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Twenty-Five Years of Lamivudine: Current and Future Use for the Treatment of HIV-1 Infection

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Abstract: Innovation in medicine is a dynamic, complex, and continuous process that cannot be isolated to a single moment in time. Anniversaries offer opportunities to commemorate crucial discoveries of modern medicine, such as penicillin (1928), polio vaccination (inactivated, 1955; oral, 1961), the surface antigen of the hepatitis B virus (1967), monoclonal antibodies (1975), and the first HIV antiretroviral drugs (zidovudine, 1987). The advent of antiretroviral drugs has had a profound effect on the progress of the epidemiology of HIV infection, transforming a terminal, irreversible disease that caused a global health crisis into a treatable but chronic disease. This result has been driven by the success of antiretroviral drug combinations that include nucleoside reverse transcriptase inhibitors such as lamivudine. Lamivudine, an L-enantiomeric analog of cytosine, potently affects HIV replication by inhibiting viral reverse transcriptase enzymes at concentrations without toxicity against human polymerases. Although lamivudine was approved more than 2 decades ago, it remains a key component of first-line therapy for HIV because of its virological efficacy and ability to be partnered with other antiretroviral agents in traditional and novel combination therapies. The prominence of lamivudine in HIV therapy is highlighted by its incorporation in recent innovative treatment strategies, such as single-tablet regimens that address challenges associated with regimen complexity and treatment adherence and 2-drug regimens being developed to mitigate cumulative drug exposure and toxicities. This review summarizes how the pharmacologic and virologic properties of lamivudine have solidified its role in contemporary HIV therapy and continue to support its use in emerging therapies.

Key Words: 3TC, lamivudine, NRTI, ART, M184V

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INTRODUCTION

Lamivudine (2′-deoxy-3′-thiacytidine, 3TC) is a first-generation nucleoside reverse transcriptase inhibitor (NRTI) that was approved for the treatment of HIV-1 infection in 1995 and hepatitis B virus (HBV) infection in 1998.[1,2] Lamivudine has been evaluated in more than 50 clinical studies registered on ClinicalTrials.gov involving >25,000 patients. Since 2002, the World Health Organization (WHO) has been recommending treatment regimens for HIV infection, and both 3TC or emtricitabine (FTC) are preferred components of nearly all fixed-dose combinations.[3] Moreover, in the updated 2016 WHO guidelines, 3TC continues to be recommended as part of fixed-dose combinations as first- and second-line antiretroviral therapy (ART) for adults, adolescents, children, and infants.[3] In addition, WHO recommends that patients coinfected with HIV and HBV use a first-line ART combination containing tenofovir disoproxil fumarate (TDF) plus 3TC or FTC. The WHO guidelines are consistent with those recommended by the International Antiviral Society,[4] European AIDS Clinical Society,[5] and the US Department of Health and Human Services.[6–8] HIV treatment has evolved to include single-tablet regimens containing potent 3- or 4-drug combinations, and 3TC has remained a well-established component in many combination strategies as HIV treatment continues to evolve.

BIOCHEMISTRY OF 3TC

Investigators first reported on the promise of the racemic mixture of BCH-189 to treat HIV-1[9] and HBV.[10] On separation of the enantiomers in this mixture, the plus and minus forms of BCH-189 were found to have activities against the replication of HIV-1 and HIV-2.[11,12] However, the minus (non-natural) enantiomer was unexpectedly more potent than the plus (natural) enantiomer, was associated with markedly less cytotoxicity in human lymphocyte cultures, and was therefore developed as an antiretroviral therapeutic.[11–13] After approval from the US Food

and Drug Administration (FDA), this levo-enantiomer was given the name "lamivudine."

Lamivudine is a nucleoside analog of 2′-deoxycytosine[9] that exerts its antiviral effects by acting as a DNA chain terminator (Fig. 1). The active anabolite of 3TC, lamivudine 5′-triphosphate, is formed from phosphorylation by intracellular kinases and competes with naturally occurring cytidine triphosphate for incorporation into DNA.[14,15] Lamivudine is a potent inhibitor of the reverse transcriptase (RT) enzymes of HIV-1, with in vitro IC_{50} in different cell lines with different HIV-1 strains ranging from 0.002 to 1.14 μM[9,11–13] and IC_{50} against HBV of 0.1 μM[10] with limited cell toxicity at concentrations >1000-fold than those effective against HIV. The unnatural nucleoside structure of 3TC, an L-(−)-enantiomer, is not recognized as a substrate by human polymerases (all natural nucleosides have a D configuration) at biologically relevant concentrations. The unique chemical structure of 3TC, which is characterized by excellent antiviral activity with little toxicity, strongly contributes to its clinical success.

