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Mouse Model of Schizophrenia Could Speed Identification of New Antipsychotic Drugs

Howard Hughes Medical Institute researchers have produced a genetically altered mouse that exhibits behavioral abnormalities that are strikingly similar to those observed in humans with schizophrenia.

The scientists report that they have already used insights from studying the mouse to identify a genetic variant associated with schizophrenia in humans.

According to the researchers, the findings could well mean that they have identified a molecular signaling pathway involved in the origin of schizophrenia, which affects about one percent of the population. If so, the search for drugs affecting that pathway could yield a new class of antipsychotic drugs that more precisely and effectively treat the disorder.

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- Susumu Tonegawa

The researchers, led by Howard Hughes Medical Institute investigator Susumu Tonegawa at the Massachusetts Institute of Technology, published their findings June 30, 2003, in two articles in the early edition of the *Proceedings of the National Academy of Sciences(PNAS)*. Tonegawa collaborated on the studies with researchers from Duke University Medical Center, The Rockefeller University and the Columbia University College of Physicians and Surgeons.

In their latest studies, Tonegawa and his colleagues built on their earlier research on a genetically engineered mouse, which had been specifically altered to knock out the gene for the enzyme calcineurin only in the animal's

forebrain. Calcineurin is an enzymatic switch that plays regulatory roles in both the immune system and the brain. Until a brain-specific knockout mouse was developed in Tonegawa's laboratory, it had not been possible to pinpoint the enzyme's function in the brain. In earlier studies, Tonegawa and his colleagues found that the mutant mouse showed severe, specific deficits in a type of short-term memory called working memory.

Although schizophrenics show similar memory deficits, said Tonegawa, there was previously “no evidence whatsoever” that the calcineurin pathway played a role in schizophrenia. That connection emerged when research scientist Tsuyoshi Miyakawa in Tonegawa's laboratory decided to explore the behavioral abnormalities observed in the knockout mice.

Those studies revealed hyperactivity in the mice that correlates with positive symptoms of schizophrenic patients. Miyakawa also noted that the mice exhibited decreased social behavior strikingly like that of human schizophrenics.

“It's very well known that schizophrenia patients keep to themselves,” said Tonegawa. “They don't interact socially. And in our studies, the mutant animals behaved similarly. For example, unlike normal animals that usually sleep together in their home cage, the knockout animals slept separately. They didn't want to be together.” The knockout animals also showed impaired nest-building and scattered nesting materials about their cage.

Most intriguing, said Tonegawa, was the behavior exhibited by the calcineurin-knockout mice in reaction to a test used to diagnose schizophrenia in humans. In the test, the mice were exposed to a startling tone preceded by a weaker tone. Normal mice and non-schizophrenic people show a reduced startle response when presented with the two tones, because the first tone prepares them for the second. However, the knockout mice - like schizophrenics with attention deficits—are significantly more startled by the second tone. The researchers also found that the mice responded to drugs affecting the NMDA receptor pathway in the same way that human schizophrenics do.

A leading theory about schizophrenia posits that it originates from a disorder of the brain-signaling pathways involving the neurotransmitter dopamine. However, analyses of the mice by co-author, former HHMI investigator Marc Caron and his colleagues at Duke revealed no alterations in the animals' dopamine pathway.

“This is the first knockout mouse to show such a comprehensive array of behavioral abnormalities that mirrors the abnormal behavior of human schizophrenia patients,” said Tonegawa.

“And with this mouse and our human studies, we have implicated an entire biochemical signaling pathway—the calcineurin pathway—that had not been

implicated in schizophrenia before. So, with this research, we have discovered a totally new target for antipsychotic drugs that is not directly related to dopamine receptors,” he said.

In the second *PNAS* article, research scientist David Gerber, also a former HHMI postdoctoral associate in Tonegawa's laboratory, and postdoctoral fellow >Diana Hall in Maria Karayiorgou's laboratory at Rockefeller University, performed a search of calcineurin-related genes to look for an association with schizophrenia.

“It is known that schizophrenia is a complex multigene disease, and that these genes are scattered in different regions, or loci, on the genome,” said Tonegawa. “And quite remarkably, many genes in the calcineurin pathway map to these loci.” Thus, he said, schizophrenia might arise from subtle mutations in numerous genes related to the calcineurin pathway that together contribute to the pathway's overall malfunction.

In an initial survey of these candidate genes in a large number of schizophrenia-affected families, Tonegawa, Karayiorgou and their colleagues discovered that a variant form of one calcineurin-related gene, called *PPP3CC* was transmitted at significantly higher frequency than expected to affected children. This gene codes for a key functional subunit of the complex calcineurin protein.

Such findings, said Tonegawa, strongly suggest that screening large libraries of candidate drugs to identify those that affect enzymes comprising the calcineurin pathway could yield an entirely new class of more effective antipsychotic drugs.