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## Targeting viral capsid assembly

By Tracey Baas, Senior Editor

Researchers at **Prosetta Antiviral Inc.** have used an *in vitro* screen to identify small molecules targeting the host-catalyzed capsid assembly pathway for the rabies virus.<sup>1</sup> The biotech has partnered with **Bristol-Myers Squibb Co.** to use the screen to identify small molecules targeting the pathway for HIV.

The viral capsid is a multiprotein complex that gives a virus its shape and protects its genome. Although once thought to form spontaneously, the viral capsid has recently been shown by Jaisri Lingappa and others to form via a pathway catalyzed by a host assembly complex.<sup>2-4</sup> Thus, directly targeting the proteins comprising the host assembly complex could be a way to block formation of the viral capsid and prevent viral infection.

Lingappa is coauthor of the paper. She is an associate professor in global health and an adjunct associate professor in medicine and microbiology at the **University of Washington**.

In the new work, Prosetta used its previously developed cell-free protein synthesis (CFPS) screen to identify compounds that blocked virus capsid assembly (*see Figure 1, “Proposed rabies capsid-assembly pathway”*). The screen consisted of host cell extract, which included the host assembly proteins, and three capsid proteins from the rabies virus. Using the system, the team identified a small set of active antiviral compounds.

Prosetta’s collaborators at the **Centers for Disease Control and Prevention** next evaluated the compounds against a strain of ‘street rabies’ isolated from a rabid gray fox. In a rabies-infected mammalian cell culture model, an optimized lead molecule showed an  $EC_{50}$  value of 15–30 nM and a 50% cytotoxicity concentration ( $CC_{50}$ ) value of about 2.5–10  $\mu$ M.

The Prosetta team then used the small molecule in affinity chromatography to isolate the host capsid assembly machinery, a multiprotein complex. Detailed analysis of the complex showed that, among other proteins, it also contained ATP-binding cassette subfamily E member 1 (ABCE1), which had been previously associated with HIV capsid assembly.

Results were published in the *Proceedings of the National Academy of Sciences*.

“Our goal is to advance one compound that inhibits the capsid formation of all viruses responsible for human respiratory disease.”

— Vishwanath Lingappa,  
Prosetta Antiviral Inc.

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“Going after a multicomponent host assembly machine requires a different way to think about screening,” said Anna Mapp, professor of chemistry and director of the program in chemical biology at the **University of Michigan** and research professor at the university’s Life Sciences Institute.

“Traditional screening methodologies look for high-affinity compounds to bind to small surfaces that participate in protein-protein interactions or aim at enzymatic components of a complex. Prosetta’s technology provides a way to identify hits based on function—in this case, disrupting the virus life cycle by impeding capsid formation—irrespective of target,” explained Mapp.

**“Going after a multicomponent host assembly machine requires a different way to think about screening.”**

—Anna Mapp,  
University of Michigan

**Clarity for the capsid**

Prosetta’s next steps toward the ultimate development of these compounds as drugs should be “to test the small set of compounds in more cell types and animals and also against different viral strains for breadth of effect,” said Stephen Goff, professor of microbiology and immunology and of biochemistry and molecular biophysics at **Columbia University**. “I’d also like details about the mechanism of action of the candidate lead for the target. What components are required, what directly binds the drugs and is the drug really impeding capsid formation?”

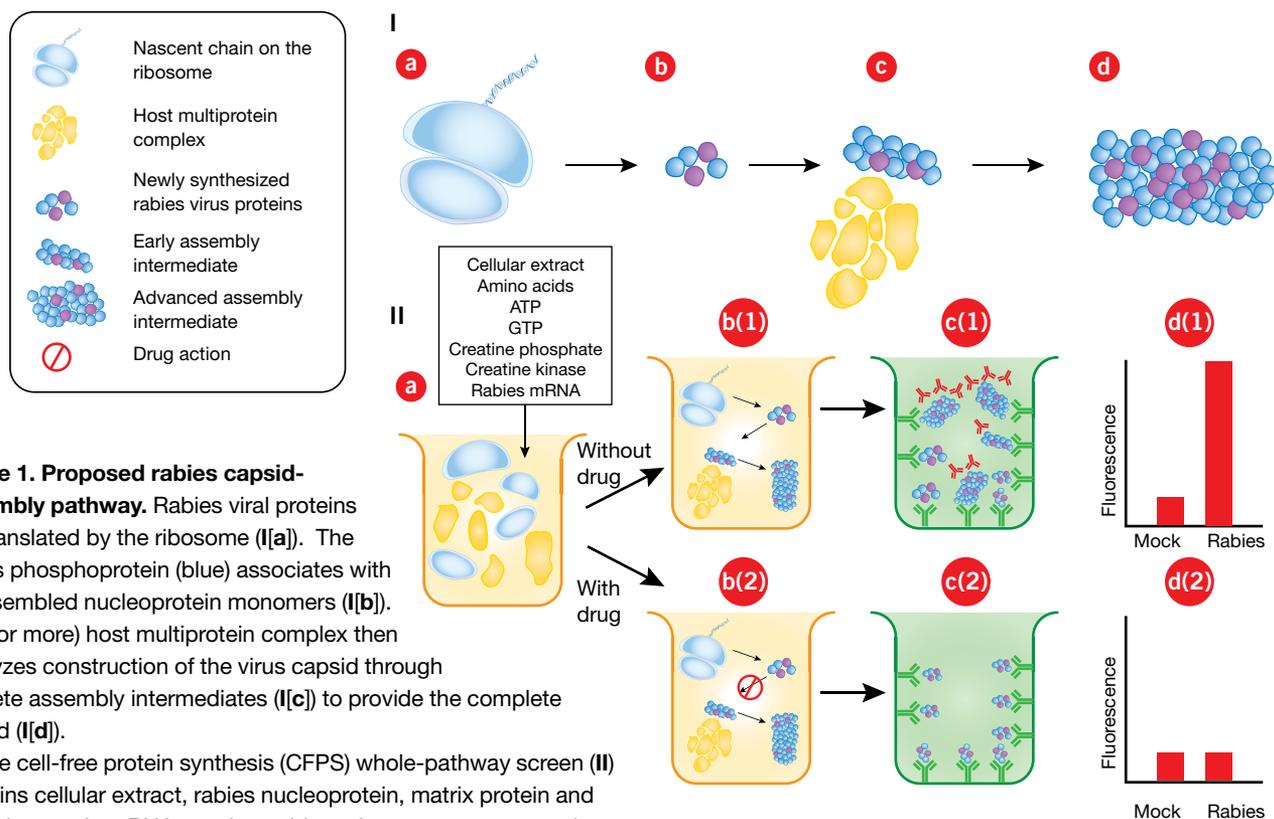
Vishwanath Lingappa, CTO and co-CEO of Prosetta, told *SciBX* that mechanistic studies to understand the compounds’ mode of action are under way at Prosetta. The company’s main focus, however, is to test the hits in animal models of rabies infection and continue using the screening platform to identify compounds that inhibit other viral infections, he said.

“We have already developed screens for 20 of the 23 virus families that cause human disease and have lead candidates targeting 14 different virus families,” he said.

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**Figure 1. Proposed rabies capsid-assembly pathway.** Rabies viral proteins are translated by the ribosome (I[a]). The rabies phosphoprotein (blue) associates with unassembled nucleoprotein monomers (I[b]). One (or more) host multiprotein complex then catalyzes construction of the virus capsid through discrete assembly intermediates (I[c]) to provide the complete capsid (I[d]).

The cell-free protein synthesis (CFPS) whole-pathway screen (II) contains cellular extract, rabies nucleoprotein, matrix protein and phosphoprotein mRNAs, amino acids and an energy-regenerating system (II[a]). The proposed synthesis and assembly of rabies capsid proteins occurs as described in (I) (II[b(1)]). The CFPS translation products are transferred to a capture plate coated with anti-rabies nucleoprotein antibody. The captured rabies assembly intermediates bind a biotinylated secondary antibody (II[c(1)]), which is used to produce a fluorescent readout that indicates capsid assembly (II[d(1)]).

