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MINIREVIEW

Antiretroviral Therapy: Reverse Transcriptase Inhibition

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INTRODUCTION

The subject of antiretroviral therapy was first reviewed in *Antimicrobial Agents and Chemotherapy* by Hirsch and Kaplan in 1987 (35). At that time, zidovudine (AZT) was emerging as the cornerstone of efforts to control human immunodeficiency virus (HIV) type 1 (HIV-1) infection. To a great extent this is still the case, as AZT remains the drug most commonly prescribed as the initial treatment and the agent with which new therapies are typically compared. However, in most other respects the field has grown enormously, with major changes having occurred in both practical and theoretical drug development. Clinically, dideoxynucleoside reverse transcriptase (RT) inhibitors remain the mainstay of practice. The results of landmark clinical trials have changed the indications and dose recommendations for AZT, and the alternative agents dideoxyinosine (ddI) and dideoxycytidine (ddC) are now available to physicians even while clinical trials to define their precise roles are ongoing. Information derived from studies of newer, nonnucleoside RT inhibitors, agents which act at other stages in the viral life cycle, and combination therapies will likely produce major changes in the chemotherapeutic approach to the control of established HIV infection.

Antiviral agent development in the field of human retrovirology is a particular challenge. In addition to the basic but often difficult principle of developing an agent that will attack a unique target in the viral life cycle but be relatively nontoxic for the human host, there are the additional hurdles posed by the integration of HIV into the host cell genome, its capacity for persistence and latency, the complexity of the viral replication scheme, and the potential for antiviral agent resistance. This challenge, however, is being met by an intensive basic research effort which has resulted in a fundamental understanding of the molecular biology of HIV (31). The present knowledge of the viral life cycle and the complex genomic organization of HIV have created opportunities for rational and unique drug design which are among the most hopeful avenues for current and future drug development. A familiarity with the replication scheme of HIV is thus crucial to a full appreciation of antiviral chemotherapy. A detailed discussion of this topic is beyond the scope of this review but, for reference, a schematic diagram of the life cycle of HIV and a list of the major genes of the virus and their products are presented in Fig. 1 and Table 1, respectively.

This minireview will be presented in two parts. The first part will focus on agents directed at the most obvious and, to date the most successful, target of antiretroviral therapy, the RT enzyme. The second part will focus on other approaches, including newer, less proven agents, strategies with great theoretical promise, and the concept of combination therapy.

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RT INHIBITORS

Nucleoside analogs. It was demonstrated in 1985 and 1986 by Mitsuya and coworkers that a family of 2′,3′-dideoxynucleoside analogs could act as inhibitors of the HIV-1 RT and therefore inhibit the replication of HIV-1 in vitro at concentrations that were 10- to 20-fold higher than concentrations causing cellular toxicity (56, 57). The first of this family to come into widespread clinical use was 3′-azido-2′,3′-dideoxythymidine (zidovudine, AZT, or ZDV), an agent originally investigated in the 1960s as a potential anticancer agent (35).

(i) AZT. AZT is sequentially phosphorylated by the cellular enzymes thymidine kinase, thymidylate kinase, and nucleoside diphosphate kinase to its active, triphosphate form. AZT triphosphate acts as a competitive inhibitor of the viral RT and, when incorporated into a growing viral strand, causes chain termination. Additional anti-HIV activity may be conferred by the ability of AZT monophosphate to inhibit the RNase H activity of the HIV RT (92). AZT was originally reported to have in vitro activity against HIV-1 in 1985, blocking lymphocyte infection at concentrations of 1 to 5 μM (57). For susceptible isolates, the 50% inhibitory doses are <0.05 μM. AZT appears to be less active against HIV-2, requiring concentrations of over 100 μM to inhibit syncytium formation in MT-2 cells in one report (73). Although initially a point of controversy, it is now accepted that AZT is active against HIV in cells of the monocyte-macrophage lineage, despite the fact that there is decreased phosphorylation of AZT in these cells (66). It appears that the ratio of the concentration of AZT triphosphate to that of the normal nucleoside triphosphate pool is the important factor in determining the intracellular activity of AZT (66).

