

THIS WEEK

ANALYSIS

COVER STORY

1 Nav-i-gating antibodies for pain

Nav1.7 entered the scene a decade ago as a pain target with wide-ranging analgesic effects, but it has been difficult to selectively target. Now, a team at Duke has succeeded with an antibody that blocks the channel in a closed state. The team is developing the antibody for pain and pruritus.

TRANSLATIONAL NOTES

4 CQDM: Canada on the brain

The Quebec Consortium for Drug Discovery and Brain Canada have announced at least C\$10 million (US\$9.2 million) in funding for a new program to develop tools for pharma partners to accelerate the discovery of neurological disease therapies.

TARGETS & MECHANISMS

6 Putting SMYD3 on the MAP

Stanford and GlaxoSmithKline report that SMYD3 acts in the cytoplasm to regulate the MAPK pathway and not in the nucleus as previously thought. The discovery implies that inhibiting the enzyme can counter activating mutations in MAPK pathway components such as *KRAS*.

9 Stromal uncertainties in pancreatic cancer

Despite the clear rationale for using hedgehog inhibitors in pancreatic cancer—they deplete the stroma to improve chemotherapy delivery—clinical results have been disappointing. New data suggest the large amounts of stroma surrounding some pancreatic cancers might actually have protective properties.

THE DISTILLERY

12 This week in therapeutics

Promoting wound healing by activating BLT2; treating AD with IP3R antagonists; improving Th1 immune response with K-class CpG oligonucleotides as vaccine adjuvants; and more...

18 This week in techniques

Engineered bone marrow-on-a-chip system; targeted, dual-drug liposomal nanoparticles; DPP-4 as a marker of CML stem cells; and more...

INDEXES

19 Company and institution index**19 Target and compound index**

Nav-i-gating antibodies for pain

By Lauren Martz, Staff Writer

Nav1.7 entered the limelight in the last decade as a pain target that could provide wide-ranging analgesia, but it has been difficult to target selectively over other voltage-gated sodium channels. Now, a team at the **Duke University Medical Center** has found a unique epitope on Nav1.7 and used it to create a highly selective antibody that blocks the channel by locking it in a closed state.¹

In mice, the antibody reduced pain and inflammation and suppressed acute and chronic itch. The Duke researchers are in discussions with companies to create a humanized form of the antibody.

Nav1.7 (SCN9A) emerged as a pain target in 2006 when a **University of Cambridge** team found loss-of-function mutations in the channel in patients with a rare congenital inability to feel pain.² Because the mutations caused no other overt pathology, drug developers jumped on the possibility that channel inhibitors could provide analgesia without causing major side effects.

At least six companies have Nav1.7-targeted compounds in clinical or preclinical development for pain (*see Table 1, “Nav1.7 small molecule inhibitors and antibodies”*).

However, the biggest challenge has been to find Nav1.7-selective compounds—which is particularly important because of the many processes triggered by other channel family members.

“There are nine subtypes of voltage-gated sodium channels in humans. They are very similar in sequence, but many sodium channel subtypes are responsible for very important but distinct roles in physiological processes,” said Seok-Yong Lee, an assistant professor of biochemistry at Duke.

For example, he said, Nav1.4 (SCN4A) and Nav1.5 (SCN5A) are responsible for muscle and cardiac action potentials, respectively. “Therefore, to reduce serious side effects, it is critical to make a subtype-specific inhibitor for sodium channels. But because of their sequence conservation, it has not been very easy,” said Lee.

In 2011, researchers at the **University of Washington** published the first crystal structure of a voltage-gated sodium channel, thus paving the way for structure-based drug design to target these channels.³

“To reduce serious side effects, it is critical to make a subtype-specific inhibitor for sodium channels. But because of their sequence conservation, it has not been very easy.”

—Seok-Yong Lee,
Duke University Medical Center

**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:** James Butcher, 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.

Now, Lee, Ru-Rong Ji and colleagues at Duke have used that structure to home in on the voltage sensor paddle region of Nav1.7—a domain that allosterically controls channel gating and differs between channel subtypes.

Ji is a professor in the Department of Anesthesiology and Neurobiology at Duke.

The team started by developing an active mAb to an epitope in the paddle region that responds to changes in membrane potential. It also created a control antibody against a different epitope that is not involved in channel gating in the same region.

In a cell line expressing human Nav1.7, the active antibody decreased sodium currents compared with the control and stabilized the closed state of the channel. The antibody had about 400-fold selectivity over subtype Nav1.6 (PN4; SCN8A) and no detectable activity against the six other channel subtypes tested.

Next, the researchers showed that the antibody suppressed formalin-induced pain and inflammation in mice after i.v., intrathecal or intraplantar administration. They noted that the i.v. and intrathecal analgesic doses were lower than morphine doses reported by others for the same model.

In a mouse model of neuropathic pain caused by chronic constriction injury, the antibody suppressed pain for several hours and did not lose its potency after repeated injections. That suggested the antibody does not cause tolerance, a problem commonly found with opioid analgesics.

Because Nav1.7 is involved in synaptic transmission of pain in nociceptive neurons in the dorsal root ganglion, and the pruriceptive neurons that mediate itch are a subset of nociceptive neurons, the team tested whether the sodium channel is also involved in the transmission of itch.

The researchers found that the Nav1.7 antibody suppressed the itch response in mouse models of acute pruritus and chronic models representing dry skin and allergic contact dermatitis.

In spinal cord sections from the pain and itch models, the antibody inhibited spontaneous excitatory postsynaptic currents in nociceptive neurons triggered by both pain and itch stimuli. These findings suggest that blocking Nav1.7 suppresses pain and itch by preventing synaptic transmission in the spinal cord.

Data were published in *Cell*.

Selective advantage

Glenn King, a professor in the Division of Chemistry and Structural Biology at **The University of Queensland's** Institute of Molecular Bioscience, told *SciBX*, "I think this is the most important paper on Nav1.7 since the original observation reported in *Nature* in 2006."

He said that it shows for the first time that pharmacological blockade of Nav1.7 provides relief for most types of pain, Nav1.7 is critically involved in transmission of pruriceptive receptors and it is possible to design a highly selective mAb against the target.

According to several experts who spoke with *SciBX*, the selectivity of the Nav1.7 antibody provides it with a significant advantage over competitor compounds.

Peter Ulrichs, senior scientist at **arGEN-X B.V.**, said, "Recent published reports have described human Nav1.7-selective small molecules and venom-derived peptides, but none have the selectivity of the human Nav1.7 antibody described in the article."

Table 1. Nav1.7 small molecule inhibitors and antibodies. Specifically blocking the Nav1.7 (SCN9A) voltage-gated sodium channel subtype suppresses pain. At least six companies have small molecule inhibitors or antibodies against Nav1.7 in clinical and preclinical testing to treat pain. Source: *BCIQ: BioCentury Online Intelligence*

Company	Product	Description	Phase of development
Convergence Pharmaceuticals Ltd.	CNV1014802	Small molecule, state-dependent Nav1.7 inhibitor	Phase II
Dainippon Sumitomo Pharma Co. Ltd. (Tokyo:4506)	DSP-2230	Small molecule inhibitor of Nav1.7 and Nav1.8 (PN3; SCN10A)	Phase I
Pfizer Inc. (NYSE:PFE)	PF-05089771	Small molecule Nav1.7 inhibitor	Phase I
SiteOne Therapeutics Inc.	ST-200	Small molecule Nav1.7 inhibitor	Discovery
arGEN-X B.V.	Nav1.7 antibodies	Antibodies against Nav1.7	Discovery
Numab AG	ND004	Antibody against Nav1.7	Discovery

According to George Miljanich, CEO of **SiteOne Therapeutics Inc.**, the difficulty of developing selective small molecules is partly caused by their hydrophobicity. John Mulcahy, director of R&D at SiteOne, added that Nav1.7-selective small molecules so far have generally shown poor pharmacokinetics, which has held up their development.

SiteOne is developing small molecule Nav1.7 blockers based on marine guanidinium toxins that are natural inhibitors of the channel.

“We know already from our interactions

with big pharma that Nav1.7 is a highly attractive and compelling target in pain and that antibodies are being taken seriously as a potential new treatment modality,” said Ulrichts.

However, Miljanich said that although antibodies against the channel might reach the market first, they will most likely be used to guide the design of small molecules that will eventually supplant them. He noted that small molecules replacing biologics is the course of drug development that has been unfolding in the autoimmune disease market.

Itching for more

Lee said that the next step will be for the Duke team to create a humanized version of the Nav1.7 antibody that retains the specificity of the mouse antibody and optimize its pharmacokinetic properties.

However, the humanization process itself can be tricky, said Ulrichts. “It would be hoped that its humanization and further engineering would not have any adverse effects on its functional properties,” he said.

Frank Zufall told *SciBX* that even if specific channel targeting is achieved with a humanized antibody, there could be side effects associated with targeting the channel itself. “Nav1.7 is important in anosmia. The people with loss-of-function mutations in Nav1.7 can’t smell. It is interesting that the same channel hits the three sensory systems—pain, itch and smell—and the goal now may be to target these systems independently,” he said.

Zufall is a professor of physiology at the **Saarland University Medical Center** and an expert in molecular medicine of sensory systems including olfaction.

However, he added that losing some sense of smell may be a tolerable risk for pain patients. “As long as you know that loss of olfaction is a side effect and the treatment effectively gets rid of pain, most patients may be OK with this side effect. Most patients will do anything to get rid of pain.”

The antibody’s ability to suppress itch could add to the commercial interest in Nav1.7 as a therapeutic target.

“Current antihistamines are insufficient to treat chronic itch. The Nav1.7 antibody can treat both histamine-dependent and -independent itch as well as chronic itch,” said Lee. The authors added that the antibody might benefit patients with skin diseases such as dermatitis.

Miljanich told *SciBX* that this is an opportunity for companies developing therapeutics targeting Nav1.7.

“The major contribution from this work is the implication that Nav1.7 is involved in the transmission of itch,” he said. “Although I have not done market research, I expect that there is a significant unmet need for itch and specifically severe chronic itch.”

He added, “We may add itch as a target indication at SiteOne because itch trials could be faster and easier than pain trials. Itch may be an easier clinical endpoint to measure.”

Lee told *SciBX* that **Duke University** has filed a patent application covering the work. The IP is available for licensing.

Martz, L. *SciBX* 7(23); doi:10.1038/scibx.2014.662
Published online June 12, 2014

REFERENCES

- Lee, J.-H. *et al. Cell*; published online May 22, 2014; doi:10.1016/j.cell.2014.03.064
Contact: Seok-Yong Lee, Duke University Medical Center, Durham, N.C.
e-mail: sylee@biochem.duke.edu
Contact: Ru-Rong Ji, same affiliation as above
e-mail: rong.ji@duke.edu
- Cox, J.J. *et al. Nature* **444**, 894–898 (2006)
- Payandeh, J. *et al. Nature* **475**, 353–358 (2011)

COMPANIES AND INSTITUTIONS MENTIONED

arGEN-X B.V., Rotterdam, the Netherlands
Duke University, Durham, N.C.
Duke University Medical Center, Durham, N.C.
Saarland University Medical Center, Homburg, Germany
SiteOne Therapeutics Inc., Redwood City, Calif.
University of Cambridge, Cambridge, U.K.
The University of Queensland, Brisbane, Queensland, Australia
University of Washington, Seattle, Washington

CQDM: Canada on the brain

By Michael J. Haas, Senior Writer

The **Quebec Consortium for Drug Discovery** and **Brain Canada** have announced at least C\$10 million (US\$9.2 million) in funding for a new program to develop tools to accelerate the discovery of neurological disease therapies.

