Abscopal benefits of localized radiotherapy depend on activated T cell trafficking and distribution between metastatic lesions

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Abstract

It remains unclear how localized radiotherapy for cancer metastases can occasionally elicit a systemic antitumor effect, known as the abscopal effect, but historically it has been speculated to reflect the generation of a host immunotherapeutic response. The ability to purposefully and reliably induce abscopal effects in metastatic tumors could meet many unmet clinical needs.

Here, we describe a mathematical model that incorporates physiological information about T cell trafficking to estimate the distribution of focal therapy-activated T cells between metastatic lesions. We integrated a dynamic model of tumor-immune interactions with systemic T cell trafficking patterns to simulate the development of metastases.

In virtual case studies, we found that the dissemination of activated T cells among multiple metastatic sites is complex and not intuitively predictable. Furthermore, we show that not all metastatic sites participate in systemic immune surveillance equally, and therefore the success in triggering the abscopal effect depends, at least in part, on which metastatic site is selected for localized therapy. Moreover, simulations revealed that seeding new metastatic sites may accelerate the growth of the primary tumor because T cell responses are partially diverted to the developing metastases, but the removal of the primary tumor can also favor the rapid growth of pre-existing metastatic lesions.

Collectively, our work provides the framework to prospectively identify anatomically-defined focal therapy targets that are most likely to trigger an immune-mediated abscopal response, and therefore may inform personalized treatment strategies in patients with metastatic disease.
**Major findings**

Using a mathematical model, we show that the distribution of activated T cells varies significantly between different metastatic sites and depends on the immune system activation site. Therefore, which metastases are chosen for localized therapy contributes to the potential for inducing systemic metastatic regression. We present a framework that could inform about patient-specific treatment targets – single or multiple – in metastatic patients, thereby supporting personalized clinical decision-making. The developed model offers additional insights into the dynamics of systemic disease, including metastases-mediated acceleration of primary tumor growth and rapid metastatic progression after surgical intervention.

**Quick Guide to Equations and Assumptions**

A general metastatic cancer setting with $N$ distinct tumors located in different organs, such as lung, liver, breast or kidney is considered. We advance a minimally parameterized model of immune system interactions within a single tumor site that was successfully compared to experimental data (1). The cancer population at each metastatic site, $C_i(t)$ for $i = 1,\ldots,N$, is assumed to follow a logistic growth with site-dependent carrying capacity, $K_i$, and growth rate, $r_i$. These populations are modulated by the predation of immunocompetent effector cells, $E_i(t)$. The equation governing growth of each metastatic site is
\[
\frac{dC_i}{dt} = r_i C_i \left( 1 - \frac{C_i}{K_i} \right) - \alpha p E_i C_i. \tag{A}
\]

A detailed description of all parameters with specific values can be found in Table 1. To limit model complexity and in the absence of evidence to the contrary, we assume the same parameter values for each metastatic site with the exception of possible differences in growth rates and carrying capacities.

Effector cells are recruited endogenously as well as in response to tumor burden, and may undergo spontaneous decay and exhaust from interactions with cancer cells. Cells that are recruited in response to the tumor are mainly cytotoxic T lymphocytes, a key component of the adaptive immune system. We extend Kuznetsov’s local model (1) to metastatic disease such that the equation governing the population of tumor-infiltrating effector cells at each of the metastatic sites is

\[
\frac{dE_i}{dt} = \lambda \left( E^* - E_i \right) - a(1 - p)E_i C_i + \sum_{j=1}^{N} \omega_{ij}(\bar{C}) \frac{fC_j}{g + C_j} E_j, \tag{B}
\]

where functions \(\omega_{ij}(C_i(t),\ldots,C_N(t))\) describe the probabilities that a cytotoxic T cell activated at site \(j\) will infiltrate the \(i\)th tumor site. The initial delay in establishing a robust immune response is incorporated in the time-dependent parameter \(f\) (compare Table 1) for each metastatic site. Probabilities \(\omega_{ij}\) depend on the anatomical distribution of metastatic sites and current tumor sizes.

