

strong physiological effects, suggesting that the peptide-cargoes could cross the blood-brain barrier.<sup>12</sup>

The article by Popiel and colleagues presents the first case of oral ingestion of a transduction peptide with an activity in the nervous system. Although these results are of great interest, an important caveat is that it is quite difficult to make the jump from flies to mammals with such technology. Even if passage across the intestinal epithelium and into the brain could be reproduced in rodents, many points would have to be resolved before transduction peptides could be used as pharmacological tools. There have been no studies of the possible toxic or mutagenic activities of these peptides. In addition, one must also be able to target this new class of pharmacological agents to the right cells and, once in the cells, to the right subcellular compartment.

There is thus plenty of work ahead. However, considering the results gathered in only a few years by a small number of research groups, it is obvious that this new technology is very promising. A main advantage of transduction peptides is that they provide access to intracellular targets, as viral vectors do, but that they remain very classical pharmacological agents. If proteins such as interferon or insulin can be used therapeutically, there is no reason to doubt the interest of approaches using transduction peptides or nonpeptidic compounds modeled after these peptides.

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# Armed Interference: Oncolytic Viruses Engineered to Carry Antitumor shRNAs

David Kirn<sup>1</sup>

doi:10.1038/sj.mt.6300089

A diverse array of oncolytic viruses are being developed for the treatment of cancer.<sup>1,2</sup> These therapeutic agents are naturally and/or genetically targeted to replicate selectively in cancer cells. The resulting “oncolysis” is a novel mechanism of action (MOA) for cancer treatment and seems in many cases to be effective against apoptosis-resistant cells. In addition to this primary MOA, oncolytic viruses can demonstrate secondary MOAs such as induction of tumor-specific cytotoxic T lymphocytes,<sup>3</sup> anti-angiogenic cytokines,<sup>4</sup> and chemosensitization.<sup>5</sup>

The next generation of oncolytic viruses have additional MOAs through therapeutic transgene “arming.”<sup>6</sup> These therapeutic payloads are expressed selectively in cancer cells during replication, resulting in complementary MOAs. Examples include JX-594 (targeted vaccinia expressing granulocyte-macrophage colony-stimulating factor (hGM-CSF), Jennerex Biotherapeutics, San Francisco, CA),<sup>7</sup> OncoVex (herpes simplex virus (HSV) expressing hGM-CSF, Biovex, Woburn, MA),<sup>8</sup> and MV-NIS (measles virus expressing the sodium iodide symporter gene, Mayo Clinic, Rochester, MN).<sup>9</sup> In addition, anti-angiogenic and antivascular gene products (*e.g.*, soluble vascular endothelial growth factor receptor (VEGF-R))

have been expressed in the context of a targeted oncolytic virus.<sup>10</sup> Therefore, these armed oncolytic viruses are designed to wage a multipronged attack against cancer.

In this issue, Yun and colleagues<sup>11</sup> report proof-of-concept studies on the expression of a small inhibitory RNA (siRNA) from an oncolytic virus. They expressed a small hairpin (sh) RNA against VEGF from an *E1A-CR2* gene region-deleted adenovirus (Ad). The authors compared this virus to important controls, including the same oncolytic Ad lacking the shRNA expression cassette and a replication-incompetent Ad expressing the anti-VEGF shRNA. They demonstrated that shRNA expression and anti-VEGF effects were greater and more prolonged in the context of the oncolytic vector as compared with the replication-deficient vector. In addition, the shRNA-armed virus demonstrated superior efficacy over the same virus without shRNA arming. An anti-angiogenic MOA was shown both *in vitro* and *in vivo*. Interestingly, the Ad *E1A* protein also showed anti-VEGF and anti-angiogenic effects.

siRNA technologies hold promise for the treatment of cancer. However, thus far this approach has had only limited success *in vivo* because of several hurdles.<sup>12</sup> These include difficulties in achieving high-level expression selectively in cancers, particularly after intravenous administration. The application of shRNA technology in the context of systemically deliverable oncolytic viruses such as Ads or vaccinia viruses may be particularly effective at overcoming such

<sup>1</sup>Jennerex Biotherapeutics, San Francisco, California, USA

**Correspondence:** David Kirn, Jennerex Biotherapeutics, 1 Market Street, Spear Tower Ste. 2260, San Francisco, California 94105, USA. E-mail: dkirn@jennerex.com

hurdles. The use of a targeted, armed oncolytic virus to deliver siRNAs has the potential to achieve a higher level of expression in a more tumor-specific fashion than with nonreplicating vector systems. Thus, safety and efficacy should be improved. By combining the shRNA therapeutic platform with the oncolytic virus platform, the promise of both therapeutic platforms may be realized.

Several questions remain with this approach. The first is whether suppression of VEGF production from tumor cells will be sufficient to treat human tumors, or whether VEGF production from normal stromal cells and from other proangiogenic factors might prevent an effective anti-angiogenic effect in the tumor. It will also be important to compare different anti-VEGF technologies in the context of an oncolytic virus. Examples include anti-VEGF antibodies (e.g., bevacizumab, Avastin; Genentech, South San Francisco, CA), soluble VEGF-R decoys, and antisense approaches. Because

VEGF has immunosuppressive properties, blocking of VEGF activity might result in enhanced immune recognition of tumors and/or the virus itself. Finally, it will be of interest to determine whether the bleeding and gastrointestinal perforation toxicities associated with bevacizumab will be seen with this approach, particularly in the context of oncolysis and the associated inflammation. Answers to these questions will have important implications for this approach and for the field.

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