BIOCENTURY
Innovations
FROM IDEA TO IND

FEBRUARY 18, 2016

PRODUCT R&D

CROSSING PATHS

By Selina Koch, Staff Writer

With its first foray into the brain, Catabasis Pharmaceuticals Inc. is using its linker technology to simultaneously target the inflammatory and oxidative stress pathways that drive neurodegeneration, starting with amyotrophic lateral sclerosis (ALS) and Friedreich's ataxia. While the two diseases affect different neurons, have different pathologies and cause different symptoms, the company believes there's enough common ground in the pathway biology to develop a single compound for both, and plans to complete IND-enabling studies by year-end.

The compound, CAT-4001, joins the NF-κB inhibitor docosahexaenoic acid (DHA), an anti-inflammatory, with the NRF2 agonist fumarate, which activates antioxidant pathways. (See "Catabasis Goes Neuro")

It came about through Catabasis' strategy of finding independent pathways that drive disease, and linking together drugs that might produce better outcomes if they hit the same cell at the same time than if delivered independently. CAT-4001 is Catabasis' fourth disclosed candidate.

Andrew Nichols, SVP of research and non-clinical development, told BioCentury CAT-4001 evolved from its CAT-1004 program, which targets the NF-κB pathway by joining DHA with the anti-inflammatory compound salicylate.

CAT-1004 is in Phase I/II for Duchenne muscular dystrophy (DMD). The other two candidates, CAT-2054 and CAT-2003, are in Phase II for hypercholesterolemia and hypertriglyceridemia, respectively.

Nichols said as the company looked further into NF-κB signaling, it found evidence in the literature that the transcription factor is chronically activated in ALS and Friedreich's ataxia.

According to Nichols, genome-wide association studies have turned up multiple hits upstream of NF-κB. "In both genetic and sporadic cases of ALS, a variety of biological pathways are activated that converge on NF-κB" to trigger inflammation, he said. "A lot of the roads lead to NF-κB, and they can come from different directions."

Once activated, the pathway turns on transcription of a variety of factors including other inflammatory molecules, creating a self-propagating feedback loop. The chronic inflammation coupled with the oxidative damage also present in the disease eventually cause motor neurons to die, said Nichols.

BIOCENTURY PRODUCT PROFILE	
INNOVATION STAGE	
Product	CAT-4001, a conjugate of fumarate and docosahexaenoic acid (DHA)
Concept	A dual-mechanism treatment for ALS and Friedreich's ataxia that inhibits both inflammation and oxidative damage to prevent loss of neurons
Disease	Amyotrophic lateral sclerosis (ALS); Friedreich's ataxia
Competition	Therapies that either inhibit inflammation or oxidative damage
Differentiation	Triggers anti-inflammatory and antioxidant responses in the same cells at the same time to produce synergistic responses
Administration	Oral
Risks	Side effects due to actions of the compound in non-neural tissues
Development status	IND-enabling studies
Patents	Patented
Company; lead investigator	Catabasis Pharmaceuticals Inc.

He emphasized, "It's really a mash of emerging biology with these molecules, and what our platform can do, that has led us into these neurodegenerative diseases."

CEO Jill Milne noted that the approach used to create CAT-4001 is in keeping with the company's strategy of targeting downstream mechanisms driving disease rather than genetic causes. "We anticipate that CAT-4001's mechanism of action will be agnostic as to the underlying mutation."

LINKING PATHWAYS

At the heart of Catabasis' platform is a set of linkers that can be used to covalently join pairs of small molecules to control at least four aspects of their biology.

First, the linkers force the two compounds to have identical tissue distributions and PK profiles.

Second, by connecting the bioactive regions of each compound to the linker, the molecules remain inactive while in circulation,