After being activated in the tumor-draining lymph node, T cells travel through the lymphatic system, enter the blood circulation system, and travel in cycles through the network of arteries, capillaries and veins. To quantify T cell movement through the blood
system we distinguish four major coarse-grain flow compartments (COMPs): pulmonary circulation (LU); gastro-intestinal tract and spleen (GIS); liver (LI); and all other organs in the systemic circulation (SO), such as breast or kidney. The distinct consideration of LU, GIS, and LI is required as venous blood from GIS flows through LI via the hepatic portal vein, before being re-oxygenated in LU (Fig. 1A). T cell trafficking between these compartments is based on the anatomy, with rates being assigned as physiological values in the form of blood flow fractions (BFFs, % of cardiac output reaching the compartment). We denote $H_{COMP}$ as the probability that the T cell will extravasate at one of the metastatic sites after entering a given compartment, i.e. $H_{COMP} = \sum_{i=1}^{N_{COMP}} P_{ij}$, where $P_{ij}$ is the probability that a T cell activated at site $j$ will infiltrate the $i^{th}$ tumor site after entering a given compartment (Fig. 1B). We calculate the probability of T cell absorption by a given compartment ($W_{COMP}$) when starting from the LU compartment using a Markov Chain approach

$$W_{COMP} = \frac{1}{\Delta} \begin{cases} H_{LU} & \text{COMP} = \text{LU}, \\ H_{LI}(1-H_{LU})(BFF_{LI} + BFF_{GIS}(1-H_{GIS})) & \text{COMP} = \text{LI}, \\ H_{GIS}BFF_{GIS}(1-H_{LU}) & \text{COMP} = \text{GIS}, \\ H_{SO}BFF_{SO}(1-H_{LU}) & \text{COMP} = \text{SO}, \end{cases}$$

where $\Delta$ is a normalizing constant such that the sum of absorption probabilities over all four compartments is equal to one (see Supplementary Material for details). Without evidence of T cell decay in the circulating compartment of the blood system, each T cell will eventually be absorbed in one of the compartments, with the average number of systemic circulation cycles before absorption

$$N_{cycles} + 1 = \frac{1}{\Delta} \frac{1}{H_{LU} + (1-H_{LU})(H_{LI}(BFF_{LI} + BFF_{GIS}(1-H_{GIS}))+H_{GIS}BFF_{GIS} + H_{SO}BFF_{SO})}.$$ (C)
Probability $P_{ij}$ is equal to the probability that the T cell will flow through the tumor site multiplied by $h_{ij}$, the probability of extravasation from blood into the tissue. We approximate the former using the relative blood flow through a specific tumor bearing organ multiplied by the fraction of organ populated by the tumor and, thus, the equation describing $P_{ij}$ is

$$P_{ij} = h_{ij} \times \frac{BFF_{\text{organ}_i}}{BFF_{\text{COMP}_i}} \times \frac{V_i}{V_{\text{organ}_i}}.$$  

Experimental studies show that T cells are programmed during the activation process in the lymph node to express homing molecules that favor trafficking back to the site of the activation (2,3). Thus, we assume $h_{ij} = h_a$ if the T cell was activated in the given organ ($\text{organ}_i = \text{organ}_j$), and $h_{ij} = h_n$ otherwise ($1 \geq h_a > h_n$). The probabilistic model defined above allows the calculation of $\omega_{ij} = W_{\text{COMP}_i}P_{ij}/H_{\text{COMP}_i}$ in the effector cell dynamics equation (B) for different anatomical distributions of metastatic disease (see Supporting Material for details).

**Introduction**

Transformed cancer cells are confronted by both innate and adaptive immune surveillance, and it is believed that tumors have evolved mechanisms to evade the immune system even before they become clinically apparent (4). The notion of increasing immune-system efficacy by therapeutic intervention in order to systemically eradicate cancer cells has long been a vision of oncologists and cancer researchers. A particularly exciting development, hailed by the editors of Science as the scientific breakthrough of
is that novel immunotherapeutic strategies show remarkable responses in some patients, especially if combined with common cytotoxic agents. Radiotherapy and chemotherapeutic agents have been shown to substantially enhance tumor-specific immune responses in well-established tumors (4,6-11). The synergy between radiation and immunotherapy stems from radiation-induced effects including (i) immunogenic cell death that locally exposes a wealth of tumor antigens, and (ii) release of danger signals, which act as endogenous immune adjuvants to stimulate the activation of dendritic cells (DCs) (12) (Fig. 1C).

Most fascinating is the observation that the stimulation of the immune system by localized radiotherapy may modulate systemic regression of metastatic nodules, known as the radiation-induced abscopal effect (13). Such abscopal responses triggered by focal radiotherapy have been reported for multiple cancer types including renal cell carcinomas (14) and papillary adenocarcinomas (15), yet due to the high volume of patients with metastatic disease being treated with radiation therapy, many still consider these reports to be nothing more than anecdotal (16). However, with a combination of both irradiation and immunotherapy, abscopal responses can be triggered more reliably (12). A strong systemic response against squamous cell carcinoma in a murine model was observed when DCs were administered intratumorally following local irradiation (17).

