

Strategic Use of Antiretroviral Therapy

Michael S. Saag, MD

Over the last decade, highly active antiretroviral therapy (HAART) has revolutionized the care of human immunodeficiency virus (HIV) infected patients. Advances in the understanding of HIV pathogenesis, the routine use of viral load and resistance testing in clinical practice, and the availability of over 23 Food and Drug Administration (FDA) approved antiretroviral (ARV) agents (Table 4-1) have created the promise for most HIV-infected patients to live, and live well, for decades such that HIV ultimately has little impact on the duration or quality of their life.¹⁻⁵ Persistent, high-level viral replication is the driving force of HIV pathogenesis, with up to 10 billion virions produced in an infected individual each day.⁶⁻⁹ Viral load measurements enable the clinician to determine the degree to which an ARV therapeutic regimen is working and, more importantly, when the regimen is failing.¹⁰⁻¹⁴ This allows therapy to be switched at the time of ARV failure rather than at the time of clinical failure. Once a regimen begins to fail, clinicians can utilize resistance test information to choose from dozens of potential alternative regimens, utilizing both existing approved drugs and experimental therapeutic agents with novel mechanisms of action now in development.¹⁵ Through the contributions of all of these developments,

ARV Drugs Approved for Use in the United States as of 2006

Table 4-1	Year Approved	Agent (Trade Name)
	1987	Zidovudine (retrovir)
	1991	Didanosine (videx)
	1992	Zalcitabine (hivid)
	1994	Stavudine (zerit)
	1995	Saquinavir (invirase) Lamivudine (epivir)
	1996	Indinavir (crivivan) Ritonavir (norvir) Nevirapine (viramune)
	1997	Delavirdine (rescriptor) Nelfinavir (viracept)
	1998	Efavirenz (sustiva) Abacavir (ziagen)
	1999	Amprenavir (agenerase)
	2000	Lopinavir/ritonavir (kaletra)
	2001	Tenofovir DF (viread)
	2003	Atazanavir (reyataz) Emtricitabine (emtriva) Enfuvirtide (fusion)
	2004	Fos-amprenavir (lexiva)
	2005	Tipranavir (aptivus)
	2006	Darunavir (prezista) Miraviroc
	2007 (in expanded access)	Raltegravir Etravirine

clinicians have the potential to achieve long-term clinical benefits by keeping the viral load as low as possible for as long as possible.

The use of modern ARV therapies has led to a striking reduction in HIV-associated mortality.^{16–18} Despite these advances, newly recognized toxicities of ARV treatment have begun to limit the long-term benefits of chronic therapy.^{19–27} Moreover, the durability of the ARV effect is quite variable. Many factors influence the ability to sustain suppression of viral replication, including pharmacokinetic properties of the regimen, tissue penetration, cellular penetration, appropriate intracellular processing, ARV drug history, tolerability of the regimen, adherence, potency of the regimen, and the development of or preexisting presence of resistant virus.^{15,28–34} To achieve the most durable effect of ARV therapy (ART), clinicians must develop a strategic approach that maximizes the likelihood of success of each given regimen through a thorough understanding of the biology of HIV disease and the principles of ART.

THE BIOLOGY OF HIV INFECTION

Since HIV was identified in 1983, several landmark discoveries have helped elucidate the mechanisms by which HIV causes the immune system dysfunction associated with AIDS. These discoveries are usually linked to the application of newly developed technology in the laboratory. Soon after discovery of the virus, investigators demonstrated the presence of HIV in virtually all tissues of the body, including the brain.^{35,36} Utilizing p24 antigen assays, an association was made between the level of p24 antigen in plasma and the stage of disease, with higher levels of viremia observed during the time of acute seroconversion and again in the later stages of advanced HIV disease.^{37–39} Most of the individuals who were asymptomatic with high CD4⁺ T-lymphocyte counts had no appreciable p24 antigenemia detected. During the late 1980s, utilizing tissue culture techniques investigators were able to titrate the amount of infectious virus in plasma.^{37,40,41}

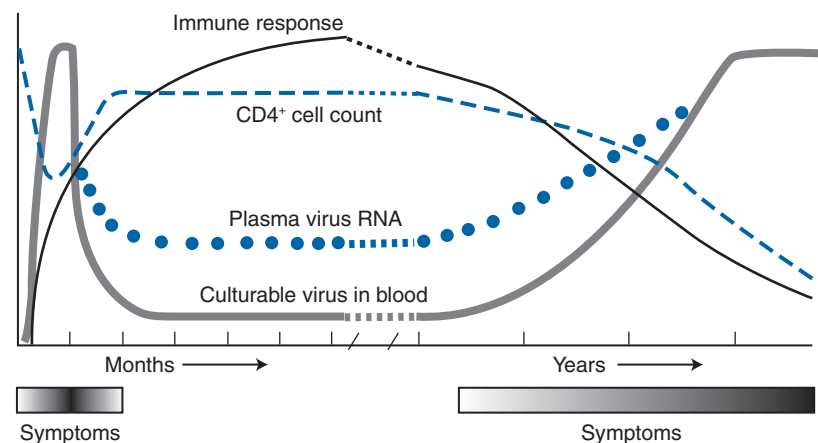
Much like p24 antigenemia, higher levels of infectious virus in plasma were noted at the time of acute seroconversion and again in later stages of disease.^{38,39,42} Although the plasma culture technique was more sensitive than the p24 antigen technique, substantial numbers of patients with asymptomatic disease, as well as those on ART, had undetectable levels of infectious virus.^{37,40,41}

Utilizing this information, a picture of HIV pathogenesis emerged whereby the virus established widespread infection early in the course of disease (at the time of seroconversion), stimulating a potent immune system response. During the period of clinical latency, which typically lasts up to 10–12 years, initial models of pathogenesis suggested that viral replication was under effective control by the immune system, only to reappear as high-level viremia during later stages of disease (Fig. 4-1).^{38,39,42} This model, however, could not explain the slow, yet progressive decline in CD4⁺ T-lymphocyte counts and immune system function that occurs during the period of clinical latency. With the advent of quantitative polymerase chain reaction (PCR) technology during the early 1990s, the association of HIV replication and immune system destruction was more completely described. Piatak and colleagues were the first to describe the presence of detectable virus at all stages of HIV infection, including the period of clinical latency.^{12,13} As was demonstrated with p24 antigen and quantitative plasma culture techniques, the highest levels of viral RNA were detected at the time of acute seroconversion and during later stages of disease.^{12,43} However, lower levels of virus were detected even at the early, asymptomatic stages of the disease, implying that viral replication is a continuous, ongoing process even during the period of clinical latency.

The application of viral load testing to determine the activity of ARV therapeutic regimens led to the opportunity to define further the nature of HIV replication *in vivo*. Even with the use of relatively weak ARV regimens, such as zidovudine monotherapy, an 80% (0.9 log) reduction in viral load was noted within 1 week of the initiation of therapy, with a relatively symmetrical return to baseline within 1 week after discontinuing treatment.⁴⁴ Based on these observations, it

Figure 4-1 ■ Natural history of HIV-1 infection over time.

Modified from Saag MA, Holodniy M, Kuritzkes DR, et al. HIV viral load markers in clinical practice. *Nature Med* 2:625, 1996. Copyright 1996 Macmillan Magazines Limited.



was apparent that viral replication was not only continuous but also quite rapid.⁴⁵ However, it was not until the dramatic responses to HIV protease and non-nucleoside reverse transcriptase (RT) inhibitors were observed that the magnitude of the rate of viral replication *in vivo* was fully appreciated.

In separate reports, Wei et al and Ho et al quantitated the rapid turnover of HIV *in vivo*, demonstrating the production and clearance of up to 10 billion virions each day (~400 million virions per hour).^{6,7} The half-life of virions in the circulation is estimated to be 1–2h or less. When new rounds of viral replication are blocked by ART, the amount of measurable virus drops by more than 99% within 2–7 days of initiating therapy. The estimated life cycle (or generation time) of HIV, which represents the time from release of a virion until it infects another cell resulting in the release of new progeny, is estimated to be 1–2 days (Fig. 4-2).^{8,46,47} These data were generated by observing the rapid decay in viral load over the first few weeks of potent therapy and represent the contribution of the acutely infected cells in the host, which have a half-life of 1–2 days. These cells represent more than 99% of the daily virus production. The remaining production of virus comes from a longer-lived population of cells that contribute to the slower, ‘second phase’ decay of plasma viremia (Fig. 4-3).^{47,48} Therefore within 8–12 weeks after initiation of potent therapy, plasma HIV RNA levels generally fall below 400 copies/mL of plasma. It usually takes several weeks longer to reach undetectable levels when utilizing ultrasensitive virologic techniques (limit of detection ~5–50 copies/mL). Most viral replication takes place in lymphoid organs, where most of the CD4+ T lymphocytes reside.^{28,49–54} The detection of virus in plasma, as measured for example by the plasma ‘viral load’, represents spillover of virus from the site of production (lymphatic tissue) into the bloodstream where it can be readily detected. In addition to the gradual reduction in absolute CD4+ T lymphocytes over time, loss of lymphoid architecture within lymph nodes also occurs

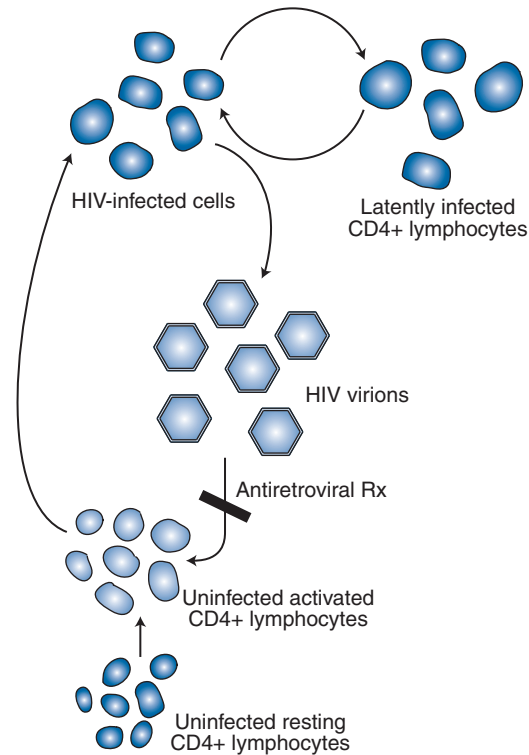


Figure 4-2 ■ HIV-1 replication is rapid *in vivo*. Plasma HIV-1 viremia, depicted here as HIV-1 virions results from spillover of recently produced virus from infected cells in lymphatic tissue. Uninfected, activated CD4+ T lymphocytes are the predominant target of HIV-1 infection. Once infected, these cells produce virus within 1–2 days and continue producing virus for an estimated 1–3 days. More than 99% of the virus detected in the bloodstream comes from recently infected CD4+ T lymphocytes. The remaining less than 1% of virus comes from chronically infected CD4+ T lymphocytes or macrophages, which have life spans ranging from a few days to several years.

Redrawn with modification from Perelson et al. HIV-1 dynamics *in vivo*: virion clearance rate, infected cell life-span, and viral generation time. *Science* 271:1582, 1996. Copyright 1996 American Association for the Advancement of Science.

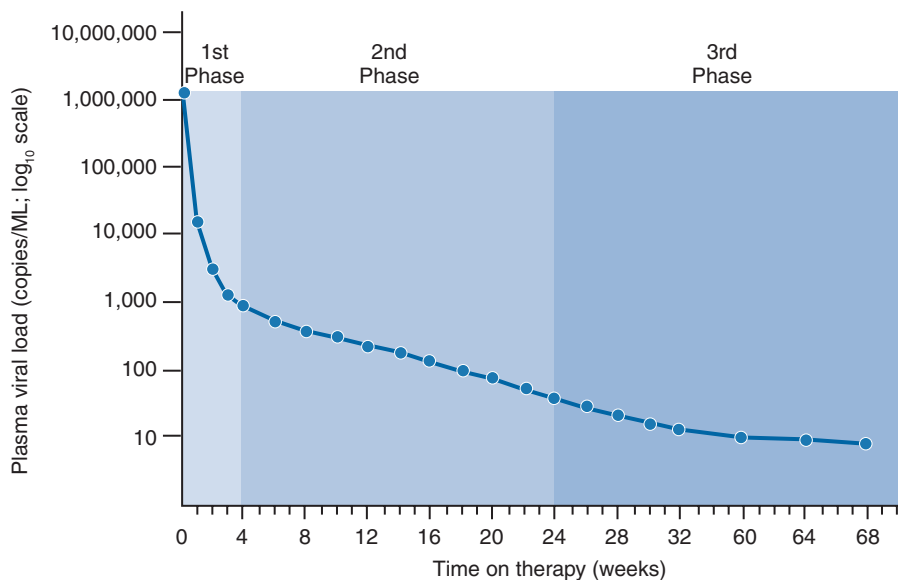


Figure 4-3 ■ Typical plasma HIV RNA response to potent ART. After initiation of treatment, a sharp decrease in viral load is observed over the first 2–4 weeks of therapy. Thereafter a steady, less steep decline in plasma viral load is observed. These differential rates of decline have been arbitrarily divided into ‘phases’ for purposes of mathematic modeling. In reality, they represent a continuum of viral decay as the life span of productively infected cells is exhausted. If all further new infection is completely blocked for a sufficient period of time (new estimates: many years) to allow all existing infected cells to die, a cure is theoretically possible.

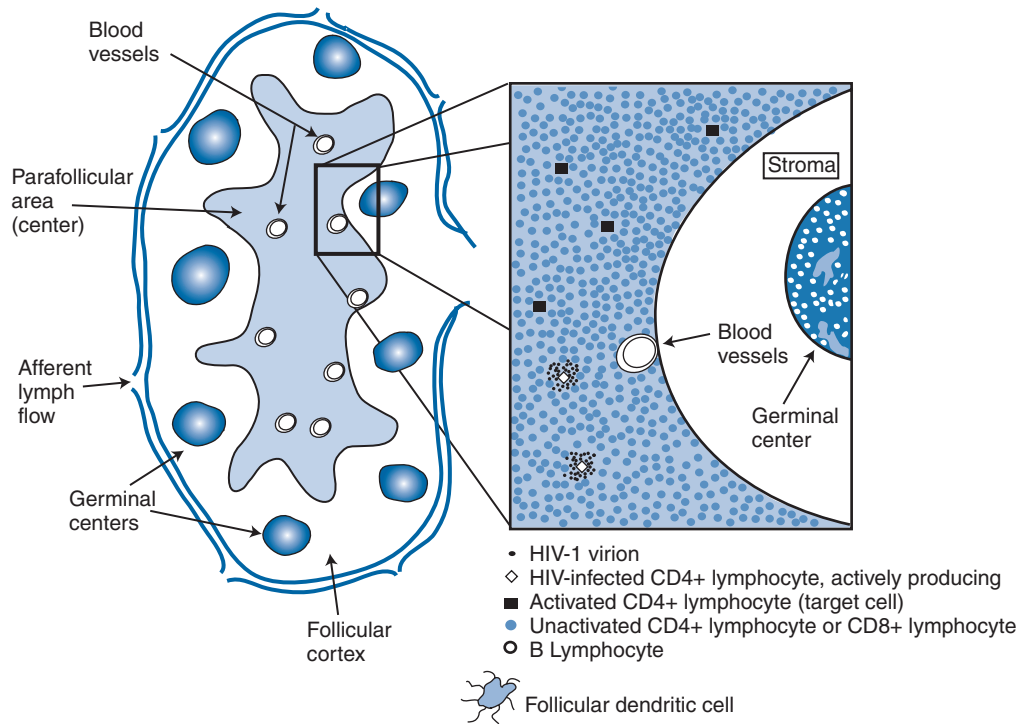


Figure 4-4 ■ Lymph node from a patient with established HIV infection. The actively producing cells (diamonds) excrete high levels of HIV virions (black dots), with an average instantaneous burst size of 4000 copies per cell. These newly produced virions are trapped by follicular dendritic cells (large cells in germinal center), are absorbed onto circulating resting CD4+ T lymphocytes (black closed circles) or activated CD4+ T lymphocytes (black squares) passing by, or spill into the circulation. The actual number of target (activated) cells infected by virus produced by any actively producing cell is small: about one new cell infected over the life span of each actively producing cell at steady state.

and is believed to contribute to the relative lack of immune system efficiency at later stages of the disease.^{49,50} Although most newly formed virions do not result in infection of a neighboring cell, on average several million new CD4+ T lymphocytes are infected each day.^{7,46} This high-level, continuous production of virus and subsequent infection of new CD4+ T lymphocytes helps explain how the CD4+ cells decline and immune system destruction occur during the time of clinical latency.

Activated CD4+ T lymphocytes are the principal targets of HIV replication. The estimated life span of an infected CD4+ T lymphocyte is 1–2 days.⁵⁵ Each infected cell has an instantaneous burst size of ~4000 copies per cell.²⁸ The calculated burst size per cell is remarkably constant and is independent of stage of disease, plasma viral load, and ARV treatment status.²⁸ Thus the plasma viral load is a direct reflection of the number of infected cells in the body producing virus at any moment in time.

