

Interferon Alfa Therapy: Toward an Improved Treatment for HIV Infection

Lucy A. McNamara^{1,2} and Kathleen L. Collins^{1,3}

¹Department of Microbiology and Immunology, ²Department of Epidemiology, and ³Department of Internal Medicine, University of Michigan, Ann Arbor

(See the major article by Azzoni et al, on pages 213–22.)

Highly active antiretroviral therapy (HAART) is an expensive, lifelong treatment for human immunodeficiency virus (HIV) infection that is associated with significant toxicity. Despite advances in treatment, there remains a need for novel therapies for HIV infection. One strategy is to achieve a functional cure, in which treatment can be safely stopped despite the presence of residual virus, by finding approaches that enhance the immune response to HIV. Such a therapy would require boosting a patient's immune system to suppress viral replication below clinically relevant levels, thereby eliminating the need for HAART. A sterilizing cure, by contrast, is one in which all functional HIV genomes within the body are eliminated. Latent HIV genomes represent a major barrier to a sterilizing cure because these genomes do not produce viral proteins and thus cannot be eliminated by the immune system or by current therapies [1].

In this issue, Azzoni and colleagues assessed the potential of pegylated (Peg) interferon alfa-2a to suppress HIV replication [2]. In what could be a first step towards a functional cure, they found that Peg–interferon alfa-2a permitted a subset of subjects to maintain low viral loads for 12–24 weeks in the absence of HAART. Furthermore, they found that in patients with a favorable virologic response to Peg interferon alfa-2a, levels of HIV provirus in peripheral blood mononuclear cells (PBMCs) were reduced. This finding suggests that Peg–interferon alfa-2a may eliminate latent HIV genomes, a possibility that would shed light on the search for a sterilizing as well as a functional cure for HIV.

Interferon alfa is used clinically for the treatment of hepatitis B and C virus infections, and several studies have assessed the capacity of this cytokine to limit HIV replication in vivo [3–9]. Multiple studies confirm that interferon alfa can reduce viral load and delay disease progression in viremic patients [3, 5–11]. However, interferon alfa's potential to suppress viremia in patients with well-controlled viral loads is less clear. Recently, one study subjected HAART-treated patients with a viral load of <400 copies/mL for at least 6 months and a CD4⁺ T-cell count of >350 cells/μL to a series of three 4-week structured treatment interruptions (STIs), during which they received interferon alfa

therapy or no treatment [4]. Treatment was then suspended until the end of the trial. Interferon alfa administration was observed to significantly blunt the viral load rebound during the 4-week STI phase, but it did not prolong the time to treatment resumption after treatment was suspended. This study also noted that interferon alfa was deleterious in patients with a low nadir CD4⁺ T-cell count [4].

This issue's study by Azzoni et al is the first to show an effect of Peg–interferon alfa-2a treatment on viral load rebound during a multimonth interval of treatment interruption and the first to identify an effect of Peg–interferon alfa-2a on the HIV proviral reservoir [2]. In this study, the authors recruited 23 HAART-treated, HIV-positive patients with HIV plasma RNA levels of <50 copies/mL [2]. All subjects received Peg–interferon alfa-2a therapy in addition to HAART for 5 weeks; then HAART but not Peg–interferon alfa-2a therapy was interrupted for 12–24 weeks [2]. Viral suppression was maintained through week 12 of treatment interruption in 45% of patients and through week 24 in 30% of patients; in both cases, the proportion of patients with suppressed viral loads was significantly greater than expected on the basis of data from historical controls [2]. The authors comment that the higher level of suppression observed in this study compared with

Received and accepted 5 September 2012; electronically published 26 October 2012.

Correspondence: Kathleen L. Collins, MD, PhD, Department of Internal Medicine, University of Michigan, Ann Arbor, 3510 MSRB1, 1150 W Medical Center Dr, Ann Arbor, MI, 48109 (kcollin@med.umich.edu).

The Journal of Infectious Diseases 2013;207:201–3

© The Author 2012. Published by Oxford University Press on behalf of the Infectious Diseases Society of America. All rights reserved. For Permissions, please e-mail: journals.permissions@oup.com.

