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Supplementary Information for

Regulation of T cell expansion by antigen presentation dynamics

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Supporting Information Text

1. Competitive binding model

The preinfection T cell repertoire specific to an antigen is typically broad consisting of many T cell clones of varying affinities. Competition between all of these T cells for binding to a limited number of pMHC complexes can reduce T cell proliferation. To make our work self-contained we derive a mathematical framework to analyze how such competition influences T cell expansion, largely following earlier work by Perelson and DeBoer (1) and Borghans et al. (2).

A. General framework. We consider the following dynamical equations to describe the expansion of multiple T cell clones competing for the cognate antigen:

$$\frac{\mathrm{d}T_i}{\mathrm{d}t} = \alpha B_i - \delta T_i, \quad [1]$$

$$\frac{\mathrm{d}C}{\mathrm{d}t} = -\mu C. \quad [2]$$

The first equation describes the dynamics of the number of T cells, T_i, in the i^{th} clone, with i running from 1 to the number of clones n that have significant affinity for the antigen. All T cells are assumed to proliferate at a rate α upon binding to pMHC complexes; thus the overall rate of proliferation of T cells in the i^{th} clone is proportional to the number of bound T cells B_i. All T cells are assumed to die at a rate δ. The second equation describes the dynamics of the number of cognate pMHCs, C. The pMHCs are presented on the surface of antigen-presenting cells and are assumed to decay at a rate μ.

The specific form of the proliferation term in Eq. 1 can be derived by considering the following scheme (1) which resembles an enzymatic reaction:

$$T_{ui} + C_u \underset{k_{di}}{\overset{k_{ai}}{\rightleftharpoons}} B_i \overset{k_{pi}}{\rightarrow} 2T_{ui} + C_u, \quad [3]$$

where T_{ui} is the number of unbound T cells in the i^{th} clone, C_u is the number of unbound pMHCs, and k_{ai}, k_{di} and k_{pi} are, respectively, rate constants for association and dissociation of the bound complex B_i and for the proliferation of T cells in i^{th} clone upon binding. After the T cell is stimulated to divide it dissociates from the APC leaving the pMHC unbound.

For mathematical simplicity we assume here that a T cell binds to a single pMHC. The derivation can be generalized to the more realistic situation in which the T cell binds on the surface of an antigen-presenting cell. In this more general setting, T cells compete for binding to different binding sites on the antigen presenting cells, where each cellular-level interaction is mediated by multiple T cell receptor - pMHC interactions (1). The association and dissociation rates k_{ai} and k_{di} then represent effective rates of assembling and disassembling an overall bound complex between a number of receptors on the T cell and a number of pMHCs on the antigen presenting cell.

The dynamical equations corresponding to the above scheme are:

$$\frac{\mathrm{d}T_{ui}}{\mathrm{d}t} = (2k_{pi} + k_{di})B_i - k_{ai}T_{ui}C_u, \quad [4]$$

$$\frac{\mathrm{d}B_i}{\mathrm{d}t} = k_{ai}T_{ui}C_u - (k_{pi} + k_{di})B_i. \quad [5]$$

The total number of T cells in the i^{th} clone is the sum of the number of unbound T cells and bound complexes $T_i = T_{ui} + B_i$, so that adding the above two equations together we have

$$\frac{\mathrm{d}T_i}{\mathrm{d}t} = k_{pi}B_i, \quad [6]$$

which gives rise to the proliferation term in Eq. 1 after setting k_{pi} to α.

Assuming that the binding/unbinding kinetics is much faster than the proliferation of T cells, we adopt the quasi-steady-state approximation $dB_i/dt = 0$ for all the bound complexes. This leads to the following equation

$$T_{ui}C_u - K_iB_i = 0, \quad [7]$$

where $K_i = (k_{pi} + k_{di})/k_{ai}$ is the saturation constant related to the affinity between the T cell receptors in the i^{th} clone and the pMHC. Note that when unbinding is fast compared to proliferation ($k_{di} \gg k_{pi}$), K_i is approximately equal to the ratio of off- to on-binding rates. Expressing the number of unbound T cells and pMHCs in terms of the total numbers of T cells and pMHCs and of bound complexes, $T_{ui} = T_i - B_i$ and $C_u = C - \sum B_i$, we finally obtain an equation relating the number of bound complexes to the total numbers of T cells T_i and of pMHCs C and the saturation constants K_i:

