

THIS WEEK

ANALYSIS

COVER STORY

1 Pre-EMP-tive strike against GBM

UCLA researchers have treated glioblastoma in mice by inhibiting EMP2. The findings open up a new indication for spinout Paganini, which has a mAb against the target in development for triple-negative breast cancer.

TARGETS & MECHANISMS

4 Positioning properdin

Researchers at the University of Leicester have shown that a recombinant properdin produced in collaboration with The Medicines Co. has markedly higher antibacterial activity than the native protein. The researchers are now working to improve the recombinant protein's activity and homogeneity.

6 Catastrophic vacuolization

Karolinska Institute scientists have attacked glioblastoma multiforme by inducing an unconventional cell death pathway involving catastrophic vacuolization. The team identified a small molecule that prolonged survival in mouse GBM, but a combination approach with a conventional anticancer agent will likely be necessary in the clinic.

TOOLS

8 Toxic assets

A tool for *in vivo* detection of liver toxicity could represent a substantial improvement over *in vitro* methods. The litmus test for the Stanford University inventors will be to show that the nanoparticle-based method can detect toxicity in compounds that previously eluded standard analysis and later failed in the clinic.

THE DISTILLERY

10 This week in therapeutics

Allosteric inhibition of thrombin to prevent blood clots and thrombosis; FXR activation by vertical sleeve gastrectomy to counter obesity; antagonism of HRD1 to treat liver cirrhosis; and more...

15 This week in techniques

A mouse model for ovarian cancer with pituitary-specific estrogen receptor knockout; CAIX-targeted small molecule-drug conjugates; small molecule inhibitors of microRNA processing; and more...

INDEXES

18 Company and institution index**18 Target and compound index**

Pre-EMP-tive strike against GBM

By Michael J. Haas, Senior Writer

University of California, Los Angeles researchers have treated glioblastoma in mice by inhibiting epithelial membrane protein 2.¹ The findings open up a new indication for spinout **Paganini Biopharma Inc.**, which has a mAb against the target in development for triple-negative breast cancer.

Epithelial membrane protein 2 (EMP2) is expressed in multiple tissues, including heart, lung, uterus and eye, in which it interacts with integrins to regulate adhesion between cells and the extracellular matrix.^{2,3}

Over the past eight years, several UCLA teams led by Madhuri Wadehra showed that EMP2 was upregulated in endometrial, ovarian and breast cancers, in which it correlated with advanced disease and poor survival,⁴⁻⁷ and that inhibiting the protein reduced ovarian and breast tumor growth in mice.^{6,7}

Based on a different group's gene expression research,⁸ Wadehra hypothesized that EMP2 also could be a target in glioblastoma multiforme (GBM)—the most common and aggressive form of brain cancer.

First, the team showed that *EMP2* upregulation directly correlated with EMP2 levels in GBM. In a panel of samples from more than 300 patients with GBM, up to 95% of primary GBM tumors had higher levels of EMP2 than the surrounding normal brain tissue.

In addition, tumor levels of EMP2 correlated positively with activation of Src—an intracellular tyrosine kinase that contributes to cancer progression—and correlated negatively with patient survival.

In human GBM cell lines, EMP2 enhanced cell invasiveness by activating the integrin $\alpha_3\beta_3$ (CD51/CD61)-focal adhesion kinase (FAK)-Src signaling pathway. In mice injected intracranially with human GBM cell lines, imaging studies showed that tumors generated from *EMP2*⁺ cells were more invasive than tumors produced from cells in which *EMP2* had been silenced with shRNA.

Lastly, the team tested two EMP2 inhibitors in mice with subcutaneous GBM xenografts. The first was Paganini's PG-101 anti-EMP2 mAb. The second was an anti-EMP2 diabody, which is a type of antibody fragment that is formed from two single-chain variable fragments and potentially has higher affinity for its target than a mAb.

In the models, i.p. administration of either agent decreased tumor growth compared with administration of inactive controls.

Data were reported in *The Journal of Biological Chemistry*.

Wadehra is an adjunct assistant professor of pathology and laboratory medicine at the University of California, Los Angeles David Geffen

**EDITORIAL****Editor-in-Chief:** Karen Bernstein, Ph.D.**Managing Editor:** Gaspar Taroncher-Oldenburg, Ph.D.**Executive Editor:** Steve Edelson**Senior Editors:** Tracey Baas, Ph.D.; Amy Donner, Ph.D.;
C. Simone Fishburn, Ph.D.**Associate Editor:** Benjamin Boettner, Ph.D.**Writers:** Chris Cain, Ph.D.; Michael J. Haas; Kai-Jye Lou; Lauren Martz;
Lev Osherovich, Ph.D.**Research Director:** Walter Yang**Research Manager:** Kevin Lehnbeuter**Production Editors:** Brandy Cafarella; Carol Evangelista; Jennifer Gustavson**Copy Editor:** Nicole DeGennaro**Editorial Assistant:** Mark Zipkin**Design:** Claudia Bentley; Miles DaviesFor inquiries, contact editorial@scibx.com**PUBLISHING****Publisher:** Peter Collins, Ph.D.**Associate Publishers:** Gaspar Taroncher-Oldenburg, Ph.D.; Eric Pierce**Marketing:** Sara Girard; Greg Monteforte**Technology:** Anthony Barrera; Julia Kulikova**Sales:** Ron Rabinowitz; Dean Sanderson; Tim Tulloch**OFFICES****BioCentury Publications, Inc.**San Francisco
PO Box 1246
San Carlos, CA 94070-1246
T: +1 650 595 5333Chicago
20 N. Wacker Drive, Suite 1465
Chicago, IL 60606-2902
T: +1 312 755 0798United Kingdom
T: +44 (0)18 6551 2184Washington, DC
2008 Q Street, NW, Suite 100
Washington, DC 20009
T: +1 202 462 9582**Nature Publishing Group**New York
75 Varick Street, 9th Floor
New York, NY 10013-1917
T: +1 212 726 9200London
The Macmillan Building
4 Crinan Street
London N1 9XW
United Kingdom
T: +44 (0)20 7833 4000Tokyo
Chiyoda Building 6F
2-37 Ichigayatamachi
Shinjuku-ku, Tokyo 162-0843
Japan
T: +81 3 3267 8751

SciBX is produced by BioCentury Publications, Inc. and Nature Publishing Group Joint Steering Committee: Karen Bernstein, Ph.D., Chairman & Editor-in-Chief, BioCentury; David Flores, President & CEO, BioCentury; Bennet Weintraub, Finance Director, BioCentury; Steven Inchcoombe, Managing Director, Nature Publishing Group; Peter Collins, Ph.D., Publishing Director, NPG; Christoph Hesselmann, Ph.D., Chief Financial Officer, NPG.

Copyright © 2014 Nature Publishing Group ALL RIGHTS RESERVED.

No part of the SciBX publication or website may be copied, reproduced, retransmitted, disseminated, sold, distributed, published, broadcast, circulated, commercially exploited or used to create derivative works without the written consent of the Publishers. Information provided by the SciBX publication and website is gathered from sources that the Publishers believe are reliable; however, the Publishers do not guarantee the accuracy, completeness, or timeliness of the information, nor do the Publishers make any warranties of any kind regarding the information. The contents of the SciBX publication and website are not intended as investment, business, tax or legal advice, and the Publishers are not responsible for any investment, business, tax or legal opinions cited therein.

School of Medicine. She also is a member of the Cancer and Stem Cell Biology Program Area at the **University of California, Los Angeles Jonsson Comprehensive Cancer Center** and a cofounder of Paganini.

The team included a researcher from the **University of California, San Diego.**

Safe harbor upstream

Wadehra told SciBX that EMP2 inhibitors could treat cancer with fewer side effects than inhibitors of the CD51/CD61-FAK-Src pathway. She said that the latter three targets are widely expressed by normal cells, whereas EMP2 is highly expressed in GBM and other tumors and has limited expression in normal tissue.

“We feel that targeting EMP2 may be a novel way to downregulate the CD51/CD61-FAK-Src pathway that has been shown to be important for tumorigenesis,” she said.

At least 16 companies have cancer compounds that inhibit CD51/CD61, FAK or Src on the market or in clinical development (see Table 1, “EMP2’s downstream crowd”).

Before Paganini decides whether to develop PG-101 or the diabody to treat GBM, Wadehra said that the UCLA team needs to compare the efficacy and delivery routes of each agent in mice with orthotopic, intracranial GBM tumors.

Initially the team will test the effect of direct intracranial administration of the antibody in the orthotopic models. “But we are also going to test various delivery strategies—including stem cell-based delivery—to determine whether we can get the diabody across the blood brain barrier,” she said.

UCLA owns a patent portfolio that covers several anti-EMP2 mAbs, their uses to treat cancer and ophthalmic indications, and diagnostic uses of

SciBX: Science–Business eXchange

*SciBX welcomes editorial queries,
comments and press releases.*

To contact the editorial team at SciBX
please e-mail editorial@scibx.com

Table 1. EMP2's downstream crowd. University of California, Los Angeles researchers have shown that in mouse models of glioblastoma multiforme (GBM) and breast cancer, epithelial membrane protein 2 (EMP2) activates the integrin $\alpha_v\beta_3$ (CD51/CD61)–focal adhesion kinase (FAK)–Src signaling pathway that drives cancer progression.¹ At least 16 companies have therapies on the market or in the clinic that target 1 of the 3 downstream components on that signaling pathway to treat cancer.