The possibility of rationally inducing abscopal effects using radiotherapy and immunotherapy has the flavor of the long-sought “magic bullet” of cancer therapy, yet generating the synergy required to provide local control and also induce the abscopal effect is difficult. A myriad of factors contribute to this challenge, including i) heterogeneity of mechanisms a tumor can use to resist immune attack; ii) heterogeneity
of the capacity for an anticancer response by the different components of the immune system; and iii) anatomic- and time-dependent treatment effects. Numerous mathematical models have been developed to describe tumor-immune interactions at different phases of tumor progression (18-20), or to look at different pathways that can be exploited for immunotherapy (21-25). However, to our knowledge, models of metastatic cancer and both local and systemic immune system interactions currently do not exist. Thus, no prominent inroads have been made to decipher the contribution of local therapy to systemic disease modulation in metastatic patients. Herein we propose a tumor-immune system interaction modeling framework that incorporates a mathematical model of activated T cells trafficking between metastatic sites. In addition to numerous biological bottlenecks such as heterogeneities in immune infiltration, immune repertoire and antigen presentation, it is conceivable that an abscopal response can only be achieved if locally activated T cells are systemically distributed among the metastatic sites, and in numbers sufficient to tip immune surveillance back in favor of tumor eradication at each of these sites (26). We show that trafficking and distribution of activated T cells strongly depends on the anatomic distribution of metastatic sites, physiologic blood flow fractions to tumor bearing organs, tumor burden in each metastatic tissue, and the strength of cues that impact immune-cell homing (2,3). It follows that different metastatic sites may have varying potential to trigger a systemic response following focal immune-activating therapy. On the basis of the T cell homing distribution we determine optimum treatment targets in a virtual patient cohort. We integrate an extended tumor-immune interaction model (1) into the framework to simulate growth of each metastasis. This provides non-intuitive insights into how the seeding of a new metastatic site can promote the growth of
a primary tumor, and offers an elegant explanation of the observation of rapid progression of metastatic disease after surgical removal of a primary tumor (27-30).

Materials and methods

The different components of the mathematical framework discussed in the Quick Guide to Equations and Assumptions are calculated or solved numerically in MATLAB (www.mathworks.com). A detailed description of the computational methods can be found in Supporting Materials.

Model parameter estimation

The physiological blood flow fraction (BFF) to the spleen is estimated to be 3% (31). The gastro-intestinal tract consists mainly of stomach and esophagus (BFF = 1%), intestines (BFF = 16%) and pancreas (BFF = 1%), which together with the BFF to the spleen, yields $BFF_{GIS} = 21\%$. It is estimated that the internal mammary arteries with an average blood flow of 59.9 mL/min (32) provide approximately 60% of breast blood supply (33). With an average cardiac output of 300 L/h (34), we estimate $BFF_{\text{breast}} = 2\%$. BFF to the kidney is estimated to be 8.5% (31).

In a study performed by Mikhak et al. (2), Ovalbumin (OVA) specific T cells isolated from transgenic mice were activated in vitro with dendritic cells (DCs) isolated from different mouse tissues including lung, thoracic lymph nodes and skin. Populations of activated T cells were then injected into naïve mice challenged with aerosolized OVA. At 24h after the last challenge mice were sacrificed and lung tissue harvested to measure
OVA-specific T cell counts. T cells that were activated with lung DCs were present in the lung in numbers three times greater than T cells activated with DCs from different organs. Thus, we estimate the extravasation probability of T cell into the tissue of activation \((h_a)\) to be approximately three times larger then extravasation into non-activation tissue sites \((h_n, h_n/h_a = 1/3)\). Values of other parameters associated with T cell trafficking, i.e. those that are necessary to calculate values of \(\omega_{ij}(C_1,\ldots,C_N)\), were taken from the literature and are summarized in Table 2.

The basic structure of equations (A) and (B) are adapted from the Kuznetsov model (1), from which we adopt the parameter values that were estimated therein (Table 1).

