pre and post intervention assessments were carried out as specified in the study design.

Recruitment of study participants was done by the researcher while the sample collection, CD4 count and other laboratory investigations (Total protein, SGOT, RG, PCV) were performed by a trained laboratory scientist in SHMCA.

One hundred enrolled participants from the four hundred eligible study participants were selected for the pilot study. Participants for the pilot intervention were randomly selected into the four study groups as illustrated in the pilot study design (Figure 3.2). Qualitative data were collected from the study participants through:

- Informal individual interviews on life style, adherence to the meal and adverse reactions
- Focus groups interview to get more objective and macro view of the intervention: Pre–ART and ART (Test versus control),
- Continuous observation of participants' response to the intervention meal
- Direct involvement of the researcher in ensuring that the study procedure is not compromised.

3.1.2.1 Sampling framework

Simple random sampling strategy was adopted in the selection of the different groups of the study participants according to the study design. In this type of probability sampling technique, there was an equal chance (probability) of selecting each group (Pre-ART and ART) from the PLWH being studied. This study was based primarily on the recognition that complete enumeration through census-based surveys imposes huge costs that are both unsustainable and unnecessary if the nature and methods of statistical sampling are properly considered. Such considerations in this research include understanding the:

- Reasons for and objectives of sampling.
- Relationship between accuracy and precision/reproducibility (pilot followed by scale-up intervention).
- Determination of safe sample sizes for surveys for effective monitoring.

3.1.2.2 Sample size calculation

This sample size calculation was used to determine the number of participants to be interviewed or assessed in order to obtain results that reflect the target population as precisely as needed. It also shows the level of precision in an existing sample (Creative Research System, 2012).

The sample size was determined as follows (Bill Godden, 2004)

Sample Size - Infinite Population (where the population is greater than 50,000)

$$SS = \frac{Z^2 \times (p) \times (1 - p)}{C^2}$$

Z = Z-value (1.96 for a 95 percent confidence level)

P = Percentage of population expressed as decimal, in the case of this study we are using 50% = 0.5 (this choice is used in this study since it is the safest when it is not possible to estimate " anticipated population proportion")

C = Confidence interval, expressed as decimal (0.05 = +/- 5 percentage points)

Z-values (Cumulative normal probability table) represent the probability that a sample will fall within a certain distribution.

The Z-values for confidence levels in this case is 1.96 (guided by the cumulative normal probability table).

$$SS = \frac{1.96^2 \times 0.5 \times 0.5}{0.5^2} \qquad SS = 384$$

5% attrition was be added = (5/100 x 384) + 384 = 403

For the pilot study, 25% of the SS was considered = 25/100 x 400

New sample size (NSS) for pilot study = 100

New sample size (NSS) for larger scale intervention = 400

In the scale-up intervention Four Hundred (N=400) study participants were randomly selected for the six months intervention according to the study design. These 400 participants comprised of the 100 participants that overlapped from the pilot to the scale-up intervention. However, due to fall outs and some incomplete results during follow up, the total number of research participants that completed the six months scale – up intervention was **three hundred and eighty four (384).**The justification of the sample size is based on the 95% confidence interval and a precision limit of 0.05 for the study. Also the Sentinel Survey in 1991 showed a prevalence of 1.8%. Subsequent sentinel surveys produced prevalence of 3.8% (1993), 4.5% (1996), 5.4% (1999), 5.8% (2001), 5.0% (2003), 4.4% (2005), 4.6% (2008) and 4.1 % (2010), a trend signaling a

general reversal of the epidemic in the country (NACA, 2012). However, the prevalence of HIV in Abuja where this study was conducted showed 9.9% in 2008 and 8.6% in 2010. The prevalence rate of HIV in Abuja is high compared to the National prevalence rate; hence the rationale for Bill Godden's formula to calculate the sample size for population greater 50,000.

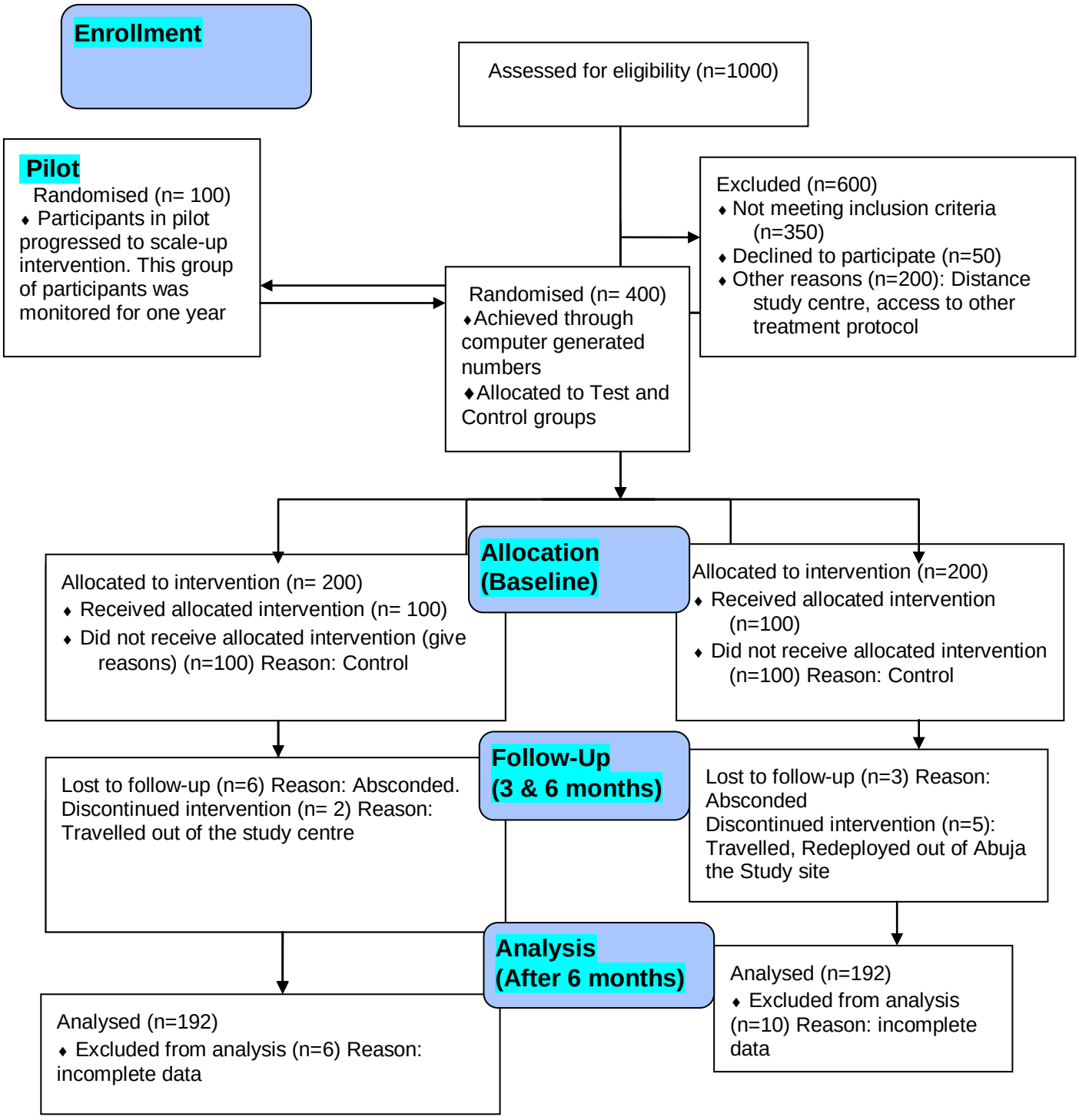


Figure 3. 1: Flow Diagram for Amtewa meal nutrition intervention (pilot and scale up)

3.1.2.3 Inclusion, exclusion criteria and confounding factors

The study was conducted in two phases (pilot and scale-up) employing the following criteria: Prospective study participants were briefed and selected using the inclusion criteria listed below as a prerequisite.

Inclusion Criteria

- PLWHA with a CD4 count of ≥200cells/mm^3. HIV infection documented by licensed ELISA test, confirmed by Western Blot or positive HIV blood culture or positive HIV serum antigen or second antibody test positive by a method other than ELISA or any two different rapid technique.
- Male or Female, age 18years and above.
- GIT – parasites free (e.g. de – wormed from GIT, worms, flukes etc).
- Willingness and ability to provide a written consent to participate in the trial.
- No life threatening opportunistic infections
- No history of lymphoma
- No pregnant women or nursing mothers
- No severe kidney or and liver dysfunction
- No HIV/AIDS complications

Exclusion Criteria

- AIDS patient with a CD4 count less than 200 cells/mm^3
- Life threatening opportunistic infections

3.1.2.4 Randomisation of study participants

Researchers in life science research demand randomisation (Suresh, 2011).
Gray and John Hopkins University, (2006), defined a Randomised Controlled Trial (RCT) as a planned experiment designed to assess the efficacy of an intervention in human beings by comparing the intervention to a control condition. Random or probability sampling is recommended as a means of informant selection because Randomisation reduces biases and allows for the extension of results to the entire sampling population (Godambe 1982, Smith 1983, Snedecor 1939, Topp *et al*., 2004). Specific plan to identify and enroll study participants involved screening and establishing criteria for number, location, and sampling method for the pilot and scale-up interventions.

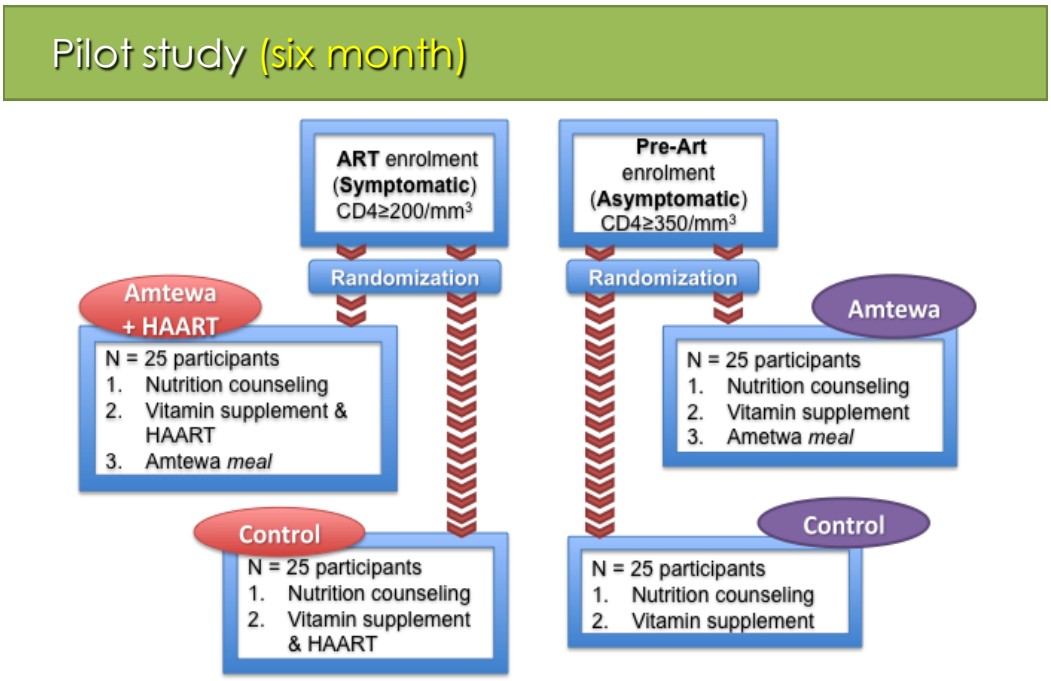
In this study, randomisation of study participants was achieved by generating randomisation schedules, obtaining the random numbers and assigning random numbers to each subject Random numbers were generated by the computer from the attendance list of PLWH who attended the briefing and had all the requirements as enumerated in the inclusion criteria above. This randomisation ensures that each participant had an equal chance of receiving any of the treatments under study, generate comparable intervention groups, which are alike in all the important aspects except for the intervention each groups receives (Suresh, 2011). The major reason for randomisation in this study design was that it eliminates the selection bias, balances the groups (Pre-ART and ART) with respect to many known and unknown confounding or prognostic variables, and forms the basis for statistical tests, a basis for an assumption of free statistical test of the equality of treatments.

The two groups (Pre-ART and ART) were randomly selected from the same population of PLWHA receiving treatment, care and support at the SHMCA and other treatment centres in Abuja. They were not only statistically equivalent to the larger group; they were also statistically equivalent to each other. The design of this simple randomised evaluation (with two groups) was that one group received the program that is being evaluated (Amtewa meal intervention) and the other did not.

The study design ensured that the research assessment tools were feasible and the study answered questions with clear variables (Anthropometric and Biochemical indices) to determine the impact of the intervention on the study population. The essential inclusion and exclusion criteria for the study population were distinct, research hypothesis (appendix 3.0) was specified, method of enrollment, follow up and rigorous monitoring were not compromised hence findings were accurately generalised and specific results to the target group as illustrated in Chapters 4 and 5.

Randomisation – Pilot Intervention

This step involved the randomisation and grouping of the research participants into the following; ART (Asymptomatic) and Pre-ART (Symptomatic) as illustrated in Figure 3.2 below



HAART: Highly Active Anti Retroviral Therapy

Figure 3.2: Illustrating the two arms of study design (Symptomatic vs Asymptomatic) and showing the patients on Amtewa meal, Amtewa + HAART versus their controls.

A. *GROUP I : Pre-ART enrolment are People Living With HIV (PLWH) but are not on HAART yet because their CD4 count value is ≥ 350/mm^3 according to WHO classification and HIV treatment guideline in Nigeria*

B. *GROUP II : ART (Anti-Retroviral Therapy) enrolment are People Living With HIV (PLWH) and are on HAART with a CD4 count value of ≥ 200/mm3 according to WHO classification and HIV treatment guideline in Nigeria. These patients are HIV patients but not full blown AIDS patients*

C. *Nutrition Counselling is provided as a routine service to People Living With HIV/AIDS (PLWH) receiving care at the SHMCA*

D. *Vitamin and minerals supplements (Box 3.1) are provided, prescribed and dispensed at a dose of one capsule daily or one capsule twice daily to PLWH receiving care at the SHMCA.*

E. *HAART (Highly Active Anti-Retroviral Therapy) are HIV medicines provided, prescribed and dispensed (according to the clinical status of the patient) to PLWH receiving treatment at SHMCA*

F. *Amtewa meal is a combination micro and macro-nutrients, carefully selected from locally available food in Abuja Nigeria, analysed and formulated into a 100g pack for daily consumption by study participants.*

Box 3.1: Vitamin and minerals supplements

Vitamin and minerals supplements :The composition of the vitamin supplement is vitamin A 3333 IU, vitamin B1 4.5mg, vitamin B2 5.1mg, vitamin B6 6mg, vitamin B12 6µg, vitamin C 180mg, vitamin D3 200 IU, vitamin E 10mg, Biotin 0.3mg, Pantothenic Acid 21mg, Folic Acid 0.2mg, Nicotinamide 57mg, Calcium 50mg, Magnesium 40mg, Phosphorus 50mg, Copper 0.4mg, Iron3.6mg, Manganese 0.5mg,Zinc 3mg, Chromium 10µg

Randomisation – Scale up intervention (n=400)

The sequence illustrated in the pilot intervention was adopted in the scale-up intervention. Figure 3.3 shows details of randomisation in the scale-up intervention.

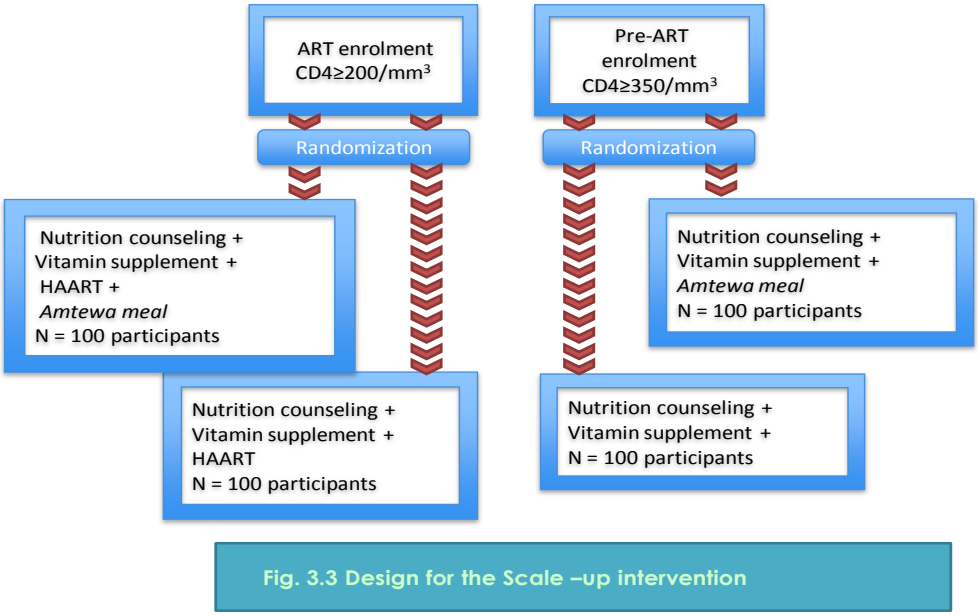


Figure 3.3: The design for the scale up of public health-nutrition intervention programme (n=400).

A. *GROUP I: Pre-ART enrolment are People Living With HIV (PLWH) but are not on HAART yet because their CD4 count value is ≥ 350/mm³ according to WHO classification and HIV treatment guideline in Nigeria*

B. *GROUP II: ART (Anti-Retroviral Therapy) enrolment are People Living With HIV (PLWH) and are on HAART with a CD4 count value of ≥ 200/mm3 according to WHO classification and HIV treatment guideline in Nigeria. These patients are HIV patients but not full blown AIDS patients*

C. *Nutrition Counselling is provided as a routine service to People Living With HIV/AIDS (PLWH) receiving care at the SHMCA*

D. *Vitamin supplements (Box 3.2) are provided, prescribed and dispensed at a dose of one capsule daily or one capsule twice daily to PLWH receiving care at the SHMCA.*

E. *HAART (Highly Active Anti-Retroviral Therapy) are HIV medicines provided, prescribed and dispensed (according to the clinical status of the patient) to PLWH receiving treatment at SHMCA*

F. *Amtewa meal is a combination micro and macro-nutrients, carefully selected from locally available food in Abuja Nigeria, analysed and formulated into a 100g pack for daily consumption by study participants. The composition of Amtewa meal is Soya beans (Glycine max), millet (Pennisetum typhoides), Carrot (Daucus carota), Moringa leaves (Moringa oleifera).*

Box 3.2: Vitamin supplements

*Vitamin supplements** :The composition of the vitamin supplement is vitamin A 3333 IU, vitamin B1 4.5mg, vitamin B2 5.1mg, vitamin B6 6mg, vitamin B12 6µg, vitamin C 180mg, vitamin D3 200 IU, vitamin E 10mg, Biotin 0.3mg, Pantothenic Acid 21mg, Folic Acid 0.2mg, Nicotinamide 57mg, Calcium 50mg, Magnesium 40mg, Phosphorus 50mg, Copper 0.4mg, Iron3.6mg, Manganese 0.5mg,Zinc 3mg, Chromium 10µg*

3.1.2.5 Confounding factors

Randomised trials are not affected by confounding by indication due to random assignment (Johnston, 2001). Confounding was less likely to occur in this study because baseline characteristics (inclusion criteria) for the different groups (Pre-ART and ART) and subgroups (Test versus Control) were the same. A reduction in the potential for the occurrence and effect of confounding factors was also

obtained by increasing the types and numbers of comparisons performed in the analysis.

Confounding variables may be categorised according to their source. The choice of measurement instrument (operational confound), situational characteristics (procedural confound), or inter-individual differences (person confound). A reduction in the potential for the occurrence and effect of operational confounding factors in this study was achieved by increasing the numbers of comparisons performed in the proximate analysis. Similarly, data from anthropometric measurements were repeated twice and witnessed by another medical personnel to ensure robustness of findings from the study and a reduction in the potential for operational confound.

Peer review in the development of the conceptual framework and at the end of the pilot intervention assisted in reducing instances of confounding, either before study implementation or after analysis had occurred. Peer review also relied on collective expertise within medical colleagues in the care and support programme for HIV to identify potential weaknesses in the study design and analysis, including ways in which results may depend on confounding. This intervention research utilised the Randomised Control Trial (RCT) design where the study population was divided randomly after briefing (Figure 3.4) in order to mitigate the chances of self-selection by participants or bias by the study designer. The technique places participants in groups (Pre-ART and ART) and subgroups (Tests versus control) relevant to criteria that fit the research question.

Figure 3. 4: Briefing of participants for the public health-nutrition intervention programme.

3.1.3 Scientific approach to develop Tailored Food Recipe (Amtewa meal)

The scientific approach involved the stepwise processes illustrated in Figure 3.5.

The Tailored Food Recipes blends were processed into powdered formulations which were also made into edible pudding by the study participants. The resulting optimised Amtewa meal is nutrient-dense, packaged and labeled appropriately for distribution according to the study design during the implementation phase.

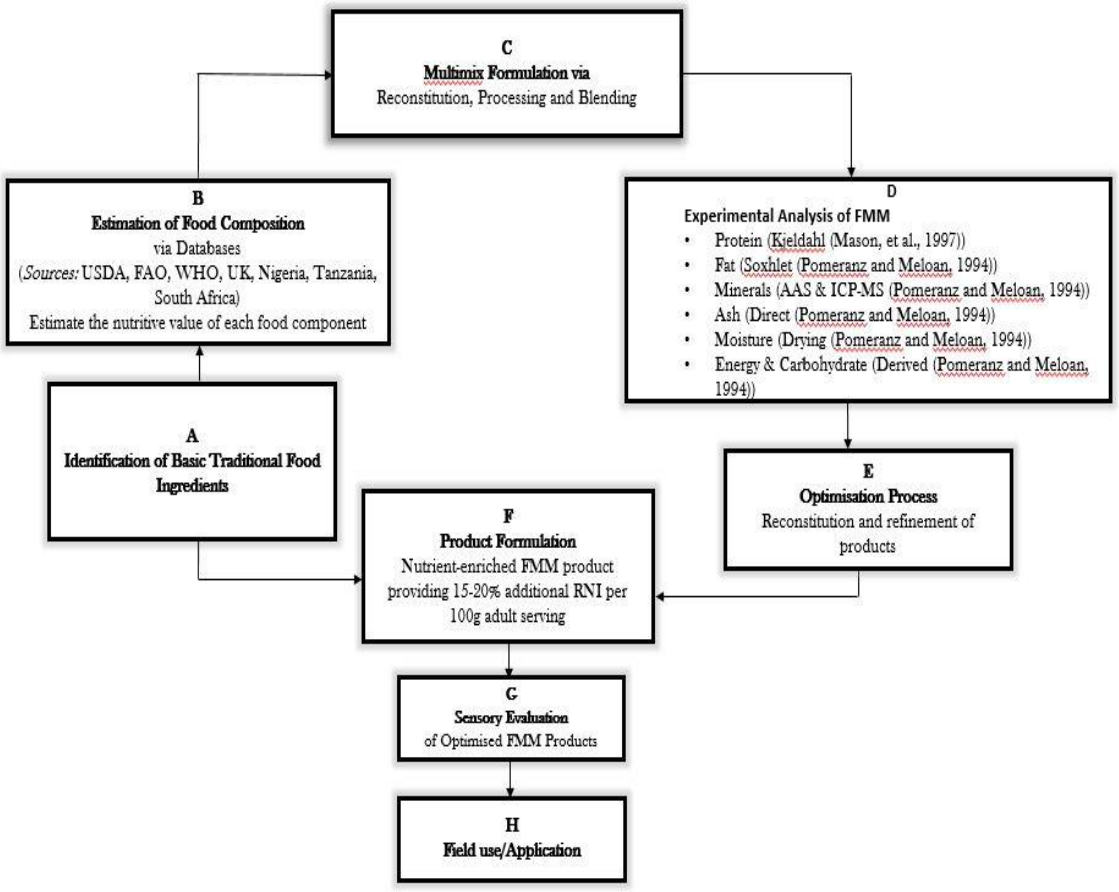


Figure 3. 5: A schematic diagram showing the stages and processes involved in the optimisation of Amtewa meal (Source: Amuna et al., 2004)

3.1.3.1 Identification, harvesting and storage of basic traditional food ingredient

Documented report on Nigeria Food Consumption and National Survey (IITA, 2004) identified some frequently consumed Nigerian food that contains specific macro and micronutrients with potentials of boosting the immune system. The presence of these macro and micronutrients were confirmed in the proximate analysis of the food samples, hence the justification for inclusion in the formulation of the intervention meal (Amtewa).

According to the IITA report (2004), the most available and frequently consumed foods/meals that are major source of energy (calories) in Nigeria were rice (14.8%), cassava (12.90%), maize (10.6%) and yam (10.1%). Cowpea,

groundnut and soya beans are major sources of plant protein (IITA, 2004). Furthermore, Nigerian's popular meals such as soyabean cake, moringa soup, millet tuwo, are also rich in both macro/micronutrients. Appraising the food composition databases enabled the selection of local, accessible, affordable, culturally accepted food ingredients. The nutritive values of each food ingredient were estimated to ascertain its inclusion in the formulation process of tailored food recipe (Amtewa meal). Optimised TFR was then employed in the randomised control trial that aimed to slow the progression of HIV/AIDS in Abuja, Nigeria. See Table 3.1 for details.

Staple and non-staple foods are available almost all the year round in Nigeria but are not affordable all the year round by the citizens. The availability – affordability gap is wider in meat and fish products. This has also affected the frequency of consumption of the staple and non-staple foods in Nigeria.

Severe food insecurity was found in over 40% of all households in Nigeria (IITA, 2004). The implication of this finding is that food security is not only in providing physical access but also economic access to food to increase food availability index of households (IITA, 2004). Judgments to identify, harvest and store food samples for analysis and the formulation of Amtewa meal were based on IITA (2004) reported and the following information on specific food sample. This information includes:

- Macro and micronutrient composition of food samples
- Sight-colour, size and shape
- Touch-texture, hardness or softness
- Smell-odour or aroma
- Taste-sweetness, sourness, bitterness
- Resonance-sound when tapped.

Receptacles in vehicles and/or containers were not used for transporting anything other than the food samples during harvesting and transportation. . Containers reserved for sample storage were marked clearly to show they are used only for the purpose of this research.

From the survey found on IITA publication in (2004) and other literature sources listed in Table 2.4, foods samples in Nigeria were identified, selected, stored, transported to UoW/LMU and analysed to ascertain their macro and micronutrient compositions (Table 3.1).

