The human immune system is extraordinarily active against infection with HIV-1, yet never eliminates the virus and rarely controls viral replication for prolonged periods without the assistance of anti-retroviral therapy (ART). In order to harness the power of the human immune response in HIV therapy, we need a better understanding of the key elements of protective immunity against the virus and how and why these ultimately fail in chronic infection.

**A New Study on Cytotoxic T Lymphocytes**

Most investigators would nominate cytotoxic T lymphocytes (CTLs) as key players in the control of HIV-1 infection, based on data accumulated over the two decades since they were first described. This evidence includes the appearance of CTLs very early in HIV-1 infection coinciding with a profound drop in plasma viral load and the dramatic rise in viremia following CTL depletion in monkey models of both acute and chronic infection with simian immunodeficiency virus [1].

In a new study in this issue of *PLoS Medicine*, Marcus Altfeld and colleagues describe the fate of CTLs responding to HIV-1 from the very earliest stages of infection—the time that most investigators believe is critical in determining the long-term outcome of HIV-1 infection [2]—through the transition to chronic infection [3]. Although the functionality and phenotype of HIV-specific CTLs showed variability both between and within patients, deterioration in the number of functions attributable to each individual T cell was consistently found in untreated patients. Thus in the 11 out of 18 patients who chose not to start ART in acute infection, the capacity of HIV-specific CTLs to secrete a range of anti-viral cytokines and chemokines as well as to generate cytotoxic granules in response to an encounter with HIV antigens declined in the face of continuing viral replication.

Deterioration of immune function as viral levels increase is well described at other stages of HIV-1 infection, but it is inevitably difficult to determine which is cause and effect in this scenario. In this new study, the authors were able to exploit another observation to examine the underlying causes of declining T cell function in their patients. By studying the evolution of the infecting virus in the first months of infection, they noted that in many instances there was an early accumulation of mutations in T cell epitopes that enabled the virus to avoid recognition by circulating CTLs. Not only did these mutations render virus-infected cells “invisible” to the responding T cells, but they also prevented repeated stimulation of the cells following contact with their target antigen. Even in untreated patients, the effect of removing CTLs from antigen exposure led to a very similar preservation of T cell function to that seen in those with a good response to ART. This maintenance of CTL function was particularly striking in untreated patients, for whom escape mutations were generated to some but not all of their repertoire of responding CTLs, thereby making it possible to discern the role of repeated antigenic stimulation in promoting T cell dysfunction.

These observations are also important in highlighting how early in HIV-1 infection immune pressure from CTLs can drive the emergence of escape mutations: this is well documented in the macaque model [4], but has not been studied systematically in human infection. CTL escape mutations were selected in nine of the untreated patients in Altfeld and colleagues’ study and could be detected as early as 61 days after initial presentation.

**Clinical Implications of the Study**

What are the clinical implications of this study? If polyfunctional HIV-specific CTLs need to be preserved long term for the fight against HIV-1 infection, then this study suggests that such preservation is best achieved by suppressing HIV-1 replication both early and efficiently. The question of whether or not to start ART in acute HIV-1 infection has been controversial.

**Funding:** The authors received no specific funding for this article.

**Competing Interests:** The authors have declared that no competing interests exist.

**Citation:** Rowland-Jones S, de Silva T (2008) Resisting immune exhaustion in HIV-1 infection. *PLoS Med* 5(5): e103. doi:10.1371/journal.pmed.0050103

**Copyright:** © 2008 Rowland-Jones and de Silva. This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

**Abbreviations:** ART, anti-retroviral therapy; CTL, cytotoxic T lymphocyte; IL-2, interleukin-2; PD-1, programmed death-1; STI, structured treatment interruption

Sarah Rowland-Jones and Thushan de Silva are with Medical Research Council Laboratories, Banjul, The Gambia.

