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In silico drug design

By Joanne Kotz, Senior Editor

Researchers at the **University of Dundee** and **The University of North Carolina at Chapel Hill School of Medicine** have created a computational algorithm that mines medicinal chemistry literature to predict new ligands that bind specific combinations of G protein–coupled receptors.¹ **Ex Scientia Ltd.** was spun out of Dundee to commercialize the findings and already has two deals in place related to the screening technology.

Developing a small molecule that binds a predefined combination of targets is at best a time-consuming medicinal chemistry effort and often has proven flat-out impossible.

One major obstacle to designing such drugs is the limited human capacity to sift through the vast accumulation of medicinal chemistry data related to multiple targets and

then identify an optimal solution.

A team led by Andrew Hopkins and Bryan Roth reasoned that a computational algorithm might be able to more effectively undertake medicinal chemistry design than a person could and thus could better identify ligands with predetermined polypharmacology, which is the modulation of multiple targets. "What we've found is that medicinal chemistry experience can be encoded and mimicked to a degree. We can now move from human judgment to exploiting vast amounts of data and come up with quite sensible-looking chemistry." —Andrew Hopkins, University of Dundee

Hopkins is chair of medicinal informatics and professor of

translational biology at Dundee and founder and managing director of Ex Scientia. Roth is a professor in the Department of Pharmacology at the UNC at Chapel Hill School of Medicine.

First, their team mined data in the ChEMBL public database, which contains compound and activity data extracted from decades of published medicinal chemistry literature. The group used this data to build Bayesian models of ligand activity across 784 human protein targets, including G protein–coupled receptors (GPCRs).

As proof of concept, the researchers decided to study the Alzheimer's disease (AD) drug Aricept donepezil, an acetylcholinesterase (AChE) inhibitor marketed by **Pfizer Inc.** and **Eisai Co. Ltd.**

The models predicted that Aricept could also act *in vitro* on the dopamine D4 receptor but not the dopamine D2 receptor, two receptors for which there are large amounts of ligand-binding data. Indeed, experimental measurements confirmed that donepezil was a moderately potent D4 receptor inverse agonist and had essentially no activity against the D2 receptor.

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The models predicted that eight isoindole-based compounds would have the desired activities. Of those, all eight had activity on both D4 and D2 receptors. The team tested the most potent compound in mice, and the molecule crossed the BBB.

Although the eight molecules had activity against the desired targets, they also had activity against targets likely to confer side effects. For example, the molecules blocked some adrenoreceptors, which is known to result in hypotension.

Thus, the team next sought to predict ligands with a more complicated combination of targets they wanted to hit and ones they wanted to avoid.

The models suggested that some benzolactam-based molecules would have activity against three particular dopamine receptors and a serotonin receptor but not against select adrenoreceptors. These predictions were confirmed experimentally.

Overall, 800 ligand-target interactions were predicted, of which about 600 (75%) were successfully confirmed experimentally.

Results were published in Nature.

"What we've found is that medicinal chemistry experience can be encoded and mimicked to a degree. We can now move from human judgment to exploiting vast amounts of data and come up with quite sensible-looking chemistry," said Hopkins.

Automating discovery

The biggest limitation of the computational drug design approach is the need for existing medicinal chemistry data, making it unlikely the method can be applied to new targets.

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"The power of this method is that it lets you leverage every bit of published data in a systematic way. You can evaluate all the possibilities in a way that you couldn't in your head. For example, it's been very difficult to design GPCR ligands against multiple targets—they have usually been designed quasi-randomly based on hope and luck," said Jon Mason, senior research fellow at GPCR drug discovery company **Heptares Therapeutics Ltd.**

Kerry Spear, VP of medicinal chemistry at **Sunovion Pharmaceuticals Inc.**, a subsidiary of **Dainippon Sumitomo Pharma Co. Ltd.**, added, "I think that this paper has the potential of being a seminal paper for the pharmaceutical industry. Interest in polypharmacology has always been clouded by the fact that you have to optimize multiple SARs. The more SARs you have, the harder the process is and the longer it takes. This outlines a framework for optimizing multiple SARs in a systematic way."

Sunovion's internal research focuses on CNS disorders.

Mason said that ligands designed against a particular target are often not assayed against more than a handful of potential off targets until fairly late in development. When the goal is to design a selective ligand, the computational models could be very useful for identifying unanticipated and undesirable off-target effects early in the drug discovery process, which "could improve efficiency up front," he said.

Combining public and in-house company data may be the best use of the models, according to Mason. "Local models trained on your own data are even better. A company could update the models overnight with any new data to have an up-to-date activity model for every target," he said.

"There are really two caveats to the method right now," noted Spear. "First, the method doesn't speak to target selection; that's a big unknown. The other caveat is that the process depends on reliable data, and a lot of it. There is the possibility that there are targets you are interested in where there isn't enough data to allow the algorithms to work effectively."

Mason agreed. "The method is very good but not the 'eureka' for everything," he said. "You need to have large datasets for the targets of interest. For new targets with no published data, you can't model and evolve compounds. You also can't explore completely unknown ways of ligands interacting with targets."

Mason said Heptares is "working now mostly on undrugged targets where there are very limited data." In this case, a structure-based approach is preferable for the primary target, he said.

The leading edge

Hopkins now is working to incorporate structural information into the algorithm. He hopes this will open the method up to identifying ligands against previously unknown targets or ligands that interact at new sites on targets. The team also is pursuing the application of the method to other target classes, such as kinases.

The team is further interested in exploring whether the computational models can be used for lead optimization. "The next challenge is can we more efficiently optimize lead matter? We are working on methods to reduce the number of compounds required to be synthesized in the lead optimization process to increase productivity," said Hopkins.

Hopkins founded Ex Scientia in July 2012. He said the company has "two deals signed and a third in the pipeline." Ex Scientia plans to apply the current algorithm and develop it further through collaborations on projects at the partner companies. Ex Scientia also hopes to develop an internal pipeline of drug candidates designed using the technology platform.

Dundee has filed a patent application on the methodology, which has been licensed to Ex Scientia.

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COMPANIES AND INSTITUTIONS MENTIONED

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TRANSLATIONAL NOTES

IMI's collaborative chemistry

By Chris Cain, Senior Writer

Europe's **Innovative Medicines Initiative** has launched seven new publicprivate partnerships with a total budget of €237 million (\$313 million). In contrast to many of the organization's disease-focused projects, three of the new consortia are taking a drug-centric approach that includes plans to optimize small molecule binding kinetics and develop new delivery systems for RNA- and protein-based therapeutics.

The projects comprise the fourth wave of initiatives launched by IMI since its 2008 founding (*see* **Table 1**, "**IMI's fourth call**"). In total, the organization now manages 37 ongoing projects with a total budget of more than \$1 billion.

Hugh Laverty, a senior scientific project manager at IMI, told *SciBX* that the push for projects focused on drug optimization and delivery was driven by **European Federation of Pharmaceutical Industries and Associations** (EFPIA) members seeking to collaborate on solutions to common problems that crop up in drug development regardless of therapeutic area.

"When companies are looking at some problems they face internally there is clearly a focus on certain areas like Alzheimer's disease or stem cell biology, and we have projects that cover those. The problems being addressed by these projects are faced at all stages of drug development, from lead identification to formulation to manufacturing," said Laverty.

