Appendix 1

We employ a backward finite difference method for solving the coupled PDEs of Eqn. set 1. Equations are made unitless by scaling the time by T = 24 h and the length by L = 3 cm. The mesh is defined by $x = x_i = i\Delta x$ and $t = t_j = j\Delta t$, where Δx = 0.017 cm and Δt = 0.5 h. The initial conditions are:

$$n(x,0) = 0.6e^{-x^2/L^2}$$
$$v(x,0) = 0.02$$

with all other compartments initially unoccupied. The diffusion term at each subsequent time point $j + 1$ for each cell type y is

$$\frac{\partial^2 y_i^{(j+1)}}{\partial x^2} = (y_{i-1}^{(j)} - 2y_i^{(j)} + y_{i+1}^{(j)})/(\Delta x)^2 ,$$

and the gradient is given as

$$\frac{\partial y_i^{(j+1)}}{\partial x} = (y_i^{(j)} - y_{i-1}^{(j)})/\Delta x .$$

For the boundaries each equation assumes the following:

$$y_i = \begin{cases} y_1 & \text{if } i = -1 \\ y_i & \text{if } 0 \le i \le L/\Delta x \\ y_{L/\Delta x - 1} & \text{if } i = L/\Delta x + 1. \end{cases}$$

Appendix 2

Understanding the appropriate bounds for some of these parameters is difficult. Some parameters are based on very limited experimental or clinical data, so the reasoning for all parameter estimations is detailed here.

A2.1 Diffusion Coefficients (D_n, D_h, D_v)

The expansion of a tumor represented by a diffusion coefficient gives a measure of motility for the cellular population as a whole. The value we use for the diffusion of the normoxic population is $D_n = 7.3 \times 10^{-9}$ cm^2⁄s, which has the same order of magnitude as similar parameters from the literature (44, 5).

Hypoxia is a promoter of invasiveness (45), so we assume that the hypoxic population has a slightly greater diffusion coefficient than the normoxic population ($D_h = 1.7D_n = 1.2 \times 10^{-8}$ cm^2/s). On the other hand, we choose the diffusion coefficient of the vascular cells to be *less* than that of the normoxic and hypoxic cells. Anderson and Chaplain use a value of $D_v = 10^{-10}$ cm^2/s (46), and Orne and Chaplain state that reasonable values for vascular diffusion should be in the range of 10^{-9} - 10^{-11} cm^2/s (47). Our diffusion coefficient for vascular cells is faster ($D_v = 0.8D_n = 5.8 \times 10^{-9}$ cm^2/s) and just outside this window but within an order of magnitude.

While D_n is a major contributor to TVDT, increasing D_h and D_v (as with increasing D_n) leads to faster growth (smaller TVDT). These values are adjusted to fit the specified TVDTs from the statistical population model. The primary diameter at the threshold of lethal metastatic burden is also affected by these values. This critical diameter is fit

better with smaller values for D_h and D_v for both the lung and the breast, so a compromise is made to fit both metrics with these values.

A2.2 Proliferation Rates (μ_n, μ_v)

Despite heterogeneity in the proliferation rates of tumor cells, we assume that all cells in the tumor and across both tissue types divide at the same rate (16 h cycle time) to focus on other aspects of the dynamics. Doubling times are observed in vitro to average around 15 h for breast cells and 24 h for lung cells (48). Values used in other models tend to stay within this window (5, 49).

Tumor vasculature is lined with actively dividing endothelial cells (6). Hobson et al. estimate a vascular doubling time of 2.4 - 13 days for mammary cancers in mice (50), so their fastest measure is nearly equal to our rate for the breast vasculature doubling time (2.1 days). The vascular doubling time we use in the lung simulation is even faster and lies farther outside this range (0.83 days), yet still slower than the doubling of the normoxic cells. Values more within this range, like the weekly vascular doubling time from Hinow et al. (5), were too slow to match the growth of the tumor determined by this equation set.

We don't consider proliferation in the hypoxic fraction. Coexistence of both hypoxia and proliferation has been observed and is most prominent at 100 – 150 μm from vessels (51), however, both oxygen and nutrients are required for cell proliferation so it is generally not expected at low oxygen tensions.

A2.3 Chemotactic Constant (χ)

The chemotactic constant also has an effect on the TVDT. Increasing χ increases the growth rate of the primary tumor. Endothelial cells migrate toward positive gradients of VEGF (6), and the resulting vascular accumulation in turn increases normoxic cell proliferation and hypoxic reoxygenation.

Orme and Chaplain use a value that is 10 times their D_n (47), but estimates are found that range from $\chi = 10^{-4} - 10^{-7}$ cm^2/s (44, 5). We use $\chi = 1.5 \times 10^{-8}$ cm^2/s, which is around twice the value of the non-directed normoxic migration.