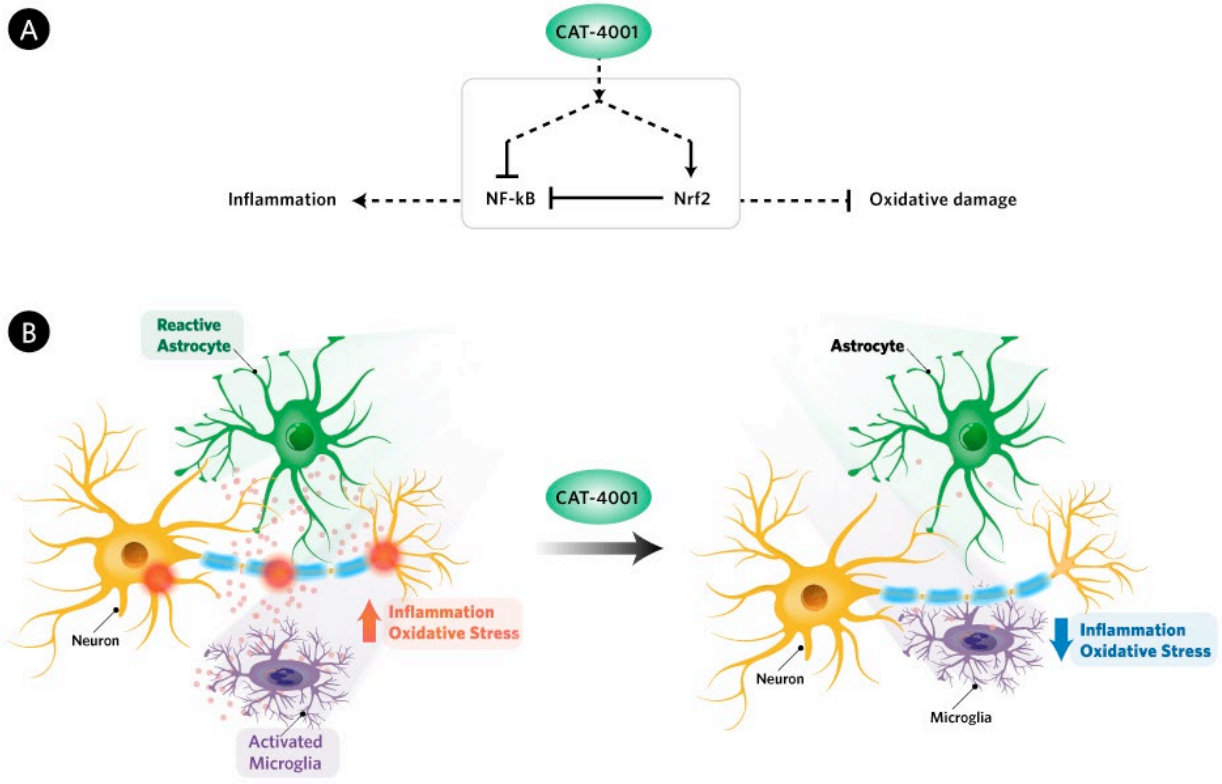
CATABASIS GOES NEURO

Catabasis Pharmaceuticals Inc., which has three compounds in the clinic for a variety of metabolic, autoimmune and musculoskeletal conditions, is stepping into the neurodegenerative disease space with its preclinical asset CAT-4001. Like the clinical candidates, CAT-4001 uses the company's linker technology to join two bioactive molecules intended to target multiple disease pathways in the same cells.

(A) CAT-4001 consists of docosahexaenoic acid (DHA), which decreases inflammation by inhibiting the NF-κB pathway, and fumarate, which activates NRF2 to up-regulate protective antioxidant pathways, and also inhibits the NF-κB pathway. The company is testing the conjugate in preclinical models of amyotrophic lateral sclerosis (ALS) and Friedreich's ataxia.

(B) ALS and Friedreich's ataxia affect different types of neurons, but the loss of motor neurons in the former and sensory neurons in the latter are both thought to be driven by excessive inflammation and oxidative stress. Literature evidence suggests the NF-κB pathway is chronically turned on in activated microglia and reactive astrocytes, which release inflammatory molecules and reactive oxygen species **(small pink dots)** that cause neuronal damage **(red spots)**, eventually leading to neuronal death and progressively worsening symptoms in patients. CAT-4001 is meant to slow disease progression by reducing NF-κB-mediated inflammation and up-regulating NRF2-dependent antioxidant pathways.

NRF2 (NFE2L2) - Nuclear factor (erythroid-derived 2)-like 2; NF-κB (NFKB1; p105; p50) - Nuclear factor of κ light polypeptide gene enhancer in B cells 1. *Reproduced with permission from Catabasis Pharmaceuticals Inc.*



which allows Catabasis to control when and where the target pathways are activated.

Third, the linkers can influence how the compounds are taken up by cells and where they are trafficked inside.