DOI: 10.1093/infdis/jis667

other trials may have resulted from co-administering Peg-interferon alfa-2a with ART prior to treatment interruption. Subjects in this study also had complete viral suppression (viral load, <50 copies/mL) and a high degree of immune reconstitution (CD4⁺ T-cell count, >450 cells/mL) [2].

Furthermore, the authors assessed the level of integrated HIV-1 DNA in PBMCs in 13 patients, 6 of whom exhibited virological failure and 7 of whom maintained virological suppression at week 12 of the treatment interruption [2]. No change in the number of copies of HIV-1 DNA per CD4⁺ T cell was observed in the patients experiencing virological failure [2], a useful finding because it demonstrates that in the presence of Peg-interferon alfa-2a, an STI can be undertaken without detectably increasing the size of the latent viral reservoir in the peripheral blood. Moreover, a significant decrease in the HIV-1 DNA load per CD4⁺ T cell was detected in the 7 subjects who maintained viral suppression at week 12 [2]. This unexpected finding suggests that when optimally effective, Peg-interferon alfa-2a may be able not only to suppress viral replication by enhancing the immune response to HIV, but also to assist in clearance of the viral reservoir.

Given the small size of this study and the necessity of using historical controls, these results must be interpreted cautiously. However, they raise several interesting questions for future studies. One question is how interferon alfa activates the immune response to both suppress viral replication and reduce the reservoir of HIV proviral DNA. Interferon alfa promotes an antiviral response to HIV that inhibits viral replication at multiple stages of the replication cycle [12]. Specifically, interferon alfa upregulates APOBEC3G, TRIM5 α , and tetherin in vitro; upregulation of both APOBEC3G and tetherin in response to interferon alfa therapy has been confirmed in vivo [8]. The upregulation of these factors in response to interferon alfa therapy

might inhibit viral replication directly by causing mutations in viral genomes (in the case of APOBEC3G) or by preventing viral uncoating (for TRIM5 α) or budding (for tetherin). Alternatively, APOBEC3G upregulation could suppress viral replication by activating other immune cells. Recent work has shown that APOBEC3G activity can enhance recognition of HIV-infected cells by CD8⁺ cytotoxic T cells [13] and natural killer (NK) cells [14]. A study showing that interferon alfa treatment in vivo enhances NK cell function provides further support for the relevance of this mechanism [15].

It is less clear how interferon alfa could contribute to a reduction in HIV proviral DNA in PBMCs. If the proviral genomes detected in this study represent latent virus, neither APOBEC3G, TRIM5 α , nor tetherin should have any impact on the amount of HIV-1 DNA detected. Instead, the finding that there is a decrease in the number of HIV-1 genomes detected may suggest that interferon alfa reactivates latent HIV-1, leading to the loss of the infected cells through the virus's cytotoxic effects or the action of the immune system. Alternatively, the decrease in proviral genomes in a subset of interferon alfa-treated individuals might indicate that a fraction of these genomes derive from ongoing replication during HAART therapy, rather than from latent viruses. In this case, the mechanism through which interferon alfa could reduce the number of proviral genomes detected is more obvious. However, although there is some evidence that ongoing replication can occur in a subset of HAART-treated patients with suppressed viral loads [16–19], the majority of the evidence suggests that residual replication is not a major source of viral persistence in these patients [20–25]. Further study is required to confirm that interferon alfa reduces the amount of HIV-1 DNA detected in the PBMCs of at least a subset of patients and to help us better understand this interesting finding.

