Original Article

IncRNA PART1 mitigates MPP+ -induced neuronal injury in SH-SY5Y cells via micRNA-106b-5p/MCL1 axis

Yue‘e Shen1, Xintao Cui2, Nan Xu3, Yuhang Hu2, Zhenyu Zhang2

Departments of 1Neurology, 2Orthopedics, The First Affiliated Hospital, Harbin Medical University, Harbin, Heilongjiang Province, China; 3Department of Anesthesiology, The Fourth Affiliated Hospital, Harbin Medical University, Harbin, Heilongjiang Province, China

Received March 9, 2021; Accepted June 8, 2021; Epub August 15, 2021; Published August 30, 2021

Abstract: Objective: Parkinson’s disease (PD) is a neurodegenerative disease caused by the loss of dopaminergic neurons. Here, we aimed to explore the function of LncRNA PART1 in PD and its underlying mechanisms. Methods: An in vivo MPTP-induced mouse model of PD was generated and the SH-SY5Y cells were treated with MPP+ to induce neuronal damage in vitro. The expressions of LncRNA PART1 and microRNA-106b-5p were assessed by RT-qPCR. The level of caspase 3 protein was detected by western blot. CCK8 assay and Annexin V/PI staining were used for detecting cell viability and survival rate, respectively. The interactions between microRNA-106b-5p and LncRNA PART1 or MCL1 were determined by RNA pull-down assay, RIP assay and DLR assay. The levels of inflammatory cytokines were assessed by ELISA, and the levels of LDH, ROS or SOD were verified using the appropriate assay kits. Results: The expression of LncRNA PART1 was decreased in PD model in vivo and in vitro (all P<0.05). In SH-SY5Y cells treated with MPP+, the overexpression of LncRNA PART1 increased cell viability and reduced cell apoptosis, the secretion of inflammatory cytokines and oxidative stress reaction (all P<0.05). Furthermore, LncRNA PART1 sponged microRNA-106b-5p which directly targeted MCL1 and thus regulated the expression of MCL1. LncRNA PART1 attenuated the injury of SH-SY5Y cells induced by MPP+ via targeting microRNA-106b-5p and enhancing MCL1 expression. Conclusion: LncRNA PART1 could alleviate the damage effects of MPP+ on SH-SY5Y cells by regulating microRNA-106b-5p/MCL1 axis, suggesting the potential therapeutic value of LncRNA PART1 as a target in PD.

Keywords: LncRNA PART1, microRNA-106b-5p, MPP+, SH-SY5Y, Parkinson’s disease

Introduction

Parkinson’s disease (PD), a chronic neurodegenerative disease in the central nervous system, affects 1-2% of individuals over the age of 60. The main pathological feature of PD is the degeneration and death of dopaminergic neurons in the substantia nigra [1]. PD has a slow progression and difficult diagnosis in early stage. With the prolongation of the onset time and gradual deterioration, the therapeutic efficacy of traditional PD drugs, such as levodopa, is unsatisfactory in recent years [2, 3]. Thus, it is of great importance to explore the pathogenesis and therapeutic targets of PD, so as to develop more effective treatments. With the progress of bioinformatics and further researches on non-coding RNAs, the functions of non-coding RNAs in the regulation of PD progression have been proved [4]. Long non-coding RNAs (IncRNAs) are endogenous non-coding RNAs with a length of about 200 nucleotides. LncRNAs regulate microRNAs (miRNAs) expression through chromatin modification or acting as endogenous sponges to silence miRNAs transcription [5]. MiRNAs, as evolutionary conserved non-coding RNAs, can inhibit protein expression by targeting their encoding mRNAs [6]. LncRNAs can compete with miRNAs for interacting with target mRNAs, thus antagonizing the function of miRNAs [7]. The roles of the LncRNAs-miRNAs-mRNA axis in PD have been demonstrated in several studies [8, 9]. Whether LncRNAs promote or inhibit PD progression depends on the role of their downstream target genes. To date, many proteins have been verified as potential targets for PD therapy. MCL1, a member of the BCL2 family, has been found to delay or block cell death in PD [10]. In addition, BCL2 and MCL1 also pro-
mote the survival of midbrain dopamine neurons [11]. Therefore, the mechanisms involved in regulating MCL1 expression in PD need to be further studied.