The new consortia that seek to optimize drug properties and delivery methods are the Collaboration on the Optimization of Macromolecular Pharmaceutical Access to Cellular Targets (COMPACT) led by **Sanofi**, Kinetics for Drug Discovery (K4DD) led by **Bayer AG** and Oral Biopharmaceutics Tools (ORBITO) led by **AstraZeneca plc**.

"These topics are the bread and butter of the pharmaceutical companies. If we see progress in developing new models or *in silico* methods to predict or measure oral bioavailability or kinetics of drug binding more accurately, you have the potential to change the way drugs are discovered and developed," said Laverty.

Ekkehard Leberer, a senior director at Sanofi and the EFPIA coordinator of COMPACT, told *SciBX* that the focus of the initiative he is overseeing is on developing new methods to specifically deliver RNA- and protein-based therapeutics to tissues including the lung and brain. This will include synthesizing and testing new liposome delivery formulations, polymeric nanoparticle carriers, cell-penetrant peptide carriers, modified oligonucleotides, exosomes and microneedle delivery systems.

He said the consortium is not interested in refining or characterizing existing formulations. "Many of our partners are working on liposomes and already have systems in place, but we are not interested in testing

Table 1. IMI's fourth call. The fourth set of projects launched by the Innovative Medicines Initiative (IMI) comprise seven projects that are a joint effort of the European Commission and the European Federation of Pharmaceutical Industries and Associations (EFPIA). The below projects include each program's expected contributions from the EFPIA and IMI. In total, the projects have a combined budget from the IMI and the EFPIA of €237 million (\$313 million).

		Contributio	n (million)
Project title	EFPIA participants ^A	EFPIA in kind	IMI
CHEM21 (Chemical Manufacturing Methods for the 21st Century Pharmaceutical Industries)	GlaxoSmithKline plc (LSE:GSK; NYSE:GSK); Bayer AG (Xetra:BAYN); Johnson & Johnson (NYSE:JNJ); Orion Corp. (HSE:ORNAV; HSE:ORNBV); Pfizer Inc. (NYSE:PFE); Sanofi (Euronext:SAN; NYSE:SNY)	€9.8 (\$13.0)	€13.6 (\$18)
COMPACT (Collaboration on the Optimization of Macromolecular Pharmaceutical Access to Cellular Targets)	Sanofi; Boehringer Ingelheim GmbH ; Bayer; Johnson & Johnson; GlaxoSmithKline; Merck KGaA (Xetra:MRK); Novo Nordisk A/S (CSE:NVO; NYSE:NVO); Pfizer	€10.2 (\$13.5)	€16.6 (\$22)
EMIF (European Medical Information Framework)	GlaxoSmithKline; Amgen Inc. (NASDAQ:AMGN); Boehringer Ingelheim; Roche (SIX:ROG; OTCQX:RHHBY); Johnson & Johnson; Novo Nordisk; Pfizer; Servier; UCB Group (Euronext:UCB)	€24.4 (\$32.3)	€24.1 (\$31.9)
eTRIKS (delivering European Translational Information and Knowledge Management Services)	AstraZeneca plc (LSE:AZN; NYSE:AZN); Bayer; Eli Lilly and Co. (NYSE:LLY); Roche; GlaxoSmithKline; H. Lundbeck A/S (CSE:LUN); Johnson & Johnson; Merck KGaA; Pfizer; Sanofi	€10.3 (\$13.6)	€10.3 (\$13.6)
K4DD (Kinetics for Drug Discovery)	Bayer; AstraZeneca; Roche; GlaxoSmithKline; Johnson & Johnson; Merck KGaA; Sanofi	€8.3 (\$11.0)	€9.8 (\$13.0)
ORBITO (Oral Biopharmaceutics Tools)	AstraZeneca; Abbott Laboratories (NYSE:ABT); Bayer; Boehringer Ingelheim; GlaxoSmithKline; H. Lundbeck; Johnson & Johnson; Merck & Co. Inc. (NYSE:MRK); Novartis AG (NYSE:NVS; SIX:NOVN); Orion; Pfizer; Sanofi	€9 (\$11.9)	€11.5 (\$15.2)
STEMBANCC (Stem Cells for Biological Assays of Novel Drugs and Predictive Toxicology)	Roche; Abbott; Boehringer Ingelheim; Eli Lilly; Johnson & Johnson; Merck KGaA; Novo Nordisk; Orion; Pfizer; Sanofi	€26 (\$34.4)	€21 (\$27.8)

^AParent company of participating subsidiaries listed.

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those systems. We want to develop new systems and new ideas, and we expect the academic postdocs and students will be involved in generating those ideas," he said.

Although the consortium will look to academia for some fresh

thinking, Leberer said the industry members will help keep the consortium focused on clinically feasible projects. "We are not just developing tools for the sake of getting publications. We have given guidance to our partners from the beginning that our goal is to develop a drug. Any formulation needs to be scalable and translatable into a manufacturing process, and industry can provide that input," he said.

Unlike COMPACT, the ORBITO and K4DD projects are not aiming to develop new drug delivery modalities. Instead, those two consortia seek to better understand how conventional small

molecules are absorbed into the bloodstream and engage their targets. ORBITO aims to define the physicochemical formulation and physiological factors that affect small molecule oral bioavailability. Laverty said this will be accomplished by sharing existing compound data accumulated by EFPIA participants, generating new complementary data and testing those compounds in new models developed by the consortium. These new models will encompass *in vitro*, *in vivo* and *in silico* tools to predict drug oral bioavailability.

AstraZeneca is lead EFPIA coordinator of ORBITO and did not respond to interview requests.

Anke Müller-Fahrnow, VP and head of lead discovery at Bayer and the EFPIA coordinator of K4DD, said her initiative is focused on improving the understanding of how small molecules interact with their targets.

"We have given guidance to our partners from the beginning that our goal is to develop a drug. Any formulation needs to be scalable and translatable into a manufacturing process, and industry can provide that input." —Ekkehard Leberer, Sanofi

"We are increasingly realizing that *in vitro* tests in a research lab may not accurately reflect the *in vivo* situation in a patient. For instance, common *in vitro* equilibrium studies may not be completely predictive in an *in vivo* system. Two similar-affinity drugs may have very different

> residence times—the actual lifetime of the drugreceptor complex—which in turn can have an effect on efficacy and also side effects," she said.

> To better understand drug binding kinetics, the initiative will begin by developing new techniques that can be used to characterize the binding of compounds within cells, particularly against targets tough to characterize *in vitro*, such as membrane-bound proteins. Müller-Fahrnow said insights from these studies will then be applied to *in vivo* pharmacology and physiology studies.

> She added that the scientific community often underestimates the importance of binding

kinetics in drug discovery. "Raising the awareness for kinetic parameters might mean that by the end of the consortium's lifetime in 2017, editors and reviewers for scientific journals could routinely ask for kinetic data and industry could do more to adopt strategies to learn about kinetics in the very early stages of the drug discovery process," she said.

