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Topics in Antiviral Medicine™

A publication of the IAS–USA

Review

Special Contribution

Invited Review

Topics in Antiviral Medicine™

Topics in Antiviral Medicine™

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On completion of this activity, which contains 3 articles, the learner will be better able to:

- Describe the causes of and trends in liver disease among people with HIV, as well as current approaches to prevention and treatment.
- List key mutations associated with resistance to antiretroviral drugs by drug class.
- Describe manifestations of potential monkeypox virus infection in the current outbreak of the disease.

This enduring material is designed for physicians who are actively involved in the medical care of people with HIV and other viral infections.

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Review

HIV and Liver Disease: A Comprehensive Update

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Despite substantial advances in the field, liver disease morbidity and mortality remain serious issues among people with HIV. The causes of liver disease are often multifactorial and include hepatitis viruses, hepatic steatosis and oxidative stress, bacterial translocation with activation of hepatic macrophages and stellate cells, and direct toxicities from alcohol and drugs of abuse. Biopsychosocial factors including a high prevalence of psychiatric disorders, food insecurity, insufficient access to care and medications, and social stigma all play roles in the persistence of liver injury and hepatic fibrosis development among people with HIV. Rising rates of hepatocellular carcinoma have been observed, suggesting that the epidemiology of liver disease is evolving.

Keywords: HIV, HCV, HBV, hepatitis, NASH, NAFLD, fatty liver, pathogenesis, opioid

Introduction

Liver disease was initially recognized as a major contributor to morbidity and mortality among people with HIV in the early 1990s and became fully manifest as a major health issue in the mid-1990s following the introduction of effective combination antiretroviral therapy (ART). The subsequent 3 decades witnessed tremendous progress in HIV care, as well as the stubborn persistence of liver disease threatening survival and quality of life. Microelimination of the hepatitis C virus (HCV) infection in people with HIV seems achievable, yet new infections and reinfections threaten progress. Hepatitis B virus (HBV) infection can be suppressed by nucleotide and nucleoside therapy, but a cure remains elusive. Limited HBV vaccine responses in people with HIV who use older vaccine products remain problematic. Newly developed HBV vaccines may help people with HIV but have not been widely studied yet in the populations that would most benefit. Nonalcoholic fatty liver disease (NAFLD) and nonalcoholic steatohepatitis (NASH) have increased in prominence as HCV treatment reduces the threat of HCV-related liver disease. ART drugs have improved in their hepatic safety, but some trends like the use of 2-drug regimens with limited HBV activity may represent a step backward for people coinfected with HBV, and weight gain associated with many integrase strand transfer inhibitors (InSTIs) is problematic as well. Barriers to treatment and prevention remain, in part because of the presence of major psychiatric comorbidities that are more prevalent in people with HIV. Emergence of the COVID-19 pandemic added complexity to the prevention and management of liver disease among people with HIV and continues to affect care and disease outcomes.

To address these issues and encourage collaboration among researchers to investigate emerging issues, the National Institute of Allergy and Infectious Diseases, in association with industry partners, has provided support for a biennial meeting to discuss the research agenda. Held in September 2021, the 8th Biennial HIV and Liver Disease Conference included key representatives from infectious diseases, hepatology, psychiatry, nutrition, and pharmacology, as well as policymakers, regulators, and basic and translational scientists focused on liver-related issues in this unique, at-risk population.

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Epidemiologic Considerations

Liver Disease and Hepatocellular Carcinoma

Few studies have addressed the epidemiology of and trends in liver disease over time in large cohorts that are diverse and have sufficient geographic representation to fully represent the at-risk populations. The NA-ACCORD (North American AIDS Cohort Collaboration on Research and Design), a US and Canadian cohort study, examined morbidity and mortality of liver disease across 3 distinct time periods that correspond broadly to eras in available choices of ART. Its results clearly documented the lack of longitudinal changes in disease mortality and progression to end-stage liver disease (ESLD) associated with coinfection with HCV, HBV, or both in people with HIV.[1] Despite this finding, more recent analyses in the NA-ACCORD identified a clear increase in rates of hepatocellular carcinoma (HCC), culminating in an incidence of 0.75 cases per 1000 person-years compared with an HCC rate in the general US population of 0.23 per 1000 person-years.[2] Interestingly, people affected by triple infection with HBV/HCV/HIV are at highest risk.

High HCC rates were also observed in the VACS (Veterans Aging Cohort Study) among nearly 35,000 veterans. Data adjusted for age, sex, race, body mass index, alcohol use, diabetes, and HBV and HCV serostatus revealed HIV virus detection and viral load as key factors associated with this outcome.[3] Presence of fatty liver disease was also identified as an important factor. In a combined analysis of 4 European cohorts of individuals coinfected with HBV/HIV on tenofovir disoproxil fumarate (TDF), cumulative time on TDF treatment was associated with a stable-to-decreased risk of HCC development, but the time off TDF therapy was highly associated with an increase in the incidence rate ratio of HCC.[4] In the same study, HCC surveillance strategies were evaluated to determine the optimal screening paradigm. In people with cirrhosis, as in HBV monoinfection, age was not found to be a factor, meaning all persons with HBV/HIV coinfection should be screened. In people without cirrhosis, an age threshold of 45 years was associated with a predetermined screening threshold of 2 events/1000 person-years.

The association of HCC development with HBV viral titer was previously well established in the Taiwanese REVEAL (Risk Evaluation of Viral Load Elevation and Associated Liver Disease/Cancer-Hepatitis B Virus) study.[5] More recently, this relationship was also noted in people with HBV/HIV coinfection; higher HCC risk was noted for persons with HBV DNA levels greater than 200 IU/mL than those with lower quantities. Longer duration of complete HBV suppression was likewise associated with decreased HCC risk.[6] Additional risk for HCC attributable to hepatitis D virus (HDV) infection was described in results from the Swiss HIV Cohort Study. The hazard ratio for HCC-free survival was 9.3 (95% CI, 3–28.6), representing a significant decrease in cancer-free survival for persons with HDV/HBV/HIV infection.[7] Recently, bulevirtide was approved in the European Union for treatment of hepatitis D virus (HDV) infection and may soon be approved in the USA, providing one tool to combat this serious challenge.

Not all news related to liver disease trends in people with HIV is negative, however. Among the first groups to document the salutary effects of effective HCV treatment in this population, Mocroft and colleagues reported significant reductions in the incidence of ESLD in people with HIV in the EuroSIDA cohort whose HCV was cleared spontaneously, versus those whose HCV was not. The lowest rates of ESLD were observed in those with spontaneous clearance, and those with treatment failure had the highest observed rates of ESLD. Successfully treated persons were normalized at an incidence rate of 1.[8] Similarly, the national US database known as the Nationwide Inpatient Sample was used to examine trends in mortality over a 15-year period within a subset of people with liver disease, namely, those with cirrhosis, either with or without HIV. Higher inpatient mortality (10.9%) was observed among people with cirrhosis and HIV than among those without HIV (9.2%; $P<.001$). However, the rate declined in both groups over the 15-year period. Liver-decompensating events decreased but infections increased in people with HIV. Thus, HIV remains an independent predictor of mortality in people with cirrhosis.[9]

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Trends in people with NAFLD and NASH remain unclear and are difficult to discern because of substantial differences in reporting methods used to define the presence or absence of disease. Varying definitions within and between diagnostic categories sharply limit the ability to draw conclusions about disease progression and make comparisons between people with HIV and cohorts without HIV. For example, the histologic definition of steatosis is the presence of 5% or more hepatocytes containing fat droplets, and the biomarker MRI-derived proton density fat fraction appears to have excellent reliability and reproducibility for defining steatosis at this level.[10] However, most studies utilize other, substantially less sensitive methodologies including controlled attenuation parameter or even echogenicity on ultrasound. The definition used for NAFLD is also variable, requiring abnormal levels of alanine aminotransferase (and with studies using varying cutoff levels) or the presence of fibrosis as determined by liver-stiffness measurement, which is itself a surrogate for fibrosis. NASH is a purely histologic diagnosis, but few studies perform enough liver biopsies to allow meaningful cross-sectional comparisons between people with HIV and control participants. This area remains key for future investigation and is especially important because of the putative relationship between some ART drugs and fat accumulation in the liver and elsewhere. This point is discussed later in more detail.

HIV Epidemiology

In 2018, several key issues in HIV epidemiology were identified. At that time, not only had progress toward ending the epidemic of new HIV infections in the United States stalled, but the opioid epidemic was driving a resurgence of new infections. Indeed, the US Centers for Disease Control and Prevention (CDC) estimated that the next decade would yield a net gain of nearly 400,000 HIV infections, based upon an incidence of greater than 38,000 new cases/year.[11] This modeling led the CDC to begin a new initiative targeting a 75% reduction in new HIV infections within 5 years and a 90% reduction in 10 years. The cornerstones of this effort included increased use of HIV preexposure prophylaxis (PrEP) and syringe exchange service programs (SSPs). Furthermore, early diagnosis and treatment with the goal of sustained viral suppression would reduce the pool of HIV index cases. Targeting was achieved by identifying 57 jurisdictions with the highest rates of HIV transmission, many in 7 rural states.

Unfortunately, data available in 2021 show little progress made to date. There were 34,800 new HIV infections in 2019, the majority (82%) among males. Black/African American and Hispanic/Latino persons account for the majority (69%) of new infections. Male-to-male sexual contact represents the highest proportion of transmissions (66%). Because 80% of infections are transmitted by people unaware they have HIV, this group remains a key priority for intervention.[12] Self-testing strategies may have an important role in increasing early diagnosis.[13] Large gaps in PrEP use between whites and other racial or ethnic groups remain, representing an area of opportunity. However, SSPs have lost ground in areas with high opioid-use risk, which poses a threat to achievement of lower HIV incidence targets. Results of several economic modeling studies suggest that SSPs are cost-effective, but political considerations may continue to limit implementation.[14]

Immunopathogenesis

The higher risk of cirrhosis in people living with HIV than in those without HIV underscores the importance of understanding the mechanisms of liver fibrosis. Most of the focus and disease burden has been on individuals with underlying HCV or HBV infection. However, some data suggest that even without a primary liver disease, HIV itself may cause liver steatosis and fibrosis. For example, results of a study of 432 people with HIV revealed that 10% of those without HBV infection, HCV infection, or self-reported excessive alcohol use had elevated values of liver stiffness (>7.1 kPa), a finding associated with HIV viral load and metabolic dysfunctions like diabetes.[15] Thus, even though the apparent net effect of HIV on liver fibrosis is most evident in the presence of a second contributor such as HCV infection, HBV infection, excessive alcohol use, or metabolic liver disease, HIV infection itself biases the liver toward fibrosis and synergistically promotes these other

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Table. Proposed Overlapping Mechanisms for HIV Potentiation of Liver Disease

Enhanced oxidative stress and TGF-beta induction
HIF-1, Hippo, YAP, LPA signaling effects
Adaptive immune dysfunction (eg, CD4+ T-cell depletion)
Alterations in Kupffer cell physiology
Lipodystrophy and adipocyte effects
Enhanced microbial translocation

Abbreviations: HIF-1, hypoxia-inducible factor-1; LPA, lysophosphatidic acid; TGF-beta, transforming growth factor-beta; YAP, yes-associated protein.

processes. Notably, some mechanisms are reversed by ART, and others continue to contribute (or are even caused by) the ART medications (discussed later).

Various mechanisms have been proposed through which HIV may potentiate or cause liver fibrosis; many overlap, and all converge on the central role of stellate cells (Table). Results of pivotal in vitro studies have shown that HIV cooperatively enhances the fibrogenesis caused by HCV accentuating reactive oxygen species and transforming growth factor (TGF)-beta induction, thereby inducing the secretion of type 1 collagen and tissue inhibitor of metalloprotease (TIMP)-1 in hepatic stellate cells as well as hepatocyte apoptosis.[16,17] Likewise, with HBV coinfection, recent evidence suggests that HIV and HBV cooperatively promote fibrosis by upregulation of hypoxia-inducible factor 1-alpha, which in turn might increase TGF-beta production.[18] Emerging evidence also suggests a role for the Hippo–yes-associated protein (YAP) pathway in hepatic fibrogenesis. When Hippo is turned off (directly or indirectly by infection), YAP is unphosphorylated and free to translocate to the stellate cell nuclei to induce genes promoting fibrosis in a manner attenuated by recognized inhibitors of fibrogenesis such as lysophosphatidic acid (LPA) and epidermal growth factor receptor (EGFR) inhibitors. Interestingly, in a murine model the YAP-mediated fibrogenesis is also attenuated by ART.

Other data point to a role for liver macrophages (Kupffer cells) in the pathogenesis of HIV-related liver disease. HIV can infect Kupffer cells and alter their cellular physiology, even though the macrophage is not thought to contribute to the latent reservoir.[19] The M2 Kupffer cell phenotype is especially relevant, in that it can activate stellate cells, a process that is correlated with the net production of a soluble protein, CD163.[20,21] Soluble CD163 was highly associated with evidence of liver injury in people with HIV and with development of hepatic fibrosis in a human cohort study.[22]

HIV infection also has myriad effects on adipocyte biology that might coordinately impact liver disease. The most obvious connection is with the accumulation of additional liver fat (steatosis), which in some instances also is associated with disease (inflammation or fibrosis). One example is the HIV accessory protein Vpr, which can inhibit peroxisome proliferator-activated receptor (PPAR)-gamma, increasing lipolysis and fat accumulation in liver.[23] Although inhibition of HIV replication would be expected to reduce that mechanism, some ART medications themselves are associated with fatty liver. Older ART medications such as stavudine (d4T) were directly toxic to cells, but even newer drugs like the InSTIs cause weight gain and possibly increased hepatic steatosis.

Chemokines and Their Receptors

Chemokines are molecules that regulate inflammation and, not surprisingly, are dysregulated in people with HIV. For example, the peripheral circulation of people with HIV/HCV coinfection contains fewer CD4+ T cells than that of individuals monoinfected with HCV, and these cells are disproportionately in an activated and "exhausted" state (PD1+, CD38+, HLADR+). Compared with T cells from people with HCV monoinfection, cells isolated from individuals with HIV/HCV coinfection are also more likely to express CXCR3, a liver-homing molecule, and to secrete chemokines and cytokines that stimulate stellate cells to produce extracellular matrix proteins and accelerate liver fibrosis (Figure 1).[24] Interestingly, in contrast to the findings for circulating blood, opposite trends occur in liver tissue (ie, more CD4+ T cells from individuals with monoinfection express CXCR3). This paradigm suggests that blocking

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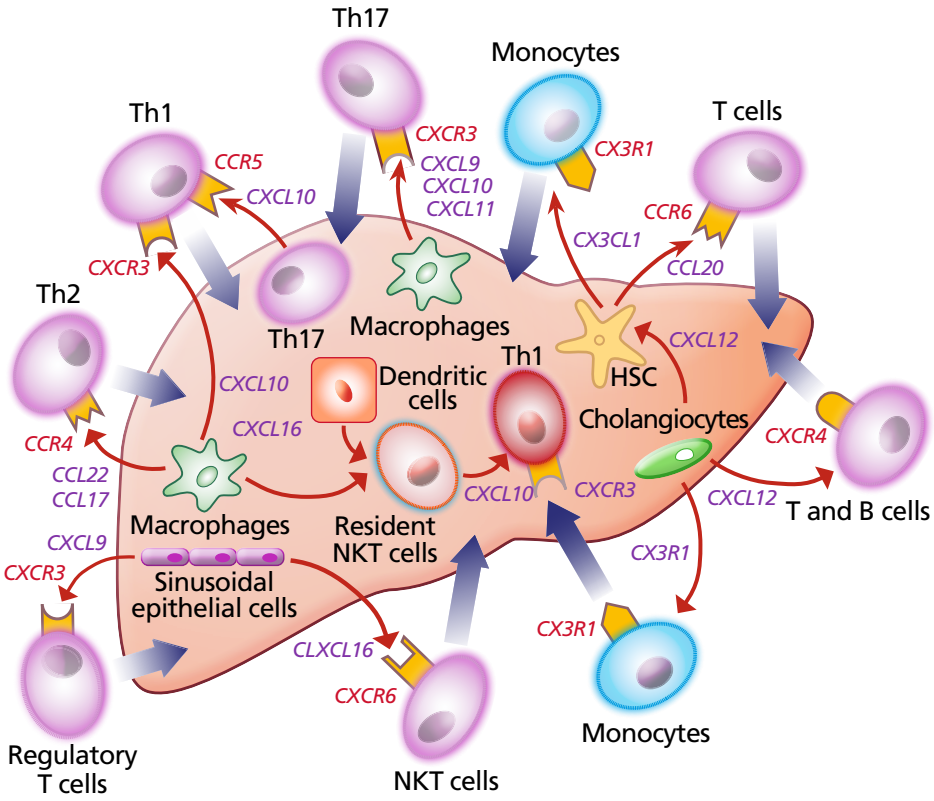

Figure 1. Schematic diagram depicting cytokines and chemokines that modulate hepatic injury and fibrosis.[53] Red arrows indicate chemokine secretion; thick arrows denote pro-inflammatory infiltration.

Abbreviations: CCL, chemokine ligand; CCR, chemokine receptor; CXCL, CXC chemokine ligand; CXCR, CXC chemokine receptor; CX3CL, CX3C chemokine ligand; CX3R, CX3 chemokine receptor; HSC, hepatic stellate cell; NKT, natural killer T cell; Th, helper T cell.

chemokine receptor 2 (CCR2) or chemokine receptor 5 (CCR5) might attenuate the increase in liver fibrosis associated with HIV.

Evidence supporting the pharmacologic blockade of these chemokine pathways comes from murine models of liver inflammation and NASH.[25,26] Some, but not all, human epidemiologic data also support the paradigm. For example, people with hemophilia and HIV/HCV coinfection who have 1 copy of the CCR5 allele known as the delta 32 mutation have lower serum liver fibrosis scores than those who are homozygous for the wild type.[27] Likewise, in a study in which people with HIV but not HCV were randomly assigned 2:1 to receive either cenicriviroc or efavirenz (plus TDF and emtricitabine), the cenicriviroc group had lower serum fibrosis scores than the efavirenz group.[27] This observational research set the stage for clinical trials to test the hypothesis. Thus far, a phase 2b study was reported in which people with NASH were randomly assigned to receive cenicriviroc either immediately or after a delay, and results of each group were compared with those of a placebo group.[28] In this relatively small study, no difference was detected in the number of participants who had both an improvement of at least 1 stage of fibrosis and no worsening of NASH.

