Dexmedetomidine attenuates the neuroinflammation and cognitive dysfunction in aged mice by targeting the SNHG14/miR-340/NF-κB axis

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Abstract. Neuroinflammation plays a key role in the pathogenesis of postoperative cognitive dysfunction (POCD). Results of our previous study demonstrated that dexmedetomidine (Dex) attenuates neuroinflammation in BV2 cells treated with lipopolysaccharide (LPS) by targeting the microRNA (miR)-340/NF-κB axis. However, the molecular mechanisms by which Dex improves POCD remain unclear. In the present study, the association between long non-coding (lnc)RNA small nucleolar RNA host gene 14 (SNHG14) and miR-340 in BV2 microglial cells was determined using a dual-luciferase reporter assay. In addition, SNHG14, miR-340 and NF-κB expression levels were measured in LPS-treated BV-2 cells and hippocampal tissues of mice with POCD, and an enzyme-linked immunosorbent assay was used to determine the levels of proinflammatory mediators. Results of the present study demonstrated that SNHG14 exhibited potential as a target of miR-340. In addition, SNHG14 knockdown increased the levels of miR-340 and reduced the levels of NF-κB in LPS-treated BV2 cells. In addition, Dex treatment significantly reduced the levels of SNHG14 and NF-κB, and elevated the levels of miR-340 in the hippocampus of aged mice with POCD. Moreover, Dex treatment notably decreased the expression levels of TNF-α, IL-1β, IL-2, IL-6, IL-8 and IL-12 in the hippocampus of aged mice with POCD by upregulating miR-340. The spatial memory impairments in aged mice with POCD were also notably increased following Dex treatment via upregulation of miR-340. Collectively, results of the present study demonstrated that Dex may protect microglia from LPS-induced neuroinflammation *in vitro* and attenuate hippocampal neuroinflammation in aged mice with POCD *in vivo* via the SNHG14/miR-340/NF-κB axis. The present study may provide further insights into the mechanisms underlying Dex in the treatment of POCD.

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Introduction

Postoperative cognitive dysfunction (POCD) is a common postoperative complication observed in elderly patients following anesthesia and surgery (1,2). POCD is often characterized by memory loss and reduced cognitive dysfunction (3,4). Results of previous studies demonstrated that hippocampal neuroinflammation induced by anesthesia or surgery is a leading contributor to POCD (5-7). Dexmedetomidine (Dex) is a highly selective α2-receptor agonist, which is commonly used as an adjuvant in general anesthesia (8,9). Notably, Dex possesses anti-inflammatory and neuroprotective properties, thereby improving POCD (10). Glumac *et al* (11) reported that preoperative administration of Dex was able to reduce the risk of POCD after surgery by ameliorating the inflammatory response. Results of our previous study reported that Dex attenuates neuroinflammation in BV2 cells treated with LPS through targeting the microRNA (miR)-340/NF-κB axis (12). However, the molecular mechanisms by which Dex improves POCD remain unclear.

Long non-coding RNAs (lncRNAs) are a class of non-coding RNA molecules that are >200 nucleotides in length (13). Results of a previous study demonstrated that lncRNAs play critical roles in the development of POCD (14). For example, Yu *et al* (15) reported that lncRNA Rian attenuates sevoflurane anesthesia-induced cognitive dysfunction in mice by regulating the miR-143/LIMK1 axis. Wei *et al* (16) indicated that lncRNA NONMMUT055714 attenuates cognitive impairment in POCD mice by targeting miR-7684. Moreover, Deng *et al* (17) demonstrated that Dex treatment improves POCD in rats by modulating lncRNA LOC102546895, suggesting that lncRNA may be involved in the protective effects of Dex against POCD. Notably, lncRNA small nucleolar

RNA host gene 14 (SNHG14) promotes neurological impairment and inflammatory responses in rats following cerebral ischemia/reperfusion injury (18). Zhang *et al* (19) revealed that SNHG14 downregulation inhibits inflammatory responses in BV2 cells exposed to oxygen-glucose deprivation. However, the role of SNHG14 in POCD has not been widely studied. Data obtained using the StarBase bioinformatics tool demonstrated that miR-340 exhibits a complementary sequence to SNHG14 (20). Moreover, Zhang *et al* (20) revealed a potential interaction between miR-340 and SNHG14.

Thus, the present study aimed to explore the role of Dex in POCD *in vitro* and *in vivo*. To investigate these aims, a mouse model of POCD was established to explore the impact of Dex treatment on sevoflurane anesthesia-induced neuroinflammation in aged mice. Meanwhile, LPS-stimulated BV2 cells were used to mimic neuroinflammation to explore the impact of Dex in LPS-induced neuroinflammation *in vitro*. Additionally, the present study also aimed to determine the potential interaction with the SNHG14/miR-340/NF-κB axis. These results may provide new insights into the mechanisms underlying Dex in the treatment of POCD.

Materials and methods

Cell culture and transfection. Mouse microglial BV2 cells were obtained from Shanghai Zhongqiao Xinzhou Biotechnology Co., Ltd. BV2 cells were cultured in Minimum Essential Medium (MEM; Gibco; Thermo Fisher Scientific, Inc.) containing 10% FBS (Gibco; Thermo Fisher Scientific, Inc.) and 1% penicillin/streptomycin at 37˚C in 5% CO_2.

