

# CD8 memory, immunodominance, and antigenic escape

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Previous theoretical work has suggested that efficient virus control or clearance requires antigen-independent persistence of memory cytotoxic T lymphocyte precursors (CTLp), and that failure to generate such memory CTLp can result in persistent infection and eventual loss of virus control. Here we use mathematical models to investigate the relationship between virus control, the clonal composition of the CTL response and the chance of the virus to evolve antigenic escape. In the presence of efficient memory CTLp, virus is controlled at very low levels by a broad CTL response directed against multiple epitopes. In this case, antigenic escape of the virus population is expected to take a very long time. On the other hand, if the CTL response is short lived in the absence of antigen, virus replicates at higher levels and is only opposed by a narrow CTL response, characterized by an immunodominant CTL clone. In this case, antigenic escape is expected to evolve in a short period of time, resulting in progressive loss of virus control. We discuss our findings in relation to data from HIV-1-infected patients.

**Key words:** Cytotoxic T lymphocyte / Memory / Immunodominance / Antigenic diversity / Mathematical models

Received	28/12/99
Revised	8/5/00
Accepted	6/6/00

## 1 Introduction

Specific CD8 cell responses are a major host defense against viral infections [1–3]. There is some evidence that the level of virus load is determined by the efficacy of CTL [3] and that differences in the CTL responsiveness to viruses can determine the outcome of the disease [2]. Many viruses, however, can establish persistent infections and escape the immune system over time, an important example being HIV [1]. Therefore, it is crucial to explore the principles which determine virus clearance versus persistence, and the processes which contribute to long-term immunological control of an infection. Since the interactions between the immune response and virus replication involve many different components, mathematical models are a useful guide to interpret experimental observations.

We recently suggested that a crucial factor determining virus clearance versus persistence is the quality of the specific CD8 memory response – traditionally associated with protection against re-infection [4]. Mathematical models predict that antigen-independent long-term per-

sistence of readily activated CTL precursors (CTLp) is required to clear viral infections [4]. If the life-span of the CTLp in the absence of antigen is short, the virus can establish a persistent infection. This can be understood intuitively as follows. When the virus population declines because of CD8-mediated activity, the CTLp population also declines, which in turn reduces the immunological pressure on the virus population and enables the virus to settle at a stable equilibrium. In contrast, with antigen-independent persistence of CTLp, the CTL population reaches a stable level, while virus load drops. This maintains continuous pressure on the declining virus population, and will drive the virus population to extinction (or to an extremely low equilibrium abundance). Thus, antigen-independent persistence of CTLp is required for long-term virus control or clearance.

Empirical studies found that the composition of the CTL response might influence the degree of immune-mediated virus control. In HIV infection, long-term non-progressors seem to have broad CTL responses, consisting of multiple CTL clones specific for different epitopes [5]. On the other hand, typical (or fast) progression is associated with narrow CTL responses against one or a few epitopes only [5]. The picture is, however, more complicated as antigenic variation can lead to broad CTL responses and loss of virus control [6].

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**Abbreviation:** CTLp: CTL precursors

Here we use mathematical models to further explore the relationship between long-term immunological control of a virus infection, the breadth of the CTL response, and the chance of the virus to escape CTL-mediated suppression. We find that the presence of a narrow CTL response, characterized by an immunodominant epitope, can be the consequence of a short life-span of the CTL response in the absence of antigen. This in turn means that the CTL response does not efficiently control the infection. In such a scenario, antigenic escape mutants are likely to evolve quickly, shifting the balance between virus replication and the immune response further in favor of the virus. On the other hand, lack of immunodominance can result from a long life-span of the CTLp response in the absence of antigen. This implies efficient long-term immunological control of the infection. In this case, the time required for the virus to evolve antigenic escape is long, and the chances of losing control of the infection are small. Hence, while the presence of an antigenically diverse virus population and progression to AIDS can induce lack of immunodominance [6], a broad CTL response in combination with low virus load indicates efficient long-term immunological control of the infection. Immunodominance is a consequence of either an impaired, short-lived CTL response, or the presence of only one or a few epitopes that can efficiently stimulate the CTL.

## 2 Results

### 2.1 A basic model of CD8 responses to viral infections

The basic interactions between the CD8 cell response and a replicating virus population can be described by ordinary differential equations (see Sect. 4) containing four variables: uninfected cells,  $x$ , infected cells,  $y$ , CTLp,  $w$ , and CTL effectors (CTLe),  $z$ . The model is explained schematically in Fig. 1 a and b. We define CTLe as CD8 cells which can inhibit viral replication, for example by lysing infected cells, at a rate  $p$ . It is assumed that CTLe differentiate from CTLp. They do not proliferate at a significant rate, and are relatively short lived [7]. CTLp are defined as CD8 cells which have been activated and have the capacity to proliferate in response to their specific antigen at a rate  $c$ . However, they do not have antiviral activity. The CTLp are characterized by an average life-span of  $1/b$ .

