

will be parameterized using this data, and will then be used to inform the design of a follow-up trial, which will include additional immunotherapies. The goal of this model is thus twofold: 1) Determine the optimum time at which to administer the DC vaccine, so as to maximize the expansion of cytotoxic T-cells specific for survivin positive tumor cells; 2) Explore the impact of administering additional immunotherapies, such as anti-PD-L1 or anti-CTLA4, which may augment the efficacy of the survivin vaccine.

The immune system is a complex mix of many cell types. To be included in the model, cell types had to meet certain criteria. First, the interactions of the cells selected must be well characterized. Second, the number of each cell type must be clinically measurable. Third, the interactions must either play a direct role in homeostatic repopulation and/or be targetable by available therapies. Finally, all cells and interactions must come together to form a model capable of realistically recapitulating the dynamics of homeostatic repopulation. The requirement that the size of each cell population be measurable will be particularly important when simulating tumor growth and treatment using individual patient data. Fortunately, we have access to a number of methods that allow us to do just that: flow cytometry will be used to measure the number of regulatory T-cells and the overall T-cell category; nanostring measurements will determine the number of survivin-positive effector T-cells, exhausted T-cells, and activated T-cells; and M protein and light chain ratios will measure the number of tumor cells. Having data at multiple time points also gives us the ability accurately estimate many of the parameters in our model.

IIB Equations

We have built a preliminary mathematical model consisting of a set of ordinary differential equations (ODEs) to describe the tumor burden and the immune response. The current model is developed for the period of time immediately following transplantation of stem and immune cells in the clinical protocol, such that the T-cells and tumor cells are both repopulating the depleted microenvironment. The system of 6 ODEs considers the following cell populations: tumor burden (P); activated T-cells (A); effector T-cells (E); tolerized T-cells (X); regulatory T-cells (R); and overall T-cells (T). The following system of ODEs was used as a preliminary model of the dendritic cell therapy.

$$\begin{aligned}
 1. \quad \dot{P} &= \overbrace{g_P P \left(1 - \frac{P}{C_P}\right)}^{\text{logistic growth}} - \overbrace{k_E E f_{PE}}^{\text{immune kill}} \\
 2. \quad \dot{A} &= \overbrace{(1 + k_v) \alpha_M M f_{PM} f_{AR}}^{\text{stimulated from memory by tumor}} + \overbrace{g_A P f_{AR}}^{\text{plasma stimulated}} - \overbrace{\delta_A A}^{\text{death}} \\
 3. \quad \dot{E} &= \overbrace{\alpha_A A f_{ER}}^{\text{development from A}} + \overbrace{(1 + k_v) g_E E f_{PE} f_{ER}}^{\text{self and tumor stimulated}} + \\
 &\quad \overbrace{s_X X S - s_R E f_{RE}}^{\text{reactivation}} + \overbrace{k_v X}^{\text{DC driven escape from tolerance}} - \overbrace{\delta_E E}^{\text{death}} \\
 4. \quad \dot{X} &= \overbrace{\alpha_A A^* f_{RE}}^{\text{development from A}} - \overbrace{s_X X S + s_R E f_{RE} - k_v X - \delta_X X}^{\text{transition to and from E}} \\
 5. \quad \dot{R} &= \overbrace{\rho_P P}^{\text{stimulated by tumor burden}} + \overbrace{\rho_E E f_{hE}}^{\text{stimulated by inflammation}} - \overbrace{\delta_R R}^{\text{death}} \\
 6. \quad \dot{T} &= \overbrace{g_T T - \left(1 - \frac{T}{C_T}\right)}^{\text{logistic homeostasis}} - \delta_T T
 \end{aligned}$$

Memory cells (M) are a constant fraction of the total T-cell population (T). The variable S is the available space in the T cell

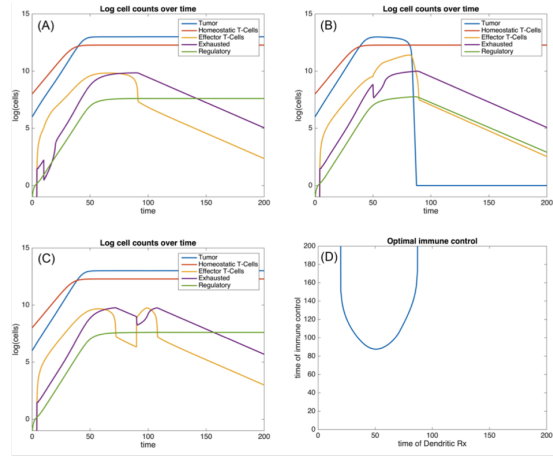


Figure 2: Results from the model showing application of dendritic cell therapy at time points 10, 50, and 90 days post transplant (panels A-C respectively). Only the therapy at 50 days leads to immune control of the tumor. Panel D shows the optimum tumor control as a function of the day of the dendritic cell therapy application. The vertical axis measures the time at which the tumor reaches its minimum post-dendritic therapy level.

