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Mathematical Modeling of The Effect of Boosting Tumor Infiltrating Lymphocyte in Immunotherapy

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Abstract: This study, we analyzed the effect of boosting tumor infiltrating lymphocyte in immunotherapy using mathematical modeling. In this model, tumor growth is described as a tumor cells population with immunotherapy. This model also describes the effect of Tumor Infiltrating Lymphocytes (TIL), interleukin-2 (IL-2) and interferon alpha (INF-α) on dynamics of tumor cells. Numerical modeling of immunotherapy with or not boosted Tumor Infiltrating Lymphocyte (TIL) are presented in this study. We obtained that boosting Tumor Infiltrating Lymphocyte (TIL) in immunotherapy have a very significant role in killing of tumor cells.

Key words: Tumor infiltrating lymphocyte, immunotherapy, mathematical modeling, interleukin-2, interferon alpha, tumor cells

INTRODUCTION

Immunotherapy is also called as biologic therapy or biotherapy. Immunotherapy become an important component in the multi-pronged treatment approach, it was developed to treat several types of tumor within a short time frame (De Pillis *et al*., 2006). Immunotherapy grouped into three categories: immune response modifiers or cytokines, monoclonal antibodies and vaccines (Rosenbaum and Rosenbaum, 2005). Cytokines are chemical that mediated both natural and specific immunity. Cytokines have an important role in responsible for lymphocyte activation, growth and differentiation (Kirschner and Panetta, 1998). Most commons of cytokines are interleukins-2 (IL-2) and interferon alpha (INF-α). Immune system has important role in fighting tumor that has been verified in the laboratory as well as with clinical experiment (Farrar *et al*., 1999; O'Byrne *et al*., 2000; Morecki *et al*., 1996; Muller *et al*., 1998; Stewart, 1996). Basic idea behind immunotherapy is boosting the immune system *in vitro*, so the body can eradicate tumor on its own. There are many ways in which the immune system can be boosted, including vaccine therapy, IL-2 and INF-α growth factor injections, as well as the direct injection of highly activated specific immune cells, such Tumor Infiltrating Lymphocyte (TIL) into the bloodstream. Tumor Infiltrating Lymphocytes (TIL) are white blood cells that have left the bloodstream and migrated into tumor. They are an important prognostic factor in melanoma (Spatz *et al*., 2007; Gallon *et al*., 2006) higher levels being associated with a better outcome.

In this study, the mathematical modeling of ordinary differential equations is based on that originally developed by de Pillis (De Pillis *et al*., 2006), but the model of tumor growth without therapy use generalized logistic equation (Spratt *et al*., 1993) while in the model of de Pillis using logistic equation (De Pillis *et al*., 2006). The generalized logistic equation is based on that originally developed by Spratt *et al*. (1993), where in this work, they observed in 448 patients suffering from tumor for 564 days. Therefore, they obtained a generalized logistic equation more accurate than a logistic equation to describe of model tumor growth without therapy. Moreover, we add the effect INF-α on the immunotherapy based Isaeva and Osiopov's model (Isaeva and Osipov, 2009). Through mathematical modeling we analyzed the effect of boosting tumor infiltrating lymphocyte in immunotherapy. The hypothesis explains that boosting tumor infiltrating lymphocyte played an important role on immunotherapy. Some researchers have never explained the important role this boosting tumor infiltrating lymphocyte. Therefore, this study will describe the important role of boosting tumor infiltrating lymphocyte in immunotherapy.