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COMPANIES AND INSTITUTIONS MENTIONED

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TARGETS & MECHANISMS

Giving the NOD2 microbiota

By Kai-Jye Lou, Staff Writer

Disruption of enteric microbiota occurs in inflammatory bowel diseases, but whether this is a cause or result of the condition is unclear. Now, European researchers have shed light on the process by showing that a deficiency in an immune system-related receptor, NOD2, can disrupt enteric microbiota and set the stage for colitis and colitisassociated cancer.1

The group now is looking for bacterial strains that could help correct the microbial imbalance and is trying to better understand why a deficiency in caspase recruitment domain family member 15 (NOD2; CARD15) causes the disruption in the first place.

NOD2 is a pattern-recognition receptor for bacterial peptidoglycans that facilitates the host immune response against pathogens. It also has been shown to regulate the composition of enteric microbiota in mice.²

Mutations in the gene encoding NOD2 predispose individuals to Crohn's disease, which is one of the two major subtypes of inflammatory bowel disease (IBD). Patients with Crohn's disease also show imbalances in their enteric microbiota^{3,4}—a condition called dysbiosis—and have an elevated risk of colorectal cancer.⁵

In the current study, a team co-led by Mathias Chamaillard sought to flesh out the relationship between NOD2 dysregulation, imbalances in enteric microbiota and susceptibility to colitis, and the development of colitis-associated colorectal carcinomas.

Chamaillard is a research director and team leader at the Center of Infection and Immunity of Lille at the Pasteur Institute in Lille. He has been studying the role and function of NOD2 and NOD2-related proteins in the host immune system, tumorigenesis and IBD for more than a decade.

The researchers showed that knockout of *Nod2* in mice increased susceptibility to chemical-induced colitis and colitis-associated colorectal carcinomas compared with no knockout.

Surprisingly, the disease-vulnerable state was transmissible. Wildtype mice cohoused with Nod2-deficient mice also showed increased susceptibility to colitis and colitis-associated colorectal carcinomas compared with wild-type mice cohoused with other wild-type mice. This result implicated the transmission of disease-predisposing bacteria as the underlying cause.

Indeed, Nod2-deficient mice treated with a broad-spectrum antibiotic showed lower susceptibility to chemical-induced colitis than untreated Nod2-deficient controls.

To confirm enteric bacteria were the culprit, the researchers carried out a series of fecal transplant experiments. Nod2-deficient mice receiving a fecal transplant from mice with functional Nod2 showed decreased susceptibility to chemical-induced colitis compared with deficient mice given transplants from other mice lacking Nod2. Conversely, mice with functional Nod2 receiving a fecal transplant from Nod2-deficient animals showed increased susceptibility to chemicalinduced colitis compared with Nod2-functional mice receiving transplants from other mice with functional Nod2.

Results were published in The Journal of Clinical Investigation.

"One of the important experimental priorities in this field is whether disruptions in the gut microbiome are causal or a result of the disease," said Peter DiStefano, SVP of R&D of Second Genome Inc. "I think this study provides a key piece of data that shows causality of the gut microbiome in IBD."

Second Genome is developing therapies that can alter the composition and activity of microbial communities in the body.

"This paper also reinforces the connection between risk factors for Crohn's disease and colitis-associated cancer and proposes a plausible role for microbial dysbiosis in connecting the two," added Bernat Olle, COO of Vedanta Biosciences Inc. and a principal at PureTech Ventures. "The described mechanisms are consistent with what has been suggested in the literature and reinforce our understanding of how certain genes, such as NOD2, can influence the gut microbial composition, which in turn can keep inflammatory responses in check."

Olle also said the findings help dispel the notion that having an at-risk genotype that predisposes one to dysbiosis and IBD is an unalterable fate.

"The host genotype may be unalterable, but the dysbiosis is certainly not, and as shown by the authors, fecal transplantation and other microbiome manipulations can reverse alterations of the gut microbiota and improve disease outcomes," he told SciBX. "In the last decades, a lot of effort in drug development in IBD has gone toward modulation of inflammatory mediators, and this has led to breakthrough treatments

like anti-TNFs. However, these approaches may not help address the underlying dysbiosis that helps drive chronic inflammation in IBD patients."

At least three anti-tumor necrosis factor (TNF) antibodies already are marketed to treat Crohn's disease. These are Humira adalimumab from AbbVie Inc., Remicade infliximab from Johnson & Johnson and Cimzia certolizumab pegol from UCB Group.

"This paper also reinforces -Bernat Olle, PureTech Ventures

Vedanta is developing an oral formulation of enteric bacteria to correct the deficiency of normal bacteria that occurs in IBD.

Bugs as drugs

With the causal relationship established, Chamaillard said the group now is screening for probiotic bacterial strains that could normalize enteric microbial populations in the IBD setting.

"The current study shows that changing the composition of microbial communities in the gut could have a beneficial effect, but exactly how one should go about selectively changing the microbiome is still unclear," said Christian Jobin, an associate professor in the Department of Medicine at The University of North Carolina at Chapel Hill School of Medicine. "They may also want to identify and go after the microbial entities that cause the increased susceptibility to disease. RNA and genome sequencing studies are going to be important for addressing these questions."

DiStefano said another avenue to search for potential therapeutic

the connection between risk factors for Crohn's disease and colitisassociated cancer and proposes a plausible role for microbial dysbiosis in connecting the two."

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strategies is the metabolites produced by enteric bacteria as such molecules may help drive or prevent disease.

"The researchers may want to look at the metabolites these bacteria produce and run studies to determine what these molecules are doing to the host and how they affect disease pathology," he said.

Indeed, Chamaillard's group is trying to identify the cellular and molecular mechanisms underlying the disruption of enteric microbiota caused by the deficiency in NOD2 and how this leads to increased disease susceptibility.

Olle wanted to see studies to determine whether there is an ideal time window during which manipulating the enteric microbiome would have benefit in IBD- or colitis-associated cancer.

"This will require prospective studies following IBD- and colitisassociated cancer patients and determining the timing and order of events that conspire to create the conditions for the disease to develop," he said. "Another important question will be to clearly define patient subsets that are most likely to respond favorably to microbiome manipulation. This information will be very useful for clinical trial design and patient selection."

The work reported in the paper is unpatented. Chamaillard said the group plans to file for IP after identifying the key bacterial strains, genes and molecular mechanisms responsible for disrupting enteric microbiota.

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AbbVie Inc. (NYSE:ABBV), Chicago, III. Johnson & Johnson (NYSE:JNJ), New Brunswick, N.J. Pasteur Institute in Lille, Lille, France PureTech Ventures, Boston, Mass. Second Genome Inc., San Francisco, Calif. UCB Group (Euronext:UCB), Brussels, Belgium The University of North Carolina at Chapel Hill School of Medicine, Chapel Hill, N.C. Vedanta Biosciences Inc., Boston, Mass.

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Sarepta's hemorrhagic mouse

By Tracey Baas, Senior Editor

A team at **Sarepta Therapeutics Inc.** has developed a mouse model of arenavirus infection that recapitulates key symptoms of the disease including hemorrhagic fever and multiorgan failure.¹ The researchers now want to use the mice to study the Lassa arenavirus.