**Virtual case studies and immunogenicity index quantification**

We create a cohort of 40 virtual metastatic patients with arbitrary combinations of breast, liver, kidney and lung metastases of random sizes between 50 and 300 mL. Given static information about patient-specific metastatic anatomy prior to therapy we investigate the homing distribution of T cells activated in response to localized therapy, such as radiotherapy applied to one of the tumors. To this extent we calculate values of \(\omega_{ij}\) for \(i=1,\ldots,N\) when activation occurred at the \(j^{th}\) site (see Eqn. (B)) for all sites of activation (without simulating the full temporal evolution model). We determine T cell dissemination quality for activation at a given site by calculating the ratio of the entropy of the established homing distribution to the entropy of uniform distribution

\[
S_j = \left( \sum_{i=1}^{N} \omega_{ij} \ln \omega_{ij} \right) / \left( \ln \frac{1}{N} \right),
\]

(D)
where \( j \) is the site of activation, and \( \omega_{ij} \) is the probability that a T cell activated at site \( j \) will infiltrate the tumor at site \( i \). Higher values of \( S_j \) indicate T cell homing distributions closer to uniform. The number of activated T cells depends on the number of tumor cells undergoing immunogenic cell death after local therapy, which in turn is dependent on tumor volume. Thus, we define the immunogenicity index of the \( i^{th} \) tumor, \( I_i \), by a combination of its homing distribution entropy with its size relative to the largest metastatic site.

\[
I_i = S_i \frac{V_i}{\max(V_1, \ldots, V_N)}.
\]  

(E)

The immunogenicity index can be considered to represent the systemic reach of activated T cells, which contributes to the probability of triggering a systemic abscopal effect. The maximum immunogenicity index of \( I_x=1 \) can only be achieved theoretically, that is if the largest tumor \( V_x \) is treated and the corresponding distribution of activated T cells is uniform between the metastatic sites (\( S_x=1 \)). For expected non-uniform distributions of activated T cells (\( S_i<1 \)), however, tumor volumes alone are insufficient to predict immunogenicity indices.

**Simulation of metastatic disease progression**

In simulations of tumor progression (solving the full model described by equations (A) and (B)), we arbitrarily initiate a breast tumor with \( 0.5 \times 10^6 \) cancer cells. After 200 days of simulated growth of this primary tumor, we simulate the onset of a metastasis by initiating a population of \( 0.5 \times 10^6 \) cancer cells in the lung, following which we simulate growth of both tumor sites for an additional 400 days. For the purpose of illustration we assume the growth rate of the metastatic site in the lung to be twice as fast as the growth
rate of the breast tumor. We assume extravasation probabilities $h_a=0.6$ and $h_n=0.2$.

*Simulation of primary tumor removal*

After simulating growth of the primary breast tumor and a lung metastasis, we mimic complete surgical removal of the primary tumor by instantaneously removing both cancer cell and T cell populations present in the breast. Parameters describing intrinsic lung metastasis dynamics remain unchanged.

**Results**

*Variation in immunogenicity between metastatic sites*

Intuitively, if T cells are unable to extravasate at any site other than the tissue of activation, i.e. $h_n/h_a=0$, then no systemic response is possible. On the other hand, if extravasation occurs at all sites with equal strength (i.e., $h_n/h_a=1$), the systemic T cell distribution is independent of the tissue of activation. Thus, it is important to investigate the intermediate values of the $h_n/h_a$ ratio. We begin with a detailed analysis of a virtual case study of breast (113 mL), liver (220 mL) and lung (270 mL) tumors. Fig. 2 shows the homing distribution of T cells for the different sites of immune system activation with $h_a=0.6$ and $h_n/h_a=1/3$. It can be seen that the distribution of activated T cells depends on the site of activation. The probability that an activated T cell will home to the breast metastasis is only larger than 10% if the immune system was activated at that site (or possibly another site located also in the breast). The highest value of normalized entropy of T cell homing distribution is obtained for activation in breast ($S_{\text{breast}}=0.85$). Activation in lung or liver attracts about 98% of all activated T cells back to these two organs ($S_{\text{lung}}=0.43$, $S_{\text{liver}}=0.66$). Fig. 3 panels A, B, and C show the distribution of T cells for
immune system activation in breast, liver and lung, respectively, for different values of parameters $h_a \in [0,1]$ and $h_n/h_a \in [0,1]$. Simulations reveal that the actual extravasation probability at the tissue of activation, $h_a$, has a negligible influence on the final systemic T cell distribution, but has a large impact on the temporal component of T cell trafficking, i.e. the average number of circulatory cycles performed before extravasation. Experimental derivation of the extravasation probability becomes imperative for the design of optimum local therapy fractionation protocols for immune response induction. Final T cell distribution is mostly determined by the ratio of extravasation probabilities at non-activation sites versus activation sites, $h_n/h_a$ (Fig. 3 panels A, B, and C). For intermediate values of $h_n/h_a$, including estimated $h_n/h_a = 1/3$, the T cells dissemination patterns vary non-intuitively across activation sites.