MiR-340 mimics (50 nM; 5'-UUAUAAAGCAAUGAG ACUGAUU-3') and mimics negative control (50 nM; NC; 5'-UUGUACUACACAAAAGUACUG-3'), miR-340 inhibitor, (100 nM; 5'-AAUCAGUCUCAUUGCUUUAUAA-3') and inhibitor NC (100 nM; 5'-UUCUCCGAACGUGUCACG UAA-3') were obtained from Guangzhou RiboBio Co., Ltd. Small interfering (si)RNA SNHG14 (si-SNHG14) were purchased from RiboBio Co., Ltd., and the sequences were as follows: Forward, 5'-GCUGAUAUUUAAGGCACUATT-3', and reverse, 5'-UAGUGCCUUAAAUAUCAGCTT-3'. A total of $2x10^5$ BV2 cells were plated into six-well plates overnight at 37˚C, and then cells were transfected with mimics NC (50 nM), miR-340 mimics (50 nM) or si-SNHG14 (50 nM) using Lipofectamine® 2000 (Invitrogen; Thermo Fisher Scientific, Inc.) for 6 h at 37˚C. Next, the culture medium was replaced with fresh MEM containing FBS, cells were then cultured for 24 h at 37˚C.

Reverse transcription-quantitative (RT-q)PCR. RT-qPCR was performed to assess SNHG14 and miR-340 expression levels in BV2 cells or in hippocampal tissues of mice. Total RNA was extracted from cells using TRIzol® reagent (Thermo Fisher Scientific, Inc.), according to the manufacturer's protocol. miRNA was reverse transcribed using the miScript II RT kit (Qiagen GmbH) according to the manufacturer's instructions, and qPCR experiments were carried out using a miScript SYBR Green PCR kit (Qiagen GmbH). In addition, reverse transcription of other genes was performed using the ReverTra Ace qPCR RT kit (Toyobo Life Science), and qPCR experiments were carried out using the Power SYBR Green PCR Master Mix (Vazyme Biotech Co., Ltd.). The cycling parameters were as follows: Denaturation at 95˚C for 5 min, followed by 45 cycles consisting for 15 sec at 94˚C, 30 sec at 55˚C and 30 sec at 70˚C. Expression levels were quantified using the $2^{-\Delta\Delta Cq}$ method (21) and normalized to the internal reference gene U6 or β-actin. The following primer pairs were used for qPCR: SNHG14 forward, 5'-CGTTGTCGAAAGCTA AAAGGA-3', and reverse, 5'-TGTTTCCATCTCACCAAA TGC-3'; β-actin forward, 5'-CTGGAACGGTGAAGGTGA CA-3', and reverse, 5'-CGGCCACATTGTGAACTTTG-3'; miR-340 forward, 5'-GTGGGGTTATAAAGCAATGAGA-3', and reverse, 5'-GTGCAGGGTCCGAGGT-3'; U6 forward, 5'-CTCGCTTCGGCAGCACAT-3', and reverse, 5'-AACGCT TCACGAATTTGCGT-3'.

Dual-luciferase reporter assay. StarBase bioinformatics tool (http://starbase.sysu.edu.cn) was used to predict the potential targets of SNHG14. To determine the relationship between SNHG14 and miR-340 in BV2 cells, dual-luciferase reporter assay was conducted. The putative miR-340-3p binding sequences of wild-type (WT) or mutant (MUT) SNHG14 were amplified and subcloned into pGL6-miR-based luciferase reporter plasmids (Beyotime Institute of Biotechnology). Subsequently, the aforementioned plasmids were co-transfected into BV2 cells with miR-340 mimics or NC using Lipofectamine® 2000 (Invitrogen; Thermo Fisher Scientific, Inc.). Relative luciferase activity was measured using a Dual-Luciferase Reporter Assay System (Promega Corporation) at 48 h. *Renilla* luciferase activity was used as a control.

Western blotting. The protein was collected from cells and tissues using the RIPA buffer (Sangon Biotech Co., Ltd.). Total protein concentration was quantified using a BCA protein assay kit (Sangon Biotech Co., Ltd.) and 20 μg/lane was separated via SDS-PAGE on a 10% gel. The separated proteins were subsequently transferred onto a PVDF membrane, and membranes were blocked with 5% non-fat milk in TBST (TBS with 0.1% Tween-20) for 1 h, and then incubated with antibodies against NF-kB p65 (1:2,000; cat. no. ab32536; Abcam) and GAPDH (1:2,000; cat. no. 60004-1-1; ProteinTech Group, Inc.) at 4˚C overnight. Following primary incubation, membranes were incubated with the HRP-conjugated goat anti-mouse secondary antibody (1:1,000; cat. no. A0216; Beyotime Institute of Biotechnology). Protein bands were visualized using the Pierce ECL Plus Western Blotting Substrate (Teasen), and protein expression was quantified using Image-Pro Plus software (v6.0; Media Cybernetics).

Enzyme-linked immunosorbent assay (ELISA). ELISA was applied to analyze the levels of cytokines in the supernatant of BV2 cells and in the hippocampus from mice. Mouse TNF-α ELISA kit (cat. no. ab208348; Abcam), mouse IL-1β ELISA kit (cat. no. ab197742; Abcam), mouse IL-6 ELISA kit (cat. no. ab222503; Abcam), mouse IL-2 ELISA kit (cat. no. ELK1150; ELK Biotechnology), mouse IL-8 ELISA kit (cat. no. EM30328S; Shanghai Weiao Biotechnology Co., Ltd.) and mouse IL-12 ELISA kit (cat. no. ELK9395; ELK Biotechnology) were used to detect the levels of TNF-α, IL-1β, IL-6, IL-2, IL-8 and IL-12 in cell culture supernatants, according to the manufacturers' protocols.

