Combining two strategies to improve perfusion and drug delivery in solid tumors

Triantafyllos Stylianopoulos[a] and Rakesh K. Jain[b,1]

[a]Department of Mechanical and Manufacturing Engineering, University of Cyprus, 1678 Nicosia, Cyprus; and [b]Edwin L. Steele Laboratory, Department of Radiation Oncology, Massachusetts General Hospital, Harvard Medical School, Boston, MA 02114

Blood perfusion in tumors can be significantly lower than that in the surrounding normal tissue owing to the leakiness and/or compression of tumor blood vessels. Impaired perfusion reduces oxygen supply and results in a hypoxic microenvironment. Hypoxia promotes tumor progression and immunosuppression, and enhances the invasive and metastatic potential of cancer cells. Furthermore, poor perfusion lowers the delivery of systemically administered drugs. Therapeutic strategies to improve perfusion include reduction in vascular permeability by vascular normalization and vascular decompression by alleviating physical forces (solid stress) inside tumors. Both strategies have shown promise, but guidelines on how to use these strategies optimally are lacking. To this end, we developed a mathematical model to guide the optimal use of these strategies. The model accounts for vascular, transvascular, and interstitial fluid and drug transport as well as the diameter and permeability of tumor vessels. Model simulations reveal an optimal perfusion region when vessels are uncompressed, but not very leaky. Within this region, intratumoral distribution of drugs is optimized, particularly for drugs 10 nm in diameter or smaller and of low binding affinity. Therefore, treatments should modify vessel diameter and/or permeability such that perfusion is optimal. Vascular normalization is more effective for hyperpermeable but largely uncompressed vessels (e.g., glioblastomas), whereas solid stress alleviation is more beneficial for compressed but less-permeable vessels (e.g., pancreatic ductal adenocarcinomas). In the case of tumors with hyperpermeable and compressed vessels (e.g., subset of mammary carcinomas), the two strategies need to be combined for improved treatment outcomes.

tumor microenvironment | vessel decompression | vessel permeability | mechanical forces | mathematical modeling

Perfused vessels are necessary for enabling adequate oxygenation and distribution of systemically administered drugs in solid tumors. However, perfusion rates in some regions of a tumor can be significantly lower than that in the peritumor normal tissue, leading to hypoxia, low pH, and inadequate drug delivery. Impaired blood perfusion in tumors could result from (*i*) excessive fluid loss from the vasculature to the interstitial space owing to vessel hyperpermeability (Fig. 1*A*), (*ii*) increased resistance to fluid flow caused by vessel tortuosity, and (*iii*) reduced effective cross-sectional area for blood flow due to vessel compression (Fig. 1*C*) (1, 2). Vessel hyperpermeability and tortuosity can be lowered using judicious doses of antiangiogenic agents (Table 1), which increases pericyte coverage and fortifies the leaky vessels without excessive pruning of vessels. This strategy—referred to as "vascular normalization"—can improve tumor perfusion and thereby increase oxygen and drug delivery as well as treatment efficacy (1, 3–6). Fig. 1 *A* and *B* shows a schematic of how vascular normalization can improve perfusion by improving the structure and composition of the vessel wall, and Table 1 summarizes evidence in various preclinical tumor models and in cancer patients.

Vessel compression is a result of physical forces—referred to as solid stress—accumulated within solid components of tumors (cancer and stromal cells, collagen, and hyaluronan) (23–25).

Stress alleviation can be achieved by depletion of any or all of these components, which can reopen compressed vessels and improve perfusion and delivery of drugs (25, 26). The schematic in Fig. 1 *C* and *D* shows how depletion of cancer or stromal cells can decompress blood vessels, and Table 2 presents therapeutic agents that have been used to deplete components of tumors and decompress tumor vessels.