Source: *BCIQ: BioCentury Online Intelligence*

Company	Product	Description	Status
Bristol-Myers Squibb Co. (NYSE: BMY)/ Otsuka Pharmaceutical Co. Ltd.	Sprycel dasatinib (BMS-354825)	Small molecule inhibitor of BCR-ABL tyrosine kinase and Src kinase	Marketed for acute lymphoblastic leukemia (ALL) and chronic myelogenous leukemia (CML); Phase II for breast and pancreatic cancers; Phase I for relapsed or refractory leukemia
Pfizer Inc. (NYSE: PFE)/ Avillion LLP	Bosulif bosutinib (PF-05208763; SKI-606)	Dual inhibitor of BCR-ABL and Src kinases	Marketed for CML
Merck KGaA (Xetra: MRK)	Cilengitide (EMD 121974)	Inhibitor of CD51/CD61 and integrin $\alpha_v\beta_3$	Phase III for brain cancer; Phase II for head and neck cancer, non-small cell lung cancer (NSCLC) and melanoma
Bristol-Myers Squibb/ Johnson & Johnson (NYSE: JNJ)	Intetumumab (BGB-101; CNTO-95)	Human mAb targeting CD51/CD61, integrin $\alpha_v\beta_3$, integrin $\alpha_v\beta_6$ and integrin $\alpha_5\beta_1$ (CD51/CD29)	Phase II for melanoma and castration-resistant prostate cancer (CRPC); Phase I for solid tumors
Kinex Pharmaceuticals LLC/Hanmi Pharmaceutical Co. Ltd. (KOSDAQ: 128940)	KX01 (KX2-391)	Small molecule, non-ATP Src inhibitor	Phase II for prostate cancer; Phase I/II for breast cancer, gastric cancer and solid tumors; Phase Ib for acute myelogenous leukemia (AML)
Pfizer/ Verastem Inc. (NASDAQ: VSTM)	Defactinib (VS-6063; formerly PF-4554878)	Inhibitor of FAK targeting cancer stem cells	Phase II for mesothelioma and NSCLC; Phase I for ovarian cancer and solid tumors
Tactic Pharma LLC	ATN-161	Inhibitor of CD51/CD61 and integrin $\alpha_5\beta_1$	Phase II for brain cancer; Phase I for head and neck cancer
BioAlliance Pharma S.A. (Euronext: BIO)	AMEP (BA-015)	Plasmid encoding a peptide targeting CD51/CD61 and integrin $\alpha_5\beta_1$	Phase I/II for melanoma
Nippon Shinyaku Co. Ltd. (Tokyo: 4516)	NS-018	Inhibitor of Src and Janus kinase-2 (JAK-2)	Phase I/II for hematologic malignancies
Verastem	VS-4718	Inhibitor of FAK targeting cancer stem cells	Phase I for solid tumors
AstraZeneca plc (LSE: AZN; NYSE: AZN)	AZD0424	Src inhibitor	Phase I for advanced solid tumors
GlaxoSmithKline plc (LSE: GSK; NYSE: GSK)	GSK2256098	Small molecule FAK inhibitor	Phase I for solid tumors
Teva Pharmaceutical Industries Ltd. (NYSE: TEVA)	CEP-37440	Inhibitor of FAK and anaplastic lymphoma kinase (ALK)	Phase I for solid tumors
ValiRx plc (LSE: VAL)	VAL201	Src inhibitor	Phase I for CRPC and other cancers

EMP2, Wadehra said. The IP portfolio is licensed to Paganini, which spun out of UCLA in 2011.

Paganini president Gary Lazar said that the biotech is in discussions with potential partners for the clinical development of PG-101 to treat triple-negative breast cancer.

Wadehra said that the team has not found a maximum tolerated dose of PG-101. "We have shown that the antibody administered at 40 mg/kg doses twice a week is safe and has no measurable toxicity," she said.

Lazar added that the anti-EMP2 diabody is not yet in Paganini's pipeline because it was developed as a research compound and has a short serum half-life.

"Additional academic studies are needed to determine its efficacy" in treating GBM or as an intravitreally administered agent to treat the ophthalmic diseases Paganini is pursuing, such as those involving aberrant proliferation of retinal pigment epithelial cells or aberrant corneal neovascularization, he said.

Haas, M.J. *SciBX* 7(14); doi:10.1038/scibx.2014.389
Published online April 10, 2014

REFERENCES

- Qin, Y. *et al. J. Biol. Chem.*; published online March 18, 2014; doi:10.1074/jbc.M113.543728
Contact: Madhuri Wadehra, University of California, Los Angeles David Geffen School of Medicine, Los Angeles, Calif.
e-mail: mwadehra@mednet.ucla.edu
- Wadehra, M. *et al. J. Biol. Chem.* **277**, 41094–41100 (2002)
- Wadehra, M. *et al. Exp. Mol. Pathol.* **74**, 106–112 (2003)
- Wadehra, M. *et al. Cancer* **107**, 90–98 (2006)
- Habeeb, O. *et al. Cancer* **116**, 4718–4726 (2010)
- Fu, M. *et al. Clin. Cancer Res.* **16**, 3954–3963 (2010)
- Fu, M. *et al. Mol. Cancer Ther.*; published online Jan. 21, 2014; doi:10.1158/1535-7163.MCT-13-0199
- Freije, W.A. *et al. Cancer Res.* **64**, 6503–6510 (2004)

COMPANIES AND INSTITUTIONS MENTIONED

Paganini Biopharma Inc., Encino, Calif.
University of California, Los Angeles, Calif.
University of California, Los Angeles, David Geffen School of Medicine, Los Angeles, Calif.
University of California, Los Angeles, Jonsson Comprehensive Cancer Center, Los Angeles, Calif.
University of California, San Diego, La Jolla, Calif.

Positioning properdin

By Kai-Jye Lou, Senior Writer

Researchers at the **University of Leicester** have shown that a recombinant properdin produced in collaboration with **The Medicines Co.** has markedly higher antibacterial activity than the native protein.¹ The researchers are now working to improve the activity and homogeneity of recombinant properdin to treat a broad range of infections.

Properdin is a glycoprotein composed entirely of thrombospondin-like repeat motifs. It is typically found in circulation as a dimer, trimer or tetramer. The protein stabilizes a key enzyme called alternative pathway C3 convertase that amplifies complement system activity.

The complement system is part of the innate immune system and helps promote the clearance of pathogens as well as potentially harmful substances such as cellular debris.

About 3.5 years ago, Wilhelm Schwaeble and his team at the university started a collaboration with The Medicines Co. to develop methods for producing recombinant human thrombospondin proteins in eukaryotic cells. The goal was to produce thrombospondin-1 (TSP-1; THBS1) for studies to evaluate the glycoprotein's effects on platelet activation and strategies to modulate thrombospondin-mediated thrombus formation. Schwaeble is a professor of immunology at the university and a Royal Society Wolfson Research Merit Award holder.

The Medicines Co.'s lead infectious disease program is oritavancin, a semisynthetic, broad-spectrum lipoglycopeptide antibiotic. In February, the company submitted an NDA to the FDA for oritavancin to treat acute bacterial skin and skin structure infections (ABSSSIs) caused by Gram-positive bacteria and an MAA to the EMA for oritavancin to treat complicated skin and skin tissue infections (cSSTIs) caused by Gram-positive bacteria. The company could not be reached for comment and does not have a disclosed therapeutic program involving properdin.

An unexpected result from the collaboration has been the generation of recombinant properdin oligomers that are much larger than those normally found in serum. According to Schwaeble, these oligomers come in a range of sizes and are comprised of more than 12 properdin monomers linked in a head-to-tail fashion.

Previously, larger properdin oligomers had been shown to stabilize the alternative pathway C3 convertase better than smaller oligomers.²

In the current study, the researchers sought to assess the activity of the large recombinant properdin oligomers against bacterial pathogens.

In vitro, recombinant properdin was about 100-fold more effective at supporting complement activation than native properdin. The large oligomers also increased complement-mediated lysis of *Neisseria meningitidis* compared with the native protein in serum.

In mice, i.p. injection of the recombinant properdin prior to a lethal challenge with *N. meningitidis* resulted in 90% survival, whereas injection of saline led to 0% survival. Mice treated with recombinant

properdin three hours after a lethal *N. meningitidis* challenge showed significantly longer survival than saline-treated controls ($p=0.0007$).

In mice infected with *Streptococcus pneumoniae*, injection of the recombinant properdin at the time of infection resulted in minimal or no disease, whereas injection of saline resulted in 70% of mice being euthanized because of development of severe disease.

Results were published in the *Proceedings of the National Academy of Sciences*. The Medicines Co. provided support for the study.

"Our recombinant properdin was found to form higher-order polymers with much higher functionality than those typically obtained from properdin purified from mouse and human serum," said corresponding author Schwaeble.

He said that the speed, strength and brevity of the properdin-induced innate immune response are the key benefits of the approach. "This is what we want in an effective immune response—we want it to come on quick and strong to clear the infection and then go away after it has fulfilled its task in killing and/or clearing the invading pathogens and before damaging healthy tissues," Schwaeble told *SciBX*.

"Aside from the positive effects observed in the tested model systems, the lack of an endotoxic shock response is particularly promising," added Dennis Hourcade, a research professor of medicine at the **Washington University in St. Louis School of Medicine**.

Endotoxic shock, or septic shock, can be triggered by endotoxins released into the bloodstream.

Indeed, Schwaeble had expected to see a septic shock response in mice because the complement-mediated bacteriolytic activity induced by recombinant properdin would release large quantities of bacterial debris into the bloodstream.

However, he said that the lack of a shock response could be because recombinant properdin potentially ramps up complement-mediated clearance of cellular debris.

Active immunotherapy for infections

Schwaeble thinks that the recombinant properdin has potential as an active immunotherapy to treat infections.

He added that although antibiotics act quickly, they do nothing to clear the cellular debris generated from killing the bacteria. Such debris could induce septic shock.

Schwaeble thus said that recombinant properdin has the potential to be used in combination with antibiotics to help promote the clearance of bacterial debris and mitigate the risks of septic shock.

Sanjay Ram, an associate professor of medicine in the division of infectious diseases and immunology at the **University of Massachusetts Medical School**, said that it is desirable to have active immunotherapies against drug-resistant bacteria.

Although he noted that the emergence of antibiotic resistance in *N. meningitidis* and *S. pneumoniae* in the U.S. has been limited thus far, Ram added, "if the approach does

prove useful in *Pseudomonas* or in other bacteria known to have become highly resistant to antibiotics, then something like this could be promising."

"If the approach does prove useful in *Pseudomonas* or in other bacteria known to have become highly resistant to antibiotics, then something like this could be promising."

**—Sanjay Ram,
University of Massachusetts
Medical School**

Both Ram and Hourcade wanted additional studies to characterize the safety of recombinant properdin given that the complement activation induced by the multimeric form of the protein is nonspecific and has the potential to damage healthy tissues.

Ram said that it will be important to assess the degree of complement activation, cytokine levels and kidney function following multiple doses with recombinant properdin.

Hourcade wanted to see studies to determine whether recombinant properdin overwhelms the effects of human complement factor H (CFH), which protects healthy tissues from complement-mediated attack. He added that it also will be important to evaluate recombinant properdin in settings in which there is a pre-existing autoimmune or inflammatory condition such as arthritis.

Finally, Hourcade wanted to see biodistribution studies of recombinant properdin, including how long the protein remains in circulation, how it is metabolized and how it is cleared from the body.

