Study Title:

BonE health in ageING Women: Improvement or prevention of changes in Bone Mineral Density by Switching Antiretroviral Agents. Is there an optimal time to intervene?

Protocol #: **IN-CA-311-3963/CTN 299**

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Sponsor-Principal Investigator: Dr. Sharon Walmsley, University Health Network

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GCP Statement

This clinical study will be conducted in accordance with applicable Health Canada regulations, International Conference on Harmonisation (ICH) guidelines on current Good Clinical Practice (GCP), and the Declaration of Helsinki.

Confidentiality Statement

This clinical study protocol contains information which is of a confidential, trade-secret or proprietary nature. The protocol is for the use of Dr. Sharon Walmsley and her designated representatives participating in the investigational trial. It is not to be disclosed to any other person or party without the prior written approval of Dr. Sharon Walmsley

APPROVED	
Sponsor-Investigator	Dr. Sharon Walmsley
Signature	
Date	

INVESTIGATOR AGREEMENT

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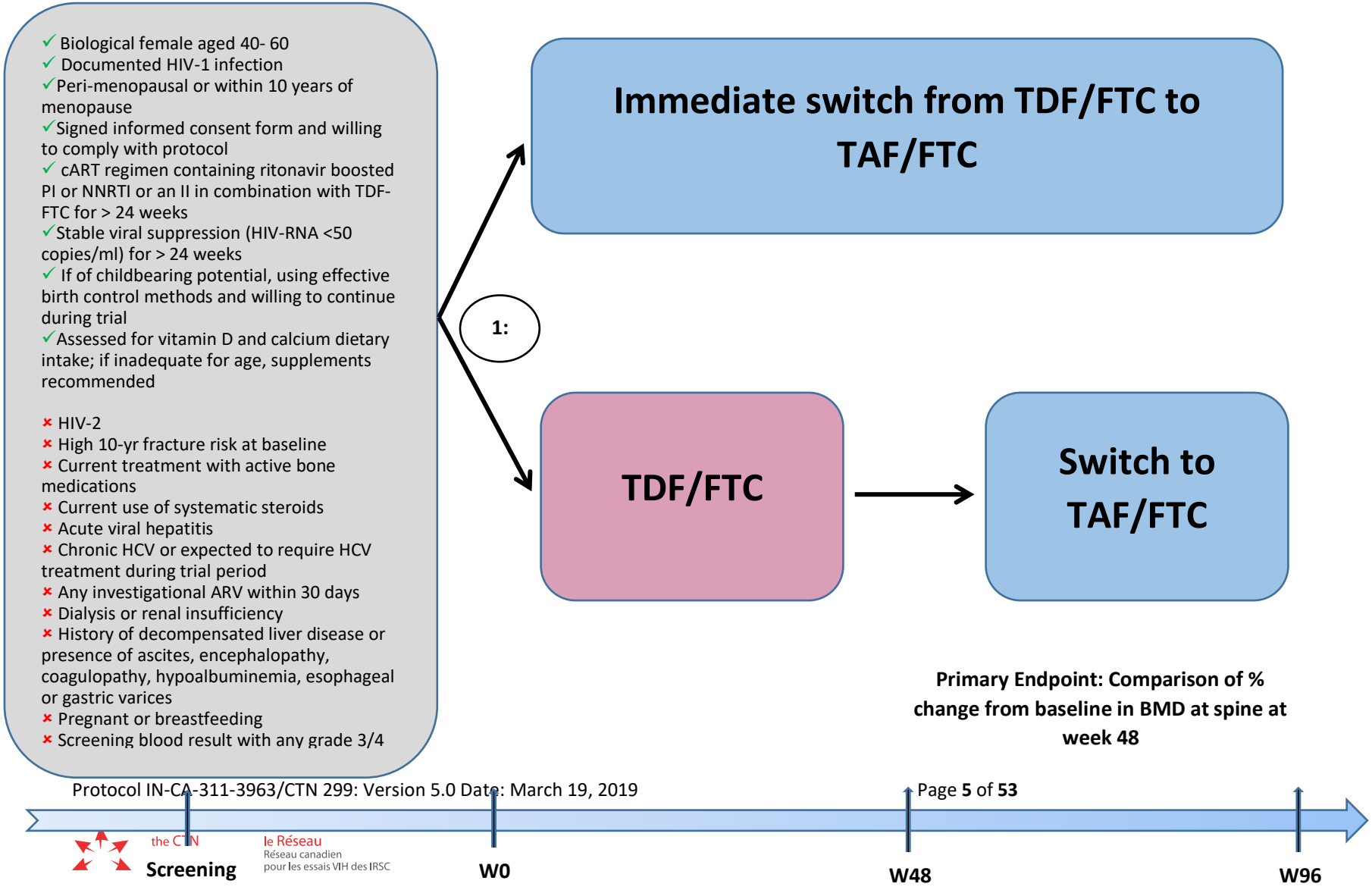
This clinical study will be conducted in accordance with applicable Health Canada, European Medicines Agency (EMA) regulations, ICH guidelines on current GCP, and the Declaration of Helsinki.

I confirm that I have read and understand this protocol and I agree to conduct this clinical study in accordance with the design and specific provisions of the protocol, with the exception of a change intended to eliminate an immediate hazard to participants. Any deviation from the study protocol will be documented in the case report form.

I agree to promptly report to the applicable ethics boards any changes in the research activity and all unanticipated problems involving risks to human participants or others. Additionally, I will not make any changes in the research without prior ethics and sponsor approval, except where necessary to ensure the safety of study participants.

Name Signature Date (dd-mmm-yyyy)

Schematic of Study Design



ABBREVIATIONS AND DEFINITIONS

Acronym / Abbreviation	Definition
AE	Adverse Event
ARV	Antiretroviral
BMD	Bone Mineral Density
CaMos	Canadian Multicentre Osteoporosis Study
cART	Combination antiretroviral therapy
DAIDS	Division of Aids (table for grading severity of adverse events)
DXA	Dual Energy X-ray Absorptiometry
F/TAF or TAF/FTC	Tenofovir alafenamide – coformulated with Emtricitabine
FTC	Emtricitabine
FRAX®	Algorithm for calculating fracture risk
HATS	HIV Adherence Treatment Scale
HCV	Hepatitis C virus
HIV	Human immunodeficiency virus
HRpQCT	High-resolution peripheral quantitative computed tomography
IATA	International Air Transportation Association
ICJME	International Committee of Medical Journal Editors
NNRTI	Non-nucleoside reverse transcriptase inhibitor
PI	Protease Inhibitor
RCT	Randomized Controlled Trial
SAE	Serious Adverse Event
SPPB	Short performance physical battery
TAF	Tenofovir alafenamide
TBS	Trabecular bone score
TDF	Tenofovir disoproxil
TDF/FTC	Tenofovir disoproxil/ emtricitabine
VAS	Visual Analogue Scale

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1.0 BACKGROUND AND SCIENTIFIC RATIONALE

1.1 Introduction

With health advances from combination antiretroviral therapy (cART) management of comorbid illness is now a major focus of research and clinical care of HIV infected persons.[1,2]. Osteoporosis is a systemic skeletal disease with an increase in bone fragility and susceptibility to fracture[3]. HIV independently contributes to low bone mineral density (BMD) through viral, immune, inflammatory and ART-related effects on bone metabolism.[4, 5]

The Canadian Multicentre Osteoporosis Study (CaMos) estimates the prevalence of osteoporosis at the lumbar spine in the general population of women > age 50 to be 12.1%.[6] The most serious manifestation is a fragility fracture, defined as a fracture occurring spontaneously or following minor trauma. The consequences of fracture include increased mortality, morbidity, institutionalization and economic costs. There is a 25% risk of death in the year following a hip fracture with excess risk continuing into the second year.[7]The HIV population is aging with > 50% now over 50 years. Osteoporosis is higher in HIV relative to the general population, is greater in severity and increases with age.[8, 9]

Although greater BMD losses are seen with initiation of cART including tenofovir (TDF) relative to other antiretroviral agents[10-2], the co-formulation of TDF/FTC (emtricitabine) is the preferred NRTI backbone for most cART regimens and hence the majority of HIV patients are exposed to TDF, often for many years.[13] Greater BMD losses are seen when cART is initiated with this agent, possibly due to a form of immune reconstitution syndrome[14]. Further BMD loss occurs with continued use likely due to an effect on osteoclastic activity[10]. Cohort studies have observed an increased fracture risk with use of this agent especially with age. [15,16] The relative importance of low BMD to clinical care differs significantly between countries and regions, based on factors such as the population at risk, access to adequate screening resources and physicians' knowledge.[17] There are no guidelines as to whether BMD should be monitored differently in HIV than the general population, whether HIV alone is an independent factor to be used in fracture risk assessment nor whether TDF should be discontinued if osteoporosis develops on therapy.[10] In many situations, it is not safe to stop TDF for risk of loss of HIV control.

1.2 Study agent

As a prodrug, tenofovir disoproxil fumarate is initially metabolised to tenofovir, which is subsequently metabolised in cells to tenofovir-diphosphate. Although intracellular tenofovir-diphosphate is responsible for the drug's antiviral activity, higher circulating plasma levels of tenofovir have been associated with an increased risk of both renal and bone toxicity. A novel tenofovir prodrug tenofovir alafenamide results in roughly four times higher intracellular concentrations of the active metabolite tenofovir-diphosphate compared with tenofovir disoproxil fumarate, allowing for much lower doses of tenofovir alafenamide versus tenofovir disoproxil fumarate. Because of tenofovir alafenamide's reduced dose and the improved stability,

plasma exposure of tenofovir is 90% lower with tenofovir alafenamide than with tenofovir disoproxil fumarate, which is believed to reduce the risk of renal and bone toxicity.[18] Initial licencing studies compared a co-formulation of TDF/FTC with an integrase inhibitor elvitegravir boosted by cobicistat (Stribild) to a similar co-formulation of elvitegravir, cobicistat and TAF/FTC[19]. The non -inferior viral suppression rates, CD4 count increases, rate and spectrum of adverse events, and resistance profiles with failure led to the approval of the latter as Genvoya[20], recently in use in Canada. Of note, Initiation of cART with TAF/FTC was shown to cause statistically significant lesser BMD losses, and smaller declines in glomerular renal function than TDF/FTC [19]. The lower serum level is thought the reason for potential less bone and renal toxicity.

