Midnolin is a novel risk factor for Parkinson's Disease and regulates expression of parkin and other causative genes

Yutaro Obara¹, Toru Imai¹, Hidenori Sato², Yuji Takeda³, Takeo Kato⁴, Kuniaki Ishii¹

¹Department of Pharmacology, Yamagata University School of Medicine, Japan, ²Genome Informatics Unit, Institution for Promotion of Medical Science Research, Yamagata University School of Medicine, Japan, ³Department of Immunology, Yamagata University School of Medicine, Japan, ⁴Department of Neurology, Hematology, Metabolism, Endocrinology and Diabetology, Yamagata University School of Medicine, Japan

Backgrounds: Parkinson’s Disease (PD) is a serious and progressive neurodegenerative disorder. Selective degeneration of dopaminergic neurons of the substantia nigra projecting into the striatum results in PD onset. Although approximately twenty causative genes have been identified, the majority is sporadic and its causes remain unclear. Therefore, it is necessary to clarify the detailed pathological mechanism.

Methods: To identify a novel PD-related gene, a hundred of healthy people and 86 patients with sporadic PD (sPD) were enrolled in Yamagata Cohort Study, and their genomic DNA was analyzed to assess single nucleotide polymorphisms and copy number variations by microarray. To examine pathophysiological roles of midnolin (MIDN), a candidate for PD-risk factor, MIDN gene in PC12 cells was knocked out by CRISPR/Cas9. These cells were also used for transcriptome analysis by RNA-sequencing.

Results: We found a significant copy number loss in MIDN with 10.5% of the patients with sPD whereas no MIDN loss was observed in healthy people. MIDN gene expression was enhanced by nerve growth factor (NGF) in PC12 cells, which was attenuated by selective inhibitors of ERK1/2 (U0126) or ERK5 (BIX02189). MIDN mainly localized in nucleus and intracellular vesicle membranes. When MIDN gene was knocked out by CRISPR/Cas9, the neurite outgrowth by NGF was abolished. Interestingly, mRNA and protein expression of Parkin, one of major PD-causative gene products, was inhibited in these cells, suggesting MIDN regulates Parkin gene expression. Mechanism of Parkin expression has been reported that it is regulated by a transcription factor, ATF4 which binds to cAMP response element (CRE) on the Parkin core promoter. In fact, CRE activity was significantly reduced in MIDN-lacking cells. Furthermore, expression of Parkin and ATF4 protein was reduced by CRISPR/Cas9 and siRNA targeting MIDN, indicating that MIDN regulates Parkin expression via ATF4. In order to clarify the role of MIDN in gene expression, we performed transcriptome analysis. MIDN positively and negatively regulated the mRNA expression of wide variety of genes including familial PD-causative genes such as α-synuclein, parkin, and EIF4G1.

Conclusions: These results suggest that MIDN is a novel risk factor for PD, and regulates expression of Parkin and possibly other PD-causative genes.