If the basic reproductive ratio of the virus is large enough, and the CTL responsiveness is sufficiently strong, the system converges towards an equilibrium describing a persistent virus infection controlled by the CD8 cell response. In this case a reduction in virus load is

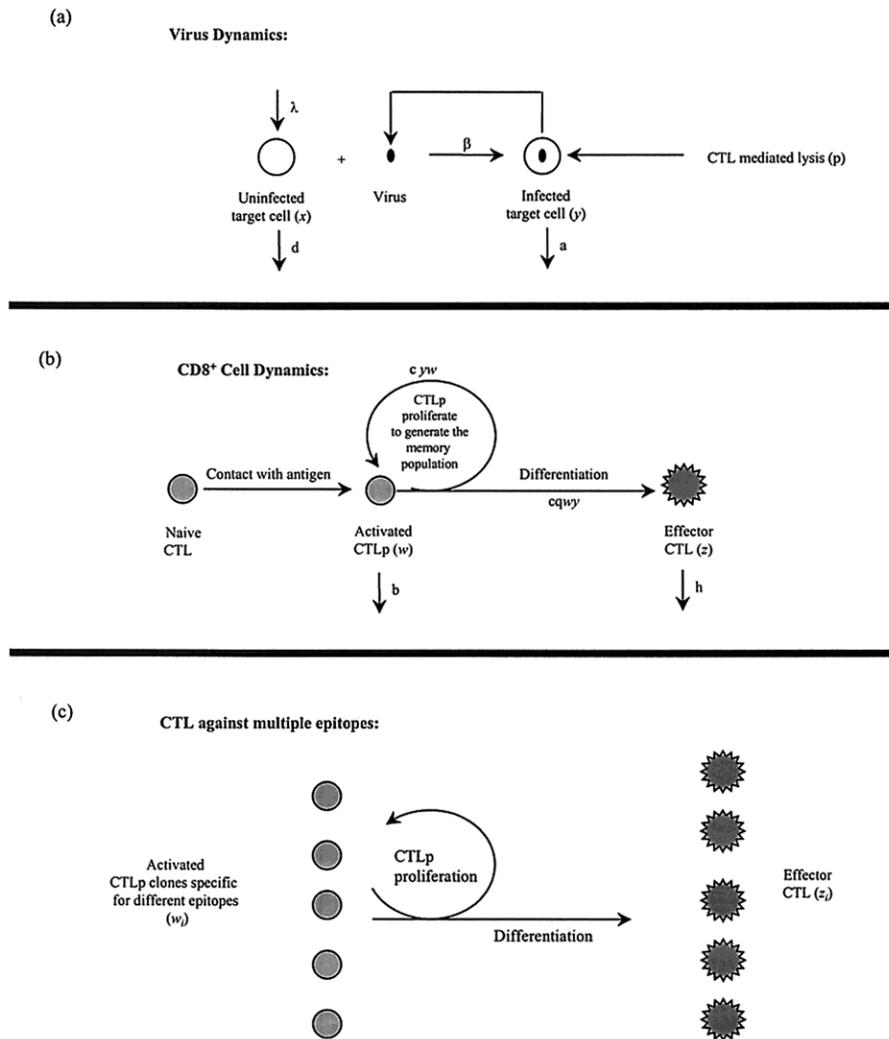
achieved by a long life-span of TCLp in the absence of antigen, and by a high activation rate of the CTLp population (high  $1/b$  and  $c$ ). Hence, virus control is associated with long-term, antigen-independent persistence of an efficient CTL memory response [4].

In addition to the equilibrium analysis, the model shows the interesting property that for a long-life-span of the CTLp response in the absence of antigen, the system remains at a quasi-equilibrium for a time span of approximately  $1/b$  (see Sect. 4). During this phase, virus load decays and the number of CTLp increases at an extremely slow rate, while the number of CTLe remains stable (Fig. 2). Virus load at the quasi-equilibrium has the same properties as virus load at the true equilibrium, although it is slightly higher [4]. Hence, denoting virus load at the true equilibrium by  $y^*$  and virus load at the quasi-equilibrium by  $\hat{y}$ , we can write  $\hat{y} = ay^*$ , where  $a > 1$ . Further mathematical details about the model are found in the appendix.