Subsequent studies that evaluated a switch from TDF/FTC to TAF/FTC in HIV infected persons with viral suppression, the absence of viral resistance and with a variety of third cART agents were associated with a small but statistically significant increase in BMD[21-3] in largely younger male populations. Although BMD assessment with dual energy x-ray absorptiometry (DXA) is well established for the diagnosis of osteoporosis and for fracture risk assessment in postmenopausal women it is not validated in young men.[14] Therefore it remains uncertain whether these changes will be clinically relevant.

1.3 Rationale for this Study

TAF (co-formulated with FTC) as a fixed tablet regimen is recently licenced in Canada[24] and Europe. It will be unclear for treating physicians and policy makers whether all HIV+ who require TDF/FTC should be switched to TAF/FTC. It is anticipated that the cost will be similar to the current price for TDF/FTC (approximately $1000 CAN per month), however generic TDF/FTC will soon be available and will be considerably cheaper. Statistically significant increases in BMD that are not linked to clinical outcomes will be insufficient to guide that decision.

Given sample size considerations and time to fracture endpoints, clinical validation studies are not possible. Further, it is unknown whether BMD can be improved in aging HIV+ women through cART switching. Demonstrating changes in BMD that correlate with fracture risk in a population of older HIV infected women in whom BMD data is validated with fracture risk will provide better evidence. To better understand the potential impact, in this study we also include new bone imaging modalities – trabecular bone score [25]and HR-pQCT[26] that collect further information about bone microarchitecture and bone strength to more fully understand changes in bone health. There is emerging evidence that changes detected by these new imaging modalities are independently associated with fracture risk. In previous work we showed an excess of fragility fractures in HIV infected women despite similar BMD scores.[27] suggesting that other parameters are important to evaluate risk. Since women experience sharp declines in BMD during menopause, the potential for improved bone health through adjustment of HIV therapy may reduce future health burdens associated with osteoporosis

1.4 Potential risks and benefits to participants

Tenofovir alafenamide as an individual agent has been administered to more than 40 HIV-positive participants in two separate studies. In study GS-US-120-1101, participants received higher doses of TAF (50 mg and 150 mg) than the TAF dose in this study. Side effects that were seen in more than two participants were headache, nausea, and gas (flatulence). In study GS-US-120-0104, adverse events observed in more than two participants were nausea and fatigue. One serious adverse event of chest pain was reported, but not considered related to study drugs.[28]

In addition, F/TAF has been given to over 3000 HIV infected participants in switching studies[21-3]. In these trials, there was no new toxicity observed relative to TDF/FTC and less than 1 % had to discontinue medication secondary to adverse events. The most frequently described adverse events occurring in more than 5% of persons included headache, diarrhea, nasal congestion, and fatigue.

A study in dogs detected eye problems (posterior uveitis- inflammation in the back of the eye) in some dogs when TAF was given at the highest doses. There have not been cases of uveitis related to TAF in clinical studies. [28]

TAF is a drug similar to Tenofovir disoproxil fumarate (TDF), Viread®, an approved medication by the FDA for the treatment of HIV and hepatitis B infection[29]. The side effects for TAF are expected to be similar to Viread®. The side effects for Viread® are listed below.

Tenofovir DF (TDF, Viread®)

Tenofovir DF has been studied in more than 12,000 HIV-infected adults for as long as 480 weeks in some patients. Common potential side effects identified in patients who received at least one dose of tenofovir DF 300 mg include diarrhea, nausea, vomiting, flatulence (intestinal gas), and dizziness. Those side effects were often mild or moderate in severity, and did not lead to discontinuation of tenofovir DF.[29]

In addition to side effects reported from clinical trials the following side effects have also been identified after tenofovir DF was approved by the FDA and Health Canada in HIV-infected patients treated with combination therapy that has included tenofovir DF and other anti-HIV drugs. Because post marketing (after regulatory licensing) reactions are reported voluntarily from a patient population of uncertain size, it is not always possible to reliably estimate how frequently these side effects occurred, or establish a causal relationship to tenofovir DF. The side effects which have been reported with tenofovir DF when taken with other anti-HIV drugs include:

- allergic reaction
- rash
- abdominal pain
- pancreatitis
- shortness of breath
- weakness
- abnormalities of hepatic (liver) function and hepatitis (inflammation of liver).

Cases of lactic acidosis, hepatosteatosis, including fatal cases, were reported in HIV-infected patients treated with anti-retroviral agents similar to tenofovir DF. The symptoms of lactic acidosis include:

- weakness
- unexpected and uncommon abdominal pain
- nausea
- vomiting

Symptoms of liver problems include:

- yellowing of the skin or whites of the eyes
- dark urine
- light colored bowel movements
- nausea
- loss of appetite
- lower abdominal pain

Cases of kidney damage have been reported in patients taking tenofovir DF who already have atherosclerosis, or specific kidney disease, and patients who, while receiving tenofovir DF, were also taking medications that may cause damage to the kidneys. Kidney damage has also been reported in patients without any of these factors. In addition, continuous or sudden kidney failure, abnormal kidney function, inflammation of the kidneys, protein in the urine, excessive urination, nephrogenic diabetes insipidus, and increased creatinine have also been reported in patients taking tenofovir DF.

Bone toxicity, including a decrease in bone mineral density was seen in animals following treatment with tenofovir DF. Decreases in bone mineral density have been seen in humans. The risk of bone fractures associated with these types of changes is unknown.

Because these events have been reported voluntarily from a population of unknown size, estimates of frequency cannot be made.

Emtricitabine (FTC, Emtriva®)[30]

The most common adverse reactions seen in greater than or equal to 10% of patients treated with FTC include:

- headache
- diarrhea
- nausea
- fatigue
- depression
- trouble sleeping
- abnormal dreams
- rash
- weakness
- increased cough
- runny nose
- dizziness
- abdominal pain

Other common side effects seen more than 1% and less than 10% with emtricitabine include: pain, weakness, vomiting, indigestion, changes in skin color primarily on the palms and/or soles, increased triglycerides, increased bilirubin, increased blood glucose, allergic reaction, increased liver enzymes , increased pancreatic enzymes and low white blood cell count.

Rare cases of lactic acidosis, and hepatosteatosis were reported in HIV-infected patients treated with anti-HIV medications similar to emtricitabine.

Allergic Reaction Risks

As with taking any drug, there is a risk of allergic reaction.

Blood Draws

Drawing blood from a vein may cause local pain, bruising, occasional light-headedness, fainting, and very rarely, infection at the site of the blood draw.

DXA

Decreases in bone mineral density have been observed in HIV-infected patients[10]. An X-Ray called dual energy X-ray absorptiometry, or a DXA scan, will be performed on spine and hip to measure changes in bone mineral density. The radiation exposure is lower than with a chest x-ray. The DXA scans are considered, in general, less harmful than a standard x-ray[31]

HRpQCT

This scan provides different types of information about bone strength and structure. Measurements are done on one forearm and one ankle and are performed when seated in a chair. This exposes participants to a small amount of radiation similar to that of the DXA scan[31]. This test will only be available at selected participating sites.

Hepatitis B and C Testing Risks

Participants will be tested for Hepatitis C and B at the screening visit, if not previously recorded. The results of these tests, if active, must be reported to the local health authority, such as Public Health.

2.0 STUDY SYNOPSIS

This is a randomized controlled clinical trial (RCT) of immediate vs delayed switch from TDF/FTC to TAF/FTC to define the impact of switching ARV on BMD in different stages of the aging trajectory in HIV infected women. Included is a geriatric assessment based on the conceptualization of health transition across menopause. The study will be conducted in five Canadian (University Health Network, Vancouver Infectious Diseases Centre, Centre Hospitalier de l'Université Laval, McGill University Health Centre, Hamilton Health Sciences Centre, and two Italian centres (Modena metabolic clinic and Milan clinic) by HIV physicians with interest and expertise in women's issues in HIV care. The Maple Leaf medical clinic in Toronto will refer all their participants to the UHN site for study procedures.

3.0 STUDY HYPOTHESIS

The primary hypothesis is that switching from TDF/FTC will improve BMD to a degree that correlates with a lower fracture risk in aging HIV+ women. We will further explore our theory that the impact is greater in those in the early stages of menopause and in those who also receive a protease inhibitor (PI) as the third ARV.

4.0 STUDY OBJECTIVES

4.1 Primary objectives:

To determine if:

1. Switching HIV+ women on TDF/FTC to TAF/FTC increases BMD at the spine at 48 weeks relative to those who continue TDF/FTC
2. To determine if any observed improvements continue or stabilize in the year after switch.

4.2. Hypothesis generating objectives:

To determine if the effect of switching from TDF/FTC to TAF/FTC on BMD varies by stage of menopause and by third ARV.

5.0 STUDY DESIGN

This study is open-label controlled randomised, 1:1, multicentre strategic trial. Patients will be randomised to immediate vs delayed switch, with randomization allocation arranged to minimize differences between treatment groups with respect to country.

5.1 Study population:

HIV positive women who are in the peri-menopausal period or those within 10 years post menopause to capture those with greatest risk of BMD loss. As menopause typically occurs earlier in HIV + women we include those aged 40 - 60 years.[32] They must be on a cART regimen containing TDF/FTC with HIV RNA <50 c/ml for at least 6 months.

5.2 Intervention:

1. Immediate switch of TDF/FTC to TAF/FTC while maintaining the third ARV agent.

2. Delayed switch of TDF/FTC to TAF/FTC at 48 weeks while maintaining the third ARV agent.

5.3 Randomization

A computer-generated randomization list will be prepared prior to study onset by a statistician/programmer at the CIHR Canadian HIV Trials Network. Randomization will be stratified by country. Site coordinators will each be given a personal code through which they

can access the web randomization system. When a site coordinator has identified an eligible, consenting participant, s/he will access the web page, register the participant and indicate non-nominal information linked to the participant. The computer will record date and time of the transaction, the participant information and allocation, which will be issued to the coordinator.