Finally, the linkers are designed to be cleaved by intracellular enzymes, which release the bioactive compounds to bind their respective targets. (See "Attacking Networks")

Last month, Catabasis disclosed its linker technology for the first time in a paper in the *Journal of Medicinal Chemistry*, and explained how it combines chemistry, cellular biology and PK to optimize the activity of the conjugates in specific disease pathways. The study focused on CAT-2054 and CAT-1004.

To create CAT-2054, which contains niacin and eicosapentaenoic acid (EPA), the company began by testing at least five linkers comprising one or two carbons and either ester or amide bonds connected to the bioactive molecules.

The linker containing two amide bonds yielded "complete plasma stability" with no sign of breakdown after two hours in mouse, rat, dog or human plasma. By contrast, less than 40% of the other conjugates, whose linkers all contained at least one ester bond, remained.

The stable conjugate displayed different cellular uptake and trafficking than unbound EPA, which enters cells by a two-step process in which it integrates into the plasma membrane and is released intracellularly by phospholipases. Then, it is trafficked to lysosomes for degradation.

However, in a human liver cell line, the niacin-EPA conjugate entered cells via endocytosis — most likely via clathrin-coated vesicles — and was then trafficked to the endoplasmic reticulum, where its amide bonds were cleaved by the integral membrane enzyme FAAH.

The authors noted that while FAAH caused the cleavage in the *in vitro* system, *in vivo* the compound may be cleaved by an array of amidases and hydrolases.

Because PK studies in rats showed the conjugate was extensively hydrolyzed in the intestine, Catabasis tweaked the linker structure to slow down the rate of hydrolysis. That produced an approximately 60-fold increase in the distribution to the liver — the compound's target organ.

In a mouse model of familial dysbetalipoproteinemia, the lead compound lowered plasma levels of PCSK9, low-density lipoprotein (LDL) cholesterol and triglycerides to a greater extent than either niacin or EPA alone, or co-administration of the two as free molecules.

ATTACKING NETWORKS

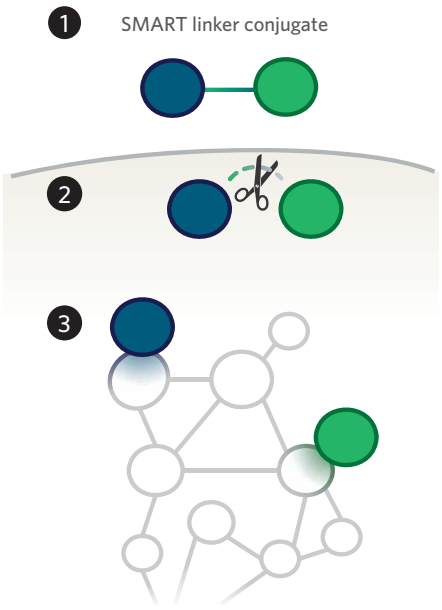
Catabasis Pharmaceuticals Inc. has developed its linker platform to deliver pairs of compounds with identical tissue distributions and PK profiles, allowing simultaneous targeting of multiple disease pathways.

(1) A selected linker **(green line)** is covalently bound to the bioactive regions of compound A **(blue)** and compound B **(green)**, which connects the two and renders both inactive while in circulation.

(2) The conjugate enters cells via endocytosis and can be cleaved by a variety of intracellular enzymes to release compounds A and B.

(3) The free compounds bind their respective targets to alter the activity of networks of disease pathways in the same cells, at the same time.

Reproduced with permission from Catabasis Pharmaceuticals Inc.



Catabasis showed similar results were achieved with conjugates of DHA and salicylate. Those too were endocytosed by liver cells, trafficked to the ER and cleaved by FAAH. In rats, the compound and its constituent molecules concentrated in the same tissues and synergistically inhibited NF-κB.