Table 3. 1: Food sources in Nigeria identified and selected for analysis

Food	Botanical name	Some popular Nigerian meals	Active Principles	Potential Inclusion in Amtewa
Soyabeans (*† ‡)	Glycine max	Soya milk Soya ogi Soya cake Soyabeans soup Soya meat etc.	Vitamins A, B, C, D, E, Iron, Phosphorus, Potassium, Sodium and Zinc	Potential ingredient for Amtewa meal
Millet (*†‡)		Millet Kunu drink Millet ogi Millet porridge Millet tuwo etc	Iron, phosphorus, B.complex, potassium, magnesium and zinc	Potential ingredient for Amtewa meal
Rice	Oryza sativa L.		Protein, carbohydrates, vitamins B1,B2, B6,folate, phosphorus, magnesium, iron, potassium, zinc, oridine, calcium manganase	
Guinea corn	Sorghum		Iron, phosphorus, B. complex, potassium, magnesium and zinc	
Carrots († ‡)	Daucus carota Linn	Carrot cake Mixed vegetable sauce with salad, rice etc	Vitamins A,B,C and D, Fe, Al, Mn, Zn	Potential ingredient for Amtewa meal
Moringa leaves (*† ‡)	Moringa oleifera	Moringa tea Moringa salad (Zogale) Moringa soup etc	Gum, moringine, moringinine, pterygos-permin, Iron, phosphorus, B. complex, potassium, magnesium, calcium, vitamin c, sodium and zinc and selenium	Potential ingredient for Amtewa meal
Ugu leaves	Telfaira occidentalis		Potassium, iron	
Bitter leaves	Vernomia amygdalina		Iron, potassium, magnesium	
Ewedu leaves	Corchorus Walcottii		Iron, calcium, vitamin c, alpha tocopherol, antioxidants	
Efo	Amaranthus gangeticus		Fatty oils	

(i) *Source of energy: (ii) † Nutraceutical boost to immune system: (iii) ‡ Food items available at low cost

3.1.3.2 Estimation of Food Composition via Databases (COMA, 1991; IITA, 2004; Anjorin et al., 2010; Omale and Ugwu, 2011; SACN, 2011)

In this study, the macro and micronutrient values reported in the Dietary Reference Values for Food Energy and Nutrients for the United Kingdom database as recommended by the Committee on Medical Aspects of Food Policy (1991) and the Scientific Advisory Committee on Nutrition (2011) were the primary nutrient data sources referenced for safe intakes. To ensure that these values were appropriate for Nigeria, references were made to other sources such as the Guidelines on Nutritional Care and Support for People Living with HIV in Nigeria, International Scientific Publications on food composition tables in Nigeria and local food composition tables (IITA, 2004). To match the foods, consideration was given to total energy content and the following nutrients (macronutrients and minerals) for fruits and vegetables: energy, carbohydrates, calcium, phosphorous, sodium and potassium; dairy: energy, protein, fat, calcium, phosphorous; cereals: energy, carbohydrates, calcium, and phosphorous; and meats and eggs: energy, protein, fat, and iron, because these nutrients were likely to be present in those food groups.

3.1.3.3 Proximate analysis (Pomeranz and Meloan, 1994; 2000)

Proximates are used in the analysis of biological materials as a decomposition of a human-consumable good into its major constituents (Pomeranz and Meloan, 1994; 2000; BradFord and Cook, 1997; Blamire, 2003). From an industry standard proximates used in the intervention study include five constituents:

- Ash – ICP (Pomeranz and Meloan, 1994; 2000)
- Moisture – Drying (Pomeranz and Meloan, 1994; 2000)
- Proteins - Kjeldahl method (Pomeranz and Meloan, 1994; 2000)
- Fat - Soxhlet method (Pomeranz and Meloan, 1994; 2000)
- Carbohydrates (Calculation)

Analytically, four of the five constituents were obtained via chemical reactions and experiments (appendixes 3.4 to 3.7). The fifth constituent, carbohydrates, was a calculation based on the determination of the four others. Proximates should nearly always add up to 100% (Pomeranz and Meloan, 1994).The proximate analysis employed in this intervention study is supported by Amuna, et al. (2004) in their study on human and economic development in developing countries: a public health dimension employing the food multimix concept.

Laboratory analyses of macronutrients (such as; Soxhlet method for fat extraction) were carried out in the laboratories of University of Westminster, London while other macro and micronutrient analyses (such as; Kjeldahl method for protein analysis; inductively coupled plasma method for analysis of minerals) were conducted at the Science Centre of the London Metropolitan University (appendixes 3.4, 3.5, 3.6 & 3.7).

3.1.3.4 Food sampling and safety aspect of Amtewa meal

The abundance of anti-nutritional factors and toxic influences in plants used as human foods and animal feeds certainly calls for concern (Omoruyi et al., 2007). Therefore, ways and means of eliminating or reducing their levels to the barest minimum was taken into consideration in this intervention.

Seasonality and variability of the nutritional composition of single food items were taken into consideration in the collection of TFR ingredients. Representative samples (n=4) were subjected to proximal analysis. Each sample was washed, irrelevant extraneous matter removed by sieving, sedimentation and flotation methods and then drained before drying in an oven heated at 78^0 C for 60 hours.

The Soya Beans sample was fermented to reduce the phytic acid content. Appropriate facilities to maintain adequate personal hygiene was ensured where necessary; surfaces in contact with food were easy to clean. Utensils and equipment in contact with food were properly cleaned and disinfected where necessary. Dried samples were size reduced with an electric blender to fine, free-flowing powders (Figure 3.6).

The dried ingredients enabled a longer shelf life of Amtewa meal package. Furthermore the food label of the package was designed in a way to address the safe food consumption of the meal as well as the mode of cooking and preparation. Consumers were advised to consume the meal once open and discard left over. This was part of the food safety briefing giving to all participants at the beginning of the intervention.

Aeration of the samples during the blending process was avoided by allowing 60 seconds of break from every 60 seconds of the blending process. Anyone known or suspected to be suffering from or to be carrier of a disease likely to be transmitted through food samples were not permitted to assist in the handling and formulation of Amtewa meal.

Additionally, study participants were strongly advise that all Amtewa meal preparations that is handled, stored, packaged, displayed or transported must be protected against any contamination likely to make it unfit for human consumption, dangerous to health, or contaminated in such a way that it would be unreasonable to expect it to be consumed in that state.

3.1.3.5 Optimisation process of Amtewa meal

The optimisation process involved the stepwise identification, combination and re-combination of macro and micronutrients obtained from the results of the proximate analysis from the local food sources in Nigeria **(Section 4.6 for details)**. The resulting optimised macro and micronutrients were formulated **(Section 3.1.3.5.1)** and sensory evaluation test conducted **(Section 3.1.3.5.2)** to produce Amtewa meal. This meal is nutrient-dense, providing on average, 10% to 20% additional energy above of the Reference Nutrient Intakes (RNIs) per adult serving of 100 g of product.

3.1.3.5.1 Formulation of Amtewa meal (the intervention meal)

Based on WHO recommendations on additional energy requirements for asymptomatic (10%) and symptomatic (20–30%) PLWH and the EARs energy for male (2500 kcal/d) and female (2000 kcal/d), the average EARs was calculated as 2250 kcal/d.

Three variations of Amtewa meal: Amtewa meal A (310 kcal/d), Amtewa meal B (354.92 kcal/d), Amtewa meal C (385.40 kcal/d) were formulated (Figure 3.7) to contain an average of 10–20% additional energy for both asymptomatic and symptomatic PLWH. These three formulations were subjected to sensory evaluation (Figure 3.8). The most suitable formulation from the sensory evaluation (Amtewa meal B 354.92kcal/d) was formulated and provided in a 100g sealed pack of Amtewa meal. The 100g Amtewa meal containing 50g soya beans, 15g moringa leaves 15g of carrot roots and 20g of millet was dispensed to PLWH for daily consumption (up to 6 months) as illustrated in the study design.

In similar studies, Amuna et al. (2004) proposed powdered blend in the development of FMM to which sugar, salt and milk could be added to increase the sensory characteristics of the product and for further energy and nutrient enrichment. Oldewage and Vaal University (2006) developed a culturally

acceptable and affordable food product using the FMM concept. The product was developed for specific target population and their nutritional need based on the staples use in the household. Also Amuna and Vaal University of Technology (2006) employed the concept in their research on the “Industrial and Dietetic application of FMM concept in meeting the nutritional need of vulnerable group in South Africa. A food-to-food fortification approach assumes that there are locally available naturally rich food sources (Rajput, 2012).

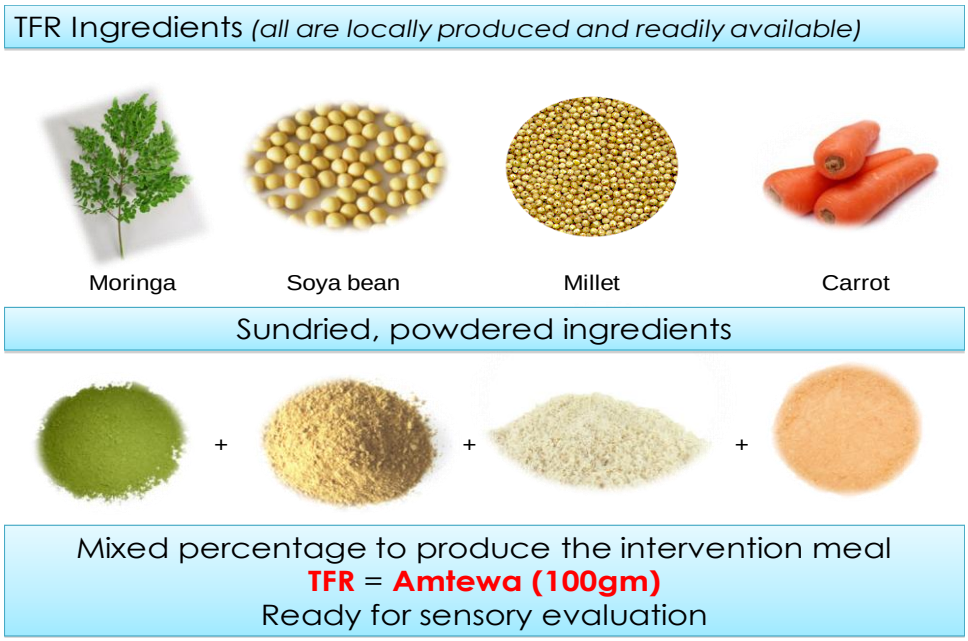


Figure 3. 6: Mixed percentage of Amtewa meal

Figure 3. 7: Amtewa meal formulated and packaged as 100g weight

3.1.3.5.2 Sensory Evaluation – Amtewa meal

A descriptive sensory analysis test (Hedonic scale) was used to determine the preferred Amtewa meal formulation out of the three variations. In this method, a ranking or ordering scales was adopted e.g. preference between products A, B, C: 1 most preferred, 2 intermediate preferred; 3 least preferred. To ensure the quality of reporting by the panelist, they were instructed to please take a drink of water before tasting Amtewa samples and to please take a sip of water between samples. Also booths were used as the sensory panel room to prevent panelists from being distracted and interacting with other panelist – unbiased evaluation. The booths were computerised and the panelists entered the answers to the sensory assessment questions (preference in terms of colour, taste, smell and texture) electronically. The booths were also illuminated with white lights to ensure that features that change environmental conditions such as different colours of light to mask the samples were avoided Figure 3.8).

The three variations of Amtewa meal: Amtewa meal A (310 kcal/d), Amtewa meal B (354.92 kcal/d, Amtewa meal C (385.40 kcal/d) were prepared according to the direction of use on the packaging. Seven participants in different booths were served with Amtewa meal A, B and C each in different containers for consumption to determine their preference in terms of colour, taste, smell and texture. Other questions that deal with quality of Amtewa meal were questions with the objective of describing liking or acceptability of Amtewa meal. These questions include: Do you like this product? How much do you like this product on a scale of 1 to 10, where 1 = dislike extremely, and 10 = like extremely? Is this product acceptable? What do you like most about this product? Is product A better than product B? Which of the three products A, B and C do you prefer?

Six of the panelists selected Amtewa meal B (354.92 kcal/d) as their preferred formula. This represents 85.71% preference compared to Amtewa meals A and C. Based on this result, Amtewa meal B (354.92 kcal/d) was formulated as the intervention meal according to the study design.

Figure 3. 8: Amtewa meal sensory evaluation test

3.1.4 Intervention Setting, Assessment tools and Ethical consideration

3.1.4.1 Intervention Setting

The setting for the study was the State House Medical Centre Abuja (SHMCA), Nigeria. SHMCA is a secondary health institution recognised by the Federal Government of Nigeria for the care and management of PLWH. Presently, the institution is involved in intervention programmes such as Voluntary Counselling and Testing (VCT), Prevention of Mother to Child Transmission of HIV (PMTCT), Paediatric Antiretroviral Treatment (PAT) and Adult Antiretroviral Treatment (AAT).

3.1.4.2 Measurements of research variables

Biochemical, anthropometric measurements and the nutritional intervention clinical trial were undertaken at Department of laboratory medicine and the HIV clinic of State House Medical Centre Abuja (SHMCA) Nigeria, one of the centres in Nigeria recognised for the care and management of HIV/AIDS patients.

3.1.4.3 Research assessment tool

The assessment tool was designed with the aim of achieving a multi-dimensional concept of quality research. The various sections in the assessment tool include

items to assess quality according to several domains including quality of reporting, methodological rigor and conceptual depth. Critical appraisal of the assessment tool was ensured after the pilot study (n=100) to ensure adequacy of reporting detail on the data sampling, data collection and analysis in the scale-up (n=400). See appendix 3.8

3.1.4.4 Study Variables/indicators (Nutritional assessment; biochemical indices; physical and anthropometric measurements)

Specific research variables (biochemical, physical and anthropometric) were selected from the assessment tool to test the hypothesis. These variables (Table 3.2) are conditions that are changeable during the course of the intervention.

Table 3. 2: Research variables and rational for inclusion

Research variables	Indications/ Rational for inclusion	Normal values	*Literature support (Box 3.3)
CD4 count	Immune status	500 – 1600 cells/mm^3	
MUAC	Nutritional status	>23.5cm and <32cm	
BMI	Nutritional status	>20kg/m^2 and <30kg/m^2	
Random glucose	Presence of Diabetes	<200g/dl	
Total protein	Immune dysregulation	60 – 85g/L	
SGOT	Liver abnormalities	5 – 35 IU/dL	
PCV	Clinical Anaemia	42 – 52% Male 35 – 47% Female	

Box 3.3: Literature support

***Encyclopedia of Surgery 2014; NIH and NIDDM, 2012; McAuley, 2012; Pribram, 2011**

3.1.4.5 Statistical analysis

Anthropometric measurements *(BMI, MUAC)* and biochemical indices *(RG, TP CD4 count, SGOT, PCV)* were conducted at the commencement of the study and in the third and sixth months of the pilot interventions. This was replicated in the larger scale study.

All collected assessment tools were revised for completeness and data analysed using SPSS Statistical Software. Simple frequencies were used for data checking. Also, simple descriptive statistics was 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.

3.1.4.6 Training of Trainers (TOT) and Quality control

The TOT programme was designed for individuals with a strong skill and experimental background in HIV care and quality management. It provided participants at the training (research team) basic knowledge on the research and exposure to some rigorous face-to-face challenges to be encountered with PLWHA. It was also designed with the demands of the HIV clinic in mind and significant portion of the TOT Program has been organised into "pre-work," a format that allows participants the flexibility to determine when they do the work (appendixes 3.9 & 3.10).

3.1.4.7 Ethical consideration

Research that involves human participants needs to go through a formal process of research ethics review (University of Leicester, 2006). Although this research required collection of in-depth high quality data from those most closely affected by HIV disease, however, thoughts that may be tempting to consider unethical research practice in order to try to obtain and/or retain some of the data was avoided (Wellcome Trust, 2014; UKRIO, 2006 – 2014).

The study design took responsibility in ensuring that:

- the level of risk is justified by the importance and relevance of the research study;
- the risk is unavoidable within the study's objectives;
- in absolute terms, the level of risk is minimised;

- participants were fully aware of the level and nature of the risk before they agree, freely, to take part in the study;
- precautions were in place to deal adequately with the effect of participation.

Basic Principles of Ethical Practice

1. Informed Consent

Participants knew exactly what they were being asked to do, and what the risks were, before they agreed to take part.

An Information Sheet was used to provide potential participants with information about the study (appendix 3.3) and was assured that the storage of data will comply with the Data Protection Act and the University of Westminster's Data Protection Code.

2. No Pressure on Individuals to Participate
3. Respect Individual Autonomy
4. Avoid Causing Harm
5. Maintain Anonymity and Confidentiality

Research ethics review forms were submitted at SHMCA and UoW and the ethical approvals from the two centres were received before commencement of the research (appendixes 3.1, & 3.2) (University of Westminster, 2013/14 v2; UKRIO, 2006 – 2014; Wellcome Trust, 2014).

Risks and Discomforts

Conventionally, HIV patients on management visit the hospital every three months for laboratory investigations such as CD4 cell count, viral load tests and other investigations (Hematology, blood chemistry and urinalysis). Naturally, the discomfort of the needle prick prior to sample collection may psychologically affect some participants that have phobia for injections.

Furthermore, participants will be subjected to a research assessment tool and anthropometric measurements. These may constitute some stress to the participants. Administration of micro and macronutrient combination to participants for daily consumption is expected to have no side effects due to the fact that the quantities to be consumed were within the limit for recommended

intake for adult; however these products may constitute an additional stress as a result of adherence to the prescription on the product.

Measures to reduce risks and Discomforts

Participants were reassured of lesser needle prick pain. Trained laboratory scientists responsible for the sample collection implemented an improved technique (e.g. immersing participants' fingers in warm water before sample collection) of sample collection with minimal pain on study participants.

Other methods adopted to reduce risks and discomforts were:

1. Educate the participants on the benefits of adhering to the recommended micro and macronutrient combination on improving their quality of life
2. Trained research team ensured compliance to assigned responsibilities in data collection with the assistant of a second member of the team to ensure accuracy.

3.1.4.8 Legal Rights

The research participants were assured that they were not waiving any of their legal rights by signing the informed consent document.

3.1.5 Intervention strategy – Planning, Implementation and Evaluation phases (Figure 3.9)

3.1.5.1 Planning Phase

The planning phase focused principally on required project planning work. The purpose of this phase was to plan all project processes and activities enumerated below:

Administrative procedures

The following documents were developed and submitted to appropriate authorities for approval before the commencement of the research:

- Registration at the University
- Development of registration document
- Ethical approvals in UK and Nigeria institutions (see appendixes 3.1 & 3.2)
- Identifying the necessary logistics before implementation – these include: sample size, study participants, teams' role and responsibilities (appendix 3.3) etc.

Visit to collaborating institutions

The institutions visited were London Metropolitan University, National Institute for Pharmaceutical Research and Development, Abuja – Nigeria and State House Medical Centre, Abuja laboratories.

London Metropolitan University superlab offers unparallelled teaching and research in nutrition, dietetics and micronutrient analysis.

National Institute for Pharmaceutical Research Development, Abuja provided training to how to undertake research and development work on, biological products including pharmaceutical raw materials from indigenous natural resources and by synthesis using appropriate science and technology methodologies in the formulation of the intervention meal.

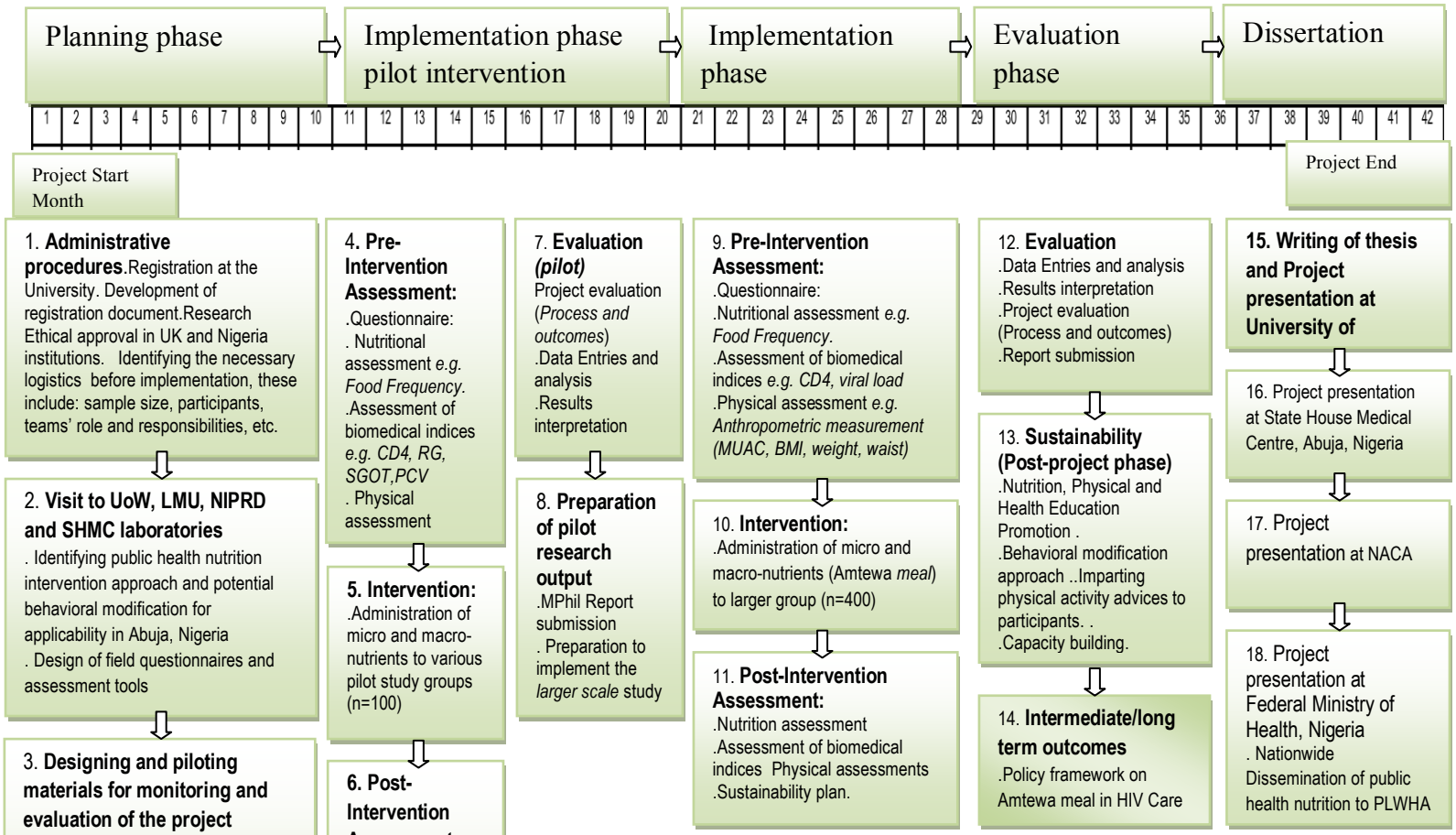


Figure 3. 9:Conceptual framework for public health nutrition intervention programmeto attenuate the progression of HIV to AIDS among PLWH in Abuja, Nigeria. Total Duration 42 months (October 2010 – March 2014).

3.1.5.2 Intervention strategy – Implementation phase

The implementation phase is associated with certain deliverables as described in sections on pilot and scale-up interventions described below:

Pilot study (n=100)

Pilot experiments are usually carried out before large-scale quantitative research in an attempt to monitor, safe time and avoid money being wasted on an inadequately designed project. This pilot study was carried out on PLWH (N=100) and most of them also progressed and formed part of the final sample size in the scale–up phase. It was a potentially valuable insight that ensured that anything missing in the pilot study was added to the scale-up intervention to improve the chances of a clear outcome.

Pre - intervention assessment – Pilot study

The following pre – intervention assessments were carried out.

1. Assessment tool: A WHO approved questionnaire for public health nutrition intervention was reviewed and adapted for the purpose of this research and developed into an assessment tool (appendix 3.8).
2. Physical assessments e.g. anthropometric measurements (MUAC, BMI, Weight, and Height) were recorded and filled on the appropriate column on assessment tool for all the study participants at the beginning of the study.
3. Assessment of biochemical indices e.g. CD4 cell counts, RG, SGOT, PCV test were conducted by the trained Laboratory Scientist and also recorded in the appropriate column on the research assessment tool form at the beginning of the study.

Intervention phase – (duration: six months)

This phase involved the following steps:

Step 1: Randomisation of research participants (Figure 3.2), administration of micro and macronutrients (Amtewa meal) to the various groups in the pilot study (n=100) and effective monitoring within the six months period of the intervention.

Step 2: Monitoring at zero, three and six months interval

Effective monitoring to ensure that the research continues to conform to approved ethical standards and procedures. This was achieved through:

- Standardised but adaptable approaches in the design of the intervention
- Local feedback from the study participants to monitor adherence to HAART and Amtewa meal intervention e.g. checking left over Amtewa meal not consumed
- Minimal data collection by the research team
- Routine surveillance (health information system) to various units in the hospital during data collection
- Community and household surveys to assess study participants' knowledge/adherence to the direction of use of Amtewa meal as indicated on the package
- Support to the research participants in the form of stipend for transportation to assist and ensure uninterrupted hospital appointment during the research.

Step 3: Post – intervention assessment – Pilot study

Re-assessment of anthropometric (BMI, MUAC) and biomedical indices (CD4 cell count, PCV, TP, RG, SGOT) for all the groups (ART & Pre- ART) at three (3) months and six (6) months interval.

Step 4: Evaluation phase

Project evaluation (process and outcomes): This phase involved the comparison of results obtained from pre–intervention assessment phase and the post–intervention assessment phase. Data entries and analysis were done after the sixth month of pilot intervention.

Post Pilot to Scale up Evaluation phase

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) (Table 3.1 and 3.3).

Table 3. 3: Post Pilot to Scale up evaluation

	Number	1-6	6-12	Duration (months)
Pilot	100	100	100	12
Scale-up	400	-	300	6
Total		100	400	

Scale up intervention (N=400).

The results of the pilot intervention are evidence-based and cost effective. This knowledge and innovative technology was demonstrated in a larger population (n=400) to ascertain the impact of the Amtewa meal in a larger sample size.

Pre - intervention assessment – Scale up study

The following pre–intervention assessments were carried out.

1. Research assessment tool: A WHO approved research assessment tool for public health nutrition intervention used for the pilot intervention was reviewed and adapted for the purpose of this research (appendix 3.8). No modification was made to the research assessment tool.
2. The research team were re-trained to ensure strict compliance to the research procedure especially during data collection
3. The use of xylocaine ointment was recommended to the Laboratory Scientist for application on the injection site during sample collection to reduce the pain for participants with phobia for injection.
4. Physical assessments e.g. anthropometric measurements (MUAC, BMI, Weight, and Height) were recorded and filled on the appropriate column on assessment tool form for all the study participants at the beginning of the study. Each measurement was done twice and double- checks by an observer to ensure that correct data were entered into the assessment tool.
5. Assessment of biomedical indices e.g. CD4 counts, RG, SGOT, PCV test were conducted by the trained Laboratory Scientist and also recorded in the appropriate column on the research assessment tool form at the beginning of the study.
6. WHO prequalified the Laboratory Medicine Department of SHMC, Abuja as an accredited centre certified for laboratory investigations.

Intervention phase – six months

This phase also involved the Randomisation of scale up research participants, administration of micro and macro – nutrients (Amtewa meal) to the various groups (n=400) and effective monitoring within the six months period of the intervention.

3.1.5.3 Intervention strategy – Evaluation phase

The evaluation phase was an ongoing phase from the commencement of the research to the completion. The participants' hospital appointments were monitored to ensure compliance, Research Team was monitored to ensure that the quality data were not compromised. Data entries were double checked, Results were analysed, interpreted and report presented for the MPhil transfer and PhD Thesis.

3.1.6 Sustainability plan

Planning for sustainability in this public health nutrition intervention was based on the results of the pilot intervention. A sustainability plan was proposed to integrate nutrition intervention in HIV care and support programme at the study centre (Figure 3.10) following the outcome of the outcome of the research.

