

## Workshop report

# Key reports from the 9th International Workshop on Adverse Drug Reactions and Lipodystrophy in HIV 2007

Ann Nestorowicz<sup>1\*</sup> and Stephen Cameron<sup>2</sup>

<sup>1</sup>International Medical Press Ltd, Sydney, NSW, Australia

<sup>2</sup>International Medical Press Ltd, London, UK

\*Corresponding author: Tel: +61 (02) 9954 4322; Fax: +61 (02) 9954 4388; E-mail: info.sy@intmedpress.com

The 9th International Workshop on Adverse Drug Reactions and Lipodystrophy in HIV provided a forum for the presentation of basic and clinical research focused on the pathogenesis and management of lipodystrophy and other adverse events associated with antiretroviral therapy. New data were reported on the contribution of both antiretroviral therapy and HIV infection itself on the development of metabolic abnormalities in patients with lipodystrophy, including insulin resistance and dyslipidaemia, which are associated with an increased risk of diabetes and cardiovascular disease. In addition, an emerging role of HIV and antiretroviral therapy in bone, liver and kidney disease were

highlighted. A major focus of the data presented in these areas concerned the identification and evaluation of risk factors and appropriate surrogate markers for defining cardiovascular disease risk as well as other outcomes of long-term treatment. The complexity of defining such risk factors was underscored by data describing the impact of race, age and gender in the progression of metabolic disease and related complications among different HIV-infected populations. Finally, advances in the development of pharmacovigilance reporting systems in resource-limited settings and their impact upon healthcare policies and the provision of patient care were also described.

## Introduction

The availability of highly active antiretroviral therapy (HAART) has dramatically changed the clinical course of human immunodeficiency virus-1 (HIV-1) infection, resulting in substantial reductions in morbidity and mortality. However, one of the major challenges for long-term treatment of HIV-1-infected patients is the management of complications associated with the use of HAART, such as dyslipidaemia, lipodystrophy, insulin resistance and glucose intolerance, bone disease, renal disease and lactic acidosis.

The International Workshop on Adverse Drug Reactions and Lipodystrophy in HIV provides a forum for the presentation of basic and clinical research in the field of metabolic complications, lipodystrophy, drug toxicities and other complications associated with HIV-1 infection and the use of antiretroviral therapy (ART). This report summarizes key findings from selected oral presentations and posters presented at the 9th Annual Workshop held recently in Sydney, Australia.

## Insulin resistance

The use of potent ART therapies, particularly protease inhibitor (PI)-based and thymidine nucleoside reverse transcriptase inhibitor (NRTI)-based regimens, has been associated with increases in the incidence of insulin resistance and glucose intolerance as well as progressive lipoatrophy and relative central fat accumulation in the HIV-infected population [1,2]. The prevalence of diabetes mellitus has been estimated at 14% in HIV-positive men on ART compared with 5% in HIV-negative men [3]. Furthermore, the incidence of diabetes mellitus in HIV-positive men on ART was more than four times that of HIV-negative controls over a 4-year period, suggesting a role for HIV itself in the pathogenesis of diabetes [3]. Although PIs have been both directly and indirectly implicated in the pathogenesis of insulin resistance [1], the impact of NRTIs upon glucose homeostasis is unclear. Van Vonderen *et al.* [4] evaluated the effects of zidovudine (AZT)/lamivudine

(3TC) on glucose metabolism and body composition in HAART-naive HIV-infected patients randomized to either AZT/3TC plus ritonavir-boosted lopinavir (LPV)/r ( $n=11$ ) or nevirapine (NVP) plus LPV/r ( $n=9$ ). After 24 months of therapy, limb fat decreased in the AZT/3TC arm and increased in the NVP arm, leading to a significant mean difference between these treatment groups of 1.3 kg ( $P<0.05$ ). Of interest, insulin-stimulated peripheral glucose disposal decreased significantly in the AZT/3TC arm from baseline to 3 months ( $P<0.05$ ), prior to limb fat loss or visceral fat gain, and was sustained for up to 24 months. This observation suggests that impairment of peripheral insulin sensitivity by certain NRTIs may occur through a body-composition independent mechanism at the level of skeletal muscle. At 12 months, insulin resistance was observed at the level of adipose tissue with increased lipolysis, whereas at 24 months resistance was observed at the liver level with increased hepatic glucose production. These findings are consistent with those of a recent study demonstrating impaired insulin sensitivity and concomitant reductions in skeletal muscle mitochondrial DNA content prior to changes in body composition in HIV-seronegative individuals after short-term (4 weeks) administration of stavudine (d4T) [5].

Differential effects of various PIs on insulin sensitivity in HIV-infected and HIV-negative populations were described in several presentations at the workshop. Lee *et al.* [6] compared the acute effects of indinavir (IDV), full-dose ritonavir (RTV) and amprenavir (APV) on endogenous glucose production (EGP) and insulin-mediated glucose disposal in randomized, placebo-controlled cross-over trials in healthy HIV-negative men. Under euglycaemic hyperinsulinaemic conditions, single-dose IDV and RTV, but not APV, blunted insulin-mediated suppression of EGP and insulin-mediated glucose disposal. In a metabolic substudy of BI 1182.33, Carr *et al.* [7] compared the effects of ritonavir-boosted tipranavir (TPV/r) (500 mg/200 mg twice daily or 500 mg/100 mg twice daily) and LPV/r, on a background of tenofovir (TDF)/3TC, on changes in body composition and metabolic parameters in ART-naive HIV-positive individuals. After 48 weeks of treatment with TPV/r or LPV/r, limb fat increased and visceral adipose tissue (VAT) decreased (5–10 cm<sup>2</sup>) relative to baseline in all three groups. In addition, significant increases in lipids (triglycerides [TG], high-density lipoprotein cholesterol [HDL-c] and total cholesterol) were associated with all treatment regimens. Despite these changes in lipid and body fat profiles, no significant changes in insulin sensitivity were observed either within or between study groups. Interestingly, plasma levels of adiponectin also increased substantially (+1,360–6,010 ng/ml) in all three study groups, with significantly greater increases occurring in both TPV/r arms relative to the LPV/r arm

( $P<0.0001$ ). It remains to be determined if these changes in circulating adiponectin levels represent a compensatory mechanism in response to the effects of PIs on lipids or other metabolic parameters. Additionally, it is unclear if the lack of lipodystrophy observed in this cohort is directly related to the absence of insulin resistance, which has been well documented with the use of other PIs.

To understand further the molecular mechanisms by which HIV infection and ART contribute to the development of insulin resistance in HIV-infected patients, Boothby *et al.* [8] analysed the expression of selected genes in subcutaneous fat from HIV-negative controls and HIV-positive patients before and after 6 months of ART. Comparison of transcript profiles of adipose tissue biopsies revealed that expression of the gene encoding 11 $\beta$ -hydroxysteroid dehydrogenase type 1 (HSD11B1), which catalyzes the conversion of inactive cortisone to cortisol, is significantly repressed in ART-naive HIV-positive patients compared with HIV-negative controls ( $P=0.006$ ). Of particular interest, HSD11B1 gene expression was significantly induced in HIV-positive patients following treatment for 6 months with efavirenz (EFV)+AZT/3TC or TDF/emtricitabine (FTC), such that levels were normalized relative to those of HIV-negative controls. Expression levels of genes involved in adipocyte differentiation and in glucose and lipid metabolism were also significantly increased in response to ART relative to baseline levels ( $P=0.0040$ – $0.0004$ ) and to HIV-negative controls ( $P=0.0001$ – $0.0130$ ), without significant differences between the two treatment arms. These findings are consistent with early increases in VAT, trunk and peripheral fat that were also observed in this cohort over 6 months of ART. Although increased HSD11B1 activity might contribute to adipocyte differentiation and changes in metabolic parameters after ART [8,9], additional time-points are needed to determine whether this and other changes in transcript profiles reflect a 'return to health' or direct effect of ART upon gene expression and to elucidate the temporal relationships between such changes with those in metabolic parameters and body fat distribution. Such studies would be of particular interest given previous data demonstrating increased HSD11B1 gene expression in abdominal subcutaneous adipose tissue of HIV-infected patients with lipodystrophy, relative to patients without lipodystrophy, following at least 18 months of HAART [10].