LncRNA PART1 (herein after referred to as PART1) is an identified lncRNA that has been reported to be a regulator in a number of diseases. Previous studies show that PART1 can impact the development of many kinds of tumors, such as glioma, and colorectal cancer [12, 13]. Also, the expression of PART1 was downregulated in patients with PD [14]. However, the roles of PART1 in PD and its underlying mechanisms were rarely investigated. In the present work, we aimed to study the effects and potential mechanism of PART1 in PD, in order to determine whether PART1 can serve as a candidate target for PD therapy.

Materials and methods

Cell culture and transfection

Human SH-SY5Y neuroblastoma cells collected from American Type Culture Collection (ATCC) were incubated with DMEM medium (Gibco, Grand Island, USA) containing 12% fetal bovine serum (FBS) (Gibco). The cells were cultured under conditions of 37°C with 5% CO2. For PART1 overexpression, lentiviral vectors containing PART1 pcDNA3.1 were transfected into SH-SY5Y cells, and the pcDNA3.1 NC was used in the control group. For overexpression or knockdown of miRNAs, microRNA-106b-5p mimics, microRNA-106b-5p inhibitor, mimics NC, and inhibitor NC were used. For dual-luciferase report (DLR) assay, the sequence of PART1 or MCL1 containing the binding sites (including wide type and mutation type) to microRNA-106b-5p were established. The above sequences are shown in Table 1. For RNA pull down assay, cell lysates were incubated with Bio-microRNA-106b-5p and Bio-NC. All the materials used for transfection were purchased from GE- NEPHARM (Suzhou, China). The transfections were diluted by serum-free medium and mixed with Lipofectamine 2,000 reagent (Invitrogen, Carlsbad, CA, USA) carefully.

Bioinformatics analysis

The binding site of PART1 to microRNA-106b-5p was predicted by Starbase 2.0 database (https://circinteractome.nia.nih.gov/index.html).

Establishment of PD model in vitro and in vivo

For in vitro PD model, SH-SY5Y cells were treated with 1 μM 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPP+) (Sigma-Aldrich, St Louis, USA) for 48 h. For in vivo PD model, the C57BL/6 mice (male, 6-8 weeks, 20-22 g, n=10/per group) (Shanghai Laboratory Animal Center of Chinese Academy of Sciences, Shanghai, China) were used. The mice received injection of 30 mg/kg 1-methyl-4-phenyl-1,2,3,6-tetrahydroxyidine-hydrochloride (MPTP) (Sigma-Aldrich) intraperitoneally for 7 days. As for control group, the mice received injection of saline solution. Mice were sacrificed after the intra- peritoneal injection of 1% pentobarbital sodium (50 mg/kg) and the brains were removed for RT-qPCR and western blot. Animal protocols in this study were performed in compliance with the Guide for Care and Use of Laboratory Animals of National Institutes of Health with approval of the ethics committee of our hospital.

Western blot

The protein extraction (RIPA buffer, Thermo Fisher Scientific, Waltham, MA, USA), concentration determination (BCA protein assay kit, Thermo Fisher Scientific), separation and transferring to nitrocellulose filter membrane (Millipore, Boston, USA), incubation with BSA and antibodies were performed as described previously. The primary antibodies for tyrosine hydroxylase (TH), α-synuclein, MCL1, caspase 3 (Cell Signaling Technology, USA; all of them were diluted by 1:1,500) and β-actin (Santac-
ruz, USA) were incubated at 4°C overnight. The secondary antibodies were obtained from Proteintech (USA). Protein expression was detected by ECL system (Amersham Pharmacia, Piscataway, NJ, USA).

Quantitative reverse transcription PCR (RT-qPCR)

Total RNA was extracted, reversely transcribed into cDNA, and amplified. This approach has been elaborated in past studies. The primers used in this study are shown in Table 2.