Early characterization of 3TC revealed that resistance to the drug develops rapidly in vitro.[16] Compared with other NRTIs, resistance to 3TC or FTC occurs via mutations (see Table, Supplemental Digital Content 1, http://links.lww.com/QAI/B131, which shows major mutations associated with resistance to NRTIs).[17] Mutations of codon 184 result in substitution of isoleucine or valine in place of the wild-type methionine via a missense mutation (M184I or M184V).[16] Amino acid 184 is located in the highly conserved tyrosine–methionine–aspartate–aspartate (YMDD) motif of HIV-1 RT (necessary for proper catalytic function of the enzyme) and confers high-level (>100-fold) resistance to 3TC.[16,18] However, the mutation was also found to increase susceptibility to zidovudine (ZDV)[19] and, subsequently, to stavudine (d4T)[20] and tenofovir.[21] In addition, by affecting the catalytic function of RT, M184V lowers processivity and enhances fidelity of HIV RT, resulting in low viral fitness. Purified M184V variant proteins have exhibited 2- to 6-fold reduced mutation frequency compared with wild-type RT,[22,23] perhaps contributing to the reduced rate of drug resistance. The combination of these effects results in residual antiviral activity, even in the presence of 3TC monotherapy, in patients who harbor the M184V/I substitution.[24] In a 48-week prospective study of 58 evaluable patients with virologic failure on ART containing 3TC and with evidence of treatment-emergent M184V substitution, immunological or clinical failure occurred in 69% of patients who discontinued ART at baseline and 41% of patients who continued ART with 3TC monotherapy ($P = 0.064$).[24] In addition, in a study of 1895 patients randomized to receive 3TC, 3TC and loviride, or placebo in combination with their current regimen of either ZDV, ZDV and didanosine (ddI), or zalcitabine (ddC), 3TC reduced the risk of HIV disease progression by 57% compared with placebo (hazard ratio, 0.42; $P < 0.0001$).[25] Because of the possible benefits of M184V, guidelines recommend considering continuation of 3TC or FTC in particular situations even if M184V has been documented.[3,26] Clinical data in 132 treatment-experienced, virally suppressed patients infected with HIV containing multiple mutations, including M184V, demonstrated that a boosted protease inhibitor [ritonavir-boosted darunavir (DRV/r) or ritonavir-boosted lopinavir (LPV/r)] plus 3TC was superior to boosted protease inhibitor monotherapy, with only 4 patients [3%; 95% confidence interval (CI) 0.8 to 7.6] experiencing viral failure after 48 weeks of treatment.[27] These data support the hypothesis that selection of M184V by 3TC results in residual antiviral activity that can be effective in controlling viral replication in combination with other antiviral agents.

PHARMACOLOGY

Lamivudine is rapidly absorbed after oral administration, with maximum serum concentrations reached between

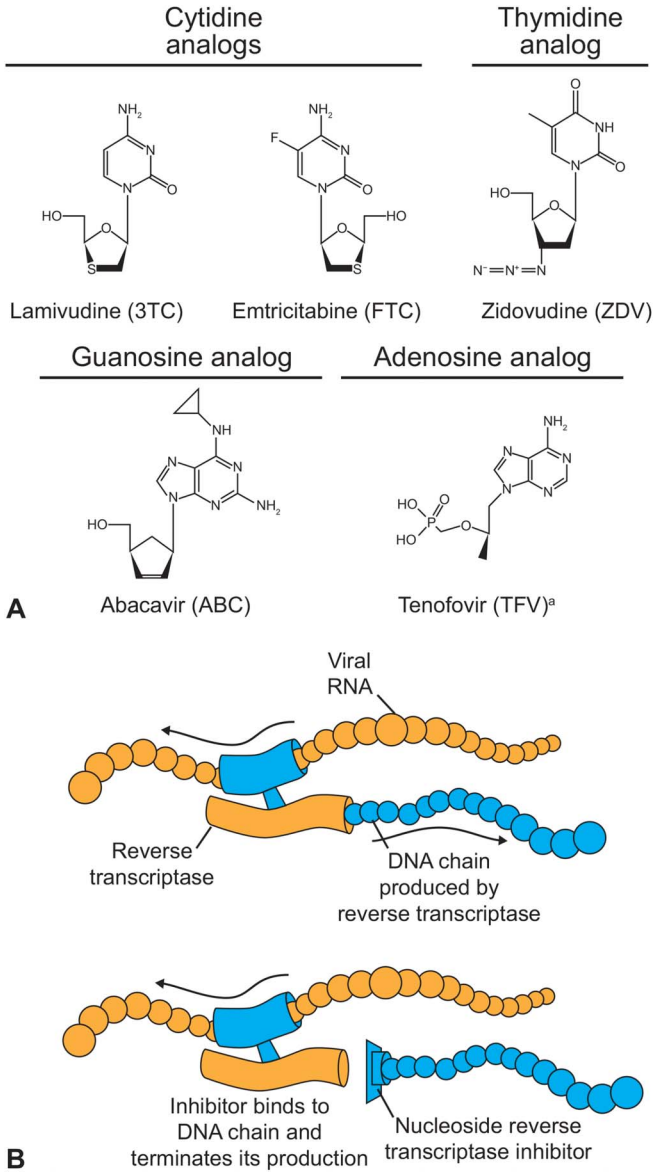

FIGURE 1. (A) Structures of lamivudine and other NRTIs used in contemporary HIV therapy. (B) Mechanism of action of NRTIs. [a]Tenofovir (TFV) is the active moiety of TDF and tenofovir alafenamide fumarate (TAF).

TABLE 1. Virological, Biochemical, and Pharmacological Characteristics of NRTIs Used in Contemporary HIV Therapy

	3TC	ABC	FTC	ZDV	TDF*	TAF*
HIV-1 reverse transcriptase in MT-4 cells†, mean (SD, n), μM[104–106]	2.1 (0.6, 7)	4.0 (1.6, 21)	0.5 (NA, 2–3)	0.040 (0.005, 51)	4.2 (0.8, 2)	0.005 (0.002, 2)
Intracellular half-life, h[75,107]	10.5–15.5	3.3	>20	3–4	>60	NA[108]‡
Plasma or serum $T_{1/2}$, h[75,107,109]	5–7	1–2	7–10	0.8–1.9	17	0.4
Reverse transcriptase K*i*§‖, mean (SE), nM[110–112]	233 (28)	10 (1)¶	430 (60)	4.4 (2)	980 (NA)#	NA
Relative mtDNA content after 25 days of treatment, mean % vs untreated control cells** (SD)[61,113]	137 (7)	134 (27)	110 (15)	118 (24)	101 (20)	107 (16)

3TC, lamivudine; ABC, abacavir; EC_{50}, drug concentration needed to inhibit 50% of viral spread; FTC, emtricitabine; IC_{50}, half-maximal inhibitory concentration; K_i, apparent inhibitor dissociation constant; mtDNA, mitochondrial DNA; n, number of determinations; NA, not available; SD, standard deviation; SE, standard error of the mean; TAF, tenofovir alafenamide fumarate; TDF, tenofovir disoproxil fumarate; ZDV, zidovudine.