5.4 Primary endpoint:

Comparison of the immediate vs delayed group in the % change from baseline in BMD at the lumbar spine at week 48.

5.5 Secondary endpoints

We will compare changes between the immediate and delayed group from data collected at screening or baseline and weeks 48 and 96.

% change from baseline in BMD at lumbar spine at 96 weeks

% change from baseline in BMD at hip at 48 weeks

% change from baseline in BMD at hip at 96 weeks

Changes in Bone architecture as determined by Trabecular bone scan (TBS)[25] and HRpQCT [26]

Changes in 10 year fracture risk determined by country specific FRAX calculator[33]

% with undetectable viral load HIV-1 RNA as per current assays. (This may be < 50/ml, < 40/ml or viral load not detected)

Change from baseline in geriatric functional measures: frailty, performance and balance

Change from baseline in muscle quality: Sarcopenia – grip strength measured by a Dynamometer and fat free mass measure (from the DXA)

Change from baseline in lipid values and Framingham cardiovascular risk scores

Changes in renal tubular and glomerular function: eGFR, Cr, urine alb/cr and protein/cr, glucosuria

Safety (clinical and laboratory adverse events)

Changes in biomarkers of inflammation, coagulation and bone metabolism

Tolerability (EuroQoL questionnaire)

5.6 Justification of Endpoints:

Primary Endpoint: BMD assessment with dual energy x-ray absorptiometry (DXA) is well established for the diagnosis of osteoporosis and for fracture risk assessment in postmenopausal women[7]. For BMD measurements to be clinically useful, they are expressed in comparison with

established normative databases. BMD is expressed as a numerical value (g/cm2). Change in BMD is considered a valid intermediate end point for efficacy of fracture risk reduction[7]. To inform the site and timing of the primary endpoint, data from studies of biphosphonates (a major therapy for osteoporosis) was used in which the increase in BMD is greatest at the lumbar spine, with one half of the gain of BMD achieved within 6–12 months followed by a lower increase over subsequent years[34]

Secondary Endpoints: New imaging modalities are increasingly incorporated into studies of osteoporosis to gain more detailed information on bone strength.

Trabecular bone score (TBS) is a recently-developed analytical tool that performs novel measurements on lumbar spine DXA images, and captures data relating to trabecular microarchitecture[25]. Low TBS is consistently associated with an increase in both prevalent and incident fractures that is partly independent of both clinical risk factors and BMD. TBS has predictive value for fracture independent of probabilities using the FRAX® algorithm.[33]

High-resolution peripheral quantitative computed tomography (HRpQCT) is a new method that can determine both bone geometric parameters (such as cortical thickness, trabecular thickness, trabecular separation and trabecular number) and BMD (such as total, cortical and trabecular volumetric BMD).This technology offers a non-invasive method to gain unique insights into bone microarchitecture that are not obtainable using routine DEXA and can provide additional 3-dimensional information about bone architecture that is associated with fracture, even among those with similar DXA results.[26] Given limited availability it will only be performed at selected participating sites.

As BMD does not add details on overall function or provide data on clinical outcomes, we include a broader geriatric assessment of study participants including:

1. **Sarcopenia** with a combined definition of fat free mass measure (from the DXA) and muscle grip strength obtained with a handheld dynamometer[35]

2. **Frailty** measured with some components from a Frailty Index.

3. **Short performance physical battery test (SPPB)** to assess balance, and walking speed [37]

4. **Fall frequency**, assessed using self-administered questionnaire validated in the 2008 National Health Interview Survey Dizziness and Balance Supplement.[38]

Increasingly **biomarkers** are studied with the goal to identify surrogate markers for use in interventional studies where clinical endpoints are less frequent. Many of the comorbidities seen with aging in HIV may be a consequence of persistent immune activation and inflammation despite control of HIV replication. Therefore we propose to collect samples to enable further analysis of putative biomarkers to be correlated with BMD changes.[4, 39-41]

1. **Biomarkers of bone formation and resorption**- to include - C-terminal cross-linked telopeptide of type I collagen (CTX) procollagen 1 N terminal extension peptide (P1NP) , vitamin D, Ca [41]

2. **Biomarkers of immune activation and inflammation** which could include- (CD38+CD8+ and CD4+T cells), inflammatory cytokines (IL-1b, IL-6, MCP-1, TNF), endothelial activation (sICAM-1, sVCAM-1) and the acute phase reactant hsCRP. [39-40]We are currently conducting a systematic review of the published literature to inform the optimal measures... Panels of the most appropriate measures will be conducted at study end.

The second major reported toxicity of TDF is renal insufficiency [42-3]**.** As improvements are reported with TAF, **measures of renal glomerular and tubular function** are included as secondary outcomes. TDF has a more favourable lipid profile than TAF, and we will also assess **lipid parameters and cardiovascular risk scores**.

6.0 ELIGIBILITY CRITERIA

6.1 Inclusion Criteria – all criteria must be met

1. Biological female aged 40 – 60
2. Documented HIV-1 infection **by means of any <u>one</u> of the following**:
    i. **Documentation of HIV diagnosis in the medical record by a licensed health care provider;**

    ii. **OR HIV-1 RNA detection by a licensed HIV-1 RNA assay demonstrating >1000 RNA copies/mL;**

    iii. **OR any licensed HIV screening antibody and/or HIV antibody/antigen combination assay confirmed by a second licensed HIV assay such as a HIV-1 Western blot confirmation or HIV rapid Multispot antibody differentiation assay.**

3. Peri-menopausal or within 10 years of menopause (based on history) history).
4. Signed Informed Consent Form and willing to comply with the protocol.
5. Receiving a cART regimen containing a ritonavir boosted PI (darunavir, atazanavir, lopinavir,) or an NNRTI (efavirenz, nevirapine or rilpivirine) or an integrase inhibitor (dolutegravir, raltegravir or elvitegravir) in combination with TDF-FTC for > 24 weeks.
    i. Participants on a single tablet regimen containing TDF/FTC are able to participate as long as they meet the virologic suppression criteria.
        1. For those who are on elvitegravir/cobicistat/tenofovir disoproxil fumarate/emtricitabine (EVG/c/TDF/FTC), they will either remain on their current regimen or switch to elvitegravir/cobicistat/tenofovir alafenamide/emtricitabine (EVG/c/F/TAF).

2. For those who are on rilpivirine/tenofovir disoproxil fumarate/emtricitabine (RPV/TDF/FTC), they will either remain on RPV/TDF/FTC or switch to rilpivirine/tenofovir alfenomide/emtricitabine (RPV/F/TAF).
3. For those on other single tablet regimens, if no component needs to be changed they will need to switch to TDF/FTC and the third agent in the STR as separate tablets prior to randomization. If the third agent needs to be changed they will need to switch to TDF/FTC plus the new third agent and show continued evidence of viral suppression for one month prior to randomization. For example: those on atripla would need to switch to TDF/FTC plus efavirenz and be randomized immediately or switch to TDF/FTC plus dolutegravir and remain suppressed for one month before randomization.

ii. Those who are on more than 3 agents are eligible provided they meet these criteria.

6. Stable viral suppression (plasma HIV-RNA<50 copies/mL for > 24weeks). Single viral blip <500 copies/mL allowed if re-suppresses.
7. If of childbearing potential, is using effective birth control methods and is willing to continue during the trial.
8. Women will be assessed for vitamin D and calcium dietary intake; if inadequate for age, supplements will be recommended.

6.2 Exclusion Criteria - if any are present the participant cannot be included

1. HIV-2
2. High 10-year fracture risk at screening (> 20%) based on country specific FRAX
3. Current treatment with active bone medications- bisphosphonates, denosumab, calcitonin, raloxifene, teriparatide, stontrium
4. Current use of systemic steroids (inhaled steroids permitted) or chemotherapeutic agents
5. Acute viral hepatitis
6. Chronic hepatitis C with AST and/or ALT ≥5 x ULN or expected to require HCV treatment during the trial period.
7. Any investigational ARV within 30 days.
8. Dialysis or renal insufficiency (creatinine clearance < 50ml/min)
9. History of decompensated liver disease (AST or ALT≥5x the upper limit of normal (ULN) or ALT ≥ 3 x ULN and bilirubin ≥ 1.5 x ULN with > 35% direct bilirubin), or the presence of ascites, encephalopathy, coagulopathy, hypoalbuminemia, esophageal or gastric varices.
10. Pregnant or breastfeeding

11. Screening blood result with any grade 3/4 toxicity according to Division of AIDS (DAIDS) grading scale, except: asymptomatic grade 3 amylase, CPK, or lipid elevation.
12. Any condition (including illicit drug use or alcohol abuse) or lab results which, in the investigator's opinion, interfere with assessments or completion of the trial.

7.0 STUDY EVALUATIONS AND PROCEDURES

7.1 Clinical Evaluations

Once a candidate for screening has been identified, study details will be carefully discussed with the potential participant. The participant will be asked to read and sign the approved informed consent form prior to any assessments being performed.

Medical history, documented HIV test results, and demographic information will be obtained at the screening visit. Data will be collected on age, race, HIV risk factor, smoking history, exercise habits, prior fractures, calcium and vitamin D intake, illicit drug use, alcohol, steroid use, ARV history, past medical illness, medications, menstrual history, obstetrical history, weight, body mass index. The FRAX calculator will be used to estimate 10 year fracture risk.

Routine investigations will include viral load, CD4, haematology, biochemistry, lipids, endocrinology, renal monitoring. These are investigations that are typically performed as part of standard of care at 3 month intervals and will not be done specifically for study purposes but the data will be collected and recorded. Hepatitis B and C status will be recorded.

A complete physical exam, including vital signs will also be performed at the screening visit. A brief physical exam, including vital signs, will be performed as per site local practices at subsequent visits. Information pertaining to medication review, and adverse events will be collected as indicated in Table 1 (Appendix A).

Information regarding the antiretroviral regimens including time of starting ART and type of drugs will be recorded. While on the study, ART switching, adherence to ART or stopping ART for any reasons will also be documented.

