Supplementary Material

## 1 Aggregated survival probability to irradiation

According to the linear-quadratic model the survival probability of one cancer cell to a dose of Gy of radiation is given by [1, 2, 3]:

where and are the constants of the linear-quadratic model.

However, in a dynamic tumor volume model, a *delay* exists between the time of *irradiation* and the time of *death* of the tumor cell and this delay is long enough for the cell to undergo a small number of mitosis during which the tumor size may continue to grow [4]. Another reason for the tumor growing in *size* but not in *number* of tumor cells, is the inflammatory process provoked by irradiation and the resulting translocation of immune effector cells inside the tumor micro-environment. Eventhough this effect has not been modeled here, it could be included by adding a certain proportion of the compartment to the tumor volume. Therefore the linear-quadratic model was modified to distribute, or spread, the death of the tumor cells over a certain time span, by defining *daily weights*: whose sum is equal to 1, and by defining the survival probability days after receiving a radiation dose of Gy as:

For a given dose regimen on days , the survival probability at time is then given by

The daily weights have been defined by using a log-normal density probability function. The simulated tumor mass obtained with that assumption could be made close to published experimental data of tumor volume after irradiation, such as those in [4], by adjusting the mean and standard deviation of the log-normal probability density function. All simulations used a mean of 5 days and a standard deviation of 1.5 days, i.e. 95% of the killed cells died between 2 and 8 days after irradiation.

Of course, the linear-quadratic model is not the only one that could be used, especially if large doses are administered (typically, in excess of 10 Gy), because of its tendency to overpredict the number of cells killed. A mathematically simple modification of the linear-quadratic model could be a switch to a linear model for doses above a certain level, as in [5]. For a comprehensive review of several possible models, see for example [4].

2 How to compute a probability of tumor rejection

This section describes how to transform the activity of the immune system against the primary tumor site into a probability of tumor rejection at a distant metastatic site, as in Figure 2, to describe the results published in [6]. Since the primary tumor volume increases at a quasi-constant rate , it follows that the activity of the immune system against the primary tumor site must be smaller than , since in the proposed model immunological attack is the only cause of cell loss (in the absence of irradiation). What is the probability of the immune system rejecting an hypothetical metastase? Since it is well known that human metastases have shorter doubling times than their primary tumor [7], which isn’t controlled by the immune system, one could object that this probability has to be zero. However, this is not what happens experimentally. To explain this, it was hypothesized that *chirurgically created*, or *synthetic* metastases, that are implanted by the experimentator in predefined locations of the animal, do not benefit from the *unusually favorable milieu into which secondary tumors tend to be seeded*, as cited in [8].

It was therefore assumed that the initial growth rate of an *experimental* metastatic site can be considered smaller than . For simplicity, it was also assumed that the immune system activity on the metastatic site is equal to . It follows that metastatic sites may be rejected if . Since metastasic sites have a certain degree of randomness in their initial properties (such as volume, localisation, interaction with their host animal...), their growth rate was assumed to follow a log-normal distribution of mean and standard deviation .

Hence, if a metastatic site is implanted on a mice N days after the implantation of the primary tumor, its rejection occurs if , where is a random variable and is given, and this event has a probability:

Where is a Gaussian density: This formula has been used to transform , the activity of the immune system at the primary tumor site on day , into a probability of tumor rejection.

## 3 Model parameters: numerical values and definitions

The table 1 contains numerical values for the parameters that were used to produce the figures provided in this paper. When a parameter is not necessary to produce a result, its value is marked as *NA* (for example, in experiments that do not use radiation, no value for or are provided, etc.). Drug concentrations are given in arbitrary units since this is a pure PD model; actual concentration units could be used at a later stage if a PK model is added.

The table 2 summarizes the model parameters and give their unit (when applicable).

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Parameter | Figure 2 | Figure 3 | Figure 4 | Figure 5 |
|  | NA | NA | 0.05 | 0.05 |
|  | NA | NA | 0.005 | 0.005 |
|  | NA | 1.8 | 2 | 0.441 |
|  | NA | 0 | 0 | 150 |
|  | 0.5 | 0.5 | 0.1 | 0.1 |
|  | NA | NA | 20 | 20 |
|  | 0.14 | 0.14 | 0.14 | 0.135 or 0.108 |
|  | 0.007 | 0.007 | 0.007 | 0.007 |
|  | 0.128 | 0 or 0.009 | 0.0314 | 0.0314 |
|  | 5 | 5 | 5 | 5 |
|  | 0.01 | 0.01 | 0.01 | 0.01 |
|  | 0.15 | 0.15 | 0.15 | 0.15 |
|  | 0.1 | 0.1 | 0.1 | 0.1 |

