miRNA inhibition of metastasis

By Michael J. Haas, Senior Writer

A U.S. team that included researchers from **Regulus Therapeutics Inc.** has reported the first successful inhibition of a microRNA—miR-10b—in tumor cells, a feat that previously had only been accomplished in normal tissue.¹ The biotech's anti-miRNA oligonucleotide prevented breast cancer metastasis in mice, showing the antimetastatic potential of targeting miR-10b.

Studies carried out over the past five years have shown that miR-10b is upregulated in invasive and metastatic cancers such as pancreatic adenocarcinoma, glioblastoma and metastatic hepatocellular carcinoma (HCC). In 2007, researchers from the **Massachusetts Institute of Technology (MIT)** and the **Memorial Sloan-Kettering Cancer Center** found that miR-10b was also highly expressed in metastatic breast tumors, in which it inhibited expression of the tumor suppressor gene *homeobox D10 (HOXD10)*.² To investigate whether miR-10b could be an antimetastasis target, the researchers teamed up with Regulus to synthesize and test an miR-10b antagonist.

In the study, now published in *Nature Biotechnology*, the team showed that in mouse models of breast cancer, Regulus' anti-miR-10b antagomir produced an 86% reduction in the occurrence of lung metastases compared with an inactive control antagomir.

The antagomir did not affect the growth of either primary breast tumors or lung metastases that did occur.

The group was led by Robert Weinberg, professor of biology at MIT and a founding member of MIT's **Whitehead Institute for Biomedical Research**. In addition to Regulus and Whitehead, the team included scientists from the **Ludwig Center for Molecular Oncology at the Massachusetts Institute of Technology** and Memorial Sloan-Kettering.

"This report is the first demonstration of effective systemic delivery of miRNA antagonists to the cells within tumors," said Alexander Pertsemlidis, assistant professor of internal medicine at **The University of Texas Southwestern Medical Center at Dallas**. "The study establishes that miR-10b is required for metastasis formation by cancer cells that are highly malignant and that it is an important target for the development of antimetastasis therapies."

Ryszard Kole, SVP of discovery research at antisense company

AVI BioPharma Inc., agreed. "MicroRNAs have been targeted before with antisense oligonucleotides, but this is the first time an miRNA has been targeted in metastatic cancer. This is an important finding, and it is encouraging that the antagomir worked *in vitro* and *in vivo*."

Regulus has antagomirs and other miRNA-targeting compounds in preclinical development to treat cancer, metabolic disease, HCV infection and other indications.

AVI's lead compound, AVI-4658, a phosphorodiamidate morpholino oligomer (PMO) that targets exon 50 of the *dystrophin (DMD)* gene to restore dystrophin protein function, has completed Phase I/IIb testing to treat Duchenne muscular dystrophy (DMD). The company plans to release additional data from the trial later in 2Q10.

Mere prevention

The results suggest that different agents might be needed to prevent both metastasis and the growth of established metastases.

Andreas Bader, associate director of research at **Mirna Therapeutics Inc.**, noted that inhibition of miR-10b did not block the growth of existing metastases. "This suggests that inhibition of miR-10b may be used to prevent tumors from becoming metastatic rather than treating established metastatic disease," he said.

Pertsemlidis agreed, adding that the findings raised the question of "when cells capable of dissemination arise within primary tumors" and thus at what point during cancer progression an miR-10b antagonist would have to be given to prevent metastasis.

He also said the Weinberg team only studied tumors derived from one cell line. "Recapitulating the results observed here in additional cell lines derived from mouse mammary tumors, with both high and low tumorigenic and metastatic ability, would greatly strengthen the support for miR-10b as a key regulator of metastasis," he said.

Mirna, a 2009 spin-out from **Asuragen Inc.**, is developing miRNA mimics that replace miRNA tumor suppressor functions that have been lost in some cancers. The company's lead candidate, an miR-34 mimic, is in preclinical development to treat prostate cancer and other solid tumors. Mirna anticipates filling an IND in 2011.

Pertsemlidis also wanted to know whether miR-10b regulated the same genes in humans as it did in mice.

Bader agreed that additional studies would help validate miR-10b as a therapeutic target. "In particular, the evaluation of other breast cancer models is needed to show that these effects are not unique to a single or limited number of cell lines," he said, adding that evaluation is needed in other types of cancer as well.

Both Pertsemlidis and Kole said a 50 mg/kg dose was higher than usual for a therapeutic and wanted to know whether the antagomir had antimetastatic effects at lower doses.

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Kole also thought dosing could be lowered by optimizing the tissue-targeting capability of the antagomir. "If they could modify its chemistry to improve uptake into target tissues, that would be useful," he said.

According to Li Ma, first author on the paper and a senior research associate at the Whitehead Institute, the team plans to test the efficacy and safety of the anti-miR-10b antagomir in other animal models of cancer and as an adjuvant to surgical resection of primary breast tumors in mice.

Although the function of miR-10b in normal cells is not well understood, Regulus CSO Peter Linsley said its strong upregulation in metastatic breast cancer cells, as compared with its expression in normal tissue, suggested that anti-miR-10b therapeutics were unlikely to have serious side effects.

Linsley said the antagomir used by Weinberg's team was developed as a research compound and noted that it would have to be optimized for efficacy and tested in standard toxicology studies before it could be considered a therapeutic candidate. Regulus has not yet decided whether to develop it further, he said.

Regulus holds an issued patent covering the use of the anti-miR-10b

antagomir and other anti-miR-10b agents to treat cancer.

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COMPANIES AND INSTITUTIONS MENTIONED

Asuragen Inc., Austin, Texas
AVI BioPharma Inc. (NASDAQ:AVII), Bothell, Wash.
Massachusetts Institute of Technology, Cambridge, Mass.
Memorial Sloan-Kettering Cancer Center, New York, N.Y.
Mirna Therapeutics Inc., Austin, Texas
Ludwig Center for Molecular Oncology at Massachusetts Institute of Technology, Cambridge, Mass.
Regulus Therapeutics Inc., Carlsbad, Calif.
The University of Texas Southwestern Medical Center at Dallas, Dallas, Texas
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