*To whom correspondence should be addressed. E-mail: sarah.rowland-jones@ndm.ox.ac.uk*
Acute HIV-1 infection is characterised by high levels of viral replication, dissemination of virus to lymphoid tissue reservoirs, and gradual depletion of circulating HIV-1-specific CD4+ T lymphocytes [5,6]. Evidence from the study of gut-associated lymphoid tissue (GALT) in animal models suggests that there is massive infection of memory CD4+ T cells in GALT and subsequent loss of over half the total memory T cell pool within two weeks of experimental simian immunodeficiency virus infection [7]. If this situation is mirrored in human infection, as suggested by the extensive depletion of GALT T cells in biopsies taken in chronic HIV-1 infection [8], the implication is that very early intervention would be needed to preserve memory T cell function. Although the use of ART in chronic HIV infection undoubtedly results in significant reductions in morbidity and mortality, reconstitution of the host immune system is rarely achieved. For example, HIV-specific CD4+ T helper cell responses, which crucially augment effector HIV-specific CD8+ responses, are poorly restored by ART in chronic infection [9,10]. Taken together, these data lead to the inevitable question of whether starting ART in acute HIV infection, and thereby minimising virus dissemination and damage to mucosal-associated lymphoid tissue, could facilitate the development and preservation of enhanced HIV-specific immunity and thus favourably alter the future course of disease.

Slow restoration of a polyfunctional CTL phenotypic profile similar to that observed by Altfeld and colleagues can also be achieved in chronic HIV-infected patients treated with ART [11]. However, the clinical significance of this improvement remains unclear in the face of evidence that suggests cessation of ART during treatment interruption in chronic HIV-1 infection results in rapid viral rebound and no long-term change in viral set point [12]. HIV-specific CTLs with strong ex vivo proliferative capacity are a feature of HIV long-term non-progressors [13], and can also be detected in acutely HIV-infected patients during the peak of viraemia, but gradually diminish during the first year of infection in the absence of therapy [14]. Preservation of CTL proliferative capacity and effector function appears to be critically dependent on interleukin-2 (IL-2) production from HIV-specific CD4+ T cells [14]; this production in turn can be preserved by early institution of ART [15].

**Structured Treatment Interruptions**

Clinicians have been understandably reluctant to commit patients diagnosed with acute HIV-1 infection to lifelong ART. An alternative strategy was based on the hypothesis that preservation of HIV-specific immunity could be achieved by starting ART in acute infection, followed by structured treatment interruptions (STIs), thereby allowing “immune boosting” by exposure to autologous virus.

This strategy involved restarting therapy if rebound plasma viraemia increased above set thresholds (more than 5,000 copies/ml for three consecutive weeks or more than 50,000 copies/ml on one occasion) and introducing further STIs once viral control was regained. Initial enthusiasm for this approach was fuelled by the observation that potent Gag-specific T helper cell responses develop in patients with acute HIV infection started on ART, at similar magnitudes to those seen in long-term non-progressors, and to significantly higher levels than are found in untreated patients with acute HIV infection or ART-treated patients with chronic HIV infection [15]. Although some patients subjected to STIs after starting ART in acute infection were initially able to control viraemia and maintain Gag-specific CD4+ responses off therapy, a detailed longitudinal analysis (median 5.3 years from infection) of this cohort showed the effect to be transient, with viral breakthrough occurring ultimately in most patients, accompanied by a similar rate of CD4+ cell loss as that seen in early chronic untreated HIV infection [16]. It seems, therefore, that although the immunological damage caused by acute HIV-1 infection may be reduced to some extent by early ART, this effect is limited to the duration of therapy and may not translate into long-term benefits.

**Can Early ART Affect Risk of Future Disease Progression?**

For clinicians to accept early and lifelong therapy for HIV-1 infection into routine practice, reliable data from controlled clinical trials are needed. To date, there have been no randomized...
and adequately powered studies addressing the issue of whether early ART can affect the risk of future HIV-1 disease progression.

