NEWS

Normalizing protein level preempts Angelman symptoms

BY **JESSA NETTING**

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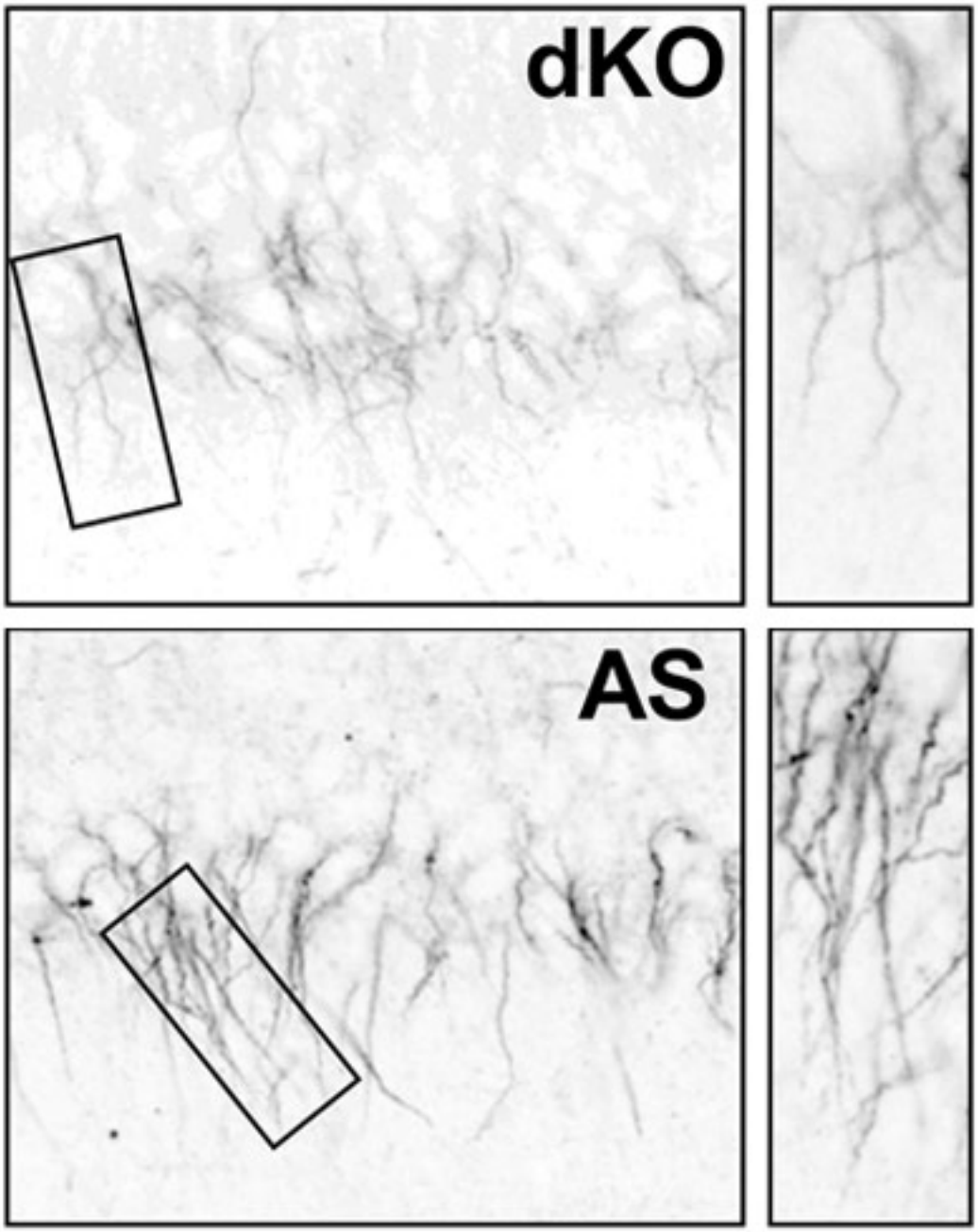
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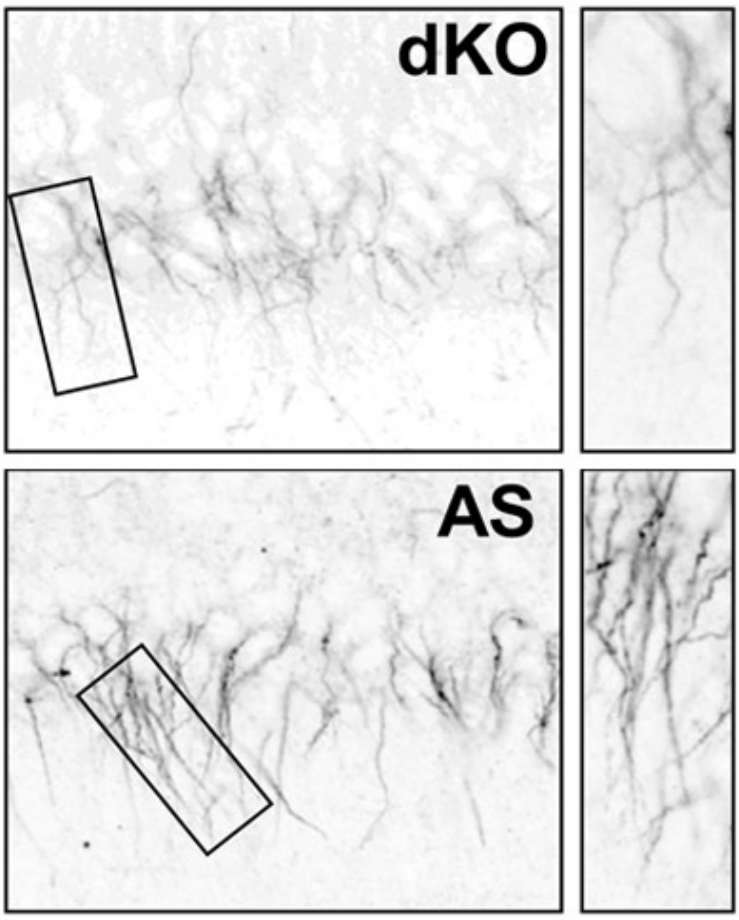

