EDITORIAL COMMENTARY

Timing, Adherence, Resistance, and . . . Persistence? New Insight Into the Mechanisms of Failure of HIV Type 1 Postexposure Prophylaxis

Gustavo H. Kijak[1,2] and Jerome H. Kim[1]

[1]US Military HIV Research Program, Walter Reed Army Institute of Research, Silver Spring; and [2]Henry M. Jackson Foundation for the Advancement of Military Medicine, Bethesda, Maryland

(See the brief report by Li et al on pages 1598–603.)

Keywords. HIV-1; postexposure prophylaxis; single-genome sequencing; viral sequestration.

Every day, hundreds of thousand healthcare workers (HCWs) worldwide are at risk of occupational exposure to human immunodeficiency virus (HIV). In the absence of a safe and effective preventive vaccine, the only way of preventing HIV infection is avoiding exposure to the virus, through safe practices and procedures and protective equipment. When occupational exposure occurs, current guidelines recommend the prompt use of postexposure prophylaxis (PEP), consisting of systemic treatment with a combination of several antiretroviral drugs [1]. Although there are no clinical trials on the use of PEP, data from case-control studies [2] show a substantial reduction of the infection rate when PEP is administered [3].

Nevertheless, failure of PEP has been documented in case-control studies [2] and case reports [4–8]. Understanding the underlying mechanism responsible for PEP can inform the modification of existing guidelines and can guide the development of new compounds, formulations, and routes of administration. As shown in nonhuman primate models, a key element for the success of PEP is the timing of the initiation and the duration of the treatment [9]; current guidelines recommend a prompt start, within 72 hours after exposure, and a treatment duration of 28 days [1]. Another key factor is the need to maintain adequate drug levels of the active form of the antiretroviral agents at the target tissues to prevent viral replication. For this reason, it is important to manage the psychological consequences of the exposure and drug tolerability, as adherence can be impacted by both factors [1, 10]. An additional documented cause of failure of occupational PEP is exposure to drug-resistant HIV [7]. In this issue of the *Journal*, Li et al suggest a novel mechanism responsible for PEP failure: the persistence of sequestered infectious viral particles.

The authors present a case report of a HCW from Australia who, while drawing blood from an HIV-infected patient, accidentally inoculated a small amount of the patient's blood in her own finger. The source patient had been infected with HIV for more than a decade and had received treatment with 3 families of antiretroviral drugs. At the time of the incident, he was receiving zidovudine, lamivudine, and nevirapine but was not adherent to the treatment. He had a plasma viral load of >100 000 copies/mL, his $CD4^+$ T-cell count was 279 cells/μL, and his virus carried a major mutation associated with nevirapine resistance, K103N, in the reverse transcriptase gene [11]. PEP (zidovudine, lamivudine, and indinavir) was initiated within 2 hours after the incident, and she completed 4 weeks of therapy, with a 4-day interruption during the third week. After PEP completion, the HCW had a negative HIV-1 enzyme-linked immunosorbent assay (ELISA) and reported no symptoms (the authors do not mention detection of viral nucleic acids in plasma). Six weeks later (11 weeks after the exposure incident), the HCW presented with symptoms consistent with acute retroviral syndrome, her plasma viral load was >2 million copies/mL, and she had a positive HIV-1 ELISA and an indeterminate HIV-1 Western blot, which set her diagnosis as acute HIV-1 infection, Fiebig stage IV [12].

The authors characterized the viral populations within the HCW, using a

Received and accepted 24 June 2013; electronically published 9 September 2013.

Correspondence: Jerome H. Kim, MD, US Military HIV Research Program, Walter Reed Army Institute of Research, 503 Robert Grant Ave, Silver Spring, MD 20910 (jkim@hivresearch.org).

The Journal of Infectious Diseases 2013;208:1542–4

DOI: 10.1093/infdis/jit486

sample obtained 81 days after exposure, and compared them to those in a sample retrieved from the source patient (obtained 8 days after the exposure incident). They used the gold-standard technique: single-genome sequencing (SGS), which consists of polymerase chain reaction–based amplification and direct sequencing preceded by sample dilution to a level where most sequences are statistically assumed to derive from a single amplifiable molecule [13]. They studied 2 subgenomic regions: the full-length *pol* gene (71 sequences from the HCW and 38 from the source patient) and the gp41-coding area in the *env* gene (100 sequences from the HCW and 23 from the source patient). The close genetic relatedness between viral sequences from both individuals, as well as the interspersed location of sequences in the phylogenetic trees, demonstrated epidemiological linkage between the source patient and the HCW. The level of genetic diversity among HIV from the source patient was consistent with that of a chronic infection, whereas the viruses from the HCW presented an altogether different profile: "multiple sets of identical or nearly identical sequences." This pattern is consistent with very recent HIV-1 infection with multiple transmitted/founder (T/F) viruses [13]. The authors were able to define at least 14–15 lineages in each of the subgenomic areas, and they also identified recombinants among the different lineages. The HCW's viral lineages were interspersed in the source patient's phylogeny, which indicates "absence of obvious selection for particular variants in the transmission event." Infections initiated by multiple T/F viral strains have been described previously [14] and tend to be more common when there is nonmucosal exposure (eg, among injection drug users) [15].

