HIV-1 infection and antiretroviral therapies: risk factors for osteoporosis and bone fracture

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Abstract

Purpose of review—Patients with HIV-1 infection/AIDS are living longer due to the success of highly active antiretroviral therapy (HAART). However, serious metabolic complications including bone loss and fractures are becoming common. Understanding the root causes of bone loss and its potential implications for aging AIDS patients will be critical to the design of effective interventions to stem a tidal wave of fractures in a population chronically exposed to HAART.

Recent findings—Paradoxically, bone loss may occur not only due to HIV/AIDS but also as a consequence of HAART. The cause and mechanisms driving these distinct forms of bone loss, however, are complex and controversial. This review examines our current understanding of the underlying causes of HIV-1 and HAART-associated bone loss, and recent findings pertaining to the relevance of the immuno-skeletal interface in this process.

Summary—It is projected that by 2015 more than half of the HIV/AIDS population in the USA will be over the age of 50 and the synergy between HIV and/or HAART-related bone loss with age-associated bone loss could lead to a significant health threat. Aggressive antiresorptive therapy may be warranted in high-risk patients.

Keywords
AIDS; HIV; OPG; osteoclast; osteoporosis; RANKL

Introduction

The management of HIV-1 infection/AIDS with highly active antiretroviral therapy (HAART) has dramatically extended life expectancy. However, with increased longevity numerous metabolic complications have begun to emerge, including a loss of bone mineral density (BMD) in many patients leading to osteopenia, the forerunner of osteoporosis. Although low BMD has been recognized in HIV/AIDS patients for more than a decade [1] the large number of independent disease and lifestyle-related risk factors, and complex HAART combinations used in clinical practice, have generated considerable confusion as to the exact causes and appropriate interventions. Ultimately, it is now becoming clear that although bone loss in HIV populations likely stems from HIV infection/disease progression, paradoxically it may continue unabated during therapy, as a consequence of HAART. This
review examines current opinion in the field pertaining to the mechanisms and cause of bone loss in patients living with HIV/AIDS and the effects of chronic exposure to HAART.

**Bone loss directly associated with HIV-1 infection/disease progression**

Osteoporosis is a devastating disease of the skeleton that dramatically increases the risk of fracture. Bone fractures incur monumental healthcare costs to patients and society. Vertebral fractures have serious consequences, including severe back pain, and deformity. Hip fractures may cause prolonged or permanent disability and almost always require major surgery with high mortality rates (~30%) in the elderly [2], and up to 75% of survivors may require nursing home placement for long-term care [3].

One study has estimated that two of every three patients presenting with HIV infection have osteopenia (low bone mass) and a 3.7 higher odds of developing osteoporosis [4]. In a recent meta-analysis Paccou et al. [5•] concluded that there is a 15% prevalence of osteoporosis in HIV patients and osteopenia in 52%. However, whether bone loss stems from a direct effect of HIV-1 infection or as a consequence of traditional osteoporosis risk factors associated with patient lifestyle (smoking and alcohol consumption) and other AIDS-associated diseases such as muscle wasting, kidney disease, and hypogonadism has clouded interpretation. Furthermore, a high rate of deficiency/insufficiency of the bone active hormone vitamin D has been identified in Swiss HIV-positive patients [6•] and vitamin D deficiency has been associated with bone loss in US cohorts [7•]. Ultimately, bone loss is likely to be multifactorial, and teasing apart the effects of HIV-1 infection and/or disease progression relative to independent risk factors in humans remains challenging.

In an attempt to dissociate lifestyle, HAART, and other therapeutic factors from bone loss intrinsic to HIV-1 infection/disease progression we recently turned to an animal model of chronic HIV-1 infection, the HIV-1 transgenic rat. Expressing a constitutively active gag-pol-deleted HIV-1 provirus, this animal is a state-of-the-art model that develops numerous immunological and metabolic abnormalities synonymous with human AIDS [8].

We recently phenotyped the skeletons of these rats and report that like their human counterparts, HIV-1 transgenic rats undergo dramatic skeletal degeneration including significant loss of BMD and declines in cortical and trabecular bone volume, and architectural properties [9••], a consequence of elevated osteoclastic bone resorption. These studies ratified the HIV-1 transgenic rat as a suitable model for investigating the mechanisms responsible for bone loss associated with HIV-infection in humans.

