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Hit 'Em Where They Live: Targeting the Cancer Stem Cell Niche

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Cancer stem cells (CSCs) are thought to be critical for initiation and propagation of many types of cancer. Because these cells are resistant to conventional therapies, they have been very difficult to eliminate. A study in this issue of *Cancer Cell* suggests that brain tumor CSCs live in a "vascular niche" that promotes their long-term growth and self-renewal. Disrupting this niche impairs CSC self-renewal and thereby significantly inhibits the growth of tumors. Targeting the unique microenvironment of CSCs may be the key to effective cancer therapy.

Once upon a time, cancer was viewed as a homogeneous mass of rapidly proliferating cells, and therapeutics were designed to eliminate highly proliferative cells. But recent studies have suggested that tumor cells are heterogeneous with respect to proliferation and differentiation, and that a cell's proliferative rate may be a poor indicator of its tumorigenic potential. In several malignancies, the capacity to initiate and maintain tumor growth has been found to reside in a small population of cells called cancer stem cells (CSCs) (Al-Hajj et al., 2004; Reya et al., 2001; Wicha et al., 2006). Like normal stem cells, CSCs have the ability to self-renew and to give rise to the variety of proliferating and differentiated cells that make up the bulk of a tumor. Importantly, CSCs are often relatively quiescent and therefore may not be affected by therapies targeting rapidly dividing cells. Elevated expression of transporters that pump out chemotherapeutic agents (Donnenberg and Donnenberg, 2005) and an increased capacity to repair DNA damage (Bao et al., 2006a) may also contribute to CSCs' ability to survive conventional modes of therapy.

The resistance of CSCs to conventional therapies may help explain why such therapies often fail: although they may destroy the bulk of a tumor, they cannot prevent the surviving CSCs from kicking into action and regenerating it again (Al-Hajj et al., 2004; Reya et al., 2001; Wicha et al., 2006). In this view, effective cancer treatment will require targeting CSCs themselves. But what are the signals that regulate CSC survival and function, and is there an effective way to subvert them?

One way to identify regulators of CSCs is to look for analogies with normal stem cells. An important characteristic of normal neural stem cells (NSCs) is that they are concentrated in regions that are rich in blood vessels, called "vascular niches" (Ramirez-Castillejo et al., 2006; Shen et al., 2004). These niches are thought to shelter NSCs from apoptotic stimuli and allow them to maintain a proper balance between self-renewal and differentiation. Endothelial cells (ECs), which line blood vessels, secrete factors that promote stem cell survival and self-renewal and are thought to be a key component of the NSC niche.

A study in this issue of *Cancer Cell* suggests that CSCs in brain tumors, similar to NSCs, reside in vascular niches, and that disrupting these niches may be the key to eliminating CSCs (Calabrese et al., 2007). By analyzing a large cohort of human brain tumors, Calabrese et al. dem-