CCK8 assay

The cell proliferation was detected by CCK8 assay. Cells (50,000/mL) were seeded and incubated in 96 well plates for indicated time. 15 μL CCK8 solution (Dojindo, Shanghai, China) was added and then reacted for 2 h. The optical density (OD) value was assessed by a microtitre plate reader (BioTek, Winooski, VT, USA) at 450 nm.

Apoptosis detection

Annexin V-FITC/PI-PerCP staining assay kit (Vazyme Biotech Co., Ltd, Nanjing, China) was used to determine the apoptosis rates. Cells were diluted in binding buffer (100 μL) and then incubated with probes (5 μL Annexin V and 5 μL PI) for 10 min. After resuspending with 300 μL of binding buffer, the fluorescence was detected by flow cytometry (FACS Calibur, BD, San Jose, CA, USA). The data were analyzed by FlowJo software (version 8.8.6; Tree Star).

Caspase 3 activity detection

Caspase 3 Activity Assay Kit (Beyotime Biotechnology, Shanghai, China) was used for caspase 3 activity detection. First, cells after indicated treatment were collected and washed by PBS. After making standard curve from standard product, cells were lysed and incubated with the reaction solution (including Ac-DEVD-pNA) at 37°C for 60 min. The OD value was assessed by a microtitre plate reader at 450 nm.

Enzyme-linked immunosorbent assay (ELISA)

The levels of inflammatory cytokines including TNF-α, IL-6 and IL-1β in SH-SY5Y cells with PART1 overexpression were determined by ELISA assay kits (Cayman Pharma, Czech Republic, Cayman Europe, Estonia). Operation was performed according to the instruction [15]. After washing, cells were lysed and then incubated with specific reacting solution. The OD value was assessed by a microtitre plate reader at 450 nm.

Reactive oxygen (ROS) detection

The production of ROS was detected by Reactive Oxygen Species Assay Kit (Beyotime Biotechnology). The cells were collected and washed by PBS twice. Then, the probes were diluted by PBS (1:1,000) and incubated with cells for 20 min in dark at 37°C. After reaction, the cells were washed and resuspended in PBS. The fluorescence was detected by flow cytometry.

Lactate dehydrogenase (LDH) detection

The production of LDH was detected by LDH Cytotoxicity Assay Kit (Beyotime Biotechnology). The cells were seeded in 96 well plates. After treatment, the culture medium was removed and the diluted LDH released reagent was added. After 1 h incubation at 37°C, the supernatants were collected and transferred to a new 96 well plate for detection. The absorbance value was then measured at 490 nm.

Superoxide dismutase (SOD) detection

Total Superoxide Dismutase Assay Kit with WST-8 (Shanghai Enzyme-linked Biotechnology Co., Shanghai, China) was used in our experiments. In brief, cells were mixed with the SOD

### Table 2. The sequence of primers

<table>
<thead>
<tr>
<th></th>
<th>Forward</th>
<th>Reverse</th>
</tr>
</thead>
<tbody>
<tr>
<td>GAPDH</td>
<td>5'-TCCGTGGTCCACGAGAACT-3'</td>
<td>5'-GAAGCATTGGCGGTGAGCAT-3'</td>
</tr>
<tr>
<td>PART1</td>
<td>5'-AAAGCCGCTGACAGACTCA-3'</td>
<td>5'-GGTGTCATCAGCCCTGGA-3'</td>
</tr>
<tr>
<td>microRNA-106b-5p</td>
<td>5'-TGGCGCAACACCAGTGATGG-3'</td>
<td>5'-CCAGTGCAGGGTCCAGGAGT3'</td>
</tr>
<tr>
<td>U6</td>
<td>5'-CTCGCTCTGCAGCCACACA-3'</td>
<td>5'-AACGCTCTCAGAATTTGGGT3'</td>
</tr>
<tr>
<td>MCL1</td>
<td>5'-GGACATCCAAAAACGAGACG-3'</td>
<td>5'-GCAGCTTTTGTGTTATG-3'</td>
</tr>
</tbody>
</table>
Role of LncRNA PART1 in neuronal injury induced by MPP⁺

working solution and then incubated at 37°C for 30 min. After that, the absorbance of each sample was measured at 450 nm.