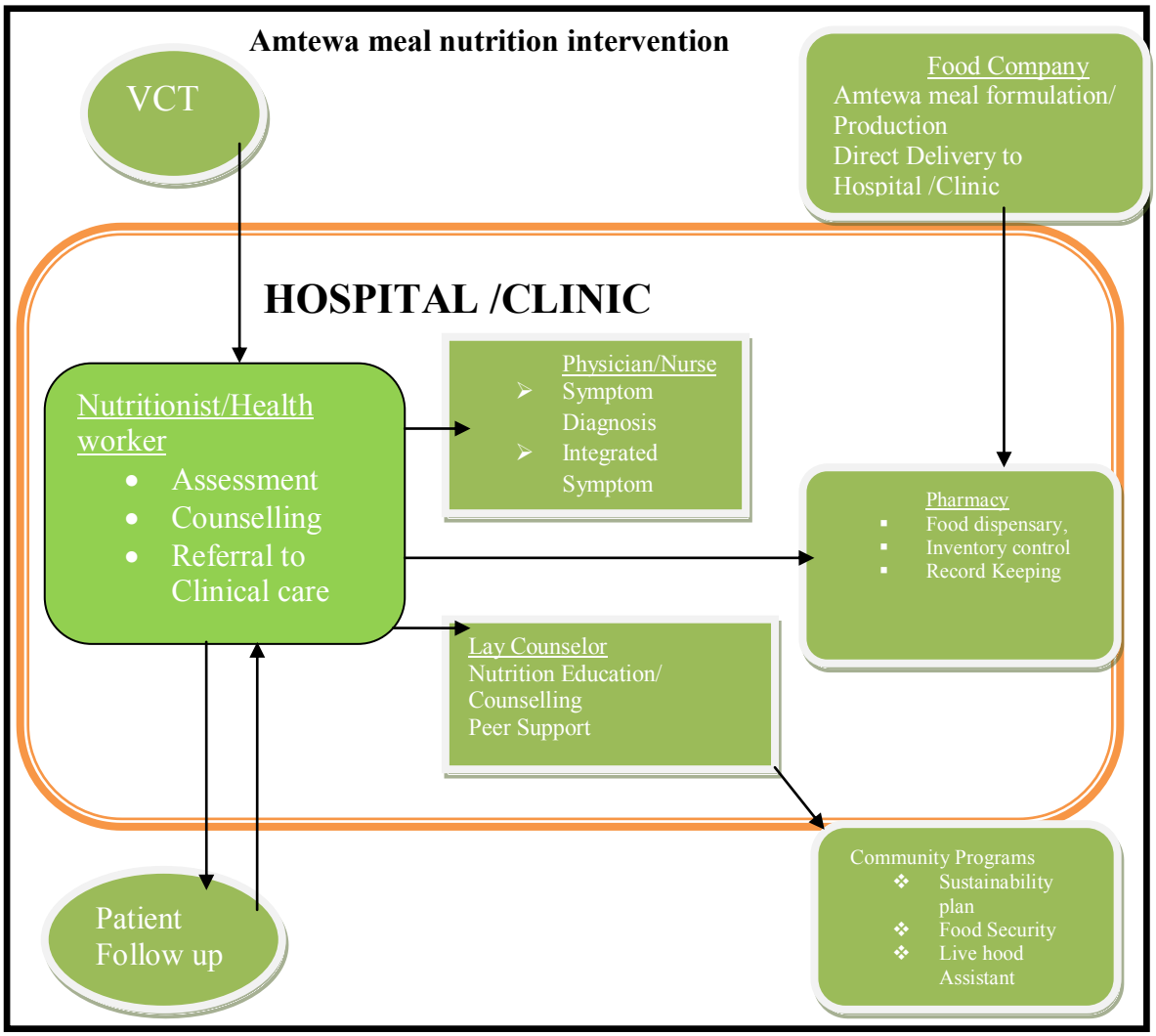


Figure 3. 10: Proposed interaction between a Nutritionist and other Health care providers in an HIV treatment site with the introduction of 'Amtewa meal'

The sustainability plan was proposed to ensure that PLWHA visiting the HIV clinic for their hospital appointments or referred from the voluntary Counselling and testing unit (VCT) would be assessed by the Nutritionist (nutritional assessment and Counselling) before referring them to the physician and other health care provider as illustrated in Figure 3.10 above.

Chapter 4

Results: Pilot intervention

4.1 Introduction

This pilot intervention was a rehearsal for subsequent full-scale intervention. It helped to fine-tune later larger study and also allow the researcher to experience the more practical aspects of implementing the study, such as determining the number of research team members needed to handle recruitment and data collection or identifying special requirements for participants' follow-up.

4.2 Summary of Materials and Methods

Enrolled participants were subjected to a comprehensive assessment (appendix 3.8), which include variables such as anthropometric measurements and biomedical indices. These study participants continue standard treatment for PLWH by the SHMCA (nutritional Counselling, vitamin supplements) and half of each arm (Pre-ART and ART) were dispensed Amtewa meal for daily consumption for six months (Figure 3.2).

Pre and post intervention assessments were carried out as specified in the study design (Chapter 3).

4.3 Results (Pilot) – Proximate Analysis

Tables 4.1, 4.2 and 4.3 demonstrate the macro and micronutrients contents in each sample employed in the optimisation and formulation of Ametwa meal.

Table 4.1 shows the weight of 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 analysed, 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 the bulk of the macronutrient and therefore 70% of Amtewa meal.

Table 4.2 shows the total weight of micronutrients in each of the analysed samples. The four samples contain micronutrients (vitamin A from carotene, vitamin B, vitamin C, Ca, Mg, K, Na, Mn, Fe, Cu, Zn and phosphorus) essential for immune boosting (Beach et al., 1992; Baum et al., 1995; WHO, 2005; Fawzi et al., 2003; 2004; 2005; Drain et al. 2007; Hurwitz et al., 2007). Soya bean seed and moringa leaves contain higher amounts of each micronutrient than carrot and millet as illustrated in the Table 4.2. 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 (COMA, 1991) 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 4.3 shows the EARs energy per day for male is 2500 kcal/d and for female is 2000 kcal/d (average 2250 kcal/d). The optimised 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 in addition to their normal daily nutritional intake.

4.4 Information on the package: Direction of use

Boil 0.5 litres of clean water in a pot. Gradually pour 100g of Amtewa meal into the boiling water, stir and maintain boiling for three to five minutes. Empty the boiled mixture in a container and allowed to cool to a temperature suitable for consumption.

Recommendation: Consume the whole content. Addition of milk and sugar is optional to improve taste. Discard left over.

4.5 Optimisation of macro and micronutrients (Amtewa meal)

Reducing or eliminating undernutrition has the potential to significantly slow progression of HIV/AIDS disease and decrease its severity (ADA, 2010). Severe food insecurity was found in over 40% of all households in Nigeria (IITA, 2004), hence the need to optimise macro and micronutrients in HIV care (Table 4.1).

Altered levels of plasma proteins, micronutrients, and other nutrition-related markers have been documented early in HIV disease process and have been associated with increased risk of mortality in HIV infection (ADA, 2010). Some of these problems may have occurred independently and before the use of ART (ADA, 2010).

Table 4. 1: Macronutrients compositions in g/100g of optimise 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
DRV[1] (g/100g)	100	50		15		35		
DRV[1] (Percent)	100	50		15		35		

[1]Dietary Reference Value for Food Energy and Nutrients for the United Kingdom (COMA, 1991).

Table 4.1 shows the macronutrient composition of optimise Amtewa meal (in 100g product as additional consumed meal) compared to the Dietary Reference Values for adults as a percentage of daily total energy intakes (percentage of food energy).

Table 4. 2: 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	2.5	0.008	352	0	0	0	0	3
MORINGA	300.4	55.2	198.6	0	0	4.23	0.09	0.5		30.6	2.84	0.4	3.08	1.23	2.60
CARROT	0.48	0.12	1.34	0.17	0.3	0.13	0.12	0.5		0	2.2	0	0	0	4.9
MILLET	12	23.1	90	5.4	0.3	6.18	0.12	0.5		59.4	0	0	0	0	0
TOTAL	451.4	218.4	1188.4	6.57	0.6	18.4	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	700000	1.0	1.3	17	40
RNI (females)[1,2]	700	270	3500	1600		14.8[3]	1.2	7.0	0.06	550	600000	0.8	1.1	13	40

[1]Dietary Reference Value for Food Energy and Nutrients for the United Kingdom. RNI for Carotene is in mcg

[2]19 years and above

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

Table 4.2 shows the micronutrient composition of optimise Amtewa meal (in 100g product as additional consumed meal) compared to the Reference Nutrition intakes daily.

Table 4. 3: 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]

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

Table 4.3 shows the daily total energy and percentage composition of Amtewa meal (in 100g product as additional consumed meal) compared to the Estimated Average Requirements for energy as recommended by Dietary Reference values for Food Energy and Nutrients for the United Kingdom.

Table 4. 4: Anthropometric and biomedical indices (see details of table title below)

Progression of participants' anthropometric and biomedical indices from inclusion to six months for participants on Amtewa meal and those without the meal

	Group (Pre-ART) Ia (Test)				Group (Pre-ART) Ib (Control)					Group (ART) IIa (Test)				Group (ART) IIb (Control)			
Time (month)	N	Min	Max	Median	N	Min	Max	Median	Time (month)	N	Min	Max	Median	n	Min	Max	Median
Group I : Weight(Kg)									**Group II : Weight(Kg)**								
0	25	49	80	64.5	25	48	97	74	0	25	46	97	64	25	47	97	61.5
3	25	51	81	65	25	50	96.5	73.5	3	25	47	96	65	25	45	96	61
6	25	53	82	66	25	51	96	72.5	6	25	49	97	69.5	25	45	94	60
Group I : BMI(Kg/m^2)									**Group II : BMI(Kg/m2)**								
0	25	18.8	32	23.63	25	18.75	32.5	26.63	0	25	17.92	34.35	22.91	25	19.6	32.55	23.68
3	25	19.32	32	23.82	25	19.53	32.27	26.5	3	25	18.28	34.35	23.09	25	18.92	32.21	23.51
6	25	20.22	32.89	24.29	25	19.92	32.1	25.97	6	25	18.28	34.73	24.24	25	18.58	31.54	23.33
Group I : MUAC(cm)									**Group II : MUAC(cm)**								
0	25	20	35	29	25	27	41	30	0	25	23	40	28.75	25	22	37	27
3	25	21	35	29.5	25	25	40	30	3	25	25	39	29.5	25	20	35	27
6	25	22	35	30	25	25	40	29.75	6	25	25	40	31	25	20	35	26
Group I: PCV (%)									**Group II: PCV (%)**								
0	25	31	45	38	25	33	45	37	0	25	31	45	37	25	30	51	35.5
3	25	32	42	38	25	34	48	38	3	25	33	46	37.5	25	31	49	36
6	25	31	43	40	25	32	45	36	6	25	34	47	39	25	30	48	36
Group I: CD4cells/mm^3									**GroupII CD4cells/mm^3**								
0	25	370	1105	573	25	340	1140	599	0	25	230	1029	399.5	25	205	820	313.5
3	25	320	1120	539.5	25	320	1282	595	3	25	190	1107	426	25	180	812	300
6	25	246	1122	560	25	325	1200	599	6	25	198	1110	442.5	25	195	800	292.5
Group I : TP(g/100ml)									**Group II : TP(g/100ml)**								
0	25	57	112	72.5	25	62	89	77.5	0	25	52	95	69	25	55	95	71.5
3	25	59	112	79	25	65	89	77	3	25	59	98	73.5	25	57	98	72
6	25	60	108	79	25	65	88	76	6	25	61	93	71.5	25	59	96	75
Group I : RGmg/100ml									**Group II: RGmg/100ml**								
0	25	70	176	109.5	25	73	211	88	0	25	69	169	107	25	68	207	98
3	25	72	140	104.5	25	72	180	95	3	25	74	136	104	25	70	200	103.5
6	25	78	140	112.5	25	70	124	94	6	25	79	125	108.5	25	71	190	95
Group I : SGOT (I.U/L)									**Group II : SGOT (I.U/L)**								
0	25	7	41	13.5	25	7	26	12	0	25	7	20	11	25	6.5	27	11
3	25	7	25	12	25	7	24	10.5	3	25	7	17	9.5	25	6	24	10
6	25	8	29	12.25	25	7.2	21	10	6	25	7	23	10	25	6	25	9

MUAC (Mid upper arm circumference), PCV (Packed cell volume), TP (Total protein), RG (Random glucose), BMI (Body mass index)

4.6 Participants' characteristics at baseline, three months and six months follow up

4.6.1 Anthropometric measurements

Median increase in MUAC of participants in the Pre-ART group (with Amtewa) was 3.4% while for the ART group (with Amtewa), it was 7.8%. On the other hand, control groups (without Amtewa) for both Pre-ART group and ART group had a decrease in their median MUAC by 0.8% and 3.7% respectively as illustrated in Table 4.4.

Similarly, Figure 4.1 shows that the mean MUAC for Pre-ART (with Amtewa) group increased by 3.25% while mean MUAC of ART (with Amtewa) group, increased by 5.58% after six months. This is statistically significant (P = 0.05).

Impact of Amtewa meal on mean MUAC

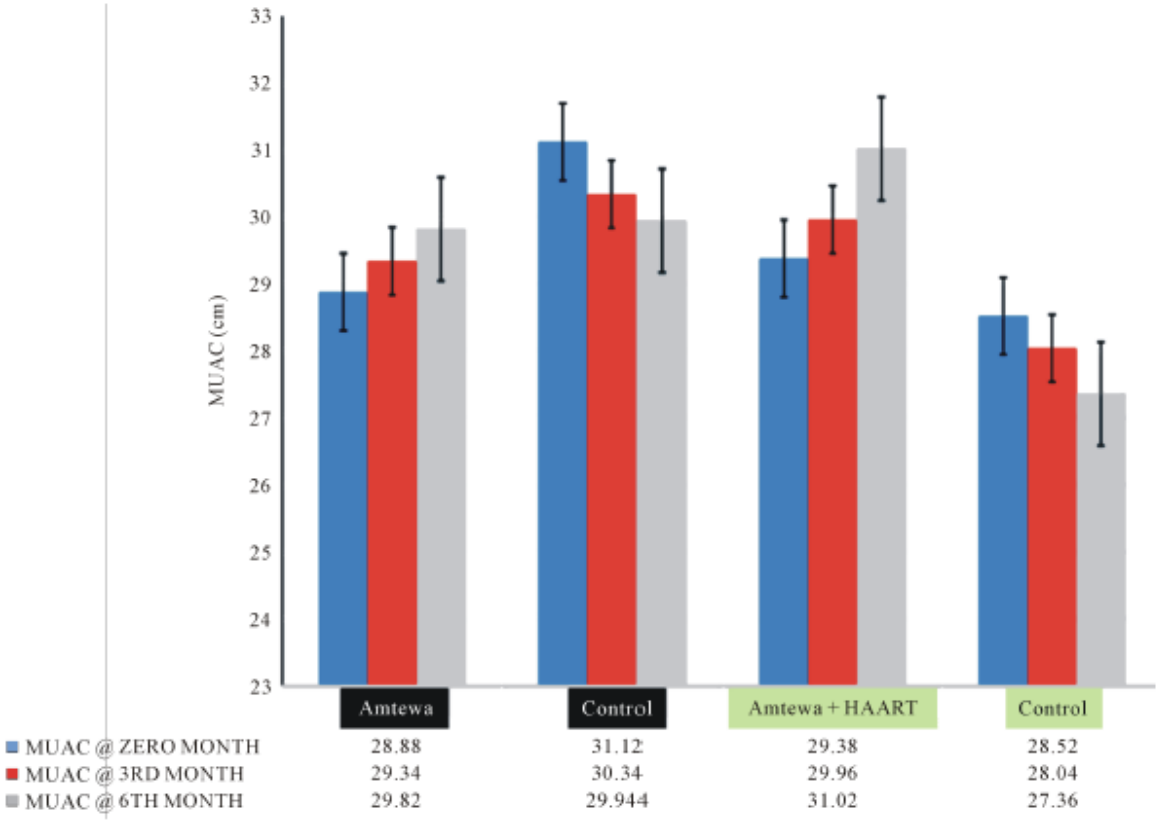


Figure 4. 1: Bar graph showing the impact of Amtewa meal on Mean MUAC (cm) of study participants

4.6.2 Immune status of study participants (pilot)

The variations of CD4 lymphocytes count for all the groups (Table 4.4) shows that the median CD4 count of participants on Pre-ART with Amtewa meal group decreased by 13cells/mm^3 (statistically significant) while that of ART with Amtewa

meal group increased by 43cells/mm^3. Also, participants in Pre-ART without Amtewa meal group maintained a median CD4 count (599cells/mm^3) while CD4 count for ART without Amtewa meal group decreased by 11cell/mm^3. Similarly, mean CD4 count for Pre-ART with Amtewa meal group increased by 0.69% and ART with Amtewa meal group increased by 9% (Figure 4.2). This indicated clinical and statistically significant differences between the two groups of participants on the meal and those without the meal.

Impact of Amtewa meal on Mean CD4 count.

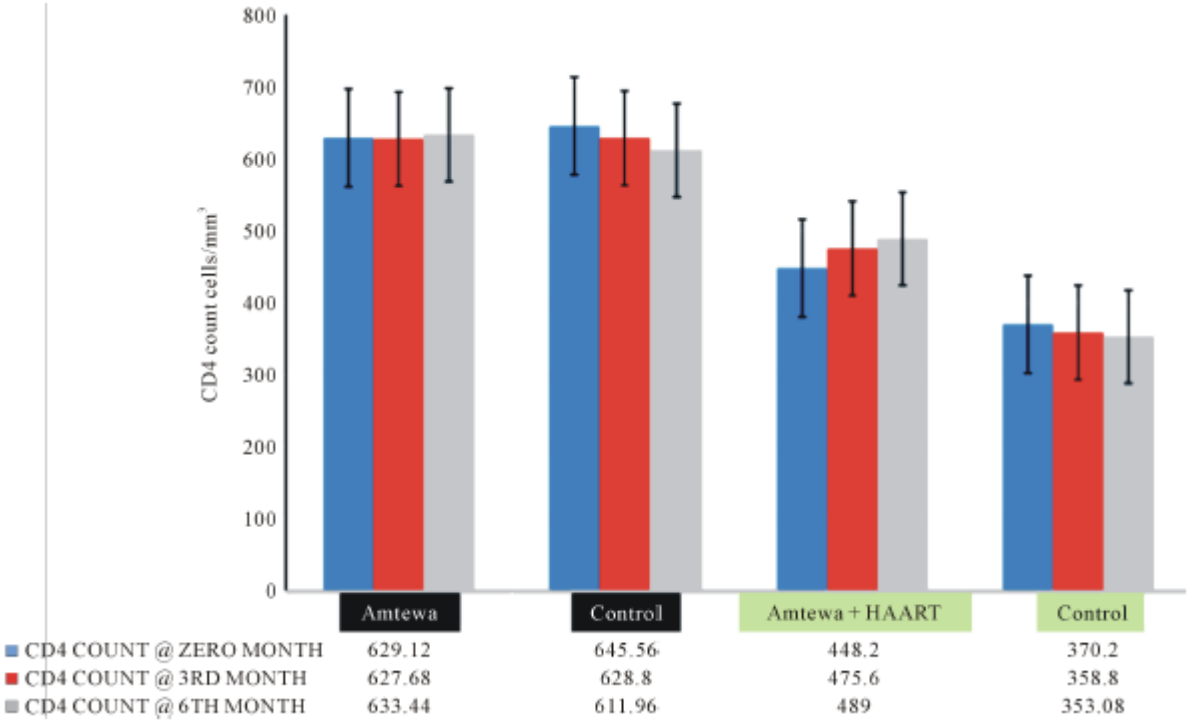


Figure 4. 2: Bar graph showing the impact of Amtewa meal on Mean CD4 (cells/mm3) cell counts of study participants (n=100)

4.6.3 Clinical end points

Table 4.4 shows the result of other biomedical indices (Random glucose, packed cell volume, total protein and serum glutamic oxaloacetic transaminase). The median random glucose (normal range=140-200mg/100ml), packed cell volume (normal value=40% female, 45% male), total protein (normal range=60-83g/100ml) and SGOT (normal range=0-12 I.U/L) were within the normal range.

4.7 Discussion (pilot intervention)

This study describes participants in the pilot intervention (n=100) that were divided into four groups of 25 participants in each group (Figure 3.2). Fifty PLWH

were monitored for adherence to Amtewa meal nutrition intervention (354.92 kcal per day) whiles the other fifty were control groups (without the meal) for six months according to the study design.

Comparison of before and after the pilot trial suggest that in two groups on the meal, there were mean increases in the CD4 lymphocyte count of the Pre–ART Test (with Amtewa meal) group and CD4 count of ART Test (with Amtewa meal) group. This increases are statistically significant (*p=0.05*) compared to the control groups without Amtewa meal with a median decrease in Pre–ART control group, mean decreases in Pre-ART control and ART control groups respectively. The effect of Amtewa meal was more pronounced in participants on HAART than those not on HAART. CD4 count increases with Amtewa meal is supported by Paton et al. (2006) in his study on the impact of malnutrition on the survival and the CD4 count response in HIV-infected patient starting ART (N=394). According to Parton et al. (2006) those on HAART showed a significantly (*P=0.03*) greater increase in CD4 count than those not on HAART.

Also, median BMI increase (Table 3.4), median MUAC increase (Table 4.4) and mean MUAC increase (3.25% in Pre-ART with Amtewa meal group and 5.58% in ART with Amtewa group see Fig. 4.1). This is supported by Chlebowski et al. (1993), Chlebowski et al. (1995), Suttmann et al. (1993) and Suttmann et al. (1995), who demonstrated a 4 and 3kg gain in body weight in their HIV studies on nutritional support (Pichard et al., 1998). Also in another randomised double-blind controlled study on HIV-infected patients (N=55) on oral nutritional supplementation enriched with fish oil and fish oil-arginine for 6 months, gain of body weight and fat mass were approximately 2 and 1kg respectively for patients on the nutritional support. Furthermore, in the study participants', median random glucose (normal range = 140-200mg/100ml), PCV (normal value=40% female, 45% male) total protein (normal range=60-83g/100ml), and SGOT (normal range=0-12 I.U/L) were within the normal range. Protein and energy improvement by Amtewa meal suggests that nutritional supplements in clinically stable patients are selectively efficient to increase nitrogen intake and stimulate protein tissue deposition (Pichard et al., 1998).

On the other hand, decreases in the mean CD4 cell count of pre ART and ART groups that were not on the meal (Fig.4.2) and decrease in median CD4 count in ART group (control) are supported by Kaiser et al. (2006) who demonstrated a

mean absolute CD4 count increase by an average of 65 cells in the micronutrient group versus a 6-cell decline in the placebo group at 12 weeks.

Although in both Pre-ART (control) and ART (control) groups, there was a decline in the median BMI (normal BMI=18.5-25kg/m^2) and mean MUAC from 28.52 to 27.36cm (normal ≥23cm for male and ≥22cm for female) at the sixth month of the study (Table 4.4, Figure 4.1), however, the mean and median values were not suggestive of underweight or undernutrition. This could be attributed to the fact that in the study design, all the groups were given nutritional counselling (Chapter 3) in addition to the multivitamin supplementation dispensed to PLWHA receiving care and support at the study centre (SHMCA). Tabi (2006) investigated the effectiveness of nutritional Counselling as an intervention to improve health outcomes for HIV positive patients in Ghana. The result shows a mean increase (from 43.86kg to 46.57kg for females and from 51.86kg to 55.23kg for males) in the body weight of the study participants (*p=0.001*). Nutrition counselling and intervention programmes must be sustained to maintain anthropometric parameters (BMI, MUAC) within normal values to decrease mortality. This is evident in the study (n=1657), Body mass index at time of HIV diagnosis: A strong and independent predictor of survival Van der Sande et al. (2004) states that a one unit decrease of BMI resulted in 21% increase in mortality rate ($p<0.001$) after controlling baseline immune status (CD4 count).

Participants' compliance was also impressive with over 95% completing the 6 months trial. This might be explained by (1) the selection of participants willing to undertake proactively a prolong action that they believed potentially able to improve the course of their infection. (2) PLWHA attend their hospital appointment every month or once in two months for prescription refill and other investigations. Hence, the intervention programme with Amtewa meal was run concurrently with the HIV clinic.

Finally, daily micronutrient (antioxidant) supplementation improved body weight and body cell mass, reduced HIV RNA level, improved CD4 cell counts and reduced the incidence of opportunistic infections in small studies of adults with AIDS, including those on antiretroviral therapy (USAID, 2004). Larger clinical trials (Fawzi, et al., 2004; Hurwitz, et al., 2007) demonstrated that daily micronutrient supplementation increased survival in adults with low CD4 cell counts. The optimal formulation of a daily multiple macro and micronutrient supplement for HIV positive individuals is evident in the innovation of Amtewa

meal. These positive outcomes qualified *Amtewa* meal to the next *scale-up* intervention phase (n=400 participants) to ascertain its effectiveness on health status of HIV infected subjects and appraise its position within the National Health Services framework as innovative approach to attenuate the progression of HIV to AIDS in Nigeria.

Consequently, the model of Amtewa meal nutrition intervention was envisage on a Nigeria population and the recipe of macro and micro–nutrients cautiously selected from locally available and accessible food in Abuja, Nigeria. The meal is a natural product that requires low technology in the formulation and with affordable training workshop to consumer could be sustainable. Unlike most intervention programmes referenced above, the authors researched on synthetic macro and micronutrients (e.g. Kupka and Fawzi, (2002) study on zinc nutrition and HIV infection; Coutsoudis et al., (1995) investigated the effects of vitamin A supplementation on the morbidity of children born to HIV-infected women).
The study design was scientific and the variables (e.g. CD4 T-cell count, MUAC, PCV, SGOT) were standardised. The inclusion and exclusion criteria were strictly followed to ensure that the positive outcome could be attributed to the additional 100g (400 kcal) of the Amtewa meal which was consumed on daily basis by each participant in the Test group.

4.8 Planning the Scale-up intervention

4.8.1 Quality control measures and good practices to be adhered to in the larger scale intervention (N=400)

- The research assessment tool was refined to fulfill the study objectives and fit the study population. Subsequent monitoring plan (input, modifications, and revisions) were developed adopted through pilot testing.
- At the study site, research team was re-trained to ensure the flow of the study and assure quality of collected data.
- Revision of data forms (checking for completeness, logical response, etc) was performed at the study site at the end of each visit of the participant by the researcher.

- Regular visits and meetings were held for the research team to monitor field progress and the need to address any potential problems arising in the field.
- Ascertain the key role of Amtewa meal in increasing the CD4 count, BMI and MUAC of HIV infected subjects.
- Sustainability plan (from outcome of conclusion and discussion) and technology transfer for delivering the package of nutrition services (assessment, Counselling and Amtewa meal support) in SHMCA and other HIV treatment centres within Abuja, Nigeria.

Chapter 5
Results: Scale-up intervention

5.1 Introduction

This presented Larger Scale Intervention (n=400) results consist of one hundred study participants in the pilot study that overlapped into the scale-up phase and additional three hundred study participants recruited based on the sample size calculation and the sampling strategy. This implied that the study participants in the pilot study were monitored over a period of one year (12-month period). This larger scale intervention was an approach for organising sustainable changes with active involvement of the researcher, the research team and the study participants throughout the six months period of data collection.

5.2 Objectives

The specific objectives of the scale-up intervention study are:

1. To appraise the outcomes of Amtewa meal on health status of HIV infected subjects (*with CD4values above 200/mm^3*) at baseline and compare these at post- scale up intervention.
2. To evaluate the effectiveness of macro and micronutrient (Amtewa meal) intervention as possible measure to attenuate the progression of HIV to AIDS.
3. Draw up a sustainability plan for outcome of discussion and conclusion.