Sarcopenia will be determined with a combined definition of fat free mass measure (from the DXA) and muscle grip strength obtained with dynamometer

Short performance physical battery test (SPPB) will be performed to assess walking speed and balance.

7.2 Participant Identification Number Assignment

Participant Identification (ID) numbers will be assigned sequentially to each participant who is eligible to participate in the study. Participants who are not eligible for the study will not be assigned a Participant ID Number. The Participant ID Number will consist of a three digit centre number and a three digit number according to chronological order of entry at each centre. Details for assigning Participant ID Numbers will be provided in the Operations Manual. Log(s) should be completed by each site to capture all participants who have consented and who have been assigned a participant identification number. If a patient discontinues from the study the Participant ID Number will not be reused.

The Participant ID Numbers will be used for identification of participants in the source documentation, CRFs, and laboratory samples. This will ensure that participant data and laboratory samples leaving the study sites will be identified and tracked.

7.3 Imaging

Participants will complete DXA scans at the local site. Specific software will be used to calculate the trabecular bone score at selected sites. Participants enrolled at selected participating sites will undergo HRpQCT.

7.4 Laboratory Evaluations

Blood samples will be obtained for clinical laboratory evaluations on the days specified in Table 1 (Appendix A). Routine investigations will include HIV viral load, CD4 cell counts, complete blood count (CBC) with differential and measurements of hepatic (AST, ALT, ALP, GGT, bilirubin, albumin) and renal function (creatinine, GFR, urinalysis, urine albumin, Ur albumin/Cr, and Ur Protein/cr), urine and serum phosphate), calcium, glucose and lipid profiles (triglyceride, LDL, HDL, TC/HDL). These are investigations that are typically performed as part of standard of care at 3 month intervals and will not be done for study purposes but the data will be collected and recorded. Plasma HIV RNA levels will be measured at local laboratories with a lower limit of detection as per current assays. (This may be < 50/ml, < 40/ml or viral load not detected). Clinical laboratory evaluations for hematology, biochemistry, CD4/CD8, viral load and serology will be performed at site local laboratories as per local practices.

7.5 Stored Research Samples

Blood samples will be aliquoted, frozen and stored in order to perform biomarker testing at a later date. As this field is rapidly evolving, not all tests are currently known or developed but the frozen samples will be used only for HIV research purposes and testing related to this study. The samples will not be made available to any commercial enterprise. Stored samples will be identified only by a code number and will be destroyed after 10 years.

7.6 Adherence

For self-reports of adherence will use the Visual Analogue Scale[44] (VAS last week and/or last month) which measures item ratings by participants in percentile terms from 0 to 100% and using a descriptor, for subjective perception of adherence in the last day, week, two weeks, and month. We will also collect self-reported adherence with the HIV Adherence Treatment Scale (HATS)[45]. We will corroborate self-reported measure with objective outcome measurements such as returned pill counts.

7.7 Questionnaires (in Appendix)

English and Italian questionnaires

Frailty measured with some components from Frailty Phenotype

Quality of life using the EuroQoL EQ-5D [46]

Fall frequency, assessed using self-administered questionnaire validated in the 2008 National Health Interview Survey Dizziness and Balance Supplement[47]

7.8 Transportation of Laboratory Samples

Stored samples for biomarkers will be transported from all sites to the UHN laboratory of Dr Rupert Kaul. Details for sample shipment will be provided in the laboratory manual. All shipments will be prepared by qualified individuals ensuring that all shipments are packed and shipped according to IATA regulations for the shipment of infectious and dangerous goods.

All samples will be clearly labelled and must be accompanied with a copy of a shipment list. Prior to shipment of samples, the sites will notify the laboratory regarding the date of shipment to ensure receipt of the shipment.

8.0 STUDY VISITS

8.1 Screening visit:

The screening visit is anticipated to require 1-2 hours of the participant's time. During this visit the following will take place:

- Informed Consent
- Assign Participant ID Number
- Demographic Information
- Medical History
- Physical Exam

- Calcium and Vitamin D intake assessment
- FRAX Score determined by country specific FRAX calculator
- Framingham Cardiovascular Risk Score
- Vital Signs including height, weight and determination of BMI
- HIV Viral load (within 1 month of screening date)
- Serum Pregnancy test and FSH (if post-menopausal)
- Hepatitis B and C (only if not previously tested)
- Hematology and biochemistry: CBC with differential and liver profile (AST, ALT, serum creatinine, Urinalysis, GFR, Urine albumin/creatinine, urine protein/creatinine) (within 1 month of screening date)
- Medication Review
- Review of Eligibility Criteria
- Dexa scan will be scheduled before baseline visit. (Dexa scan should be completed prior to Randomization.).

8.2 Baseline Visit for Participants

Day 1- Week 0 (no more than 30 days after screening visit date)

This visit must occur within 30 days of the screening visit. (**Note:** If baseline visit is within 2 weeks of screening labwork, then available screening lab results may be used and **duplicate** lab work need not be recollected). This visit is anticipated to require 2 hours of the participant's time. A baseline visit should be scheduled if the eligibility criteria met, at which the following events will take place:

- Confirmation that eligibility criteria have been met
- Confirm Dexa Scan has been completed.
- Urine Pregnancy test (if premenopausal)
- Randomization
- Targeted Physical Exam
- Vital Signs
- Framingham Cardiovascular Risk Score
- Hematology and biochemistry: CBC with differential and liver profile (AST, ALT), serum creatinine, calcium, phosphate, Urinalysis, GFR,

Urine albumin/creatinine, urine protein/creatinine, lipids- cholesterol, LDL, HDL, TG, glucose)

- CD4/CD8
- HIV Viral Load
- Research Blood Sample
- Study Drug dispensation
- Questionnaires: all questionnaires/adherence scales should be completed to include, quality of life, frailty, and falls
- Medication review
- Trabecular bone score (TBS) will be determined from the screening DXA at selected participating sites
- Participants at selected participating sites will have HRpQCT scan
- Grip strength with the dynamometer
- Tests of balance and gait - balance tests, gait speed, chair stand

8.3 Follow-up Visits for Participants

These will occur at weeks 4, 12, 24, 36, 48, 52, 60, 72, 84 and 96. At weeks 4, 12, 24, 36, 52, 60, 72, and 84, the participants will have routine investigations as per the study site, and will complete targeted physical exams. **Complete** physical exams, labwork as outlined in the schedule of Assessments on pages 25 – 26, and FSH (if premenopausal) will be required at weeks 48 and 96. Adverse events will be captured at each study visit as will any changes in clinical conditions or medications. Pill counts of returned study drug except week 52 will also be captured. The accepted windows around study visits are +/- 7 days. The procedures will last about 60 minutes.

If they agree, participants will continue to be followed even if they decide to stop the study medication

At weeks 48 and 96 all participants will complete the study imaging, collection of plasma specimens for marker banking, quality of life questionnaires, FRAX score as determined by country specific FRAX calculator, Framingham cardiovascular risk score, and geriatric measures as above. The additional procedures will last

approximately 30 minutes plus imaging time. The imaging is to be scheduled within 4 weeks of these visit dates.

Drug dispensation will be done every 3 months during the regular visits at W0, 12, W24, W36, W48, W60, W72, and 84. At week 48, all participants will receive open label TAF/FTC.

8.4 Final Study Visit (Week 96 +/- 14 days)

The duration of the study is 2 years per participant. Participants will be recruited during the first year of the study and will be followed for 96 weeks. The final visit will be the participant's last follow-up visit and the following assessments will take place:

- Complete Physical Exam
- Vital Signs
- Hematology and biochemistry: CBC with differential and liver profile (AST, ALT), serum creatinine, calcium, phosphate, Urinalysis, GFR, Urine albumin/creatinine, urine protein/creatinine, lipids- cholesterol, LDL, HDL, TG, glucose. CD4/CD8
- Urine pregnancy test and FSH (if premenopausal)
- HIV Viral Load
- Research Blood Sample
- Questionnaires- quality of life, adherence, falls, balance
- Frax Score determined by country specific FRAX calculator
- Framingham Cardiovascular Risk Score
-
- Medication review: ART and Other Concomitant Medications
- Imaging as per study site: DXA, TBS, HRpQCT
- Geriatric assessment, with questionnaires (frailty, quality of life, balance, falls), dynamometer, performance walking test.

To collect any information on potential adverse event reaction, patients will be called 4 weeks after their last visit at 96 weeks +/- 7 days.

8.5 Early Termination

If a participant withdraws from the study early, documentation for reason of early termination is required. Investigations will be completed as per final visit.

8.6 Re-contact of Participants after Study Termination

Study results will be made known to the site investigators. As per site practices and IRB/REB requirements, site investigators will be responsible for disseminating the information to participants who participated in the study.