Schwaeble said that his group is trying to further increase the functional activity of recombinant properdin and produce it under better-controlled conditions and according to pharmacological standards.

“Our goal is to manufacture a more homogeneous and stable product in a large-scale production process,” he said.

The group also is evaluating recombinant properdin in additional models of bacterial infection, including *Pseudomonas*, as well as in models of parasitic infection.

The University of Leicester and The Medicines Co. have cofiled a patent covering the use of the recombinant properdin for treating or preventing properdin-related diseases and disorders, such as *N. meningitidis* infection. The work is available for licensing.

Lou, K.-J. *SciBX* 7(14); doi:10.1038/scibx.2014.390
Published online April 10, 2014

REFERENCES

1. Ali, Y.M. *et al. Proc. Natl. Acad. Sci. USA*; published online March 24, 2014; doi:10.1073/pnas.1401011111
Contact: Wilhelm J. Schwaeble, University of Leicester, Leicester, U.K.
e-mail: ws5@le.ac.uk
2. Higgins, J.M. *et al. J. Immunol.* **155**, 5777–5785 (1995)

COMPANIES AND INSTITUTIONS MENTIONED

The Medicines Co. (NASDAQ:MDCO), Parsippany, N.J.
University of Leicester, Leicester, U.K.
University of Massachusetts Medical School, Worcester, Mass.
Washington University in St. Louis School of Medicine, St. Louis, Mo.



SciBX: Science-Business eXchange

“Understanding the business context and commercial relevance of new science is the key to lowering investment risk and stimulating industry innovation”

Become a subscriber today!

Visit scibx.com for details on how to subscribe to *SciBX*

Catastrophic vacuolization

By Tracey Baas, Senior Editor

Karolinska Institute scientists have found a way to attack glioblastoma multiforme—one of the most deadly forms of brain cancer—by inducing an unconventional cell death pathway that triggers catastrophic vacuolization.¹ The team identified a small molecule that prolonged survival in a mouse model of glioblastoma without affecting normal brain tissue, but it will likely need to combine the compound with a conventional anticancer agent to translate it for clinical use.

The Karolinska team looked for a nonconventional therapy for glioblastoma multiforme (GBM) because compounds that target tumorigenic pathways have barely made a dent in the disease.

Standard GBM treatment is surgical resection coupled with chemotherapy and radiotherapy, but only 3%–5% of patients survive longer than 3 years before the disease recurs.²

Although genetic studies have revealed many potential targets in the disease, the pathways involved are complex and diverse, and many GBM tumors have multiple mutations.^{3,4} That has made it difficult to develop agents that can block tumor progression because inhibiting any one pathway has little impact.

Patrik Ernfors and colleagues at Karolinska started with the assumption that GBM cells might accumulate genetic alterations and gain functions not necessarily involved in cancer that could affect cellular processes.

Ernfors is head of office at the Laboratory of Molecular Neurobiology and a professor of tissue biology and molecular and developmental biology at Karolinska.

The team set up a phenotypic screen to find compounds that would affect visible morphological changes in two patient-derived glioblastoma cell lines but not in mouse embryonic stem cells or human fibroblasts.

The initial screen against a 1,364-compound library yielded 63 compounds, which were confirmed as active in 7 other patient-derived glioblastoma cell lines. A panel of cell-based assays—including cytotoxicity, cell viability and apoptosis—combined with *in silico* ADME analyses reduced the list to 17 hits.

Using zebrafish embryo toxicity and cardiotoxicity screens, the team identified three compounds that lacked toxicity and also decreased tumor size compared with vehicle in a zebrafish model of GBM.

The top hit, containing a quinoline-alcohol scaffold, was named vacquinol-1 and showed an IC_{50} value of 2.36 μ M in cell viability assays. Vacquinol-1 was about 60-fold more potent than the glioma drug temozolomide, which had an IC_{50} value of 139 μ M.

Cancer Research Technology Ltd. and **Merck & Co. Inc.**'s Temodal temozolomide and **Reliance Life Sciences'** TemoRel temozolomide are marketed to treat brain cancer.

Vacquinol-1 was ineffective on bladder, prostate, breast and neuroblastoma cancer cell lines, suggesting that the compound might work by targeting cellular vulnerability found in the glioblastoma cell lines.

Next, the researchers looked for vacquinol-1's mechanism of action.

In live cell imaging, vacquinol-1-treated glioblastoma cells showed dose-dependent cell rounding, membrane ruffle formation and rupture of the plasma membrane, which led to necrotic-like cell death. The cell rupture was attributed to the production of large vacuoles induced by massive macropinocytosis.

The team then performed an unbiased shRNA screen with vacquinol-1 in glioblastoma cells and found that those expressing *mitogen-activated protein kinase kinase 4* (*MAP2K4*; *MKK4*) shRNA were more resistant than cells expressing other shRNAs. The researchers concluded that *MKK4* acts as a critical node in vacquinol-1-induced catastrophic vacuolization.

Finally, the team used a mouse model of GBM to evaluate the efficacy of vacquinol-1 on tumors that had been allowed to develop for six or seven weeks.

Despite the advanced stage of cancer, vacquinol-1 administered intracranially or orally abolished tumors or decreased tumor size without affecting normal brain tissue. The mouse glioblastoma cells showed necrosis and accumulation of macropinocytic vacuoles similar to those seen *in vitro*. By contrast, vehicle-treated mice had large tumors, with brain tissue showing necrosis and hemorrhage.

Vacquinol-1 prolonged survival in GBM mice that received the oral compound once daily for five days starting two weeks after engraftment. In the treated mice, 6 out of 8 survived for at least 80

days, whereas all the vehicle-treated mice died and had a median survival of 31.5 days.

Results were published in *Cell*.

The team's next steps are to identify a dosing regimen and perform toxicity studies to move into clinical trials.

Opening the therapeutic window

Ernfors told *SciBX* that he believes vacquinol-1 would be a good addition to the GBM armamentarium because the compound can act on cells with different proliferation rates, whereas temozolomide and radiotherapy act primarily on rapidly dividing cells.

"The compound's macropinocytosis action works independently of cell proliferation and should also be able to target relatively quiescent stem cell-like, tumor-initiating cells or drug-resistant GBM," he said.

However, Paul Dent, a professor of biochemistry and molecular biology at **Virginia Commonwealth University**, thinks much more data on preclinical toxicology and tissue distribution need to be obtained to support an IND filing.

Dent's laboratory is working on *IL-24* (*MDA7*) as a gene therapy to sensitize malignant glioma to ionizing radiation.

"Ideally, a more efficacious form of the agent needs to be developed. It would then be a logical next step to perform Phase I studies in recurrent GBM patients," he said. He noted that the team has developed an agent with an IC_{50} value of about 0.39 μ M, but they report that unrelated

"The compound's macropinocytosis action works independently of cell proliferation and should also be able to target relatively quiescent stem cell-like, tumor-initiating cells or drug-resistant GBM."

—Patrik Ernfors,
Karolinska Institute

toxicity begins at 15 μ M. That therapeutic window might not be enough for translation to the clinic.

Ernfors' team is looking for better compounds. "Using chemical synthesis and SAR studies, we were able to show that vacquinol-1 is critically dependent on a conserved 4-(piperidin-2-ylmethanol)-quinoline scaffold and produced a series of structural analogs with increased potency," he said.

"We are interested in optimizing the compounds and also pursuing related compounds that modulate other targets within the vacuolization pathway," he added.

Dent wanted to see more details in the mechanisms underlying the *MKK4*-induced catastrophic vacuolization. "The identification of *MKK4* can only be considered a preliminary step in developing an understanding of the mechanism of agent action," he said. "Thus, an unbiased approach examining multiple signaling pathways at the level of activity—phosphorylation and acetylation—will be required to define key target pathways."

Kevin Roth, a professor and chair of pathology at The University of Alabama at Birmingham School of Medicine, thought that inducing GBM macropinocytosis and cell death by targeting *MKK4* might be complicated by the fact that

MKK4 has been reported to have both pro- and antitumorigenic properties. Other targets of macropinocytosis should be investigated.

Roth's laboratory research focuses on the regulation of neuronal apoptosis and development of autophagy-targeted therapies for brain cancers.

"Macropinocytosis is a topic of great interest recently," said Roth. "Normal cellular processes such as macropinocytosis or autophagy can promote the tumor cells' survival or progression but at the same time make the tumor cells vulnerable to agents that are relatively harmless to normal cells," he said.

Previous studies have shown examples of chemical agents that induce neuronal or cancer cell death through stimulation of macropinocytosis.⁵⁻⁸ The nonapoptotic cell death pathway described in those papers "appears similar if not identical to what is described by the Karolinska team," noted Roth.

"The identification of *MKK4* can only be considered a preliminary step in developing an understanding of the mechanism of agent action. Thus, an unbiased approach examining multiple signaling pathways at the level of activity—phosphorylation and acetylation—will be required to define key target pathways."

—Paul Dent,
Virginia Commonwealth University

"To more fully understand the mechanism of action of vacquinol-1, Ernfor's team might want to interrogate the entire macrocytic pathway, from endosomal maturation through lysosome fusion and degradation," said Roth.

According to Dent, translation to the clinic might require finding an effective combination of this compound with an established anticancer drug.

"Empiric combinations of established drugs with this agent could possibly yield interesting results," Dent told *SciBX*. "From a translational perspective, it will be important to determine the effect of the agent in combination with standard-of-care drugs in GBM such as BiCNU [1,3-bis(chloroethyl)-1-nitrosourea] and temozolomide, as well as with ionizing radiation."

BiCNU carmustine is marketed by **Bristol-Myers Squibb Co.** and **Emcure Pharmaceuticals Ltd.** for brain cancer, Hodgkin's disease, non-Hodgkin's lymphoma (NHL) and multiple myeloma (MM).

"Only by fully understanding the mechanisms of action of vacquinol-1 can progress be made to permit a more rational combination with other clinically relevant agents," said Dent.

A patent application has been filed in the U.S. and Sweden and is available for licensing from **Karolinska Institute Innovations AB**.

