

An enriched environment promotes cognitive recovery and cerebral blood flow in aged mice under sevoflurane anaesthesia

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Abstract

Sevoflurane is an inhalation anaesthetic agent widely used in clinical settings. Despite good surgical outcomes using sevoflurane, patients frequently develop postoperative cognitive dysfunction (POCD). An enriched environment (EE), as a rehabilitation technique, could provide objects and tools to facilitate neuromotor and visual stimuli and brain activity, and is reported to improve cognitive functions. We aim to investigate the impairments of sevoflurane inhalation on cognitive function in mice and determine the benefits of EE in ameliorating POCD. Eighteen-month-old mice were exposed to sevoflurane inhalation for 2 h and then placed in standard environment (SE) or EE cages. The mice without sevoflurane exposure in standard or EE cages were used as controls. The behavioural tests include Morris water maze, Y maze and novel object recognition. Magnetic resonance imaging (MRI) was used to determine the blood circulation in the brains. The proangiogenic factors (CD31, angiopoietin-1, vascular endothelial growth factor, and N-cadherin) and neurotrophic (brain-derived neurotrophic factor, post-synaptic density protein 95) expression in hippocampus of aged mice were evaluated by Western blotting and RT-PCR analysis. Sevoflurane-exposed mice demonstrated reduced performance in learning, memory and spatial memory tests. Enriched environment improved the behavioural performance of sevoflurane-exposed animals. Sevoflurane exposure reduced the blood flow in the brains, and these effects were ameliorated by EE habitation. The EE also promoted the expression of angiogenic and neurotrophic factors in sevoflurane-exposed animals. In summary, EE is effective in ameliorating the side-effects of sevoflurane exposure in aged mice.

Key words: sevoflurane, memory, enriched environment, neuroinflammation.

Introduction

Safety and efficacy of sevoflurane, as an inhalation anaesthetic in clinical settings, have been tested in numerous studies [2]. Sevoflurane was reported to have cardioprotective [12] and liver protective [13] effects in hypoxia environments, contributing to high survival rates in sevoflurane-managed surgical interventions. However, cognitive disorders have also been reported

following anaesthesia events using sevoflurane, particularly in aged and paediatric patients, forming a complex of symptoms including progressive hypomnesia (deficiency of short-term memory and recollection), personality alteration or accentuation following surgery, defined as postoperative cognitive dysfunction (POCD) [1,18]. The detailed mechanisms of POCD after sevoflurane sedation are not entirely understood. Ani-

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mal studies indicate microglia activation and microglia-mediated pyroptosis of the neural cells in sevoflurane-exposed animals [21]. The increased expression of proinflammatory mediators and apoptosis markers *in vivo* was found in the brains of the rats after sevoflurane exposure [4]. Brain blood flow is also closely linked to cognitive function. The decoupling of cerebral blood flow (CBF) with brain metabolic demands due to vasodilation was reported in sevoflurane application [16]. Brain tissue hypoxia could result in the hyperphosphorylation of Tau protein [15].

An enriched environment (EE) is a set of objects and tools to provide neuromotor and visual stimuli to facilitate brain activity. Enriched environment was reported to alleviate senile cognitive impairments [26] and protect against post-stroke cognitive dysfunction [24]. After a neurological injury, EE could help the brain induce a positive response by enhancing the synaptic plasticity or triggering neurogenesis [5]. Furthermore, EE increased cerebral blood flow after an ischemic event [27]. The current study investigates if EE has beneficial effects against sevoflurane-induced POCD.

Material and methods

Animals

An animal experiment was granted by the Quanzhou First Hospital Affiliated to Fujian Medical University. C57BL/6J mice were obtained from GemPharmatech (Nanjing, China). Mice were fed water and normal chow diets *ad libitum*.

Sevoflurane anaesthesia model

The mice were placed in the plexiglass container connected to the EZ-7000 anaesthesia device (World Precision Instruments, Sarasota, FL). The mice were placed on the heating pad maintained at 37°C and subjected to continuous inhalation of sevoflurane (3%, Sigma, St. Louis, MO) in pure oxygen for 2 h. Following the sevoflurane exposure, mice undergo standard recovery inside the cage on the heating pad maintained at 38°C for 2 h until sternal recumbency. Naïve mice were used as controls.

Enriched environment modelling

In the EE group, the home cage (65 × 75 × 25 cm) contained small toys, running wheels, climbing ladders, and plastic tunnels and tubes. To maintain environmental novelty, the objects were often changed. In the standard environment (SE) group, the home cage (21 × 27 × 16 cm) had no objects. Mice were housed in their SE or EE cages for 28 days before the assessment was conducted.

Novel object recognition (NOR) test

The mice were placed in the testing cage before the experiments to habituate. During the training phase, an individual mouse was placed in the centre of the apparatus. The mouse was returned to the home cage after 4 min of free exploration. Then, 2 h or 24 h later, the mouse was exposed to a novel object and an identical copy of the object. Animal's behaviour was video recorded and analysed.

Y maze test

Spontaneous alternation and novel arm exploration in a Y-maze (SD Instruments, San Diego, CA) were used to evaluate the learning and memory function. The experimental procedures were conducted as reported in the literature [31].