5.3 Summary of method

Enrolled participants (1000) were invited to participate in the research from all the HIV treatment centres in Abuja, Nigeria. Based on the sample size calculation, inclusion and exclusion criteria, 400 participants were selected through simple Randomisation (Figure 3.1). The selected participants were subjected to an "assessment tool'' (appendix 3.8), which includes demographic, anthropometric and biomedical indices. These study participants continued standard treatment for PLWH by the SHMCA (nutritional Counselling, vitamin supplements for Pre-ART, nutritional Counselling, and vitamin supplements for the ART group). Half of each group (Pre-ART and ART) were dispensed Amtewa meal for daily consumption for six months.

Pre and post intervention assessments were carried out as specified in the study design (Details in Chapter 3).

5.4 Results

This result section in the scale-up intervention was divided into results obtained from the demographic characteristics, anthropometric and biochemical indices of study participants.

5.4.1 Demographic Characteristics of the study participants

Sample size

Table 5.1 shows four hundred (400) PLWH were recruited for the intervention programme, however sixteen participants could not complete the six months duration of the research. All the research participants were above 18 years of age. Gender, age range and marital status of the various groups of the study participants are illustrated in the sections below.

Table 5. 1: Demographic characteristics of study participants (n=400)

	GENDER		AGE (years)					AGE RANGE (years)			MARITAL STATUS				
GROUPS	MALE	FEMALE	MEAN	MEDIAN	STD DEV	MIN	MAX	< = 30	31 - 50	> = 51	DIV	M	S	WID	WIDR
PRE ART TEST	36	60	45.57	45	9.965	20	65	8	56	32	0	68	26	2	0
PRE ART CONTROL	33	63	41.89	41.5	8.763	25	64	10	70	16	0	46	43	5	2
ART TEST	40	56	45.22	45	9.784	26	67	7	61	28	1	72	20	3	0
ART CONTROL	40	56	40.61	40	9.535	25	66	14	66	16	0	40	53	3	0
TOTAL	**149**	**235**	**NA**	**NA**	**NA**	**NA**	**NA**	**39**	**253**	**92**	**1**	**226**	**142**	**13**	**2**

Key: STD DEV: STANDARD DEVIATION; MIN: MINIMUM; MAX: MAXIMUM; DIV: DIVORCED; M: MARRIED; S: SINGLE; WID: WIDOW; WIDR: WIDOWER; NA: NOT APPLICABLE

Marital status frequency distribution of study participants

Figure 5.1 shows the marital status frequency distribution. It shows that 0.26% of the participants were divorced, 36.98% were single, 58.85% were married, 3.39% were widow and 0.52% was widower. The percentage of married participants may be a factor to explain the degree of adherence why most of the

participants completed the six months or one year intervention program. All marital status were represented with the highest representation from the married (n=226) and the lowest representation from the divorced (n=1). The cumulative frequency was 100%.

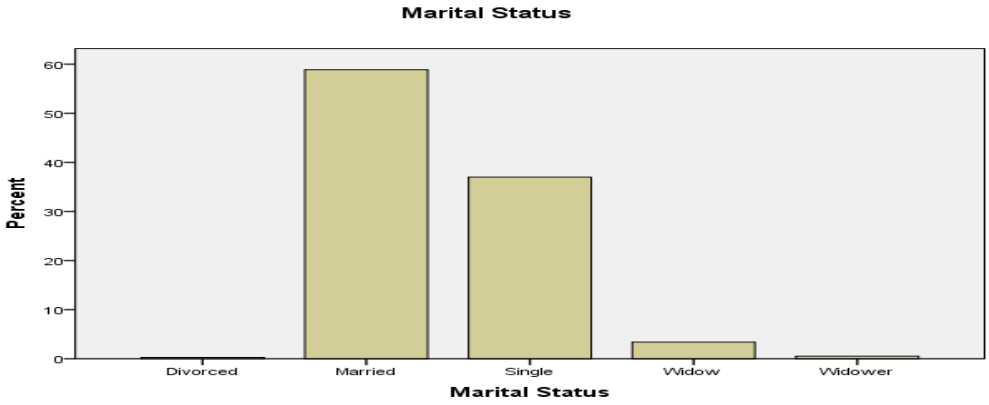


Figure 5. 1: Bar graph showing the marital status distribution of participants

The Bar graph above shows that 95.83% of participants were either married or single. The remaining 4.17% participants were the Divorced, Widow and Widower. This result reflects the inclusion criteria that excluded participants with a CD4 count less than 200cells/mm^3 (PLWAIDS) who may have lost either of their spouses from the complications of HIV which is often the case when the CD4 count is less than 200cells/mm^3.

Sex distribution of study participants in the scale-up intervention

Figure 5.2 shows the sex distribution of the study participants in the scale up intervention. 61.2% were female while 38.8% were male. This shows that more women are receiving treatment care and support in Abuja, Nigeria or women are interested in intervention programmes that will slow the progression of their disease condition (HIV) than men.

Figure 5. 2: Bar graph showing sex distribution of study participants in the scale-up intervention

The greater percentage of 22.4% of women more than men in the research as shown on the bar graph above was also an indication that a positive outcome in the research is likely to be sustained because they have the skill to educate their spouses. Also women are more likely to adhere to the intervention approach than men. Therefore, positive outcome in the nutrition intervention programme.

Age distribution of study participants in the scale-up intervention

Figure 5.3 shows that 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=400) was 43.32 years, median age (n=400) was 43years and the standard deviation (SD) was 9.721.

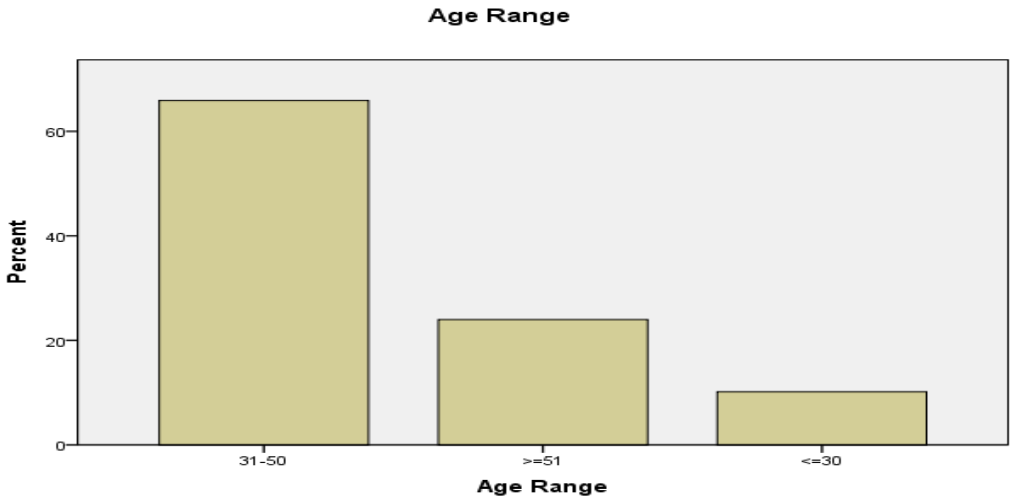


Figure 5. 3: Bar graph showing the percentage age range in years of study participants in the scale-up intervention

The bar graph above shows 89.9% of participants were above 30 years of age. This was an indication that participants above 30 years of age were more interested in the intervention approach and are less likely to be affected by stigma associated with HIV intervention programmes. The 10.1% of participants less than 30 years of age compared to the 89.9% (30 years above) were the group that was most likely affected by stigma. Therefore, intervention programmes 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.

5.4.2 Anthropometric parameters and Biochemical indices

Table 5. 2: Results showing Biochemical indices and Anthropometric parameters

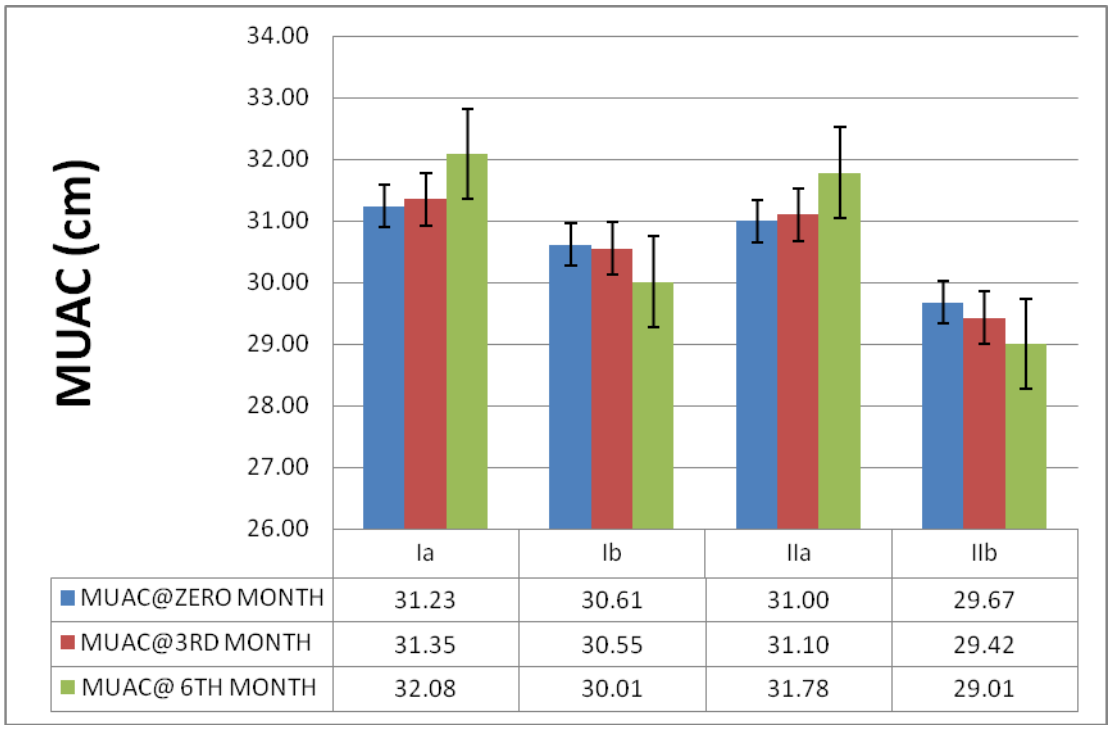
Anthropometric and Biochemical indices of study participants from inclusion to 6 months of the Amtewa meal nutrition intervention n=400

	Test (1a)	Control (1b)				Test (11a)		Control (11b)	
Duration (months)	**N**	**Mean**	**N**	**Mean**	**Time (months)**	**N**	**Mean**	**N**	**Mean**
Pre ART Weight (Kg)					**ART Weight (Kg)**				
0	96	68.20	96	69.73	0	96	66.27	96	65.24
3	96	68.81	96	68.59	3	96	67.47	96	64.80
6	96	70.83	96	67.39	6	96	69.15	96	63.76
Pre ART Height (m)					**ART Height (m)**				
0	96	1.66	96	1.65	0	96	1.64	96	1.63
3	96	1.66	96	1.65	3	96	1.64	96	1.63
6	96	1.66	96	1.65	6	96	1.64	96	1.63
Pre ART BMI (kg/m^2)					**ART BMI (kg/m^2)**				
0	96	24.75	96	25.54	0	96	24.64	96	24.55
3	96	24.97	96	25.12	3	96	25.08	96	24.39
6	96	25.70	96	24.68	6	96	25.71	96	24.00
Pre ART MUAC (cm)					**ART MUAC (cm)**				
0	96	31.23	96	30.61	0	96	31.00	96	29.67
3	96	31.35	96	30.55	3	96	31.10	96	29.42
6	96	32.08	96	30.01	6	96	31.78	96	29.01
Pre ART PCV (%)					**ART PCV (%)**				
0	96	39.81	96	37.73	0	96	38.74	96	37.54
3	96	39.74	96	37.82	3	96	40.39	96	37.84
6	96	43.18	96	37.71	6	96	41.84	96	37.57
Pre ART CD4 ($cells/mm^3$)					**ART CD4 ($cells/mm^3$)**				
0	96	621.09	96	600.34	0	96	418.53	96	351.81
3	96	635.36	96	559.79	3	96	443.93	96	322.65
6	96	660.26	96	536.01	6	96	469.26	96	306.07
Pre ART TP (g/100ml)					**ART TP (g/100ml)**				
0	96	73.23	96	72.48	0	96	70.44	96	67.93
3	96	74.56	96	74.15	3	96	71.98	96	68.06
6	96	76.10	96	74.89	6	96	72.85	96	71.18
Pre ART RG (mg/100ml)					**ART RG (mg/100ml)**				
0	96	131.61	96	125.44	0	96	123.14	96	117.13
3	96	143.46	96	136.66	3	96	134.21	96	127.08
6	96	147.31	96	138.09	6	96	133.28	96	131.41
Pre ART SGOT (I.U/L)					**ART SGOT (I.U/L)**				
0	96	9.08	96	8.87	0	96	9.21	96	9.10
3	96	8.24	96	8.84	3	96	8.51	96	8.67
6	96	8.76	96	9.48	6	96	9.52	96	9.44

Table 5.2 above shows the mean values of the variables at baseline to the sixth month of the nutrition intervention for all the groups (Pre-ART and ART) and subgroups (Test versus control) of participants. The sample size (n=400) represents the number of participants enrolled into the intervention research. However 384 participants completed the intervention programme for six months. 16 participants had incomplete data, hence incomplete data were not analysed. In the analysis of the results, emphasis was placed on MUAC as a tool to assess the nutritional status and CD4 cell counts as a tool to assess the immune status. The *t*-test analysis was used to compare the difference between two means (Test versus Control groups) in relation to the variation in the data.

Mid Upper Arm Circumference (MUAC)

Figure 5.4 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. The increase in mean MUAC in the scale–up intervention (n=400) is comparable to the increase in mean MUAC in the pilot intervention (n=100). As illustrated in Figure 4.1 (pilot result), mean MUAC for Pre-ART Test (Amtewa meal) group increased by 3.25%and ART Test (Amtewa meal) group, mean MUAC increased by 5.58% after six months. This is statistically significant (p=0.05).



	Ia	Ib	IIa	IIb
MUAC@ZERO MONTH	31.23	30.61	31.00	29.67
MUAC@3RD MONTH	31.35	30.55	31.10	29.42
MUAC@ 6TH MONTH	32.08	30.01	31.78	29.01

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

Figure 5. 4: The impact of Amtewa meal on Mean MUAC (cm) at zero, three and six months intervals of the scale-up intervention (n=400)

5.4.3 Biochemical indices

Regular biochemical assessments are good markers to assess the progression of HIV to AIDS. Potential deficit in micronutrients were also assessed by specific biochemical markers during routine blood tests as specified in the study design. These biochemical markers include the following as illustrated in the Table 5.2 and include the following:

Packed cell volume (PCV)

Table 5.2 shows the results of the PCV for the Pre-ART and ART groups. In both groups (Test and Control), the mean range in PCV from inclusion to the sixth month of the intervention was between 37. 54% to 43.18%. Reference values are 42-52% for males and 35-47% for females (Encyclopedia of Surgery). The hematocrit is usually about 3 times the hemoglobin value (assuming there is no marked hypochromia). The average error in hematocrit is about 1-2%. The hematocrit may be changed by altitude, position, and heavy smoking, in the same manner as the hemoglobin may be changed. The results obtained were within the normal range for all the groups (Test and Control).

Random plasma glucose (RPG)

From the results on Table 5.2, the mean random plasma glucose (RPG) in all the groups range between 117.13 to 147.31mg/100ml from inclusion to six months of the intervention. This shows that the participants RPG were within the normal range. The range in the mean RPG can also be explained by the fact that the RPG test were usually done in the morning between 7am to 8am local time to enable the participants on routine HIV clinic appointment see the physician with the results. Most individuals may not have taken their breakfast within this period. Hence what was considered RPG may be fasting plasma glucose in reality.

Total protein

The result in Table 5.2 shows that the mean Total protein range for all the groups was between 67.93 to 76.10g/dl from their inclusion to the sixth month of the intervention. The reference range for total protein is typically 60-85g/L (It is also sometimes reported as 6.0-8.5g/dl) (McAuley, 2012). Concentration below the reference range usually reflect low albumin concentration, for instance in liver disease, poor nutrition, malabsorption, diarrhea, or severe burns and sometimes immunodeficiency. Similarly, concentrations above the reference range are found in paraproteinaemia, Hodgkins's lymphoma, leukemia, liver disease, chronic infections, alcoholism, tuberculosis or any condition causing increase in immunoglobulins.

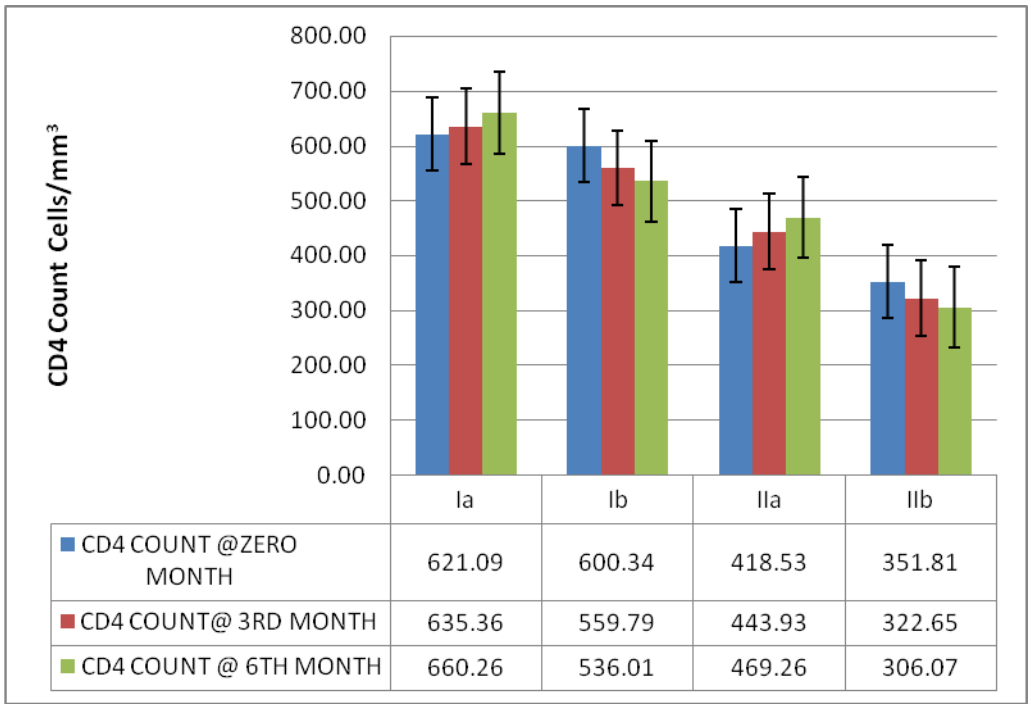
Serum glutamic oxaloacetic transaminase

The SGOT result in Table 5.2 shows results within normal range of SGOT (Normal range of SGOT is 5-35 IU/dL) in all the group of participants. This again could be attributed to the inclusion and exclusion criteria. PLWH with any HIV complication according to WHO clinical staging of HIV disease were excluded. The normal SGOT value also indicates that Amtewa meal may not be toxic to the liver.

CD4 Count

Figures 5.5 shows increase in mean CD4 count of the Test groups (Amtewa meal intervention groups) of participants in the scale–up intervention. In the Pre–ART group, the percentage increase in the mean CD4 count at three and six months were 2.3% and 6.31% respectively. Similarly, in the ART Test group percentage increase at three and six months were 6.07% and 12.12%. In the

control groups, mean CD4 count decreased by 64.33cells/mm^3 in the Pre–ART group and 45.74cells/mm^3 in the ART group. This result was a further confirmation of the result obtained in the pilot intervention.



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

Figure 5. 5: The impact of Amtewa meal on Mean CD4 counts (cells/mm3) at zero, three and six months intervals of the scale-up intervention (n=400)

Percentage increase in Mean MUAC (cm) and Mean CD4 counts (cells/mm^3) (Test groups)

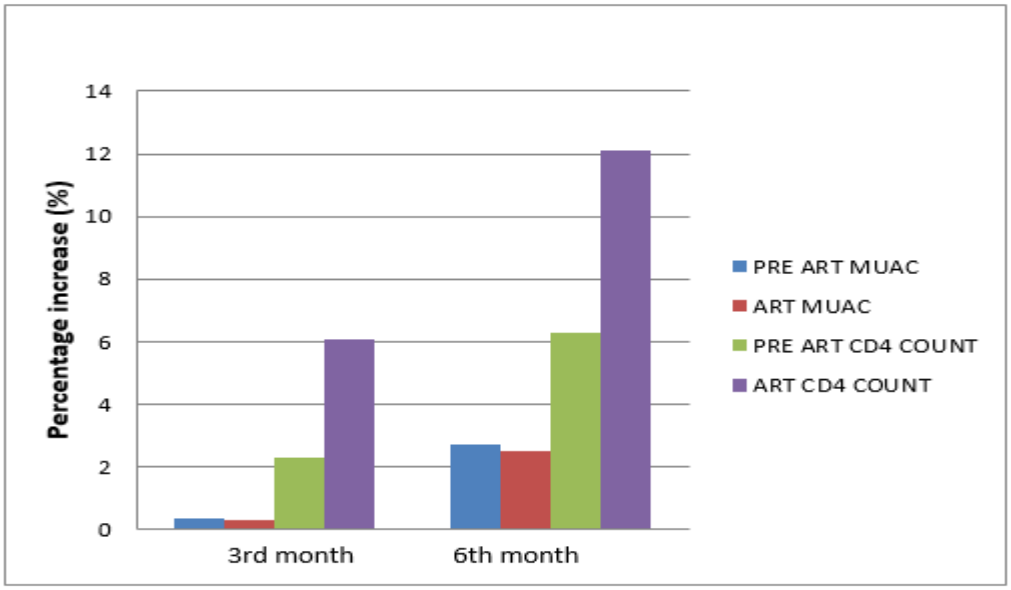


Figure 5. 6: Percentage increase in participants' CD4 Count and MUAC over six (6) months period (n=400)

Table 5. 3: Variance on percentage increase

Percentage Increase (%)				
Months	**Pre-ART MUAC**	**ART MUAC**	**Pre-ART CD4 count cells**	**ART CD4 count cells**
0 - 3	0.38	0.32	2.30	6.07
6	2.72	2.52	6.31	12.12

The graph (Figure 5.6) and Table 5.3 shows steady increases in the mean CD4 counts (Pre-ART and ART Test groups) in the third and sixth months respectively when compared to the mean CD4 count at baseline (0 month). Pre–ART Test group increased by 6.31% while the ART Test group increased by 12.12%. Mean MUAC increased by 2.72% and 2.52% for both groups (Pre–ART and ART Test groups) within the same period.

5.5 Qualitative assessment

Qualitative data were collected through direct encounter (interview) with the study participants. Specific interview questions were focused on participants' opinion, feelings and experience as illustrated in Table 5.4.

Table 5. 4: Qualitative assessment (interview) of study participants

	Finished their Amtewa meal before the next Hospital appointment date ®		**Difficult to prepare the meal**		**Shared intervention meal with others ©**		**Adverse reactions**	
	MALE	**FEMALE**	**MALE**	**FEMALE**	**MALE**	**FEMALE**	**MALE**	**FEMALE**
PRE ART TEST	7	3	0	0	2	6	0	0
ART TEST	5	4	0	0	4	4	0	1
TOTAL	12	7	0	0	6	10	0	1

Keys : ® = Not more than 2 appointments, © =Not more than 2 occasions

The Table 5.4 above shows 9.5% of participants (N=400, male =6%, female =3.5%) in the Test group at some point within the six month duration of the intervention but not more than two occasions, finished their meal before the next

hospital appointment date. The direction of use of the meal was quite explicit; hence there was no record of any participant who did not understand the method of preparation of the meal. The method of preparation is similar to the method of preparation of some local meals such as “Akamu” (a traditional oatmeal made from cereals) in Nigeria. Therefore the participants understood the concept.
Similarly, 8% of the participants (male=3%, female=5%) shared their intervention meal with others (spouses, participants in Control groups etc) probably due to the positive effect of the meal while 0.5% of the participants reacted (nausea and vomiting) to the meal and discontinued the intervention programme.

5.6 One year follow up from Pilot to Scale–up (12 months) intervention

Fifty (50) Test (Pre-ART and ART) participants out of the one hundred sample size in the pilot intervention (n=100) were followed up for another six months in the scale-up intervention to establish the impact of Amtewa meal on PLWH over a period of twelve (12) months. The results of the overlapped participants (n=100) illustrated in Tables 5.5 further confirmed the independent results obtained from the pilot and scale-up interventions. The major indicators assessed in the one year follow-up were the CD4 cell counts for immune status and the MUAC measurement for the nutritional status.

Table 5. 5: X2 T Test analysis: One year follow-up for study participants (pilot to scale-up n=100) anthropometric and biochemical indices

		Test				Test	
Time (months)		n	Mean	Time (months)		n	Mean
Pre ART Weight(Kg)				ART Weight(Kg)			
0		25	64.14	0		25	65.78
3		25	65.04	3		25	66.60
6		25	66.20	6		25	68.64
9		25	65.68	9		25	67.92
12		25	67.60	12		25	69.44
Pre ART Height(cm)				ART Height(cm)			
0		25	162.02	0		25	164.88
3		25	161.98	3		25	164.88
6		25	162.00	6		25	164.88
9		25	161.98	9		25	164.88
12		25	162.00	12		25	164.88
Pre ART MUAC(cm)				ART MUAC(cm)			
0		25	28.88	0		25	29.38
3		25	29.34	3		25	29.96
6		25	29.82	6		25	31.02
9		25	29.08	9		25	30.56
12		25	29.48	12		25	30.54
Pre ART PCV(%)				ART PCV(%)			
0		25	38.16	0		25	37.06
3		25	38.35	3		25	37.74
6		25	38.86	6		25	38.86
9		25	38.88	9		25	39.01
12		25	39.04	12		25	39.20
Pre ART CD4(cells/mm3)				ART CD4(cells/mm3)			
0		25	629.12	0		25	448.20
3		25	627.68	3		25	475.60
6		25	633.44	6		25	489.00
9		25	618.40	9		25	486.00
12		25	643.00	12		25	502.60
Pre ART TP(g/100ml)				ART TP(g/100ml)			
0		25	76.16	0		25	70.36
3		25	79.28	3		25	73.40
6		25	78.88	6		25	74.68
9		25	75.08	9		25	74.08
12		25	77.72	12		25	73.40
Pre ART RG(mg/100ml)				ART RG(mg/100ml)			
0		25	106.08	0		25	107.68
3		25	104.56	3		25	106.88
6		25	107.36	6		25	105.60
9		25	130.20	9		25	127.56
12		25	133.40	12		25	124.80
Pre ART SGOT(I.U/L)				ART SGOT(I.U/L)			
0		25	14.44	0		25	11.46
3		25	13.56	3		25	10.47
6		25	13.38	6		25	11.17
9		25	10.60	9		25	10.00
12		25	11.52	12		25	11.00

Table 5.5 shows a 12 month follow up on participants (n=100) that participated in the pilot intervention and overlapped into the scale-up intervention. The result above shows that the longer the duration of the intervention meal the more the improvement in the anthropometric and biochemical indices.