$$(T_i - B_i)(C - \sum_{j=1}^{n} B_j) - K_iB_i = 0. \quad [8]$$

The above set of equations (with i running from 1 to n) can be readily solved numerically to find $\{B_i\}$ given $\{T_i\}$, C and $\{K_i\}$. Analytical solutions are also possible in special cases, e.g. when competition only involves one or two T cell clones or for certain limiting cases (see SI Text B to D).

B. Competitive binding for a single T cell clone. Following the general framework, we first consider the simplest case of competitive binding within a single population of T cells, all with equal affinity for the cognate antigen. This case can be relevant in practice. For example, in experiments a single clone of transgenic T cells and cognate antigens with high specificity for each other may be transferred together (3), or in a natural setting one of the T cell clones in a preinfection repertoire may have a much higher specificity for a particular pathogen-derived antigen than the other clones, so that competition occurs mainly within this highly specific population of T cells.

From Eqs. 1 and 2, the dynamical equations for the expansion of a single T cell clone are

$$\frac{\mathrm{d}T}{\mathrm{d}t} = \alpha B(T, C, K) - \delta T, \quad [9]$$

$$\frac{\mathrm{d}C}{\mathrm{d}t} = -\mu C, \quad [10]$$

where the analytical expression $B(T, C, K)$ can be derived from the single-clone version of Eq. 8:

$$B^2 - (K + T + C)B + TC = 0. \quad [11]$$

This quadratic equation has the solution

$$B = \frac{1}{2}\left[K + T + C - \sqrt{(K + T + C)^2 - 4TC}\right], \quad [12]$$

where the choice of the minus sign is determined by the conditions that the number of bound complexes is smaller than the total numbers of T cells and pMHCs, $B < T$ and $B < C$. When $TC \ll (K + T + C)^2$, we can expand the exact solution Eq. 12 with respect to the small parameter $TC/(K + T + C)^2$. This leads to a convenient approximate solution for B:

$$B = \frac{TC}{K + T + C}. \quad [13]$$

Eq. 13 has been obtained previously in (1). (The largest deviation between the exact and approximate expressions occurs when $K \to 0$ and $T = C$, where there is a factor of 2 difference between the exact solution $B = T$ and the approximate solution $B = T/2$.) Substituting the approximate solution Eq. 13 into Eq. 9, we obtain Eqs. 1 and 2 in the main text for the dynamics of the T cell and pMHC numbers.

C. Competitive binding for two T cell clones. T cell dynamics becomes more nuanced when two T cell clones with different affinities compete for binding to the same antigen. When the antigens are abundant, all T cells can proliferate freely. However, as the level of antigen decays, competition can differentially reduce the proliferation of T cells in the different clones. The expansions of two competing T cell clones are described by Eqs. 1 and 2 with the total number of clones $n = 2$. The specific forms of the numbers of the bound complexes B_1 and B_2 in the dynamical equations can be derived from Eq. 8:

$$(T_1 - B_1)(C - B_1 - B_2) - K_1 B_1 = 0, \quad [14]$$

$$(T_2 - B_2)(C - B_1 - B_2) - K_2 B_2 = 0. \quad [15]$$

Since the above two equations are symmetric with respect to interchange of the subscript indices 1 and 2, the analytical expressions for B_1 and B_2 must preserve the symmetry, i.e., $B_1 = f(T_1, T_2, C, K_1, K_2)$ and $B_2 = f(T_2, T_1, C, K_2, K_1)$. Therefore, we first obtain the solution for B_1, and the solution for B_2 can then be found by interchanging the subscripts 1 and 2 in B_1. Combining Eqs. 14 and 15, we obtain a cubic equation for B_1:

$$aB_1^3 + bB_1^2 + cB_1 + d = 0, \quad [16]$$

where

$$\begin{aligned} a &= K_1 - K_2, \\ b &= K_1 T_2 + K_2 T_1 - (K_1 - K_2)(K_1 + C + T_1), \\ c &= -T_1\left[K_1 T_2 + K_2 T_1 + K_1 K_2 + C(2K_2 - K_1)\right], \\ d &= K_2 C T_1^2. \end{aligned} \quad [17]$$

