NEWS

Fast-evolving gene is key player in brain development

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Knocked down: Zebrafish lacking AUTS2 (right), a gene linked to autism, have fewer neurons in the mid-brain region compared with controls (left).

A gene that changed rapidly after the human genome diverged from that of Neanderthals plays a critical role in brain development, according to unpublished results presented Thursday at the **International Congress of Human Genetics** in Montreal, Canada.

Neanderthals are the closest evolutionary relatives of present-day humans. In 2001, researchers first identified mutations in the gene, autism susceptibility candidate 2 or **AUTS2**, which is located on **chromosome 7**, in a pair of identical twins with autism[1].

Since then, AUTS2 has also been linked to **attention deficit hyperactivity disorder**, **epilepsy** and mental retardation.

A mouse study last year reported that AUTS2 is expressed at high levels in developing neurons of certain brain regions, notably the frontal cortex and cerebellum[2].

Last year, a study published in *Science* pinpointed the gene as containing a genomic sequence that differentiated humans from Neanderthals early in human history[3].

Still, the function of AUTS2 has remained elusive until now.

Researchers at the University of California, San Francisco presented the first functional study of the gene, which they identified while searching for genes important in development.

“We were looking for regions in the genome that have a lot of evolutionary conservation, which usually indicates an important developmental gene that needs tight regulation,” says lead investigator **Nadav Ahituv**, assistant professor of bioengineering and therapeutic sciences at the University of California, San Francisco.

Ahituv’s team began characterizing the function of AUTS2 before the Neanderthal genome study came out last year. The prominence of AUTS2 as a fast-evolving gene in that study added excitement to the team’s efforts to illuminate the gene’s function.

“We were on a fishing expedition and we caught a big fish,” says Ahituv.

Fish tale:

In the new study, the researchers used morpholinos — molecules that prevent translation of particular RNA sequences — to block expression of AUTS2 in **zebrafish**. Loss of AUTS2 leads to fish with smaller heads, smaller eyes, fewer neurons in the midbrain region and deficits in motor neurons.

Normally, zebrafish quickly swim away when prodded, but those lacking AUTS2 move sluggishly, if at all. Inserting the RNA for human AUTS2 into the mutants corrects these defects.

The researchers then went looking for enhancers — short regions of DNA that regulate gene expression — in the region of AUTS2 that differs between humans and Neanderthals.

They focused on the first half of the AUTS2 gene, which contains a stretch of nearly 300 single nucleotide polymorphisms (SNPs), variations of a single nucleotide, between the genome of Neanderthals and that of humans.

They also looked for enhancers in ‘human accelerated regions’ of the gene, which are conserved throughout vertebrate evolution, but have radically changed since humans and chimpanzees split from their common ancestor between 5 to 7 million years ago.

Finally, they honed in on enhancers in intron 4, a region of AUTS2 that does not code for protein, and which a previous study had found to be deleted in an individual with autism[4].

Using a zebrafish cell line, the researchers identified 21 nucleotide sequences that appear to regulate AUTS2 expression in the central nervous system, and 10 that enhance expression in the

brain.

Several of these sequences also regulate central nervous system structure and function in mice, the researchers found. The zebrafish and mouse studies together indicate that AUTS2 serves an important purpose in neurodevelopment, the researchers say.

The researchers plan to check whether deletions of the gene found in individuals with autism are in enhancer regions. “We could then test the enhancer with the mutations that an individual might have, and see if that affects enhancer activity,” says Nir Oksensberg, a graduate student in Ahituv’s lab who presented the results at the conference.

Although the enhancers tested thus far are all within the gene, it is possible that other enhancers lie outside its boundaries, says Ahituv.

These results are intriguing, says **Janine LaSalle**, professor of medical microbiology and immunology at the University of California, Davis, who is not involved with the study.

“I’m fascinated by the Neanderthal stuff,” LaSalle says. Sequences selected for late in evolution like the ones in AUTS2 would have been advantageous for our ancestors, she notes. “It’s got to have a reproductive or survival advantage in order to be selected.”

LaSalle’s work has shown that AUTS2 is highly methylated in the brain. Methylation involves the addition of a methyl group, which tweaks gene expression without altering the genetic sequence.

None of the SNPs in the new study are in protein-coding regions, La Salle notes. “It’s possible that more recently evolved changes to the genome are regulatory in nature.”

References:

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3: Green R.E. *et al. Science* **328**, 710-722 (2010) **Article**

4: Pinto D. *et al. Nature***466**, 368-372 (2010) **PubMed**