*Active moiety is tenofovir diphosphate.

†IC_{50} for 3TC, ABC, FTC, and ZDV; EC_{50} for TDF and TAF.

‡Could not be estimated.

§Inhibition constants for the triphosphate analogs of 3TC, ABC, FTC, and ZDV.

‖Values were generated using a homopolymeric RNA/DNA template for 3TC; DNA template for TDF, FTC, and ZDV; and RNA template for ABC.

¶In the form of carbovir, the active metabolite of ABC.

#Average standard error for K_i values for TDF and its analogs was 16%.

**Changes were observed after 25 days of treatment in HepG2 cells for 3TC, ABC, FTC, and ZDV; changes were observed after 10 days of treatment in MT-2 cells for TDF and TAF.

0.5 and 1.5 hours after dosing and a dominant elimination half-life of approximately 5–7 hours (Table 1).[14] The absolute bioavailability of 3TC is 82% in adults and 68% in children. Systemic exposure is not affected when 3TC is administered with food and is consistent across sex and race. The in vitro intracellular half-life of lamivudine 5′-triphosphate is 10.5–15.5 hours in HIV-infected cell lines and 17–19 hours in HBV-infected cell lines, which supports a minimum dosing interval of 12 hours. Approximately 5.2% of 3TC is metabolized and excreted as a minor product, with the majority remaining unchanged and undergoing active organic cationic secretion through the kidneys.[28]

Considering 3TC's low metabolic clearance, minimal binding to plasma protein, and no observed effects on hepatic metabolism, it is expected that 3TC would have few clinically relevant pharmacological interactions with concomitantly administered drugs.[14] Of note, ZDV, ddI, cotrimoxazole, and interferon-α-2b have all been shown to result in minor pharmacokinetic changes that do not require a dose adjustment.[14] In a 2012 study, an unexpected interaction was observed between oral solutions of abacavir (ABC) and oral solutions of 3TC,[29] which led to a 2017 study to determine whether sorbitol, an excipient of other antiviral liquid formulations, alters 3TC pharmacokinetics (PK) by altering osmolarity in the intestine, thus reducing the absorption.[30] Maximal concentrations of plasma 3TC were shown to be reduced in a dose-dependent manner as much as 55% when coadministered with sorbitol 13.4 g; plasma 3TC exposure was reduced by 36%–44% in the presence of sorbitol 13.4 g.[30] Decreases in plasma exposure corresponded to increased apparent oral clearance by 57% with sorbitol 13.4 g. Therefore, it is likely that avoiding coadministration of 3TC and sorbitol-containing medicines will be recommended, requiring a switch to tablet regimens.

Because the potential viral replication in reservoir sites, such as the genital tract and the central nervous system, is an important challenge to preventing transmission and the future possibility of cure, it is important to understand the drug's PK in these sites.[31] On the basis of PK modeling, concentrations of 3TC and the active triphosphate analog 3TC-TP with the first dose were greater than the targeted IC_{50} in seminal mononuclear cells and peripheral blood mononuclear cells (PBMCs), partially because of the long half-life in seminal mononuclear cells (159 hours) compared with PBMCs (5 hours). Another PK model estimated that the ratio of 3TC exposure in the cervicovaginal fluid to that in the blood plasma was between 4.6 and 9.9 (at first dose and multiple doses) depending on the strength of the dose, suggesting a high level of 3TC disposition into the female genital tract.[32] By contrast, study results on 3TC penetration into the central nervous system have been inconsistent. In a preclinical study in primary human fetal astrocytes and astrocyte cell lines, 3TC was less potent in astrocytes compared with PBMCs, resulting in an estimated 90% inhibitory concentration for 3TC in astrocytes being 12-fold greater than the maximum 3TC concentration that could occur in the cerebrospinal fluid (CSF).[33] However, in a PK study of children aged 8–18 years infected with HIV, the median (interquartile range) CSF concentration of 3TC was 97 (51–144) ng/mL, which is much higher than the reported IC_{50} of 0.5–3.4 ng/mL, suggesting that 3TC demonstrates antiviral potency in the CSF in children.[34]

EARLY DRUG DEVELOPMENT

When 3TC was in clinical development, HIV infection was treated with first-generation NRTI monotherapy using ZDV, ddI, ddC, or d4T. The limited clinical success of this approach motivated the investigation of 2-drug combinations. Clinical trials of ZDV in combination with ddC or ddI were the first to demonstrate that concurrent or alternating administration of 2 NRTIs was superior to administration of ZDV alone.[35–37]

Lamivudine was evaluated in clinical studies alone and in combination with ZDV. The NUCA 3001 and NUCA 3002 studies in North America and the NUCB 3001 and NUCB