MATHEMATICAL MODELING

The model is a system of Ordinary Differential Equation (ODE) whose state variables are populations of tumor cells, specific and non-specific immune cells and concentrations of therapeutic interventions. In this research, the model describes the kinetics of population

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tumor cells and three types of immune cells (NK cells, CD8+T cells, circulating lymphocytes), as well as two drug concentrations in the bloodstream, the equations are expressed by:

$$\frac{dT}{dt} = aT\left(1-\left(\frac{T}{b}\right)^{e}\right) - cNT - DT - c'TL \quad (1)$$

$$\frac{dN}{dt} = eC - fN + g\frac{T^2}{h+T^2}N - pNT \quad (2)$$

$$\frac{dL}{dt} = -mL + j\frac{D^2T^2}{k+D^2T^2}L - qLT + (r_1N + r_2C)T - uNL^2 + p_l\frac{LI}{g_I+I} + v_L(t) \quad (3)$$

$$\frac{dC}{dt} = \alpha - \beta C \quad (4)$$

$$\frac{dI}{dt} = -\mu_I I + v_I(t) \quad (5)$$

$$\frac{dI_\alpha}{dt} = -\mu_\alpha I_\alpha + v_{I_\alpha}(t) \quad (6)$$

$$D = d\frac{(L/T)^1}{s+(L/T)^1} \quad (7)$$

$$c' = c_{CTL}\left(2 - e^{-\frac{I_\alpha}{I_{\alpha 0}}}\right) \quad (8)$$

The populations are denoted by:

- T(t) = Tumor cell population at time t
- N(t) = Total NK cell effectiveness at time t
- L(t) = Total CD8+T cell effectiveness at time t
- C(t) = Number of circulating lymphocytes (or white blood cells) at time t
- I(t) = Immunotherapy interleukin 2 drug concentration in the bloodstream at time t
- Iα(t) = Immunotherapy interferon alpha drug concentration in the bloodstream at time t

Term VL(t) represent unction of boosting tumor infiltrating lymphocyte in immunotherapy. While $V_I(t)$ and $v_{I_\alpha}(t)$, respectively represent drug intervention term are functions of time denoted of interleukin and interferon.

PARAMETER DERIVATION

To complete the simulation and analysis, it necessary to obtain accurate parameters. The model is very sensitive to the choice of parameters. Tumor size was measured as volume (in mm^3) while this model considers population of cells. In the following we will assume that 1 mm^3 corresponds to 10^6 cells and will consider cells. Most of parameters in this model obtained from Pillis's model (De Pillis *et al*., 2006) and also several parameters were taken from Isaeva and Osipov's model (Isaeva and Osipov, 2009), as well as from Spratt' work (Spratt *et al*., 1993). Table 1 describes all parameters to run simulation our model.

NUMERICAL RESULTS

Here, we simulated the effect boosted Tumor Infiltrating Lymphocyte (TIL) in immunotherapy. In this simulation, we denoted initial tumor burden of 2×10^6 cells, since a tumor consisting of less than 10^6 cells is considered to be undetectable. In this simulation we also denoted as an initially value with 10^3 NK cells, 10 CD8+T cells and 6×10^8 circulating lymphocytes.

Figure 1-4 shown that immunotherapy without boosting tumor infiltrating cannot effectively kill tumor cells, where in these simulations IL-2 is administered in 6 pulses at strength 5×10^6, 5×10^{10}, 5×10^{20} and 5×10^{50} from day 8-12, respectively.

Figure 5-8 show that the effect boosting tumor infiltrating lymphocyte in immunotherapy. In these simulations, tumor infiltrating lymphocyte at strength 6.6440×10^7 boosted in the body in variation in day. Tumor infiltrating lymphocyte at strength 6.6440×10^7 boosted every weeks until four weeks, respectively, these simulations shown that at strength 6.6440×10^7 is not effective to kill tumor cells.

Figure 9-12 shown that simulation with boosting tumor infiltrating lymphocyte higher is at strength 6.6439×10^6. Figure 9 show that tumor initially decreased at day 10 then increased at day 28 leading to a dangerous level. Tumor effective kills at day 22 and never relapses again with boosting tumor infiltrating lymphocyte at strength 6.6439×10^6 in the second, third and fourth week as shown in Fig. 10-12, respectively. The last results shown the effect boosting tumor infiltrating lymphocyte higher is at strength 6.6440×10^8 boosted in every week.