One challenge now is to better understand under what conditions vascular normalization and vessel decompression improve perfusion in solid tumors and to develop guidelines for optimal use alone and in combination. To this end, we use a mathematical model for fluid flow and drug transport in tumors that accounts explicitly for the geometry and leakiness of the blood vessels. Our model accounts for preclinical and clinical data and suggests guidelines for effective use of these two therapeutic strategies. The tumor vasculature is represented as a percolation network with one inlet and one outlet (31, 32) (Fig. S1). The baseline value for the diameter of the vessels is set at 15 μm (1), whereas the leakiness is defined by the size (i.e., diameter) of the pores of the vessel wall. The pore size in our analysis varies from 50 nm (poorly permeable tumor vessels) to 400 nm (hyperpermeable vessels) (33). To quantify vascular efficiency we calculate the fraction of perfused vessels and the effective vascular density. Perfused vessels are considered to be those with velocities greater than 0.1 mm/s (34), whereas the effective vascular density is the density of the perfused vessels. Perfusion is considered optimal when these two parameters are maximal.

Results

Vessel Decompression Improves Perfusion in Tumors with Abundant Compressed but Poorly or Moderately Permeable Vessels. We first investigated under what conditions decompression of blood vessels with stress-alleviation treatment can improve tumor perfusion. We modeled vessel decompression by increasing the diameter of the vessels to the baseline value. The effects for different vessel wall pore sizes are shown in Fig. 2, and in Figs. S2

Significance

Vascular normalization and stress-alleviation treatments are promising strategies to improve tumor perfusion and delivery of drugs. Vascular normalization is more effective for tumors with hyperpermeable but uncompressed vessels, whereas stress alleviation is more beneficial for compressed but low-permeable vessels. In the case of tumors with hyperpermeable and compressed vessels, the two strategies need to be combined for improved treatment.
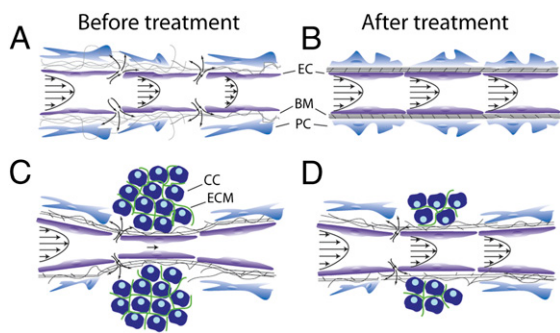

Fig. 1. Schematic of therapeutic strategies to improve tumor perfusion. (*A*) Abnormalities in interendothelial junctions, pericyte coverage, and/or basement membrane lead to hyperpermeability of tumor blood vessels and excessive fluid leakage that slows down blood flow. (*B*) Vascular normalization fortifies the vessel wall, resulting in smaller interendothelial gaps ("pores") and improved perfusion. (*C*) Structural components of the tumor microenvironment exert forces on blood vessels, resulting in vessel compression and reduced blood flow. (*D*) Alleviation of these forces by selective depletion of tumor constituents (e.g., cells or extracellular matrix) can decompress the vessels and improve vessel perfusion. BM, basement membrane; CC, cancer and/or stromal cell; EC, endothelial cell; ECM, extracellular matrix; PC, pericyte.

and S3 for compressed central regions occupying 80% and 30% of the tumor, respectively. The figures show that our results are qualitatively independent from the region of vessel compression. For pore sizes less than 200 nm (low or moderately permeable tumors), there is a strong dependence of the fraction of perfused vessels and the effective vascular density on increases in vessel diameter. Recently, we found that stress-alleviation therapy in murine pancreatic ductal adenocarcinomas increased vessel diameter by 20% (from 10 to 12 μm) and the perfused vessel fraction by 47% (from 0.32 to 0.47) (25). Our model predictions are within this range and, moreover, they show that perfusion is optimal for uncompressed vessels with low permeability. Therefore, vessel decompression would be most beneficial for tumors with abundant compressed but poorly or moderately permeable vessels (Fig. 2, dashed line), such as pancreatic ductal adenocarcinomas. However, decompression of tumor vessels is

reversible and vessels can revert to their compressed state unless stress-alleviation treatment is continued.

