HIV-1 Dynamics in Vivo: Virion Clearance Rate, Infected Cell Life-Span, and Viral Generation Time

Alan S. Perelson, Avidan U. Neumann, Martin Markowitz, John M. Leonard, David D. Ho*

$= 1/c + 1/\delta$, can be shown formally (Table 2). The average value of τ for the patients was 2.6 ± 0.8 days (Table 2).

By a heuristic procedure, we found minimal estimates for the average duration of the HIV-1 life cycle and of its intracellular or eclipse phase (from virion binding to the release of the first progeny). The duration of the HIV-1 life cycle, S, is defined as the time from the release of a virion until the release of its first progeny virus; we estimated S by the lag in the decay of HIV-1 RNA in plasma (Fig. 1) after the pharmacologic delay (Table 1) is subtracted. The shoulder in the RNA decay curve is explained by the fact that virions produced before the pharmacologic effect of ritonavir are still infectious and capable of producing, for a single cycle, viral particles that would be detected by the RNA assay. Thus, the drop in RNA concentration should begin when target cells interact with drug-affected virions and do not produce new virions. These "missing virions" would first have been produced at a time equal to the minimum time for infection plus the minimum time for production of new progeny. The estimated values for S were quite consistent for the five patients, with a mean duration of 1.2 ± 0.1 days (Table 2). In steady state, $1/c = 1/NkT_0$ is the average time for infection (Table 2, legend); if this average time is assumed to be greater than the minimal time for infection, then a minimal estimate of the average duration of the intracellular phase of the HIV-1 life cycle is given by $S - (1/c) =$ 0.9 days (*16*).

Previous studies that used potent antiretroviral agents to perturb the quasi steady state in vivo provided a crude estimate of the $t_{1/2}$ of viral decay in which the life-span of productively infected cells could not be separated from that of plasma virions (*1*, *2*). Our results show that the average life-span of a productively infected cell (presumably an activated CD4 lymphocyte) is 2.2 days; thus, such cells are lost with an average $t_{1/2}$ of ~1.6 days (Fig. 2). The life-spans of productively infected cells were not markedly different among the five patients (Table 2), even though individuals with low CD4 lymphocyte counts generally have decreased numbers of virus-specific, major histocompatibility complex class I–restricted cytotoxic T lymphocytes (*17*).

The average life-span of a virion in blood was calculated to be 0.3 days. Therefore, a population of plasma virions is cleared with a $t_{1/2}$ of 0.24 days; that is, on average, half of the population of plasma virions turns over approximately every 6 hours (Fig. 2). Because our analysis assumed that the antiviral effect of ritonavir was complete and that target cells did not recover during treatment, our estimates of the virion clearance rate and infected cell loss rate are minimal estimates (*12*, *16*). Consequently, the true virion $t_{1/2}$ may be shorter than 6 hours. For example, Nathanson and Harrington (*18*) found that monkeys clear the Langat virus from their circulation on a time scale of ~30 min. Thus, the total number of virions produced and released into the extracellular fluid is at least 10.3×10^9 particles per day (*14*); this rate is about 15 times our previous minimum estimate (*1*). At least 99% of this large pool of virus is produced by recently infected cells (*1*, *2*) (Fig. 2). At quasi steady state, the virion clearance rate cV equals the virion production rate $N\delta T^*$. Because c has similar values for all patients studied (Table 1), the degree of plasma viremia is a reflection of the total virion production, which in turn is proportional to the number of productively infected cells T^* and their viral burst size N. The average generation time of HIV-1 was determined to be 2.6 days, which suggests that ~140 viral replication cycles occur each year, about half the number estimated by Coffin (*19*).

It is now apparent that the repetitive replication of HIV-1 (left side of Fig. 2) accounts for ≥99% of the plasma viruses in infected individuals (*1*, *2*, *19*), as well as for the high destruction rate of CD4 lymphocytes. The demonstration of the highly dynamic nature of this cyclic process provides several theoretical principles to guide the development of treatment strategies:

1) An effective antiviral agent should detectably lower the viral load in plasma after only a few days of treatment.

2) On the basis of previous estimates of the viral dynamics (*1*, *2*) and data on the mutation rate of HIV-1 (3.4×10^{-5} per base pair per replication cycle) (*20*) and the genome size (10^4 base pairs), Coffin has cogently argued that, on average, every mutation at every position in the genome would occur numerous times each day (*19*). The larger turnover rate of HIV-1 described in our study makes this type of consideration even more applicable. Therefore, the failure of the current generation of antiviral agents,