Figure 13-16 show that tumor effective kills at day 28 and never relapses. Figure 14-16, respectively show that tumor effective kills at day 22 and never relapses.

DISCUSSION AND CONCLUSION

In this study, we analyzed the effect boosting tumor infiltrating lymphocyte using our mathematical model which has never been done in previous researchers' work. From our result, we can show the effect of boosting tumor infiltrating lymphocyte in immunotherapy. Immunotherapy

Table 1: Parameter values used for numerical simulation

Parameter	Description	References
$a = 4.31\times10^{-1}$ (day^{-1})	Tumor growth rate	Diefenbach *et al.* (2001)
$b = 1/1.02\times10^{-9}$ ($cell^{-1}$)	Tumor carrying capacity	Diefenbach *et al.* (2001)
$c = 6.41\times10^{-11}$ ($day^{-1}.cell^{-1}$)	Fractional (non) ligand transduced tumor cell kill by NK cells	Dudley *et al.* (2002) and Diefenbach *et al.* (2001)
$d = 2.34$ (day^{-1})	Saturation level of fractional tumor cell kills by CD8+T Cells. Primed with ligand-transduced cells, challenged with ligand-transduced	Dudley *et al.* (2002)
$e = 2.08\times10^{-7}$ (day^{-1})	Fraction of circulating lymphocytes that became NK cells	Kuznetsov *et al.* (1994)
$\varepsilon = 1.65$ (dimensionless)	Parameter which characterizes the shape of the sigmoidal growth curve	Spratt *et al.* (1993)
$l = 2.09$ (dimensionless)	Exponent of fractional tumor cell kill by CD8+T cells. Fractional tumor cell kill by chemotherapy	Dudley *et al.* (2002)
$f = 4.12\times10^{-2}$ (day^{-1}	Date rate of NK cells	Kuznetsov *et al.* (1994)
$g = 1.25\times10^{-2}$ (day^{-1})	Maximum NK cells recruitment by ligand-transduced tumor cells	Dudley *et al.* (2002) and Diefenbach *et al.* (2001)
$h = 2.02\times10^{7}$ ($cell^{2}$)	Steepness coefficient of the NK cell recruitment curve	Kuznetsov *et al.* (1994)
$j = 2.49\times10^{-2}$ (day^{-1})	Maximum CD8+T cell recruitment rate. Primed with ligand-transduced cells	Dudley *et al.* (2002) and Diefenbach *et al.* (2001)
$k = 3.66\times10^{7}$ ($cell^{2}.day^{-2}$)	Steepness coefficient of the CD8+T cell recruitment curve	Dudley *et al.* (2002) and Diefenbach *et al.* (2001)
$m = 2.04\times10^{-1}$ (day^{-1})	Death rate of CD8+T cells	Yates and Callard (2001)
$q = 1.42\times10^{-6}$ ($day^{-1}.cell^{-1}$)	CD8+T cell inactivation rate by tumor cells	Kuznetsov *et al.* (1994)
$p = 3.42\times10^{-6}$ ($day^{-1}.cell^{-1}$)	NK cell inactivation rate by tumor cells	Diefenbach *et al.* (2001)
$s = 8.39\times10^{-2}$ (dimensionless)	Steepness coefficient of tumor-(CD8+T cell) lysis term D. Primed with ligand-transduced cells, challenged with ligand-transduced.	Dudley *et al.* (2002)
$r_1 = 1.10\times10^{-7}$ ($day^{-1}.cell^{-1}$)	Rate of which CD8+T cells are stimulated to be produced as a result a tumor cells killed by NK cells	Yates and Callard (2001)
$r_2 = 6.50\times10^{-11}$ ($cell^{-1}.day^{-1}$)	Rate of which CD8+T cells are stimulated to be produced as a result a tumor cells interaction with circulating lymphocytes	-
$u = 3.00\times10^{-10}$ ($cell^{-2}.day^{-1}$)	Regulatory function by NK cells of CD8+T cells	-
$\alpha = 7.50\times10^{8}$ ($cell.day^{-1}$)	Constant source of circulating lymphocytes	Hauser (2001)
$\beta = 1.20\times10^{-2}$ (day^{-1})	Natural death and differentiation of circulating lymphocytes	Hauser (2001)
$\gamma = 9.00\times10^{-1}$ (day^{-1})	Rate of chemotherapy drug decay	Calabresi and Schein (1993)
$p_1 = 1.25\times10^{-1}$ (day^{-1})	Maximum CD8+T cell recruitment curve by IL-2	Kirschner and Panetta (1998)
$g_1 = 2.00\times10^{2}$ ($cells^{2}$)	Constant	-
$\mu_i = 1.00\times10^{1}$ (day^{-1})	Rate of IL-2 drug decay	Kirschner and Panetta (1998)
$\mu_\alpha = 1.7$ (day^{-1})	Decay rate of therapeutic INF-α	Isaeva and Osipov (2009)
$c_{CTL} = 4.4\times10^{-9}$ ($cell^{-1}day^{-1}$)	Rate of tumor cells inactivation by CD8+T cells	Isaeva and Osipov (2009)
I_{α_0} units	Initial Interferon	Isaeva and Osipov (2009)