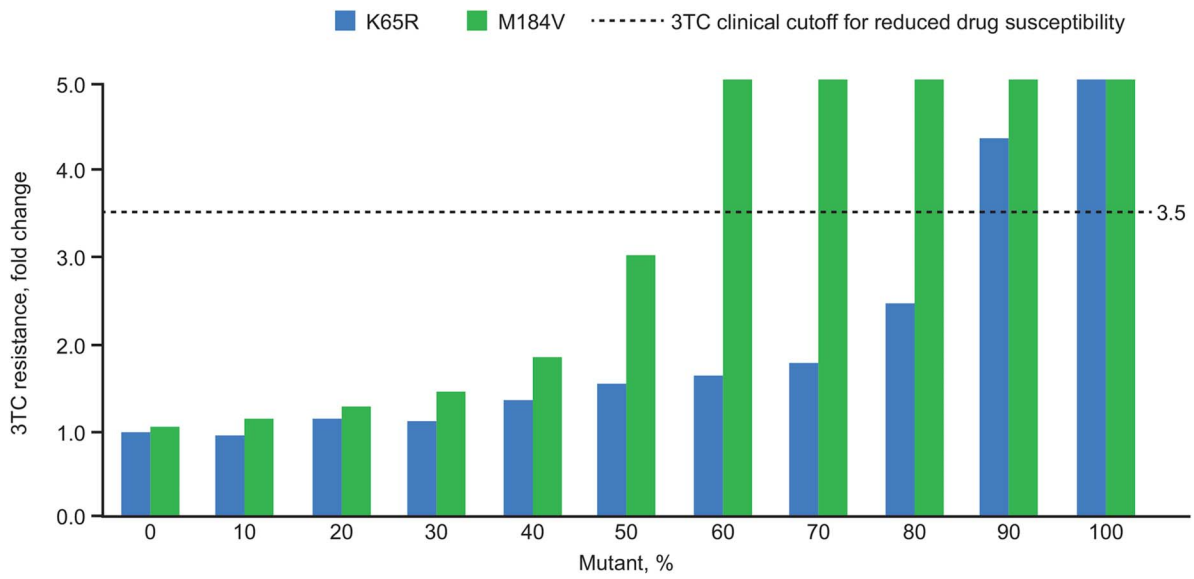

FIGURE 2. Effect of differing relative concentrations of either M184V or K65R mutations when combined with wild-type HIV. 3TC, lamivudine. Reproduced from Underwood MR, Ross LL, Irlbeck DM, et al. Sensitivity of phenotypic susceptibility analyses for nonthymidine nucleoside analogs conferred by K65R or M184V in mixtures with wild-type HIV-1. *J Infect Dis*. 2009;199(1):84–88, by permission of the Infectious Diseases Society of America.

3002 studies in Europe conducted between 1993 and 1994 were the first randomized controlled trials to evaluate the efficacy of 3TC.[38–41] The trials compared the combination of 3TC and ZDV with ZDV monotherapy[38–40] and ZDV in combination with ddC,[42] with change from baseline in $CD4^+$ cell counts as the primary end point; reduction in plasma viral load was evaluated as an exploratory end point. The results of these trials at 52 weeks showed increased $CD4^+$ cell counts in the combination treatment groups, whereas $CD4^+$ counts were slightly decreased or similar to baseline in the ZDV or 3TC monotherapy treatment groups, respectively. More importantly, viral load was significantly reduced at 52 weeks in the combination group compared with smaller decreases in either monotherapy group.[39] The trials that studied 2 different doses of 3TC found no significant differences between the 150-mg twice-daily and 300-mg twice-daily doses in viral load reduction and $CD4^+$ cell counts.[39,40] A subsequent trial showed similar efficacy in patients receiving 3TC 300 mg once-daily versus 3TC 150 mg twice-daily when combined with ZDV and efavirenz (EFV).[43]

RESISTANCE

The efficacy of 3TC monotherapy was limited by the early development of a mutation causing the M184I substitution in RT (followed by emergence of M184V), detected within the first month of 3TC monotherapy in both treatment-naive and treatment-experienced patients.[28,44] Although HIV-1 RNA reduction of 1.3 logs occurred in the first 2–4 weeks of 3TC monotherapy, selection of the M184V mutant virus resulted in a subsequent rebound in viral load, leading to an overall viral load reduction of 0.5 logs at 48 weeks.[28] An in vitro analysis of heterogeneous mixtures of M184V and wild-type HIV showed that the relative concentration of M184V had to be at least 20% to result in a >1-fold increase in 3TC resistance and more than 50% to result in a fold change greater than 3TC's clinical cutoff of 3.5; these results were consistent with the reported assay validation data (Fig. 2).[45] Interestingly, after selection of M184V, some in vitro activity of 3TC was retained when it was combined with other agents, even in the presence of full resistance to 3TC.[46] In vitro studies showed that the M184V variant preferentially incorporates deoxycytidine triphosphate instead of lamivudine 5′-triphosphate 20- to 100-fold more frequently than wild-type virus.[15] This mechanism of resistance contrasts with the mechanism of ZDV resistance. Reverse transcriptase carrying ZDV resistance mutations still incorporates ZDV, but chain termination is relieved by excision of the terminal nucleotide analog; ZDV resistance mutations accelerate the rate of excision reaction.[47] The M184V mutation sensitizes HIV-1 ZDV and reverses the effect of ZDV resistance mutations by reducing the rate of ZDV excision.[47] Therefore, the observed sustained response to 3TC/ZDV combination therapy is explained in part by the sensitization to ZDV by the M184V mutation.[48] The synergy between 3TC and ZDV played a crucial role in early suppressive therapy against HIV and heralded the beginning of the combination ART era with 3TC/ZDV, the first antiretroviral fixed-dose combination.

Mechanistic studies have also demonstrated that a reduction in HIV replicative capacity by an estimated 10% leads to a reduction in fitness associated with the M184V variant.[48] The presence of the M184V substitution further reduces the viral fitness of HIV viruses carrying other drug resistance mutations known to affect viral fitness (eg, K65R in RT and R263K in integrase).[49,50] Slower replication of mutant viruses may further reduce viral fitness in $CD4^+$ lymphocytes, which possess low concentrations of deoxynucleoside triphosphates.[46,51] Interestingly, 3TC has shown