Several studies have previously demonstrated altered adipocyte morphology and macrophage infiltration in adipose tissue from HIV-infected patients with ART-related lipodystrophy [11–13]. Avettand-Fenoel *et al.* [14] reported dramatic increases in the expression of specific markers of pro-inflammatory M1 (CD68 and CD14) and anti-inflammatory M2 (CD163, chitinase

and CHI3L1) macrophages in abdominal subcutaneous adipose tissue from HIV-infected patients with lipodystrophy compared with non-obese controls. Adipose tissue biopsies taken after 6 months of ART interruption, however, showed significant decreases in the number of M1 ( $P=0.003$ ) macrophages and expression of M1 markers relative to baseline but not of M2 macrophages or specific markers. Stratification of patient samples by treatment regimen revealed that the observed decreases in M1 macrophages were mostly attributable to discontinuation of treatment with thymidine analogues (AZT and d4T). Although recruitment of pro-inflammatory M1 but not resident anti-inflammatory M2 macrophages has been implicated in the development of insulin resistance [15,16], it is unclear whether recruitment of M1 macrophages in lipodystrophic tissue from HIV-infected individuals reflects a causal relationship or is secondary to insulin resistance. Additional studies are also needed to clarify the role of M2 macrophages in adipocyte function and whole-body insulin resistance and other metabolic complications in HIV infection.

Although certain studies have provided evidence for dysregulation of adiponectin expression in HIV-infected subjects [17], other data suggest that PIs may increase adiponectin levels [18]. The relationship between fat redistribution and adipokines in HIV-infected patients receiving ART was examined by Kosmiski *et al.* [19], who evaluated leptin and adiponectin levels and regional adiposity in 1,143 HIV-infected individuals and 286 controls from the Fat Redistribution and Metabolic Change in HIV Infection (FRAM) study [20]. Previous analyses of this cohort have shown that HIV-infected men have significantly less total body fat ( $P<0.0001$ ) and subcutaneous fat ( $P<0.0001$ ) in all depots than non-HIV-infected controls. The expected positive association between leptin levels and total body fat as well as individual fat depots (VAT, leg subcutaneous adipose tissue [SAT], upper trunk SAT, lower trunk SAT and arm SAT) was apparent in both control men and women and in HIV-infected men and women ( $P<0.0001$ ). Although the predicted negative association between adiponectin and total body fat was observed for control uninfected men ( $P<0.01$ ) and women ( $P<0.0001$ ), no such association was apparent for HIV-infected men or women. Whereas VAT and upper trunk SAT were negatively associated with adiponectin levels as expected, a paradoxical positive association was found for leg SAT, lower trunk and arm SAT in HIV-infected men and leg SAT in HIV-infected women. Multivariate analysis also revealed a positive and expected association between leptin and regional fat depots (VAT, leg SAT and trunk SAT;  $P<0.0001$ ) and a paradoxical positive association

between adiponectin and leg SAT and lower trunk SAT. Similar results demonstrating a positive relationship between extremity fat and adiponectin levels were seen in previous smaller studies of HIV-infected patients [21]. However, adiponectin levels were negatively associated with VAT. These relationships between adiponectin and total and regional adiposity in HIV-infected individuals may reflect changes in adipocyte function associated with lipodystrophy or other metabolic complications associated with ART and/or HIV infection [19].

## Cardiovascular disease

The use of PIs in HIV-infected patients has been associated with increased cardiovascular (CV) events and deleterious effects on multiple coronary heart disease (CHD) risk factors, including dyslipidaemia, endothelial dysfunction and insulin resistance [22,23]. Other components of ART, such as NRTIs and non-nucleoside reverse transcriptase inhibitors (NNRTIs), could also affect CV risk factors [23–25]. Consequently, one of the key questions in the long-term management of HIV infection is whether traditional cardiovascular disease (CVD) risk factors defined for the general population are also applicable to the HIV-positive patient population. At this year's workshop, a number of presentations described studies to identify and evaluate potential CVD risk factors in HIV-positive patients.

Kingsley *et al.* [26] compared the prevalence and extent of coronary artery calcification (CAC), a marker of subclinical atherosclerosis, in HIV-infected and HIV-seronegative individuals from the Multicenter AIDS Cohort Study (MACS). The overall prevalence of CAC (Agatston score [AS]  $>10$ ), as assessed by computed tomography (CT), increased progressively with age from ~12% in individuals  $<45$  years of age to ~73% in those  $\geq 60$  years of age. After adjustment for age, multivariate analyses indicated that individuals on long-term HAART ( $\geq 8$  years) had an increased likelihood of having CAC (odds ratio [OR] 1.52; 95% confidence interval [95% CI] 1.04–2.25) relative to HIV-negative individuals. This association was not significant, however, upon further adjustment for race and age, smoking, family history of CAD, lipid levels and other known risk factors. By contrast, long-term HAART usage ( $\geq 8$  years) was significantly associated with a reduction in the extent of CAC (OR 0.62; 95% CI 0.42–0.94; adjusted for age and race). Surprisingly, the effect of HAART on the extent of CAC reduction was more pronounced in those not receiving lipid-lowering therapy. The combined data suggest that evaluation of CAC might not be an appropriate surrogate marker of increased cardiovascular risk in this population. In

addition, the results of this study also raise questions about whether there are factors specific to HIV infection that affect, either directly or indirectly, the prognostic value of various surrogate measures of CVD. As quantification of CAC (Agatston score) by CT does not involve assessment of plaque burden in non-calcified lesions, which might also be relevant in this cohort, further studies with newer multislice CT techniques [27,28] to determine plaque burden in addition to Agatston score are necessary to fully evaluate CVD risk and the impact of HAART on various markers for risk. Follow-up studies are in progress to determine the precise role of lipid-lowering therapy in CAC and to identify other putative risk factors for incident subclinical coronary atherosclerosis in this cohort.

Young *et al.* [29] developed a hierarchical model, based on drug exposure parameters, drug class and adjusted for confounders, to estimate the rate at which treatment-naïve patients develop metabolic syndrome (MetS) after commencing ART. Application of this model to 1,218 patients in the Swiss HIV Cohort Study estimated the prevalence of MetS, as defined by the International Diabetes Federation (IDF) [30], at approximately 20% (median follow up of 27 months) and an incidence rate of 7.5 cases per 100 patient years. In those receiving NRTIs, MetS was less likely to develop with the use of didanosine (OR 0.82; 95% CI 0.64–1.05 per 6 months of use) and more likely with d4T (OR 1.07; 95% CI 0.88–1.31). Among PIs, atazanavir (ATZ) (OR 0.37; 95% CI 0.18–0.78) or ATZ/r (OR 0.76; 95% CI 0.48–1.21) were less likely to be associated with MetS, whereas IDV/r was more likely (OR 1.17; 95% CI 0.83–1.65) to be associated with MetS. By contrast, MetS appears relatively unlikely to develop with the NNRTIs, EFV or NVP. Together, these data suggest that specific drugs within each drug class may have differential effects on the development of MetS, which could have important implications for the management of CVD risk in the HIV-infected population. A possible caveat to these findings, however, is whether the definition of MetS used in this study, which is based on a criterion of increased waist circumference [30], adequately reflects the metabolic and anthropometric abnormalities specific to the HIV-infected population and, if not, whether a revised definition of MetS might affect the prevalence data estimated using this hierarchical model.

The importance of CVD in HIV-infected patients was also highlighted at this year's workshop by the inclusion of a roundtable session providing feedback from the recent American Heart Association (AHA)/American Academy of HIV Medicine (AAHIVM) sponsored conference 'HIV & CVD: Decreasing Risks, Improving Care' (28–30 June 2007, Chicago, IL, USA). The objectives of this interdisciplinary conference were threefold:

to evaluate evidence linking CVD to HIV infection; to assess potential mechanisms for increased CVD; and to consider possible treatment strategies to reduce CVD risk in the HIV-infected population. Brief summaries of the key findings from this conference were presented and emphasized that increased CVD risk in the HIV-infected population arises from a complex interplay of multiple HIV- and ART-associated mechanisms. Such mechanisms encompass ART-associated metabolic and anthropometric abnormalities, which appear to be drug-specific rather than class-specific. Importantly, the finding that ART-associated changes in metabolic and anthropometric parameters are dynamic, with both acute and long-term changes that differ according to the treatment regimen, has implications for evaluating the effect of these abnormalities on long-term CVD outcomes. In addition, both ART and HIV infection itself may contribute to increased CVD risk by direct or indirect effects on myocardial function and the vasculature. Although the available data were also found to support an association between ART, particularly the PI class, and increased risk of myocardial infarction (MI), there was insufficient data to differentiate between specific drugs within the PI class with respect to CVD outcomes. Furthermore, the clinical significance of these effects of ART on the risk of MI may depend on the underlying CV risk. In this regard, it was also concluded that traditional CVD risk factors that have been defined for the general population appear to be relevant to the HIV-infected population. However, there is uncertainty regarding the possible role of HIV infection itself and the existence of specific independent risk factors for CVD. Finally, recommendations arising from this conference included further investigation and validation of risk factors to facilitate the development of appropriate CVD risk prediction models for the HIV-infected population. Such factors might also represent targets for therapeutic intervention to reduce CVD risk.