Morris water maze

This test was established in a quiet room (background noise below 20 dB). All tests were conducted under indirect illumination of 20 lux. The maze consisted of a circular pool with a diameter of 120 cm and 30 cm deep. Each quadrant's midpoint was used as a mouse water entry point. Three signs with different shapes and colours were placed in each quadrant lacking the escape platform to serve as the reference points. The escape platform quadrant lacked the sign. The mice were acclimated to the water maze room the night before the experiment. During the training, the mouse was randomly placed into any of the 3 quadrants lacking the escape platform. After the mouse stayed on the escape platform for 15 seconds, the mouse was removed from the platform and placed into the next entry quadrant. Each mouse was trained in a different orientation once a day. The video recording determined escape latency and the mouse movement trajectory. After each training, the mice were dried and placed under a 150W incandescent lamp for 3-5 min. The platform was removed on the fifth day of the experiment. The area of the platform was marked on the bottom of the maze with black tape. The mouse was placed into the pool from the bottom left quadrant (farthest quadrant) from where the platform was present during the training. The 60s video of the mouse movement was recorded. The average swim speed, time spent in the escape quadrant, and crossings of the escape platform area were then determined.

Magnetic resonance imaging

The magnetic resonance imaging (MRI) was conducted as reported previously [9], using the MRI equipment (GE Healthcare). Each mouse was anesthetized

by an intraperitoneal injection of 20 mg/kg xylazine 20 mg/kg and 50 mg/kg ketamine, and placed in a supine position. The same parameters were used for the scanning for all the mice.

Experimental endpoint and tissue collection

At the endpoint, mice were euthanized by CO₂ inhalation with secondary cervical dislocation. After decapitation, the brains were carefully removed from the skull, then was cut longitudinally. The hippocampus tissue complexes were harvested by spatula. The hippocampus tissues intended for Western blotting were lysed with radioimmunoprecipitation assay buffer with 1 : 100 of Halt Protease and Phosphatase Inhibitor Cocktail (78440, Thermo Fisher Scientific) and stored until use at -80°C. The tissues intended for RNA extraction were preserved with RNAlater solution at -80°C until RNA isolation.

Western blotting

A BCA kit was employed to determine the total protein concentrations. Protein lysate (50 µg) was subjected to SDS-PAGE, which was further transferred to PVDF membranes (EMD Millipore) as a routine procedure. After blocking, the membranes were incubated with the corresponding primary antibody, including vascular endothelial growth factor (VEGF, Santa Cruz, Dallas, TX), CD31 (Thermo Fisher), N-cadherin (Thermo Fisher), angiopoietin-1 (Ang-1, Thermo Fisher), brain-derived neurotrophic factor (BDNF, Abcam, Cambridge, MA),

β-actin (C4) (Santa Cruz), post-synaptic density protein 95 (PSD-95, Santa Cruz) overnight at 4°C.

RNA isolation and quantitative RT-PCR (qRT-PCR)

A RNeasy Plus Mini Kit (Qiagen, Germantown, MD) was employed to extract the total RNA. Power SYBR Green Master Mix (Thermo Fisher Scientific) was used to detect the gene expression. Gene expression was normalized to the expression of GAPDH, and was calculated using the 2^{-ΔΔCT} method.

Statistics

The data were presented as mean and standard deviation (SD). Statistical analysis was performed by GraphPad Prism software. Two-way ANOVA followed Tukey's multiple comparisons test, and one-way ANOVA followed Dunn's multiple comparisons test were employed for the analysis.

Results

Enriched environment ameliorated sevoflurane-induced recognition memory impairments of aged mice

The sevoflurane exposure has significantly reduced NOR scores at 2 h (Fig. 1A) and 24 h (Fig. 1B) after exposure to the new object at the indicated experimental time points compared to control mice housed in standard and enriched environments without sevoflurane exposure. The EE has significantly increased the novel object rec-