Under the three-year Focus on Brain program, the Quebec Consortium (CQDM) and the not-for-profit Brain Canada will fund collaborative projects at research institutions and small companies across Canada. The goal is to generate tools, technologies and platforms that could enable industry to overcome some of the barriers that impede drug discovery in neurology.

Those problems, said Diane Gosselin, include the challenge of delivering therapies across the blood brain barrier, the lack of preclinical models that predict the efficacy of therapies to treat Alzheimer's disease (AD) and other neurodegenerative indications, and the need for markers to stratify patients for clinical trials and identify who is likely to respond to a therapy.

Gosselin is president and CEO of CQDM.

The partners expect to fund about seven projects and will provide a total of up to C\$500,000 (US\$458,000) annually to each project.

"We have been somewhat vague about the total amount of funding and number of projects because our two organizations have the ability to do more" depending on the quality of project proposals received, Gosselin said.

CQDM receives two-thirds of its funding from the federal and provincial governments and one-third from its pharma sponsors and other partner organizations. Thus, Gosselin said, the consortium can match every \$1 a pharma invests in it with about \$19 additional dollars, thereby giving the pharma \$20 worth of R&D.

"The more pharmas we have, the more leverage we'll have—so it makes sense for us to have as many pharmas as possible," she said.

This month, she said, the consortium expects to disclose the amount of funding a third partner, the **Ontario Brain Institute** (OBI), will provide to the program on top of the C\$10 million (US\$9.2 million) total from CQDM and Brain Canada.

And today, CQDM announced that **Sanofi** joined the consortium as its eighth pharma sponsor.

"We believe that R&D is not a one-company matter anymore," said Marc Bonnefoi, head of the North America R&D hub and VP of the disposition, safety and animal research scientific core platform at Sanofi. Thus, "for the past two to three years, we have been focusing our efforts to tap external innovation in the EU and U.S. Now we want to explore the opportunities for external innovation in Canada, where our R&D has been limited primarily to running clinical trials."

Bonnefoi added, "We chose CQDM because their precompetitive platform merges the interest of academic institutions, small companies and pharmas, and they have very good funding programs."

Pan-Canadian brainstorm

CQDM and Brain Canada are now accepting project proposals for the Focus on Brain program and expect to award the first grants by April 2015.

Gosselin said that Focus on Brain was inspired by the 2012 Quebec/Ontario funding program that CQDM established in partnership with OBI, the **Ontario Centres of Excellence** and **MaRS Innovation** to develop tools for biopharma research. The program also was the consortium's first step in building a network of Canadian researchers outside Quebec.

At that time, CQDM wanted to get more involved in funding neurology projects, she said. Besides a shared interest in neuroscience, "we could see that CQDM and OBI have many of the same goals, including funding of translational research, fostering innovation and working with pharmas."

The two organizations decided to launch a pan-Canadian neuroscience initiative but wanted another partner with experience and connections across Canada. "This is where Brain Canada came in. They have the ability to fund research across Canada, and they have the networks and expertise to help us organize a neuroscience program," Gosselin said.

She added, "CQDM's links with pharma were attractive to Brain Canada because it had been funding mostly academic R&D and wanted to get more into industry and translational research."

Haifa Staiti, director of research programs at Brain Canada, agreed. "This is the first initiative of its kind in which a funding program is encouraging collaborations not only across Canada but also between academia and industry," she said.

Indeed, the scope of Focus on Brain is in line with CQDM's goal of continuing to raise the visibility of Canadian research and innovation in the eyes of the global pharmaceutical industry, Gosselin said.

"The problems confronting biopharmaceutical R&D are so complex that it would not be wise for us to duplicate the efforts of others by trying to solve them on our own," she said. "Instead, we want to work with groups outside Canada. But in order to position ourselves among other initiatives around the world and form partnerships with them, we first need a strong network of

researchers within Canada" that demonstrates what the country can offer on the global stage.

CQDM already has funded six neurology projects outside of the Focus on Brain program. Of these, ongoing projects include one by researchers at the **University Institute of Geriatrics of Montreal** for noninvasive imaging of oxidative metabolism in the brain to stratify patients with AD and another by researchers at **Laval University** and **McMaster University** to validate catecholamine-regulated protein 40 (CRP40) as a marker for the early diagnosis of Parkinson's disease (PD). CRP40 is an alternate, 40 kDa splice variant of heat shock 70 kDa protein 9 (HspA9; mortalin-2).

Gosselin noted that CQDM is not involved with the **NIH's** Brain Research through Advancing Innovative Nanotechnologies (BRAIN) Initiative—which aims to map the human brain to show how individual

"In order to position ourselves among other initiatives around the world and form partnerships with them, we first need a strong network of researchers within Canada."

*—Diane Gosselin,
Quebec Consortium for Drug Discovery*

cells and neural circuits interact—because the initiative will not produce a translation tool or technology that has immediate utility in solving neurology drug discovery problems. But as part of its plan for international expansion, the consortium will continue to evaluate how or whether it could contribute to such large-scale initiatives outside Canada, she said.

Five years and counting

Since 2008, CQDM has funded a total of 32 projects under 4 of its existing programs, which are called Focus, Explore, Quebec/Ontario and Quebec/France.

Focus is for large-scale, multi-institutional projects within Quebec that will produce immediately usable results. Explore is for innovative and high-risk projects within Quebec that have the potential for a major breakthrough.

The Quebec/Ontario program funds Focus- and Explore-like projects by researchers in both provinces. The Quebec/France program also supports Focus- and Explore-like collaborations, with CQDM funding the research conducted in Quebec and the **Alsace BioValley** cluster funding the research conducted in France.

Of 11 completed projects, 10 achieved the target milestones and deliverables, Gosselin said. She added that results from eight of those projects are in use by one or more of the consortium's pharma partners.

Completed Focus projects include biosensors that predict the efficacy and side effects of drugs targeting GPCRs. The biosensors were developed by researchers at the **University of Montreal**, **McGill University**, **Sherbrooke University** and the **Montreal Heart Institute**. Another completed Focus project is a noninvasive method for stratifying patients with major psychiatric disorders based on retinal responses to photostimulation (electroretinography). The technology was developed by researchers at Laval University and **NDEI Inc.**

Completed Explore projects include a McGill study that found somatic mutations that cause autoimmune diseases such as rheumatoid arthritis (RA) and a computer platform for optimizing the specific interactions between antibodies and their target antigens that was developed by a team at the **National Research Council Canada**.

The one completed Quebec/France project, by researchers from **Caprion Proteomics Inc.** and the **Centre National de la Recherche Scientifique** (CNRS), identified tumor-secreted proteins as new diagnostic and prognostic markers for neuroendocrine cancers.

Gosselin also said that 25 partnerships have been formed between CQDM's academic and industry members outside the scope of CQDM projects. As an example, she cited an ongoing Focus project mentored by **Merck & Co. Inc.** in which researchers at the Montreal Heart Institute and the **National Institute of Scientific Research** are developing radio-labeled peptides as imaging agents for the early detection of pulmonary hypertension. Based on the team's results so far, Merck has established a separate partnership with the researchers to develop a PET version.

In another partnership, **Pfizer Inc.** and the University of Montreal researcher who led the GPCR biosensor project are developing a drug discovery platform based on that biosensor technology.

To date, cash and in-kind investments resulting from partnerships among the consortium's members—beyond the scope of a CQDM project—have totaled more than C\$15 million (US\$13.7 million), Gosselin said. In addition, five startup companies have been founded based on the results of CQDM-funded projects.

In addition to newcomer Sanofi, CQDM includes its three founding pharmas—**AstraZeneca plc**, Merck and Pfizer—as well as **Boehringer Ingelheim GmbH**, **Eli Lilly and Co.**, **GlaxoSmithKline plc** and **Novartis AG**.

Pharma sponsors invest equally in CQDM, and the money is applied equally to all projects. In return, the pharmas gain nonexclusive rights to the project results.¹

CQDM also includes researchers at 21 academic institutions and hospitals and 25 private companies in Quebec, Ontario and France.

All CQDM-funded projects are assigned one or more senior scientist mentors from the consortium's pharma sponsors to ensure

that projects are progressing in line with industry needs. Gosselin said that Focus on Brain projects will have at least three mentors—each from a different pharma—and could have more depending on the level of interest the remaining pharmas have in a project.

Haas, M.J. *SciBX* 7(23); doi:10.1038/scibx.2014.663
Published online June 12, 2014

REFERENCES

1. Haas, M.J. *SciBX* 4(41); doi:10.1038/scibx.2011.1135

COMPANIES AND INSTITUTIONS MENTIONED

Alsace BioValley, Illkirch, France
AstraZeneca plc (LSE:AZN; NYSE:AZN), London, U.K.
Boehringer Ingelheim GmbH, Ingelheim, Germany
Brain Canada, Montreal, Quebec, Canada
Caprion Proteomics Inc., Montreal, Quebec, Canada
Centre National de la Recherche Scientifique, Strasbourg, France
Eli Lilly and Co. (NYSE:LLY), Indianapolis, Ind.
GlaxoSmithKline plc (LSE:GSK; NYSE:GSK), London, U.K.
Laval University, Quebec City, Quebec, Canada
MaRS Innovation, Toronto, Ontario, Canada
McGill University, Montreal, Quebec, Canada
McMaster University, Hamilton, Ontario, Canada
Merck & Co. Inc. (NYSE:MRK), Whitehouse Station, N.J.
Montreal Heart Institute, Montreal, Quebec, Canada
National Institute of Scientific Research, Montreal, Quebec, Canada
National Institutes of Health, Bethesda, Md.
National Research Council Canada, Ottawa, Ontario, Canada
NDEI Inc., Sainte-Foy, Quebec, Canada
Novartis AG (NYSE:NVS; SIX:NOVN), Basel, Switzerland
Ontario Brain Institute, Toronto, Ontario, Canada
Ontario Centres of Excellence, Toronto, Ontario, Canada
Pfizer Inc. (NYSE:PFE), New York, N.Y.
Quebec Consortium for Drug Discovery, Montreal, Quebec, Canada
Sanofi (Euronext:SAN; NYSE:SNY), Paris, France
Sherbrooke University, Sherbrooke, Quebec, Canada
University Institute of Geriatrics of Montreal, Montreal, Quebec, Canada
University of Montreal, Montreal, Quebec, Canada

Putting SMYD3 on the MAP

By Lev Osherovich, Senior Writer

Stanford University and GlaxoSmithKline plc researchers have uncovered the mechanism of action of SMYD3, a histone lysine methyltransferase overexpressed in many lung and pancreatic tumors.¹ The surprise was that SMYD3 acts in the cytoplasm to regulate the MAPK pathway and not in the nucleus as previously thought. The discovery implies that inhibiting the enzyme can counter activating mutations in MAPK pathway components such as K-Ras.

SMYD3 (SET and MYND domain containing 3) is a member of a family of histone lysine methyltransferases that typically are epigenetic regulators of chromatin structure in the nucleus.²

Now, a team co-led by Julien Sage and Or Gozani has found a previously unknown function for SMYD3 in cancer. Rather than working in the nucleus as expected, SMYD3 promotes cancer growth by regulating a branch of the cytoplasmic MAPK signaling pathway.