AZT is well absorbed orally but undergoes first-pass hepatic glucuronidation, resulting in a bioavailability of about 63% (97). The serum half-life is 1.1 h (97), and the intracellular half-life for the active, triphosphate form is 3 h (27). Unlike many nucleosides, which require cellular uptake by active transport systems, AZT can passively diffuse across cellular membranes (104). The cerebrospinal fluid (CSF)/plasma AZT ratio averages 0.6 but may be as low as 0.1 in some patients. AZT is excreted by the kidneys, approximately 20% as AZT and the remainder as the glucuronide metabolite (60, 97). Because of the importance of hepatic glucuronidation, there has been concern about potential drug interactions with other medications, such as acetaminophen, which are metabolized in this manner. Early clinical experience with AZT raised the concern of increased

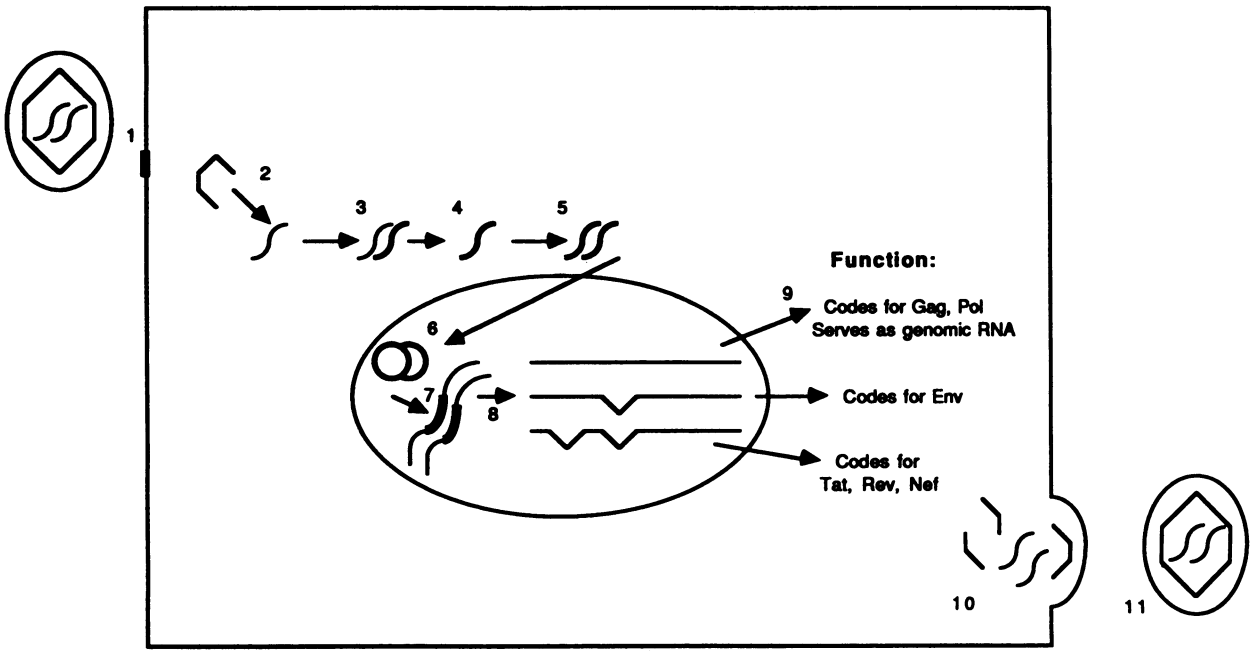

FIG. 1. Schematic diagram of the replication cycle of HIV. Stages in the cycle are identified by numbers, as follows. (1) Attachment of the virion to a specific receptor (CD4) on the cell surface; the virion is pictured with two copies of genomic, single-stranded RNA, a nucleocapsid, and an envelope. (2) Uncoating of the nucleocapsid to release viral genomic RNA. (3) Reverse transcription to produce a single-stranded proviral DNA copy (shown in bold face type). (4) Degradation of the viral RNA strand to leave a single-stranded proviral DNA copy. (5) DNA replication to produce a double-stranded proviral DNA copy. (6) Translocation to the nucleus and circularization of the proviral DNA. (7) Integration of the proviral DNA into the host cell genome. (8) Transcription of the proviral genome to produce multiply spliced (2-kb), singly spliced (4.3-kb), and unspliced (9.2-kb) mRNA species. (9) Differential export of the mRNA species (functions listed) and translation to produce viral proteins. (10) Maturation and release at the cell membrane. (11) Mature virion in the extracellular space.

AZT toxicity in patients taking acetaminophen (76); however, pharmacologic studies of this combination have not shown increases in concentrations in blood of AZT or the glucuronide metabolite with concomitant acetaminophen administration (82, 89). One drug which clearly does affect AZT pharmacokinetics by decreasing renal excretion and hepatic glucuronidation is probenecid; AZT levels are increased, and the serum half-life is prolonged (17). Although of potential utility, probenecid therapy may be problematic, as it can affect the metabolism of numerous other medications, may increase the levels of toxic AZT metabolites (16), and may potentiate drug hypersensitivity.