Animal experiments. The Ethics Committee of Jiading District Central Hospital Affiliated Shanghai University of Medicine & Health Sciences approved the animal experiments (approval no. 20190018). A total of 50 healthy aged C57BL/6 male mice (age, 18 months; 28-32 g) were obtained from the SPF (Beijing) Biotechnology Co., Ltd. (animal license no. SCXK (Beijing) 2019-0010), and housed in a specific pathogen-free environment (23±2˚C and 55±5% humidified atmosphere) under a 12/12 h light-dark cycle with free access to food/water in accordance with the recommended procedures of The National Institutes of Health Guide for the Care and Use of Laboratory Animals. Animals were randomly divided into 5 groups (n=10/group) as follows: Control, POCD, POCD + Dex, POCD + miR-340 inhibitor and POCD + miR-340 inhibitor + Dex groups. In POCD groups, mice were anesthetized using an anesthesia machine (R500; RWD Life Technologies Inc.) with sevoflurane (induction dose, 3%; maintenance dose, 2%). Subsequently, a 1.5-cm longitudinal incision was created along the midline of the abdomen. The abdomen was explored for 5 min, and the incision was closed using 5-0 and 4-0 sterile surgical sutures. Mice in the Dex treatment groups were intravenously treated with Dex (25 μg/kg) 30 min prior to POCD surgery. Mice in the miR-340 inhibitor groups were injected with a miR-340 inhibitor in the hippocampus 48 h prior to the POCD operation. Animals in the control group were not treated. After 48 h of operation, all mice were sacrificed using CO_2 at a displacement rate of 40% volume/min, and hippocampus tissues were collected.

Hematoxylin & Eosin (H&E) staining analysis. Hippocampus tissues were fixed in 4% paraformaldehyde overnight at 4˚C and then embedded in paraffin and sliced into 4-μm thick sections. Subsequently, sections were stained with hematoxylin reagent for 8 min and then stained with eosin reagent for 3 min at room temperature and examined under a light microscope.

Morris water maze (MWM). The MWM test was used to evaluate spatial learning and memory. MWM equipment (Chengdu Techman Technology Co., Ltd.) includes a round pool with a diameter of 150-cm that is divided into four quadrants. A movable circular platform (diameter, 9-cm) was placed 2-cm below the water surface in the fourth quadrant. During the experiment, mice entered the water facing the wall at each of the four starting locations and were trained to locate the hidden platform. If mice were unable to board the platform in 120 sec, rats were guided to the platform and the latency period was noted as 120 sec. A camera attached to a computer system (Chengdu Techman Technology Co., Ltd.) was used to record the trajectory of the mice. Following examination for 5 days, the platform was removed on Day 6. Animals were released into the water and the escape latency was recorded.

Statistical analysis. All experiments were repeated at least three times independently. Data are presented as the mean ± standard deviation (SD). Data were analyzed using GraphPad Prism software (version 7.0; GraphPad Software, Inc.; Dotmatics). One-way analysis of variance (ANOVA) followed by Tukey's post hoc tests were used to determine the differences between three or more groups. $P<0.05$ was considered to indicate a statistically significant difference.

Results

Dex attenuates neuroinflammation in LPS-treated BV2 cells by regulating the SNHG14/miR-340/NF-κB axis. To investigate whether the SNHG14/miR-340/NF-κB axis is involved in the development of POCD, the StarBase bioinformatics tool was used to predict whether miR-340 is a potential target of SNHG14. The data indicated that miR-340 had a complementary sequence to SNHG14 (Fig. 1A). In addition, results of the present study demonstrated that transfection with miR-340 mimics significantly reduced the luciferase activity of the SNHG14-WT vector in BV2 cells compared with the SNHG14-WT + mimics NC group (Fig. 1B). Collectively, these results demonstrated that SNHG14 exhibits potential as a target of miR-340.

Moreover, transfection with the miR-340 mimics notably elevated the expression levels of miR-340 in BV2 cells compared with the control group, which was confirmed using an RT-qPCR assay (Fig. 1C). In addition, transfection with si-SNHG14 significantly reduced SNHG14 expression levels in BV2 cells compared with the control group (Fig. 1D). LPS treatment significantly decreased miR-340 expression levels in BV2 cells compared with the control; however, this was significantly reversed following transfection with si-SNHG14, miR-340 mimics or Dex treatment (Fig. 1E). Notably, miR-340 expression levels were increased following si-SNHG14 transfection in LPS-treated BV2 cells (Fig. 1E). These results demonstrated that SNHG14 negatively regulated the expression of miR-340.

Results of previous studies demonstrate that surgery or anesthesia-induced neuroinflammation play critical roles in the progression of POCD (5,22). Thus, the levels of NF-κB protein and pro-inflammatory cytokines (TNF-α, IL-1β, IL-2, IL-6, IL-8 and IL-12) in LPS-treated BV2 cells were detected in the present study. As presented in Fig. 1F-G and 2A-F, LPS treatment significantly upregulated the levels of NF-κB, TNF-α, IL-1β, IL-2, IL-6, IL-8 and IL-12 in BV2 cells. However, si-SNHG14, miR-340 mimics or Dex treatment significantly decreased the levels of NF-κB and the aforementioned pro-inflammatory cytokines in LPS-treated BV2 cells compared with the LPS group (Figs. 1F-G and 2A-F). Collectively, results of the present study demonstrated that Dex treatment attenuated neuroinflammation in LPS-treated BV2 cells by regulating the SNHG14/miR-340/NF-κB axis.

Dex attenuates hippocampal damage in aged mice with POCD by regulating the SNHG14/miR-340/NF-κB axis. To explore whether Dex attenuated hippocampal damage in aged mice with POCD by regulating the SNHG14/miR-340/NF-κB axis, the expression levels of miR-340, SNHG14 and NF-κB were detected. As presented in Fig. 3A-C, miR-340 expression levels were significantly decreased and SNHG14 and NF-κB expression levels were notably increased in the hippocampus of aged mice with POCD compared with the control. In addition, this was partially reversed following treatment with Dex. In addition, results of the H&E staining assay demonstrated that the hippocampal neurons in the control group were neatly arranged and the cell structure was complete (Fig. 4A). By contrast, the hippocampal neurons in the POCD group demonstrated inflammatory cell infiltration and nuclear condensation,