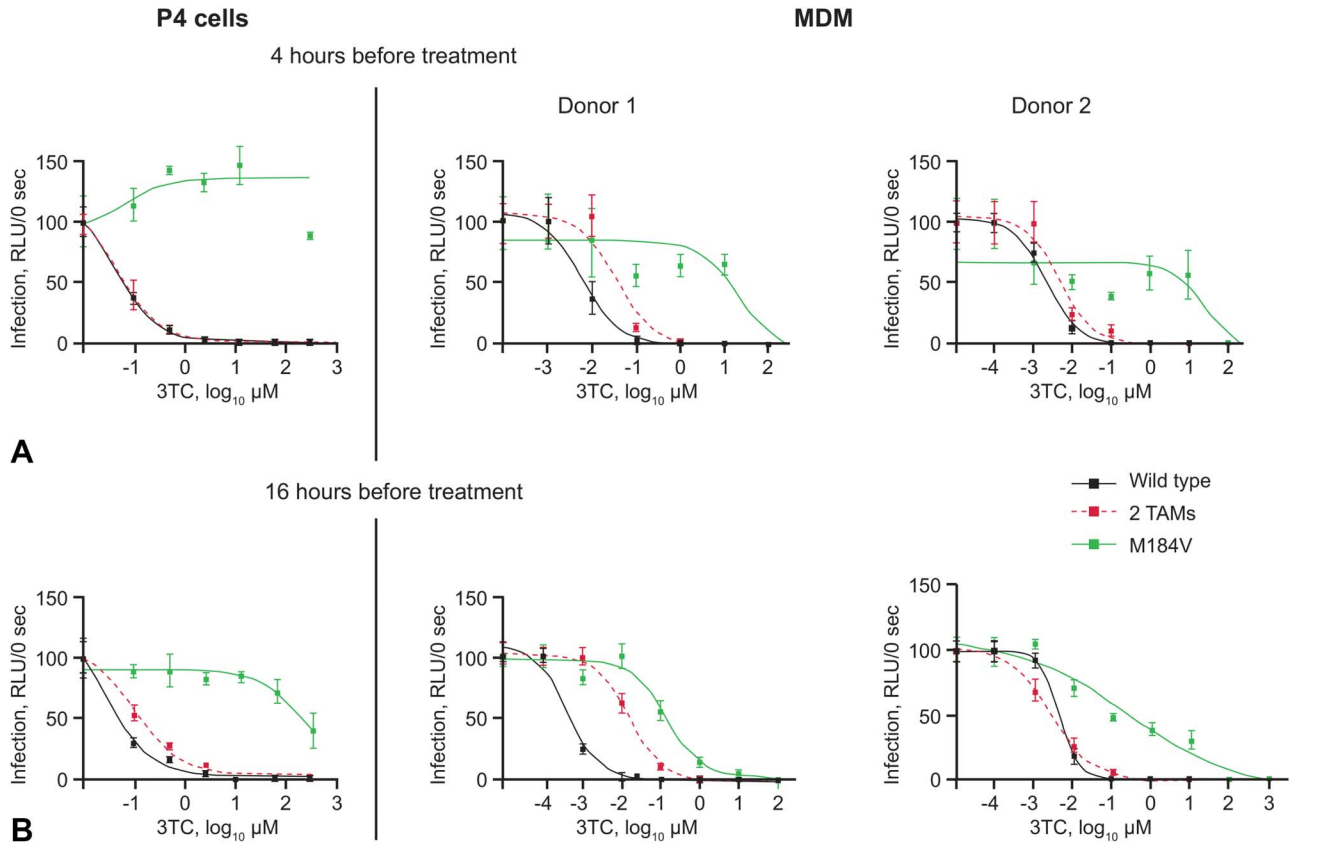

FIGURE 3. In vitro inhibition of wild-type, two-TAM, and M184V HIV-1 by 3TC in P4 cells and MDMs (A) 4 hours pretreatment and (B) 16 hours pretreatment. Data represent mean values of quadruplicate wells. Independent experiments (n = 11) were performed with MDMs from different donors. 3TC, lamivudine; MDM, monocyte-derived macrophage; RLU, relative light unit; TAM, thymidine analog mutation. Republished with permission of American Society for Microbiology—Journals, from Perez-Bercoff D, Wurtzer S, Compain S, Benech H, Clavel F. Human immunodeficiency virus type 1: resistance to nucleoside analogues and replicative capacity in primary human macrophages. *J Virol*. 2007;81(9):4540–4550; permission conveyed through Copyright Clearance Center, Inc.

residual in vitro activity in monocyte-derived macrophages (MDMs) in the presence of the M184V substitution that was potentially driven by specific conditions of these HIV target cells (Fig. 3).[52] In MDMs, resistance to 3TC via the M184V pathway occurs more slowly than in other cells. Thus, selection for M184V by 3TC is weaker in MDMs compared with other cell types, leading to the possibility of residual activity of 3TC in cells displaying less frequent mitosis compared with cultured tumor cells.[52] The M184I reverse transcriptase isolated from the serum of a single patient infected with HIV-1 exhibited decreased DNA polymerization in the presence of low concentrations of deoxynucleotide triphosphate (dNTP) in vitro compared with wild-type HIV reverse transcriptase, which was explained by the low binding affinity of M184I to dNTPs.[53] Macrophages have lower dNTP concentrations compared with other cell types (4 and 20 times less than in resting and activated T cells, respectively).[54] Interestingly, the HIV M184I virus was unable to transduce macrophages but was able to transduce cell types with higher dNTP concentrations. These biochemical and preclinical data suggest that preservation of M184V by continued selection pressure of 3TC may be clinically beneficial by preventing the overgrowth of wild-type virus,[46] especially in cells with low dNTP concentrations such as macrophages.