Sage is an associate professor of pediatrics and genetics at the Stanford University School of Medicine. Gozani is an associate professor of biology at Stanford University.

The findings suggest that SMYD3 inhibitors could complement MAPK pathway inhibitors such as GSK's Mekinist trametinib. Mekinist is marketed to treat melanoma with activating mutations in the upstream MAPK pathway regulator BRAF. The drug is in Phase II testing for solid tumors including lung cancers with K-Ras (KRAS)-activating

“The beauty and the weakness of hitting SMYD3 is that this is not an essential protein for MAPK signaling. Thus, drugs against SMYD3 are not going to have as many side effects as direct inhibition of the pathway. On the other hand, they may not be as effective as drugs that hit the main components of the pathway.”

—Julien Sage,
Stanford University School of Medicine

Figure 1. SMYD3 in cancer. Mazur *et al.* have identified the role of SET and MYND domain containing 3 (SMYD3) in K-Ras (KRAS)-driven lung and pancreatic cancers.

In certain lung and pancreatic tumors, activating mutations in *KRAS* or *BRAF* (a) lead to excessive signaling through downstream MAPK signaling pathways. One branch of the pathway utilizes mitogen-activated protein kinase kinase 2 (MAP3K2) (b) to activate downstream effectors mitogen-activated protein kinase kinase 5 (MAP2K5; MEK5) and MAP kinase 7 (MAPK7; BMK1; ERK-5) (c), which drive tumor growth (d).

The team uncovered that the histone lysine methyltransferase SMYD3 can methylate (CH₃) MAP3K2 (e), leaving the kinase stuck in an active state to promote hyperactive proliferative signaling.

mutations. At least 18 cancer therapeutics targeting the MAPK pathway are in development or on the market (see Table 1, “Selected MAPK pathway compounds in cancer”).

“SMYD3 is acting in the cytoplasm to methylate a specific target in the MAPK cascade,” said Robert Copeland, EVP and CSO of Epizyme Inc. “The demonstration of a broader role for this enzyme beyond just methylating histones speaks to the interplay between chromatin-remodeling factors and other important signaling pathways.”

Under a 2011 deal, Epizyme and GSK are discovering compounds against three undisclosed epigenetic targets.

SMYD range

As part of an ongoing collaboration to characterize the function of epigenetic targets, the Stanford-GSK team started by analyzing gene expression data for 54 histone lysine methyltransferases in a panel of pancreatic cancers with high KRAS activity.

SMYD3 levels were consistently higher in the tumors than in healthy tissue, suggesting that the enzyme might contribute to cancer growth. To test this possibility, the team introduced conditionally activated *Kras* mutations into a *Smyd3* knockout mouse. The animals were less susceptible to pancreatic and lung tumors caused by hyperactive *Kras* than controls with normal *Smyd3*. The team observed similar effects

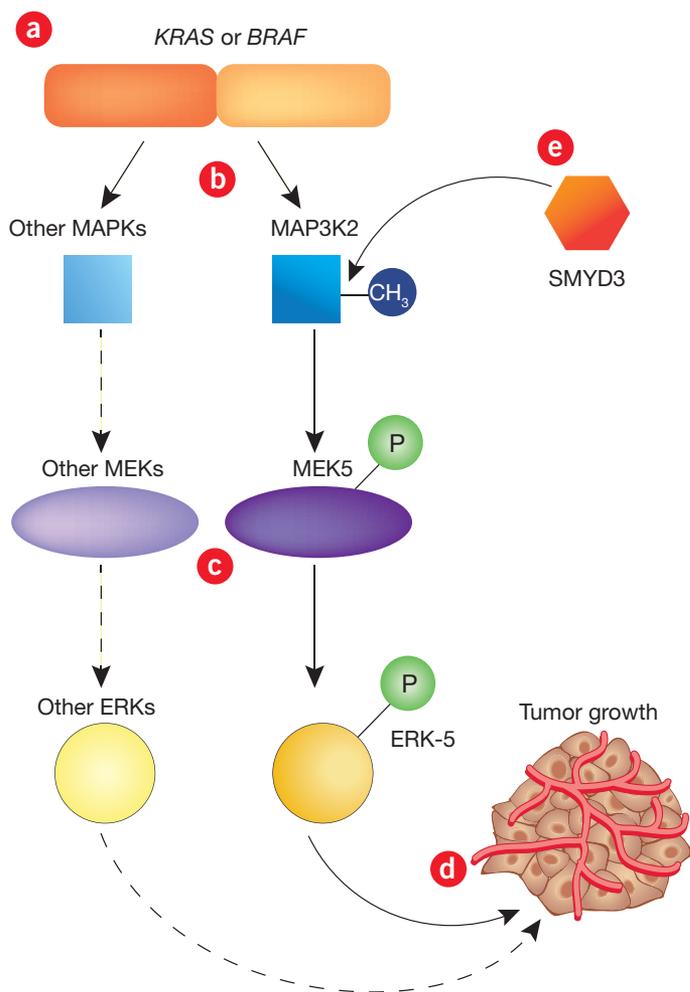


Table 1. Selected MAPK pathway compounds in cancer.

Source: BCIQ: BioCentury Online Intelligence

Company	Compound	Indication	Target	Status
Amgen Inc. (NASDAQ:AMGN)	Nexavar sorafenib	Advanced cancers including colorectal, liver, breast, renal and ovarian cancers	CRAF (RAF1); VEGF receptor	Marketed
GlaxoSmithKline plc (LSE:GSK; NYSE:GSK)	Tafinlar dabrafenib	Melanoma with a BRAF V600E or V600K mutation	BRAF	Marketed
	Mekinist trametinib	Melanoma with a BRAF V600E or V600K mutation	MAP kinase kinase 1 (MAP2K1; MEK1); MEK2 (MAP2K2)	Marketed
Roche (SIX:ROG; OTCQX:RHHBY); Daiichi Sankyo Co. Ltd. (Tokyo:4568); Chugai Pharmaceutical Co. Ltd. (Tokyo:4519)	Zelboraf vemurafenib	Melanoma with a BRAF V600E mutation	BRAF	Marketed
Amgen	Nexavar sorafenib	Thyroid cancers	CRAF; VEGF receptor	Approved
GlaxoSmithKline	Tafinlar dabrafenib plus Mekinist trametinib	Melanoma with a BRAF V600E or V600K mutation	MEK1; MEK2; BRAF	Registration
Amgen	Nexavar sorafenib	Breast, ovarian and liver cancers; colorectal cancer (CRC)	CRAF; VEGF receptor	Phase III
AstraZeneca plc (LSE:AZN; NYSE:AZN); Array BioPharma Inc. (NASDAQ:ARRY)	Selumetinib (AZD6244)	Non-small cell lung cancer (NSCLC)	MEK1; MEK2	Phase III
Exelixis Inc. (NASDAQ:EXEL); Genentech Inc. unit of Roche	Cobimetinib (RG7421)	Melanoma with a BRAF V600E mutation	MEK1; MEK2	Phase III
GlaxoSmithKline	Tafinlar dabrafenib	Solid tumors with BRAF mutation	BRAF	Phase III
Novartis AG (NYSE:NVS; SIX:NOVN)	Encorafenib (LGX818)	Melanoma with BRAF mutations	BRAF	Phase III
Novartis; Array BioPharma	Binimetinib (MEK162)	Melanoma with BRAF mutations; ovarian cancer	MEK1; MEK2	Phase III
AstraZeneca; Bayer AG (Xetra:DAYN)	Refametinib (BAY 86-9766)	Liver cancer	MEK1; MEK2	Phase II
AstraZeneca; Array BioPharma	Selumetinib (AZD6244)	Leukemia; melanoma; thyroid cancer; breast cancer; CRC	MEK1; MEK2	Phase II
GlaxoSmithKline	Tafinlar dabrafenib	NSCLC with BRAF mutations	BRAF	Phase II
	Tafinlar dabrafenib plus Mekinist trametinib	CRC	MEK1; MEK2; BRAF	Phase II
	Mekinist trametinib	Relapsed or refractory solid tumors Second-line treatment of NSCLC with <i>K-Ras</i> (<i>KRAS</i>) mutations Pancreatic cancer	MEK1; MEK2	Phase II
Merck KGaA (Xetra:MRK)	Pimasertib (AS703026)	Melanoma; pancreatic and ovarian cancer	MEK1; MEK2	Phase II
Novartis	Encorafenib (LGX818)	Solid tumors	BRAF	Phase II
Roche; Daiichi Sankyo; Chugai Pharmaceutical	Zelboraf vemurafenib	Thyroid cancer with a BRAF V600E mutation	BRAF	Phase II
AstraZeneca; Bayer	Refametinib (BAY 86-9766)	Pancreatic and biliary duct cancers	MEK1; MEK2	Phase I/II
BioMed Valley Discoveries Inc.	BVD-523	Solid tumors	MAP kinase 1 (MAPK1; ERK-2); MAPK3 (ERK-1)	Phase I/II
GlaxoSmithKline	Tafinlar dabrafenib	Metastatic CRC (mCRC)	BRAF	Phase I/II
	Mekinist trametinib	mCRC Solid tumors with BRAF V600 mutations	MEK1; MEK2	Phase I/II
Novartis	Encorafenib (LGX818)	CRC	BRAF	Phase I/II

(Continues on p. 8)

Table 1. Selected MAPK pathway compounds in cancer. (continued)

Company	Compound	Indication	Target	Status
Teva Pharmaceutical Industries Ltd. (NYSE:TEVA); Ambit Biosciences Corp. (NASDAQ:AMBI)	CEP-32496	Melanoma	BRAF	Phase I/II
Eli Lilly and Co. (NYSE:LLY); Deciphera Pharmaceuticals LLC	LY3009120 (DP-4978)	Melanoma; CRC	BRAF; CRAF; ARAF	Phase I
Exelixis; Genentech	Cobimetinib (RG7421)	Solid tumors	MEK1; MEK2	Phase I
Merck; BeiGene Co. Ltd.	BeiGene-283	<i>BRAF</i> - or <i>KRAS</i> -mutation-positive cancers	BRAF	Phase I
Pfizer Inc. (NYSE:PFE)	PD-0325901 plus PF-05212384	Solid tumors	MEK1; MEK2; mammalian target of rapamycin (mTOR; FRAP; RAFT1); phosphoinositide 3-kinase (PI3K)	Phase I
Roche; Chugai Pharmaceutical	RG7304	Solid tumors	MEK1; CRAF	Phase I
	RG7167	Solid tumors	MEK1; MEK2	Phase I
Takeda Pharmaceutical Co. Ltd. (Tokyo:4502)	TAK-733	Solid tumors	MEK1; MEK2	Phase I

with shRNA knockdown of *SMYD3* in cultured, *KRAS*-driven human pancreatic and lung cancer cells and in mouse xenografts.

The next step was to figure out how *SMYD3* promotes tumor growth. The team knew that the enzyme methylated something, but identifying the substrates required informed guesswork. One clue came from immunohistochemical experiments in murine pancreatic tumors that showed *Smyd3* was predominantly localized in the cytoplasm, not the nucleus.

An *in vitro* screen for cytoplasmic proteins that could be methylated by *SMYD3* pointed to mitogen-activated protein kinase kinase kinase 2 (*MAP3K2*), a key component of a *MAPK* pathway branch that is hyperactive in some tumors.