Over growth: Lowering the levels of a key protein normalizes the length and thickness of nerve fibers (top) in the hippocampus of mice that model Angelman syndrome (bottom).

The origins of **Angelman syndrome** seem simple — the loss of the maternal copy of a single gene, **UBE3A**[1] — but the cascade of disruptions that follow are anything but. Research groups have tried to reconstruct the complex pathway by pinpointing the effects[2] or by unmasking UBE3A's many targets[3].

One team has now shown that depleting excess levels of a molecule involved in neuronal firing prevents many of the symptoms in a mouse model of the syndrome[4]. They published their results 15 August in *Cell Reports*.

By ferreting out the molecule that drives physical changes in neurons, the researchers were able to prevent both the physical changes and the behavioral symptoms, says lead investigator **Eric**

Klann, professor of neuroscience at New York University.

Identifying UBE3A's many targets is among the biggest challenges in understanding Angelman syndrome. The key target in this case is a protein with the unwieldy name of alpha-1 subunit of the sodium-potassium ATPase (alpha-1-NaKA). The protein helps regulate the flow of sodium and potassium in and out of a neuron as it prepares to fire. A shift in this ion balance was what first tipped the researchers off about where to look.

In mice that model Angelman syndrome, the protein builds up in the brain and seems to set off changes, such as making parts of neurons too long. This may lead to other problems, such as difficulties in learning and memory.

Breeding Angelman mice that make just enough of alpha-1-NaKA corrects many of the mice's symptoms, such as trouble learning to navigate a water maze, the researchers found.