### 2.2 CTL responses against multiple epitopes

We extend the above model to include multiple CTL clones ( $i = 1..n$ ) directed against different epitopes of the same virus population. We assume that the CTL clones are characterized by different responsiveness to the antigen,  $c_i$ , and by different rates of target cell killing,  $p_i$ . For simplicity we assume that the CTL responsiveness is positively correlated with the rate of target cell killing, since both parameters are determined by recognition of antigen in conjunction with MHC. However, the results do not depend on this assumption. The model is explained schematically in Fig. 1 c. In such a scenario, the CTL clones are essentially in competition with each other [6]. Competitive ability correlates with the CTL responsiveness of the epitope,  $c_i$ . The CTL clone directed at the epitope with the largest  $c_i$  is the most superior competitor. The outcome of these competitive interactions depends the life-span of the CTL response in the absence of antigen, i. e. on the parameter  $1/b$ .

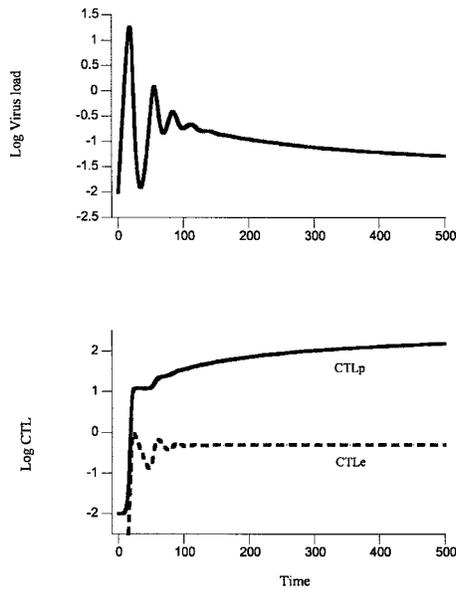
According to the equilibrium expressions of the model (see Sect. 4), only the clone with the highest CTL responsiveness,  $c_i$ , can survive at significant levels. Mathematically speaking, all other CD8 clones go extinct, although in practical terms, the spatial environment of the immune system could result in persistence of these clones at low levels. The reason is that the competitively superior CTL clone reduces virus load to levels too low to stimulate the weaker CTL clones. This result will be obtained if the life-span of the CTLp response in the absence of antigen ( $1/b$ ) is short. Thus, lack of efficient CTL-mediated control of the infection correlates with the presence of an immunodominant CTL clone (Fig. 3 a).



**Fig. 1.** Schematic representation of mathematical models. For mathematical details, see Sect. 4. (a) Basic virus infection model. Uninfected cells react with virus to form infected cells, and infected cells produce new virus particles. (b) CD8 T cell dynamics. CTLp proliferate in response to antigen and differentiate into effector cells. Effector cells exert antiviral activity by lysing infected cells. However, the conclusions drawn from this model remain identical for non-lytic activity of CD8 T cells. (c) Dynamics of multiple CTL clones directed against different epitopes. All CTL clones proliferate and differentiate in response to the same virus population, and are therefore in competition with each other.

On the other hand, if the life-span of the CTLp response in the absence of antigen is long (high  $1/b$ ), the dynamics between virus replication and the CTL response converge to a quasi-equilibrium. Similar to the basic model discussed above, virus load at the quasi equilibrium ( $\hat{y}$ ) is higher than virus load at the true equilibrium ( $y^*$ ), *i.e.*  $\hat{y} = ay^*$ , where  $a > 1$ . Now, coexistence of multiple CTL clones directed against different epitopes is possible (Fig. 3 b). This is because the competitively superior CTL clone does not reduce virus load down to the true equilibrium, but only to the quasi-equilibrium, where virus load may still be sufficient to stimulate the weaker CTL

clones. We can define this more precisely by ranking the CTL clones against different epitopes according to their competitive ability, *i.e.*  $c_1 > c_2 > c_3 > \dots > c_n$ . If the life-span of the CTLp is long, then the dynamics of the CTL clones directed against the different epitopes can be described by  $w_i(t) = \exp[-bt(1-ac_i/c_1)]$ . From this it follows that the CTL clone directed against epitope  $i$ ,  $w_i$ , persists during the quasi-equilibrium if  $c_i a > c_1$ . (For further details, see Sect. 4). In summary, efficient long-term virus control does not lead to immunodominance, but to a broad CTL response directed against multiple epitopes.

