News Release

Title

Single-cell trajectory analysis of human homogenous neurons carrying a rare *RELN* variant

Key Points

- Reelin is a protein encoded by the *RELN* gene that controls neuronal migration in the developing brain. Human genetic studies have suggested that a rare variant in *RELN* gene is one of factors contributing to neurodevelopmental impairments relevant for mental disorders. However, the biological significance of rare *RELN* variants in human neurons remains unknown.
- We generated iPSC cells derived from subjects carrying congenital a rare *RELN* variant (RELN-del) and isogenic RELN-del iPSC lines using targeted genome editing.
- Single-cell trajectory analysis revealed that RELN-del causes a loss of directional stability during neuronal migration.
- Our approach, focusing on individual human neuronal migration, will be useful for understanding the significance of *RELN* variant in human brain and evaluating mental disorder susceptibility.

Summary

Prof. Norio Ozaki at Department of Psychiatry, Nagoya University Graduate School of Medicine (Dean: Kenji Kadomatsu); Dr. Yuko Arioka (first author) at Center for Advanced Medicine and Clinical Research, Nagoya University Hospital; Dr. Daisuke Mori (corresponding author) at Brain and Mind Research Center, Nagoya University revealed that the rare *RELN* variant, initially identified in schizophrenic patient, causes a loss of directional stability during neuronal migration. This study was achieved thorough collaborative research work with Keio University.

For the functional nervous system, in brain embryonic neurons migrate from their place of origin to their designated place. Reelin, encoded by the *RELN* gene, play an important role in this neuronal migration. Human genetic studies of patients with mental disorders have identified several possible factors contributing to neurodevelopmental impairments, one of which is a rare variant in the *RELN* gene. Nevertheless, it remains unknown what effects rare *RELN* variants have on human neuronal cells. To address this, we generated iPSC cells derived from subjects carrying congenital a rare *RELN* variant (RELN-del) and isogenic RELN-del iPSC lines using targeted genome editing; iPS cells were further differentiated into highly homogenous dopaminergic neurons. Single-cell trajectory analysis using these neurons revealed

that control neurons possessed directional migration, while RELN-del neurons demonstrated a wandering type of migration. This study, focusing on the directionality of individual cell migration, provides a foundation for the elucidation of reelin function in neural development and may contribute to novel therapeutic strategies for mitigating mental disorder susceptibility.

Research Background

Brain organization during development requires the rigid regulation of neuronal migration. Reelin, encoded by the *RELN* gene, is a secretory protein and plays an important role in this process. Recent many genomic studies in the world including ours (Kushima et al., 2016) have shown that *RELN* gene variant is involved in the development of psychiatric disorders such as schizophrenia. However, it has been unsolved yet how the *RELN* gene variants identified in patients with mental disorders affects the movement of neurons in the human brain related to the onset of mental illness.

Research Results

In order to clarify the biological effect of variant identified in *RELN* gene on human nerve cells, we established the iPS cells from the subjects in whom this variant was identified. In addition, we also generated the artificial iPS cell lines mimicking the subject by genomic editing techniques for the experimental controls. Next, we differentiated highly homogeneous dopaminergic neurons from both these congenital and isogenic iPS cells and analyzed their common dynamic characters due to the *RELN* gene variant, comparing with the neurons from healthy controls. As a result, we found that the dopamine neurons with the *RELN* gene variant had decreased Reelin protein, disturbed its downstream signals, and impaired the gene expressions involved in cell movement. Furthermore, we performed the time-lapse analysis for the migration of each neuron on the dish, resulting that control neurons possessed directional migration, while RELN-del neurons demonstrated a wandering type of migration.

Research Summary and Future Perspective

This is the first demonstration of the biological significance of the *RELN* gene variant in human neurons. This concept got led to the elucidation of mechanisms related to the onset of schizophrenia involving in the disturbance of Reelin signals. Here, we are trying to apply this analysis system to develop the drugs for treatment of schizophrenia.

Publication

Single-cell trajectory analysis of human homogenous neurons carrying a rare *RELN* variant. *Translational Psychiatry*. in press (published online on July 19, 2018)

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