We find the solution for B_1 following standard methods for solving cubic equations,

$$B_1 = -\frac{1}{3a}\left[b + \mathrm{Re}\left((-1 + \sqrt{3}i)\Omega\right)\right], \quad [18]$$

where Re means real part, and

$$\Omega = \sqrt[3]{\frac{Q \pm i\sqrt{4P^3 - Q^2}}{2}} \quad [19]$$

with $P = b^2 - 3ac$ and $Q = 2b^3 - 9abc + 27a^2 d$. We use the plus sign in Ω when $K_2 > K_1$ and the minus sign when $K_1 < K_2$.

These analytical expressions were employed to generate the results shown in Fig. 3 in the main text.

D. Competitive binding in limiting cases. Beyond the simplest cases of competitive binding for one or two T cell clones, analytical solutions for Eq. 8 are also possible in certain limiting cases.

In the non-competitive limit, when the number of pMHCs bound to T cells is much smaller than the total number of pMHCs, $\sum B_i \ll C$, we can neglect $\sum B_j$ in the term $C - \sum B_j$ in Eq. 8 and obtain

$$B_i \approx \frac{T_i C}{K_i + C}. \quad [20]$$

Expressing the condition $\sum B_i \ll C$ in terms of the known parameters C, T_i and K_i, the assumption underlying Eq. 20 can be rewritten as $\sum T_i/(K_i + C) \ll 1$. The expression 20 corresponds to independent proliferation of T cells in all clones, as expected in the non-competitive limit. A less crude approximation obtained by only dropping the $B_i B_j$ terms in Eq. 8 is provided in (1).

In the highly competitive limit, when the number of bound T cells is much smaller than the total number of T cells in each clone, $B_i \ll T_i$, we can neglect B_i in the term $T_i - B_i$ in Eq. 8 and obtain

$$B_i \approx C \left(1 + \sum_{j=1}^{n} \frac{T_j}{K_j}\right)^{-1} \frac{T_i}{K_i}. \quad [21]$$

Expressing the condition $B_i \ll T_i$ in terms of the known parameters, the assumption underlying Eq. 21 can be rewritten as $C/K_i \ll 1 + \sum(T_j/K_j)$. Note that $C(1 + \sum(T_j/K_j))^{-1}$ in Eq. 21 is a constant prefactor for all the bound species. Therefore, the fraction of bound T cells in each clone is inversely proportional to their saturation constant. Interestingly, as long as T cell numbers are sufficiently large that $B_i \ll T_i$, Eq. 21 holds even when $C \gg K_i$, where naively one might have expected the competition to be affinity independent.

2. Influence of initial antigen dynamics

The dynamics of antigen presentation can be more complex than simple exponential decay from an initial level. Complications include the uptake and processing of antigens by the antigen presenting cells or continued generation of new antigens, e.g. due to pathogen replication during a natural infection. How might these complications in antigen dynamics influence the power-law dependence of fold expansion on the initial T cell number?

Here, we show that the power-law scaling continues to hold for a larger class of antigen dynamics. Specifically, we define a time t' after which there is no new processing of antigens into pMHCs. The initial antigen dynamics before t' can be arbitrary as long as the number of antigens remains above the saturating level for T cell proliferation. This ensures independent proliferation of T cells during the initial stages, and hence a constant ratio between $T(t')$ and $T(0)$ for all precursor numbers. Antigen dynamics beyond t' will obey exponential decay $C(t) = C(t')e^{-\mu(t-t')}$ as there is no new generation of pMHCs. Since the number of antigens remains saturating until t', the transition from T cell proliferation to decay happens at a time $t^\star$ later than t' for all precursor numbers. Therefore, in the competition-limited regime the power-law relation in Eq. 4 in the main text holds with $C(t')$ and $T(t')$ in place of $C(0)$ and $T(0)$,

$$\frac{T(t^\star)}{T(t')} \approx \left(\frac{C(t')}{T(t')}\right)^{(\alpha-\delta)/(\alpha-\delta+\mu)}. \quad [22]$$

As $T(t')$ and $T(0)$ only differ by a constant factor, the full fold expansion preserves the inverse power-law dependence on the initial T cell number: $T(t^\star)/T(0) \propto T(0)^{-(\alpha-\delta)/(\alpha-\delta+\mu)}$.