The ease with which the interstitial fluid percolates through the extravascular space of a tissue is referred to as its hydraulic conductivity. Hydraulic conductivity affects blood velocity because of the communication between vascular and interstitial fluid (35). High hydraulic conductivity allows fluid to rapidly flow in the interstitial space and be drained by peripheral lymphatic vessels. This prevents accumulation of interstitial fluid and thus more fluid can leak from the vessel wall, which in turn decreases perfusion. Results shown in Fig. 2 and Fig. S2 suggest that tumors with a relatively high hydraulic conductivity, usually tumors with low collagen and hyaluronan levels such as some melanomas, would be less responsive to stress-alleviation treatment.

Vascular Normalization Improves Perfusion of Tumors with Hyperpermeable and Uncompressed Vessels. In parallel, we tested under what conditions vascular normalization can improve perfusion (Fig. 3). The tumor vessel was assigned an initial diameter of 15 μm and a vascular pore size of 400 nm (circle in the top right corner of Fig. 3). An ideal treatment to optimize perfusion in such a vessel would aim at decreasing its permeability while keeping the vessel open. In our previous animal study (36), the vessel diameter changed from 14.5 μm to 7.3, 9.5, and 9.5 μm at days 1, 3, and 5 after anti-VEGF treatment, a predominant angiogenic growth factor in many tumors. The solid lines in the figure show the potential paths of the treatment as the diameter decreased from 14.5 μm to 7.3 μm (day 1) or 9.5 μm (days 3 and 5). Even though the decrease in pore size was not measured, there is a large range of pore sizes for which treatment falls into the optimal perfusion region. One should note that vascular normalization is reversible and if anti-VEGF treatment is stopped the vascular structure will return to an abnormal, less-functional state (37). Indeed, in our clinical trial in recurrent glioblastoma patients we saw a similar reversal in tumor vessel structure and function (38).

In another study (10), we found that the increase in tumor perfusion was dependent on the dose of anti-VEGF receptor 2 (VEGFR2) antibody used. The lowest dose of anti-VEGFR2 antibody caused an increase in the perfused vessel fraction from 0.2 to 0.45, whereas the highest dose did not increase perfusion. These values of perfused vessel fraction are within the range of our study—the highest dose of the antibody led to a significant decrease in vessel diameter and/or pruning of tumor vessels, yielding

Table 1. Studies showing vascular normalization improves perfusion measured as improved oxygenation

Therapeutic agent	Tumor model	Effect on oxygenation (refs.)
Antibody		
Bevacizumab	Melanoma, breast and ovarian carcinomas, GBM	↑ (7, 8)
DC101	GBM, mammary carcinoma	↑ (9, 10)
TKI		
Cediranib	GBM	↑ (11–13)*
Sunitinib	Squamous carcinoma	↑ (14)
Semaxanib	Melanoma	↑ (15)
Other therapies		
PI-103 (PI3K inhibitor)	Fibrosarcoma, squamous carcinoma	↑ (16)
FTIs (Ras inhibitors)	Prostate carcinoma, bladder carcinoma, glioma, fibrosarcoma, squamous carcinoma	↑ (16–18)
Nelfinavir (AKT inhibitor)	Fibrosarcoma, squamous carcinoma	↑ (16)
PHD2 down-regulation	Lung carcinomas	↑ (19)
R-Ras up-regulation	Melanomas and lung carcinomas	↑ (20)
Cancer cells nitric oxide synthesis inhibition	Glioblastomas	↑ (21)
VE-PTP inhibition	Breast carcinomas	↑ (22)

Information given in the table is updated from ref. 5. FTI, farnesyltransferase inhibitors; GBM, glioblastoma multiforme; PHD2, prolyl hydroxylase domain protein 2; TKI, tyrosine kinase inhibitor; VE-PTP, vascular endothelial protein tyrosine phosphatase.
*Clinical evidence.