8.7 Schedule of Procedures

Study Weeks		**W0**	**W4**	**W12**	**W24**	**W36**	**W48**	**W52**	**W60**	**W72**	**W84**	**W96/End of Study**	**W100**
Study Visits	Screening (-30 days)	Baseline (screening + 30 days											
Visit Duration	1-2 hours	2 hours	1 hour	1 hour	1 hour	1 hour	1-2 hours	1 hour	1 hour	1 hour	1 hour	1-2 hours	Phone call
Informed Consent Form	X												
Assign Subject ID number	X												
Randomization		X											
Demographics	X												
FRAX and Framingham Score	X						X					X	
Medical History	X												
Physical Exam[1] Signs/ Symptoms	X (complete)	X	X	X	X	X	X (complete)	X	X	X	X	X (complete)	
Vital Signs including height, weight and determination of BMI	X	X	X	X	X	X	X	X	X	X	X	X	
Pregnancy test (if premenopausal), FSH	X[2]	X	X	X	X	X	X[2]	X	X	X	X	X[2]	
HIV viral load [3]	X	X[3]	X[3]	X	**X**	**X**	X[3]	X[3]	**X**	**X**	**X**	X[3]	
CD4/CD8 count[3]		X[3]		X	**X**	**X**	X[3]	X	**X**	**X**	**X**	X[3]	
Hematology and biochemistry: CBC with differential and liver profile (AST, ALT, serum creatinine, calcium, phosphate, Urinalysis, GFR, Urine albumin/creatinine)[3]	*X	X[3]	X	X	X	X	X[3]	X	X	X	X	X[3]	
HBsAg, anti-HBcAb, HBsAb, HCV antibody[4]	X[4]												

Study Weeks		**W0**	**W4**	**W12**	**W24**	**W36**	**W48**	**W52**	**W60**	**W72**	**W84**	**W96/End of Study**	**W100**
Fasting glucose/cholesterol[3]		X[3]					X[3]					X[3]	
Research blood sample		X					X					X	
Study Drug dispensation		X		X	X	X	X		X	X	X	X	
Study Drug Accountability (pill counts)			X	X	X	X	X		X	X	X	X	
Questionnaires/assessments (calcium & vitamin D[5], quality of life, adherence, frailty, falls, balance)	X[5]	X					X					X	
Concomitant Medication review	X	X	X	X	X	X	X	X	X	X	X	X	
Dexa scan[6]	X[6]						X					X	
Trabecular bone score determined from DXA (**participating sites)**		X					X					X	
HRQCT scan (**participating sites**)		X					X					X	
Grip strength with dynamometer		X					X					X	
Test of balance and gait-balance tests, gait speed, chair stand		X					X					X	
Capture adverse events or changes in clinical conditions/medications		X	X	X	X	X	X	X	X	X	X	X	X

[1] Targeted physical exam except at Screening, Weeks 48 and 96/end of study

[2] Serum pregnancy and FSH for all participants at screening. Urine pregnancy at all other visits, and FSH at Weeks 48 and 96/end of study **for pre-menopausal women only**

[3] Screening lab results may be within 1 month of screening visit. Lab collection and results as per site Standard of Care, except at Weeks 0, 48 and 96 (end of study) where lab collection and results are required. Repeat viral load and safety assessments should be done at Weeks 4 and 52.

[4] HBV and HCV status to be tested at screening **only** if **NOT** previously done

[5] Calcium & Vitamin D assessment should be done at Screening. All other questionnaires at Weeks 0, 48 & 96/end of study

[6] Baseline Dexa Scan should be completed prior to baseline visit

9.0. STUDY DRUG

Participants will take a single tablet of FTC/TDF (300/200mg) or FTC/TAF (200/**10** mg) if the third agent is a ritonavir boosted protease inhibitor, or 200mg/**25** mg if the third antiretroviral agent is an NNRTI, raltegravir or dolutegravir) [24] once daily orally with the recommended dose of the co-administered third agent. For those previously on EVG/c/TDF/FTC or RPV/TDF/FTC, you will continue to receive EVG/c/TDF/FTC or RPV/TDF/FTC, or switch to EVG/c/F/TAF or RPV/F/TAF.

After 48 weeks all participants will take a single tablet of FTC/TAF (200mg/10 mg if the third agent is a ritonavir boosted protease inhibitor or 200mg/25 mg if the third agent is an NNRTI, raltegravir or dolutegravir), EVG/c/F/TAF, or RPV/F/TAF once daily.

Study drug will be donated by Gilead. Study drug will be packaged in bottles containing 30 tablets, silica gel desiccant and polyester packing material. Each bottle will be enclosed with a polypropylene screw cap. To ensure stability the drug should not be stored in any other container than that supplied. The study drug bottles will be labelled to meet all the applicable requirements of the EU guidelines to good manufacturing practice, Health Canada and the US food and drug administration. The bottles should be stored at a controlled room temperature of 25C (77F) with excursions permitted to between 15C and 30C). Storage conditions will be specified on the labels.

Until dispensed to participants, all bottles of study drug will be stored in a securely locked area, accessible only to authorized study personnel.

The third ARV agent of the participants pre-existing regimen should be taken and stored according to the approved product labeling for each particular component.

9.1 Study Drug Accountability

The investigator at each site is responsible to ensure adequate accountability of all used and unused study products. The investigator or designated central pharmacy will acknowledge receipt of the study drugs from Gilead after reviewing the shipment's content and condition. Study sites will acknowledge receipt of study drugs from the UHN Central Pharmacy after reviewing the contents and condition of the shipment. The investigator or designee will be responsible for maintaining an accurate inventory of the dates and quantities of all study drugs received, dispensed and returned. Each dose of the study drug administered by the qualified study centre personnel will be accurately recorded on logs which indicate the date and quantity of all doses dispensed. These logs will be available for inspection by study monitors. Study medication supplies must be accounted for by the study monitor prior to destruction or return.

10. EARLY TERMINATION

Participation in the study will be terminated early if: the participant refuses further treatment and/or follow-up evaluations; the study staff or participant's medical provider determines that further participation in the study would be detrimental to the participant's health or well-being; the participant is non-compliant with the study requirements in a manner that is either detrimental to her health or interferes with the validity of the study results; the patient has viral failure and requires an alternative antiretroviral regimen (the viral load that requires a change in therapy will be at discretion of the investigator), or the sponsor terminates the study.

11. ASSESSMENT OF SAFETY

11.1 Adverse Events (AEs)

Adverse events (AEs) are defined as any untoward medical occurrence in a participant that does not necessarily have a causal relationship with the study medication/intervention. An AE can therefore be any unfavourable and unintended sign (including abnormal laboratory finding), symptom, or disease temporally associated with the use of study drug/intervention, whether or not it is related to the medication/intervention.

Toxicities will be graded on a 4-point scale according to standard guidelines outlined in the "Division of AIDS Table for Grading the Severity of Adult and Pediatric Adverse Events" [48][Appendix C]. In general, grade 1 and 2 events will not be considered adverse events, but details should be documented in the source documents. Fractures will be considered and captured as an adverse event regardless of grading.

Stable chronic conditions which are present prior to clinical study entry and do not worsen are not considered adverse events and will be accounted for in the participant's medical history.

Participants who have an adverse event should be followed and treated by the site Investigator until the abnormal parameter or symptom has resolved or stabilized. It is up to the clinician to determine that the AE is either resolved or that is has reached a stable state, after which no further follow-up is necessary. There should also be source documentation to support this determination.

Rates of adverse events, serious adverse events and hospitalisations will be compared between the two arms. Specific attention will be paid to HIV RNA (see below), CD4 cell counts, and metabolic parameters such as fasting glucose, and lipid profiles.

The most common adverse reactions of moderate to severe intensity and incidence ≥2% in those receiving TAF are headache, fatigue, nasal congestion and diarrhea.

11.2 Recording/ Documentation of AEs

At each contact with the participant, information regarding adverse events will be elicited by appropriate questioning and examinations, and will be recorded immediately on a source

document (e.g., progress notes, laboratory reports, survey tools, and data collection tools). The start date, stop date, severity of each reportable event, and the PI's judgment of the AE's relationship to the study medication/intervention will also be recorded in the case report form (CRF).

11.3 Serious Adverse Events (SAEs)

A Serious Adverse Event is defined as an AE meeting one of the following:

- Death during the period of protocol-defined surveillance
- Life Threatening Event (immediate risk of death at the time of the event)
- In-patient hospitalization or prolongation of existing hospitalization during the period of protocol-defined surveillance
- Results in congenital anomaly or birth defect
- Results in a persistent or significant disability/incapacity
- Any other important medical event that may not result in one of the above outcomes may be considered a serious adverse event when: based upon appropriate medical judgment, the event may jeopardize the participant, and may require medical or surgical intervention to prevent one of the outcomes listed above. Examples of such medical events include allergic bronchospasm requiring intensive treatment in an emergency room or at home, blood dyscrasias, or convulsions that do not result in in-patient hospitalization, or the development of drug dependency or drug abuse or any event of possible drug-induced liver injury with hyperbilirubinemia defined as ALT>3xULN and bilirubin >2xULN (>35% direct)

Any AE that occurs between baseline and 30 days following the last study visit at Week 96 (or at the time of early discontinuation of the participant from the study for any reason) is to be recorded in the source documentation (patient will be called). Surveillance for AEs will end at 30 days following Week 96. If an SAE is ongoing at the time a participant discontinues/completes the trial the SAE will be followed until the Investigator agrees that the event is satisfactorily resolved, becomes chronic, or that no further follow-up is required.

11.4 AE/SAE Grading, Relationship Assignment and Reporting Procedures

11.4.1 AE/SAE Grading

The intensity (severity) of each adverse/serious event will be graded according to the toxicity table and toxicity guidelines provided by the Division of AIDS (DAIDS) Table for Grading the Severity of Adult and Pediatric Adverse Events. The DAIDS Table will be provided with the Operations Manual.

11.4.2 Relationship Assessment

For all collected AEs (including SAEs), the clinician who examines and evaluates the participant will determine the AE's causality based on temporal relationship and his/her clinical judgment. The degree of certainty about causality will be graded using the categories below:

Definitely Related: There is clear evidence to suggest a causal relationship, and other possible contributing factors can be ruled out. The clinical event, including an abnormal laboratory test result, occurs in a plausible time relationship to drug administration and cannot be explained by concurrent disease or other drugs or chemicals. The response to withdrawal of the drug (de-challenge) should be clinically plausible. The event must be pharmacologically or phenomenologically definitive, with use of a satisfactory re-challenge procedure if necessary.

Probably Related: There is evidence to suggest a causal relationship, and the influence of other factors is unlikely. The clinical event, including an abnormal laboratory test result, occurs within a reasonable time sequence to administration of the drug, is unlikely to be attributed to concurrent disease or other drugs or chemicals, and follows a clinically reasonable response on withdrawal (de-challenge). Re-challenge information is not required to fulfill this definition.

Possibly Related: There is some evidence to suggest a causal relationship (e.g., the event occurred within a reasonable time after administration of the trial medication). However, the influence of other factors may have contributed to the event (e.g., the participant's clinical condition, other concomitant events). Although an adverse drug event may rate only as "possibly related" soon after discovery, it can be flagged as requiring more information and later be upgraded to "probably related" or "definitely related", as appropriate.

Unlikely: A clinical event, including an abnormal laboratory test result, whose temporal relationship to drug administration makes a causal relationship improbable (e.g., the event did not occur within a reasonable time after administration of the trial medication) and in which other drugs or chemicals or underlying disease provides plausible explanations (e.g., the participant's clinical condition, other concomitant treatments).