PARALLEL PATHS IN ALS

The table lists companies that are directly targeting inflammation or oxidative stress pathways that drive neurodegeneration in amyotrophic lateral sclerosis (ALS), indicating there are at least 11 anti-inflammatory and 5 antioxidant compounds in development. CAT-4001 from **Catabasis Pharmaceuticals Inc.** (NASDAQ:CATB) is designed to treat both pathologies and to treat all patients regardless of mutation status, whereas therapies targeting mutant superoxide dismutase (SOD1) would be restricted to a subset of patients. *Source: BCIQ: BioCentury Online Intelligence, ClinicalTrials.gov, company documents and websites*

ANTI-INFLAMMATORY COMPOUNDS			
Company	**Product**	**Description**	**Status**
MediciNova Inc. (NASDAQ:MNOV; JASDAQ:4875) / **Kyorin Pharmaceutical Co. Ltd.** (Tokyo:4569)	Ibudilast (MN-166, AV411)	Oral small molecule inhibitor of phosphodiesterase-4 (PDE-4), PDE-10 and macrophage migration inhibitory factor (MIF) that suppresses pro-inflammatory cytokines	Phase II
Neuraltus Pharmaceuticals Inc.	NP001	Small molecule regulator of macrophage activation	Phase II
Global Neurotech (GNT) Pharma Co. Ltd.	AAD-2004	Microsomal prostaglandin E synthase-1 (PTGES; mPGES-1) inhibitor	Phase I
Anida Pharma Inc.	Neuroprotectin D1	Resolvin derived from the oxidation of the omega-3 fatty acid docosahexaenoic acid (DHA)	Preclinical
ArmaGen Inc.	AGT-110	IgG-decoy receptor fusion protein that inhibits tumor necrosis factor α (TNFα)	Preclinical
Catabasis Pharmaceuticals	CAT-4001	Conjugate of DHA and fumarate	Preclinical
FPRT Bio Inc. / **Xencor Inc.** (NASDAQ:XNCR)	XPro1595	Dominant-negative inhibitor of TNFα	Preclinical
ImStar Therapeutics Inc.	IMS-088	Small molecule derived from withaferin A	Preclinical
miRagen Therapeutics Inc.	MRG-107	MicroRNA-155 (miR-155) antagonist	Preclinical
Regenesance B.V.	Regenemab	mAb against complement 6 (C6)	Preclinical
Tarix Orphan LLC	TXA127	Formulation of the angiotensin (1-7) peptide	Preclinical
COMPOUNDS TARGETING OXIDATIVE STRESS PATHWAYS			
Company	**Product**	**Description**	**Status**
Treeway B.V.	TW001	Oral formulation of edaravone, a free radical scavenger cerebral neuroprotectant	Phase I
Biogen Inc. (NASDAQ:BIIB) / **Ionis Pharmaceuticals Inc.** (NASDAQ:IONS)	IONIS-SOD1Rx (BIIB067)	Antisense inhibitor of SOD1	Phase I
Catabasis Pharmaceuticals	CAT-4001	Conjugate of DHA and fumarate	Preclinical
ProMIS Neurosciences Inc. (TSX:PMN)	mAb against SOD1	mAb against misfolded SOD1	Preclinical
Voyager Therapeutics Inc. (NASDAQ:VYGR)	VY-SOD101	Adeno-associated virus (AAV)-based gene therapy designed to knock down mutant SOD1	Preclinical

SYSTEM UPGRADES

However, while the company has shown its linkers can ensure conjugates remain inactive outside cells and be used to manipulate tissue distribution, it hasn't yet determined how to target specific cell types, such as microglia, the motor neurons that die in ALS or the sensory neurons lost in Friedreich's ataxia.

But Nichols is optimistic, and said making conjugates that are selectively cleaved in target cells is "definitely within the realm of possibility."

"What you need to do is identify an enzyme that's only in those cell types and make linkers that can be cleaved by that enzyme," he said, adding that the company has "lots of ideas" for how to do that.

Additional upgrades in the works include making trifunctional conjugates.

"There's nothing that says that the linker itself has to be inert. We're looking at building molecules where we have two bioactives conjugated by a linker that's also bioactive," said Nichols.

That would allow Catabasis to modulate a wider network of disease pathways. It would also give it the flexibility to retain dual functionality while replacing one of the compounds in the conjugate with a molecule that isn't therapeutic but enables uptake into specific cell types, said Nichols.

In the meantime, the company is moving ahead with CAT-4001, which crosses the blood-brain barrier to enter neurons and microglia but is also distributed throughout the body.

> "There's actually only been a few things that have shown any real level of efficacy in well-designed animal model studies of ALS, and all of them were anti-inflammatories of some sort."
>
> Andrew Nichols, Catabasis

The team is taking the recent Phase I results for CAT-1004, which shares its DHA component with CAT-4001, as a positive sign. The company reported no serious adverse events or treatment discontinuations in the Phase I portion of its Phase I/II trial of CAT-1004 to treat boys of ages four to seven who have DMD.