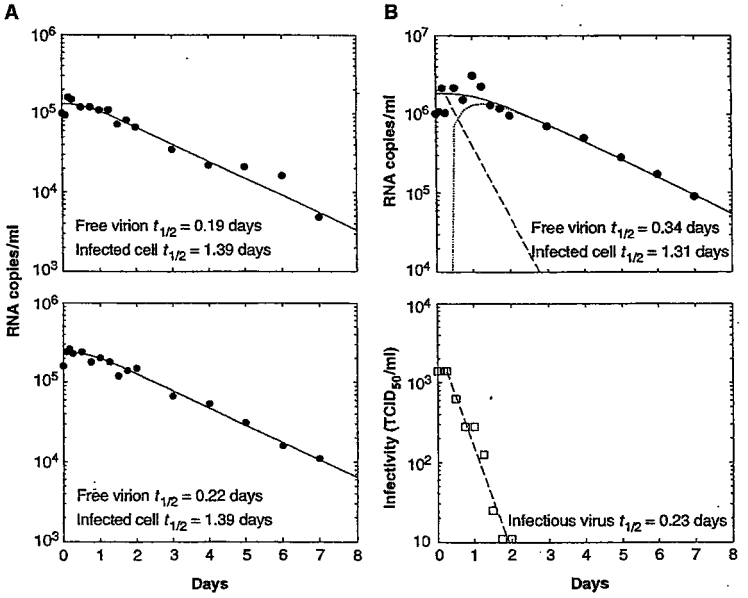
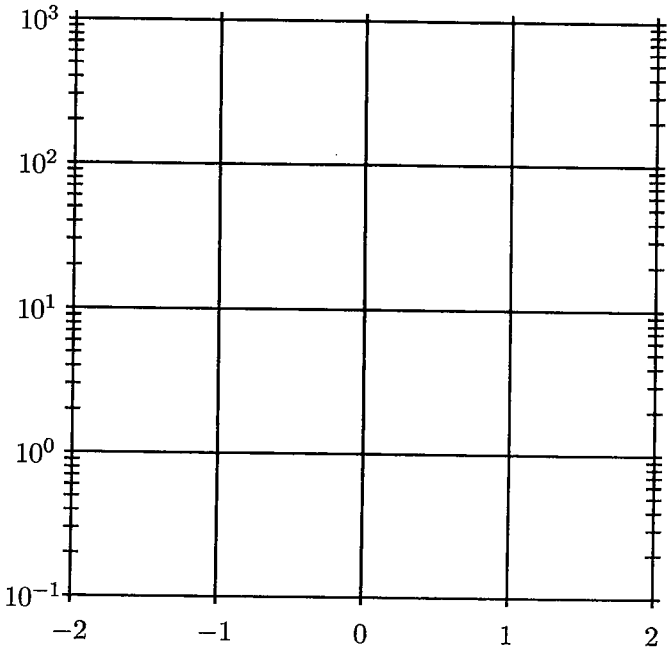


Fig. 1. (**A**) Plasma concentrations (copies per milliliter) of HIV-1 RNA (circles) for two representative patients (upper panel, patient 104; lower panel, patient 107) after ritonavir treatment was begun on day 0. The theoretical curve (solid line) was obtained by nonlinear least squares fitting of Eq. 6 to the data. The parameters c (virion clearance rate), δ (rate of loss of infected cells), and V_0 (initial viral load) were simultaneously estimated. To account for the pharmacokinetic delay, we assumed $t = 0$ in Eq. 6 to correspond to the time of the pharmacokinetic delay (if measured) or selected 2, 4, or 6 hours as the best-fit value (see Table 1). The logarithm of the experimental data was fitted to the logarithm of Eq. 6 by a nonlinear least squares method with the use of the subroutine DNLS1 from the Common Los Alamos Software Library, which is based on a finite difference Levenberg-Marquardt algorithm. The best fit, with the smallest sum of squares per data point, was chosen after eliminating the worst outlying data point for each patient with the use of the jackknife method. (**B**) Plasma concentrations of HIV-1 RNA (upper panel; circles) and the plasma infectivity titer (lower panel; squares) for patient 105. (Top panel) The solid curve is the best fit of Eq. 6 to the RNA data; the dotted line is the curve of the noninfectious pool of virions, $V_{NI}(t)$; and the dashed line is the curve of the infectious pool of virions, $V_I(t)$. (Bottom panel) The dashed line is the best fit of the equation for $V_I(t)$ to the plasma infectivity data. $TCID_{50}$, 50% tissue culture infectious dose.

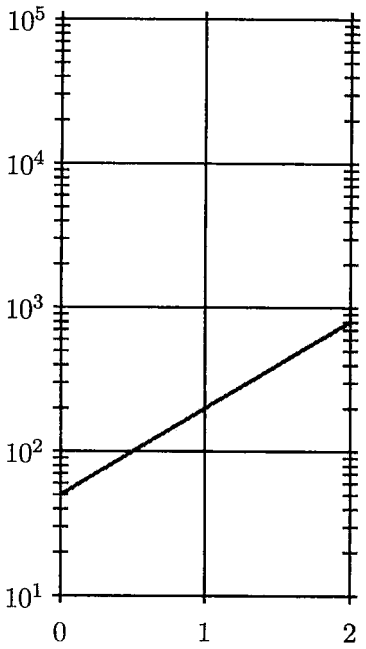
1. Suppose that x and y are related by the expression

$$y = 4 \cdot 10^{-x/2}$$

Use a logarithmic transformation to find a linear relationship between the given quantities and graph the resulting linear relationship on a log-linear (or semilog) plot.



2. Given the semilog plot below, find a functional relationship between x and y.



3. Given the double-log (or log-log) plot below, find a functional relationship between x and y.