

HIV-1 CCR5 gene therapy will fail unless it is combined with a suicide gene

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Supplementary Information

Table S1. A summary of the viral load observed in the study of Tebas et al.¹

Patient	Set-point VL (log ₁₀ copies/ml)	VL after gene therapy (log ₁₀ copies/ml)	Comments
201	3.27	3.99	Moderate increase
203	4.08	3.92	Mild decrease
204	4.73	4.78	ART re-initiated because of high VL
205	5.21	1.53	Individual was heterozygous for CCR5Δ32
251	4.72	5.22	ART re-initiated because of high VL
253	4.16	4.11	Mild decrease

The patients shown here underwent a 12-week interruption to ART, 4 weeks after receiving the gene therapy. A marked decline of the viral load was only observed in patient 205, who was later discovered to be heterozygous for CCR5Δ32.¹ Thus, the gene therapy potentially resulted in homozygous CCR5 disruption in modified CD4+ T cells, indicating that complete disruption of CCR5 gene can drastically decrease the viral load. In the other five patients the viral load went up (3/5) or mildly down (2/5). The viral load values were obtained using WebPlotDigitizer (<http://arohatgi.info/WebPlotDigitizer>) from data given in Tebas et al.¹

References

1. Tebas, P. *et al.* Gene editing of CCR5 in autologous CD4 T cells of persons infected with HIV. *New England Journal of Medicine* **370**, 901–910 (2014).

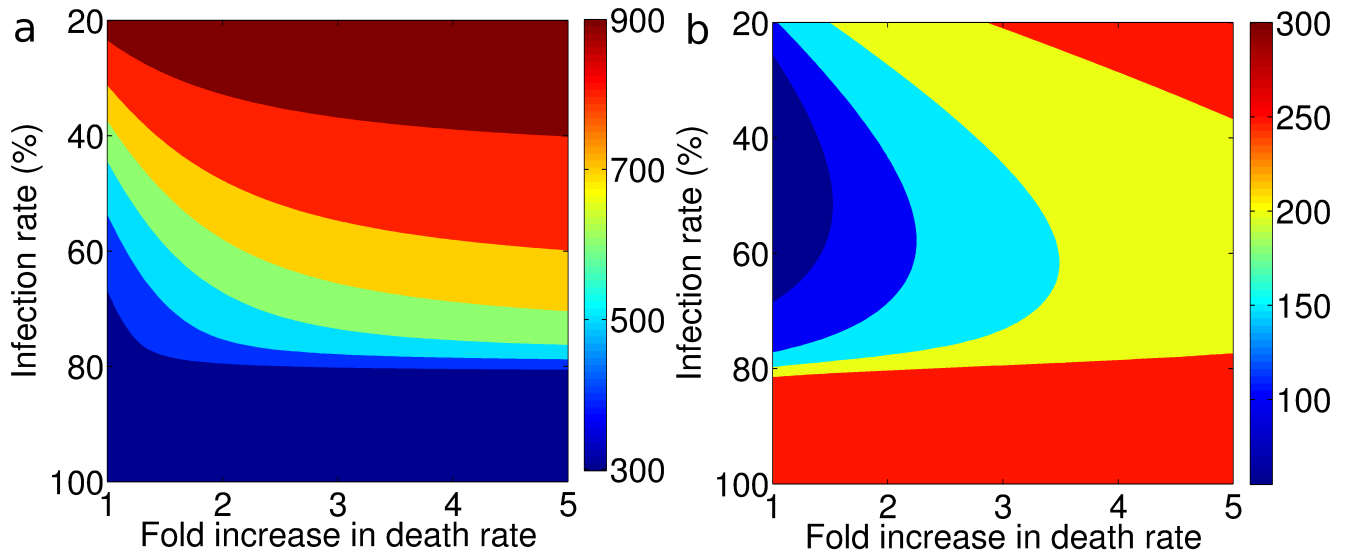


Figure S1. The effect of decreasing the infection rate (β_g) and increasing the death rate (δ_g) of genetically modified cells on the steady state: (a) total CD4+ T cell count ($\overline{T}_n + \overline{T}_g$), and (b) normal CD4+ T cell count (\overline{T}_n). The colors indicate steady state T cell counts, (a) $\overline{T}_n + \overline{T}_g$ or (b) \overline{T}_n , for the same parameter values as considered in Figure 2a. A decrease in β_g can decrease the counts and hence the diversity of the normal T cells, T_n (Panel b). A concomitant increase in δ_g retains the counts (and hence the diversity) of the T_n cells much better (Panel b).