The team went on to show that *SMYD3* methylates *MAP3K2* in tumor cells, leading to higher *MAPK* pathway signaling than unmethylated *MAP3K2*. Surprisingly, the team found that methylation of *MAP3K2* by *SMYD3* did not directly activate the kinase. Rather, methylation prevented *MAP3K2* from being inactivated by a downstream phosphatase called protein phosphatase 2 (*PPP2CA*; *PP2A*).

“We expected that the methylation mark would recruit something, but in fact it’s the opposite—exclusion of the inhibitor *PP2A*,” said Sage.

Altogether, the findings suggest that *SMYD3* renders *MAP3K2* constitutively active, leading to broad effects on downstream effector proteins that drive tumor growth (see Figure 1, “*SMYD3* in cancer”).

Results were reported in *Nature* and are not patented.

Been there, *SMYD* that

SMYD3 thus appears to be a good target for a subset of *KRAS*-driven tumors. “If you have cancers driven by *KRAS* and have high levels of *SMYD3* or methylation of *MAP3K2*, these are good candidates for *SMYD3* inhibition,” said Gozani.

“This study defines a patient population that might benefit from *SMYD3* inhibitors,” said coauthor Olena Barbash, an oncology R&D investigator at GSK.

For drug developers, the key question is how targeting *SMYD3* compares to hitting *MAP3K2* or other targets in the *MAPK* pathway. “The beauty and the weakness of hitting *SMYD3* is that this is not an essential protein for *MAPK* signaling,” said Sage. “Thus, drugs against *SMYD3* are not going to have as many side effects as direct inhibition of the pathway. On the other hand, they may not be as effective as drugs that hit the main components of the pathway.”

One concern is that *SMYD3* could be a player in just one branch of the multipronged *MAPK* pathway. If so, compensatory changes to other branches of the pathway could render *SMYD3* inhibitors ineffective.

However, the Stanford-GSK team showed that *SMYD3* knockdown reduced phosphorylation of multiple downstream *MAPK* targets, suggesting that *SMYD3* plays a broad role in amplifying proliferative signaling. Thus, it is possible *SMYD3* has other downstream effectors besides *MAP3K2*.

The principal advantage of hitting *SMYD3* over *MAP3K2* may be a better safety profile. Sage said that *Smyd3* knockout mice appear to be healthy, so pharmacological blockade of the target is likely to be well tolerated.

Sage and Gozani think that *SMYD3* inhibitors should be paired with compounds such as Mekinist that target core components of *MAPK* signaling. Indeed, in the *Nature* study Sage cited evidence that *SMYD3*⁻ cells were especially sensitive to Mekinist. *In vitro* and in mouse xenografts, growth of tumor cells lacking *SMYD3* was inhibited by a smaller dose of the compound than that needed for wild-type tumor cells.

“We showed deleting *SMYD3* could improve the effective dose of *MEK* inhibitors,” said Sage. He noted that drug resistance is

(Continues on p. 9)

“If you have cancers driven by *KRAS* and have high levels of *SMYD3* or methylation of *MAP3K2*, these are good candidates for *SMYD3* inhibition.”

—Or Gozani, Stanford University

Stromal uncertainties in pancreatic cancer

By Kai-Jye Lou, Senior Writer

Despite the clear rationale for using hedgehog inhibitors in pancreatic cancer—they deplete the stroma to improve chemotherapy delivery—clinical results have been disappointing. New data suggest that the large amounts of stroma surrounding some pancreatic cancers might actually have protective properties.^{1,2}

The findings could explain the negative clinical data for some hedgehog inhibitors in pancreatic cancer, but it remains to be seen whether the cautionary tale applies to other classes of stroma-depleting therapies. In addition, the data provide a rationale to retry two other classes of cancer drugs in pancreatic cancer—angiogenesis inhibitors and immune checkpoint inhibitors (see Box 1, “Revisiting the pancreas”).

Pancreatic ductal adenocarcinomas (PDACs) account for the majority of pancreatic cancers. PDACs usually have poor perfusion and vascularization and often are surrounded by abundant stroma, which is thought to supply factors that support tumor growth and hinder drug delivery.³

In 2009, an international team led by researchers at **Cancer Research UK** published preclinical data in *Science* suggesting that hedgehog pathway inhibitors could deplete the tumor stroma. The group showed that the small molecule smoothed (SMO) inhibitor saridegib (IPI-926) from **Infinity Pharmaceuticals Inc.** enhanced delivery of the chemotherapeutic gemcitabine and improved survival in mouse models of PDAC.⁴

SMO is a key mediator of hedgehog pathway signaling.

The following year, Infinity began a Phase I/IIb trial of saridegib plus gemcitabine in patients with pancreatic cancer. In January 2012, Infinity stopped the trial after interim data showed that patients receiving the

combination had higher rates of progressive disease and lower overall survival than patients receiving placebo plus gemcitabine. The company discontinued development of saridegib that year after interim analyses suggested it also was going to miss the primary endpoint in a Phase II trial in chondrosarcoma and would not meet prespecified criteria for expansion of a Phase II trial in myelofibrosis.

At least two other SMO antagonists are still in clinical trials for pancreatic cancer. LDE225, from **Novartis AG**, is in a Phase Ib trial in combination with gemcitabine. Erivedge vismodegib, from the **Genentech Inc.** unit of **Roche**, is being tested in combination with various drugs in multiple investigator-led Phase II and Phase I trials.

Interim data from one of those trials showed that Erivedge plus gemcitabine yielded small gains in median progression-free survival (PFS) and overall survival (OS) versus gemcitabine plus placebo.

However, patients on the combination had no complete or partial responses and 49 cases of stable disease. There were 3 complete responses, 11 partial responses and 31 cases of stable disease in patients receiving gemcitabine alone.

Genentech markets Erivedge to treat metastatic basal cell carcinoma. The company said that it does not have any clinical trials of Erivedge in pancreatic cancer and declined to comment further.

Novartis did not respond to requests for comment, and Infinity declined to comment.

“I think these new data mean we need to be cognizant of the possibility that some classes of anti-stromal compounds have the potential to make pancreatic cancers more aggressive.”

—**Kenneth Olive,**
Columbia University Medical Center

Stromal uncertainties

A pair of papers in *Cancer Cell* could explain the weak showing for SMO inhibitors in pancreatic cancer.

In a study co-led by researchers from the **Columbia University Medical Center (CUMC)** and **Perelman School of Medicine at the University of Pennsylvania**, *sonic hedgehog homolog (Shh)* knockout or chronic treatment with saridegib in a genetic mouse model of PDAC decreased the tumor stroma compared with no alteration or treatment. However, both approaches led to the development of aggressive, poorly

(Continues on p. 10)

(Continued from “Putting SMYD3 on the MAP,” p. 8)

more likely to develop when a single drug is given at high doses, so achieving a potent antiproliferative effect with a lower dose of Mekinist “together with SMYD3 inhibitors could reduce the potential for resistance.”

The next biology question is whether SMYD3 plays a role in other tumors besides the *KRAS*-mutant pancreatic and lung cancers examined in the *Nature* paper. “It will be important to understand whether the modulation of MAPK activity by SMYD3 could be used outside of *KRAS*-driven tumors,” said Barbash. She noted that MAPK pathway abnormalities also occur in some liver cancers.

Osherovich, L. *SciBX* 7(23); doi:10.1038/scibx.2014.664
Published online June 12, 2014

REFERENCES

- Mazur, P.K. *et al. Nature*; published online May 21, 2014; doi:10.1038/nature13320
Contact: Julien Sage, Stanford University School of Medicine, Stanford, Calif.
e-mail: julsage@stanford.edu
Contact: Or Gozani, Stanford University, Stanford, Calif.
e-mail: ogozani@stanford.edu
- Cain, C. *SciBX* 7(19); doi:10.1038/scibx.2014.545

COMPANIES AND INSTITUTIONS MENTIONED

Epizyme Inc. (NASDAQ:EPZM), Cambridge, Mass.
GlaxoSmithKline plc (LSE:GSK; NYSE:GSK), London, U.K.
Stanford University, Stanford, Calif.
Stanford University School of Medicine, Stanford, Calif.

Box 1. Revisiting the pancreas.

The insights into the nature of these aggressive, poorly differentiated pancreatic tumors suggest a niche for two other classes of cancer treatments—angiogenesis inhibitors and immune checkpoint inhibitors.

Additional data published in the **Columbia University Medical Center–Perelman School of Medicine at the University of Pennsylvania** study showed that the poorly differentiated tumors in *sonic hedgehog homolog (Shh)*-deficient or saridegib-treated mice were more vascularized than those from nondeficient mice or vehicle-treated controls.¹ In the *Shh*^{-/-} mice, a VEGF receptor–blocking mAb increased survival compared with IgG control.

Ben Stanger, an assistant professor of medicine at the Perelman School of Medicine, noted that even though antiangiogenic therapies have failed in the overall PDAC patient population, the mouse data in his study suggest that the subset of patients with poorly differentiated tumors could benefit.

“In PDAC, about 10% of the patients have poorly differentiated tumors, and based on what we’ve seen in our mouse studies, such tumors might also have higher blood vessel density and be more dependent on the vascular network. If this is the case, it might mean that such tumors will be responsive to antiangiogenic therapies,” he said.

Meanwhile, **The University of Texas MD Anderson Cancer Center** researchers showed that depletion of actin $\alpha 2$ smooth aorta muscle (*Acta2*; α -Sma)-positive myofibroblasts in the mouse PDAC model resulted in a more immunosuppressive tumor microenvironment and increased expression of the immune checkpoint protein *Ctla-4* (Cd152) compared with no alteration.² In the myofibroblast-depleted mice, an anti-CTLA4 mAb decreased PDAC progression and increased survival compared with IgG control.

Raghu Kalluri, chair of the Department of Cancer Biology at MD Anderson, noted that the additional data from his group suggest that stroma depletion could create an immune profile in PDAC tumors that renders them sensitive to inhibitors against CTLA-4 and other immune checkpoint proteins. He said that stratifying patients based on their stroma could help identify those that would benefit most from such therapies.

Data from a Phase II trial published in 2010 showed that monotherapy with the anti-CTLA-4 mAb Yervoy ipilimumab was ineffective in advanced PDAC.⁷ **Bristol-Myers Squibb Co.** markets Yervoy to treat unresectable or metastatic melanoma. —K-JL

differentiated tumors, and treated animals had lower survival rates than controls. In the mouse PDAC model, gemcitabine plus saridegib failed to increase survival compared with gemcitabine alone.

The reason why the findings run counter to the 2009 *Science* paper likely relates to when the mice started treatment. In the earlier study, researchers began treating the mice after large PDAC tumors developed. Treatment duration was limited to a few weeks because of the short survival time of these animals. In the new study, mice received treatment earlier—they had precancerous pancreatic lesions as opposed to established PDAC tumors.

Kenneth Olive said that the goal of the work published in 2009 was to recapitulate the clinical situation in PDAC, in which most patients are diagnosed with advanced disease. However, he noted that the mice were ill-suited for evaluating longer-term treatment effects as they rapidly succumbed to disease.

Olive is a member of the Herbert Irving Comprehensive Cancer Center at CUMC and an assistant professor of medicine and pathology at CUMC. He was the lead author on the 2009 *Science* paper and co-corresponding author on the current CUMC-UPenn study.

Olive said that his group started trying to figure out why saridegib failed in pancreatic cancer within hours after Infinity announced the trial halt. The team homed in on the possibility of long-term treatment effects several months later.