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Table 2. Studies showing stress alleviation decompresses vessels and improves perfusion

Therapeutic agent	Target	Tumor model	Effect on vessel diameter/density (refs.)	Effect on perfusion (refs.)
Diptheria toxin	Cancer cells	Soft tissue sarcoma	↑ (24)	Not reported
Taxane	Cancer cells	Soft tissue sarcoma	↑ (23)	Not reported
Saridegib	Stromal cells	Pancreatic ductal carcinoma	↑ (25, 27)	↑ (25, 27)
PEGPH20	Hyaluronan	Pancreatic ductal carcinoma	↑ (28, 29)	↑ (28, 29)
1D11	Collagen	Mammary carcinoma	↑ (30)	↑ (30)
Losartan	Stromal cells, hyaluronan, collagen	Pancreatic ductal carcinoma	↑ (26)	↑ (26)

Diphtheria toxin is preferentially toxic to human cancer cells and thus preferentially kills cancer cells in a human tumor xenograft model. Taxane is a chemotherapeutic agent. Saridegib is an inhibitor of the Hedgehog cellular signaling pathway. PEGPH20 is a PEGylated human recombinant hyaluronidase. 1D11 is a TGF-β neutralizing antibody. Losartan is an angiotensin receptor blocker.

compromised perfusion (Figs. 3 and 4). Fig. 4 shows changes in perfusion as a function of time and dose. The region of improved perfusion is related to the window of normalization, and thus the higher the dose of treatment, the shorter the window of normalization, and excessive pruning would lead to hypoxia.

Other parameters that affect vascular normalization are the degree of vessel compression and the hydraulic conductivity. Our results suggest that effective anti-VEGF treatment requires tumor vessels to be open (Fig. 3). In cases where tumor vessels are compressed normalization of abnormal vessels and the resulting decrease in vascular permeability will not be sufficient to improve perfusion. Fig. S4 depicts the fraction of perfused vessels for three different values of the hydraulic conductivity. As for vessel decompression, a high conductivity compromises the therapeutic outcome. Taken all together, vascular normalization is expected to be more effective for tumors with hyperpermeable and uncompressed vessels with low or moderate values of hydraulic conductivity, such as a subset of glioblastomas, melanomas, and ovarian carcinomas.

Combined Treatment Improves Perfusion in Tumors with Compressed Yet Hyperpermeable Vessels. Our analysis thus far suggests that neither vessel decompression nor vascular normalization would be effective in treating tumors with compressed and hyperpermeable vessels, such as those seen in a subset of mammary carcinomas. In such a case, a combined treatment might be beneficial. A treatment to alleviate solid stress (depletion of cells or extracellular matrix) to decompress blood vessels would increase the effective vascular density without affecting significantly the fraction of perfused vessels (Fig. 2 and Fig. S2). Then, a subsequent or concurrent vascular normalization treatment would have the potential to improve perfusion (Fig. 3). Attention should be given, however, to the fact that stress-alleviation therapies, which aim to selectively deplete structural components of tumors, might increase the hydraulic conductivity of the interstitial space and thus reduce the therapeutic potential of vascular normalization. Indeed, the hydraulic conductivity depends on the concentration of collagen and hyaluronan, and empirical correlations to describe this relationship exist (39–41). In Fig. S5, we used these methodologies to calculate the hydraulic conductivity of tumors. Assuming that the optimal perfusion region is severely affected by conductivity values higher than 5×10^{-7} cm^2/mmHg-s (Fig. S4), data in Fig. S5 suggest that collagen or hyaluronan can be significantly decreased and still the tumors be responsive to anti-VEGF treatment. The schematic in Fig. 5 summarizes the guidelines for improving perfusion with these two strategies based on our baseline values. Fig. S6 provides examples from preclinical studies.