TABLE 1. Genes and gene products of HIV

Gene	Gene product or effect
gag	Group-specific antigens—internal structural proteins (p24, p17, and p7)
pol	RT (p66); integrase (p32); protease (p11)
env	Envelope glycoproteins (gp120 and gp41)
tat	Transactivating protein (p14); increases the efficiency of transcriptional elongation; possible posttranscriptional activity
rev	Regulator of virion expression (p19); facilitates nuclear export of unspliced and singly spliced mRNAs
nef	Originally negative regulatory factor (p27); function now unclear
vif	Virion infectivity factor (p23); important for cell-free viral infectivity; cysteine protease activity
vpu	Viral protein U (p16); important in assembly or maturation (HIV-1 only)
vpr	Viral protein R (p15); structural/regulatory protein; accelerates replication
vpx	Viral protein X (p14); structural protein; potentiates an early step in viral infectivity (HIV-2 only)
tev (*tnv*)	Regulatory fusion protein (p28) with Tat and Rev activities

A phase I trial of AZT involving 19 patients with AIDS or AIDS-related complex (ARC) was begun in 1985 and demonstrated improvements in CD4 cell counts, weight gain, a return of delayed-type cutaneous hypersensitivity reactions, and general well-being (103). Subsequently, a double-blind, randomized, placebo-controlled, multicenter phase II trial involving 282 adult patients with AIDS following a first episode of *Pneumocystis carinii* pneumonia or advanced ARC showed that AZT at a dose of 250 mg every 4 h (1,500 mg/day) reduced morbidity and mortality (25). Improvements in CD4 lymphocyte counts were also noted, although these tended to return to the baseline after 20 weeks in the patients with AIDS. The beneficial clinical effect of AZT therapy was subsequently confirmed and extended in several other studies (15, 18, 24, 75, 87, 90).

Once AZT was shown to have clinical efficacy in patients with advanced symptomatic disease, studies were quickly designed to investigate the efficacy of AZT in earlier stages of disease and to find the optimum dose regimen. In this regard, two National Institutes of Health-sponsored AIDS Clinical Trials Group (ACTG) studies were designed to examine the efficacy of AZT in HIV-positive patients who were mildly symptomatic (ACTG 016) or asymptomatic (ACTG 019).

In ACTG 016 (26), subjects had CD4 lymphocyte counts of between 200 and 800/mm^3 and mild ARC, as evidenced by oral candidiasis, oral hairy leukoplakia, herpes zoster, weight loss, diarrhea, chronic seborrhea or folliculitis, or chronic fatigue. A total of 351 subjects received placebo, and 360 received AZT at 200 mg orally every 4 h (1,200 mg/day). A total of 51 subjects developed AIDS or advanced ARC or died. In the subgroup of patients with CD4 lymphocyte counts of <500/mm^3, there were 34 events in the placebo group and 12 in the AZT group ($P = 0.0002$). In the placebo group, CD4 lymphocyte counts decreased, whereas in the AZT group, CD4 lymphocytes counts increased and, although they subsequently decreased back to baseline levels after 1 year, remained higher than those in the placebo group. A greater than 50% drop in p24 antigenemia from that at entry was seen in 65% of the AZT group and 22% of the placebo group. There was no difference in the rate of progression to Kaposi's sarcoma. Because of the significant benefit of AZT in the group with <500 CD4 cells per mm^3, the study was terminated. The mean follow-up was 11 months, although there was no obvious increase in the rate of disease progression for patients on AZT monitored for up to 22 months. For patients with CD4 counts of >500/mm^3, there were no significant differences between AZT and control groups, but there were only 198 subjects in this subgroup.

In ACTG 019 (94), patients had asymptomatic HIV infection with CD4 lymphocyte counts of <500/mm^3. In this three-armed study, patients were randomized to receive placebo ($n = 428$), AZT at 500 mg/day ($n = 453$), or AZT at 1,500 mg/day ($n = 457$). After a mean follow-up of 55 weeks, 33 patients in the placebo group had progressed to AIDS or advanced ARC, as compared with 11 in the 500-mg/day AZT group ($p = 0.002$) and 14 in the 1,500-mg/day AZT group ($p = 0.05$). Subjects receiving AZT were more likely to have a decrease in p24 antigenemia and a slowing in the rate of decline of CD4 lymphocyte levels.

Despite the statistically significant results obtained in these two large, well-conducted trials, the issue of the use of AZT in early HIV disease has remained somewhat controversial. A central question has been the following: as the vast majority of the patients did not progress and no effect on survival was demonstrated, is the widespread use of AZT justified given the cost and potential risk of inducing drug resistance? Supportive data have thus been sought from other studies. One trial (reported in abstract form) which supports the findings of ACTG 016 and ACTG 019 was conducted with individuals who had <500 CD4 lymphocytes per mm^3, were asymptomatic, or had persistent generalized adenopathy (30). Patients received either placebo or AZT at 800 mg/day. The study was terminated early because of the results of ACTG 019, but with a mean follow-up of 26 weeks, the AZT group had fewer study endpoints and higher median CD4 lymphocyte counts.