The other half of CAT-4001, fumarate, is similar to Biogen Inc.'s multiple sclerosis (MS) drug Tecfidera dimethyl fumarate, which carries a small risk of progressive multifocal leukoencephalopathy (PML).

ROAD MOST TRAVELED

Nichols told BioCentury the company is testing CAT-4001 in a transgenic mouse model of ALS that harbors mutations in SOD1 linked to familial forms of the disease.

While he acknowledged other compounds have produced promising results in SOD1 mice only to later fail clinical trials, he said the model can yield reliable results "if it's run robustly."

According to Nichols, some of the previous studies used too few animals and didn't properly control for age or use equal proportions of males and females across groups. "You have to be very careful when you run the model to make sure that, if you're seeing an effect, the only variable that's actually being studied is truly the drug."

After weeding out the rigorous studies from the sloppy ones, he said, "there's actually only been a few things that have shown any real level of efficacy in well-designed animal model studies of ALS, and all of them were anti-inflammatories of some sort."

However, he noted, "most of the things currently being developed for ALS are directed towards specific aspects of inflammation as opposed to inhibition of NF-κB, which is a more broadly applicable approach to the inhibition of the inflammation."

At least 11 compounds have been disclosed that target inflammation, and at least 5 targeting oxidative stress, for ALS. (See "Parallel paths in ALS")

Nichols noted that the situation in Friedreich's ataxia is similar to ALS, but oxidative stress plays a larger role and inflammation a smaller one, and probably also occurs in several other neurodegenerative diseases including Rett syndrome and Parkinson's disease (PD).

"I don't think the role of CAT-4001 is limited just to ALS and Friedreich's ataxia," he said, but added that the company is sticking with those two indications for now because it thinks it could make a significant difference in them.

"Inhibition of NF-κB would not be a cure in the sense that it would not remove the initial stimulus, but it would definitely be disease-modifying in that it would significantly impact a major part of the process that drives the loss of neurons," said Nichols.

Milne added that the two indications also fit squarely into the biotech's rare disease strategy, and the company has no plans to partner. "For us, as a small company, rare diseases make a lot of sense because we can apply our technology to discover product candidates in-house and take them all the way through to commercialization."

COMPANIES AND INSTITUTIONS MENTIONED

Biogen Inc. (NASDAQ:BIIB), Cambridge, Mass.

Catabasis Pharmaceuticals Inc. (NASDAQ:CATB), Cambridge, Mass.

TARGETS AND COMPOUNDS

FAAH - Fatty acid amide hydrolase

NF-κB (NFKB1; p105; p50) - Nuclear factor of κ light polypeptide gene enhancer in B cells 1

NRF2 (NFE2L2) - Nuclear factor (erythroid-derived 2)-like 2

PCSK9 - Proprotein convertase subtilisin/kexin type 9

SOD1 - Superoxide dismutase 1

REFERENCES

Vu, C., et al. "Synthesis and characterization of fatty acid conjugates of niacin and salicylic acid." *Journal of Medicinal Chemistry* **(2016)**

BIOCENTURY
Innovations
FROM IDEA TO IND

EDITORIAL & RESEARCH

NEWSROOM:
pressreleases@biocentury.com

SAN CARLOS, CA:
+1 650-595-5333; Fax: +1 650-595-5589

CHICAGO:
+1 312-755-0798; Fax: +1 650-595-5589

WASHINGTON, DC:
+1 202-462-9582; Fax: +1 202-667-2922

UNITED KINGDOM:
+44 (0)1865-512184; Fax: +1 650-595-5589

Editor: C. Simone Fishburn, Ph.D.

Associate Editor: Michael J. Haas

Senior Writer: Lauren Martz

Staff Writers: Selina Koch, Ph.D.; Mary Romeo; Karen Tkach, Ph.D.; Mark Zipkin

Director of Research: Walter Yang

Copy Editor: Claire Quang

CORPORATE, SUBSCRIPTIONS & PRIVACY

BioCentury's mission is to provide value-added business information & analysis for life science companies, investors, academia and government on the strategic issues essential to the formation, development and sustainability of life science ventures.

BioCentury Publications, Inc.
BioCentury International Inc.

MAIN OFFICES
PO Box 1246
San Carlos CA 94070-1246
+1 650-595-5333; Fax: +1 650-595-5589