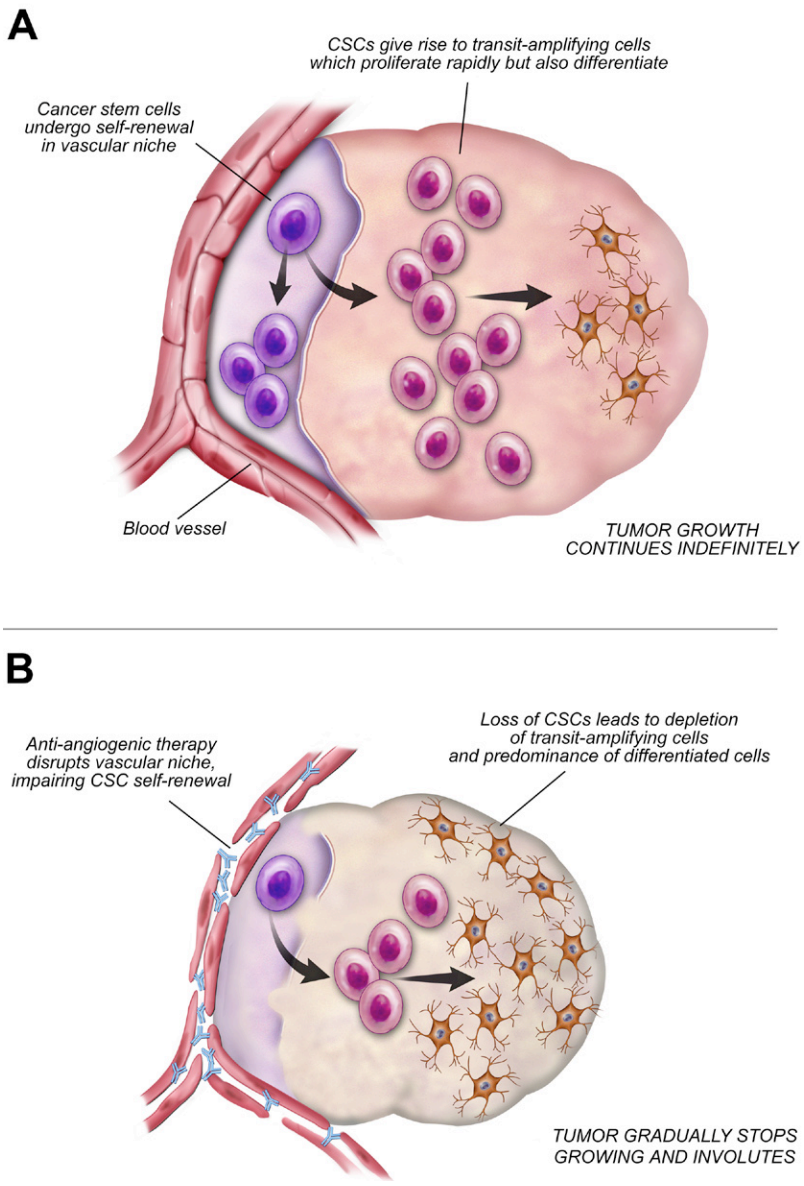

Figure 1. Model for the Role of the Vascular Niche in Cancer

(A) Cancer stem cells (CSCs) reside in close proximity to blood vessels, where they receive signals that allow them to self-renew and to generate transit-amplifying cells. Transit-amplifying cells proliferate rapidly and make up the bulk of the tumor but cannot self-renew and only give rise to differentiated (postmitotic) cells. The continued generation of transit-amplifying cells from CSCs allows the tumor to keep growing.

(B) Antiangiogenic therapies disrupt blood vessels, leading to disintegration of the vascular niche. Without this niche, CSCs cannot self-renew and instead only differentiate into transit-amplifying cells. As these cells exhaust themselves, the tumor gradually stops growing and involutes.

Illustration by Stan Coffman/Medmedia Solutions.

onstrated that cells expressing markers of CSCs represent a very small fraction of tumor tissue and are frequently located close to capillaries within the tumor. Moreover, when cultured with primary human endothelial cells, CSCs rapidly and selectively associate with these cells, while the majority of tumor cells do not. Importantly, ECs also enhance the self-renewal capacity of CSCs in vitro. To examine the consequences of this interaction for tumor growth in vivo, Calabrese et al. transplanted human medulloblastoma cells, with or without ECs, into immunocompromised mice. In both cases the transplanted cells formed tumors. However, medulloblastoma cells transplanted with ECs grew more rapidly and formed much larger tumors than those transplanted alone. Moreover, tumors that were established in the presence of ECs contained up to 25 times more CSCs. Thus, ECs can enhance the self-renewal of CSCs in vitro and promote the growth of brain tumors in vivo.

Calabrese et al. then investigated whether elimination of ECs could prevent tumor growth. Previous studies had demonstrated that medulloblastomas often overexpress ERBB2, which leads to increased production of vascular endothelial growth factor (VEGF), a critical regulator of EC survival and proliferation. Consistent with a role for ECs in maintaining CSCs, medulloblastoma cells overexpressing ERBB2 formed tumors more rapidly than control cells, and these tumors contained a higher proportion of CSCs. Importantly, treatment of tumor-bearing mice with inhibitors of either ERBB2 or VEGF signaling depleted blood vessels and caused a dramatic reduction in the number of CSCs and in the growth rate of the tumor. Interestingly, these drugs had little effect on the proliferation or survival of most of the cells in the tumor, suggesting that they were specifically acting on the CSCs. Similar results were seen with glioblastoma cells, raising the possibility that inhibition of blood vessel growth may be an effective method for eliminating CSCs in many types of brain tumors.