Whereas the instantaneous burst size, defined as the number of measurable virions produced by an infected cell at any given moment, is ~4000 copies per cell, the effective burst size, defined as the number of viruses produced that result in productive infection of a neighboring cell, at steady state is ~1 virion per cell (Fig. 4-4). At steady state, each patient establishes an equilibrium between the production and

destruction of virus producing cells such that as each productive cell is destroyed or stops producing virus it is replaced by another newly infected, productive cell. Clinically, this equilibrium establishes a relatively stable viral load value over time, defined as the viral ‘set point’.^{8,56} Mellors and colleagues have demonstrated a direct relation between the viral load, or set point, and the rate of CD4+ T-lymphocyte count decline.⁵⁷ When viewed in the context of the direct relation between plasma viral load and the number of productively infected cells at any moment in time, these findings are not surprising. Yet, when this concept is applied to individual patients the ability of the plasma viral load to predict the rate of CD4+ T-lymphocyte decline for a specific patient is quite poor, indicating that other factors, as yet unmeasured, are playing a role in causing the loss of T cells in HIV-infected patients.⁵⁸

Much attention has focused on how actively producing cells are eliminated.^{59,60} The leading possibilities are the direct cytopathic effect of the virus (or deleterious effects of virions budding from the cell membrane), virus-induced cell apoptosis, or direct destruction via an effective immune system response. Initial experience with tissue culture growth of the virus supported the concept of a direct cytopathic effect.⁶¹ Under the stimulation of phytohemagglutinin and interleukin-2 (IL-2), most virus-infected cells died within several days of

becoming infected in tissue culture, often through the development of large syncytia. Syncytia have never been demonstrated *in vivo*, and so the concept of death of cells via this mechanism remains uncertain. Apoptosis, or programmed cell death, is a natural mechanism for eliminating lymphocytes of the immune system. Viral proteins, or cytokines released in response to viral infection, may trigger the apoptotic pathway in actively producing cells, although the degree to which this occurs is not known. What is clear, however, is the tremendous loss of CD4+ T lymphocytes in the gut during the first several days to weeks after initial infection.^{62–64} Studies in both animals (infected with SIV) and humans with acute HIV infection demonstrate profound loss of immune cells and disruption of normal architecture of the gut in the first several days after infection. These data suggest strong consideration of aggressive use of ART during the period of acute infection.

A large body of evidence supports the role of HIV-specific immune system responses to eliminate virus-producing cells.^{65–69} The early and rapid development of the HIV quasi-species is a direct consequence of HIV-specific immunity, both cellular and humoral. Neutralizing antibody studies have demonstrated a very dynamic virologic escape phenomena within the first several weeks of HIV infection, with persistent generation of novel viral variants in response to antibody responses over the life of infection.^{70,71} Enhanced HIV-specific immunologic responses were postulated to be responsible for the lower virologic set points observed among seroconverters who were treated within days to a couple of weeks after the onset of symptoms of their initial HIV infection and had therapy periodically withdrawn.⁷² High levels of anti-HIV-specific CD4+ T-lymphocyte activity were demonstrated in association with control of replication. This level of CD4+ T-lymphocyte help is typical in ‘long-term nonprogressor’ patients, who naturally control viral infection and do not suffer CD4+ T lymphopenia despite chronic HIV infection. In contrast, when therapeutic withdrawal of treatment was performed in patients with chronic, well-established HIV infection who were not treated within 12–20 weeks of acute infection, viral load values typically rebound back to high levels.^{73,74} These patients have extremely low or absent HIV-specific CD4+ T-helper lymphocyte responses. Longer-term follow-up of the treated acute seroconversion patients revealed a return to higher viral load set points indicating some loss of the initial immunologic priming from therapy. Of more concern, one patient in the initial intermittent therapy study became superinfected with virus from another patient following sexual exposure while off therapy. This story has created a challenge and some controversy for those working on development of a preventative HIV vaccine.

Taken together, HIV pathogenesis is best depicted as a vicious cycle of production of large numbers of HIV virions that infect activated CD4+ target cells, which in turn produce more viruses that infect additional cells (Fig. 4-5).⁹ The function of the targeted CD4+ T lymphocytes, ironically, is to create an HIV-specific, coordinated response against the virus that is attacking them. Current thinking supports the concept that cytotoxic CD8+ T lymphocytes, under the support of an

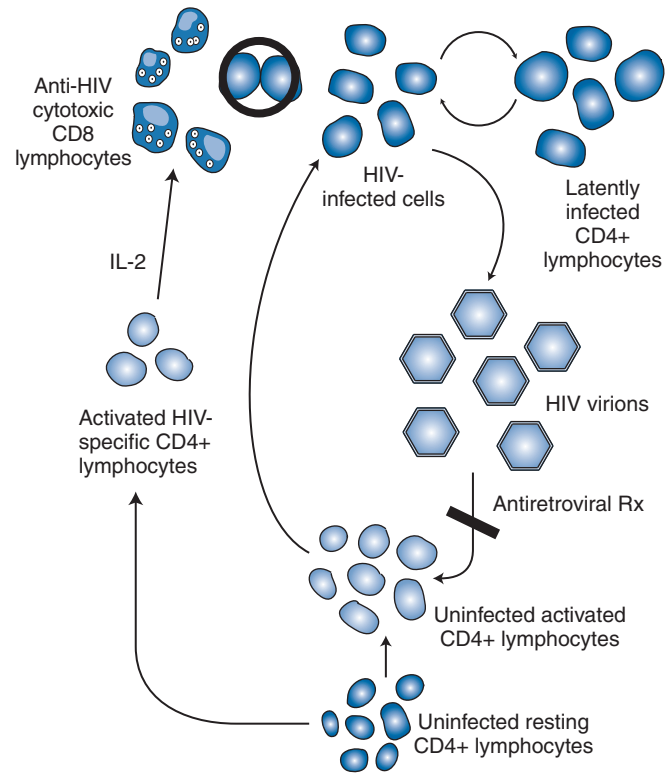


Figure 4-5 ■ More complete picture of HIV pathogenesis. Actively producing CD4+ lymphocytes are killed, at least in part, by HIV-specific CD8+ cytotoxic lymphocytes. These cytotoxic cells are stimulated by cytokines (e.g., IL-2) produced by HIV-specific CD4+ lymphocytes. In the presence of functioning CD4+ T lymphocytes, the inherent anti-HIV immune response is capable of controlling viral replication in an efficient manner. Unfortunately, early during the course of infection (soon after the time of seroconversion), most of the HIV-specific CD4+ activity is lost, and the immune response is much less efficient. Early treatment, prior to the time of seroconversion, helps preserve the HIV-specific immune response, potentially abrogating the need for ART.

HIV-specific CD4+ T-lymphocyte response, are responsible, at least in part, for eliminating the active virus-producing HIV-infected cells.^{68–72,75–78} The HIV-specific CD4+ T-helper lymphocyte responses appear to be lost early (within the first few weeks after infection), perhaps never to return. The continuous infection of other activated CD4+ cells is believed to lead to impairment of the immune responses to other antigens/pathogens, thereby creating potential deficits in the immune system’s response to opportunistic processes. The goal of effective ART therefore is to block, as completely as possible, the ability of the virus to infect uninfected CD4+ T lymphocytes, thereby inhibiting *de novo* production of virus and, at the same time, preserving immune system competence.⁶¹

STRATEGIC USE OF ART

Simply stated, the goal of ART is to inhibit completely viral replication *in vivo* and sustain the effect for as long as possible.

Theoretically, if complete suppression is sustained for a long enough time to allow the population of chronically infected cells to decay to extinction, eradication of HIV (or a true cure) is possible. Whether a cure is truly achievable is critical to establishing the foundation of an ARV therapeutic strategy. If cure is indeed possible, all patients should be treated with the most potent agents early and aggressively during the course of their infection, much like the treatment of acute lymphocytic leukemia. If a cure with ART alone is not a realistic possibility, other strategic approaches must be considered, more like the treatment of chronic lymphocytic leukemia.

Whether eradication is possible depends in large part on the life span of longer-lived, chronically infected cells and whether complete (or relatively complete) suppression of viral replication is achievable. About 99% of the plasma viremia is generated by actively producing CD4+ T lymphocytes with a relatively short half-life (1–2 days), the remaining 1% or less of plasma viremia is produced by longer-lived cells that previously were predicted to have an estimated average half-life of 14–28 days, although some of the cells could have a life span as short as 2–4 days or as long as more than 400 days (Fig. 4-2).^{46,48,79} If the life span of these long-lived cells is 28 days or less and all *de novo* infection is completely blocked, eradication may be achieved as early as 3–5 years after initiation of therapy^{46,47}; however, if the life span of the longer-lived chronically infected cells is substantially longer (on the order of 400 days) or if latently infected cells (that contain proviral DNA) are able to circulate, multiply or expand, and express virus at a later time, the time required for complete eradication of HIV could be on the order of decades and may not be achievable, even with complete block of *de novo* rounds of replication sustained over a prolonged period of time.^{50,67} Data generated during the latter portion of the 1990s demonstrated that the half-life of the chronically infected population of cells was not 14–28 days as originally assumed but, rather, 6 months at the shortest or over 44 months at the longest.^{29,60,80–82} Thus the time to eradication under the assumption of complete arrest of all *de novo* infection now ranges from 12 to well over 60 years of continuous, maximally effective ART.

As indicated above, the concept of eradicating HIV from an infected individual requires chronically infected cells to have a relatively short half-life but also requires complete block of all new rounds of *de novo* infection and this block must be sustained for a long enough time to allow all chronically infected, productive cells within the body to be destroyed or die off. To achieve eradication, inhibitory levels of ART must be maintained above inhibitory concentrations within all body compartments and within all susceptible target cells, ideally on a continuous basis. Thus a ‘cure’ for HIV infection assumes complete penetration of ARV drugs into the cells in all compartments of the body where the virus is replicating and strict adherence to the regimen for a period long enough to allow the infected cells to die off. An important issue in the discussion of a potential ‘cure’ relates to the relation of ‘undetectable’ levels of plasma virus to complete suppression of viral replication. ‘Undetectable’ levels of HIV

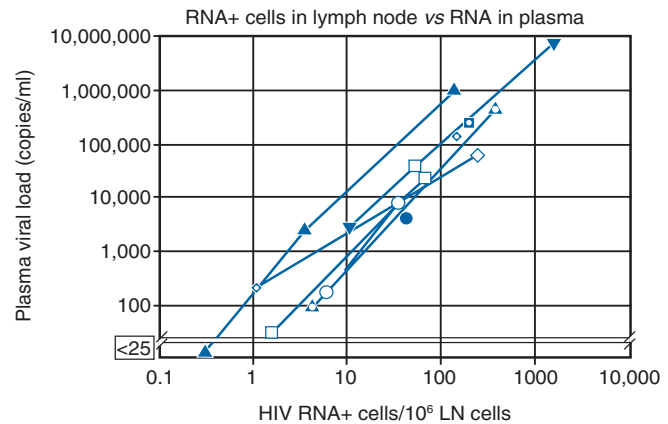


Figure 4-6 ■ A log–log plot of the relation between plasma viral load (copies per milliliter) and the number of cells actively producing virus in lymphatic tissue. Each point represents a separate biopsy time point paired with the plasma viral load obtained at the time of biopsy. The lines connecting points with the same symbol represent values obtained from the same patient before and after initiation of potent ART. The slope of this line is 1.6, and the Pearson correlation coefficient (R) is 0.95, indicating a strong relation between the number of cells producing virus in the body and the plasma viral load.

Modified from Hockett RD, Kilby JM, Derdeyn CA, et al. Constant mean viral copy number per infected cell in tissues regardless of high, low, or undetectable plasma HIV RNA. *J Exp Med* 189:1545, 1999.

in plasma are a function of a laboratory assay; complete suppression is a biologic phenomenon. All too often patients and clinicians assume that achieving an undetectable level of virus is synonymous with complete suppression. Even when plasma viral load values are less than a theoretical 1 log₁₀ copy/mL, an estimated 75 000–150 000 cells are still producing virus in lymphoid tissue (Fig. 4-6).²⁸ Zhang et al reported evidence of genetic drift in the envelope region in virus isolated from a patient who was treated at the time of seroconversion and sustained levels of less than 50 copies/mL for more than 20 months, supporting the notion of ongoing low-level replication when plasma viral load values are undetectable.²⁹ Therefore low-level viral replication appears to be ongoing even in the face of potent, yet incomplete viral suppression.⁵⁹ This implies that breakthrough viremia may ultimately occur in any given patient with sustained plasma HIV RNA levels below 50 copies/mL, although the likelihood of such breakthrough is a function of the probability of emergent resistant virus in the face of strong selective drug pressure (as discussed in the following section). Inevitably, more sensitive virologic assays will become commercially available and help narrow the gap between ‘undetectable’ and ‘complete suppression’. At the present time, however, it does not appear that currently available ARV agents, even when used in combination, are potent enough to achieve and maintain truly complete suppression. Thus a gap continues to exist between the ideal goal of ART (defined as complete suppression of *de novo* infection) and the achievable goal of ART (defined as

achieving undetectable levels of virus by available viral load techniques). With earlier, less potent regimens, many patients were unable to achieve undetectable levels of virus.^{83–85} In such circumstances, the ability to achieve undetectable levels of virus was more difficult among patients with higher baseline viral load levels and heavier treatment experience.^{86–92} However, with more potent and tolerable regimens available today, over 80–85% of patients are able to achieve <50 c/mL with their initial regimens and increasingly the <50 c/mL target can be achieved even among very heavily treatment experienced patients.^{93–95}

Taken together, evidence of chronically infected cells living for extended periods of time combined with evidence of ongoing, low-level replication in the face of ‘undetectable’ levels of plasma viremia indicates that eradication of HIV is not likely to occur with potent ART alone. Although eradication of HIV remains the ultimate goal of therapy, a more realistic goal at the present time would be to sustain suppression as completely as possible for as long as possible utilizing existing therapy over time.

Initiation of Therapy

The ultimate success of ART is total eradication, or cure, as discussed above. In the case of healthcare workers who have been exposed to infected blood through percutaneous needle-stick exposure, the use of ART appears to help abort early infection and, in this regard, is the best example of eradication occurring as a result of ART.^{96,97} Similarly, prevention of perinatal infection through the use of prepartum and peripartum treatment of the mother and postpartum treatment of the neonate is another example where early infection is either prevented or aborted after exposure through the use of ART.⁹⁸ Other than these two situations, success with ART in total eradication or cure has not been achieved. Rather, antiretroviral ‘success’ is defined more as the absence of ARV failure rather than as success *per se*. Therefore, prevention of ARV failure becomes the principal goal of ART.

With the development of a large number of more potent ARV agents, there has been a paradigm shift in the approach to therapy: the focus of therapy has changed from keeping patients alive from this year to the next to keeping patients alive from this decade to the next. The goals of chronic administration of ART can be seen as twofold: to prevent clinical progression and to prevent or delay development of resistance. Over the last decade the guidelines for treatment of HIV infection have vacillated, owing in large part to the emerging realization that a cure was not readily achievable with ART alone.⁹⁹ The ‘treat early, treat hard’ approach to therapy was initially linked with the idea that complete, maintained viral suppression could result in the eradication of HIV from the body within a short time.⁶¹ In addition, confusion regarding the goals of therapy at different stages of disease contributes to the apparent ‘change’ in guidelines. To prevent the emergence of viral resistance, relatively complete viral suppression is required. Therefore, in all newly treated patients the principal goal of treatment should be to achieve

virologic suppression to undetectable levels.^{1,100} While clinical benefit can still be realized with less than maximal suppression of viral load,^{83,89,101,102} use of more modern drugs has led to the emergence of a new treatment paradigm whereby the target of ARV therapy ideally should be <50 c/mL even in heavily experienced patients.^{1,4} Yet, for those patients who cannot achieve this degree of virologic suppression, a reduction in HIV RNA levels of 0.5 log₁₀ copies/mL below baseline is associated with relative maintenance of the CD4+ T-lymphocyte count over time, provided this degree of viral suppression is sustained.^{83,89,102} Therefore for patients who have experienced multiple regimens and cannot achieve <50 c/mL, the goal of therapy changes from prevention of further resistance mutations to prevention of clinical progression. In this setting, a reasonable target is to achieve and sustain viral load values at least 0.5 log₁₀ copies/mL, and preferably 1.0 log₁₀ copies/mL below baseline.