## Endothelial dysfunction

Several presentations at the workshop focused on the role of ART and HIV in endothelial dysfunction, an early marker of atherosclerosis, in HIV-infected subjects. Multiple factors have been implicated in the pathophysiology of endothelial dysfunction, including HIV infection, the immune response to infection, metabolic factors, lipodystrophy and direct and indirect effects of HAART, particularly PIs [31–33]. Dubé *et al.* [34] compared the effects of short-term treatment with ATZ or LPV/r on endothelial function in healthy HIV-seronegative individuals. Subjects were randomized to ATZ 400 mg once daily ( $n=9$ ), LPV/r 400/100 mg twice daily ( $n=9$ ) or placebo ( $n=12$ ). At week 4 of treatment, no significant differences in any of the treatment arms

were observed in either endothelium-dependent vasodilation (EDV) or endothelium-independent vasodilation relative to baseline, as measured by change in leg blood flow (LBF) in response to methacholine or nitroprusside, respectively. Furthermore, hyperinsulinaemic euglycaemic clamp studies revealed that insulin sensitivity was not impaired by treatment with either ATZ or LPV/r. These results contrast with those of previous studies in which the PI indinavir directly impaired EDV [35] and insulin sensitivity [36,37] in healthy HIV-seronegative individuals, suggesting that adverse effects of PIs on insulin sensitivity and endothelial function might be drug-specific rather than class-specific.

In a presentation by Torriani *et al.* [38], evidence was provided for an association between viral load suppression and improvement in endothelial function. On ACTG A5152s, a substudy of A5142, treatment-naive subjects were randomized to either a PI-sparing (NRTI+EFV;  $n=23$ ), an NNRTI-sparing (NRTI+LPV/r;  $n=31$ ) or an NRTI-sparing (EFV+LPV;  $n=28$ ) regimen. Endothelial function, as measured by brachial artery flow-mediated dilation (FMD), was impaired before treatment (4.0%; normal range >7.0%). With the exceptions of low-density lipoprotein cholesterol (LDL-c) and TG in the NRTI/EFV arm, and LDL-c in the NRTI/LPV/r arm, significant increases in lipids (HDL-c, TG and total cholesterol) relative to baseline were observed in all treatment arms at weeks 4 and 24 ( $P=0.01-0.001$ ). Despite these increases in proatherogenic lipids, endothelial function improved significantly in all treatment arms by week 4 (1.1%;  $P=0.003$ ) relative to baseline, with further increases by week 24 (1.9%;  $P<0.001$ ). Among those receiving NRTIs, significant improvements in FMD were observed in subjects receiving TDF ( $P<0.005$ ) or AZT ( $P<0.001$ ), but not d4T. Of those variables examined, only viral load suppression at week 24 was associated with improvements in FMD, suggesting that the improved endothelial function resulted from the control of HIV infection. At present, it is unknown whether this association reflects a causal relationship between viral load suppression and endothelial dysfunction and if the observed improvements in endothelial function result in reduced CVD risk for this population.

As described above, HIV infection is associated with impaired FMD whereas ART results in some improvement, irrespective of the treatment regimen used [38]. To identify additional factors mediating endothelial dysfunction during prolonged ART, Dubé *et al.* [39] presented preliminary data from studies assessing the relationship between endothelial dysfunction and ART, body composition and metabolic dysregulation (dyslipidaemia, insulin resistance and other factors). In this cross-sectional study, endothelial function was assessed by brachial artery FMD in 96 HIV-infected individuals.

Of the 49 (51%) individuals receiving ART, 28 were on a PI and 21 were on a non-PI-based regimen. No significant associations were apparent between endothelial dysfunction and ART, PI use, CD4<sup>+</sup> T-cell count or viral load in either ART-treated or untreated individuals. Among ART-treated subjects, VAT/SAT was not associated with FMD. Interestingly, decreased limb fat was associated with worse FMD, suggesting that lipodystrophy could be an important contributor to endothelial dysfunction in HIV-infected patients receiving ART. Further analyses of these and other metabolic parameters in this study are required to determine whether there is a mechanistic association between lipodystrophy and endothelial dysfunction in HIV-infected individuals.

In contrast to certain PIs, which are associated with proatherogenic lipid changes, the NNRTIs NVP and EFV have been associated with increases in plasma HDL-c [40,41]. Sankatsing *et al.* [42] explored the molecular mechanisms by which NVP induces increases in HDL-c cholesterol. In the Nevirapine Intensive Lipid Evaluation (NILE) study, treatment of 13 HIV-infected patients on AZT/3TC/abacavir (ABC) with NVP for 24 weeks resulted in significant increases in median apolipoprotein A-I (apoA-I; 14%;  $P<0.01$ ) and HDL-c (19%;  $P<0.01$ ) levels relative to baseline. Although no inhibition of cholesterol ester transfer protein (CETP) or other major HDL-c-modulating enzymes was observed, the absolute production rate of apoA-I was significantly increased relative to baseline ( $P<0.05$ ), suggesting that NVP increases apoA-I and HDL-c by promoting apoA-I production without affecting HDL-c catabolism. As the increases in HDL-c and apoA-I in these treatment-experienced patients are lower than those reported for treatment-naive patients [40], it is possible that a 'return to health' effect might contribute to the early increases in HDL observed with NVP in treatment-naive patients, whereas specific drug-induced effects could be responsible for HDL increases observed at later stages of treatment [42].

Flint *et al.* [43,44] investigated the mechanisms underlying EFV-mediated increases in plasma HDL-c. In these *in vitro* studies, EFV was shown to antagonize the activity of LXR, a transcription factor that has a major role in regulating lipid synthesis. EFV abrogated LXR-agonist-induced *de novo* synthesis of cholesterol and TG in human hepatoma cells. In addition, the expression of LXR downstream target genes involved in cholesterol metabolism, such as CETP, SREBP-1c and SREBP-2, was also attenuated by EFV *in vitro*. Similarly, EFV inhibited LXR-agonist-mediated induction of CETP protein and activity in transgenic mice expressing the human CETP gene (APOE\*3\* Leiden hCETP). However, EFV-treatment of this mouse model did not recapitulate the EFV-mediated increases in TG and HDL-c that have been observed in humans.

Further studies are needed to clarify the reasons for this discrepancy and to determine whether modulation of CETP expression also occurs during EFV treatment of HIV-infected individuals.

### Adverse drug reactions in resource-limited and other settings

Within the context of developing countries, major challenges in the management of HIV-1 infection include an increased burden of ART-related adverse drug reactions (ADRs) and implementation of protocols that minimize the risk for such side-effects. Mehta *et al.* [45] reviewed the design and preliminary findings of an ART pharmacovigilance reporting system recently established in the Western Cape, South Africa. Over a 2-year period, a total of 600 potentially ART-related ADRs (excluding immune reconstitution inflammatory syndrome) were reported, with the majority occurring in adults (585 events; 96%). Lactic acidosis and symptomatic hyperlactataemia (LASH) represented ~50% (301 events) of reported ADRs and, with the exception of three cases, occurred exclusively in patients on d4T-based regimens. LASH ADRs also occurred more frequently in women compared with non-LASH ADRs (81% versus 72%;  $P=0.031$ ) and in patients with a higher body mass ( $\leq 60$  kg 35%;  $>75$  kg 62%;  $P=0.001$ ). On the basis of these data, local HIV treatment guidelines were modified to recommend (1) dose reduction of d4T from 40 to 30 mg twice daily and (2) use of AZT instead of d4T in women with a body mass index (BMI)  $>28$  kg/m<sup>2</sup>. Other frequent ADRs included hepatotoxicity (62 events), peripheral neuropathy (107) and lipodystrophy (85). Although this pharmacovigilance system is still evolving and certain challenges remain, such as improving the rate and quality of reporting, it is an inexpensive system that has provided outcomes data for healthcare policy decisions. As such, it provides a valuable model for the feasibility of developing similar pharmacovigilance systems in other resource-limited settings.