The CFPS system components are combined, and the drug is added  $\leq$  six hours later (II[a]). A drug that blocks the formation of large capsid multimers (II[b(2)]) will lead to the capture plate being coated with monomeric units of rabies proteins (II[c(2)]) and inhibition of the fluorescent readout (II[d(2)]). (Figure based on Figures 2 and 3 from ref. 1.)

“One of our main projects is to find compounds that inhibit the capsid formation of multiple respiratory viruses. Right now, we have a subset of compounds that inhibits the six different virus families responsible for viral respiratory infection,” said Vishwanath Lingappa. “Our goal is to advance one compound that inhibits the capsid formation of all viruses responsible for human respiratory disease.”

In order to optimize the compound, he added, “we have functional assays in the form of cell-free assembly of the capsid via the host-catalyzed pathway for each viral family, so we can use the activity in that assay to track the SAR and simply corroborate improvements in activity with improvements in activity against the real virus.”

Prosetta has filed patent applications for the small molecules and the screening strategy.

Prosetta and Bristol-Myers Squibb (BMS) entered into a multiyear collaboration in July 2012 that includes a research program to discover and advance compounds shown to block HIV capsid assembly using the screening platform.

BMS has the right to develop and commercialize products arising from the research program. Prosetta will receive an upfront payment and multiyear research funding, and the biotech is eligible to receive milestone payments and royalties based on worldwide sales of drugs emerging from the collaboration.

BMS declined requests for interviews.

Prosetta is in discussions with two other undisclosed pharmas for partnering on different viral indications.

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## COMPANIES AND INSTITUTIONS MENTIONED

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**Prosetta Antiviral Inc.**, San Francisco, Calif.  
**University of Michigan**, Ann Arbor, Mich.  
**University of Washington**, Seattle, Wash.

# Making TRAIL

By Chris Cain, Senior Writer

Researchers at the **Penn State Milton S. Hershey Medical Center** have identified a small molecule inducer of TRAIL expression that has anti-tumor effects in multiple mouse models.<sup>1</sup> **Oncocotics Inc.** has licensed the findings and plans to advance the compound into the clinic this year.

Tumor necrosis factor-related apoptosis-inducing ligand (TRAIL) is an immune cytokine that induces apoptosis by binding tumor necrosis factor receptor superfamily member 10a (TNFRSF10A; DR4; TRAILR1; CD261) and DR5 (TNFRSF10B; TRAILR2; CD262), two receptors expressed on the surface of tumor cells.

Companies have developed recombinant TRAIL derivatives and antibodies targeting DR4 or DR5 to selectively induce apoptosis in tumor cells. However, none of those approaches has advanced into Phase III testing due to poor efficacy.<sup>2,3</sup>

The Penn State researchers took a fresh approach to modulating the TRAIL pathway by screening for small molecule inducers of TRAIL expression. Corresponding author Wafik El-Deiry told *SciBX* that his team hypothesized that small molecules could be more efficacious than biologics because their superior bioavailability and biodistribution properties would induce high levels of TRAIL in cells throughout the tumor and its surrounding microenvironment.

El-Deiry is professor of medicine and chief of hematology and oncology at the medical center.

The team used a cell-based reporter assay to screen a **National Cancer Institute** library of about 2,000 compounds for their ability to induce TRAIL expression. The screen identified a small molecule, dubbed TRAIL-inducing compound 10 (TIC10), which led to TRAIL mRNA and protein expression in a panel of cancer cell lines and triggered TRAIL-dependent cell death at low micromolar concentrations. In mice, the same molecule induced TRAIL expression in tissues including the brain, kidney and spleen.

The next step was to evaluate the efficacy of TIC10 in a series of mouse models for cancer.

In xenograft mice bearing breast or colon cancer cell lines, injection of TIC10 decreased tumor growth compared with vehicle control injection, and its effectiveness was equal or superior to that of recombinant TRAIL. Oral dosing of the compound at 25 mg/kg also decreased tumor growth compared with vehicle dosing.

In a mouse model for spontaneous lymphoma driven by c-Myc (MYC) overexpression, orally delivered TIC10 prolonged survival.

Finally, the researchers turned to an orthotopic mouse model for glioblastoma multiforme (GBM). Oral delivery of TIC10 decreased tumor growth and doubled overall survival compared with vehicle. When TIC10 was combined with the anti-VEGF antibody Avastin bevacizumab, overall survival tripled.

**Roche's Genentech Inc.** unit and **Chugai Pharmaceutical Co. Ltd.** market Avastin to treat several cancers including GBM.

Results were published in *Science Translational Medicine*.

## Pathway finding

Penn State has exclusively licensed the findings to Oncocotics, for which El-Deiry is cofounder and CSO. The company was founded in 2009 to develop small molecules with activity against p53-deficient cancers.

TIC10 was effective against both p53-wild-type and p53-mutant cells *in vitro* and in mouse models. El-Deiry said that the discovery of the compound was a natural extension of his lab's work on understanding tumor-suppressing pathways downstream of p53. "We got into this in the mid-to-late 1990s when we discovered that the TRAIL death receptors were regulated by p53, and we subsequently showed that TRAIL itself is regulated by p53," he said. "Once we knew that, it became pretty straightforward for us to conceptualize and go after small molecules that could induce TRAIL independent of p53."

Oncocotics now plans to advance TIC10, which has been renamed ONC201, into a Phase I/II trial in patients with solid tumors, which will include GBM. The trial is being funded by a Pennsylvania Department of Health grant

to Oncocotics. "If it looks safe in the early study, we would like to move into Phase II studies in combination with other targeted cancer therapies in responsive tumor types," El-Deiry said.

He said his lab will continue to work out the precise mechanism by which TIC10 induces TRAIL expression and kills cancer cells.

In the paper, the researchers reported that TIC10 indirectly inhibited protein kinase B (PKB; PKBA; AKT; AKT1) and MEK-MAP kinase (MAPK; ERK) signaling and required the downstream transcription factor forkhead box O3 (FOXO3; FOXO3a) to induce TRAIL expression, but its molecular target or targets were not identified.

"We are absolutely committed to identifying the precise molecular target," El-Deiry said. "We have unpublished evidence that there are some tumor cell lines that respond to TIC10 as a single agent but not to the combination of small molecules that target Akt and ERK, and we plan to study this further to understand why this occurs."

Carl Ware, professor and director of the Infectious and Inflammatory Disease Center at the **Sanford-Burnham Medical Research Institute**, wanted further confirmation that TIC10's anticancer effects are mediated by TRAIL.

"The authors provide one experiment utilizing a blocking antibody to TRAIL to demonstrate that the TIC10-induced apoptosis is TRAIL dependent, but the experiment was limited to the original tumor line used to screen for TIC," said Ware. "The use of this or a similar antibody would provide an approach to discriminate between TIC10-induced TRAIL expressed in transplanted human tumors and the bystander effect from TIC10-induced mouse TRAIL. Such experiments would provide more convincing evidence that TRAIL is involved in the action of TIC10 *in vivo*."

Ware's lab is currently focusing on the role TRAIL plays in the immune system to control viral infection.

**"We have unpublished evidence that there are some tumor cell lines that respond to TIC10 as a single agent but not to the combination of small molecules that target Akt and ERK, and we plan to study this further to understand why this occurs."**

—Wafik El-Deiry,  
Penn State Milton S. Hershey  
Medical Center

(Continues on p. 5)

# Islet transplants find the adrenal gland

By Lauren Martz, Staff Writer

German and U.S. researchers have shown that transplantation into the adrenal gland of islet cells preconditioned with an improved peptide agonist of growth hormone–releasing hormone may help normalize glucose levels in diabetic mice better than existing islet transplantation protocols.<sup>1</sup> The agonist has been licensed to **Biscayne Pharmaceuticals Inc.** for all indications, and the benefits of the intra-adrenal transplantation protocol will next be confirmed in large animal models.

Although transplantation of pancreatic islet cells has the potential to permanently correct dysfunctional insulin production and treat type 1 diabetes, current procedures require long-term immunosuppressive treatment, and progressive loss of islet functionality poses a big challenge.