The work of Calabrese et al. highlights the importance of the vascular microenvironment in brain tumor growth (see Figure 1). Their observation that cotransplantation of tumor cells with ECs leads to more rapid tumor formation raises the possibility that the vascular niche might contribute to tumor initiation. For example, stem cells or progenitors that develop mutations within the vascular niche might be more likely to give rise to tumors than those outside the niche. Similarly, the

finding that disruption of angiogenesis leads to a reduction in growth of fully formed tumors suggests that the vascular niche may also be critical for tumor maintenance. Once a tumor is established, CSCs that find a vascular niche may continue to self-renew, while those that cannot may differentiate into transit-amplifying cells that contribute to the bulk of the tumor but not to its long-term maintenance. Identification of the signals that allow ECs to regulate CSC self-renewal may shed light on precisely when and how the niche regulates tumorigenesis.

Interestingly, recent evidence suggests that the relationship between CSCs and the vascular niche may be bidirectional: just as the niche can support the growth and renewal of CSCs, CSCs may contribute to maintenance of the niche. Bao et al. (2006b) show that CSCs from gliomas secrete markedly elevated levels of VEGF, which significantly increases EC migration and tube formation. This reciprocal relationship raises the question of how the niche itself is generated: are CSCs attracted to pre-existing blood vessels, or do they create a vascular network to support themselves? Either way, the interdependence of CSCs and ECs makes the vascular niche an important target for therapy.

Of course, the use of antiangiogenic therapy to target cancer is not a new idea. But while numerous studies have suggested that preventing the growth of new blood vessels can inhibit tumor growth, the mechanism by which this therapy works remains a subject of debate (Jain, 2001; Lin and Sessa, 2004). The work of Calabrese et al. suggests that one mechanism by which antiangiogenic drugs might act is by disrupting a vascular niche that is necessary for CSC self-renewal. The notion that antiangiogenic therapy targets CSCs has important implications for evaluating and optimizing the use of antiangiogenic drugs in cancer. For example, it is notable that Calabrese et al. observe that antiangiogenic agents have a striking effect on CSC renewal but have little effect on proliferation or apoptosis of the majority of tumor cells. This suggests that in evaluating antiangiogenic therapies, it may not be sufficient to look for rapid tumor regression or shrinkage. Rather, more careful examination of tumor morphology and functional properties of tumor cells may be required to determine whether a particular agent is having an effect.

In general, therapies that target CSCs may have unique properties compared to therapies that target the bulk of a tumor. Assuming that CSCs represent only a small proportion of the entire tumor, killing them may, in the short term, have little impact on the size of the tumor as a whole. However, over time the tumor would be expected to exhaust itself and wither away, because it has lost the capacity for long-term self-renewal. From a clinical standpoint, it remains to be seen whether such therapies are effective on their own; it is possible that, for some cancers, continued proliferation of transit-amplifying cells that make up the bulk of the tumor may be sufficient to cause irreversible histological and physiological damage. In that sense, it may be critical to combine CSC targeting with conventional agents that debulk the tumor. Indeed, combinations of antiangiogenic drugs with conventional chemotherapies have proven to be more effective than either mode of therapy alone (Tozer et al., 2005). Finding the appropriate combinations of therapeutic agents to achieve both debulking and CSC elimination will be an important challenge in the clinic.

The theory that tumors depend on a small population of CSCs for their long-term growth and propagation has profound implications for our understanding and treatment of cancer. But until recently, this theory has largely served to explain why conventional therapies may fail. The work of Calabrese et al. takes a critical next step, identifying unique properties and vulnerabilities of CSCs that may allow us to target them. Only by delving into the biology of CSCs—both their intrinsic properties and their microenvironment—will we find ways to prevent their longevity from interfering with our own.

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