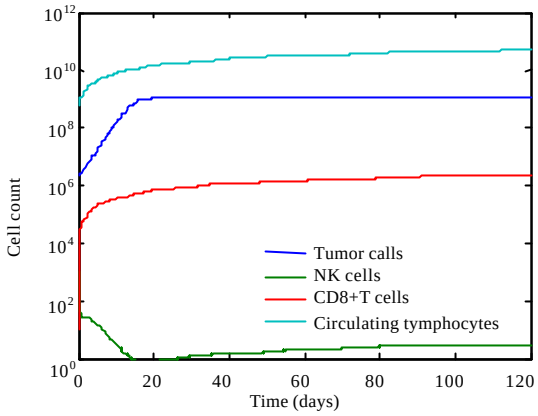

Fig. 1: Simulation of immunotherapy in absence boosting tumor infiltrating lymphocyte, IL-2 is administered in 6 pulses at strength 5×10^{6} from day 8-12 and INF-α is administered in 4 pulses at strength 5 from day 1-34

without boosting tumor infiltrating cannot effectively kill tumor cells. In these simulations, IL-2 is administered in 6 pulses at strength 5×10^{6}, 5×10^{10}, 5×10^{20} and 5×10^{50} from day 8-12 as pictured in Figure 1-4, respectively. Figure 5-8

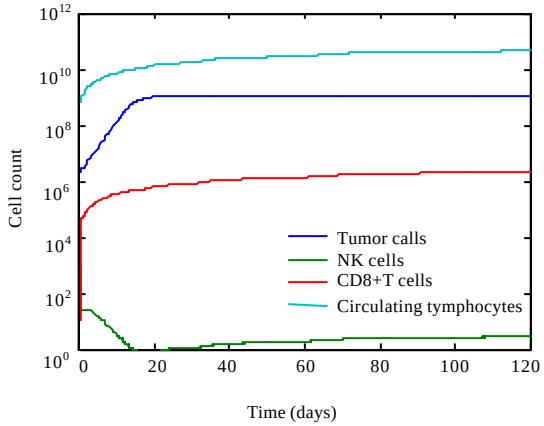

Fig. 2: Simulation of immunotherapy in absence boosting tumor infiltrating lymphocyte, IL-2 is administered in 6 pulses at strength 5×10^{10} from day 8-12 and INF-α is administered in 4 pulses at strength 5 from day 1-34