Baas, T. *SciBX* 7(14); doi:10.1038/scibx.2014.391
Published online April 10, 2014

REFERENCES

1. Kitambi, S.S. *et al. Cell*; published online March 18, 2014; doi:10.1016/j.cell.2014.02.021
Contact: Patrik Ernfor, Karolinska Institute, Stockholm, Sweden
e-mail: patrik.ernfors@ki.se
2. Dolecek, T.A. *et al. Neuro-oncol.* **14**, v1-v49 (2012)
3. Dent, P. *et al. Cancer Biol. Ther.* **7**, 1335-1340 (2008)
4. Polivka, J. Jr. *et al. Anticancer Res.* **32**, 2935-2946 (2012)
5. Kaul, A. *et al. Cell Signal.* **19**, 1034-1043 (2007)
6. Overmeyer, J.H. *et al. Mol. Cancer Res.* **6**, 965-977 (2008)
7. Robinson, M.W. *et al. J. Med. Chem.* **55**, 1940-1956 (2012)
8. Trabbic, C.J. *et al. ACS Med. Chem. Lett.* **5**, 73-77 (2014)

COMPANIES AND INSTITUTIONS MENTIONED

Bristol-Myers Squibb Co. (NYSE:BMJ), New York, N.Y.
Cancer Research Technology Ltd., London, U.K.
Emcure Pharmaceuticals Ltd., Pune, India
Karolinska Institute, Stockholm, Sweden
Karolinska Institute Innovations AB, Stockholm, Sweden
Merck & Co. Inc. (NYSE:MRK), Whitehouse Station, N.J.
Reliance Life Sciences, Rabale, India
The University of Alabama at Birmingham School of Medicine, Birmingham, Ala.
Virginia Commonwealth University, Richmond, Va.

Toxic assets

By C. Simone Fishburn, Senior Editor

A new tool for *in vivo* detection of liver toxicity could represent a significant improvement over routine methods that only work *in vitro* or on tissue slices.¹ The litmus test for the **Stanford University School of Medicine** inventors will be to show that the nanoparticle-based method can detect toxicity in compounds that previously eluded standard analysis and later failed in the clinic.

Current preclinical toxicity testing methods are clearly suboptimal, as more than 20% of compounds end up failing clinical trials because of toxicity issues.²

Standard toxicology analysis involves early stage screening using *in vitro* microsomes, hepatocytes and liver slices followed by IND-enabling histopathology and plasma biomarker studies on animals treated for two weeks or longer.

IND submissions also require additional assessments such as genotoxicity and immunotoxicity.

Multiple tools have been tried in discovery research, but few have been incorporated into routine preclinical toxicology assessment.

Michael Taylor, founder and principal of **NonClinical Safety Assessment**, told *SciBX* that the field often is resistant to disruptive innovation. Companies prefer to focus resources on techniques currently accepted by the FDA, and toxicologists want to see a validated improvement over existing methods before switching to a new one.

“Toxicology is in general a very risk-averse field, so it’s a tough nut to crack with this space—even for new tools that are exciting,” he said.

Attitudes aside, it is very difficult to monitor short-lived metabolites *in vivo*; thus, toxicity analysis relies on detecting biomarkers in isolated plasma or tissue samples.

Jianghong Rao, an associate professor of radiology and chemistry at the Stanford University School of Medicine, encountered that very problem about three years ago when he was studying markers related to inflammation.

He—like other researchers—was trying to find short-lived markers of toxicity. “The big challenge was that you could only apply reagents locally or in cell culture, but you couldn’t apply them systemically to see inflammatory sites *in vivo*,” he told *SciBX*.

As luck would have it, others in his lab had developed a technology based on semiconducting polymer nanoparticles (SPNs) for deep-tissue imaging of tumors. Those particles could detect signals with high sensitivity and had good *in vivo* stability and distribution properties.

That led the team, which included researchers from the **University of Toronto**, to adapt the technology to detect—in live animals—short-lived biomarkers that are inaccessible to standard techniques.

Interactive sensors

Because drug-induced toxicity occurs primarily in the liver, Rao’s team focused on reactive metabolites formed as transient by-products of hepatic clearance enzymatic reactions.

Reactive oxygen species (ROS) are generated by oxidative enzymes such as cytochrome P450 (p450) or by reactions of drug metabolites with oxygen. Reactive nitrogen species (RNS) are the result of drug

metabolite-induced disruption of the electron transport chain, which leads to mitochondrial toxicity.

ROS and RNS thus represent two potential biomarkers of toxicity produced by different mechanisms that have different biological effects.

To enable real-time simultaneous monitoring of hepatic ROS and RNS in live rats, the team designed SPNs containing two matrix polymers, two sensors and a liver-targeting, galactose-conjugated polyethylene glycol (PEG) tail.

The ROS sensor in the matrix is a chemiluminescent substrate that breaks down in the presence of the ROS hydrogen peroxide to produce a reaction intermediary that excites nearby dye molecules to emit light.

Via chemiluminescence resonance energy transfer (CRET), the light emitted serves as an excitation source for the fluorescent RNS sensor.

When no RNS is present, the fluorescent sensor embedded in the matrix responds to the excitation source and emits light at wavelengths of 680 nm and 820 nm via fluorescence resonance energy transfer (FRET). However, when the RNS peroxynitrite is present, the sensor dye decomposes and the FRET is lost—along with the 820 nm emission. The 680 nm emission increases as a consequence.

Thus, ROS measurement involves direct quantitation of the luminescence signal, and RNS measurement is based on alterations in the ratio of the 680 nm and 820 nm emissions.

To test the nanoparticles, the researchers gave mice subtoxic to toxic doses of acetaminophen. Overdoses of that drug cause necrotic cell death through overproduction of ROS and RNS.

The team captured sequential fluorescence and luminescence images for 80 minutes after dosing. ROS and RNS were detected 18 minutes and 53 minutes after a toxic 300 mg/kg acetaminophen dose. Only baseline signals were seen at all the lower doses.

That threshold type of toxicity—in which toxicity only occurs above a specific dose level—is characteristic of acetaminophen and differs from dose-dependent toxicity in which increasing levels of toxic products are formed as the dose increases.

The nanoparticle method produced evidence of toxicity earlier than immunohistochemical and histological analysis of liver tissue. Those methods showed no changes at 45 minutes and only much later—at 180 minutes—showed signs of necrosis.

Glutathione, a scavenger of reactive metabolites, reversed the signals produced by the acetaminophen-generated ROS and RNS. That finding suggested that the technique also could be used to assess agents that help reverse or recover from toxicity.

Finally, the researchers tested whether the SPNs could detect ROS and RNS produced by isoniazid, a widely used tuberculosis drug that causes significant hepatotoxicity by poorly understood mechanisms.

Because the kinetics and sequence of ROS and RNS generation depend on which metabolic pathway is involved, the group expected that a time-course study would provide clues to the mechanism of toxicity.

As with acetaminophen, the SPNs detected both hydrogen peroxide and peroxynitrite from isoniazid. Unlike acetaminophen, the two toxicity readouts produced very different time profiles. The results suggested that isoniazid toxicity occurs mainly through a nitrosylation pathway, whereas acetaminophen metabolism involves oxidation and nitrosylation.

The kinetics also showed that isoniazid toxicity is dose dependent—in contrast to the threshold type of toxicity seen for acetaminophen.

The authors concluded that the SPNs can be used to help understand mechanisms of toxicity and that the technology can be used for *in vivo* monitoring and early detection of potentially toxic preclinical candidate compounds.

Results were published in *Nature Biotechnology*.

Translational toxicology

Rao told *SciBX* that the team is testing the technology on a range of molecules in animals. He also plans to develop it for clinical use, in particular for monitoring liver damage in patients waiting to receive transplants.

According to Jonathan Sorger, the technology is a big step toward solving the problem of finding biomarkers for liver imaging. “Everyone is trying to come up with new biomarkers: early, predictive and noninvasive are the key properties to have,” he said.

Sorger said that the ability to take serial measurements in intact animals is a key advantage of the new method.

“I’m not sure if this will be the way to do it in the end, but it’s a better way than what we’re doing now,” he said. He added that in drug screening, the SPN technology would most likely be used after standard *in vitro* tests with microsomes and hepatocytes.

Sorger is director of medical research at **Intuitive Surgical Inc.**, a medical device company with imaging technologies for noninvasive surgery.

Alexandra de Lille agreed that the noninvasive aspect of the new technique is one of its biggest advantages over current methods as it can detect toxicity biomarkers and perform kinetic assessments without “slicing and dicing of tissues.”

She said that in addition to providing mechanistic clues underlying a toxic readout—such as whether nitrosylation or oxidation pathways are dominant—the information from kinetic monitoring can translate to dosing decisions.

de Lille is director of technical applications for *in vivo* imaging at Caliper Life Sciences, a unit of **PerkinElmer Inc.**

She also liked that the technology can monitor multiple toxic metabolites in parallel.

“It’s very much in the pioneer stage to use multiple-reporter systems in the same animal—you get two outcomes from one mouse. Technically we could probably get five readings out of one mouse with a system like this. In the past it has been one reading per mouse,” she told *SciBX*.

According to Sorger, the signal intensity used in the study

works well in mice but would not be sufficient for use in humans.

“This makes a lot of sense in the drug screening world, but in humans it will be hard to get the necessary tissue penetration,” he said. “In humans, photons [in this wavelength range] don’t penetrate tissue far

enough. You can go to 1 cm depth, but the liver is deeper than that. You would need to use a different emitter or a radioisotope.”

de Ville agreed that use in humans is further in the future.

Taylor said that the method still has challenges to meet before it is used in animals because a more sophisticated readout does not necessarily translate to better predictive power.

He said that the team compared its method with standard histopathology but did not compare it with plasma biomarkers used in routine toxicology assessments.

The standard to beat, according to Taylor, is biomarkers of hepatotoxicity such as transaminase and superoxide dismutase (SOD).

He said that one way to show superiority would be to draw plasma and look at transaminase and SOD in addition to histopathology to see whether the Stanford group’s method is more sensitive.

“Sometimes there’s a disconnect between biomarkers such as transaminase and SOD and the visual histopathological tissue markers, so if they could show the technique can be a better predictor of tissue damage, that would be an advantage,” said Taylor.

Another meaningful benefit, he said, would be getting reliable information faster than current methods.

“A two-week full rat toxicology study can approach \$100,000. So if you can get a signal sooner than that, there would be significant savings,” he said.

Those savings involve not only time-related costs of the study but also the amount of material that needs to be synthesized, which is one of the most costly and rate-limiting aspects of preclinical development, he said. “This is one of the first big financial jumps for a small company,” he added.

Taylor said that the real test will be to take compounds that were dropped in the clinic because of toxicity in humans and see if the new method can produce a preclinical toxicity signal where the current methods failed.