In order to investigate pathological bone turnover, it must first be recognized that physiologically the skeleton is a highly dynamic organ that is continually regenerated by the process of homeostatic bone remodeling. Bone renewal involves the removal (resorption) of old worn bone by osteoclasts and resynthesis of new bone by osteoblasts [10]. Osteoclasts form from precursors which derive from the monocyctic lineage and express the surface molecule, receptor activator of NFκB (RANK). Under the influence of the key osteoclastogenic cytokine RANK Ligand (RANKL) these cells differentiate into bone resorbing osteoclasts [11]. The osteoclastogenic and proresorptive activity of RANKL is moderated by its physiological decoy receptor osteoprotegerin (OPG) [12,13]. In both humans and animals the ratio of RANKL to OPG is considered to be the final arbiter of the rate of osteoclastic bone resorption, and an inappropriate balance between these two factors is a key factor in the bone loss associated with numerous skeletal diseases.

Indeed, histological analyses of bones from HIV-1 transgenic rats and in-vitro bone marrow cultures revealed that bone loss in this model was a consequence of enhanced osteoclastic
Bone resorption, driven by an increase in osteoclast precursor number, and amplified by an elevated RANKL/OPG ratio [9••].

Bone loss in HIV/AIDS and role of the immuno-skeletal interface

In recent years it has become evident that the immune and skeletal systems are interlinked, and consequently changes in the immune system potently affect skeletal metabolism [14]. Not only are osteoclast precursors derived from monocytic cells, but activated lymphocytes secrete osteoclastogenic cytokines including RANKL, tumor necrosis factor-α (TNF-α), secreted osteoclastogenic factor of activated T cells (SOFAT) [15•], and others that drive up osteoclastic bone resorption in inflammatory conditions including rheumatoid arthritis [16], periodontitis [17,18], and during estrogen deficiency [14]. In contrast, under basal physiological conditions, B cells act as critical stabilizers of peak BMD in vivo. This is a consequence of the fact that both human [19] and mouse [20] B cells are a source of OPG. In fact, in mice B cells and their precursors contribute 64% of total bone marrow OPG concentrations [20]. B cell OPG production is further sustained by interactions with T cells, in part through CD40 ligand (CD40L) co-stimulation [19,20], and consequently animal models of B-cell deficiency, T-cell deficiency, and CD40, and CD40L deficiency all display severe bone loss and significantly diminished total and B-cell OPG production [20].

Although CD4 depletion is synonymous with HIV-1 infection, the humoral immune response is also dramatically impacted and severely impaired with significant declines in resting memory B cells along with a concomitant increase in activated and exhausted memory B-cell populations and an increase in the frequency of immature/transitional B cells [21,22••].

With this in mind we further investigated the source of elevated RANKL and diminished OPG in HIV-transgenic rats and identified a switch from production of bone sparing OPG by B cells, to production of bone destroying RANKL [9••]. The immunological regulation of osteoclastogenesis by B cells and T cells and our model of bone loss driven by alterations in the immuno-skeletal interface is presented diagrammatically in Fig. 1.

Interestingly, Chakravarti et al. [23] have reported that serum OPG concentrations are diminished in AIDS patients and that human peripheral blood T cells may also be a potential source of OPG. Interestingly, treatment of peripheral blood CD4 T cells in vitro with the HIV-1 coat protein gp120, caused a decline in T-cell OPG production. However, such actions have yet to be ratified in HIV-1-infected patients.

Another central cytokine involved in the immuno-skeletal interface is IFNγ. Under inflammatory conditions IFNγ has been found to promote bone loss by up regulating the activity of antigen-presenting cells (especially macrophages) and leading to T-cell activation and osteoclastogenic cytokine production [24]. On the contrary, IFNγ has also been shown to be a potent direct inhibitor of osteoclast formation [24,25] by antagonizing the downstream signal transduction from the RANKL receptor, RANK. Specifically, IFNγ induces degradation of tumor necrosis factor receptor-associated factor-6 (TRAF-6) [25], a critical adapter protein that links RANK to downstream osteoclastogenic transcription factors including NF-κB and c-Jun N-terminal kinase (JNK). Th1 T cells are a major source of IFNγ and interestingly, Yadav et al. [26••] have recently reported that the suppressor of cytokine signaling- 1 (SOCS-1), a potent inhibitor of IFNγ signal transduction is significantly elevated in cells derived from individuals with progressive HIV infection, as well as in the HIV-1 transgenic rat. It is speculated that up regulated SOCS-1 may contribute to elevated osteoclastogenesis by removing the dampening effect of IFNγ on the differentiation of osteoclast precursors in the context of HIV/AIDS (Reid W, University of Maryland School of Medicine, personal communication).
Overall, current data suggest that alterations in the immuno-skeletal interface may account for much of the loss of BMD observed in HIV patients naive to antiretroviral therapy.