Improved Perfusion Enhances Delivery of Drugs. To elucidate the relationship between perfusion and drug delivery, we calculated the transport of drugs with sizes ranging from 1 nm (e.g., chemotherapeutic agents) to 10 nm (e.g., antibodies, nab-paclitaxel following disintegration in plasma) to 60 nm (e.g., liposomes and

micelles). The model accounts for the transport of the drug from the vessels to the tumor interstitial space and for drug binding to and internalized by cancer cells (details are given in *Supporting Information*) (42–44). Fig. 6 shows the intratumoral distribution of the drug with low or high binding affinity as a function of the perfused vessel fraction. Intratumoral distribution is defined as the fraction of the tumor in which the drug is bound to cancer cells in amounts higher than 1% of the concentration at the inlet of the vascular network. Delivery is improved with improved perfusion for drugs of all sizes, and the effect is more prominent for small drugs with low binding affinity. Small drugs diffuse quickly in the tumor extracellular matrix and thus their transport is perfusion-limited. By contrast, large-size particles cannot effectively penetrate into the tumor tissue and a rate-limiting step is the interstitial transport. Binding to cells or matrix is an additional barrier to interstitial transport. The higher the binding affinity, the more difficult it is for the drug to spread into the tumor. Interestingly, for drugs binding with a high affinity, the distribution of the 10-nm and 60-nm drugs is the same. This suggests that drug binding to cells dominates interstitial diffusion, whereas for low binding affinity interstitial diffusion is important and for that reason 10-nm particles distribute better than 60-nm particles. Our results are in agreement with previous studies showing that delivery of 10-nm particles is superior compared with that of 60-nm and 120-nm particles in tumors following vascular normalization (32). In general, the smaller the drug, the better it will distribute into the tumor, unless binding dominates. This may explain why nab-paclitaxel (10 nm in size)
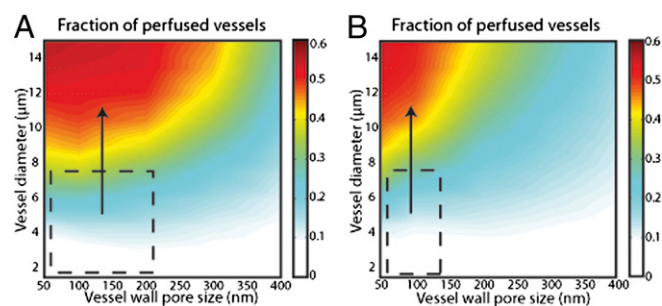
Fig. 2. Results for the fraction of perfused vessels as a function of the vessel diameter and the vessel wall pore size, a determinant of vascular permeability. Two values of the hydraulic conductivity of the interstitial space were used: (A) 1×10^{-7} cm^2/mmHg-s and (B) 1×10^{-6} cm^2/mmHg-s. The compressed central region occupies 80% of the tumor. The colors represent the values of the fraction of perfused vessels and the dashed line depicts the region within which vessel decompression is beneficial. The fraction of the well-perfused vessels becomes optimal for uncompressed (larger diameter) and low-permeable (smaller vessel wall pore size) vessels. The fraction of perfused vessels decreases if the tumor vessels are compressed or hyperpermeable. Vessel decompression is beneficial for compressed and low-permeable vessels.
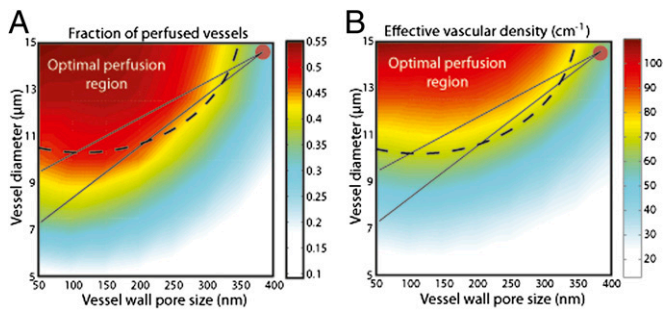
Fig. 3. Results for (*A*) fraction of perfused vessels and (*B*) effective vascular density as a function of vessel diameter and vessel wall pore size. The dashed line depicts the region within which vascular normalization is beneficial (referred to as optimal perfusion region), the circle shows the initial values, and the solid lines show potential paths of the treatment. The fraction of the well-perfused vessels and the effective vascular density become optimal for uncompressed (larger diameter) and low-permeable (smaller vessel wall pore size) vessels. The fraction of perfused vessels decreases if the tumor vessels are compressed or hyperpermeable. Vascular normalization is beneficial for compressed and hyperpermeable vessels.

improves overall survival of metastatic breast and pancreatic adenocarcinoma patients, whereas pegylated liposomal doxorubicin (100 nm in size) does not robustly improve survival over conventional chemotherapy (45–47).