“To be a game changer, it needs to increase the chances of detecting toxicity better than other methods,” he said.

Stanford University has filed a patent on the findings. The technology is available for licensing.

Fishburn, C.S. *SciBX* 7(14); doi:10.1038/scibx.2014.392
Published online April 10, 2014

REFERENCES

- Shuhendler, A.J. *et al. Nat. Biotechnol.*; published online March 23, 2014; doi:10.1038/nbt.2838
Contact: Jianghong Rao, Stanford University School of Medicine, Stanford, Calif.
e-mail: jrao@stanford.edu
- Kola, I. & Landis, J. *Nat. Rev. Drug Discov.* 3, 711–715 (2004)

COMPANIES AND INSTITUTIONS

Intuitive Surgical Inc. (NASDAQ:ISRG) Sunnyvale, Calif.
NonClinical Safety Assessment, Mountain View, Calif.
PerkinElmer Inc. (NYSE:PKI), Waltham, Mass.
Stanford University, Stanford, Calif.
Stanford University School of Medicine, Stanford, Calif.
University of Toronto, Toronto, Ontario, Canada

“It’s very much in the pioneer stage to use multiple-reporter systems in the same animal—you get two outcomes from one mouse. Technically we could probably get five readings out of one mouse with a system like this. In the past it has been one reading per mouse.”

**—Alexandra de Lille,
PerkinElmer Inc.**

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
Cancer				
Brain cancer	Epithelial membrane protein 2 (EMP2)	Patient sample and mouse studies suggest targeting EMP2 could be used to treat glioblastoma multiforme (GBM). In brain tumor samples, EMP2 expression was higher than that in healthy brain tissue, and elevated EMP2 expression correlated with poor survival. In two heterotopic xenograft mouse models of GBM, an antibody fragment or IgG targeting EMP2 led to decreased tumor growth compared with fragment or IgG controls. Next steps include testing the EMP2-targeting antibody fragment for brain penetration in intracranial mouse models of GBM (see <i>Pre-EMP-tive strike against GBM</i> , page 1).	Patented; licensed to University of California, Los Angeles start-up company Paganini Biopharma Inc.	Qin, Y. <i>et al. J. Biol. Chem.</i> ; published online March 18, 2014; doi:10.1074/jbc.M113.543728 Contact: Madhuri Wadehra, University of California, Los Angeles David Geffen School of Medicine, Los Angeles, Calif. e-mail: mwadehra@mednet.ucla.edu
SciBX 7(14); doi:10.1038/scibx.2014.393 Published online April 10, 2014				
Cancer	Anoctamin 1 calcium activated chloride channel (ANO1)	<i>In vitro</i> studies suggest compounds that promote ANO1 degradation, as opposed to ANO1 antagonists, could be useful for treating cancer. ANO1 is a critical survival factor for cancer cells. In ANO1-amplified human cancer cell lines, the ANO1 inhibitor CaCC _{inh} -A01 decreased proliferation compared with two other ANO1 inhibitors, a result that suggested functions other than the conductance of ions across ANO1 can affect tumor survival. <i>In vitro</i> studies showed that CaCC _{inh} -A01 decreased ANO1 protein levels by promoting endoplasmic reticulum-associated proteasomal degradation of the protein. Next steps could include developing assays to screen for compounds that promote ANO1 degradation. CaCC _{inh} -A01 is a research compound.	Patent and licensing status unavailable	Bill, A. <i>et al. J. Biol. Chem.</i> ; published online March 5, 2014; doi:10.1074/jbc.M114.549188 Contact: L. Alex Gaither, Novartis Institutes for BioMedical Research, Cambridge, Mass. e-mail: alex.gaither@novartis.com
SciBX 7(14); doi:10.1038/scibx.2014.394 Published online April 10, 2014				
Chronic lymphocytic leukemia (CLL)	Not applicable	<i>In vitro</i> and mouse studies suggest the rheumatoid arthritis (RA) drug Ridaura auranofin could be used to treat CLL. In cell culture, Ridaura induced apoptosis in a series of human CLL cells with different genomic alterations but did not induce apoptosis in normal CD34 ⁺ hematopoietic stem cells. In a transgenic mouse model of CLL, Ridaura administration over 2 weeks decreased CLL cell counts by >90% and increased survival compared with no treatment. Next steps include testing Ridaura in combination with other drugs for inhibition of CLL and mantle cell lymphoma (MCL) in mouse models and human samples. The Leukemia & Lymphoma Society, Kansas Bioscience Authority and the National Center for Advancing Translational Sciences Therapeutics for Rare and Neglected Diseases program have Ridaura in Phase I/II trials to treat CLL. Nestle S.A.'s Prometheus Laboratories Inc. unit markets Ridaura to treat RA.	Unpatented; licensing status not applicable	Fiskus, W. <i>et al. Cancer Res.</i> ; published online March 5, 2014; doi:10.1158/0008-5472.CAN-13-2033 Contact: Kapil N. Bhalla, Houston Methodist Research Institute, Houston, Texas e-mail: knbhalla@tmhs.org
SciBX 7(14); doi:10.1038/scibx.2014.395 Published online April 10, 2014				

This week in therapeutics (continued)

Indication	Target/marker/pathway	Summary	Licensing status	Publication and contact information
Colorectal cancer	Splice variant v6 of CD44 (CD44v6); phosphoinositide 3-kinase (PI3K)	<i>In vitro</i> and mouse studies suggest inhibiting CD44v6 and PI3K could be useful for preventing metastasis in colorectal cancer. In mice implanted with patient-isolated colorectal cancer stem cells (CR-CSCs), shRNA against <i>CD44v6</i> inhibited metastasis, whereas control shRNA did not. In CR-CSCs, expression of <i>CD44v6</i> was associated with upregulated PI3K signaling. In mice implanted with human CR-CSCs, PI3K inhibition decreased metastatic potential compared with vehicle treatment. Next steps could include evaluating small molecule PI3K inhibitors in models of metastatic colorectal cancer. SciBX 7(14); doi:10.1038/scibx.2014.396 Published online April 10, 2014	Patent and licensing status unavailable	Todaro, M. <i>et al. Cell Stem Cell</i> ; published online March 6, 2014; doi:10.1016/j.stem.2014.01.009 Contact: Giorgio Stassi, University of Palermo, Palermo, Italy e-mail: giorgio.stassi@unipa.it Contact: Ruggero De Maria, Elena National Cancer Institute, Rome, Italy e-mail: demaria@ifo.it
Lymphoma	Jagged 1 (JAG1); notch 2 (NOTCH2); fibroblast growth factor 4 (FGF4); fibroblast growth factor receptor 1 (FGFR1; CD331)	Cell culture and mouse studies suggest inhibiting JAG1-NOTCH2 or FGF4-FGFR1 signaling could help treat Burkitt lymphoma. In cocultures of human endothelial cells and Burkitt lymphoma cells from patients or mouse models, paracrine signaling between endothelial JAG1 and lymphoma NOTCH2, as well as between lymphoma FGF4 and endothelial FGFR1, induced a feed-forward loop that increased lymphoma cell growth, invasiveness and chemotherapeutic resistance compared with what was seen in control cultures. In mouse models of Burkitt lymphoma, <i>Jag1</i> ⁻ or <i>Fgfr1</i> ⁻ mice or <i>Notch2</i> ⁻ tumors had less tumor growth and greater survival and tumor sensitivity to chemotherapy than nondeficient controls. Ongoing work includes identifying other endothelial cell proteins that drive lymphoma tumor growth. OncoMed Pharmaceuticals Inc. has OMP-59R5 (Anti-Notch2/3), a huCAL mAb that binds NOTCH2 and notch 3 (NOTCH3), in Phase I/II trials to treat pancreatic and small cell lung cancers and Phase I testing to treat solid tumors. SciBX 7(14); doi:10.1038/scibx.2014.397 Published online April 10, 2014	Unpatented; unlicensed	Cao, Z. <i>et al. Cancer Cell</i> ; published online March 17, 2014; doi:10.1016/j.ccr.2014.02.005 Contact: Shahin Rafii, Weill Cornell Medical College, New York, N.Y. e-mail: srafi@med.cornell.edu Contact: Bi-Sen Ding, same affiliation as above e-mail: bid2004@med.cornell.edu
Non-small cell lung cancer (NSCLC); bone cancer	Ubiquitin specific peptidase 1 (USP1); WD repeat domain 48 (WDR48; UAF1)	Cell-based studies suggest inhibiting the USP1-UAF1 complex could help increase NSCLC and bone cancer sensitivity to platinum-based chemotherapies. In a cisplatin-resistant NSCLC cell line, a USP1-UAF1 inhibitor combined with cisplatin decreased cell proliferation compared with either of the agents alone or vehicle control. In a human osteosarcoma cell line, the inhibitor plus cisplatin decreased proliferation compared with cisplatin alone. Next steps include testing the inhibitor in mouse models of cancer. SciBX 7(14); doi:10.1038/scibx.2014.398 Published online April 10, 2014	Patent filed by the NIH and University of Delaware; available for licensing	Liang, Q. <i>et al. Nat. Chem. Biol.</i> ; published online Feb. 16, 2014; doi:10.1038/nchembio.1455 Contact: Zhihao Zhuang, University of Delaware, Newark, Del. e-mail: zzhuang@udel.edu Contact: David J. Maloney, National Institutes of Health, Bethesda, Md. e-mail: maloneyd@mail.nih.gov
Prostate cancer	Sterol O-acyltransferase 1 (SOAT1; ACAT1)	Studies in patient samples, mice and cell culture suggest inhibiting ACAT1 prevents cholesteryl ester (CE) synthesis and could help treat prostate cancer. In patient tissue samples, CE-filled lipid droplets were more abundant in high-grade and metastatic prostate cancers than other cancers or matched healthy tissues. In prostate cancer cell lines, pharmacological inhibition of ACAT1 increased cell cycle arrest and apoptosis and decreased cell migration and invasiveness compared with vehicle treatment. In a mouse xenograft model of metastatic prostate cancer, pharmacological inhibition of ACAT1 decreased tumor weight compared with treatment using vehicle or inhibitors of other lipid-synthesizing enzymes. Next steps include developing an injectable formulation of an ACAT1 inhibitor for intratumoral delivery. Atterocor Inc. has an inhibitor of ACAT1 in Phase I testing to treat adenocarcinoma. SciBX 7(14); doi:10.1038/scibx.2014.399 Published online April 10, 2014	Patent application filed; available for licensing	Yue, S. <i>et al. Cell Metab.</i> ; published online March 4, 2014; doi:10.1016/j.cmet.2014.01.019 Contact: Ji-Xin Cheng, Purdue University, West Lafayette, Ind. e-mail: jcheng@purdue.edu