It has been postulated that the lower replication activity of M184V variants could result in a reduction in HIV transmissibility. It has been noted that although M184V is commonly acquired during treatment with NRTIs, it is rarely detected in treatment-naive patients, suggesting that M184V variants are rarely transmitted.[55] According to 1 study that used population genetic sequencing, 67.5% of patients with virologic failure on first-line treatment harbored M184V variants compared with only 7% of those who were treatment naive.[55] However, when a polymerase chain reaction–based method was used to detect minority subpopulations of HIV variants in the sample, 23.4% of treatment-naive patients harbored M184V variants, whereas the method of detection did not substantially affect the reported frequency of other variants. The differences in the results of the 2 methods reflect the rate at which M184V is overcome by other genotypes within the viral population. In 3 individual samples from treatment-naive patients with M184V originally present, the percentage of M184V decreased over time and was no longer detectable between 40 and 61 weeks after the infection. The combination of these results suggests that the replication

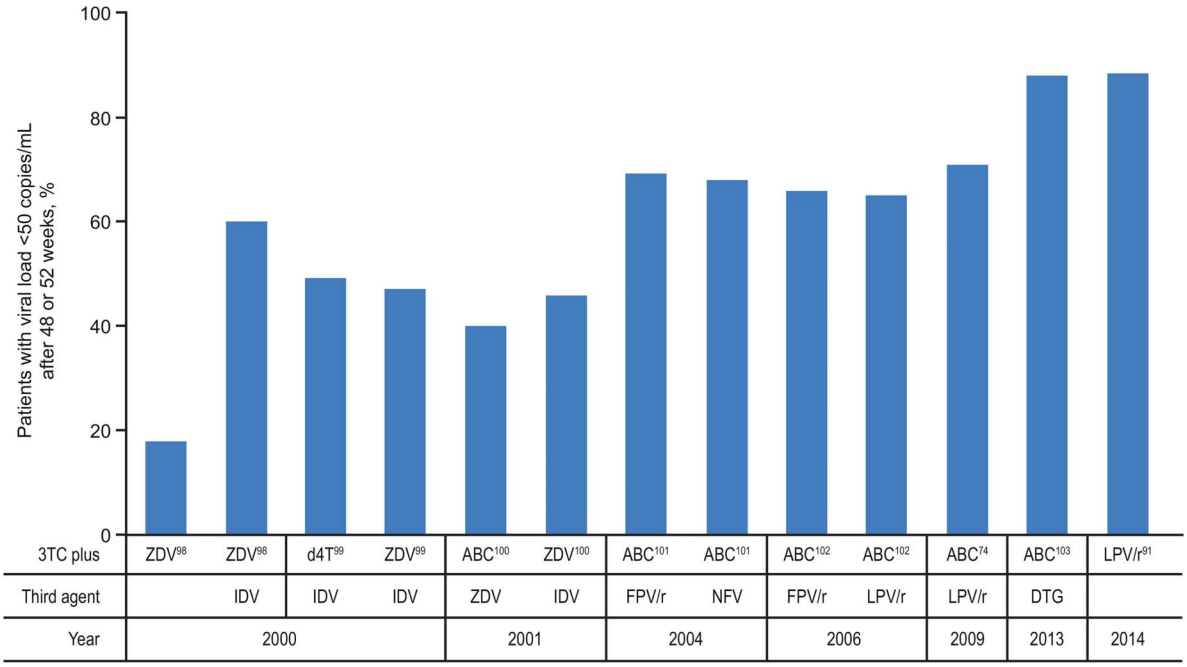

FIGURE 4. Proportion of patients in individual studies who achieved viral load <50 copies per milliliter after 48 or 52 weeks on therapy. Treatment was lamivudine in combination with antiretroviral drugs listed below the x-axis. ABC, abacavir; d4T, stavudine; DTG, dolutegravir; FPV/r, fosamprenavir/ritonavir; IDV, indinavir; LPV/r, lopinavir/ritonavir; NFV, nelfinavir; ZDV, zidovudine.

defect of M184V does not substantially affect HIV transmission, but the reduced fitness causes the elimination of M184V in the absence of selection by 3TC or FTC.

Studies of mono- and dual-NRTI regimens suggest residual clinical benefit of 3TC despite the presence of M184V mutants.[39,56,57] For example, in NUCA 3001, plasma HIV-1 RNA levels remained >0.5 $\log_{10}$ below baseline in participants who received 3TC monotherapy despite emergence of the M184V substitution and high-level phenotypic resistance.[56] Moreover, in a small pilot study, withdrawal of 3TC in viremic patients with multidrug-resistant HIV and the M184V substitution led to a 0.5 $\log_{10}$ increase in viral load at 6 weeks, suggesting a persistent antiviral effect of 3TC despite the presence of the M184V substitution.[58] These and other clinical observations suggest a benefit of continuing 3TC even in the presence of M184V mutants; however, determining the clinical relevance of this benefit may require further study.[57,59]

SAFETY

Since the approval of NRTIs, postmarketing reports of adverse reactions have been collected,[1] and numerous toxicities were associated with prolonged use of first-generation NRTIs. A widely accepted hypothesis is that many of these toxicities result from inhibition of mitochondrial DNA (mtDNA) polymerase γ.

The potential for mitochondrial toxicity of first-generation NRTIs was generally high, with ddI, d4T, and ddC most strongly associated with toxicities and ZDV showing a weaker, but still relevant, association.[60,61] With the emergence of combination ART, 3TC was combined with other NRTIs in early trials, which made it difficult to identify the drug that caused any given AE. However, preliminary safety data from the NUC trials revealed that the addition of 3TC did not result in toxicities beyond those associated with other components of the combination regimen.[39,40] Similar safety results have been demonstrated in more recent clinical studies.[62] These data suggested that the safety profile of 3TC may be different from that of other NRTIs. Active 3TC metabolites are not concentrated in the mitochondria, which may explain why a subsequent in vitro analysis revealed that 3TC had no effect on cell growth, lactate production, intracellular lipids, mtDNA, and mtDNA-encoded respiratory chain subunit II of cytochrome c oxidase after a long-term treatment.[61] Another explanation for this lack of mitochondrial toxicity is the weaker inhibition constant of the minus enantiomer of 3TC for mitochondrial DNA polymerase γ.[63] Similarly, the minus form's weaker inhibition of DNA polymerase α translates to less toxicity caused by off-target inhibition of nuclear DNA synthesis. In a case control retrospective analysis, there was an 87% risk reduction (odds ratio, 0.13; 95% CI: 0.06 to 0.26; $P < 0.001$) in experiencing hyperlactataemea/lactic acidosis while taking 3TC compared with taking other NRTIs (including TDF, d4T, ABC, ddI, and ZDV), suggesting that the risk of developing lactic acidosis is minimal for patients treated with 3TC.[64] Thus, from both in vitro and clinical studies, there is little evidence of a negative effect of 3TC on mitochondrial biology.