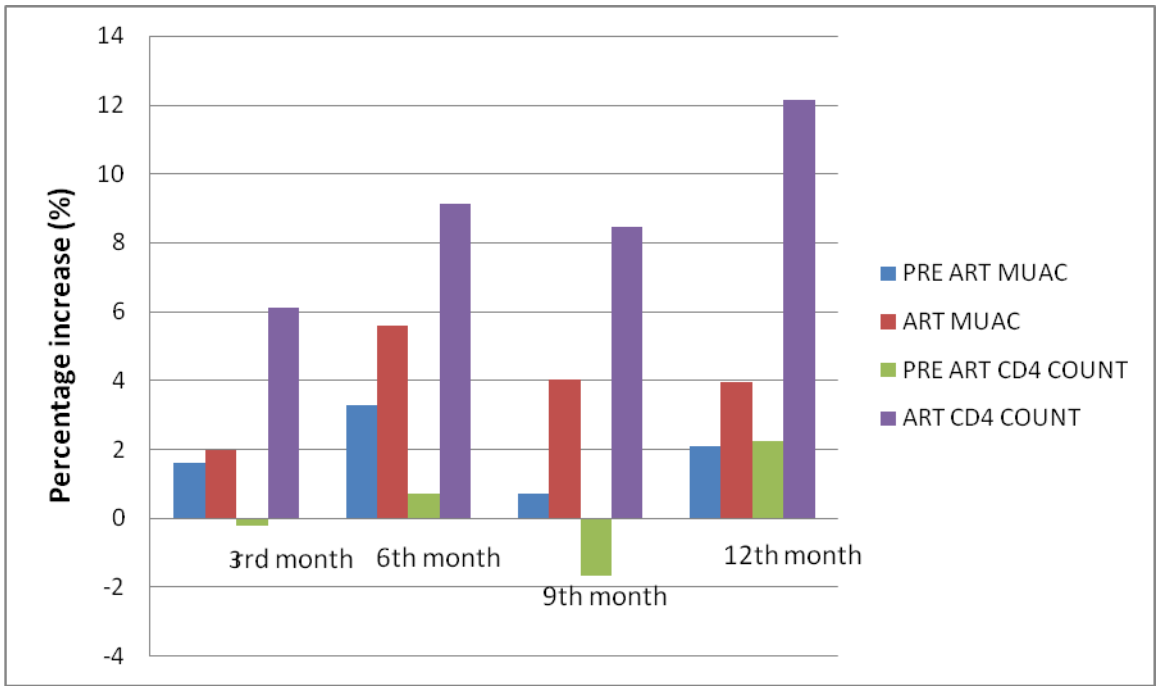


Figure 5. 7: Percentage increase in participants' CD4 Count and MUAC over twelve (12) months period (n=100)

Table 5. 6: Variance on percentage increase for twelve (12) months

Percentage (%) increase/decrease				
Months	Pre-ART MUAC	ART MUAC	Pre-ART CD4 count cells	ART CD4 count cells
0 - 3	1.59	1.97	-0.23	6.11
6	3.25	5.58	0.69	9.10
9	0.69	4.02	-1.70	8.43
12	2.08	3.95	2.21	12.14

Figure 5.7 and Table 5.6 shows the significant increases (p=0.05) in CD4 count and MUAC at twelve month duration from baseline to 12 months of the research. The increase in the mean CD4 count in the ART Test group was 54.40cells/mm^3 (over twelve-month period). Mean MUAC decreased from 29.82cm at the sixth month to 29.48cm at the 12 month. Pribram, (2011) classification of MUAC values (MUAC > 32cm, BMI > 30kg/m^2 =obesity; MUAC < 23.5cm, BMI < 20kg/m^2 =underweight) shows that these participants were neither obese nor underweight. However, the 0.24cm drop in the MUAC may be due to general information on healthy life style to all patients assessing treatment in State House Medical Centre, Abuja.

Chapter 6

Critical Analysis of Amtewa meal nutrition intervention

6.1 Author's Critical view on the different phases of the Intervention

In the planning phase of this research, vegetables and grains rich in carbohydrates, protein and mineral elements needed for normal body function, maintenance and reproduction were selected and analysed. In the analysis of these grains and vegetables, some contained high amount of carbohydrates, protein, vitamins and minerals while some had low amount of these nutrients. It was obvious from the results obtained in this investigation that intakes of these nutrients in different combination is essential for the maintenance of healthy life and normal body functioning of PLWH as recommended by WHO. In the article on comparative studies on the protein and mineral composition of some selected Nigerian vegetables, the authors Omale and Ugwu (2011) recommended further investigations to determine the effects of cooking and storage conditions on the valuable nutrients in the vegetables studied. The nutrients in Amtewa meal is neither affected by cooking nor storage conditions because the formulation processes took the necessary precautionary measures as explained in chapter 3 to ensure that the product was sterile and safe for human consumption.

Scientific advances have allowed researchers to better characterise the biological basis of disease states, understand the metabolism of food at the cellular level, and identify the role of bioactive components in food and assess their impact on metabolic processes. The tenet "Let food be thy medicine and medicine be thy food" espoused by Hippocrates nearly 2,500 years ago is receiving renewed interest under the term 'nutrition sensitive food sciences'. Avorn et al. (1994) published a randomised, double-blind, placebo-controlled clinical trial. The positive outcome of their publication measured a baseline and one month intervals data that investigated the effect of regular intake of cranberry juice beverage on bacteriuria and pyuria in elderly women. McClements and Decker (2009) advanced in research method to study food sensory perception, digestion and absorption. They reported that the breakdown of food structures in the gastrointestinal tract has a major impact on the sensory properties and nutritional quality of foods. Amuna et al. (2004) demonstrated the processes involved in the development of a suitable foodmultimix - FMM and how such products can provide sufficient nutrients in the daily composite diets of underprivileged, food-insecure impoverished communities as a short-term nutrient intervention. Amtewa meal nutrition intervention adapted the theoretical

approaches of these and other scientists, improved on their scopes by designing and implementing the intervention program in a Randomised Control Trial (i.e. putting theory into practice). The Amtewa-RCT was designed to compare study participants (allocated to treatment/intervention or control/placebo groups) using a random mechanism. Furthermore, Amtewa-RCT was best for studying the effect of an intervention, unbiased distribution of confounders, blinding more likely and randomisation facilitates statistical analysis. Cohort study, cross over, cross sectional or other forms of study designs were not used in Amtewa meal nutrition intervention to avoid wash-out period, unequal distribution of confounders and potential bias associated with these study designs.

Adherence to ART by PLWH is an essential component of programmatic treatment success. Adherence to ART is required to achieve adequate and sustained viral suppression and prevent the emergence of drug-resistant viral strains. Adherence rates exceeding 95% are necessary in order to maximize the benefits of ART. Even with adequate regimen adherence, there is a significant risk of ART-induced toxic effects and metabolic dysfunction. Thus, complete control of HIV over time using ART is unlikely and pharmacotherapeutic limitations leave a significant void in the treatment of HIV. Despite promising findings in the pilot study that Amtewa meal may improve nutritional status and immune functioning, definitive evidence of its impact in the scale–up intervention suggests this meal as an adjunct treatment in the care and support treatment of PLWH in Nigeria. The success of the Amtewa meal nutrition intervention depended on the education of research participants before the initiation of the meal, an assessment of their understanding of the intervention meal and their readiness to participate in the intervention programme.

6.2 Author's critical view on the other published intervention in relation to employed intervention.

The article, randomised trial of multivitamin supplements and HIV disease progression and mortality, the authors: Fawzi et al. (2004) proposed micronutrient supplements as low-cost immunomodulating interventions that may slow the progression of HIV disease. In this study, the progression of the disease was related to the WHO disease staging of HIV and the study participants' CD4 cell count was not a criterion for inclusion in the research. The authors' data suggests that multivitamins delay the onset of disease progression and thus the

time to the initiation of antiretroviral therapy may be delayed. This study provided information on the number of enrolled pregnant women of which by WHO recommendations all pregnant women must be on HAART to prevent mother to child transmission of HIV. Fawzi et al. (2004) study did not provide information on the group of participants who were Pre-ART hence their conclusion that multivitamin supplements are a means of delaying the initiation of antiretroviral therapy in HIV-infected women may not be justified.

In the Amtewa meal nutrition intervention, similar results to Fawzi et al. (2004) were obtained but the PLWHA with CD4 cell counts of >200cells/mm^3 were excluded from the research according to the inclusion and exclusion criteria of the research. Also the study design identified the Pre–ART group to justify the findings that Amtewa meal nutrition intervention was a means of delaying the initiation of ART. Hence, the originality of this reported data.

In Baum et al. (1995), deficiencies of vitamin A or vitamin B12 was associated with a decline in CD4 cell count (P =0.0255 and 0.0377, respectively), while normalization of vitamin A, vitamin B12 and zinc was associated with higher CD4 cell counts (P =0.0492, 0.0061 and 0.0112, respectively). Baum et al.'s results were based on vitamins A, vitamin B12 and zinc only. Amtewa meal contains varieties of macro and micronutrients with proven efficacy to improve nutritional status and immune function of PLWH in Nigeria.

6.3 Author's critical view on the employed meal (Amtewa) compared to other employed meal or nutritional fact on Nutrition in support of HIV in the existing body of knowledge

The World Health Organization (2005) reported HIV infection and undernutrition rates are raising to alarming levels in sub-Saharan Africa. There are complex interactions between nutrition and HIV/AIDS; lack of nutrition weakens the immune systems of people living with HIV/AIDS, increasing their susceptibility to other infections and worsening weight loss. To address these issues, the World Health Organization convened a Consultation on Nutrition and HIV/AIDS in Africa. The goal of the consultation was to:

- Develop strategies that were both evidence-based and feasible to help improve the health status of people living with HIV/AIDS in southern and eastern African countries.

- Review and disseminate the latest evidence on nutrition and HIV/AIDS, and thereby help ensure nutrition is integrated as part of a comprehensive response to HIV/AIDS.

Amtewa meal as a competent tool in nutrition intervention programme may be suggested as the latest evidence on the clinical nutrition - HIV interplay.

In the book, Nutrition and HIV, the author Pribram (2011) provided health professionals with a source of comprehensive information on the nutritional care and treatment of people living with HIV. This includes people of different age groups and varying states of health in a wide range of settings. The author gave excellent evidence to support other Nutritional experts who now see HIV patients with conditions as diverse as advanced wasting, obesity, cancers, renal failure, metabolic disorders, and many other illnesses. In support of Pribram and other related articles, nutrition has always been an important aspect of HIV care. In the early days of the pandemic, clinical nutrition was of necessity, experimental and required urgent responses to extreme and often fatal weight loss and wasting, associated with HIV disease progression. HIV has become a long-term chronic condition and the knowledge and skills required to manage this have changed rapidly and profoundly.

In the UNAIDS (2008) global report on HIV, regions of the world mostly affected by HIV infection simultaneously are affected by undernutrition. In Schaible and Kaufmann article on undernutrition and infection: complex mechanisms and global impacts, the author's reported that undernutrition is responsible, directly or indirectly, for 54% of the 10.8 million deaths per year in children under 5 and contributes to every second death (53%) associated with infectious diseases among children under 5 years of age in developing countries. While severe undernutrition resulting from acute, life threatening complications is still a major problem in sub-Saharan Africa, undernutrition in HIV may cause changes in immune function and have negative effect on each other. This provides a baseline setting on which the HIV pandemic has been imposed in many areas of sub-Saharan Africa. In regions with good access to ARVs, undernutrition and wasting associated with HIV disease are still highly prevalent and various forms of nutrition support are routinely required. Resource constraints often exist in these regions at household level where PLWH are often affected by unstable

immigration and poor socio-economic status like Nigeria. Nutritional care in HIV infection involves more than treatment of disease states. It has become increasingly apparent that optimum nutrition and healthy lifestyle interventions are essential to help enable people with HIV to lead long and healthy lives. Amtewa is not only accessible but also convenient and affordable by low socio-economic people in the Nigerian community.

Interestingly, in the article macronutrients and HIV/AIDS: a review, the authors; Hsu et al. (2005) reported a WHO working group recommended intake above 10% of expected energy for asymptomatic HIV infection. The authors also reported that in a recovery phase after opportunistic infections, nutritional requirements may be increased by 20 to 50% in both children and adults. Hsu JW.C et al. supports WHO (2003) and FANTA/USAID (2007) report on additional nutrient intake for asymptomatic and symptomatic PLWH. Although Ockenga et al. (2006) reported that the role of nutrition support in HIV infection has been poorly investigated, so is a paucity of evidence to demonstrate improved clinical outcomes through dietary interventions, however scientific evidence by WHO, UNAIDS, Mahlungulu et al. (2007), Fawzi et al. (2004), Fawzi et al. (2005) and other scientists suggests that dietary support may improve clinical outcomes in individuals with HIV infection by reducing the incidence of HIV–associated complications and attenuating progression of HIV disease, thereby improving quality of life and ultimately reducing disease–related mortality. The WHO report on additional nutrient intake for PLWH formed the baseline evidence for the calculation and formulation of Amtewa meal nutrition intervention.

Furthermore, in the article nutrients intake and health status of HIV/AIDS patients, the authors; Temitope et al. (2011) reported that nutrient intake is specific to a study population and depends on a variety of factors that are difficult to control. These factors include cultural, socioeconomic, environmental and geographical determinants. Therefore, the concept of Amtewa meal nutrition intervention was envisaged on a Nigeria population and the combination of macro and micronutrients carefully selected from locally available food in Abuja, Nigeria. The meal is a natural product that requires low technology in the formulation and with affordable training workshop to consumer could be sustainable.

Kupka and Fawzi (2002) reported low plasma zinc values in up to 26% of HIV-infected asymptomatic subjects without clinical evidence of nutritional

deficiencies; low plasma zinc was seen in up to 96% of AIDS/AIDS-related complex patients. Some investigators also reported a significant association between plasma zinc levels and immune parameters such as CD4 cell counts and HIV-RNA levels when data from HIV-positive subjects were evaluated cross-sectionally. Although Kupka and Fawzi's observational epidemiologic studies have provided conflicting results on the role of zinc status in HIV disease progression. However, they recommended that randomised, placebo-controlled trials are needed to resolve this controversy. Amtewa meal nutrition intervention is RCT to support other authors on the role of zinc in the progression of HIV. Zinc is a constituent of Amtewa meal (3.88mg in Amtewa meal) and the RNI for zinc is 9.5mg (male) and 7.0mg (female).

Kotler (1992), Van Standen et al. (1998), Piwoz (2004), FANTA (2004), WHO (2010) and other authors also supported the evidence that nutrient deficiencies associated with HIV are: total calories, proteins, vitamin A,B,C,E, selenium , iron, and zinc, however most of the nutrition intervention studies proposed the use of synthetic macro and micronutrients for example, micronutrient supplementation increases CD4 cell count in HIV–infected individual on HAART (Kaiser et al., 2006).

Contrary to Kaiser et al. (2006), Amtewa meal nutrition intervention supports Thiel (2000) article on natural vitamins that may be superior to synthetic ones. In this article, the author reported that vitamins originate primarily in plant tissues. In the *United States Pharmacopoeia* (USP) synthetic vitamin isolates are not naturally included in the diet, they do not necessarily originate primarily in plant tissues, nor have all of them been proven to safely and fully replace all natural vitamin activities. USP vitamins are not food, even though they are often called 'natural' and are sometimes added to foods. USP vitamins are synthesised, standardised chemical isolates. Also, in Thiel (2000) report, an animal study found synthetic vitamin A in the form of retinyl acetate significantly reduced vitamin E utilization. Another animal study found that natural food complex vitamin B6 was absorbed 2.54 times more into the blood and was retained 1.56 times more in the liver than an isolated USP form. Thiel's conclusion suggests that the bioavailability of natural food complex vitamins is better than most isolated USP vitamins. Amuna et al. (2004) took the view, whereas food fortification and food supplementation may be important alternatives that

complement food-based approaches to satisfy nutritional needs in poor communities in developing countries, the bioavailability of some synthetic supplements and fortificants may be limited. They are also expensive, may be unaffordable and may, thus, be less cost-effective as a long-term strategy. This formed the basis for the macro and micronutrients in Amtewa meal which originate primarily in plant tissues, is affordable and the bioavailability of this natural food complex (Amtewa meal) is better than the synthetic supplements.

Additionally, in the article on Food insecurity and HIV/AIDS: Current knowledge, gaps and research priorities; the authors, Anema et al. reported that food insecurity is associated with decreased ART adherence, reduced baseline CD4 cell count, incomplete virologic suppression, and decreased survival. Integration of food security interventions into HIV/AIDS treatment programs is essential to curtail the HIV/AIDS epidemic and improve health and quality of life among those infected. Hence, the selection of Amtewa meal constituents was based on:

1. Literature sources and sound scientific researches on Nigeria indigenous plant with known macro and micronutrient compositions.
2. Plant based food sources in Nigeria that are readily available and affordable to ensure sustainability of the intervention program.

Food security with Amtewa meal nutrition intervention is a strategy to build the capacity of households and communities affected by HIV and AIDS to cope with the disease. Many other services and intervention programs exist, which are relevant but not primarily designed to address nutrient deficiencies HIV/AIDS with natural food sources available in Nigerian community.

Ten (10) indigenous food samples from Nigeria were selected and analysed based on their macro and micronutrient compositions and other reasons mentioned above. However, only four (4) of the samples constitutes the composition of the Amtewa meal formulation. The justification for this selection was based on the macro and micronutrient strengths, stability of the formulation and the sensory evaluation. Although proximate analysis of some of the samples such as Corchorus walcottii (Ewedu leave) and Telfairaocci dentalis (Ugu leaves) show that these samples contain iron (Iron deficiency anaemia is the most widespread nutritional problem in the world and it is the main cause of anaemia) and antioxidants which are essential micronutrients to slow the progression of

HIV disease, but were excluded in the formulation to avoid stability and sensory evaluation challenges.

6.4 Author's critical view on Amtewa meal and HAART interactions

Food and drug interactions are an important issue for effectiveness and tolerability of HAART regimens. Thus, in the Amtewa meal nutrition intervention, HAART regimens with documented evidence of interaction with food were monitored and the study participants on such regimen were counsel to use their medication 2-3hrs before or after the intervention meal. The presence of Amtewa meal in the gastrointestinal tract can influence the absorption of several HIV medications such as didanosine, indinavir, saquinavir, and nelfinavir. Drug-food interactions can influence serum drug concentrations, thus increasing the likelihood of side effects of the ART when serum concentrations are too high and increasing the risk for viral resistance and loss of durable viral suppression when serum concentrations are too low.

In summary, the National Guidelines for HIV and AIDS treatment and Care in Adolescents and Adults, the Federal Ministry of Health, Nigeria recommended strategies for improving and monitoring nutritional status. These recommendations include:

- Close weight monitoring
- Nutrition education and Counselling
- Prompt treatment of opportunistic infections (mouth disorders, diarrhea)
- Nutritional Support (macro and micronutrient)
- Economic empowerment

Although the guidelines recommended nutritional support with macro and micronutrients but no evidence of any intervention meal in all the treatment centres in Abuja, Nigeria to ensure that the nutrient requirements for PLWH as recommended by WHO is achieved. Thus, Amtewa meal nutrition intervention if embedded into the guidelines on nutritional care and support for PLWH in Nigeria may suppress the progression of HIV and provide indirect improvement of CD4 count. The results support the use of Amtewa meal as a tailored functional recipe that is simple, inexpensive, and safe adjunct therapy in HIV disease management.

Chapter 7

Overall Discussion

7.1 Introduction to the Discussion of results

The findings from this Amtewa meal nutrition intervention confirm that the 75% of Nigerians infected with HIV who does not require ART, but nutritional assistance to maintain their immune system. This group of PLWH is guaranteed decrease in progression of their HIV status to AIDS. Similarly, the remaining 25% of HIV infected subjects in Nigeria who are currently on the HIV care and support programmes can be sustained by obviating their progression to AIDS. This discussion will be addressed under the following:

7.2 Sample size

According to Cochran (1997) our knowledge, our attitudes and our actions are based to a very large extent on samples. Sample surveys can be classified broadly into two types – descriptive and analytical (Cochran, 1997). This research was an analytical survey where comparisons were made between different sub-groups of a population in order to discover whether difference exist among sub-groups (Test group versus control) to verify the hypothesis (appendix 3.0). Larger sample size (n=400) compared to the sample size in the pilot intervention (n=100) generally led to increased precision (Bartlett et al., 2001). Also the sample size was based on the quality of the resulting estimates to have 95% confidence interval. In Abbie Griffin and John Hauser article, the importance of a quality sample report 20-30 in-depth interviews are necessary to uncover 90-95% of all customer needs for the product categories studied (Unite for Sight, 2013). This number is corroborated by a clinical research that identified sample size larger than 30 and less than 500 as appropriate for most research, (Unite for Sight, 2013). Also, Sandelowski (2007) reported that adequate sample size in qualitative research is ultimately a matter of judgment and experience in evaluating the quality of the information collected against the uses to which it will be put, the particular research method and purposive sampling strategy employed, and the research product intended. This research employed the RCT strategy for the calculated sample size of 400 study participants recruited for the study.

7.3 Marital status frequency distribution of study participants – Social dimension.

Having a main partner has been associated with better HIV outcomes in some studies, including higher HIV survival rates. This was evident in Amtewa meal nutrition intervention results (58.9% of the participants were married). Relationships characterised by mutual exchange, or reciprocity, of support are associated with positive appraisals of support, and with better mental and physical health outcomes (Knowlton et al., 2011). Also, Roach et al., (1981) reported marital satisfaction scale: Development of a measure for intervention research. They defined marital satisfaction as an attitude of greater or larger favorability towards one's own marital relationship. Couples on this intervention study encouraged one another to ensure adherence to the meal. In Knowlton et al. (2011) investigation, spouses or boy/girlfriends are often important sources of informal care and social support to the chronically ill, supports the outcome of this research. This study encouraged the monthly support group meetings which was an avenue for social interactions between PLWHA. Main partner support and care–giving may be especially important to the health of individuals vulnerable to family alienation and failed HAART.

7.4 Sex distribution of study participants in the scale-up intervention

The findings of this study suggest the merit of interventions targeting men and their informal caregivers, particularly main partners, and gender-specific, contextually tailored strategies to promote HAART adherence (Knowlton et al., 2011). The findings also complement findings from a prior study of HAART adherence among women in the study community, and suggest that gender differences in support exchange in this community affect their HAART adherence. This study means to encourage clinicians to plan specific, gender-focused support for enhancing adherence as supported by Ubbiali et al. (2008). The result suggests that more women were interested in HIV nutrition intervention than men in Abuja, Nigeria.

7.5 Age distribution of study participants in the scale-up intervention

In Rosenberg et al. (1994) age-specific data on the seroincidence of the human immunodeficiency virus (HIV) are difficult to obtain, however in this intervention research the age distribution results ascertained the response of specific age group to the intervention. In Detels et al. (1998) cohort studies that followed up

persons with human immunodeficiency virus (HIV) infection in periods characterised by different therapies, offered the opportunity to estimate therapy effectiveness at the population level. Similarly, in the calendar period when Amtewa meal was introduced with or without (Pre-ART and ART groups) antiretroviral therapy, the time to development of AIDS and time to death were extended and rate of CD4 cell count increase was more beneficial to the age group 31 – 50 years which constituted 65.9% of the study population. The SD of 9.721 indicates that over 97% of the values lie in the symmetric intervals as stated by Chebyshev's inequality from Weisstein (2014) that the minimum population Number of standard deviation from mean is six (6) for a 97% Confidence Interval.

7.6 Biochemical Indices of the study participants in the scale–up intervention

PCV results within normal range (Reference values: 42-52% for males and 35-47% for females) could be as a result of the exclusion criteria in the intervention. Although anaemia is more common and more severe with advanced HIV disease progression however, several longitudinal studies have reported either a significant increase in haemoglobin concentration or a significant decrease in clinical anaemia one year after HIV-positive persons began ART (Pribram, 2011). Participants monitored for one year did not show any clinical sign of anaemia. Pribram (2011) reported a multivariate analysis in which BMI, opportunistic infections and sex were adjusted for, mean heamoglobin concentrations increased significantly by 0.223g/L per month in HIV-positive person receiving ART. This may explain the significant increase in Pre-ART Test group from 39.81% to 43.18% within six months. Although some data reported that increased iron intake may be detrimental in HIV infection but conclusive evidence is lacking (Pribram, 2011). In Fawzi, et al. (2007) children born to mothers who received multivitamins had a 63% reduced risk of anemia (RR=0,37, 95% CI: 0,18, 0,79, p=0.01) compared to the placebo group. Therefore, multivitamin supplementation provided significant improvements in hemoglobin status among HIV-infected women and their children (Fawzi, et al., 2007). Amtewa meal nutrition intervention provided significant improvement in hemoglobin status of the study participants.

Furthermore, diagnostic criteria for diabetes are defined as: The presence of classical diabetes symptoms plus either a random venous plasma glucose concentration >11.1 mmol/L (>200g/dl) or a fasting venous plasma glucose concentration >7.0 mmol/L or >11.1 mmol/L as part of an oral glucose tolerance test (OGTT) (WHO, 1999; Pribram, 2011; NIH and NIDDK, 2012). Any test used to diagnose diabetes requires confirmation with a second measurement unless clear symptoms of diabetes exist (NIH and NIDDK, 2012). In this intervention, the random plasma glucose (RPG) test was used because it is sometimes used to diagnose diabetes during a regular health checkup (Pribram, 2011; NIH and NIDDK, 2012). If the RPG measures 200 micrograms per deciliter or above and the individual also shows symptoms of diabetes, then a health care provider may diagnose diabetes (NIH and NIDDK, 2012). Any test used to diagnose diabetes requires confirmation with a second measurement unless clear symptoms of diabetes exist. However, in the HIV population, the prevalence of diabetes is estimated at between 8 and 10% and results from decompensated insulin resistance (Pribram, 2011). Participants with history of diabetes mellitus were strongly advised to adhere to their medications. In this study, RPG was monitored and there was no need for further confirmation with a second measurement in all the participants.

Immune dysregulation in terms of hyper and hypo-gammaglobulinaemia was part of definition of immunodeficiency that characterised AIDS. This results in either raised level of total plasma/serum protein or low level of plasma/serum protein depending on which component of immune dysregulation predominates (Audu, et al., 2004). Sarro (2010) reported hypo-albuminemia and hyperproteinemia to be associated with a polyclonal ã-globulinemia in HIV seropositive patients as compared to HIV seronegative patients. Total protein test measures the total amount of two kinds of protein in the body: albumin and globulin. It was used as part of the investigations in this research. Study participants were recommended to repeat this test when experiencing unexpected weight loss, fatigue, or have symptoms of a kidney or liver disease but there was no need for such since their results was within the normal range. These symptoms are frequently experienced in HIV disease progression to AIDS.

Also, in HIV low serum albumin which is a component of TP remains a marker for mortality (Pribram, 2011). A serum albumin of level of less than 35g/l, along with

a history of prolong inadequate nutritional intake and other physical signs of undernutrition indicates severe undernutrition (Pribram, 2011).

The most widely used liver enzymes that are sensitive to abnormalities in liver and most commonly measured are the aminotransferases. The two aminotransferases are the alanine aminotransferase (ALT or SGPT) and aspartate aminotransferase (AST or SGOT). In this study, only the SGOT was checked. SGOT is normally present in a number of tissues such as heart, liver, muscle, brain and kidney. It is released into the blood stream whenever any of these tissues gets damaged. Normal range of SGOT is 5-35 IU/dL (McAuley, 2012). Results on SGOT obtained in this study shows that they were within normal range.

