Benztropine for MS

By Lauren Martz, Staff Writer

Although the secondary progressive multiple sclerosis space is filling up with new compounds, few have shown the ability to remyelinate neurons. A new study suggests that an ancient Parkinson's disease drug—benztropine—could accomplish that task in secondary progressive multiple sclerosis, although it might require reformulation to improve activity and specificity.¹

There are multiple immunomodulatory therapies on the market that stop or slow the autoimmune attack on neurons by myelin-specific T cells in relapsing-remitting MS (RRMS). Nevertheless, within 20 years of diagnosis, the vast majority of patients will progress to secondary progressive MS (SPMS). At that point, available immunomodulatory agents are no longer able to prevent further neuronal degeneration.

The only drug on the market for SPMS is mitoxantrone, a generic chemotherapeutic that carries a boxed warning about potential cardiotoxicity.

The SPMS pipeline includes seven compounds in clinical development. A few compounds, including the antibodies BIIB033 and HlgM22, potentially can remyelinate axons and thus reverse the course of the disease.

BIIB033, from **Biogen Idec Inc.**, targets leucine-rich repeat neuronal protein 1 (LINGO-1) and is in Phase II testing. HlgM22, a remyelinating antibody from **Acorda Therapeutics Inc.**, is in Phase I testing.

Now, a group of researchers from **The Scripps Research Institute** and the **California Institute for Biomedical Research** (Calibr) have developed a screen for small molecules that can accomplish remyelination.

Specifically, the screen identifies compounds that promote the differentiation of oligodendrocyte precursor cells (OPCs) into myelin-promoting mature oligodendrocytes. In MS, the differentiation process is impaired.^{2,3}

The team was led by Peter Schultz, a professor of chemistry at Scripps and founding director of Calibr; Luke Lairson, an assistant professor at Scripps and a principal investigator at Calibr; and Brian Lawson, an assistant professor of immunology at Scripps. The team also included researchers from the **Salk Institute for Biological Sciences** and **Hokkaido University**.

The group screened about 100,000 small molecules in rat OPCs using an imaging assay that detects expression of myelin basic protein

(MBP). MBP expression indicates that the OPCs have differentiated into mature oligodendrocytes.

From the screen's hits, the team selected benztropine for additional studies because it is approved for PD, is orally bioavailable and crosses the blood brain barrier.

In coculture of mouse or rat OPCs with mouse neurons, benztropine induced OPC differentiation and axon remyelination by increasing the number of myelinating oligodendrocytes.

Benzotropine functions through multiple mechanisms including acting as an anticholinergic and antihistamine and inhibiting dopamine reuptake. The group blocked each mechanism to determine which one was responsible for OPC differentiation.

They found that a muscarinic acetylcholine receptor agonist, but not nicotinic acetylcholine, histamine or dopamine agonists, blocked the OPC-differentiating effect of benztropine.

Muscarinic receptor antagonists induced OPC differentiation in cultures, thus suggesting that the muscarinic antagonist effect of benztropine, specifically M1 and M3 antagonism, is responsible for OPC differentiation.

In a mouse model of experimental autoimmune encephalomyelitis (EAE), intraperitoneal injection of 10 mg/kg a day of benztropine decreased clinical severity compared with vehicle control injection and prevented the disease relapse phase. Benztropine also decreased disease severity comparably to or better than the marketed

MS immunosuppressants Avonex IFN- β -1a and Gilenya fingolimod (FTY720).

Biogen Idec markets Avonex IFN- β -1a and **Bayer AG** markets Betaseron IFN- β -1b to treat RRMS. **Novartis AG** and **Mitsubishi Tanabe Pharma Corp.** market Gilenya, a sphingosine 1-phosphate receptor antagonist, to treat RRMS.

In spinal cords from EAE mice that received prophylactic benztropine, immune cell infiltration and demyelination were the same as those in vehicle-treated mice. However, the number of mature oligodendrocytes increased in the mice given benztropine. These findings suggest that benztropine induced remyelination without altering the immune response.

In a mouse model of cuprizone-induced, T cell-independent demyelination, 10 mg/kg per day of benztropine accelerated remyelination following cuprizone withdrawal, whereas vehicle had no effect.

Finally, the team tested the effects of benztropine in combination with immunosuppressive MS therapeutics. In EAE mice, 2.5 mg/kg of benztropine plus 1 mg/kg of Gilenya was better than Gilenya alone in multiple measures of disease and comparable to Gilenya monotherapy in others.

Also, 2.5 mg/kg of benztropine plus a 0.1 mg/kg dose of Gilenya produced decreases in disease severity that were comparable to 1 mg/kg of Gilenya alone.

The results were published in *Nature*.

"This approach represents a much-needed addition to MS therapeutics. We need immune modulators, neuroprotectants and remyelinating agents to all work together."

—Spyros Deftereos, Biovista Inc.