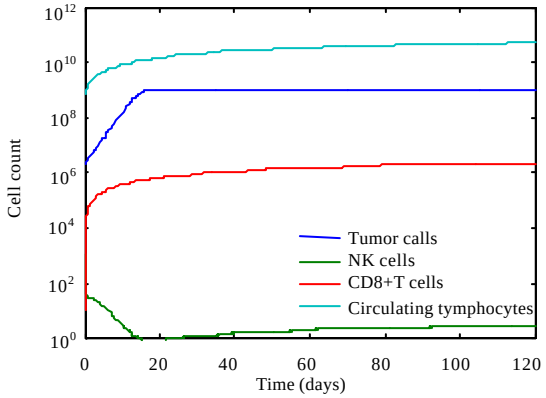

Fig. 3: Simulation of immunotherapy in absence boosting tumor infiltrating lymphocyte, IL-2 is administered in 6 pulses at strength 5×10^{20} from day 8-12 and INF-α is administered in 4 pulses at strength 5 from day 1-34

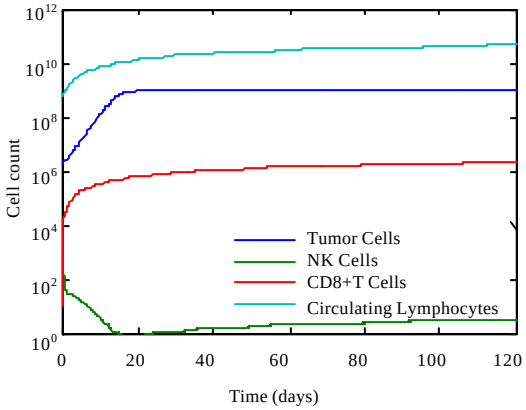

Fig. 4: Simulation of immunotherapy in absence boosting tumor infiltrating lymphocyte, IL-2 is administered in 6 pulses at strength 5×10^{50} from day 8-12 and INF-α is administered in 4 pulses at strength 5 from day 1-34

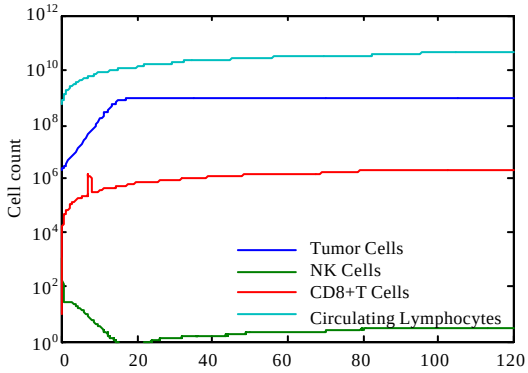

Fig. 5: Simulation of immunotherapy in present boosting tumor infiltrating lymphocyte with IL-2 is administered in 6 pulses at strength 5×10^6 from day 8-12, INF-α is administered in 4 pulses at strength 5 from day 1-34 and 6.6440×10^7 of TILs boosted from day 7 through 8

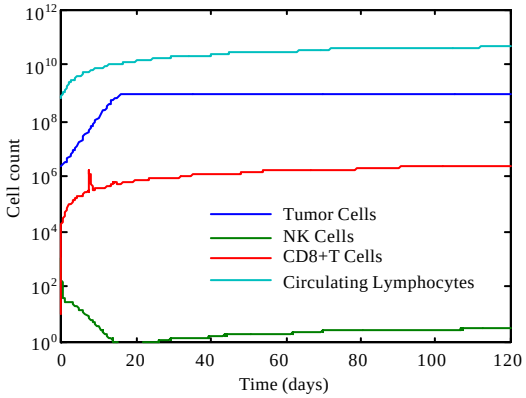

Fig. 6: Simulation of immunotherapy in present boosting tumor infiltrating lymphocyte with IL-2 is administered in 6 pulses at strength 5×10^6 from day 8-12, INF-α is administered in 4 pulses at strength 5 from day 1-34 and 6.6440×10^7 of TILs boosted from day 7 through 8 and day 14 through 15