A number of observational studies of ART used for a limited period in early HIV-1 infection present contrasting conclusions. One multicentre observational study compared surrogate markers of disease progression at 24, 48, and 72 weeks of untreated observation in 58 ART-treated patients (13 with “acute” infection within two weeks of seroconversion and 45 with “early” infection within six months of seroconversion) and 337 untreated patients with primary HIV infection [17]. Lower viral loads and higher CD4 counts were observed at 24 weeks following cessation of ART in both the acute and early treatment groups, although a longer-term benefit at 72 weeks was less clear. Despite these promising results, the variable ART duration (median 1.5 years) and lack of randomisation, amongst other factors, make interpretation of this study difficult. In contrast, short-term ART (for 24 weeks) failed to show any benefit in CD4 counts or viral loads at six months after treatment discontinuation in a smaller observational study [18]. Enhanced interferon-γ and CD107a expression on HIV-specific CD8+ T cells at 12 months in the treated group did not result in lower viral load set points.

Although these, along with other such studies, may hint at the potential benefits of using short-term ART in acute HIV-1 infection, data are needed from adequately powered and controlled studies, such as the ongoing Short Pulse Anti Retroviral Therapy at HIV Seroconversion (SPARTAC) study (http://www.ctu.mrc.ac.uk/studies/spartac.asp). This is an international randomised controlled trial comparing combination anti-retroviral therapy given for 48 weeks or 12 weeks, the effect of combination anti-retroviral therapy compared to the duration of therapy, then the benefits of early ART are limited with a no-intervention arm.

The possibility that ART may need to be started as early as possible and continued indefinitely raises many concerns. How would clinicians balance the potential impact on disease progression against the cost, drug toxicity, and the risk of drug resistance entailed by prolonged ART, particularly at a time in early HIV-1 infection when most patients would be probably be asymptomatic without therapy? If it is important to treat within days of primary infection, how should we best identify newly infected patients at the optimum time for instituting therapy? The need to provide long-term therapy for participants in vaccine trials who acquire primary HIV-1 infection during the trial would have major cost and logistic implications that could make phase III vaccine trials virtually impracticable.

A Way Forward

Probably the best way forward would be to develop a therapeutic strategy that combines early viral suppression using ART with immunotherapy to augment HIV-specific immune responses in a way that does not expose the host immune system to the damaging consequences of continued HIV-1 replication (see Figure 1). Although both therapeutic vaccination and passive monoclonal antibody infusion in this setting have so far failed to show absolute benefit [22–24], there are some data to suggest that therapeutic vaccination in chronic HIV-1 infection can lead to a restoration of a broad and fully functional CTL response [25]. In Altfeld and colleagues’ study [3], a reliable marker of failing CTLs was expression of the molecule.

Five Key Papers in the Field

- **Streeck et al., 2008** [3] Epitope-specific CD8+ T cells in acute HIV infection progressively lose their polyfunctional capacity following repeated exposure to antigen in acute HIV infection, but this exhausted phenotype is reversible either with anti-retroviral therapy or reduction in epitope-specific antigen load due to cytotoxic T cell escape mutations.

- **Day et al., 2006** [27] The inhibitory receptor PD-1 is significantly up-regulated on HIV-specific T cells in chronic HIV infection, and expression correlates with impaired cytotoxic T cell function and predictors of disease progression. Blockade of the PD-1 pathway enhances HIV-specific CD4+ and CD8+ cellular function.

- **Mattapallil et al., 2005** [7] 30%–60% of CD4+ memory cells in most tissues are infected during the peak of experimental acute simian immunodeficiency virus infection in macaques, resulting in catastrophic early depletion of these cells by direct viral infection.

- **Lichterfeld et al., 2004** [14] The loss of HIV-specific CD8+ T cell function in chronic HIV-1 infection correlates with disease progression and is critically dependent on IL-2 secretion from HIV-specific CD4+ T helper cells. This functional deficit may be reversible with immunotherapeutic interventions.

