News Release

Title
Rare Genetic Variants in the Gene Encoding Histone Lysine Demethylase 4C (KDM4C) and Their Contributions to Susceptibility to Schizophrenia and Autism Spectrum Disorder

Key Points
- There was a significant association between CNVs in KDM4C and schizophrenia (SCZ) and autism spectrum disorder (ASD) respectively.
- There was also a significant association between deletions in KDM4C and SCZ.
- Immunoblotting analysis using lymphoblastoid cell lines (LCLs) from a case with KDM4C deletion revealed reduced KDM4C protein expression and altered histone methylation patterns of H3K9 and H3K36.
- These findings strengthen the evidence for associations between KDM4C CNVs and these two disorders and for their potential functional effect on histone methylation patterns.

Summary
Prof. Norio Ozaki, Dr. Itaru Kushima and Dr. Hidekazu Kato (Department of Psychiatry, Nagoya University Graduate School of Medicine) and collaborators performed analysis of copy number variation (CNV) in Japanese population. They revealed a significant association between CNVs in KDM4C and schizophrenia (SCZ) and autism spectrum disorder (ASD). Then, they confirmed a significant association between KDM4C deletion and SCZ. Furthermore, they performed immunoblotting analysis using lymphoblastoid cell lines (LCLs) from a SCZ case with KDM4C deletion and revealed reduced KDM4C protein expression and altered histone methylation patterns.

SCZ and ASD are considered clinically different psychiatric disorders. Nevertheless, recent studies indicate an overlap between the two disorders in genetic risk factors, biological pathways and phenotypic features. Dysregulation of epigenetic processes involving histone methylation induces neurodevelopmental impairments and has been implicated in both disorders. KDM4C is expressed from fetal to adult stages and regulates differentiation of neural stem cells. Kdm4c hypomorphic mutant mice exhibit abnormal behaviors such as hyperactivity, persistence, and learning and memory deficits. These findings suggest the significance of KDM4C for early neural development.

This study strengthens the evidence for associations between KDM4C CNVs and these two disorders. Then, it revealed that KDM4C deletion may confer susceptibility to the pathogenesis of SCZ through dysregulation of histone methylation patterns.

Research Background
The diagnostic criteria for schizophrenia (SCZ) and autism spectrum disorder (ASD) are based on subjective symptoms of patients, and diagnostic methods and treatments based on
pathophysiology have not been fully developed. Strong evidence indicates that genetic factors contribute substantially to the etiology of SCZ and ASD. Recent genome-wide studies of both disorders revealed that rare copy number variations (CNVs) may have large effect sizes and that elucidating the pathophysiology of these disorders may be possible. Previous studies have reported that KDM4C gene, which plays an important role in histone methylation modification, is involved in the regulation of neural stem cell differentiation and that model mice exhibit phenotypes associated with both disorders. These findings suggest that KDM4C plays an important role in neurodevelopment. In addition, KDM4C CNVs have been associated with both disorders. However, the association between KDM4C CNVs and each of the two disorders has not been fully evaluated, and the functional impact of KDM4C CNVs has not been studied using cells derived from clinical cases with such variants.

**Research Results**

KDM4C CNVs were analyzed in patients with SCZ, ASD, and controls (more than 6,000 in total). Exonic CNVs of KDM4C in nine SCZ cases and three ASD cases were identified, but none were detected in the controls (Figure 1). Eight of 12 CNVs were deletions (six in SCZ and two in ASD). KDM4C CNVs were significantly associated with both disorders. A significant association between deletion and SCZ was also observed. Through the evaluation of the clinical characteristics of patients with KDM4C CNVs, about half of SCZ cases showed treatment resistance despite high-dose antipsychotics.

