

MAY 30, 2008

Rett Syndrome: One Mutation Affects a Cast of Thousands

Rett syndrome arises in early childhood and robs affected children of the ability to speak and control their movements. The disorder is caused by a mutation in a single gene, but scientists have not understood how that mutation disrupts neurological function. Now, Howard Hughes Medical Institute (HHMI) scientists have discovered that the mutation interferes with the regulation of 2,500 other genes.

The researchers said their discovery offers a promising pathway for understanding the broad and variable symptoms of Rett syndrome, which include language and growth retardation, breathing problems, seizures, motor dysfunction, hand-wringing and social impairment.

"The duplication syndrome and Rett syndrome may share many clinical symptoms, but on the level of the neuron they are totally different, and so the treatment would be totally different."

— **Huda Y. Zoghbi**

Huda Y. Zoghbi, an HHMI investigator at Baylor College of Medicine, Maria Chahrour, a graduate student in Zoghbi's lab, and their colleagues reported their findings May 30, 2008, in the journal *Science*.

Rett syndrome is a devastating disease, says Zoghbi. Think about what it takes to function normally: You have to be coordinated, you must be able to think, you have to be able to communicate, and you need to move smoothly and with balance. The symptoms of other neurological diseases affect some of these functions, she says, but Rett syndrome affects them all.

Zoghbi started her career as a pediatric neurologist, and became fascinated by Rett syndrome during her training. It's very hard not to be intrigued by this disease, she says. We all are familiar with neurological diseases; I'm sure everyone has seen some one with epilepsy, or Parkinson's disease, or bipolar disorder or schizophrenia. But when you see symptoms of all these diseases in one individual, you are struck by this. Rett syndrome is particularly heartbreaking, she says, because it develops after the child has already

learned to walk, and perhaps even say a few words. That's part of what drove her to study it. To lose all that, and gradually develop a symptom from almost every neurological disease in the book is quite mind boggling, she says.

In 1999, Zoghbi's group showed that a mutation in the *MECP2* gene causes Rett syndrome, which affects one in about 10,000 girls. The *MECP2* gene resides on the far end of the longest arm of the X chromosome. During development, certain genes that are to be silenced, or prevented from being expressed, are physically marked by the addition of methyl groups. *MECP2* encodes a protein named methyl-CpG-binding protein 2, or MeCP2, that binds to these methylated genes. Once bound, MeCP2 attracts other proteins that prevent access to the cell's DNA transcription machinery, thereby facilitating gene silencing.

Zoghbi and other researchers have worked to understand how a single genetic mutation can be responsible for Rett syndrome's many symptoms, but they have met with little success. The protein was known to orchestrate gene expression in neurons, she says, but the question that remained was What genes is this regulator talking to?

Recently researchers discovered a mutation that adds a duplicate copy of *MECP2* to the genome. This doubles the amount of the MeCP2 protein in cells rather than eliminating it, as in Rett syndrome — but otherwise causes similar symptoms. The human *MECP2* duplication syndrome was described so recently that no one knows how common it is, or why having too much of the protein is just as harmful as not having it at all.

As researchers search for the genes that are regulated by MeCP2, they typically compare gene activity patterns in the brains of mice that lack the protein to patterns in the brains of normal mice. Those kinds of studies, Zoghbi says, have identified changes in the activity in a handful of genes, or even nothing at all—not the sort of widespread disruption that one might expect to be responsible for the variety of symptoms associated with Rett syndrome.

Zoghbi thought researchers might be getting those results because they were casting their nets too wide. As a result, they were not seeing changes because they were lost against the vast backdrop of gene activity in the whole brain. So, she took an educated guess and narrowed her search to a small area of the brain called the hypothalamus. In humans, the hypothalamus is about the size of an almond. In mice, it's no larger than a split pea. It's responsible for many metabolic processes, says Zoghbi. When you get thirsty, there's a hormone produced in the hypothalamus that tells you to drink water. It controls how much stress hormones you have; when it's time to shiver because you're cold. It's a command center. With its broad purview, Zoghbi says, it's reasonable that misregulation within the hypothalamus could be responsible for many of the symptoms of Rett syndrome.

Zoghbi measured gene expression in the hypothalami of normal mice, as well as mice with Rett syndrome or the *MECP2* duplication syndrome to increase the likelihood of finding MeCP2 targets. She found that the disorders

changed the expression of about 2,500 genes. In the animals with Rett syndrome, 2,200 were less active than they were in normal mice, while the remainder showed increased activity. The numbers were precisely reversed in the duplication syndrome. These findings were surprising because they show that MeCP2 can activate genes.

From a practical viewpoint, knowing how the two syndromes behave on a molecular level is really important, said Zoghbi. The duplication syndrome and Rett syndrome may share many clinical symptoms, but on the level of the neuron they are totally different, and so the treatment would be totally different.

In addition, knowing just how many genes are affected is important in and of itself. If only a few genes were affected, she says, it might be possible to artificially control those proteins directly. As it is, we're going to have to find a way to co-opt another master regulator to fill in the gaps, she says.