The results of the CUMC-UPenn study were corroborated in a separate study in PDAC mice led by researchers at **The University of Texas MD Anderson Cancer Center**. The Texas group showed that depletion of the major subset of hedgehog-responsive cells in the tumor stroma resulted in decreased survival and the development of aggressive, poorly differentiated tumors. Specifically, the subset was myofibroblasts that were positive for actin $\alpha 2$ smooth aorta muscle (*ACTA2*; α -SMA).

The MD Anderson–led group also analyzed tumor samples from a cohort of 53 patients with PDAC and showed that low levels of α -SMA were associated with decreased overall survival ($p=0.0053$).

“Our original hypothesis had been that if we eliminated stromal fibroblasts, we would decrease cancer progression and metastasis, but the results we got were the complete opposite of that,” said Raghu Kalluri, a professor and chair of the Department of Cancer Biology at MD Anderson and corresponding author on the MD Anderson study.

“I think these new data mean we need to be cognizant of the possibility that some classes of anti-stromal compounds have the potential to make pancreatic cancers more aggressive and that the effects you see in the mouse over days and weeks may not be the same as what you see over the longer term,” Olive told *SciBX*.

“The past work has suggested that if you briefly deplete stroma, you could get benefit from improved drug delivery, but the current studies suggest the consequences on tumor biology from long-term stroma depletion outweigh the delivery benefit,” added Ben Stanger, an assistant professor of medicine at the Perelman School of Medicine and the other co-corresponding author on the CUMC-UPenn study.

David Tuveson cautioned against generalizing the observations to other classes of stroma-depleting therapies being developed for pancreatic cancer.

“It is not yet known whether going after other targets in the tumor stroma would have the same effect as depletion of stromal fibroblasts or

“The current studies suggest the consequences on tumor biology from long-term stroma depletion outweigh the delivery benefit.”

—Ben Stanger,
Perelman School of Medicine at the
University of Pennsylvania

hedgehog inhibition,” said Tuveson. He is director of the **Lustgarten Foundation** Pancreatic Cancer Research Laboratory, a professor at **Cold Spring Harbor Laboratory** and was senior author on the 2009 *Science* paper.

Edward Kim, an assistant professor of medicine at the **University of California, Davis Comprehensive Cancer Center**, said that the current data raise the question of whether the detrimental effects of targeting stromal cells is due to depletion of the stroma itself or other effects. “Attacking the stromal cells results in decreased stroma, but this clearly leads to many more changes than just decreased stroma. These results show targeting stromal cells may not be the answer but fail to elucidate whether removing the acellular stroma may still provide benefit by improving drug delivery,” he said.

At least one company is developing a stroma-depleting therapy in pancreatic cancer that goes after an acellular stromal component. Last year, **Halozyne Therapeutics Inc.** reported data from the Phase Ib portion of a Phase I/II trial in which i.v. PEGPH20 plus gemcitabine resulted in an overall response rate of 42% in 24 evaluable patients. In a post hoc subgroup analysis, patients with high levels of tumor-associated hyaluronan showed the greatest survival benefit, with double the PFS and triple the OS of patients with low levels of tumor-associated hyaluronan.

Hyaluronan is a glycosaminoglycan found throughout the body and a component of the tumor stroma in PDAC.

PEGPH20 is a recombinant human PH20 hyaluronidase (sperm adhesion molecule 1; SPAM1; PH20) enzyme conjugated to polyethylene glycol (PEG). Hyaluronidase digests hyaluronan to decrease interstitial fluid pressure around the tumor, leading to improved blood flow and drug delivery.

Halozyne VP and CSO H. Michael Shepard thinks that the negative effects described in the *Cancer Cell* papers are a cautionary tale for new therapies that target myofibroblasts but probably not for PEGPH20.

He said that the roles of hyaluronan and myofibroblasts in the tumor stroma are very distinct, citing recent studies showing that PEGPH20-mediated depletion of hyaluronan plus gemcitabine increased survival in the same mouse PDAC models.^{5,6}

New insights

Kalluri said that his group is now working to elucidate the role of various stromal cell populations in PDAC.

“There are multiple types of fibroblasts in the pancreatic cancer stroma, so we want to determine whether all stromal fibroblasts have a protective role or just the α -SMA fibroblasts. We also want to determine what various other cellular populations in the stroma are doing and how they affect tumor immunosurveillance,” he said.

“Without a better understanding of what the stroma is doing in patients, we can’t really determine where anti-stromal therapies will be helpful and where they could be harmful.”

—Mert Erkan,
Technical University Munich

“Without a better understanding of what the stroma is doing in patients, we can’t really determine where anti-stromal therapies will be helpful and where they could be harmful,” added Mert Erkan, a consultant surgeon and head of the Division of Pancreatic Surgery at **Technical University Munich**.

Erkan noted that the tumor stroma in patients with pancreatic cancer is very heterogeneous—both in amount and composition—and that the consequences of such differences are unclear.

The findings reported in the two new studies are not patented.

Lou, K.-J. *SciBX* 7(23); doi:10.1038/scibx.2014.665
Published online June 12, 2014

REFERENCES

- Rhim, A.D. *et al. Cancer Cell*; published online May 22, 2014; doi:10.1016/j.ccr.2014.04.021
Contact: Ben Z. Stanger, Perelman School of Medicine at the University of Pennsylvania, Philadelphia, Pa.
e-mail: bstanger@exchange.upenn.edu
Contact: Kenneth P. Olive, Columbia University Medical Center, New York, N.Y.
e-mail: kenolive@columbia.edu
- Özdemir, B.C. *et al. Cancer Cell*; published online May 22, 2014; doi:10.1016/j.ccr.2014.04.005
Contact: Raghu Kalluri, The University of Texas MD Anderson Cancer Center, Houston, Texas
e-mail: rkalluri@mdanderson.org
- Erkan, M. *et al. Nat. Rev. Gastroenterol. Hepatol.* **9**, 454–467 (2012)
- Olive, K.P. *et al. Science* **324**, 1457–1461 (2009)
- Jacobetz, M.A. *et al. Gut* **62**, 112–120 (2013)
- Provenzano, P.P. *et al. Cancer Cell* **21**, 418–429 (2012)
- Royal, R.E. *et al. J. Immunother.* **33**, 828–833 (2010)

COMPANIES AND INSTITUTIONS MENTIONED

Bristol-Myers Squibb Co. (NYSE:BMJ), New York, N.Y.
Cancer Research UK, London, U.K.
Cold Spring Harbor Laboratory, Cold Spring Harbor, N.Y.
Columbia University Medical Center, New York, N.Y.
Genentech Inc., South San Francisco, Calif.
Halozyne Therapeutics Inc. (NASDAQ:HALO), San Diego, Calif.
Infinity Pharmaceuticals Inc. (NASDAQ:INFI), Cambridge, Mass.
Lustgarten Foundation, New York, N.Y.
Novartis AG (NYSE:NVS; SIX:NOVN), Basel, Switzerland
Roche (SIX:ROG; OTCQX:RHHBY), Basel, Switzerland
Perelman School of Medicine at the University of Pennsylvania, Philadelphia, Pa.
Technical University Munich, Munich, Germany
University of California, Davis Comprehensive Cancer Center, Davis, Calif.
The University of Texas MD Anderson Cancer Center, Houston, Texas

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
Autoimmune disease				
Multiple sclerosis (MS)	Lipocalin (LCN2; NGAL)	<i>In vitro</i> and mouse studies suggest inhibiting LCN2 could help treat MS. In an experimental autoimmune encephalomyelitis (EAE) mouse model of MS, compared with wild-type mice, <i>Lcn2</i> expression was increased in the spinal cord, lymph nodes and spleen. In the EAE model, <i>Lcn2</i> knockout decreased disease severity, inflammatory cell infiltration into the spinal cord and demyelination compared with no alteration. Next steps include developing pharmacological inhibitors of LCN2 signaling.	Findings unpatented; available for licensing	Nam, Y. <i>et al. J. Biol. Chem.</i> ; published online May 7, 2014; doi:10.1074/jbc.M113.542282 Contact: Kyoungsook Suk, Kyungpook National University School of Medicine, Daegu, South Korea e-mail: ksuk@knu.ac.kr
SciBX 7(23); doi:10.1038/scibx.2014.666 Published online June 12, 2014				
Cancer				
Breast cancer	Complement component 5a receptor 1 (C5AR1; C5AR)	Mouse studies suggest inhibiting C5AR could help prevent breast cancer metastasis. In a mouse model of metastatic breast cancer, knockout or pharmacological inhibition of C5ar1 decreased metastasis to the lung and liver compared with no knockout or inhibition. Next steps include investigating whether C5AR mediates the recruitment of myeloid-derived suppressor cells and metastasis to other organs including the brain.	Patent application filed; unavailable for licensing	Vadrevu, S.K. <i>et al. Cancer Res.</i> ; published online May 1, 2014; doi:10.1158/0008-5472.CAN-14-0157 Contact: Maciej M. Markiewski, Texas Tech University Health Sciences Center, Abilene, Texas e-mail: maciej.markiewski@ttuhsc.edu
SciBX 7(23); doi:10.1038/scibx.2014.667 Published online June 12, 2014				
Cancer	3-Oxoacyl-ACP synthase mitochondrial (OXSM); fatty acid synthase (FASN; FAS)	Cell culture studies suggest increasing OXSM signaling with antioxidants such as α -lipoic acid could help protect normal cells from the toxic effects of C75, a FAS inhibitor known to have anticancer activity. In a normal human embryonic kidney cell line, α -lipoic acid supplementation decreased C75 challenge-induced mitochondrial dysfunction and increased cell viability compared with no supplementation. In cell culture, vector-induced OXSM overexpression also protected against C75 challenge-induced mitochondrial dysfunction and toxicity compared with wild-type OXSM expression. Next steps include confirmatory preclinical studies in mouse models. C75 is a research reagent.	Patent application filed covering use of α -lipoic acid and C75 in cancer treatment; unavailable for licensing	Chen, C. <i>et al. J. Biol. Chem.</i> ; published online May 1, 2014; doi:10.1074/jbc.M114.550806 Contact: Zhihui Feng, Xi'an Jiaotong University, Xi'an, China e-mail: zhfeng@mail.xjtu.edu.cn Contact: Jiankang Liu, same affiliation as above e-mail: j.liu@mail.xjtu.edu.cn
SciBX 7(23); doi:10.1038/scibx.2014.668 Published online June 12, 2014				
Cancer	ADAM10; ADAM17	<i>In vitro</i> studies suggest inhibiting ADAM10 and ADAM17 could help stimulate an NK cell immune response to treat cancer. In cultured cancer cell lines, ADAM inhibitors or siRNAs targeting ADAM10 or ADAM17 increased cell surface expression of an NK cell target, B7-H6 tumor cell ligand, compared with vehicle or control siRNAs. Next steps include evaluating the effect of ADAM10 and ADAM17 inhibitors in animal models.	Patent and licensing status unavailable	Schlecker, E. <i>et al. Cancer Res.</i> ; published online April 29, 2014; doi:10.1158/0008-5472.CAN-13-3017 Contact: Adelheid Cerwenka, German Cancer Research Center, Heidelberg, Germany e-mail: a.cerwenka@dkfz.de
SciBX 7(23); doi:10.1038/scibx.2014.669 Published online June 12, 2014				

This week in therapeutics (continued)