Not related: The AE is completely independent of study drug administration, and/or evidence exists that the event is definitely related to another etiology. There must be an alternative, definitive etiology documented by the clinician.

11.4.3 SAE Reporting Procedures

The Investigator or designee must report any SAE occurring in a patient receiving study drug to the Sponsor within 24 hours, even if the SAE is not drug-related. This should be done by telephone and by sending a faxed or emailed copy of the Serious Adverse Event form plus other supporting documentation, as required.

SAEs will be reported to:

Immunodeficiency Clinic

University Health Network

Attention: Rosemarie Clarke, Research Nurse Manager

Phone: 416-340-4800 ext. 6723 or 416-340-5077
Fax: 416-340-9105

E-mail: rosemarie.clarke@uhn.ca

All additional follow-up evaluations must also be reported as soon as possible in the same manner. All SAEs will be followed until the Investigator agrees that the event is satisfactorily resolved, becomes chronic, or that no further follow-up is required.

The Sponsor or designee will be responsible for notifying the relevant authorities of any reportable SAE as outlined in the Canadian Food and Drug Regulations and ICH Guidelines.

The Sponsor will also ensure that other participating Investigators are notified of any SAEs. The Investigator is responsible for ensuring that their local Research Ethics Board (REB) is notified of any SAEs.

11.5 AEs/SAEs monitoring

Any AE/SAE that occurs between baseline and the time the participant departs the study at the end of the final follow-up visit (or at the time of early discontinuation of the participant from the study for any reason) will be captured and recorded in the source documentation. At each contact with the participant, the investigator (or designate) must seek information on adverse events by specific questioning and, as appropriate, by examination.

Adverse events that had previously been reported by the study participant will also be reassessed for duration, intensity and possible reoccurrence. Assessment of safety will include clinical observation and monitoring of routine hematology and biochemistry.

Surveillance for AEs will end 30 days post Week 96 (or at the time of early discontinuation of the participant from the study for any reason). If an SAE is ongoing at the time a participant discontinues/completes the study, the SAE will be followed up until the Investigator agrees that the event is satisfactorily resolved, becomes chronic, or that no further follow-up is required.

11.6 Toxicities/ Adverse Event Management

In the event of a discontinuation of TAF/FTC for suspected renal impairment, or other clinically significant chemistry elevations, participants should not restart TDF/FTC or TAF/FTC due to the risk of a recurrent reaction; such participants should be withdrawn from study and seek/be reviewed for alternative antiretroviral therapy. If they agree, participants will continue to be followed for the duration of the study.

11.7 Management of HIV Virologic failure.

Virologic failure must be considered for any patient with plasma HIV RNA ≥50 c/mL at any time following randomization. For the purposes of clinical management in this study, virologic failure is defined as:

Two consecutive plasma HIV RNA ≥50 c/mL within four weeks of the initial suspected virologic failure.

Cases of confirmed virologic failure will trigger genotypic virologic resistance testing, including integrase resistance testing, when plasma HIV RNA >400c/mL. The virologic failure definition is intended for guidance and Investigators should use their discretion as to the most appropriate clinical management of their patients. The choice of antiretroviral therapy will be left to the discretion of the treating physician. In some cases, e.g. if there is no evidence of acquired resistance to the current regimen, treatment may be continued with increased efforts at adherence support.

11.8 Pregnancy Management

As this study is enrolling peri and post-menopausal women the risk of pregnancy is extremely low.

11.9 Action to be taken if pregnancy occurs

Any participant who becomes pregnant during the study must be immediately withdrawn from TAF/FTC to eliminate further exposure to the embryo/foetus.

11.10 Pregnancy Reporting

If pregnancy occurs during the period of protocol-defined surveillance, while it is not considered an adverse event or serious adverse event requires monitoring and follow up. The investigator must report the pregnancy information for trial participants using a pregnancy CRF. The pregnancy should be followed up by the investigator until delivery. To ensure participant safety, each pregnancy must be reported within one week of learning of its occurrence. The pregnancy must be followed up to determine outcome (including premature termination) and status of mother and child, which must also be reported using a pregnancy outcome CRF. Pregnancy complications and elective terminations for medical reasons must be reported as an AE or SAE. Spontaneous abortions must be reported as an SAE.

Any SAE occurring in association with a pregnancy brought to the investigator's attention after the participant has completed the study and considered by the investigator as possibly related to the investigational product, must be promptly reported.

[Investigators are also encouraged to report any pregnancy that occurs during study participation to the Antiretroviral Pregnancy Registry. More information including copies of applicable CRFs and fax numbers are available at www.apregistry.com.]

12. TRIAL MANAGEMENT

12.1 Local Coordination

Each collaborating investigator will be responsible for recruiting study personnel, obtaining local REB/IRB approval, ensuring local clinic staff are informed about the study, and supervising local study activities. The study personnel at the site will be responsible for: identifying participants; obtaining informed consent; conducting study visits; ensuring participants have regular follow-up; encouraging attendance at visits; telephoning participants if visits are missed; and collecting participant data. A study manual will outline methods for CRF completion and record management. Personnel will also be trained on study procedures prior to and during the study. The informed consent forms and study questionnaires will be translated into Italian and French.

12.2 Central Coordination

A Project Manager will coordinate the day-to-day management of the study. A computerized study database will be created by an appointed Data Manager and managed at the data management centre at the CIHR Canadian HIV Trials Network. Electronic CRFs will be used.

12.3 Communication

Regular investigator meetings will be planned by teleconference to keep study personnel informed of study progress, and to encourage ongoing recruitment. Brief updates will be provided to meet enrolment targets. The information on criteria for study participation will be posted on the websites of the different clinical centres.

13.0 DETAILS OF STATISTICAL ANALYSIS

13.1. Sample size considerations

128 women, 64 per group, are required to detect a difference of 1% in the annual change in spine BMD at 48 weeks between those randomized to immediate switch to TAF/FTC vs those randomized to delayed switch to TAF/FTC with a two sample t test, assuming a standard deviation (SD) of 2%, 80% power and a significance level of .05. The difference of 1% is believed to be a clinically important difference to detect. The expected rate of loss in BMD in peri- and post-menopausal women is 1-3% per year.

The SD in the % change in BMD was estimated from several studies of BMD in HIV+ individuals. Yin et al [49] compared longitudinal changes in BMD in pre and post-menopausal HIV+ and uninfected women in two separate cohorts. In the premenopausal period annual % decrease in BMD was not different in HIV+ and HIV- at the lumbar spine (LS) (-0.8 +/-2.1% vs 0.4 +/-1.9%, p=.20) and remained similar after adjusting for traditional risk factors. In the postmenopausal period annualized rates of bone loss adjusted for baseline BMD were higher in HIV+ women by 2.4 fold at the LS (-1.2 vs -0.5 %, p<.00009). In a randomized study, where participants were 90% men, mean age 40, switching from TDF to TAF containing regimens

showed a statistically significant increase in BMD (median(IQR) + 1.79 % (-.22%, 3.38%)vs 0.28% loss(-2.25,1.5%), p<.001).[21] Another study (85 % men, median age 49) showed a mean increase in BMD of 1.5% at 48 weeks with a switch from TDF to TAF compared to a .2% loss in those continuing TDF. [29]A third study in persons with impaired renal function switching from a TDF to TAF containing regimen resulted in a mean increase in BMD of 2.29% at the spine and 1.47% increase at the hip.[22]

13.2 Primary analysis:

Generalized linear mixed models (GLMM) will be used to estimate the effect of immediate vs delayed switch from TDF/FTC to TAF/FTC on the percent change in BMD at the spine at 48 weeks.

In the GLMM model of percent change in BMD at the spine, each patient shall contribute two data points: the percent change in BMD at the spine at 48 weeks and the percent change in BMD at the spine at 96 weeks. A variogram will be used to determine the amount of error due to random effects and serial correlation. If random effects are the source of a significant amount of variation, terms for random intercept and random time shall be included in the model. If serial correlation is determined to be the main source of error, a single parameter will be required to model the correlation between the week 48 and week 96 values from a single patient, since there will be only two BMD values per patient after enrolment.

The model shall contain four covariates: (1) treatment arm, (2) a binary indicator for whether or not the data point corresponds to week 96, (3) the interaction between the treatment arm and the 96 week indicator and (4) a binary for country (Canada vs Italy). If sufficient woman are receiving hormone replacement therapy, an additional binary covariate may be included to capture whether or not the woman was receiving hormone replacement therapy at randomization.

The GLMM model can be expressed as follows:

$$Y = \beta_0 + \beta_1 \text{ treatment} + \beta_2 \text{ week 96} + \beta_3 \text{ treatment* week 96} \quad (1)$$

where Y is the percent change in BMD at the spine.

The hypothesis test for the primary analysis is H_0: $\beta_1 = 0$, which corresponds to the primary hypothesis of the effect of switching from TDF to TAF on percent change in BMD at the spine at 48 weeks.

13.3 Secondary analyses:

The secondary endpoints of greatest importance are: the percent change in BMD at the hip at week 48, the percent change in BMD at the hip at week 96, and the percentage change at the spine at week 96. GLMM will be used to estimate the effect of immediate switch to TAF/FTC on these outcomes

The hypothesis test for the secondary analysis is H_0: $\beta_3 = 0$ in model (1) above, which corresponds to the secondary hypothesis of the effect of switching from TDF to TAF on percent change in BMD at the spine at 96 weeks.

A similar model will be fit for the outcome of percent change in the spine. In that model, the hypothesis tests of H_0: $\beta_1 = 0$ and H_0: $\beta_3 = 0$ will both be of interest, corresponding to the secondary hypotheses of the effects of switching from TDF to TAF on percent change in BMD at the hip at 48 and 96 weeks, respectively.

Interactions terms will be used to determine if the effect of switching from TDF/FTC to TAF/FTC on 48 week endpoints varies between peri and post–menopausal women or according to whether a protease inhibitor is used as the third ARV.