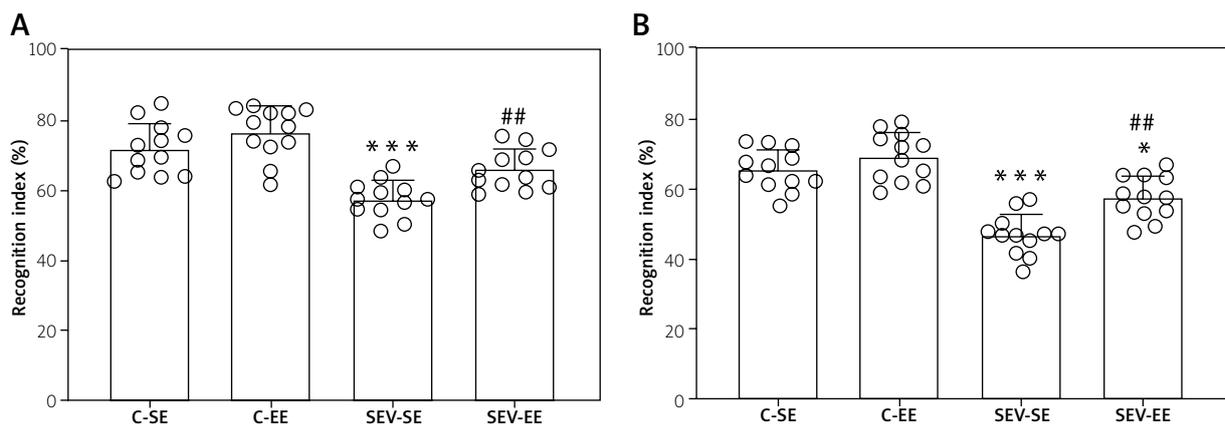


Fig. 1. Enriched environment ameliorated sevoflurane-induced recognition memory impairments of aged mice. Novel object recognition (NOR) tasks were carried out 2 h (A) and 24 h (B) after exposure to objects. The recognition index was calculated as the proportion of time with the target or novel object out of the total time. Data were presented as mean ± SD. **p* < 0.05, ****p* < 0.001 compared to C-SE, ***p* < 0.01 compared to SEV-SE. One-way ANOVA followed Dunn's multiple comparisons test. *N* = 12 mice for each group.

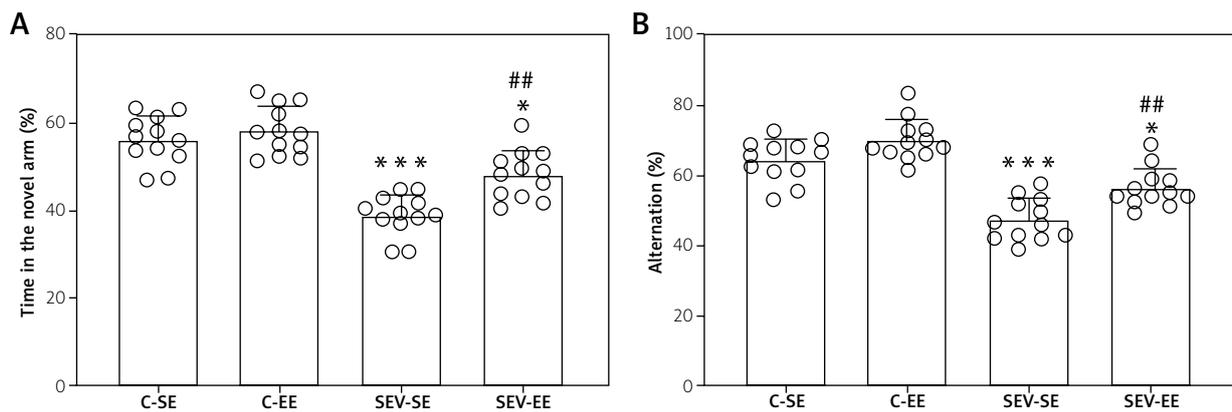


Fig. 2. Enriched environment ameliorated sevoflurane-induced learning and memory function impairments of aged mice. Time spent in the novel arm (A) and spontaneous alternation (B) in the Y-maze test. Data were presented as mean \pm SD. * $p < 0.05$, *** $p < 0.001$ compared to C-SE, ## $p < 0.01$ compared to SEV-SE. One-way ANOVA followed Dunn's multiple comparisons test. $N = 12$ mice for each group.

ognition in sevoflurane-exposed mice in the enriched environment (SEV-EE) compared to sevoflurane-exposed mice in the standard environment (SEV-SE). No significant differences were identified between mice in SE and EE without sevoflurane exposure, $p > 0.05$. Interestingly, however, there was a slight tendency to have an increased score in control mice without sevoflurane exposure in an EE environment. This tendency was observed in several subsequent experiments.

Enriched environment ameliorated sevoflurane-induced learning memory impairments of aged mice

The sevoflurane exposure had negative effects on both learning and memory function with the time spent in the novel arm of the Y-maze (Fig. 2A) and spontaneous alterations between the maze arms (Fig. 2B), which were significantly decreased when compared to controls. The SEV-EE mice demonstrated significantly ($p < 0.01$) higher scores for the same experiment. An enriched environment has benefits for reducing the deficit of learning and memory function caused by sevoflurane, as evidenced by the results of the Y-maze test.

An enriched environment improved cognitive recovery and the spatial memory of sevoflurane-exposed aged mice

Compared to control mice, sevoflurane exposure has significantly increased escape latency in the training sessions (Fig. 3A). The EE, however, significantly decreased the escape latency curve to nearly normal values by day 4 of the training. No noticeable differ-

ence was found in the escape latency curve in the control mice without sevoflurane exposure. Interestingly the mice's average swimming speed was also affected by sevoflurane exposure (Fig. 3B), while sevoflurane is not reported to have substantial lasting effects on muscle strength or physical endurance. However, reduced cognitive function and overall animal confusion on the direction of swimming may have affected this result as SEV-EE mice have demonstrated significantly ($p < 0.01$) improved performance in the swimming speed. The effects of sevoflurane on spatial memory are even more apparent, with SEV-SE having a significant ($p < 0.001$) decrease in mice reaching the water maze area where the escape platform is expected to be (Fig. 3C) and a similar decline in the time spend in the quadrant of the escape platform indicating a deficit in spatial learning and memory (Fig. 3D). Consistent with other observations of beneficial effects of the environmental enrichment, the SEV-EE mice demonstrated significantly improved performance in reaching the escape platform area and spent significantly more time in the escape platform quadrant.