The results of two other major studies which address this issue are eagerly awaited. The European Concorde trial, which is examining the effect of 1,000 mg of AZT per day versus placebo in asymptomatic HIV-infected individuals, has undergone interim analyses and safety reviews as a result of the ACTG 019 results and has been continued. Veterans Administration Cooperative Study 298 differs from ACTG 016 and ACTG 019 in that patients had overall more advanced disease than patients in ACTG 016, the arms of the study were early versus delayed AZT therapy, with AZT being administered in the latter group when the CD4 count fell to below 200/mm^3, and the dose of AZT was 1,500 mg/day. A preliminary summary of the data was published (33, 61), and analysis of the study population as a whole confirmed the benefit of early AZT in preventing disease progression. A concern was raised, however, when analysis of the Black and Hispanic study subgroups did not reveal the same benefit and in fact revealed more deaths in the group receiving early AZT therapy (33, 61). Reassuringly, a reanalysis of the data from ACTG 016 and ACTG 019 confirmed the benefit of AZT in the minority subgroups (61). The significance of the unexpected finding of the Veterans Administration Cooperative Study is unclear; the finding may be a statistical anomaly or may relate to other, as-yet-unidentified factors (61).

With respect to the issue of dose finding in patients with advanced HIV disease, ACTG 002 was designed to evaluate the efficacy of a reduced dose of AZT in patients with AIDS following a first episode of *P. carinii* pneumonia (23). Patients received either 250 mg of AZT orally every 4 h (1,500 mg/day) or 200 mg orally every 4 h (1,200 mg/day) for 4 weeks followed by 100 mg orally every 4 h (600 mg/day) thereafter. In an intention-to-treat analysis, no differences were noted between the two groups in the time to development of another opportunistic infection, changes in the CD4 lymphocyte count, or decreases in HIV antigenemia. There was a better survival rate in the lower-dose group. Progression of HIV disease during long-term therapy was seen in both groups. The authors cautioned that the efficacy of 600 mg/day for patients with neurologic disease could not be assessed; theoretically higher levels in blood might be important in achieving therapeutic levels in the central nervous system. Another recent study (12) has suggested that AZT may have clinical benefits and an in vivo antiviral effect at doses as low as 300 mg/day, but further study is necessary before this dose can be generally recommended.

Current recommendations, promulgated following a National Institutes of Health Consensus Conference which considered the results of ACTG 002, ACTG 016, and ACTG 019, are to initiate AZT treatment at a dose of 500 to 600 mg/day in adult patients with HIV infection and CD4 lymphocyte counts of <500/mm^3 (62). Studies to evaluate the efficacy of this "lower-dose" recommendation in HIV-related neurologic disease and pediatric HIV infection are ongoing.

In controlled and uncontrolled trials published to date, AZT has been shown to be of clinical benefit in the treatment of HIV-related neurologic disease (84, 96, 102) and to improve HIV-related thrombocytopenia (59, 91). Studies of children with symptomatic HIV disease have also demonstrated a benefit from AZT therapy (7, 50, 51, 64, 68). In a recently reported open study of 88 children (aged 4 months to 11 years) with advanced HIV disease and treated with oral AZT for 24 weeks at a dose of 180 mg/m^2 every 6 h, improvements were seen in weight gain, cognitive function, serum and CSF p24 antigen levels, and CSF viral cultures. CD4 lymphocyte counts improved over the first 12 weeks, but this improvement was not sustained over the full 24 weeks of the trial (50).

With the early reports of clinical efficacy, AZT has also been considered for use as a prophylactic agent. To date, this use has primarily been considered for occupationally exposed health care workers, and guidelines for the use of AZT under these circumstances have been published (11). A number of difficulties have arisen with this approach and include the mixed results in animal studies, the low risk of HIV acquisition with most exposures, which makes efficacy studies nearly impossible and will result in exposing normal

individuals to a potentially toxic drug unnecessarily, and the failure of the drug in isolated but well-described reports (19, 44, 48). Each accidental exposure should thus be considered on an individual basis, with AZT administration being considered in those instances with the greatest attendant risk. The second intervention for which AZT prophylaxis has been considered is in attempting to interrupt vertical transmission from an HIV-infected mother to her fetus. No human data exist as yet to address the efficacy of AZT in this situation, but a trial (ACTG 076) has been designed to examine this question. In the future, other alternatives for prophylaxis, such as the recombinant soluble CD4 derivatives, drugs that are more potent than AZT, or combination treatments, may be available.