Although numerous studies are available on HIV- and ART-associated metabolic complications in adults, there are limited data on these complications in young HIV-infected individuals. Preliminary data from the Pediatric Aids Clinical Trial Group (PACTG) 1045 study addressing these questions were presented by Aldrovandi *et al.* [46], who analysed the distribution and prevalence of metabolic and morphological outcomes in vertically infected children and youth using PI-based ( $n=161$ ; 90 males and 71 females) and PI-sparing ( $n=79$ ; 37 males and 42 females) treatment regimens. Body composition analyses, using dual X-ray absorptiometry (DEXA), revealed significant reductions in total and limb fat in both HIV-patient

treatment groups ( $P<0.05$ ) compared with HIV-negative controls. In addition, the PI-sparing treatment group also had significantly reduced BMI and trunk fat compared with HIV-negative controls. Although both HIV-infected groups also had increased TG relative to HIV-negative individuals ( $P<0.001$ ), dyslipidaemia was more pronounced in subjects on PI-based regimens, with significant elevations in total cholesterol, LDL-c and non-HDL-c ( $P<0.001$ – $0.002$ ) as well as reduced HDL-c ( $P<0.001$ ) relative to HIV-negative controls. Unadjusted fasting insulin levels were ~40% higher in both HIV-infected groups and, consequently, homeostasis model assessment of insulin resistance (HOMA-IR) scores were significantly increased in both HIV-infected groups relative to controls. Taken together, the observed dyslipidaemia and insulin resistance may accelerate the lifetime risk of CVD in this population, which already has extensive exposure to ART and will probably face a continued lifetime exposure to ART.

Given the tremendous racial diversity among the HIV-infected population, an important consideration in long-term treatment is whether racial differences exist in ART-related changes in metabolic and body composition parameters. To address this question, Gibert *et al.* evaluated changes in metabolic parameters [47] and body composition [48] among Latino ( $n=43$ ), African-American ( $n=243$ ) and Caucasian ( $n=112$ ) ART-naïve subjects initiating HAART. In this long-term metabolic subanalysis of the Flexible Initial Retrovirus Suppressive Therapies (FIRST) study cohort, mean changes in TG increased significantly over time (median follow up of 62 months) relative to baseline values in all three racial groups ( $P<0.01$ ), with more pronounced effects in Latinos and Caucasians versus African-Americans. Early, non-sustained rapid increases in LDL-c occurred in all groups but to a greater extent in Latinos and Caucasians, whereas early increases in HDL-c were sustained over time, with no significant differences among the groups. In contrast to these early changes in lipid profiles, plasma glucose levels increased dramatically in Latinos after 2 years of treatment ( $P<0.01$  versus baseline) and to a lesser extent in African-Americans ( $P<0.01$ ), with no significant increases in Caucasians. Development of insulin resistance showed a temporal profile similar to glucose among the racial groups, which is consistent with the known genetic predisposition of the Latino general population to diabetes and insulin resistance [49–51]. Anthropometric assessments of body composition (median follow up of 60 months) revealed that all racial groups exhibited an early and beneficial effect of ART, with significant increases in thigh and mid-arm non-subcutaneous tissue area as well as waist visceral tissue area. However, prolonged treatment resulted in

significant decreases in mid-thigh and mid-arm subcutaneous tissue area (fat) in all groups relative to baseline ( $P<0.01$ ). Latinos had the greatest loss in both waist and mid-arm subcutaneous tissue areas (fat) ( $P<0.01$  versus baseline), suggesting that this group might have an increased risk for lipoatrophy. Together, these differences in metabolic parameters and body composition changes among racial groups in response to ART could have clinical implications with respect to monitoring of lipids, glucose and body composition changes and, possibly, the choice of ART.

In dose-reduction studies in Asians, different PIs display good antiviral efficacy despite dose reductions of up to 50% [52–54]. To determine whether such dose reductions decrease the severity and/or frequency of PI-associated lipodystrophy and lipid abnormalities in Asians, van der Lugt *et al.* [55] assessed metabolic and body composition outcomes in treatment-naive, HIV-infected, Thai patients commencing double-boosted PI regimens. In this pilot study, 48 ART-naive patients were randomized to (1) LPV/r 400/100 mg twice daily + SQV 1000 mg twice daily (standard dose); (2) LPV/r 400/100 mg twice daily + SQV 600 mg twice daily; (3) LPV/r 266/66 mg twice daily + SQV 1000 mg twice daily; or (4) LPV/r 266/66 mg twice daily + SQV 600 mg twice daily. With the exception of LDL-c in group 3, significant increases in total cholesterol, TG, HDL-c and LDL-c were observed in all treatment groups at week 24 relative to baseline levels ( $P<0.0001$ – $0.046$ ). Dyslipidaemia was most pronounced when LPV/r and SQV were combined at standard doses. By contrast, no alterations in glucose metabolism, as measured by serum glucose and insulin, were apparent. Although there was no evidence of peripheral fat loss in any of the treatment groups, full-dose LPV/r+SQV was associated with increases in VAT and trunk fat at 24 weeks relative to baseline ( $P=0.023$ ) and to both low-dose groups of LPV/r ( $P=0.015$ – $0.020$ ). These findings suggest that metabolic and body composition changes related to PI use are exposure-related and that reductions in PI dose may alleviate, at least in part, the severity of these complications. However, whether these findings can be extrapolated to other populations and antiretroviral therapies remains to be clarified.

## Other toxicities

### Bone disease

Osteopenia and osteoporosis have recently emerged as complications of HIV infection and the use of ART. The estimated prevalence of osteoporosis in HIV-infected individuals is more than threefold higher than in HIV-negative controls [56]. Furthermore, the prevalence of osteopenia is higher in treated than in non-treated patients, suggesting a role for ART in the

pathogenesis of this disorder [56–58]. Preliminary results from a prospective study comparing early changes in bone metabolism in treatment-naive HIV-infected patients commencing HAART were presented by Bonnet *et al.* [59]. In this study, patients were randomized to receive two NRTIs in combination with either a PI ( $n=36$ ) or an NNRTI ( $n=36$ ). Interestingly, significant increases in markers of bone formation, bone alkaline phosphatase ( $P<0.0001$ ) and osteocalcin ( $P<0.0002$ ) as well as bone resorption ( $\beta$ -cross-laps;  $P<0.0002$ ) occurred as early as 3 months after commencing HAART. No significant differences in the increases of any biochemical markers of bone metabolism were observed between the treatment groups. Changes in bone markers were accompanied by global decreases in total bone mineral density (BMD) ( $P=0.003$ ) and L2–L4 BMD ( $P<0.0001$ ). After 9 months of treatment, slight but significant reductions in L2–L4 BMD relative to baseline were observed in the PI group ( $P=0.002$ ), but not in the NNRTI group ( $P=0.21$ ). Additional longer term measurements of BMD are needed to verify this lack of effect on bone metabolism by NNRTIs.

Briot *et al.* [60] evaluated the effect of a single-drug (LPV/r) ( $n=83$ ) versus a triple-therapy regimen (LPV/r+AZT/3TC) ( $n=53$ ) on changes in BMD in HIV-infected patients from the Monotherapy Antiretroviral Kaletra (MONARK) trial. Following 1 year of treatment, lumbar spine BMD (as measured by DEXA) was significantly decreased in both treatment arms at week 48, with a significant 4.1% decrease (interquartile range [IQR]  $-5.15$  to  $-2.1$ ;  $P\leq 0.001$ ) in the LPV/r group and a 3.32% decrease (IQR  $-5.0$  to  $-1.72$ ;  $P\leq 0.0001$ ) in the LPV/r + AZT/3TC group. Although no significant differences in loss of BMD were observed between the treatment arms at 1 year, longer term follow up is required to determine whether individual classes of ART or specific antiretrovirals have deleterious effects on BMD.