Given the complexity of the arterial tree, trafficking through a specific site is a relatively rare event (evident from the BFFs reported in Table 2). In addition, $h_a$ and ratio $h_n/h_a$ strongly impact the number of cycles that the T cells will remain in transit in the circulatory system before extravasating at a tumor-harboring site (Fig. 3D). It follows from equation (C) that the average number of cycles increases with smaller values of $h_a$. In the case of immune system activation in the breast, if $h_a=1$ and $h_n/h_a=1$, T cells will perform an average of approximately 8 circulatory transit cycles before extravasation. With a blood recirculation time of 10 to 25 seconds (35), this translates to around 2 minutes. For lower values of $h_a=0.05$ and $h_n/h_a=0$, our model suggests that a T cell could complete approximately 4,500 circulatory cycles (~32 hours) before extravasation.
Comparing the entropy between T cell homing distributions (equation (D)) shows that, in this virtual patient example, activated T cells distribute most uniformly after treating the breast tumor metastasis with focal radiotherapy as the immune system activating agent (Fig. 4A), regardless of extravasation probabilities $h_d$ and $h_n$. Targeting the lung tumor, however, yields a heavily skewed distribution of T cells towards the lung, given the large $BFF$ to this site. Integration of tumor volumes to calculate the immunogenicity index (equation (E)), the metric we propose to support clinical decision making, reveals that the liver metastasis has the highest immunogenicity index for $h_d/h_a$ values between 15% and 60%, including the estimated value of $h_d/h_a = 33\%$ ($I_{\text{liver}}=0.49$, $I_{\text{breast}}=0.38$, $I_{\text{lung}}=0.34$) (Fig. 4B).

We additionally calculated which site has the highest immunogenicity index in a virtual patient cohort for the estimated values of $h_d/h_a = 1/3$ and $h_d=0.6$ (Fig. 4C). Calculations suggest that largest tumor volume or a specific combination of metastases is not necessarily always an indicator of maximum system response or optimal treatment target for an individual patient. A much smaller breast tumor (53 mL) has a higher immunogenicity index than the much larger kidney tumor (254 mL) in virtual patient 5 (Fig 4C). Virtual patients 20 and 21 have the same combination of virtual sites of metastatic tumors, but the tumor with the highest immunogenicity index is in different organs. Therefore, the identification of treatment targets with the highest likelihood of inducing a systemic abscopal response is highly patient-specific and not predictable from the average response of historical cases.
Systemic interdependence of metastatic sites through activated T cell trafficking

Simulation of primary tumor growth for 200 days shows that a primary tumor can be modulated by a cytotoxic immune surveillance, keeping the tumor in a dormant state in equilibrium with the immune system (26) and below the imposed carrying capacity (Fig 5A). Seeding of a metastatic site disrupts the dynamic equilibrium between T cells and cancer cells in the primary site. Before the immune response against the metastasis is established locally, some of the T cells generated in the primary breast tumor traffic and extravasate at the metastatic site, allowing for transient immune escape and progression of the primary tumor (Fig 5B). After T cells activated by the lung metastasis begin trafficking in the circulatory system, the total number of T cells increases and an increase in T cells surveilling the primary breast cancer can be observed. However, because of the attained equilibrium of homing distributions (Fig 5C and 5D), the now lower proportion of T cells penetrating the breast allows the tumor to attain dormancy with cell numbers larger than during the time period prior to metastasis formation. Interestingly, the lung metastasis is also kept at a dormant state despite its larger growth rate, mostly due to T cells that are generated in the breast and trafficking to the lung. It is worth pointing out that due to its assigned large proliferation rate, a single tumor site in the lung without a pre-existing breast tumor would escape immune surveillance and grow close to its imposed carrying capacity (Figure S2). Thus, surgical removal of the breast tumor results in removal of locally infiltrated T cells as well as prevention of future T cells activated in the breast. This leads to the escape of the lung metastasis from immune surveillance due to the rapid decrease of the effector-to-cancer cell ratio in the lung (Fig. 5A).
Discussion