7.7 Multidimensional advantages of Amtewa meal nutrition intervention

The combination of Amtewa meal consumption in addition to the daily nutrient intake (DNI) has significantly improved the overall nutritional status of PLWH. This multi-levels improvement can be summarised as follows:

- Meeting the daily requirements of some essential minerals and vitamins (selenium, zinc, iron etc and vitamins - A, B, C, D, and E).
- Preserving or increasing fat-free mass
- Achieving and maintaining an ideal body weight
- Decreasing functional impairment from under-nutrition (muscular fatigue, bedridden state and work incapacity).
- Improving immune function.
- Improved quality of life

Additionally, Amtewa meal may also assist in limiting disease-specific complications, improving tolerance to antiretroviral treatment, provide symptomatic relief and alleviating gastrointestinal symptoms of HIV illness (nausea, diarrhea and bloating) and improving quality of life and survival of PLWH in Nigeria.

The mechanism of improved immune and nutritional status achieved by Amtewa meal nutrition intervention has qualified Amtewa meal to assist in multidimensional facet as illustrated in Figure 7.1 below:

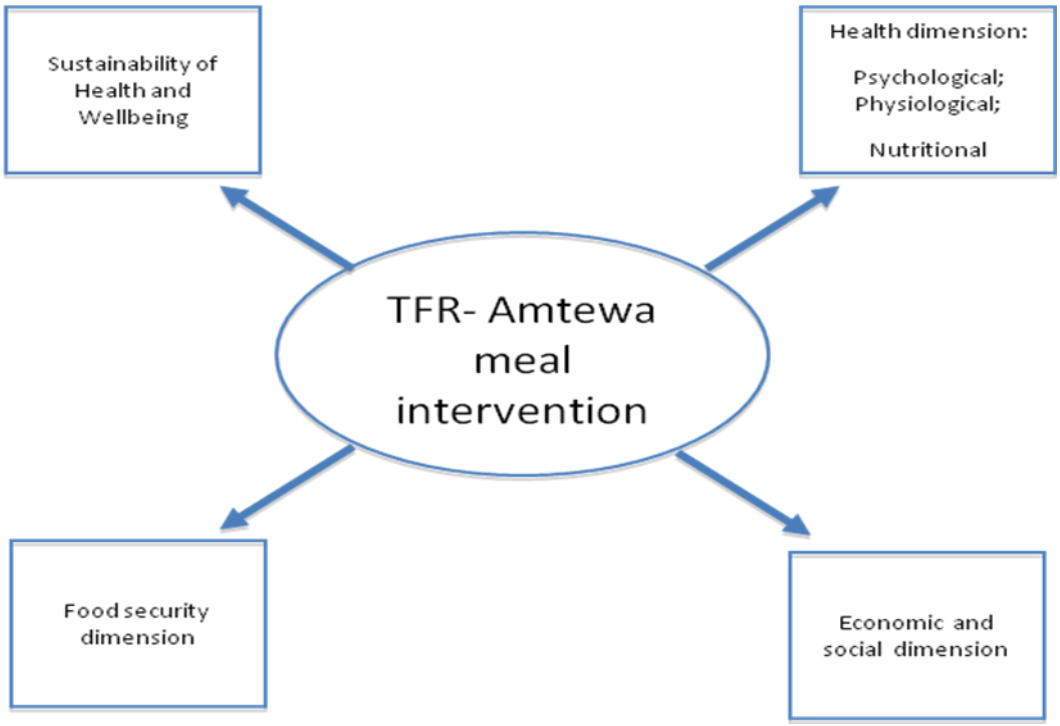


Figure 7. 1: The Multi-dimensional advantages of Tailored Functional Meal - AMETWA

7.7.1 Health Dimension

The Amtewa meal nutrition intervention increased CD4 cell counts and MUAC of PLWH. This supports other trials that have shown positive effect of vitamins B, C and E supplements on the immune status of HIV-infected persons and the relationships between micronutrients status and HIV disease progression among adults and children (Fawzi, 2003). The finding of this study was similar to Kupka et al. (2004) report on increased plasma selenium levels. It shows that selenium in Amtewa meal may be related to a decreased risk of mortality. Although plasma selenium levels was not associated with time to progression to CD4 cell count < 200 cells/mm, but were weakly and positively related to CD4 cell count in the one year of follow up. Selenium (present in moringa leaves) status is important for clinical outcomes related to HIV disease in sub-Saharan Africa (Kupka et al., 2004).

Also, Diplock et al. (1998) identified human diet to contain an array of different compounds that possess antioxidant activities or have been suggested to scavenge reactive oxygen species (ROS). 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 Amtewa meal may be responsible for scavenging this ROS.

According to Fridovich (1986) and Halliwell (1996), there are various sources of specific ROS in the human organism. However, the superoxide radical anion appears to play central role, since other reactive intermediates are formed in reaction sequences starting with O_2^-. 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 defence systems is operative in biological systems including enzymic and non-enzymic antioxidants (Diplock et al., 1998). This antioxidants (present in Amtewa meal) are substances when present in low concentration compared to that of an oxidizable substrate significantly delays or inhibits the oxidation of that substrate (Halliwell and Gutteridge, 1989).

Vitamin C (present in carrot, moringa and soyabeans) is the most powerful, least toxic natural antioxidant (Bendich and Langseth, 1995). It is a water-soluble vitamin and is found in high concentration in many tissues. Vitamin E a generic description for all tocols and tocotrienol derivatives exhibits the biological activity of alpha tocopherol (Diplock et al., 1998).The richest sources of vitamin E in the diet are vegetables oils such as soyabean, maize, cottonseed and safflower seed (Diplock et al., 1998).

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, spinach. Flavonoids are considered polyphenolic antioxidants that occur in several fruits, vegetables and beverages. Enzymes such as glutathione peroxidase and superoxide dismutase, which require a dietary supply selenium, copper and zinc respectively, contributed to the overall oxidative defence mechanism (Diplock et al., 1998) achieved by the intervention meal.

Local food substances in Nigeria (soyabean, millet, carrot and moringa) that contain vitamin C, vitamin E, carotenoids and flavonoids provided antioxidant effect in the formulation of Amtewa meal. Therefore, the first strategy to balance oxidative damage and antioxidant defence of human cells and tissues in PLWH was to enhance the antioxidant capacity by carefully selecting and optimising the dietary intake (macro and micronutrients) of antioxidants.

Furthermore, micronutrients (zinc, copper, selenium) and vitamins (A, C, E, B6 and B12) contained in Amtewa meal are essential for maintaining proper immunological function and they perform very important roles in the body's physiological and psychological (fear of death, anger, anxiety etc) processes and the regulation of energy metabolism (Williams, 1999; Drain et al., 2007; Pribram, 2011).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, advance HIV-related diseases. Also, faster disease progression or HIV-related mortality among HIV-positive persons not receiving ART was observed (Drain et al., 2007; Pribram, 2011).

Some morbidity characteristics, such as diarrhea, nausea or vomiting, lower respiratory tract infections, oral ulcers; thrush, severe anemia, and low CD4+ are related to a higher risk of wasting. Paton et al. (2006) reported moderate to severe undernutrition was present among 16% of patient at the time of starting ART, and was a significant predictor of death. Villamor et al. (2005) indentified vitamin B and vitamins C and E (contained in Amtewa meal) as essential vitamins that can reduce the risk of wasting. Jones et al. (2006) also reported increased serum zinc levels (as optimised in Amtewa meal) are associated with improved virologic control.

Interestingly, Austin et al. (2006) investigated a prospective double blind placebo controlled trial on the impact of carotenoids (which is a constituent of Amtewa meal) supplements on survival and health of AIDS patients. Their result shows mortality was increased in participants who did not receive carotenoids treatment compared to those who did. In the multivariate analysis survival was significantly and independently improved in those with higher baseline serum carotene concentration (p=.04) or higher baseline CD4 T lymphocyte counts (p=.005). It shows that low serum carotene concentration is common in AIDS patients and can predict death among advanced AIDS patients. The presence of carotenoids in Amtewa meal may account for the delay in the progression of HIV to AIDS in this study as justified by Austin et al. (2006). Supplementation as used in this intervention can correct micronutrient deficiency, improve survival and the study

participants were less likely to have progression to advanced stages of HIV disease with a better preservation of CD4+ (Fawzi et al., 2004; Austin et al., 2006).

In Kaiser et al. (2006) investigation on absolute CD4 count increase 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 micronutrients supply as proposed in the study improved CD4 cells count reconstitution in HIV-infected person taking HAART. 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 finding of Amtewa meal nutrition intervention was further supported by the following authors: Kruzich et al. (2004) reported that the effect of inadequate intake of vitamin A and E and zinc is related to impaired immune function and metabolic complications of the disease in the population. O'Brien et al. (2005) analysis using adjusted models also showed an association between anemia and an increased risk of all cause mortality and AIDS-related mortality, independent of CD4 cell count, WHO clinical stage, age, pregnancy, vitamin supplementation, and BMI. Papathakis et al. (2007) identified micronutrients deficiencies are associated with disease progression, reduced CD4 cell counts, and increased morbidity and mortality in HIV-positive persons.

According to Pribram (2011) 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 oedema than other anthropometric measurements. Bishop et al. (1981) 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. Although the MUAC results (Test versus control) in the scale-up intervention indicated that both groups are within the normal range as specified by Bishop et al. (1981) however there was a consistent decrease in MUAC in the control (Pre-ART and ART) groups.

Although Pribram (2011) identified time constraints and lack of practitioner training, along with inter and intra operator variation and inaccuracies that informed practice in paediatric units within the UK, to not routinely include this as an assessment tool, however this intervention research excluded paediatric participants, the research team were periodically trained and measurements monitored by a second observer to ensure accurate readings. The MUAC results provided a measure of acute nutrition status in adults (USAID and Measure Evaluating PRH) and this indicator is part of the linked set of 'Harmonised indicators for Nutrition and HIV Care' which track and provide comparative and trend data on the number and proportion of undernourished individuals receiving various program services (FANTA, 2009). The percentage increase in the pilot and scale–up interventions was used to tract the improvement in the nutritional status of the study participants. The validity of this indicator is also supported by USAID and Measure Evaluating PRH who states that this indicator can be used by donors and international organizations to track the extent to which nutrition interventions are improving client status and to identify countries or regions where more focused efforts may be required.

In support of our findings, Arpadi (2005) reported that lean body mass is lower in HIV-infected children than in HIV-negative control children when assessed by arm muscle circumference. Grinspoon et al. (2003) reported that HIV infection lead to wasting, particularly loss of metabolically active lean tissue, even in the era of highly active antiretroviral therapy. Similarly, Berhane et al. (1997) identified HIV-infected Ghanaian children with poor weight gain in infancy have a fivefold increase in the risk for death by age 2 years. Fontana et al. (1999) estimated that the relative risk of death was increased fivefold in HIV-infected children with the lowest quantities of fat-free mass. This implies that there is a strong association between poor growth and mortality in HIV which may further be investigated in future work with Amtewa meal. In Berhane et al. (1997) they proposed a possibility that poor nutritional status accelerates the progression from asymptomatic HIV infection to AIDS. Likewise, Centeville et al. (2005) reinforced the link between nutritional status and clinical progression of HIV/AIDS. Villamor et al. (2005) reported that multivitamins alone significantly reduced the risk of a first episode of a low mid-upper arm circumference. Therefore, Amtewa meal which is a natural source of macro and micronutrients

may give better outcome in future comparative studies with studies done with synthetic multivitamins.

From the results of 12 months follow-up (pilot to scale-up), the CD4 count in the Pre-ART Test group increased by 2.16% over a period of 12 months. Similarly, in the ART Test group the CD4 count increased by 10.8% over the same period. This increase is statistically significant (*p=0.05*). The result suggests that ART Test participants had 5 folds increase more than the Pre-ART Test group. Additionally, the result obtained in the 12 months follow up is comparable to pilot intervention that reported a 0.69% increase in the Pre-ART Test group and a 9% increase in the ART Test group. Although Paton et al. (2006) justified these results (pilot and scale-up intervention) however, Fawzi et al. (2005) caution the use of micronutrient supplementation as an alternative treatment, instead recommend micronutrient supplementation as a complementary intervention to antiretroviral therapy (ART). Therefore, Amtewa meal nutrition intervention is not a substitute to ART for PLWH who are eligible for HAART but complimentary in HIV care and support.

Similarly, the 12 months pilot to scale-up intervention results also shows the results of the MUAC increased by 2.04% in the Pre-ART Test group from inclusion to the study to 12 months. Also, in the ART Test group, MUAC increased by 3.8% within the same period. Tang (2003) reported body weight, fat free mass and body mass index improved in patient receiving nutritional intervention compared with the patient receiving placebo.

The Malnutrition Universal Screening Tool (MUST) developed by the British Association of Parenteral and Enteral Nutrition (BAPEN) and supported by many professional bodies, including the British Dietetic Association, Royal College of Physicians and National Institute for Health and Clinical Excellence (NICE) (Pribram, 2011) classified MUAC values using the following reference tool:

- If MUAC < 23.5cm, BMI < 20 kg/m^2 (underweight)
- If MUAC > 32cm, BMI > 30 kg/m^2 (obese)

From the above reference tool, the MUAC intervention results reveals that the participants monitored over a period of 12 and 6 months were not underweight but the controls consistently losing weight calls for an urgent intervention

(Amtewa meal intervention) because Tang et al. (2005) studies have shown that among HIV-positive persons 5% weight loss in 6 months is markedly associated with an increased risk of death.

7.7.2 Economic and Social dimensions

The traditional diets of most societies in developing countries (like Nigeria) are good (FAO, 2014). Scientific approach in the development of a functional meal like Amtewa will help to protect, support and help preserve the many excellent existing local food sources nutritionally valuable in the management of some disease conditions like HIV. Although synthetic multivitamins are present in the Nigerian market, however, functional food such as Amtewa meal constituents (soya beans, millet, carrot and moringa leaves) are readily available, accessible, affordable and more bio-available than the synthetic products. Synthetic multivitamins are very expensive food supplements which are not affordable by an average Nigerian, and if the country continues to import them, then foreign exchange is unnecessarily spent. The production of food comes mainly from agriculture. Nigeria has the human and material resources for the cultivation and production of the constituents of Amtewa meal. High-yielding varieties of the important cereals in Amtewa meal (soyabeans, millet) have been successfully developed, and much progress has been made in increasing food yields per hectare of land.

The suggested association on prolong use of Amtewa meal over 12 months period may slow down the progression of HIV to AIDS thereby improving the economic and social status of PLWH by:

- Improving the high manpower-intensive sectors of the economy that are most affected by HIV disease in Nigeria. This includes the agricultural, educational and health sectors as well as the rural economy.
- Decreasing the number of orphans and decreasing the cost of achieving set developmental goals by increasing the size of the workforce.
- Improving the quality of life of Nigerians thereby contributing positively to the economic development.
- Decreasing cumulative deaths which represent an incalculable loss of human potential associated with enduring trauma in households and communities.

- PLWHIV with a CD4 count above 200 cells/mm^3 can be continually productive in their community economy and sustain a little disrupted family life.

7.7.3 Food security

Household food security depends on a nutritionally adequate and safe food supply in Nigeria, at the household level and for each individual; a fair degree of stability in the food availability to the household both during the year and from year to year; and access of each family member to sufficient food to meet nutritional requirements (FAO, 2014). This last criterion includes not only physical access but also economic and social access to foods that are culturally acceptable (Amtewa meal). Incomes received from cash crops or wage earnings and prices paid for purchased items influence a rural population's food security (FAO, 2014). However, IITA (2004) reported that average household expenditure on non-staples in Nigeria was highest on fish (N140.84 approximately $1) followed by meat products (N81.54 approximately half a dollar). Here, the least weekly expenditure was on fruit (N13.62), followed by weekly expenditure on the leafy vegetables (N20.88). This suggests that most Nigerians have vegetable gardens in their neighborhood were these vegetables such as moringa leaves are easily accessed and may not need to purchase from the market. Food security can be threatened by increased prices, job loss, income reduction, rent increases, larger numbers of dependent persons (more children, or relatives moving into the household) and other factors (FAO, 2014; Ivers, 2009). Nonetheless, IITA (2004) report on frequently consumed staple and non-staple foods in Nigeria suggests that the constituents of Amtewa meal (soyabeans, millet, carrot and moringa leaves) are available and affordable.

The concept of food security in this intervention research is to ensure both physical and economic access to food that meets dietary needs (in the form of a functional meal - Amtewa) of PLWH. Wang et al. (2011) reported that food insecurity associated with unsuppressed viral load and may render treatment less effective among HIV-infected participants receiving antiretroviral medications.

Similarly, Palar (2012) reported improvements in work and mental health status were identified as potential pathways through which ART may improve food

security. Therefore, Amtewa meal nutrition intervention may guarantee food security due to the fact that the intervention meal meets the three pillars on which food security is built. These are:

- Amtewa meal constituents are readily available: sufficient quantities of soyabeans, moringa leaves, carrot and millet are available on a consistent basis (IITA, 2004 report on Nigeria Food Survey).
- Amtewa meal constituents are accessible: having sufficient resources to obtain /afford the constituents for a nutrition intervention (FAO, 2014; IITA, 2004).
- Amtewa meal formulation requires low technology: appropriate use based on knowledge of basic HIV nutrition and care, as well as adequate training on TFR concept (Amuna et al., 2004).

7.7.4 Sustainability and health promotion

The relationship between undernutrition and infection has been extensively studied and documented. There is no doubt that common infections such as diarrhoea, respiratory disease, intestinal worms, measles and HIV/AIDS are important causes of undernutrition (FAO, 2014). However, the need to sustain the gain of Amtewa meal nutrition intervention in slowing down the progression of HIV to AIDS is imperative.

Planning for sustainability in this public health nutrition intervention required a clear understanding of the concept of sustainability and operational indicators that may be used in monitoring sustainability over time within State House Medical Centre, Abuja and the community. In addition, it also required the use of programmatic approaches and strategies that favored long-term program maintenance in a resource limited setting. The current HIV clinic structure in SHMCA, identifies some key personnel (Health professionals), management staff of SHMCA, the monthly support group meeting of the PLWH and the HIV programme implementing partners. Sustainability will be assured if these groups of people are incorporated into the intervention programme. Broad-based support from this cross-section of service providers is necessary to overcome reasons related to limited financial resources, territoriality concern, local politics, limited time and lack of hospital management interest. Quality control procedures and HIV treatment protocols implemented in HIV care and support programmes will

incorporate Amtewa meal nutrition intervention. However, the sustainability and health promotion approach focused primarily on the following:

- Three training-workshops to empower (capacity building) the PLWH community: Topics covered in these workshops were: Basic health education on HIV/AIDS; Nutrition education on the macro/micronutrient contents of local fruit and vegetables; formulation/preparation of Amtewa meal at home standardised but adaptable approaches in the design of the intervention.
- maintenance of research outcome (pilot and scale-up) health benefits achieved through the intervention program.
- Presentation of research outcome at the Federal Ministry of Health Nigeria, National Agency for the Control of AIDS Nigeria, and publication in international journals.
- Level of institutionalization of a program within the health settings by incorporating Amtewa meal nutrition intervention in the HIV care program as a policy framework.
- Replication of Amtewa meal nutrition intervention in other HIV treatment centre in collaboration with the National Agency for the Control of AIDS (NACA) in Nigeria.

In summary, the multi-faceted dimensional benefits of amtewa meal nutritional intervention is justifiable and should be an integral part of HIV care and support services in Nigeria.

Chapter 8

Limitations, Conclusion and Future work

8.1 Limitations to the study

This presented research is an implementation of previous research papers on the subject with a view of developing a culturally acceptable, affordable and accessible nutrition intervention meal (Amtewa) to slow the progression of HIV to AID amongst PLWHIV in Nigeria. However, some limitations include:

- The shortage of trained research active/oriented staff was challenging in the health setting particularly nutrition and dietician trained staff.
- Risk of sample contamination (Un-intentional collusion between the control and Test group while on HIV care hospital appointments).
- The undivided attention of the Principal investigator (researcher) to manage a large sample size (logistics and funding) in the scale-up intervention.
- Financial support: Lack of funds stalled the inclusion of viral load test as an assessment tool in the study design. Viral load test is very expensive.
- Time period: The time period from the points of participants' assessment, data collection, blood sample collection and collection of the Amtewa meal was a challenge to some participants who were in a hurry to report in their respective work places.
- Large population (sample size): Recruiting and monitoring participants in the ART group was not challenging compared to the Pre–ART group. Participants in the Pre–ART group are not on HAART according to WHO recommendations; hence they are not mandated to the monthly or bimonthly hospital appointment prescribed for ART group of participants. The Pre–ART irregular hospital appointment was a challenge in tracking this group of participants.

8.2 Conclusions

The success of recent nutrition interventions in HIV demonstrate the progress made in HIV care and support. The idea to adapt principles and technologies from the TFR concept to the development of a functional recipe to slow the progression of HIV to AIDS proves to be meaningful and realisable in Amtewa meal nutrition intervention approach.

Although the achieved results take the form of specific technology, it suggests that a prolong consumption of the intervention meal (Amtewa) will be suitable to sustain the gained improvements in the anthropometric and biochemical indices. The research highlighted crucial issues and identified key design parameters that require further attention and research in developing countries like Nigerian context.

Overall, it underpins the synergistic relationship between nutrition and HIV infection, the nutritional requirement and nutritional care and support for PLWH in Nigeria. While the nutrition intervention demonstrated a positive effect, the study also suggests that the initial visit of a newly diagnosed HIV-positive patient should include screening for nutritional status, identify risks and offer appropriate nutrition Counselling which was not in existence but currently has been imbedded as "Nutritional Framework" within the HIV care and support programme in SHMCA, Nigeria. This drafted Nutritional Framework has provided information to PLWH on their HIV medication and food interactions and about nutritional screening tools available in the health setting.

Finally, the research provides evidence which may be used as a basis for policy makers to incorporate Amtewa meal nutrition intervention in HIV care and support programme in other HIV treatment centres in Nigeria with a view of attenuating the progression of HIV to AIDS amongst PLWH in Nigeria.

8.3 Future work

The conducted intervention identified areas for further research that may significantly support the government initiative to combat HIV in Nigeria. Future researches are necessary and must expand upon past programmes and this presented intervention. The following intervention programmes warrant clinical investigation. These can be summarised as:

- Amtewa meal nutrition intervention approach in the management of adult patients with AIDS (CD4 <200cells/mm^3).
- Amtewa meal nutrition intervention approach in the management of paediatric patients living with HIV.
- Amtewa meal nutrition intervention approach in management of pregnant women living with HIV.

Chapter 9

References and Appendices

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Appendices

Appendix 3.0

Research Hypothesis

Daily intake of optimised Amtewa meal which contains immune-boosting macro and micronutrients from indigenous food sources in Nigeria delays progression of HIV to AIDS by ameliorating and sustaining the nutritional status (*BMI, MUAC*) as well as improving the biochemical indices (*CD4 count, PCV, RG, SGOT, and TP*) in People Living with HIV (with a CD4 count above 200cells/mm^3).

Appendix 3.1

STATE HOUSE MEDICAL CENTRE,
ABUJA,
NIGERIA.
TEL: 09-6252594/5

Reference: SHMC/REC/0001/2009 1st November, 2010

Dear Dr Abraham Amlogu,
Department of Pharmacy
State House Medical Centre, Abuja, Nigeria.

SHMC Ethics Reference No: *Please quote this ref on all correspondence*	**SHMC/REC/0001/2009**
Project Title:	Assessing the effectiveness of public health-nutrition intervention programme to attenuate the progression of human immuno-deficiency virus (HIV) to AIDS (*acquired immuno-deficiency disease syndrome*) among people living with HIV in Abuja, Nigeria'
Researchers Name(s):	**DR ABRAHAM AMLOGU**

Thank you for submitting your application and request for ethical clearance for the above-named study which was considered at the SHMC Research and Ethics Committee. The following documents were reviewed:

1. Request for ethical clearance
2. Research Plan
3. Informed Consent Form
4. Participant Information Sheet (PIS)

The State House Medical Centre Research and Ethics Committee (SHMC REC) hereby approves this study from an ethical point of view.

Approval is hereby given till 1st November, 2011 renewable yearly for three years. However, if the research has not commenced/finished before the expiration date, a request for an extension must be re-submitted to SHMC REC.

You must also inform the SHMC REC when the research has been completed. If you are unable to complete your research within the stipulated validation period, you will be required to write to SHMC REC.

Any serious adverse events or significant change which occurs in connection with this study and/or which may alter its ethical consideration must be reported immediately to the SHMC REC, for an appropriate Ethical Amendment.

Approval is given on the understanding that the **'NATIONAL CODE OF HEALTH RESEARCH ETHICS'** are adhered to.

Yours sincerely,

STATE HOUSE MEDICAL CENTRE
State House, Abuja.

1/11/10

DR M.N AJANI, MBBS, FWACS
Chairperson
SHMC Research and Ethics Committee

STATE HOUSE,
ABUJA,
NIGERIA.

29th September, 2009

Reference:

Tajalli Keshavarz
Professor of Biotechnology,
Research Director,
School of Biosciences, University of Westminster,
115 New Cavendish Street, London W1W 6UW

RESEARCH PROPOSAL
RE: POTENTIAL IMPACT OF MACRO- AND MICRO NUTRIENTS ON PEOPLE LIVING WITH HIV IN ABUJA, NIGERIA

The above subject matter refers.
At Dr Abraham Amlogu's request, we are writing this letter to you.
We write to confirm that the State House Medical Centre (SHMC), a secondary medical facility, has the requisite capacity for CD4 lymphocyte count, viral load analysis, and other laboratory investigations needed in the above-named research.
Furthermore, we would like to give assurance that SHMC would provide all the necessary facilities and assistance that the researcher would require for the whole duration of the period of study in this centre.
In the event that you require any further information, please do not hesitate to contact the undersigned.

Thank you.

Dr. M. N. AJANI (MBBS, FWACS)
Chairman, Medical Advisory Committee
Chairperson, State House Medical Centre, Research and Ethics Committee (SHMC, REC)
Tel: +234 9 6252597 (Office)

Appendix 3.2

UNIVERSITY OF
FORWARD
THINKING
WESTMINSTER

05 August 2011

Dear Abraham

App. No. 10/11/18
Abraham Amlogu: School of Life Sciences
Mode: MPhil/PhD
Supervisor: Ihab Tewfik

Assessing the effectiveness of public health-nutrition intervention programme to attenuate the progression of human immune-deficiency virus (HIV) to AIDS (*acquired immuno-deficiency disease syndrome*) among People Living With HIV (PLWH) in Abuja, Nigeria

I am writing to inform you that your application (condition set to you) was considered on 05 August 2011. The proposal was **approved**.

If your protocol changes significantly in the meantime, please contact me immediately, in case of further ethical requirements.