An enriched environment ameliorates sevoflurane exposure-induced reduction in the brain blood flow in the brain

Magnetic resonance imaging analysis of the brain circulation indicated a significant reduction ($p < 0.001$) in CBF of sevoflurane-exposed mice maintained in the standard environment (Fig. 4A) in both right (Fig. 4B) and left hemispheres (Fig. 4C). The mice, after sevoflurane exposure, habituated to the enriched environment and demonstrated a significant ($p < 0.05$) increase in brain circulation.

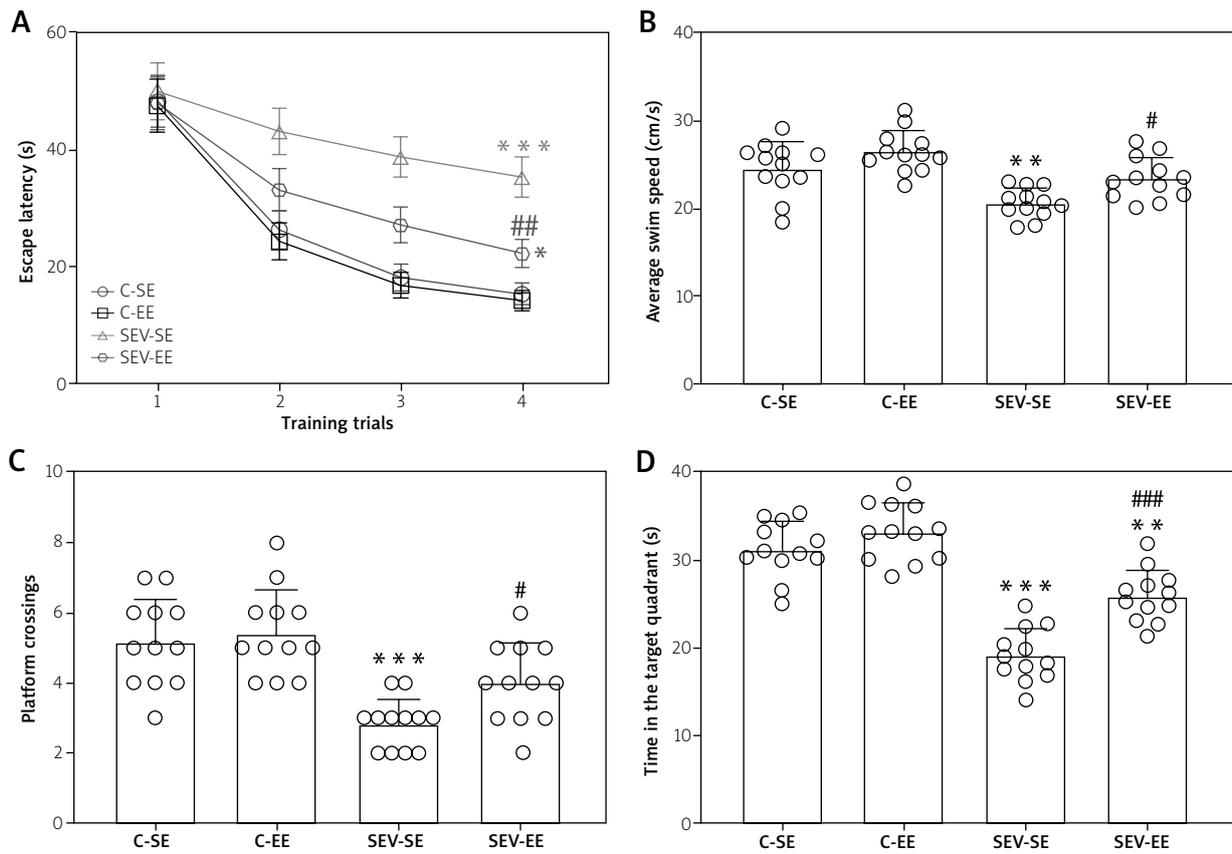


Fig. 3. By the Morris Water Maze test, an enriched environment promoted cognitive recovery in aged mice under sevoflurane anaesthesia. In 4 training sessions, the mouse escape latencies (A) and the average swim speed (B) were recorded. The probe trial recorded the number of platform site crossings (C) and the time in the target quadrant in 60 s (D). $N = 12$ mice for each group. Data were presented as mean \pm SD. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$ compared to C-SE, # $p < 0.05$, ## $p < 0.01$, ### $p < 0.001$ compared to SEV-SE. Two-way ANOVA followed Tukey's multiple comparisons test, and One-way ANOVA followed Dunn's multiple comparisons test.