Progression from localized to metastatic disease sharply diminishes patient prognosis. For example, in 10-15% of breast cancer patients, the second most common cancer diagnosed worldwide, metastasis will develop within 3 years of diagnosis (36) decreasing the 5-year survival from >80% to around 26% (37). When targeting both local and metastatic disease, immunotherapy has been shown to synergize with local radiation (6-12), which is currently being explored in more than 10 active clinical trials (12). The biological and physical principles underlying this synergy are yet to be completely understood. The complexity arises from the dynamic interplay between the immune system and the tumor, which is further confounded when examining local as well as systemic dynamics. The increasing number of case reports of abscopal effects in metastatic tumors distant to the areas targeted by radiation (14,15) encourages further investigation into the possibility of intentionally triggering such responses. We hypothesized that different metastatic sites in individual patients may have varying potential to induce a systemic response, and that mathematical modeling could help to identify the most promising treatment targets prior to intervention on a per patient basis.

A myriad of complex biological cascades are without doubt involved in the development of abscopal effects. While T cell trafficking may only be one contributing mechanism to an abscopal response, it follows physiologic blood flow and hence has an element of predictability. Identifying local treatment targets with the highest systemic reach of activated T cells promises to increase the likelihood of purposely inducing abscopal
effects. We propose a systemic tumor-immune interaction framework, which accounts for activated T cell trafficking through the host circulatory system. Patient-specific distribution of metastatic sites can be acquired from routine radiology scans including CT, MRI, and PET/CT. When combined with physiological information about T cell trafficking, the model estimates the distribution of focal therapy-activated T cells for each metastatic site. Using a virtual patient cohort we showed that the activated T cell distribution is dependent on (i) the anatomic distribution of metastatic sites, (ii) the tumor volume of each metastasis, and (iii) the site of activation. In the clinic, once patient-specific anatomic metastatic distribution and individual tumor volumes are obtained from radiology, the presented framework can calculate proposed immunogenicity indices for each of the metastatic sites. Those values may support clinical decision making as to which metastatic sites serve as the most promising local treatment targets to induce systemic abscopal responses, which can be further enhanced through various immunotherapeutic approaches (12).

Radiotherapy may be the most intuitive local treatment choice as the abscopal effect has been observed clinically after focal radiation both with and without immunotherapy (14,15). The framework may be equally applicable to other potentially immune-activating therapies such as radiofrequency ablation (high frequency current to heat and coagulate tissues), cryoablation (extreme cold to cause tumor destruction) or photodynamic therapy (drug activated by light of suitable wavelength) (38). Before translation into a prospective clinical trial to validate clinical decision support, however, the model needs to be validated with retrospective clinical data, and estimated parameters need to be calibrated.
with the results of experimental studies. Of utmost importance is the tracking of labeled T cells and measurement of their distribution amongst different tissues and metastatic sites, as well as quantification of the homing rate. This information would allow the determination of optimum time intervals between radiotherapy fractions to facilitate a strong immune response. In future work, the SO compartment may be further divided into anatomically distinct and important areas such as bone, skin, muscle and central nervous system. Bone in particular is often treated with focal radiation for definitive or palliative control (39), and thus the explicit simulation of T cell trafficking to the bone is of clinical importance.

The presented framework reproduced several established key features of local tumor-immune interactions including long-term tumor dormancy (19,26), but additionally revealed a previously unappreciated interdependence of metastatic sites through activated T cell trafficking. First, the onset of a new metastatic site can boost the growth of the primary tumor by diverting T cells to the distant metastasis and therefore facilitating a transient escape from immune-induced dormancy. This might offer yet another angle to the Norton hypothesis that tumors may not metastasize because they are large, but may be large because they are (self-)metastatic (40). Second, immune system activation and activated T cell trafficking from the primary tumor may impede the growth of a metastatic site by mounting an immune response in the form of movement of T cells from the primary to the metastasis, a process that is degraded at the time of resection of the primary. This augments an understanding of the modulation of distant tumors from the primary, a phenomena recognized more than 100 years ago and termed ‘concomitant
immunity’ (41). Third, surgical removal of the primary tumor may trigger progression of existing metastatic sites which has been seen in the clinic (27-30) as well as in animal experiments (42,43) and mathematical models (44,45). With 93% of breast cancer (early stage I and II), 98% of colon cancer (stage I, II & III), and 71% of non-small cell lung cancer (early stage I and II) patients undergoing surgery as a part of their treatment (46), systemic perturbation by the presumed local treatment needs to be explored further.