- **Rosenberg et al., 2000** [15] Successful treatment of acute HIV infection with anti-retroviral therapy leads to preservation of HIV-specific CD4+ T helper cell responses. Maintenance of virus-specific HIV CD8+ and CD4+ responses, along with viraemic control, can be seen in the short term even when therapy is subsequently stopped in STIs.
programmed death-1 (PD-1), which has been associated with “exhausted” and dysfunctional T cells in chronic infection [26]. Up-regulation of PD-1 on HIV-specific CTLs correlates with plasma viral load [27] and can be reversed when viral replication is controlled both in chronic and acute infection [3,28]. It has recently been shown in a murine model of lymphohoriomeningitis virus that a combination of therapeutic vaccination and blockade of PD-1’s interaction with its ligand, PD-L1, both enhanced the function of responding T cells and significantly improved viral control [29]. Perhaps a similar strategy may enhance the response to therapeutic immunisation in early HIV-1 infection and facilitate long-term viral control without resorting to lifelong drug therapy. ■

References
1. McMichael AJ, Rowland-Jones SL (2001) Cellular immune responses to HIV. Nature 410: 980-987.
2. Mellors JW, Kingsley LA, Rinaldo CR Jr, Todd JA, Hooe BS, et al. (1995) Quantitation of HIV-1 RNA in plasma predicts outcome after seroconversion. Ann Intern Med 122: 573-579.
3. Streeck H, Brumme ZL, Anastario M, Cohen KW, John JS, et al. (2008) Antigen load and viral sequence diversification determine the functional profile of HIV-1-specific CD8+ T cells. PLoS Med 5: e100. doi:10.1371/journal.pmed.0050100
4. Allen TM, O’Connor DH, Jing P, Dzirius JS, Mothi BR, et al. (2000) Tat-specific cytotoxic T lymphocytes select for SIV escape variants during resolution of primary viremia. Nature 407: 386-390. Comment in: Nature 407: 315-316.
5. Clark SJ, Saag MS, Becker WD, Campbell-Hill S, Roberson JL, et al. (1991) High titers of cytotoxic virus in plasma of patients with symptomatic primary HIV-1 infection. N Engl J Med 324(14): 934-939.
6. Daar ES, Moudgil T, Meyer RD, Ho DD (1991) Transient high levels of viremia in patients with primary human immunodeficiency virus type 1 infection. N Engl J Med 324: 961-964.
7. Mattapallil JJ, Donohue DC, Hill B, Nishimura Y, Martin M, et al. (2005) Massive infection and loss of memory CD4+ T cells in multiple tissues during acute SIV infection. Nature 434: 1093-1097.
8. Brenchley JM, Schacker TW, Ruff LE, Price DA, Taylor JH, et al. (2004) CD4+ T cell depletion during all stages of HIV disease occurs predominantly in the gastrointestinal tract. J Exp Med 200: 1397-1404.
9. Autran B, Carcelain G, Li TS, Blanc C, Mathez D, et al. (1997) Positive effects of combined antiretroviral therapy on CD4+ T cell homeostasis and function in advanced HIV disease. Science 277: 112-116.
10. Rinaldo CR Jr, Liebmann JM, Huang X, Fan Z, Al-Shboul Q, et al. (1999) Prolonged suppression of human immunodeficiency virus type 1 (HIV-1) viremia in persons with advanced disease results in enhancement of CD4 T cell reactivity to microbial antigens but not to HIV-1 antigens. J Infect Dis 179: 329-336.
11. Rehr M, Cabierzzi F, Pietsch A, Price DA, Gostick E, et al. (2008) Emergence of polyfunctional CD8+ T cells after prolonged suppression of human immunodeficiency virus replication by antiretroviral therapy. J Virol 82: 3391-3404.