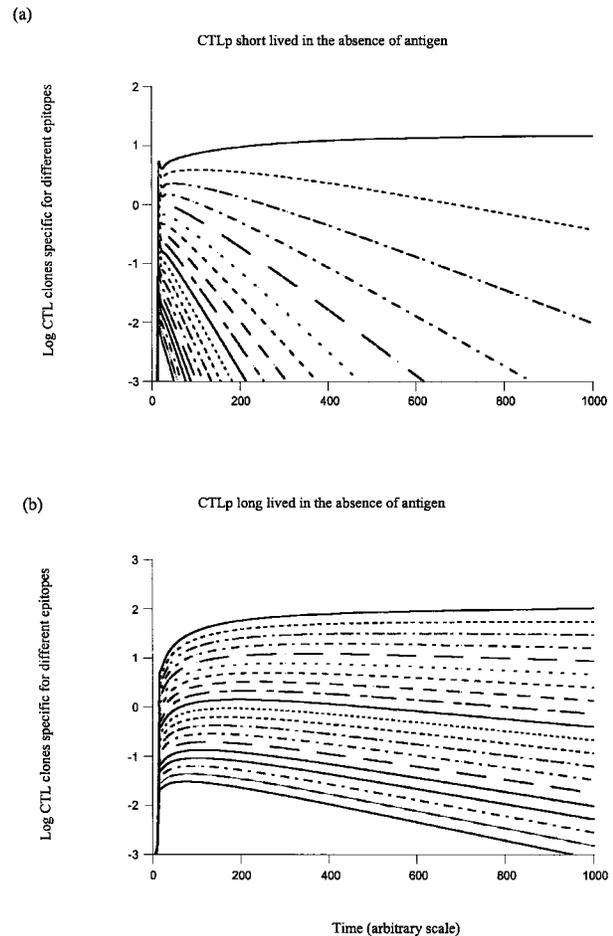


**Fig. 2.** Quasi-equilibrium dynamics in the basic model assuming a homogeneous CTL population that is relatively long-lived at low levels of antigen (low  $b$ ). After an initial and transient phase of oscillations, the system attains a quasi-equilibrium at which virus load declines at a very slow rate, and the number of CTLp increases at a very slow rate. The true equilibrium is reached after a time span of approximately  $1/b$ . Parameters were chosen as follows:  $\lambda = 10$ ;  $d = 0.1$ ;  $\beta = 0.01$ ;  $a = 0.5$ ;  $p = 1$ ;  $c = 0.063$ ;  $b = 0.0015$ ;  $q = 0.1$ ;  $h = 0.1$ .

### 2.3 Antigenic variation and immune escape

If the host establishes a CTL response that can efficiently control virus replication and prevent development of disease, the question is how long this control can be maintained, especially with highly variable pathogens such as HIV [1]. Antigenic variation could result in escape from the CTL response. In this section, we analyze the chances of antigenic escape mutants evolving, depending on the life-span of the CTLp response in the absence of antigen. As discussed in the previous section, a slow decay rate of the CTLp response in the absence of antigen results in improved virus control and the development of a broad CTL response. We can derive the rate at which escape mutations are generated, and thus the average time until the virus population remains under CTL-mediated control.

We consider two scenarios. On the one hand, the virus may not require activation of its target cells to complete its replication cycle. An example is macrophage infection by HIV [8]. On the other hand, target cell activation can be a requirement for successful viral replication. This is especially true for CD4 T cell-infecting viruses, such as