_Dual-luciferase report (DLR) assay_

To examine whether microRNA-106b-5p targeted PART1 or MCL1 directly, PART1-WT, PART1-MUT, MCL1-WT and MCL1-MUT reporter vectors were purchased from Promega (Madison, WI, USA). The assays were described previously [16]. Cells were culture in 96-well plates until the confluence reached 50-70%. Using Lipofectamine 2000 (Invitrogen), cells were co-transfected with reporter vectors and microRNA-106b-5p-NC or microRNA-106b-5p mimic. Following 48 h of post-transfection, the luciferase activity was estimated by Dual-Luciferase Reporter Assay System (Promega).

_RNA binding protein immunoprecipitation (RIP) assay_

The RIP experiments were carried out using an EZ-Magna RIP kit (Millipore, USA). After lysis, magnetic beads with antibody targeting Ago2 or IgG as well as RIP buffer were added. Then, proteinase K was employed to culture with these magnetic beads, and the precipitated RNA was subsequently eluted and verified using PCR.

_RNA pull-down assays_

MicroRNA-106b-5p was biotinylated, and oligo probe was used as control. The indicated cells were collected and lysed. The microRNA-106b-5p probe (Tsingke, Wuhan, China) and streptavidin magnetic beads (Life Technologies, USA) were co-incubated at room temperature for 2 h, which was then incubated with cell lysate at 4°C overnight. The beads were washed twice. The Trizol reagent was used for collection (TRizol reagent, Takara, Otsu, Japan) and the production was analyzed by RT-qPCR assay.

_Statistical analysis_

SPSS 18.0 software was used for statistical analyses, and data are presented as the mean ± SD from at least three independent experiments. Statistical analysis was performed with analysis of variance (ANOVA). The significance was determined through the Newman-Keuls post hoc test. Significant differences were determined at P<0.05.

Results

The expression of PART1 was decreased in PD

First, we investigated the expression of PART1 in a mouse PD model. As shown in Figure 1A, the expression of tyrosine hydroxylase (TH) was decreased, while the expression of α-synuclein was upregulated in MPTP group, confirming the successful establishment of PD model. The PART1 expression was also downregulated in MPTP group (Figure 1B). Then in vitro PD model was constructed by treating SH-SY5Y cells with 0, 0.5, 1 and 2 μM of MPP⁺ and the expression of PART1 was decreased in a concentration-dependent manner (Figure 1C). After treatment with 1 μM of MPP⁺ for 0, 12, 24 and 48 h, the expression of PART1 was reduced in a time-dependent manner (Figure 1C). Thus, the expression of PART1 was downregulated in PD.

Overexpression of PART1 alleviated neuronal injury

Various indicators can be used to evaluate neuronal abnormalities in PD, including the cell survival and viability, the secretion of inflammatory cytokines and the production of oxidative stress factors [17]. After transfecting with PART1 pcDNA3.1, the expression of PART1 was significantly increased (Figure 2A). MPP⁺ treatment reduced cell viability and increased the apoptosis rates, which could be blocked by PART1 overexpression (Figure 2B, 2C). MPP⁺ promoted the expression and activity of cleaved caspase 3, reflecting the initiation of apoptosis signaling (Figure 2D, 2E). The effects of MPP⁺ on caspase 3 cleavage was suppressed by PART1 (Figure 2D, 2E). In addition, MPP⁺ promoted the secretion of inflammatory cytokines such as TNF-α, IL-6 and IL-1β (Figure 2F-H). MPP⁺ also increased the level of oxidative stress factors, including LDH, ROS, and decreased the level of SOD (Figure 2I-K). However, after overexpression of PART1, inflammation and oxidative stress were weakened. Therefore, it was indicated that overexpression of PART1 alleviated the injury of SH-SY5Y cells treated with MPP⁺.