The most significant toxicity of AZT in both adults and children has been hematologic, primarily macrocytic anemia and leukopenia (18, 76). In one study of individuals with AIDS or ARC given 1,200 mg of AZT per day, approximately 80% of the patients required an interruption or modification of therapy during the first 6 months (18). In the original placebo-controlled trial, toxicity in the AZT group (1,500 mg/day) included hemoglobin levels of <7.5 g/dl in 24%, a multiple transfusion requirement in 21%, and neutropenia of <500 neutrophils per mm^3 in 16%. These side effects generally have appeared more frequently in patients with more advanced disease or with baseline anemia, neutropenia, or low serum B_{12} levels prior to therapy (76). At lower doses of AZT, the rate of observed hematologic toxicity is substantially lower (23), although some patients are unable to tolerate AZT at any dose. Subsequent studies of AZT therapy in individuals earlier in the course of disease have confirmed the observation of Richman et al. (76) that the risk of hematologic toxicity is lower in this setting. In ACTG 016 (26) (early symptomatic disease), severe anemia was seen in 5% and leukopenia was seen in 4%. In ACTG 019 (94) (asymptomatic infection), the rate of severe anemia or leukopenia in the 500-mg/day AZT group was not statistically different from that in the placebo group; in the 1,500-mg/day AZT group, anemia was seen in 6.3% and neutropenia was seen in 6.3%.

There also appears to be an increased risk of myopathy in patients taking AZT, particularly with prolonged use. The etiology may relate to a depletion of muscle cell mitochondrial DNA with a proliferation of abnormal mitochondria which is reversible with the withdrawal of AZT (1). Headache, insomnia, nausea, and nail pigmentation (28) are more minor side effects that have been noted in patients taking AZT. Isolated reports of other side effects, such as esophageal ulceration (20), macular edema (42), and meningoencephalitis with acute drug withdrawal (34), require additional confirmation. As patients have survived longer with AZT therapy and the improved management of opportunistic infections, an increased incidence of lymphoma has been reported (69). This increased incidence has raised concern about the potential long-term toxicity of AZT, but the lymphoma incidence more likely reflects the longer survival of a population with an inherently elevated risk of this complication as a result of the underlying immunocompromised state.

In addition to dose reduction, promising approaches to ameliorate the toxicity of AZT are the use of recombinant erythropoietin to counteract AZT-associated anemia (22) and the use of granulocyte-macrophage or granulocyte colony-stimulating factors (GM-CSF or G-CSF, respectively) to treat AZT-related neutropenia (32, 70). These compounds are now available for clinical use.

Despite the clear therapeutic benefits of AZT therapy, it is evident that disease progression is slowed but not halted with this drug. This phenomenon of clinical resistance to the drug may in some patients be attributed to dose reductions or interruptions due to toxicity (18); in others, this loss of efficacy is observed despite continued AZT dosing (24).

The question of the development of documentable in vitro resistance of HIV to AZT was brought into sharp focus by Larder et al. in 1989 (46). They reported the isolation of HIV with a reduced susceptibility (100-fold increase in the 50% inhibitory dose) to AZT in vitro from patients with AIDS or ARC and long-term exposure to AZT. A study of a number of these patient isolates revealed that specific amino acid substitutions in the RT enzyme (positions 67, 70, 215, and 219) were associated with high-level resistance (47). Mutations in position 215 (substituting tyrosine or phenylalanine for threonine) are consistently associated with resistance, and it appears that high-level AZT resistance is directly proportional to the number of alterations in these amino acid sites (4). Mutations in other amino acid positions in the RT enzyme have also been reported, however (38). The precise mechanism of resistance is unclear, as there is no decrease in the affinity of the mutated RT enzyme for AZT triphosphate. The possibility has been raised that a decreased sensitivity of RNase H to AZT monophosphate may be responsible for the in vitro resistance observed in certain isolates (92). In vitro resistance appears to develop more commonly in patients with more advanced disease (74, 77). In this population, resistance is relatively uncommon until after 6 months of therapy but then becomes common over the next 6 to 18 months (74, 77). Although likely to be clinically significant and an explanation for some of the clinical failures of AZT, such a clear-cut correlation with in vitro resistance has yet to be firmly established. As noted above, a major concern has been whether the widespread use of AZT in patients with early HIV disease would lead to a greater incidence of AZT resistance. Encouragingly, when isolates from patients in ACTG 016 and ACTG 019 were analyzed, there was little development of high-level AZT resistance over a period of therapy of 18 months (77).

AZT-resistant HIV strains are generally resistant to related azidonucleosides but retain their susceptibility to other nucleosides, such as ddC and ddI (45), and to nonnucleoside RT inhibitors (see below). One study has reported cross resistance to 2′,3′-didehydro-2′,3′-dideoxythymidine (d4T) (79), however. Interestingly, with time off AZT therapy, the in vitro susceptibility of AZT-resistant isolates may improve (3, 52, 88).