One of the original and innovative aspects of the current study is that sampling multiple strains through SGS allowed the authors to calculate the time to the most recent common ancestor (MRCA), based on a model that assumes exponentially growing population in the absence of differential selection [16]. They found that the time to the MRCA of the main HCW lineages was 14–37 days before sampling (ie, 44–67 days after the exposure incident). This surprising finding may suggest that "each sequence lineage evolved from a discrete T/F virus that began to replicate only after PEP was discontinued."

Why was it that at least 15 T/F viruses established infection in the setting of prompt and potent combination antiretroviral therapy? The report of the case and the molecular evidence indicates that the classical factors—timing of treatment initiation, adherence to treatment, and viral resistance—were likely not the reasons for PEP failure: (1) PEP was initiated within 2 hours after exposure, (2) the HCW completed the prescribed 4-week treatment (the authors report a 4-day interruption of treatment, but they do not comment on the cause), or (3) the HCW's viral sequences did not carry mutations in the *pol* gene that would confer resistance to the prescribed antiretroviral PEP, just the aforementioned nevirapine resistance–associated mutation that was already present in the source patient. This argues against the transmission or emergence of drug-resistant viruses as the reason for PEP failure.

The authors suggest this is a case of virus sequestration and associated evolutionary arrest and suggest a number of possible mechanisms. They disfavored preintegration or postintegration latency, because the former was not consistent with the life span of preintegration complexes (ie, in the order of days [17]) and the latter would have required the infection of an immense number of $CD4^+$ T cells during the initial hours after exposure to generate enough latently infected memory $CD4^+$ T cells to give rise to all of the observed viral lineages. They also dismissed incomplete suppression of virus replication, because this would be inconsistent with the low level of genetic diversity within each of the lineages and the absence of mutations associated with resistance to the PEP regimen.

The authors favor the hypothesis that the virus was trapped and sequestered by follicular dendritic cells (DCs) or other antigen-presenting cells, which would be supported by the observation in animal models and ex vivo experiments with human cells that indicate that trapped HIV-1 particles can remain infectious for several months [18]. Interestingly, the trapping of HIV-1 by follicular DCs is mediated by Fc gamma receptors on the surface of these cells and requires the viral particles to be in the form of immune complexes with viral-specific antibodies [19]. A requirement of this plausible hypothesis is the presence of preformed antibody-virus complexes, rather than free viral particles, in the source patient at the time of exposure. This is a hypothesis that can be tested in nonhuman primate models by simulating occupational exposure to either free viral particles or antibody-virus complexes and then administering PEP. If corroborated, this would signify an important change in the way PEP is studied. Moreover, this could also have implications for other related fields, such as vaccines or preexposure prophylaxis. For example, in a very recent trial conducted by the Bangkok Tenofovir Study Group, in which the daily use of tenofovir reduced the risk of HIV acquisition among injection-drug users by 48.9%, poor adherence was only partly responsible for the lack of protection in the treated arm [20]. Did viral sequestration and evolutionary arrest manifest itself in these patients?

While this study does not constitute the first report of the failure of highly active antiretroviral therapy as PEP, it is the first to use an in-depth and systematic molecular analysis that presents testable hypotheses for nonhuman primate studies that could, when more fully delineated, make the case for changes in the recommendations for PEP.

Notes

Disclaimer. The views expressed are those of the authors and should not be construed to represent the positions of the US Army or Department of Defense.

Financial support. This work was supported by the Henry M. Jackson Foundation for the Advancement of Military Medicine and the US Department of Defense (cooperative agreement W81XWH-11-2-0174), as well as by the National Institute of Allergy and Infectious Diseases.

Potential conflicts of interest. All authors: No reported conflicts.

All authors have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Conflicts that the editors consider relevant to the content of the manuscript have been disclosed.