PART1 targets microRNA-106b-5p directly

Through bioinformatics analysis using Starbase 2.0, we found that PART1 had the binding site for microRNA-106b-5p (Figure 3A). Overexpression of PART1 could suppress the
Role of LncRNA PART1 in neuronal injury induced by MPP⁺

expression of miR-106b-5p (Figure 3B). Increased expression of microRNA-106b-5p was observed in SH-SY5Y cells transfected with microRNA-106b-5p mimics (Figure 3C). In order to further validate the interaction between microRNA-106b-5p and PART1, luciferase reporter assay was performed. As shown in Figure 3D, microRNA-106b-5p mimics decreased the activity of luciferase in SH-SY5Y cells transfected with PART1-WT plasmid but not those transfected with PART1-MUT plasmid. RNA pull-down assay and RIP assay also confirmed that microRNA-106b-5p could bind to PART1 (Figure 3E, 3F). Moreover, the enhanced expression of microRNA-106b-5p could be detected in the PD model in vitro (Figure 3G) and in vivo (Figure 3H), showing the aberrant increase of microRNA-106b-5p in PD. Furthermore, there was a negative correlation between PART1 and microRNA-106b-5p in mouse model of PD (Figure 3I). These data suggested that PART1 targeted microRNA-106b-5p and regulated its expression negatively.

**MCL1 is the direct target of microRNA-106b-5p**

MiRNAs play the regulatory roles in the expression of mRNAs by binding to their 3'-UTR and promoting the post-transcriptional modification [18]. The bioinformatics tools (Starbase 2.0) predicted that microRNA-106b-5p could interact with MCL1, which was also confirmed by DLR assay (Figure 4A). In SH-SY5Y cells transfected with MCL1-MUT plasmid, the decreased luciferase activity was blocked compared with those transfected with MCL1-WT. Moreover, microRNA-106b-5p mimics inhibited the expression of MCL1 mRNA, while microRNA-106b-5p inhibitor increased the expression of MCL1 mRNA (Figure 4B), indicating the negative regulation of microRNA-106b-5p on MCL1. The results of western blot also proved the same trend at protein levels (Figure 4C). Next, the expression of MCL1 was assessed in PD model in vitro and in vivo. As shown in Figure 4D, 4E, the expression of MCL1 was decreased transcriptionally and translationally. Similarly, in MPTP-induced mouse model, MCL1 expression was also reduced (Figure 4F, 4G). The results suggested that the expression of MCL1 was downregulated in PD, which was negatively regulated by microRNA-106b-5p.

**PART1 attenuated the injury of SH-SY5Y cells induced by MPP⁺ via microRNA-106b-5p/MCL1 axis**

Finally, we explored whether miR-106b-5p/MCL1 contributed to the regulatory function of
Role of LncRNA PART1 in neuronal injury induced by MPP⁺

A

![Graph A]

B

![Graph B]

C

![Graph C]

D

![Graph D]

E

![Graph E]

F

![Graph F]

G

![Graph G]

H

![Graph H]

I

![Graph I]

J

![Graph J]

K

![Graph K]
Role of LncRNA PART1 in neuronal injury induced by MPP⁺

Figure 2. Overexpression of PART1 alleviated neuronal injury. SH-SY5Y cells transfected with pcDNA3.1-NC or PART1 pcDNA3.1 were treated with 1 μM MPP⁺ for 48 h. A: The expression of PART1 was examined by RT-qPCR; B: The cell viability was assessed by CCK8 assay; C: The cell apoptosis rate was detected by Annexin V-FITC/PI assay; D: The expression of cleaved caspase 3 was detected by western blot; E: The activity of cleaved caspase 3 was detected; F-H: The levels of inflammatory cytokines were examined by ELISA; I-K: The levels of LDH, ROS and SOD were examined. *P<0.05, **P<0.01. MPP⁺: 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine; LDH: Lactate dehydrogenase; ROS: Reactive oxygen; SOD: Superoxide dismutase.