The ‘treat early, treat hard’ approach to therapy was grounded in concepts other than eradication.^{61,99} Most clinical trials confirm that the first treatment represents the ‘best shot’ at achieving profound suppression of viral replication. Based on current knowledge of HIV pathogenesis, early and profound suppression also prevents the development of resistance by limiting replication, preserving immune system integrity (before there is loss of critical clones of responsive cells), and creating a higher virologic hurdle for emergence of viral resistance.^{103–105} Although this rationale is clearly sound, the approach rests on assumptions concerning adherence, toxicity, pharmacokinetics/pharmacodynamics, and the absence of meaningful immune system recovery. First, complete adherence to complex ARV regimens is difficult for most patients to maintain.^{34,106,107} Multiple studies demonstrate a striking relation between adherence and success of ARV therapeutic regimens. Second, although serious toxicity is relatively uncommon with initial treatment for early disease, prolonged exposure to treatment is associated with a number of metabolic and hepatic complications.^{20,21,108–114} Third, drug pharmacokinetics and pharmacodynamics are subject to variability that can reduce the effectiveness of treatment.^{31,115} Not all patients are able to achieve and sustain similar levels of intracellular concentrations of drug or are able to metabolize drugs in a predictable fashion.^{32,116,117} Fourth, although treatment that is initiated late in the course of disease (e.g., at CD4+ T-lymphocyte counts of less than 100 cells/μL) is associated with a worse outcome, treatment started at moderate CD4+ T-lymphocyte counts (e.g., 350 cells/μL) can be accompanied by preservation of immune competence that does not seem to be clinically distinguishable from that seen when starting therapy at earlier stages (e.g., CD4+ T-lymphocyte count of 600 cells/μL).^{87,118,119}

There are additional considerations that argue against universal application of very early treatment.¹²⁰ Much of the data on progression of HIV disease from the Multicenter AIDS Cohort Study (MACS), which has been used to support earlier intervention, is derived from untreated individuals (Fig. 4-7A).⁵⁶ The use of data from untreated cohorts, although providing information on the natural history of

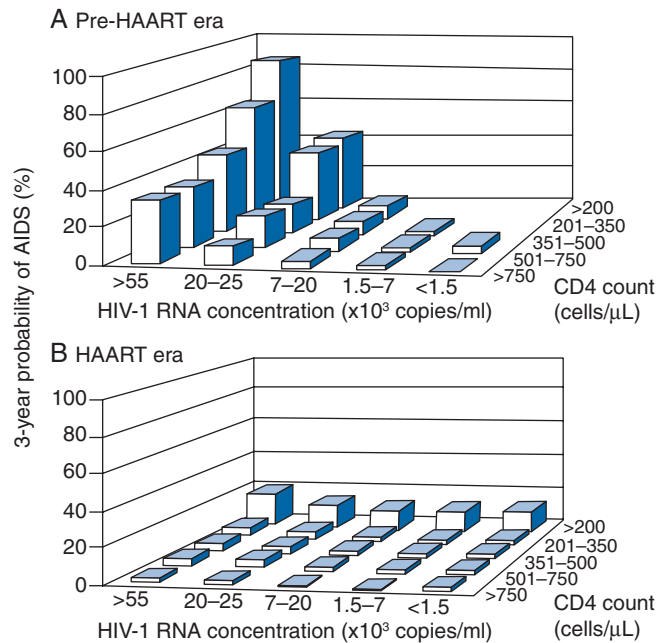


Figure 4-7 ■ Likelihood of progression to AIDS or death in the pretreatment (A) and treatment (B) era.

Used from Egger M, May M, Chene G, et al. Prognosis of HIV-1 infected drug naive patients starting potent antiretroviral therapy: a collaborative analysis of prospective studies. *Lancet* 360:119–29, 2002, as presented in US Department of Health and Human Services Panel on Clinical Practices for Treatment of HIV Infection. Guidelines for the Use of Antiretroviral Agents in HIV-1 Infected Adults and Adolescents. Washington, DC, DHHS. Available: <http://aidsinfo.nih.gov/Guidelines/GuidelinesDetail.aspx?MenuItem=Guidelines&Search=Off&GuidelineID=7&ClassID=1> 10 Oct 2006.

disease prior to the influence of ART, is anachronistic during the treatment era. Because the impact of treatment on the viral load determinants of progression is immediate, its predictive value is negated once potent treatment is initiated. Moreover, most clinicians do not idly observe progression of disease in their patients without introducing some intervention prior to the onset of profound CD4+ T-lymphocyte depletion or the development of clinical disease. More recent data sets derived from clinical cohorts of treated individuals are more valuable for the ‘treatment era’ (Figs 4-7B and 4-8).¹¹⁹ Data from the ART-Cohort Collaborative, a compilation of treatment experience of over 23 000 patients from over 50 clinical sites, demonstrate little short-term disease progression among patients who initiated therapy with more moderate CD4+ T-lymphocyte counts (e.g., 350–500 cells/ μ L), even if their viral load values at the time of treatment initiation are high (e.g., >100 000 copies/mL).^{87,118}

Based on such considerations, along with the increased frequency of long-term adverse events of treatment, a more conservative approach than ‘treat early, treat hard’ is advocated currently, although as therapies become better tolerated the recommendations continue to be in flux (Table 4-2).^{1,4} Among asymptomatic patients, treatment may be initiated relatively early (e.g., at CD4+ T-lymphocyte counts of

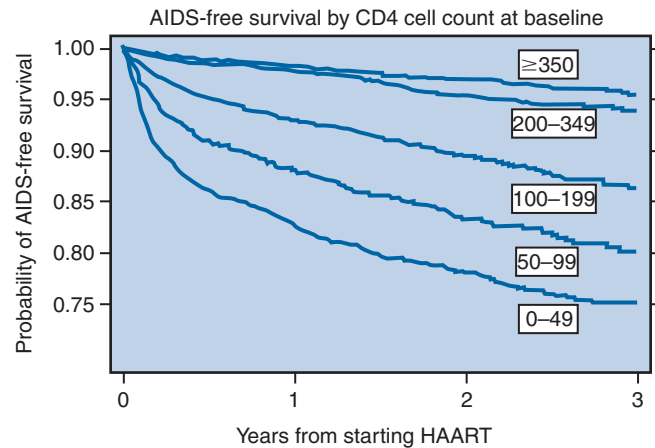


Figure 4-8 ■ Progression of HIV disease to AIDS or death by initial CD4 count.

Used from Egger M, May M, Chene G, et al. Prognosis of HIV-1 infected drug naive patients starting potent antiretroviral therapy: a collaborative analysis of prospective studies. *Lancet* 360:119–29, 2002.

350–500 cells/ μ L; while guidelines suggest consideration of treatment at these CD4+ T-lymphocyte counts, some experts now recommend treatment at this level) rather than very early (e.g., CD4 counts >500 cells/ μ L) using ARV combinations that are likely to reduce the viral load below 50 copies/mL.^{1,4,5} In contrast, all symptomatic patients should have treatment initiated regardless of CD4+ count and viral load. Similarly, all HIV-infected pregnant women should have treatment initiated by the beginning of the second trimester, regardless of viral load or CD4+ count, to eliminate the transmission of HIV to her baby. Once the mother has delivered, therapy can be stopped among those women who had initial CD4+ counts above 350–500 cells/ μ L and who are not breast-feeding (Table 4-2).^{1,4}

Selection of the specific regimen is based on the predicted likelihood of patient tolerance and adherence, with consideration of short- and long-term toxicities. The dramatically improved outcomes of more modern initial regimens, whereby over 85% of patients achieve target values of <50c/mL at 1 year of treatment, is due in large part to improved tolerability. Simply put, adherence is most directly related to drug tolerance. Medications and regimens that create even low-grade symptoms, such as intermittent nausea, mild headache, or occasional crampy diarrhea, ultimately lead to skipped doses. These skipped doses result in resolution of the offending symptoms, thus reinforcing the nonadherent behavior. Over time, skipped doses become more common, ultimately leading to the emergence of drug-resistant virus and regimen failure.

Drugs cannot work if they are not taken. Creating a regimen that is relatively easy for the patient to take therefore is essential when designing initial treatment (Fig. 4-9A). Moreover, frequently missed doses not only decrease the ability of the drugs to work, it is a recipe for the development of ARV resistance. A study comparing outcomes among prisoners versus patients who participated in clinical trials using the

Recommendations for Initiating ART in Treatment-Naive Adults^a with Chronic HIV Infection

Table 4-2

Parameter	Recommendation: IAS-USA ¹	Recommendation: HHS ⁴
Symptomatic HIV Disease	ART is recommended	ART is recommended
Asymptomatic HIV Disease		
CD4+ \leq 200 cells/ μ L	ART is recommended	ART is recommended
CD4+ <350 cells/ μ L but >200 cells/ μ L	ART should be considered ^b and the decision is individualized (see text); monitoring and counseling for HIV transmission prevention should continue if therapy is deferred	Treatment should be offered following discussion of pros and cons of therapy
CD4+ >350 cells/ μ L but \leq 500 cells/ μ L	ART is generally not recommended ^c ; monitoring and counseling for HIV transmission prevention should continue	VL > 100 000 c/mL, most clinicians recommend deferring ART, but some would treat; VL < 100 000 c/mL, defer therapy
CD4+ >500 cells/ μ L	ART is generally not recommended; monitoring and counseling for HIV transmission prevention should continue	VL > 100 000 c/mL, most clinicians recommend deferring ART, but some would treat; VL < 100 000 c/mL, defer therapy

^aNonpregnant adults.

^bThe closer the CD4+ cell count is to 200/ μ L, the stronger the recommendation, particularly if the plasma viral load is high (>100 000 copies/mL) or if the CD4+ count is declining rapidly (>100 cells μ L⁻¹ year⁻¹).

^cConsider treatment for patients with high plasma viral load or with rapid decline of CD4+ cell count.

Adapted from IAS-USA Guidelines 2006¹

identical regimens from the clinical trials, demonstrated that the prisoners, who had drugs administered under conditions of directly observed therapy, had substantially better virologic outcomes (90% vs 75% achieved viral load values of less than 400 copies/mL).¹²¹ These data underscore the importance of drug-taking behavior, commitment to the regimen, simplicity of regimens, and overall adherence to the success of the initial regimen.

Other considerations in the selection of the initial regimen include the availability of the treatments and, indirectly, their costs. In the past, selection of the initial regimen was made with a focus on keeping subsequent treatment options open in the case of failure of the first regimen, which was believed to be inevitable for most patients. However, with more modern regimens that have reduced pill burdens (some requiring only one pill once a day) and markedly improved tolerability, the concept of 'planning for failure' has been replaced with a strategy of 'planning for success', wherein regimens are tailored and adjusted for each patient on an individual basis. Once a regimen is initiated, frequent questioning regarding tolerance, for even mild or intermittent symptoms related to the regimen, should occur at each clinic visit and the regimen should be modified to eliminate the offending ARV agent. Most importantly, to ensure the best chance for success, patients should understand the rationale for treatment prior to treatment initiation and genuinely be 'ready to start' therapy.

The potency of the initial regimen is critical to long-term success. Regimens for initial treatment should be of sufficient potency to ensure a high likelihood of achieving viral load levels below the limit of detection (<50 c/ μ L). Preexisting drug resistant virus, which has been reported with increased

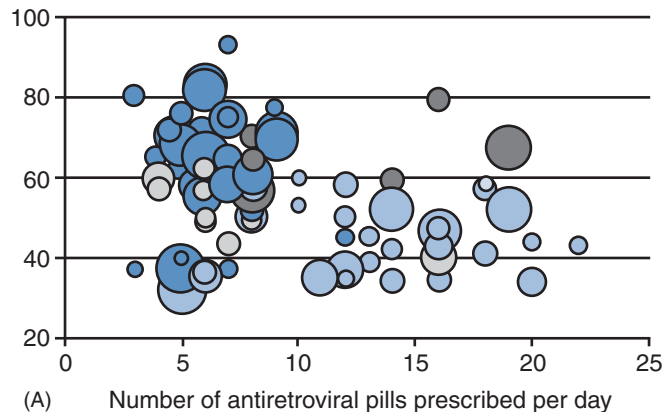


Figure 4-9 ■ Proportion of patients achieving virologic success based on number of pills required per day in the regimen (Pill Burden). These data were derived from a meta-analysis of 53 trials that enrolled 14 264 patients into 90 treatment arms. Each regimen evaluated contained two nucleosides plus either a boosted-BPI (grey), an NNRTI (dark blue), a third nucleoside (blue), or a nonboosted PI (very light blue). Overall 55% of patients had plasma HIV RNA levels <50 copies/mL at week 48. Pill count correlated with the percentage of patients with plasma HIV RNA levels <50 copies/mL at week 48 (A). In this figure, the size of the circles represents the standard error associated with the point estimate. Significantly greater numbers of patients receiving NNRTI (64%) and boosted-PI (BPI; 64%) had RNA <50 copies/mL at 48 weeks compared to those receiving triple nucleoside (54%) or nonboosted PI (43%) regimens. However, when the individual trials are listed, more NNRTI-based treatment regimens are associated with success (B).

Adapted from Bartlett JA, Fath MJ, Demasi R, et al. An updated systematic overview of triple combination therapy in antiretroviral-naive HIV-infected adults. *AIDS* 20:2051–64, 2006.

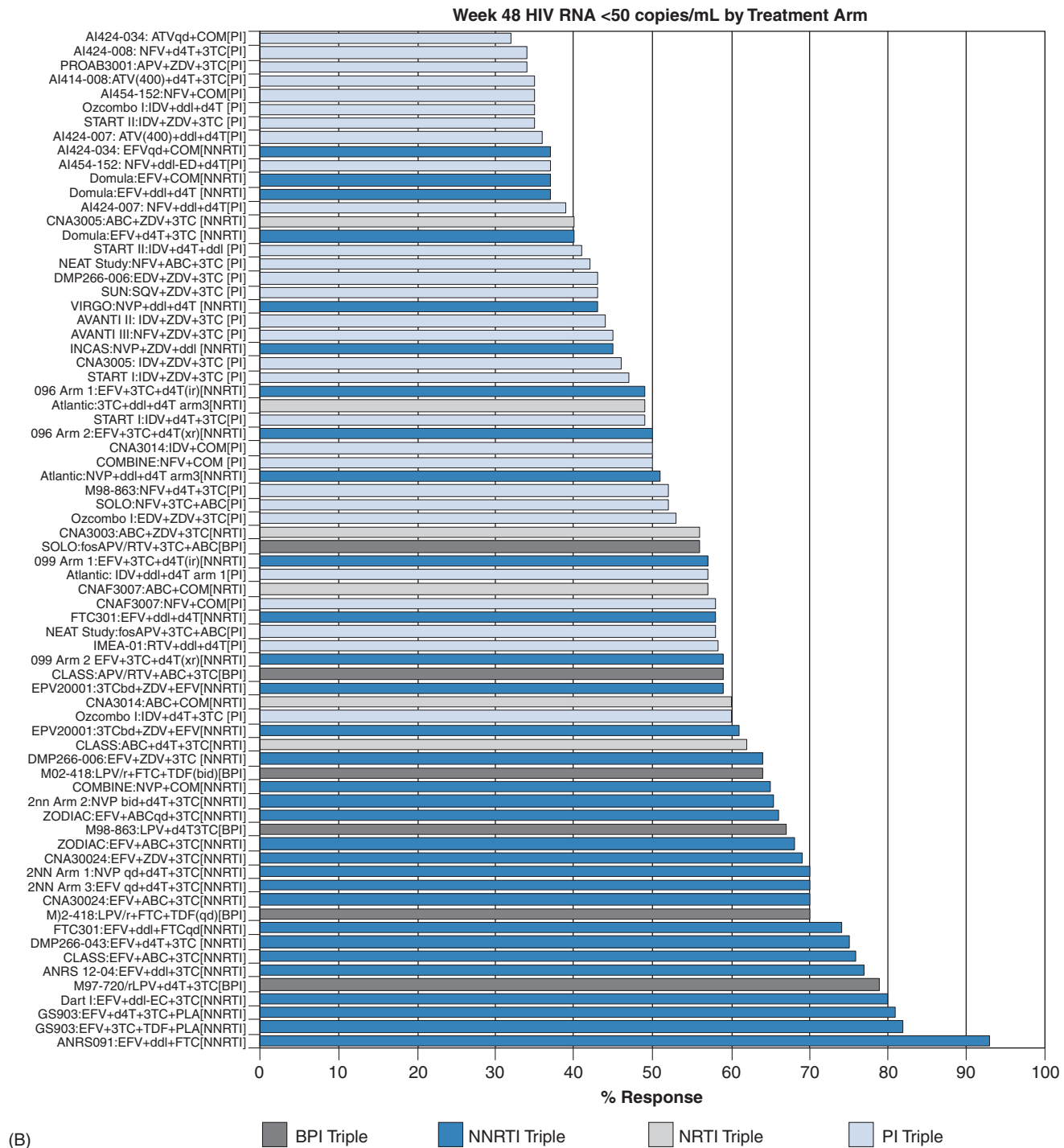


Figure 4-9 ■ Continued

frequency (between <3% and 14%), significantly hampers the potency of the initial regimen. Current guidelines suggest obtaining a pre-ARV treatment genotypic resistance test for any patient living in an area where a high prevalence (>5%) of resistant virus is known to exist, for patients presenting with acute seroconversion syndrome, and for pregnant women in selected cases.