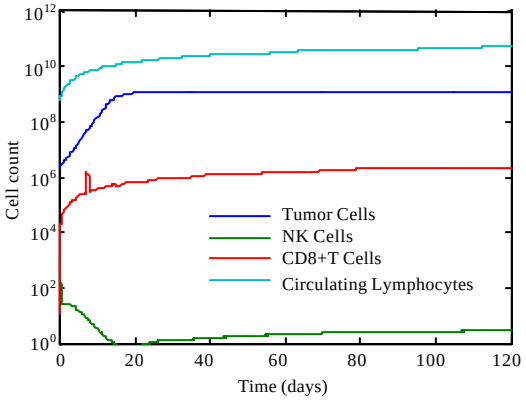

Fig. 7: Simulation of immunotherapy in present boosting tumor infiltrating lymphocyte with IL-2 is administered in 6 pulses at strength 5×10^6 from day 8-12 and INF-α is administered in 4 pulses at strength 5 from day 1-34 and 6.6440×10^7 of TILs boosted from day 7 through 8, day 14 through 15 and day 20 through 21

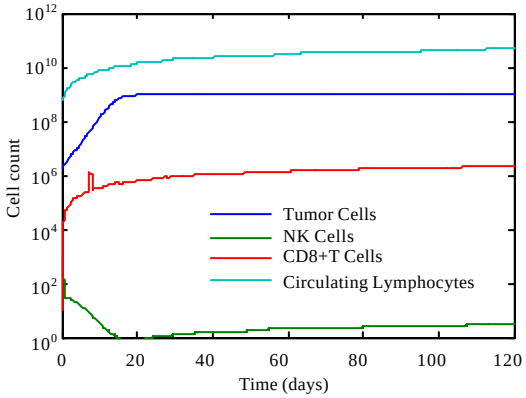

Fig. 8: Simulation of immunotherapy in present boosting tumor infiltrating lymphocyte with IL-2 is administered in 6 pulses at strength 5×10^6 from day 8-12 and INF-α is administered in 4 pulses at strength 5 from day 1-34 and 6.6440×10^7 of TILs boosted from day 7 through 8, day 14 through 15, day 20 through 21 and day 27 through 28

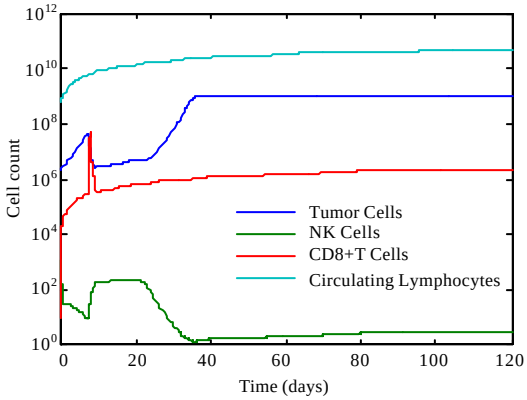

Fig. 9: Simulation of immunotherapy in present boosting tumor infiltrating lymphocyte with IL-2 is administered in 6 pulses at strength 5×10^6 from day 8-12, INF-α is administered in 4 pulses at strength 5 from day 1-34 and 6.6439×10^8 of TILs boosted from day 7 through 8

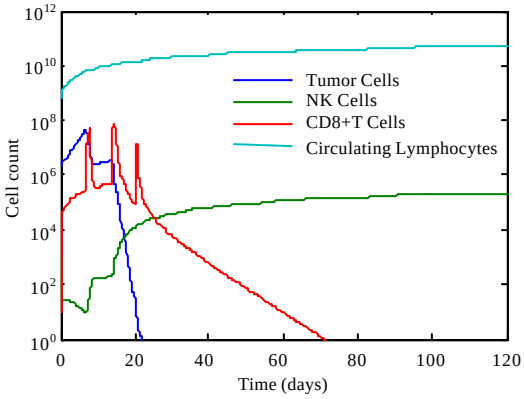

Fig. 10: Simulation of immunotherapy in present boosting tumor infiltrating lymphocyte with IL-2 is administered in 6 pulses at strength 5×10^6 from day 8-12, INF-α is administered in 4 pulses at strength 5 from day 1-34 and 6.6439×10^8 of TILs boosted from day 7 through 8 and day 14 through 15