Figure 3. PART1 targets microRNA-106b-5p directly. (A) The predictive binding site of PART1 was predicted by Starbase 2.0; (B) The expression of microRNA-106b-5p was examined by RT-qPCR in SH-SY5Y cells transfected with PART1 pcDNA3.1; (C) The expression of microRNA-106b-5p was examined by RT-qPCR in the SH-SY5Y cells transfected with mimics NC and microRNA-106b-5p mimics; (D) The DLR assay was used to detect the binding relationship between microRNA-106b-5p and PART1 in SH-SY5Y cells co-transfected with PART1-WT or PART1-MUT and mimics NC or microRNA-106b-5p mimics; (E, F) The RIP assay (E) and RNA pull down assay (F) were used to examine the interaction between PART1 and microRNA-106b-5p in SH-SY5Y cells; (G, H) The expressions of microRNA-106b-5p in SH-SY5Y cells (G) and PD mouse model (H) were examined by RT-qPCR; (I) The correlation analysis of PART1 and microRNA-106b-5p in PD mouse model. **P<0.01. DLR: Dual-luciferase report; PD: Parkinson’s disease.

PART1. As shown in Figure 5A, the expression of MCL1 was decreased after MPP⁺ treatment, which could be reversed by PART1 overexpression. However, after co-treatment with MPP⁺, PART1 pcDNA3.1 and microRNA-106b-5p mimics, the expression of MCL1 was decreased. We observed the similar regulatory effect of microRNA-106b-5p/MCL1 on PART1 by assess-
Role of LncRNA PART1 in neuronal injury induced by MPP+

Figure 4. microRNA-106b-5p targets MCL1 directly. (A) The predictive binding site of microRNA-106b-5p was predicted by Starbase 2.0. The DLR assay was used to detect the interaction between microRNA-106b-5p and MCL1 in SH-SY5Y cells co-transfected with MCL1-WT or MCL1-MUT and mimics NC or microRNA-106b-5p mimics; (B, C) The expression of MCL1 was detected by RT-qPCR (B) and western blot (C) in SH-SY5Y cells transfected with microRNA-106b-5p mimics or microRNA-106b-5p inhibitor; (D, E) The expression of MCL1 was detected by RT-qPCR (D) and western blot (E) in in vitro PD model constructed by treating SH-SY5Y cells with MPP+; (F, G) The expression of MCL1 was detected by RT-qPCR (F) and western blot (G) in in vivo PD model constructed by treating mice with MPTP. *P<0.05, **P<0.01. miRNAs: microRNAs; DLR: Dual-luciferase report; PD: Parkinson’s disease; MPP+: 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine; MPTP: 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine-hydrochloride.

Figure 5B, cell apoptosis (Figure 5C), the function of cleaved-caspase 3 (Figure 5D, 5E), the secretion of inflammatory cytokines (Figure 5F-H) including TNF-α, IL-6 and IL-1β, and the production of oxidative stress factors (Figure 5I-K) including LDH, ROS and SOD. The results showed that the injury of SH-SY5Y cells was inhibited by PART1 overex-
Figure 5. PART1 attenuated the injury of SH-SY5Y cells induced by MPP⁺ via microRNA-106b-5p/MCL1 axis. After transfection with PART1 pcDNA3.1, microRNA-106b-5p mimics, and MCL1 siRNA, SH-SY5Y cells were treated with 1 μM MPP⁺ for 48 h. A: The expression of MCL1 was examined by western blot; B: The cell viability was assessed by CCK8 assay; C: The cell apoptosis rate was detected by Annexin V-FITC/PI assay; D: The expression of cleaved caspase 3 was detected by western blot; E: The activity of cleaved caspase 3 was detected; F-H: The levels of inflammatory cytokines were examined by ELISA; I-K: The levels of LDH, ROS and SOD were examined. *P<0.05, **P<0.01. MPP⁺: 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine.
Role of LncRNA PART1 in neuronal injury induced by MPP⁺

![Figure 6. The schematic diagram of PART1/microRNA-106b-5p/MCL1 axis.](image)

Expression as demonstrated by the above assays. However, after transfection with microRNA-106b-5p mimics or knockdown of MCL1, the protective effects of PART1 on SH-SY5Y cells were abrogated. The schematic diagram of our research is summarized in Figure 6. PART1 could negatively target microRNA-106b-5p which negatively target MCL1. In summary, these results proved that PART1 attenuated neuronal injury in vitro by modulating microRNA-106b-5p/MCL1 axis.

