News Release

Title

Discovery of a novel gene that strongly associates with the onset of schizophrenia. A multi-perspective analyses by integrating CNV analysis, model mouse creation, and iPSCs.

Key Points

•Genome-wide copy number variation (CNV) analysis of Japanese patients with schizophrenia (SCZ) has identified the genetic association between the onset of SCZ and the "double hit" case of CNVs found in the *ARHGAP10* gene.

•We established the SCZ model mouse by creating the mouse that mimics the SCZ patient who carried the CNV and the simultaneous single nucleotide variant (SNV) on the other allele and found the phenotypic abnormalities (immature neurites, decreased spine density, increased anxious behavior).

•With the agreement of the patients, we established induced pluripotent stem cells(iPSCs) and differentiated into neuron. The neuron derived from the patient iPSCs showed the immature dendric spines which was also observed in the neurons in the model mouse.

•Our findings indicate that ARHGAP10 is an important regulatory gene in Rho signaling pathway, thus making it a promising drug discovery target for SCZ treatment.

Summary

Researchers at Nagoya University has found the strong association between a novel CNV of *ARHGAP10* gene and the pathogenesis of SCZ by conducting comprehensive studies including genome-wide CNV analysis, the model mouse studies, and iPSCs induced by the patients who carried the CNVs. *ARHGAP10*, Rho GTPase activating protein 10, is a GTPase-activating protein which inactivates a small G-protein such as RhoA and Cdc42. *ARHGAP10* is widely expressed throughout our body including brain and heart, and play an important role in the physiological regulation such as cytoskeletal actin regulation through Rho-kinase activity which works as an effector. However, the association between the pathogenesis of SCZ and the function of ARHGAP10 has not been investigated. The large-scale genome-wide study of Japanese SCZ patients revealed the statistical association between the *ARHGAP10* gene variant and the onset of SCZ. In the CNV analysis, we identified one case with CNV in one allele and single nucleotide variant (SNV) on the other allele, which had a severe clinical phenotype. We established the ARHGAP10 model mouse that mimics this particular case. The model mouse showed the phenotypic abnormalities: 1. Immature neurite length 2. Decreased spine density. 3. Increased anxious behavior, pertaining to the SCZ model mouse. Furthermore, with the agreement of the patients, we established the iPCs and differentiated into neuron. Those neurons also had the immature neurite length which was the same phenotypic abnormality found in the model mice.

In this study, integrated findings from the CNV analyses, model mice studies, and iPSCs analyses indicated the *ARHGAP10* is a novel gene that accounts for the pathogenesis of SCZ. Furthermore, we showed that the Rho-signaling pathway can be an important drug target in a treatment of SCZ, thus enabling

us to make a transition to drug development targeting this signaling pathway in cooperation with pharmaceutical companies using the bioresources established throughout this study.

Research Background

The contribution of genetic factors in the pathogenesis of SCZ has long been investigated, and several genetic variants have been identified. A variant is a difference in genotype found in the same biological species. Genetic differences that exist within a same species are called "variant". However, the relationship between this genetic variant and the disease onset are unclear, and the establishment of mice and cells that mimic SCZ genotype which enables the drug development have long been the subject of the study in the field.

Research Results

In this study, the genome-wide CNV analysis of approximately 3,000 Japanese patients with SCZ had identified 7 CNVs of ARHGAP10: 6 deletions, 1 duplication, and revealed the association between the onset of SCZ and statistically significant CNVs (Figure 1). Of these, we identified one case with “double hit”, the deletion of ARHGAP10 in one allele and a single nucleotide variant (SNV),a missense mutation (Ser490→Pro) in the other allele, on a RhoGAP domain. Further biochemical study revealed the ARHGAP10’s decreased binding ability with the substrate, the activated RhoA. Then, we established the model mouse and iPSCs that mimic the genotype of this patient and found the same phenotypic abnormalities in both iPSCs and model mouse (Figure 2), that is, decreased neurites length in brain development.