Table 1: Values of parameters used in the model.   
*NA: Not Applicable, or (equivalently) set to zero*.

|  |  |  |
| --- | --- | --- |
| Parameter | Description | Unit |
|  | linear radiosensitivity of tumor cells | Gy |
|  | quadratic radiosensitivity of tumor cells | Gy |
|  | concentration of immunotherapy treatment (PD1-PD-L1 axis) | *none* |
|  | concentration of immunotherapy treatment (CTLA4 pathway) | *none* |
|  | natural daily rate of production of tumor antigens | *none* |
|  | daily rate of production of tumor antigens by radiation-killed tumor cells | *none* |
|  | daily growth rate of the tumor | *none* |
|  | sensitivity of the *primary* immune response to immune effectors | *none* |
|  | max value of the propensity of the *primary* response to induce a *secondary* response | *none* |
|  | min value of the propensity of the *primary* response to induce a *secondary* response | *none* |
|  | sensitivity of the *primary* immune response to tumor down-regulation | *none* |
|  | daily transformation of tumor antigens into immune effectors | *none* |
|  | daily wash-out of the immune effectors | *none* |
| Variable | Description | Unit |
|  | tumor mass | g |
|  | tumor antigens | *none* |
|  | immune effectors | *none* |
|  | activity of the primary immune respone | *none* |
|  | activity of the secondary immune response | *none* |
|  | ratio of tumor cells that survive on day to irradiation received up to day | *none* |

Table 2: Definitions of parameters and variables used in the model.  
: concentrations have been left without unit because this is a pure PD model, but actual concentrations units would be used if a PK model was added.

## 4 Discussion on the mathematical formulation of Equation 4

Let us imagine a chemical reaction by which these immunomodulatory factors combine with immune effectors to produce *exhausted* or *deactivated* effectors that have lost their anti-tumor activity. As a first approximation, it seems reasonable to assume that the law of Guldberg and Waage holds for this chemical equilibrium, i.e, if denotes the fraction of free, or active immune effectors, there exists a constant such as:

it follows that the proportion of active immune effectors is:

If we include the constant inside , we obtain Equation 4 for .

Let us now assume that an immunotherapy drug in concentration binds to these immunomodulatory factors and inactivate them. This drug is either an anti-PD1 or anti-PD-L1 monoclonal antibody, depending of the context. For an anti-PD-L1, the fraction of free immunomodulatory factors is , where is the constant from the law of Guldberg and Waage. Without loss of generality, we can include inside and we obtain Equation 4 with a concentration of an anti-PD-L1 drug. Symetrically, for an anti-PD1 drug, the same reasoning holds if we assume that is proportionnal to the fraction of free PD1 receptors, since contains the above described constant which describes the affinity between immunomodulatory factors and their receptors on the immune effectors. Hence, with an anti-PD1 drug, has to decreases in proportion with the number of free receptors and must be replaced by , where is again another equilibrium constant. This allows us to derive the same mathematical formulation for an anti-PD1 and for an anti-PD-L1 drug.

## 5 Concurrent blockade of PD1 and PDL1

Though this concurrent blockade is relatively less synergistic it may be considered sometimes and we provide here a formula derived from the previous discussion. If and are the concentrations of the drugs, Equation 4 becomes:

Where as already discussed, equilibrium constants and have been included in and .

## 6 Modelling of redundant immunomodulatory factors: the example of PD-L1 and PD-L2

Since it is known that PD-L2 is another ligand of PD-1, with similar immunomodulatory properties [9], it is reasonable to question the validity of Equation 4 in the context of more than one immunomodulatory factors. For this question to be relevant, we must assume that the anti-PD-L1 drug in concentration has a different affinity for the ligand PD-L2. For both simplicity and conservatism, we assume that the anti-PD-L1 drug does not bind at all to PD-L2.

The quantity of immunomodulatory factors produced by the tumor is now the sum of the quantities of PD-L1 and PD-L2 ligands:

Let be the ratio of free or active immune effectors, that do not bind to neither PD-L1 nor PD-L2, let be the proportion of inactive immune effectors that bind to PD-L1 and let be the proportion of inactive immune effectors that bind to PD-L2. Let us assume that the chemical equilibrium for each reaction is governed by the law of Guldberg and Waage whose and are the associated constants. We can write:

We can solve directly the ratio of active effectors:

Which proves that the general form of Equation 4 is still valid if one adds PD-L2. By recurrence, Equation 4 is valid for any number of ligands that would have a similar mechanism of action than PD-L1.