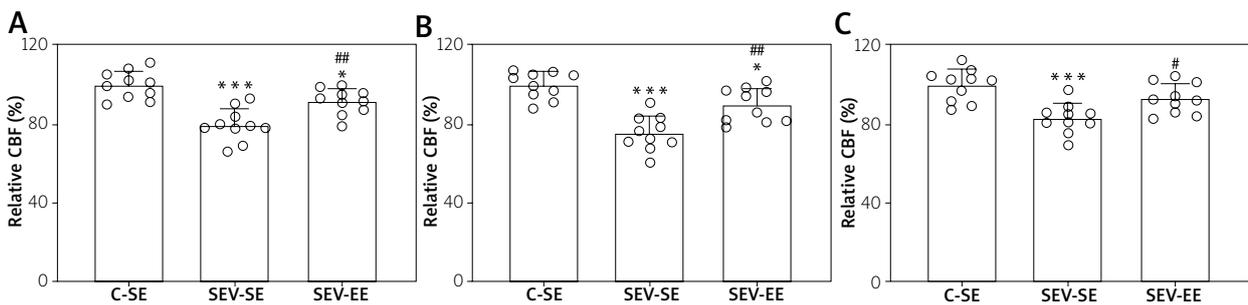


Fig. 4. An enriched environment promoted cerebral blood flow in aged mice under sevoflurane anaesthesia. The relative cerebral blood flows from the whole brain (A), right hemisphere (B) and left hemisphere (C) were normalized to C-SE. $N = 10$ mice for each group. Data were presented as mean \pm SD. * $p < 0.05$, *** $p < 0.001$ compared to C-SE, # $p < 0.05$, ## $p < 0.01$ compared to SEV-SE. One-way ANOVA followed Dunn's multiple comparisons test.

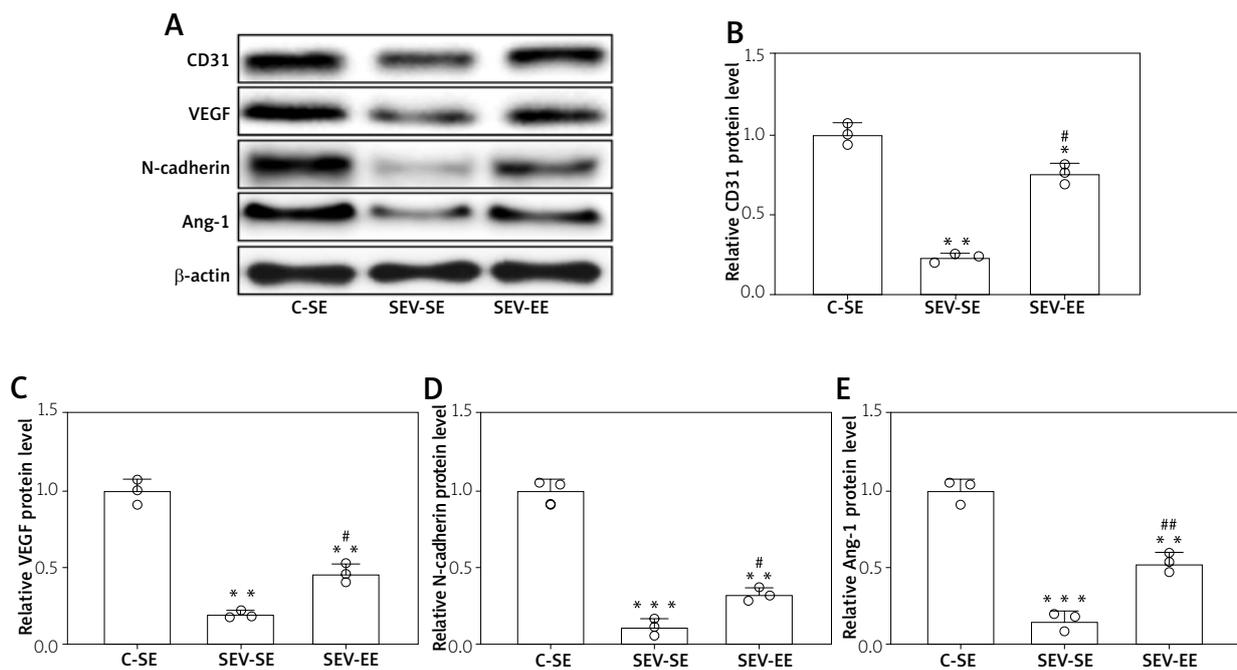


Fig. 5. An enriched environment promoted the expressions of proangiogenic factors in the hippocampi of aged mice under sevoflurane anaesthesia. Western blotting showed protein levels of CD31, VEGF, N-cadherin and Ang-1 from different groups (A). β -actin was used as a loading control, and the expressions were normalized to C-SE (B-E). $N = 3$ repeats for each group (10 hippocampus homogenates were mixed for each group). Data were presented as mean \pm SD. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$ compared to C-SE, # $p < 0.05$, ## $p < 0.01$ compared to SEV-SE. One-way ANOVA followed Dunn's multiple comparisons test.

An enriched environment improves sevoflurane exposure and reduces angiogenic factors and N-cadherin expression

The Western blotting analysis of the hippocampus tissues indicates that sevoflurane exposure significantly reduced the expression of CD31 (Fig. 5A, B), VEGF (Fig. 5A, C), N-cadherin (Fig. 5A, D) and angiotensin-1 (Fig. 5A, E). The enriched environment significantly ameliorated the decrease in expression of these factors. These results indicate that sevoflurane exposure reduces angiogenic factors in the hippocampus of aged mice, consistent with the observed reduced brain circulation in sevoflurane-exposed mice *in vivo*.

An enriched environment ameliorates sevoflurane exposure-induced decrease in BDNF and PSD-95

Western blotting of the hippocampus tissues also revealed that sevoflurane exposure in aged mice habituated in a standard environment induces a significant reduction in the expression of PSD-95 ($p < 0.001$, Fig. 6A, B) and neurotrophic BDNF (Fig. 6A, C). RT-PCR

further confirmed these results on the RNA transcription level and was fully consistent with the protein level observation, with both *Psd95* and *Bdnf* transcripts significantly reduced (Fig. 6E). The mice exposed to sevoflurane but habituated in the enriched environment cages demonstrated an ameliorative effect on PSD95 and BDNF expression.