Another question is why Peg-interferon alfa-2a therapy was effective in suppressing viral replication only for some individuals in this study. Analogous to a recent study on the control of hepatitis C virus infection, in which Ge et al found that the response to interferon alfa treatment is associated with a polymorphism in the *IL28B* gene [26], the authors examined multiple polymorphisms associated with improved or reduced hepatitis C virus or HIV control. No association between any genotype and response to Peg-interferon alfa-2a was found; however, this study was too small for any but the strongest association between genotype and treatment outcome to be detected. Alternative biomarkers for response to interferon alfa should also be investigated, such as variation in levels of APOBEC3G [14, 27], TRIM5 α , or tetherin. Finally, HIV characteristics may determine which individuals can respond to interferon alfa. This possibility would again be analogous to hepatitis C virus infection, in which patients infected with viral genotype 1 are less likely to respond to interferon alfa treatment than are patients infected with viral genotype 2 or 3 [28].

Although the results of the present study need to be confirmed in a larger cohort, the clinical implications of this research are promising. Since the SMART study, which demonstrated that STIs were associated with a significantly increased risk of opportunistic disease or death [29], it has been difficult to assess the efficacy of therapies designed to limit viral rebound without concerns about endangering the health of study participants. In the current study, however, Azzoni et al have demonstrated that it is possible to subject participants to an STI without increasing the reservoir of HIV DNA in the peripheral blood. Although HIV DNA and adverse health outcomes are very different outcome measurements, this result suggests that interferon alfa administration before and during STIs could provide a

safer way to evaluate viral control in the absence of HAART. Thus, further study of interferon alfa therapy in HAART-treated patients will at once enhance our understanding of how viral replication can be limited in the absence of HAART and provide us with a tool to study alternative approaches to a cure.

Notes

Funding. L.A.M. is supported by a University of Michigan Rackham Predoctoral Fellowship. This work was funded by the Burroughs Wellcome Foundation and the National Institutes of Health RO1 AI096962.