Microbiome

The microbiome refers to all the microorganisms (bacteria, viruses, fungi, and archaea) that live within an ecosystem. In humans, the gut is the most studied ecosystem, and in health, homeostasis exists between the microbiome and intestinal mucosa. HIV disrupts this homeostasis, and evidence indicates that HIV affects the mucosal interface. Specifically, during acute HIV infection, there is marked depletion of the CD4+ T cells that line the intestinal lumen.[29] More pronounced effects are the depletion of a subset of Th17+ T cells and disruption of mucosal barriers. This breach in the gut mucosa is thought to promote translocation of bacteria from the gut microbiome, and correlates of that process (and macrophage activation) such as soluble CD14 are associated with HIV-related immune activation and even mortality.[30]

There is also evidence that the components of the microbiome are altered in people with HIV. Results of some studies adjusting for HIV risk exposure and other confounders show differences linked to HIV infection itself. Specifically enriched in people with HIV are some Proteobacteria and Enterobacteria species, with a corresponding reduction in Bacteroides species.[31] A dynamic metabolic

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mechanism has been proposed, with increased production of kynurenine from tryptophan associated with disease in a manner that contributes to depletion of Th17+ cells and disruptions in mucosal protection. Results of a recent study provided evidence for this paradigm in peripheral blood. In people with HIV receiving ART, fragments of enteric microbes and specifically *Serratia* species were correlated with proinflammatory cytokines and CD4+ T-cell increases in the first treatment year. Subsequently, lower *Serratia* species DNA abundance was associated with more favorable outcomes. DNA fragments traced to *Pseudomonas* species had the opposite associations.[32]

The link with liver pathogenesis is assumed to be related to the drainage of organisms that are translocated from the gut to the liver via the portal blood.[33,34] Collectively, this work raises the question of whether the microbiome could be manipulated therapeutically to improve HIV and liver outcomes. There is precedence in the use of rifaximin and lactulose to reduce encephalopathy in people with cirrhosis. Although probiotics and synbiotics have been studied in NAFLD, none has yet been convincingly translated into improving patient care.[35]

Prevention and Treatment of Liver Disease in People With HIV

Prevention and treatment of HIV-related liver disease logically targets the underlying causes, beginning with prevention and treatment of HIV itself. The clear medical benefits of PrEP shift the focus to implementation, overcoming barriers, and reaching those at risk. Notably, prevention of disease need not be siloed. Harm-reduction strategies to reduce the risks of illicit drug use also prevent HCV and HIV infections. These benefits were observed in Scott County, Indiana, where changes in behavior were reported after introduction of SSPs as part of the public health response to an HIV outbreak associated with shared injection paraphernalia among opioid users.[36] Unfortunately, county officials ended the program in mid-2021, following ongoing local and national debate over providing such services.[37]

As mentioned, ART improves some drivers of liver disease and is indicated for all people with HIV. Individuals for whom specific components of ART clearly need to differ are those with HBV/HIV coinfection, for whom compounds that are also active against HBV, such as tenofovir analogues, are recommended. This principle became especially relevant with the 2021 US Food and Drug Administration (FDA) approval of a long-acting HIV regimen composed of cabotegravir and rilpivirine, which lacks activity against HBV. People with HIV should not be switched from a TDF-based regimen without knowledge of each individual's HBV serostatus.[38] Similarly, a dolutegravir/lamivudine combination is not effective for long-term suppression of HBV, as resistance emerges quickly when HBV is present. An additional recent consideration concerns the association of marked steatosis with the weight gain that may accompany InSTI treatment. If confirmed, this association might justify switching people with NAFLD from an InSTI-based treatment to alternative regimens (eg, ritonavir boosted or darunavir based).[39]

Improved understanding of the immunopathogenesis of liver disease in people with HIV has yet to translate into HIV-specific treatments other than ART. Medical manipulation and restoration of the specific molecular pathways described earlier are theoretically possible; various potential approaches are shown in Figure 2. However, large, randomized clinical trials are still needed to establish their efficacy.

Beyond treatment of HIV, the current approach for preventing and treating liver disease in people with HIV is essentially the same as in those without HIV: reducing or eliminating alcohol ingestion, reducing weight in people with high body mass index values, and updating vaccinations as needed to protect against other forms of liver disease like that related to HBV or hepatitis A virus.

Since 2017, 2 new HBV vaccines have been approved by the FDA. The first was a 2-dose recombinant, adjuvanted HBV vaccine that is more immunogenetic than historic recombinant vaccines and is undergoing testing in people with HIV in a large multicenter, multinational clinical trial. Single-

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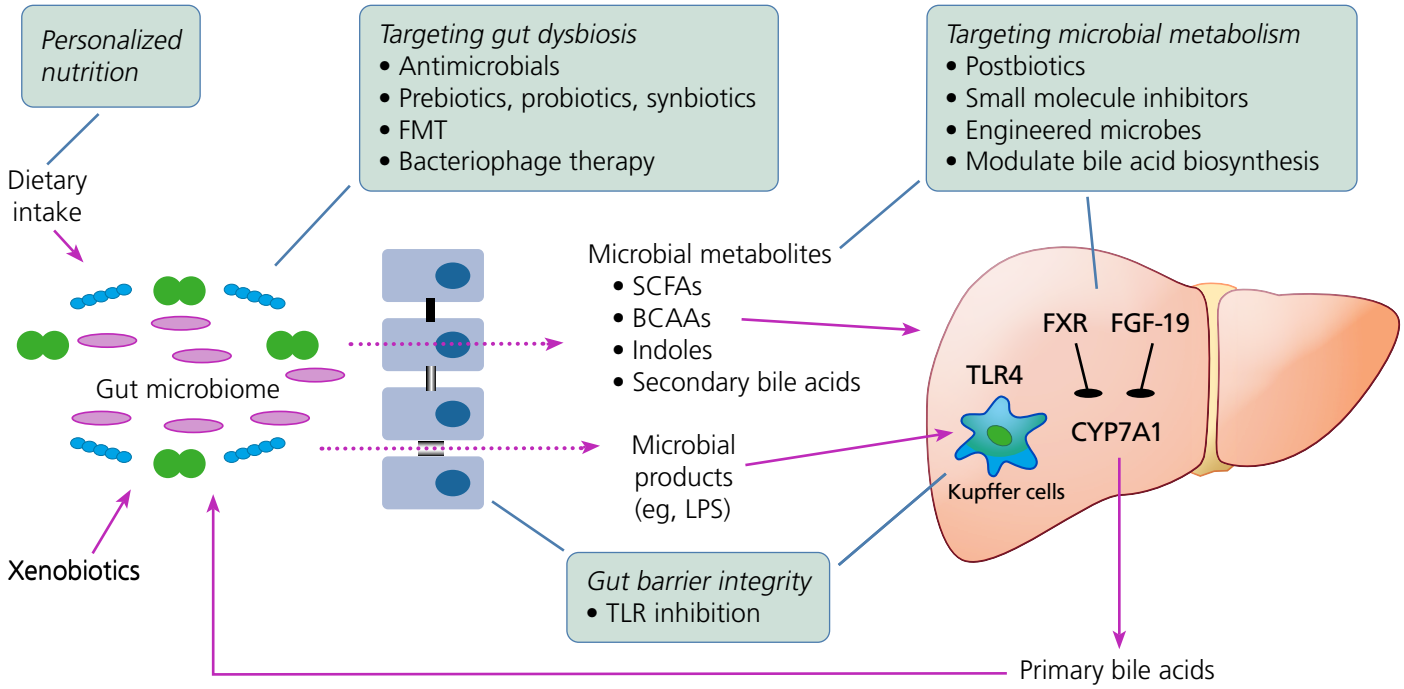

Figure 2. Schematic showing potential targets for decreasing liver injury (boxed text) that are not disease specific and may find application in people with HIV.[54] Solid arrows indicate impact on disease pathogenesis; dotted arrows denote gut microbiome metabolite leakage into portal circulation. BCAA, branched-chain amino acid; CYP, cytochrome P450; FGF, fibroblast growth factor; FMT, fecal microbiota transplantation; FXR, farnesoid X receptor; LPS, lipopolysaccharide; SCFA, short-chain fatty acid; TLR, toll-like receptor.

center retrospective results have been reported that suggest improved efficacy compared with older vaccine products.[40,41] The second vaccine, approved in November 2021, is a recombinant hepatitis B vaccine that is composed of recombinant forms of surface and 2 larger presurface envelope proteins and is more immunogenetic than historic recombinant vaccines when given in 3 doses.[42] Studies in people with HIV are needed to determine whether these new products (1) overcome the lower responses to historic recombinant vaccines in people with HIV, and (2) permit revaccination with desirable responses among people who did not respond to older vaccine preparations.

Although interferon alfa–based treatments for HCV originally differed for people coinfected with HIV, the current era of direct-acting antiviral (DAA) HCV therapies now provides similarly high efficacy and effectiveness for sustained virologic response in both groups.[43] The same treatments are recommended for HCV without regard for an individual's HIV serostatus. With available DAA treatments, even liver transplantation outcomes are similar between people coinfected with HIV and those with HCV monoinfection.[44]

Biopsychosocial Barriers and Responses

The prevention and treatment of liver disease in people with HIV are inhibited by the presence of numerous barriers to care, some of which are intrinsic to this population and others that are more broadly distributed in the general population and thus impact those with HIV as well. These barriers have their basis in socioeconomic and political processes that affect equal application of current scientific principles of prevention and care.[45] Cultural and religious differences among people at risk also lead to imbalances in the application of interventions. For example, hepatitis and HIV harm-reduction programs, including SSPs and health-education and screening programs, are limited or not available in many jurisdictions in the United States, despite evidence from Australia and Europe that they reduce

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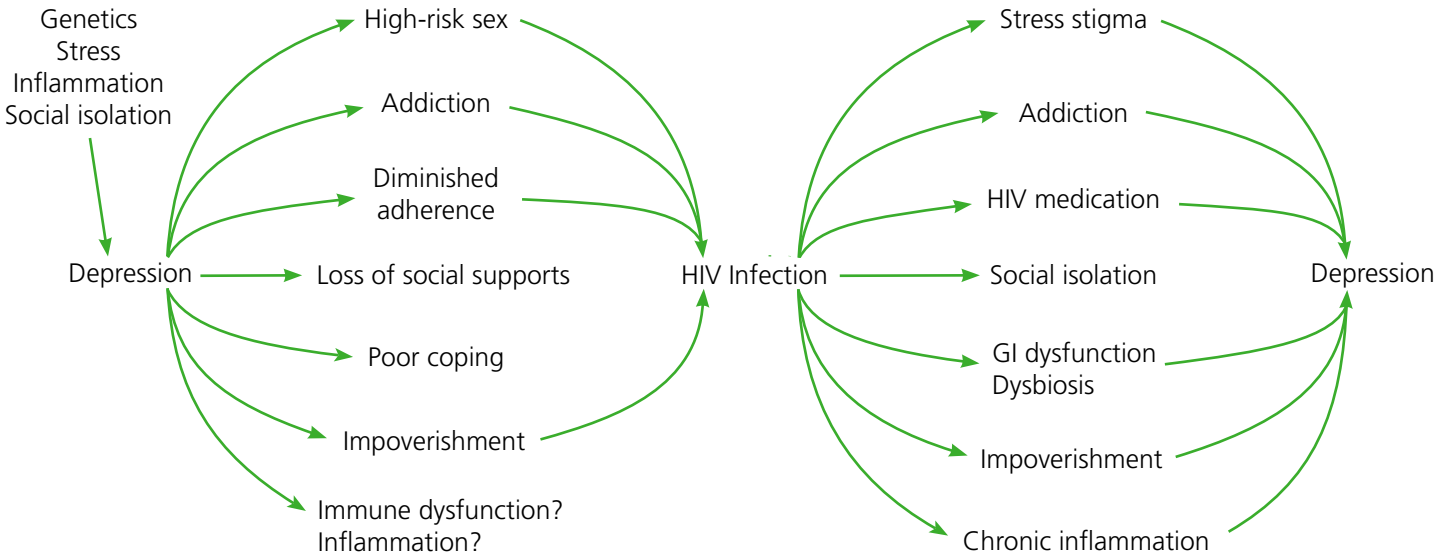

Figure 3. Diagram summarizing the complex interplay between HIV infection and depression and other manifestations of psychiatric and behavior disease processes. (Diagram courtesy of Glenn Treisman, MD, PhD.)

spread of chronic viral infections.[46,47] Funding opportunities vary between state and local levels. Even use of Ryan White HIV/AIDS Program funds for HCV treatment is allowed only in some states for people coinfected with HIV. Rules for the use of DAAs vary by state as well, with some requiring evaluation by a subspecialist before prescriptions for treatment can be filled. Although liver transplantation has been shown with few caveats to be safe and efficacious for people with HIV, only a subset of transplant centers will consider this population for transplant.[47,48]

Comorbid psychiatric and behavioral disorders are common among people with HIV. Axis 1 disorders may be present in nearly 50% of individuals with HIV.[49] These disorders include psychiatric diseases such as depression, bipolar disorder, and schizophrenia; behavior disorders like addictions; problems of endowment including personality and cognitive disorders; and problems of lived experiences including poor coping, limited choices, and poverty. A complex interaction exists among factors that lead to HIV, systemic immune activation, and depression and other psychiatric manifestations of disease (Figure 3).

Behavioral interventions appear to represent a key step in limiting the epidemic and managing individual patients. Implementing this step requires the development of integrated care systems that include medical, psychiatric, and substance-use expertise.[50,51] Economic models suggest that this approach is cost-effective, but it requires investment and infrastructure, and sadly, implementation of such systems has been absent from most care venues. Randomized trials are needed to confirm the efficacy of these approaches.[52]

Research Agenda: 2022 and Beyond

A key deliverable from the 8th Biennial HIV and Liver Disease Conference was the identification of a research agenda encompassing near- and long-term priorities for HIV and liver disease; a synthesis is presented herein. For HIV infection itself, targeted programs have been put in place as part of disease-elimination plans in the United States, but ongoing assessments of their efficacy and adjustments of implementation methods are still needed. HIV and viral hepatitis continue to be spread via parenteral exposure. Syringe exchange service programs work as an element of risk reduction, but backsliding is observed in many parts of the country. Economic modeling and education are needed to provide information on the cost-effectiveness of SSPs and other early-intervention programs that limit disease transmission.

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Highly effective treatment regimens for HIV are readily available but are not curative and may engender adverse effects with long-term implications for liver disease. Additional research is clearly needed to clarify the role of InSTIs in weight gain and the linkage to NAFLD, NASH, and other complications of metabolic syndrome. Up to 35% of people with HIV may have NAFLD, including NASH, yet data are limited regarding new treatment modalities for NAFLD and NASH in this group, and many researchers are still using diagnostic modalities like ultrasound that lack both sensitivity and specificity for disease identification. Lean disease appears to be more prevalent than NAFLD and NASH in people with HIV and may have a different mechanism that requires different treatment. Despite many efforts to utilize large electronic medical record databases to study these issues, more refined and integrated tools to do so are needed. The use of International Classification of Diseases (ICD)-10 codes is at best a blunt instrument when trying to determine disease incidence and prevalence.

Basic and translational science research continues to identify new pathways for liver injury and fibrosis (eg, hypoxia-inducible factor-1, yes-associated protein-1). These pathways represent potential new targets for therapeutic intervention. To this end, results of several clinical studies suggest a central role for chemokines as mediators of liver injury, stellate-cell activation, and remodeling. Thus far, however, the results are mixed for blockade of receptors like CCR5 and CCR2, and whether this lack of clarity stems from efficacy or study design is unclear.

Results of other studies point toward manipulation of the microbiome as a promising intervention strategy, but progress is hindered by the lack of agreement on the best methods for identifying microbial populations as well as clearly defined risks of such manipulation. Data suggest that a conceptually simple idea like addressing food insecurity can impact the microbiome and reduce downstream hepatic injury by affecting translocation and macrophage activation. However, any beneficial strategies for changing diet would require an understanding of the economic and social drivers of dietary choice and lack of choice.

Hepatic viral infections continue as ongoing sources of injury and progressive liver disease, even in 2022. HBV cure still seems remote despite the availability of valid targets and medications. Long-standing definitions of treatment response need to be updated, and current measures used to define response require revalidation with newer endpoints or replacement with newer biomarkers. The ability to separate host-integrated HBV DNA from covalently closed circular DNA (cccDNA) remains a challenge and may be key to definitions of functional cure. HBV vaccination outcomes remain suboptimal. Studies are needed to identify optimal vaccine strategies, which may rely on newer vaccine products and better population-based adherence to useful preventive vaccine regimens. Infection with HCV is now easily curable with DAAs, but new infections continue to occur, and men who have sex with men, with or without HIV infection, remain at high risk. An effective HCV vaccine is still elusive; development of new RNA vaccine technologies such as those used for SARS-CoV-2 may lead to development of new HCV vaccines. In addition, human challenge studies such as used for SARS-CoV-2, dengue, malaria, and other vaccines may promote HCV vaccine testing. Infection with HDV currently receives little attention but may emerge as a key issue with new treatment interventions approaching approval. However, availability of testing for HDV RNA is limited, and studies are needed to confirm the value of reflex-testing strategies following HBV detection.

Long-acting treatments for HCV and HBV infections may well transform their treatment and prevention, as they have for HIV. For HCV in particular, long-acting treatments might provide the ability to cure infection in a single encounter, opening the paradigm to test-and-cure public health approaches to elimination. In contrast, long-acting treatments for HBV infection might provide another "pill-free" option for maintaining treatment, especially useful when adherence is challenging. The dual activity of tenofovir against HIV and HBV makes development of those long-acting approaches of interest.

Although use of illicit drugs and high alcohol intake are known as key factors in the promotion and maintenance of HIV disease and viral hepatitis,

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recent data suggest they have substantial direct effects on liver injury and fibrosis progression. Cocaine may promote fibrosis independently of its effects on HIV. Fentanyl and other opioids may increase viral loads of HCV and HIV and thus alter both disease epidemiology and clinical presentation in individuals with either or both infections. Studies of these cofactors are needed to further elucidate their roles in individuals who use drugs.

The linkage between HIV infection and liver disease is multifactorial and remains a key driver of morbidity and mortality. Ongoing research in a variety of areas provides the opportunity to change the current landscape and improve the health of people with HIV.

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References

1. Klein MB, Althoff KN, Jing Y, et al. Risk of end-stage liver disease in HIV-viral hepatitis coinfected persons in North America from the early to modern antiretroviral therapy eras. *Clin Infect Dis*. 2016;63(9):1160-1167.
2. Sun J, Althoff KN, Jing Y, et al. Trends in hepatocellular carcinoma incidence and risk among persons with HIV in the US and Canada, 1996-2015. *JAMA Netw Open.* 2021;4(2):e2037512.
3. Torgersen J, Kallan MJ, Carbonari DM, et al. HIV RNA, CD4+ percentage, and risk of hepatocellular carcinoma by cirrhosis status. *J Natl Cancer Inst*. 2020;112(7):747-755.
4. Wandeler G, Mauron E, Atkinson A, et al. Incidence of hepatocellular carcinoma in HIV/HBV-coinfected patients on tenofovir therapy: relevance for screening strategies. *J Hepatol*. 2019;71(2):274-280.
5. Chen CJ, Iloeje UH, Yang HI. Long-term outcomes in hepatitis B: the REVEAL-HBV study. *Clin Liver Dis*. 2007;11(4):797-816, viii.
6. Kim HN, Newcomb CW, Carbonari DM, et al. Risk of HCC with hepatitis B viremia among HIV/HBV-coinfected persons in North America. *Hepatology.* 2021;74(3):1190-1202.
7. Béguelin C, Moradpour D, Sahli R, et al. Hepatitis delta-associated mortality in HIV/HBV-coinfected patients. *J Hepatol*. 2017;66(2):297-303.
8. Mocroft A, Lundgren J, Gerstoft J, et al. Clinical outcomes in persons coinfected with human immunodeficiency virus and hepatitis C virus: impact of hepatitis C virus treatment. *Clin Infect Dis*. 2020;70(10):2131-2140.
9. Kaplan A. Declining mortality for patients with human immunodeficiency virus (HIV) and cirrhosis: an analysis of national trends. Presented at the Eighth Biennial HIV and Liver Disease Conference. September 2021; Teton Village, WY.
10. Qu Y, Li M, Hamilton G, Zhang YN, Song B. Diagnostic accuracy of hepatic proton density fat fraction measured by magnetic resonance imaging for the evaluation of liver steatosis with histology as reference standard: a meta-analysis. *Eur Radiol*. 2019;29(10):5180-5189.
11. Centers for Disease Control and Prevention (CDC). Estimated HIV incidence and prevalence in the United States, 2014-2018. https://stacks.cdc.gov/view/cdc/87841. Accessed August 12, 2022.
12. Dailey AF, Hoots BE, Hall HI, et al. Vital signs: human immunodeficiency virus testing and diagnosis delays - United States. *MMWR Morb Mortal Wkly Rep*. 2017;66(47):1300-1306.
13. MacGowan RJ, Chavez PR, Mermin JH. Implementation of HIV self-testing program in New York City-reply. *JAMA Intern Med*. 2020;180(4):616-617.
14. Ruiz MS, O'Rourke A, Allen ST, et al. Using interrupted time series analysis to measure the impact of legalized syringe exchange on HIV diagnoses in Baltimore and Philadelphia. *J Acquir Immune Defic Syndr*. 2019;82 Suppl 2:S148-S154.
15. Mohr R, Schierwagen R, Schwarze-Zander C, et al. Liver fibrosis in HIV patients receiving a modern cART: which factors play a role? *Medicine (Baltimore)*. 2015;94(50):e2127.
16. Lin W, Tsai WL, Shao RX, et al. Hepatitis C virus regulates transforming growth factor beta1 production through the generation of reactive oxygen species in