Discussion

Our study confirms the cognitive deficit and neurological changes in the brains of the aged mice after prolonged exposure to sevoflurane. The EE is found to have an ameliorative effect on both behavioural performances, brain circulation, and the expression of the angiogenic and neurotrophic factors PSD95 and BDNF in the hippocampus of sevoflurane-exposed animals. As a structural scaffolding protein, PSD95 modulates the synaptic localization of various signalling proteins, channels and receptors [8]. The dysfunction of PSD95 was implicated in many neurological disorders, such as Alzheimer's disease [20], and schizophrenia [3]. Indeed, our behavioural experiment observation indicates similarities in the reduced cognitive performance of the sevoflurane-exposed mice to early dementia in the Alz-

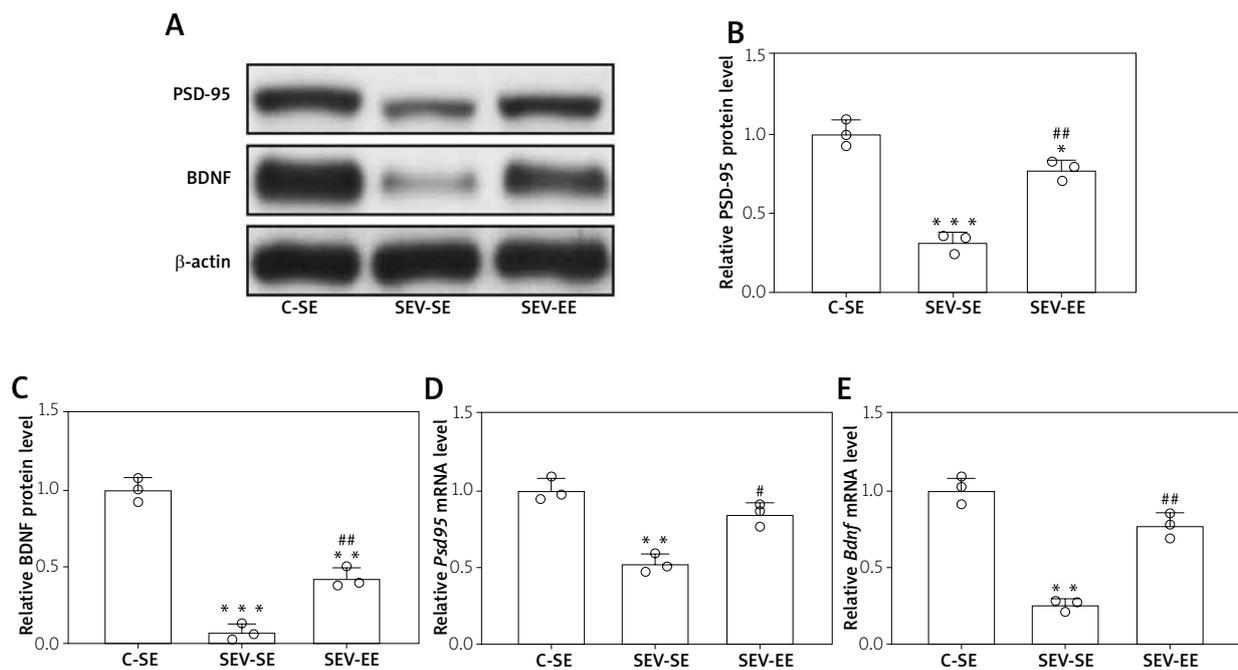


Fig. 6. An enriched environment promoted the expressions of postsynaptic density-95 (PSD-95) and brain-derived neurotrophic factor (BDNF) in the hippocampi of aged mice under sevoflurane anaesthesia. Western blotting showed protein levels of PSD-95 and BDNF from different groups (A). β -actin was used as a loading control, and the expressions were normalized to C-SE (B, C). The mRNA levels of *Psd95* (D) and *Bdnf* (E) in the hippocampus of aged mice after sevoflurane anaesthesia were tested by RT-PCR. $N = 3$ repeats for each group (10 hippocampus homogenates were mixed for each group). Data were presented as mean \pm SD. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$ compared to C-SE, # $p < 0.05$, ## $p < 0.01$ compared to SEV-SE. One-way ANOVA followed Dunn's multiple comparisons test.

heimer's disease model animals such as 3xTg-AD mice [19]. The decrease in the expression of BDNF indicates a reduced neuronal function and potential cell death in the hippocampus of the sevoflurane-exposed mice. The neuronal cell death was confirmed in sevoflurane-exposed animals [30], and correlated well with the clinical reports on memory and learning deficits observed in patients [22].

There are several potential mechanisms of neuronal loss due to sevoflurane exposure. The increase of pro-inflammatory cytokine and microglia activation were recorded in animal studies of sevoflurane exposure. The microglia inhibitor minocycline demonstrated ameliorative effects on sevoflurane-induced cognitive impairment, associating microglia activation with the observed cognitive deficits [7].

