MOLECULAR BIOLOGY

Sowing the seeds of Alzheimer's

A better understanding of how the condition spreads in the brain could help uncover ways to stop it in its tracks.

BY MOHEB COSTANDI

Researchers who have been scrambling to develop effective treatments for Alzheimer's disease have so far come away empty handed. Most have focused their attention and resources on a peptide called amyloid-β, which aggregates in brain tissue to form plaques — an established hallmark of the condition (see page S4). As success continues to elude them, a growing proportion of the research community is exploring other aspects of the condition in search of alternative molecular targets. And in trying to understand how Alzheimer's disease spreads through the brain, they might have stumbled on a way of stopping it in its tracks.

As well as the accumulation of amyloid-β around neurons, Alzheimer's disease is characterized by the aggregation of microtubule-associated protein tau into neurofibrillary tangles inside the cells. Pharmaceutical companies have spent vast amounts of money on developing drugs that target amyloid-β in various ways, but their efforts have yielded little of therapeutic value: although many such approaches worked well in mice, none has succeeded in clinical trials.

Plaques and tangles begin to form in the brain many years before any cognitive symptoms appear (S2). They emerge in the entorhinal cortex, a densely connected region of the medial temporal lobe that is crucial for memory and spatial navigation. From there, the insoluble clumps spread through the brain in a predictable way, moving into neighbouring regions of the temporal and frontal lobes, where they affect areas involved in speech and executive functions such as planning, before spreading throughout the cerebral cortex.

This pattern of progression, in which hubs of neurons in the brain become hotspots of neurodegeneration, is also seen in other neurological conditions, such as Parkinson's disease and Huntington's disease. Researchers have suggested two explanations for this phenomenon. Some propose that the high levels of neural activity and related energy demands of the hubs make them vulnerable to degeneration, whereas others think that the hubs are compromised by a deficiency in growth factors that promote cell survival.

Now, a third idea is gaining traction. Cellular hallmarks of Alzheimer's disease — especially those formed by tau — might propagate from neuron to neuron, and therefore disperse easily through well-connected neural circuits. This concept is known as transneuronal spreading.

"The experimental data on the spread of tau is compelling," says John Hardy, a neurogeneticist at University College London. "It does seem plausible that the characteristic distribution of the pathology is related, in part at least, to transmission along neuronal networks." The discovery could indicate a point of therapeutic

Aggregates of the protein tau (green) can travel across synapses to seed tau aggregation in neighbouring neurons (bottom).

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intervention. Already, several pharmaceutical companies are testing antibodies that target tau in people with Alzheimer's disease, and hopes are high that this change in approach might lead to the treatment breakthrough that researchers have been seeking for so long.

SPREADING EVIDENCE

The first hints of transneuronal spreading were uncovered in 2009, when researchers showed that aggregates of tau are readily taken up by neurons growing in culture. Inside these cells, the clumps then act as seeds, inducing other tau molecules to misfold and form fresh tangles[1].

Subsequent studies in cells and animal models added weight to the idea. Neurons in culture were shown to take up tau aggregates by a common cellular process called endocytosis[2]. And when injected into the brains of healthy mice, tau aggregates could spread across synapses into connected cells — and then on to neighbouring regions[3].

Further evidence comes from work in strains of genetically engineered mice that were created independently by two international teams of researchers. These animals express a disease-causing form of human tau exclusively in their entorhinal cortices, enabling researchers to closely examine how tangles spread from that part of the brain. Both teams found that, over the course of up to two years, tau aggregates spread first to neighbouring neurons, and then to 'downstream' regions of the brain that receive input from the entorhinal cortex[4,5].

Given that tau is thought to be toxic to synapses, the connections between neurons through which it spreads, its propagation could help to explain the widespread loss of these connections over time in Alzheimer's disease, which correlates closely with the cognitive decline that is observed in people with the condition.

In February this year, Thomas Cope, a clinical neurologist at the University of Cambridge, and his colleagues published evidence to support the transneuronal spread of tau in the brains of people with Alzheimer's disease[6].

According to the hypothesis of transneuronal spreading, when tau spreads from the entorhinal cortex, it should move first into strongly connected regions. Other hypotheses predict that tau traverses the brain according to a different pattern. Using a combination of positron emission tomography imaging and magnetic resonance imaging, the team was able to measure the density and distribution of tangles in the brains of 17 people with Alzheimer's disease, as well as the relative strength of the connections between regions of their brains.

"We demonstrated that tau was arranged based upon the number and strength of connections that a brain region possessed," says Cope. "That's what you'd expect if tau trafficked transneuronally."

TREATMENT GOALS

The spread of tau seems to be closely related to cognitive decline before the onset of Alzheimer's disease, and tangles are likely to be better indicators of the progression and severity of the condition than are plaques. Blocking tau's spread could therefore stop the condition progressing — or at least hinder its progress. "There's good evidence that tau in some form is toxic," says Cope, "so locking it up in one part of the brain and preventing it from transiting to other parts would likely reduce its toxicity."