12. Okunishi A, Price DA, Gunaward HF, Dawson SJ, Fagard C, et al. (2002) Stimulation of HIV-specific cellular immunity by structured treatment interruption fails to enhance viral control in chronic HIV infection. Proc Natl Acad Sci U S A 99: 13747-13752.
13. Migueles SA, Laborico AC, Shupert WL, Sabibaghian MS, Rabia R, et al. (2002) HIV-specific CD4+ T cell proliferation is coupled to perforn expression and is maintained in nonprogressors. Nat Immunol 3: 1061-1068.
14. Lichterfeld M, Kaufmann DE, Xu XY, Mui SK, Addo MM, et al. (2004) Loss of HIV-1-specific CD8+ T cell proliferation after acute HIV-1 infection and restoration by vaccine-induced HIV-1-specific CD4+ T cells. J Exp Med 200: 701-712.
15. Rosenberg ES, Alfheid M, Poon SH, Phillips MN, Wilkes BM, et al. (2000) Immune control of HIV-1 after early treatment of acute infection. Nature 407: 523-526.
16. Kaufmann DE, Lichterfeld M, Alfheid M, Addo MM, Johnston MN, et al. (2004) Limited durability of viral control following treated acute HIV infection. PLoS Med 1: e36. doi:10.1371/journal.pmed.0010036
17. Hecht FM, Wang L, Collier A, Little S, Markowitz M, et al. (2006) A multicenter observational study of the potential benefits of initiating combination antiretroviral therapy during acute HIV infection. J Infect Dis 194: 725-735.
18. Strecker H, Jessen H, Alter G, Teigen N, Schacker T, et al. (2000) Lack of mucosal immune reconstitution during prolonged treatment of acute and early HIV infection. J Infect Dis 190: 594-601.
19. Prazuck T, Lafeuillade A, Hocqueloux L, Parrot E, Joos B, et al. (2007) Adjunctive passive immunotherapy in human immunodeficiency virus type 1-infected individuals treated with antiviral therapy during acute and early infection. J Virol 81: 11016-11025.
20. Markowitz M, Jin X, Hurley A, Simon V, D’Cruz R, et al. (2002) Discontinuation of antiretroviral therapy commenced early during the course of human immunodeficiency virus type 1 infection, with or without adjunctive vaccination. J Infect Dis 186: 634-645.
21. Kimlodelle I, Hoen B, Smith DE, Autran B, Lampe FC, et al. (2005) Impact of therapeutic immunization on HIV-1 viremia after discontinuation of antiretroviral therapy initiated during acute infection. J Infect Dis 192: 607-617.
22. Mehandru S, Vellens B, Lin D, Perng Y, MarraRnam B, et al. (2007) Adjunctive passive immunotherapy in human immunodeficiency virus type 1-infected individuals treated with antiretroviral therapy during acute and early infection. J Virol 81: 11016-11025.
23. Yang H, Dong T, Turnbull E, Ranainge S, Caudwell B, et al. (2007) Broad TCR usage in functional HIV-1-specific CD8+ T cell expansions driven by vaccination during highly active antiretroviral therapy. J Immunol 177: 607-606.
24. Barber DL, Wherry EJ, Masopust D, Zhu B, Allison JP, et al. (2006) Restoring function in exhausted CD8 T cells during chronic viral infection. Nature 439: 692-697.
25. D’Cruz R, Kaufmann DE, Kiepiela D, Brown JA, Moodley ES, et al. (2006) PD-1 expression on HIV-specific T cells is associated with T-cell exhaustion and disease progression. Nature 439: 350-354.
26. Trautmann L, Joos B, et al. (2006) Upregulation of PD-1 expression on HIV-specific CD8+ T cells leads to reversible immune dysfunction. Nat Med 12: 1198-1202.
27. Ha SJ, Mueller SN, Wherry EJ, Barber DL, Aubert RD, et al. (2006) Enhancing therapeutic vaccine by blocking PD-1-mediated inhibitory signals during chronic infection. J Exp Med 205: 543-555.