

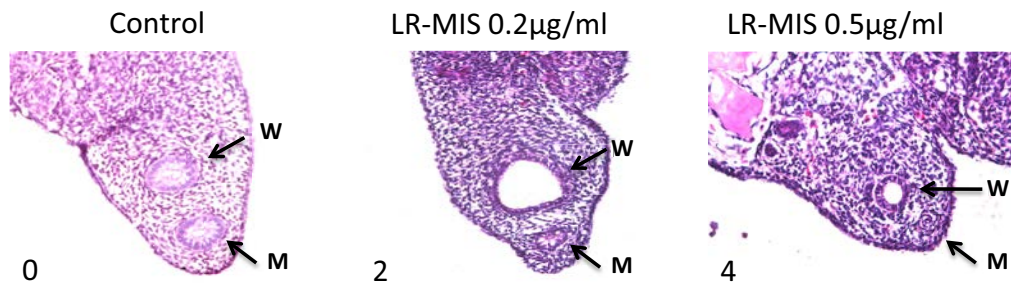
PRESS RELEASE

Modifications to a recombinant Mullerian inhibiting substance brings this new biological therapeutic one step closer to clinical testing in ovarian cancer patients

Sept. 17, 2013 — Mullerian inhibiting substance, or MIS, is a reproductive hormone produced in fetal testes, which inhibits the development of female secondary sexual structures in males. Before sexual differentiation, the fetus is bipotential, and the developmental choice of male Wolffian ducts (i.e. prostate, vas deferens) over female Mullerian ducts (i.e. Fallopian tubes, uterus, vagina) in the male is controlled in part by MIS. Because MIS is such a potent inhibitor of the development of Mullerian-derived tissue, it has been proposed as a potential therapeutic of Mullerian-derived tumors such as uterine, Fallopian, cervical, and ovarian cancers.

MIS belongs to the TGF-beta superfamily, a class of proteins involved in many pathologies including cancer. Recombinant TGF-beta proteins have been very difficult to produce because they require dimerization, activating cleavage and disulfide bonding for activity. Because of these peculiarities, MIS can only be feasibly produced in mammalian cells, and not *E. coli* or yeast, where production yields are much higher, and industrial scaling more straightforward. In mammalian cells, yields and homogeneity of the product can be significant barriers to industrial scaling and ultimate entry into clinical trials. For example, proteolytic degradation was a contributing factor to the failure of topical TGF-B3 in early clinical trials against chemotherapy-induced oral mucositis in patients with lymphomas and solid tumors. Recombinant BMP-2 in a paste form remains the only TGF-B family ligand used in the clinic, and is limited to the specific indication of autologous bone grafting. Progress in the technology of production and purification of TGF-Beta recombinant proteins could help many candidates, including MIS, to achieve their therapeutic potential in the clinic.

A team of researchers from the Massachusetts General Hospital (MGH) and the Massachusetts Eye and Ear Infirmary report that modifications to the protein sequence at the activating cleavage site of MIS to enhance maturation into the active form, and the addition of a leader sequence from albumin, the most highly secreted protein in the blood, results in higher production yield of cleaved active MIS which does not suffer from unwanted proteolytic degradation.



Fetal rat urogenital ridges treated with the new constructs of MIS show Mullerian duct (4+) regression as a measure of bioactivity at lower concentrations and thus increased potency (1µ/ml) (right) when prohormone cleavage is enhanced, compared to wild type (WT) MIS (left) which shows a retained Mullerian Duct (M) (0 regression). The adjacent Wolfian duct (W), shown for comparison, are unaffected (200X).

The report appears in the inaugural edition of the journal *Technology*, and is entitled "An albumin leader sequence coupled with a cleavage site modification enhances the yield of recombinant c-terminal Mullerian Inhibiting Substance."

"This technology is the culmination of our efforts to scale the production of MIS beyond the lab and into the clinic" says Dr. Donahoe, of MGH, the paper's senior author. "We show that not only can we produce high levels of this protein, but unexpectedly our modifications increase activating cleavage of MIS while improving the homogeneity of the product, all of which are necessary for translation to the clinic". The enhanced cleavage of MIS resulted in much greater activity when it was tested for its ability to induce Mullerian duct regression *ex vivo*. These modifications can be incorporated in other technologies such as viral vectors for gene therapy.

"The newly engineered recombinant MIS shows great promise and is a necessary component before clinical application of a biological such as MIS", said Dr. Austen, the Edward D. Churchill Distinguished Professor of Surgery at Harvard Medical School and Massachusetts General Hospital (MGH) Surgeon-in-Chief Emeritus, and Chairman of the MGH Chief's Council, who was not involved with this study.

Jacques-Pierre Moreau, PhD, CEO of Mulleris Therapeutics, Inc., who is dedicated to developing MIS as a therapeutic, states, "the therapeutic potential of MIS has, so far, been hindered by the scarcity of clinical grade product. The publication of Pepin, *et al.* represents a major advance in providing a reliable source of this regulatory hormone, thus making clinical development in oncology, endocrinology, and neurology possible".

Family advocates from the McBride Foundation and Commons Development have long provided support to the Pediatric Surgical Research Laboratories, Jack McBride, states, "we feel that our support of Dr. Donahoe's research over the years is now reaching a state where patients may profit".

The technology surrounding the modification of the MIS peptide is not only applicable to ovarian cancer, but has shown strong promise for other tumor types such as uterine, breast, and cervical cancers. "We choose to go after ovarian cancer, first, since it is the most lethal of these gynecological malignancies, with the fewest treatment options", says Dr. Donahoe. There is also early promise of using MIS gene therapy to treat neurodegenerative diseases using these modified constructs.

Additional co-authors of the *Technology* paper are David Pepin, Mien van Hoang, Fotini Nicolaou, Kathryn Hendren, Leo Andrew Benedict, Ahmed Al-Moujahed, Amanda Sosulski, Anna Marmalidou, and Demtrios Vavvas.

This work was funded in part by the Ovarian Cancer Research Fund Ann Schreiber Mentored Investigator Award (DP), a Sudna Gar Foundation fellowship (DP), the Harvard Ophthalmology Department Support (DGV, MH), a Research to Prevent Blindness unrestricted grant to MEEI, and an RPB Physician Scientist Award (DGV), Foundation Lions Eye Research Fund (DGV), The Yeatts Family Foundation (DGV); a Department of Surgery grant (PKD), a Department of Defense translational grant (#OC110078) to (PKD); and gifts from the McBride Family Foundation, the Austen Foundation, and the Common Development (PKD).

Corresponding author for this study in [TECHNOLOGY](https://doi.org/10.1002/tech.10001) is dpepin@partners.org

About TECHNOLOGY

Fashioned as a high-impact, high-visibility, top-echelon publication, this new ground-breaking journal — TECHNOLOGY — will feature the development of cutting-edge new technologies in a broad array of emerging fields of science and engineering. The content will have an applied science and technological slant with a focus on both innovation and application to daily lives. It will cover diverse disciplines such as health and life science, energy and environment, advanced materials, technology-based manufacturing, information science and technology, and marine and transportations technologies.

About World Scientific Publishing Co.

World Scientific Publishing is a leading independent publisher of books and journals for the scholarly, research and professional communities. The company publishes about 500 books annually and more than 120 journals in various fields. World Scientific collaborates with prestigious organisations like the Nobel Foundation, US National Academies Press, as well as its subsidiary, the Imperial College Press, amongst others, to bring high quality academic and professional content to researchers and academics worldwide. To find out more about World Scientific, please visit www.worldscientific.com.