Arenavirus infection symptoms vary from virus to virus, but general symptoms are fever, malaise, body aches and rash. For arenaviruses that cause hemorrhagic disease, symptoms also include low platelet count, internal and external hemorrhage and multiorgan failure.²

Arenaviruses are divided into two serogroups: Old World and New World, which differ genetically and by geographical distribution. Historically, only the New World hemorrhagic fever arenaviruses appear to cause a set of neurologic disease symptoms.²

Arenavirus infection typically is treated with best supportive care and the generic antiviral ribavirin. Targeted therapies do not exist and have

"It would also be very important to determine if Lassa-infected FVB chimeric mice show disease that is similar to Lassa-infected nonhuman primates. This would make compound screening in the chimeric mice more useful to translation in nonhuman primates."

-Juan Carlos de la Torre, The Scripps Research Institute been difficult to develop because no rodent model accurately mimics human arenavirus infection.

Sarepta's initial plan was to characterize its own morpholinobased arenavirus antiviral, dubbed AVI-7012, in mice expressing a fluorescent transgene. Those mice were derived from the FVB mouse strain, which is named for its susceptibility to the B strain of Friend leukemia virus and has enhanced susceptibility to a variety of viruses.³

When the researchers infected

the FVB mice with an arenavirus called lymphocytic choriomeningitis virus 13 (LCMV-13), the animals showed signs of mucosal, cutaneous and organ hemorrhaging. That result was unexpected because LCMV is a prototype laboratory arenavirus strain that typically causes only mild infection in humans.

To confirm that the fluorescent transgene was not causing the hemorrhaging, the team repeated the experiment using FVB mice not expressing the transgene. LCMV-13 infection again caused hemorrhagic fever-like symptoms—platelet loss, cutaneous hemorrhaging and hepatic dysfunction. Seven of eight mice died six to eight days postinfection.

The researchers next used the hemorrhagic mice to test the antiviral activity of AVI-7012. The compound decreased viral load compared with saline but surprisingly did not result in decreased mortality.

In animals with T cell depletion or Il-17 receptor C (Il17rc) knockdown, LCMV-13 infection resulted in decreased viral loads, organ damage and death compared with what was seen in wild-type mice. These results suggest that arenavirus alone does not cause the pathology

and that T cell–produced Il-17 may exacerbate a renaviral hemorrhagic fever–like disease in FVB mice.

Finally, the Sarepta team generated a third chimeric mouse strain by crossing the FVB mice with standard C57BL/6 lab mice. The result was a mouse strain compatible with available mouse immunological reagents and with a better correlation between increased virus replication and increased disease symptoms.

High virus infectivity resulted in 100% death, medium virus infectivity resulted in 66% death and low virus infectivity resulted in no death.

Results were reported in PLoS Pathogens.

More mouse, different virus

"We have ongoing work using the model in-house to explore hostvirus interactions that contribute to hemorrhagic symptoms," said Dan Mourich, director of immunology at Sarepta and principal investigator on the manuscript. "Our goal is to use the chimeric mouse to study other types of arenaviruses, such as Junin or Lassa, which cause hemorrhagic fever in humans. But in order to do that, we will have to rely on collaborators that have access to laboratories with proper biosafety containment."

Lassa, an Old World strain, is the most prevalent hemorrhagic feverinducing arenavirus found in West Africa.

Sarepta previously has worked with the **U.S. Department of Defense** to investigate antivirals in mouse models of Ebola and Marburg infection.

Juan Carlos de la Torre, professor of immunology and microbial science at **The Scripps Research Institute**, thinks the next steps should be showing that chimeric mice infected with Lassa virus (LASV) recapitulate key features of Lassa fever disease, with increased viral load correlating to increased disease symptoms, and pathology unrelated to T cell immune–mediated damage of infected cells and tissues.

"It would provide tremendous possibilities for the investigation of disease mechanisms and therapies. Current mouse models of Lassa infection are rather artificial. They lack a fully functional immune system or require the use of rather specific viral strains, which raises some issues about whether information derived from their use would accurately reflect the nature of LASV infection," he said. "It would also be very important to determine if Lassa-infected FVB chimeric mice show disease that is similar to Lassa-infected nonhuman primates. This would make compound screening in the chimeric mice more useful to translation in nonhuman primates."

De la Torre did say two issues presented in Sarepta's study of LCMV-13 may limit the use of these chimeric mice to study Lassa virus.

"First, they show that T cells play a critical role in arenavirus-induced disease pathology. This is in contrast to what is known about human patients succumbing to Lassa virus infection. Patients show very limited immune responses, immune cell infiltrates and tissue damage," de la Torre said. "Second, the study shows limited correlation between viral load and disease outcome. In humans, levels of virus in the blood are a good predictor of disease outcome for Lassa fever."

"It is possible that the model does not fully reflect the human disease; however, making precise comparisons of immunological and viral endpoints in a mouse model to those observed in the clinic are difficult at best," said Mourich. "The exact time of infection, inoculum size, peak of virus production in all tissues as well as the peak and specific nature of

T cell responses in a clinically manifested disease are rarely measurable or measured."

"In our mouse model we are stating that the level of virus dose does not directly predict disease outcome," Mourich continued. "We would not discount that detection of virus in the blood of a human patient would not be predictive of Lassa disease severity."

Eric Vela, senior research scientist at **Battelle Biomedical Research Center**, would like to see the group use the chimeric mouse model to evaluate if there are differences between Old World and New World arenavirus disease outcomes.

"Sarepta did not show if there was a neurological component in the chimeric mice infected with LCMV-13. Researchers have previously shown hemorrhagic symptoms and neurologic disease—tremors, and shaking and rocking behavior—in Syrian golden hamsters infected with the New World Pirital virus," he noted.

"If neurological disease is found to occur in the FVB mice, they might be considered better suited to model New World arenavirus infections. If neurological disease is absent, they might be considered better suited to model Old World arenavirus infections," Vela added.

"We would hope to explore if there exists a neurological component in the mouse disease model," Mourich said. "Observation of virus in the brain might be an initial clue that there is some disruption in the blood brain barrier either caused by the virus or as a result of the profound inflammatory damage."

The chimeric mouse model of arenavirus infection is patented by Sarepta and is available for licensing.

Baas, T. *SciBX* 6(3); doi:10.1038/scibx.2013.53 Published online Jan. 24, 2013

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 Contact: Dan V. Mourich, Sarepta Therapeutics Inc., Cambridge, Mass. e-mail: dmourich@sareptatherapeutics.com
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COMPANIES AND INSTITUTIONS MENTIONED

Battelle Biomedical Research Center, Columbus, Ohio Sarepta Therapeutics Inc. (NASDAQ:SRPT), Cambridge, Mass. The Scripps Research Institute, La Jolla, Calif. U.S. Department of Defense, Washington, D.C.