This week in therapeutics (continued)

Indication	Target/marker/ pathway	Summary	Licensing status	Publication and contact information
Cardiovascular disease				
Atherosclerosis	Peroxisome proliferation-activated receptor- γ (PPARG; PPAR γ); myeloid-lymphoma or mixed-lineage 5 (MLL5); 5'-3' exoribonuclease 2 (XRN2)	<i>In vitro</i> and mouse studies suggest activating genes involved in the plasma cholesterol-lowering response could help treat atherosclerosis. In a mouse model of atherosclerosis with elevated low-density lipoprotein levels, blocking hepatic synthesis of lipoproteins to lower plasma cholesterol induced regression of plaques and stimulated different gene expression networks controlled by PPAR γ in early stages of plaque expansion and by MLL5 and XRN2 in later stages. In an <i>in vitro</i> model of atherosclerosis, knockdown of these individual regulatory transcription factors increased cholesterol esterase accumulation by 12%–21%. Next steps include validating the role of the regulatory genes in plaque formation. SciBX 7(14); doi:10.1038/scibx.2014.400 Published online April 10, 2014	Findings unpatented; licensing status not applicable	Björkegren, J.L.M. <i>et al. PLoS Genet.</i> ; published online Feb. 27, 2014; doi:10.1371/journal.pgen.1004201 Contact: Josefin Skogsberg, Karolinska Institute, Stockholm, Sweden e-mail: josefin.skogsberg@ki.se
Blood clots; thrombosis	Thrombin (factor IIa; F2)	<i>In vitro</i> studies suggest an allosteric thrombin inhibitor could prevent blood clots and thrombosis. Chemical synthesis and <i>in vitro</i> testing identified a lead compound that bound the allosteric exosite 2 on thrombin and inhibited its activity at a low nanomolar IC ₅₀ value. In a human plasma-based anticoagulation assay, the compound increased time to clot formation compared with vehicle. In platelet-rich human plasma, the compound decreased thrombin-induced platelet aggregation compared with no treatment. Ongoing work includes testing the compound in mouse models of venous thrombosis. SciBX 7(14); doi:10.1038/scibx.2014.401 Published online April 10, 2014	Patent application filed; available for licensing	Mehta, A.Y. <i>et al. J. Med. Chem.</i> ; published online March 17, 2014; doi:10.1021/jm4020026 Contact: Umesh R. Desai, Virginia Commonwealth University, Richmond, Va. e-mail: urdesai@vcu.edu
Endocrine/metabolic disease				
Obesity	Farnesoid X receptor (FXR; NRIH4)	Mouse studies suggest FXR is required to mediate the effects of vertical sleeve gastrectomy—a type of bariatric surgery—on obesity. In wild-type, obese mice that received vertical sleeve gastrectomy, body fat and food intake were lower at 11 weeks after surgery than those in sham-surgery controls. However, in <i>Fxr</i> ^{-/-} , obese mice receiving the surgery, food intake increased beginning 4 weeks after surgery and there was no loss of body fat 11 weeks after surgery. Ongoing work includes elucidating how FXR signaling regulates food intake and induces weight loss after vertical sleeve gastrectomy. SciBX 7(14); doi:10.1038/scibx.2014.402 Published online April 10, 2014	Unpatented; unlicensed	Ryan, K.K. <i>et al. Nature</i> ; published online March 26, 2014; doi:10.1038/nature13135 Contact: Randy J. Seeley, University of Cincinnati, Cincinnati, Ohio e-mail: randy.seeley@uc.edu
Hepatic disease				
Cirrhosis	Synoviolin (SYVN1; HRD1)	Studies in human samples and mice suggest inhibiting HRD1 could help treat liver cirrhosis. In tissue samples from patients with cirrhosis and in a mouse model of liver cirrhosis, levels of x-box binding protein 1 (XBP1) and HRD1, components of the unfolded protein response, were higher and levels of nuclear factor (erythroid-derived 2)-like 2 (NFE2L2; NRF2) were lower than those in samples from healthy controls. In the mouse model, an HRD1 inhibitor decreased cirrhosis compared with no treatment. Next steps include improving the potency of the HRD1 inhibitor. SciBX 7(14); doi:10.1038/scibx.2014.403 Published online April 10, 2014	Patented; licensing status undisclosed	Wu, T. <i>et al. Genes Dev.</i> ; published online March 17, 2014; doi:10.1101/gad.238246.114 Contact: Donna D. Zhang, The University of Arizona, Tucson, Ariz. e-mail: dzhang@pharmacy.arizona.edu Contact: Deyu Fang, Northwestern University Feinberg School of Medicine, Chicago, Ill. e-mail: fangd@northwestern.edu

This week in therapeutics (continued)

Indication	Target/marker/ pathway	Summary	Licensing status	Publication and contact information
Infectious disease				
Bacterial infection	Pannexin 1 (PANX1)	<i>In vitro</i> studies suggest avoiding PANX1 inhibition could prevent the toxic side effects of some quinolone antibiotics. Pfizer Inc.'s quinolone antibiotic Trovan trovafloxacin showed potent antibacterial effects by targeting bacterial topoisomerases but was linked to severe toxicity and patient deaths. Cell-based assays identified Trovan as an inhibitor of the transmembrane channel PANX1. In cultured cells undergoing apoptosis, quinolones lacking a fluorinated ring at position N1, such as ciprofloxacin or levofloxacin, did not inhibit small molecule uptake by PANX1, whereas quinolones containing a fluorinated ring at that position, such as Trovan, difloxacin or tosufloxacin, inhibited or partially inhibited small molecule uptake by PANX1. Next steps could include conducting studies to identify specific PANX1-quinolone interactions and determining underlying mechanisms of toxicity. Ciprofloxacin and levofloxacin are generic antibiotics. Difloxacin and tosufloxacin are commercially available in some Asian countries to treat bacterial infections.	Patent application filed; available for licensing	Poon, I.K.H. <i>et al. Nature</i> ; published online March 12, 2014; doi:10.1038/nature13147 Contact: Kodi S. Ravichandran, University of Virginia, Charlottesville, Va. e-mail: ravi@virginia.edu
SciBX 7(14); doi:10.1038/scibx.2014.404 Published online April 10, 2014				
Malaria	Peroxisome proliferation-activated receptor- γ (PPARG; PPAR γ)	Studies in mice and human samples suggest combining PPAR γ agonists with antimalarial therapy could improve outcomes in cerebral malaria. In a mouse model of cerebral malaria, antimalarial drugs plus the PPAR γ agonist Avandia rosiglitazone increased brain levels of neuroprotectants including brain-derived neurotrophic factor (Bdnf) compared with the antimalarial alone. In this mouse model, the combination improved survival and prevented infection-induced cognitive impairments and brain atrophy. In patients who have uncomplicated <i>Plasmodium falciparum</i> infection, the antimalarial regimen plus rosiglitazone decreased inflammatory markers and increased BDNF in plasma compared with antimalarial drugs alone. Next steps include clinical testing in patients with cerebral malaria. GlaxoSmithKline plc markets Avandia rosiglitazone to treat diabetes.	Findings unpatented; available for partnering	Serghides, L. <i>et al. PLoS Pathog.</i> ; published online March 6, 2014; doi:10.1371/journal.ppat.1003980 Contact: Lena Serghides, University Health Network, Toronto, Ontario, Canada e-mail: lena.serghides@utoronto.ca
SciBX 7(14); doi:10.1038/scibx.2014.405 Published online April 10, 2014				
Meningitis; pneumonia	Properdin	Mouse studies suggest recombinant properdin could help protect against bacterial meningitis and pneumonia. In mice, intraperitoneal injection of recombinant properdin prior to lethal <i>Neisseria meningitidis</i> challenge resulted in 90% survival, whereas saline led to 0% survival. In mice, injection of recombinant properdin at time of <i>Streptococcus pneumoniae</i> infection decreased the development of disease compared with injection of saline. Next steps include evaluating the recombinant properdin in additional models of bacterial and parasitic infections and improving the activity and homogeneity of the manufactured protein (see Positioning properdin , page 6).	Patent application cofiled by University of Leicester and The Medicines Co.; available for licensing	Ali, Y.M. <i>et al. Proc. Natl. Acad. Sci. USA</i> ; published online March 24, 2014; doi:10.1073/pnas.1401011111 Contact: Wilhelm J. Schwaeble, University of Leicester, Leicester, U.K. e-mail: ws5@le.ac.uk
SciBX 7(14); doi:10.1038/scibx.2014.406 Published online April 10, 2014				

This week in therapeutics (continued)

Indication	Target/marker/ pathway	Summary	Licensing status	Publication and contact information
Inflammation				
Inflammation	Leukotriene A4 hydrolase (LTA4H)	Structure-based design studies led to an LTA4H inhibitor that could be useful as a lead for new anti-inflammatory agents. LTA4H is a dual-acting enzyme that catalyzes the formation of proinflammatory leukotriene B4 (LTB4) and hydrolyzes the chemotactic peptide Pro-Gly-Pro. Mass spectrometry and X-ray structural analyses of LTA4H bound to a tripeptide analog revealed the mechanism for peptide hydrolysis. The structure aided the rational design of an LTA4H inhibitor that decreased LTB4 catalysis in cultured human neutrophils with an IC ₅₀ value of about 0.5 μM without affecting Pro-Gly-Pro hydrolysis. Next steps include medicinal chemistry studies to optimize the new inhibitor. Celtaxsys Inc. has the LTA4H inhibitor CTX-4430 in Phase I testing to treat cystic fibrosis (CF). <i>SciBX</i> 7(14); doi:10.1038/scibx.2014.407 Published online April 10, 2014	Patent and licensing status unavailable	Stsiapanava, A. <i>et al. Proc. Natl. Acad. Sci. USA</i> ; published online March 3, 2014; doi:10.1073/pnas.1402136111 Contact: Jesper Z. Haeggström, Karolinska Institute, Stockholm, Sweden e-mail: jesper.haeggstrom@ki.se Contact: Bengt Samuelsson, same affiliation as above e-mail: bengt.samuelsson@ki.se