Once a decision has been made to initiate therapy the proper choice of regimen is essential. Meta-analyses have

shown that efavirenz plus two nucleoside reverse transcriptase inhibitors (NRTIs) demonstrates ARV effectiveness activity comparable, if not superior, to that of ritonavir boosted protease inhibitor (PI)-containing regimens (Fig. 4-9B).^{30,122,127} However, ritonavir-boosted regimens are more likely to be associated with hypertriglyceridemia and possibly insulin resistance and lipodystrophy, reinforcing the trend for use of PI-sparing regimens as initial therapy. Unboosted PI-containing regimens are universally inferior to boosted-PI

Initial Treatment (HHS Guidelines⁺)

Table 4-3

Preferred Option	
NNRTI Option	NRTI Options
<ul style="list-style-type: none"> Efavirenz* 	Tenofovir + emtricitabine**
OR	OR
PI Options <ul style="list-style-type: none"> Atazanavir + ritonavir Fosamprenavir + ritonavir (BID) Lopinavir/ritonavir (BID) 	Zidovudine + lamivudine**
Alternative Options	
NNRTI Option	NRTI Options
<ul style="list-style-type: none"> Nevirapine[#] 	Abacavir + lamivudine
OR	OR
PI Options <ul style="list-style-type: none"> Atazanavir^{##} Fosamprenavir Fosamprenavir + ritonavir (1x/day) Lopinavir/ritonavir (1x/day) 	Didanosine + (emtricitabine or lamivudine)

⁺ As of October 2006; reference 4.

* Avoid in pregnant women and women with significant pregnancy potential.

** Emtricitabine can be used in place of lamivudine and vice versa.

[#] Nevirapine should not be initiated in women with CD4 counts >250 cells/mm³ or men with CD4 counts >400 cells/mm³.

^{##} Atazanavir must be boosted with ritonavir if used in combination with tenofovir.

and most non-nucleoside reverse transcriptase inhibitor (NNRTI) containing regimens and should be used only in selected clinical situations, such as in some active intravenous drug users, in those with significant hepatic compromise, or in patients requiring therapy for concomitant diseases where the drugs interact with ritonavir or NNRTI agents. In head-to-head studies, efavirenz has generally been more effective (by intent to treat) than nevirapine, although the outcomes were at the margin of statistical significance. Nevirapine should not be used in patients with higher CD4⁺ T-lymphocyte counts (>250 cells/ μ L for women; >350 cells/ μ L for men) owing to a higher risk of hepatic toxicity.²⁷ Efavirenz should generally be avoided in pregnant women, especially in the first trimester when the neural tube maturation is occurring. The preferred and alternative regimens according to the HHS guidelines are shown in Table 4-3.

Notwithstanding all of the considerations listed above, the most important issue related to the long-term success of ART is the degree to which the patient desires to start therapy.¹²⁸ Patients' 'buy in' is based in large part on their understanding of the rationale of treatment (i.e., why should they start) and the degree to which they believe they have a role to play

when selecting of the regimen. The 'best' choice of therapy must encompass not just potency, appropriate pharmacokinetics, minimal toxicity, and relative ease of administration, it must include strong consideration of how the regimen fits into the patients' day-to-day activities and their beliefs regarding the likelihood that the regimen will work. Taken together, the timing of therapy initiation is a complex decision process that requires an understanding of HIV biology, pathogenesis, natural history, pharmacology, toxicology, psychology, and behavior. In this regard, therapy is genuinely 'tailored' for each patient, making guidelines for the initiation of therapy entirely relative rather than absolute.

Management of ARV Failure

ART 'failure' is a relative term that can be defined in terms of both clinical and virologic parameters. Clinical failure is the easiest to define: progression of clinical symptomatology or the development of a new opportunistic infection or death in the face of ART. From a virologic standpoint, ARV failure simply means the loss of control of viral replication; however, this loss of control is often incremental and in most cases is

only partially lost. When viral load testing was first used in clinical practice, virologic failure was originally defined as a return toward (within 0.3–0.5 log¹⁰ copies/mL) the original baseline viral load.¹⁰ Soon thereafter the definition was more stringently defined as a confirmed return to detectable levels of virus (>200–500 c/mL) or the inability to achieve undetectable HIV RNA after 16–20 weeks of therapy.¹²⁹ This definition of failure was deemed most appropriate for a patient's first regimen, with the goal of therapy being driven predominantly by the desire to avoid the development of resistance. For patients who have experienced multiple regimen failures, the new target of therapy is the same as the first regimen. However, not all patients can achieve this level of suppression. Since the development of clinical progression (clinical failure) is infrequent provided the viral load level is maintained below 0.5 log¹⁰ copies/mL (threefold) of the original viral load set point,^{89,101} the definition of virologic 'failure' becomes dependent on the clinical setting and must be individualized for each patient, taking into account the history of ART, baseline viral load, response to therapy, and availability of options for treatment once 'failure' has occurred. Once the definitions of success and failure have been delineated, the choice of subsequent regimens depends on the prior exposure to ARV agents, previous adverse experiences, potential drug–drug interactions, underlying disease, CD4+ T-lymphocyte count/viral load status, and the perspective of the patient regarding the ability to adhere to a complex drug regimen. Fortunately, with the newer generation of drugs, the ability to achieve <50 c/mL even in experienced patients is easier than in the past.^{93–95}

The reasons for ART failure are multifactorial. The four primary reasons for failure include toxicity/intolerance, development of resistance, pharmacodynamics, and nonadherence. Each of these factors is discussed individually below.

Toxicity/Intolerance

Each pharmaceutical agent is associated with a unique set of toxicities. The degree to which a given toxicity is tolerated by the patient depends on a number of factors, including the nature and severity of the toxicity, the degree to which the toxicity interferes with day-to-day living, and the willingness of the patient to live with the toxicity. Generally, a patient who is totally asymptomatic is less willing to tolerate an adverse experience from a medication than a patient who has more advanced, symptomatic disease or has previously experienced disease progression. In addition, the degree to which a patient believes the regimen will provide benefit is proportional to the patient's willingness to tolerate an adverse experience.

With the use of combination therapy, the individual adverse experience profile for a given agent is compounded by drug–drug interactions, altered pharmacokinetic profiles, and synergistic toxicities of the drugs. Some adverse experiences develop soon after initiation of therapy, and the symptoms may wane after several weeks (tachyphylaxis). Depending on the nature of the toxicity, its severity, the tendency of the toxicity to develop tachyphylaxis, and the willingness of the patient to tolerate the toxicity, the decision to

discontinue a specific agent (or not) must be individualized. In general, intolerance reported on a patient's first regimen is of special concern owing to the association with nonadherence. Dose reduction or withholding a single agent for even a short period of time generally is not an option based on concern of inducing resistant virus to the agent while using a suboptimal dose or exposing the virus to a partially suppressive regimen with inadequate potency from the remaining agents in the regimen.

Development of Resistance

Resistance is a function of two principal conditions: (1) innate susceptibility of the virus (typically expressed in terms of 50% (IC⁵⁰) or 90% (IC⁹⁰) inhibitory concentration from an *in vitro* assay); and (2) the achievable level of drug in susceptible target cells, where viral replication takes place. In the case of antibacterial therapy, the difference between the *in vitro* susceptibility and the tissue concentration of drug is often as much as 100–1000-fold; however, in the case of antiviral therapy, the therapeutic window is generally much smaller. The maximum tolerated dose of most ARV agents limits tissue levels of drug to single-digit multiples of the IC₉₀ (e.g., three- to sevenfold) at trough levels. Therefore any genetic mutation that reduces the susceptibility of the virus to even a modest degree may have significant clinical implications.

To manage resistance clinically, the biology of how resistance develops should be understood. HIV exists *in vivo* as a quasi-species.¹³⁰ After initial infection with a predominant genotype, viral replication ensues at an extraordinary rate, resulting in the production of progenitor viruses that are highly related yet genetically distinct. The major reason for the generation of such enormous diversity is a combination of immune system pressure and the relative infidelity of the RT enzyme. RT is a relatively error-prone enzyme, creating a transcriptional error every 3000–4000 basepairs (bp) transcribed.^{131,132} Because HIV is a 9000-bp virus, on average one to two transcriptional errors occur with each replication cycle. With the generation of up to 10 billion virions per day, a wide variety of genetic mutants are theoretically produced on a daily basis. Most of these transcriptional errors are believed to lead to stop codons and defective virions, although critical point mutations that confer resistance to antiviral therapy may be generated. Under the conditions of selective pressure of either the immune system or drug therapy, these mutated viruses become the predominant genotype. Although most of these errors are neutral or lead to stop codons, several mutations may lead to the development of new virions with altered fitness or selective advantage.¹³³ When the mutation results in reduced fitness, the mutant virus grows less well and becomes a minor component of the quasi-species. However, when the mutant virus is more fit and has a selective growth advantage, it may dominate the quasi-species rapidly (within days).⁶ Therefore a major difference between antibacterial susceptibility testing and ARV testing becomes immediately obvious: antibacterial tests are performed against a single clone of bacteria isolated from an infected source, whereas ARV

tests are performed on a swarm of viruses that coexist in different frequencies and change in rapid fashion upon challenge with a new selective pressure (e.g., initiation of a new ARV regimen).

The likelihood of the development of resistance mutations *in vivo* is a function of two factors: the relative potency of the ARV regimen and the degree of ongoing replication occurring while the regimen is being administered. A regimen that has relatively poor potency creates little selective pressure on the virus to mutate; in such cases, resistance mutations are unlikely to develop (Fig. 4-10). In contrast, a highly active regimen generates substantial selective pressure for the virus to mutate. The likelihood of resistance development then becomes a function of the degree of replication allowed to occur while the regimen is being administered: the more replication allowed to occur under strong selective pressure, the faster resistance develops. Theoretically, if a regimen is completely suppressive, no resistance develops because no replication is taking place and resistance mutations do not have the chance to develop.

Virally encoded RT and protease enzymes, the two primary targets of most ARTs, are highly plastic molecules. Some of the mutations induced by one agent may lead to reduced susceptibility of the virus to other agents (cross-resistance), especially to agents with the same mechanism of action (e.g., NNRTIs and PIs). Conversely, some mutations that confer high-level resistance to one agent may increase the susceptibility of the virus to another, resulting in a so-called 'hypersusceptible' virus to the other agent.¹⁵ Additionally, many resistance-conferring mutations cause substantially less

efficient replication capacity than wild-type virus, resulting in a virus that is less 'fit' than wild-type virus.¹³³

Common mutations selected by NRTIs, NNRTIs and PIs are shown in Figure 4-11.¹³⁴ Although there is general concordance between laboratory-selected mutations induced by serial passage of the virus in the presence of drug and those mutations observed in clinical isolates, some mutations selected *in vitro* have not been identified from clinical specimens among patients receiving drug.¹⁵ In either case, mutations can be viewed as primary or secondary. Primary mutations are defined as those that have a demonstrable effect on the degree of resistance to the given agent. Secondary mutations often have no discernible effect on the susceptibility of the virus to the drug but are likely selected on the basis of their ability to improve the fitness of the virus, usually as a compensatory mutation that allows more effective viral replication or enzyme function (*see* Chapter 28).

Variable degrees of resistance are conferred by single point mutations, depending on the specific agent and the ease of resistance development. For example, a single point mutation at amino acid position 181 of the RT enzyme (e.g., Y181C) can lead to profound reduction in susceptibility to most NNRTIs, whereas single amino acid changes in RT (e.g., M41L or T215Y) may lead to only partial reduction in susceptibility to zidovudine.^{135,136} Upon development of subsequent mutations, the degree of resistance increases incrementally, ultimately resulting in high-level resistance when four or five resistance-conferring mutations are present.^{137,138} A similar pattern of sequential mutation acquisition within the protease gene product region is required for the development of high-level PI resistance.^{139,140} To complicate matters, although the sequential acquisition of mutations may be common among agents within a particular class, the precise mutation responsible for conferring resistance is generally agent-specific with varying degrees of cross-reactivity (depending on the drugs and the mutation in question). As an example, a single point mutation at amino acid position 30 (D30N) of the protease gene product leads to a several-fold reduction in susceptibility of the virus to nelfinavir but not other PIs.^{141,142} Subsequent acquisition of other mutations, at amino acid positions 82 or 84 of the protease gene product leads to high-level resistance to nelfinavir and other PIs as well. To complicate matters further, some point mutations create secondary changes in the functional gene product, leading to improved fitness or some degree of reversal of resistance.¹³³ In the case of lamivudine and entricitabine, an M184V mutation results in substantial reduction of the susceptibility of these drugs while partially reversing zidovudine resistance or delaying the emergence of zidovudine resistance when the two agents are used together in a regimen.¹⁴³ Taken together, drug-induced mutations occur under the influence of multiple factors, often resulting in variable phenotypic expression and clinical consequences.

In addition to *de novo* development of resistance, some viral variants with resistance-conferring mutations may pre-exist as subpopulations within infected T lymphocytes and macrophages.^{91,144} Under the conditions of strong selective

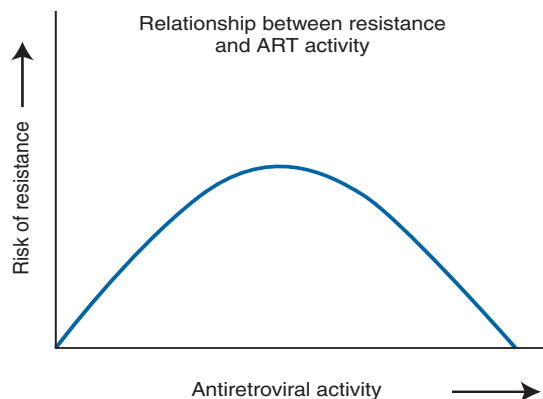


Figure 4-10 ■ Likelihood of developing resistance is directly related to the relative activity of ART. In the absence of meaningful activity (far left-hand side) there is no selective pressure exerted that can lead to resistance. Similarly, in the absence of viral replication (complete suppression of virus; far right-hand side), no resistance can develop because there is no ongoing replication. However, when regimens are only partially suppressive (middle portion of figure), there is ongoing replication in the presence of ample selective pressure, creating a higher likelihood of resistance.

Modified from presentations by Doug Richman and Emilio Emini.

A

MUTATIONS IN THE REVERSE TRANSCRIPTASE GENE ASSOCIATED WITH RESISTANCE TO REVERSE TRANSCRIPTASE INHIBITORS

Nucleoside and Nucleotide Reverse Transcriptase Inhibitors (nRTIs)

Multi-nRTI Resistance: 69 Insertion Complex (affects all nRTIs currently approved by the US FDA)

M	A	▼	K				L	T	K
41	62	69	70				210	215	219
L	V	Insert	R				W	Y	Q
								F	E

Multi-nRTI Resistance: 151 Complex (affects all nRTIs currently approved by the US FDA except tenofovir)

	A		V	F		F	Q		
	62		75	77		116	151		
	V		I	L		Y	M		

Multi-nRTI Resistance: Thymidine Analogue-associated Mutations (TAMs; affects all nRTIs currently approved by the US FDA)

M	D	K					L	T	K
41	67	70					210	215	219
L	N	R					W	Y	Q
								F	E

Abacavir

	K	L		Y	M
65	74		115	184	
R	V		F	V	

Didanosine

	K	L
65	74	
R	V	

Emtricitabine

	K		M
65			184
R			V
			I

Lamivudine

	K		M
65			184
R			V
			I

Stavudine

M	D	K				L	T	K
41	67	70				210	215	219
L	N	R				W	Y	Q
							F	E

Tenofovir

	K	K
65	70	
R	E	

Zidovudine

M	D	K				L	T	K
41	67	70				210	215	219
L	N	R				W	Y	Q
							F	E

B

Nonnucleoside Reverse Transcriptase Inhibitors (NNRTIs)

Efavirenz

		L	K	V	V		Y	Y	G		P
100	103	106	108			181	188	190		225	
		I	N	M	I		C	L	S		H
							I	A			

Etravirine (expanded access)

	V	A	L	K	V		V	Y	G
90	98	100	101	106		179	181	190	
	I	G	I	E	I		D	C	S
			P				F	I	A
							V		

Nevirapine

	L	K	V	V		Y	Y	G
100	103	106	108		181	188	190	
	I	N	A	I		C	C	A
			M			I	L	
							H	

Figure 4-11 ■ Common point mutations conferred by use of PIs (A), nucleoside RT inhibitors (B), and non-nucleoside RT inhibitors (C), mutations associated with decreased susceptibility to PIs. For each amino acid residue listed, the letter above the listing represents the wild-type virus, and the letter below the listing represents the mutation. Amino acids: A, alanine; C, cysteine; D, aspartate; E, glutamate; F, phenylalanine; G, glycine; H, histidine; I, isoleucine; K, lysine; L, leucine; M, methionine; N, asparagine; P, proline; Q, glutamine; R, arginine; S, serine; T, threonine; V, valine; W, tryptophan; Y, tyrosine. For full details of footnotes refer to chapter.

Reprinted from the International AIDS Society-USA. Johnson VA, Brun-Vezinet F, Clotet B, et al. Update of the drug resistance mutations in HIV-1: fall 2006. Top HIV Med 14:125–30, 2006. Updated information and thorough explanatory notes is available at <http://www.iasusa.org>.