a nuclear factor kappaB-dependent manner. *Gastroenterology*. 2010;138(7):2509-2518, 2518.e1.
17. Salloum S, Holmes JA, Jindal R, et al. Exposure to human immunodeficiency virus/hepatitis C virus in hepatic and stellate cell lines reveals cooperative profibrotic transcriptional activation between viruses and cell types. *Hepatology*. 2016;64(6):1951-1968.
18. Liu PJ, Harris JM, Marchi E, et al. Author Correction: Hypoxic gene expression in chronic hepatitis B virus infected patients is not observed in state-of-the-art in vitro and mouse infection models. *Sci Rep*. 2020; 10(1):19332.
19. Kandathil AJ, Sugawara S, Goyal A, et al. No recovery of replication-competent HIV-1 from human liver macrophages. *J Clin Invest*. 2018;128(10):4501-4509.
20. Kazankov K, Barrera F, Møller HJ, et al. Soluble CD163, a macrophage activation marker, is independently associated with fibrosis in patients with chronic viral hepatitis B and C. *Hepatology*. 2014;60(2):521-530.
21. Lidofsky A, Holmes JA, Feeney ER, et al. Macrophage activation marker soluble CD163 is a dynamic marker of liver fibrogenesis in human immunodeficiency virus/hepatitis C virus coinfection. *J Infect Dis*. 2018;218(9):1394-1403.
22. Sherman KE, Meeds HL, Rouster SD, et al. Soluble CD163 identifies those at risk for increased hepatic inflammation & fibrosis. *Open Forum Infect Dis*. 2021; 8(6):ofab203.
23. Agarwal N, Iyer D, Gabbi C, et al. HIV-1 viral protein R (Vpr) induces fatty liver in mice via LXRalpha and PPARalpha dysregulation: implications for HIV-specific pathogenesis of NAFLD. *Sci Rep*. 2017;7(1):13362.
24. Shrivastava S, Kottilil S, Sherman KE, Masur H, Tang L. CCR5+ T-cells homed to the liver exhibit inflammatory and profibrogenic signatures in chronic HIV/HCV-coinfected patients. *Viruses*. 2021;13(10):2074.
25. Bartneck M, Koppe C, Fech V, et al. Roles of CCR2 and CCR5 for hepatic macrophage polarization in mice with liver parenchymal cell-specific NEMO deletion. *Cell Mol Gastroenterol Hepatol*. 2021;11(2):327-347.
26. Lefebvre E, Moyle G, Reshef R, et al. Antifibrotic effects of the dual CCR2/CCR5 antagonist cenicriviroc in animal models of liver and kidney fibrosis. *PLoS One*. 2016;11(6):e0158156.
27. Sherman KE, Abdel-Hameed E, Rouster SD, et al. Improvement in hepatic fibrosis biomarkers associated with chemokine receptor inactivation through mutation or therapeutic blockade. *Clin Infect Dis*. 2019 May 17;68(11):1911-1918.
28. Ratziu V, Sanyal A, Harrison SA, et al. Cenicriviroc treatment for adults with nonalcoholic steatohepatitis and fibrosis: final analysis of the phase 2b CENTAUR study. *Hepatology*. 2020;72(3):892-905.
29. Schuetz A, Deleage C, Sereti I, et al. Initiation of ART during early acute HIV infection preserves mucosal Th17 function and reverses HIV-related immune activation. *PLoS Pathog*. 2014;10(12):e1004543.
30. Brenchley JM, Price DA, Schacker TW, et al. Microbial translocation is a cause of systemic immune activation in chronic HIV infection. *Nat Med*. 2006;12(12): 1365-1371.
31. Vujkovic-Cvijin I, Dunham RM, Iwai S, et al. Dysbiosis of the gut microbiota is associated with HIV disease progression and tryptophan catabolism. *Sci Transl Med*. 2013;5(193):193ra91.
32. Nganou-Makamdop K, Talla A, Sharma AA, et al. Translocated microbiome composition determines immunological outcome in treated HIV infection. *Cell.* 2021;184(15):3899-3914.e16.
33. Balagopal A, Philp FH, Astemborski J, et al. Human immunodeficiency virus-related microbial translocation and progression of hepatitis C. *Gastroenterology*. 2008;135(1):226-233.
34. Kardashian A, Ma Y, Yin MT, et al. High kynurenine:tryptophan ratio is associated with liver fibrosis in HIV-monoinfected and HIV/hepatitis C virus-coinfected women. *Open Forum Infect Dis*. 2019; 6(7):ofz281.
35. Sharpton SR, Maraj B, Harding-Theobald E, Vittinghoff E, Terrault NA. Gut microbiome-targeted therapies in nonalcoholic fatty liver disease: a systematic review, meta-analysis, and meta-regression. *Am J Clin Nutr*. 2019;110(1):139-149.
36. Dasgupta S, Broz D, Tanner M, et al. Changes in reported injection behaviors following the public health response to an HIV outbreak among people who inject drugs: Indiana, 2016. *AIDS Behav*. 2019;23(12): 3257-3266.
37. Knowles H. Rural Indiana county ends needle swap that helped fight HIV — sparking fears of another outbreak. *The Washington Post.* June 5, 2021.
38. Pintado C, Delaugerre C, Molina JM. Acute hepatitis B infection after a switch to long-acting cabotegravir and rilpivirine. *Open Forum Infect Dis*. 2020;7(9):ofaa367.
39. Bischoff J, Gu W, Schwarze-Zander C, et al. Stratifying the risk of NAFLD in patients with HIV under combination antiretroviral therapy (cART). *EClinicalMedicine.* 2021;40:101116.
40. Khaimova R, Fischetti B, Cope R, Berkowitz L, Bakshi A. Serological response with Heplisav-B® in prior Hepatitis B vaccine non-responders living with HIV. *Vaccine.* 2021;39(44):6529-6534.
41. Schnittman SR, Zepf R, Cocohoba J, Sears D. Brief report: heplisav-B seroprotection in people with HIV: a single-center experience. *J Acquir Immune Defic Syndr.* 2021;86(4):445-449.
42. Vesikari T, Langley JM, Segall N, et al. Immunogenicity and safety of a tri-antigenic versus a mono-antigenic hepatitis B vaccine in adults (PROTECT): a randomised, double-blind, phase 3 trial. *Lancet Infect Dis*. 2021;21(9):1271-1281.
43. Naggie S, Cooper C, Saag M, et al. Ledipasvir and sofosbuvir for HCV in patients coinfected with HIV-1. *N Engl J Med*. 2015;373(8):705-713.
44. Shimada S, Ivanics T, Kitajima T, et al. Improvements in liver transplant outcomes in patients with HCV/HIV coinfection after the introduction of direct-acting antiviral therapies. *Transpl Infect Dis*. 2022;24(2)e13808.
45. Saab S, Le L, Saggi S, Sundaram V, Tong MJ. Toward the elimination of hepatitis C in the United States. *Hepatology*. 2018;67(6):2449-2459.

46. Bretaña NA, Gray RR, Cunningham EB, et al. Combined treatment and prevention strategies for hepatitis C virus elimination in the prisons in New South Wales: a modelling study. *Addiction*. 2020;115(5):901-913.

47. van Santen DK, Boyd A, Matser A, et al. The effect of needle and syringe program and opioid agonist therapy on the risk of HIV, hepatitis B and C virus infection for people who inject drugs in Amsterdam, the Netherlands: findings from an emulated target trial. *Addiction.* 2021; 116(11):3115-3126.

48. Wall A, Lee GH, Maldonado J, Magnus D. Medical contraindications to transplant listing in the USA: a survey of adult and pediatric heart, kidney, liver, and lung programs. *World J Surg*. 2019;43(9):2300-2308.

49. Ahmed S, Algarin AB, Thadar H, et al. Comorbidities among persons living with HIV (PLWH) in Florida: a network analysis. *AIDS Care*. doi:10.1080/09540121.2022.2038363.

50. Seval N, Frank CA, Litwin AH, et al. Design and methods of a multi-site randomized controlled trial of an integrated care model of long-acting injectable buprenorphine with infectious disease treatment among persons hospitalized with infections and opioid use disorder. *Contemp Clin Trials*. 2021;105:106394.

51. Sudjaritruk T, Aurpibul L, Songtaweesin WN, et al. Integration of mental health services into HIV healthcare facilities among Thai adolescents and young adults living with HIV. *J Int AIDS Soc*. 2021;24(2):e25668.

52. Weaver MR, Conover CJ, Proescholdbell RJ, et al. Cost-effectiveness analysis of integrated care for people with HIV, chronic mental illness and substance abuse disorders. *J Ment Health Policy Econ*. 2009;12(1):33-46.

53. Czaja AJ. Review article: chemokines as orchestrators of autoimmune hepatitis and potential therapeutic targets. *Aliment Pharmacol Ther*. 2014;40(3):261-279.

54. Sharpton SR, Schnabl B, Knight R, Loomba R. Current concepts, opportunities, and challenges of gut microbiome-based personalized medicine in nonalcoholic fatty liver disease. *Cell Metab*. 2021;33(1):21-32.

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Special Contribution

2022 Update of the Drug Resistance Mutations in HIV-1

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*The 2022 edition of the IAS–USA drug resistance mutations list updates the **Figure** last published in September 2019. The mutations listed are those that have been identified by specific criteria for evidence and drugs described. The **Figure** is designed to assist practitioners to identify key mutations associated with resistance to antiretroviral drugs, and therefore, in making clinical decisions regarding antiretroviral therapy.*

Keywords: HIV, antiretroviral, drug resistance, TAM, therapy, mutations

The 2022 edition of the International Antiviral Society–USA (IAS–USA) drug resistance mutations list updates the **Figure** last published in September 2019.[1] In this update:

- Cabotegravir, fostemsavir, and ibalizumab have now been approved by regulatory agencies in many countries are all now included. The capsid inhibitor lenacapavir (GS 6207) has been added to the **Figure**.[2]
- A new section on specific drugs and details has been added to this update for information on recently approved drugs, that may not been added to the **Figure**.
- Several changes were made to drugs already on the **Figure**. Several changes were made to the **Figure Bars** of the integrase strand transfer inhibitors (InSTIs) cabotegravir and dolutegravir, the protease inhibitors atazanavir and lopinavir, and the nonnucleoside analogue reverse transcriptase (NNRTI) inhibitor doravirine.
- The user notes for tenofovir have been modified as recent clinical data suggest that the K65R plus M184V mutational profile is of less clinical relevance if tenofovir with either lamivudine or emtricitabine is prescribed in combination with a boosted protease inhibitor or one of the second generation InSTIs bictegravir or dolutegravir.
- For antiretroviral drugs that are no longer recommended, the associated **Figure Bars** are listed at the bottom of the drug class and are shaded in gray. Their user notes are retained for historical significance.

Specific Drugs and Details

Cabotegravir (formerly GSK-1265744) was approved by the US Food and Drug Administration (FDA) in December 2021 in combination with rilpivirine for the treatment of HIV-1 infection in adults who are virologically suppressed on a stable antiretroviral regimen with no history of treatment

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failure and with no known or suspected resistance to either cabotegravir or rilpivirine. Cabotegravir is available for the treatment of HIV-1 infection as oral formulation or as an extended-release injectable suspension copackaged with rilpivirine.[3,4] Cabotegravir suspension was also approved as a long-acting injectable for the use of preexposure prophylaxis (PrEP).[5]

Fostemsavir (formerly GSK-3684934) was approved by the FDA in February 2020 as a first-in-class oral attachment inhibitor binding to gp120.[6] It is licensed for the treatment of HIV-1 infection in combination with other antiretroviral drugs in heavily treatment-experienced adults with multidrug-resistant HIV-1 infection in whom their current regimen has failed due to resistance, intolerance, or safety considerations.[2,7] Fostemsavir shows high variation of in vitro susceptibility, but susceptibility is not dependent on tropism or on subtype with the exception of CRF01_AE, which shows intrinsic resistance.[8,9] In areas where CRF01_AE is prevalent, subtyping is recommended. No correlation between baseline resistance and treatment success has yet been established. For this reason, resistance testing for gp120 is not currently recommended. Fostemsavir-associated resistance does not cause cross-resistance to other entry or attachment inhibitors such as ibalizumab and maraviroc.[10]

Ibalizumab, a humanized monoclonal antibody and noncompetitive CD4 post-attachment inhibitor, is approved for treatment in patients with multiclass drug-resistant virus.[11] Since the mechanism of action of ibalizumab requires a previous attachment of HIV-gp120 to the CD4 receptor, ibalizumab does not interfere with the functional capacity of CD4 receptors unbound to HIV-1. Loss of N-linked glycosylation sites in the V5 loop reduce the activity of this compound by preventing HIV-1 gp120 conformational changes and gp41 rearrangements required for the virus to enter target cells.[12-14] There are no mutations depicted on the **Figure Bars** for fostemsavir, ibalizumab, or maraviroc. As such, genotypic testing to predict resistance to these drugs is not recommended in clinical practice. In rare occasions phenotypic testing may be performed, if available.

Methods

The IAS–USA Drug Resistance Mutations Group is an independent, volunteer panel of experts charged with delivering accurate, unbiased, and evidence-based information on drug resistance–associated mutations for HIV clinical practitioners. The group reviews new data on HIV drug resistance to maintain a current list of mutations associated with clinical resistance to HIV-1. This list includes mutations that may contribute to a reduced virologic response to a drug.

The group considers only data that have been published or have been presented at a scientific conference. Table 1 provides the list of amino acids and the abbreviations used. Drugs that have been approved by the US FDA and are generally recommended, as well as any drugs available in development with expectation of approval in the next few years are included (listed in alphabetic order by drug class). Drugs that are no longer recommended are listed at the bottom of the class and are shaded in gray. User notes provide additional information. Although the Drug Resistance Mutations Group works to maintain a complete and current list of these mutations, it cannot be assumed that the list presented here is definitive.

The magnitude of the reduction in susceptibility conferred by drug resistance mutations varies widely, and is modulated by the genetic context of the HIV sequence in which the mutation occurs. Despite the fact that mutations result in a spectrum of degrees of resistance, mutations have been arbitrarily designated as major (bolded) or minor (not bolded) (see Figure 1). Those defined as major tend to occur earlier during treatment failure and generally confer larger reductions in susceptibility. Those defined as minor tend to accrue after the emergence of a major mutation, confer some incremental resistance, may occur as well as polymorphisms in wild-type virus, and in some cases do not reduce susceptibility but restore replication fitness to viruses with resistance mutations

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Table 1. Amino acids and their abbreviations.

Alanine	A	Methionine	M
Cysteine	C	Asparagine	N
Aspartate	D	Proline	P
Glutamate	E	Glutamine	Q
Phenylalanine	F	Arginine	R
Glycine	G	Serine	S
Histidine	H	Threonine	T
Isoleucine	I	Valine	V
Lysine	K	Tryptophan	W
Leucine	L	Tryosine	Y

that impair fitness. In general, a major mutation should raise concern that a drug is at least partially compromised; a minor mutation on its own may not raise such a concern, but it should add concern in the presence of other mutations. The delineation between major and minor is often not clear-cut.

Identification of Mutations

The mutations listed are those that have been identified by 1 or more of the following criteria: (1) in vitro passage experiments with validation of contribution to resistance by using site-directed mutagenesis; (2) susceptibility testing of laboratory or clinical isolates; (3) nucleotide sequencing of viruses from patients in whom the drug is failing; (4) association studies between genotype at baseline and virologic response in patients exposed to the drug.

The development of more recently approved drugs that cannot always be tested as monotherapy precludes assessment of the impact of resistance on antiretroviral activity that is not seriously confounded by the activity of other drug components in the background regimen. Readers are encouraged to consult the literature and experts in the field for clarification or more information about specific mutations and their clinical impact. Polymorphisms associated with impaired treatment responses that occur in otherwise wild-type viruses should not be used in epidemiologic analyses to identify transmitted HIV-1 drug resistance. Consequently, only some of the resistance mutations depicted on the Figure can be used to identify transmitted drug resistance.[15]

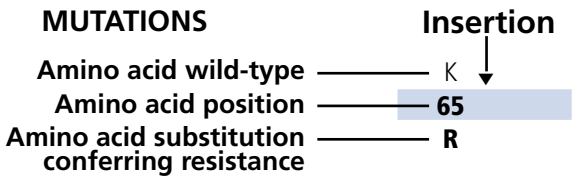

Figure 1. Display of the Figure Bar: Amino acid position, wild type, mutation conferring resistance, and indication of insertion mutation.

Clinical Context

The **Figure** is designed for practitioners to use in identifying key mutations associated with antiretroviral drug resistance and in selecting therapeutic regimens. In the context of making clinical decisions regarding antiretroviral therapy, evaluating the results of HIV-1 genotypic testing includes: (1) assessing whether the pattern or absence of a pattern in the mutations is consistent with the patient's history of antiretroviral therapy; and (2) recognizing that resistant strains may be present at levels below the limit of detection of the test after discontinuation or during poor adherence of the regimen that conferred the selection pressure. Analyzing stored samples, collected under selection pressure, could be useful in this setting; and (3) recognizing that virologic failure of a first-line regimen typically involves HIV-1 isolates with resistance to only 1 or 2 of the drugs in the regimen. In this setting, resistance emerges most commonly to lamivudine or emtricitabine, NNRTIs, or first-generation InSTIs (elvitegravir, raltegravir).

The absence of detectable viral resistance after treatment failure may result from any combination of the following factors: the presence of drug-resistant minority viral populations, a prolonged interval between the time of antiretroviral drug discontinuation and genotypic testing, nonadherence to medications, laboratory error, lack of current knowledge of the association of certain mutations with drug resistance, the occurrence of relevant mutations outside the regions targeted by routine resistance assays, drug-drug interactions leading to subtherapeutic drug levels, and possibly the consequence of drugs not reaching optimal levels in specific anatomic compartments.

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For more in-depth reading and an extensive reference list, see the 2018 IAS–USA panel recommendations for resistance testing[16] and 2020 IAS–USA panel recommendations for antiretroviral therapy.[17] Updates to the **Figure** are posted periodically at www.iasusa.org.