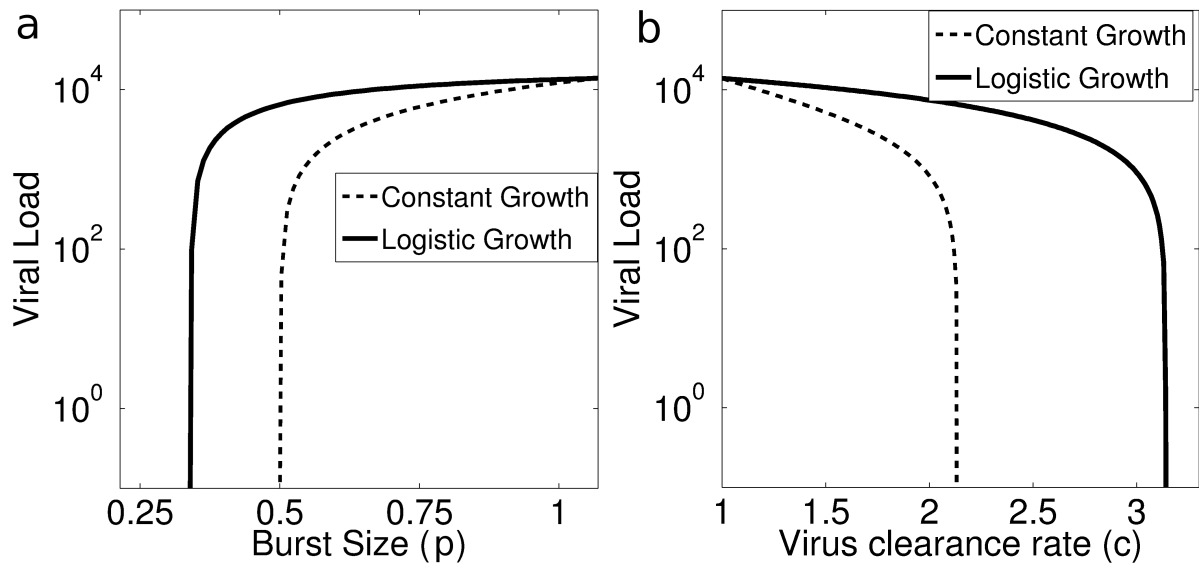


Figure S2. The effect of changing (a) the burst size, p , and (b) the virus clearance rate, c , on viral load in the typical HIV model (Eqs. 1-3).