References

1. Panlilio AL, Cardo DM, Grohskopf LA, Heneine W, Ross CS. Updated U.S. Public Health Service guidelines for the management of occupational exposures to HIV and recommendations for postexposure prophylaxis. MMWR Recomm Rep **2005**; 54:1–17.
2. Cardo DM, Culver DH, Ciesielski CA, et al. A case-control study of HIV seroconversion in health care workers after percutaneous exposure. Centers for Disease Control and Prevention Needlestick Surveillance Group. N Engl J Med **1997**; 337:1485–90.
3. Agins BD, Bacon O, Balano K, et al. Section 3: health care maintenance and disease prevention. In: Coffey S, ed. Guide for HIV/AIDS clinical care. Rockville, MD: U.S. Department of Health and Human Services, Health Resources and Services Administration, HIV/AIDS Bureau, **2011**.
4. Wang SA, Panlilio AL, Doi PA, White AD, Stek M, Jr., Saah A. Experience of healthcare workers taking postexposure prophylaxis after occupational HIV exposures: findings of the HIV Postexposure Prophylaxis Registry. Infect Control Hosp Epidemiol **2000**; 21:780–5.
5. Jochimsen EM. Failures of zidovudine postexposure prophylaxis. Am J Med **1997**; 102:52–5; discussion 6–7.
6. Pratt RD, Shapiro JF, McKinney N, Kwok S, Spector SA. Virologic characterization of primary human immunodeficiency virus type 1 infection in a health care worker following needlestick injury. J Infect Dis **1995**; 172:851–4.
7. Beltrami EM, Luo CC, de la Torre N, Cardo DM. Transmission of drug-resistant HIV after an occupational exposure despite postexposure prophylaxis with a combination drug regimen. Infect Control Hosp Epidemiol **2002**; 23:345–8.
8. Tokars JI, Marcus R, Culver DH, et al. Surveillance of HIV infection and zidovudine use among health care workers after occupational exposure to HIV-infected blood. The CDC Cooperative Needlestick Surveillance Group. Ann Intern Med **1993**; 118:913–9.
9. Tsai CC, Emau P, Follis KE, et al. Effectiveness of postinoculation (R)-9-(2-phosphonylmethoxypropyl) adenine treatment for prevention of persistent simian immunodeficiency virus SIVmne infection depends critically on timing of initiation and duration of treatment. J Virol **1998**; 72:4265–73.
10. de la Tribonniere X, Dufresne MD, Alfandari S, et al. Tolerance, compliance and psychological consequences of post-exposure prophylaxis in health-care workers. Int J STD AIDS **1998**; 9:591–4.
11. Johnson VA, Calvez V, Gunthard HF, et al. Update of the drug resistance mutations in HIV-1: March 2013. Top Antivir Med **2013**; 21:6–14.
12. Fiebig EW, Wright DJ, Rawal BD, et al. Dynamics of HIV viremia and antibody seroconversion in plasma donors: implications for diagnosis and staging of primary HIV infection. Aids **2003**; 17:1871–9.
13. Keele BF, Giorgi EE, Salazar-Gonzalez JF, et al. Identification and characterization of transmitted and early founder virus envelopes in primary HIV-1 infection. Proc Natl Acad Sci U S A **2008**; 105:7552–7.
14. Shaw GM, Hunter E. HIV transmission. Cold Spring Harb Perspect Med **2012**; 2.
15. Bar KJ, Li H, Chamberland A, et al. Wide variation in the multiplicity of HIV-1 infection among injection drug users. J Virol **2010**; 84:6241–7.
16. Lee HY, Giorgi EE, Keele BF, et al. Modeling sequence evolution in acute HIV-1 infection. J Theor Biol **2009**; 261:341–60.
17. Pierson TC, Zhou Y, Kieffer TL, Ruff CT, Buck C, Siliciano RF. Molecular characterization of preintegration latency in human immunodeficiency virus type 1 infection. J Virol **2002**; 76:8518–31.
18. Smith BA, Gartner S, Liu Y, et al. Persistence of infectious HIV on follicular dendritic cells. J Immunol **2001**; 166:690–6.
19. Smith-Franklin BA, Keele BF, Tew JG, et al. Follicular dendritic cells and the persistence of HIV infectivity: the role of antibodies and Fcgamma receptors. J Immunol **2002**; 168:2408–14.
20. Choopanya K, Martin M, Suntharasamai P, et al. Antiretroviral prophylaxis for HIV infection in injecting drug users in Bangkok, Thailand (the Bangkok Tenofovir Study): a randomised, double-blind, placebo-controlled phase 3 trial. Lancet **2013**; 381: 2083–90.