C MUTATIONS IN THE PROTEASE GENE ASSOCIATED WITH RESISTANCE TO PROTEASE INHIBITORS

Atazanavir +/- ritonavir	L 10	G 16	K 20	L 24	V 32	L 33	E 34	M 36	M 46	G 48	I 50	F 53	I 54	D 60	I 62	I 64	A 71	G 73	V 82	I 84	I 85	N 88	L 90	I 93
	I F V C	E M I T V	R I		I F V	I Q L V			I L	V	L Y	L V M T A		E V L M V	V L T L	V C S T A		A T F I	V V S	M	L		M	
Fosamprenavir/ ritonavir	L 10				V 32				M 46	I 47	I 50	I 54					G 73	L 76	V 82	I 84			L 90	
	F I R V				I				I L	V	V	L V M					S V	V	A F S T	V			M	
Darunavir/ ritonavir	V 11				V 32	L 33			I 47	I 50	I 54						G 73	L 76	I 84			L 89		
	I				I F			V	V	M L						S V	V	V	V			V		
Indinavir/ ritonavir	L 10	K 20	L 24		V 32	L 36			M 46			I 54					A 71	G 73	L 76	V 77	V 82	I 84	L 90	
	I R V	M R	I		I	I			I L		V	V					V T	S A	V I	A F T	V		M	
Lopinavir/ ritonavir	L 10	K 20	L 24		V 32	L 33			M 46	I 47	I 50	F 53	I 54				L 63	A 71	G 73	L 76	V 82	I 84	L 90	
	F I R V	M R	I		I F				I L	V A	V L	V L A M T S					P T	S V	V	A F T S	V		M	
Nelfinavir	L 10			D 30		M 36			M 46								A 71		V 77	V 82	I 84	N 88	L 90	
	F I			N	I				I L								V T		I A F T S	V D S			M	
Saquinavir/ ritonavir	L 10		L 24							G 48		I 54		I 62			A 71	G 73	V 77	V 82	I 84		L 90	
	I R V		I							V	V	L		V		V T	S	V	I A F T S	V			M	
Tipranavir/ ritonavir	L 1013	I 20	K		L 33	E 35	M 36		K 43	M 46	I 47		I 54	Q 58		H 69	T 74		V 82	N 83	I 84		L 90	
	V V	M R			F G I				T L	L V			A M V	E		K	P		L D V				M	

MUTATIONS IN THE ENVELOPE GENE ASSOCIATED WITH RESISTANCE TO ENTRY INHIBITORS

Enfuvirtide				G 36	I 37	V 38	Q 39	Q 40	N 42	N 43
				D S	V	A M E	R	H T	D	
Maraviroc										

MUTATIONS IN THE INTEGRASE GENE ASSOCIATED WITH RESISTANCE TO INTEGRASE INHIBITORS

Raltegravir (expanded access)		Q 148	N 155
		H K R	H

Amino acid abbreviations: A, alanine; C, cysteine; D, aspartate; E, glutamate; F, phenylalanine; G, glycine; H, histidine; I, isoleucine; K, lysine; L, leucine; M, methionine; N, asparagine; P, proline; Q, glutamine; R, arginine; S, serine; T, threonine; V, valine; W, tryptophan; Y, tyrosine.

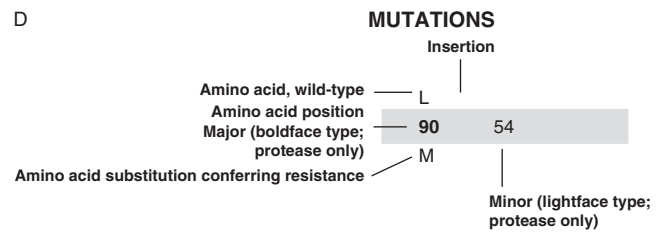


Figure 4-11 ■ Continued

pressure, these preexistent mutants may rapidly become the predominant species expressed *in vivo*. The best example of this phenomenon is the development of rapid resistance observed when NNRTIs are administered as monotherapy.^{6,144} Resistant viral variants have been observed as early as 5 days after the initiation of NNRTI monotherapy, with complete conversion of the predominant genotype in plasma from wild-type to resistant within 14–28 days after initiation of therapy.⁶ Monotherapy with NRTIs (e.g., lamivudine) typically emerges within 2–4 weeks of therapy and for PIs (e.g., indinavir) within 4–12 weeks of treatment initiation.^{6,145}

The concept of preexistent viral mutants plays a critical role in the sequencing of ARV regimens and the ability of a new regimen to succeed after a previous regimen has failed. At the time a regimen fails because of the development of resistance, resistant genotypes predominate in the plasma. As the regimen is changed, if these virions are susceptible to the new regimen, a shift in the population generally occurs, with suppression of the previous resistant viruses (ideally to undetectable levels).^{6,144} If the new regimen is not completely suppressive, some degree of replication continues, potentially utilizing the preexisting resistant mutants as substrates for the ongoing production of virus. This may lead to the creation of individual virions with multiple resistance mutations. This scenario is most likely to occur in the context of so-called sequential monotherapy, whereby a single agent is added to an existing failing regimen. A common example of sequential monotherapy is the addition of indinavir to a failing regimen of zidovudine/lamivudine. In this case, resistance mutations to zidovudine and lamivudine already exist at the time indinavir is added. If the new regimen is only partially suppressive, mutations that confer resistance to indinavir begin to appear sequentially on the background of zidovudine and lamivudine resistance mutations, potentially leading to multi-drug-resistant virions.¹³⁹

The development of resistance is often an incremental process, perhaps best thought of as a function of chance; that is, the likelihood of resistance development is directly related to the degree of ongoing replication and the chance that a resistance mutant may be precisely the virus that infects a susceptible, activated CD4+ lymphocyte passing by at that moment in time. Even with regimens that are highly potent, yet not completely suppressive, a single point mutation may occur that confers partial reduction in susceptibility.^{139,146} This results in a slightly higher degree of viral replication, thereby increasing the likelihood of the development of a second resistance-conferring mutation, leading to even further reduction in susceptibility and increased replication in the face of drug selective pressure. Therefore mutations occurring after the first mutation often appear more rapidly because of the higher degree of replication that occurs after development of the first mutation. The resultant end-product is a highly resistant mutant that has multiple coexisting resistance mutations. From a clinical perspective, the more rapid acquisition of subsequent mutations suggests that regimens should be changed early in the course of virologic failure to avoid the development of higher-level resistance,

assuming that viable therapeutic options exist for the patient in question.

Pharmacologic Aspects

For ARV drugs to have activity *in vivo*, the drugs must be absorbed and delivered to the site where viral replication is occurring (e.g., CD4+ T lymphocytes) and be appropriately processed by the target cell into an active moiety (e.g., the triphosphate derivative of nucleosides). The relative amount of drug in plasma may or may not reflect the amount of drug at the active, intracellular site of replication; therefore measuring plasma levels may only partially describe the relative activity of an agent. Just like toxicity profiles and resistance patterns, each drug has its own unique characteristics of absorption, metabolism, tissue penetration (including penetration into various body compartments, such as the central nervous system), intracellular processing, intracellular half-life, and mechanism of elimination (including the first-pass effect; *see* Chapter 79). Not only is the pharmacologic profile unique for each agent, but the absorption and metabolic processing may vary from patient to patient. This interpatient variability, due to weight, volume of distribution, hepatic metabolism, renal function, or genetic polymorphisms influencing drug absorption and clearance, becomes critical when interpreting data from studies that involve large populations of patients receiving combination ARV regimens. Even when patients within a given population adhere completely to the regimen, a fixed-dose regimen of a drug given every 8 h may lead to adequate concentrations at the target site for most individuals but be too low to sustain complete suppression in some or too high in others, resulting in early drug failure or excess toxicity, respectively.¹⁴⁷ Because most ARV agents have narrow therapeutic windows, these issues become critical when trying to implement a strategy of complete suppression of viral replication for all patients. Thus many current studies focus on measuring drug levels as part of a strategy to achieve improved virologic outcome with less toxicity. To date, not enough data have been generated to recommend this approach in clinical practice.

Adherence

To achieve and maintain complete suppression of viral replication as a goal of therapy, patients must take all of the agents comprising a combination regimen at the prescribed time and under the proper conditions. Because of drug–drug interactions and the interference of food with the absorption of some agents, many patients must adhere to complicated regimens that require careful planning of meals, sometimes in conjunction with taking multiple tablets at different times during the day. Even the most dedicated and committed patients find it difficult to remember to take each pill as prescribed every day over several months to years. Based on experience with other chronic diseases, such as hypertension, up to one-third of patients are able to adhere to their regimen 90% of the time, but most patients adhere poorly or

intermittently.^{148–150} For diseases in which the consequences of nonadherence result in some degree of morbidity, such as the use of insulin in brittle diabetics, the adherence is much better but still not 100%. In most instances, HIV infection is more like hypertension than diabetes; that is, most individuals with HIV infection are relatively asymptomatic and even those with symptoms usually do not experience the consequences of missed doses of their medicines. Because the objective of therapy is to maintain complete suppression and to achieve this objective drug levels must remain above the IC₉₀ at the target site throughout the entire viral life cycle, intermittent dosing that allows some degree of replication to occur in the face of high selective pressure becomes a recipe for the development of resistance and ultimate ARV failure.

Several factors may contribute to poor or intermittent adherence. The most common factor is poor instructions given to the patient regarding the regimen and its possible adverse experiences.¹⁵⁰ The development of side effects and their severity have a significant impact on a patient's willingness to take medications as prescribed or to continue a given regimen. The degree to which a patient is willing to tolerate adverse effects of medications is related to several factors, including the nature of the side effect, how much it interferes with the patient's ability to carry on daily activities, the severity of the patient's underlying condition (the more serious the disease, the higher the rate of adherence), and the patient's belief that the regimen is likely to be effective.¹⁵¹ Even mild or intermittent symptoms, such as low-grade nausea or headache, can lead to skipped doses that result in reinforcement of skipped dose behavior, especially among patients asymptomatic to their HIV infection. Regimens given once or twice daily have a higher degree of adherence, and therefore effectiveness, than three or four times a day regimens or those that require strict timing of drug administration in relation to meals (Fig. 4-9A). Other factors, such as level of education, socioeconomic status, and underlying substance abuse, generally do not predict adherence behavior.³⁴

STRATEGIES FOR CHANGING THERAPY

Failure of ART can generally be divided into two categories: failure resulting from toxicity and that resulting from virologic escape. The strategies for managing toxicity and virologic escape are quite different and are discussed separately.

Strategies for Changing Therapy because of Toxicity

Toxicity may be due to a single agent in a regimen or to multiple agents. Adverse events may manifest either through the inherent toxicity of the offending agent(s), through additive toxicity between two or more agents, or through adverse drug–drug interactions. Even though most of the approved agents have been in widespread use for only a few years, the most common adverse experiences are well established and fairly distinctive for each drug. In cases where a side effect

occurs that is commonly associated with a particular drug in the regimen, it is relatively easy to ascribe the toxicity to the most likely offending agent and adjust the regimen accordingly. However, in other cases, overlapping toxicities or uncommon side effects occur that are difficult to ascribe to a particular drug. In such cases it is best to stop the entire regimen, wait for the adverse effect to abate or decrease in severity, and reinstate therapy with a new regimen that substitutes one or more new drugs for the most likely offending agent(s).

When adjusting regimens because of toxic effects, several guidelines should be followed. When toxicities are noted within 2–4 weeks after initiating a new regimen, the most likely offending agent can be removed and a new drug without overlapping toxicity added to the remaining drugs in the regimen. For example, the development of rash from nevirapine usually occurs 14–28 days after initiating the regimen, in which case another agent of similar potency can be substituted into the regimen without changing the nucleoside backbone. Similarly, early manifestations of distal symmetric peripheral neuropathy (DSPN) in patients who are taking didanosine or stavudine or anemia among patients taking zidovudine should prompt the substitution of another NRTI for the offending agent. When toxicity to an agent develops weeks to months after the regimen had been initiated and the virologic response has achieved and maintained undetectable levels of virus, substitution of a new drug for the most likely offending agent is still an appropriate approach. In this case it is assumed that the existing regimen has suppressed viral replication sufficiently to prevent the development of resistance-conferring mutations to the drugs remaining in the regimen as the regimen is changed. Perhaps the best example of this is the development of metabolic or lipid abnormalities among patients taking a PI-containing regimen who replace the PI with an NNRTI agent while maintaining the nucleoside backbone (Table 4-4). Multiple small studies have been reported demonstrating successful maintenance of virologic control with improvement in lipid profiles but variable success in correcting insulin resistance or body composition abnormalities. Conversely, when side effects are noted weeks to months after starting a regimen that has not successfully achieved an optimal virologic response, it is best to change at least two, and possibly all, of the agents in the regimen (including the drug most likely responsible for the adverse effect) based on resistance testing results because of concerns regarding rapid development of resistance to a new drug when it is added as 'sequential monotherapy'.

Strategies for Changing Therapy because of Virologic Failure

Virologic failure may result from an inability to achieve the desired level of viral suppression initially or from the return of plasma HIV RNA to unacceptable levels after having achieved and sustained the targeted degree of viral suppression for months or years previously. In either case, the existence of the undesirable plasma HIV RNA value should be confirmed with repeat testing before any change in therapy

Effects of Switching from Initial Regimens to Alternative Regimens

Table 4-4

Study	No.	Follow-Up (week)	TGs	Chol	Glu/IR	Body Change	Comments
2NRTI + PI to 2NRTI + efavirenz ¹⁵⁴	33	40	NC	NC	NC	NC	Subset analysis of a cohort of 624 patients evaluated for body fat, lipid, and glucose abnormalities
2NRTI + PI to 2NRTI + efavirenz ¹⁵⁵	39	24	<↓	NC	NC	NC	Virologic control maintained. Modest increase in HDL-cholesterol
2NRTI + PI to 2NRTI + efavirenz ¹⁵⁶	43	24	<↓	NC	-	NC	Viral load remained <50 copies/mL in all patients. HDL-cholesterol unchanged
2NRTI + PI to 2NRTI + efavirenz ¹⁵⁷	25	24	<↑	NC	-	NC	Randomized to NVP, EFV, or control. Only one patient had rebound VL in NEV group vs 2 EFV vs 1 PI
2NRTI + PI to 2NRTI + efavirenz ¹⁵⁸	25	24	<↓	<↓	↑	<pVAT	All patients remained at <500 copies/mL 2NRTI + efavirenz ¹⁴³
2NRTI + PI to 2NRTI + efavirenz ¹⁵⁹	165	24	-	NC	-	-	Improvement in HDL-cholesterol in EFV group
2NRTI + PI to 2NRTI (+ABC) + efavirenz ¹⁶⁰	27	36	<↑	<↑	<↑	NC	Some overall fat loss by BIA (2.5 kg), but no change in symptoms of fat redistribution. Virologic failure in one patient
2NRTI + PI to 2NRTI ¹⁶¹	56	24	↑	↓HDL _L	-	NC	No virologic failure. Some increase in lipotrophy (five patients)
2NRTI + PI to 2NRTI + efavirenz ¹⁶²	45	48	↑	<↑	-	-	Virologic failure in two patients
2NRTI + PI to 2NRTI + efavirenz ¹⁶³	20	24	NC	NC	NC	NC	No virologic failures. Subjective improvement in morphologic appearance but no change in anthropometric studies
2NRTI + PI to 2NRTI + efavirenz ¹⁶⁴	93	52	↑	NC	↑	↑WHR ↑VAT	Switch (46) vs controls (47) Moderate increase in HDL with EFV; no difference in VL outcome. SQ fat loss no different
2NRTI + PI to 2NRTI + efavirenz ¹⁶⁵	41	52	-	-	NC	-	Patients with lipodystrophy syndrome; only IR and Glu tolerance evaluated
2NRTI + PI to 2NRTI + EFV or NVP ¹⁶⁶	100	52	↑	↑	-	NC	VL suppression maintained in 80%. No difference between EFV and NVP groups

Comparison of Genotypic and Phenotypic Resistance Assays

Table 4-5	Issues	Genotypic Assays	Phenotypic Assays
	Availability	Generally available	Restricted availability
	Time to results	Days	Weeks
	Technical issues	Straightforward	Demanding
	Susceptibility measurement	Indirect	Direct
	Sensitivity (minor species)	Poor	Poor
	Interpretation	Complex	Straightforward
	Cost	Moderately expensive	Expensive
	Major limitations	May not correlate with phenotype	Cutoff values poorly defined

is initiated. This point has been emphasized with results from many studies, where solitary increases in viral load ('blips') up to 1000 c/mL were noted among several patients who had otherwise maintained undetectable levels of virus.^{152,153} The blips were followed by a second value <50 c/mL and were not associated with an increased risk of long-term virologic failure or with an increased risk of developing resistance. Even patients who experience multiple isolated blips during the course of the therapy experience no apparent long-term adverse consequences.