Discussion

In this article, we used a mathematical framework for blood perfusion in solid tumors focusing on the effects of vessel leakiness and compression. In agreement with our model predictions,
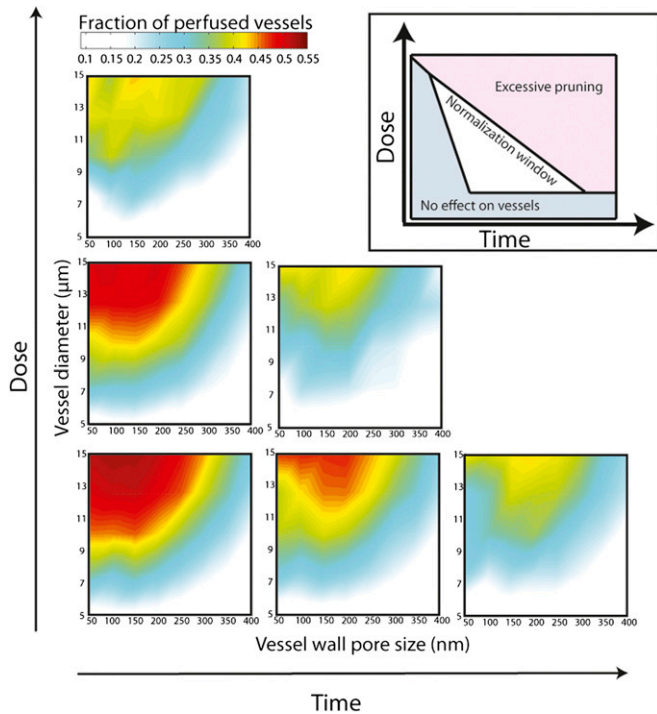

Fig. 4. Normalization window as a function of dose and time. Higher doses of antiangiogenic treatment over time increase vessel pruning, which in turn decreases the effective vascular density and impairs vascular efficiency. As a result the fraction of perfused vessels and the region of optimal perfusion decrease. These dose- and time-dependent effects create a normalization window within which drug delivery is optimized. (*Inset*) Schematic presents the effects of dose and time on the normalization window (adapted from ref. 2).
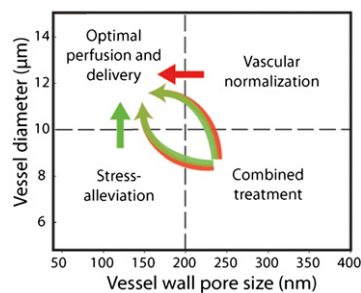

Fig. 5. Schematic of the proposed therapeutic strategies to improve perfusion and drug delivery. Tumors with hyperpermeable and uncompressed vessels benefit from vascular normalization strategy (red arrow). Tumors with compressed and low or moderately permeable vessels benefit from vessel decompression/stress-alleviation strategy (green arrow). Tumors with compressed and hyperpermeable vessels benefit from the application of stress-alleviation treatment to decompress vessels along with vascular normalization treatment to decrease permeability (two-color arrows).