Complementary remyelination

Lairson told *SciBX* that his team is working on pharmacokinetic tissue-distribution studies. "Upon completion of these studies we plan to meet with a clinical consultant to determine our best course," said Lairson.

He said that could include studies of benztropine in MS either alone or in combination with immunosuppressive therapy.

"Benztropine seems to have little or no impact on the immune system but rather seems to drive differentiation of the precursors of myelin cells to actual myelinating cells, stimulating myelin production," said Mike Gresser, CSO of the **Myelin Repair Foundation**.

"This approach represents a much-needed addition to MS therapeutics," added Spyros Deftereos, SVP of drug discovery at **Biovista Inc.** "It is important to treat the neurological deficits when patients relapse, and in addition to dampening the neuron-damaging immune response in these patients, we need a way of restoring the myelin to repair the damage. We need immune modulators, neuroprotectants and remyelinating agents to all work together."

Biovista's BVA101 and BVA201 are in preclinical testing to treat MS. The mechanisms are undisclosed.

"The goal is not just to stabilize patients by blocking further damage but also to positively improve their disability," said Don Healey, CSO of **Opexa Therapeutics Inc.**

Opexa's Tcelna, a patient-specific T cell immunotherapy that decreases myelin-reactive T cells, is in Phase IIb testing to treat SPMS.

Andrew Blight, CSO of Acorda, said that it may be worthwhile to try combinations of remyelinating agents.

In addition to developing HIGM22, Acorda markets Ampyra dalfampridine, a sustained-release formulation of 4-aminopyridine (4-AP), to improve walking and other neurological conditions in patients with MS.

Beyond benztropine

There are two main roadblocks for benztropine—one is molecule specific, and the other is indication specific.

For the disease in general, results in EAE models do not consistently predict results in patients with MS.

"One reason that EAE is not predictive of MS is that animal models are conducted over a short time frame of typically a month or two. By comparison, MS is truly a chronic disease taking many years to evolve to a progressive stage that exhibits changes from acute attacks that translate to relapses through to chronic progression, which may be driven by different disease processes. Relapses can be characterized by cellular proinflammatory events, whereas progression bears the hallmarks of chronic inflammation, possibly involving innate immune mechanisms," said Healey.

He added that the big problem in the field is coming up with a model for SPMS, which goes beyond the typical proinflammatory events that characterize chronic relapsing EAE. No models to date truly capture the immunopathology of progressive MS, he said.

Deftereos told *SciBX* that "the mouse data show that benztropine is more potent than existing drugs such as FTY720 and IFN- β , which is good, but it also shows that the existing drugs are equally effective in mice. In humans with MS, FTY720 is much more potent than IFN- β , so it isn't yet clear how reliable the data are or how effective

benztropine may be in humans."

Despite the problems with the EAE model, Lawrence Steinman, a professor of neurology and of neurological sciences and pediatrics at **Stanford University**, added that effective and approved drugs for MS, including Tysabri natalizumab and Gilenya, have all come out of the EAE model. Natalizumab is marketed by **Elan Corp. plc** to treat MS.

"EAE, when used creatively, can give some potential drug candidates that will make a difference in MS treatment," he added.

Lairson countered that the team also showed strong results in the cuprizone model, although he acknowledged that "it hasn't really been evaluated whether results in this model will translate. There are not any effective remyelinating agents in humans at this point to use as indicators."

Healey said that the downside of the cuprizone model is that although it enables a state of demyelination to be generated, which is reversible and leads to spontaneous remyelination that was accelerated by benztropine, it is achieved in the absence of an opposing chronic autoimmune response that typifies progressive MS.

Regardless of the model, Deftereos said that the data shown suggest that benztropine is effective at much higher doses than those approved for human use and causes neurological symptoms like confusion at high doses.

Healey added that the dose-limiting neurological adverse events represent a significant challenge, but the possibility of defining a new chemical entity targeting the associated signaling pathway in oligodendrocytes with lower toxicity would be an enviable goal.

Blight added, "The dosing used in preclinical models was high relative to the maximum tolerable dose in humans, and the drug was delivered by intraperitoneal injection rather than as an oral medicine."

"It will be important to investigate whether the concentrations in the brains of patients taking approved doses of benztropine are parallel to the efficacious tissue drug levels achieved in the brains of EAE models and *in vitro* assays," said Gresser.

Lairson said that medicinal chemistry could improve the therapeutic index of benztropine.

Gresser said that a path forward could involve "identifying the molecular targets of the promyelinating effects of benztropine and then finding a compound that has the desired effect on this target at efficacious doses free of the adverse effects of benztropine."

Lairson said that in addition to benztropine, his team has identified six other clinical candidates using the screen that could become remyelinating therapeutics.

Scripps has filed a patent application covering the use of a broad panel of neurotransmitter receptor-modulating agents in MS, including benztropine. The IP is available for licensing.

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COMPANIES AND INSTITUTIONS MENTIONED

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