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References

1. Wensing AM, Calvez V, Ceccherini-Silberstein F, et al. 2019 update of the drug resistance mutations in HIV-1. *Top Antivir Med*. 2019;27(3):111-121.
2. Dvory-Sobol H, Shaik N, Callebaut C, Rhee MS. Lenacapavir: a first-in-class HIV-1 capsid inhibitor. *Curr Opin HIV AIDS*. 2022;17(1):15-21.
3. ViiV Healthcare. Vocabria [prescribing information]. 2021.Research Triangle Park, NC, Viiv Healthcare.
4. ViiV Healthcare. Cabenuva [prescribing information]. 2021.Research Triangle Park, Viiv Healthcare.
5. ViiV Healthcare. Apretude [prescribing information]. 2021.Research Triangle Park, NC, ViiV Healthcare.
6. Lataillade M, Lalezari JP, Kozal M, et al. Safety and efficacy of the HIV-1 attachment inhibitor prodrug fostemsavir in heavily treatment-experienced individuals: week 96 results of the phase 3 BRIGHTE study. *Lancet HIV*. 2020;7(11):e740-e751.
7. ViiV Healthcare. Rukobia [prescribing information]. 2020.Research Triangle Park, NC, ViiV Healthcare.
8. Nowicka-Sans B, Gong YF, McAuliffe B, et al. In vitro antiviral characteristics of HIV-1 attachment inhibitor BMS-626529, the active component of the prodrug BMS-663068. *Antimicrob Agents Chemother.* 2012; 56(7):3498-3507.
9. Montaner LJ, Lynn K, Azzoni L, et al. Susceptibility to 3BNC117 and 10-1074 in ART-suppressed chronically infected persons [CROI Abstract 503]. In Special Issue: Abstracts From the 2022 Conference on Retroviruses and Opportunistic Infections. *Top Antivir Med*. 2022;30(1s):192-193.
10. Rose R, Gartland M, Li Z, et al. Clinical evidence for a lack of cross-resistance between temsavir and ibalizumab or maraviroc. *AIDS*. 2022;36(1):11-18.
11. Canada Theratechnologies Inc. Trogarzo [prescribing information]. 2018.Montréal, Québec Canada, Canada Theratechnologies Inc.
12. Emu B, Fessel J, Schrader S, et al. Phase 3 study of ibalizumab for multidrug-resistant HIV-1. *N Engl J Med*. 2018;379(7):645-654.
13. Pace CS, Fordyce MW, Franco D, Kao CY, Seaman MS, Ho DD. Anti-CD4 monoclonal antibody ibalizumab exhibits breadth and potency against HIV-1, with natural resistance mediated by the loss of a V5 glycan in envelope. *JAIDS*. 2013;62(1):1-9.
14. Toma J, Weinheimer SP, Stawiski E, et al. Loss of asparagine-linked glycosylation sites in variable region 5 of human immunodeficiency virus type 1 envelope is associated with resistance to CD4 antibody ibalizumab. *J Virol*. 2011;85(8):3872-3880.
15. Pingen M, Nijhuis M, de Bruijn JA, Boucher CA, Wensing AM. Evolutionary pathways of transmitted drug-resistant HIV-1. *J Antimicrob Chemother*. 2011;66(7): 1467-1480.
16. Gunthard HF, Calvez V, Paredes R, et al. Human immunodeficiency virus drug resistance: 2018 recommendations of the International Antiviral Society–USA panel. *Clin Infect Dis*. 2018;67:1-11.
17. Saag MS, Gandhi RT, Hoy JF, et al. Antiretroviral drugs for treatment and prevention of HIV infection in adults: 2020 recommendations of the International Antiviral Society-USA Panel. *JAMA*. 2020;324(16):1651-1669.

Top Antivir Med. 2022;30(4):559-574.

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

Nucleoside and Nucleotide Analogue Reverse Transcriptase Inhibitors (nRTIs)[1]

69 Insertion Complex[2] (affects all nRTIs currently approved by the US FDA)

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

151 Complex[3] (affects all nRTIs currently approved by the US FDA except tenofovir)

	A	V	F	F	Q
Multi-nRTI Resistance	**62**	**75**	**77**	**116**	**151**
	V	**I**	**L**	**Y**	**M**

Thymidine Analogue-Associated Mutations[4,5] (TAMs; affect all nRTIs currently approved by the US FDA other than emtricitabine and lamivudine)

	M	K	L	T	K
Multi-nRTI Resistance	**41**	**70**	**210**	**215**	**219**
	L	**R**	**W**	**Y**	**Q**
				F	**E**

	K	L	Y	M
Abacavir[1,6]	**65**	**74**	**115**	**184**
	R	**V**	**F**	**V**
	E			
	N			

	K	M
Emtricitabine/ Lamivudine	**65**	**184**
	R	**V**
	E	**I**
	N	

	K	K
Tenofovir[1,7]	**65**	**70**
	R	**E**
	E	
	N	

	M	D	K	L	T	K
Zidovudine[4,5,8,9]	**41**	**67**	**70**	**210**	**215**	**219**
	L	**N**	**R**	**W**	**Y**	**Q**
					F	**E**

	K	L
Didanosine[1,10,21]	**65**	**74**
	R	**V**
	E	
	N	

	M	K	D	K	L	T	K
Stavudine[1,4,5,8,9]	**41**	**65**	**67**	**70**	**210**	**215**	**219**
	L	**R**	**N**	**R**	**W**	**Y**	**Q**
		E				**F**	**E**
		N					

Nonnucleoside Analogue Reverse Transcriptase Inhibitors (NNRTIs)[1,11]

	V	Y	G	P	F	M	L	Y
Doravirine[12]	**106**	**188**	190	225	**227**	**230**	234	**318**
	A	**L**	E	H	**C**	**L**	I	**F**
	I				I			
	M				**L**			
	T				R			
					V			

	L	K	K	V	V	Y	Y	G	P	M
Efavirenz	**100**	**101**	**103**	**106**	**108**	**181**	**188**	**190**	**225**	**230**
	I	**P**	**N**	**M**	**I**	**C**	**L**	**S**	**H**	**L**
			S			**I**		**A**		

	V	A	L	K	V	E	V	Y	G	M
Etravirine[13]	90	98	**100**	**101**	106	138	179	**181**	190	230
	I	G	**I**	E	I	A	D	**C**	S	L
				H		G	F	**I**	A	
				P		K	T	**V**		
						Q				

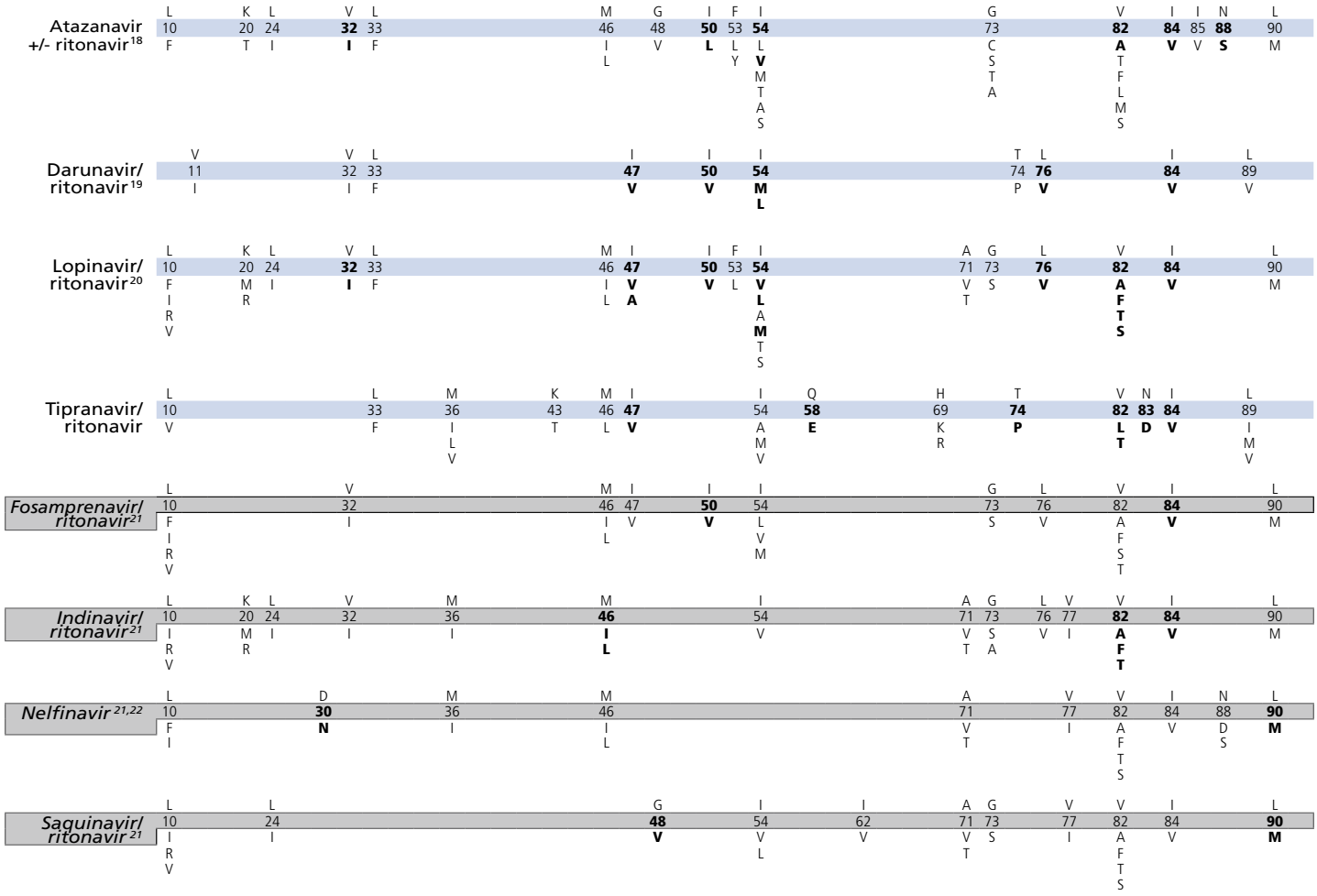
	L	K	K	V	V	Y	Y	G	M
Nevirapine	**100**	**101**	**103**	**106**	**108**	**181**	**188**	**190**	**230**
	I	**P**	**N**	**A**	**I**	**C**	**C**	**A**	**L**
			S	**M**		**I**	**L**		
							H		

	L	K	E	V	Y	Y	H	F	M
Rilpivirine[14]	**100**	**101**	**138**	**179**	**181**	**188**	**221**	**227**	**230**
	I	**E**	**A**	**L**	**C**	**L**	**Y**	**C**	**I**
		P	**G**		**I**				**L**
			K		**V**				
			Q						
			R						

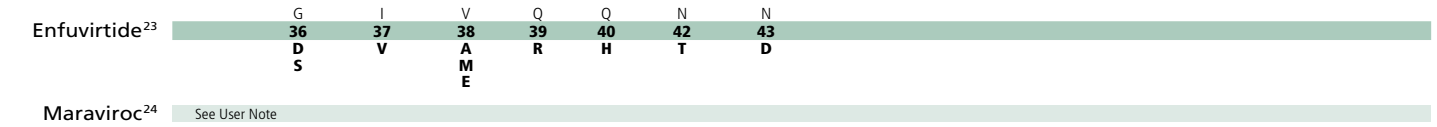
MUTATIONS IN THE CAPSID GENE ASSOCIATED WITH RESISTANCE TO CAPSID INHIBITORS

	L	M	Q	K	N	A	T
Lenacapavir[31]	56	**66**	**67**	70	74	105	107
	I	**I**	**H**	N	D	T	N
				S	S		
				R			

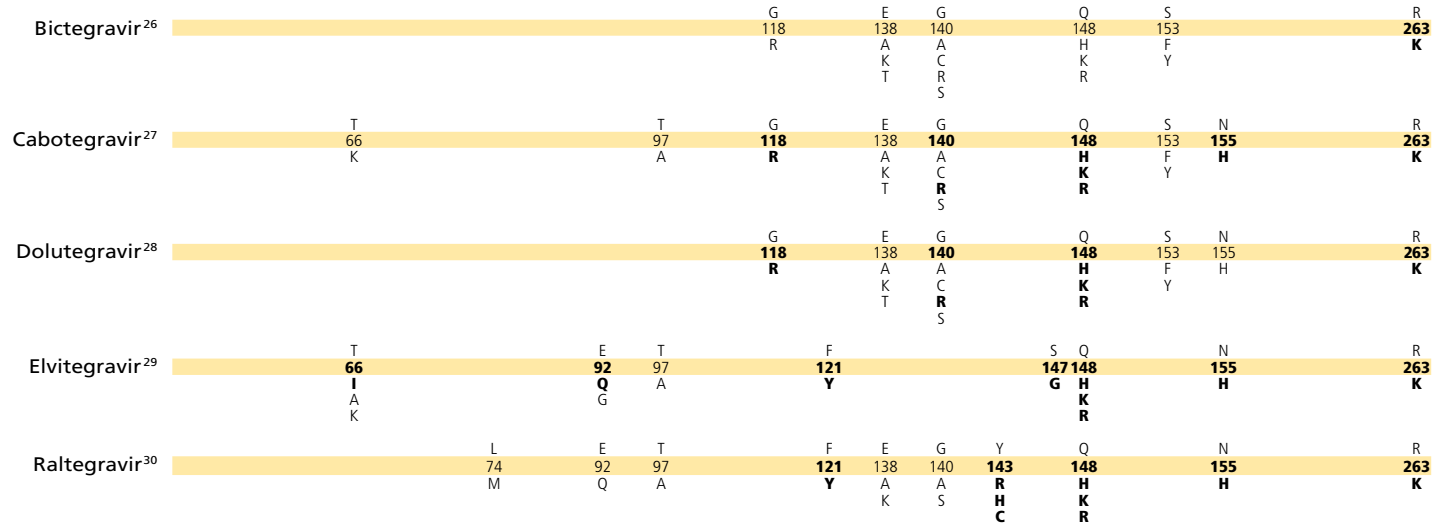
MUTATIONS IN THE PROTEASE GENE ASSOCIATED WITH RESISTANCE TO PROTEASE INHIBITORS (PIs)[15,16,17]



MUTATIONS IN THE ENVELOPE GENE ASSOCIATED WITH RESISTANCE TO ENTRY INHIBITORS



MUTATIONS IN THE INTEGRASE GENE ASSOCIATED WITH RESISTANCE TO INTEGRASE STRAND TRANSFER INHIBITORS[25]



User Notes

1. Mutations at the C-terminal reverse transcriptase domains (amino acids 293-560) outside of regions depicted on the **Figure Bar** may contribute to nucleoside (or nucleotide) analogue reverse transcriptase inhibitor (nRTI) and nonnucleoside analogue reverse transcriptase inhibitors (NNRTI) HIV-1 drug resistance. The clinical relevance of these connection domain mutations arises mostly in conjunction with thymidine analogue–associated mutations (TAMs) and M184V and they have not been associated with increased rates of virologic failure of etravirine or rilpivirine in clinical trials.[1-3] K65E/N/R variants are reported in patients experiencing treatment failure of tenofovir (ie, tenofovir disoproxil fumarate [TDF] or tenofovir alafenamide [TAF]), stavudine, or didanosine. The K65R/N variants may be selected by tenofovir, didanosine, abacavir, or stavudine and are associated with decreased viral susceptibility to these drugs.[4-8] The K65R may be more easily selected in subtype C clades.[9] K65E usually occurs in mixtures with wild-type virus. Patient-derived viruses with K65E and site-directed mutations replicate very poorly in vitro; as such, no susceptibility testing can be performed.[10,11] Some nRTI mutations, like T215Y and H208Y,[12] may lead to viral hypersusceptibility to NNRTIs, including etravirine.[13] The presence of these mutations may improve subsequent virologic response to NNRTI-containing regimens (nevirapine or efavirenz) in NNRTI-naive individuals;[14-18] no clinical data exist for improved response to etravirine in NNRTI-experienced individuals.

2. The 69 insertion complex consists of a substitution at codon 69 (typically T69S) and an insertion of 2 or more amino acids (S-S, S-A, S-G, or others). The 69 insertion complex is associated with resistance to all nRTIs currently approved by the US Food and Drug Administration (FDA) when present with 1 or more TAMs at codons 41, 210, or 215.[4] Some other amino acid changes from the wild-type T at codon 69 without the insertion may be associated with broad nRTI resistance.

3. Since no differences in resistance patterns have been observed between TDF and TAF, both drugs are referred to as "tenofovir" on the **Figure Bar**.[19] Tenofovir retains activity against the Q151M complex of mutations.[4] Q151M is the most important mutation in the complex (ie, the other mutations in the complex [A62V, V75I, F77L, and F116Y] in isolation may not reflect multi-nucleoside resistance).

4. Mutations known to be selected by TAMs (ie, M41L, D67N, K70R, L210W, T215Y/F, and K219Q/E) also confer reduced susceptibility to all currently approved nRTIs[20] except emtricitabine and lamivudine, which in fact reverse the magnitude of resistance and are recommended with tenofovir or zidovudine in the presence of TAMs. The degree to which cross-resistance is observed depends on the specific mutations and number of mutations involved.[21-24]

5. Although reverse transcriptase changes associated with the E44D and V118I mutations may have an accessory role in increased resistance to nRTIs in the presence of TAMs, their clinical relevance is very limited.[25-27]

6. The M184V mutation alone does not appear to be associated with a reduced virologic response to abacavir in vivo. When associated with TAMs, M184V increases abacavir resistance.[5,28]

7. If resistance develops specifically to tenofovir, the most common drug resistance mutation to emerge is K65R. It is associated with about 2-fold reduced tenofovir susceptibility, which is clinically significant. However, when K65R occurs in combination with the lamivudine/emtricitabine resistance mutation M184V/I, the reduction in tenofovir susceptibility is less than 1.5 fold, a reduction in susceptibility that is less clinically significant. This is particularly the case in patients who are treated with the combination of tenofovir, a cytosine analogue, and a highly potent third drug such as the integrase strand transfer inhibitors (InSTIs) bictegravir and dolutegravir or a boosted protease inhibitor (PI).[29,30]

A reduced response also occurs in the presence of 3 or more TAMs inclusive of either M41L or L210W.[4] The presence of TAMs or combined treatment with zidovudine prevents the emergence of K65R in the presence of tenofovir.[31-33]

8. The presence of M184V appears to delay or prevent emergence of TAMs.[34] This effect may be overcome by an accumulation of TAMs.

9. The T215A/C/D/E/G/H/I/L/N/S/V substitutions are revertant mutations at codon 215 that confer increased risk of virologic failure of zidovudine or stavudine in antiretroviral-naive patients.[35,36] The T215Y mutant may emerge quickly from one of these mutations in the presence of zidovudine or stavudine.[37]

10. The presence of 3 of the following mutations—M41L, D67N, L210W, T215Y/F, K219Q/E—is associated with resistance to didanosine.[38] The presence of K70R or M184V alone does not decrease virologic response to didanosine.[39] However, the mutations depicted on the **Figure Bar** cannot be considered comprehensive because little relevant research has been reported in recent years to update the resistance and cross-resistance patterns for this drug.

11. There is no evidence for the utility of efavirenz, nevirapine, or rilpivirine in patients with NNRTI resistant virus.[40]

12. Doravirine is active in vitro against variants containing the common NNRTI mutations K103N, E138K, Y181C, and G190A.[41,42] Doravirine selects for mutations at positions 106, 108, 227, and 234, with more than 1 mutation usually required for substantial levels of resistance.[43] Mutations V106A, Y188L, and M230L are associated with a 10- or greater fold reduced susceptibility to doravirine. V106A and Y188L have also been selected in vivo.[44,45] In 1 clinical isolate, G190E was associated with about 20-fold reduced susceptibility to doravirine.[42] Furthermore, the double

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and triple mutants V106A and F227L; V106A and L234I; V106A and F227L and L234I; and V106A and 190A and F227L, are all associated with substantial resistance to doravirine.[41,43,46]

13. Resistance to etravirine has been extensively studied only in the context of coadministration with ritonavir-boosted darunavir. Mutations associated with virologic outcome were assessed and their relative weights (or magnitudes of impact) assigned. Phenotypic cutoff values were calculated, and assessments of genotype-phenotype correlations from a large clinical database have determined the relative importance of the various mutations. These 2 approaches are in agreement for many, but not all, mutations and weights.[47-49] The single mutations L100I, K101P, and Y181C/I/V have high relative weights with regard to reduced susceptibility and reduced clinical response compared with other mutations.[50,51] The presence of K103N alone does not affect etravirine response.[51] Accumulation of several mutations results in greater reductions in susceptibility and virologic response than do single mutations.[52-54]

14. Sixteen mutations have been associated with decreased rilpivirine susceptibility (K101E/P, E138A/G/K/Q/R, V179L, Y181C/I/V, Y188L, H221Y, F227C, and M230I/L).[55-57] The K101P and Y181I/V mutations reduce rilpivirine susceptibility approximately 50 fold and 15 fold, respectively, but are not commonly observed in patients receiving rilpivirine.[58-60] Mutations at position 138 (most notably E138A) may occur as natural polymorphisms, especially in non-B subtype virus.[61] The K101E, E138K, and Y181C mutations, each of which reduces rilpivirine susceptibility 2.5 fold to 3 fold, occur commonly in patients receiving rilpivirine. E138K and to a lesser extent K101E usually occur in combination with the nRTI resistance–associated mutation M184I, which alone does not reduce rilpivirine susceptibility. When M184I is combined with E138K or K101E, rilpivirine susceptibility is reduced about 7 fold and 4.5 fold, respectively.[60,62-64] The combinations of reverse transcriptase–associated mutations L100I plus K103N/S and L100I plus K103R plus V179D were strongly associated with reduced susceptibility to rilpivirine; however, for isolates harboring the K103N/R/S or V179D as single mutations, no reduction in susceptibility was detected.[57,65]

15. Often, several mutations are necessary to substantially impact virologic response to a ritonavir-boosted PI.[66]

16. Mutations in Gag cleavage sites may confer or contribute to resistance to PIs and may even emerge before mutations in protease.[67] A large proportion of virus samples from patients with confirmed virologic failure on a PI-containing regimen is not found to have PI resistance–associated mutations, attributable to poor adherence.