Indication	Target/marker/pathway	Summary	Licensing status	Publication and contact information
Cancer	AT rich interactive domain 1A (ARID1A); ARID1B	Cell culture studies suggest inhibiting ARID1A or ARID1B could sensitize tumors to radiotherapy and chemotherapy. In cultured cells, siRNA knockdown of <i>ARID1A</i> , <i>ARID1B</i> and other SWI/SNF complex genes decreased accumulation of repair proteins on DNA in response to radiation and increased cisplatin-induced cell death compared with control siRNA. Next steps could include determining whether tumors with mutations in <i>ARID1A</i> or <i>ARID1B</i> are more sensitive to DNA-damaging therapies in animal models. SciBX 7(23); doi:10.1038/scibx.2014.670 Published online June 12, 2014	Patent and licensing status unavailable	Watanabe, R. <i>et al. Cancer Res.</i> ; published online May 1, 2014; doi:10.1158/0008-5472.CAN-13-3608 Contact: Akira Yasui, Tohoku University, Sendai, Japan e-mail: ayasui@idac.tohoku.ac.jp
Cancer	CXC chemokine receptor 2 (CXCR2; IL8RB); programmed cell death 1 (PDCD1; PD-1; CD279)	<i>In vitro</i> and mouse studies suggest CXCR2-targeted antibodies could improve the therapeutic efficacy of PD-1 antibodies in cancer. Immunosuppressive cells in the tumor microenvironment such as myeloid-derived suppressor cells can limit the efficacy of PD-1 antibodies against cancer. In a mouse model of rhabdomyosarcoma, numbers of Cxcr2-expressing, myeloid-derived suppressor cells increased as the tumors developed. In the animals, a CXCR2 antibody plus PD-1 antibody improved survival and caused more potent tumor growth inhibition than either antibody alone. Next steps include evaluating CXCR2 blockade with other cancer immunotherapies. Dompe Farmaceutici S.p.A. has the CXCR2 inhibitor reperixin in Phase III testing to treat graft rejection. At least four other companies have CXCR2 antagonists in Phase II or earlier testing to treat various pulmonary and inflammatory indications. Merck & Co. Inc. has the PD-1 antibody lambrolizumab under FDA review to treat melanoma. Ono Pharmaceutical Co. Ltd. and Bristol-Myers Squibb Co. have the PD-1 antibody nivolumab under review in Japan to treat melanoma and under FDA review for non-small cell lung cancer (NSCLC). At least five other companies have anti-PD-1 antibodies in Phase II or earlier testing to treat various cancers. SciBX 7(23); doi:10.1038/scibx.2014.671 Published online June 12, 2014	Findings unpatented; licensing status not applicable	Highfill, S.L. <i>et al. Sci. Transl. Med.</i> ; published online May 21, 2014; doi:10.1126/scitranslmed.3007974 Contact: Crystal L. Mackall, National Institutes of Health, Bethesda, Md. e-mail: cm35c@nih.gov Contact: Steven L. Highfill, same affiliation as above e-mail: steven.highfill@nih.gov
Cancer	Hypoxia-inducible factor 1 (HIF1); E1A binding protein p300 (EP300; p300)	<i>In vitro</i> and mouse studies suggest inhibiting the HIF1-p300 interaction could help treat cancer. <i>In vitro</i> , an oxopiperizine mimic for a helix at the HIF1-p300 interface bound to p300 with a K_D of 38 nM. In cultured human cancer cell lines, the helical mimic decreased expression of HIF1-dependent genes compared with inactive analogs. In a mouse xenograft model of breast cancer, the mimic decreased tumor cell proliferation and tumor size compared with saline. Next steps include using a computational approach and non-natural amino acids to improve the potency of the compound. At least seven companies have compounds that inhibit HIF1 in Phase II or earlier testing to treat various cancers. SciBX 7(23); doi:10.1038/scibx.2014.672 Published online June 12, 2014	Patent applications filed covering nonpeptidic helical mimics and their application to modulation of hypoxia signaling; licensing status not applicable	Lao, B.B. <i>et al. Proc. Natl. Acad. Sci. USA</i> ; published online May 12, 2014; doi:10.1073/pnas.1402393111 Contact: Paramjit S. Arora, New York University, New York, N.Y. e-mail: arora@nyu.edu Contact: Bogdan Z. Olenyuk, University of Southern California, Los Angeles, Calif. e-mail: bogdan@usc.edu

This week in therapeutics (continued)

Indication	Target/marker/pathway	Summary	Licensing status	Publication and contact information
Cancer	Insulin-like growth factor binding protein 2 (IGFBP2)	<p>Mouse studies suggest an N-terminal IGFBP2 vaccine could help treat IGFBP2-overexpressing tumors. In mice with such tumors, a vaccine encoding an epitope of the IGFBP2 N-terminal region induced an antitumor immune response and decreased tumor volume compared with vehicle or vaccines encoding full-length IGFBP2 or a C-terminal epitope. An investigator-led Phase I trial of the vaccine in patients with ovarian cancer is ongoing.</p> <p>SciBX 7(23); doi:10.1038/scibx.2014.673 Published online June 12, 2014</p>	Patent application filed; technology under negotiation for licensing by an undisclosed company; available for licensing	<p>Cecil, D.L. <i>et al. Cancer Res.</i>; published online April 28, 2014; doi:10.1158/0008-5472.CAN-13-3286 Contact: Mary L. Disis, University of Washington, Seattle, Wash. e-mail: ndisis@uw.edu</p>
Colorectal cancer	Pre-mRNA processing factor 6 (PRPF6)	<p>Mouse and cell culture studies suggest inhibiting PRPF6 signaling could help treat colon cancer. In 10 of 11 human colon cancer cell lines with high PRPF6 protein levels, PRPF6-targeting siRNA decreased growth compared with nontargeting siRNA. In a mouse xenograft model of established colon cancers with high PRPF6 levels, PRPF6-targeting siRNA induced tumor shrinkage and decreased tumor growth compared with nontargeted shRNA. Transcriptome analysis showed that inhibiting PRPF6 signaling alters alternative splicing of multiple genes, including an oncogenic <i>sterile α-motif and leucine zipper containing kinase</i> AZK (ZAK) kinase isoform. Researchers did not disclose next steps, which could include screening for pharmacological inhibitors of PRPF6.</p> <p>SciBX 7(23); doi:10.1038/scibx.2014.674 Published online June 12, 2014</p>	Patent and licensing status undisclosed	<p>Adler, A.S. <i>et al. Genes Dev.</i>; published online May 1, 2014; doi:10.1101/gad.237206.113 Contact: Ron Firestein, Genentech Inc., South San Francisco, Calif. e-mail: ronf@gene.com</p>
Glioblastoma	Gremlin 1 (GREM1)	<p><i>In vitro</i> and mouse studies suggest inhibiting GREM1 could help treat glioblastoma. In human glioma cancer stem cells (CSCs), expression of the bone morphogenetic protein (BMP) antagonist GREM1 was greater than that in glioma non-CSCs. In human glioma CSCs, GREM1 suppressed BMP2-mediated inhibition of proliferation. In mice receiving an intracranial injection of human CSCs, GREM1-targeted shRNA prevented tumor formation and increased survival compared with nontargeted shRNA. Next steps could include screening for a pharmacological GREM1 inhibitor.</p> <p>SciBX 7(23); doi:10.1038/scibx.2014.675 Published online June 12, 2014</p>	Patent and licensing status unavailable	<p>Yan, K. <i>et al. Genes Dev.</i>; published online May 1, 2014; doi:10.1101/gad.235515.113 Contact: Jeremy N. Rich, Cleveland Clinic, Cleveland, Ohio e-mail: richj@ccf.org</p>
Hematologic malignancies	Serine/threonine kinase 4 (STK4); yes-associated protein 1 (YAP1)	<p><i>In vitro</i> and mouse studies suggest inhibiting STK4 could help treat hematological malignancies, which often have downregulated expression of the proapoptotic protein YAP1. In multiple myeloma (MM) cells, lentiviral delivery of YAP1 restored YAP1-mediated apoptotic pathways and induced cancer cell death. In mice injected with human MM cells, shRNA knockdown of STK4 restored YAP1 expression and prevented tumor growth, whereas scrambled shRNA resulted in the development of tumors. Next steps include designing STK4 inhibitors.</p> <p>SciBX 7(23); doi:10.1038/scibx.2014.676 Published online June 12, 2014</p>	Patent application filed; available for licensing	<p>Cottini, F. <i>et al. Nat. Med.</i>; published online May 11, 2014; doi:10.1038/nm.3562 Contact: Giovanni Tonon, Scientific Institute for Hospitalization and Care, San Raffaele Scientific Institute, Milan, Italy e-mail: tonon.giovanni@hsr.it Contact: Kenneth C. Anderson, Harvard Medical School, Boston, Mass. e-mail: kenneth_anderson@dfci.harvard.edu</p>

This week in therapeutics (continued)

Indication	Target/marker/pathway	Summary	Licensing status	Publication and contact information
Multiple myeloma (MM)	CD28	<i>In vitro</i> and mouse studies suggest blocking the interaction between CD28 and its ligands could help prevent chemotherapy resistance in MM. In a coculture of MM and dendritic cells, an antibody that blocks the interaction between CD28 on MM cells and its ligand on dendritic cells resensitized the former to melphalan chemotherapy. In a mouse model of MM, melphalan plus the antibody decreased tumor burden compared with either treatment alone. Next steps include evaluating antibodies that block CD28 activity in patients with MM. Bristol-Myers Squibb Co. markets Orenzia abatacept, an antibody that targets CD28 ligands, to treat rheumatoid arthritis (RA). Effimune S.A.S. has the anti-CD28 antibody fragment FR104 in preclinical development to treat organ transplant rejection.	Patent application filed; available for licensing	Murray, M.E. <i>et al. Blood</i> ; published online April 29, 2014; doi:10.1182/blood-2013-10-530964 Contact: Kelvin P. Lee, Roswell Park Cancer Institute, Buffalo, N.Y. e-mail: kelvin.lee@roswellpark.org
Cardiovascular disease				
Cardiovascular disease; myocardial infarction (MI)	Triiodothyronine (T3); insulin-like growth factor-1 (IGF-1)	Mouse studies suggest IGF-1 or the thyroid hormone T3 could help promote cardiac repair in pediatric patients. In normal preadolescent mice that were 14–18 days old, elevated levels of cardiac Igf-1 or serum T3 increased the number of cardiomyocytes by 40% compared with baseline. Mice subjected to MI on postnatal day 15 developed smaller infarcts and retained higher cardiac function than mice subjected to MI on postnatal day 21. Ongoing work includes testing T3 and Igf-1 therapy in preadolescent mouse models of cardiac injury.	Unpatented; licensing status not applicable	Naqvi, N. <i>et al. Cell</i> ; published online May 8, 2014; doi:10.1016/j.cell.2014.03.035 Contact: Ahsan Husain, Emory University School of Medicine, Atlanta, Ga. e-mail: ahusai2@emory.edu Contact: Robert M. Graham, Victor Chang Cardiac Research Institute, Darlinghurst, New South Wales, Australia e-mail: b.graham@victorchang.edu.au
Dermatology				
Wounds	Leukotriene B4 type 2 receptor (BLT2)	Mouse studies suggest BLT2 agonists could help promote wound healing. In mice, <i>Bltl2</i> knockout or inhibition of Blt2 signaling with aspirin delayed wound healing. In diabetic mice, a synthetic BLT2 agonist accelerated wound healing, whereas vehicle control did not. Next steps include screening a chemical library for a more potent BLT2 agonist and evaluating its safety and efficacy.	Patent status undisclosed; available for licensing	Liu, M. <i>et al. J. Exp. Med.</i> ; published online May 12, 2014; doi:10.1084/jem.20132063 Contact: Takehiko Yokomizo, Juntendo University School of Medicine, Tokyo, Japan e-mail: yokomizo-tyk@umin.ac.jp Contact: Kazuko Saeki, same affiliation as above e-mail: ksaeki@juntendo.ac.jp
Endocrine/metabolic disease				
Diabetes	Abhydrolase domain containing 6 (ABHD6)	Cell culture and mouse studies suggest inhibiting ABHD6 could help treat diabetes. In cultured β cells, <i>ABHD6</i> -targeted siRNA increased glucose-stimulated insulin secretion compared with control siRNA. In a mouse model of diabetes, inhibition of <i>Abhd6</i> normalized blood glucose levels and increased insulin secretion and glucose tolerance compared with no inhibition. Next steps include developing pharmacological ABHD6 inhibitors and mechanistic studies to better understand the relationship between ABHD6 and insulin secretion.	Patent and licensing status undisclosed	Zhao, S. <i>et al. Cell Metab.</i> ; published online May 8, 2014; doi:10.1016/j.cmet.2014.04.003 Contact: Marc Prentki, University of Montreal, Montreal, Quebec, Canada e-mail: marc.prentki@umontreal.ca Contact: S.R. Murthy Madiraju, same affiliation as above e-mail: murthy.madiraju@crchum.qc.ca