(ii) ddC. ddC is approximately 10 times more potent than AZT in vitro (56), inhibiting de novo lymphocyte infection at a level of 0.5 μM. ddC is sequentially phosphorylated to its active triphosphate form by the cellular enzymes deoxycytidine kinase, cytidine monophosphate kinase, and nucleoside diphosphate kinase. It is well absorbed, with an oral bioavailability of 87%, a plasma half-life of 0.34 h, and an intracellular half-life of the triphosphate form of 2.6 h. The CSF/plasma ddC ratio is approximately 0.2 (97).

A phase I study of ddC in 20 patients with AIDS or ARC revealed improvement in surrogate markers of HIV infection (101). ddC administration resulted in some weight gain and an increased energy level in the study population, without the major hematologic toxicity associated with AZT. However, this study was limited by the common appearance at doses of greater than 0.09 mg/kg daily of a severe, painful peripheral neuropathy which appeared to be related to both the daily and the cumulative doses of ddC. It was also

observed that at 4 to 6 weeks into therapy, many patients suffered a syndrome of maculovesicular cutaneous eruptions, aphthous oral ulcers, fever, and malaise, although these symptoms often subsided with continued therapy. As a result of these problems, ddC development was slowed. Subsequently, ACTG 012 demonstrated in 15 patients with AIDS or ARC that ddC had an antiviral effect at a lower dose (0.005 mg/kg every 4 h), with much less toxicity (neuropathy in 2 of 15 patients) (53, 54). ddC is now being tested comparatively with AZT in patients who are intolerant of AZT or who have not responded to treatment with AZT (5) and in combination with AZT (see second part [13a]). The dose of ddC now generally used for adults is 0.75 mg every 8 h. In these trials, adverse effects not previously reported with ddC—pancreatitis and possibly esophageal ulceration and cardiomyopathy—have been noted.

One report of ddC resistance developing with treatment has appeared (36). In this instance, an isolate from a patient 17 months into therapy was found to have a Thr→Asp mutation at amino acid position 69 in the viral RT.

(iii) ddI. ddI (didanosine) is a purine analog whose pathway of anabolic phosphorylation is not completely defined. One likely predominant pathway is phosphorylation intracellularly by 5′-nucleotidase, conversion to 2′,3′-dideoxyadenosine (ddA) monophosphate by adenylosuccinate synthetase and adenylosuccinate lyase, and finally triphosphorylation to the active antiretroviral moiety, ddA triphosphate (49). ddA is also an effective antiretroviral agent in vitro, but in humans ddI as a prodrug of ddATP is preferable, because orally administered ddA is cleaved by acid in the stomach to form adenine and dideoxyribose, and the adenine product may cause nephrotoxicity. Like AZT, ddI may enter cells passively (49). ddI has been shown to be effective in vitro against HIV-1, HIV-2, human T-cell lymphotropic virus type 1, and simian immunodeficiency virus (49). ddI inhibits HIV infection in vitro at 1 to 10 μM, levels which are at least 10- to 20-fold lower than toxic levels (56).

Because of the susceptibility of ddI to stomach acid, the oral availability of ddI is lower than that of AZT or ddC but may be improved to about 40% with the addition of a buffer or the concomitant use of antacids and by taking ddI in the fasting state. The bioavailability may be widely variable for this drug; in the future, plasma ddI levels may prove useful. There is little information about drug interactions to date, but the buffered ddI preparation may interfere with the absorption of dapsone and other drugs dependent on gastric acidity for absorption, such as ketaconazole. The administration of such drugs 2 h prior to the administration of ddI is therefore recommended. The plasma half-life is short, 0.5 h, but the intracellular half-life of the active drug is 12 h or more, allowing for less frequent dosing of ddI than of AZT or ddC. The CSF/plasma ddI ratio is 0.2 (97). ddI is actively excreted by the renal tubules and may be partly metabolized through an alternate salvage pathway via a hypoxanthine intermediate.

In a series of phase I, short-term trials involving a total of 92 patients with AIDS or ARC (14, 43, 80, 98), ddI was shown to be biologically active, with an improvement in CD4 lymphocyte counts, a reduction in p24 antigenemia, weight gain, and the return of cutaneous hypersensitivity reactions. The response was generally greater among patients with ARC and in those who had not had prior AZT therapy and less clear among those with more advanced disease (99). A study of ddI in 43 children over 24 weeks showed decreases in p24 antigenemia and improvements in CD4 lymphocyte counts, with the greatest improvement in those with higher counts at entry (9). Subjects gained weight and had decreased adenopathy and organomegaly. The plasma ddI levels correlated with the improvements seen in IQ scores and the decreases in p24 antigenemia.