17. Ritonavir is not listed separately, as it is currently used only at low doses as a pharmacologic booster of other PIs.

18. Several mutations are associated with atazanavir resistance. Their impacts differ, with I50L, I84V, and N88S having the greatest effect. Mutations that are selected during unboosted atazanavir are not different from those selected during boosted atazanavir, but the relative frequency of mutations may differ. Higher atazanavir levels obtained with ritonavir boosting increase the number of mutations required for loss of activity. The presence of M46I plus L76V might increase susceptibility to atazanavir when no other related mutations are present.[68]

19. Virologic response to ritonavir-boosted darunavir correlates with baseline susceptibility and the presence of several specific PI resistance–associated mutations. Reductions in response are associated with increasing numbers of the mutations indicated on the **Figure Bar**. The negative impact of the protease mutations I47V, I54M, T74P, and I84V and the positive impact of the protease mutation V82A on virologic response to ritonavir-boosted darunavir were shown independently in 2 data sets.[69,70] Some of these mutations appear to have a greater effect on susceptibility than others (eg, I50V vs V11I). The presence at baseline of 2 or more of the substitutions V11I, V32I, L33F, I47V, I50V, I54L/M, T74P, L76V, I84V, or L89V was associated with a decreased virologic response to ritonavir-boosted darunavir.[71]

20. Virologic response to ritonavir-boosted lopinavir is affected by the presence of 3 or more of the following amino acid substitutions in protease at baseline: L10F/I/R/V, K20M/N/R, L24I, L33F, M36I, I47V, G48V, I54L/T/V, V82A/C/F/S/T, and I84V. In addition, the combination of 47A/V with V32I is associated with high-level resistance.[68,72-78] I50V is only occasionally selected in vivo but has a clear impact on susceptibility.[12,79-81] Subtype C patterns with M46L, I54V, L76V, and V82A are frequently observed in patients receiving ritonavir-boosted lopinavir.

21. The mutations depicted on the **Figure Bar** cannot be considered comprehensive because little relevant research has been reported in recent years to update the resistance and cross-resistance patterns for this drug.

22. In some nonsubtype-B HIV-1, D30N is selected less frequently than are other PI resistance–associated mutations.[82]

23. Resistance to enfuvirtide is associated primarily with mutations in the first heptad repeat (HR1) region of the gp41 envelope gene. However, mutations or polymorphisms in other regions of the env (eg, the HR2 region or those yet to be identified), as well as coreceptor usage and density, may affect susceptibility to enfuvirtide.[83-85]

24. The activity of CC chemokine receptor 5 (CCR5) antagonists is limited to patients with virus that use only CCR5 for entry (R5 virus). Viruses that use both CCR5 and CXC chemokine receptor 4 (CXCR4; termed dual/mixed [D/M] virus) or only CXCR4 (X4 virus) do not respond to treatment with CCR5 antagonists. Virologic failure of these drugs is frequently associated with outgrowth of D/M or X4 virus from a preexisting minority population present at levels below the limit of assay detection. Mutations in HIV-1 gp120 that allow the virus to bind to the drug-

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bound form of CCR5 have been described in viruses from some patients whose virus remained R5 after virologic failure of a CCR5 antagonist. Most of these mutations are found in the V3 loop, the major determinant of viral tropism.[86] There is as yet no consensus on specific signature mutations for CCR5 antagonist resistance, so they are not depicted on the **Figure Bar**. Some CCR5 antagonist–resistant viruses selected in vitro have shown mutations in gp41 without mutations in V3;[87] the clinical significance of such mutations is not yet known.

25. With their low genetic barrier to resistance and the high level of cross-resistance, the InSTIs elvitegravir and raltegravir are no longer generally recommended in an initial therapy for most people with HIV.[88] A second-generation InSTIs (dolutegravir, bictegravir, and cabotegravir) is recommended for most treatment situations.

26. In vitro susceptibility data indicate relatively small quantitative reductions in most cases for dolutegravir and bictegravir for single mutations in integrase.[89-91] Consequently, the **Figure Bar** listing the mutations or indicating them as bold is somewhat arbitrary in the absence of clinical data. The listing of mutations is based in most cases on in vitro selection data and testing single mutations seen mostly with first-generation InSTI failure in vitro. Several mutations were selected by dolutegravir, primarily during monotherapy trials or as add-on therapy to failing regimens.[92] Failure with the emergence of resistance to bictegravir, which is only available as a fixed-dose formulation with TAF and emtricitabine for individuals with no known InSTI resistance, has not been well documented. The only clinical data for treatment of individuals with InSTI resistance comes from the VIKING Study, in which even double doses of dolutegravir combined with the best available background regimen had higher failure rates against Q148K with 2 or more additional mutations in integrase.[93] Failure with emergence of resistance to bictegravir in a first-line regimen has been very rarely observed.[94] In vitro data suggest that these double mutants might have compromised the efficacy of bictegravir in one study but not another.[90,91] Multiple mutations are not displayed in the **Figure Bar**.

27. Cabotegravir is a long-acting InSTI. In clinical trials in individuals receiving HIV treatement or PrEP, several resistance mutations were observed in integrase associated with in vitro cabotegravir resistance.[95-97] A multivariate analysis showed that the presence of at least 2 factors among archived rilpivirine resistance-associated mutations at baseline, HIV-1 subtype A6/A1, or body mass index of at least 30 kg/m^2, was associated with increased risk of confirmed virologic failure.[98] The A6/A1 subtype frequently harbors the L74I polymorphism. A recent study showed that L74I conferred greater replication capacity to recombinant viruses expressing HIV-1 A6 integrase when present together with InSTI resistance mutations at positions 118, 140, 148, and 263. This finding may explain in part the association of this subtype to virologic failures of long-acting cabotegravir/rilpivirine.[99]

Although knowledge from clinical studies thus far is limited, in vitro studies indicate that multiple integrase substitutions including compensatory mutations enhance resistance to cabotegravir.[100]

28. Several mutations are required in HIV integrase to confer high-level resistance to dolutegravir.[100,101] Cross-resistance studies with raltegravir- and elvitegravir-resistant viruses indicate that Q148H/R and G140S in combination with mutations L74I/M, E92Q, T97A, E138A/K, G140A, or N155H are associated with 5-fold to 20-fold reduced dolutegravir susceptibility[102] and reduced virologic suppression in patients.[103-106]

29. Seven elvitegravir codon mutations have been observed in InSTI treatment–naive and –experienced patients in whom therapy is failing.[107-113] T97A, which may occur as a polymorphism,[114] results in only a 2-fold change in elvitegravir susceptibility and may require additional mutations for resistance.[112,113] The sequential use of elvitegravir and raltegravir (in either order) is not recommended because of cross-resistance between these drugs.[112]

30. Raltegravir failure is associated with integrase mutations in at least 3 distinct, but not exclusive, genetic pathways defined by 2 or more mutations including (1) a mutation at Q148H/K/R, N155H, or Y143R/H/C; and (2) 1 or more additional minor mutations. Minor mutations described in the Q148H/K/R pathway include L74M plus E138A, E138K, or G140S. The most common mutational pattern in this pathway is Q148H plus G140S, which also confers the greatest loss of drug susceptibility. Mutations described in the N155H pathway include this major mutation plus either L74M, E92Q, T97A, E92Q plus T97A, Y143H, G163K/R, V151I, or D232N.[115] The Y143R/H/C mutation is uncommon.[116-120] E92Q alone reduces susceptibility to elvitegravir more than 20 fold and causes limited (<5 fold) cross-resistance to raltegravir.[121-123] N155H mutants tend to predominate early in the course of raltegravir failure, but are gradually replaced with continuing raltegravir treatment by viruses with higher resistance, often bearing mutations G140S plus Q148H/R/K.

31. The emergence of resistance with lenacapavir was characterized with in vitro selection, which identified several variants in the capsid (CA) portion of Gag (L56I, M66I, Q67H, K70N, N74D/S, and T107N), with 20-fold to 1000-fold reduced susceptibility in vitro with Q67H+N74S, Q67H+T107N, L56I (204), Q67H+M66I, Q67H+N74D, M66I (>2,700), and reduced replication capacity for most mutant viruses.[124-126]

None of these mutations were found to be polymorphic suggesting there is no need for resistance testing before treatment with lenacapavir.[127] In a phase Ib study, post-monotherapy analyses revealed the emergence of mutation Q67H at the lowest lenacapavir doses.[125,126] In highly treatment-experienced patients with lenacapavir failure, M66I was

observed alone or in combination with other mutations. In all cases, the failures were initially associated with the selection of M66I.[30,128]

In highly treatment-experienced patients experiencing treatment failure in the CAPELLA study, the M66I mutation was most frequently observed.[129] In treatment-naive individuals in the CALIBRATE trial mutations 67H (fold change 7) and 70R were selected.[130,131]

References to the User Notes

1. von Wyl V, Ehteshami M, Demeter LM, et al. HIV-1 reverse transcriptase connection domain mutations: dynamics of emergence and implications for success of combination antiretroviral therapy. *Clin Infect Dis*. 2010;51(5):620-628.
2. Gupta S, Vingerhoets J, Fransen S, et al. Connection domain mutations in HIV-1 reverse transcriptase do not impact etravirine susceptibility and virologic responses to etravirine-containing regimens. *Antimicrob Agents Chemother.* 2011;55(6):2872-2879.
3. Rimsky L, Van Eygen V, Vingerhoets J, Leijskens E, Picchio G. Reverse transcriptase connection domain mutations were not associated with virological failure or phenotypic resistance in rilpivirine-treated patients from the ECHO and THRIVE Phase III trials (week 96 analysis). *Antivir Ther*. 2012;17(Suppl 1):A36.
4. Miller MD, Margot N, Lu B, et al. Genotypic and phenotypic predictors of the magnitude of response to tenofovir disoproxil fumarate treatment in antiretroviral-experienced patients. *J Infect Dis*. 2004; 189(5):837-846.
5. Harrigan PR, Stone C, Griffin P, et al. Resistance profile of the human immunodeficiency virus type 1 reverse transcriptase inhibitor abacavir (1592U89) after monotherapy and combination therapy. CNA2001 Investigative Group. *J Infect Dis*. 2000;181(3):912-920.
6. Winters MA, Shafer RW, Jellinger RA, Mamtora G, Gingeras T, Merigan TC. Human immunodeficiency virus type 1 reverse transcriptase genotype and drug susceptibility changes in infected individuals receiving dideoxyinosine monotherapy for 1 to 2 years. *Antimicrob Agents Chemother.* 1997;41(4):757-762.
7. Svarovskaia ES, Margot NA, Bae AS, et al. Low-level K65R mutation in HIV-1 reverse transcriptase of treatment-experienced patients exposed to abacavir or didanosine. *JAIDS.* 2007;46(2):174-180.
8. Hawkins CA, Chaplin B, Idoko J, et al. Clinical and genotypic findings in HIV-infected patients with the K65R mutation failing first-line antiretroviral therapy in Nigeria. *JAIDS.* 2009;52(2):228-234.
9. Brenner BG, Oliveira M, Doualla-Bell F, et al. HIV-1 subtype C viruses rapidly develop K65R resistance to tenofovir in cell culture. *AIDS*. 2006; 20:F9-F13.
10. Fourati S, Visseaux B, Armenia D, et al. Identification of a rare mutation at reverse transcriptase Lys65 (K65E) in HIV-1–infected patients failing on nucleos(t)ide reverse transcriptase inhibitors. *J Antimicrob Chemother*. 2013;68(10): 2199-2204.
11. Chunduri H, Crumpacker C, Sharma PL. Reverse transcriptase mutation K65N confers a decreased replication capacity to HIV-1 in comparison to K65R due to a decreased RT processivity. *Virology.* 2011;414(1):34-41.
12. Clark SA, Shulman NS, Bosch RJ, Mellors JW. Reverse transcriptase mutations 118I, 208Y, and 215Y cause HIV-1 hypersusceptibility to non-nucleoside reverse transcriptase inhibitors. *AIDS.* 2006; 20(7):981-984.
13. Picchio G, Vingerhoets J, Parkin N, Azijn H, de Bethune MP. Nucleoside-associated mutations cause hypersusceptibility to etravirine. *Antivir Ther*. 2008;13(Suppl 3):A25.
14. Shulman NS, Bosch RJ, Mellors JW, Albrecht MA, Katzenstein DA. Genetic correlates of efavirenz hypersusceptibility. *AIDS.* 2004;18(13):1781-1785.
15. Demeter LM, DeGruttola V, Lustgarten S, et al. Association of efavirenz hypersusceptibility with virologic response in ACTG 368, a randomized trial of abacavir (ABC) in combination with efavirenz (EFV) and indinavir (IDV) in HIV-infected subjects with prior nucleoside analog experience. *HIV Clin Trials*. 2008;9(1):11-25.
16. Haubrich RH, Kemper CA, Hellmann NS, et al. The clinical relevance of non-nucleoside reverse transcriptase inhibitor hypersusceptibility: a prospective co hort analysis. *AIDS.* 2002;16(15): F33-F40.
17. Tozzi V, Zaccarelli M, Narciso P, et al. Mutations in HIV-1 reverse transcriptase potentially associated with hypersusceptibility to nonnucleoside reverse-transcriptase inhibitors: effect on response to efavirenz-based therapy in an urban observational cohort. *J Infect Dis*. 2004;189(9):1688-1695.
18. Katzenstein DA, Bosch RJ, Hellmann N, et al. Phenotypic susceptibility and virological outcome in nucleoside-experienced patients receiving three or four antiretroviral drugs. *AIDS*. 2003;17(6): l821-830.
19. Margot N, Cox S, Das M, McCallister S, Miller MD, Callebaut C. Infrequent development of drug resistance in HIV-1-infected treatment-naive subjects after 96 weeks of treatment with elvitegravir/cobicistat/emtricitabine/tenofovir alafenamide or elvitegravir/cobicistat/emtricitabine/tenofovir disoproxil fumarate. *Antivir Ther*. 2017;22(5):443-446.
20. Whitcomb JM, Parkin NT, Chappey C, Hellman NS, Petropoulos CJ. Broad nucleoside reverse-transcriptase inhibitor cross-resistance in human immunodeficiency virus type 1 clinical isolates. *J Infect Dis.* 2003;188(7):992-1000.
21. Larder BA, Kemp SD. Multiple mutations in HIV-1 reverse transcriptase confer high-level resistance to zidovudine (AZT). *Science.* 1989;246(4934):1155-1158.
22. Kellam P, Boucher CA, Larder BA. Fifth mutation in human immunodeficiency virus type 1 reverse transcriptase contributes to the development of high-level resistance to zidovudine. *Proc Natl Acad Sci USA*. 1992;89(5): 1934-1938.
23. Calvez V, Costagliola D, Descamps D, et al. Impact of stavudine phe-

notype and thymidine analogues mutations on viral response to stavudine plus lamivudine in ALTIS 2 ANRS trial. *Antivir Ther*. 2002;7(3):211-218.

24. Kuritzkes DR, Bassett RL, Hazelwood JD, et al. Rate of thymidine analogue resistance mutation accumulation with zidovudine- or stavudine-based regimens. *JAIDS.* 2004;36(1):600-603.

25. Romano L, Venturi G, Bloor S, et al. Broad nucleoside-analogue resistance implications for human immunodeficiency virus type 1 reverse-transcriptase mutations at codons 44 and 118. *J Infect Dis.* 2002;185(7):898-904.

26. Walter H, Schmidt B, Werwein M, Schwingel E, Korn K. Prediction of abacavir resistance from genotypic data: impact of zidovudine and lamivudine resistance in vitro and in vivo. *Antimicrob Agents Chemother.* 2002;46(1):89-94.

27. Mihailidis C, Dunn D, Pillay D, Pozniak A. Effect of isolated V118I mutation in reverse transcriptase on response to first-line antiretroviral therapy. *AIDS.* 2008;22(3):427-430.

28. Lanier ER, Ait-Khaled M, Scott J, et al. Antiviral efficacy of abacavir in antiretroviral therapy-experienced adults harbouring HIV-1 with specific patterns of resistance to nucleoside reverse transcriptase inhibitors. *Antivir Ther.* 2004;9(1):37-45.

29. Paton NI, Musaazi J, Kityo C, et al. Dolutegravir or darunavir in combination with zidovudine or tenofovir to treat HIV. *N Engl J Med.* 2021;385(4):330-341.

30. Segal-Maurer S, DeJesus E, Stellbrink HJ, et al. Capsid inhibition with lenacapavir in multidrug-resistant HIV-1 infection. *N Engl J Med*. 2022;386(19):1793-1803.

31. Parikh UM, Zelina S, Sluis-Cremer N, Mellors JW. Molecular mechanisms of bidirectional antagonism between K65R and thymidine analog mutations in HIV-1 reverse transcriptase. *AIDS*. 2007;21(11):1405-1414.

32. Parikh UM, Barnas DC, Faruki H, Mellors JW. Antagonism between the HIV-1 reverse-transcriptase mutation K65R and thymidine-analogue mutations at the genomic level. *J Infect Dis*. 2006;194(5):651-660.

33. von Wyl V, Yerly S, Böni J, et al. Factors associated with the emergence of K65R in patients with HIV-1 infection treated with combination antiretroviral therapy containing tenofovir. *Clin Infect Dis*. 2008;46(8):1299-1309.

34. Kuritzkes DR, Quinn JB, Benoit SL, et al. Drug resistance and virologic response in NUCA 3001, a randomized trial of lamivudine (3TC) versus zidovudine (ZDV) versus ZDV plus 3TC in previously untreated patients. *AIDS.* 1996;10(9):975-981.

35. Violin M, Cozzi-Lepri A, Velleca R, et al. Risk of failure in patients with 215 HIV-1 revertants starting their first thymidine analog-containing highly active antiretroviral therapy. *AIDS.* 2004;18(2):227-235.

36. Chappey C, Wrin T, Deeks S, Petropoulos CJ. Evolution of amino acid 215 in HIV-1 reverse transcriptase in response to intermittent drug selection. *Antivir Ther.* 2003;8:S37.

37. Garcia-Lerma JG, MacInnes H, Bennett D, Weinstock H, Heneine W. Transmitted human immunodeficiency virus type 1 carrying the D67N or K219Q/E mutation evolves rapidly to zidovudine resistance in vitro and shows a high replicative fitness in the presence of zidovudine. *J Virol.* 2004;78(14):7545-7552.

38. Marcelin AG, Flandre P, Pavie J, et al. Clinically relevant genotype interpretation of resistance to didanosine. *Antimicrob Agents Chemother*. 2005;49(5):1739-1744.

39. Molina JM, Marcelin AG, Pavie J, et al. Didanosine in HIV-1-infected patients experiencing failure of antiretroviral therapy: a randomized placebo-controlled trial. *J Infect Dis.* 2005;191(6):840-847.

40. Antinori A, Zaccarelli M, Cingolani A, et al. Cross-resistance among nonnucleoside reverse transcriptase inhibitors limits recycling efavirenz after nevirapine failure. *AIDS Res Hum Retroviruses*. 2002;18(12):835-838.