This is the first study to directly implicate alpha-1-NaKA in Angelman syndrome, but other studies have shown that restoring normal levels of other UBE3A targets also alleviates symptoms of the disorder. The new work suggests a therapeutic option for Angelman syndrome in people, adding to the tantalizing possibility this year of a **cancer drug** to treat the disorder.

"I think the study is very well executed, but there is more to understand about how changes in UBE3A levels can regulate alpha-1-NaKA," says **Lawrence Reiter**, associate professor of neurology, anatomy and neurobiology at the University of Tennessee in Memphis. Reiter was not involved in this study, but searches for proteins related to UBE3A in fruit flies. "All good research promotes more questions."

Protein tracking:

Angelman syndrome is known to severely impair language and cognitive development, produce seizures and frequent laughter, and often cause autism. It begins with a DNA-copying snafu that deletes the mother's copy of UBE3A, the gene that makes ubiquitin ligase.

This enzyme regulates **communication between neurons** and normally tags other proteins for destruction when they build up or outlast their usefulness. With this triage protein out of commission, proteins such as alpha-1-NaKA can accumulate unchecked, with serious consequences.

Last year, the researchers showed that mice that model Angelman syndrome have increased levels of several proteins, including alpha-1-NaKA[5]. This study found that, of these, only alpha-1-NaKA increases before changes appear in the neurons of Angelman mice.

The mice have defects in neurons of the hippocampus involved in learning and memory, called

pyramidal cells, and alterations of the neurons' membranes that dampen their tendency to fire. They also show longer initial segments of axons — where the arms of the neuron branch from its body and electrical signals originate — compared with controls.

The researchers noticed that the expression of a number of proteins shoots up where these axonal segments are longer. But early on, at 2 and 4 weeks of age, only alpha-1-NaKA is expressed at higher levels than in controls.

"So our idea was that maybe this [protein] was what was driving this change in the axon initial segment, the expression of these other proteins and the changes in the membrane properties of the animals," says Klann.

Cross-breeding mice that have one faulty copy of UBE3A with mice that have one faulty copy of alpha-1-NaKA confirmed the researchers' hypothesis. One-quarter of the mice that result from this cross-breeding have a disabled UBE3A gene (so they can't clear excess alpha-1-NaKA) but also make half the normal amount of alpha-1-NaKA, so it never builds up in the first place.

These mice also have short axonal initial segments, and neural membranes that perform normally. They have none of the learning and memory deficits seen in the Angelman mice. For example, the mice learn how to move through a water maze and retain memory of how to escape it, whereas Angelman mice must figure out the route each time anew.

"To find that [reducing the protein] not only corrects that change but also corrects behavioral changes is very exciting," says Klann.

The paper offers proof-of-concept that compensating for the missing UBE3A gene can prevent the symptoms from developing in the first place, but that won't be possible in people, notes **Seth Margolis**, assistant professor of biological chemistry at? Johns Hopkins University in Baltimore, who was not involved in the new study.

The next step is to identify drugs that can lower the levels of this or another UBE3A target, and perhaps correct some of the symptoms in people already diagnosed with the disorder, Margolis says.

"It is likely that there are many targets," says Margolis. He is working with various pharmaceutical companies to find drugs to block UBE3A's direct targets or others farther down the pathway.

"It is not a simple idea," he says, "but I don't think that there are many simple ideas when comes to cognitive disorders."

References:

1. Kishino T. *et al. Nat. Genet*. **15**, 70-73 (1997) **PubMed**

2. Jensen L. *et al. PLoS One* **8**, e61952 (2013) **PubMed**

3. Riday T.T. *et al. J. Clin. Invest.* **122**, 4544-4554(2012) **PubMed**

4. Kaphzan *et al. Cell Reports* **4**, 405-412 (2013) **PubMed**

5. Kaphzan *et al. J. Neurosci.* **31**, 17637-17648 (2011) **PubMed**