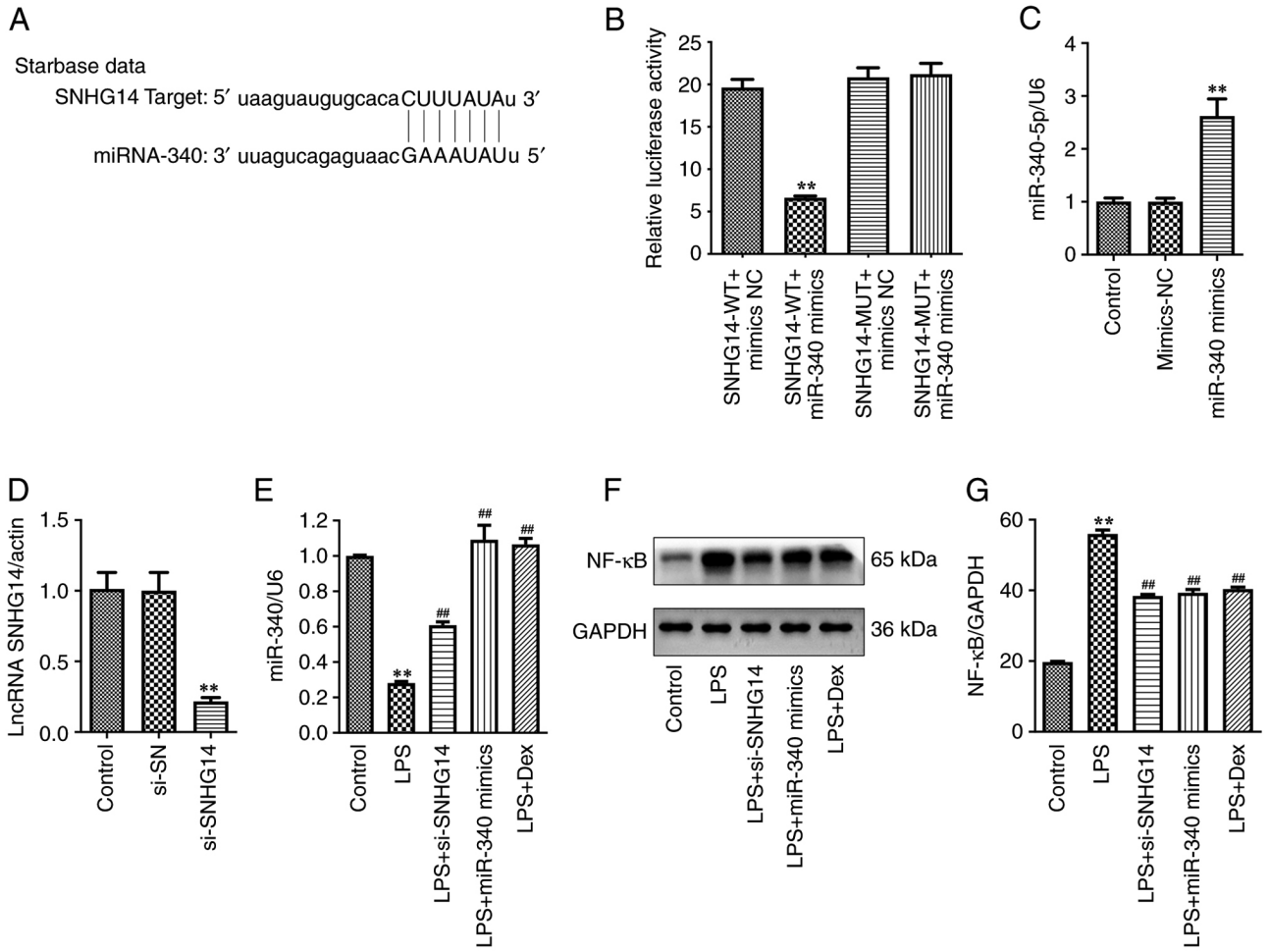

Figure 1. MiR-340 is a direct target of SNHG14 in BV2 cells. (A) Putative binding sites of miR-340 on SNHG14. (B) Luciferase assay of BV2 cells transfected with SNHG14-WT or SNHG14-MUT reporter together with miR-340. (C) RT-qPCR was applied to measure miR-340 level in BV2 cells transfected with miR-340 mimics. (D) SNHG14 level in si-SHNG14-transfected BV2 cells was detected using RT-qPCR. (E) BV2 cells were transfected with miR-340 mimics and si-SHNG14 for 24 h, and then treated with Dex for 30 min, followed by stimulation with 10 μg/ml LPS treatment for 24 h. The miR-340 level in BV2 cells was detected using RT-qPCR. (F and G) Western blotting was used to determine NF-κB expression in BV2 cells. **P<0.01 vs. control group; $^{\#\#}$P<0.01 vs. LPS group. MiR, microRNA; SNHG14, small nucleolar RNA host gene 14; WT, wild-type; MUT, mutated; RT-qPCR, reverse transcription-quantitative PCR; si, small interfering; Dex, dexmedetomidine; LPS, lipopolysaccharide; lncRNA, long non-coding RNA.

whereas the opposite results were observed following Dex treatment (Fig. 4A). Meanwhile, compared with the inhibitor NC group, miR-340 inhibitor significantly reduced miR-340 level in the hippocampus of aged mice (Fig. S1). Dex treatment significantly elevated miR-340 expression levels and reduced NF-κB expressions levels in the hippocampus of aged mice with POCD compared with the POCD group. Notably, the opposite results were observed following transfection with the miR-340 inhibitor (Fig. 4B and C). Collectively, results of the present study demonstrated that treatment with Dex attenuated hippocampal damage in aged mice with POCD by regulating the SNHG14/miR-340/NF-κB axis.