Other secondary endpoints such as the change in TBS at the hip, the proportions of patients with viral load < 50 copies/mL and the mean eGFR at 48 and 96 weeks will be summarized over time by treatment group to provide preliminary information but will not be compared formally to avoid type 1 errors due to multiple comparisons.

13.4. Interim analysis: no interim analyses are planned

13.5 Feasibility: The number of women on TDF/FTC aged 45-55 at UHN, MLMC, Modena and Milan are 48, 114, 105 and 80 respectively, for a total of 347. We expect that 90% (n=312) will be eligible and of those, that 80**%** (n=250) women would agree to participate. In addition each of the additional four Canadian sites have agreed to recruit between 5-10 participants. Therefore, it should thus be feasible to recruit the required 128 women within 12 months.

13.6 Timeline: 6 months for start- up and ethics approval, 12 months for enrollment; 24 months for follow-up; 6 months for analysis/write-up

13.7. Loss to follow-up: We expect that 10% of randomized patients will drop out before 48 weeks. Primary analyses will be conducted assuming data are missing at random. Pattern mixture models and selection models will be used as sensitivity analyses to determine the range of possible treatment effects under the assumption of not missing at random.[50]

14.0 Quality control and quality assurance

Checks will be built into the database to alert the data manager to inconsistent data at the time of data entry. Site personnel will then be prompted to double-check the data. Source documentation will not be verified by an on-site monitor visit during the study. It is the responsibility of the Sponsor Investigator with the CIHR Canadian HIV Trials Network Data Management Centre to assure the quality of computerized data for this study.

15. ETHICS AND PROTECTION OF HUMAN PARTICIPANTS

15.1 ICH Guidance E6:

Good Clinical Practice: Consolidating Guideline/ Declaration of Helsinki. The conduct of this study will conform to the International Conference for Harmonization and Good Clinical Practice (ICH-GCP) regulations and guidelines and the current revision of the Declaration of Helsinki.

15.2 Research Ethics Board/ Institutional Review Board

A copy of the protocol (including protocol amendments), all versions of the informed consents, other information to be completed by participants such as questionnaires, and any proposed advertising/ recruitment materials must be reviewed and approved by the REB/IRB of each participating centre prior to implementation of the study. The site investigator will be responsible for obtaining REB/IRB approval of the annual Continuing Review throughout the duration of the study. The site investigator will notify the REB/IRB of violations from the protocol and serious adverse events.

15.3 Informed consent process

All participants will be given detailed oral and written information about the study. Consent forms describing in detail the study medications/intervention(s), study procedures, anticipated benefits and potential risks will be given to each participant and written documentation of informed consent is required prior to starting the study. Participants must voluntarily sign and date an informed consent document that has been approved by a participating centre's REB/IRB prior to any procedures being done specifically for the trial. Each participant should have sufficient opportunity to discuss the study and consider the information in the consent process prior to agreeing to participate. Participants may withdraw consent at any time during the course of the trial. The informed consent will be signed and dated by the participant, the person who conducted the informed consent discussion and the investigator. The original signed informed consent form will be retained in the participant's study files and a copy will be provided to the subject.

15.4 Subject confidentiality

All participant related information including the CRFs, laboratory samples, evaluation forms, etc. will be kept strictly confidential. All records will be kept in a secure, locked location and only research staff will have access to the records. Participants will be identified only by means of a coded number specific to each participant, and a participant letter code. All computerized databases will identify participants by numeric codes only, and will be password protected. Upon request, clinical information may be reviewed by or released to study monitors, auditors, or regulatory agencies.

15.5 Early Termination of the Protocol

The REB/IRB or other government organizations, as part of their duties to ensure that research participants are protected may discontinue the study at any time. Regulatory authorities and the study Sponsor retain the authority to suspend additional enrollment for the entire study as applicable.

15.6 Record Retention

The Investigator will maintain all study records according to the ICH-GCP and applicable regulatory requirement(s). Records will be retained for 25 years, in accordance with applicable regulatory requirement(s). If the Investigator withdraws from the responsibility of keeping the study records, custody must be transferred to a person willing to accept the responsibility and the Sponsor notified. The Investigator should ensure that no destruction of medical records occurs without the written approval of the Sponsor.

Samples collected and sent to the central laboratory will be retained for 10 years, after which they will be destroyed.

16.0 PROTOCOL DEVIATIONS

No deviations from this protocol will be permitted without the prior written approval of the Sponsor, except when the modification is needed to eliminate an immediate hazard or hazards to participants. Any deviations that may affect a participant's treatment or informed consent, especially those increasing potential risks, must receive prior approval from the REB unless performed to remove an immediate safety risk to the participants. In this case it will be reported to the REB and the Sponsor immediately thereafter. Any departures from the protocol must be documented.

17.0 STUDY CONDUCT AND MONITORING

The study site agrees to allow monitors direct access to the study records and medical records from those participants enrolled in the clinical study as well as drug accountability records. Monitoring for this study will consist of data management review focused on erroneous data, illogical entries, and missing data and central monitoring of clinical and operational data reviewed for outliers and trends. On-site monitoring will be targeted based on results of the data management and central monitoring reviews as necessary.

18.0 DATA MANAGEMENT RESPONSIBILITIES

Instructions concerning the recording of study data on case report forms will be provided by the CIHR Canadian HIV Trials Network. Each study site is responsible for submitting the data in a timely fashion. It is the responsibility of the Data Management Centre to assure the quality of computerized data for this study. This role extends from protocol development to generation of the final study databases.

19.0 SOURCE DOCUMENTS AND ACCESS TO SOURCE DATA DOCUMENTS

Each participating site must maintain appropriate medical and research records for this study and regulatory/ institutional requirements for the protection of confidentiality of study participants. The Principal Investigator is responsible for assuring that the data collected are complete, accurate, and recorded in a timely manner.

Study data will be collected on study specific CRFs. Source documentation should support the data collected on the CRFs. Source documents include all recordings of observations or notations of clinical activities and all reports and records necessary for the evaluation and reconstruction of the clinical trial. Data for CRFs will be collected during participant visits, phone calls with participants and health care providers, completed questionnaires and abstracted from the participant's medical records. It is not acceptable for the CRF to be the only record of participation in the trial.

Clear and detailed instruction explaining CRF completion and where, when and how CRFs are to be sent to the data centre will be included in the study Operations Manual.

20. DISCLOSURE AND PUBLICATION POLICY

Publication of the final study report is planned. The Sponsor-Investigator (Dr. Walmsley) will determine authorship for each manuscript based on contributions to the study design, study execution, and manuscript completion. No author will be included without prior authorization but the intention is to be broadly inclusive of all study investigators who make active contributions as outlined by the ICJME criteria for authorship (Authorship credit should be based on 1) substantial contributions to conception and design, acquisition of data, or analysis and interpretation of data; 2) drafting the article or revising it critically for important intellectual content; and 3) final approval of the version to be published. Authors should meet conditions 1, 2, and 3).

21. STUDY TEAM ROLES AND RESPONSIBILITIES

Dr. Walmsley, MD is a clinician investigator with a strong and sustained (over 25 year) track record in the conduct of multicentre RCT in clinic HIV management. Her interest and knowledge translation in the field of HIV, aging and women, and her clinical practice which includes HIV infected aging women on TDF will strengthen recruitment and interest in this study. She directs a clinical research team who will conduct this study at the Toronto site. The team has participated in over 150 HIV related studies and have excellent recruitment and retention records. She has collaborations with HIV immunologist (Dr Kaul) for completion of the inflammatory marker studies.

Dr. Guaraldi, MD is a clinician investigator, with extensive expertise, interest and experience in HIV and women and aging. He has developed and oversees the Modena HIV Metabolic clinic (MHMC) data base which includes 680 HIV+ women, including 131 women who he has evaluated in the pre-menopausal, transition, early, and late menopausal period for a total of 1577 person-year follow-up. Approximately 60% of these women are on TDF containing regimens. Analysis of this data provided the framework for this study. He has expertise in conducting clinical trials and will supervise the participation of subjects in Italy. He has interest and has published on the aging and frailty measures that will be incorporated into this study. He will also supervise the development, maintenance and cleaning of the database for this study. In order to ensure timely recruitment to the study, **Dr Castagna, MD** who has a large population of HIV infected woman in Milan has agreed to serve as an enrolling centre for this study. Her site will be supervised by Dr. Guaraldi.

Dr. Mona Loutfy, MD is an infectious diseases and HIV clinician scientist who has many ongoing studies and cohorts addressing issues important to HIV infected women. In addition to her clinical trial expertise she has many collaborations with the infected community in Toronto and a strong focus and record in KTE activities. She will refer patients to the Toronto site, assist in the interpretation of the data, and direct the KTE resulting from this project.

Dr. Janet Raboud, PhD statistician holds a Chair in Biostatistics from the Ontario HIV Treatment Network. She has a long history of analysis of HIV research projects and clinical trials many in collaboration with Dr. Walmsley. She supervises a statistical team with content expertise who will be responsible for the analysis of this RCT.

Dr. Joel Singer, PhD statistician is a professor in the School of Population and Public Health at University of British Columbia, and has been the head of Methods and Statistics at the CIHR Canadian HIV Trial Network since its inception in 1990. He has a long history as a methodologist in HIV clinical trials as well as in other clinical disciplines. He has also served as member or chair of many data and safety monitoring committees

Dr. Angela Cheung, MD, is the head of the osteoporosis unit at the University Health Network. She has participated on developing the Canadian guidelines on this topic. She has expertise in

the newer imaging modalities. She has collaborated on previous HIV related projects with the team. She will advise on the imaging, bone biomarkers and outcomes.

All members of the team will collaborate on presentations and publications arising from this project.