### Renal disease

In addition to HIV- and ART-associated adverse effects upon body composition, bone and metabolic parameters, renal complications are also frequent in the HIV-infected population. Proteinuria occurs in up to 30% of HIV-infected individuals and approximately 25% of patients develop increased serum creatinine levels as well as reduced glomerular filtration rates (GFR) [61]. Based on the findings that (1) dipstick proteinuria and increased serum creatinine predict HIV-1 disease progression [62–64], and (2) that tubulointerstitial inflammation in HIV-associated nephropathy correlates with the severity of renal dysfunction [65], Gupta *et al.* [66] evaluated whether dipstick proteinuria and reduced creatinine clearance

(CrCl) are markers of increased levels of activated T-cells in ART-naïve HIV-infected individuals. In this cross-sectional study of ACTG 384, A5095 and A5001, the prevalence of dipstick proteinuria (defined as  $\geq 1+$ ) and reduced CrCl ( $< 90$  ml/min; Cockcroft-Gault equation) in the total cohort at baseline was 7% and 18%, respectively. Comparison of individuals with and without proteinuria revealed that immune activation, as evidenced by an increased median percentage of CD8<sup>+</sup>/CD38<sup>+</sup>/HLA-DR<sup>+</sup> cells, was significantly higher in those with dipstick proteinuria ( $P=0.01$ ). Interestingly, this association between increased immune activation and dipstick proteinuria was present in the ACTG 384 subgroup ( $P<0.005$ ), but not the A5095 subgroup. In multivariate analyses, proteinuria in the ACTG 384 cohort was associated not only with immune activation (OR 1.15; 95% CI 1.04–1.29) but also lower haemoglobin levels (OR 0.11; 95% CI 0.03–0.37) and black race/ethnicity (OR 2.37; 95% CI 1.14–4.95). By contrast, viral load was a strong predictor of proteinuria (OR 2.58; 95% CI 1.40–4.47) in the A5095 subgroup, but no association was found with immune activation, decreased haemoglobin or black race. Immune activation was not significantly associated with CrCl (either  $< 90$  ml/min or greater) in the total cohort, possibly reflecting a relatively preserved glomerular function overall. Longitudinal population-based studies that include individuals with more severe renal disease are needed to fully evaluate whether CrCl and other markers of renal function are predictive of overall mortality as well as risk for disease progression to AIDs. Further studies are also necessary to define factors responsible for the observed differences between the study subgroups.

#### Liver disease

HIV–HCV coinfection is one of the leading causes of liver disease worldwide, with HCV coinfection occurring in an estimated ~30% of HIV-infected individuals [67–70]. Furthermore, evidence from several studies suggests that such patients have a more aggressive and rapid progression to cirrhosis and end-stage liver disease than patients infected with HCV alone [69,71–73]. Although the pathogenesis and underlying mechanisms of accelerated fibrosis have not been precisely defined, factors influencing liver disease progression in this population include ART-associated hepatotoxicity and immune suppression [74]. Barreiro *et al.* [75] evaluated several risk factors for liver disease progression in a cohort of 489 HIV–HCV co-infected patients. Of these patients, 37% had advanced liver fibrosis (ALF), defined as liver stiffness  $> 9.5$  kPa (Metavir F3–F4) by ultrasonic transient elastometry (FibroScan). Factors

significantly associated with ALF included increased age (OR 2.1; 95% CI 1.1–4.0 per decade), past or current alcohol abuse (OR 2.3; 95% CI 1.3–4.0), episodes of ALT levels  $> 100$  IU/ml (OR 1.04; 95% CI 1.03–1.06), increased glucose levels (OR 1.1; 95% CI 1.0–1.2 per 10 mg/dl), insulin resistance (HOMA scores; OR 1.2; 95% CI 1.1–1.4) and reduced CD4<sup>+</sup> T-cell counts (OR 0.8; 95% CI 0.7–0.9 per 100 cells/ml). Importantly, duration of exposure to PIs was correlated with greater liver fibrosis ( $\rho$  0.11;  $P=0.01$ ), with more pronounced effects occurring in those receiving RTV-boosted PIs ( $\rho$  0.18;  $P<0.001$ ). By contrast, no such correlations were observed with specific NRTIs or NNRTIs. Together, these findings suggest that, in addition to host factors, PI-based regimens may have both potentially positive and detrimental effects upon liver disease progression in the HIV–HCV co-infected population.

#### Hypersensitivity reactions

Current treatment guidelines recommend against the use of NVP in HIV-infected patients with nadir CD4<sup>+</sup> T-cell counts  $> 250$  cells/ $\mu$ l in females and  $> 400$  cells/ $\mu$ l in males, owing to an increased risk for severe and potentially life-threatening hypersensitivity reactions (HSR). Such reactions are characterized by an increased incidence of symptomatic hepatic events and skin rashes. However, CD4<sup>+</sup> T-cell thresholds for NVP use are primarily based on data from treatment-naïve patients at the start of NVP-based HAART and few data are available on the incidence of NVP-associated HSR in treatment-experienced patients. Wit *et al.* [76] presented a retrospective analysis of the AIDS Therapy Evaluation in the Netherlands (ATHENA) cohort in which they examined the relationship between NVP-associated HSR and CD4<sup>+</sup> T-cell counts as well as other possible HSR risk factors in treatment-experienced patients. Of the 3,752 treatment-experienced and treatment-naïve patients who used NVP, 231 (6.2%) discontinued treatment due to adverse events compatible with HSR within 18 weeks of starting therapy. Skin rashes without concomitant elevations in liver enzymes occurred in the majority of patients who experienced NVP-related HSRs (182; 78%); hepatotoxicity was present in 57 patients (25%) and 8 patients experienced both rash and hepatotoxicity. When compared to treatment-naïve patients with low nadir CD4<sup>+</sup> T-cell counts at baseline, treatment-experienced patients with both low nadir and current CD4<sup>+</sup> T-cell counts have a similar risk of HSR (OR 1.02; 95% CI 0.67–1.57), whereas those with low nadir and high current CD4<sup>+</sup> T-cell counts are at increased risk for HSR (OR 1.77; 95% CI 1.10–2.85;  $P<0.02$ ). Undetectable viral load at

baseline (start of NVP) in the treatment-experienced group with low nadir and high current CD4<sup>+</sup> T-cell counts reduced the risk of developing NVP-related HSR in this population (OR 1.03; 95% CI 0.66–1.61). Other independent risk factors for NVP-related HSR include female gender (OR 1.39; 95% CI 1.02–1.90), with women of Asian descent having a particularly high risk of HSR (OR 6.15; 95% CI 2.96–12.69;  $P < 0.0001$ ). Further studies are needed to confirm if viral load and other risk factors for NVP-associated HSR defined in this work are applicable to different cohorts or populations.

## Intervention strategies

Various interventions for the prevention and/or treatment of lipodystrophy and other complications of HIV infection and ART are currently under investigation. Such interventions include surgical correction, uridine supplementation, and the use of growth hormone and growth-hormone-releasing factor analogues, metformin, thiazolidinediones and statins.

### Growth hormone-releasing factor analogues

Administration of TH9507, a growth hormone-releasing factor (GRF) analogue, has been demonstrated to decrease truncal and visceral fat without reductions in subcutaneous fat. Moreover, TH9507 has been shown to improve lipid profiles in HIV patients with abdominal fat accumulation [77]. Falutz *et al.* [78] reported follow up data on the time-course of these changes in VAT in response to TH9507 treatment. Significant decreases in VAT (-12.1%;  $P < 0.001$  versus placebo) in TH9507-treated groups were mostly achieved by week 13 of treatment, with smaller additional decreases occurring by week 26 (-15.2%;  $P < 0.001$  versus placebo). Absolute reductions in VAT were related to the magnitude of VAT accumulation at baseline, such that changes in VAT were greatest among patients with larger VAT at baseline. Similar temporal profiles and association with baseline VAT were observed for TH9507-induced increases in insulin-like growth factor 1 (IGF-1). In addition to increases in IGF-1, TH9507 induced significant increases in adiponectin levels ( $P = 0.029$  versus placebo). Although other studies from the same group [79] found no evidence for significant changes in body size parameters at week 26, TH9507 treatment significantly improved body dysmorphia, with patients reporting improvements in belly appearance distress ( $P = 0.03$  versus placebo) and composite body distress. Both patients ( $P = 0.03$ ) and physicians ( $P = 0.04$ ) reported improved belly profile scores together with concomitant improvements in health-related quality of life scores.