![Figure 1](image-url) **KDM4C CNVs detected in patients with schizophrenia and ASD**

In mRNA expression analysis using LCLs, the mRNA transcript level was significantly lower in LCLs from the patient with the KDM4C deletion than in LCLs from SCZ patients without CNVs and healthy controls. Immunoblotting analysis using LCLs from a case with KDM4C deletion revealed reduced KDM4C protein expression and the elevation of methylation levels of H3K9 and H3K36 (Figure 2). These findings suggested that KDM4C deletion may confer susceptibility to the pathogenesis of SCZ through haploinsufficiency of KDM4C and dysregulation of histone methylation modification.
Research Summary and Future Perspective

This study revealed the association between $KDM4C$ CNVs and SCZ and ASD, respectively, and the potential effects of $KDM4C$ deletion on histone methylation patterns. In future research, it should be examined how $KDM4C$ regulates the spatio-temporal expression of other genes during neural development and how KDM4C contributes to neurogenesis and the pathogenesis of both disorders.
Publication

Translational Psychiatry

Rare Genetic Variants in the Gene Encoding Histone Lysine Demethylase 4C (KDM4C) and Their Contributions to Susceptibility to Schizophrenia and Autism Spectrum Disorder

Hidekazu Kato1, MD, Itaru Kushima1,2, MD, PhD, Daisuke Mori1,3, PhD, Akira Yoshimi4, PhD, Branko Aleksic1, MD, PhD, Yoshihiro Nawa1, MD, Miho Toyama1, MMedSc, Sho Furuta1, MD, Yanjie Yu1, MD, PhD, Kanako Ishizuka1, MD, PhD, Hiroki Kimura1, MD, PhD, Yuko Arioka1,5, PhD, Keita Tsujimura1,6, PhD, Mako Morikawa1, MD, PhD, Takashi Okada1, MD, PhD, Toshiya Inada1, MD, PhD, Masahiro Nakatochi7, PhD, Keiko Shinjo8, MD, PhD, Yutaka Kondo8, MD, PhD, Kozo Kaibuchi9, MD, PhD, Yasuko Funabiki9, MD, PhD, Ryosuke Kimura10, MD, PhD, Toshimitsu Suzuki11,12, PhD, Kazuhiro Yamakawa11,13, PhD, Masashi Ikeda14, MD, PhD, Nakao Iwata14, MD, PhD, Tsutomu Takahashi15, 16, MD, PhD, Michio Suzuki15, 16, MD, PhD, Yutaka Okahisa17, MD, PhD, Manabu Takaki17, MD, PhD, Jun Egawa18, MD, PhD, Toshiyuki Someya18, MD, PhD and Norio Ozaki1, MD, PhD

1 Department of Psychiatry, Nagoya University Graduate School of Medicine, Nagoya, Japan
2 Medical Genomics Center, Nagoya University Hospital, Nagoya, Japan
3 Brain and Mind Research Center, Nagoya University, Nagoya, Japan
4 Division of Clinical Sciences and Neuropsychopharmacology, Faculty and Graduate School of Pharmacy, Meijo University, Nagoya, Japan
5 Center for Advanced Medicine and Clinical Research, Nagoya University Hospital, Nagoya, Japan
6 Innovative Research Unit for Developmental Disorders, Institute of Advanced Research, Nagoya University, Nagoya, Japan
7 Public Health Informatics Unit, Department of Integrated Health Sciences, Nagoya University Graduate School of Medicine, Nagoya, Japan
8 Division of Cancer Biology, Nagoya University Graduate School of Medicine, Nagoya, Japan
9 Department of Cell Pharmacology, Nagoya University Graduate School of Medicine, Nagoya, Japan
10 Department of Cognitive and Behavioral Science, Graduate School of Human and Environmental Studies, Kyoto University, Kyoto, Japan
11 Department of Anatomy and Developmental Biology, Graduate School of Medicine, Kyoto University, Kyoto, Japan
12 Department of Neurodevelopmental Disorder Genetics, Institute of Brain Science, Nagoya City University Graduate School of Medical Sciences, Nagoya, Japan
13 Laboratory for Neurogenetics, RIKEN Center for Brain Science, Saitama, Japan
14 Department of Psychiatry, Fujita Health University School of Medicine, Toyoake, Japan
15 Department of Neuropsychiatry, University of Toyama Graduate School of Medicine and Pharmaceutical Sciences, Toyama, Japan
16 Research Center for Idling Brain Science, University of Toyama, Toyama, Japan
17 Department of Neuropsychiatry, Okayama University Graduate School of Medicine, Dentistry and Pharmaceutical Sciences, Okayama, Japan
18 Department of Psychiatry, Niigata University Graduate School of Medical and Dental Sciences, Niigata, Japan

DOI: https://doi.org/10.1038/s41398-020-01107-7

Japanese ver.