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22. APPENDIX A

METHODOLOGY

During the study visit at Baseline and at week 48 and 96 a partial assessment will be performed using the following outcome measurements:

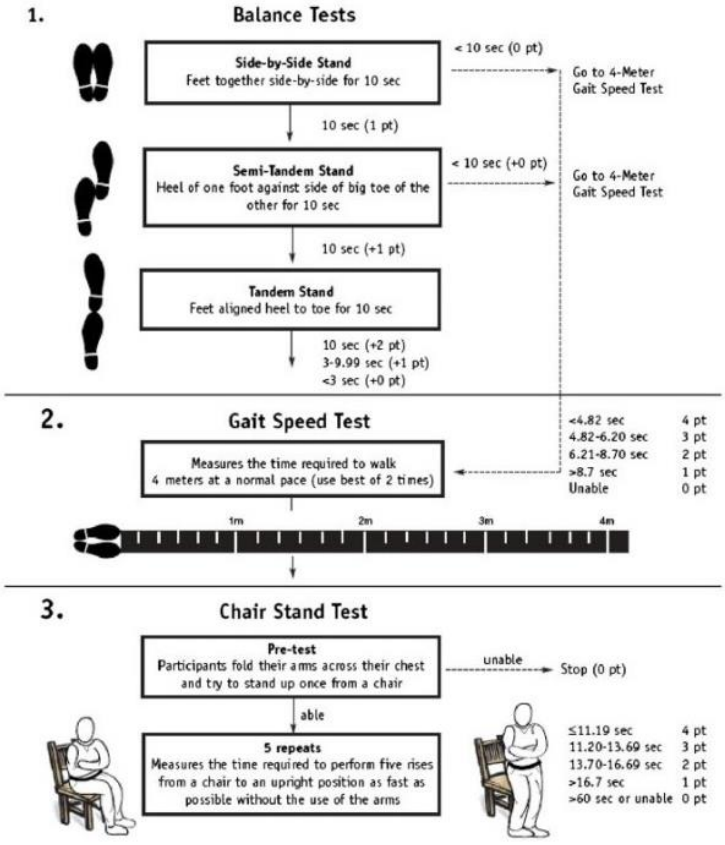
1. Physical Function.

   This is assessed using Short Performance Physical Battery (SPPB). This test consists of three components: balance, timed 4 m walk, and chair stands. The standing balance portion required participants to maintain a side-by-side, semi-tandem, and tandem stance for 10 seconds, with scores ranging from 0 to 4 (maximum score) [1].

   The fastest time of two 4 m usual-pace walk attempts was used. The chair stands required participants to rise from a chair with arms across their chest for five repetitions.

   A score of less than 9 is highly predictive for subsequent disability and was considered low function, 9–11 points moderate function, and 12 points (no deficits) high function.

   The figure represents test task and score system:



2. SARCOPENIA

Sarcopenia will be assessed using the following measurements and cut off:

1. Muscle mass using DEXA

Criterion	Measurement method	Cut-off points by gender	Reference group defined	Ref
Muscle mass	DXA	Skeletal muscle mass index (SMI) (Appendicular skeletal muscle mass/height^2) Men: 7.26 kg/m^2 Women: 5.5 kg/m^2	Based on 2 SD below mean of young adults (Rosetta Study)	[66]

2. Muscle strength using grip strength with handheld dynamometry using age- and gender specific cut-off reported in the table:

Criterion	Measurement method	Cut-off points by gender	Reference group defined	Ref
Muscle strength	Handgrip strength	Men: <30 kg Women: <20 kg	Based on statistical analysis of study group (n = 1,030)	[13]
		Men: BMI ≤ 24 ≤ 29 kg BMI 24.1–26 ≤ 30 kg BMI 26.1–28 ≤ 30 kg BMI > 28 ≤ 32 kg Women: BMI ≤ 23 ≤ 17 kg BMI 23.1–26 ≤ 17.3 kg BMI 26.1–29 ≤ 18 kg BMI > 29 ≤ 21 kg	Based on quartiles of study group (n = 5,317)	[27]

3. Performance, using SPPB (see above) with the following cut off points

Criterion	Measurement method	Cut-off points by gender	Reference group defined	Ref
Physical performance	SPPB	SPPB ≤8 SPPB 0–6 Low performance SPPB 7–9 Intermediate performance SPPB 10–12 High Performance	SPPB score is a summation of scores on three tests: Balance, Gait Speed and Chair Stand. Each test is weighted equally with scores between 0 and 4—quartiles generated from Established Populations for Epidemiologic Studies of the Elderly (EPESE) data (n = 6,534). The maximum score on the SPPB is 12	[62]

3. Frailty

Some components of frailty will be collected as follows:

The study data base will also embed most variables utilized in a **Frailty Index** (FI) based on available measures of age-related health deficits. Based on studies of HIV-negative older adults, standard criteria were established for candidate health deficits in a FI. It was determined that each deficit should be associated with adverse health outcomes and with age, have a prevalence of at least 1%, and

Unemployment	Self-report
High or low BMI	<18, 19 – 24, or >25 kg/m2
Abnormal white blood cell counts	<4000 cells/ml
Anemia	If female: <10 g/dl If male: <12 g/dl
Low platelets	<150 billion/l
Hepatitis C coinfection	Positive
Hepatitis B coinfection	Hepatitis B antigen positive
Elevated aspartate transaminase	>31 U/l
Elevated alanine transaminase	>31 U/l
Abnormal alkaline phosphatase	<38 or >126 U/l
Abnormal phosphorus	<2.5 or >5.1 mg/dl

Elevated total bilirubin	>1.10 mg/dl
Polypharmacy	>5 drug classes (excluding antiretroviral therapy)
Current CD4 cell count	<500 cells/ml
Nadir CD4 cell count	<200 cells/ml
HIV viral load	>40 copies/ml
CD4/CD8 cell ratio	<1.0
Duration of HIV infection	>10 years
Pre-HAART start	Start of antiretroviral therapy before 1 January 1997
History of AIDS	History of CDC category C HIV disease [42]
ART failure	History of viral load > 1000 copes/ml while on ART

4. Fall Frequency

The following questionnaire will be used to capture Fall Frequency

QLW0247(A5322)/01-31-14

ACTG A5322 FALL HISTORY QUESTIONNAIRE

NIAID AIDS CLINICAL TRIALS GROUP

Page 1 of 2

Patient Number | Date of Patient Visit/Contact mmm dd yyyy

Protocol Number A 5 3 2 2

Institution Code

Form Week | *Seq No. | **Step No. | Key Operator Code

* Enter a '1' if this is the first of this form for this date. Designate subsequent forms on the same date with a 2, 3, etc.
** Enter the subject's current study step number. Enter '1' if the study does not have multiple steps.

FOR OFFICE USE ONLY - TEAR OFF SHEET

SITE PERSONNEL INSTRUCTIONS:

The following interview asks the participant about falls that he/she may have experienced during his/her usual daily activities. **The interview should be conducted prior to the clinical exam and preferably in a quiet secluded area (e.g., exam room or other office). This form must be interviewer-administered.**

It is important to be familiar with the content and format of the interview before administering it to the study participants. At the visit, please begin by telling the participant:

"We are now going to ask you some questions about falls that may have happened during your usual daily activities. For the following questions, by "a fall" or "falling", we mean an unexpected event, including a slip or trip, in which you lost your balance and landed on the floor, ground or lower level, or hit an object like a table or chair. Falls that result from a major medical event (for example, a stroke, or seizure) or an overwhelming external hazard (for example, hit by a truck or pushed) should not be included."

The interview is very brief and should take no more than 10 minutes to complete. Complete the header prior to interviewing the participant.

INSTRUCTIONS TO THE INTERVIEWER:

PLEASE COMPLETE THE FOLLOWING ITEMS AFTER THE PARTICIPANT COMPLETES THE QUESTIONNAIRE OR AFTER YOU ASCERTAIN THAT THIS IS NOT POSSIBLE.

1. Was the interview completed? ..... (1-Yes, 2-No)
   If Yes, go to question 2.
   If No, complete 'a' and **STOP**.
   a. Indicate reason ..... 1-Participant declined
   2-Not enough time to complete form in clinic
   9-Other, specify
   Specify [70]:

2. Enter the language used to complete the form. Refer to Appendix 80 for Language Codes.
   Language:

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QLW0247(A5322)/01-31-14

ACTG A5322 FALL HISTORY QUESTIONNAIRE

NIAID AIDS CLINICAL TRIALS GROUP

Page 2 of 2

Patient Number ☐☐☐☐☐☐☐ Date of Patient Visit/Contact ☐☐☐ mmm ☐☐ dd ☐☐☐☐ yyyy

Protocol Number A 5 3 2 2 ☐ Institution Code ☐☐☐☐☐☐

Form Week ☐☐☐☐ * Seq. No. ☐☐ ** Step No. ☐☐ Key Operator Code ☐☐☐

* Enter a '1' if this is the first of this form for this date. Designate subsequent forms on the same date with a 2, 3, etc.
** Enter the subject's current study step number. Enter '1' if the study does not have multiple steps.

1. In the past 6 months, have you been concerned with losing your balance and falling while doing your usual daily activities? Would you say not at all, a little, quite a bit or very much? ☐
   1-Not at all
   2-A little
   3-Quite a bit
   4-Very much
   8-Unknown

   INTERVIEWER NOTE: Do not read 'Unknown' to participant.

2. In the past 6 months, have you had a fall? ☐
   1-Yes
   2-No
   8-Unknown
   If No or Unknown, STOP. This questionnaire is complete.
   If Yes, continue.

3. How many times have you fallen in the past 6 months? ☐
   1-(1) time
   2-(2) times
   3-(3-5) times
   4-More than 5 times
   8-Unknown

4. Did you seek medical attention after any of these falls (such as calling 911, visiting an on-site nurse, going to the emergency room or to a doctor's office)? ☐
   1-Yes
   2-No
   8-Unknown
   If No, STOP. This questionnaire is complete.
   If Yes or Unknown, continue.

   INTERVIEWER NOTE: Answer "No", if the participant did not actually see a medical provider (nurse, physician, paramedic, etc.) in-person. For example, if the participant asked a friend or neighbor for advice, or if the participant contacted a medical provider, but was not actually seen in the office, answer "No".

5. Did any of these falls result in a broken bone? ☐
   1-Yes
   2-No
   8-Unknown
   If Yes, report the fracture on the ACTG A5322 Focused Diagnosis, Progression and Treatment Event Report (DXW0037).

Thank you very much for completing this questionnaire.

01-31-14

Date Form Keyed (DO NOT KEY): _____/_____/_____

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