Text S3. Effects of endogenous pMHC

We modify the ODE model presented in the main text to investigate the effects of a large concentration of identical pMHC molecules. In this calculation, we assume the contact interface to be a flat disc (radius 5 μm) containing a pMHC concentration of $M = 500$ μm^{-2} and a TCR concentration of $R = 100$ μm^{-2}. We first reformulate the model from the perspective of a single TCR interacting with a homogeneous pMHC distribution. Next, we use this modified model to calculate the probability of productive signaling for a single TCR and assuming TCR do not compete for pMHC (i.e. M $\gg$ R), we calculate the probability that at least 1 TCR (out of 7854) at the T cell-APC contact interface has transduced a productive signal from interacting with 39270 pMHC.

In this model, we introduce additional states, denoted as $\hat{U}_j$, that track the probability of finding a TCR without pMHC (i.e. once it diffuses away) in an intermediate state. In this state, a different pMHC may bind TCR and resume signaling from where a previous pMHC left off (provided μ is sufficiently small). The ODE system describing this modified model is,

$$
\begin{aligned}
\partial B_0/\partial t &= -\overline{k}_{\text{on}}^{\text{c}} B_0 + k_{\text{off}}^{\text{c}} B_0^c + k_{\text{on}} U_0 - (k_{\text{off}} + k_{\text{p}}) B_0 \\
\partial B_0^c/\partial t &= \overline{k}_{\text{on}}^{\text{c}} B_0 - k_{\text{off}}^{\text{c}} B_0^c + k_{\text{on}} U_0^c - (k_{\text{off}} + k_{\text{p}}) B_0^c \\
\partial U_0/\partial t &= -\overline{k}_{\text{on}}^{\text{c}} U_0 + k_{\text{off}}^{\text{c}} U_0^c - (k_- + \overline{k}_{\text{on}}) U_0 + k_+ M \hat{U}_0 + k_{\text{off}} B_0 + \mu \sum_{j=1}^{S-1} U_j \\
\partial U_0^c/\partial t &= \overline{k}_{\text{on}}^{\text{c}} U_0 - k_{\text{off}}^{\text{c}} U_0^c - \overline{k}_{\text{on}} U_0^c + k_{\text{off}} B_0^c + \mu \sum_{j=1}^{S-1} U_j^c \\
\partial \hat{U}_0/\partial t &= k_- U_0 - k_+ M \hat{U}_0 + \mu \sum_{j=1}^{S-1} \hat{U}_j \\
\partial B_j/\partial t &= -\overline{k}_{\text{on}}^{\text{c}} B_j + k_{\text{off}}^{\text{c}} B_j^c + k_{\text{p}} B_{j-1} - (k_{\text{p}} + k_{\text{off}}) B_j + \overline{k}_{\text{on}} U_j \\
\partial B_j^c/\partial t &= \overline{k}_{\text{on}}^{\text{c}} B_j - k_{\text{off}}^{\text{c}} B_j^c + k_{\text{p}} B_{j-1}^c - (k_{\text{p}} + k_{\text{off}}) B_j^c + \overline{k}_{\text{on}} U_j^c \\
\partial U_j/\partial t &= -\overline{k}_{\text{on}}^{\text{c}} U_j + k_{\text{off}}^{\text{c}} U_j^c - (k_- + \overline{k}_{\text{on}} + \mu) U_j + k_+ M \hat{U}_j + k_{\text{off}} B_j \\
\partial U_j^c/\partial t &= \overline{k}_{\text{on}}^{\text{c}} U_j - k_{\text{off}}^{\text{c}} U_j^c - (\overline{k}_{\text{on}} + \mu) U_j^c + k_{\text{off}} B_j^c \\
\partial \hat{U}_j/\partial t &= k_- U_j - k_+ M \hat{U}_j - \mu \hat{U}_j \\
\partial B_S/\partial t &= k_{\text{p}} B_{S-1} \\
\partial B_S^c/\partial t &= k_{\text{p}} B_{S-1}^c
\end{aligned}
$$

where M is the pMHC concentration and all other parameters are defined in the main text. The probability of productive signaling through a single TCR is $B_S + B_S^c$ and the probability that at least 1 TCR at the T cell-APC contact interface has transduced a productive signal is simply $1 - (1 - B_S - B_S^c)^{7854}$. We plot this quantity in Figure 5 in the main text. As an example, consider an endogenous pMHC with $k_{\text{off}} = 5$ s^{-1} and $k_{\text{on}} = 0.001$ μm^{-2}. When $\mu = 100$ s^{-1} this pMHC does not produce a productive signal (Figure 5C) despite forming $\sim 100,000$ TCR/pMHC bonds during 30 s $(= 1/(1/k_{\text{off}} + 1/(k_{\text{on}}[\text{pMHC}]))(7853$ TCR$)(30\text{s}) = 107,100)$.