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Tumor Tropism: A Silver Bullet?

What if there was a magic pill that could cure all cancers? Certainly, this might not be realistic, but the idea is one that merits more consideration and is in a way, the goal of all cancer researchers. If there were a general solution to cancers and tumors that could be administered systemically, such a solution would be ideal. Several exciting therapies are the topic of current research that could, with further discoveries, could be just such a magic pill, or at the very least, contribute to the discovery of such a cure. Of particular interest are vectors that display tumor tropism: this behavior allows for the administration of a variety of therapies in a localized fashion, ranging from the delivery of viral particles to gene therapy (Carroll). Specifically, three cases seem promising: mesenchymal stem cells for cancers and tumors of the lung, liver and bone, neural stem cells for the central nervous system, and alphavirus vectors as general oncolytic agents, such as the Sindbis virus. Each of these methods has generated a significant amount of interest as a result of their apparent ability to home to the tumor site, but they are certainly not flawless as yet, and will be the object of much future research (Kerin).

Mesenchymal Stem Cells

Bone marrow contains mesenchymal stem cells (MSCs), which support hematopoietic stem cells and provide progenitors for several mesenchymal tissues. Interestingly, comparable

adult stem cells have also been isolated from a wide variety of tissues (Kerin). These cells can differentiate into specialized cells with a phenotype distinct from that of the precursor under the influence of appropriate signals. They also have several other attractive therapeutic features: not only can they be isolated and expanded with high efficiency, but also they are not completely developmentally committed. In fact, MSCs can give rise to nonmesenchymal cells such as neural cells or epithelial cells as well as a variety of mesenchymal phenotypes such as bone, cartilage, fat, and muscle (Suk-Kee).

Although MSCs have potential uses in regenerative medicine and a number of different disease models, they also have enormous potential for targeted delivery of suicide genes, oncolytic viruses and secreted therapeutic proteins due to their tumor tropic tendencies (Kerin). The mechanism for the tumor tropism is not yet fully characterized, but it is likely related to the inflammation of tumor sites, along with the presence soluble factors such as epidermal growth factor, vascular endothelial growth factor-A, fibroblast growth factor, platelet-derived growth factor, stromal-derived growth factor-1α, and many more (Kerin). These homing MSCs can be modified to express a variety of therapeutic proteins, include IFNβ, IL-2, IL-12, pigment epithelium-derived factor, NK, TNF-related apoptosis inducing ligand (TRAIL), and cytosine deaminase. Each of these proteins is capable of antitumoral activity: MSCs expressing IFNβ have been shown to result in increased animal survival, and IL-12 expressing cells embedded in a matrix adjacent to tumors were also reported to have a significant therapeutic effect (S H Seo). Importantly, regional secretion is required for both of these proteins to have effect: systemic administration alone was not enough for any therapeutic result (S H Seo). In another case, MSCs expressing TRAIL induce caspase-mediated apoptosis in tumor cells that over express the receptor. The power of this method is

its stability: like most healthy tissues, MSCs are resistant to TRAIL-induced apoptosis due to their very low levels of active receptors, which allows for safe antitumoral activity. Additionally, MSCs secreting IL-2 or IL-12 were shown to elicit an immunological reaction, and to stimulate inflammatory cell infiltration of the tumor tissue. The result is an immune attack on the nearby tissues: however, the MSCs are importantly immunoprivileged, and are protected by a variety of different factors (Suk Kee).

MSCs may also be an ideal vehicle for virus delivery, especially since they are easily genetically modified. According to a 2010 paper:

> Modification of the adenovirus fiber or knob domain has been used to improve adenovirus-mediated transgene expression. Incorporation of an arginine-glysine-aspartate motif into the adenovirus fiber or the 5/3 knob domain of human adenovirus serotype 3 supports coxsackie and adenovirus receptor-independent transfer and improves MSC transduction efficiency (Dembinski)

After these modifications, MSCs can be used to increase the viral load at the tumor. Furthermore, the kinetics of adenoviruses in MSCs is relatively slow, solving the problem of toxicity prior to engraftment. This would ultimately allow for the delivery of oncolytic viruses with little collateral damage to surrounding tissues.

Since variations in MSC engraftment have been observed in different tumor models, attempts are being made to improve tumor tropism and infiltration through modification of the MSC surface. In one study where native MSC tropism for the tumor of interest was not detected, MSCs were engineered to overexpress the epidermal growth factor receptor. The result was that transduced MSCs had enhanced migratory properties towards GL261 gliomas

or B16 melanoma in vivo. Also, immobilized sialyl Lewis X on MSCs was shown to induce cell rolling, which could also have potential in improving tumor tropism (Sato H).

The use of MSCs in cancer therapy is not without caveats, however. The potential role of MSCs in tumor initiation or promotion is a significant concern that must be addressed fully to allow MSC-mediated therapy for cancer to realize its full potential. MSCs may serve as precursors for carcinoma-associated fibroblasts and pericytes, as well as having the potential to spontaneously transform following long-term passage (Kerin). Also, the immunoprivileged MSCs may support the growth of tumor cells by protecting the cancer cells from immune detection.