vascular normalization has been shown to improve perfusion and therapeutic outcomes in both animal and human studies (9–13, 22, 32, 48) (Table 1). It is, however, effective mainly in the setting of leaky tumor vessels with open lumen (e.g., glioblastomas) and might not benefit compressed vessels. This would explain why antiangiogenic agents have failed thus far in desmoplastic tumors such as pancreatic ductal adenocarcinomas, with abundant compressed vessels (49, 50). Furthermore, anti-VEGF treatment has a dose-dependent effect, and a high dose or long-term treatment with anti-VEGF agents can cause excessive pruning of vessels and impairment of vascular efficiency (Fig. 4 and refs. 2 and 10). This could explain why in many studies anti-angiogenic strategies reduce drug delivery (51–55). Monitoring of blood perfusion or oxygenation in tumors during treatment could be used to identify the right dose. This could be done, for instance, with a novel functional MRI approach termed "vessel architecture imaging" (13). Additionally, anti-VEGF treatment has a size-dependent effect because the decrease in vessel wall pore size might exclude the delivery of nanoscale drugs larger than the pore size. Indeed, we have recently shown that normalization treatment improved the delivery of small particles with a size of 10 nm but not the delivery of 60-nm and 120-nm particles (32). In our analysis anti-VEGF agents were considered to cause a decrease in vessel diameter and vascular density, which compromises the benefit of reducing vessel leakiness on perfusion. One notable exception is when vessels are normalized with restoring nitric oxide gradients, which increases both vessel density and diameter (21, 22, 56). Finally, vessel leakiness is a hallmark of inflammation as well. In inflammation, however, plasma extravasation and leukocyte adhesion occur in a coordinated manner to enable the immune response and to maintain tissue perfusion.

Our model suggests that treatment to alleviate solid stress in tumors has the potential to enhance blood flow. Many studies, indeed, support the idea that decompression of blood vessels by alleviating solid stress improves perfusion and treatment efficacy (Table 2). We have recently shown that depletion of hyaluronan or collagen can lower solid stress levels in a variety of tumors, including pancreatic cancers (25, 26). This reduction in solid stress may explain how enzymatic degradation of hyaluronan combined with cytotoxic agents can improve perfusion and the overall survival of mice bearing pancreatic ductal adenocarcinomas (28, 29). Furthermore, depletion of collagen using losartan, an angiotensin receptor blocker, or an anti–TGF-β antibody improved the in vivo delivery of therapeutics in a variety of tumors in mice (26, 30, 57). In addition, pharmacological depletion of stromal cells with saridegib, an inhibitor of the Sonic Hedgehog pathway, has been shown to decrease solid stress (25) and improve drug delivery and overall survival of mice bearing pancreatic tumors (27). This drug, however, recently failed in a phase-II
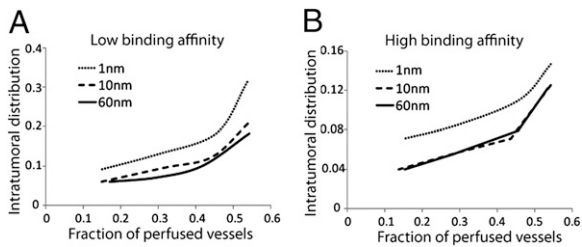
Fig. 6. Intratumoral distribution of therapeutic agents as a function of perfused vessel fraction for drugs with (*A*) low binding affinity and (*B*) high binding affinity (details in *Supporting Information*). Three curves in each panel show the intratumoral distribution of drugs of three sizes (1, 10, and 60 nm) versus the fraction of perfused vessels. The curves demonstrate that the intratumoral distribution of drugs of all three sizes increases with the fraction of perfused vessels, but in a nonlinear manner. The increase is more substantial for smaller drugs than for larger drugs. Intratumoral distribution is defined as the fraction of the tumor extravascular space that the drug is bound to cancer cells in amounts higher than 1% of the concentration at the inlet of the vascular network.

clinical trial for pancreatic cancer patients when combined with gemcitabine (58), presumably owing to intrinsic resistance to gemcitabine by these tumors. Increased delivery of a drug might not benefit patients if cancer cells are resistant to that drug.