The initial studies on the cerebral blood during sevoflurane anaesthesia indicate decreased oxygen consumption and increased cerebral blood flow, leading to the expectation of neuroprotective properties of sevoflurane [14]. However, the subsequent subacute neuroinflammation induced by sevoflurane microves-

sel damage by the microglia cytokine release and local perivascular brain oedema may contribute to the reduced blood perfusion through the brain of the affected animals we observed in the MRI blood flow evaluation. *In vitro* studies indicated that sevoflurane had an inhibitory effect on the proangiogenic factors, including VEGF [25]. The reduced regulation of the microvasculature network in the brain may further contribute to neuronal hypoxia and death.

Recent studies demonstrated that EE exhibited anti-inflammatory effects in the brains [29]. Enriched environment ameliorated lipopolysaccharide-induced impairments of spatial learning and memory [11], the over-production of inflammatory cytokines, and brain infection with the influenza virus [10]. The molecular mechanisms of EE on neuroinflammation are not entirely understood. However, reports showed that EE chronically enhanced β adrenergic signalling, then reduced the microglia activation [28]. Neuromodulatory effects on circulating immune cells such as T lymphocytes and NK cells were also reported [17]. Furthermore, EE modulated the brain plasticity in Alzheimer's

disease model animals [6]. Consistent with our Western blotting results, EE successfully upregulated several neurotrophic signalling pathways [6]. These neurotrophic factors are also associated with the increase in cAMP response element-binding (CREB) transcripts, which is believed to be a critical step for the initiation of learning and memory [23]. These reports correlate well with the observed increase in learning and spatial memory performance of sevoflurane-exposed animals habituated in the EE in the current study.

Our study has several limitations. First, the pre-synaptic events have not been explored under EE treatment. Second, electrophysiological recording has not been employed to verify the effects of EE on the synaptic transmission.

Conclusions

Our findings suggest that sevoflurane should be used with caution and that prolonged sevoflurane anaesthesia should be avoided, especially in geriatric patients or patients with underlying neurological or psychiatric disorders. Patients who undertook sevoflurane anaesthesia should be followed up with a neurological assessment, and the cognitive deficit symptoms after sevoflurane anaesthesia should be managed as mild neuroinflammation. EE can be an efficient therapeutic strategy to ameliorate or mitigate the negative effects of sevoflurane exposure.

Disclosure

The authors report no conflict of interest.

References

- Bittner EA, Yue Y, Xie Z. Brief review: anesthetic neurotoxicity in the elderly, cognitive dysfunction and Alzheimer's disease. *Can J Anaesth* 2011; 58: 216-223.
- Brioni JD, Varughese S, Ahmed R, Bein B. A clinical review of inhalation anesthesia with sevoflurane: from early research to emerging topics. *J Anesth* 2017; 31: 764-778.
- Coley AA, Gao WJ. PSD95: A synaptic protein implicated in schizophrenia or autism? *Prog Neuropsychopharmacol Biol Psychiatry* 2018; 82: 187-194.
- Cui RS, Wang K, Wang ZL. Sevoflurane anesthesia alters cognitive function by activating inflammation and cell death in rats. *Exp Ther Med* 2018; 15: 4127-4130.
- Han Y, Yuan M, Guo YS, Shen XY, Gao ZK, Bi X. The role of enriched environment in neural development and repair. *Front Cell Neurosci* 2022; 16: 890666.
- Hu YS, Long N, Pigino G, Brady ST, Lazarov O. Molecular mechanisms of environmental enrichment: impairments in Akt/GSK-3beta, neurotrophin-3 and CREB signaling. *PLoS One* 2013; 8: e64460.
- Huang X, Ying J, Yang D, Fang P, Wang X, Zhou B, Zhang L, Fang Y, Yu W, Liu X, Zhen Q, Hua F. The Mechanisms of sevoflurane-induced neuroinflammation. *Front Aging Neurosci* 2021; 13: 717745.
- Jeong J, Pandey S, Li Y, Badger JD, 2nd, Lu W, Roche KW. PSD-95 binding dynamically regulates NLGN1 trafficking and function. *Proc Natl Acad Sci U S A* 2019; 116: 12035-12044.
- Jing Z, Shi C, Zhu L, Xiang Y, Chen P, Xiong Z, Li W, Ruan Y, Huang L. Chronic cerebral hypoperfusion induces vascular plasticity and hemodynamics but also neuronal degeneration and cognitive impairment. *J Cereb Blood Flow Metab* 2015; 35: 1249-1259.
- Jurgens HA, Johnson RW. Environmental enrichment attenuates hippocampal neuroinflammation and improves cognitive function during influenza infection. *Brain Behav Immun* 2012; 26: 1006-1016.
- Keymoradzadeh A, Hedayati Ch M, Abedinzade M, Gazor R, Rostampour M, Taleghani BK. Enriched environment effect on lipopolysaccharide-induced spatial learning, memory impairment and hippocampal inflammatory cytokine levels in male rats. *Behav Brain Res* 2020; 394: 112814.
- Li F, Yuan Y. Meta-analysis of the cardioprotective effect of sevoflurane versus propofol during cardiac surgery. *BMC Anesthesiol* 2015; 15: 128.
- Li J, Yuan T, Zhao X, Lv GY, Liu HQ. Protective effects of sevoflurane in hepatic ischemia-reperfusion injury. *Int J Immunopathol Pharmacol* 2016; 29: 300-307.
- Oshima T, Karasawa F, Okazaki Y, Wada H, Satoh T. Effects of sevoflurane on cerebral blood flow and cerebral metabolic rate of oxygen in human beings: a comparison with isoflurane. *Eur J Anaesthesiol* 2003; 20: 543-547.
- Raz L, Bhaskar K, Weaver J, Marini S, Zhang Q, Thompson JF, Espinoza C, Iqbal S, Maphis NM, Weston L, Sillerud LO, Caprihan A, Pesko JC, Erhardt EB, Rosenberg GA. Hypoxia promotes tau hyperphosphorylation with associated neuropathology in vascular dysfunction. *Neurobiol Dis* 2019; 126: 124-136.
- Reinsfelt B, Westerlind A, Ricksten SE. The effects of sevoflurane on cerebral blood flow autoregulation and flow-metabolism coupling during cardiopulmonary bypass. *Acta Anaesthesiol Scand* 2011; 55: 118-123.
- Singhal G, Jaehne EJ, Corrigan F, Baune BT. Cellular and molecular mechanisms of immunomodulation in the brain through environmental enrichment. *Front Cell Neurosci* 2014; 8: 97.
- Steinmetz J, Christensen KB, Lund T, Lohse N, Rasmussen LS, Group I. Long-term consequences of postoperative cognitive dysfunction. *Anesthesiology* 2009; 110: 548-555.
- Stover KR, Campbell MA, Van Winssen CM, Brown RE. Early detection of cognitive deficits in the 3xTg-AD mouse model of Alzheimer's disease. *Behav Brain Res* 2015; 289: 29-38.
- Sultana R, Banks WA, Butterfield DA. Decreased levels of PSD95 and two associated proteins and increased levels of BCL2 and caspase 3 in hippocampus from subjects with amnesic mild cognitive impairment: Insights into their potential roles for loss of synapses and memory, accumulation of Abeta, and neurodegeneration in a prodromal stage of Alzheimer's disease. *J Neurosci Res* 2010; 88: 469-477.
- Tian Y, Guo S, Wu X, Ma L, Zhao X. Minocycline alleviates sevoflurane-induced cognitive impairment in aged rats. *Cell Mol Neurobiol* 2015; 35: 585-594.
- Wang CM, Chen WC, Zhang Y, Lin S, He HF. Update on the mechanism and treatment of sevoflurane-induced postoperative cognitive dysfunction. *Front Aging Neurosci* 2021; 13: 702231.
- Wang H, Xu J, Lazarovici P, Quirion R, Zheng W. cAMP Response element-binding protein (CREB): A possible signaling molecule link in the pathophysiology of schizophrenia. *Front Mol Neurosci* 2018; 11: 255.