Besides destructive autoimmunity, additional causes of post-transplant islet death include cell damage during isolation from donors and a lack of vasculature and oxygen supply at the hepatic transplantation site. Indeed, the standard intraportal infusion method of transplantation results in the liver quickly killing as much as 60% of the islet cell mass.<sup>2</sup>

(Continued from "Making TRAIL," p. 4)

Donald Buchsbaum, professor and director of the Division of Radiation Biology at **The University of Alabama at Birmingham**, wanted to see experiments in TRAIL-resistant cells. "It would have been informative if this paper had reported results for TRAIL-resistant cell lines and their sensitivity to TIC10," he said. "I believe the major limitation to successful clinical translation of TRAIL and agonistic death receptor antibody therapy has been the high incidence of resistance in the majority of patient tumors due to the overexpression of modulators of apoptosis."

The University of Alabama at Birmingham is leading a Phase II trial of **Daiichi Sankyo Co. Ltd.**'s DR5-agonist antibody tigatuzumab with or without Abraxane nab-paclitaxel in patients with triple-negative breast cancer. **Celgene Corp.**'s Abraxane is approved to treat non-small cell lung cancer (NSCLC) and metastatic breast cancer.

Aside from tigatuzumab, the fate of programs targeting TRAIL remains unclear.

**Human Genome Sciences Inc.** had been developing two antibodies before it was acquired by **GlaxoSmithKline plc** last year—the DR5 agonist lexatumumab and the DR4 agonist mapatumumab. However, GSK had previously obtained rights to the programs under a licensing agreement with HGS that GSK terminated in 2008, calling into question whether the pharma is now interested in pursuing their development. GSK declined to confirm the programs' development status.

Genentech discontinued development of its DR5 agonist antibody RG7425 and recombinant TRAIL in 2009 and currently lists no TRAIL- or death receptor-targeted drugs in its pipeline. **Amgen Inc.** declined to confirm the status of its recombinant TRAIL (dulcanermin) and does not list any TRAIL programs in its pipeline.

To improve the long-term functioning of islet grafts, diabetes researchers have begun focusing on new sites of transplantation and new pharmacological approaches to improving islet survival.

In 2010, a team led by Andrew Schally and including most of the same principal German coauthors as the current study found that treatment of insulinoma or islet cells with an agonist of growth hormone–releasing hormone (GHRH) increased cell viability and decreased apoptosis in culture compared with vehicle treatment. The group also found that transplantation of GHRH agonist-treated islets into diabetic mice improved glucose tolerance better than transplantation of untreated cells.<sup>3</sup>

The team has now built on the previous work and designed a more potent GHRH agonist, MR403, and shown that the adrenal gland is a potential alternative transplantation site. The researchers chose the adrenal gland because it has easy surgical access, dense vascularization and high oxygen tension. It also provides an anti-inflammatory and immunoprotective environment.

Schally is head of the Endocrine, Polypeptide and Cancer Institute at the **Veterans Affairs Medical Center** and a professor of medicine at the **University of Miami Miller School of Medicine**. The team included researchers from the **University Hospital of Carl Gustav Carus** and the **German Center for Diabetes Research**.

The team initially found that MR403 increased the survival and proliferation of cultured insulinoma cells and decreased apoptosis  
(Continues on p. 6)

**Pennsylvania State University** has filed a patent application covering the composition and use of TIC10 to treat cancer, and Oncoceutics exclusively licensed worldwide rights to the compound.

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## COMPANIES AND INSTITUTIONS MENTIONED

**Amgen Inc.** (NASDAQ:AMGN), Thousand Oaks, Calif.  
**Celgene Corp.** (NASDAQ:CELG), Summit, N.J.  
**Chugai Pharmaceutical Co. Ltd.** (Tokyo:4519), Tokyo, Japan  
**Daiichi Sankyo Co. Ltd.** (Tokyo:4568; Osaka:4568), Tokyo, Japan  
**Genentech Inc.**, South San Francisco, Calif.  
**GlaxoSmithKline plc** (LSE:GSK; NYSE:GSK), London, U.K.  
**Human Genome Sciences Inc.**, Rockville, Md.  
**National Cancer Institute**, Frederick, Md.  
**Oncoceutics Inc.**, Hummelstown, Pa.  
**Penn State Milton S. Hershey Medical Center**, Hershey, Pa.  
**Pennsylvania State University**, University Park, Pa.  
**Roche** (SIX:ROG; OTCQX:RHHBY), Basel, Switzerland  
**Sanford-Burnham Medical Research Institute**, La Jolla, Calif.  
**The University of Alabama at Birmingham**, Birmingham, Ala.

compared with vehicle. Coculture of agonist-treated rat islet cells and rat adrenal cells led to further survival improvements.

In nonobese diabetic mice, transplantation of islet cells pretreated with MR403 into the adrenal gland led to normal glucose levels a few days after transplantation that lasted until the grafts were removed at 30 days post-transplantation. The short-term transplantation efficiency was comparable to that of MR403-pretreated islet transplantation under the kidney capsule, the standard site of transplantation in rodent diabetes models. Adrenal gland transplantation also did not damage adrenal tissue, cause apoptosis of the islets or induce an inflammatory reaction.

Results were published in the *Proceedings of the National Academy of Sciences*.

“The shortage of donor islets and the need for lifelong immunosuppressive therapy remain major barriers to the adoption of islet cell transplantation,” said coauthor Stefan Bornstein, who is professor of medicine and director and chair of the Department of Medicine at the **Dresden University of Technology**. He added that the GHRH agonists and other modifications to the protocol may help overcome these obstacles and make islet cell transplantation a feasible therapeutic option.

#### Next steps

Diabetes researchers believe the new work is a step toward an improved protocol for islet cell transplantation but also say further *in vivo* testing is needed to confirm the benefits of the approach.

“As encouraging as these results are, GHRH-enhanced islet cell transplantation strategies require identifying suitable islet cell donors as well as subjecting diabetes patients to invasive procedures,” Hubert Chen, VP of clinical development at **Aileron Therapeutics Inc.**, told *SciBX*.

But, added Chen, it could also turn out that because GHRH may enhance the quality of islet cell harvests, the pool of eligible donors will be expanded.

With regard to the invasiveness of the procedure, coauthor Barbara Ludwig said that “transplantation into the adrenal gland could be envisioned either by laparoscopic procedure or via an intravascular approach. These options will be tested in the near future in a large animal model.” But she also acknowledged that “compared to the transhepatic approach that is currently performed in most centers for intraportal islet transplantation, this would generally be more invasive.”

Ludwig is a medical doctor at the University Hospital of Carl Gustav Carus.

Chen suggested that the invasiveness of the procedure could be reduced by directing islet cells toward the adrenal glands with X-ray-guided catheterization procedures, which are routinely used in hospitals.

**“The shortage of donor islets and the need for lifelong immunosuppressive therapy remain major barriers to the adoption of islet cell transplantation.”**

**—Dresden University of Technology**

Aileron’s long-acting GHRH analog, ALRN-5821, is in preclinical testing to treat growth hormone deficiency, abdominal obesity and cardiometabolic conditions including lipodystrophy.

Finally, Ludwig added that long-term *in vivo* experiments to prove that GHRH pretreatment increases the survival of the transplants are ongoing.

#### Biscayne

The University of Miami has licensed several of Schally’s GHRH-targeting compounds to Biscayne Pharmaceuticals. The company first plans to pursue indications in cardiovascular disease and cancer rather than diabetes because these indications have the most experimental characterization and documentation.

“We are in preclinical stages and getting ready for clinical testing. We have a peptide agonist that is being tested for ischemic heart disease and an antagonist for solid tumors,” CEO Samuel Reich told *SciBX*.

Biscayne has raised \$1.5 million in seed-round financing for preclinical testing and expects to raise more financing in the next 12–18 months to move into clinical trials.

“Right now we want to remain as focused as possible on the chosen indications,” said Reich.