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This week in therapeutics

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

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

Indication	Target/marker/ pathway	Summary	Licensing status	Publication and contact information
Cancer	Pyruvate kinase M2 isozyme	Cell culture and mouse studies suggest targeting PKM2 may not be sufficient to inhibit tumor growth. Studies in mouse	Patent and licensing	Cortés-Cros, M. et al. Proc. Natl. Acad. Sci. USA; published online
	(PKM2); PKM1	tumor xenograft models have previously shown that PKM2 is required for initial growth and establishment of tumors. In cultured colon carcinoma or adenocarcinoma cells, small hairpin RNA-mediated knockdown of <i>PKM1</i> and <i>PKM2</i> , but not <i>PKM1</i> or <i>PKM2</i> alone, decreased pyruvate kinase activity and proliferation compared with no knockdown. However, in mouse xenograft models of established colon carcinoma or adenocarcinoma, individual or dual inhibition of <i>PKM1</i> and <i>PKM2</i> failed to decrease tumor growth. Next steps could include testing PKM2 modulators plus other antitumor compounds in mouse xenograft models to identify effective combination treatments.	status undisclosed	Dec. 24, 2012; doi:10.1073/pnas.1212780110 Contact: Marta Cortés-Cros, Novartis Institutes for BioMedical Research, Basel, Switzerland e-mail: marta.cortes-cros@novartis.com
		<i>SciBX</i> 6(3); doi:10.1038/scibx.2013.54 Published online Jan. 24, 2013		
Cardiovascul	ar disease			
Hypertension	Apelin (APLN); microRNA-424 (miR-424); miR-503	<i>In vitro</i> and rodent studies suggest miR-424 and miR-503 could help treat pulmonary arterial hypertension (PAH). In human pulmonary artery endothelial cells, decreased expression of miR-424 and miR-503 was associated with increased proliferation of pulmonary artery endothelial and smooth muscle cells, which contributes to PAH. In two rat models of PAH, lentiviral-mediated lung delivery of miR-424 and the rat homolog of miR-503 decreased pulmonary hypertension compared with a control vector. Next steps include developing a nonviral method for microRNA delivery to the lungs.	Patent application filed; available for licensing	Kim, J. <i>et al. Nat. Med.</i> ; published online Dec. 23, 2012; doi:10.1038/nm.3040 Contact: Hyung Chun, Yale School of Medicine, New Haven, Conn. e-mail: hyung.chun@yale.edu
		SciBX 6(3); doi:10.1038/scibx.2013.55 Published online Jan. 24, 2013		
Endocrine/me	etabolic disease			
Mitochondrial disease	Not applicable	Nuclear transfer between oocytes could prevent transmission of disorders related to mutations in mitochondrial DNA. In unfertilized denucleated oocytes from female donors, fusion with oocyte nuclear DNA from another donor generated cells with exchanged nuclear genotype and less than 1% carryover of mitochondrial DNA. The resulting oocytes showed normal differentiation and mitochondrial function and did not show detrimental spontaneous oocyte activation. Next steps could include testing the strategy in larger samples.	Patent and licensing status unavailable	Paull, D. <i>et al. Nature</i> ; published online Dec. 19, 2012; doi:10.1038/nature11800 Contact: Dieter Egli, The New York Stem Cell Foundation Laboratory, New York, N.Y. e-mail: d.egli@nyscf.org Contact: Mark V. Sauer, Columbia University, New York, N.Y.
		<i>SciBX</i> 6(3); doi:10.1038/scibx.2013.56 Published online Jan. 24, 2013		e-mail: mvs9@columbia.edu
Infectious dis	ease			
Clostridium	Calcitonin receptor-like (CALCRL; CRLR)	Rat studies suggest decreasing CRLR expression in the intestines could help treat <i>Clostridium difficile</i> infection. In rats, double-stranded RNA-mediated inhibition of CRLR expression in the ileum prior to injection of <i>C. difficile</i> toxin A decreased both intestinal secretions and toxin-induced inflammation compared with no inhibition. Next steps could include testing the effects of decreasing CRLR in additional models of <i>C. difficile</i> infection.	Patent and licensing status unavailable	Bhargava, A. <i>et al. Proc. Natl.</i> <i>Acad. Sci. USA</i> ; published online Dec. 24, 2012; doi:10.1073/pnas.1219733110 Contact: Susan E. Leeman, Boston University School of Medicine, Boston, Mass. e-mail: sleeman@bu.edu
		SciBX 6(3); doi:10.1038/scibx.2013.57 Published online Jan. 24, 2013		-

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This week in therapeutics (continued)