24. Wang X, Meng ZX, Chen YZ, Li YP, Zhou HY, Yang M, Zhao TT, Gong YL, Wu Y, Liu T. Enriched environment enhances histone acetylation of NMDA receptor in the hippocampus and improves cognitive dysfunction in aged mice. *Neural Regen Res* 2020; 15: 2327-2334.
25. Wang X, Yao Y, Gao J. Sevoflurane inhibits growth factor-induced angiogenesis through suppressing Rac1/paxillin/FAK and Ras/Akt/mTOR. *Future Oncol* 2020; 16: 1619-1627.
26. Wang Y, Wu J, Xie H, Wang L, Wang C, Wu Y. Enriched environment promotes cognitive function recovery following cerebral ischemic injury via upregulating GABAergic and glutamatergic systems in the contralateral hippocampus. *Evid Based Complement Alternat Med* 2020; 2020: 8850119.
27. Xie H, Zhang Q, Zhou N, Li C, Yu K, Liu G, Wu J, Jiang C, Hu R, Wu Y. Environmental enrichment enhances post-ischemic cerebral blood flow and functional hyperemia in the ipsilesional somatosensory cortex. *Brain Res Bull* 2020; 160: 91-97.
28. Xu H, Rajsombath MM, Weikop P, Selkoe DJ. Enriched environment enhances beta-adrenergic signaling to prevent microglia inflammation by amyloid-beta. *EMBO Mol Med* 2018; 10: e17061.
29. Zhou HY, Huai YP, Jin X, Yan P, Tang XJ, Wang JY, Shi N, Niu M, Meng ZX, Wang X. An enriched environment reduces hippocampal inflammatory response and improves cognitive function in a mouse model of stroke. *Neural Regen Res* 2022; 17: 2497-2503.
30. Zhou X, Li W, Chen X, Yang X, Zhou Z, Lu D, Feng X. Dose-dependent effects of sevoflurane exposure during early lifetime on apoptosis in hippocampus and neurocognitive outcomes in Sprague-Dawley rats. *Int J Physiol Pathophysiol Pharmacol* 2016; 8: 111-119.
31. Zhu Y, Gao M, Huang H, Gao SH, Liao LY, Tao Y, Cheng H, Gao CY. p75NTR Ectodomain ameliorates cognitive deficits and pathologies in a rapid eye movement sleep deprivation mice model. *Neuroscience* 2022; 496: 27-37.