“As those programs advance, Biscayne expects to develop other applications of the GHRH technology, including potentially its role in islet cell regeneration and diabetes,” said Biscayne scientific advisor Schally.

Schally told *SciBX* that the University of Miami has patents pending covering composition of matter of the peptides and methods of use in clinical applications including diabetes. The IP is licensed to Biscayne, and there may be opportunities to collaborate or partner.

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#### COMPANIES AND INSTITUTIONS MENTIONED

**Aileron Therapeutics Inc.**, Cambridge, Mass.  
**Biscayne Pharmaceuticals Inc.**, Miami, Fla.  
**Dresden University of Technology**, Dresden, Germany  
**German Center for Diabetes Research**, Dresden, Germany  
**University Hospital of Carl Gustav Carus**, Dresden, Germany  
**University of Miami Miller School of Medicine**, Miami, Fla.  
**Veterans Affairs Medical Center**, Miami, Fla.

# Cardiomyocytes acting like adults

By Tracey Baas, Senior Editor

A collaboration between **Sanford-Burnham Medical Research Institute** and **The Johns Hopkins University School of Medicine** cardiology teams has developed a model for arrhythmogenic right ventricular dysplasia/cardiomyopathy that for the first time is able to replicate the phenotype of the disease.<sup>1</sup> The key innovation was getting the induced pluripotent stem cell–based model to behave more like adult heart cells than fetal heart cells.

The team expects that the model could be used to identify disease-modifying therapeutics and thinks researchers working with induced pluripotent stem (iPS) cell–based models of other disease should consider maturing the final cell type when needed.

Arrhythmogenic right ventricular dysplasia/cardiomyopathy (ARVD/C) occurs in about 1 in 5,000 people, with onset typically occurring at 20–40 years of age. The disease is characterized by progressive right ventricular myocardium degeneration and apoptosis, ventricular arrhythmias, fibrous-fatty replacement of cardiac muscle within the right ventricular wall and increased risk of sudden death.<sup>2</sup>

Understanding the pathogenesis of ARVD/C has been challenging. For example, it is hard to obtain tissues from patients with ARVD/C at various stages of the disease because cardiac biopsies are dangerous and patients are commonly diagnosed at late stages of disease or even postmortem.

“In cardiac drug development, the vast majority of preclinical testing is conducted with animal or immortalized cell lines that do not resemble

the human heart. In most cases, the first time a pharmacological compound interacts directly with a human cardiomyocyte is in a Phase I trial,” said Joseph Wu, co-director of the Stanford Cardiovascular Institute and associate professor of medicine and radiology at the **Stanford University School of Medicine**.

To remedy this, the Sanford-Burnham group, with help from the Johns Hopkins Center

for Inherited Heart Diseases, opted to produce iPS cell–derived cardiomyocytes. The team generated iPS cells from the fibroblasts of patients with ARVD/C who had plakophilin 2 (*PKP2*) mutations and then differentiated the cells into cardiomyocytes using previously published protocols.<sup>3,4</sup>

Mutations in junction proteins, called desmosomes, are associated with ARVD/C. In addition to *PKP2*, ARVD/C-associated mutations also have been identified in subunits of desmosomes on cardiac cells that help glue the cells together, including junction plakoglobin (JUP), desmoplakin (DSP), desmoglein 2 (DSG2) and desmocollin 2 (DSC2).<sup>2</sup>

Curiously, the mutant *PKP2* cardiomyocytes did not reproduce

pathological ARVD/C. The group attributed the lack of a pathologic phenotype to an absence of exaggerated lipogenesis and apoptosis, which are found in adult-like but not fetal-like cardiomyocytes.

Indeed, the mutant *PKP2* cardiomyocytes had qualities that were similar to those of fetal-like cells, including using mostly glycolysis for energy production rather than fatty acid oxidation.<sup>5,6</sup>

To induce adult-like metabolism and accelerate pathogenesis in the mutant *PKP2* iPS cell–derived cardiomyocytes, the team created a lipogenic cocktail. The cocktail resulted in mutant *PKP2* iPS cell–derived cardiomyocytes with adult-like metabolism, exaggerated lipogenesis and pronounced apoptosis.

The cardiomyocytes with pathologies also showed impaired calcium relaxation and electrophysiological properties in normal media, which may contribute to the cellular pathology and arrhythmia in patients with ARVD/C.

The group’s final step was making the iPS cell–derived cardiomyocytes specifically resemble cells found in the right cardiac ventricle—the predominant pathological location of ARVD/C. To do so, the group added 6-bromoindirubin-3’-oxime to the cocktail. This reagent increases levels of islet 1–positive cardiac progenitor cells and subsequent cardiomyocytes, which form the right ventricle.<sup>7</sup>

Mutant cardiomyocytes derived from enriched islet 1–positive cardiac progenitor cells showed significantly greater lipogenesis and apoptosis than cells derived from unenriched progenitor cells.

Results were published in *Nature*.

“The approach that the team employs is especially interesting because the authors induce maturation in the iPS cell–derived cardiomyocytes in order to observe the disease phenotype,” said Wu. “This approach may be employed to model both cardiac and noncardiac diseases in the future given that many of the embryonic stem cell and iPS cell derivatives made today in laboratories resemble fetal cells more closely than adult counterparts.”

“That they found a metabolic switch to transition from fetal-like cardiac cells to adult-like cardiac cells is a major breakthrough and will provide a very valuable model,” said Roberto Iacone, laboratory head of the stem cell, cardiovascular and metabolism group at **Roche**. “It is encouraging to see this advance. We are using a similar approach to develop our own stem cell–based models and are genetically and pharmacologically altering the insulin pathway in order to provide a more translational cardiomyocyte platform to study the signaling pathways responsible for type 2 diabetes [-related] cardiovascular complications.”

## Taken to heart

Huei-Sheng Vincent Chen, associate professor of neuroscience, aging and stem cell research at Sanford-Burnham and principal investigator of the new study, thinks maturation will probably be needed for many iPS cell–based systems—especially those used to represent adult-onset diseases.

“Although a number of iPS cell–derived neurons in a dish show some disease phenotypes—such as those for Alzheimer’s, Parkinson’s and Huntington’s—we don’t know if this is as good as it gets because no detailed genetic, metabolic or cellular maps are established to guide the maturation induction from human fetal neurons to adult neurons, especially regarding their metabolic states,” Chen noted.

**“In cardiac drug development, the vast majority of preclinical testing is conducted with animal or immortalized cell lines that do not resemble the human heart.”**

—Joseph Wu,  
Stanford University School of  
Medicine

“A number of groups have provided iPS cell–based models for Alzheimer’s and long QT syndrome, but it would be interesting to see if incorporating a maturation step would provide a more pathogenic phenotype with more similarities to the human disease,” added Sheng Ding, a senior investigator at the **Gladstone Institute of Cardiovascular Disease**, a professor of pharmaceutical chemistry at the **University of California, San Francisco** and a cofounder of stem cell company **Fate Therapeutics Inc.**

Currently, Chen’s team has only checked one box in the list of ways to make iPS cell–derived cardiomyocytes mature.

“Right now, we have only dug into one aspect of maturation—metabolism. In fact, adequate human disease modeling with iPS cell derivatives might require many other aspects of maturation induction, which could include transverse tubule networks that provide structure, gap junction distribution, cell size and morphology,” said Chen. “I would be extremely happy if other teams could use what we learned from cardiomyocytes to develop other types of mature cells derived from iPS cells. We still have quite a distance to go.”

Chris Parker, VP and chief commercial officer at stem cell company **Cellular Dynamics International Inc.**, cautioned that it is possible to go overboard in the maturation of cardiomyocytes. “We’ve been using our own media cocktails since 2009 that switch newly differentiated cardiomyocytes into a functional cardiomyocyte phenotype,” he said. “Our functional cardiomyocyte phenotype resembles many aspects of cells found in an adult human heart, but they are adult-like and not adult. You may not want a fully adult cardiomyocyte because they do not spontaneously beat in culture, and this spontaneous beating is an important characteristic needed to screen for efficacy and toxicity of potential therapeutics.”