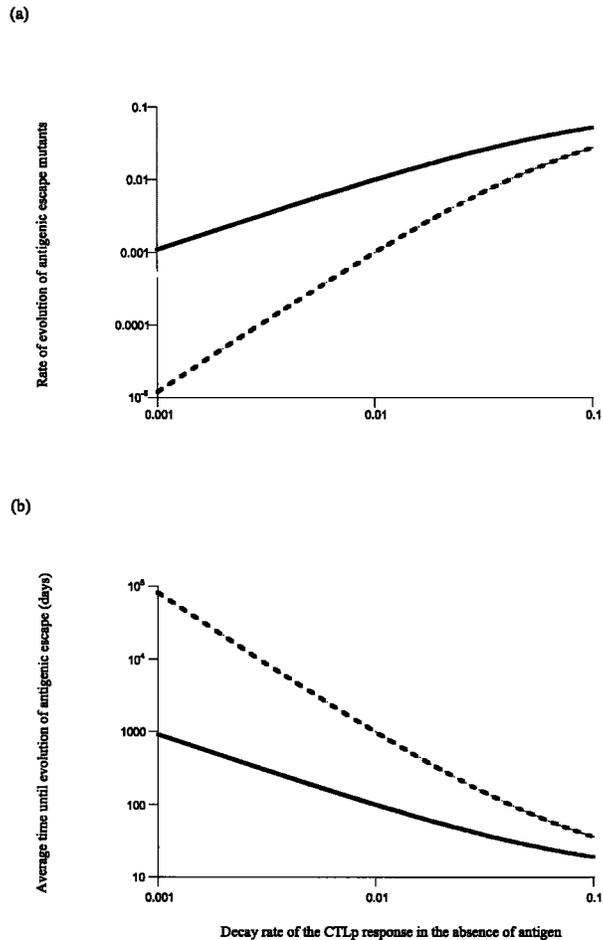
**Fig. 3.** Relationship between the longevity of the CTL response in the absence of antigen and the clonal composition of the CTL response. (a) If CTLp decay at a fast rate in the absence of antigen (large  $b$ ), most CTL clones are out-competed by the most immunogenic clone, resulting in immunodominance. (b) If CTLp decay at a slow rate in the absence of antigen (small  $b$ ), we observe coexistence of multiple CTL clones directed against different epitopes. Baseline parameters were chosen as follows:  $n = 20$ ;  $\lambda = 10$ ;  $d = 0.1$ ;  $\beta = 0.01$ ;  $a = 0.5$ ;  $q = 0.1$ ;  $h = 1$ ;  $c_i = 1 + 0.1i$ ;  $p_i = c_i$ . For (a),  $b = 0.1$ ; for (b)  $b = 0.007$ .

HIV or HTLV [8]. For details regarding mathematical expressions, see Sect. 4.

#### 2.3.1 Viral replication without target cell activation

In the model, the rate of generation of escape mutants is proportional to  $\beta\mu x^*y^*$ . The quantity  $x^*$  denotes the number of uninfected cells at equilibrium, while  $y^*$  denotes virus load at equilibrium. The parameter  $\beta$  represents the rate of viral replication and  $\mu$  stands for the mutation rate

of the virus. Fig. 4 shows the rate of mutation depending on the decay rate of the CTLp response in the absence of antigen. Decreasing the decay rate of the CTL response in the absence of antigen reduces virus load to



**Fig. 4.** Relationship between the longevity of the CTL response in the absence of antigen and the evolution of antigenic escape mutants. This is shown both for viruses requiring cell activation for infection (dashed line) and viruses that can replicate independently of cell activation (solid line). (a) Rate of evolution of escape mutants. The slower the decay rate of CTLp in the absence of antigen, and the better the degree of virus control, the slower the generation of escape mutants. This effect is more pronounced for viruses requiring cell activation for replication. (b) Average time span until the emergence of escape mutants. The slower the decay rate of CTLp in the absence of antigen, the longer the time until escape emerges, and the longer the time the virus remains under efficient immunological control. This effect is more pronounced for viruses requiring target cell activation for infection. Baseline parameters were chosen as follows:  $\lambda = 10$ ;  $d = 0.1$ ;  $\beta = 0.1$ ;  $a = 0.2$ ;  $q = 0.1$ ;  $h = 1$ ;  $c = 0.1$ ;  $p = 1$ ;  $\mu = 0.01$ .

low levels, while increasing the number of uninfected cells asymptotically towards its natural set point. Hence, the longer lived the CTLp response, the slower the rate of generation of escape mutants because the overall level of viral replication is reduced (Fig. 4). The average time period required to evolve escape is proportional to  $1/\beta\mu x^*y^*$ . Thus, the longer the life-span of the CTLp response in the absence of antigen, the lower the virus load, the broader the CTL response, and the longer the average time span in which escape does not evolve and the virus remains under efficient immunological control (Fig. 4). Note that in the model the slow rate of evolution of escape is not a direct consequence of the broad CTL response. It is a direct consequence of the long life-span of memory CTLp, and this also promotes a broad CTL response.

### 2.3.2 Viral replication requires target cell activation

Many viruses need cell activation to complete their replication cycle. A typical example is T cell infection by HTLV and HIV. This adds another dimension to the dynamics of viral replication: activation of the target cells is increased by the presence of viral antigen. A reduction of virus load to low levels will be accompanied by a reduction of the number of activated uninfected T cells. Thus, low virus load is accompanied by limited amounts of target cells in which the virus can replicate and mutate. Hence, the rate generation of escape mutants will be approximately proportional to  $\beta\mu x^*y^{*2}$ , and the time-period required to evolve escape is approximately proportional to  $1/\beta\mu x^*y^{*2}$ . If the CTL response is long-lived in the absence of antigen, the time until escape emerges is longer than predicted by the model in which viral replication is not dependent on target cell activation (Fig. 4). This is because both the number of infected and susceptible cells become limiting. Again, the slow rate of emergence of escape mutants in this model is a consequence of the long life-span of the memory CTLp (limiting the number of susceptible cells), and this is associated with a broad CTL response.