A similar analysis also applies to the affinity-limited regime.

3. Grazing model

In the competitive binding model, the power-law scaling between the fold expansion and the initial T cell number is ensured by the termination of expansion at a fixed relative level of T cells and pMHC complexes. A similar outcome can also be achieved in a model in which T cells actively degrade pMHCs upon binding, based on the observation that T cells that bind to pMHCs have some probabilities of acquiring the pMHCs (4, 5). This process of active acquisition has been termed *T cell grazing* (6).

In the grazing model, the dynamics of T cells and pMHCs are described by the following equations:

$$\frac{\mathrm{d}T}{\mathrm{d}t} = \alpha \frac{TC}{K + T + C} - \delta T, \quad [23]$$

$$\frac{\mathrm{d}C}{\mathrm{d}t} = -\mu C - \beta \frac{TC}{K + T + C}. \quad [24]$$

The last term in Eq. 24 describes the loss of pMHCs through grazing, where the prefactor β is the rate of grazing upon binding. For simplicity, we assume in the following that grazing limits proliferation before competitive binding sets in, i.e. we assume $T \ll K + C$ so that the number of bound complexes can be approximated by $TC/(K + C)$. The model differs from the previous grazing model in (6) in two major ways: In Eqs. 23 and 24, the T cell proliferation rate saturates at high pMHC concentrations, and the rate of grazing has a simple linear dependence on the T cell number. The grazing term in Eq. 24 leads to an increased

loss of pMHCs at high T cell numbers. This T cell-induced loss of pMHCs is also compatible with mechanisms other than grazing. In particular, the "grazing" term might also be interpreted as a reduction of pMHCs through T cell-induced pMHC endocytosis followed by lysosomal degradation, which has recently been reported (7).

The grazing model also yields an inverse power-law dependence of the fold expansion on the initial T cell number. Here, we provide a simple explanation following our analysis of the competitive binding model. At small times, the pMHC concentration is saturating for T cell binding, $C(t) \gg K$, so that T cells proliferate exponentially $T(t) = T(0)e^{(\alpha-\delta)t}$. The initial loss of pMHCs is mainly due to natural decay, as the rate of grazing of pMHCs is proportional to T cell number, which is initially small. Mathematically this means that $C(t) = C(0)e^{-\mu t}$ as $\beta T(t) \ll \mu C(t)$. The condition $\beta T(t) \ll \mu C(t)$ holds until a characteristic time

$$t^\star = \frac{1}{\alpha - \delta + \mu} \ln \frac{\mu C(0)}{\beta T(0)}. \quad [25]$$

After this time grazing leads to a rapid decline in the pMHC level. Therefore T cell proliferation ceases and the total number of T cells after expansion can be approximated by $T(t^\star) = T(0)e^{(\alpha-\delta)t^\star}$. The fold expansion is thus approximately

$$\frac{T(t^\star)}{T(0)} \approx \left(\frac{\mu C(0)}{\beta T(0)} \right)^{(\alpha-\delta)/(\alpha-\delta+\mu)}, \quad [26]$$

displaying an inverse power-law dependence on the initial T cell number. In both the grazing and the competitive binding model, the exponential increase of T cell number, the exponential decay of pMHCs, and the termination of expansion at a fixed relative level of T cells and pMHC complexes are the essential ingredients leading to power laws in the fold expansion of T cells.

4. Numerical simulations

The differential equations describing the T cell and pMHC dynamics were integrated using a Runge-Kutta method of order 4(5) due to Dormand and Prince (routine *dopri5* in scipy (8)). Parameters were determined using least-square fits to log-transformed mean cell numbers weighted by the number of repeat experiments. Asymptotic standard errors of the parameters were calculated based on the residual covariance matrix. An upper bound for K was defined by the value of K which leads to a unit increase of the sum of squared residuals (9).