Herein we present the first attempt to quantify how local tumor-immune interactions may propagate systemically, which may lead to the prediction of patient-specific treatment targets to trigger abscopal effects. For illustration purposes we utilized the established Kuznetsov model of the interaction of local tumor with the immune system, starting with parameter values developed using data obtained from a mouse lymphoma model (1). This module of the framework, however, can be interchanged for other models of tumor growth and immune modulation. In future work, either the Kuznetsov model or its replacement will need to be calibrated with organ-specific tumor growth kinetics and immune surveillance to fully integrate local and systemic dynamics and confidently support personalized decision making in the clinic. Furthermore, the model of tumor-immune interactions for each local site may be expanded to include other cell types, as well as tissue specific parameterizations of carrying capacity, growth rates, and immune interaction parameters. The blood flow associated trafficking model can be also utilized to explore the metastatic dissemination patterns, which is currently being investigated by others (47).
Acknowledgements

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References


### Tables

**Table 1.** Parameter values for the differential part of the model, equations (A) and (B), taken from (1).

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
<th>Unit</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>$r$</td>
<td>0.19 (for tumor located in breast)</td>
<td>1/day</td>
<td>Maximal tumor growth rate</td>
</tr>
<tr>
<td></td>
<td>0.38 (for tumor located in lung)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>$K$</td>
<td>$531.9 \times 10^6$ cells</td>
<td></td>
<td>Carrying capacity</td>
</tr>
<tr>
<td>$a$</td>
<td>$0.14 \times 10^{-6}$ 1/day/cells</td>
<td></td>
<td>T cell – cancer cell interactions constant</td>
</tr>
<tr>
<td>$p$</td>
<td>0.998</td>
<td>non-dimensional</td>
<td>Probability that during the interaction between T cell and cancer cell the latter will be killed</td>
</tr>
<tr>
<td>$\lambda$</td>
<td>0.59</td>
<td>1/day</td>
<td>Effector cells decay rate</td>
</tr>
<tr>
<td>$E^*$</td>
<td>$0.3 \times 10^6$ cells</td>
<td></td>
<td>Physiological level of effector cells</td>
</tr>
<tr>
<td>$f$</td>
<td>0.53 if time since metastatic site creation $&gt; 28$ days and 0</td>
<td>1/day/cells</td>
<td>Magnitude of immune system stimulation by the presence of cancer cells</td>
</tr>
</tbody>
</table>
otherwise
\[ g = 0.16 \times 10^6 \text{ cells} \]

Immune stimulation damping coefficient

**NOTE:** \( C(t) \) measures cell number, but in the T cell trafficking part of the model its value is used in terms of tumor volume in milliliters according to the formula \( V(C(t)) = C(t)\times(\frac{4}{3}\pi r^3)\times10^{-12} \text{ mL} \), where \( r \) is the diameter of the cell assumed to be 10 micrometers.

**Table 2.** Parameter values for the T cell trafficking part of the model.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
<th>Description</th>
<th>Reference</th>
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</thead>
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<tr>
<td>( BFF_{LI} )</td>
<td>6.5%</td>
<td>Blood flow to liver</td>
<td>(31)</td>
</tr>
<tr>
<td>( BFF_{GIS} )</td>
<td>21%</td>
<td>Blood flow to gastrointestinal tract estimated</td>
<td></td>
</tr>
<tr>
<td>( BFF_{SO} )</td>
<td>72.5%</td>
<td>Blood flow to SO compartment by definition = 100% - ( BFF_{LI} - BFF_{GIS} )</td>
<td></td>
</tr>
<tr>
<td>( BFF_{breast} )</td>
<td>2%</td>
<td>Blood flow to the breast estimated</td>
<td></td>
</tr>
<tr>
<td>( BFF_{kidney} )</td>
<td>8.5%</td>
<td>Blood flow to the kidney</td>
<td>(31)</td>
</tr>
<tr>
<td>器官</td>
<td>容积 (mL)</td>
<td>平均容积</td>
<td>参考文献</td>
</tr>
<tr>
<td>-------</td>
<td>----------</td>
<td>----------</td>
<td>----------</td>
</tr>
<tr>
<td>肝脏</td>
<td>$V_{\text{liver}}$</td>
<td>1493 mL</td>
<td>(48)</td>
</tr>
<tr>
<td>乳腺</td>
<td>$V_{\text{breast}}$</td>
<td>500 mL</td>
<td>(34)</td>
</tr>
<tr>
<td>肺</td>
<td>$V_{\text{lungs}}$</td>
<td>3679 mL</td>
<td>(49)</td>
</tr>
<tr>
<td>肾脏</td>
<td>$V_{\text{kidney}}$</td>
<td>249 mL</td>
<td>(50)</td>
</tr>
</tbody>
</table>
Figure legends