41. Feng M, Wang D, Grobler JA, Hazuda DJ, Miller MD, Lai MT. In vitro resistance selection with doravirine (MK-1439), a novel nonnucleoside reverse transcriptase inhibitor with distinct mutation development pathways. *Antimicrob Agents Chemother*. 2015;59(1):590-598.

42. Martin EA, Lai MT, Ngo W, et al. Review of doravirine resistance patterns identified in participants during clinical development. *J Acquir Immune Defic Syndr.* 2020;85(5):635-642.

43. Lai MT, Feng M, Falgueyret JP, et al. In vitro characterization of MK-1439, a novel HIV-1 nonnucleoside reverse transcriptase inhibitor. *Antimicrob Agents Chemother*. 2014;58(3):1652-1663.

44. Orkin C, Squires KE, Molina JM, et al. Doravirine/lamivudine/tenofovir disoproxil fumarate is noninferior to efavirenz/emtricitabine/tenofovir disoproxil fumarate in treatment-naive adults with human immunodeficiency virus-1 infection: week 48 results of the DRIVE-AHEAD trial. *Clin Infect Dis*. 2019;68(4):535-544.

45. Molina JM, Squires K, Sax PE, et al. Doravirine versus ritonavir-boosted darunavir in antiretroviral-naive adults with HIV-1 (DRIVE-FORWARD): 48-week results of a randomised, double-blind, phase 3, non-inferiority trial. *Lancet HIV.* 2018;5(5):e211-e220.

46. Smith SJ, Pauly GT, Akram A, et al. Rilpivirine and doravirine have complementary efficacies against NNRTI-resistant HIV-1 mutants. *JAIDS*. 2016;72(5):485-491.

47. Benhamida J, Chappey C, Coakley E, Parkin NT. HIV-1 genotype algorithms for prediction of etravirine susceptibility: novel mutations and weighting factors identified through correlations to phenotype. *Antivir Ther.* 2008;13(Suppl 3):A142.

48. Coakley E, Chappey C, Benhamida J, et al. Biological and clinical cut-off analyses for etravirine in the PhenoSense HIV assay. *Antivir Ther*. 2008;13(Suppl 3):A134.

49. Vingerhoets J, Tambuyzer L, Azijn H, et al. Resistance profile of

etravirine: combined analysis of baseline genotypic and phenotypic data from the randomized, controlled Phase III clinical studies. *AIDS.* 2010;24(4):503-514.

50. Haddad M, Stawiski E, Benhamida J, Coakley E. Improved genotypic algorithm for predicting etravirine susceptibility: comprehensive list of mutations identified through correlation with matched phenotype. Poster presented at: 17th Conference on Retroviruses and Opportunistic Infections (CROI); February 16-19, 2010; San Francisco, CA.

51. Janssen Therapeutics. Etravirine [prescribing information]. 2013. Titusville, NJ, Janssen Therapeutics.

52. Scherrer AU, Hasse B, Von Wyl V, et al. Prevalence of etravirine mutations and impact on response to treatment in routine clinical care: the Swiss HIV Cohort Study (SHCS). *HIV Med*. 2009;10(10):647-656.

53. Tambuyzer L, Nijs S, Daems B, Picchio G, Vingerhoets J. Effect of mutations at position E138 in HIV-1 reverse transcriptase on phenotypic susceptibility and virologic response to etravirine. *JAIDS*. 2011; 58(1):18-22.

54. Tudor-Williams G, Cahn P, Chokephaibulkit K, et al. Etravirine in treatment-experienced, HIV-1-infected children and adolescents: 48-week safety, efficacy and resistance analysis of the phase II PIANO study. *HIV Med.* 2014;15(9): 513-524.

55. Janseen Therapeutics. Rilpivirine [prescribing information]. 2015. Titusville, NJ, Janseen Therapeutics.

56. Azijn H, Tirry I, Vingerhoets J, et al. TMC278, a next-generation nonnucleoside reverse transcriptase inhibitor (NNRTI), active against wild-type and NNRTI-resistant HIV-1. *Antimicrob Agents Chemother.* 2010;54(2):718-727.

57. Picchio GR, Rimsky LT, Van Eygen V, et al. Prevalence in the USA of rilpivirine resistance-associated mutations in clinical samples and effects on phenotypic susceptibility to rilpivirine and etravirine. *Antivir Ther.* 2014;19(8):819-823.

58. Cohen CJ, Andrade-Villanueva J, Clotet B, et al. Rilpivirine versus efavirenz with two background nucleoside or nucleotide reverse transcriptase inhibitors in treatment-naive adults infected with HIV-1 (THRIVE): a phase 3, randomised, non-inferiority trial. *Lancet*. 2011;378(9787):229-237.

59. Molina JM, Cahn P, Grinsztejn B, et al. Rilpivirine versus efavirenz with tenofovir and emtricitabine in treatment-naive adults infected with HIV-1 (ECHO): a phase 3 randomised double-blind active-controlled trial. *Lancet.* 2011;378(9787):238-246.

60. Rimsky L, Vingerhoets J, Van Eygen V, et al. Genotypic and phenotypic characterization of HIV-1 isolates obtained from patients on rilpivirine therapy experiencing virologic failure in the phase 3 ECHO and THRIVE studies: 48-week analysis. *JAIDS*. 2012;59(1): 39-46.

61. Hofstra LM, Sauvageot N, Albert J, et al. Transmission of HIV drug resistance and the predicted effect on current first-line regimens in Europe. *Clin Infect Dis.* 2016; 62(5):655-663.

62. Kulkarni R, Babaoglu K, Lansdon EB, et al. The HIV-1 reverse transcriptase M184I mutation enhances the E138K-associated resistance to rilpivirine and decreases viral fitness. *JAIDS*. 2012;59(1): 47-54.

63. Hu Z, Kuritzkes DR. Interaction of reverse transcriptase (RT) mutations conferring resistance to lamivudine and etravirine: effects on fitness and RT activity of human immunodeficiency virus type 1. *J Virol*. 2011;85(21):11309-11314.

64. Xu HT, Asahchop EL, Oliveira M, et al. Compensation by the E138K mutation in HIV-1 reverse transcriptase for deficits in viral replication capacity and enzyme processivity associated with the M184I/V mutations. *J Virol*. 2011; 85(21):11300-11308.

65. Haddad M, Napolitano LA, Frantzell A, et al. Combinations of HIV-1 reverse transcriptase mutations L100I + K103N/S and L100I + K103R + V179D reduce susceptibility to rilpivirine. Poster presented at: 53rd Interscience Conference on Antimicrobial Agents and Chemotherapy (ICAAC); September 10-13, 2013; Denver, CO.

66. Hirsch MS, Günthard HF, Schapiro JM, et al. Antiretroviral drug resistance testing in adult HIV-1 infection: 2008 recommendations of an International AIDS Society-USA panel. *Clin Infect Dis*. 2008; 47(2):266-285.

67. Fun A, Wensing AM, Verheyen J, Nijhuis M. Human immunodeficiency virus gag and protease: partners in resistance. *Retrovirology.* 2012;9:63.

68. Young TP, Parkin NT, Stawiski E, et al. Prevalence, mutation patterns, and effects on protease inhibitor susceptibility of the L76V mutation in HIV-1 protease. *Antimicrob Agents Chemother*. 2010; 54(11):4903-4906.

69. De Meyer S, Descamps D, Van Baelen B, et al. Confirmation of the negative impact of protease mutations I47V, I54M, T74P and I84V and the positive impact of protease mutation V82A on virological response to darunavir/ritonavir. *Antivir Ther*. 2009;14(Suppl 1):A147.

70. Descamps D, Lambert-Niclot S, Marcelin AG, et al. Mutations associated with virological response to darunavir/ritonavir in HIV-1-infected protease inhibitor-experienced patients. *J Antimicrob Chemother*. 2009;63(3):585-592.

71. Janssen Therapeutics. Darunavir [prescribing information]. 2015. Titusville, NJ, Janssen Therapeutics.

72. Masquelier B, Breilh D, Neau D, et al. Human immunodeficiency virus type 1 genotypic and pharmacokinetic determinants of the virological response to lopinavir-ritonavir-containing therapy in protease inhibitor-experienced patients. *Antimicrob Agents Chemother.* 2002;46(9):2926-2932.

73. Kempf DJ, Isaacson JD, King MS, et al. Identification of genotypic changes in human immunodeficiency virus protease that correlate with reduced susceptibility to the protease inhibitor lopinavir among viral isolates from protease

inhibitor-experienced patients. *J Virol*. 2001;75(16):7462-7469.

74. AbbVie Inc. Lopinavir/ritonavir [prescribing information]. 2015. Abbott Park, IL, AbbVie Inc.

75. Mo H, King MS, King K, Molla A, Brun S, Kempf DJ. Selection of resistance in protease inhibitor-experienced, human immunodeficiency virus type 1-infected subjects failing lopinavir- and ritonavir-based therapy: mutation patterns and baseline correlates. *J Virol*. 2005;79(6):3329-3338.

76. Kagan RM, Shenderovich M, Heseltine PN, Ramnarayan K. Structural analysis of an HIV-1 protease I47A mutant resistant to the protease inhibitor lopinavir. *Protein Sci*. 2005;14(7):1870-1878.

77. AbbVie Inc. KALETRA (lopinavir and ritonavir) [prescribing information]. 2018. Abbott Park, IL, AbbVie Inc.

78. Friend J, Parkin N, Liegler T, Martin JN, Deeks SG. Isolated lopinavir resistance after virological rebound of a ritonavir/lopinavir-based regimen. *AIDS*. 2004;18(14):1965-1966.

79. Lam E, Parkin NT. Amprenavir resistance imparted by the I50V mutation in HIV-1 protease can be suppressed by the N88S mutation. *Clin Infect Dis*. 2003;37(9):1273-1274.

80. Rhee SY, Taylor J, Fessel WJ, et al. HIV-1 protease mutations and protease inhibitor cross-resistance. *Antimicrob Agents Chemother*. 2010;54(10):4253-4261.

81. Hermans LE, Steegen K, ter Heine R, et al. PI drug-level testing as screening tool for drug resistance in 2nd-line ART failure. *Top Antivir Med*. 2019;27(1s):169s.

82. Gonzalez LM, Brindeiro RM, Aguiar RS, et al. Impact of nelfinavir resistance mutations on in vitro phenotype, fitness, and replication capacity of human immunodeficiency virus type 1 with subtype B and C proteases. *Antimicrob Agents Chemother*. 2004;48(9):3552-3555.

83. Reeves JD, Gallo SA, Ahmad N, et al. Sensitivity of HIV-1 to entry inhibitors correlates with envelope/coreceptor affinity, receptor density, and fusion kinetics. *Proc Natl Acad Sci USA*. 2002;99(25):16249-16254.

84. Reeves JD, Miamidian JL, Biscone MJ, et al. Impact of mutations in the coreceptor binding site on human immunodeficiency virus type 1 fusion, infection, and entry inhibitor sensitivity. *J Virol*. 2004;78(10):5476-5485.

85. Xu L, Pozniak A, Wildfire A, et al. Emergence and evolution of enfuvirtide resistance following long-term therapy involves heptad repeat 2 mutations within gp41. *Antimicrob Agents Chemother*. 2005;49(3):1113-1119.

86. ViiV Healthcare. Maraviroc [prescribing information]. 2015. Research Triangle Park, NC, ViiV Healthcare.

87. Anastassopoulou CG, Ketas TJ, Sanders RW, Klasse PJ, Moore JP. Effects of sequence changes in the HIV-1 gp41 fusion peptide on CCR5 inhibitor resistance. *Virology*. 2012;428(2):86-97.

88. Saag MS, Gandhi RT, Hoy JF, et al. Antiretroviral drugs for treatment and prevention of HIV infection in adults: 2020 recommendations of the International Antiviral Society-USA Panel. *JAMA*. 2020;324(16):1651-1669.

89. Oliveira M, Ibanescu RI, Anstett K, et al. Selective resistance profiles emerging in patient-derived clinical isolates with cabotegravir, bictegravir, dolutegravir, and elvitegravir. *Retrovirology*. 2018;15(1):56.

90. Tsiang M, Jones GS, Goldsmith J, et al. Antiviral activity of bictegravir (GS-9883), a novel potent HIV-1 integrase strand transfer inhibitor with an improved resistance profile. *Antimicrob Agents Chemother*. 2016;60(12):7086-7097.

91. Smith SJ, Zhao XZ, Burke TR, Jr., Hughes SH. Efficacies of cabotegravir and bictegravir against drug-resistant HIV-1 integrase mutants. *Retrovirology*. 2018;15(1):37.

92. Rhee SY, Grant PM, Tzou PL, et al. A systematic review of the genetic mechanisms of dolutegravir resistance. *J Antimicrob Chemother*. 2019;74(11):3135-3149.

93. Castagna A, Maggiolo F, Penco G, et al. Dolutegravir in antiretroviral-experienced patients with raltegravir- and/or elvitegravir-resistant HIV-1: 24-week results of the phase III VIKING-3 Study. *J Infect Dis*. 2014;210(3):354-362.

94. Chamberlain N, Mena L, Brock JB. Case report: emergent resistance in a treatment-naive person with human immunodeficiency virus under bictegravir-based therapy. *Open Forum Infect Dis*. 2021;8(6):ofab297.

95. Overton ET, Richmond G, Rizzardini G, et al. Long-acting cabotegravir and rilpivirine dosed every 2 months in adults with HIV-1 infection (ATLAS-2M), 48-week results: a randomised, multicentre, open-label, phase 3b, non-inferiority study. *Lancet*. 2021;396(10267):1994-2005.

96. Marzinke MA, Grinsztejn B, Fogel JM, et al. Characterization of Human Immunodeficiency Virus (HIV) Infection in Cisgender Men and Transgender Women Who Have Sex With Men Receiving Injectable Cabotegravir for HIV Prevention: HPTN 083. *J Infect Dis*. 2021;224(9):1581-1592.

97. Overton ET, Richmond G, Rizzardini G, et al. Long-acting cabotegravir + rilpivirine every 2 months: ATLAS-2M week 152 results [CROI Abstract 479]. In Special Issue: Abstracts From the 2022 Conference on Retroviruses and Opportunistic Infections. *Top Antivir Med*. 2022;30(1s):183.

98. Cutrell AG, Schapiro JM, Perno CF, et al. Exploring predictors of HIV-1 virologic failure to long-acting cabotegravir and rilpivirine: a multivariable analysis. *AIDS*. 2021;35(9):1333-1342.

99. Hu Z, Cordwell T, Jeffrey J, Kuritzkes D. Effect of L74I polymorphism on fitness of IHV-1 subtype A6 resistant to cabotegravir [CROI Abstract 506]. In Special Issue: Abstracts From the 2022 Conference on Retroviruses and Opportunistic Infections. *Top Antivir Med*. 2022;30(1s):193-194.

100. Cheung PK, Shahid A, Dong W, of clinically observed HIV integrase mutations on phenotypic resistance to integrase strand

transfer inhibitors (INSTIs): a molecular study. *J Antimicrob Chemother*. 2022;77(4):979-988.

101. Frantzell A, Petropoulos C, Huang W. Dolutegravir resistance requires multiple primary mutations in HIV-1 integrase. *Top Antivir Med.* 2015; 23(e-1):51.

102. Kobayashi M, Yoshinaga T, Seki T, et al. In vitro antiretroviral properties of S/GSK1349572, a next-generation HIV integrase inhibitor. *Antimicrob Agents Chemother.* 2011;55(2):813-821.

103. Raffi F, Rachlis A, Stellbrink HJ, et al. Once-daily dolutegravir versus raltegravir in antiretroviral-naive adults with HIV-1 infection: 48 week results from the randomised, double-blind, non-inferiority SPRING-2 study. *Lancet.* 2013;381(9868): 735-743.

104. Eron JJ, Clotet B, Durant J, et al. Safety and efficacy of dolutegravir in treatment-experienced subjects with raltegravir-resistant HIV type 1 infection: 24-week results of the VIKING Study. *J Infect Dis*. 2013;207(5):740-748.

105. Seki T, Suyama-Kagitani A, Kawauchi-Miki S, et al. Effects of raltegravir or elvitegravir resistance signature mutations on the barrier to dolutegravir resistance in vitro. *Antimicrob Agents Chemother*. 2015;59(5): 2596-2606.

106. DeAnda F, Hightower KE, Nolte RT, et al. Dolutegravir interactions with HIV-1 integrase-DNA: structural rationale for drug resistance and dissociation kinetics. *PLoS One*. 2013;8(10):e77448.

107. Goodman D, Hluhanich R, Waters J, et al. Integrase inhibitor resistance involves complex interactions among primary and second resistance mutations: a novel mutation L68V/I associates with E92Q and increases resistance. *Antivir Ther*. 2008; 13(Suppl 3):A15.

108. Waters J, Margot N, Hluhanich R, et al. Evolution of resistance to the HIV integrase inhibitor (INI) elvitegravir can involve genotypic switching among primary INI resistance patterns. Fort Myers, FL. *Antivir Ther*. 2009; 14(Supp 1):A137.

109. Doyle T, Dunn DT, Ceccherini-Silberstein F, et al. Integrase inhibitor (INI) genotypic resistance in treatment-naive and raltegravir-experienced patients infected with diverse HIV-1 clades. *J Antimicrob Chemother*. 2015; 70(11):3080-3086.

110. Sax PE, DeJesus E, Mills A, et al. Co-formulated elvitegravir, cobicistat, emtricitabine, and tenofovir versus co-formulated efavirenz, emtricitabine, and tenofovir for initial treatment of HIV-1 infection: a randomised, double-blind, phase 3 trial, analysis of results after 48 weeks. *Lancet.* 2012;379(9835): 2439-2448.

111. DeJesus E, Rockstroh J, Henry K, et al. Co-formulated elvitegravir, cobicistat, emtricitabine, and tenofovir disoproxil fumarate versus ritonavir-boosted atazanavir plus co-formulated emtricitabine and tenofovir disoproxil fumarate for initial treatment of HIV-1 infection: a randomised, double-blind, phase 3, non-inferiority trial. *Lancet*. 2012;379(9835):2429-2438.

112. Abram ME, Hluhanich RM, Goodman DD, et al. Impact of primary elvitegravir resistance-associated mutations in HIV-1 integrase on drug susceptibility and viral replication fitness. *Antimicrob Agents Chemother.* 2013;57(6):2654-2663.

113. White K, Kulkarni R, Miller MD. Analysis of early resistance development at the first failure timepoint in elvitegravir/cobicistat/emtricitabine/tenofovir disoproxil fumarate-treated patients. *J Antimicrob Chemother.* 2015;70(9):2632-2638.

114. Scherrer AU, Yang WL, Kouyos RD, et al. Successful prevention of transmission of integrase resistance in the Swiss HIV Cohort Study. *J Infect Dis*. 2016;214(3):399-402.

115. Hazuda DF, Miller MD, Nguyen BY, Zhao J, for the P005 Study Team. Resistance to the HIV-integrase inhibitor raltegravir: analysis of protocol 005, a phase II study in patients with triple-class resistant HIV-1 infection. *Antivir Ther*. 2007;12:S10.

116. Gatell JM, Katlama C, Grinsztejn B, et al. Long-term efficacy and safety of the HIV integrase inhibitor raltegravir in patients with limited treatment options in a Phase II study. *JAIDS*. 2010; 53(4):456-463.

117. Fransen S, Gupta S, Danovich R, et al. Loss of raltegravir susceptibility by human immunodeficiency virus type 1 is conferred via multiple nonoverlapping genetic pathways. *J Virol.* 2009;83(22):11440-11446.

118. Hatano H, Lampiris H, Fransen S, et al. Evolution of integrase resistance during failure of integrase inhibitor-based antiretroviral therapy. *J Acquir Immune Defic Syndr.* 2010;54(4): 389-393.

119. Wittkop L, Breilh D, Da Silva D, et al. Virological and immunological response in HIV-1-infected patients with multiple treatment failures receiving raltegravir and optimized background therapy, ANRS CO3 Aquitaine Cohort. *J Antimicrob Chemother.* 2009; 63(6):1251-1255.