## 3 Discussion

We have used mathematical models to investigate the relationship between virus control, immunodominance, and the chance for the virus to escape the CTL response. As in previous studies [4] we found that virus control is achieved by antigen-independent persistence of readily activated CTLp. Our models predict that a short life-span of the CTLp in the absence of antigen results in less efficient immunological control, associated

with the presence of an immunodominant CTL clone. This is because the CTL clone with the highest immune responsiveness can out-compete all the inferior CTL clones. In this situation, escape mutants are expected to evolve in a short period of time. On the other hand, a long life-span of the CTLp in the absence of antigen leads to efficient long-term virus control and does not result in immunodominance. Instead we observe the coexistence of multiple CTL clones directed against different viral epitopes. In addition, the time period required to evolve antigenic escape is long. The more virus load is suppressed by CTL, the slower the evolution of antigenic escape, and the longer the time span during which the infection remains under control. Low virus load can result in the reduced availability of susceptible target cells, and this further slows down the overall rate of viral replication. Antigenic escape and progressive increase in virus load is most likely if CTL-mediated virus control is inefficient in the first place.

These predictions are supported by data from HIV-infected individuals. One patient, a long-term non-progressor, had strong CTL responses, low viral load, and a stable CD4 T cell count of  $> 500$  cells/ $\mu$ l 15 years after infection [5]. There was a broad CTL response directed against multiple epitopes and maintained at high levels despite the low level of viraemia [5]. This supports the prediction that broad CTL responses can result from a long life-span of CTLp in the absence or at low levels of antigen. On the other hand, faster progressing patients are characterized by narrow CTL responses [9]. Although CTL are present during the asymptomatic phase, these CTL seem short-lived in the absence of antigen [10]. Starting anti-retroviral therapy in these patients results first in a transient increase of CTLp, followed by a rapid decline to low levels. The temporary increase in CTL numbers is likely to be caused by a trade-off between antigenic stimulation and virus-induced immune impairment. When viral replication is stopped, the amount of immune impairment is reduced, and before virus load has significantly declined, the number of CTL can increase. Once virus load drops to low levels the antigenic stimulus is removed. The observation that the level of CTLp then also declines indicates that many of the CTLp seen in HIV-infected patients are short lived at low levels of antigen.

Previous theoretical work [6] suggested that lack of immunodominance can be a consequence of an antigenically heterogeneous virus population. This hypothesis was supported by data from an HIV-infected patient with a relatively low CD4 T cell count of around 200 cells/ $\mu$ l [6]. Hence, the presence of CTL directed against multiple epitopes can indicate two alternative situations, depending on the level of virus load found in the patient:

(i) if virus load is low, a broad CTL response results from the presence of efficient memory CTL that are long-lived in the absence of antigen, and have the potential to control virus replication in the long-term; (ii) if virus load is high, the presence of CTL directed against multiple epitopes indicates heterogeneity in the virus population caused by antigenic variation. The same study [6] also suggested that a narrow CTL response indicates the presence of a homogeneous virus population and stable virus control. This hypothesis was supported by a patient with a higher CD4 T cell count. However, although the CD4 T cell count declined slowly in this patient, it reached about 300 cells/ $\mu$ l after about 50 months. Hence, this patient cannot be classified a long-term non-progressor, and corresponds better to a typical progressor. According to the findings presented here, the narrow CTL response in this patient indicates that the CTLp are short-lived in the absence of antigen, which in turn could be the reason for the steady loss of virus control and decline of the CD4 T cell count. Indeed, there is evidence that in the presence of narrow CTL responses, antigenic escape mutants are likely to arise [9]. Emergence of escape mutants can result in increased viral loads, and evolution of antigenic escape has been associated with the development of AIDS [9].

However, as discussed above, antigenic heterogeneity can result in coexistence of CTL clones directed against multiple epitopes. Hence, the narrow CTL response in patients with weak CTL memory might only be transient, and likely to be observed relatively early in the disease process. As virus replication increases while the patient progresses towards AIDS, accumulation of antigenic diversity could in principle trigger a broadening of the responses, associated with increased viral loads. However, this hypothesis needs further investigation. Since progressing patients are characterized by the absence of significant levels of T cell help, and by an overall weakened immune system, it is difficult to tell whether significant levels of CTL would have the capacity to expand in response to antigenic variation. Furthermore, if anti-viral immunity is weak, competition among the different virus strains could lead to the dominance of fast replicating strains despite the overall presence of antigenic heterogeneity, and this could again skew the CTL response towards immunodominance.