He also said that a maturation cocktail that is useful for cardiomyocytes “would not be useful for other cell types. For each of our cell types—

cardiomyocytes, endothelial cells, hepatocytes and neurons—we have specific media to provide cells to customers with appropriate functional phenotypes.”

Chen’s team has filed for patents covering its work. The licensing status of the IP is undisclosed. Chen said he would like to find a partner to distribute the group’s model iPS cells so that they can be used to develop ARVD/C-modifying therapeutics.

**Baas, T. *SciBX* 6(8); doi:10.1038/scibx.2013.183  
Published online Feb. 28, 2013**

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**Contact:** Huei-Sheng Vincent Chen, Sanford-Burnham Medical Research Institute, La Jolla, Calif.  
e-mail: [hsv\\_chen@burnham.org](mailto:hsv_chen@burnham.org)
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#### COMPANIES AND INSTITUTIONS MENTIONED

**Cellular Dynamics International Inc.**, Madison, Wis.  
**Fate Therapeutics Inc.**, San Diego, Calif.  
**Gladstone Institute of Cardiovascular Disease**, San Francisco, Calif.  
**The Johns Hopkins University School of Medicine**, Baltimore, Md.  
**Roche** (SIX:ROG; OTCQX:RHHBY), Basel, Switzerland  
**Sanford-Burnham Medical Research Institute**, La Jolla, Calif.  
**Stanford University School of Medicine**, Stanford, Calif.  
**University of California, San Francisco**, Calif.

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## This week in therapeutics

**THE DISTILLERY** brings you this week's most essential scientific findings in therapeutics, distilled by *SciBX* editors from a weekly review of more than 400 papers in 41 of the highest-impact journals in the fields of biotechnology, the life sciences and chemistry. The Distillery goes beyond the abstracts to explain the commercial relevance of featured research, including licensing status and companies working in the field, where applicable.

This week in therapeutics includes important research findings on targets and compounds, grouped first by disease class and then alphabetically by indication.

Indication	Target/marker/pathway	Summary	Licensing status	Publication and contact information
<b>Autoimmune disease</b>				
Systemic lupus erythematosus (SLE)	Solute carrier family 15 member 4 (SLC15A4; PHT1)	<p>Mouse studies suggest antagonizing SLC15A4 could help treat SLE. In a mouse model for SLE, a loss-of-function mutation in <i>Slc15a4</i> prolonged survival and decreased type I interferon and autoimmune antibody production compared with what was seen in mice with wild-type <i>Slc15a4</i>. Next steps include developing a high throughput assay for inhibitors of SLC15A4 or its interaction with endosomal toll-like receptors (TLRs), which are thought to mediate the effects of SLC15A4 activity.</p> <p><b>SciBX 6(8); doi:10.1038/scibx.2013.184</b>  <b>Published online Feb. 28, 2013</b></p>	Patent pending; licensed to an undisclosed company	<p>Baccala, R. <i>et al. Proc. Natl. Acad. Sci. USA</i>; published online Feb. 4, 2013;  doi:10.1073/pnas.1222798110  <b>Contact:</b> Argyrios N. Theofilopoulos, The Scripps Research Institute, La Jolla, Calif.  e-mail: <a href="mailto:argyrio@scripps.edu">argyrio@scripps.edu</a>  <b>Contact:</b> Roberto Baccala, same affiliation as above  e-mail: <a href="mailto:rbaccala@scripps.edu">rbaccala@scripps.edu</a>  <b>Contact:</b> Bruce Beutler, The University of Texas Southwestern Medical Center, Dallas, Texas  e-mail: <a href="mailto:bruce.beutler@utsouthwestern.edu">bruce.beutler@utsouthwestern.edu</a></p>
<b>Cancer</b>				
Acute lymphoblastic leukemia (ALL)	5'-Nucleotidase cytosolic II (NT5C2)	<p><i>In vitro</i> studies suggest inhibiting NT5C2 could help treat chemotherapy-resistant ALL. In the first study, whole-exome sequencing of 103 relapsed T cell and 35 B cell ALL samples identified 21 with mutations in <i>NT5C2</i> that increased nucleotidase activity and were absent at diagnosis. In the second study, gain-of-function mutations in <i>NT5C2</i> were identified in 2 of 10 pediatric patients with B cell ALL at relapse that were absent at diagnosis and in 5 of 61 additional relapsed samples in a follow-up study. Next steps include developing diagnostic assays to identify the mutations and developing NT5C2 inhibitors.</p> <p><b>SciBX 6(8); doi:10.1038/scibx.2013.185</b>  <b>Published online Feb. 28, 2013</b></p>	<p>Patent application filed covering findings in first study; available for licensing for diagnostic and therapeutic applications</p> <p>Patent and licensing status unavailable for findings in second study</p>	<p>Tzoneva, G. <i>et al. Nat. Med.</i>; published online Feb. 3, 2013;  doi:10.1038/nm.3078  <b>Contact:</b> Adolfo Ferrando, Columbia University, New York, N.Y.  e-mail: <a href="mailto:af2196@columbia.edu">af2196@columbia.edu</a></p> <p>Meyer, J.A. <i>et al. Nat. Genet.</i>; published online Feb. 3, 2013;  doi:10.1038/ng.2558  <b>Contact:</b> William L. Carroll, NYU Cancer Institute, New York, N.Y.  e-mail: <a href="mailto:william.carroll@nyumc.org">william.carroll@nyumc.org</a></p>
Acute myelogenous leukemia (AML)	Notch 1 (NOTCH1)	<p>Two studies <i>in vitro</i> and in mice suggest NOTCH pathway activation could help treat AML. In human AML samples and in a mouse model for the cancer, NOTCH-mediated activation of target genes and NOTCH1 mRNA were lower than those in samples from healthy controls. In AML mouse models, increasing NOTCH pathway signaling led to greater cancer cell apoptosis, differentiation and cell cycle arrest than control treatments. Next steps include developing a stable peptide or small molecule agonist of the NOTCH pathway.</p> <p><b>SciBX 6(8); doi:10.1038/scibx.2013.186</b>  <b>Published online Feb. 28, 2013</b></p>	<p>Patent and licensing status unavailable for findings in first study</p> <p>Findings in second study unpatented; available for licensing</p>	<p>Lobry, C. <i>et al. J. Exp. Med.</i>; published online Jan. 28, 2013;  doi:10.1084/jem.20121484  <b>Contact:</b> Iannis Aifantis, New York University School of Medicine, New York, N.Y.  e-mail: <a href="mailto:iannis.aifantis@nyumc.org">iannis.aifantis@nyumc.org</a>  <b>Contact:</b> Camille Lobry, same affiliation as above  e-mail: <a href="mailto:camille.lobry@nyumc.org">camille.lobry@nyumc.org</a></p> <p>Kannan, S. <i>et al. J. Exp. Med.</i>; published online Jan. 28, 2013;  doi:10.1084/jem.20121527  <b>Contact:</b> Patrick A. Zweidler-McKay, The University of Texas MD Anderson Cancer Center, Houston, Texas  e-mail: <a href="mailto:pzweidler@mdanderson.org">pzweidler@mdanderson.org</a></p>