HAART-induced bone loss in HIV/AIDS patients

Whereas it is now clear that the majority of antiretroviral naïve HIV-infected individuals have diminished BMD at presentation, surprisingly HAART does not appear to prevent further bone loss and may even exacerbate skeletal deterioration. In fact, HAART has long been suspected of influencing bone turnover independently of the bone loss associated with HIV-1 infection itself [1], although it has been suggested that this effect may be relatively modest in relation to the loss of BMD associated with other established osteoporosis risk factors [27,28]. Initiation of HAART has been consistently associated with up to a 6% reduction in hip BMD, a common site of fracture, over the first 48–96 weeks of therapy [29,30,31••]. However, some HAART regimens may be associated with more pronounced bone loss. In a recent meta-analysis of cross-sectional studies, HIV infected patients receiving protease inhibitors had a higher prevalence of osteoporosis compared to those receiving nonprotease inhibitor regimens [4]. However, with the available studies, the influence of other important factors such as disease severity and prior HAART history, could not be determined and interpretation of studies addressing HAART-associated bone loss have been further confounded by the wide range of antiretroviral regimens utilized in clinical practice, inadequate controlling of traditional osteoporosis risk factors, other disease-related effects, and variability in anatomical sites chosen for BMD analysis [4]. In the meta-analysis of Paccou et al. [5•] conventional risk factors for osteoporotic fractures, including wasting syndrome, hypogonadism, disorders in calcium and phosphate metabolism, and HIV infection per se accounted for bone loss with no evidence of a HAART-related contribution. Recently, Yin et al. [7•] report that premenopausal HIV-positive women display lower BMD than comparable HIV-negative women; however, BMD did not correlate with use or class of antiretrovirals. Similar data were reported by Libois et al. [32•] who found that osteopenia and osteoporosis was highly prevalent among HIV-infected premenopausal women, but loss of BMD was not associated with antiretroviral therapy. Furthermore, Brown et al. [33•] reported that loss of BMD after HAART initiation occurred independently of the antiretroviral regimen, and Grund et al. [34•] reported that continuous HAART was associated with a decline in BMD and possibly more fractures relative to intermittent HAART, but likewise reported no consistent drug-specific association.

By contrast, Duvivier et al. [35••] report that in a randomized clinical trial in which patients received either a non-nucleoside reverse transcriptase inhibitor (NNRTI) and a protease inhibitor, two nucleoside reverse transcriptase inhibitors (NRTIs) with a protease inhibitor, or a combination of one NNRTI with two NRTIs, 48 weeks of treatment revealed an overall combined (average of all treatment groups) significant decline in BMD of −4.1% at lumbar spine and −2.8% at hip. Importantly, the decrease in BMD at lumbar spine was significantly higher in the protease inhibitor-containing arms [−4.4% for protease inhibitor + NNRTI and −5.8% in the protease inhibitor + NRTIs arm, compared with (−1.5%) in the NNRTI + NRTIs arm] [35••]. Although this study suggests a protease inhibitor-specific effect, van Vonderen et al. [36•] reported greater bone loss in patients treated with the protease inhibitors lopinavir and ritonavir when combined with the NRTIs zidovudine and lamivudine, than when combined with the NNRTI nevirapine.

Attempts to understand the pathophysiology of HAART-associated bone loss are further confounded by the inability to replicate in-vivo effects of HAART in vitro. For example in vitro the protease inhibitor ritonavir inhibits osteoclastogenesis [37], whereas NRTIs are reported to have no effect on osteoclastogenesis but instead to suppress the activity of bone
forming osteoblasts [38]. In another in-vitro study fosamprenavir showed a significant increase in OPG and was associated with a RANKL decrease. However, the protease inhibitors atazanavir, saquinavir, and indinavir failed to impair the OPG/RANKL system at early times and at optimal concentrations [39•]. In another study tenofovir was reported to downregulate expression of Gnas, Got2 and Snord32a genes in osteoclasts, potentially impacting their function [40].