This week in therapeutics (continued)

Indication	Target/marker/pathway	Summary	Licensing status	Publication and contact information
Obesity	Poly(ADP-ribose) polymerase (PARP)	<p><i>In vitro</i> and mouse studies suggest PARP inhibition could help treat muscle dysfunction caused by mitochondrial defects. In mice fed a high-fat diet, a PARP inhibitor decreased weight gain and increased energy expenditure compared with vehicle and did not alter food intake. In the mouse model, long-term treatment with a PARP inhibitor did not cause genomic instability or observable toxicities, which are potential concerns with use of PARP inhibition in indications other than cancer. In human skin fibroblasts with impaired mitochondrial activity and skeletal muscle cells from obese patients, a PARP inhibitor increased mitochondrial function compared with vehicle. Next steps could include testing PARP inhibitors in additional models of metabolic dysfunction.</p> <p>AstraZeneca plc has the PARP inhibitor Olaparib in FDA and EMA review to treat ovarian cancer. At least 12 other companies have PARP inhibitors in Phase III or earlier testing to treat various cancers.</p> <p>SciBX 7(23); doi:10.1038/scibx.2014.681 Published online June 12, 2014</p>	Patent and licensing status unavailable	<p>Pirinen, E. <i>et al. Cell Metab.</i>; published online May 8, 2014; doi:10.1016/j.cmet.2014.04.002 Contact: Johan Auwerx, Swiss Federal Institute of Technology Lausanne, Lausanne, Switzerland e-mail: admin.auwerx@epfl.ch Contact: Carles Cantó, Nestlé Institute of Health Sciences, Lausanne, Switzerland e-mail: carlos.cantoalvarez@rd.nestle.com</p>
Neurology				
Alzheimer's disease (AD)	Inositol 1,4,5-triphosphate receptor (ITPR; IP3R)	<p>Mouse studies suggest antagonizing IP3R could be useful for treating AD. In two mouse models of AD, an engineered loss-of-function mutation that lowered <i>Ip3r</i> expression by 50% improved hippocampal function and decreased pathological intracellular calcium levels compared with wild-type <i>Ip3r</i> expression. In one of the models, lower <i>Ip3r</i> expression decreased levels of the AD markers β-amyloid (Aβ) and hyperphosphorylated microtubule-associated protein-τ (MAPT; tau; FTDP-17) compared with those seen in wild-type controls. Next steps could include examining the effect of <i>Ip3r</i> mutations on neurodegeneration and evaluating IP3R antagonists in AD models.</p> <p>SciBX 7(23); doi:10.1038/scibx.2014.682 Published online June 12, 2014</p>	Patent and licensing status undisclosed	<p>Shilling, D. <i>et al. J. Neurosci.</i>; published online May 14, 2014; doi:10.1523/JNEUROSCI.5441-13.2014 Contact: J. Kevin Foskett, University of Pennsylvania, Philadelphia, Pa. e-mail: foskett@mail.med.upenn.edu</p>
Other				
Adjuvant	Not applicable	<p><i>In vitro</i> and mouse studies suggest modified K-class CpG oligodeoxynucleotides (K-ODNs) could be useful as vaccine adjuvants. In mice vaccinated against foot and mouth disease, a modified K-ODN nanoring adjuvant induced a stronger T helper type 1 (Th1) immune response than unmodified K-ODN or K-ODN mixed with a human peptide. In mice receiving a tumor vaccine, the K-ODN nanoring adjuvant induced a more potent antitumor response than unmodified K-ODN. Next steps include testing the adjuvants in primate models.</p> <p>SciBX 7(23); doi:10.1038/scibx.2014.683 Published online June 12, 2014</p>	Patent status available from principal investigator; available for licensing	<p>Gungor, B. <i>et al. Sci. Transl. Med.</i>; published online May 7, 2014; doi:10.1126/scitranslmed.3007909 Contact: Mayda Gursel, Middle East Technical University, Ankara, Turkey e-mail: mgursel@metu.edu.tr</p>

This week in therapeutics (continued)

Indication	Target/marker/pathway	Summary	Licensing status	Publication and contact information
Various				
Atherosclerosis; obesity	Inhibitor of κ -light polypeptide gene enhancer in B cells kinase- β (IKBKB; IKK2)	<p>Mouse studies suggest inhibiting IKBKB could help treat atherosclerosis. In a mouse model of atherosclerosis, knockout of <i>Ikkbb</i> in smooth muscle and pre-adipose cells decreased the size of atherosclerotic lesions and expression of proinflammatory genes within the lesions compared with no knockout. In mice, <i>Ikkbb</i> knockout also prevented high-fat diet-induced obesity and obesity-associated pathology including glucose intolerance and hyperlipidemia. Next steps include developing small molecules or biologics that inhibit IKBKB.</p> <p>Sanofi's IKBKB inhibitor, SAR113945, is in Phase II testing to treat osteoarthritis (OA).</p> <p>IMMD Inc. has the oral IKBKB inhibitor IMD-1041 in Phase II testing to treat chronic obstructive pulmonary disease (COPD) and pulmonary fibrosis.</p> <p>EntreChem S.L. has the selective IKBKB inhibitor EC-70124 in preclinical development for cancer.</p> <p>SciBX 7(23); doi:10.1038/scibx.2014.684 Published online June 12, 2014</p>	Unpatented; licensing status not applicable	<p>Sui, Y. <i>et al. J. Exp. Med.</i>; published online May 5, 2014; doi:10.1084/jem.20131281 Contact: Changcheng Zhou, University of Kentucky, Lexington, K.Y. e-mail: c.zhou@uky.edu</p>
Itch; pain	Nav1.7 (SCN9A)	<p><i>In vitro</i> and mouse studies suggest an anti-Nav1.7 antibody could help treat itch and pain. In human embryonic kidney cells expressing Nav channel subtypes, an antibody targeting the voltage sensor paddle of Nav1.7 specifically inhibited sodium channel currents through that channel subtype. In mouse models of inflammatory and neuropathic pain, intrathecal or i.v. delivery of the antibody decreased pain compared with delivery of a control antibody. In mouse models of acute and chronic itch, the antibody decreased itch compared with a control antibody. Next steps include creating a humanized mAb with improved half-life.</p> <p>Convergence Pharmaceuticals Ltd. has CNV1014802, a Nav1.7 inhibitor, in Phase II testing to treat pain. At least four other companies have Nav1.7 inhibitors in Phase I or earlier testing to treat pain (<i>see Nav-i-gating antibodies for pain, page 1</i>).</p> <p>SciBX 7(23); doi:10.1038/scibx.2014.685 Published online June 12, 2014</p>	Patent application filed; available for licensing	<p>Lee, J.-H. <i>et al. Cell</i>; published online May 22, 2014; doi:10.1016/j.cell.2014.03.064 Contact: Seok-Yong Lee, Duke University Medical Center, Durham, N.C. e-mail: sylee@biochem.duke.edu Contact: Ru-Rong Ji, same affiliation as above e-mail: rong.ji@duke.edu</p>
Neurology; musculoskeletal disease	Growth differentiation factor 11 (GDF11)	<p>Mouse studies suggest GDF11 could be useful for treating age-related neurological disease and regenerating skeletal muscle. In an experimental model system in which young and aged mice share a circulatory system, long-term exposure to young blood increased muscle regeneration, neurovascular function, neurogenesis and cognition in the aged mice compared with what was seen in controls receiving blood from aged mice. In aged mice, recombinant GDF11 partially recapitulated the effects of young blood infusion. Next steps could include testing GDF11 in models of age-related neurological and muscular disease.</p> <p>SciBX 7(23); doi:10.1038/scibx.2014.686 Published online June 12, 2014</p>	For findings from both studies, patents pending; licensing status undisclosed	<p>Katsimpardi, L. <i>et al. Science</i>; published online May 5, 2014; doi:10.1126/science.1251141 Contact: Lee L. Rubin, Harvard University, Cambridge, Mass. e-mail: lee_rubin@harvard.edu Contact: Lida Katsimpardi, same affiliation as above e-mail: lida_katsimpardi@harvard.edu</p> <p>Sinha, M. <i>et al. Science</i>; published online May 5, 2014; doi:10.1126/science.1251152 Contact: Amy J. Wagers, Harvard University, Cambridge, Mass. e-mail: amy_wagers@harvard.edu</p>