## This week in therapeutics (continued)

Indication	Target/marker/pathway	Summary	Licensing status	Publication and contact information
Cancer	Ataxia telangiectasia and Rad3 related (ATR; FRP1)	Rodent and cell culture studies identified sulfonyl-morpholino-pyrimidine-based ATR inhibitors that could help treat cancer. In a human colorectal cancer cell line, the lead compound caused 50% growth inhibition at nanomolar concentrations. In a mouse xenograft model for human colorectal cancer, oral treatment with the compound decreased tumor growth compared with vehicle treatment. Next steps could include testing the lead ATR inhibitor in models for other cancer types.  <b>SciBX 6(8); doi:10.1038/scibx.2013.187</b> <b>Published online Feb. 28, 2013</b>	Patent application filed; licensing status unavailable	Foote, K.M. <i>et al. J. Med. Chem.</i> ; published online Feb. 11, 2013; doi:10.1021/jm301859s <b>Contact:</b> Kevin Michael Foote, AstraZeneca plc, Cheshire, U.K. e-mail: <a href="mailto:kevin.foote@astrazeneca.com">kevin.foote@astrazeneca.com</a>
Cancer	BCL2-associated X protein (BAX); BCL2-antagonist/killer 1 (BAK1; BAK); B cell lymphoma 2 (BCL-2; BCL2); BCL2 homology domain 3 (BH3)	Structural studies have identified regions of BAK that could guide the development of apoptosis-inducing compounds that could help treat cancer. Crystal structures of BCL2 have guided the development of BH3-mimetic compounds that bind and inhibit prosurvival proteins, but it was unclear how the BH3 domains bind and activate distinct proapoptotic proteins, including BAX and BAK. <i>In vitro</i> , full-length BAK protein was purified and photochemical crosslinking identified a BH3 binding site on BAK that was structurally distinct from the BH3 binding site of BAX. Ongoing work includes optimizing the potency and specificity of BAX and BAK modulators. Abbott Laboratories and Roche's Genentech Inc. have navitoclax (ABT-263), a pan-inhibitor of BCL2-family proteins, in Phase I/II testing for small cell lung cancer and Phase I testing for other cancers. At least six other companies have antagonists of BCL2-family proteins in Phase II testing or earlier to treat various cancers.  <b>SciBX 6(8); doi:10.1038/scibx.2013.188</b> <b>Published online Feb. 28, 2013</b>	Stapled BH3 peptides that directly engage BAX, BAK and antiapoptotic BCL-2 family targets patented; licensed to Aileron Therapeutics Inc.; small molecules that directly engage BAX patented; available for licensing	Leshchiner, E.S. <i>et al. Proc. Natl. Acad. Sci. USA</i> ; published online Feb. 12, 2013; doi:10.1073/pnas.1214313110 <b>Contact:</b> Loren D. Walensky, Dana-Farber Cancer Institute, Boston, Mass. e-mail: <a href="mailto:loren_walensky@dfci.harvard.edu">loren_walensky@dfci.harvard.edu</a>
Cancer	Receptor-interacting serine-threonine kinase 4 (RIPK4; RIP4)	Studies in cell culture, mice and human tissue samples suggest inhibiting RIPK4 could help treat cancers driven by increased wingless-type MMTV integration site (WNT) signaling. In human cells, RIPK4 overexpression increased WNT signaling compared with no overexpression. In a xenograft mouse model for cancer with WNT pathway activation, <i>RIPK4</i> depletion suppressed tumor growth. In patient tissue samples, <i>RIPK4</i> mRNA was increased in some ovarian, skin and colorectal tumors compared with matched healthy tissue. Next steps include testing whether inhibiting RIPK4 would have efficacy in tumors with WNT pathway activation that lack downstream activating mutations.  <b>SciBX 6(8); doi:10.1038/scibx.2013.189</b> <b>Published online Feb. 28, 2013</b>	Patent and licensing status undisclosed	Huang, X. <i>et al. Science</i> ; published online Jan. 31, 2013; doi:10.1126/science.1232253 <b>Contact:</b> Vishva M. Dixit, Genentech Inc., South San Francisco, Calif. e-mail: <a href="mailto:dixit@gene.com">dixit@gene.com</a>
Cancer	Tumor necrosis factor-related apoptosis-inducing ligand (TRAIL)	Cell culture and mouse studies have identified a small molecule inducer of TRAIL expression that could help treat cancer. A cell-based screen identified a compound, TRAIL-inducing compound 10 (TIC10), that upregulated TRAIL expression at low micromolar concentrations and induced apoptosis in a panel of cancer cell lines. In multiple mouse xenograft models of cancer, TIC10 decreased tumor growth and increased survival compared with vehicle control. Next steps include an IND filing at Oncoceutics Inc., for which the corresponding author is founder and CSO ( <i>see Making TRAIL, page 4</i> ).  <b>SciBX 6(8); doi:10.1038/scibx.2013.190</b> <b>Published online Feb. 28, 2013</b>	Patent application filed; exclusively licensed to Oncoceutics	Allen, J.E. <i>et al. Sci. Transl. Med.</i> ; published online Feb. 6, 2013; doi:10.1126/scitranslmed.3004828 <b>Contact:</b> Wafik S. El-Deiry, Penn State Milton S. Hershey Medical Center, Hershey, Pa. e-mail: <a href="mailto:wafk.eldeiry@gmail.com">wafk.eldeiry@gmail.com</a>

## This week in therapeutics (continued)

Indication	Target/marker/pathway	Summary	Licensing status	Publication and contact information
Chronic lymphocytic leukemia (CLL)	Yippee-like 5 (YPEL5); protein phosphatase 1 catalytic subunit $\beta$ -isozyme (PPP1CB)	Patient sample and cell culture studies have identified a <i>YPEL5-PPP1CB</i> fusion transcript that could be a marker or therapeutic target in the disease. The <i>YPEL5-PPP1CB</i> fusion transcript was detected in 97 of 103 samples from patients with CLL but not in any healthy controls. <i>In vitro</i> , the fusion transcript produced a truncated PPP1CB protein with less phosphatase activity than the wild-type protein. In cultured CLL cells, small hairpin RNA against <i>PPP1CB</i> increased cell proliferation compared with control shRNA. Next steps include determining when and how this fusion transcript is produced in patients with CLL.  <b>SciBX 6(8); doi:10.1038/scibx.2013.191</b> <b>Published online Feb. 28, 2013</b>	Patent and licensing status undisclosed	Velusamy, T. <i>et al. Proc. Natl. Acad. Sci. USA</i> ; published online Feb. 4, 2013; doi:10.1073/pnas.1214326110 <b>Contact:</b> Kojo S.J. Elenitoba-Johnson, University of Michigan, Ann Arbor, Mich. e-mail: <a href="mailto:kojoelen@med.umich.edu">kojoelen@med.umich.edu</a> <b>Contact:</b> Arul M. Chinnaiyan, same affiliation as above e-mail: <a href="mailto:arul@umich.edu">arul@umich.edu</a>
Colorectal cancer	Jagged 1 (JAG1)	Cell culture studies suggest blocking soluble JAG1 could help treat colorectal cancer by targeting cancer stem cells. In human colorectal cancer cell lines incubated in endothelial cell-conditioned medium, antibody-mediated depletion of soluble JAG1 decreased the formation of cells with a cancer stem cell phenotype compared with no depletion. Next steps could include evaluating JAG1-targeting antibodies in combination with existing therapies in mouse models of colorectal cancer.  <b>SciBX 6(8); doi:10.1038/scibx.2013.192</b> <b>Published online Feb. 28, 2013</b>	Patent and licensing status unavailable	Lu, J. <i>et al. Cancer Cell</i> ; published online Jan. 31, 2013; doi:10.1016/j.ccr.2012.12.021 <b>Contact:</b> Lee M. Ellis, The University of Texas MD Anderson Cancer Center, Houston, Texas e-mail: <a href="mailto:lellis@mdanderson.org">lellis@mdanderson.org</a>
Leukemia	2-Hydroxyglutarate (2-HG); isocitrate dehydrogenase 1 (IDH1)	Cell culture studies suggest inhibiting the ( <i>R</i> )-enantiomer of 2-HG could help treat leukemia. In two human leukemia cell lines, a membrane-permeable version of ( <i>R</i> )-2-HG promoted leukemic transformation, whereas ( <i>S</i> )-2-HG and vehicle did not. In the two cell lines, a compound that blocked 2-HG production by a cancer-associated mutant <i>IDH1</i> reversed the markers of leukemic transformation, whereas vehicle did not. Next steps include evaluating the inhibition of ( <i>R</i> )-2-HG signaling in leukemia mouse models and in other types of IDH1-driven cancers.  <b>SciBX 6(8); doi:10.1038/scibx.2013.193</b> <b>Published online Feb. 28, 2013</b>	Unpatented; licensing status not applicable	Losman, J.-A. <i>et al. Science</i> ; published online Feb. 7, 2013; doi:10.1126/science.1231677 <b>Contact:</b> William Kaelin Jr., Dana-Farber Cancer Institute, Boston, Mass. e-mail: <a href="mailto:william_kaelin@dfci.harvard.edu">william_kaelin@dfci.harvard.edu</a>
<b>Dermatology</b>				
Wounds	MicroRNA-198 (miR-198); follistatin-like 1 (FSTL1)	Patient sample and cell culture studies suggest inhibiting miR-198 could help treat chronic diabetic wounds. miR-198 is encoded within an exon of <i>FSTL1</i> . In tissue samples from nonhealing ulcers on patients with diabetes, miR-198 expression was greater and FSTL1 expression was lower than that seen in tissue samples from wounded healthy skin. In cell culture, small interfering RNA knockdown of <i>FSTL1</i> or overexpression of <i>miR-198</i> decreased wound healing compared with control treatment. Next steps include conducting additional studies to understand the regulatory mechanisms controlling the switch between <i>FSTL1</i> and <i>miR-198</i> expression and developing antagomirs against miR-198.  <b>SciBX 6(8); doi:10.1038/scibx.2013.194</b> <b>Published online Feb. 28, 2013</b>	Patent and licensing status undisclosed	Sundaram, G.M. <i>et al. Nature</i> ; published online Feb. 10, 2013; doi:10.1038/nature11890 <b>Contact:</b> Prabha Sampath, Institute of Medical Biology, Agency for Science, Technology and Research (A*STAR), Singapore e-mail: <a href="mailto:prabha.sampath@imb.a-star.edu.sg">prabha.sampath@imb.a-star.edu.sg</a>