### Autologous fat transplants

Several surgical options to treat lipodystrophy have recently become available, including autologous fat transplantation and implantation or injection of synthetic bulking agents, collagen and silicone. Disadvantages of these approaches include a lack of durability, resulting in a requirement for repeated injections over time. In an observational study, Fontdevila *et al.* [80] evaluated the safety and durability of autologous fat grafts in 44 HIV-infected patients with facial lipodystrophy. Mean volumes of facial fat, as assessed by CT, progressively increased from 2.9 cm<sup>3</sup> (95% CI 2.4–3.4) at baseline to 6.3 cm<sup>3</sup> (95% CI 5.8–6.8) at 12 months ( $P < 0.0001$ ). Only one case of fat reabsorption was reported and the procedure was well-tolerated, suggesting that autologous fat grafts could achieve durable results for at least 12 months. In a similar study, Orlando *et al.* [81] assessed the effectiveness and long-term durability of autologous fat transplants (AFT) in a longer-term 104-week follow up of 45 patients. Of these patients, 23 (51.1%) had a single surgical treatment and 22 (48.9%) needed re-intervention with AFT and/or poly-L-lactic acid. At 2 years after the first AFT, cheek thickness had increased significantly in all patients ( $P < 0.0001$ ), with concomitant important improvements in face (+3.3 ± 2.8;  $P < 0.0001$ ) and body image satisfaction scores (-0.8 ± 0.7;  $P < 0.0001$ ). Although AFT appears to be safe and effective for the treatment of facial lipodystrophy, the requirement for re-intervention in long-term follow up in almost half of patients poses a major limitation of this approach.

### Thiazolidinediones

As both peroxisome proliferator-activated receptor- $\gamma$  (PPAR- $\gamma$ ) agonists and exercise training have been demonstrated to have beneficial effects upon insulin resistance, proatherogenic lipid profiles and other CVD risk factors, and because exercise has been shown to decrease visceral and peripheral fat [82–88], Reeds *et al.* [89] compared the effectiveness of the PPAR- $\gamma$  agonist pioglitazone plus exercise training with pioglitazone alone on insulin sensitivity, hepatic lipid content and regional adiposity in a pilot study of 15 HIV-infected patients. With respect to adiposity, modest increases in trunk (0.13 ± 0.57 kg) and appendicular (0.48 ± 0.21 kg) fat occurred in the pioglitazone-treated group, whereas pioglitazone plus exercise resulted in decreased trunk (-1.84 ± 0.99 kg) and appendicular (-0.31 ± 0.50 kg) fat. No significant effects of these treatment regimens were observed on lipid profiles (HDL, LDL and TG or very low-density lipoprotein). Insulin sensitivity was improved by both treatment regimens, as evidenced by a greater increase in peripheral insulin sensitivity with pioglitazone plus exercise

(+92%) than with pioglitazone alone (+62%;  $P=0.05$ ), and a similar enhancement of insulin-mediated suppression of EGP (23% for pioglitazone alone; 31% for pioglitazone plus exercise) that was significantly correlated ( $P<0.002$ ) with reductions (~3.0–6%) in hepatic lipid content in both groups. Further follow-up studies in larger cohorts are required to determine whether there are differential effects of these regimens upon long-term CVD risk in HIV-infected individuals.

## Conclusions

The availability of potent antiretroviral therapies for the treatment of HIV-infected patients has not only altered the clinical course of HIV infection, but has also brought new challenges and issues arising from long-term drug toxicities that can cause significant morbidity and mortality. As a consequence, numerous questions and new areas of research have arisen in the field. Several of these areas were highlighted by data presented at the 9th International Workshop on Adverse Reactions and Lipodystrophy in HIV, including the need for a greater understanding of the underlying mechanisms and pathogenesis of HIV- and ART-associated metabolic, anthropomorphic and cardiovascular abnormalities. Other presentations raised the important question of whether risk factors that have been defined in the general population for cardiovascular, liver, kidney or metabolic disease are also applicable to the HIV-infected population. Defining such risk factors could be further complicated by racial, age and gender differences among HIV-infected populations. There is also a vital need to develop validated surrogate markers to assess and manage CVD risk as well as other ART-related adverse events. Several studies provided evidence for differential effects of individual drugs within drug classes upon metabolic parameters, raising the possibility of improved treatment regimens in the future. Finally, further research into the molecular mechanisms responsible for these differential effects may yield alternative approaches for the development of novel therapeutics that display improved safety and efficacy for the treatment of HIV infection.

## Acknowledgements

This report was made possible by an unrestricted educational grant from Boehringer Ingelheim International GmbH.

## References

1. Florescu D, Kotler DP. Insulin resistance, glucose intolerance and diabetes mellitus in HIV-infected patients. *Antivir Ther* 2007; **12**:149–162.

2. Koutkia P, Grinspoon S. HIV-associated lipodystrophy: pathogenesis, prognosis, treatment, and controversies. *Annu Rev Med* 2004; **55**:303–317.
3. Brown TT, Cole SR, Li X, *et al.* Antiretroviral therapy and the prevalence and incidence of diabetes mellitus in the multicenter AIDS cohort study. *Arch Intern Med* 2005; **165**:1179–1184.
4. van Vonderen MGA, Blumer RME, Hassink E, *et al.* Zidovudine/lamivudine persistently contributes to peripheral insulin resistance by a body composition-independent mechanism demonstrated by repeated clamp studies during 2 years of first-line ART with zidovudine/lamivudine/lopinavir/ritonavir. *Antivir Ther* 2007; **12**:L13.
5. Fleischman A, Johnsen S, Systrom DM, *et al.* Effects of a nucleoside reverse transcriptase inhibitor, stavudine, on glucose disposal and mitochondrial function in muscle of healthy adults. *Am J Physiol Endocrinol Metab* 2007; **292**:E1666–E1673.
6. Lee GA, Schwarz JM, Patzek S, *et al.* The acute effects of HIV protease inhibitors on glucose production in healthy HIV-negative men. *Antivir Ther* 2007; **12**:L46.
7. Carr A, Zajdenverg R, Workman C, *et al.* Effects of tipranavir/r (500/200 or 500/100 mg BID) in comparison with lopinavir/r (400/100 mg BID) on changes in body composition and metabolic parameters in ARV-naive patients over 48 weeks. *Antivir Ther* 2007; **12**:L8.
8. Boothby M, Tomlinson JW, McGee KC, *et al.* The effect of antiretroviral therapy on genes involved with glucose and lipid metabolism. *Antivir Ther* 2007; **12**:L12.
9. Sukhija R, Kakar P, Mehta V, *et al.* Enhanced 11 $\beta$ -hydroxysteroid dehydrogenase activity, the metabolic syndrome, and systemic hypertension. *Am J Cardiol* 2006; **98**:544–548.
10. Sutinen J, Kannisto K, Korshennikova E, *et al.* In the lipodystrophy associated with highly active antiretroviral therapy, pseudo-Cushing's syndrome is associated with increased regeneration of cortisol by 11 $\beta$ -hydroxysteroid dehydrogenase type 1 in adipose tissue. *Diabetologia* 2004; **47**:1668–1671.
11. Bastard JP, Caron M, Vidal H, *et al.* Association between altered expression of adipogenic factor SREBP1 in lipotrophic adipose tissue from HIV-1-infected patients and abnormal adipocyte differentiation and insulin resistance. *Lancet* 2002; **359**:1026–1031.
12. Jan V, Cervera P, Maachi M, *et al.* Altered fat differentiation and adipocytokine expression are inter-related and linked to morphological changes and insulin resistance in HIV-1-infected lipodystrophic patients. *Antivir Ther* 2004; **9**:555–564.
13. Villarroya F, Domingo P, Giral M. Lipodystrophy in HIV 1-infected patients: lessons for obesity research. *Int J Obes (Lond)* 2007; in press.
14. Avettand-Fenoel V, Kim M, Antuna B, *et al.* Macrophage recruitment in adipose tissue from HIV-infected patients under ART: concomitant presence of classically activated pro-inflammatory M1 and alternatively activated M2 macrophages. *Antivir Ther* 2007; **12**:L12.
15. Lumeng CN, Deyoung SM, Saltiel AR. Macrophages block insulin action in adipocytes by altering expression of signalling and glucose transport proteins. *Am J Physiol Endocrinol Metab* 2007; **292**:E166–E174.
16. Lumeng CN, Bodzin JL, Saltiel AR. Obesity induces a phenotypic switch in adipose tissue macrophage polarization. *J Clin Invest* 2007; **117**:175–184.
17. Addy CL, Gavrila A, Tsioutras S, *et al.* Hypoadiponectinemia is associated with insulin resistance, hypertriglyceridemia, and fat redistribution in human immunodeficiency virus-infected patients treated with highly active antiretroviral therapy. *J Clin Endocrinol Metab* 2003; **88**:627–636.
18. Lee GA, Mafong DD, Noor MA, *et al.* HIV protease inhibitors increase adiponectin levels in HIV-negative men. *J Acquir Immune Defic Syndr* 2004; **36**:645–647.