Three major phase II/III ACTG studies have been undertaken to compare the effectiveness of ddI with that of AZT in patients who have symptomatic or asymptomatic HIV infection and who have received <4 months of AZT (ACTG 116), to randomize individuals who have already received AZT for >4 months to either continued AZT or to ddI to try to clarify the issue of AZT resistance (ACTG 117), and to examine different dose regimens for ddI in patients who are intolerant of AZT (ACTG 118). ddI had also been made available by the manufacturer as part of an expanded-access program which permitted patients who did not qualify for available protocols or who were geographically isolated from study centers to receive this agent. As of March 1991, the manufacturer reported that a total of over 24,000 patients had taken ddI, including 5,035 in phase II/III trials, 14,114 in the U.S. Treatment Investigational New Drug program, and 4,624 in the open-label "compassionate-use" track (6). Parallel-track availability had the great advantage of providing an alternative antiretroviral agent for patients who could not take AZT, but the larger number of patients on ddI outside the ACTG trials raised concerns that valuable information about the clinical efficacy of ddI might be delayed. Nevertheless, on the basis of phase I data and a preliminary analysis of CD4 responses in ACTG 117, ddI was recently licensed for adults and children (>6 months old) with advanced HIV infection and either intolerance of AZT or a lack of response to treatment with AZT. It is available as a chewable buffered tablet and as a buffered powder for oral solution. Dosing is by weight, with the recommended dose for adults of 50 to 74 kg being 200 mg twice daily and 250 mg twice daily for the two preparations, respectively. The final results of ACTG 116, ACTG 117, and ACTG 118 will be crucial in refining the therapeutic indications for ddI as single-agent therapy. As with ddC, there is considerable interest in examining combination regimens which alternate or combine ddI with AZT (see second part [13a]).

ddI may also be beneficial to patients with certain forms of HIV-related neurologic disease (9, 100). Five patients with dementia due to HIV and treated with ddI showed improvement in memory and attention in one report (100). In a study of 64 children with symptomatic HIV infection, 21% showed improvement in IQ scores over the first 6 months of treatment; 70% remained unchanged (95). Little subsequent change was noted in the second 6-month period. Interpatient differences in drug bioavailability were again important in this pediatric trial (95). Further evaluation of ddI for this indication is needed and, as ddI can cause peripheral neuropathy (see below), its efficacy against HIV-related neurologic disease remains to be confirmed.

ddI is less toxic than AZT for bone marrow progenitor cells and has been associated with relatively little hematologic toxicity, even in patients with prior hematologic intolerance of AZT (13, 14, 43, 80, 98, 99). The dose-limiting toxicity of ddI has been a painful peripheral neuropathy, which is more common at higher daily and greater cumulative doses. Neuropathy has generally been more readily reversible with cessation of ddI therapy when recognized early. Additionally, there has been an increased incidence of pancreatitis, ranging from mild to life threatening, which may be dose related. Because of these toxicities, it is recommended that ddI not be used in patients with preex-

isting neuropathy or a history of pancreatitis and that the use of other drugs known to cause these conditions be limited. Patient monitoring should include serial amylase determinations, and there is a suggestion that serum lipase may be a more sensitive indicator in some cases. Elevated triglycerides have also been noted in some patients with ddI-associated pancreatitis; as yet it is unknown whether this elevation is a cause or an effect of the pancreatitis. Diarrhea is the most commonly noted adverse clinical event, but is usually tolerable. Elevated uric acid levels are also commonly noted but have not yet been associated with clinical consequences of gout or nephropathy. Other problems noted have included minor liver function abnormalities, although a case of fatal hepatitis in which ddI may have been contributory has been reported (41), and transient subjective complaints of insomnia or headache. Patients on long-term therapy often complain of xerostomia, and some of these patients have had elevations of serum salivary amylase. Seizures have been reported in association with ddI therapy, but the complexity of these cases has made it difficult to be certain of a direct etiologic link.

The now well-described phenomenon of AZT resistance has led to an intense search for evidence of ddI resistance. To date, the >100-fold decrease in susceptibility seen in isolates from patients with advanced disease on AZT treatment has not been described in isolates obtained from patients on long-term ddI therapy, but a tendency toward diminishing ddI susceptibility with time has been seen (3, 38, 52, 72, 88). This resistance is associated with a Leu→Val substitution at position 74 in the HIV RT which also confers decreased susceptibility to ddC (88). Interestingly, this mutation improves AZT susceptibility in isolates with one or more AZT resistance-associated codon changes (i.e., at positions 67, 70, 215, and 219). This result correlates with reports of improved AZT susceptibility in isolates with diminished ddI sensitivity (3, 52, 88).