If, as previously assumed, the immunotherapy drug in concentration binds specifically to PD-L1 and has zero affinity for PD-L2, by the same reasoning, the ratio of free PD-L1 is , where is again a constant of chemical equilibrium, that we can integrate inside and we can rewrite :

In summary, to take into account the existence of more immunomodulatory factors with a similar mechanism than PD-L1, such as PD-L2, it suffices to rewrite Equation 4 as:

Where is a new constant.

## 7 Discussion on the mathematical formulation of Equation 5

This section explains the rationale for the term . Let denote the number of Antigen Presenting Cells and let be the number of CD4+ T cells. We assume that all Antigen Presenting Cells considered here are loaded with tumor antigens, that they present to cells. There are three possibilities:

• APC can engage the inhibitory CTLA4 receptor of a CD4+ T cell, with an affinity , which yields a deactivated CD4+ T cell, whose proportion is denoted by

• APC can engage the CD28 co-stimulatory receptor of CD4+ T cell, with an affinity , which yields an activated CD4+ T cell, whose proportion is denoted by

• APC may not interact with any CD4+ T cells, and the proportion of these *free* T cells is denoted by

Note that the affinity of APC is greater for the inhibitory CTLA4 than for the co-stimulatory CD28 (), which explains that the activation of CD4+ T cells requires that the antigenic stimulation exceeds some threshold before triggering the CD4 response; this is maybe a stabilizing mechanism of the immune response that helps inhibiting auto-immunity. If one assumes that a chemical equilibrium holds and that the law of Guldberg and Waage gives a good approximation of it, we can write that:

Hence:

Now, if one assumes that an anti-CTLA4 agent in concentration binds to the CTLA4 receptors and hides them from the APC, then the affinity is expected to decrease along with the number of free CTLA4 receptors. Assuming again that a chemical equilibrium holds for this reaction, one can replace by where K is the constant of this equilibrium. Then, the new proportion of activated CD4+ T cells become:

Which proves the general form of Equation 5 if we include the constant of equilibrium K in the concentration . Since , this also proves that the constant in Equation 5 is greater than 2. Also, the larger the ratio between the affinity of APC for inhibitory and co-stimulatory receptors, the bigger the necessary concentration of anti-CTLA4 to activate a significant proportion of CD4+ T cell. Hence the greater this concentration, the greater the risk of unwanted activation of CD4+ T cell specific of antigens that are totally unrelated to the tumor, for example auto-antigens, and the bigger the risk of auto-immune disease.

One could object that , the proportion of free CD4+ T cell, is unlikely to be constant over time. For simplicity, it was nevertheless assumed. Another reason could be that : if the antigenic stimulation is large enough, APC could engage almost all CD4+ T cell and the vast majority of them should be either in the activated or in the inhibited state.

References

[1] G. Barendsen, “Dose fractionation, dose rate and iso-eﬀect relationships for normal tissue responses,” International Journal of Radiation Oncology\* Biology\* Physics, vol. 8, no. 11, pp. 1981–1997, 1982.

[2] R. G. Dale, “The application of the linear-quadratic dose-eﬀect equation to fractionated and protracted radiotherapy,” The British journal of radiology, vol. 58, no. 690, pp. 515–528, 1985.

[3] J. F. Fowler, “The linear-quadratic formula and progress in fractionated radiotherapy,” The British journal of radiology, vol. 62, no. 740, pp. 679–694, 1989.

[4] M. Tubiana, J. Dutreix, and A. Wambersie, “Radiobiologie. hermann,” 1986.

[5] C. Park, L. Papiez, S. Zhang, M. Story, and R. D. Timmerman, “Universal survival curve and single fraction equivalent dose: useful tools in understanding potency of ablative radiotherapy,” International Journal of Radiation Oncology\* Biology\* Physics, vol. 70, no. 3, pp. 847–852, 2008.

[6] J. Vaage, “Inﬂuence of tumor antigen on maintenance versus depression of tumor-speciﬁc immunity,” Cancer research, vol. 33, no. 3, pp. 493–503, 1973.

[7] A. Charbit, E. Malaise, and M. Tubiana, “Relation between the pathological nature and the growth rate of human tumors,” European Journal of Cancer (1965), vol. 7, no. 4, pp. 307–315, 1971.

[8] E. J. Hall and A. J. Giaccia, Radiobiology for the Radiologist. Lippincott Williams & Wilkins, 1993.

[9] Y. Latchman, C. R. Wood, T. Chernova, D. Chaudhary, M. Borde, I. Chernova, Y. Iwai, A. J. Long, J. A. Brown, R. Nunes, et al., “Pd-l2 is a second ligand for pd-1 and inhibits t cell activation,” Nature immunology, vol. 2, no. 3, pp. 261–268, 2001.