Neural Stem Cells

Neural stem cells (NSCs) are cells that give rise to neurons and glial cells. Like other adult stem cells, they have potential for regenerative medicine, but it is their tumor tropic behavior that is promising, especially when taken in consideration with the advancements in the use of monoclonal antibodies as tools for cancer therapy. One of the problems with antibody therapy is that, despite its promise, the large molecular size of antibodies limits their ability to efficiently penetrate solid tumors and precludes efficient crossing of the blood-brain-barrier into the central nervous system (CNS) (Aboody). As a result, antibodies cannot treat any cancers that CNS metastases or poorly vascularized tumors effectively. However, intravenously delivered NSCs preferentially migrate to primary and metastatic tumor sites within and outside the CNS, which may mean that they could be vehicles for delivering antibodies to malignant tumors (Carroll). Once again, the exact molecular basis of the tumor-tropism of the neural stem cells is not well understood, but factors such as stromal cell-

derived factor-1, vascular endothelial growth factor, and macrophage chemotactic protein-1 expressed by tumor cells likely play a chemotactic role (Cheung).

In a 2009 study, human NSCs were modified to target HER2-overexpressing breast cancer cells. HER2 overexpression in breast cancer is highly correlated with CNS metastases, which are inaccessible to current drug therapies. NSCs were engineered to secrete anti-HER2 immunoglobulin molecules that mimicked trastuzumab, the current monoclonal antibody therapy for HER2 breast cancer. It was found that these NSC-secreted antibodies assembled properly, possessed tumor cell-binding affinity and specificity, and effectively inhibited the proliferation of HER2-overexpressing breast cancer cells in vitro (Cheung). Furthermore, these cells were able to target tumor foci and deliver the anti-HER2 antibody in vivo to xenografted mice. This is what makes stem cell immunotherapy most attractive: antigens on tumor cells can likely be safely targeted, regardless of whether they are present in healthy tissues. These results suggest that NSCs could be a novel platform for delivering antibodies to metastasized cancers (Cheung).

Alphavirus Vectors

Alphaviruses contain a single stranded positive sense RNA as their genome, which allows them to be easily modified to express genes at very high levels in many different kinds of cells. “The high expression levels, induction of apoptosis, and activation of type I IFN response are the key features that have made alphavirus vectors very attractive for cancer treatment and vaccination” (Smerdou). Already, alphavirus vectors have been successfully used as vaccines to produce immune in animal models of mastocytoma, melanoma,

mammary, prostate, and virally induced tumors. However, their potential as oncolytic agents is particularly interesting.

Specifically, the Sindbis virus (SIN) has a natural tropism for tumor cells, and has been demonstrated to be able to reach metastatic tumors when administered systemically. While most alphaviruses are neurotropic, SIN preferentially infects mammalian cells at the high-affinity laminin receptor (LAMR), which is upregulated in many tumor cells. Many studies have shown that SIN vectors were able to inhibit tumor growth in tumor xenografts implanted subcutaneously, intrapancreatically, intraperitoneally, or in the lungs of mice by inducing apoptosis within the cells (Dembinski). However, this method still requires more research: in many experiments, tumors were not completely eradicated and the use of immunodeficient mice bearing tumors highly susceptible to SIN infection is somewhat suspect (Smerdou).

Efforts are going into harnessing these alphaviruses to target specific tumors, usually involving modification of the envelope protein. Some extremely elegant solutions have been found: by deleting residues 71–74 of E2 and introducing two IgG-binding domains of protein A, SIN vectors with minimal infectivity for many human cell lines have been generated. Interestingly, when these vectors were incubated with monoclonal antibodies specific for surface antigens, these “chimerical” viruses were able to infect cells expressing those antigens (Smerdou). It was then possible to retarget the alphaviruses to a variety of specific cell lines. All of these results suggest that immunotherapy with alphavirus vectors expressing cytokines represents a very attractive strategy.

Once again, however, the strategy is not without its pitfalls. For one, most experiments involved immunodeficient animals, and efficacy is much lower in immunocompetent animals.

This suggests that antiviral immune responses will hinder the use of these strategies in patients. Also, when dealing with viruses, pathogenicity is always a problem (Smerdou). As such, when expressing certain cytokines, like IL-4, from propagating vectors, caution is required. Expression of this factor by a recombinant mousepoxvirus increased its pathogenicity due to suppression of cytolitic lymphocyte responses against the virus, which is potentially harmful (Smerdou).

Conclusions

Unfortunately, in the realm of cancer research, there appears to be no silver bullet. However, the therapies considered here each represent advances with remarkable potential. Mesenchymal stem cells are extremely versatile, able to deliver therapeutic proteins and viruses while safely avoiding collateral damage. Neural stem cells are particularly mobile: not only can they cross the blood-brain barrier, but also they are able to efficiently penetrate poorly vascularized tumors. This allows for localized delivery of therapeutic antibodies, a growing field with similarly exciting potential. Finally, alphavirus vectors, specifically the Sindbis virus, have several interesting features that could lead to future therapies, especially their natural tropisms and ease of modification. Taking these therapies, one just can't help but be optimistic about the fact that one day, cancer will be cured.

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