(iv) Other nucleosides. d4T is another nucleoside analog which has entered clinical trials. This compound is somewhat less potent than AZT but is much less toxic for bone marrow progenitor cells in vitro, perhaps because it may be less likely than AZT to deplete cellular stores of thymidine triphosphate. It is acid stable and highly bioavailable. In a phase I trial, patients showed improvement in symptoms, increases in CD4 lymphocyte counts, and decreases in serum p24 antigenemia (8, 86). The major dose-limiting toxicity to date has been peripheral neuropathy. 3′-Azido-2′,3′-dideoxyuridine has also been shown to have antiviral effects in small phase I trials (58, 71); as yet, the toxicity profile is not well described. There are numerous other candidate nucleoside analogs which are under investigation, with chemical modifications designed to maximize activity against the viral RT, improve the pharmacokinetic profile, and minimize toxicity. Among these are halogenated nucleosides (85), nucleotide dimers (83), and a series of acyclic and carbocyclic nucleoside analogs, such as carbovir (carbocyclic 2′,3′-didehydro-2,3-dideoxyguanosine) (10) and HEPT {1-[(2-hydroxyethoxy)methyl]-6-phenylthiothymine} (93). HEPT derivatives are nucleoside analogs that do not require anabolic phosphorylation for their activity and are HIV-1 specific (2); thus, they possess the characteristics of some of the nonnucleoside RT inhibitors (see below).

Nonnucleoside RT Inhibitors. (i) Phosphonoformate. Phosphonoformate (foscarnet sodium) is a PP_i analog that is active against herpesviruses as well as HIV. It is a noncompetitive inhibitor of the HIV RT and has been shown to have anti-HIV-1 activity (81) and to exhibit synergism with AZT in vitro (21, 39). Studies of patients who have HIV infection and who have received foscarnet for cytomegalovirus infection have revealed some evidence of antiretroviral activity, such as decreased serum p24 antigenemia, as have small trials of foscarnet as a primary antiretroviral therapy (37). Progress in its use as an antiretroviral agent has been slowed by the requirement for intravenous administration and the toxicity profile, which includes nephrotoxicity and altered calcium and phosphorus metabolism. Its clinical role will likely prove to be more in the treatment of acyclovir-resistant herpes simplex and varicella-zoster virus infections and in the treatment of cytomegalovirus infections. Foscarnet was recently licensed for treatment of cytomegalovirus retinitis.

(ii) TIBO Compounds. A search for other compounds with antiretroviral activity led to the discovery of a series of TIBO {tetrahydro-imidazo[4,5,1-jk][1,4]-benzodiazepin-2(1H)-one and -thione} derivatives, nonnucleosides that inhibit HIV-1 in vitro at nanomolar concentrations, which are 10,000- to 100,000-fold lower than the concentrations causing cellular toxicity (65). These agents inhibit HIV-1 but not HIV-2, other retroviruses, or other RNA and DNA viruses tested to date. A pilot trial of R82913 (a representative of this class) in 22 patients with AIDS or ARC at intravenous doses of 10 to 300 mg daily demonstrated that the agent was well tolerated for up to 50 weeks and had a half-life of 3 days (67). Subtle effects on CD4 cell counts and serum p24 antigen concentrations were seen, and no overt clinical benefit was noted in this phase I trial.

(iii) Nevirapine (BI-RG-587). This recently described compound has stimulated a great deal of interest. It is a dipyridodiazepinone that inhibits HIV-1 in vitro at nanomolar concentrations but does not inhibit HIV-2. The antiretroviral activity of this compound was discovered during a screening of many compounds and was not predicted on the basis of its structure and the known structure of the RT enzyme (40). It appears to act through noncompetitive binding at a site different from the template and nucleoside binding site (55). In studies published to date, it has shown synergy with AZT (78) and has been effective against AZT-resistant isolates. Nevirapine also shows partial inhibition of HIV RNase H activity. The drug is nontoxic for human cells, including bone marrow progenitor cells, in vitro at doses several thousand times higher than effective doses (40) and does not appear to inhibit human DNA polymerases. Clinical trials of this agent alone and in combination with AZT have been initiated.

(iv) L-697,639 and L-697,661. L-697,639 and L-697,661 are pyridinone derivatives that are HIV-1 specific, inhibiting viral replication at nanomolar concentrations (29). They have been shown to have activity against both laboratory and clinical isolates, including AZT-resistant strains (29). Synergy with AZT or ddI has also been demonstrated (29). Although structurally distinct from TIBO compounds and nevirapine, L-697,639 and L-697,661 are also noncompetitive inhibitors of the HIV-1 RT and bind to the same site on the RT-template-primer complex. Viral isolates resistant to this class of compounds can be readily selected in vitro and exhibit cross-resistance to the TIBO compounds and nevirapine (29, 63). Clinical trials of these agents have begun, but concern for the rapid emergence of resistance has given impetus to an examination of these drugs in combination with other agents (e.g., AZT).

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