MG-CoA ductase	Mouse studies suggest statins could help prevent malaria- and sepsis-associated cognitive dysfunction. In mouse models of malaria infection, the generic malaria drug chloroquine plus Mevacor lovastatin decreased neuroinflammation and cognitive impairment compared with chloroquine alone. In a mouse model of bacterial sepsis, antibiotics plus Mevacor decreased cognitive impairment compared with antibiotics alone. Next steps include starting clinical trials of statin drugs to prevent malaria or bacterial sepsis–associated cognitive impairment. Merck & Co. Inc. markets Mevacor, an HMG-CoA reductase inhibitor, to treat hypercholesterolemia and reduce the risk of	Unpatented; licensing status not applicable	Reis, P.A. <i>et al. PLoS Pathog.</i> ; published online Dec. 27, 2012; doi:10.1371/journal.ppat.1003099 Contact: Patricia A. Reis, Oswaldo Cruz Foundation, Rio de Janeiro, Brazil e-mail: reispa@gmail.com
	cardiovascular diseases. SciBX 6(3); doi:10.1038/scibx.2013.58		
	Published online Jan. 24, 2013		
ctadherin IFGE8; HMFG)	Mouse studies suggest increasing MFGE8 levels could help treat asthma. In two mouse models of allergen-induced asthma, <i>Mfge8</i> knockout increased asthma-associated airway hyperresponsiveness compared with no knockout. Tracheal rings from <i>Mfge8</i> knockout mice showed greater sensitivity to cytokines that cause airway hyperresponsiveness than rings from wild-type mice. In tracheal rings from humans and <i>Mfge8</i> knockout mice, recombinant MFGE8 prevented the cytokine- induced muscle contractions. Next steps include optimizing the recombinant protein and testing it in animal models.	Findings unpatented; available for licensing	Kudo, M. <i>et al. Proc. Natl. Acad.</i> <i>Sci. USA</i> ; published online Dec. 26, 2012; doi:10.1073/pnas.1216673110 Contact: Kamran Atabai, University of California, San Francisco, Calif. e-mail: kamran.atabai@ucsf.edu
	<i>SciBX</i> 6(3); doi:10.1038/scibx.2013.59 Published online Jan. 24, 2013		
eural precursor Il expressed velopmentally wnregulated 8 (EDD8); NEDD8 tivating enzyme (AE); ubiquitin- njugating zyme E2M (BE2M; UBC12)	Mouse and cell culture studies suggest inhibiting the NEDD8 signaling pathway could help prevent inflammation. In cell culture, the NAE inhibitor MLN4924 or small hairpin RNA against Ubc12, a component of the NEDD8 pathway, decreased CD4 ⁺ T cell activation and proliferation compared with no treatment or control shRNA. In a mouse model of airway inflammation, transfer of T cells with Ubc12 knockdown caused less T cell-mediated lung inflammation than transfer of T cells with no knockdown. Next steps could include evaluating clinical-stage NEDD8 pathway inhibitors in additional inflammation models. Takeda Pharmaceutical Co. Ltd.'s Millennium Pharmaceuticals Inc. unit has MLN4924 in Phase I testing to treat various cancers.	Patent and licensing status unavailable	Jin, Hs. <i>et al. Proc. Natl. Acad.</i> <i>Sci. USA</i> ; published online Dec. 24, 2012; doi:10.1073/pnas.1213819110 Contact: Yun-Cai Liu, La Jolla Institute for Allergy & Immunology, La Jolla, Calif. e-mail: yuncail@liai.org
	Sc <i>iBX</i> 6 (3); doi:10.1038/scibx.2013.60 Published online Jan. 24, 2013		
ot applicable	Rodent studies identified hapten-protein conjugates that could be used as oxycodone immunogens to help treat opioid addiction. In rats, vaccination with the hapten-protein conjugates increased oxycodone-specific antibody titers in serum compared with vaccination using an unconjugated control compound. In mice and rats given oxycodone or hydrocodone, vaccination with the hapten-protein conjugates decreased the brain concentrations and antinociceptive effects of the drugs compared with vaccination using the unconjugated hapten. Next steps could include testing conjugates in large animal models of opioid addiction. <i>SciBX</i> 6(3): doi:10.1038/scibx.2013.61	Patent and licensing status unavailable	Pravetoni, M. <i>et al. J. Med. Chem.</i> ; published online Dec. 18, 2012; doi:10.1021/jm3013745 Contact: Marco Pravetoni, Minneapolis Medical Research Foundation, Minneapolis, Minn. e-mail: prave001@umn.edu
eu ll Vvww IE IA n Z IE	applicable	ral precursor expressedMouse and cell culture studies suggest inhibiting the NEDD8 signaling pathway could help prevent inflammation. In cell culture, the NAE inhibitor MLN4924 or small hairpin RNA against Ubc12, a component of the NEDD8 pathway, decreased CD04* T cell activation and proliferation compared with no vating enzyme tE); ubiquitin- jugating caused less T cell-mediated lung inflammation than transfer of T cells with no knockdown. Next steps could include evaluating clinical-stage NEDD8 pathway inhibitors in additional inflammation models. Takeda Pharmaceutical Co. Ltd's Millennium Pharmaceuticals Inc. unit has MLN4924 in Phase I testing to treat various cancers.sciBX 6(3); doi:10.1038/scibx.2013.60 Published online Jan. 24, 2013applicableRodent studies identified hapten-protein conjugates that could be used as oxycodone immunogens to help treat opioid addiction. In rats, vaccination with the hapten-protein conjugates increased oxycodone-specific antibody titers in serum compared with vaccination using an unconjugates decreased the brain concentrations and antinociceptive effects of the drugs compared with vaccination using the unconjugates in large animal models of opioid addiction. SciBX 6(3); doi:10.1038/scibx.2013.61	ral precursor expressed signaling pathway could help prevent inflammation. In cell culture, the NAE inhibitor MLN4924 or small hairpin RNA against Ubc12, a component of the NEDD8 pathway, decreased DD98); NEDD8 CD4* T cell activation and proliferation compared with no treatment or control shRNA. In a mouse model of airway inflammation, transfer of T cells with Ubc12 knockdown caused less T cell-mediated lung inflammation than transfer of T cells with no knockdown. Next steps could include evaluating clinical-stage NEDD8 pathway inhibitors in additional inflammation models. Takeda Pharmaceutical Co. Ltd's Millennium Pharmaceuticals Inc. unit has MLN4924 in Phase I testing to treat various cancers. <i>SciBX</i> 6(3); doi:10.1038/scibx.2013.60 Published online Jan. 24, 2013 applicable Rodent studies identified hapten-protein conjugates that could be used as oxycodone immunogens to help treat opioid addiction. In rats, vaccination with the hapten-protein conjugates increased oxycodone-specific antibody titers in serum compared with vaccination using an unconjugates decreased the brain concentrations and antinociceptive effects of the drugs compared with vaccination using the unconjugates in large animal models of opioid addiction. <i>SciBX</i> 6(3); doi:10.1038/scibx.2013.61

This week in therapeutics (continued)

Indication	Target/marker/ pathway	Summary	Licensing status	Publication and contact information
Alzheimer's disease (AD)	NLR family pyrin domain containing 3 (NLRP3; NALP3); caspase-1 (CASP1)	Human tissue, cell culture and mouse studies suggest antagonizing NALP3 or CASP1 could help prevent AD. In postmortem patient brain tissue samples, CASP1 activity was higher than that in tissue from healthy controls. In a transgenic mouse model of AD, deletion of <i>Nalp3</i> or <i>Casp1</i> increased learning and memory and decreased both neuroinflammation and accumulation of β -amyloid (A β) aggregates compared with no deletion. Next steps include identifying compounds that inhibit <i>Nalp3</i> or <i>Casp1</i> and testing them in mouse models of AD. SciBX 6(3) ; doi:10.1038/scibx.2013.62	Unpatented; licensing status not applicable	Heneka, M.T. <i>et al. Nature</i> ; published online Dec. 19, 2012; doi:10.1038/nature11729 Contact: Eicke Latz, University of Bonn, Bonn, Germany e-mail: eicke.latz@uni-bonn.de Contact: Michael T. Heneka, same affiliation as above e-mail: michael.heneka@ukb.uni-bonn.de
Autism	Eukaryotic translation initiation factor 4E (eIF4E)	 Published Online Jan. 24, 2013 Mouse studies suggest inhibiting eIF4E could help treat autism spectrum disorder (ASD). In mice, greater eif4e expression led to increased protein translation, electrophysiological abnormalities and autistic behaviors compared with normal eif4e expression. In those autistic mice, a small molecule inhibitor of eIF4E signaling decreased protein translation and autistic behaviors compared with vehicle and reversed electrophysiological abnormalities. Next steps include testing whether targeting eif4e in mouse models of fragile X syndrome corrects disease-associated autistic behaviors and developing a method to measure the translation of various proteins in mouse models of autism. Isis Pharmaceuticals Inc.'s eIF-4E ASO, a second-generation antisense compound targeting eIF4E, is in Phase II testing to treat prostate cancer and non-small cell lung cancer (NSCLC). Clavis Pharma ASA and Translational Therapeutics Inc. have TRX-201, a Lipid Vector Technology (LVT) derivative of ribavirin that inhibits eIF4E, in preclinical development to treat thyroid cancer. SciBX 6(3); doi:10.1038/scibx.2013.63 	Patent and licensing status unavailable	Santini, E. <i>et al. Nature</i> ; published online Dec. 23, 2012; doi:10.1038/nature11782 Contact : Eric Klann, New York University, New York, N.Y. e-mail: eklann@cns.nyu.edu
Multiple sclerosis (MS)	Annexin A1 (ANXA1)	Published online Jan. 24, 2013 Patient sample and mouse studies suggest ANXA1 could help restore blood brain barrier (BBB) integrity in patients with MS. In mice, <i>Anxa1</i> knockout increased BBB permeability compared with no knockout. In the same mice, i.v. infusion of recombinant human ANXA1 decreased BBB permeability compared with no treatment. Cerebral capillaries from patients who have MS showed selective loss of ANXA1, whereas capillaries from healthy donors did not. Next steps include analyzing ANXA1 levels in blood samples from patients at different stages of MS to see if the protein could be a useful biomarker. SciBX 6(3); doi:10.1038/scibx.2013.64 Published online Jan. 24, 2013	Work unpatented; models available for licensing	Cristante, E. <i>et al. Proc. Natl. Acad.</i> <i>Sci. USA</i> ; published online Dec. 31, 2012; doi:10.1073/pnas.1209362110 Contact: Egle Solito, University of London, London, U.K. e-mail: e.solito@qmul.ac.uk