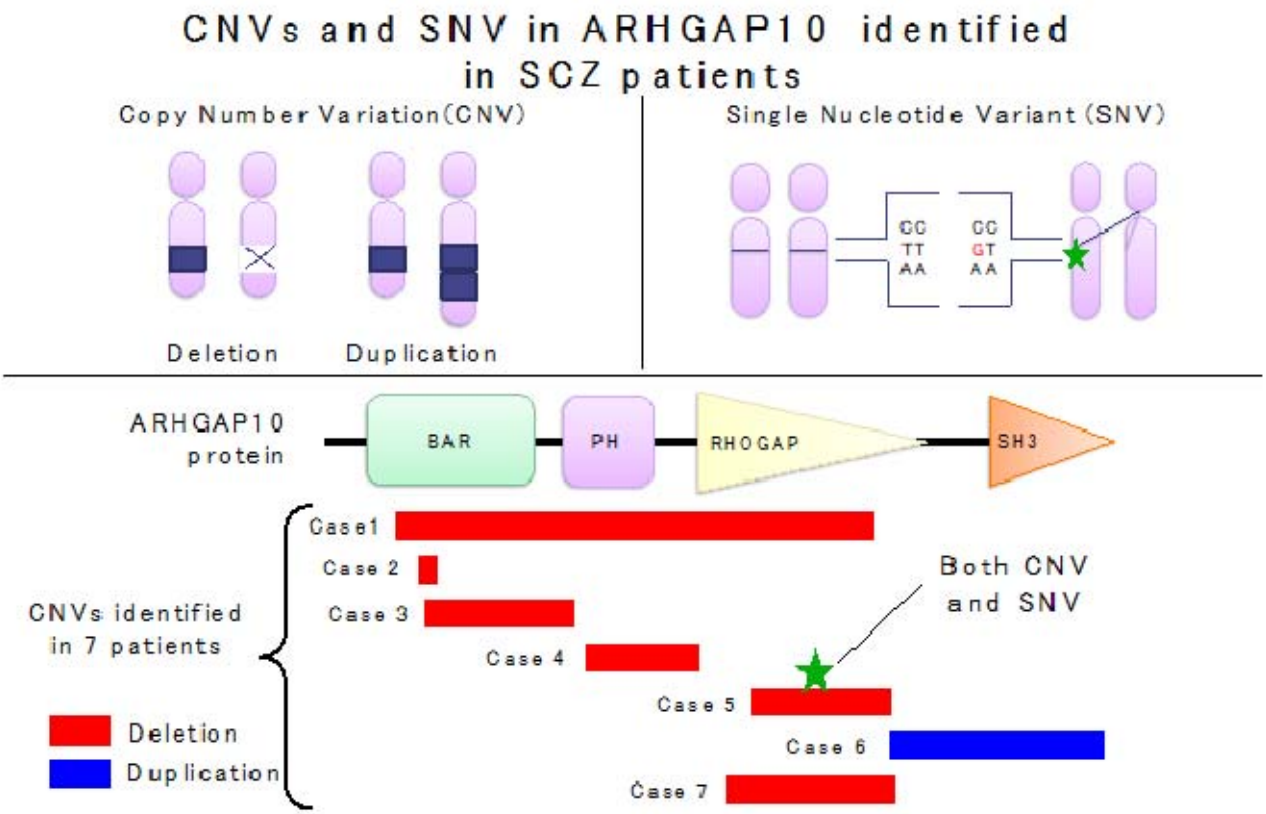

Figure 1: Gene map of 7 patients with ARHGAP10 CNV identified by genemic analysis of schizophrenia. One of the 7 patients (Case 5) had a deletion as well as a single nucleotide variant (SNV) that caused an amino acid substitution in the RhoGAP domain of the ARHGAP10 protein. This SNV had a very important functional meaning (the effect of strongly reducing the binding to activated RhoA).