Yours sincerely

Huzma Kelly
Senior Research Officer (Policy and Governance)
Secretary, Research Ethics sub Committee

cc Dr. John Colwell, (Chair) Research Ethics sub Committee
Dr. Ihab Tewfik
Dr. Keith Redway
Mike Fisher

I am advised by the Committee to remind you of the following points:

1. Your responsibility to notify the Research Ethics sub Committee immediately of any information received by you, or of which you become aware, which would cast doubt upon, or alter, any information contained in the original application, or a later amendment, submitted to the Research Ethics sub Committee and/or which would raise questions about the safety and/or continued conduct of the research.
2. The need to comply with the Data Protection Act 1998
3. The need to comply, throughout the conduct of the study, with good research practice standards
4. The need to refer proposed amendments to the protocol to the Research Ethics sub Committee for further review and to obtain Research Ethics sub Committee approval thereto prior to implementation (except only in cases of emergency when the welfare of the subject is paramount).
5. You are authorised to present this University of Westminster Ethics Committee letter of approval to outside bodies, e.g. NHS Research Ethics Committees, in support of any application for further research clearance.
6. The requirement to furnish the Research Ethics sub Committee with details of the conclusion and outcome of the project, and to inform the Research Ethics sub Committee should the research be discontinued. The Committee would prefer a concise summary of the conclusion and outcome of the project, which would fit no more than one side of A4 paper, please.
7. The desirability of including full details of the consent form in an appendix to your research, and of addressing specifically ethical issues in your methodological discussion.

Appendix 3.3

STATE HOUSE MEDICAL CENTRE, ABUJA RESEARCH AND ETHICS COMMITTEE(REC)

Informed Consent Document And Participant information sheet

TITLE OF RESEARCH: 'Assessing the effectiveness of public health-nutrition intervention programme to attenuate the progression of human immuno-deficiency virus (HIV) to AIDS (*acquired immuno-deficiency disease syndrome*) among People Living with HIV (PLWH) in Abuja, Nigeria'

NAME(S) AND AFFILIATIONS OF RESEARCHER(S): UNIVERSITY OF WESTMINSTER

PRINCIPAL INVESTIGATOR(S): AMLOGU MAINAJI ABRAHAM

SPONSOR(S) OF RESEARCH: AMLOGU MAINAJI ABRAHAM

PURPOSE(S) OF RESEARCH: INTERVENTION ON PATIENT MANAGEMENT

PROCEDURE OF THE RESEARCH:

Eligible People Living with HIV (PLWH) will be recruited, and given the right to decline participation without jeopardizing receipt of care at the State House Medical Centre, Abuja. Prior intervention, 'consent form' will be signed and 'project information' sheet will be given and explained to all participants.

Once enrolled, participants will be subjected to a research assessment tool, which include some variables; *demographic information, specific questions on lifestyle, health habits and nutritional practices.*
Furthermore, participants shall be grouped into four groups according to the study design. Nutrition Counselling, Highly Active Antiretroviral Therapy *(provided as standard mode of treatment for PLWH by the State House Medical Centre),* vitamin supplements, specific micro and macro-nutrient combination will be administered to the groups' accordingly.
At the onset of the research, one hundred (100) participants will be recruited for the pilot study. Thereafter, a larger scale intervention that will recruit four hundred (400) participants will follow to determine the impact of the research on a larger population.

Anthropometric measurements*(e.g. body mass index, skin fold measurement, waist circumference),* clinical assessments *(e.g. review of dietary history, blood pressure measurement)* and laboratory investigations *(e.g. blood glucose level, lipid profile,CD4 count)* will be conducted at the commencement of the study, third and sixth months of the pilot and larger scale studies respectively.

All collected questionnaire will be revised for completeness and data will be analysed.

Risks and Discomforts

Conventionally, HIV clients on management visit the hospital every three months for laboratory investigations such as CD4 count and viral load tests. Naturally, the discomfort of the needle prick prior to sample collection may psychologically affect some participants that have phobia for injections.

Furthermore, participants will be subjected to a research assessment tool and anthropometric measurements. These may constitute some stress to the participants.

Administration of micro and macro-nutrient combination to participants for daily consumption is expected to have no side effects due to the fact that the quantities to be consumed will be within the limit for recommended intake for adult; however these products may constitute an additional stress as a result of adherence to the prescription on the product.

Measures to reduce risks and Discomforts

Participants will be reassured of lesser needle prick pain.
To discuss with the trained laboratory scientists responsible for the sample collection on an improve technique (e.g. immersing participants fingers in warm water before sample collection) that will not inflict much pain on study participants.

3. Educate the participants on the benefits of adhering to the recommended micro and macro-nutrient combination on improving their quality of life
4. Trained interviewers will be assisting in filling the research assessment tool under the supervision of the researcher.

Legal Rights

The research participant is not waiving any of his/her legal rights by signing this informed consent document.
Please initial your choice(s) below:
___ I agree to allow my samples to be kept and used for future research
___ I do not agree to allow my samples to be kept and used for future research
___ I wish to be notified if my samples are going to be used for future research.

Signatures
Your signature below indicates that you agree to participate in this study. You will receive a copy of this signed document.

Signature of Participant Date

Signature of investigator or other person obtaining consent Date

Signature of Witness Date

The consent process must include a witness unless the PI requests and justifies, and the SHMC REC approves a waiver of the requirement.
The person administering the consent (e.g., study coordinator) cannot sign as the witness.

Signature of Investigator reviewing consent document Date

STATE HOUSE MEDICAL CENTRE, ABUJA
RESEARCH AND ETHICS COMMITTEE (REC)

AUTHORIZATION FOR USE/DISCLOSURE OF HEALTH INFORMATION AND PARTICIPANT INFORMATION SHEET FOR RESEARCH

What is the purpose of this form? You are being asked to sign this form so that SHMC may use and release your health information for research**.** Participation in research is voluntary. If you choose to participate in the research, you must sign this form so that your health information may be used for the research.

Participant Name: SHMC REC Protocol Number:

Research Protocol: Principal Investigator:

Sponsor:

What health information do the researchers want to use? All medical information and personal identifiers include past, present, and future history, examinations, laboratory results, imaging studies and reports and treatments of whatever kind related to or collected for use in the research protocol.

Why do the researchers want my health information? The researchers want to use your health information as part of the research protocol listed above and described to you in the Informed Consent document.

Who will disclose, use and/or receive my health information? The physicians, pharmacists, nurses, laboratory scientists and staff working on the research protocol (whether at SHMC, University of Westminster, London or elsewhere).

How will my health information be protected once it is given to others? Your health information that is given to the researcher will coded and remain private to the extent possible, even though the researcher is not required to follow the federal privacy laws. However, once your information is given to other organizations that are not required to follow federal privacy laws, we cannot assure that the information will remain protected.

How long will this Authorization last? Your authorization for the uses and disclosures described in this Authorization does not have an expiration date.
Can I cancel the Authorization? You may cancel this Authorization at any time by notifying the Principal investigator in writing, referencing your Research Protocol Number. If you cancel this Authorization, the researcher will not use any new health information for the research. However, researchers may continue to use the health information that was provided before you cancelled your authorization.
Can I see my health information? You have a right to request to see your health information. However, to ensure the scientific integrity of the research, you will not be able to review the research information until after the research protocol has been completed.

Signature of participant
Date:
Or participant's legally authorised representative: Date:
Printed Name of participant's representative:
Relationship to the participant:

Appendix 3.4

Determining Moisture Content in Food Samples (Pomeranz and Meloan, 1994)

Oven Drying Methods

Materials: Food samples, analytical balance, Mortar and pestles, Petri dishes, Blender, Spatula, Oven

Method: With oven drying, a known weight of each sample was heated at 78^0C for 60hrs.The loss of weight was used to calculate the moisture content of the samples.

Moisture Content Calculations: For oven-drying methodologies

$$\% \text{ Moisture (wt/wt)} = \frac{\text{(wt of wet sample - wt of dry sample)}}{\text{wt of wet sample}} \times 100$$

Appendix 3.5

Fat (Soxhlet extraction of Fat from Food Samples) (Pomeranz and Meloan, 1994)

Purpose

This experiment involves the extraction of fat from food samples in Nigeria by an exhaustive extraction with solvent using a Soxhlet extractor apparatus as shown below

Materials

• Organic solvent
• Sodium sulfate
• Thimble (cellulose or slitted glass)
• Food samples from Nigeria
• Teflon® boiling chips or glass beads
• Glass wool
• Nitrogen gas
• Mortar and pestle
• Micro-Soxhlet glassware with 25-mL round-bottomed flask
• Heating mantle
• Concentrator (evaporation) apparatus
• Rubber tubing
• Analytical balance

Handling and Disposal

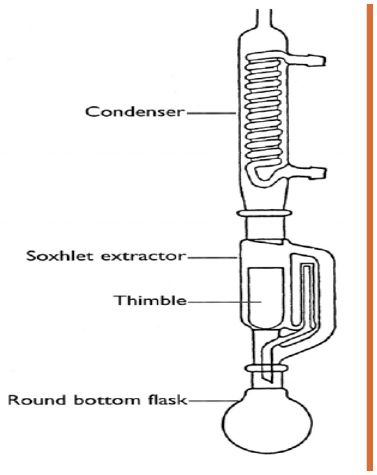
Specifically the University of Westminster Standard Operating Procedures (SOPs) and guidelines on safe handling and storage of all chemicals and equipment were strictly followed in these activities. This includes determining and using the appropriate personal protective equipment (e.g., goggles, gloves, and apron).

This experiment uses hazardous solvents that were disposed of in appropriate waste containers. Organic solvents tend to be highly flammable. Appropriate precautions (Extraction and post extraction evaporation steps)were performed in a vented hood.

Method

Glasswares were cleaned with petroleum spirit, drained, and dried in an oven at 102 º C for 30 minutes and cooled in desiccators. A piece of cotton wool was placed in the bottom of a 100ml beaker and also a plug of cotton wool was placed in the bottom of an extraction thimble. The thimble was made to stand in the beaker.

5 g of sample was accurately weighed into the thimble. 1 - 1.5 g of sand was added and mixed with the sample with a glass rod. Dried sample was allowed to cool in desiccators. The thimble was inserted into in a Soxhlet liquid/solid extractor as shown below.



Soxhlet extraction of fat

A clean dried 150ml round bottom flask was accurately weighed and 90ml of petroleum ether was poured into the flask. The extraction unit was assembled over either an electric heating a water bath. The solvent in the flask was heated until it boils. The heat source was adjusted at intervals to avoid solvent drips from the condenser into the sample chamber at the rate of about 6 drops per second. The extraction process continued for 6 hours. After 6hrsthe heat source was removed the solvent drained from the extractor in the flask. The thimble was removed from the extractor and the sample transferred to a 100ml beaker. With the aid of a glass rod, the sample was broken, returned to the thimble and replaced in the extractor. The beaker with the petroleum ether was rinsed and poured into the extract. Extraction continued for a further two hours. After 2hrs of further extraction, the heat source was removed and the extraction unit and condenser were detached.

The flask was replaced on the heat source to evaporate off the solvent. (The solvent may be distilled and recovered). The flask was placed in an oven at 102ºC to dry the contents until a constant weight was reached (1-2 hours). The flask with the content was cooled in desiccators and weighed.

Weight of empty flask (g) = W1

Weight of flask and extracted fat (g) = W2

Weight of sample = S

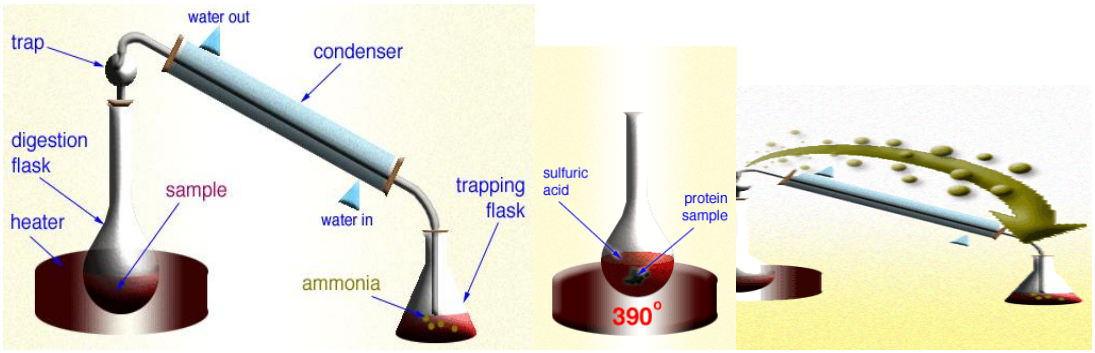
% Crude fat = (W2 – W1) x 100/S

Appendix 3.6

Protein (Kjeldahl extraction) (Blamire, 2003)

The Kjeldahl method consists of three steps, which were carefully carried out in sequence:

1. The sample was first digested in strong sulfuric acid in the presence of a catalyst, which helps in the conversion of the amine nitrogen to ammonium ions,
2. The ammonium ions were then converted into ammonia gas, heated and distilled. The ammonia gas was led into a trapping solution where it dissolved and became an ammonium ion once again,
3. Finally, the amount of the ammonia that was been trapped was determined by titration with a standard solution, and a calculation made.



Step 1: Digestion

Approximately 1gm of sample containing protein was weighed, placed into a digestion flask, containing 12-15 ml of concentrated sulfuric acid (H_2SO_4). Seven grams of potassium sulfate and a copper catalyst were added. The mixtures in the flask were heated (about 370^oC to 400^oC) using a heating a block until white fumes were seen, and then the heating continued for about 60-90 minutes. The flask was cooled cautiously by adding 250ml of water.

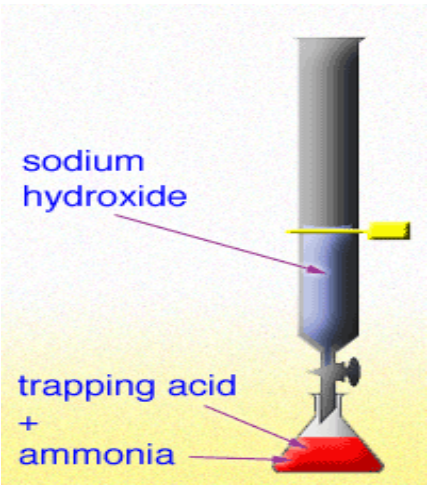
Step 2: Distillation

The pH of the mixture was raised using sodium hydroxide (45% NaOH solution). This has the effect of changing the ammonium (NH_4^+) ions (which were dissolved in the liquid) to ammonia (NH_3), which is a gas. The nitrogen was separated away from the digestion mixture by distilling the ammonia (converting it to a volatile gas, by raising the temperature to boiling point) and then trapping the distilled vapors in a special trapping solution of about 15 ml HCl (hydrochloric acid) in 70 ml of water. The trapping flask was removed and the condenser rinsed with water to ensure that all the ammonia has been dissolved.

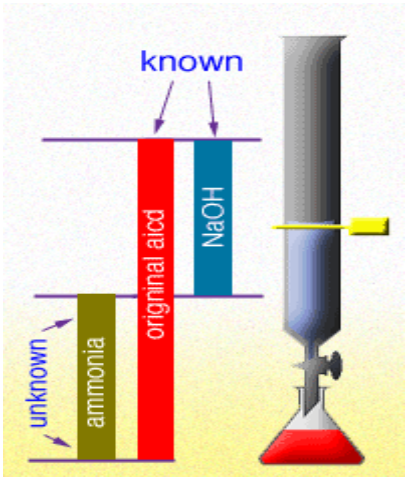
Step 3: Titration

An indicator dye was added to the acid/ammonia trapping solution. This dye turned a strong color, indicating that a significant amount of the original trapping acid was still present. A standard solution of NaOH (sodium hydroxide) was poured into the burette (a long tube with a tap at the end), and slowly, slowly adding small amounts of the sodium hydroxide solution to the acid solution with the dye. This process continued to the point at which the dye turned orange, indicating that the "endpoint" has been reached and that all the acid has been neutralised by the base.

The volume of the neutralizing base(sodium hydroxide solution) that was necessary to reach the endpoint was recorded.Amount of ammonia, and thus nitrogen that came from the original sample was calculated.



Calculations:



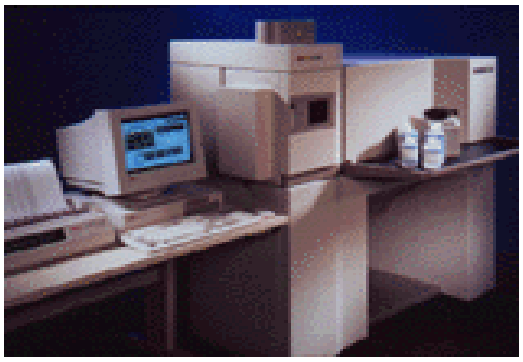
- moles of acid = molarity of acid x volume used in flask (moles A = M x V)
- moles of base = molarity of base x volume added from the burette (molesB = M x V)
- gms nitrogen = moles nitrogen x atomic mass (gN = molesN x 14.0067)
- %nitrogen = (gms nitrogen / gms sample) x 100 %N = (gN / gS) x 100
- Amount of crude protein in the sample (**CP**) can be found by multiplying the percent Nitrogen by a factor (usually 6.25).
- **CP = %N x 6.25**

Appendix 3.7

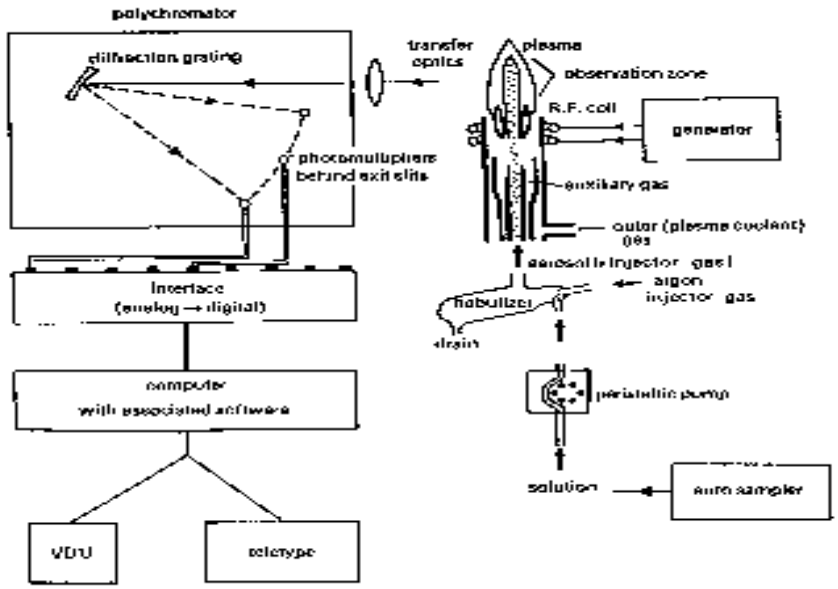
Minerals (Inductively coupled plasma analysis of micronutrients) (Pomeranz and Meloan, 1994; Brad & Cook, 1997; Xiandeng and Bradley, 2000)

An ICP typically includes the following components:

- sample introduction system (nebulizer)
- ICP torch
- High frequency generator
- Transfer optics and spectrometer
- Computer interface



An ICP



Schematic of an ICP system

Aqueous or organic solutions of samples were obtained and injected into the ICP. The nebulizer transformed the aqueous solution into an aerosol. The light

emitted by the atoms of an element in the ICP was converted to an electrical signal that was measured quantitatively. This was accomplished by resolving the light into its component radiation (nearly always by means of a diffraction grating). The light intensity was measured with a photomultiplier tube at the specific wavelength for each element line. The light emitted by the atoms or ions in the ICP was converted to electrical signals by the photomultiplier in the spectrometer. The intensity of the electron signal was compared to previous measured intensities of known concentration of the element and a concentration was computed. Each element had many specific wavelengths in the spectrum which could be used for analysis. However, the best line with the assistance from an experience technician with considerable experience of ICP wavelengths was obtained.

Recommended wavelengths and estimated instrumental detection limits

Element	Wavelength (nm)	Estimated Detection Limit (µg/L)
Aluminum	308.215	45
Antimony	206.833	32
Arsenic	193.696	53
Barium	455.403	2
Beryllium	313.042	0.3
Boron	249.773	5
Cadmium	226.502	4
Calcium	317.933	10
Chromium	267.716	7
Cobalt	228.616	7
Copper	324.754	6
Iron	259.940	7
Lead	220.353	42
Magnesium	279.079	30
Manganese	257.610	2
Molybdenum	202.030	8
Nickel	231.604	15
Potassium	766.491	See note c
Selenium	196.026	75
Silicon	288.158	58
Silver	328.068	7
Sodium	588.995	29
Thallium	190.864	40
Vanadium	292.402	8
Zinc	213.856	2

[c]Highly dependent on operating conditions and plasma position.

Appendix 3.8: Public Health Nutrition intervention Assessment Tool

Cxc ANNEX I: DEMOGRAPHY INITIALS OF PATIENTS

HOSPITAL NUMBER : _____

☐☐☐☐☐☐

Date of visit dd mm yy

IMPORTANT:
1) Make sure that written informed consent has been obtained at this point before Proceeding.

2) This screening assessment should take place within 7 days (1 week) before the Baseline
(day 0) visits.

Weight, kg **Height**,cm **Temperature**, degrees Celsius

Body Mass Index, kg

Date of birth dd mm yy

Sex
Male
Female

Status single
married divorced
widow

Risk factors (more than one box may be ticked)
Heterosexual contact
Homosexual contact
Blood product or transfusion recipient
I.V. drug use
Females
post-menopause
Sterile
Pre-menarche
Potentially able to bear children (determine serum β-HCG)
Not applicable

For females of childbearing potential, please tick the type of birth control used
oral contraceptive
intra-uterine contraceptive device
depot contraceptive (implants/injectables)

Mid-Upper Arm Circumference (MUAC) cm =

Other: _____
Not applicable
None

Tuberculosis screening
Chest X-ray date dd mm yy

Are there radio-diagnostic indications that pulmonary tuberculosis may be present?
Yes
No
PPD skin test date
dd mm yy
PPD skin test positive?
Yes
No
If PPD skin test is positive, the patient may only be included if the following conditions are met:
1: no signs of systemic or pulmonary tuberculosis;
2: the patient is willing to take INH secondary prophylaxis according to protocol guidelines (section 3.3)

Peripheral neuropathy screening

Does the patient have symptoms indicating peripheral neuropathy > grade 1?
(See Adverse Events section for Grading Scale)

Yes (=exclusion criteria)
No

For females of child-bearing potential only:
Date of pregnancy test pregnancy test outcome positive
(serum or urine β-HCG) dd mm yy (=exclusion criteria)

Negative

ANNEX II: HIV-RELATED ILLNESSES INITIALS OF PARTICIPANT

HOSPITAL NUMBER:_____

Please indicate if the patient has experienced any of the following HIV-related illnesses at any time, whether the diagnosis was definitive (Def) or presumptive (Pres) and if the illness is still present at screening.

Code	CDC-B Event	Def	Pres	Date first experienced (dd/mm/yy)	Still present Yes	No
B1	Bacillary angiomatosis					
B2	Candidiasis oropharyngeal					
B3	Candidiasis vulvovaginal (persistent, Recurrent, or unresponsive to therapy)					
B4	Cervical dysplasia (moderate or severe) / Cervical carcinoma in situ					
B5	Herpes zoster (shingles), involving at least two distinct episodes or more than one dermatome					
B6	Idiopathic thrombocytopenia purpura					
B7	Listeriosis					
B8	Oral hairy leukoplakia					
B9	Pelvic inflammatory disease					
B10	Peripheral neuropathy					
B11	Persistent fever (> 1 month duration)					
B12	Persistent diarrhoea (>1 month duration)					
B13	Pruritic papular eruptions					
B14	Other, specify :					

Code	CDC-C Event	Def	Pres	Date first experienced (dd/mm/yy)	Still present Yes	No
C1	Candidiasis of bronchi, traches, or lungs					
C2	Cervical cancer, invasive					
C3	Coccidioidomycosis, disseminated or extrapulmonary					
C4	Cryptococcosis, extrapulmonary					
C5	Cryptosporidiosis, chronic intestinal (> 1 month duration) two distinct episodes or more than one dermatone					
C6	Cytormegalovirus disease (other than liver, speen or nodes).					
C7	Cytomegalovirus retinitis (with loss of vision)					
C8	Herpes simplex : chronic ulcer(s) (> 1 month duration) ; or bronchitis, pneumonitis, or oesophagitis					
C9	Histoplasmosis, disseminated or					

	extrapulmonary					

Code	CDC-B Event	Def	Pres	Date first experienced (dd/mm/yy)	Still present Yes No	
C10	HIV-related encephalopathy					
C11	HIV wasting syndrome (more than 10% bodyweight loss with chronic diarrhea and/or fever, otherwise unexplained)					
C12	Isosporiasis, chronic intestinal (>1month duration)					
C13	Kaposi's sarcoma					
C14	Lymphoma, Burkitt's (or equivalent term)					
C15	Lymphoma, immunoblastic (or equivalent term)					
C16	Lymphoma, primary, of the brain					
C17	M.avium complex or M.Kansasii, disseminated or extrapulmonary					
C18	M.tuberculosis, any site (pulmonary or extrapulomonary)					
C19	Mycobacterium, other or unidentified species, disseminated or extrapulmonary					
C20	Oesophageal candidiasis					
C21	Pneumocystis carinii pneumonia					
C22	Pneumonia, recurrent (2 distinct episodes)					
C23	Progressive multifocal leukoencephatopathy					
C24	Salmonella septicemia, recurrent					
C25	Toxoplasmosis of the brain					

The following additional events, although not currently part of the CDC Classification System, will be
Reported as HIV-related illnesses:

Code	Other HIV-related Events	Def	Pres	Date first experienced (dd/mm/yy)	Still present Yes No	
D1	Aspergillosis					
D2	Isosporiasis					
D3	Leishmaniasis					
D4	Microsporidiosis					
D5	Nocardiasis					
D6	Penicillium marneffei, disseminated					

ANNEX III: LABORATORY INVESTIGATIONS **INITIALS OF PARTICIPANT**

HOSPITAL NUMBER :_____

Storage samples **Date of collection** ☐☐☐☐☐☐
dd mm yy

Have the following blood samples been collected for serum and plasma storage?

I large serum tube ☐ ☐ and 2 large EDTA tubes ☐ ☐
yes no yes no

Hematology **Date of collection** ☐☐☐☐☐☐
dd mm yy

Hemoglobin g/dl ☐☐☐ Neutrophile % ☐☐ ☐☐
Hematocrit % ☐☐☐ Lymphocytes % ☐☐ ☐☐
WBCcells/µl ☐☐☐ Monocytes. % ☐☐ ☐☐
Platelets 10^9/l ☐☐☐ Eosinophils % ☐☐ ☐☐
Basophils % ☐☐☐

Immunology **Date of collection** ☐☐☐☐☐☐
dd mm yy

CD4+cells /mm^3 ☐☐☐ CD4+cells % . % ☐☐☐☐
CD8+cells /mm^3 ☐☐☐ CD8+cells % . % ☐☐☐☐

HIV Virology **Date of collection** ☐☐☐☐☐☐
dd mm yy

HIV RNA ☐☐☐☐☐☐☐ copies/ml

EKG ☐ pass **Date of collection** ☐☐☐☐☐☐
dd mm yy

Laboratory Investigations (Continues)

Storage samples **Date of collection**

dd mm yy

Have the following blood samples been collected for serum and plasma storage?