Dex attenuates hippocampal neuroinflammation and improves spatial memory impairments in aged mice with POCD by regulating the SNHG14/miR-340/NF-κB axis. To evaluate the effects of Dex on hippocampal inflammation in a mouse model of POCD, the levels of pro-inflammatory cytokines were measured using ELISA. As displayed in Fig. 5A-F, miR-340 inhibitor obviously increased, but Dex treatment significantly decreased the levels of TNF-α, IL-1β, IL-2, IL-6, IL-8 and IL-12 in the hippocampus of aged mice with POCD compared with the POCD group. However, compared with the POCD + Dex group, the inhibitory effects of Dex on the levels of these pro-inflammatory cytokines in the hippocampus of aged mice with POCD were reversed by miR-340 inhibitor (Fig. 5A-F). In addition, results of the MWM analysis demonstrated that Dex treatment significantly decreased latency to escape in aged mice with POCD, and these results were reversed following transfection with the miR-340 inhibitor (Fig. 6A and B). Collectively, results of the present study demonstrated that Dex attenuated hippocampal neuroinflammation and improved spatial memory impairments in aged mice with POCD by regulating the SNHG14/miR-340/NF-κB axis.

Discussion

The present study revealed that Dex could protect microglia from LPS-induced neuroinflammation *in vitro* and attenuate hippocampal neuroinflammation in aged mice with POCD *in vivo*. Mechanistically, Dex could ameliorate

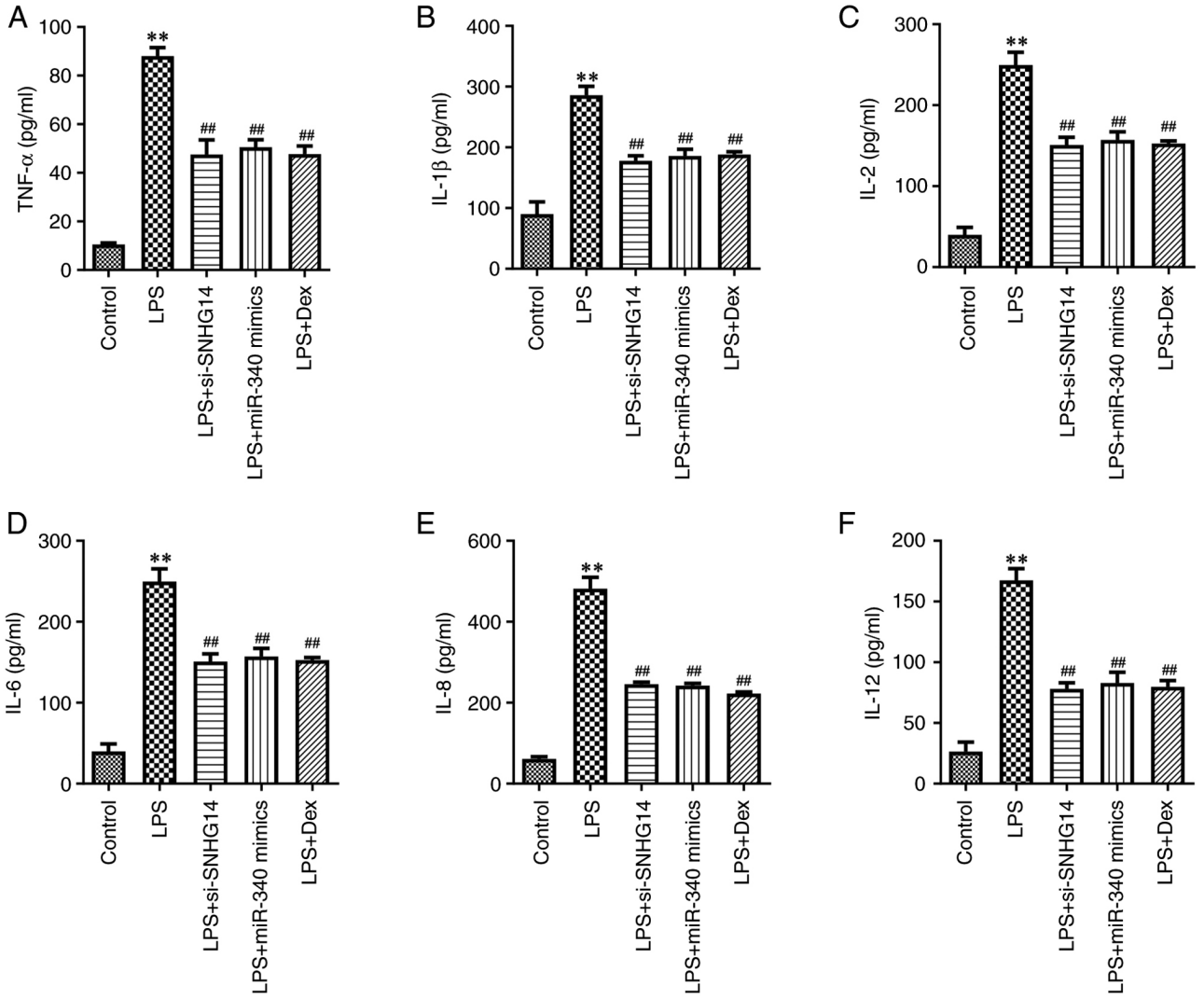

Figure 2. Dex attenuates neuroinflammation in LPS-stimulated BV2 cells by regulating the SNHG14/miR-340/NF-κB axis. BV2 cells were transfected with miR-340 mimics and si-SHNG14 for 24 h, and then treated with Dex for 30 min, followed by stimulation with 10 μg/ml LPS treatment for 24 h. ELISA assay was conducted to detect the level of (A) TNF-α, (B) IL-1β, (C) IL-2, (D) IL-6, (E) IL-8 and (F) IL-12 in the culture supernatant of BV2 cells. **P<0.01 vs. control group; $^{\#\#}$P<0.01 vs. LPS group. MiR, microRNA; SNHG14, small nucleolar RNA host gene 14; si, small interfering; Dex, dexmedetomidine; LPS, lipopolysaccharide.

inflammatory injury of hippocampal microglia cells through modulating the SNHG14/miR-340/NF-κB axis.