120. Armenia D, Vandenbroucke I, Fabeni L, et al. Study of genotypic and phenotypic HIV-1 dynamics of integrase mutations during raltegravir treatment: a refined analysis by ultra-deep 454 pyrosequencing. *J Infect Dis.* 2012;205(4):557-567.

121. Cooper DA, Steigbigel RT, Gatell JM, et al. Subgroup and resistance analyses of raltegravir for resistant HIV-1 infection. *N Engl J Med*. 2008;359(4):355-365.

122. Malet I, Delelis O, Valantin MA, et al. Mutations associated with failure of raltegravir treatment affect integrase sensitivity to the inhibitor in vitro. *Antimicrob Agents Chemother.* 2008;52(4):1351-1358.

123. Blanco JL, Varghese V, Rhee SY, Gatell JM, Shafer RW. HIV-1 integrase inhibitor resistance and its clinical implications. *J Infect Dis.* 2011;203(9):1204-1214.

124. Link JO, Rhee MS, Tse WC, et al. Clinical targeting of HIV

capsid protein with a long-acting small molecule. *Nature.* 2020;584(7822):614-618.

125. Margot N, VanderVeen L, Naik V, et al. Phenotypic resistance to lenacapavir and monotherapy efficacy in a proof-of-concept clinical study. *J Antimicrob Chemother.* 2022;77(4):989-995.

126. Callebaut CVL, Margot N, Naik V, Rhee M. Activity and resistance characterization of the HIV CAPSID inhibitor lenacapavir [CROI Abstract 128]. In Special Issue: Abstracts From the 2021 Conference on Retroviruses and Opportunistic Infections. *Top Antivir Med*. 2021;29(1):35.

127. Marcelin AG, Charpentier C, Jary A, et al. Frequency of capsid substitutions associated with GS-6207 in vitro resistance in HIV-1 from antiretroviral-naive and -experienced patients. *J Antimicrob Chemother*. 2020; 75(6):1588-1590.

128. Margot N, VaderVeen L, Naik V, et al. Resistance analysis of long-acting lenacapavir in highly treatment-experienced people with HIV after 26 weeks of treatment. Poster presented at: 18th European AIDS Conference (EACS); October 27-30, 2021; London, UK.

129. Ogbuagu O, Segal-Maurer S, Brinson C, et al. Long-acting lenacapavir in people with multidrug resistant HIV-1: week 52 results [CROI Abstract 491]. In Special Issue: Abstracts From the 2022 Conference on Retroviruses and Opportunistic Infections. *Top Antivir Med*. 2022; 30(1s):188.

130. VanderVeen L, Margot N, Naik V, et al. Interim-resistance analysis of long-acting lenacapavir in treatment-naive people with HIV at 28 weeks (CALIBRATE). Poster presented at: IDWeek 2021 Virtual Conference; September 29-October 3, 2021.

131. Gupta S, Sims J, Brinson C, et al. Lenacapavir as part of a combination regimen in treatment naive PWH: week 54 results [CROI Abstract 138]. In Special Issue: Abstracts From the 2021 Conference on Retroviruses and Opportunistic Infections. *Top HIV Med.* 2022;30(1s):53.

Invited Review

Approaching Monkeypox: A Guide for Clinicians

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The 2022 outbreak of monkeypox virus infection has expanded far beyond regions in which the disease was previously endemic. Monkeypox has a wide range of manifestations, some of which are unique to this outbreak. Novel clinical presentations, testing limitations, and a lack of available treatments have contributed to delays in recognition, diagnosis, and treatment of monkeypox. As health care workers and governments fight this rare viral infection, which may become a routine diagnosis, early recognition of potential signs and symptoms along with appropriate testing is essential to prevent continuing spread and potential endemicity.

Keywords: monkeypox, MPX, lesions, tecovirimat, Tpoxx, JYNNEOS, proctitis

Many parts of the world are currently experiencing an outbreak of monkeypox virus infection, with more than 100 countries reporting cases.[1] Monkeypox is a rare viral infection that can cause fever, chills, lymphadenopathy, fatigue, myalgia, cough, and, characteristically, skin lesions or rash.[1,2] Patients may present with prodromal symptoms, rash, or lesions, or sometimes unexpected manifestations such as proctitis, urethritis, or conjunctivitis (Figure 1).[3,4] The skin lesions seen in monkeypox can vary widely in appearance and may include papules, pustules, umbilicated lesions, ulcers, or morbilliform rash (Figures 2, 3, 4, and 5). Classically, most lesions are deep-seated and well circumscribed and progress through a series of stages.[1,2] Lesions or rash can appear anywhere on the body, with predominance in the genital and anorectal areas in the current outbreak.[5,6] Interestingly, monkeypox virus has been detected on rectal swabs of at-risk but asymptomatic individuals despite the absence of skin lesions, raising concern about the possibility of asymptomatic transmission of the virus.[7]

The evolving definition of suspected monkeypox according to the Centers for Disease Control and Prevention (CDC) is a rash consistent with monkeypox and a high degree of clinical suspicion in patients who, within 21 days of symptom onset, had contact with a suspected or known case; close contact with men who have sex with men, especially if contact was made via dating or hookup websites, apps, or parties; or traveled to an area where there are confirmed cases or the disease is endemic. Given that tens of thousands of cases have been identified across the United States, having traveled abroad is no longer an essential clue to diagnosis.

Testing for monkeypox virus should be ordered for any suspected case. Although testing for the presence of monkeypox virus can be performed with cerebrospinal fluid, urine, blood, breast milk, or genital swabs in the absence of lesions, the only type of testing that has been cleared by the US Food and Drug Administration (FDA) to date is lesion based because of insufficient clinical data to support other sample types and the risk of inaccurate results.[8] This means that, under current recommendations, if a patient presents with prodromal symptoms but without rash or skin changes, they should be

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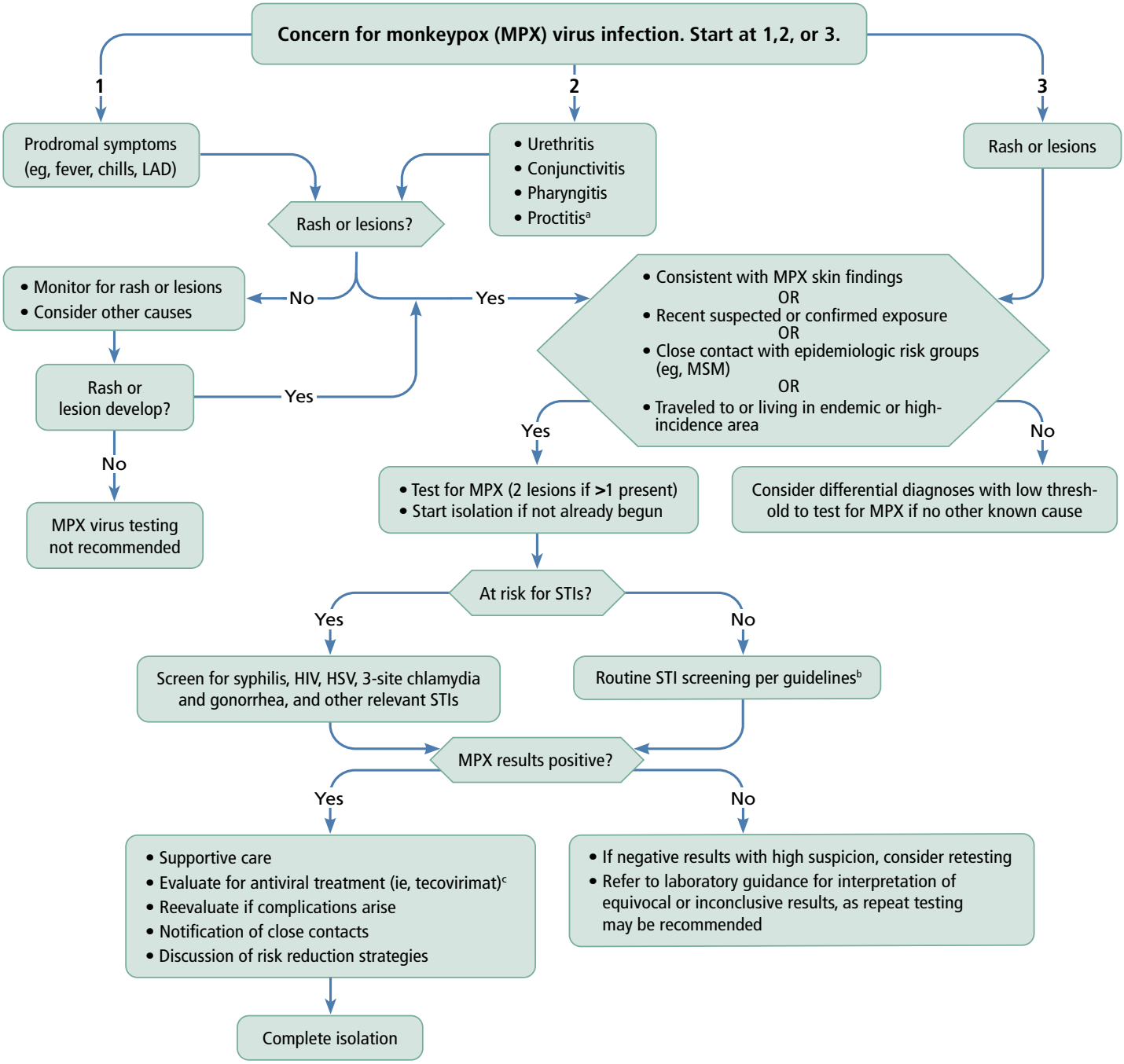

Figure 1. Flowchart for an approach to monkeypox virus infection.

Abbreviations: HSV, herpes simplex virus; LAD, lymphadenopathy; MSM, men who have sex with men; STI, sexually transmitted infection.

[a]Some presentations of MPX can start without prodromal symptoms or visible skin changes. As current testing recommends sampling of skin lesions, if a patient presents with symptoms such as urethritis, conjunctivitis, pharyngitis, or particularly proctitis, perform careful evaluation for lesions at areas such as the urinary meatus, inner eyelids, mouth, pharynx, and perirectal region. If no lesions are found, in cases of proctitis consider a rectal mucosal swab to test for monkeypox virus if testing is available.

[b]Refer to Centers for Disease Control and Prevention STI Treatment Guidelines, 2021.

[c]Refer to Centers for Disease Control and Prevention guidance for antiviral eligibility criteria.

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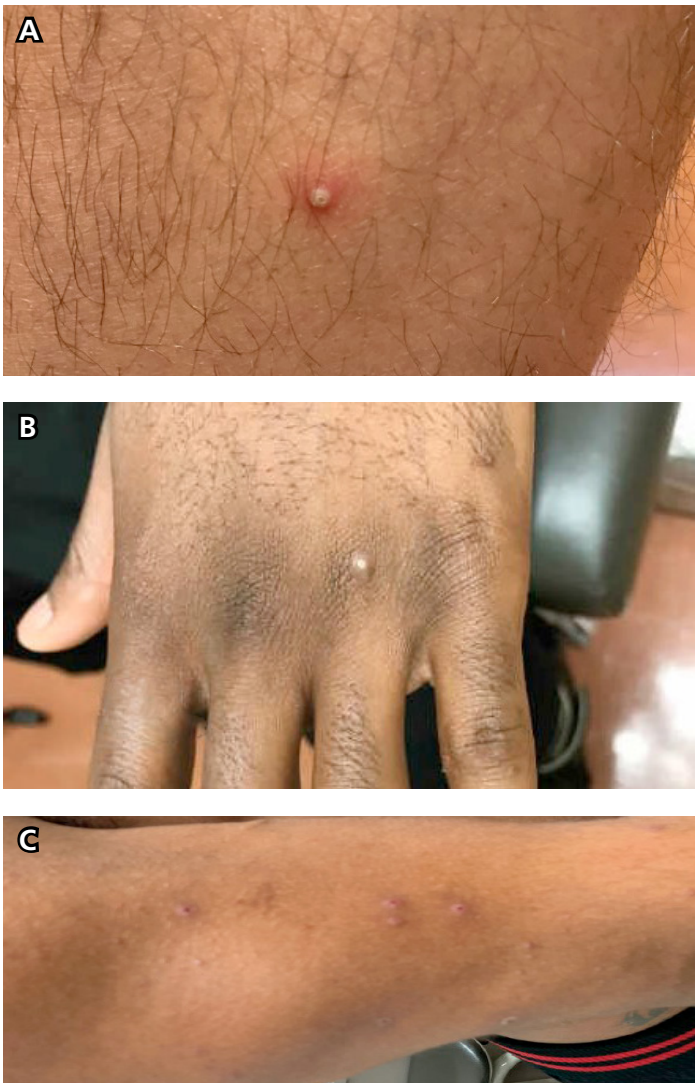

Figure 2. A and B: Pustule-like lesions on a limb and dorsal hand. C: Cluster of papules, some with umbilication.

monitored for the development of lesions. If none appear within approximately 5 days, they are unlikely to be infected with monkeypox, and testing is not recommended according to current CDC guidelines.[1] Similarly, if a patient presents with a manifestation such as proctitis, urethritis, conjunctivitis, or pharyngitis, testing is recommended if they have lesions. A thorough physical examination should be performed in these cases to identify any nonapparent lesions, which may be occult in the inner eyelids, mouth, pharynx, perirectal region, urinary meatus, or other areas, depending on the location of symptoms. An anoscopy to look for deeper lesions in the

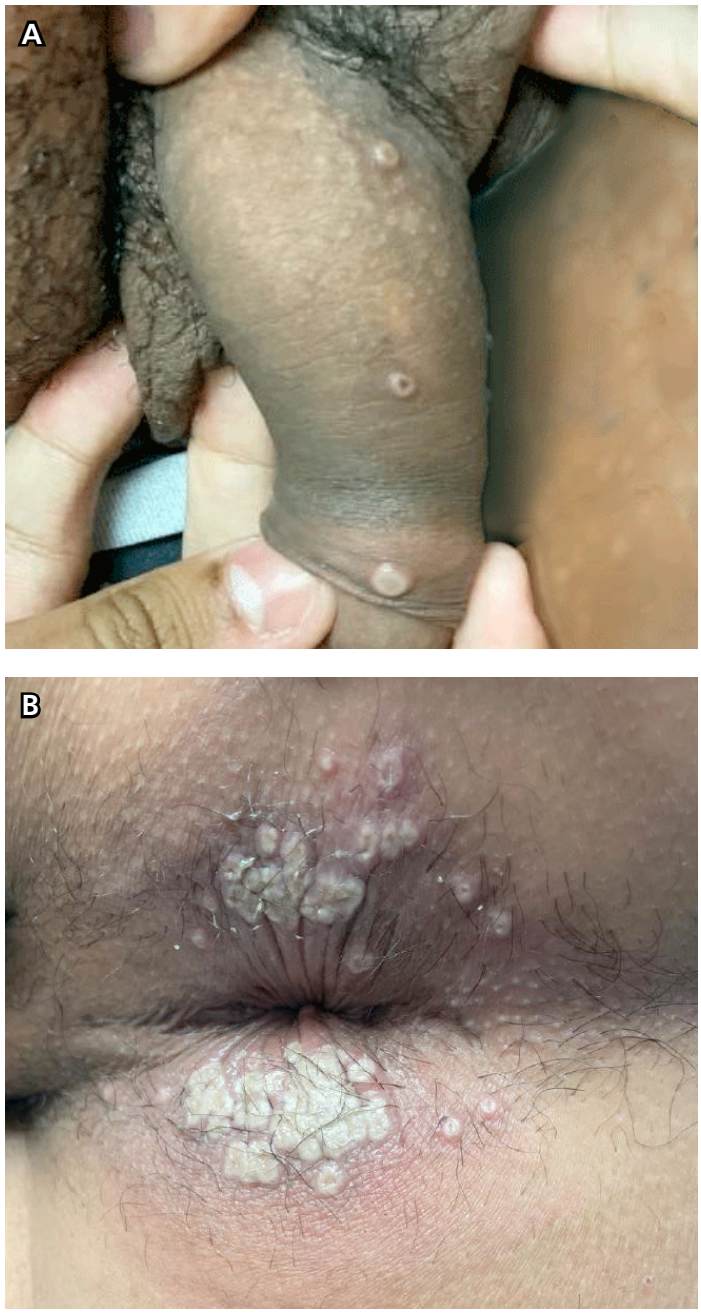

Figure 3. A: Umbilicated lesion on penis. B: Cluster of papules in the perianal region, many with umbilication.

anal mucosa can be considered if a patient presents with proctitis without visible lesions in the perianal region.[4] If no lesions are identified, these patients should be monitored for lesion development in accordance with testing guidance from the CDC. This guidance does have limitations in cases of occult lesions or delayed appearance of lesions, or if

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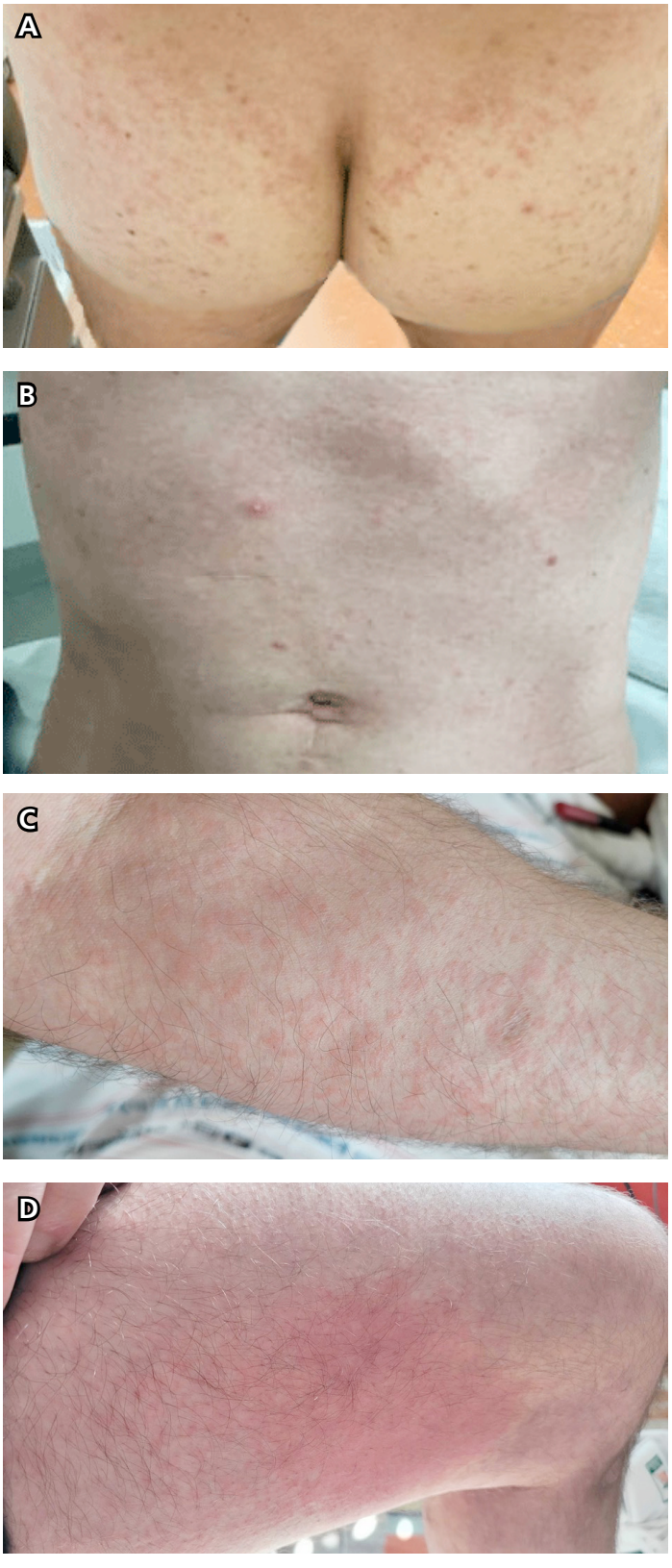

Figure 4. Morbilliform rash on buttocks (A), lower trunk (B), and forearm (C). D: Morbilliform rash on thigh with area of confluent rash.