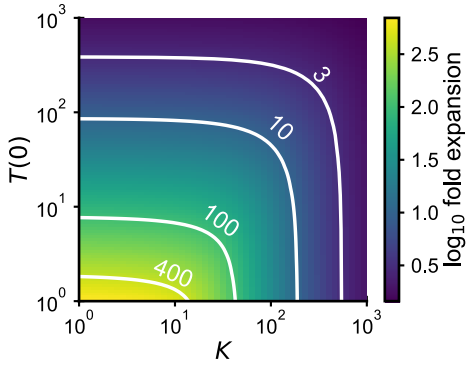

Fig. S1. Dependence of fold expansion at day 7 on precursor number $T(0)$ and saturation parameter K. Parameters as in Fig. 3.

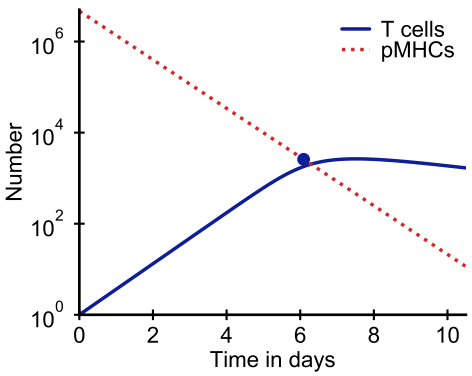

Fig. S2. Time courses of T cell and pMHC numbers in the model starting from the lowest experimentally used T cell precursor number (using the model parameters from Fig. 1). The estimates of peak time $t^\star$ and peak value $T(t^\star)$ of the T cell population from Eqs. 3 and 4 (blue dot) provide a good approximation to those of the full dynamics. Note that even for this smallest precursor number the peak T cell population size is reached by day 7. This allows us to approximate $T(t^\star)/T(0)$ by the fold expansion between day 0 and day 7 (Fig. 1).

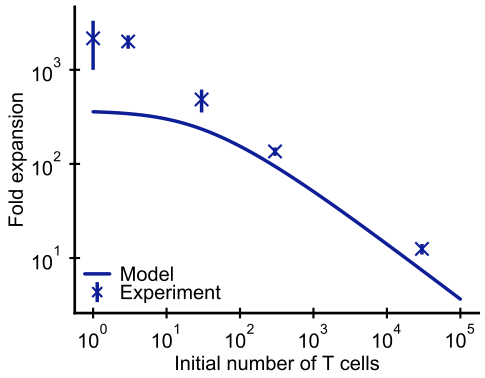

Fig. S3. Modeled T cell dynamics as in Fig. 1 with a saturation parameter $K = 10^4$ above the upper bound inferred from the data. At low precursor numbers, T cell expansion becomes affinity-limited, which caps fold expansion and leads to deviations from the observed power-law behavior. Mathematically, a crossover into the affinity-limited regime happens when $C(t^\star)$ is no longer larger than K. The absence of any such deviation in the experimental data even for the smallest precursor numbers sets an upper bound on K, which is $K \ll C(0)^{(\alpha-\delta)/(\alpha-\delta+\mu)} T(0)^{\mu/(\alpha-\delta+\mu)}$.

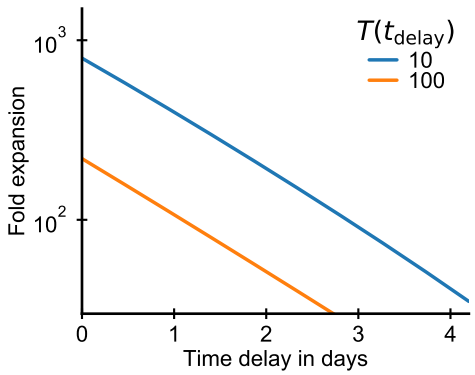

Fig. S4. Transfer of transgenic T cells after a time delay relative to antigen administration is predicted to decrease fold expansion. Modeled factor of expansion at day 7 after antigen administration as a function of the time delay for initial T cell numbers 10 (blue) and 100 (orange). Parameters as in Fig. 1.

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