The inflammatory activation of hippocampal microglia plays a key role in the progression of POCD (23-25). In addition, microglia are resident immune cells in the central nervous system, and are key players in age-related neurodegenerative and neuroinflammatory diseases (26,27). It has been shown that NF-κB signaling exhibits a key role in POCD (28). Activation of NF-κB signaling in microglial cells is critical during neuroinflammation in neurodegenerative diseases (29,30). Zheng *et al* (31) revealed that NF-κB expression levels are upregulated in the hippocampal zone in rats following sevoflurane anesthesia (31). In addition, inactivating NF-κB signaling attenuates neuronal apoptosis and inflammatory responses in the hippocampus of aged rats with POCD (32). Moreover, Ding *et al* (33) found that andrographolide an alleviate POCD progression by modulating NF-κB/MAPK signaling. These data demonstrate that inhibiting neuroinflammation via the NF-κB signaling pathway may exhibit potential in alleviating POCD.

Zhou *et al* (34) indicated that Dex attenuates inflammatory responses in the hippocampus of aged mice with POCD by suppressing the TLR4-NF-κB signaling pathway. In addition, results of our previous study demonstrated that Dex reduces NF-κB expression levels in LPS-treated BV2 cells by upregulating miR-340, thereby attenuating microglial-mediated neuroinflammation (12). Thus, the present study aimed to explore whether Dex could attenuate neuroinflammation and cognitive impairment in aged mice with POCD *in vivo*. Results of the present study demonstrated that Dex treatment upregulated miR-340 expression levels and reduced NF-κB expression levels in the hippocampus of aged mice with POCD. In addition, Dex treatment decreased the levels of pro-inflammatory cytokines in the hippocampus of aged mice with POCD by upregulating miR-340. Moreover, Dex notably improved spatial memory impairments in aged mice with POCD via upregulation of miR-340. Collectively, these results revealed that Dex attenuated neuroinflammation and improved spatial memory impairments in aged mice with POCD by regulating the miR-340/NF-κB axis.

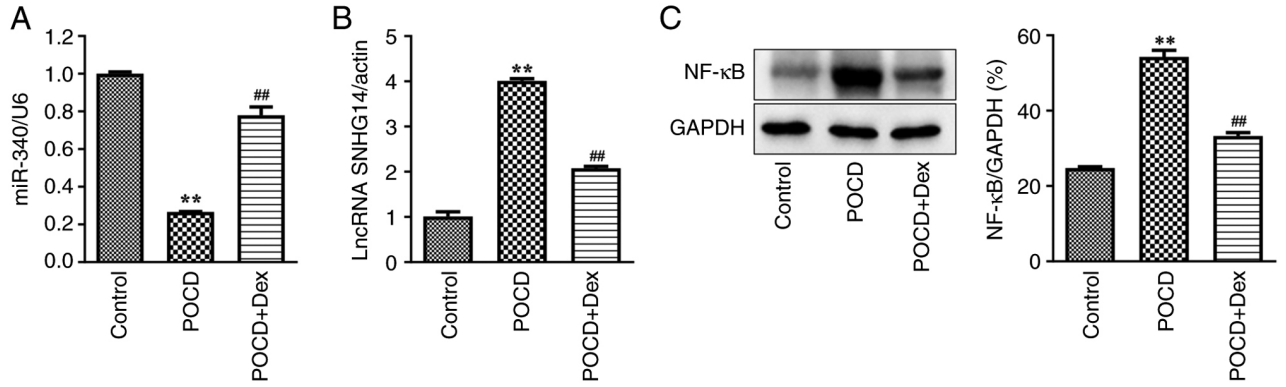

Figure 3. Dex downregulates the level of SNHG14 and NF-κB and upregulates the level of miR-340 in the hippocampus in aged mice with POCD. (A) MiR-340 and (B) SNHG14 level in the hippocampus were determined with reverse transcription-quantitative PCR. (C) Expression of NF-κB in the hippocampus was determined using western blotting. **P<0.01 vs. control group; $^{\#\#}$P<0.01 vs. POCD group. MiR, microRNA; SNHG14, small nucleolar RNA host gene 14; Dex, dexmedetomidine; POCD, postoperative cognitive dysfunction.