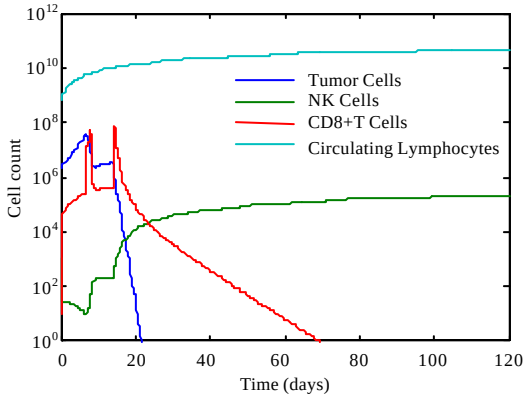

Fig. 11: Simulation of immunotherapy in present boosting tumor infiltrating lymphocyte with IL-2 is administered in 6 pulses at strength 5×10^6 from day 8-12, INF-α is administered in 4 pulses at strength 5 from day 1-34 and 6.6439×10^8 of TILs boosted from day 7 through 8, day 14 through 15 and day 20 through 21

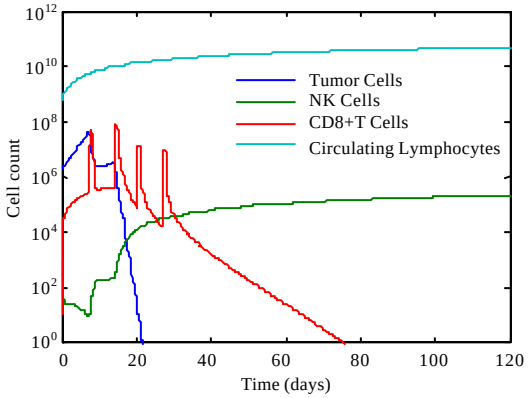

Fig. 12: Simulation of immunotherapy in present boosting tumor infiltrating lymphocyte with IL-2 is administered in 6 pulses at strength 5×10^6 from day 8-12, INF-α is administered in 4 pulses at strength 5 from day 1-34 and 6.6439×10^8 of TILs boosted from day 7 through 8, day 14 through 15, day 20 through 21 and day 27 through 28

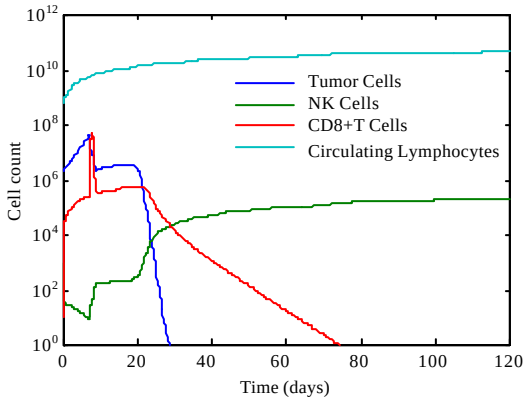

Fig. 13: Simulation of immunotherapy in present boosting tumor infiltrating lymphocyte with IL-2 is administered in 6 pulses at strength 5×10^6 from day 8-12, INF-α is administered in 4 pulses at strength 5 from day 1-34 and 6.6440×10^8 of TILs boosted from day 7 through 8

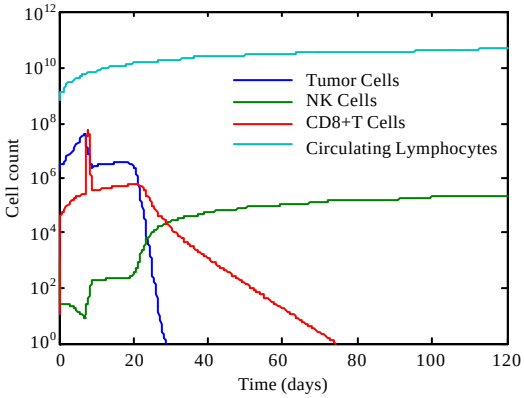

Fig. 14: Simulation of immunotherapy in present boosting tumor infiltrating lymphocyte with IL-2 is administered in 6 pulses at strength 5×10^6 from day 8-12, INF-α is administered in 4 pulses at strength 5 from day 1-34 and 6.6440×10^8 of TILs boosted from day 7 through 8 and day 14 through 15