To this end, several clinical trials that aim to determine whether antibodies that bind to tau might benefit people with Alzheimer's disease are already under way. "The idea is that they'll capture tau and reduce its spread," says Thomas Jahn, a biochemist at the University of Heidelberg in Germany, and head of neuroscience discovery at AbbVie, a biopharmaceutical company in Ludwigshafen, Germany, that has developed and is testing one such antibody.

Known as ABBV-8E12, and developed in collaboration with C_2N Diagnostics of St Louis, Missouri, the antibody binds to tau aggregates outside cells. Researchers have shown that it blocks the aggregation of tau in neurons in culture[7,8] and prevents tangle formation in mice that express mutant human tau — alleviating cognitive symptoms[8,9].

A successful phase I trial of the drug was performed in 30 people with progressive supranuclear palsy (PSP) — a rare brain disorder in which tau mutations cause tangles to form mainly in the brain stem. Ongoing phase II trials that began in 2016 are set to involve 400 participants with Alzheimer's disease, who are being recruited from North America and seven countries in Europe, as well as Australia and New Zealand. The study is expected to run until 2021.

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Several other companies have launched similar trials. Biogen of Cambridge, Massachusetts, has two tau-targeting antibody candidate drugs, one of which has been licensed from Bristol-Myers Squibb in New York, and is in phase II trials in people with Alzheimer's disease and PSP. Genentech in South San Francisco, California, is testing the safety of a tau-binding antibody in healthy people and people with Alzheimer's disease.

"It's clear that there's spreading of tau, and there's little question that this spreading plays a role in disease," says Jahn. He adds, however, that because the consequences of the spread of tau are still unknown, it remains to be seen whether blocking the process will actually benefit those with Alzheimer's disease.

The results from the trials should help to answer this question. Meanwhile, a team of researchers in the United States has identified a small molecule that seems to inhibit tau's ability to move from cell to cell.

Tina Bilousova, a biochemist at the University of California, Los Angeles, and her colleagues identified cambinol, as well as several other candidates, from a screen of more than 200,000 compounds. The molecule inhibits an enzyme called nSMase2, which plays an important part in the release of vesicles in which tau crosses the synapse. In May, Bilousova's team showed that this inhibition blocks the transfer of tau aggregates between neurons in culture, and that cambinol reduces the enzyme's activity in the brains of live mice[10].

"We are now trying to determine if cambinol blocks the spread of tau in animal models and alleviates cognitive symptoms in live animals," says Varghese John, a medicinal chemist and director of the Drug Discovery Lab at the University of California, Los Angeles. "But compounds related to or derived from cambinol may work even better, so we are also testing these other candidates."

John says that other recent advances, including the X-ray crystal structure of nSMase2, will hasten their efforts. "We now have a very clear understanding of the molecular mechanisms by which cambinol and nSMase2 interact," he says, "and that should help us to identify more molecules that block the spread of tau."

This strategy could prove to be challenging, however. "We know there are multiple ways that tau can transfer from cell to cell," says Selina Wray, a molecular neuroscientist at University College London, "so although a small molecule could reduce tau propagation, it will only be blocking one of many mechanisms."

There is also the risk of side effects. "All of these mechanisms probably have physiological functions, so shutting down a whole pathway might have a big impact on the cell," she adds. "The advantage of an antibody is that it specifically targets tau."

Just as drugs targeting amyloid-β have failed to bear therapeutic fruit, Wray and others think that targeting tau alone will not be enough to treat Alzheimer's disease. "Even if we stop tau from spreading, there are other things happening in the brain, such as inflammation and microglial-cell dysfunction," she says. But as part of a strategy to hit several aspects of Alzheimer's disease, drugs that target tau could have an important role in the first effective treatments. "We need to tackle more of these things to see more benefit." ■

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1. Frost, B., Jacks, R. L. & Diamond, M. I. *J. Biol. Chem.* **284**, 12845–12852 (2009).
2. Wu, J. W. *et al. J. Biol. Chem.* **288**, 1856–1870 (2013).
3. Clavaguera, F. *et al. Nature Cell Biol.* **11**, 909–913 (2009).
4. Liu, L. *et al. PLoS ONE* **7**, e31302 (2012).
5. de Calignon, A. *et al. Neuron* **73**, 685–697 (2012).
6. Cope, T. E. *et al. Brain* **141**, 550–567 (2018).
7. Kfoury, N., Holmes, B. B., Jiang, H., Holtzman, D. M. & Diamond, M. I. *J. Biol. Chem.* **287**, 19440–19451 (2012).
8. Yanamandra, K. *et al. Neuron* **80**, 402–414 (2013).
9. Yanamandra, K. *et al. Ann. Clin. Trans. Neurol.* **2**, 278–288 (2015).
10. Bilousova, T. *et al. Biochem. Biophys. Res. Comm.* **499**, 751–757 (2018).