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			
Adapted <i>Plasmodium</i> <i>knowlesi</i> parasites that infect cultured human red blood cells	Culture of modified <i>P. knowlesi</i> strains could help identify new malaria therapies. Wild-type <i>P. knowlesi</i> survive and replicate in macaque but not human red blood cells. The parasites were adapted to infect human red blood cells by culturing them in a 4:1 mixture of human and macaque cells. After eight months of continuous coculture, the adapted parasites infected and replicated in cultured human red blood cells. Next steps include using the adapted parasite to screen for therapeutics and vaccines against <i>P. knowlesi</i> and related parasites. <i>SciBX</i> 6(3); doi:10.1038/scibx.2013.65 Published online Jan. 24, 2013	Unpatented; licensing status not applicable	Moon, R.W. <i>et al. Proc. Natl. Acad. Sci.</i> <i>USA</i> ; published online Dec. 24, 2012; doi:10.1073/pnas.1216457110 Contact: Michael J. Blackman, Medical Research Council National Institute for Medical Research, London, U.K. e-mail: mblackm@nimr.mrc.ac.uk Contact: Anthony A. Holder, same affiliation as above e-mail: aholder@nimr.mrc.ac.uk Contact: Robert W. Moon, same affiliation as above e-mail:
Multiple annealing and looping-based amplification cycles (MALBAC) to identify single- nucleotide and copy- number variations in single human cells	MALBAC could be used to identify genomic variations at the single cell level. MALBAC uses a genome amplification step that introduces less bias than current sequencing methods. In single cells from a human colon adenocarcinoma cell line, MALBAC sequencing provided 85%–93% genome coverage, whereas standard sequencing methods provided 72% coverage. In the same cells, MALBAC could detect single-nucleotide variations with 76% efficiency compared with 41% for standard sequencing. Next steps include applying MALBAC-based single-cell genome sequencing in cases for which sample size is limited, such as circulating tumor cells. SciBX 6(3); doi:10.1038/scibx.2013.66	Patent application filed; Harvard University's Office of Technology Development is in licensing and commercialization discussions with undisclosed entities	Zong, C. <i>et al. Science</i> ; published online Dec. 21, 2012; doi:10.1126/science.1229164 Contact: X. Sunney Xie, Harvard University, Cambridge, Mass. e-mail: xie@chemistry.harvard.edu
Serum protein panel for identifying type 1 diabetes	Published online Jan. 24, 2013 An assay of serum peptides could help identify patients with type 1 diabetes. In serum samples from patients with type 1 diabetes and healthy controls, proteomics analyses identified 24 proteins that were significantly associated with the disease (<i>p</i> <0.05). A multiplexed peptide assay was created to detect and quantify 52 peptides that are surrogates for the 24 identified proteins. In blinded serum samples from independent patient cohorts, the peptide assay distinguished patients with type 1 diabetes from healthy subjects and from patients with type 2 diabetes. Next steps include validating the predictive capabilities of the peptide assay in additional patient cohorts. <i>SciBX</i> 6(3); doi:10.1038/scibx.2013.67 Published online Jan. 24, 2013	Patent application filed; available for licensing from Pacific Northwest National Laboratory Technology Transfer Contact: Ron Thomas, Pacific Northwest National Laboratory, Richland, Wash. e-mail: ron.thomas@pnnl.gov	Zhang, Q. <i>et al. J. Exp. Med.</i> ; published online Dec. 31, 2012; doi:10.1084/jem.20111843 Contact: Thomas O. Metz, Pacific Northwest National Laboratory, Richland, Wash. e-mail: thomas.metz@pnnl.gov Contact: Qibin Zhang, same affiliation as above e-mail: qibin.zhang@pnnl.gov

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This week in techniques (continued)

Approach	Summary	Licensing status	Publication and contact information
Chemistry			
Photosensitive small molecule-mediated cleavage of RNA	Photosensitive small molecules that bind and cleave RNA could help treat diseases driven by aberrant RNA transcripts, such as myotonic dystrophy type 1 (DM1). DM1 is caused by expanded r(CUG) repeats in dystrophia myotonica-protein kinase (DMPK) mRNA. An <i>N</i> -hydroxypyridine-2(1 <i>H</i>)-thione was developed that specifically bound expanded DMPK mRNA and released hydroxyl radicals that cleaved r(CUG) repeats when photoactivated. In cell culture studies, the compound improved a DM1-associated splicing defect and decreased the level of expanded r(CUG) repeats compared with an r(CUG) repeat-binding control compound. Next steps include developing photoactivated small molecules that can target cancer-associated RNA in tumors. <i>SciBX</i> 6(3); doi:10.1038/scibx.2013.68	Covered by issued and pending patents; available for licensing; undisclosed aspects of technology licensed by SMaRT Therapeutics Inc.	Guan, L. & Disney, M.D. Angew. Chem. Int. Ed.; published online Dec. 20, 2012; doi:10.1002/anie.201206888 Contact : Matthew D. Disney, Scripps Florida, Jupiter, Fla. e-mail: disney@scripps.edu
	Published online Jan. 24, 2013		
Disease models Sortilin 1 (Sort1) knockout mouse model for Alzheimer's disease (AD)	<i>Sort1</i> knockout mice could be useful models for AD pathophysiology. Two existing transgenic mouse AD models were crossed with <i>Sort1</i> knockout mice to generate AD-prone mice with a <i>Sort1</i> deficiency. The <i>Sort1</i> -deficient mice had impaired amyloidogenic processing and accelerated plaque formation and had higher β-amyloid (Aβ) levels in the brain the properties of the levels in the brain.	Patent and licensing status unavailable	Carlo, AS. <i>et al. J. Neurosci.</i> ; published online Jan. 2, 2013; doi:10.1523/JNEUROSCI.2425-12.2013 Contact: Thomas E. Willnow, Max Delbrueck Center for Molecular Medicine, Berlin, Germany
	knockout mice to evaluate AD therapeutics.		e-mail: willnow@mdc-berlin.de
	<i>SciBX</i> 6(3); doi:10.1038/scibx.2013.69 Published online Jan. 24, 2013		
Drug delivery			
Polyisoprene nanoparticles for cancer drug delivery	Polyisoprene nanoparticles could be used to deliver drugs to cancer cells. Polyisoprene nanoparticles were conjugated to alkoxyamine-coupled Gemzar gemcitabine using nitroxide-mediated polymerization. In a panel of four human cancer cell lines, the drug-loaded nanoparticles had nanomolar IC_{50} values. In a mouse xenograft model of human pancreatic cancer, the drug-loaded nanoparticles decreased tumor volume compared with free gemcitabine, non-drug-loaded nanoparticles or saline. Next steps could include conjugating the particles with other chemotherapies and evaluating them in additional cancer models. Eli Lilly and Co. markets the nucleoside analog Gemzar to treat pancreatic and other cancers.	Patent and licensing status unavailable	Harrisson, S. <i>et al. Angew. Chem. Int. Ed.</i> ; published online Dec. 17, 2012; doi:10.1002/anie.201207297 Contact: Julien Nicolas, University of Paris-Sud 11, Châtenay-Malabry, France e-mail: julien.nicolas@u-psud.fr
	<i>SciBX</i> 6(3); doi:10.1038/scibx.2013.70 Published online Jan. 24, 2013		
Drug platforms			
Cell-based therapy for photoreceptor layer reconstruction to treat age- related macular degeneration (AMD) and retinitis	Transplantation of rod precursors could help reconstruct the outer nuclear layer of photoreceptors to treat AMD and retinitis. In a mouse model of severe human retinitis pigmentosa, subretinal transplantation of photoreceptor precursor cells from postnatal mice led to regeneration of the outer retinal layer of lost photoreceptors. In the same mice, the precursor cells differentiated into mature light-sensing rods that integrated with neuronal synapses, leading to visual function as characterized by the pupil light response. Next steps include differentiating embryonic stem cells to rod precursors for use in subretinal transplantation.	Unpatented; licensing status not applicable	Singh, M.S. <i>et al. Proc. Natl. Acad. Sci.</i> <i>USA</i> ; published online Jan. 3, 2013; doi:10.1073/pnas.1119416110 Contact: Robert E. MacLaren, University of Oxford, Oxford, U.K. e-mail: enquiries@eye.ox.ac.uk
	Published online Jan. 24, 2013		