The Scientific Acumen of Nature Publishing Group
plus
The Business Intelligence of BioCentury Publications, Inc.
in a single publication

Can you afford not to subscribe?
Visit **scibx.com** for details on how to subscribe to *SciBX*

BioCentury

nature publishing group 

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
Assays & screens			
Nanoparticles for <i>in vivo</i> detection of drug-induced hepatotoxicity	<p>Mouse studies suggest semiconducting polymer nanoparticles (SPNs) could be used for real-time <i>in vivo</i> monitoring of drug-induced hepatotoxicity. SPNs were generated containing a liver-targeting galactose residue, a chemiluminescent substrate and a fluorescent sensor that produces signals when detecting reactive oxygen species (ROS) and reactive nitrogen species (RNS) simultaneously. In mice, the SPNs detected ROS and RNS induced by toxic doses—but not by subtoxic doses—of acetaminophen or isoniazid earlier than histopathological changes occurred. Next steps include testing toxicity of other small molecules and virus particles (<i>see Toxic assests, page 8</i>).</p> <p>SciBX 7(14); doi:10.1038/scibx.2014.408 Published online April 10, 2014</p>	Patent application filed; available for licensing	<p>Shuhendler, A.J. <i>et al. Nat. Biotechnol.</i>; published online March 23, 2014; doi:10.1038/nbt.2838 Contact: Jianghong Rao, Stanford University School of Medicine, Stanford, Calif. e-mail: jrao@stanford.edu</p>
Polymerized antibody-tagged magnetic beads for isolation of circulating tumor cells (CTCs)	<p>Magnetic beads tagged with polymerized antibodies or antibody-like molecules could help capture CTCs for cancer research purposes. A method of forming isopeptide bonds between peptide-tagged proteins enabled synthesis of polymeric chains of antibody-like molecules targeting epidermal growth factor receptor 1 (EGFR1; HER1; ErbB1) or HER2 (EGFR2; ErbB2; neu). In cultures of human breast cancer cell lines expressing HER1 or in blood samples spiked with HER2⁺ breast cancer cells, magnetic beads tagged with the polymerized antibody-like molecules captured cells better than magnetic beads tagged with monomeric antibody-like molecules. Planned work includes using the technology to capture CTCs from patients with breast cancer enrolled in a clinical trial of an undisclosed antiangiogenic therapy.</p> <p>SciBX 7(14); doi:10.1038/scibx.2014.409 Published online April 10, 2014</p>	Patent application filed by University of Oxford for method of isopeptide bond formation; available for licensing	<p>Fierer, J.O. <i>et al. Proc. Natl. Acad. Sci. USA</i>; published online March 17, 2014; doi:10.1073/pnas.1315776111 Contact: Mark Howarth, University of Oxford, Oxford, U.K. e-mail: mark.howarth@bioch.ox.ac.uk</p>
Protein misfolding cyclic amplification (PMCA) technique to create α -synuclein (SNCA) aggregates for drug screening	<p><i>In vitro</i> studies suggest SNCA aggregates formed using PMCA could be used to screen antiaggregating agents for Parkinson's disease (PD). The technique uses cycles of incubation to grow fibrils and cycles of sonication to break fibrils. The method resulted in faster formation of SNCA aggregates than traditional techniques, and drugs known to block aggregation were effective in the assay. Next steps could include using the approach to develop a cellular assay for screening.</p> <p>SciBX 7(14); doi:10.1038/scibx.2014.410 Published online April 10, 2014</p>	Findings unpatented; available for licensing and partnerships	<p>Herva, M.E. <i>et al. J. Biol. Chem.</i>; published online Feb. 28, 2014; doi:10.1074/jbc.M113.542340 Contact: Maria Grazia Spillantini, University of Cambridge, Cambridge, U.K. e-mail: mgs11@cam.ac.uk Contact: Maria Eugenia Herva, same affiliation as above e-mail: meh67@cam.ac.uk</p>

This week in techniques (continued)

Approach	Summary	Licensing status	Publication and contact information
Disease models			
Mice with human innate immune cells	Humanized mice with functional human innate immune cells could be used as models to study cancer in the presence of an intact human immune system. Previous humanized mouse models could not support development of human monocytes, macrophages or NK cells. In immunodeficient mice expressing human macrophage colony-stimulating factor 1 (CSF1; M-CSF), IL-3, granulocyte macrophage colony-stimulating factor (GM-CSF; CSF2), thrombopoietin (TPO) and signal regulatory protein- α (SIRPA), irradiation followed by transplantation of human fetal liver- or adult-derived CD34 ⁺ progenitor cells resulted in development of functional human monocytes, macrophages and NK cells but led to the destruction of red blood cells, which caused anemia within two to three weeks. In mice with functional human monocytes, macrophages and NK cells, compared with mice lacking such cells, engrafted human melanoma tumors showed increased infiltration by human macrophages and decreased tumor volume. Ongoing work includes using the mice to evaluate the behavior of hematologic malignancies, solid tumors and autoimmune diseases. SciBX 7(14); doi:10.1038/scibx.2014.411 Published online April 10, 2014	Patent application filed; available for licensing	Rongvaux, A. <i>et al. Nat. Biotechnol.</i> ; published online March 16, 2014; doi:10.1038/nbt.2858 Contact: Richard A. Flavell, Yale University, New Haven, Conn. e-mail: richard.flavell@yale.edu Contact: Markus G. Manz, University Hospital Zurich, Zurich, Switzerland e-mail: markus.manz@usz.ch
Mouse model of amyotrophic lateral sclerosis (ALS) caused by partial loss of TAR DNA binding protein 43 (Tdp-43; Tardbp) function	Mice with partial loss of Tdp-43 function could be useful as models to study and screen for new compounds to treat ALS. TDP-43 aggregation is associated with ALS. Mice were engineered to express artificial microRNA to knock down expression of <i>Tdp-43</i> . The engineered mice recapitulated ALS-like pathophysiology including progressive neuromuscular impairments that eventually led to paralysis and death, and neurodegeneration in the forebrain and spinal cord. Next steps could include using the mouse model to evaluate therapeutic candidates for ALS. SciBX 7(14); doi:10.1038/scibx.2014.412 Published online April 10, 2014	Patent and licensing status unavailable	Yang, C. <i>et al. Proc. Natl. Acad. Sci. USA</i> ; published online March 10, 2014; doi:10.1073/pnas.1322641111 Contact: Zuoshang Xu, University of Massachusetts Medical School, Worcester, Mass. e-mail: zuoshang.xu@umassmed.edu
Pituitary ablation of estrogen receptor expression as a mouse model for ovarian cancer	A genetic mouse model for estrogen-dependent ovarian cancer could be used to study disease progression and evaluate therapeutic candidates. In the mice, progesterone receptor-driven Cre recombinase was used to knock out estrogen receptor expression in the anterior pituitary. The resulting mice showed disruption of the hypothalamic-pituitary-ovarian hormonal axis, elevated estrogen production in ovaries, emergence of ovarian tumors and oncogenic transformation of ovarian surface epithelial cells. In the ovarian tumor-bearing mice, treatment with the aromatase inhibitor letrozole over 3 months decreased tumor volume by 60% compared with a sham treatment. Next steps could include using the model to study drug interactions. SciBX 7(14); doi:10.1038/scibx.2014.413 Published online April 10, 2014	Patent and licensing status unavailable	Laws, M.J. <i>et al. PLoS Genet.</i> ; published online March 6, 2014; doi:10.1371/journal.pgen.1004230 Contact: Indrani C. Bagchi, University of Illinois at Urbana-Champaign, Urbana, Ill. e-mail: ibagchi@illinois.edu Contact: Milan K. Bagchi, same affiliation as above e-mail: mbagchi@life.illinois.edu
Drug platforms			
Carbonic anhydrase IX (CAIX)-targeted small molecule-drug conjugates	CAIX-targeted small molecule conjugates could be useful for imaging and treating cancer. A series of small molecule ligands of CAIX, which is overexpressed in many types of cancer, were conjugated to fluorescent dyes or cytotoxic agents with a linker molecule. In a mouse xenograft model of human renal cell carcinoma (RCC), several CAIX-targeted ligands conjugated to fluorescent dye selectively accumulated at the tumor site. In the model, a CAIX-targeted ligand conjugated to the cytotoxic agent mertansine caused more potent tumor growth inhibition than untargeted conjugates or the RCC drugs Nexavar sorafenib or Sutent sunitinib. Next steps include studies to identify a lead candidate for clinical development. Pfizer Inc. markets the receptor tyrosine kinase (RTK) inhibitor Sutent to treat gastrointestinal stromal tumors (GISTs), pancreatic neuroendocrine tumors and advanced RCC. Amgen Inc. and Bayer AG market Nexavar to treat hepatocellular carcinoma and advanced RCC. SciBX 7(14); doi:10.1038/scibx.2014.414 Published online April 10, 2014	Patented; licensing status undisclosed	Krall, N. <i>et al. Angew. Chem. Int. Ed.</i> ; published online March 12, 2014; doi:10.1002/anie.201310709 Contact: Dario Neri, Swiss Federal Institute of Technology Zurich (ETHZ), Zurich, Switzerland e-mail: dario.neri@pharma.ethz.ch

This week in techniques (continued)