19. Kosmiski L, Kotler DP, Lewis CE, *et al.* Relationship of fat distribution with adipokines in HIV-infection: the FRAM study. *Antivir Ther* 2007; **12**:L16.
20. Bacchetti P, Gripshover B, Grunfeld C, *et al.* Fat distribution in men with HIV infection. *J Acquir Immune Defic Syndr* 2005; **40**:121–131.
21. Tong Q, Sankale JL, Hadigan CM, *et al.* Regulation of adiponectin in human immunodeficiency virus-infected patients: relationship to body composition and metabolic indices. *J Clin Endocrinol Metab* 2003; **88**:1559–1564.
22. Friis-Moller N, Reiss P, Sabin CA, *et al.* Class of antiretroviral drugs and the risk of myocardial infarction. *N Engl J Med* 2007; **356**:1723–1735.
23. Mondy K, Tebas P. Cardiovascular risks of antiretroviral therapies. *Annu Rev Med* 2007; **58**:141–155.
24. Currier JS, Kendall MA, Henry WK, *et al.* Progression of carotid artery intima-media thickening in HIV-infected and uninfected adults. *AIDS* 2007; **21**:1137–1145.
25. Stein JH, Klein MA, Bellehumeur JL, *et al.* Use of human immunodeficiency virus-1 protease inhibitors is associated with atherogenic lipoprotein changes and endothelial dysfunction. *Circulation* 2001; **104**:257–262.
26. Kingsley LA, Cuervo J, Munoz A, *et al.* Subclinical coronary atherosclerosis, HIV-infection and antiretroviral therapy; results from the multicentre AIDS cohort study. *Antivir Ther* 2007; **12**:L11.
27. Budoff MJ, Achenbach S, Blumenthal RS, *et al.* Assessment of coronary artery disease by cardiac computed tomography: a scientific statement from the American Heart Association Committee on Cardiovascular Imaging and Intervention, Council on Cardiovascular Radiology and Intervention, and Committee on Cardiac Imaging, Council on Clinical Cardiology. *Circulation* 2006; **114**:1761–1791.
28. Budoff MJ, Gopal A, Gopalakrishnan D. Cardiac computed tomography: diagnostic utility and integration in clinical practice. *Clin Cardiol* 2006; **29**:14–14.
29. Young J, Glass T, Weber R, *et al.* The rate at which therapy-naïve patients develop metabolic syndrome when treated and its associations with different components of antiretroviral therapy: the Swiss Cohort Study. *Antivir Ther* 2007; **12**:L10.
30. Alberti KG, Zimmet P, Shaw J. The metabolic syndrome — a new worldwide definition. *Lancet* 2005; **366**:1059–1062.
31. Andrade AC, Cotter BR. Endothelial function and cardiovascular diseases in HIV infected patient. *Braz J Infect Dis* 2006; **10**:139–145.
32. Cotter BR. Endothelial dysfunction in HIV infection. *Curr HIV/AIDS Rep* 2006; **3**:126–131.
33. Wang X, Chai H, Yao Q, *et al.* Molecular mechanisms of HIV protease inhibitor-induced endothelial dysfunction. *J Acquir Immune Defic Syndr* 2007; **44**:493–499.
34. Dubé M, Shen C, Greenwald ML, *et al.* Effects of 4 weeks of atazanavir, lopinavir/ritonavir or placebo on endothelial function and insulin sensitivity in healthy men. *Antivir Ther* 2007; **12**:L14.
35. Shankar SS, Dubé MP, Gorski JC, *et al.* Indinavir impairs endothelial function in healthy HIV-negative men. *Am Heart J* 2005; **150**:933.
36. Noor MA, Lo JC, Mulligan K, *et al.* Metabolic effects of indinavir in healthy HIV-seronegative men. *AIDS* 2001; **15**:F11–F18.
37. Noor MA, Seneviratne T, Aweeka FT, *et al.* Indinavir acutely inhibits insulin-stimulated glucose disposal in humans: a randomized, placebo-controlled study. *AIDS* 2002; **16**:F1–F8.
38. Torriani FJ, Komarow L, Cotter BR, *et al.* Control of HIV viral replication is associated with rapid improvement in endothelial function sustained over 24 weeks: A5152s, a substudy of A5142. *Antivir Ther* 2007; **12**:L15.
39. Dubé M, Shen C, Waltz JS, *et al.* Relationship of body composition, antiretroviral use, and HIV disease factors to endothelial dysfunction in HIV-infected subjects. *Antivir Ther* 2007; **12**:L15.
40. van der Valk M, Kastelein JJ, Murphy RL, *et al.* Nevirapine-containing antiretroviral therapy in HIV-1 infected patients results in an anti-atherogenic lipid profile. *AIDS* 2001; **15**:2407–2414.
41. Negro E, Ribalta J, Ferre R, *et al.* Efavirenz induces a striking and generalized increase of HDL-cholesterol in HIV-infected patients. *AIDS* 2004; **18**:819–821.
42. Sankatsing RR, Franssen R, Hassink E, *et al.* Nevirapine increases high density lipoprotein-cholesterol by stimulation of apolipoprotein A1 synthesis. *Antivir Ther* 2007; **12**:L5.
43. Flint OP, Bellamine A, Noor MA, *et al.* Effects of efavirenz on lipid metabolism in APOE\*3\*Leiden hCETP double-transgenic mice: evidence for antagonism of LXR pathway. *Antivir Ther* 2007; **12**:L5.
44. Flint OP, Bellamine A, Noor MA, *et al.* Molecular mechanisms for efavirenz effects on lipid metabolism. *Antivir Ther* 2007; **12**:L6.
45. Mehta U, Durrheim DN, Cohen K, *et al.* Design and outcomes of an antiretroviral pharmacovigilance programme in South Africa. *Antivir Ther* 2007; **12**:L7.
46. Aldrovandi GM, Lindsey JC, Jacobsen D, *et al.* Dyslipidemia in vertically infected children and youth on protease-inhibitor (PI)-containing antiretroviral therapy (ART): preliminary results of PACTG 1045. *Antivir Ther* 2007; **12**:L6.
47. Gibert CL, Shlay JC, Sharma S, *et al.* Racial differences in long-term changes in metabolic parameters in antiretroviral-naïve persons initiating HAART. *Antivir Ther* 2007; **12**:L19.
48. Gibert CL, Shlay JC, Sharma S, *et al.* Racial differences in long-term changes in body composition in antiretroviral-naïve persons initiating HAART. *Antivir Ther* 2007; **12**:L19.
49. Karter AJ, Ferrara A, Liu JY, *et al.* Ethnic disparities in diabetic complications in an insured population. *JAMA* 2002; **287**:2519–2527.
50. Harris MI. Racial and ethnic differences in health care access and health outcomes for adults with type 2 diabetes. *Diabetes Care* 2001; **24**:454–459.
51. Pugh JA, Stern MP, Haffner SM, *et al.* Excess incidence of treatment of end-stage renal disease in Mexican Americans. *Am J Epidemiol* 1988; **127**:135–144.
52. Burger D, Boyd M, Duncombe C, *et al.* Pharmacokinetics and pharmacodynamics of indinavir with or without low-dose ritonavir in HIV-infected Thai patients. *J Antimicrob Chemother* 2003; **51**:1231–1238.
53. Boyd M, Moosikapun P, Burger D, *et al.* Pharmacokinetics of reduced-dose indinavir/ritonavir 400/100 mg twice daily in HIV-1-infected Thai patients. *Antivir Ther* 2005; **10**:301–307.
54. Ananworanich J, Hill A, Siangphoe U, *et al.* A prospective study of efficacy and safety of once-daily saquinavir/ritonavir plus two nucleoside reverse transcriptase inhibitors in treatment-naïve Thai patients. *Antivir Ther* 2005; **10**:761–767.
55. van der Lugt J, Ruxrungtham K, Autar S, *et al.* Metabolic changes in a Thai treatment-naïve population starting double-boosted protease inhibitor therapy. *Antivir Ther* 2007; **12**:L7.
56. Brown TT, Qaqish RB. Antiretroviral therapy and the prevalence of osteopenia and osteoporosis: a meta-analytic review. *AIDS* 2006; **20**:2165–2174.
57. Bongiovanni M, Fausto A, Cicconi P, *et al.* Non-nucleoside reverse-transcriptase-inhibitor-based HAART and osteoporosis in HIV-infected subjects. *J Antimicrob Chemother* 2006; **58**:485–486.
58. Brown TT, McComsey GA. Osteopenia and osteoporosis in patients with HIV: a review of current concepts. *Curr Infect Dis Rep* 2006; **8**:162–170.
59. Bonnet E, Mabile L, Ruidavets JB, *et al.* Important changes in bone metabolism soon after commencing HAART. *Antivir Ther* 2007; **12**:L17.