Once an elevated plasma HIV RNA level has been confirmed, resistance testing can help identify which drugs, if any, may be contributing to the rebounding viral load (Table 4-5) (*see* Chapter 28).¹⁵ The absence of resistance mutations in the face of rising viral load implies problems with adherence or potential pharmacologic difficulties, such as poor absorption, increased metabolism (possibly due to a drug–drug interaction), decreased intracellular processing, or increased extrusion of drug from the intracellular compartment (e.g., as occurs with induction of P-glycoprotein pumps). Additionally, it is vital to confirm that the patient was still on the failing regimen at the time the sample for resistance testing was obtained, otherwise the viral quasi-species may have reverted back to wild-type falsely indicating that resistance mutations were not the cause of the virologic failure. When only one of the drugs in the regimen has induced detectable resistance mutations, changing only the single drug is acceptable; however, some consideration should be given to changing some of the other drugs in the regimen owing to the theoretical presence of low-level resistance to the other agents in the form of subpopulations. The activity of the next regimen should be at least as potent as the original regimen and ideally of more potency. In the absence of resistance testing, failure should be associated with the entire regimen rather than attempting to ascribe the failure to a particular drug in the regimen. In general, intensification with the addition of a single agent added to a failing regimen should be avoided except in circumstances where a substantial reduction in viral load has been achieved but not to undetectable levels within 16–20 weeks of initiating therapy. Even when these conditions are met, great care should be used to ensure that the persistent detectable viral load is not due to problems with adherence, which would be aggravated

with the addition of yet another pill to take. The addition of single new drugs, 'sequential monotherapy', is less likely to achieve either the targeted virologic effect or a durable response and is more likely to lead to the development of multidrug-resistant isolates. Conversely, in cases where adherence is judged to be the reason for failure, the substitution of ritonavir-boosted PIs (e.g., low-dose ritonavir added to saquinavir, indinavir, amprenavir, or lopinavir) for single PIs enable the regimen to be given less frequently. Several studies with regimens of boosted PIs given twice daily have reported comparable virologic results, lower total drug costs, greater tolerability, and improved adherence.

Selection of the next regimen must be individualized based, in relatively equal parts, on the last regimen, the resistance test results obtained while the patient is still taking the failing regimen, and prior exposure to other agents, with emphasis on tolerability and potential residual (archived) resistant viruses that are being harbored in latently infected cells. As when selecting the initial regimen, consideration must be given to what the patient is willing to take and likely to tolerate. In situations where options are limited because of previous toxicity or prior failure of available agents, it may be necessary to 'recycle' an ARV drug back into a new regimen. When this is necessary, it is best to use agents that have not been utilized in the last two or three regimens and to avoid recycling two or three agents from a single previous regimen back together. Ideally, the new regimen should consist of two or more agents deemed to be active, preferably with drugs that have not been used together in previous regimens. Most often it is prudent to maintain the presence of either lamivudine or emtricitabine in the next regimen owing to both the observation that these drugs still exert a substantial antiviral effect (0.5–0.8 log reduction in viral load) even in the face of an M184V mutation and the high degree of tolerability of these drugs.^{145,179}

Pharmacokinetic enhancement, such as boosting PI levels with low-dose ritonavir, should be exploited whenever possible in the setting of multiple-regimen failure. The use of 'double-boosted PIs' (two PI agents along with low-dose ritonavir) has not been shown to have any benefit over single-boosted PI therapy and is associated with increased toxicity and unpredictable drug–drug interactions.¹ Similarly, the use of multidrug (six or more drugs) rescue therapy has shown variable ARV activity.^{180–182} However, this approach often

leads to substantial toxicity and problems with adherence, drug–drug interactions, and cost. With newer, more potent drugs available, such as tipranavir, darunavir, and enfuvirtide, multiple drug regimens (so-called ‘mega-HAART’) is no longer a meaningful consideration.

Except in cases of toxicity, it is preferable to continue even ‘failing’ drug regimens that maintain selective pressure on the viruses rather than permanently discontinuing all ART, especially in settings in which the CD4+ T-lymphocyte counts are maintained despite a rebound in viral replication.¹⁷⁹ In cases where the regimen is changed, the same concepts of defining the target plasma RNA values that define success and failure of the regimen in the current clinical setting still apply and should be discussed with the patient.

Treatment Interruptions

Use of a ‘supervised’ treatment interruption (STI) has been postulated to be of clinical benefit among certain patient populations. In the setting of multidrug-resistant virus, STIs have been used in an attempt to exploit the rapid reversion to wild-type viruses.^{183–185} Such a strategy should be undertaken with great caution owing to the rapid increase in viral load and marked reduction in CD4+ T-lymphocyte count in association with reversion to wild-type virus around week eight of the STI.¹⁸³ Studies of this approach have yielded mixed results, mostly due to differences in the timing of reinitiation of therapy (prior to week 12 of the STI vs at week 16).^{186–190} Therefore, if an STI is employed in the setting of multiply resistant virus, frequent assessments should be performed during the STI with early resumption of treatment with any sign of plummeting CD4+ T-lymphocyte count. Of significant concern is the higher incidence of cardiovascular events in studies of planned treatment interruptions.¹⁸⁶ This unexpected safety outcome occurred most commonly among those patients who stopped ART with lower CD4+ counts and those who had a lower CD4 count nadir.

STIs have also been postulated to be a strategy among successfully treated patients in an attempt to reduce drug exposure, long-term toxicities, and costs. Several studies that employed this strategy have demonstrated harm associated with routine use of STIs in this setting.¹⁸⁶ In particular, the SMART study, which randomized patients to standard of care (continuous treatment) versus routine cessation of ARV therapy when their CD4+ count rose above 400 cells/ μ L and have treatment reinstated when the CD4+ count dropped below 250 cells/ μ L, demonstrated clear harm associated with the STI strategy. The study was stopped early by a Data Safety and Monitoring Board owing to a significantly higher degree of clinical events, including cardiac events, among the STI managed patients.

Another proposed use of an STI is to enhance and stimulate immunologic responses to HIV with the goal of reducing the need for ARV therapy. The concept is to use periodic treatment interruptions as a natural vaccine using autologous virus as the immunogen.^{191,192} The few studies that have been conducted to evaluate this concept have demonstrated some

evidence of partial lowering of the viral load set-point, but the magnitude of response is quite meager and this approach cannot be recommended.

The most common and perhaps only appropriate use of an STI is in the management of treatment toxicities as discussed above.

Other ARV Management Issues

On occasion, patients present with a ‘discordant’ response, whereby the viral load is successfully suppressed but the CD4+ T-lymphocyte count fails to increase. Several options exist for managing a ‘discordant’ response, but because the reasons for the discordant responses are not well understood there is no clearly defined or optimal approach to management. Drug-related toxic effects, inhibition of *de novo* CD4+ T-lymphocyte synthesis, sequestration of cells within lymphoid tissue, and interference with clonal expansion of memory CD4+ T lymphocytes have been proposed as potential mechanisms. The approach to management depends on the suspected reason for the discordant response. For those patients with viral load values <50c/mL, it is best to continue the current regimen and evaluate for other causes of bone marrow or cellular toxicity. For example, substituting specific drugs in the regimen, removing potentially cytotoxic drugs, and eliminating hydroxyurea-containing regimens are valid options when bone marrow toxicity is suspected. Alternatively, the use of cytokines, such as IL-2 or granulocyte- and granulocyte/macrophage colony-stimulating factors, are potential approaches to expand the cell populations directly (*see* Chapter 30). However there are not enough data to recommend cytokine therapy at this time. Trials of IL-2 (3.0–4.5 million units twice daily for 5 days every 2 months) are being evaluated among patients with low (SILCAAT) and high (ESPRIT) CD4+ T-lymphocyte counts in an attempt to raise the CD4+ T-lymphocyte cell counts among patients who have achieved successful suppression of HIV but did not have concomitant increases in their CD4+ T-lymphocyte counts. To date, these trials have not yielded definitive recommendations on the use of IL-2; but it is important to note the high degree of toxicity associated with IL-2 treatment and the low degree of observed disease progression among patients with low CD4 T-lymphocyte counts and persistent viral load values <50c/mL.

The use of therapeutic drug monitoring (TDM) has been proposed as a means of enhancing virologic outcomes and assessing reasons for virologic failure or toxicity (*see* Chapter 79). TDM is a process whereby drug levels are measured at fixed time intervals after a dose of medicine has been ingested. The PK curve of a given drug can be modeled with as few as one or two measurements. Once determined, the peak and trough levels can be estimated and used to assess the likelihood of virologic suppression or development of toxicity. Although the concept is attractive on face value, multiple assumptions limit the usefulness of TDM in practice. For example, to model accurately, the time of dosing must be known precisely. Relying on verbal reports of dosing is inaccurate. Moreover, the modeling assumes that the patient

is at steady state, which means that all previous doses were taken correctly and on time, which may not be the case. The modeling is inaccurate if it utilizes levels determined on samples obtained within the first 2 h (absorption/distribution phase) of dosing; therefore, only levels for samples obtained 'postpeak' can be used. Finally, it is unclear which 'level' is the optimal target for best virologic response for each individual drug. Because TDM is based solely on plasma levels, there is no consideration for intracellular drug concentrations or intracellular processing, creating further limitations of the technique. Nonetheless, TDM can be a useful adjunct to therapy when assessing the possibility of nonadherence or poor absorption (e.g., absence of drug in the bloodstream) or in cases of extremely advanced disease where higher doses of drug are being considered to maximize ARV benefit. Outside of these two circumstances, TDM remains a research tool.

REFERENCES

1. Hammer SM, Saag MS, Schechter M, et al. Treatment of adult HIV infection: 2006 recommendations of the International AIDS Society-USA panel. *JAMA* 296:827–883, 2006.
2. National Institutes of Health Panel to define principles of therapy of HIV infection. Guidelines for the use of antiretroviral agents in HIV-infected adults and adolescents. *Ann Intern Med* 128:1079–100, 1998.
3. Gazzard B, Moyle G, BHIVA Guidelines Writing Committee. Revision to the British HIV Association guidelines for antiretroviral treatment of HIV seropositive individuals. *Lancet* 352:314–16, 1998.
4. US Department of Health and Human Services Panel on Clinical Practices for Treatment of HIV Infection. Guidelines for the use of antiretroviral agents in HIV-1 infected adults and adolescents. Washington, DC: DHHS. Available: <http://aidsinfo.nih.gov/Guidelines/GuidelineDetail.aspx?MenuItem=Guidelines&Search=Off&GuidelineID=7&ClassID=1> 10 Oct 2006.
5. US Department of Health and Human Services Panel on Clinical Practices for Treatment of HIV Infection. Guidelines for the use of antiretroviral agents in HIV-1 infected adults and adolescents. *MMWR Morb Mortal Wkly Rep* 47(RR-5):1–41, 1998.
6. Wei X, Ghosh SK, Taylor ME, et al. Viral dynamics in HIV-1 infection. *Nature* 373:117–22, 1995.
7. Ho DD, Neumann AU, Perelson AS, et al. Rapid turnover of plasma virions and CD4+ lymphocytes in HIV-1 infection. *Nature* 373:123–26, 1995.
8. Coffin JM. HIV population dynamics in vivo: implications for genetic variation, pathogenesis, and therapy. *Science* 267:483–9, 1995.
9. Wain-Hobson S. AIDS: virological mayhem. *Nature* 373:102, 1995.
10. Saag MS, Holodniy M, Kuritzkes DR, et al. HIV viral load markers in clinical practice. *Nat Med* 2:625–9, 1996.
11. Holodniy M, Katzenstein DA, Sengupta S. Detection and quantification of human immunodeficiency virus RNA in patient serum by use of the polymerase chain reaction. *J Infect Dis* 163:862–6, 1991.
12. Piatak M, Saag MS, Yang LC, et al. High levels of HIV-1 in plasma during all stages of infection determined by competitive PCR. *Science* 259:1749–54, 1993.
13. Piatak M Jr, Saag MS, Yang LC, et al. Determination of plasma viral load in HIV-1 infection by quantitative competitive polymerase chain reaction. *AIDS* 7(Suppl 2):S65–71, 1993.
14. Cao Y, Ho DD, Todd J, et al. Clinical evaluation of branched DNA signal amplification for quantifying HIV type 1 in human plasma. *AIDS Res Hum Retroviruses* 11:353–61, 1995.
15. Hirsch MS, Brun-Vézinet F, Clotet B, et al. Antiretroviral drug resistance testing in adults infected with human immunodeficiency virus type 1: 2003 recommendations of an International AIDS Society-USA panel. *Clin Infect Dis* 37:113–28, 2003.
16. Hogg RS, Heath KV, Yip B, et al. Improved survival among HIV-infected individuals following initiation of antiretroviral therapy. *JAMA* 279:450–54, 1998.
17. Palella FJ, Delaney KM, Moorman AC, et al. Declining morbidity and mortality among patients with advanced human immunodeficiency virus infection. *N Engl J Med* 338:853–60, 1998.
18. Palella F, Moorman A, Chmiel J, et al. Continued low morbidity and mortality among patients with advanced HIV infection and their patterns of highly active antiretroviral therapy (HAART) usage. Presented at the 7th Conference on Retroviruses and Opportunistic Infections, 2000, p 216.
19. Carr A, Samaras K, Burton S, et al. A syndrome of peripheral lipodystrophy, hyperlipidaemia and insulin resistance in patients receiving HIV protease inhibitors. *AIDS* 12:F51–8, 1998.
20. Carr A, Miller J, Law M, Cooper DA. A syndrome of lipodystrophy, lactic acidemia and liver dysfunction associated with HIV nucleoside analogue therapy: contribution to protease inhibitor-related lipodystrophy syndrome. *AIDS* 14:F25–32, 2000.
21. Carr A, Samaras K, Chisholm DJ, Cooper DA. Pathogenesis of HIV-1-protease inhibitor-associated peripheral lipodystrophy, hyperlipidaemia, and insulin resistance. *Lancet* 351:1881–3, 1998.
22. Yarasheski KE, Tebas P, Sigmund C, et al. Insulin resistance in HIV protease inhibitor-associated diabetes. *J Acquir Immune Defic Syndr* 21:209–16, 1999.
23. Schambelan M, Benson C, Carpenter CCJ, et al. Metabolic complications guidelines: recommendation of the International AIDS Society-USA panel. *JAIDS* 31:257–75, 2002.
24. Masur H, Miller KD, Jones EC, et al. High prevalence of avascular necrosis (AVN) of the hip in HIV infection: magnetic resonance imaging of 339 asymptomatic patients. Presented at the 38th Annual Meeting of the Infectious Diseases Society of America, 2000.
25. 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* 40:1559–85, 2005.
26. Lo JC, Kazemi MR, Hsue PY, et al. The relationship between nucleoside analogue treatment duration, insulin resistance, and fasting arterial lactate level in patients with HIV infection. *Clin Infect Dis* 41:1335–40, 2005.
27. Sanne I, Mommeja-Marin H, Hinkle J, et al. Severe hepatotoxicity associated with nevirapine use in HIV-infected subjects. *J Infect Dis* 192:545–6, 2005.
28. Hockett RD, Kilby JM, Derdeyn CA, et al. Constant mean viral copy number per infected cell in tissues regardless of high, low, or undetectable plasma HIV RNA. *J Exp Med* 189:1545–59, 1999.
29. Zhang L, Ramratnam B, Tenner-Racz K, et al. Quantifying residual HIV-1 replication in patients receiving combination antiretroviral therapy. *N Engl J Med* 340:1605–13, 1999.
30. Acosta EP, Henry K, Baken L, et al. Indinavir concentrations and antiviral effect. *Pharmacotherapy* 19:708–12, 1999.
31. Acosta EP, Kakuda TN, Brundage RC, et al. Pharmacodynamics of human immunodeficiency virus type 1 protease inhibitors. *Clin Infect Dis* 30(Suppl 2):S151–9, 2000.
32. Sommadossi JP, Zhou XJ, Moore J, et al. Impairment of stavudine phosphorylation in patients receiving a combination of zidovudine and stavudine. Presented at the 5th Conference on Retroviruses and Opportunistic Infections, 1998.