Consequently, after more than a decade of investigation no agreement exists as to the direct effects of HAART or their components on bone cells in vivo, or their mechanisms of action on the skeleton. In fact, consensus is beginning to emerge that all HAART formulation may be inherently detrimental to the skeleton[41]and that bone loss following HAART may be a general phenomenon related to the realignment of metabolism, as a consequence of disease reversal, and hence initiated by all HAART regimens, rather than due to direct effects of specific HAART constituents on bone cells. In fact, the relative potency of certain regimens of HAART such as those containing protease inhibitors in the disease reversal process may potentially explain, in part, their apparent impact on bone turnover.

Osteoporosis and bone fracture in the aging HIV/AIDS population

The cumulative number of AIDS cases in persons aged 50 years or older increased five-fold between 1990 and 2001 [42]. The Center for Disease Control and Prevention (CDC) has estimated that from 2001 to 2007 the number of people 50 years and older with AIDS nearly doubled, and currently, more than 70% of those with HIV are over age 40. It is projected that by 2015 more than half of the HIV-infected population in the USA will be over the age of 50 and the collision of HIV/HAART and age-related bone loss could lead to an epidemic of fractures, as these patients continue to live longer on therapy [43••]. A clinical study comparing fracture prevalence in HIV-infected and HIV-negative individuals has revealed that overall fracture prevalence was increased by 62% in HIV-infected patients. Fracture prevalence was relatively higher in HIV-infected patients across all age categories (30–79 years of age) and increased with age in both HIV-infected men and women [44]. Furthermore, in a study of BMD and fracture rates in HIV-infected older men (>49 years of age) HIV infection was independently associated with modestly reduced BMD and a 38% (although not statistically significant) increase in fracture rate [45].

Although men account for the majority of HIV-infected older adults, older women are acquiring HIV infection at a higher rate than older men [46,47].

Yin et al. [31••] recently investigated BMD and bone turnover in postmenopausal HIV-infected women and examined, among other indices, BMD, fracture prevalence, and bone turnover markers in 92 HIV-infected and 95 HIV-negative postmenopausal Hispanic and African- American women. HIV-infected women were found to have significantly lower T scores at the spine (78 vs. 64%), total hip (45 vs. 29%), and femoral neck (64 vs. 46%) and displayed significantly lower BMI-adjusted Z scores at the same sites. In addition, HIV-infected women had significantly higher serum bone resorption marker relative to HIV-negative women. HIV seropositive status independently and negatively correlated with spine and hip BMD after adjustment for age, ethnicity, BMI, and alcohol consumption. The investigators concluded that the lower BMD, higher prevalence of low BMD, and higher levels of bone turnover markers detected in HIV-infected postmenopausal women could place them at high risk for future fractures [31••].

Amelioration of osteoporosis in the HIV-infected population

A number of unresolved issues surround osteoporosis management in the setting of HIV infection. For example, what is the optimal measure of bone loss in this context? When
should antiresorptive intervention begin? How pharmacologically compatible are the antiresorptives with HAART? Several antiresorptive agents including bisphosphonates, calcitonin, synthetic estrogen receptor modulators (SERMs), and Denosumab (a monoclonal antibody against RANKL) are currently available for the management of osteoporosis, as is one anabolic agent, teriparatide (a fragment of human parathyroid hormone that promotes bone formation). Bisphosphonates are the most commonly prescribed class of antiresorptive agents and typically elicit a 60–70% decrease in markers of bone resorption [48]. Bisphosphonates are pyrophosphate analogs that attach strongly to hydroxyapatite, the major mineral component of bone, and promote apoptosis of resorbing osteoclasts by inhibiting farnesyl pyrophosphate synthase, a key enzyme in the mevalonate pathway [49]. Several oral analogs of this class of compound as well as an intravenous third-generation long-acting bisphosphonate (zoledronic acid) are currently licensed for the treatment of osteoporosis. Clinical trials of these agents in HIV-infected patients receiving antiretroviral therapy seem to suggest they are compatible with HAART.

Whereas studies on the use of teriparatide are in progress, in the context of HIV-1/AIDS, management of osteoporosis has primarily involved the use of bisphosphonates. A meta-analysis by Clay et al. [54] concludes that based on available safety and efficacy data from four randomized controlled trials assessing the utility of the bisphosphonate alendronate in patients with HIV-infection and osteopenia or osteoporosis [50–53], alendronate, an oral bisphosphonate should currently be considered the preferred agent for this application.