60. Briot K, Kolta S, Flandre P, *et al.* One-year bone mineral density changes in antiretroviral-naïve HIV-infected patients treated by a triple versus a single-agent regimen, with lopinavir/ritonavir in the Monark trial. *Antivir Ther* 2007; **12**:L62.
61. Gupta SK, Eustace JA, Winston JA, *et al.* Guidelines for the management of chronic kidney disease in HIV-infected patients: recommendations of the HIV Medicine Association of the Infectious Diseases Society of America. *Clin Infect Dis* 2005; **40**:1559–1585.
62. Szczech LA, Gupta SK, Habash R, *et al.* The clinical epidemiology and course of the spectrum of renal diseases associated with HIV infection. *Kidney Int* 2004; **66**:1145–1152.
63. Szczech LA, Hoover DR, Feldman JG, *et al.* Association between renal disease and outcomes among HIV-infected women receiving or not receiving antiretroviral therapy. *Clin Infect Dis* 2004; **39**:1199–1206.
64. Gupta SK, Mamlin BW, Johnson CS, *et al.* Prevalence of proteinuria and the development of chronic kidney disease in HIV-infected patients. *Clin Nephrol* 2004; **61**:1–6.
65. Kimmel PL. HIV-associated nephropathy: virological issues related to renal sclerosis. *Nephrol Dial Transplant* 2003; **18 Suppl 6**:vi59–vi63.
66. Gupta SK, Komarow L, Gulick RM, *et al.* Proteinuria, creatinine clearance and immune activation in HIV-infected subjects: a secondary analysis of treatment-naïve studies ACTG 384, A5095 and A5001. *Antivir Ther* 2007; **12**:L17.
67. Jones R, Dunning J, Nelson M. HIV and hepatitis C co-infection. *Int J Clin Pract* 2005; **59**:1082–1087.
68. Rockstroh J. HIV/hepatitis co-infection. *J HIV Ther* 2007; **12**:1.
69. Rockstroh JK. Management of hepatitis C/HIV coinfection. *Curr Opin Infect Dis* 2006; **19**:8–13.
70. Leen CL. Hepatitis C and HIV co-infection. *Int J STD AIDS* 2004; **15**:289–294.
71. Soto B, Sanchez-Quijano A, Rodrigo L, *et al.* Human immunodeficiency virus infection modifies the natural history of chronic parenterally-acquired hepatitis C with an unusually rapid progression to cirrhosis. *J Hepatol* 1997; **26**:1–5.
72. Greub G, Ledergerber B, Battegay M, *et al.* Clinical progression, survival, and immune recovery during antiretroviral therapy in patients with HIV-1 and hepatitis C virus coinfection: the Swiss HIV Cohort Study. *Lancet* 2000; **356**:1800–1805.
73. Vallet-Pichard A, Pol S. Natural history and predictors of severity of chronic hepatitis C virus (HCV) and human immunodeficiency virus (HIV) co-infection. *J Hepatol* 2006; **44**:S28–S34.
74. Petrovic LM. HIV/HCV co-infection: histopathologic findings, natural history, fibrosis, and impact of antiretroviral treatment: a review article. *Liver Int* 2007; **27**:598–606.
75. Barreiro P, Blanco F, de Mendoza C, *et al.* Metabolic syndrome and progression of liver fibrosis in HIV/HCV-coinfected patients on HAART. *Antivir Ther* 2007; **12**:L64.
76. Wit F, Kesserling A, Gras L, *et al.* Incidence of hypersensitivity reactions associated with nevirapine-containing HAART in patients with prior treatment experience may differ from that in treatment-naïve patients: the ATHENA cohort study. *Antivir Ther* 2007; **12**:L4.
77. Tomlinson B. Drug evaluation: tesamorelin, a synthetic human growth hormone releasing factor. *Curr Opin Investig Drugs* 2006; **7**:936–945.
78. Falutz J, Allas S, Mamputu J-C, *et al.* Further data on the effects of tesamorelin (TH9507), a growth hormone-releasing factor analogue, on body composition and metabolic parameters in HIV-infected patients with abdominal fat accumulation. *Antivir Ther* 2007; **12**:L9.
79. Turner RR, Falutz J, Testa MA, *et al.* The impact of tesamorelin (TH9507), a growth hormone releasing factor analogue, on body image and health-related quality of life in HIV-infected patients with abdominal fat accumulation. *Antivir Ther* 2007; **12**:L33.
80. Fontdevila J, Berenguer J, Prades E, *et al.* Autologous fat grafts are safe and durable in HIV-infected adults with facial lipoatrophy. *Antivir Ther* 2007; **12**:L34.
81. Orlando G, Guaraldi G, Squillace N, *et al.* Effectiveness and long-term durability of autologous fat transplants for HIV-related face lipoatrophy. *Antivir Ther* 2007; **12**:L35.
82. Rector RS, Warner SO, Liu Y, *et al.* Exercise and diet induced weight loss improves measures of oxidative stress and insulin sensitivity in adults with characteristics of the metabolic syndrome. *Am J Physiol Endocrinol Metab* 2007; **293**:E500–E506.
83. Perez-Martin A, Raynaud E, Mercier J. Insulin resistance and associated metabolic abnormalities in muscle: effects of exercise. *Obes Rev* 2001; **2**:47–59.
84. Kadoglou NP, Iliadis F, Liapis CD, *et al.* Beneficial effects of combined treatment with rosiglitazone and exercise on cardiovascular risk factors in patients with type 2 diabetes. *Diabetes Care* 2007; in press.
85. Roberts CK, Ng C, Hama S, *et al.* Effect of a short-term diet and exercise intervention on inflammatory/anti-inflammatory properties of HDL in overweight/obese men with cardiovascular risk factors. *J Appl Physiol* 2006; **101**:1727–1732.
86. Duncan GE. Exercise, fitness, and cardiovascular disease risk in type 2 diabetes and the metabolic syndrome. *Curr Diab Rep* 2006; **6**:29–35.
87. Panunti B, Fonseca V. Effects of PPAR gamma agonists on cardiovascular function in obese, non-diabetic patients. *Vascul Pharmacol* 2006; **45**:29–35.
88. Takano H, Hasegawa H, Zou Y, *et al.* Pleiotropic actions of PPAR gamma activators thiazolidinediones in cardiovascular diseases. *Curr Pharm Des* 2004; **10**:2779–2786.
89. Reeds DN, Cade WT, Mondy K, *et al.* Pioglitazone with or without exercise training reduces liver lipid content and improves insulin sensitivity in HIV with impaired glucose tolerance (IGT). *Antivir Ther* 2007; **12**:L14.

Accepted for publication 23 August 2007