33. Hsu A, Granneman GR, Bertz RJ. Ritonavir-clinical pharmacokinetics and interactions with other anti-HIV agents. *Clin Pharmacokinet* 35:275–91, 1998.
34. Chesney MA, Morin M, Sherr L. Adherence to HIV combination therapy. *Soc Sci Med* 50:1599–605, 2000.
35. Shaw GM, Harper ME, Hahn BH, et al. HTLV-III infection in brains of children and adults with AIDS encephalopathy. *Science* 227:177–81, 1985.
36. Bagasra O, Lavi E, Bobroski L. Cellular reservoirs of HIV-1 in the central nervous system of infected individuals: identification by the combination of in situ polymerase chain reaction and immunohistochemistry. *AIDS* 10:573–85, 1996.
37. Coombs RW, Welles SL, Hooper C. Association of plasma human immunodeficiency virus type-1 RNA level with risk of clinical progression in patients with advanced infection. *J Infect Dis* 174:704–12, 1996.
38. Daar ES, Moudgil T, Meyer RD, Ho DD. Transient high levels of viremia in patients with primary human immunodeficiency virus type 1 infection. *N Engl J Med* 324:961–4, 1991.
39. Clark SJ, Saag MS, Decker WD, et al. High titers of cytopathic virus in plasma of patients with symptomatic primary HIV-1 infection. *N Engl J Med* 324:954–60, 1991.
40. Ho DD, Moudgil T, Alam M. Quantitation of human immunodeficiency virus type 1 in the blood of infected persons. *N Engl J Med* 321:1621–5, 1989.
41. Saag MS, Crain MJ, Decker WD. High-level viremia in adults and children infected with human immunodeficiency virus: relation to disease stage and CD4+ lymphocyte levels. *J Infect Dis* 164:72–80, 1991.
42. Clark SJ, Shaw GM. Acute retroviral syndrome and the pathogenesis of HIV-1 infection. *Semin Immunol* 5:149–55, 1993.
43. Schacker T, Hughes JP, Shea T, et al. Biological and virologic characteristics of primary HIV infection. *Ann Intern Med* 128:613–20, 1998.
44. Kappes JC, Saag MS, Shaw GM, et al. Assessment of antiretroviral therapy by plasma viral load testing: standard and ICD HIV-1 p24 antigen and viral RNA (QC-PCR) assays compared. *Acquir Immune Defic Syndr Hum Retrovir* 10:139–49, 1995.
45. Saag MS, Emini EA, Laskin OL. Short-term clinical evaluation of L-697,661, a nonnucleoside inhibitor of HIV-1 reverse transcriptase. *N Engl J Med* 329:1065–72, 1993.
46. Perelson AS, Neumann AU, Markowitz M, et al. HIV-1 dynamics in vivo: virion clearance rate, infected cell life-span, and viral generation time. *Science* 271:1582–6, 1996.
47. Perelson AS, Essunger P, Cao Y, et al. Decay characteristics of HIV-1 infected compartments during combination therapy. *Nature* 387:188–91, 1997.
48. Perelson AS, Essunger P, Ho DD. Dynamics of HIV-1 and CD4 lymphocytes in vivo. *AIDS* 11(Suppl A):517–24, 1997.
49. Fauci AS. Host factors and the pathogenesis of HIV-induced disease. *Nature* 384:529–34, 1996.
50. Chun TW, Fauci AS. Latent reservoirs of HIV: obstacles to the eradication of virus. *Proc Natl Acad Sci USA* 96:10958–61, 1999.
51. Wong JK, Gunthard HF, Havlir DV, et al. Reduction of HIV in blood and lymph nodes after potent antiretroviral therapy and the virologic correlates of treatment failure. *Proc Natl Acad Sci USA* 94:12574–9, 1997.
52. Dybul M, Chun TW, Ward DJ, et al. Evaluation of lymph node virus burden in human immunodeficiency virus-infected patients receiving efavirenz-based protease inhibitor-sparing highly active antiretroviral therapy. *J Infect Dis* 181:1273–9, 2000.
53. Boucher C, Nijhuis M, Schipper P, et al. Reduction of HIV in blood and lymph nodes after potent antiretroviral therapy. Presented at the 4th Conference on Retroviruses and Opportunistic Infections, 1997.
54. Cavert W, Notermans DW, Staskus K, et al. Kinetics of response in lymphoid tissues to antiretroviral therapy of HIV-1 infection. *Science* 276:960–4, 1997.
55. Nelson PW, Mittler JE, Perelson A. Effect of drug efficacy and the eclipse phase of the viral life cycle on estimates of HIV viral dynamic parameters. *J Acquir Defic Syndr Hum Retrovir* 26:405–12, 2001.
56. Mellors JW, Rinaldo CR Jr, Gupta P, et al. Prognosis in HIV-1 infection predicted by the quantity of virus in plasma. *Science* 272:1167–70, 1996.
57. Mellors JW, Munoz AM, Giorgi JV, et al. Plasma viral load and CD4+ lymphocytes as prognostic markers of HIV-1 infection. *Ann Intern Med* 126:946–54, 1997.
58. Rodriguez B, Sethi AK, Cheruvu VK, et al. Predictive value of plasma HIV RNA level on rate of CD4 T-cell decline in untreated HIV infection. *JAMA* 296:1498–506, 2006.
59. Ferguson NM, deWolf F, Ghani AC, et al. Antigen-driven CD4+ T cell and HIV-1 dynamics: residual viral replication under highly active antiretroviral therapy. *Proc Natl Acad Sci USA* 96:15167–72, 1999.
60. Finzi D, Siliciano RF. Viral dynamics in HIV-1 infection. *Cell* 93:665–71, 1998.
61. Ho DD. Time to hit HIV, early and hard. *N Engl J Med* 333:450–1, 1995.
62. Brechley JM, Price DA, Douek DC. HIV disease: fallout from a mucosal catastrophe? *Nat Immunol* 7:235–9, 2006.
63. Brechley JM, Schacker TW, Ruff LE, et al. CD4+ T cell depletion during all stages of HIV disease occurs predominantly in the gastrointestinal tract. *J Exp Med* 200:749–59, 2004.
64. Mattapallil JJ, Douek DC, Hill B, et al. Massive infection and loss of memory CD4+ T cells in multiple tissues during acute SIV infection. *Nature* 434:1093–7, 2005.
65. Autran B, Carcelain G, Li TS, et al. Positive effects of combined antiretroviral therapy on CD4+ T cell homeostasis and function in advanced HIV disease. *Science* 277:112–16, 1997.
66. Bucy RP. Immune clearance of HIV type 1 replication-active cells: a model of two patterns of steady state HIV infection. *AIDS Res Hum Retroviruses* 15:223–7, 1999.
67. Saag MS, Kilby JM. HIV-1 and HAART: a time to cure, a time to kill. *Nat Med* 5:609–11, 1999.
68. Rosenberg ES, Billingsley JM, Caliendo AM, et al. Vigorous HIV-1-specific CD4+ T cell responses associated with control of viremia. *Science* 278:1447–50, 1997.
69. Rosenberg ES, Walker BD. HIV type 1-specific helper T cells: a critical host defense. *AIDS Res Hum Retroviruses* 14(Suppl 2):S143–7, 1998.
70. Wei X, Decker JM, Wang S, et al. Antibody neutralization and escape by HIV-1. *Nature* Mar 20; 422:307–12, 2003.
71. Decker JM, Bibollet-Ruche F, Wei X, et al. Antigenic conservation and immunogenicity of the HIV co-receptor binding site. *J Exp Med* 201:1407–19, 2005.
72. Rosenberg ES, Altfield M, Poon SH, et al. Immune control of HIV-1 after early treatment of acute infection. *Nature* 407:523–6, 2000.
73. Davey RT Jr, Bhat N, Yoder C, et al. HIV-1 and T cell dynamics after interruption of highly active antiretroviral therapy (HAART) in patients with a history of sustained viral suppression. *Proc Natl Acad Sci USA* 96:15109–14, 1999.
74. Ruiz L, Martinez-Picado J, Romeo J, et al. Structured treatment interruption in chronically HIV-1 infected patients after long-term viral suppression. *AIDS* 14:397–403, 2000.
75. Borrow P, Lewicki H, Wei X, et al. Antiviral pressure exerted by HIV-1-specific cytotoxic T lymphocytes (CTLs) during primary infection demonstrated by rapid selection of CTL escape virus. *Nat Med* 3:205–11, 1997.

76. Borrow P, Lewicki H, Hahn BH, et al. Virus-specific CD8+ cytotoxic T-lymphocyte activity associated with control of viremia in primary human immunodeficiency virus type 1 infection. *J Virol* 68:6103–10, 1994.
77. Altfeld M, Rosenberg ES, Mukherjee J, et al. Enhancement of HIV-1 specific CTL responses during structured treatment interruptions (STI) following treated acute HIV-1-infection is associated with control of HIV-1 viremia. Presented at the XIII International AIDS Conference, 2000.
78. Walker CM, Moody DJ, Stites DP, Levy JA. CD8+ lymphocytes can control HIV infection in vitro by suppressing virus replication. *Science* 234:1563–6, 1986.
79. Mclean AR, Michie CA. In vivo estimates of division and death rates of human T-lymphocytes. *Proc Natl Acad Sci USA* 92:3707–11, 1995.
80. Chun TW, Carruth L, Finzi D, et al. Quantification of latent tissue reservoirs and total body fat viral load in HIV-1 infection. *Nature* 387:183–8, 1997.
81. Wong JK, Hezareh M, Gunthard HF, et al. Recovery of replication-competent HIV despite prolonged suppression of plasma viremia. *Science* 278:1291–5, 1997.
82. Finzi D, Hermankova M, Pierson T, et al. Identification of a reservoir for HIV-1 in patients on highly active antiretroviral therapy. *Science* 278:1295–300, 1997.
83. Deeks SG, Hecht FM, Swanson M, et al. HIV RNA and CD4 cell count response to protease inhibitor therapy in an urban AIDS clinic: response to both initial and salvage therapy. *AIDS* 13:F35–43, 1999.
84. Hammer SM, Squires KE, Hughes MD, et al. A controlled trial of two nucleoside analogues plus indinavir in persons with human immunodeficiency virus infection and CD4 cell counts of 200/ μ l or less. *N Engl J Med* 337:725–33, 1997.
85. Gulick RM, Mellors JW, Havlir D, et al. Treatment with indinavir, zidovudine, and lamivudine in adults with human immunodeficiency virus infection and prior antiretroviral therapy. *N Engl J Med* 337:734–9, 1997.
86. Chaisson RE, Keruly JC, Moore RD. Association of initial CD4 cell count and viral load with response to highly active antiretroviral therapy. *JAMA* 284:3128–9, 2000.
87. Hogg RS, Yip B, Wood E, et al. Diminished effectiveness of antiretroviral therapy among patients initiating therapy with CD4+ cell counts below 200/ mm^3 . Presented at the 8th Conference on Retroviruses and Opportunistic Infections, 2001.
88. Moore R, Keruly J, Bartlett J, Chaisson R. Start HAART early (CD4 > 350 cells/ μ l) or later? Evidence for greater effectiveness if started early. Presented at the 7th Conference on Retroviruses and Opportunistic Infections, 2000, p 174.
89. Marschner IC, Collier AC, Coombs RW, et al. Use of changes in plasma levels of human immunodeficiency virus type 1 RNA to assess clinical benefit to antiretroviral therapy. *J Infect Dis* 177:40–7, 1998.
90. Yerly S, Kaiser L, Perneger TV, et al. Time of initiation of antiretroviral therapy: impact on HIV-1 viraemia: the Swiss HIV Cohort Study. *AIDS* 14:243–9, 2000.
91. Chun TW, Davey RT Jr, Ostrowski M, et al. Relationship between pre-existing viral reservoirs and the re-emergence of plasma viremia after discontinuation of highly active antiretroviral therapy. *Nat Med* 6:757–61, 2000.
92. Kempf DJ, Rode RA, Xu Y, et al. The duration of viral suppression during protease inhibitor therapy for HIV-1 infection is predicted by plasma HIV-1 RNA at the nadir. *AIDS* 12:F9–14, 1998.
93. Cahn P. 24-week data from RESIST 2: phase 3 study of the efficacy and safety of either tipranavir/ritonavir (TPV/r) or an optimized ritonavir (RTV)-boosted standard-of-care (SOC) comparator PI (CPI) in a large randomized multicenter trial in treatment-experienced HIV+ patients. 7th International Congress on Drug Therapy in HIV Infection, Glasgow, Scotland, 14–18 Nov 2004, abstract PL14.3.
94. Katlama C, Carvalho MT, Cooper D, et al. TMC114/r outperforms investigator-selected PI(s) in 3-class-experienced patients: week 24 primary analysis of POWER 1 (TMC114-C213). 3rd IAS Conference on HIV Pathogenesis and Treatment, Rio de Janeiro, Brazil, 24–27 Jul 2005, abstract WeOaLB0102.
95. Wilkin T, Haubrich R, Steinhard CR, et al. TMC114/r superior to standard of care in 3-class-experienced patients: 24-wks primary analysis of the Power 2 Study (C202). 45th Interscience Conference on Antimicrobial Agents and Chemotherapy, Washington, DC, 16–19 Dec 2005, abstract H-413.
96. Clerici M, Levin JM, Kessler HA. HIV-specific T-helper activity in seronegative health care workers exposed to contaminated blood. *JAMA* 271:42–6, 1994.
97. Centers for Disease Control and Prevention. Public health service guidelines for the management of health-care worker exposures to HIV and recommendations for post-exposure prophylaxis. *MMWR Morb Mortal Wkly Rep* 47(RR-7):1–33, 1998.
98. Connor EM, Sperling RS, Gelber R, et al. Reduction of maternal-infant transmission of human immunodeficiency virus type 1 with zidovudine treatment: pediatric AIDS Clinical Trials Group Protocol 076 Study Group. *N Engl J Med* 331:1173–80, 1994.
99. Saag MS, Schooley RT. Initiation of antiretroviral therapy: current controversies in when and with what to start. *Top HIV Med* 8:8–13, 2000.
100. Raboud JM, Rae S, Hogg RS, et al. Suppression of plasma viral load below the detection limit of a human immunodeficiency virus kit is associated with longer virologic response than suppression below the limit of quantitation. *J Infect Dis* 180:1347–50, 1999.
101. Deeks S, Barbour J, Martin JN, et al. Sustained CD4+ T cell response after virologic failure of protease inhibitor-based regimens in patients with human immunodeficiency virus infection. *J Infect Dis* 181:946–53, 2000.
102. Deeks SG, Barbour JD, Martin JN, Grant RM. Delayed immunologic deterioration among patients who virologically fail protease inhibitor-based therapy. Presented at the 7th Conference on Retroviruses and Opportunistic Infections, 2000, p 120.
103. Emini EA, Graham DJ, Gotlib L. HIV and multidrug resistance. *Nature* 364:679, 1993.
104. Richman DD. Resistance of clinical isolates of human immunodeficiency virus to antiretroviral agents. *Antimicrob Agents Chemother* 37:1207–13, 1993.
105. Richman DD, Havlir D, Corbeil J, et al. Nevirapine resistance mutations of human immunodeficiency virus type 1 selected during therapy. *J Virol* 68:1660–6, 1994.
106. Bangsberg DR, Hecht FM, Charlebois ED, et al. Adherence to protease inhibitors, HIV-1 viral load, and development of drug resistance in an indigent population. *AIDS* 14:357–66, 2000.
107. Arnsten J, Demas P, Gourevitch M, et al. Adherence and viral load in HIV-infected drug users: comparison of self-report and medication event monitors (MEMS). Presented at the 7th Conference on Retroviruses and Opportunistic Infections, 2000, p 88.
108. Carr A, Samaras K, Thorisdottir A, et al. Diagnosis, prediction, and natural course of HIV-1 protease inhibitor-associated lipodystrophy, hyperlipidaemia, and diabetes mellitus: a cohort study. *Lancet* 353:2093–9, 1999.
109. Hadigan C, Meigs JB, Corcoran C, et al. Metabolic abnormalities and cardiovascular disease risk factors in adults with human immunodeficiency virus infection and lipodystrophy. *Clin Infect Dis* 32:130–9, 2001.
110. Blacksin MF, Kloser PC, Simon J. Avascular necrosis of bone in human immunodeficiency virus infected patients. *Clin Imaging* 23:314–18, 1999.