A2.4 Deoxygenation and Reoxygenation (α_n, α_h)

As the tumor grows larger and more dense, the build-up of pressure causes blood vessels within to collapse. Temporary occlusions may lead to reversible hypoxia. Lanzen et al. measure acute hypoxia caused by fluctuating vascular occlusions, recording frequencies of more than 1 cycle every 3 minutes (52), but this fast rate is more of an individual vessel measure than one that can be used with continuous cellular densities. Hinow et al. estimate that half of the cell population will become hypoxic within 7 h (5). This model's values for the rate of deoxygenation range from $4 \times 10^{-3} - 6 \times 10^{-5}$ h^{-1}, which are much smaller than the aformentioned estimates because they are switched on from the outset and remain turning over, instead of switching on and off due to a critical threshold of oxygen concentration. We choose reoxygenation to be somewhat slower ($\alpha_h = 0.7\alpha_n$) than the rate of deoxygenation.

A2.5 Intravasation (β_n, β_h)

Metastasis might be an approach to find a more suitable environment or to overcome hypoxia by losing adhesiveness and increasing motility (34). It may also occur by being at the right place at the right time. Though it seems that hypoxic cells might get more easily drawn into the circulation, the normoxic cells are already in contact with the vasculature. Further analysis was done to determine how to adjust these parameters.

The landscapes in Fig. 7 show how the relation between total rate of entry into the circulation and the fraction of this total that comes from the normoxic population affects the diameter of the primary at which lethal metastases are shed. Once again, for the breast tumor, there are several combinations that lead to the same results. For the lung tumor, as long as the rate of entry is large enough, it doesn't seem to matter which compartment they are coming from. The lung tumor simulation, though, does have a slight shift toward smaller diameters when β_n is greater than β_h, so this influences our choice for the larger entry rate from the hypoxic population.

A2.6 Death Rates (δ_n, δ_h, δ_v)

The high interstitial pressure within the tumor can lead to apoptotic signals and collapsing blood vessels, which may be followed by cellular and vascular death (53). It is difficult to measure cell death, as its rarity and inconsistency makes it difficult to capture and measure a rate, but cell loss is most pronounced in the hypoxic fraction (54). Chronically hypoxic cells are viable from hours to days, with an upper limit around 4 days (54, 55, 38, 56). This model works on a continuous population, so single cell metrics will overestimate the rates.

In the model, increasing δ_h slows the growth of the primary for both the lung and breast simulations by moving more of the hypoxic fraction that summons the vasculature into an irreversible necrotic state. We adjust this value to make the best fit, and we set the death rate of the hypoxic cells to be greater than that of the normoxic and vascular portions.

Appendix 3

Obtaining a TVDT from exponential growth, as seen with the lung simulation, is straightforward. With exponential growth ($V = V_0 e^{rt}$) the growth rate is $r = \ln(2)/\tau$ in terms of the doubling time τ. We simply calculate

$$\tau = \frac{\ln 2}{r} \qquad (1s)$$

for the doubling time after fitting the tumor growth to an exponential.

To calculate a dynamic TVDT from power law growth we first consider the volume at time t_1 and then again at time t_2 when the volume has doubled. So we have

$$V_1 = ct_1^n$$
$$2V_1 = ct_2^n,$$

assuming constant c and n. After scaling one by the other and rearranging, we get $t_2 = t_1 2^{1/n}$. The TVDT then is given by TVDT = $t_2 - t_1 = t_1(2^{1/n} - 1)$. In terms of the diameter of the spherical tumor, we have

$$\mathrm{TVDT} = \left(\frac{\pi d^3}{6c}\right)^{1/n} (2^{1/n} - 1), \quad \text{(2s)}$$

where c and n are both found from fitting the power law curve. We can then relate the dynamic TVDT of the tissue model to the static TVDT of the statistical population model by calculating the doubling time at an average tumor size (d) of around 3 cm.

Appendix 4

The total hypoxic turnover is given by the 2nd equation in Eqn. set 1. Ignoring the spatial component (the diffusion term), we simplify this to:

$$\frac{\partial h}{\partial t} \to \frac{dh}{dt} = -\alpha_h h v + \alpha_n n(1-v) - \beta_h h v - \delta_h h.$$

Solutions for the hypoxic turnover for both the breast and lung tumors are larger at the leading edge but nearly zero within the interior. For the interior, we approximate a zero flux ($dh/dt \sim 0$) and rearrange, solving for the vascular density:

$$v \sim \frac{1 - \sigma\left(\frac{\delta_h}{\alpha_h}\right)}{1 + \sigma\left(1 + \frac{\beta}{\alpha_h}\right)}$$

where $\sigma = \alpha_h h/\alpha_n n$ represents the reoxygenation flux over the deoxygenation flux. This equation can be further simplified by recognizing that the rate of entry into the circulation is much less than the rate of reoxygenation ($\beta_h/\alpha_h << 1$), which always holds true by several orders of magnitude (c.f. Table 1). The resulting equation is:

$$v \sim \frac{1-\sigma\left(\frac{\delta_h}{\alpha_h}\right)}{1+\sigma}.$$