Approach	Summary	Licensing status	Publication and contact information
Crystal structure of purinergic receptor P2Y G protein-coupled 12 (P2RY12; P2Y12) bound to an antagonist	<i>In vitro</i> structural studies of P2RY12 bound to an antagonist could aid antithrombotic drug design. P2RY12 is activated by nucleotide binding and regulates platelet activation and thrombus formation. A 2.6 Å-resolution crystal structure showed that antagonist-bound P2RY12 adopted a canonical seven-transmembrane bundle of helices and that the extracellular cavity of the receptor is divided into two pockets with the antagonist bound in one. <i>In silico</i> analyses predict that both pockets could be bound by dinucleotide or chemically related molecules. Next steps include understanding how binding in the two pockets impacts affinity and side effects associated with P2RY12 antagonists. P2RY12 antagonists marketed to treat cardiovascular disease include AstraZeneca plc and The Medicines Co.'s Brilinta ticagrelor, Daiichi Sankyo Co. Ltd. and Eli Lilly and Co.'s Effient prasugrel and Sanofi and Bristol-Myers Squibb Co.'s Plavix clopidogrel. At least five companies have P2RY12 antagonists in Phase III or earlier testing to treat cardiovascular disease. SciBX 7(14); doi:10.1038/scibx.2014.415 Published online April 10, 2014	Unpatented; licensing status not applicable	Zhang, K. <i>et al. Nature</i> ; published online March 23, 2014; doi:10.1038/nature13083 Contact: Qiang Zhao, Shanghai Institute of Materia Medica, Chinese Academy of Sciences, Shanghai, China e-mail: zhaoq@simm.ac.cn
Enhanced antigen-specific T cell activation using nanoscale artificial antigen-presenting cells (aAPCs) in a magnetic field	<i>In vitro</i> and mouse studies suggest magnetic-field culturing methods could increase activation of antigen-specific T cells by nanoscale aAPCs. In T cells cultured with nanoscale aAPCs, exposure to a magnetic field induced aAPC aggregation to increase T cell receptor cluster size, which led to increased T cell expansion and activation compared with no exposure to a magnetic field. In mice with subcutaneous melanoma tumors, adoptive transfer of tumor-specific T cells activated by aAPCs in a magnetic field caused decreased tumor growth compared with adoptive transfer of cells cultured without the magnetic field. Next steps include developing GMP-grade materials for use in humans. SciBX 7(14); doi:10.1038/scibx.2014.416 Published online April 10, 2014	Patent applications filed; exclusively licensed to NexImmune Inc. for immune regulation; available for licensing in indications other than immunology	Perica, K. <i>et al. ACS Nano</i> ; published online Feb. 24, 2014; doi:10.1021/nn405520d Contact: Jonathan P. Schneck, The Johns Hopkins University School of Medicine, Baltimore, Md. e-mail: jschnecl@jhmi.edu
Small molecule inhibitors of microRNA processing	An <i>in vitro</i> screening system could be used to identify small molecule inhibitors of miRNAs. Structural calculations of human miRNA hairpin precursors were analyzed in conjunction with a database of predicted interactions between small molecules and RNA motifs to identify small molecules that could bind to processing sites required for miRNA maturation. In a human cell line, one computationally identified compound inhibited the production of miRNA-96 (miR-96) and stimulated apoptosis. Next steps could include using the method to identify small molecules to inhibit miRNAs or other RNAs of therapeutic interest. SciBX 7(14); doi:10.1038/scibx.2014.417 Published online April 10, 2014	Patent and licensing status unavailable	Velagapudi, S.P. <i>et al. Nat. Chem. Biol.</i> ; published online Feb. 9, 2014; doi:10.1038/nchembio.1452 Contact: Matthew D. Disney, Scripps Florida, Jupiter, Fla. e-mail: disney@scripps.edu

SciBX: Science–Business eXchange

Kick-start your knowledge management—and leave your competitors behind...

Can you afford not to subscribe?

Visit scibx.com for details on how to subscribe to SciBX

Company and institution index**A**

Amgen Inc. 16
 AstraZeneca plc 3,17
 Atterocor Inc. 11
 Avillion LLP 3

B

Bayer AG 16
 BioAlliance Pharma S.A. 3
 Bristol-Myers Squibb Co. 3,7,17

C

Cancer Research Technology Ltd. 6
 Celtaxsys Inc. 14

D

Daiichi Sankyo Co. Ltd. 17

E

Eli Lilly and Co. 17
 Emcure Pharmaceuticals Ltd. 7

G

GlaxoSmithKline plc 3,13

H

Hanmi Pharmaceutical Co. Ltd. 3

I

Intuitive Surgical Inc. 9

J

Johnson & Johnson 3

K

Kansas Bioscience Authority 10
 Karolinska Institute 6
 Karolinska Institute Innovations AB 7
 Kinex Pharmaceuticals LLC 3

L

Leukemia & Lymphoma Society 10

M

Medicines Co. 4,13,17
 Merck & Co. Inc. 6
 Merck KGaA 3

N

National Center for Advancing Translational Sciences 10
 National Institutes of Health 11
 Nestle S.A. 10
 NexImmune Inc. 17
 Nippon Shinyaku Co. Ltd. 3
 NonClinical Safety Assessment 8

O

OncoMed Pharmaceuticals Inc. 11
 Otsuka Pharmaceutical Co. Ltd. 3

P

Paganini Biopharma Inc. 1,10

PerkinElmer Inc. 9
 Pfizer Inc. 3,13,16
 Prometheus Laboratories Inc. 10

R

Reliance Life Sciences 6

S

Sanofi 17
 Stanford University 9
 Stanford University School of Medicine 8

T

Tactic Pharma LLC 3
 Teva Pharmaceutical Industries Ltd. 3

U

University of Alabama at Birmingham School of Medicine 7
 University of California, Los Angeles 1,10
 University of California, Los Angeles David Geffen School of Medicine 1
 University of California, Los Angeles Jonsson Comprehensive Cancer Center 2
 University of California, San Diego 2
 University of Delaware 11
 University of Leicester 4,13
 University of Massachusetts Medical School 4
 University of Oxford 15
 University of Toronto 8

V

ValiRx plc 3
 Verastem Inc. 3
 Virginia Commonwealth University 6

W

Washington University in St. Louis School of Medicine 4

.....

Target and compound index

1,3-Bis(chloroethyl)-1-nitrosourea 7
 4-(Piperidin-2-ylmethanol)-quinoline 7
 5'-3' Exoribonuclease 2 12

A

α -Synuclein 15
 ACAT1 11
 Acetaminophen 8,15
 ALK 3
 Alternative pathway C3 convertase 4
 AMEP 3
 Anaplastic lymphoma kinase ANO1 3
 Anoctamin 1 calcium activated chloride channel 10

Anti-Notch2/3 11
 Aromatase 16
 ATN-161 3
 Auranofin 10
 Avandia 13
 AZD0424 3

B

BA-015 3
 BCR-ABL tyrosine kinase 3
 Bdnf 13
 BGB-101 3
 BiCNU 7
 BMS-354825 3
 Bosulif 3
 Bosutinib 3
 Brain-derived neurotrophic factor 13
 Brilinta 17

C

CaCC_{inh}-A01 10
 CAIX 16
 Carbonic anhydrase IX 16
 Carmustine 7
 CD331 11
 CD34 10,16
 CD44v6 11
 CD51/CD29 3
 CD51/CD61 1
 CEP-37440 3
 CFH 5
 Cilengitide 3
 Ciprofloxacin 13
 Cisplatin 11
 Clopidogrel 17
 CNTO-95 3
 Complement factor H 5
 CSF1 16
 CSF2 16
 CTX-4430 14
 Cytochrome P450 8

D

Dasatinib 3
 Defactinib 3
 Difloxacin 13

E

Effient 17
 EGFR1 15
 EGFR2 15
 EMD 121974 3
 EMP2 1,10
 Epidermal growth factor receptor 1 15
 Epithelial membrane protein 2 1,10
 ErbB1 15
 ErbB2 15
 Estrogen receptor 16

F

F2 12
 Factor IIa 12
 FAK 1
 Farnesoid X receptor 12
 FGF4 11
 FGFR1 11
 Fibroblast growth factor 4 11
 Fibroblast growth factor

receptor 1 11
 Focal adhesion kinase 1
 FXR 12

G

Galactose 8,15
 GM-CSF 16
 Granulocyte macrophage colony-stimulating factor 16
 GSK2256098 3

H

HER1 15
 HER2 15
 HRD1 12

I

IL-24 6
 IL-3 16
 Integrin $\alpha_5\beta_1$ 3
 Integrin $\alpha_v\beta_1$ 3
 Integrin $\alpha_v\beta_3$ 1
 Integrin $\alpha_v\beta_5$ 3
 Integrin $\alpha_v\beta_6$ 3
 Intetumumab 3
 Isoniazid 8,15

J

JAG1 11
 Jagged 1 11
 JAK-2 3
 Janus kinase-2 3

K

KX01 3
 KX2-391 3

L

Letrozole 16
 Leukotriene A4 hydrolase 14
 Leukotriene B4 14
 Levofloxacin 13
 LTA4H 14
 LTB4 14

M

M-CSF 16
 Macrophage colony-stimulating factor 1 16
 MAP2K4 6
 MDA7 6
 Mertansine 16
 miR-96 17
 miRNA-96 17

Mitogen-activated protein kinase kinase 4 6
 MKK4 6
 MLL5 12
 Myeloid-lymphoma or mixed-lineage 5 12

N

Neu 15
 Nexavar 16
 NFE2L2 12
 Notch 2 11
 Notch 3 11
 NOTCH2 11
 NOTCH3 11
 NR1H4 12
 NRF2 12
 NS-018 3

Nuclear factor (erythroid-derived 2)-like 2	12	Progesterone receptor	16	Src	1	Transaminase	9
O		Properdin	4,13	Sterol O-acyltransferase 1	11	Trovaflaxacin	13
OMP-59R5	11	Purinergic receptor P2Y G protein-coupled 12	17	Sunitinib	16	Trovan	13
Oritavancin	4	Q		Superoxide dismutase	9	TSP-1	4
P		Quinoline	6	Sutent	16	U	
P2RY12	17	R		Synoviolin	12	UAF1	11
P2Y12	17	Receptor tyrosine kinase	16	SYVN1	12	Ubiquitin specific peptidase 1	11
p450	8	Ridaura	10	T		USP1	11
Pannexin 1	13	Rosiglitazone	13	TAR DNA binding protein 43	16	V	
PANX1	13	RTK	16	Tardbp	16	Vacquinol-1	6
Peroxisome proliferation-activated receptor- γ	12,13	S		Tdp-43	16	VAL201	3
PF-05208763	3	Signal regulatory protein- α	16	Temodal	6	VS-4718	3
PF-4554878	3	SIRPA	16	TemoRel	6	VS-6063	3
PG-101	1	SKI-606	3	THBS1	4	W	
Phosphoinositide 3-kinase	11	SNCA	15	Thrombin	12	WD repeat domain 48	11
PI3K	11	SOAT1	11	Thrombopoietin	16	WDR48	11
Plavix	17	SOD	9	Thrombospondin	4	X	
PPAR γ	12,13	Sorafenib	16	Thrombospondin-1	4	X-box binding protein 1	12
PPARG	12,13	Splice variant v6 of CD44	11	Ticagrelor	17	XBP1	12
Prasugrel	17	Sprycel	3	Tosufloxacin	13	XRN2	12
				TPO	16		

SciBX: Science-Business eXchange

SciBX welcomes editorial queries, comments and press releases.

To contact the editorial team at *SciBX* please e-mail editorial@scibx.com