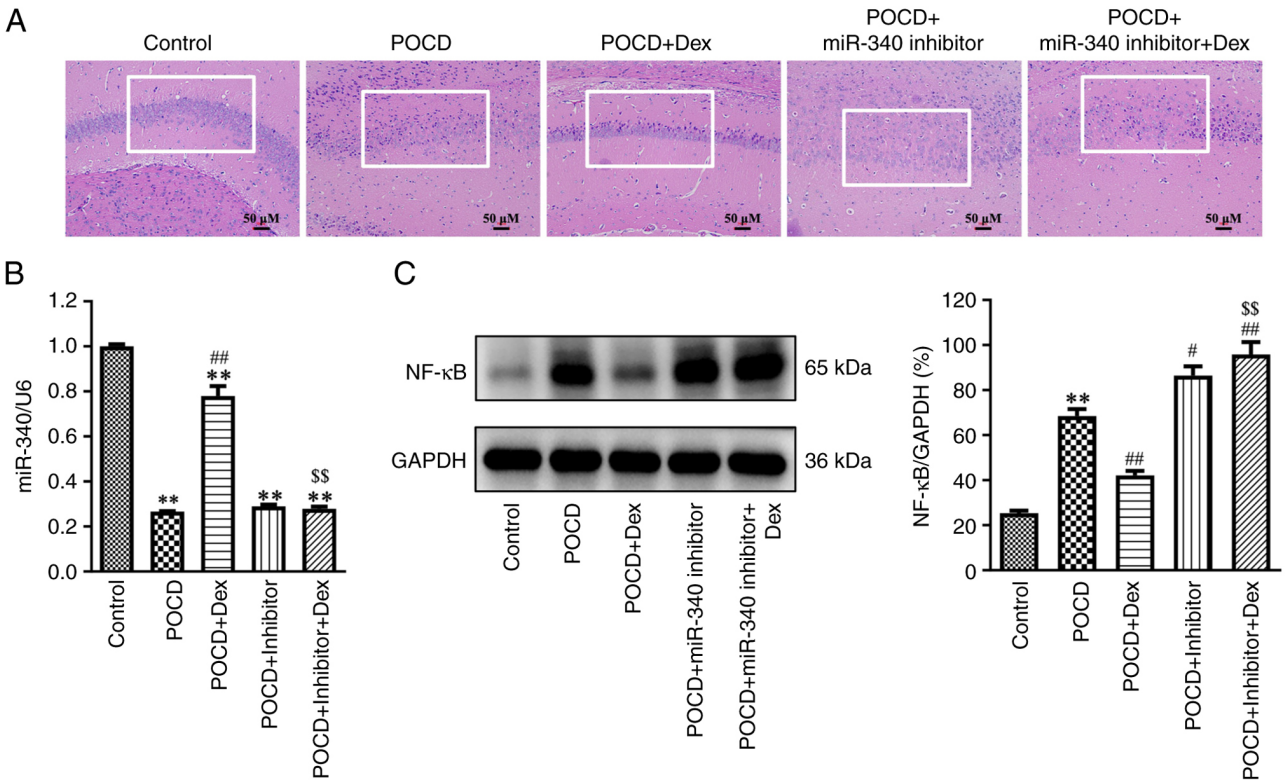

Figure 4. Dex attenuates hippocampal damage in aged mice with POCD by regulating the miR-340/NF-κB axis. (A) Hematoxylin and eosin-staining assay was performed to observe the pathological histology of the hippocampus (scale bar, 50 μM). (B) MiR-340 level in the hippocampus was measured with reverse transcription-quantitative PCR. (C) Expression of NF-κB in the hippocampus was measured using western blotting. **P<0.01 vs. control group; $^{\#}$P<0.05, $^{\#\#}$P<0.01 vs. POCD group; $^{\$\$}$P<0.01 vs. POCD + Dex group. Dex, dexmedetomidine; POCD, postoperative cognitive dysfunction; miR, microRNA.

However, the molecular mechanisms by which Dex improves POCD remain unclear. The lncRNA-miRNA-mRNA axis plays key roles in POCD development (15,16). In addition, lncRNAs, as important regulators in biological processes, are involved in the pathogenesis of nervous system disease (15,35). Mechanistically, lncRNAs regulate gene expression by sponging miRNAs (36). Tian *et al* (37) demonstrated that lncRNA SNHG8 may inhibit microglial inflammatory responses in mice subjected to ischemic injury through targeting the miR-425/SIRT1/NF-κB axis. Cao *et al* (38) reported that downregulation of lncRNA HOXA11-AS attenuates neuroinflammation and neuronal apoptosis in mice with Parkinson's disease via the miR-124/FSTL1/NF-κB axis. These data indicate that lncRNAs may modulate NF-κB expression by targeting miRNAs. Results of the present study demonstrated that SNHG14 exhibited potential as a target of miR-340. In addition, SNHG14 knockdown increased miR-340 levels and reduced NF-κB levels in LPS-treated BV2 cells *in vitro*. Moreover, Dex treatment reduced SNHG14 levels in the hippocampus of aged mice with POCD *in vivo*. Collectively, results of the present study demonstrated that Dex ameliorated