Discussion

PD is a common degenerative neurological disease that occurs most often in older patients [19]. More and more studies have revealed the important roles of LncRNAs in neurodegenerative diseases [20]. In the present work, we demonstrated that PART1 overexpression alleviated the neuronal injury, which was achieved by sponging microRNA-106b-5p and then upregulating MCL1 expression. Therefore, the overexpression of microRNA-106b-5p or knockdown of MCL1 could block the effects of PART1 on PD.

The functions of PART1 have been demonstrated in a variety of diseases, such as intervertebral disc degeneration and knee osteoarthritis [21, 22]. The most popular understanding of PART1 is its association with tumors. PART1 exerts its role in various tumors via regulating cell proliferation, suppression of cell apoptosis, or enhancement of cell migration [16, 23, 24]. PART1 could promote tumor development. In neurodegenerative diseases such as PD, whether PART1 may promote tumor development, thereby maintaining the survival of neuronal cells remains unknown. Previous study showed that PART1 was downregulated in PD as compared with healthy control and may play an essential role in PD progression [14]. Therefore, promoting the expression of PART1 might be beneficial in the treatment of PD. Our research for the first time validated that PART1 overexpression could attenuate the injury of SH-SY5Y cells, increase cell viability and suppress the production of inflammatory cytokines and oxidative stress factors. All these results indicated that PART1 contributed to cell survival, suggesting the potential protective ability of PART1 in neuron cells.

Mechanistically, we proved that PART1 targeted microRNA-106b-5p directly and regulated the expression of MCL1. Previous study showed that miR-106b-5p was a biomarker in PD diagnosis [25, 26]. Our data indicated that miR-106b-5p was upregulated in PD model both in vivo and in vitro, suggesting that the expression of miR-106b-5p was associated with PD progression. Unfortunately, due to the lack of clinical patient samples, the expression levels of
PART1 and microRNA-106b-5p were not verified. In human cancers, the effects of microRNA-106b-5p display a tumor type-specific manner. For example, in hepatocellular carcinoma, microRNA-106b-5p promotes cancer progression, while in colorectal cancer, microRNA-106b-5p inhibits the ability of cell migration and invasion [27, 28]. However, few studies have focused on the function of microRNA-106b-5p in PD to date. In our research, miR-106b-5p promoted neuronal injury, which was achieved by targeting MCL1 and thus inhibiting its expression. Overexpression of microRNA-106b-5p blocked the neuronal protective effects of PART1. MCL1 belongs to BCL2 family and is regarded as an anti-apoptotic protein. The overexpression of MCL1 is associated with the poor prognosis in cancer patients [29]. However, MCL1 promotes cell survival of midbrain dopamine neurons and is regarded as the therapeutical target of PD [10, 11]. It was also reported that MCL1 protected against cell apoptosis induced by MPP+ [30]. In MPP+-treated mice, MCL1 expression was found to be decreased [31]. These evidences suggested the potential protective role of MCL1 in PD. Here, we verified the beneficial effect of MCL1 on MPP+-induced neuronal injury and its associated mechanism.

In conclusion, this study identified PART1 as a novel protective factor in PD by sponging microRNA-106b-5p, thus suggesting the potential therapeutic value of PART1 in PD.

Disclosure of conflict of interest

None.

Address correspondence to: Zhenyu Zhang, Department of Orthopedics, The First Affiliated Hospital, Harbin Medical University, No. 23 Youzheng Steet, Nangang District, Harbin 150001, Heilongjiang Province, China. Tel: +86-15004623629; E-mail: zhangzhenyu_zzy@126.com

References