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. Geeraert L, Kraus G, Pomerantz RJ. Hide-and-seek: the challenge of viral persistence in HIV-1 infection. *Annu Rev Med* **2008**; 59:487–501.
2. Azzoni L, Foulkes AS, Papasavvas E, et al. Pegylated interferon alfa-2a monotherapy results in suppression of HIV type 1 replication and decreased cell-associated HIV DNA integration. *J Infect Dis* **2012**; In press.
3. Asmuth DM, Murphy RL, Rosenkranz SL, et al. Safety, tolerability, and mechanisms of antiretroviral activity of pegylated interferon alfa-2a in HIV-1-monoinfected participants: a phase II clinical trial. *J Infect Dis* **2010**; 201:1686–96.
4. Boué F, Reynes J, Rouzioux C, et al. Alpha interferon administration during structured interruptions of combination antiretroviral therapy in patients with chronic HIV-1 infection: INTERVAC ANRS 105 trial. *AIDS* **2011**; 25:115–8.
5. Dianzani F, Rozera G, Abbate I, et al. Interferon may prevent HIV viral rebound after HAART interruption in HIV patients. *J Interferon Cytokine Res* **2008**; 28:1–3.
6. Lane HC, Kovacs JA, Feinberg J, et al. Antiretroviral effects of interferon-alpha in AIDS-associated Kaposi's sarcoma. *Lancet* **1988**; 2:1218–22.
7. Lane HC, DAvey V, Kovacs JA, et al. Interferon- α in patients with asymptomatic human immunodeficiency virus (HIV) infection. *Ann Intern Med* **1990**; 112:805–11.
8. Pillai SK, Abdel-Mohsen M, Guatelli J, et al. Role of retroviral restriction factors in the interferon- α -mediated suppression of HIV-1 in vivo. *Proc Natl Acad Sci U S A* **2012**; 109:3035–40.
9. Tavel JA, Huang C-Y, Shen J, et al. Interferon- α produces significant decreases in HIV load. *J Interferon Cytokine Res* **2010**; 30: 461–4.
10. Rivero J, Limonta M, Aguilera A, et al. Use of recombinant interferon- α in human immunodeficiency virus (HIV)-infected individuals. *Biotherapy* **1994**; 8:23–31.
11. Rivero J, Fraga M, Cancio I, et al. Long-term treatment with recombinant interferon alpha-2b prolongs survival of asymptomatic HIV-infected individuals. *Biotherapy* **1997**; 10:107–13.
12. Neil S, Bieniasz P. Human immunodeficiency virus, restriction factors, and interferon. *J Interferon Cytokine Res* **2009**; 29:569–80.
13. Casartelli N, Guivel-Benhassine F, Bouziat R, et al. The antiviral factor APOBEC3G improves CTL recognition of cultured HIV-infected T cells. *J Exp Med* **2010**; 207:39–49.
14. Norman JM, Mashiba M, McNamara LA, et al. The antiviral factor APOBEC3G enhances the recognition of HIV-infected primary T cells by natural killer cells. *Nature Immunol* **2011**; 12:975–83.
15. Portales P, Reynes J, Pinet V, et al. Interferon- α restores HIV-induced alteration of natural killer cell perforin expression in vivo. *AIDS* **2003**; 17:495–504.
16. Buzón MJ, Massanella M, Llibre JM, et al. HIV-1 replication and immune dynamics are affected by raltegravir intensification of HAART-suppressed subjects. *Nature Med* **2010**; 16:460–5.
17. Shiu C, Cunningham CK, Greenough T, et al. Identification of ongoing human immunodeficiency virus type 1 (HIV-1) replication in residual viremia during recombinant HIV-1 poxvirus immunizations in patients with clinically undetectable viral loads on durable suppressive highly active antiretroviral therapy. *J Virol* **2009**; 83: 9731–42.
18. Sigal A, Kim JT, Balazs AB, et al. Cell-to-cell spread of HIV permits ongoing replication despite antiretroviral therapy. *Nature* **2011**; 477:95–8.
19. Tobin NH, Learn GH, Holte SE, et al. Evidence that low-level viremia during effective highly active antiretroviral therapy result from two processes: expression of archival virus and replication of virus. *J Virol* **2005**; 79:9625–34.
20. Besson GJ, McMahon D, Maldarelli F, Mellors JW. Short-course raltegravir intensification does not increase 2 long terminal repeat episomal HIV-1 DNA in patients on effective antiretroviral therapy. *Clin Infect Dis* **2012**; 54:451–3.
21. Dinoso JB, Kim SY, Wiegand AM, et al. Treatment intensification does not reduce residual HIV-1 viremia in patients on highly active antiretroviral therapy. *Proc Natl Acad Sci U S A* **2009**; 106:9403–8.
22. Gandhi RT, Zheng L, Bosch RJ, et al. The effect of raltegravir intensification on low-level residual viremia in HIV-infected patients on antiretroviral therapy: a randomized controlled trial. *PLoS Med* **2010**; 7: e1000321.
23. Hatano H, Hayes TL, Dahl V, et al. A randomized, controlled trial of raltegravir intensification in antiretroviral-treated, HIV-infected patients with a suboptimal CD4⁺ T cell response. *J Infect Dis* **2011**; 203: 960–8.
24. Joos B, Fischer M, Kuster H, et al. HIV rebounds from latently infected cells, rather than from continuing low-level replication. *Proc Natl Acad Sci U S A* **2008**; 105: 16725–30.
25. Kieffer TL, Finucane MM, Nettles RE, et al. Genotypic analysis of HIV-1 drug resistance at the limit of detection: virus production without evolution in treated adults with undetectable HIV loads. *J Infect Dis* **2004**; 190:1452–65.
26. Ge D, Fellay J, Thompson AJ, et al. Genetic variation in IL28B predicts hepatitis C treatment-induced viral clearance. *Nature* **2009**; 461:399–401.
27. Koning FA, Newman ENC, Kim E-Y, et al. Defining APOBEC3 expression patterns in human tissues and Hematopoietic cell subsets. *J Virol* **2009**; 83:9474–85.
28. Zeuzem S. Interferon-based therapy for chronic hepatitis C: current and future perspectives. *Nature Clin Pract Gastroenterol Hepatol* **2008**; 5:610–22.
29. Strategies for Management of Antiretroviral Therapy (SMART) Study Group. CD4⁺ count-guided interruption of antiretroviral treatment. *N Engl J Med* **2006**; 355: 2283–96.