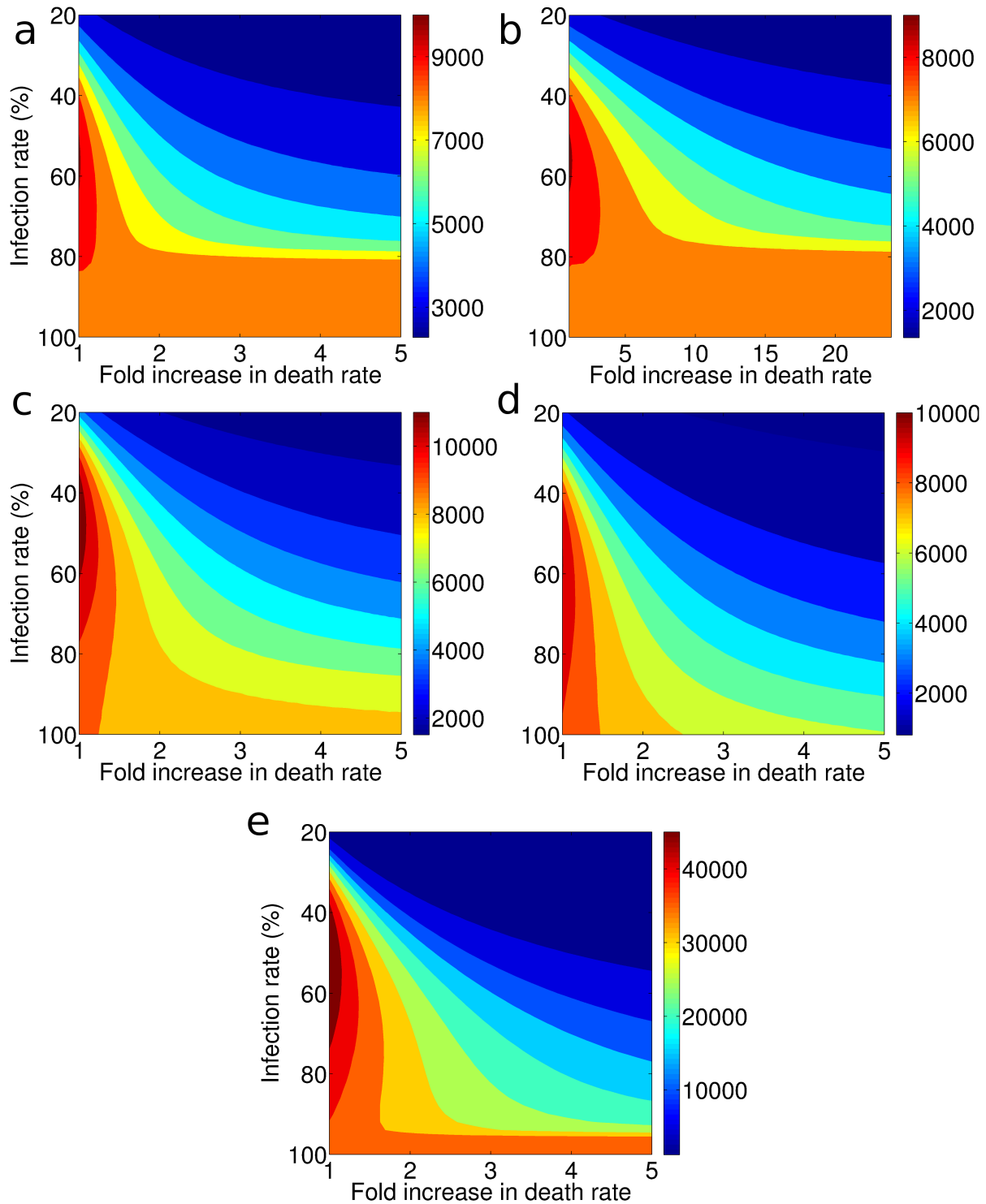


Figure S3. The effect of decreasing the infection rate (β_g) and increasing the death rate (δ_g) of genetically modified cells in: (a) the *Latent model* (Eqs. 6, 7, 10-14); (b) the *CTL model* (Eqs. 6, 7, 10, 15, and 16); (c) the model considering 25% hematopoietic stem cells to be genetically modified ($\lambda_2 = 0.33\lambda_1$); (d) the model considering 50% hematopoietic stem cells to be genetically modified ($\lambda_2 = \lambda_1$ in Eqs. 6, 8-10, and 20); and (e) the model considering different stages of infected cells described in Methods (Eqs. 21-33).