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
Disease models			
Engineered bone marrow-on-a-chip system	An engineered bone marrow-on-a-chip system could be useful in screening drug candidates for hematopoietic toxicity. The microchip consists of an engineered bone and bone marrow construct grown in mice that is inserted into an <i>in vitro</i> microfluidic culture system. The engineered bone marrow on the microchip retained the composition of natural bone marrow and recapitulated features of the hematopoietic niche. It also showed responses to radiation exposure and granulocyte colony-stimulating factor (G-CSF; CSF3) that were similar to those of bone marrow in live mice. Next steps include developing a humanized version of the bone marrow-on-a-chip system. SciBX 7(23); doi:10.1038/scibx.2014.687 Published online June 12, 2014	Patent application filed; licensing discussions under way with undisclosed parties	Torisawa, Y.-s. <i>et al. Nat. Methods</i> ; published online May 4, 2014; doi:10.1038/nmeth.2938 Contact: Donald E. Ingber, Harvard University, Cambridge, Mass. e-mail: don.ingber@wyss.harvard.edu
Heart-on-a-chip model for cardiomyopathy caused by Barth syndrome	A heart-on-a-chip model for Barth syndrome-associated cardiomyopathy could help identify new treatments for the indication. The heart-on-a-chip model was designed using patient induced pluripotent stem (iPS) cell-derived cardiomyocytes that self-assembled into myocardial tissue. Resulting cardiomyocytes showed disrupted sarcomere organization and contractile abnormalities when organized into myocardial tissue, and they had decreased mitochondrial function compared with cardiomyocytes derived from iPS cells from healthy individuals. In the model system, linoleic acid or a reactive oxygen species scavenger increased sarcomere organization and decreased contractile abnormalities compared with vehicle. Next steps include developing a high throughput screening platform using the model and doing quality control. SciBX 7(23); doi:10.1038/scibx.2014.688 Published online June 12, 2014	Covered by issued and filed patents; available for licensing	Wang, G. <i>et al. Nat. Med.</i> ; published online May 11, 2014; doi:10.1038/nm.3545 Contact: William T. Pu, Boston Children's Hospital, Boston, Mass. e-mail: wpu@enders.tch.harvard.edu Contact: Kevin Kit Parker, Harvard University, Cambridge, Mass. e-mail: kkparker@seas.harvard.edu
Drug delivery			
Targeted, dual-drug liposomal nanoparticles	Dual-drug liposomal nanoparticles could be useful for delivering combinations of cancer therapeutics. The liposomes were coated with folate to facilitate cancer cell targeting and loaded with Tarceva erlotinib in their hydrophobic regions and doxorubicin in their hydrophilic regions. In mouse xenograft models of triple-negative breast cancer (TNBC) and non-small cell lung cancer (NSCLC), the nanoparticles caused tumor regression, whereas targeted, single-drug nanoparticles resulted in continued tumor growth. In human TNBC and NSCLC cell lines, folate-targeted liposomes loaded with doxorubicin or cisplatin and various receptor tyrosine kinase (RTK) inhibitors generally showed greater cytotoxicity than liposomes containing single drugs. Next steps could include evaluating the liposomal nanoparticles for the delivery of additional combinations of cancer drugs. Astellas Pharma Inc., Chugai Pharmaceutical Co. Ltd. and Roche market the small molecule epidermal growth factor receptor (EGFR) inhibitor Tarceva to treat liver cancer, pancreatic cancer and NSCLC. SciBX 7(23); doi:10.1038/scibx.2014.689 Published online June 12, 2014	Patent application filed; licensing status unavailable	Morton, S.W. <i>et al. Sci. Signal.</i> ; published online May 13, 2014; doi:10.1126/scisignal.2005261 Contact: Paula T. Hammond, Massachusetts Institute of Technology, Cambridge, Mass. e-mail: hammond@mit.edu
Markers			
Dipeptidyl peptidase-4 (DPP-4; CD26) as a marker of chronic myelogenous leukemia (CML) stem cells	DPP-4 could be useful as a marker for detecting CML stem cells. In a leukemia stem cell-enriched cellular fraction from patients with CML, gene array and PCR analyses showed that DPP-4 expression was almost entirely restricted to the leukemia stem cell population. Almost 100% of the DPP-4 ⁺ cells isolated from this population expressed the oncogenic BCR-ABL tyrosine kinase fusion protein versus none of the DPP-4 ⁻ cells. Next steps could include validating DPP-4 as a selective and specific marker for CML stem cells in larger patient cohorts. SciBX 7(23); doi:10.1038/scibx.2014.690 Published online June 12, 2014	Patent and licensing status unavailable	Herrmann, H. <i>et al. Blood</i> ; published online April 28, 2014; doi:10.1182/blood-2013-10-536078 Contact: Peter Valent, Medical University of Vienna, Vienna, Austria e-mail: peter.valent@meduniwien.ac.at

Company and institution index

A		N		ADAM10	12	FASN	12
Alsace BioValley	5	National Institute of Scientific Research	5	ADAM17	12	Fatty acid synthase	12
Ambit Biosciences Corp.	8	National Institutes of Health	4	ARAF	8	Folate	18
Amgen Inc.	7	National Research Council		ARID1A	13	FR104	15
arGEN-X B.V.	2	Canada	5	ARID1B	13	FRAP	8
Array BioPharma Inc.	7	NDEI Inc.	5	AS703026	7	FTDP-17	16
Astellas Pharma Inc.	18	Novartis AG	5,7,9	AT rich interactive domain 1A	13		
AstraZeneca plc	5,7,16	Numab AG	3	AZD6244	7	G	
B		O		B		G-CSF	18
Bayer AG	7	Ono Pharmaceutical Co. Ltd.	13	β-Amyloid	16	GDF11	17
BeiGene Co. Ltd.	8	Ontario Brain Institute	4	B7-H6 tumor cell ligand	12	Gemcitabine	9
BioMed Valley Discoveries Inc.	7	Ontario Centres of Excellence	4	BAY 86-9766	7	Glucose	15,17
Boehringer Ingelheim GmbH	5	P		BCR-ABL tyrosine kinase	18	GPCR	5
Brain Canada	4	Perelman School of Medicine at the University of Pennsylvania	9	BeiGene-283	8	Granulocyte colony-stimulating factor	18
Bristol-Myers Squibb Co.	10,13,15	Pfizer Inc.	3,5,8	Binimetinib	7	GREM1	14
C		Q		BLT2	15	Gremlin 1	14
Cancer Research UK	9	Quebec Consortium for Drug Discovery	4	BMK1	6	Growth differentiation factor 11	17
Caprion Proteomics Inc.	5	R		BMP	14	H	
Centre National de la Recherche Scientifique	5	Roche	7,9,18	BMP2	14	Heat shock 70 kDa protein 9	4
Chugai Pharmaceutical Co. Ltd.	7,18	S		Bone morphogenetic protein	14	Hedgehog	9
Cold Spring Harbor Laboratory	11	Saarland University Medical Center	3	BRAF	6	HIF1	13
Columbia University Medical Center	9	Sanofi	4,17	BVD-523	7	HspA9	4
Convergence Pharmaceuticals Ltd.	3,17	Sherbrooke University	5	C		Hyaluronan	11
D		SiteOne Therapeutics Inc.	3	C5AR1	12	Hypoxia-inducible factor 1	13
Daiichi Sankyo Co. Ltd.	7	Stanford University	6	C5AR	12	I	
Dainippon Sumitomo Pharma Co. Ltd.	3	Stanford University School of Medicine	6	C75	12	IGF-1	15
Deciphera Pharmaceuticals LLC	8	T		Catecholamine-regulated protein 40	4	IGFBP2	14
Dompe Farmaceutici S.p.A.	13	Takeda Pharmaceutical Co. Ltd.	8	Cd152	10	IKKB	17
Duke University	3	Technical University Munich	11	CD26	18	IKK2	17
Duke University Medical Center	1	Teva Pharmaceutical Industries Ltd.	8	CD279	13	IL8RB	13
E		U		CD28	15	IMD-1041	17
Effimune S.A.S.	15	University Institute of Geriatrics of Montreal	4	CEP-32496	8	Inhibitor of κ-light polypeptide gene enhancer in B cells kinase-β	17
Eli Lilly and Co.	5,8	University of California, Davis		Cisplatin	13,18	Inositol 1,4,5-triphosphate receptor	16
EntreChem S.L.	17	Comprehensive Cancer Center	11	CNV1014802	3,17	Insulin-like growth factor binding protein 2	14
Epizyme Inc.	6	University of Cambridge	1	Cobimetinib	7	Insulin-like growth factor-1	15
Exelixis Inc.	7	University of Montreal	5	Complement component 5a receptor 1	12	Insulin	15
G		University of Queensland	2	CRAF	7	IP3R	16
Genentech Inc.	7,9	University of Texas MD Anderson Cancer Center	10	CRP40	4	IP1-926	9
GlaxoSmithKline plc	5,6	University of Washington	1	CSF3	18	Ipilimumab	10
H			Ctla-4	10	ITPR	16
Halozyne Therapeutics Inc.	11	Targets and compounds		CXC chemokine receptor 2	13	K	
I		3-Oxoacyl-ACP synthase mitochondrial	12	CXCR2	13	K-class CpG oligodeoxynucleotide	16
IMMD Inc.	17	A		Dabrafenib	7	K-ODN	16
Infinity Pharmaceuticals Inc.	9	α-Lipoic acid	12	DP-4978	8	K-Ras	6
L		α-Sma	10	DPP-4	18	KRAS	6
Laval University	4	Aβ	16	DSP-2230	3	L	
Lustgarten Foundation	11	Abatacept	15	E		Lambrolizumab	13
M		ABHD6	15	E1A binding protein p300	13	LCN2	12
MaRS Innovation	4	Abhydrolase domain containing 6	15	EC-70124	17	LDE225	9
McGill University	5	ACTA2	10	EGFR	18	Leukotriene B4 type 2 receptor	15
McMaster University	4	Actin α2 smooth aorta muscle	10	Encorafenib	7	LGX818	7
Merck & Co. Inc.	5,13			EP300	13	Linoleic acid	18
Merck KGaA	7			Epidermal growth factor receptor	18	Lipocalin	12
Montreal Heart Institute	5			Erivedge	9	LY3009120	8
				ERK-1	7	M	
				ERK-2	7	Mammalian target of rapamycin	8
				ERK-5	6	MAP kinase 1	7
				Erlotinib	18	MAP kinase 7	6
				F			
				FAS	12		

MAP kinase kinase 1	7	ND004	3	PPP2CA	8	Smoothened	9
MAP2K1	7	Nexavar	7	Pre-mRNA processing factor 6	14	SMYD3	6
MAP2K2	6	NGAL	12	Programmed cell death 1	13	Sperm adhesion molecule 1	11
MAP2K5	6	Nivolumab	13	Protein phosphatase 2	8	SPAM1	11
MAP3K2	8			PRPF6	14	<i>Sonic hedgehog homolog</i>	9
MAPK1	7	O				Sorafenib	7
MAPK3	7	Olaparib	16	R		ST-200	3
MAPK7	6	Orencia	15	RAF1	7	<i>Sterile α-motif and leucine zipper containing kinase AZK</i>	14
MAPK	6	Oxopiperizine	13	RAFT1	8	STK4	14
MAPT	16	OXSM	12	Receptor tyrosine kinase	18		
MEK162	7			Refametinib	7		
MEK1	7	P		Reperixin	13	T	
MEK2	7	p300	13	RG7167	8	T3	15
MEK5	6	PARP	16	RG7304	8	Tafinlar	7
Mekinist	6	PD-0325901	8	RG7421	7	TAK-733	8
Melphalan	15	PD-1	13	RTK	18	Tarceva	18
Microtubule-associated protein- τ	16	PDCD1	13			Tau	16
Mitogen-activated protein kinase kinase 5	6	PEG	11	S		Trametinib	6
Mitogen-activated protein kinase kinase kinase 2	6	PEGPH20	11	SAR113945	17	Triiodothyronine	15
Mortalin-2	4	PF-05089771	3	Saridegib	9		
mTOR	8	PF-05212384	8	SCN10A	3	V	
		PH20	11	SCN4A	1	VEGF receptor	7,10
		PH20 hyaluronidase	11	SCN5A	1	Vemurafenib	7
		Phosphoinositide 3-kinase	8	SCN8A	2	Vismodegib	9
		PI3K	8	SCN9A	1,17		
		Pimasertib	7	Selumetinib	7	Y	
		PN3	3	Serine/threonine kinase 4	14	YAP1	14
		PN4	2	SET and MYND domain containing 3	6	Yervoy	10
		Poly(ADP-ribose) polymerase	16	<i>Shh</i>	9	Yes-associated protein 1	14
		Polyethylene glycol	11	SMO	9		
		PP2A	8			Z	
						ZAK	14
						Zelboraf	7