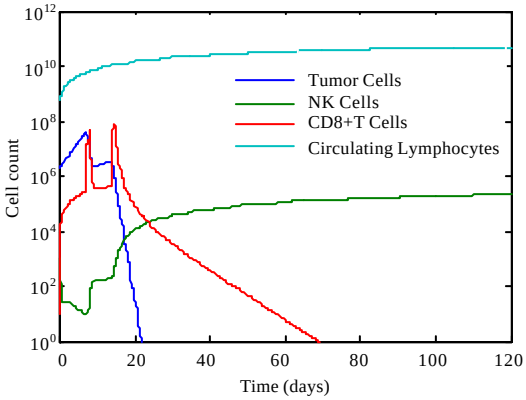

Fig. 15: Simulation of immunotherapy in present boosting tumor infiltrating lymphocyte with IL-2 is administered in 6 pulses at strength 5×10^6 from day 8-12 and INF-α is administered in 4 pulses at strength 5 from day 1-34 and 6.6440×10^8 of TILs boosted from day 7 through 8, day 14 through 15 and day 20 through 21

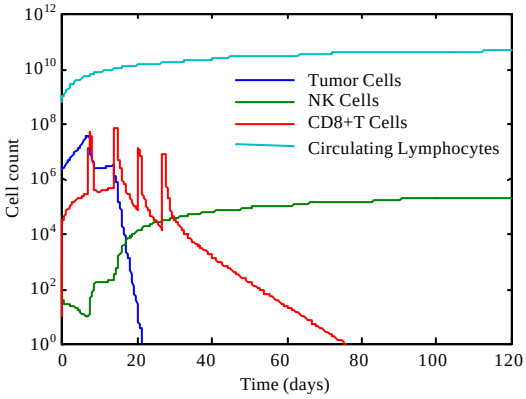

Fig. 16: Simulation of immunotherapy in present boosting tumor infiltrating lymphocyte with IL-2 is administered in 6 pulses at strength 5×10^6 from day 8-12, INF-α is administered in 4 pulses at strength 5 from day 1-34 and 6.6440×10^8 of TILs boosted from day 7 through 8, day 14 through 15, day 20 through 21 and day 27 through 28

shown that the effect boosting tumor infiltrating lymphocyte be analyzed in immunotherapy, these simulations tumor infiltrating lymphocyte at strength 6.6440×10^{7} boosted in the body in variation in day. Tumor infiltrating lymphocyte at strength 6.6440×10^{7} boosted every weeks until four weeks as pictured in Fig. 5-8, respectively, these simulations shown that at strength cannot effective to kill tumor cells. In Fig. 9-12, simulation with boosting tumor infiltrating lymphocyte higher is at strength 6.6439×10^{8}. In Fig. 9, tumor cells decreased at day 10 then increased at day 28 leading to a dangerous level. Tumor effective kills at day 22 and never relapses again with boosting tumor infiltrating lymphocyte at strength 6.6439×10^{6} in the second, third and fourth week as shown in Fig. 10-12, respectively. The last results shown the effect boosting tumor infiltrating lymphocyte higher is at strength 6.6440×10^{8} in every week. Figure 13 show that tumor effective kills at day 28 and never relapses. Figure 14-16 show that tumor effective kills at day 22 and never relapses.

Based on our simulation results, we can conclude that the boosting of tumor infiltrating lymphocyte on immunotherapy has a very significant role in killing tumor cells. Boosting tumor infiltrating lymphocyte in immunotherapy is more effective in two week at strength 6.6439×10^{8}.

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