**Figure 1. Activated T cell trafficking.** (A) Activated cytotoxic T cells (CTLs) enter the blood system via the great veins, flow through the pulmonary circulation and then continue into systemic circulation. Venous blood from the gastrointestinal tract and spleen goes to the liver through the hepatic portal vein. (B) CTL can flow through each compartment without reaching any of the tumor sites (Mi, circles), or flow through one of them. At the metastatic site T cell has a certain probability to extravasate, $h_a$ if it has been activated by that site and $h_n$ otherwise. We denote by $H_{\text{compartment}}$ the overall probability that T cell will extravasate in the given compartment. (C) Radiotherapy (RT) triggers immunogenic cell death that activates dendritic cells (DCs) that subsequently travel to the lymph node. DCs transform naïve T cells into CTLs, which, in case of non-metastatic disease, traffic through the blood system back to the tumor site for cancer cell eradication.

**Figure 2. Model-predicted homing distribution for different activation sites.** Results for a virtual patient with breast (113 mL), liver (220 mL) and lung (270 mL) metastases with $h_a = 0.6$, $h_n/h_a = 1/3$. Activation in breast yields the most uniform distribution ($S_{\text{breast}} = 0.85$ vs $S_{\text{liver}}=0.66$ vs $S_{\text{lung}}=0.43$). Values of other parameters used in calculations are reported in Table 1.

**Figure 3. T cell homing distribution for different activation sites and extravasation probabilities.** Model-predicted homing distributions between metastatic sites in a virtual
patient with breast (113 mL), liver (220 mL) and lung (270 mL) metastases for different sites of activation (A) breast, (B) liver, (C) lung and for different values of extravasation probabilities $h_n$ and $h_a$. Rectangles correspond to the narrow ranges around the value of $h_n/h_a$ estimated from the literature. (D) The average number of transitions between model compartments before extravasation at one of the metastatic sites for different sites of activation and different extravasation probabilities $h_n$ and $h_a$. Calculations were performed using parameters reported in Table 2.

Figure 4. T cell trafficking and extravasation for different activation sites in a virtual patient with breast (113 mL), liver (220 mL) and lung (270 mL) metastases (A) normalized entropy values (equation (D)). (B) immunogenicity indices (equation (E)) for different extravasation probabilities $h_n$ and $h_a$. (C) Analysis of 40 virtual case studies of possible metastatic tumors in lung, liver, kidney and the breast. Circles denote existence of the metastatic site and radii correspond to tumor sizes. Black background identifies the tumor with the highest immunogenicity index for $h_n/h_a = 1/3$ and $h_a=0.6$. Calculations were performed using parameters reported in Table 2.

Figure 5. Partial removal of T cell surveillance and metastatic progression after surgery. (A) Model-predicted growth of a primary breast tumor before and after the onset of a lung metastasis at $t = 200$ days, and surgical removal of the primary breast tumor and its local effector cells at day 600. (B) Corresponding T cell/tumor cell ratios for the tumors shown in panel (A). (C) and (D) Homing distributions of T cells that are
activated in the breast tumor and lung metastasis, respectively. Simulations were performed with $h_a = 0.6$, $h_n = 0.2$, and parameters reported in Tables 1 and 2. $r_{\text{breast}}=0.19$/day, $r_{\text{lung}}=0.38$/day.
A Pulmonary | Systemic
---|---
GI TRACT AND Spleen | LIVER
OTHER ORGANS | Compartment

B

C

(1) RT TUMOR
(2) Activated DCs
(3) CTLs
BLOOD SYSTEM
LYMPH NODE

Incoming CTLs

1-h
1-ha
1-H compartment

Figure 2

- Activation in breast: 31% (Lung), 57% (Liver), 12% (Breast)
- Activation in liver: 2% (Lung), 34% (Liver), 13.6% (Breast)
- Activation in lung: 1.5% (Lung), 84.9% (Liver), 0% (Breast)
Abscopal benefits of localized radiotherapy depend on activated T cell trafficking and distribution between metastatic lesions

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