111. Brinkman K, Smeitink JA, Romijn JA, Reiss P. Mitochondrial toxicity induced by nucleoside-analogue reverse-transcriptase inhibitors is a key factor in the pathogenesis of antiretroviral-therapy-related lipodystrophy. *Lancet* 354:1112–15, 1999.
112. Tebas P, Powderly WG, Claxton S, et al. Accelerated bone mineral loss in HIV-infected patients receiving potent antiretroviral therapy. *AIDS* 14:F63–7, 2000.
113. Hoy J, Hudson J, Law M, Cooper DA. Osteopenia in a randomized multicenter study of protease inhibitor (PI) substitution in patients with the lipodystrophy syndrome and well-controlled HIV viremia. Presented at the 7th Conference on Retroviruses and Opportunistic Infections, 2000, p 114.
114. Glesby MJ, Hoover PR, Vaamonde CM. Osteonecrosis in patients infected with HIV: a case-control study. *J Infect Dis* 184:519–23, 2001.
115. Fletcher CV, Anderson PL, Kakuda TN, et al. A novel approach to integrate pharmacologic and virologic characteristics: an in vivo potency (IVP) index for antiretroviral agents. Presented at the 8th Conference on Retroviruses and Opportunistic Infections, 2001.
116. Flexner C, Speck RR. Role of multidrug transporters in HIV pathogenesis. Presented at the 8th Conference on Retroviruses and Opportunistic Infections, 2001, p 281.
117. Haas DW, Ribaldo HJ, Kim RB, et al. Pharmacogenetics of efavirenz and central nervous system side effects: an Adult AIDS Clinical Trials Group study. *AIDS* 18:2391–400, 2004.
118. Chen R, Westfall A, Cloud G, et al. Long-term survival after initiation of antiretroviral therapy. Presented at the 8th Conference on Retroviruses and Opportunistic Infections, 2001.
119. Egger M, May M, Chene G, et al. Prognosis of HIV-1 infected drug naive patients starting potent antiretroviral therapy: a collaborative analysis of prospective studies. *Lancet* 360:119–29, 2002.
120. Henry K. The case for more cautious, patient-focused antiretroviral therapy. *Ann Intern Med* 132:306–22, 2000.
121. Kirkland LR, Fischl MA, Tashira KT, et al. Response to lamivudine-zidovudine plus abacavir twice daily in ART-NAIVE, incarcerated patients with HIV taking directly observed treatment. *Clin Infect Dis* 34:511–18, 2002.
122. Staszewski S, Morales-Ramirez J, Tashima KT, et al. Efavirenz plus zidovudine and lamivudine, efavirenz plus indinavir, and indinavir plus zidovudine and lamivudine in the treatment of HIV-1 infection in adults: study 006 team. *N Engl J Med* 341:1865–73, 1999.
123. Acosta EP, Gulick R, Katzenstein D, et al. Pharmacokinetic (PK) evaluation of saquinavir soft gel capsules (SQV)/ritonavir (RTV) or SQV/nelfinavir (NFV) in combination with delavirdine (DLV) and/or adefovir dipivoxil (ADV)-ACTG 359. Presented at the 6th Conference on Retroviruses and Opportunistic Infections, 1999.
124. Andrade A, Flexner C. HIV-related drug metabolism and cytochrome P450 enzymes. *AIDS Clin Care* 12:91–5, 2000.
125. Benson C, Brun S, King M, et al. Two year follow-up of ABT378/ritonavir (ABT-378/r) in antiretroviral naive HIV + patients. Presented at the 40th Interscience Conference on Antimicrobial Agents and Chemotherapy, 2000, p 282.
126. Cameron DW, Japour AJ, Xu Y, et al. Ritonavir and saquinavir combination therapy for the treatment of HIV infection. *AIDS* 13:213–24, 1999.
127. Casado JL, Moreno A, Marti-Belda P, et al. Increased indinavir levels using twice daily ritonavir/indinavir at 100/800mg improves virological response even after multiple failure. Presented at the 40th Interscience Conference on Antimicrobial Agents and Chemotherapy, 2000, p 301.
128. Hammer SM. Clinical practice. Management of newly diagnosed HIV infection. *N Engl J Med* 353:1702–10, 2005.
129. Carpenter CCJ, Fischl MA, Hammer SM, et al. Antiretroviral therapy for HIV infection in 1998: updated recommendations of the International AIDS Society-USA panel. *JAMA* 280:78–86, 1998.
130. Saag MS, Hahn BH, Gibbons J, et al. Extensive variation of human immunodeficiency virus type-1 in vivo. *Nature* 334:440–4, 1988.
131. Drake JW. Rates of spontaneous mutation among RNA viruses. *Proc Natl Acad Sci USA* 90:4171–5, 1993.
132. Mansky LM, Temin HM. Lower in vivo mutation rate of human immunodeficiency virus type 1 than that predicted from the fidelity of purified reverse transcriptase. *J Virol* 69:5087–94, 1995.
133. Deeks SG, Wrin T, Ducey E, et al. Decreased HIV-1 fitness after long-term virologic failure of protease inhibitor-based therapy: relationship to immunologic response. Presented at the 7th Conference on Retroviruses and Opportunistic Infections, 2000, p 104.
134. International AIDS Society-USA Resistance Mutations Project Panel. Update on drug resistance mutations in HIV-1. *Top HIV Med* 14:125–30, 2006.
135. Kuritzkes DR, Bell S, Shugarts D, Abrams D. Development of resistance to lamivudine (3TC) in NUCA 3001, a phase II comparative study of 3TC versus zidovudine versus 3TC plus zidovudine. Presented at the 2nd National Conference on Human Retroviruses and Related Infections, 1995.
136. Saag MS, Emini EA, Laskin OL, et al. A short-term clinical evaluation of L-697,661, a non-nucleoside inhibitor of HIV-1 reverse transcriptase: L-697,661 working group. *N Engl J Med* 329:1065–72, 1993.
137. Larder BA, Darby G, Richman DD. HIV with reduced sensitivity to zidovudine (AZT) isolated during prolonged therapy. *Science* 243:1731–4, 1989.
138. Larder BA, Chesebro B, Richman DD. Susceptibilities of zidovudine-susceptible and -resistant human immunodeficiency virus isolates to antiviral agents determined by using a quantitative plaque reduction assay. *Antimicrob Agents Chemother* 34:436–41, 1990.
139. Condra JH, Schleif WA, Blahy OM. In vivo emergence of HIV-1 variants resistant to multiple protease inhibitors. *Nature* 374:569–71, 1995.
140. Condra JH, Holder DJ, Schleif WA, et al. Genetic correlates of in vivo viral resistance to indinavir, a human immunodeficiency virus type 1 protease inhibitor. *J Virol* 70:8270–6, 1996.
141. Patrick A, Mo H, Markowitz M. Antiviral and resistance studies of AG1343, an orally bioavailable inhibitor of human immunodeficiency virus protease. *Antimicrob Agents Chemother* 40:292–7, 1996.
142. Kravcik S, Farnsworth A, Patick A, et al. Long-term follow-up of combination protease inhibitor therapy with nelfinavir and saquinavir (soft gel) in HIV infection. Presented at the 5th Conference on Retroviruses and Opportunistic Infections, 1998.
143. Larder BA, Kemp SD, Harrigan PR. Antiviral potency of AZT + 3TC combination therapy supports virological observations. Presented at the 2nd National Conference on Human Retroviruses and Related Infections, 1995.
144. Havlir DV, Gamst A, Eastman S, Richman DD. Nevirapine-resistant human immunodeficiency virus: kinetics of replication and estimated prevalence in untreated patients. *J Virol* 70:7894–9, 1996.
145. Eron JJ, Benoit SL, Jemsek J, et al. Treatment with lamivudine, zidovudine, or both in HIV-positive patients with 200 to 500 CD4+ cells per cubic millimeter. *N Engl J Med* 333:1662–9, 1995.
146. Larder BA, Kellam P, Kemp SD. Convergent combination therapy can select viable multidrug-resistant HIV-1 in vitro. *Nature* 365:451–3, 1993.
147. Murphy RL, Sommadossi JP, Lamson M, et al. Antiviral effect and pharmacokinetic interaction between nevirapine and

- indinavir in persons infected with human immunodeficiency virus type 1. *J Infect Dis* 179:1116–23, 1999.
148. Urquhart J. Partial compliance in cardiovascular disease: risk implications. *Br J Clin Pract* 73(Suppl):2, 1994.
 149. Greenberg RN. Overview of patient compliance with medication dosing: a literature review. *Clin Ther* 65:590, 1984.
 150. Wright EC. Non-compliance: or how many aunts has Matilda? *Lancet* 342:909, 1993.
 151. Urquhart J. Patient non-compliance with drug regimens: measurement, clinical correlates, economic impact. *Eur Heart J* 17(Suppl A):8, 1996.
 152. Havlir D, Levitan D, Bassett R, et al. Prevalence and predictive value of intermittent viraemia in patients with viral suppression. *Antiviral Ther* 5(Suppl 3):89, 2000.
 153. Havlir DV, Marschner IC, Hirsch MS, et al. Maintenance antiretroviral therapies in HIV infected patients with undetectable plasma HIV RNA after triple-drug therapy: AIDS Clinical Trials Group Study 343 team. *N Engl J Med* 339:1261–8, 1998.
 154. Gharakhanian S, Salhi Y, Adda N, et al. Identification of fat redistribution/metabolic anomalies in a cohort treated by 2 NRTIs + 1 PI, and absence of significant modification following PI substitution. Presented at the 7th Conference on Retroviruses and Opportunistic Infections, 2000, p 84.
 155. Viciano P, Alarcon A, Martin D, et al. Partial improvement of lipodystrophy after switching from HIV-1 protease inhibitors (PI) to efavirenz (EFV). Presented at the 7th Conference on Retroviruses and Opportunistic Infections, 2000, p 84.
 156. Bonnet E, Lepec R, Bluteau M, et al. Evolution of lipodystrophy syndrome and lipidic profile in HIV patients after switching from protease inhibitors to efavirenz. Presented at the 7th Conference on Retroviruses and Opportunistic Infections, 2000, p 84.
 157. Negro E, Cruz L, Ruiz L, et al. Impact of switching from protease inhibitors (PI) to nevirapine (NVP) or efavirenz (EFV) in patients with viral suppression. Presented at the 40th Interscience Conference on Antimicrobial Agents and Chemotherapy, 2000, p 277.
 158. Moyle GJ, Baldwin C, Dent N, et al. Management of protease inhibitor (PI)-associated lipodystrophy by substitution with efavirenz (EFV) in virologically controlled HIV-infected persons. Presented at the 39th Interscience Conference on Antimicrobial Agents and Chemotherapy, 1999, p 526.
 159. Katlama C. Successful substitution of protease inhibitors with Sustiva (efavirenz) in patients with undetectable plasma HIV-1 RNA levels: results of a prospective, randomized, multicenter, open-label study (DMP 266–027). Presented at the XIII International AIDS Conference, 2000.
 160. Bickel M, Rickerts V, Klauke S, et al. The Protra study: switch from PI to abacavir (ABC) and efavirenz (EFV) in HIV-1 infected adults previously treated with 2 NRTIs and a PI with undetectable HIV-RNA levels (vRNA). Presented at the 40th Interscience Conference on Antimicrobial Agents and Chemotherapy, 2000, p 324.
 161. Knechten H, Stürner KH, Hohn C, Braun P. 24-Week follow-up of patients switching from a protease inhibitor (PI) containing regimen with lamivudine (3TC) and stavudine (d4T) or zidovudine (AZT) to an efavirenz (EFV) based therapy. Presented at the 40th Interscience Conference on Antimicrobial Agents and Chemotherapy, 2000, p 324.
 162. Maggiolo F, Migliorino M, Pravettoni G, et al. Management of PI-associated metabolic changes by substitution with efavirenz in virologically controlled HIV + persons. Presented at the 40th Interscience Conference on Antimicrobial Agents and Chemotherapy, 2000, p 325.
 163. Lafon E, Bani Sadr F, Chandemerle C, et al. LIPSTOP study: evolution of clinical lipodystrophy (LD), blood lipids, visceral (VAT) and subcutaneous (SAT) adipose tissue after switching from protease inhibitor (PI) to efavirenz (EFV) in HIV-1 infected patients. Presented at the 40th Interscience Conference on Antimicrobial Agents and Chemotherapy, 2000, p 325.
 164. Martinez E, Romeu J, Garcia-Viejo MA, et al. An open randomized study on the replacement of HIV-1 protease inhibitors by efavirenz in chronically suppressed HIV-1-infected patients with lipodystrophy. Presented at the 8th Conference on Retroviruses and Opportunistic Infections, 2001.
 165. Estrada V, De Villar NGP, Martínez-Larrad T, et al. Switching to efavirenz from protease inhibitor-based therapy does not improve insulin resistance after one year in HIV patients with lipodystrophy syndrome. Presented at the 8th Conference on Retroviruses and Opportunistic Infections, 2001.
 166. Casado JL, Arrizabalaga J, Antela A, et al. Long-term efficacy and tolerance of switching the protease inhibitor for nonnucleoside reverse transcriptase inhibitors: a 52-week, multicenter, prospective study. Presented at the 8th Conference on Retroviruses and Opportunistic Infections, 2001.
 167. Martinez E, Conget I, Lozano L, et al. Reversion of metabolic abnormalities after switching from HIV-1 protease inhibitors to nevirapine. *AIDS* 13:805–10, 1999.
 168. Barreiro P, Soriano V, Blanco F, et al. Risks and benefits of replacing protease inhibitors by nevirapine in HIV-infected subjects under long-term successful triple combination therapy. *AIDS* 14:807–12, 2000.
 169. Ruiz L, Negro E, Domingo P, et al. Clinical, virological, and immunological benefit of switching the protease inhibitor (PI) by nevirapine (NVP) in HAART-experienced patients suffering lipodystrophy (LD): 36-week follow-up. Presented at the 7th Conference on Retroviruses and Opportunistic Infections, 2000, p 114.
 170. Carr A, Hudson J, Chuah J, et al. HIV protease inhibitor substitution in patients with lipodystrophy: a randomised, multicentre, open-label study. *AIDS* 15:1811–15, 2001.
 171. Munoz V, Casado JL, Moreno A, et al. Persistent viral suppression after switching a protease inhibitor (PI)-containing regimen to a nonnucleoside reverse transcriptase inhibitor (NNRTI)-based therapy (BEGIN study). Presented at the 39th Interscience Conference on Antimicrobial Agents and Chemotherapy, 1999, p 524.
 172. Tebas P, Yarasheski K, Powderly WG, et al. A prospective open-label pilot trial of a maintenance nevirapine (NVP)-containing regimen in patients with undetectable viral loads (VL) on protease inhibitor (PI) regimens for at least 6 months. Presented at the 7th Conference on Retroviruses and Opportunistic Infections, 2000, p 83.
 173. Raffi F, Esnault JL, Reliquet V, et al. The maintavir study, substitution of a nonnucleoside reverse transcriptase inhibitor (NNRTI) for a protease inhibitor (PI) in patients with undetectable plasma HIV-1 RNA: 18 months follow-up. Presented at the 40th Interscience Conference on Antimicrobial Agents and Chemotherapy, 2000, p 277.
 174. Buisson M, Grappin M, Piroth L, et al. Simplified maintenance therapy with NNRTI (nevirapine) in patients with long-term suppression of HIV-1 RNA: first results of a cohort study. Presented at the 40th Interscience Conference on Antimicrobial Agents and Chemotherapy, 2000, p 327.
 175. Saint-Marc T, Partisani M, Poizot-Martin I, Touraine JL. Reversibility of peripheral fat wasting (lipoatrophy) on stopping stavudine therapy. Presented at the 7th Conference on Retroviruses and Opportunistic Infections, 2000, p 85.
 176. Goebel FD, Walli RK. A novel use of abacavir to simplify therapy in PI-experienced patients successfully treated with HAART: CNA30017. Presented at the 7th Conference on Retroviruses and Opportunistic Infections, 2000, p 84.
 177. Opravil M, Hirschel B, Lazzarin A, et al. Simplified maintenance therapy with abacavir + lamivudine + zidovudine in patients with HAART-induced long-term suppression of

- HIV-1 RNA: final results. Presented at the 40th Interscience Conference on Antimicrobial Agents and Chemotherapy, 2000, p 278.
178. Montaner JSG. A novel use of abacavir to simplify therapy and reduce toxicity in PI experienced patients successfully treated with HAART: 48-week results (CNA30017). Presented at the 40th Interscience Conference on Antimicrobial Agents and Chemotherapy, 2000, p 278.
 179. Deeks SG, Hoh R, Neilands TB, et al. Interruption of treatment with individual therapeutic drug classes in adults with multidrug-resistant HIV-1 infection. *J Infect Dis* 192:1537–44, 2005.
 180. Miller V, Gute P, Carlebach A, et al. Baseline resistance and virological response to mega-HAART salvage therapies. Presented at the 6th Conference on Retroviruses and Opportunistic Infections, 1999.
 181. Workman C, Mussen R, Sullivan J. Salvage therapy using six drugs in heavily pretreated patients. Presented at the 5th Conference on Retroviruses and Opportunistic Infections, 1998.
 182. Montaner JSG, Harrigan R, Jahnke N, et al. Multi-drug rescue therapy (MDRT) following failure to multiple regimens: preliminary results [abstract 76c]. *Antiviral Ther* 3(Suppl 2):80, 1998.
 183. Deeks SG, Wrin T, Liegler T, et al. Virologic and immunologic consequences of discontinuing combination antiretroviral-drug therapy in HIV-infected patients with detectable viremia. *N Engl J Med* 344:472–80, 2001.
 184. Miller V, Sabin C, Hertogs K, et al. Virological and immunological effects of treatment interruptions in HIV-1 infected patients with treatment failure. *AIDS* 14:2857–67, 2000.
 185. Miller V, Sabin C, Hertogs K, et al. Antiretroviral treatment interruptions in patients with treatment failure: analyses from the Frankfurt HIV cohort [abstract 25]. *Antivir Ther* 5(Suppl 2):22, 2000.
 186. El-Sadr W, Neaton J. Episodic CD4-guided use of ART is inferior to continuous therapy: results of the SMART study. *NEJM* 355:2283–2296, 2006.
 187. Ananworanich J, Gayet-Ageron A, Lebraz M, et al. CD4 guided scheduled treatment interruption compared to continuous therapy: Results of the Staccato Trial. 13th Conference on Retroviruses and Opportunistic Infections, Denver, CO, 5–8 Feb 2006, abstract 102.
 188. Marchou B, Tangre P, Charreau I, et al. Structured treatment interruptions in HIV-infected patients with high CD4 cell counts and virologic suppression: results of a prospective, randomized, open-label trial (Window-ANRS 106). 13th Conference on Retroviruses and Opportunistic Infections, Denver, CO, 5–8 Feb 2006, abstract 104.
 189. Danel C, Moh R, Sorho S, et al. The CD4-guided strategy arm stopped in a randomized structured treatment interruption trial in West-African adults: ANRS 1269 Trivacan Trial. 13th Conference on Retroviruses and Opportunistic Infections, Denver, CO, 5–8 Feb 2006, abstract 105LB.
 190. Palmisano L, Giuliano M, Bucciardini R, et al. Final results of a randomized, controlled trial of structured treatment interruptions vs continuous HAART in chronic HIV-infected subjects with persistent suppression of viral replication. 13th Conference on Retroviruses and Opportunistic Infections, Denver, CO, 5–8 Feb 2006, abstract 103.
 191. Jacobson JM, Bucy PR, Spritzler J, et al. Evidence that intermittent structured treatment interruption, but not immunization with ALVAC-HIV vCP1452, promotes host control of HIV replication: the results of AIDS Clinical Trials Group 5068. *J Infect Dis* 194:623–32, 2006.
 192. Kilby JM, Bucy RP, Mildvan D, et al. A randomized, partially blinded phase 2 trial of antiretroviral therapy, HIV-specific immunizations, and interleukin-2 cycles to promote efficient control of viral replication (ACTG A5024). *J Infect Dis* 194:1672–6, 2006.