Retrospective analysis of clinical data has shown that patients with pancreatic ductal adenocarcinomas receiving angiotensin receptor blockers survive approximately 6 mo longer than those who do not (59). Similar retrospective analyses have shown increased survival in patients with lung or renal cancers treated with such drugs (60, 61). Indeed, based on our preclinical findings (26), our Massachusetts General Hospital colleagues have initiated a phase-II trial with losartan and FOLFIRINOX in pancreatic ductal adenocarcinoma patients (clinicaltrials.gov identifier NCT01821729). Finally, depletion of cells with chemotherapeutics (or fractionated radiation) is another way to alleviate stresses and decompress vessels, which might explain the greater tumor response to chemotherapy in a subset of pancreatic cancer patients who are able to tolerate a combination of highly toxic drugs (62).

Our current study is of particular importance because it proposes new considerations for the use of treatments that modify the tumor microenvironment. Vessel compression is caused by physical forces exerted by the cells and extracellular matrix (25). Therefore, desmoplastic tumors should have a large amount of compressed vessels and thus anti-VEGF drugs (Table 1) most likely would not work, unless these drugs also alleviate solid stress by depleting stromal elements. If not, stress-alleviation drugs (Table 2) to decompress vessels should be considered either alone or in combination with anti-VEGF agents (Table 1). Less-desmoplastic tumors are expected to have a low fraction of compressed vessels, and thus stress-alleviation treatment would not be required. In this case, the use of anti-VEGF treatment with agents that target cancer cells should be considered (Table 3). To turn our theoretical predictions into practice, one needs to identify which tumors are leaky, compressed, both, or neither. This is a challenging task. Although we can make some broad statements (e.g., pancreatic ductal adenocarcinomas are desmoplastic), there are many tumors such as breast cancers in which the degree of desmoplasia is highly variable from one tumor to the next and potentially from the primary site to the metastatic site, and thus it could be hard to choose an appropriate strategy until the state of that individual tumor is known. Emerging imaging approaches have the potential to help in this selection (13).

It is conceivable that opening blood vessels could bring more nutrients to the tumor and increase its growth rate. Also, the opened vessels could allow more metastatic cells to leave the

Table 3. Summary of barriers to and strategies for improved drug delivery

Tumor microenvironment	Strategy
Desmoplastic, moderately permeable tumors (e.g., pancreatic ductal adenocarcinomas)	Stress alleviation
Hyperpermeable, nondesmoplastic tumors (e.g., subset of glioblastomas and ovarian carcinomas)	Vascular normalization
Hyperpermeable, desmoplastic tumors (e.g., a subset of mammary carcinomas)	Combined treatment
Drug	Delivery barrier
Small size ($\leq$ 10 nm), low binding affinity	Perfusion-limited transport. Improved perfusion increases delivery.
Large size (> 10 nm), low binding affinity	Perfusion, transvascular, and interstitial limited transport. Improved perfusion increases delivery.
Any size, high binding affinity	Additional barrier to interstitial transport. Improved perfusion increases delivery but benefit may be compromised.

primary tumor and increase metastases. Indeed, this has been shown in a number of preclinical studies (63, 64). Therefore, drugs that open vessels should be given only with concurrent cytotoxic treatments, such as chemotherapy, radiation therapy, immune therapy, or another cancer-cell-targeted treatment. Finally, apart from vessel compression and hyperpermeability, other causes of decreased blood flow in tumors include heterogeneous distribution of blood vessels, intravascular coagulation/thrombosis, and formation of vascular shunts (65, 66). Our model accounts for vascular heterogeneity with the use of an irregular network structure and by permitting vessel pruning. Mathematically, thrombosis is similar to vascular compression because both phenomena reduce the effective cross-sectional area of tumor blood vessels. Vascular shunts are short, high-flow vascular pathways that bypass long downstream pathways and thus exclude downstream regions from blood supply (66). Our model does not recapitulate the high heterogeneity of path lengths of the vessels as well as the enlargement of short flow pathways.

Methods

The current model builds on our recently published framework (32, 67). The baseline parameters of the model are summarized in Table S1. Description of model equations, methodology, and limitations (Figs. S7 and S8) can be found in *Supporting Information*.

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