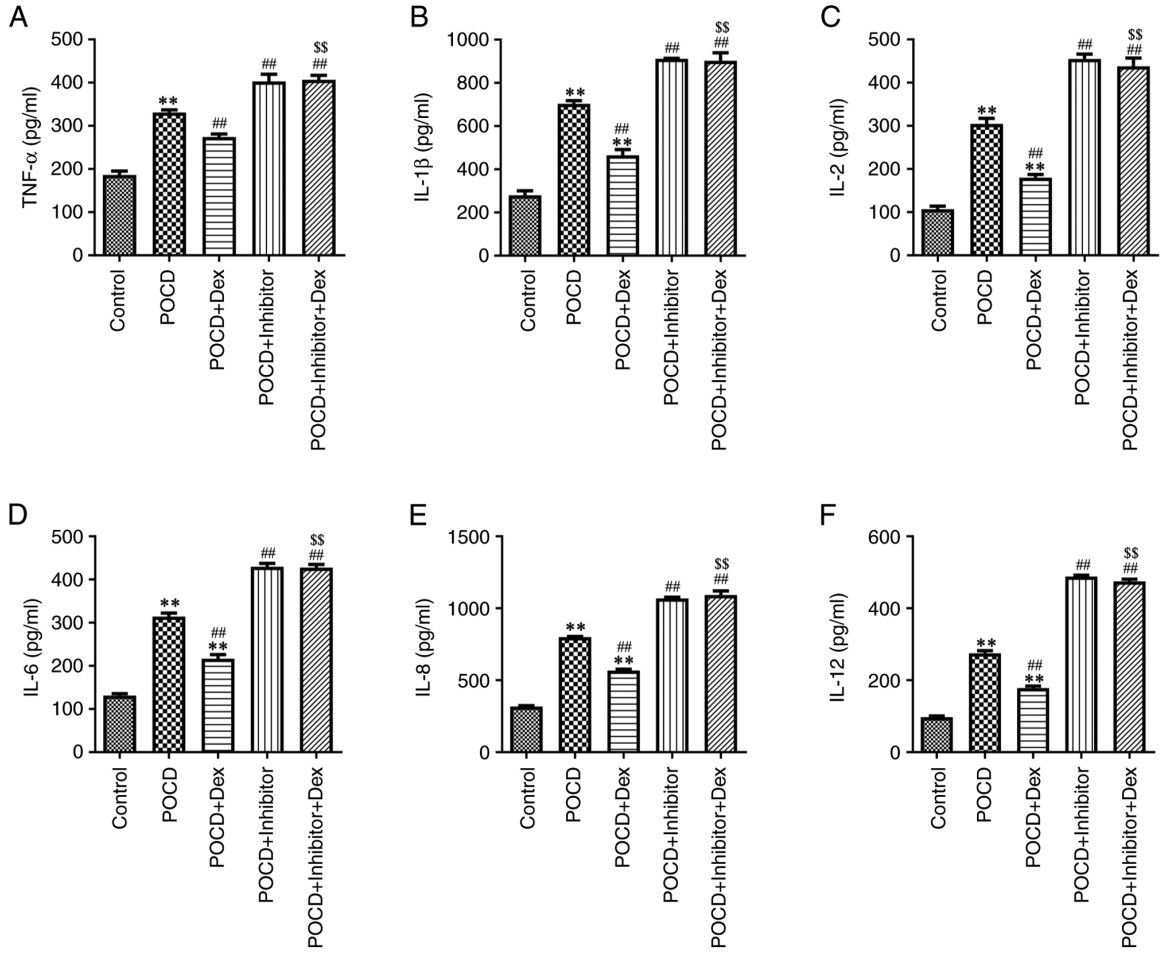

Figure 5. Dex attenuates neuroinflammation in the hippocampus in aged mice with POCD by regulating the miR-340/NF-κB axis. ELISA assay was applied to determine the level of (A) TNF-α, (B) IL-1β, (C) IL-2, (D) IL-6, (E) IL-8 and (F) IL-12 in the hippocampus. **P<0.01 vs. control group; $^{\#\#}$P<0.01 vs. POCD group; $^{\$\$}$P<0.01 vs. POCD + Dex group. Dex, dexmedetomidine; POCD, postoperative cognitive dysfunction; miR, microRNA.

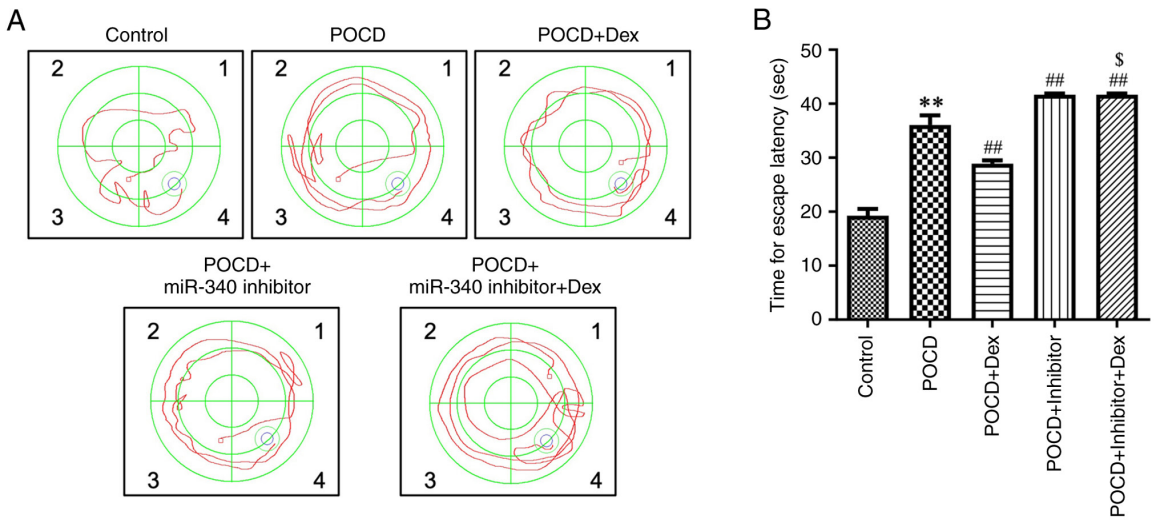

Figure 6. Dex improves spatial memory impairments in a mouse model of POCD by regulating the miR-340/NF-κB axis. (A and B) The learning and memory ability in POCD mice were evaluate using the Morris water maze test. **P<0.01 vs. control group; $^{\#\#}$P<0.01 vs. POCD group; $^{\$}$P<0.05 vs. POCD + Dex group. Dex, dexmedetomidine; POCD, postoperative cognitive dysfunction; miR, microRNA.

neuroinflammation and cognitive dysfunction in aged mice with POCD through targeting the SNHG14/miR-340/NF-κB axis. However, except for the SNHG14/miR-340/NF-κB axis, other signaling pathways involved in the neuroprotective effects of Dex in POCD are needed to be further investigated in the future.

Results of the present study demonstrated that Dex may protect microglia from LPS-induced neuroinflammation *in vitro* and attenuate hippocampal neuroinflammation in aged mice with POCD *in vivo* via the SNHG14/miR-340/NF-κB axis. The present study may provide further insights into the mechanisms underlying Dex in the treatment of POCD.

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Availability of data and materials

All data generated or analyzed during this study are included in this published article.

Authors' contributions

GH, HN, KW and HG were responsible for data investigation, data acquisition, data analysis, data interpretation and manuscript revision. YL, JG, XN, ZW and YB made substantial contributions to conception and design of the study and revised the manuscript. All authors agreed to be accountable for all aspects of the work. All authors confirmed the authenticity of all the raw data and read and approved the final manuscript.

Ethics approval and consent to participate

The Ethics Committee of Jiading District Central Hospital Affiliated Shanghai University of Medicine & Health Sciences approved the protocol of animal experiments (approval no. 20190018).

Patient consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

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