compartment, calculated as $(1 - \frac{T}{C_T})$. The final term of Eqs. 2-6 represents the natural death of that compartment. The term $(1 + k_v)$ represents the effect of the dendritic cell therapy. In all equations, the following shorthand is used:

$$f_{BC} = \frac{B}{B + C}$$

The tumor burden (Eq. 1) is increased by logistic growth (first term) and decreased through killing by the effector T-cells (second term). Activated T-cells (Eq. 2) are stimulated (first term) from memory cells by increased tumor burden; high Treg numbers suppress their stimulation and self proliferation (first and second term). Effector T-cells (Eq. 3) develop from the activated T-cells (first term) and through self proliferation based on tumor burden (second term). The third and fourth terms represent transition to and from the tolerized state, affected by space and Treg numbers. Dendritic cell therapy enhances the escape from tolerance (fifth term).

Tolerized T-cells (Eq. 4) are derived from activated T-cells when Treg numbers are high (first term). Terms 2-4 are the transition to and from the effector T-cell compartment. Tregs (Eq. 5) are promoted by the tumor burden (first term) and the amount of inflammation in the system (second term). The total T-cell compartment (Eq. 6) is modeled as logistic homeostasis. See Figure 1 for a schematic illustrating these interactions.

III ILLUSTRATIVE RESULTS OF APPLICATION OF METHODS

A sample result of our model's ability to simulate the effect of timing of the vaccine can be found in Figure 2. These simulations highlight the importance timing has on the ability of the vaccine to stimulate a sufficient anti-tumor immune response. In panels A and C, the vaccine was given too early (day 10) and too late (90), respectively. In these cases, treatment fails because it misses the window of opportunity in which homeostatic repopulation can be exploited to increase expansion of survivin specific cytotoxic T-cells. Giving the vaccine too early will fail because there are an insufficient number of T-cells to stimulate, while administering it too late it will fail due to the return of peripheral tolerance. However, there is a sweet spot, where the T-cell population is neither too large nor too small, during which the vaccine will stimulate an immune

response strong enough to eliminate the tumor (panel D). In these simulations, this ideal time to administer the vaccine occurs 50 days post AHCT (panel B). The optimum is sensitive to the parameters and initial conditions, suggesting that the model can be used in a patient-specific way to predict individual optima for vaccination.

IV CLOSING REMARKS

The nature of multiple myeloma and the ability to collect key data at multiple time points provides the unique opportunity to develop a highly parameterized model of homeostatic T-cell repopulation, tumor growth, and the efficacy of immunotherapy. It is our hope that the model developed here will serve as a powerful tool for clinicians designing future clinical trials.

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References

- [1] DC Altieri. Survivin, versatile modulation of cell division and apoptosis in cancer. *Oncogene*, 22(53):8581–8589, 2003.
- [2] G Ambrosini, C Adida, and DC Altieri. A novel anti-apoptosis gene, survivin, expressed in cancer and lymphoma. *Nat Med.*, 3(8):917–21, 1997.
- [3] B Barlogie, J Shaughnessy, and G Tricot. Treatment of multiple myeloma. *Blood*, 103:20–32, 2004.
- [4] H Kawasaki, DC Altieri, CD Lu, M Toyoda, T Tenjo, and N Tanigawa. Inhibition of apoptosis by survivin predicts shorter survival rates in colorectal cancer. *Cancer Res.*, 58(22):5071–5074, 1998.
- [5] SK Knauer, W Mann, and Stauber RH. Survivin’s dual role: An export’s view. *Cell Cycle*, 5(5):518–21, 2007.
- [6] Y Nakagawa, S Abe, and Kurata M. Iap family protein expression correlates with poor outcome of multiple myeloma patients in association with chemotherapy-induced overexpression of multidrug resistance genes. *Am J Hematol*, 81:824–31, 2006.
- [7] A Palumbo, JS Miguel, and Sonneveld P. Lenalidomide: A new therapy for multiple myeloma. *Cancer Treat Rev*, 34:283–91, 2008.
- [8] AC Rawstron. Minimal residual disease detection in myeloma: No more molecular remissions? *Haematologica*, 90, 2005.
- [9] Schlossman R Richardson PG, Mitsiades C. Bortezomib in the front-line treatment of multiple myeloma. *Expert Rev Anticancer Ther*, 8:1053–72, 2008.
- [10] American Cancer Society. Cancer facts and figures 2016. Atlanta, GA, 2016.
- [11] RH Stauber, W Mann, and SK Knauer. Nuclear and cytoplasmic survivin: Molecular mechanism, prognostic, and therapeutic potential. *Cancer Res*, 67(13):5999–6002, 2007.
- [12] HS Swana, D Grossman, JN Anthony, RM Weiss, and Altieri DC. Tumor content of the antiapoptosis molecule survivin and recurrence of bladder cancer. *N Engl J Med*, 341(6):452–53, 1999.
- [13] K Tanaka, S Iwamoto, G Gon, T Nohara, M Iwamoto, and Tanigawa N. Expression of survivin and its relationship to loss of apoptosis in breast carcinoma. *Clin Cancer Res*, 6(1):127–134, 2000.