Although morphologic changes (lipoatrophy and lipohypertrophy) and insulin resistance are considered to be clinical features of HIV, some ART regimens are believed to

contribute to these metabolic findings. In contrast with results using thymidine-containing NRTIs, clinical studies of 3TC plus ABC combination therapy demonstrated small increases in lipohypertrophy and no increased risk of lipoatrophy.[65,66] Similarly, the risk of developing insulin resistance is reduced with 3TC plus ABC combination therapy compared with a thymidine-containing combination.[66] These data suggested that 3TC may have a favorable toxicity profile compared with other NRTIs.

PEDIATRICS

Children infected with HIV at birth are an example of a population that needs nuanced care. Lamivudine is approved for children aged 3 months to 16 years when the dosage is scaled to body weight,[1] and it is included as an NRTI backbone agent in every WHO-recommended first-line regimen for treatment of children, adolescents, and pregnant or breastfeeding women at any $CD4^+$ cell count, regardless of the clinical stage.[3] The inclusion of 3TC in recommended regimens is attributable to its well-established record of efficacy and safety in children living with HIV.[67–69]

Lamivudine has been evaluated as a component of combination therapy for its capacity to decrease the risk of mother-to-child HIV transmission and infant mortality.[70] Antiretroviral therapy prevented HIV transmission to an estimated 670,000 children in low- and middle-income countries from 2009 to 2012.[71] The Kesho Bora study demonstrated that infants of mothers treated with 3TC-containing ART during pregnancy and breastfeeding had a decreased risk of HIV transmission (43% reduction, $P = 0.029$), mortality, and HIV transmission or death (36% reduction, $P = 0.017$) at 12 months compared with mothers treated with ART that did not include 3TC.[72] An even greater decrease in HIV transmission risk (48% reduction, $P = 0.02$) was seen in infants whose mothers were treated with 3TC and intended to breastfeed compared with non–3TC-treated mothers who intended to breastfeed.

COMPARISON OF 3TC AND FTC

Like 3TC, FTC is a dideoxynucleoside analog of cytosine used to treat HIV infection, and the 2 drugs share similar characteristics and safety profiles as shown in head-to-head studies.[73,74] Emtricitabine is more potent than 3TC in vitro and has a longer intracellular half-life (Table 1).[75] By contrast, 3TC achieves higher intracellular levels of the active triphosphate analog than does FTC.[76] The terminal half-life of FTC 200 mg once-daily is 7.4 hours,[77] which is similar to that of 3TC 300 mg administered once-daily (7.9 hours).[78] Similar to 3TC, single-agent FTC has been shown to select for M184V in vitro approximately 2 weeks after initiation.[79] In a pooled analysis of trials in which the treatment-naive patients received EFV plus 2 NRTIs that included either 3TC or FTC, the frequency of patients with observed M184V/I variants was 1.0% in patients treated with FTC and 3.2% in patients treated with 3TC ($P = 0.009$).[80] However, conclusions from this analysis were limited by the nature of the cross-trial comparison, including different NRTI partners.

Emtricitabine and 3TC are usually considered clinically equivalent. A phase II dose-escalation study demonstrated that a daily 200-mg dose of FTC had superior virologic efficacy compared with 150 mg of 3TC administered twice-daily.[81] However, in the context of combination ART, 3TC- and FTC-containing regimens have comparable efficacy.[82] A recent review evaluated clinical trials that studied 3TC and FTC as part of combination therapy for treatment-naive or treatment-experienced adults infected with HIV from 2002 to 2013.[82] Twelve clinical trials provided 15 different randomized comparisons, providing data on 2251 patients receiving 3TC and 2662 patients receiving FTC. In the 12 trials, treatment outcomes were not significantly different. In 3 trials that compared 3TC and FTC directly, the relative risk of treatment success was nonsignificant (RR, 1.03; 95% CI: 0.96 to 1.10; $P = 0.3$). For all 12 trials, the pooled relative risk of treatment success was not significantly different (RR, 1.00; 95% CI: 0.97 to 1.02). Similarly, there was no difference in the pooled relative risk of treatment failure (RR, 1.08; 95% CI: 0.94 to 1.22). A retrospective, exploratory analysis of pooled data from 3 phase III studies that evaluated the safety and efficacy of dolutegravir-based regimens provided further support for the clinical similarity of the 2 agents.[83] A Kaplan–Meier estimate of time to efficacy-related discontinuation or failure (ERDF), stratified by NRTI backbone and HIV-1 RNA level at baseline ($\leq$100,000 or $>$100,000 copies per mL), supported a conclusion that risk of ERDF was not significantly different for participants treated with ABC/3TC versus TDF/FTC (regardless of third agent) overall (HR 0.90, 95% CI: 0.58 to 1.38, $P = 0.63$) or in patients with baseline HIV-1 RNA $>$100,000 copies per milliliter (HR 0.95, 95% CI: 0.55 to 1.65, $P = 0.86$).

By contrast, a study of 4740 treatment-naive patients infected with HIV-1 without baseline resistance from the ATHENA cohort in the Netherlands who were treated with 3TC or FTC plus an NNRTI (EFV or nevirapine) and tenofovir found that virologic failure occurred more often in patients treated with 3TC than in patients treated with FTC [odds ratio (OR; 95% CI) with EFV/tenofovir, 1.78 (1.11 to 2.84), $P = 0.016$; with nevirapine/tenofovir, 2.09 (1.25–3.52), $P = 0.005$].[84] However, in another study from the ATHENA cohort of 1582 treatment-naive patients with HIV who initiated treatment with 3TC or FTC plus a boosted PI and TDF, no significant differences in rates of virologic failure were observed in patients taking 3TC compared with patients taking FTC.[85] Collectively, these findings support the current US treatment guidelines to consider FTC and 3TC as interchangeable for initial therapy,[6] with the possibility that further head-to-head prospective studies may be warranted to evaluate whether the interchangeable nature of FTC and 3TC should be context specific based on ART regimen.