The implications of the breadth of the CTL response are summarized in Table 1. Our main conclusion is that a broad CTL response can develop in the presence of an efficient CTL memory response, even if the virus population is homogeneous. On the other hand, in the absence of significant CTL memory, a homogeneous virus population results in dominance of one or only a few CTL clones. Apart from the above considerations, a narrow

**Table 1.** Breadth of the CTL response and virus control

	Narrow CTL response	Broad CTL response
Homogenous virus population	<ul style="list-style-type: none"> <li>• CTLp short lived in the absence of antigen and inefficient virus control;</li> <li>• Super-immunogenic epitope</li> </ul>	CTLp long-lived in the absence of antigen and efficient virus control
Heterogeneous virus population	CTLp short lived in the absence of antigen and inefficient virus control;	<ul style="list-style-type: none"> <li>• If virus load low: CTLp long-lived in the absence of antigen and efficient virus control</li> <li>• If virus load high: loss of virus control due to antigenic variation</li> </ul>

CTL response could also arise in specialized circumstances that have not been taken into account in our models. In HTLV-1 infection, a single dominant CTL response directed against the TAX protein is observed [11]. While this could result from a CTLp response that is short-lived in the absence of antigen, another explanation is that TAX is the only antigen exposed to CTL, and that infected cells are killed before any other viral proteins can be presented on the cell surface (C. R. M. Bangham, personal communication). In addition, the presence of specific viral antigens that are highly immunogenic compared to alternative viral antigens could result in a narrow CTL response, even if it is efficient and can control viral replication in the long term.

Our models provide insights for improving the design of treatment regimes that may result in long-term immunological control of HIV [12, 13]. Recent experimental evidence [14] suggests that in primary HIV infection, the high virus loads results in significant impairment of the T helper cell response, measured by anti-viral CD4 cell proliferative responses. Data, together with theoretical findings [15] suggest that the absence of CD4 cell help in the primary infection results in the failure to establish significant levels of CTL memory, defined by long-term persistence of CTLp in the absence of antigen. Recent studies on SIV-infected macaques [13] and HIV-infected patients [12] suggest that treatment during primary infection, in combination with regimes involving intermittent therapy, can turn fast progressors into a similar state as long-term nonprogressors. Virus control in these cases is

associated with more efficient CD4 cell proliferative responses. Theory suggests that these treatment regimes can facilitate the establishment of an efficient CTL memory response which can potentially control virus replication at low levels and prevent virus-induced pathogenesis. In accordance with our current findings, subjects which successfully control virus replication are characterized by broad CTL responses [12]. This indicates that these treatment regimes results in the development of virus-specific CTL that have a long life-span in the absence of antigen. These findings suggest that improved T helper cell responses brought about by therapy can convert and expanding primary CTL population that is short-lived into a memory CTL response that is maintained in the absence of antigen [15]. The hypothesis could be tested in HIV-infected patients or SIV-infected macaques undergoing intermittent therapy regimes. After these treatment schedules have resulted in improved virus control, subjects should be put back on therapy and the decay rate of CTLp at low levels of antigen monitored. If our hypothesis is correct, the decay rate of CTLp at low levels of antigen should diminish with repeated cycles of intermittent therapy and improved virus control.

## 4 Materials and methods

### 4.1 General remarks

Due to space constraints, only essential details are given in this section. Complete mathematical details can be found on the following web page: <http://www.admin.ias.edu/ptb/>

### 4.2 Basic dynamics between CD8 cells and virus replication

To analyze the dynamics of anti-viral CTL responses, we use the basic virus infection model taking into account uninfected ( $x$ ) and infected ( $y$ ) host cells. We assume that the CTL pool consists of two populations: the precursors ( $w$ ) and the effectors ( $z$ ). The model is given by the following set of differential equations:

$$\begin{aligned}
 \dot{x} &= \lambda - dx - \beta xy \\
 \dot{y} &= \beta xy - ay - pyz \\
 \dot{w} &= cyw(1-q) - bw \\
 \dot{z} &= cqyw - hz
 \end{aligned} \tag{1}$$

Target cells are produced at a rate  $\lambda$ , die at a rate  $dx$  and become infected by virus at a rate  $\beta xy$ . Infected cells die at a rate  $ay$  and are killed by CTL effector cells at a rate  $pyz$ . Upon contact with antigen, CTLp proliferate at a rate  $cyw$  and differentiate into effector cells at a rate  $cqyw$ . CTL precursors die at a rate  $bw$ , and effectors die at a rate  $hz$ . If the basic reproductive ratio of the virus ( $R_0 = \beta\lambda/da$ ) is greater than unity, the virus may establish a persistent infection.