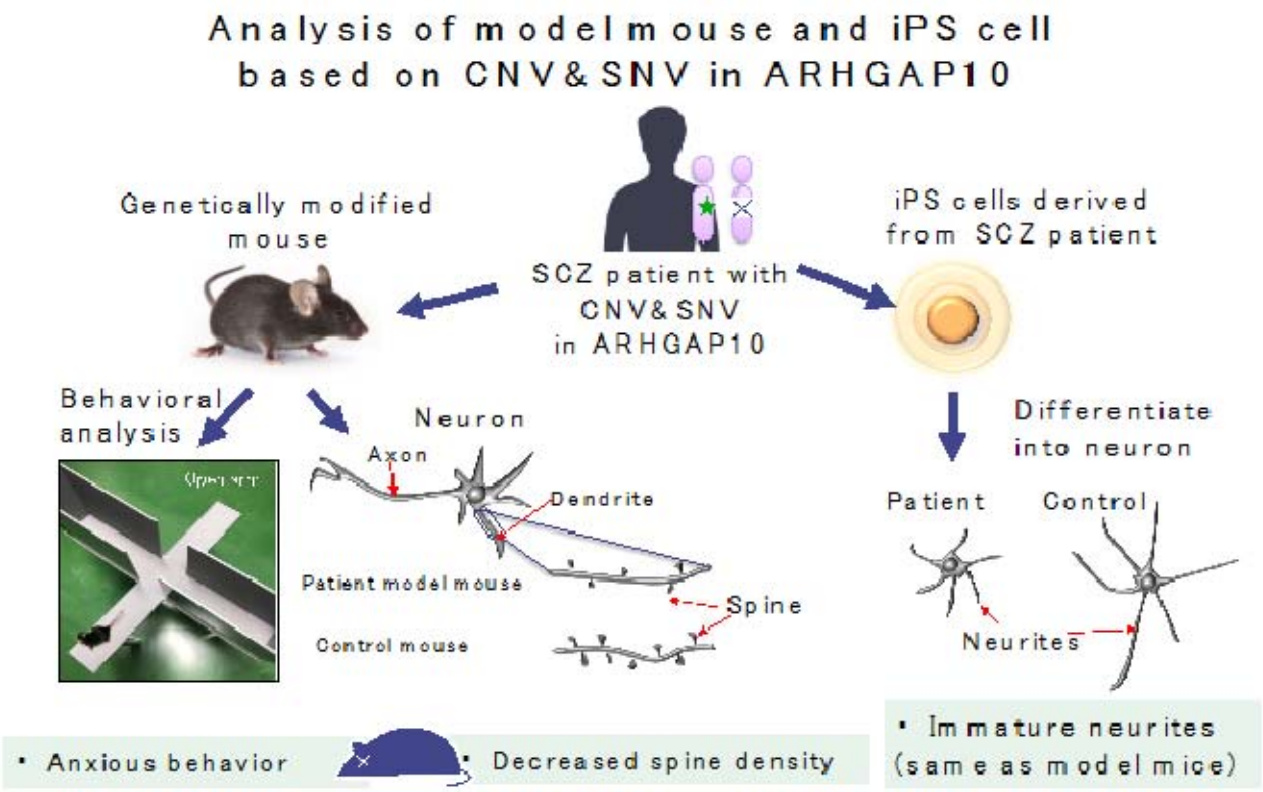

Figure 2: Generation of a novel model mouse carrying both single nucleotide variant: Ser490→Pro amino acid mutation and the CNV of ARHGAP10. Then phenotypic abnormalities of the mouse were investigated. As a result of the behavioral analysis of the model mouse, "promotion of anxiety-like behavior" was shown. In addition, when the brain tissue of a model mouse was collected and the neurons that compose the brain were observed under a microscope, the density of spines, which are dendritic spine-like structures, decreased. Furthermore, when iPS cells were established from patients with ARHGAP10 variant, differentiated into neurons and examined for neurite length, a phenotypic abnormality was confirmed in which neurites were shorter than iPS cells established from healthy subjects. The phenotypic abnormalities of shortening neurites was also observed in model mice. In future research, we have a plan to clarify the detailed molecular mechanisms of these phenotypic abnormalities and how they are associated with the pathophysiology of schizophrenia patients, leading to drug discovery and treatment.

Research Summary and Future Perspective

ARHGAP10 is an important regulatory gene in Rho signaling pathway. Future plan is to proceed to drug inventory in cooperation with pharmaceutical companies.

Publication

ARHGAP10, which encodes Rho GTPase-activating protein 10, is a novel gene for schizophrenia risk" by Daisuke Mori, Mariko Sekiguchi, Akira Sobue, Itaru Kushima, Wang Chenyao, Yuko Arioka, Hidekazu Kato, Akiko Kodama, Hisako Kubo, Norimichi Ito, Masahito Sawahata, Kazuhiro Hada, Ryosuke Ikeda, Mio Shinno, Chikara Mizukoshi, Keita Tsujimura, Akira Yoshimi, Kanako Ishizuka, Yuto Takasaki, Hiroki

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