Mystery gene reveals new mechanism for anxiety disorders

May 15 2012

A novel mechanism for anxiety behaviors, including a previously unrecognized inhibitory brain signal, may inspire new strategies for treating psychiatric disorders, University of Chicago researchers report.

By testing the controversial role of a gene called Glo1 in anxiety, scientists uncovered a new inhibitory factor in the brain: the metabolic by-product methylglyoxal. The system offers a tantalizing new target for drugs designed to treat conditions such as anxiety disorder, epilepsy, and sleep disorders.

The study, published in the Journal of Clinical Investigation, found that animals with multiple copies of the Glo1 gene were more likely to exhibit anxiety-like behavior in laboratory tests. Further experiments showed that Glo1 increased anxiety-like behavior by lowering levels of methylglyoxal (MG). Conversely, inhibiting Glo1 or raising MG levels reduced anxiety behaviors.

"Animals transgenic for Glo1 had different levels of anxiety-like behavior, and more copies made them more anxious," said Abraham Palmer, PhD, assistant professor of human genetics at the University of Chicago Medicine and senior author of the study. "We showed that Glo1 was causally related to anxiety-like behavior, rather than merely correlated."

In 2005, a comparison of different mouse strains found a link between anxiety-like behaviors and Glo1, the gene encoding the metabolic

enzyme glyoxylase 1. However, subsequent studies questioned the link, and the lack of an obvious connection between glyoxylase 1 and brain function or behavior made some scientists skeptical.

"When people discover a gene, they're always most comfortable when they discover something they already knew," Palmer said. "The alarming thing here was there was a discovery of something that nobody knew, and therefore it seemed less likely to actually be correct."

A 2009 study from Palmer's laboratory suggested that differences in Glo1 expression between mouse strains were due to copy number variants, where the segment of the genome containing the gene is repeated multiple times. To test this hypothesis, lead author Margaret Distler inserted two, eight or ten copies of the Glo1 gene into mouse lines. She then ran experiments such as the open field test, in which researchers measure how much time a mouse spends in the center of an arena versus along the walls, to detect changes in anxiety behavior.

The results confirmed a causative role for Glo1 copy number variants, as mice with more copies of the Glo1 gene exhibited higher anxiety-like behavior in their experiments.

"It's the first study to show that it's the copy number variant that has the potential to change Glo1 expression and behavior," said Distler, an MD/PhD student in the Pritzker School of Medicine's Medical Scientist Training Program. "Our study was a physiological representation of what it means to increase Glo1 expression for anxiety."

The researchers then set about answering the mystery of how Glo1 expression influences anxiety-like behaviors. The primary function of glyoxylase 1 is to metabolize and lower cellular levels of methylglyoxal, a waste product of glycolysis. Distler produced the opposite effect by injecting MG to artificially increase its levels in the brain, finding that

raising MG levels quickly reduced anxiety symptoms in mice.

"Methylglyoxal changed behavior within 10 minutes of administration, which means it's a rapid onset. It's not changing gene expression, and it's not having long-term downstream effects," Distler said. "That was our first breakthrough."

The short time course suggested that MG might have a direct effect on neuronal activity. MG also demonstrated sedative effects at high doses, a hallmark of drugs that activate inhibitory GABA receptors on neurons. In collaboration with Leigh Plant, now at Brandeis University, the researchers demonstrated that MG activated GABA-A receptors on neurons, a previously unknown inhibitory mechanism.

"It's a completely different system that is tying neuronal inhibitory tone into metabolic activity," Palmer said. "That's potentially really exciting in terms of re-evaluating what we thought we knew about inhibitory tone in the CNS. It turns out now that methylglyoxal, which has been around ever since glycolysis evolved, was also acting at these receptors, and nobody knew that."

Conventionally, anxiety has been treated with drugs that activate the GABA-A receptor, such as benzodiazepines and barbiturates, which are prone to abuse and dangerous side effects. The researchers theorized that targeting the Glo1/MG interaction could provide a more selective strategy for reducing anxiety symptoms by subtly influencing inhibitory tone.

"The GABA-A receptor agents already out there have a lot of side effects, such as sedation and hypothermia, as well as a high abuse liability," Distler said. "It's possible that taking a Glo1 inhibitor will increase only MG levels to a certain maximum. You could have the potential for more specificity, given that you're activating a system that's

already in place, not just dumping methylglyoxal or some other GABA-A receptor agent throughout the brain."

Preliminary experiments with a small molecule inhibitor of Glo1 supported the theory. Injections of the inhibitor, developed by John Termini at the Beckman Research Institute of the City of Hope, reduced anxiety-like symptoms in mice.

"It's a different way of hitting these GABA-A receptors," Palmer said. "We have yet to determine if that's a better way of doing it, but it's certainly different, and it gives us a unique angle of attack on this system and potential advantages that we have yet to evaluate."

Such a drug may also be useful in treating epilepsy and sleep disorders, where GABA-A drugs have shown success. While the therapeutic potential of manipulating this system is yet to be determined, the research clears the fog around the role of Glo1 in anxiety by adding behavioral and cellular evidence.

"What's neat is that we started with exploratory, open-ended genetic studies in mice, and we've now gotten into some fundamental new physiology that nobody had appreciated or put together before," Palmer said. "Now we're starting to reap some of the fruit from those types of genetic studies to enrich our understanding of more classical aspects of biology."

More information: The paper, "Glyoxalase 1 increases anxiety by reducing GABAA receptor agonist methylglyoxal," will be published online May 15 by the *Journal of Clinical Investigation.*

Provided by University of Chicago Medical Center