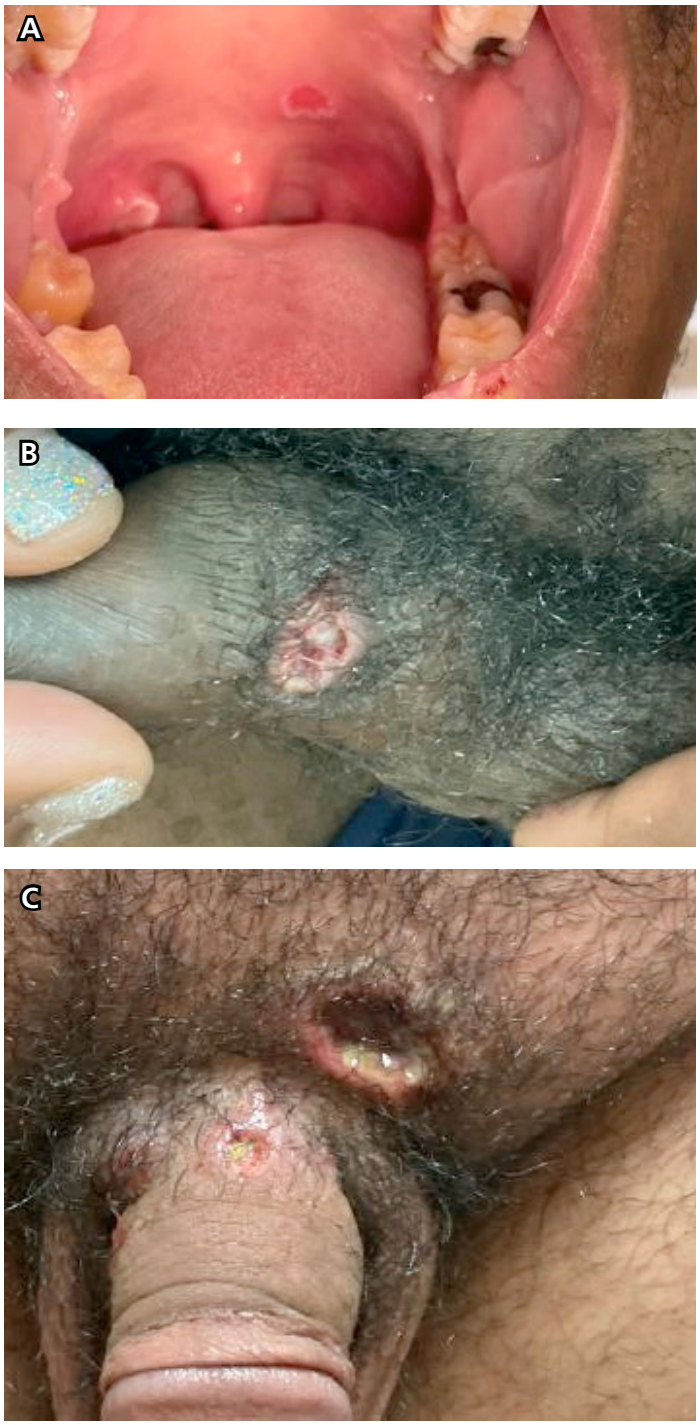

Figure 5. A: Ulcerations on palate and tonsil. B: Ulceration on penis. C: Ulcerations on penis and pubis.

asymptomatic transmission of the virus is confirmed. Therefore, while nonlesion monkeypox tests are not currently approved by the FDA, their use could be considered on a case-by-case basis, depending on

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provider suspicion. However, if a patient does not have major risk factors for monkeypox and their illness seems inconsistent with the disease, differential diagnoses to consider include syphilis, herpes simplex virus, primary varicella virus, varicella zoster virus, molluscum contagiosum, lymphogranuloma venereum, gonorrhea, enterovirus (including coxsackievirus) infection, and fungal infections.

Once a patient is suspected to have a monkeypox virus infection, they should begin isolation while test results are pending. Given high rates of concurrent sexually transmitted infections, it is recommended that patients who are at risk of such infections (ie, those with multiple sexual partners, anonymous partners, or new partners) also be screened for coinfections, which can include syphilis, HIV infection, herpes simplex virus infection, and triple-site pharyngeal, urinary, and rectal gonorrhea and chlamydia.[1,5]

In suspected or confirmed cases of monkeypox, the mainstay of treatment is symptomatic management. Therapy should target oral, genital, and anorectal pain; pruritus; fever; proctitis, if present; and keeping skin lesions clean. Treatment options include oral salt and anesthetic rinses, topical anesthetics, sitz or oatmeal baths, stool softeners, pain medication, oral or topical antihistamines, or other nonirritant gels or lotions such as calamine or menthol.[9] Some patients develop severe pain requiring multimodal oral pain management regimens that may include opioids. Most patients recover with symptom management alone, but those who are at high risk of severe disease, have involvement of anatomic areas that might result in serious sequelae from scarring or strictures, or experience complications should be evaluated for eligibility to receive the antiviral tecovirimat, currently available through a CDC-sponsored expanded access program or clinical trials.[10,11] Although tecovirimat may shorten the duration of illness, it is not approved by the FDA, its efficacy is unknown, and its low genetic barrier to resistance has raised concerns.[12]

Typical illness courses of monkeypox last, on average, 2 to 4 weeks.[1,2,6] Complications of this viral infection can include bacterial superinfection of lesions, gastroenteritis, bronchopneumonia, encephalomyelitis, sepsis, uncontrolled pain, spread of infection to eyes, scars, and strictures (Figure 6).[1,5,9] Any symptoms that raise concerns about complications should prompt reevaluation. If a

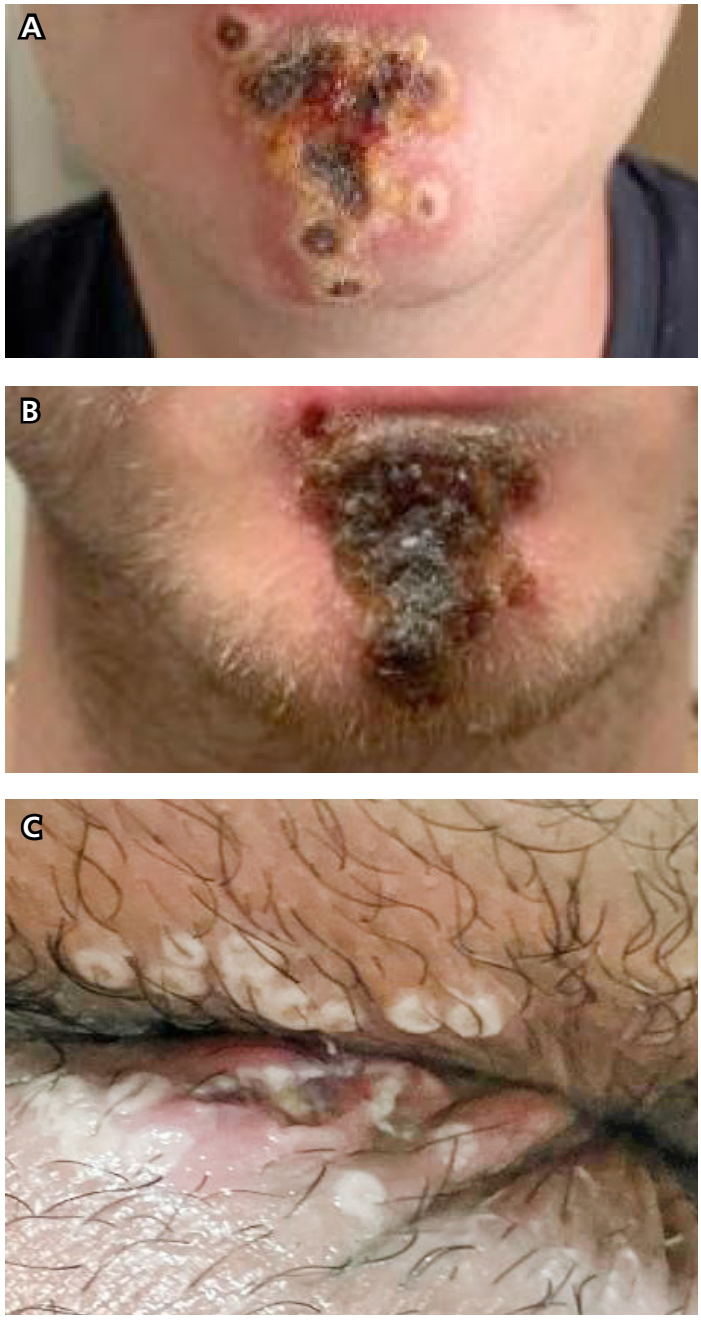

Figure 6. Complications of monkeypox. A: Secondary infection of chin lesions. B: Appearance of infection shown in panel A 5 days later. C: Fissure and ulceration in the perianal region surrounded by papules and umbilicated lesions.

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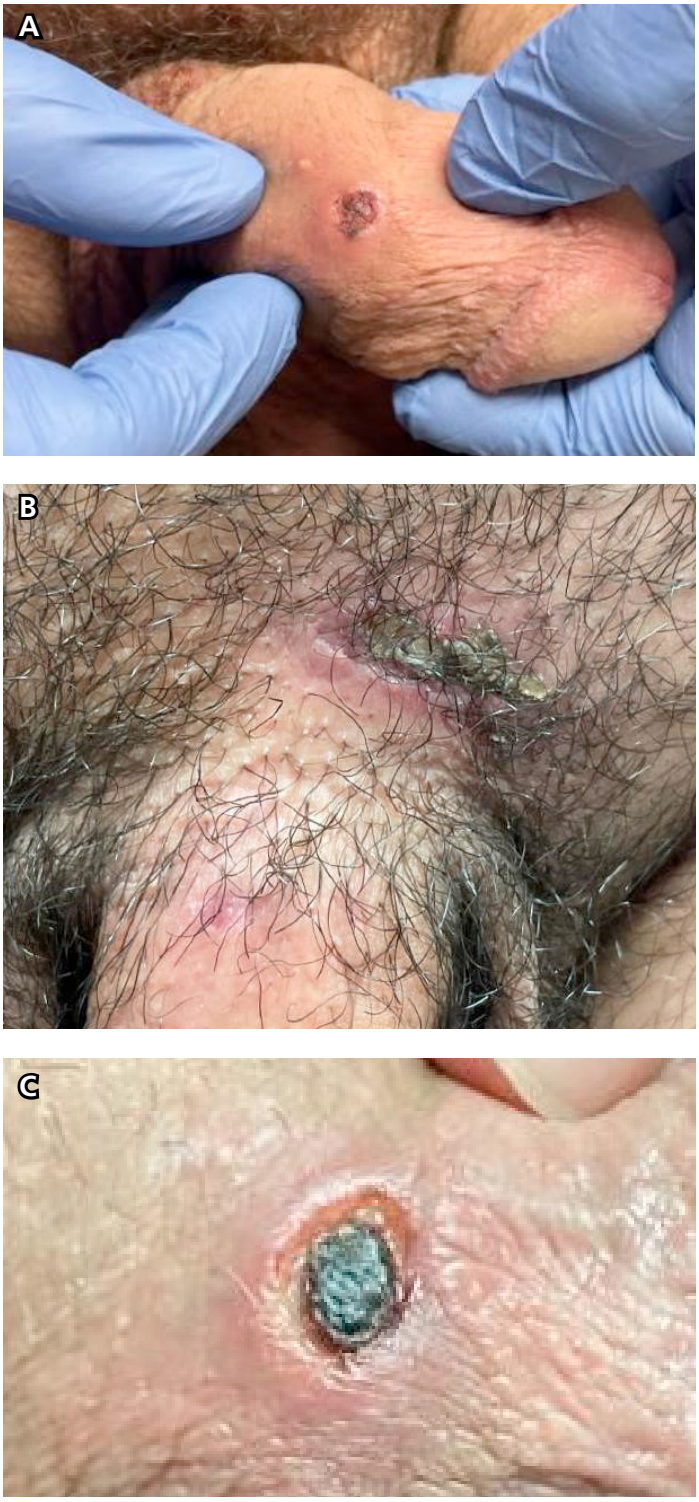

Figure 7. Scabbed lesions. A: On penis; note the varying stages of lesions present with adjacent pustule. B: Progression of ulcerated lesions shown in Figure 5C; note the reepithelization of the penile shaft lesion and scabbing of the pubic ulceration. C: Scabbed lesion.

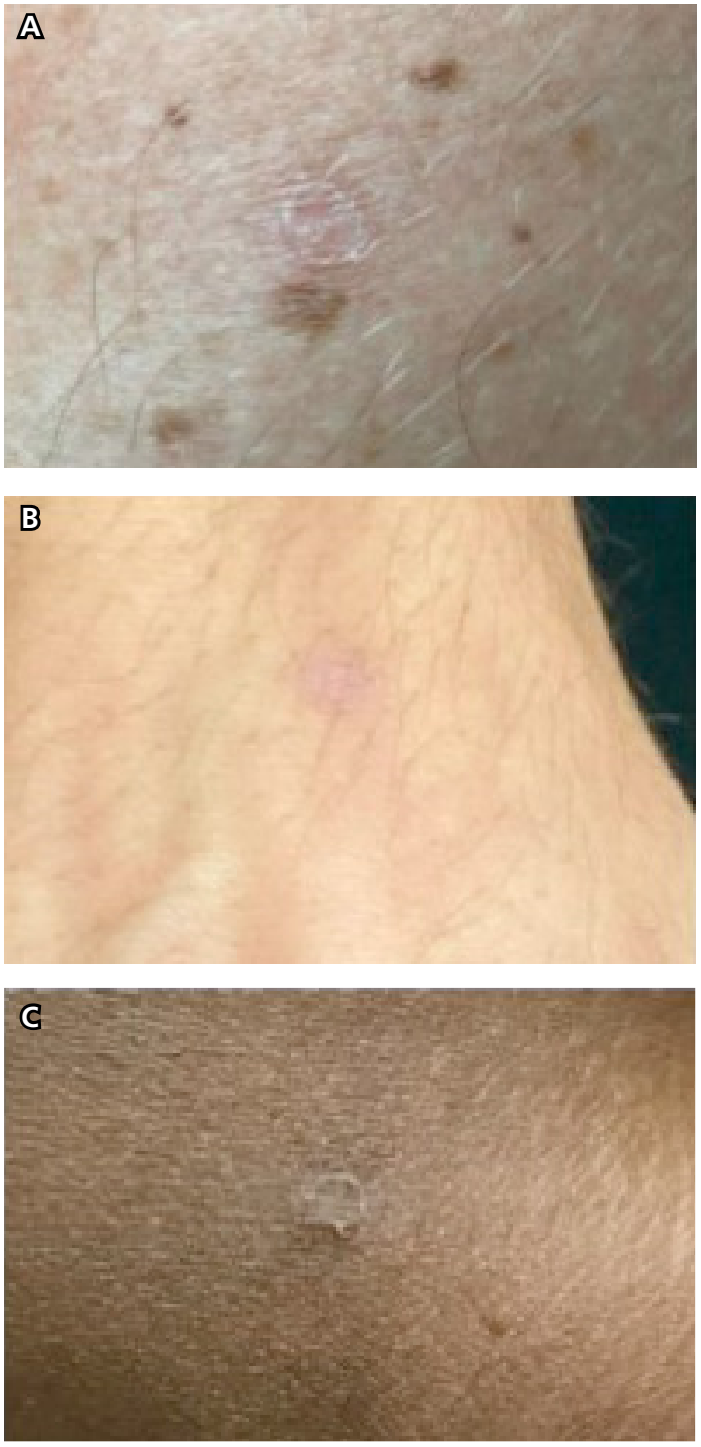

Figure 8. A, B, and C: Resolution of lesions.

patient's sample tests positive, they should inform recent close contacts so those individuals may monitor themselves for symptoms and be evaluated for postexposure prophylaxis, which in most cases is accomplished with prompt vaccination.

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Methods to prevent the spread of monkeypox include vaccination for those who are eligible as well as behavioral changes regarding sex practices to reduce infection risk. JYNNEOS, a live nonreplicating virus vaccine, is the primary vaccine in use in the United States during the current outbreak. Although the efficacy of JYNNEOS in preventing monkeypox virus infection in this outbreak is unknown, early data suggest that there is a lower incidence of infection among individuals with even 1 dose of the 2-dose vaccine series compared with unvaccinated individuals.[1,13] Other infection risk reduction strategies include discussing symptoms of monkeypox with partners, avoiding or delaying close or intimate contact with someone who may be symptomatic or has a pending monkeypox test, and avoiding sharing linens, sex toys, and personal grooming items.[1,9] Those infected with monkeypox should isolate until all skin lesions have scabbed (Figure 7) and fallen off, with the formation of new skin (Figure 8).

Given the many challenges that arose in the early part of this outbreak, some of which continue today, it is unlikely that this infection will be eradicated, which means that ongoing knowledge of and ability to manage monkeypox infections may become part of everyday clinical care.

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References

1. Monkeypox. Centers for Disease Control and Prevention. https://www.cdc.gov/poxvirus/monkeypox/. 2022. Accessed October 15, 2022.
2. Monkeypox. World Health Organization. https://www.who.int/news-room/fact-sheets/detail/monkeypox. 2022. Accessed October 15, 2022.
3. Benatti SV, Venturelli S, Comi N, Borghi F, Paolucci S, Baldanti F. Ophthalmic manifestation of monkeypox infection. *Lancet Infect Dis*. 2022;22(9):1397.
4. Basgoz N, Brown CM, Smole SC, et al. Case 24-2022: a 31-year-old man with perianal and penile ulcers, rectal pain, and rash. *N Engl J Med*. 2022;387(6):547-556.
5. Patrocinio-Jesus R, Peruzzu F. Monkeypox genital lesions. *N Engl J Med*. 2022;387(1):66.
6. Thornhill JP, Barkati S, Walmsley S, et al. Monkeypox virus infection in humans across 16 countries — April-June 2022. *N Engl J Med*. 2022;387(8):679-691.
7. De Baetselier I, Van Dijck C, Kenyon C, et al. Retrospective detection of asymptomatic monkeypox virus infections among male sexual health clinic attendees in Belgium. *Nat Med*. Published online August 12, 2022. doi:10.1038/s41591-022-02004-w
8. Monkeypox update: FDA takes significant action to help expand access to testing. https://www.fda.gov/news-events/press-announcements/monkeypox-update-fda-takes-significant-action-help-expand-access-testing. 2022. Accessed September 17, 2022.
9. Monkeypox: what to do when sick. NYC Health. https://www1.nyc.gov/site/doh/health/health-topics/monkeypox-when-sick.page. 2022. Accessed September 14, 2022.
10. O'Laughlin K, Tobolowsky FA, Elmor R, et al. Clinical use of tecovirimat (Tpoxx) for treatment of monkeypox under an investigational new drug protocol–United States, May-August 2022. *Morb Mortal Wkly Rep*. 2022;71(37):1190-1195.
11. Study of tecovirimat for human monkeypox virus (STOMP). ClinicalTrials.gov identifier: NCT05534984. https://clinicaltrials.gov/ct2/show/NCT05534984. September 10, 2022. Accessed September 20, 2022.
12. FDA Monkeypox response. U.S. Food and Drug Administration. https://www.fda.gov/emergency-preparedness-and-response/mcm-issues/fda-monkeypox-response#therapeutics. 2022. Accessed September 16, 2022.
13. Payne AB, Ray LC, Kugeler KJ, et al. Incidence of monkeypox among unvaccinated persons compared with persons receiving ≥1 JYNNEOS vaccine dose — 32 U.S. jurisdictions, July 31–September 3, 2022. *MMWR Morb Mortal Wkly Rep*. 2022;71(40):1278–1282.

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