## This week in therapeutics (continued)

Indication	Target/marker/ pathway	Summary	Licensing status	Publication and contact information
<b>Endocrine/metabolic disease</b>				
Diabetes; obesity	HNF1 homeobox B (HNF1B); microRNA-802 (miR-802)	Mouse and human studies suggest inhibiting miR-802 or increasing HNF1B signaling could help treat obesity and type 2 diabetes. In overweight humans and in genetic and diet-induced obesity mouse models, miR-802 expression was greater in the liver than that in livers from lean individuals and nonobese control mice. In the mouse obesity models, locked nucleic acid (LNA) antagonists of miR-802 expression increased insulin resistance and glucose tolerance compared with control LNAs. In the mouse models, vector-induced overexpression of <i>Hnf1b</i> , the identified target of miR-802, also improved insulin resistance and glucose tolerance. Next steps could include developing a screen to identify molecules that inhibit miR-802 and/or promote HNF1B signaling.	Patent and licensing status unavailable	Kornfeld, J.-W. <i>et al. Nature</i> ; published online Feb. 6, 2013; doi:10.1038/nature11793 <b>Contact:</b> Jens Brüning, Max Planck Institute for Neurological Research, Cologne, Germany e-mail: <a href="mailto:bruening@nf.mpg.de">bruening@nf.mpg.de</a>
<b>SciBX 6(8); doi:10.1038/scibx.2013.195</b> Published online Feb. 28, 2013				
<b>Hematology</b>				
Thalassemia; sickle cell disease	Lysine-specific histone demethylase 1 (KDM1A; LSD1)	<i>In vitro</i> and mouse studies suggest inhibiting LSD1 could help treat sickle cell disease and $\beta$ -thalassemia. Inducing hemoglobin- $\gamma$ A (HBG1) expression reduces symptoms of $\beta$ -globinopathies including $\beta$ -thalassemia and sickle cell disease, but current inducers have limited efficacy and can cause toxicity. In transgenic mice expressing human hemoglobin- $\beta$ (HBB) to model the diseases, inhibition of the HBG1 transcriptional repressor LSD1 with tranlycypromine increased HBG1 expression compared with no inhibition. Next steps could include testing LSD1 inhibition in additional models. Tranlycypromine is a generic monoamine oxidase inhibitor (MAOI) marketed to treat depression. Oryzon Genomics S.A.'s LSD1 inhibitor, ORY-1001, is in Phase I/II testing to treat acute myelogenous leukemia (AML).	Patent and licensing status unavailable	Shi, L. <i>et al. Nat. Med.</i> ; published online Feb. 17, 2013; doi:10.1038/nm.3101 <b>Contact:</b> Osamu Tanabe, University of Michigan Medical School, Ann Arbor, Michigan e-mail: <a href="mailto:otanabe@umich.edu">otanabe@umich.edu</a> <b>Contact:</b> James D. Engel, same affiliation as above e-mail: <a href="mailto:engel@umich.edu">engel@umich.edu</a>
<b>SciBX 6(8); doi:10.1038/scibx.2013.196</b> Published online Feb. 28, 2013				
<b>Infectious disease</b>				
Influenza virus	Influenza A virus hemagglutinin (HA)	Mouse studies suggest a liposome formulation of a sialic acid-based sugar could help prevent or treat influenza infection. The sugar sialylneolacto- <i>N</i> -tetraose c blocks the interaction of HA with host cells. In mice infected with influenza A virus, liposome-mediated delivery of the sugar during viral challenge led to greater survival than delivery of control liposomes. Next steps include optimizing the sugar chemistry and liposome formulation.	Patent and licensing status undisclosed	Hendricks, G.L. <i>et al. J. Biol. Chem.</i> ; published online Jan. 28, 2013; doi:10.1074/jbc.M112.437202 <b>Contact:</b> Jennifer P. Wang, University of Massachusetts Medical School, Worcester, Mass. e-mail: <a href="mailto:jennifer.wang@umassmed.edu">jennifer.wang@umassmed.edu</a>
<b>SciBX 6(8); doi:10.1038/scibx.2013.197</b> Published online Feb. 28, 2013				
<b>Inflammation</b>				
Asthma	<i>Forkhead box P3</i> (FOXP3)	Mice studies suggest modified <i>FOXP3</i> mRNA could help prevent or treat allergy-induced asthma. In mice, intratracheal delivery of <i>Foxp3</i> mRNA modified with 2-thiouridine and 5-methylcytidine led to greater <i>Foxp3</i> expression in immune cells in the lung and lower allergen-induced lung inflammation than intratracheal delivery of saline. Next steps include optimizing mRNA modifications, formulations and delivery approaches, including development of a powder for inhalation.	Unpatented; licensing status not applicable	Mays, L.E. <i>et al. J. Clin. Invest.</i> ; published online Feb. 8, 2013; doi:10.1172/JCI65351 <b>Contact:</b> Michael S.D. Kormann, University of Tuebingen, Tuebingen, Germany e-mail: <a href="mailto:kormann.michael@gmail.com">kormann.michael@gmail.com</a>
<b>SciBX 6(8); doi:10.1038/scibx.2013.198</b> Published online Feb. 28, 2013				