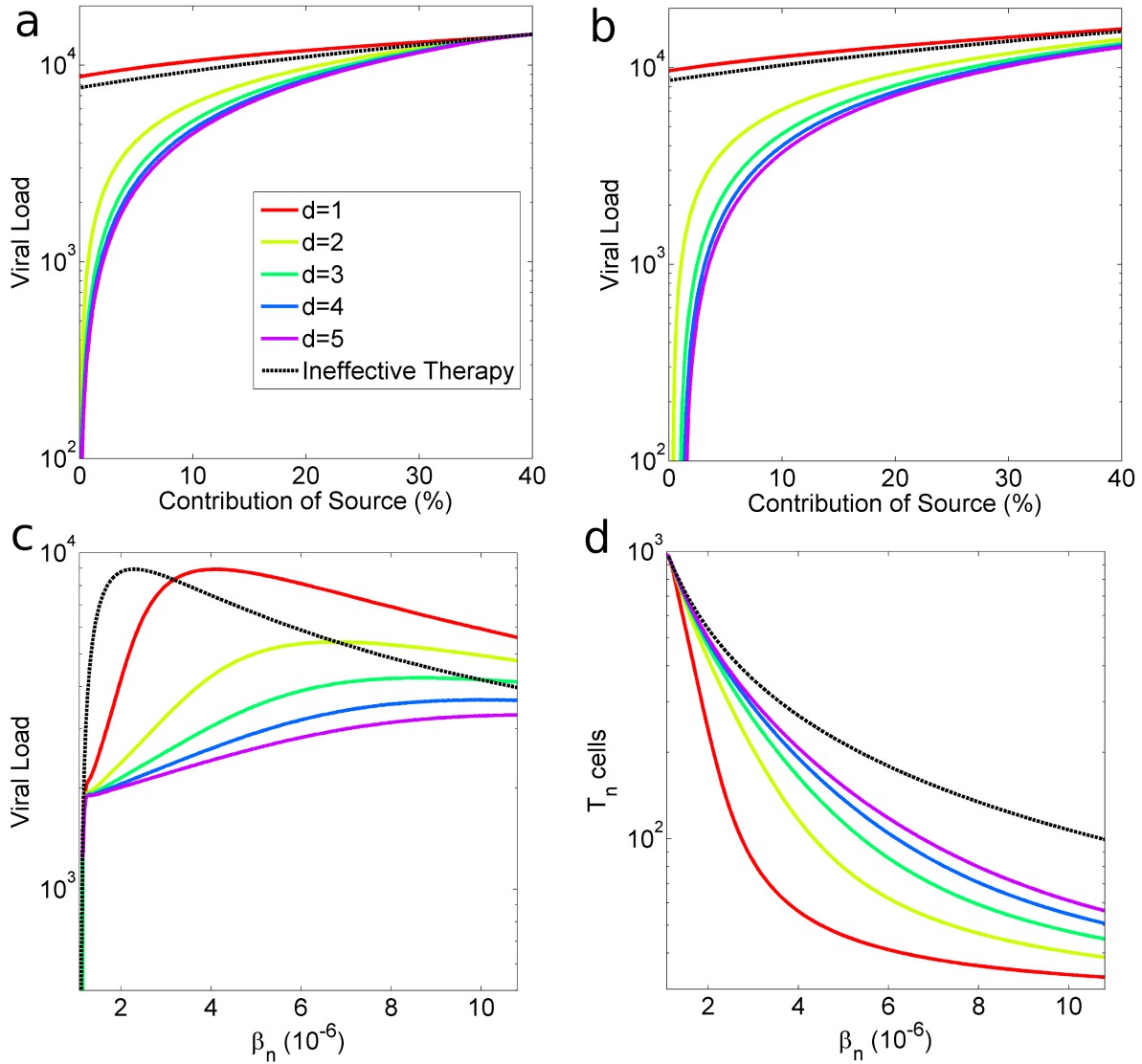


Figure S4. Parameter sensitivity analysis. In Figure 2, we considered the case where 5% of the normal uninfected T cells (T_n) come from the source (naive T cells) and 95% of the T_n cells are formed via self-renewal (i.e. $\lambda = 1$ cells μl^{-1} day $^{-1}$, $r = 0.057$ and $K = 1500$ in Eq. 1). Here we study the effect of the contribution from the source on a CCR5 suicide gene therapy. We performed a parameter sensitivity analysis considering $r = 0.06$ and $\bar{T}_n = 1000$ (Fig. 1) for the virus free steady state, and varied λ and K solving K from $K = r\bar{T}_n^2 / (\lambda + (r - \delta_T)\bar{T}_n)$ (derived from Eq. 1). In panel (a) we show the effect of the relative contribution from the source for T_n cells with a heterozygous CCR5 disruption (i.e. $\beta_g = 0.5\beta_n$) considering normal gene therapy i.e. 0% contribution from the source for T_g cells. In panel (b) we repeat the analysis considering a stem cell based gene therapy i.e. 5% contribution from the source for T_g cells. In panels (c) and (d) we perform a similar parameter sensitivity analysis by varying the rate of infection of the unmodified T cell β_n , setting other parameters the same as Fig. 2. In panel (c) we show that for large range of β_n values a suicide gene always results in a reduction of the virus load compared to the current CCR5 gene therapy (red line). In panel (d) we show that an increase in β_n results in a decrease of the T_n cells. Since we are simulating the chronic stage of HIV-1 infected individuals, we consider normal T cell counts of about 300 cells μl^{-1} . For high rates of infection ($\beta_n \geq 5.4 \times 10^{-6}$), T_n is less than 200 cells μl^{-1} which is too low for a normal chronic stage, as one expects AIDS to develop. Nevertheless, a suicide gene would always improve the normal T cell count compared to the current CCR5 gene therapy (red line).