3TC IN NOVEL 2-DRUG REGIMENS

A new clinical strategy has been proposed to increase patient adherence and reduce toxicities associated with chronic ART to improve patient outcomes by switching from 3- or 4-drug combinations to 2-drug regimens. This approach is recommended by treatment guidelines in specific

situations.[5,6] Clinical trials with treatment-experienced patients have analyzed the safety and efficacy of switching to a 2-drug regimen, including several combinations that include 3TC (see Table, Supplemental Digital Content 2, http://links.lww.com/QAI/B131, which details clinical studies involving 2-drug regimens containing 3TC and an NRTI, non-NRTI, integrase inhibitor, or boosted protease inhibitor). In the SALT trial, ATV/r plus 3TC was noninferior to ATV/r plus 2 NRTIs and had significantly fewer treatment discontinuations.[86] The similarly designed ATLAS study showed that ATV/r plus 3TC exhibited superior efficacy to ATV/r plus 2 NRTIs at 48 weeks ($P = 0.027$).[87] In the OLE trial, 3TC paired with LPV/r showed similar efficacy and tolerability compared with LPV/r plus 2 NRTIs in virologically suppressed participants.[88] Highly ART-experienced adults (n = 27) switched to DTG plus 3TC in the DOLULAM study maintained viral suppression (<50 copies per milliliter) for 96 weeks with no serious or clinical adverse events.[89] Interestingly, ultradeep sequencing analysis determined that the M184I/V substitution was observed in 63% (n = 17) of patients enrolled in DOLULAM, yet no virologic failure was observed over the course of the study.[90]

The 2-drug treatment strategy with 3TC has demonstrated success with treatment-naive patients as well. Lamivudine plus LPV/r was noninferior to the 3-drug regimen of LPV/r plus 2 NRTIs at 48 weeks in the GARDEL trial.[91] Because DRV/r is better tolerated than LPV/r,[92] the ANDES trial compared DRV/r plus 3TC with DRV/r plus 3TC/TDF, and the interim analysis indicated that the study achieved a secondary end point, noninferiority of DRV/r plus 3TC in patients achieving HIV-1 RNA <400 copies per milliliter at 24 weeks.[93] The primary end point, viral load <50 copies per milliliter at 48 weeks, will be reported after the next phase of the study is complete. There were 5 serious adverse events (DRV/r + 3TC, 3; DRV/r + 3TC/TDF, 2), none of which were considered drug related, and 5 discontinuations from the study (DRV/r + 3TC, 4; DRV/r + 3TC/TDF, 1). The PADDLE pilot study evaluated DTG and 3TC as a 2-drug regimen in treatment-naive patients with HIV-1 infection and showed that 90% of patients were virologically suppressed at 48 weeks,[94] and 100% of patients (n = 18) who were included in the extension phase maintained virologic suppression at 96 weeks.[95] To extend the findings of PADDLE, 3TC was paired with DTG in patients with HIV-1 RNA <500,000 copies per milliliter in the phase II 52-week ACTG A5353 pilot study.[96] After 24 weeks, DTG plus 3TC demonstrated potent virologic efficacy, with 90% of participants (n = 108) meeting the primary end point for virologic success (HIV-1 RNA <50 copies per milliliter); 3 participants experienced protocol-defined virologic failure, including 1 participant with mutations resulting in R263K and M184V detected at week 14. Pharmacokinetic data showed DTG concentrations below the limit of detection at ≥1 visit. Two participants experienced grade 3 adverse events considered to be treatment related but did not discontinue the study. Last, in the open-label ASPIRE trial with 89 patients with virologic suppression on 3-drug ART, 93.2% of patients who switched to DTG plus 3TC maintained virologic suppression compared with 91.1% of patients who remained on their initial regimen ($P = 0.71$) at the 24-week primary analysis, demonstrating noninferiority of DTG plus 3TC.[97] These proof-of-concept studies provide the rationale for 2 phase III trials, GEMINI-1 (clinicaltrials.gov identifier, NCT02831673) and GEMINI-2 (clinicaltrials.gov identifier, NCT02831764), which compare DTG/3TC with DTG plus TDF/FTC in treatment-naive patients. The outcomes of these studies will provide strong clinical trial evidence for evaluating this innovative strategy.

CONCLUSIONS

Despite its 25-year history as a component of antiretroviral treatment regimens, 3TC continues to be studied, and new aspects of its fascinating virology and mechanism of action continue to be uncovered. Over the past 2 decades, treatment options associated with 3TC have improved as the combinations have become more potent, safer, and more convenient.[74,91,98–103] (Fig. 4) As new drugs and drug classes have been approved, regimens that contain 3TC continue to be widely used in first-line therapy. In developing countries, 3TC is critical to HIV care because of its excellent efficacy and safety profile and the availability of low-cost generic versions. Indeed, 3TC is recommended in nearly all first-line and a majority of second-line combination regimens for both adults and children. In addition, 3TC has a well-established PK profile with few drug–drug interactions, which is important not only for the convenience of combining 3TC with other drugs in single-tablet regimens but also because of the prevalence of multiple comedications in certain populations such as the elderly or hepatitis-coinfected patients. Among all drugs first approved more than 20 years ago for HIV treatment, only 3TC continues to be recommended in the most recent worldwide guidelines. With few drug–drug interactions and low cost, 3TC continues to emerge and play a role in new treatment strategies worldwide in combination with a new generation of antiretroviral drugs and remains an attractive component for inclusion in future drug combinations.

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