This week in techniques (continued)

Approach	Summary	Licensing status	Publication and contact information
Crystal structure of αβ-tubulin in complex with anticancer microtubule- stabilizing agents	Protein crystal structure studies identified the molecular mechanism of microtubule-stabilizing cancer therapeutics, which could help guide the development of new treatments. Crystallography studies of $\alpha\beta$ -tubulin complexed with two microtubule-stabilizing cancer therapies—zampanolide and epothilone A—showed that the compounds bind to a hydrophobic pocket on the tubulin- β subunit known as the taxane pocket. Next steps include using the structural data to develop analogs of zampanolide and epothilone A and investigating the mechanism of additional tubulin-targeting compounds. Zampanolide and epothilone A are research reagents.	Unpatented; licensing status not applicable	Prota, A.E. <i>et al. Science</i> ; published online Jan. 3, 2013; doi:10.1126/science.1230582 Contact : Michel O. Steinmetz, Paul Scherrer Institute, Villigen, Switzerland e-mail: michel.steinmetz@psi.ch
	<i>SciBX</i> 6(3); doi:10.1038/scibx.2013.72 Published online Jan. 24, 2013		
Crystal structure of the antifibrotic halofuginone bound to the prolyl-tRNA synthetase domain of human glutamyl- prolyl-tRNA synthetase (EPRS)	The crystal structure of the antifibrotic halofuginone bound to the prolyl-tRNA synthetase domain of human EPRS could aid the development of new compounds to target the synthetase. Halofuginone is a halogenated derivative of the alkaloid febrifugine, which is the active compound in a Chinese herb used to treat malaria-induced fever. The crystal structure showed that halofuginone is an ATP-dependent inhibitor. Next steps could include optimizing the halofuginone analogs and determining how such compounds bind to other tRNA synthetases.	Unpatented; aTyr Pharma Inc. has an exclusive option to license the work	Zhou, H. <i>et al. Nature</i> ; published online Dec. 23, 2012; doi:10.1038/nature11774 Contact: Paul Schimmel, The Scripps Research Institute, La Jolla, Calif. e-mail: schimmel@scripps.edu
	<i>SciBX</i> 6(3); doi:10.1038/scibx.2013.73 Published online Jan. 24, 2013		
Genetic correction of spinal muscular atrophy (SMA) neurons with a nonviral, nonintegrating vector	<i>In vitro</i> and mouse studies suggest genetic modification of patient-derived neurons with a nonviral, nonintegrating vector could help treat SMA. In induced pluripotent stem (iPS) cells derived from the fibroblasts of a patient with SMA, an episomal vector was used to convert the gene encoding <i>survival</i> <i>of motor neuron 2 centromeric</i> (<i>SMN2</i>) into a <i>survival of motor</i> <i>neuron 1 telomeric</i> (<i>SMN1</i>)-like gene that produced the full- length SMN protein. In a mouse model of SMA, spinal cord engraftment of neurons derived from the vector-treated human iPS cells decreased disease severity and increased lifespan and neuron engraftment compared with engraftment of cells derived from untreated iPS cells. Next steps could include testing the strategy in animal models of more advanced SMA. Isis Pharmaceuticals Inc. and Biogen Idec Inc. have ISIS- SMNRx, an antisense oligonucleotide modulating the splicing of <i>SMN2</i> pre-mRNA, in Phase I/II testing to treat SMA.	Patent and licensing status unavailable	Corti, S. <i>et al. Sci. Transl. Med.</i> ; published online Dec. 19, 2012; doi:10.1126/scitranslmed.3004108 Contact: Giacomo P. Comi, University of Milan, Milan, Italy e-mail: giacomo.comi@unimi.it
	<i>SciBX</i> 6(3); doi:10.1038/scibx.2013.74 Published online Jan. 24, 2013		
Micelle drug delivery with a modified tumor- targeting Tat peptide	<i>In vitro</i> and mouse studies identified modified Tat peptides that could improve drug delivery to tumors. Tat can be used to target compounds to tumors, but the peptide's positive charges can cause nonspecific blood interactions and cardiotoxicity. In tumor cells, micelles functionalized with Tat where positively charged lysine residues were replaced with succinylamides showed greater tumor cell penetration than micelles functionalized with unmodified Tat. In a mouse xenograft model of human breast cancer, doxorubicin-loaded micelles with the modified Tat had longer blood circulation times and led to less tumor growth and cardiotoxicity than micelles using unmodified Tat. Next steps could include using the modified Tat peptide to enhance delivery of other cancer drugs. <i>SciBX</i> 6(3); doi:10.1038/scibx.2013.75	Patent and licensing status unavailable	Jin, E. <i>et al. J. Am. Chem. Soc.</i> ; published online Dec. 19, 2012; doi:10.1021/ja311180x Contact: Maciej Radosz, University of Wyoming, Laramie, Wyo. e-mail: radosz@uwyo.edu

This week in techniques (continued)

Published online Jan. 24, 2013

Approach	Summary	Licensing status	Publication and contact information
Imaging			
Diffuse reflectance spectroscopy to detect microcalcified breast cancer lesions	Studies of breast cancer samples suggest diffuse reflectance spectroscopy could help detect microcalcified breast cancer lesions during standard biopsies. Breast cancer biopsy techniques target microcalcified lesions because lesions lacking microcalcifications can result in false negative hits. In core biopsy samples from 23 patients, diffuse reflectance spectra were obtained for cancerous lesions with and without microcalcifications and for benign lesions. A computational algorithm incorporating the spectra detected microcalcified lesions with 97% positive predictive power and 88% negative predictive power. Next steps include devising a miniature optical fiber probe to conduct the studies with the needle device currently used in breast biopsies and conducting a large-scale clinical trial.	Patent application filed by Massachusetts Institutes of Technology and co-inventors; will be available for licensing in the near future	Soares, J.S. <i>et al. Proc. Natl. Acad. Sci.</i> <i>USA</i> ; published online Dec. 24, 2012; doi:10.1073/pnas.1215473110 Contact: Maryann Fitzmaurice, Case Western Reserve University, Cleveland, Ohio e-mail: maryann.fitzmaurice@case.edu
	SCIDA 0(3); UUI: 1U. 1U30/SCIDX.2013.76		

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