Oral bisphosphonate use in the setting of HIV infection may, however, be limited by gastrointestinal side-effects and contribution to an already high pill burden of HAART and other medications used for prophylaxis or treatment of opportunistic infections. As an alternative, annual zoledronic acid infusion has been shown to be well tolerated and to significantly increase BMD in HIV-infected individuals with osteoporosis [55,56••]. In a recent double-blinded, randomized, placebo-controlled trial 27 HIV-infected men and 3 women with osteopenia and osteoporosis were treated with a single intravenous 5mg dose of zoledronate for 12 months. BMD T scores were found to significantly increase at 6 and 12 months at lumbar spine and by 12 months at the hip, whereas markers of bone resorption significantly decreased as compared to placebo controls [56••]. Surprisingly bone formation markers were not observed to change significantly during the 12 months of the study. Importantly, this study confirmed that zoledronic acid was well tolerated with few side-effects and was compatible with simultaneous use of HAART.

The standard of care in postmenopausal osteoporosis, the archetypal osteoporotic condition, is to delay pharmacological intervention until the clinical definition of osteoporosis [based on World Health Organization (WHO) T score criteria of less than -2.5 SD from the mean optimal BMD reference range] have been met. This same conservative standard has generally been applied to HIV patients, and currently condemns them to development of osteoporosis before therapy is initiated. In the context of a rapidly growing aging HIV-infected population the potential for HIV/AIDS-associated bone loss to collide with bone loss associated with aging imposes a unique health threat in this population. Consequently, the conservative strategy of waiting for osteoporosis to develop prior to beginning antiresorptive therapy may be deleterious in this setting. Despite the apparent efficacy of antiresorptive agents at improving BMD, this class of compounds is generally inefficient at restoring lost bone mass because bone formation and bone resorption work as a ‘coupled’ unit. Consequently, numerous clinical studies have revealed that anticitabatic agents such as bisphosphonates also lead to an unfortunate concurrent decline in new bone formation along with suppression of bone resorption [57]. Although bisphosphonate treatment promotes a net balance in favor of bone formation relative to resorption, allowing some weak recovery of BMD to occur, typically up to 7–8% [48], antiresorptive agents are generally inefficient in
restoring lost bone mass, and once lost, BMD can almost never be restored to normal healthy levels. Furthermore, although DXA is cheap, well tolerated and noninvasive, BMD is a very imprecise measure of bone strength as bone architectural properties, which are a more pertinent reflection of bone quality and load bearing strength, cannot be extrapolated from this technique. Additionally, 70% of the skeleton comprises cortical bone, whereas trabecular bone, which is often rapidly degraded under most pathological conditions, comprises only 30% of total bone mass. Consequently, trabecular deterioration can be grossly under-estimated by DXA measurements. This can be a serious clinical problem as BMD may not reflect the severity of bone decline in teams of bone quality and load bearing capacity. The extent of this inadequacy is reflected by the fact that the WHO has estimated that over half of all fracture cases occur in individuals without osteoporosis [58]. Taken together, more aggressive screening and therapeutic measures may be warranted to delay the onset of osteoporosis in HIV patients who are comparatively younger and may have to live with skeletal complications for considerably longer than other aged populations.

Conclusion

It is now clear that bone loss occurs as a consequence of HIV infection and AIDS progression and that HAART likely contributes in part to this process, although more likely due to factors associated with disease reversal rather than through direct skeletal effects. It is also emerging that alterations in the immuno-skeletal interface are likely key driving forces in skeletal deterioration. Irrespective of the mechanisms involved a renewed emphasis on diagnosis and therapy may be indicated to avoid a significant new threat to the health and independence of the aging HIV/AIDS community.

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References and recommended reading

Papers of particular interest, published within the annual period of review, have been highlighted as:

• of special interest

• of outstanding interest

Additional references related to this topic can also be found in the Current World Literature section in this issue (pp. 569–570).


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Figure 1. Model of B-cell regulation of basal osteoclastogenesis through osteoprotegerin production and HIV-1-induced bone loss

B-cell production of OPG, regulated in part through CD40 to CD40 ligand co-stimulation by T cells, counteracts the key osteoclastogenic cytokine RANKL, moderating osteoclast formation and activity. HIV-1 interfering with/AIDS leads to a disruption of the immunoskeletal interface disrupting T cell to B-cell communication and leading to elevated RANKL and diminished OPG production by B cells. The elevated RANKL/OPG ratio is biased in favor of increased osteoclast formation. OPG, osteoprotegerin.