Virus replication in the absence of immunity is described by equilibrium E1

$$x^{(1)} = \alpha/\beta, y^{(1)} = \lambda/a - d/\beta, w^{(1)} = 0, z^{(1)} = 0.$$

If the CTL response is strong enough to expand [i.e. if  $c(1-q)y^{(1)} > b$ ], then the system converges to the following equilibrium.

$$x^{(2)} = \frac{\lambda c(1-q)}{dc(1-q) + b\beta}, y^{(2)} = \frac{b}{c(1-q)}, w^{(2)} = \frac{z^{(2)}h(1-q)}{bq},$$

$$z^{(2)} = \frac{\beta x^{(2)} - a}{p}$$

For low values of  $b$ , the system takes a long time to equilibrate. After an initial transient phase, the dynamics lead to a quasi-equilibrium at  $(y^1)$  at which virus load decays only at a very small rate. Virus load at the quasi-equilibrium is higher than at the true equilibrium, but has similar properties. Hence, virus load at the quasi-equilibrium can be approximated by  $y^* = ay^{(2)}$ , where  $\alpha > 1$ . After a time period of  $1/b$ , the system approaches the true equilibrium,  $y^{(2)}$ .

### 4.3 Multiple epitopes

The basic model can be extended to include multiple CTL clones ( $i = 1..n$ ) specific for different viral epitopes of the same virus population. We assume that the CTL clones differ in their responsiveness to the antigen,  $c_i$ , and the rate of target cell lysis,  $p_i$ , where  $c_i$  and  $p_i$  are likely to be correlated. The model is given by the following set of differential equations:

$$\dot{x} = \lambda - dx - \beta xy$$

$$\dot{y} = \beta xy - ay - y \sum_{i=1..n} p_i z_i$$

$$\dot{w}_i = c_i y w_i (1-q) - b w_i$$

$$\dot{z}_i = c_i q y w_i - h z_i$$

Assuming the existence of  $n$  CTL clones specific for different epitopes, we can rank them according to their competitive abilities, expressed by the value of  $c_i$ :  $c_1 > c_2 > c_3 > \dots > c_n$ . If  $y$  has equilibrated,  $w_i$  declines with an exponential rate, described by  $w_i(t) = e^{-b \left(1 - \frac{c_i}{c_1}\right) t}$ .

Only the most superior competitor survives. However, if  $b$  is small, the system takes a very long time to equilibrate. The system will remain at a quasi-equilibrium for a time-span of approximately  $1/b$ . Virus load at the quasi-equilibrium has the same properties as virus load at equilibrium, but is characterized by a higher value. That is  $y^* = \alpha \frac{b}{c(1-q)}$ , where  $\alpha > 1$ . In this case,  $w_i$  declines at a rate  $w_i(t) = e^{-b \left(1 - \frac{c_i}{c_1}\right) t}$ . Hence, the CTL clone  $w_i$  persists if  $c_i \alpha > c_1$ .

### 4.4 A model including target cell activation

Certain viruses require activation of their target cells in order to complete their replication cycle. This is especially true for retroviruses with a tropism for CD4 T cells. We can extend the basic model describing the dynamics between virus replication and CTL to take this additional feature into account. The model is given by the following set of equations.

$$\dot{x} = \frac{\lambda y}{\epsilon y + 1} - dx - \beta xy$$

$$\dot{y} = \beta xy - ay - p y z$$

$$\dot{w} = c y w (1-q) - b w$$

$$\dot{z} = c q y w - h z \quad (3)$$

In this model, the rate of target cell production is not constant, but depends on the level of virus load providing an activation stimulus. This model has the interesting property that the establishment of an infection depends on a complex balance of host and viral parameters and initial conditions. This is explored in detail in [16]. Here, we are interested in the equilibrium expression describing persistent virus replication in the presence of a CTL response. They are given by:

$$x^* = \frac{\lambda b c (1-q)}{[c(1-q) + b] [dc(1-q) + b\beta]}, y^* = \frac{b}{c(1-q)}, w^* = \frac{z^* h}{c p y^*},$$

$$z^* = \frac{\beta x^* - a}{p}$$

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