I large serum tube and 2 large EDTA tubes

yes no yes no

Biochemistry **Date of collection**

dd mm yy

SGOT	U/l	
SGPT	U/l	
Bilirubin	mg/dl	
Albumin	mg/dl	
Globulin	mg/dl	
FBS	mg/dl	
BUN	mg/dl	
Creatinine	mg/dl	
Sodium	mmol/l	
Potassium	mmol/l	
Carbonate ion	mmol/l	
Chloride ion	mmol/l	
Amylase	U/l	
Protein electrophoresis		
Total Protein		

ANNEX IV:
OTHER MEDICATIONS DURING STUDY/DIETARY SUPPLEMENT/ HIVMEAL NUTRITION
INITIALS OF PARTICIPANT_____
HOSPITALNUMBER : _____

Date	**Items**	Dose

ANNEX V:
FOLLOW UP VISITS **INITIALS OF PARTICIPANT**

HOSPITAL NUMBER: _____

Please report all relevant medical history, all abnormalities found
Upon physical examination at screening, and all lab-abnormalities grade 3 or 4, that have<u>not</u> been listed in the "HIV Related Illnesses" section. In each column record Grading scale of each symptom

VISITS

1 2 3 4 5 6 7 8 9 10 11

Please, give code from 'Adverse Events Coding and Grading Scale"	Please, give name from 'Adverse Events Coding and Grading Scale"	Still present Yes No		Still present Yes No		Still present Yes No		Still present Yes No		Still present Yes No		Still present Yes No		Still present Yes No		Still present Yes No		Still present Yes No		Still present Yes No		Still present Yes No	
E1	Fever																						
E2	Sore throat																						
E3	Diarrhea																						
E4	Anorexia																						
E5	Weight loss																						
E6	Candidiasis																						
E7	Skin rash																						
E8	Herpes zoster																						
E9	Herpes simplex																						

E10	Cough																						
E11	Cough with blood																						
E12	Headache																						
E13	Insomnia																						
E14	Other symptoms specify																						

ANNEX VI: KARNOFSKY PERFORMANCE SCALE

FIRST VISIT

___100	Normal, no complaints, no evidence of disease
___90	Able to carry on normal activity : minor symptoms of disease
___80	Normal activity with effort: some symptoms of disease
___70	Cares for self: unable to carry on normal activity or active work
___60	Requires occasional assistance but is able to care for needs
___50	Requires considerable assistance and frequent medical care
___40	Disabled: require special care and assistance
___30	Severely disabled: hospitalizations indicated, death not imminent
___20	Very sick, hospitalization necessary: active treatment necessary
___10	Moribund, fatal process progressing rapidly

SECOND VISIT

___100	Normal, no complaints, no evidence of disease
___90	Able to carry on normal activity : minor symptoms of disease
___80	Normal activity with effort: some symptoms of disease
___70	Cares for self: unable to carry on normal activity or active work
___60	Requires occasional assistance but is able to care for needs
___50	Requires considerable assistance and frequent medical care
___40	Disabled: require special care and assistance
___30	Severely disabled: hospitalizations indicated, death not imminent
___20	Very sick, hospitalization necessary: active treatment necessary
___10	Moribund, fatal process progressing rapidly

THIRD VISIT

___100	Normal, no complaints, no evidence of disease
___90	Able to carry on normal activity : minor symptoms of disease
___80	Normal activity with effort: some symptoms of disease
___70	Cares for self: unable to carry on normal activity or active work
___60	Requires occasional assistance but is able to care for needs
___50	Requires considerable assistance and frequent medical care
___40	Disabled: require special care and assistance
___30	Severely disabled: hospitalizations indicated, death not imminent
___20	Very sick, hospitalization necessary: active treatment necessary
___10	Moribund, fatal process progressing rapidly

ANNEX VII:

WHO CLINICAL STAGING SYSTEM FOR HIV INFECTION AND DISEASE

The WHO Global Program on AIDS has issued the following proposed clinical staging system for HIV infection and disease. Based primarily on clinical criteria, the system is organised into four prognostic categories. It also incorporates a performance scale based on the Eastern Cooperative Oncology Group score.

Clinical Stage 1:

Asymptomatic

Persistent generalised lymphadenopathy (PGL)
Performance scale 1: asymptotic, normal activity

Clinical stage 2:

Weight loss of <10 % of body weight

Minor mucocutaneous manifestation (seborrhoeic dermatitis, prurigo, fungal nail infections, current oral ulcerations, angular shelties).

Herpes zoster, within the last 5 years

Recurrent upper respiratory tract infection (i.e., bacterial sinusitis) and/or Performance scale 2: symptomatic, normal activity

Clinical stage 3:

Weight loss of > 10 % of body weight

Unexplained chronic diarrhoea, < 1 month

Unexplained prolonged fever (intermittent or constant), < 1 month

Oral hairy candidiasis (thrush)

Oral hairy leukoplakia

Pulmonary tuberculosis, within the pass year

Severe bacterial infections (i.e., pneumonia, pyomyositis) and/or Performance scale 3: bed-ridden <50 % of the day during the last month

Clinical stage 4:

HIV wasting syndrome, as defined by the Centres for Disease Control (CDC)[2]

Pneumocystis carinii pneumonia

Toxoplasmosis of the brain

Cryptosporidiosis with diarrhoea, > 1 month

Cryptococcosis, extrapulmonary

Cytomegalovirus (CMV) disease of an organ other than liver, spleen, or lymph nodes

Herpes simplex virus (HSV) infection, mucocutaneous (> 1 month) or visceral (any duration)

Progressive multifocal leukoencephalopathy (PML)

Any disseminated endemic mycosis (i.e., histoplasmosis, coccidioidomycosis)

Candidiasis of the oesophagus, trachea, bronchi, or lungs

Atypical mycobacteriosis, disseminated

Non-typhoid Salmonella septicaemia

Extrapulmonary tuberculosis

Lymphoma

Karposis sarcoma (KS)

HIV encephalopathy, as defined by CDC[1]
and/or Performance scale 4: bedridden □50 % of the day during the last month.

(Note: Both definitive and presumptive diagnoses are acceptable).

[2] HIV wasting syndrome : Weight loss of 10 % of body weight, plus either unexplained chronic diarrhoea (1 month) or chornic waekness and unexplained prolonged fever (1 month).
[1] HIV encephalopathy: Clinical findings of disabling cognitive and/or motor dysfunction interfering with activities of daily living, progressing over weeks to months, in the absence of a current illness or condition other than HIV infection that could explain the findings.

ANNEX VIII: TOXICITY GRADING SCALE

	GRADE 1	**GRADE 2**	**GRADE 3**	**GRADE 4**
SYMPTOMS	Asymptomatic, easily tolerated, transient	Mild tolerable symptoms, short duration, normal activity	Moderate symptoms, poorly tolerated, sustained, interferes with normal activity	Severe symptoms, intolerable, sustained, incapacitating, life threatening, fatal, permanently, results in congenital abnormalities, cancer, overdose
TREATMENT	Not required	Not required except when noted	Required, responds to Rx	No response to Rx, hospitalisation required
ALLERGY	**GRADE 1**	**GRADE 2**	**GRADE 3**	**GRADE 4**
ALLERGIC REACTION	Transient	URTICARIA, DRUG FEVER >38º C, 100.4ºF. Mild bronchosphasm	Serum sickness, bronchosphasm requiring parental Rx	Anaphylaxis with hypotension
FEVER WITH DRUG (Absence of infection)	37.1-38ºC, 98.7-100ºF	38.1-40ºC, 100.5-104ºF	>40ºc, >104ºF FOR >24 hours despite antipyretic Rx	>40.ºC, >104ºF despite Rx for >24 hours, or any fever associated with hypotension
CARDIOVASCULAR	**GRADE 1**	**GRADE 2**	**GRADE 3**	**GRADE 4**
CARDIAC SYMPTOMS	Mild or transient	Symptoms on exertion, recurrent or persistent, no Rx required	Symptoms at rest, requires Rx	Severe symptoms, unresponsive to Rx
ARRHYTHMIA	Asymptomatic, transient, no Rx required	Recurrent or persistent, no Rx required	Requires Rx	Requires monitoring; or hypertension, or ventricular tachycard or fibrillation
CARDIAC BIOPSY (Index)	0.5	1.0	1.5	>1.5
CARDIAC FUNCTION	Asynptomatic, decreased ejection fraction by 20% of baseline	Asymptomatic decreased ejection fraction by >20% of baseline	Mild CHF, responsive to Rx	Severe refractory CHF

CARDIOVACULAIRE cont.	GRADE 1	GRADE 2	GRADE 3	GRADE4
OEDEMA (e.g. Peripheral capillary leak syndrome) also see Pulmonary	Minimal ankle pitting oedema	Ankle pitting oedema and weight gain 5kg	Peripheral oedema, weight gain 5-10kg, pleural effusion with no pulmonary function deficit	Anascara, severe pleural effusion with pulmonary function deficit, ascities, pulmonary oedema, weight gain>8kg.
HYPERTENSION	Asymptomatic, transient increases 20mm Hg or 150/100 if previously WNL. No Rx required	Recurrent or persistent (>1 hr) 20mm Hg or 150/100 if previously WNL. No Rx required	Persistent increase >20mm Hg or>150/100 if previously WNL. Rx required	Hypertension crisis
HYPERTENSION	10-20% decrease, systolic, no Rx required (includes transient orthostactic	21-50% decrease systolic, requiring fluids or other Rx but no hospitalisation	21-50% decrease systolic, requires pressors and hospitalisation, resolves within 48 hours	>50% decrease systolic, requiring hospitalisation, unresponsive to pressors, requires >48 hours to resolve after stopping agent
ISCHAEMIA	Non-specific T-wave flattening; stable EKG	Asymptomatic, EKG change; ST and T-wave change suggests ischaemia	New onset angina without evidence of infraction; clinical signification EKG	Acute EKG change diagnostic for myocardial infraction
PERICARDIAL EFFUSION	Asymptomatic, No Rx required	Pericarditis (rub, chest pain, EKG changes)	Symptomatic; large effusion, drainage required, no tamponade, responsive to drainage	Large effusion, temponade, drainage urgently required
CNS	**GRADE 1**	**GRAGE 2**	**GRADE 3**	**GRADE 4**
AFFECT ABNORMALITY	Transient panic/apathy; Mild anxiety/depression	Sustained panic/apathy; Moderate anxiety/depression	Sustained panic/apathy; severe anxiety/depression; Requiring Rx	Sustained panic/apathy; Unreason to Rx; Suicidal ideation
ATAXIA 9Cerebellar)	Mild/transient gait or limb ataxia; slight incoordination, dysdiachokinesia	Intention tremor, nystagmus, dysmetria	Moderate gait or limb ataxia	Disabling ataxia, cerebella necrosis
AUTOMATIC DYSFUNCTION	Abnormal sweating	Impotence	Asymptomatic arrhythmia, orthostatic hypertension	Symptomatic arrhythmia, orthostate hypertension
BLADER DYSFUNCTION		Dysfunction not requiring catheter	Dysfunction requiring catheter	Dysfunction requiring permanent catheter
COGNITIVE DEFFECT	Slow, accurate	Impaired memory or new learning	Global deficiency	Unresponsive
CONSTIPATION AUTOMATIC	Mild, no Rx required	Occasionally requiring cathartics	Daily cathartics/enema required	Abdominal distension, vomiting; Ileum>96 hours

CNS cont.	GRADE 1	GRADE 2	GRADE 3	GRADE 4
CORTICAL	Mild somnolence or agitation, easily arousable	Moderate somnolence or agitation ; responds to loud verbal or tactile stimuli	Severe somnolence, agitation, confusion, disorientation, or hallucinations ; responds to pain only	Coma, grand male seizures (current), toxic psychosis
FOCAL SEIZURES	Isolated	<2 per day	>3 per day	Status epileptics
GENERALISED SEIZURES	Isolated	<2 per day	>3 per day	Epilepsy partials continua
HEADACHE	Mild	Moderate or severe but transient	Unrelenting and severe	
HEARING LOSS	Transient decrease, loss by audiometer only	Tinnitus, moderate loss	Interferes with functions, correctable	Deaf, despite hearing aid
LANGUAGE ABNORMALITY	Inattentivness, slurring	Motor or communicative aphasia <2 hours	Motor or communicative aphasia >hours	Global aphasia
MOTOR DEFICIT	Mild/transient subjective weakness	Moderate objective weakness, ambulatory	Non-ambulatory; objective weakness	Complete paralysis
MOVEMENT DISORDERS	Transient abnormal limb movement	Moderate limb/gait disorder	Severe and reversible parkinsonism, dystonia or tremor	Permanent parkinsonism dystonia or tremor
SENSORY DEFICIT	Mild parenthesis, decreased DTR's	Mild to moderate objective sensory loss; absent DTR's moderate	Severe paresthesia, severe objective sensory loss; interferes with function	Complete loss of sensation
SPEECH ABNORMALITY	Mild slurring	Moderate slurring	Unintelligible	Mute
VERTIGO	Mild, transient	Moderate, nausea	Associates with nausea and vomiting	Disabling intractable
VISION ABNORMALITY	Slightly reduces acuity	Symptomatic, correctable	Symptomatic, unable to correct	Blind
DERMATOLOGIC	**GRADE 1**	**GRADE 2**	**GRADE 3**	**GRADE 4**

ALOPECIA	Partial loss	Complete loss	Non-reversible	
CHEILITIS	Chapping	Fissures	Bleeding	Necrosis
SKIN REACTION	Dry skin, mild or transient rash, scattered asymptomatic macular or papular eruption or erythema	Dry desquamation, scattered macular or papular eruption or erythema with pruritius or other associated symptoms	Moist desquamation, bulbous disease, generalised symptomatic macular, papular or vesicular eruption	Exfoliate dermatitis; requires surgical Rx
GASTROINTESTINAL	GRADE 1	GRADE 2	**GRADE 3**	**GRADE 4**
DIARRHOEA	Transient, >2-3 stools per day over baseline	Tolerable, >4-6 stools per day over baseline, or nocturnal stools, moderate cramping	Intolerable, >7-9 stools per day over baseline, or incontinence, severe cramping	Haemorrhagic, >10 stools per day over baseline, dehydration, requires parenteral Rx.
NAUSEA	Able to eat, reasonable intake	Able to eat, decreased intake	No significant intake	
VOMITING	1 x in 24 hours	2-5 x in 24 hours	6-10 x in 24 hours	>10 x in 24 hours, or requires parenteral support
STOMATITIS	Milk soreness, erythema, painless ulcers	Painful erythema, patchy oedema or ulcers, but can eat	Confluent ulcers, painful erythema, necrosis, requires parenteral or entereal support	Haemorrhagic ulceration, necrosis, requires parenteral or entereal support
GENERAL	**GRADE 1**	**GRADE 2**	**GRADE 3**	**GRADE 4**
MYALGIA	Myalgia	Myalgia requiring treatment	Severe, CK 2.0-5,0 X ULN	Intractable, CK >5 x ULN
CHILLS	Mild	Moderate		
LOCAL	Pain	Pain and swelling, inflammation or phlebitis	Ulceration	Plastic surgery indicated
HAEMATOLOGICAL	**GRADE 1**	**GRADE 2**	**GRADE 3**	**GRADE 4**

Anaemia (gHgb)	10 – normal	8,0 - 10	6,5 – 7,9	<6.5
GRANULOCYTOPENIA (X 10 μL)	1-5.9 and □20% decrease from baseline	1.0- 1.4and □35% decrease from baseline	0.5 – 0.9 and □50% decrease from baseline	□0.5 and >75% decrease from baseline
LEUKOPOENIA (X10 μl)	3.0 – 3.9 and >20% decrease from baseline	2.0 – 2.9 and >35% decrease from baseline	1.0 – 1.9 and >50% decrease form baseline	<1.0 and > 75% decrease from baseline
THROMBOCYTOPOENIA (X10 μL)	75-99 and >20% decrease from baseline	50-74 and >35% decrease from baseline	25-49 and >50% decrease from baseline	<25 and >75% decrease from baseline
HAEMORRHAGE	Petechiae, minimal blood loss, no transfusion required, mild	Gross, transfusion required, 1-2 U	Gross, transfusion required 3-4 U	Massive transfusion required, ≥4 U
HAEPATIC	GRADE 1	GRADE 2	GRADE 3	GRADE 4
ALKALINE PHOSPHATASE INCREASE	1.5 –2.5 X ULN1	>1.5 – 2.5 X ULN1	>5.0 – 10.0 X ULN1	>10.0 X ULN1
BILIRUBIN INCREASE		1.3 – 1.5 X ULN1	>1.5 – 3.0 X ULN1	>3.0 X ULN1
HEPATIC SYMPTOMS			Pre-coma	Hepatic Coma
TRANSMINASE INCREASE	1.5 – 2.5 X ULN1	>2.5 – 5.0 X ULN1	>5.0 – 20 X ULN1	>20 X ULN1
INFECTION	GRADE 1	GRADE 2	GRADE 3	GRADE 4
INFECTION	Mild Infection, Unknown origin	Moderate infection	Major organ infection	Disseminated infection, multi-lobar pneumonia; life-threatening sepsis
FEVER	37.1-38°C	38.1-40°C	>40°C, >104°F for 24 hours despite antipyretic Rx	>40°C, >104°F for >24hours or a fever associated with hypotension

PULMONARY	GRADE 1	GRADE 2	GRADE 3	GRADE 4
PULMONARY FUNCTION ABNORMALITY	FVC 70-80% of predicted FEV, or DLCO 60-80% of predicted; 15-25% decrease from abnormal baseline	FVC 50-69% of predicted FEV, or DLCO 40-59% or predicted; 50% decrease from abnormal baseline	FVC <50% of predicted FEV, or DLCO <40% of predicted; 50% decrease from abnormal baseline	Unable to perform test due to respiratory distress
RESPIRATORY SYMPTOMS	Mild or transient, asymptomatic with PFT (Pulmonary Function Tests) abnormal	Dyspnea on significant exertion	Symptoms during normal activity; persistent Dyspnea	Severe symptoms a1 rest, non-responsive to Rx
CHEST X-RAY	<10% lung fields show infiltrate or effusion	10-20% lung fields show infiltrate or effusion	21-50% lung fields show infiltrate or effusion	>50% lung fields show infiltrate or effusion
ABG	PAO2 <95 on room air	PAO2 <80 on room air	PAO2 <60 on room air	PAO2 <60 on supplemental oxygen
RENAL	GRADE 1	GRADE2	GRADE 3	GRADE 4
CREATININE INCREASE (mg/dL)	1.25-2.5 X ULN1	>2.5-5.0 X ULN1	>5.0-10.0 X ULN1	>10 X ULN1; requiring dialysis)≥8.0), irreversible loss of >20%
CLACULATED CREATININE CLEARANCE	70-80% of baseline	50-69% of baseline	30-49% of baseline	<30% of baseline
DYSURIA	Mild	Moderate	Severe	
HAEMATURIA	6-10 RBC/HPF2	11-50 RBC/HPF2	>50 RBC/HPF2	Requires transfusion
PROTEINURIA	1; <3.0g%, <3g/l	2-3; >0.3-1, 0g%, 3-10gl	4; >1.0G%, >10g/l	Nephritic syndrome

METABOLIC	GRADE 1	GRADE 2	GRADE 3	GRADE 4
HYPERGLYCAEMIA	140-160MG/dL	161-250MG/dL	251-500mg/dL	500MG/dL; ketoacidosis
HYPOGLYCAEMIA (Random)	55-64ML/dL	40-54mg-dL	30-39MG/Dl	>30MG/dL
MYLASE	>1.5 XUNL1	1.5-2.0 XUNL1	2.1 –5.0 XULN1	>5.0 XULN1
HUPERCALCAEMIA	10.6-11.5MG/dL	11.6-125MG/dL	12.6-13.5MG/dL	>13.5MG/dL
HYPOCLACAEMIA	8.4-7.8mg/dL	7.7-7.0MG/dL	6.9-6.81MG/dL	>6.1MG/dL
HYPOMAGNESEMIA	1.4-1.2mg/dL	1.1-0.9mg/dL	08-06mg/Dl	≤0.5mg/Dl
HYPOALBUMINEMIA	2.8mg/dL	2.4-2.7mg/dL	2.0-2.3mg/Dl	≤2.0mg/Dl
COAGULATION	GRADE 1	GRADE 2	GRADE 3	GRADE 4
FIBRINOGEN	0.99-0.75 XUNL1	0.74-0.5 XUNL1	049-0.25 XUNL1	≤0.24 XUNL1
PROTHROMBIN TIME	1.1-25 XULN1	1.26 XULN1	1.51 –2.0 XULN1	>2.0 XULN1
PARTIAL THROMBOPLASTIN TIME	1.1 – 1.66 X UNL1	1.67-2.33 XULN1	2.34.3.0 XULN1	>3.0 X ULN1

KEY:

ULN – Upper Limit of Normal
RBC/HPF – Red Blood Cells/High Power Field

Appendix 3.9

Names, profession/designation and responsibilities of the research team

Research Team			
S/No.	Study Personnel	Profession/Designation & Role in the Project	Remarks
1.	Dr. Abraham Amlogu (Principal Investigator)	Consultant Clinical Pharmacist: Recruiting patients, general supervision and monitoring laboratory investigations; writing and submitting reports as required.	
2.	Pharm. Dawuda Bage (Logistics Management)	Pharmacist: Co-ordination of product supply and dispensing Amtewa meal and HAART	
3.	Dr. Victoria Ogala-Akogwu (Project Coordinator)	Senior Registrar (Medicine): Recruiting patients & coordinating clinic activities, investigations and home visits.	
4.	Dr. T. Acho	Physician: Recruiting patients & assisting with patients' follow up.	
5.	Matron Aishatu Jakada	Nurse & Counsellor: Counselling, nursing care of patients and home visiting.	
6.	Mrs Bello	Nurse: Nursing care of patients and home visiting.	
7.	Mr. Evaristus Ibekwe	Med. Lab. Scientist: CD4 count, and monitoring other biochemical indices.	
8.	Mr Ifeanyi Ikete	Med. Lab. Scientist: CD4 count, and monitoring other biochemical indices.	
9.	Mrs. Ibilanke Yomi	Nutritionist: Monitor the nutritional status of patients.	
10.	Mr Mohammed Maigari	Medical Records officer: Collate participants' case files for routine assessment, investigations and evaluation	
11.	Mr. Henry Ukachi	Statistician: Statistical analysis of results	

Appendix 3.10

Programme of Events for the TOT Training

Time	Activity	Facilitator
8.00am	Registration	Research Assistant
8:35am	Welcome address, Introduction of the research aim, objectives and expected outcome	Principal Investigator
8:45am	Introduction, ground rules and appointment of a rapporteur	Research Assistant
9:00am	General research data collection	Medical Records Officer
9:30am	Nursing HIV care and the significance of patient follow up in a research setting	Nurse & Counsellor
10:00am	Nutritional status and anthropometric measurements in HIV care	Nutritionist
10:30am	HIV & Nutrition – Gaps	Physician
11:00am	Laboratory investigations in HIV care and support	Laboratory Scientist
11:30am	Adherence to HAART and Amtewa meal in HIV care	Pharmacist
12:00pm	Statistical analysis of variables in the research	Statistician
12:30pm	Review of the research assessment tool	Principal investigator
1:00pm	Interactive session	ALL
1:30pm	Lunch and closing	ALL

Appendix 3.11

List of Publications based on this study

1**. Amlogu**, M. A., Tewfik, S., Wambebe, C., Godden, K. and Tewfik, I. (2011) Conceptual framework of public health-nutrition intervention programme to attenuate the progression of HIV to AIDS among People Living with HIV (PLWH) in Abuja, Nigeria. In Sharing Knowledge Making a Difference: The Role of International Scientific Cooperation, World Sustainable Development Outlook 2011. ISBN 978-1-907106-12-5, 11-20.

2. **Amlogu**, A.M., Godden, K., Tewfik, S., Wambebe, C. & Tewfik, I. (2012). Tailored Food Recipe – TFR: Employing the European perspective on functional food science (FUFOSE) to promote effective dietary intervention in Africa. International Journal of Food, Nutrition & Public Health, 5 (1/2/3), 1-10.

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TAILORED FOOD RECIPES-TFR: EMPLOYING THE EUROPEAN PERSPECTIVE ON FUNCTIONAL FOOD SCIENCE (FUFOSE) TO PROMOTE EFFECTIVE DIETARY INTERVENTION IN AFRICA

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Abstract: *Purpose:* Because of increasing interest in the concept of "Functional Foods" and "Health Claims", the European Union set up a European Commission Concerted Action on Functional Food Science in Europe (FUFOSE). The report takes the position that functional foods should be in the form of normal foods and they must demonstrate their effects in amounts that can normally be expected to be consumed in the diet.
Methodology: Currently, health concerns of communicable and non-communicable diseases have necessitated investigating into options for dietary interventions

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PUBLIC HEALTH-NUTRITION INTERVENTION PROGRAMME TO ATTENUATE THE PROGRESSION OF HIV TO AIDS AMONG PEOPLE LIVING WITH HIV (PLWH) IN ABUJA, NIGERIA: A CONCEPTUAL FRAMEWORK

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Abstract
Purpose: This presented pilot intervention provides evidence that suggests the use of local resources as therapeutic nutrition. The latter can act as a fundamental part of the comprehensive package of care at the country level.
Background: HIV/AIDS is a pandemic disease and its scourge has had a devastating impact on health, nutrition, food security and overall socio-

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Tailored Functional Recipe (TFR) Approach to Delay the Progression of HIV to AIDS among People Living with HIV (PLWH) in Abuja, Nigeria

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Abstract

Background: HIV/AIDS is a pandemic disease and its scourge has had a devastating impact on health, nutrition, food security and overall socioeconomic development in affected countries. Moreover, intervention programmes, which simply employ antiretroviral drugs, have been found to lack effectiveness particularly when the patient is under-nourished. Aim and Purpose: This presented pilot intervention provides evidence that suggests use of local resources as therapeutic nutrition. This can act as a fundamental part of the comprehensive package of care at the country level. Methodology: Local ingredients, which were known for their availability, accessibility, micro and macro-nutrient strengths were selected and optimised into a nutritional functional meal (*Amtewa*). Daily consumption was ascertained to assess its effects on nutritional status and bio-medical indices of the study participants (n = 100) who were/were not taking Highly Active Anti-retroviral Therapy (HAART). Findings: Mean CD4 count for ART-Test group at baseline and sixth months increased by 40.8 cells/mm^3 while the ART-Control group decreased 18.12 cells/mm^3. This positive outcome qualified *Amtewa* meal to the next phase of intervention (400 participants) to ascertain its effectiveness on health status of HIV infected subjects and appraise its position within the National Health Services framework as innovative approach to attenuate the progression of HIV to AIDS in Nigeria. Conclusion: Amtewa-based approach in HIV management is innovative, culturally relevant, reliable and requiring low technology in order to assure compliance, sus-

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Appendix 3.12

List of International Conferences and Presentation

S/N	DATE	CONFERENCE/PRESENTATION	VENUE	TOPIC	REMARKS
1	4TH JULY, 2011	3RD HIV SYMPOSIUM	The Royal London Hospital	Public Health Nutrition Intervention to attenuate the progression of HIV to AIDS among People Living with HIV virus in Abuja, Nigeria	Poster
2	26TH – 28TH OCT.2011	World Association for Sustainable Development 9th International Conference, Atlantic City, USA, 2011	Atlantic City, USA	Same as above: Conceptual Framework	Oral presentation Published
3	4th Dec. 2011	Royal Society of Tropical Medicine and Hygiene Research in Progress 2011: Short presentations and poster	University of London	Same as above	Oral presentation
4	11th - 12th May, 2013	13th International Conference on Functional Food in Health and Disease Kyoto, Japan	University of Medicine, Kyoto, Japan	Tailored Food Recipes-TFR: Employing the European perspective on functional food science (FUFOSE) to promote effective dietary intervention in Africa	Poster Abstract
5.	31st August – 5th September, 2013	Federation of International Pharmacist (FIP) World Congress 2013	Dublin, Ireland	Tailored Functional Recipe (TFR) approach to delay the progression of HIV to AID among PLWHIV in Abuja, Nigeria.	Poster Abstract. Approved no: 418