## This week in therapeutics (continued)

Indication	Target/marker/ pathway	Summary	Licensing status	Publication and contact information
<b>Neurology</b>				
Alzheimer's disease (AD)	Prion protein (PrNP; PrP; CD230)	<i>In vitro</i> and mouse studies suggest the soluble N1 fragment of PrP could help treat AD. <i>In vitro</i> , both the N1 fragment and full-length PrP bound synthetic $\beta$ -amyloid ( $A\beta$ ) oligomers and inhibited fibril formation. In a mouse model for $A\beta$ toxicity, intracerebroventricular microinfusion of $A\beta$ oligomers impaired cognition and behavior, whereas microinfusion of a mixture of $A\beta$ oligomers and N1 did not. Next steps include designing N1-derived peptides with improved CNS penetration.  <b>SciBX 6(8); doi:10.1038/scibx.2013.199</b> <b>Published online Feb. 28, 2013</b>	Findings unpatented; licensing status not applicable	Fluharty, B.R. <i>et al. J. Biol. Chem.</i> ; published online Jan. 28, 2013; doi:10.1074/jbc.M112.423954 <b>Contact:</b> Emiliano Biasini, Boston University School of Medicine, Boston, Mass. e-mail: <a href="mailto:biasini@bu.edu">biasini@bu.edu</a>
Amyotrophic lateral sclerosis (ALS); frontal temporal dementia	Chromosome 9 open reading frame 72 (C9ORF72)	Studies <i>in vitro</i> and in patient tissue samples have identified peptides translated from C9ORF72 that could be biomarkers for frontal temporal lobar dementia (FTLD) and ALS or could be targeted to treat the conditions. Expansion of GGGGCC repeats in the noncoding region of C9ORF72 is the most common genetic marker associated with ALS and FTLD. In the first study, non-ATG (RAN) translation of the repeat expansions created insoluble protein aggregates, including poly-(Gly-Ala) dipeptide-repeat proteins, which were detected in cytoplasmic inclusions in CNS samples from patients with C9ORF72 repeats but not in healthy controls. In the second study, the aggregates were detected in neuronal inclusions in CNS tissues from patients with FTLD and ALS carrying the C9ORF72 repeats but not in peripheral tissues from the patients or CNS tissues from patients without the mutation. Next steps could include screening for compounds that interfere with the production or aggregation of these proteins.  <b>SciBX 6(8); doi:10.1038/scibx.2013.200</b> <b>Published online Feb. 28, 2013</b>	Patent application filed for findings in first study; available for licensing  Patent and licensing status undisclosed for findings in second study	Mori, K. <i>et al. Science</i> ; published online Feb. 7, 2013; doi:10.1126/science.1232927 <b>Contact:</b> Dieter Edbauer, Ludwig Maximilian University of Munich, Munich, Germany e-mail: <a href="mailto:dieter.edbauer@dzne.de">dieter.edbauer@dzne.de</a>  Ash, P.E.A. <i>et al. Neuron</i> ; published online Feb. 12, 2013; doi:10.1016/j.neuron.2013.02.004 <b>Contact:</b> Leonard Petrucelli, Mayo Clinic Florida, Jacksonville, Fla. e-mail: <a href="mailto:petrucelli.leonard@mayo.edu">petrucelli.leonard@mayo.edu</a>
<b>Transplantation</b>				
Bone marrow transplant	Epidermal growth factor (EGF)	Mouse studies suggest EGF could be useful in aiding recovery after bone marrow transplant. In irradiated mice, EGF increased hematopoietic stem cell (HSC) regeneration and survival compared with vehicle. In these mice, genetic or pharmacological disruption of EGF signaling decreased HSC regeneration and survival compared with no disruption. Next steps include scaling up EGF manufacturing and conducting preclinical studies comparing its efficacy with that of granulocyte colony-stimulating factor (G-CSF; CSF3), the standard of care for HSC mobilization after myeloablative therapy.  <b>SciBX 6(8); doi:10.1038/scibx.2013.201</b> <b>Published online Feb. 28, 2013</b>	Patent pending; available for licensing	Doan, P.L. <i>et al. Nat. Med.</i> ; published online Feb. 3, 2013; doi:10.1038/nm.3070 <b>Contact:</b> John P. Chute, Duke University, Durham, N.C. e-mail: <a href="mailto:john.chute@duke.edu">john.chute@duke.edu</a>

## This week in techniques

**THE DISTILLERY** brings you this week's most essential scientific findings in techniques, distilled by *SciBX* editors from a weekly review of more than 400 papers in 41 of the highest-impact journals in the fields of biotechnology, the life sciences and chemistry. The Distillery goes beyond the abstracts to explain the commercial relevance of featured research, including licensing status and companies working in the field, where applicable.

This week in techniques includes findings about research tools, disease models and manufacturing processes that have the potential to enable or improve all stages of drug discovery and development.

Approach	Summary	Licensing status	Publication and contact information
<b>Disease models</b>			
Comprehensive library of human genome-targeted transcription activator-like effector nucleases (TALENs)	A library of TALENs designed to disrupt every protein-coding region of the human genome could be used to generate new disease models. A computational algorithm was developed to identify unique 40-base-pair sites in the 5' end of protein-coding genes that have minimal potential for off-target effects. Using this information, about 18,000 gene-targeting TALEN pairs were cloned and assembled in 96-well plates. In cultured cells, a test set of 126 TALENs induced site-specific mutations with a 98.4% success rate. Next steps include further optimizing the TALENs and using this approach to perform functional knockout studies.	Patented; TALEN plasmids available from Seoul National University through its <a href="#">TALEN library</a> ; TALEN-modified cells available for purchase from ToolGen= Inc.	Kim, Y. <i>et al. Nat. Biotechnol.</i> ; published online Feb. 17, 2013; doi:10.1038/nbt.2517 <b>Contact:</b> Jin-Soo Kim, Seoul National University, Seoul, South Korea e-mail: <a href="mailto:jskim01@snu.ac.kr">jskim01@snu.ac.kr</a> <b>Contact:</b> Seokjoong Kim, ToolGen Inc., Seoul, South Korea e-mail: <a href="mailto:sj.kim@toolgen.com">sj.kim@toolgen.com</a>
	<b>SciBX 6(8); doi:10.1038/scibx.2013.202</b> Published online Feb. 28, 2013		
Induced pluripotent stem (iPS) cell-derived astrocytes from patients with amyotrophic lateral sclerosis (ALS)	Astrocytes derived from patient iPS cells could be useful for evaluating therapeutic candidates to treat ALS. Astrocytes carrying an ALS-causing mutation in TAR DNA binding protein 43 (TDP-43; TARDBP) were generated from an iPS cell line derived from a patient with familial ALS. In culture, the astrocytes showed mislocalization of the protein in the cytoplasm and increased TDP-43 levels and decreased survival compared with astrocytes generated from control iPS cell lines. Next steps could include using the cell model to evaluate therapeutic candidates for ALS.	Patent and licensing status unavailable	Serio, A. <i>et al. Proc. Natl. Acad. Sci. USA</i> ; published online Feb. 11, 2013; doi:10.1073/pnas.1300398110 <b>Contact:</b> Siddharthan Chandran, The University of Edinburgh, Edinburgh, U.K. e-mail: <a href="mailto:siddharthan.chandran@ed.ac.uk">siddharthan.chandran@ed.ac.uk</a> <b>Contact:</b> Tom Maniatis, Columbia University, New York, N.Y. e-mail: <a href="mailto:tm2472@columbia.edu">tm2472@columbia.edu</a>
	<b>SciBX 6(8); doi:10.1038/scibx.2013.203</b> Published online Feb. 28, 2013		
<b>Drug delivery</b>			
Apolipoprotein E (APOE)-derived peptide for drug delivery across the blood brain barrier (BBB)	<i>In vitro</i> and mouse studies suggest an APOE-derived peptide could help deliver therapeutics across the BBB. <i>In vitro</i> assays identified two peptides derived from APOE that bound low-density lipoprotein-related protein 1 $\alpha$ -2-macroglobulin receptor (LRP1; CD91), a receptor on the BBB surface, and underwent transcytosis. In a mouse model for the lysosomal storage disease mucopolysaccharidosis I (MPS I), which involves loss of $\alpha$ -L-iduronidase (IDUA) function in the CNS, delivery of transgenes encoding a fusion between the APOE-derived peptide and IDUA increased levels of the enzyme in the brain compared with delivery of transgenes encoding a control peptide-IDUA fusion. Next steps include testing the effects on neurological deficits in additional animal models of MPS I. Angiochem Inc. has LRP1-binding peptide-enzyme fusions in preclinical development to treat lysosomal storage disorders.	Patent application filed; available for licensing	Wang, D. <i>et al. Proc. Natl. Acad. Sci. USA</i> ; published online Feb. 4, 2013; doi:10.1073/pnas.1222742110 <b>Contact:</b> Dao Pan, Cincinnati Children's Hospital Medical Center, Cincinnati, Ohio e-mail: <a href="mailto:dao.pan@cchmc.org">dao.pan@cchmc.org</a> <b>Contact:</b> Roscoe O. Brady, National Institutes of Health, Bethesda, Md. e-mail: <a href="mailto:bradyr@ninds.nih.gov">bradyr@ninds.nih.gov</a>
	<b>SciBX 6(8); doi:10.1038/scibx.2013.204</b> Published online Feb. 28, 2013		

