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GCN5L1 Regulates Anaesthesia-Induced Nerve Injury in Newborn Through FoxO1 by Oxidation Effects

Hui Li, Xiao-ling Jiao, Jing-jing Ge and Yan-jie Ding*

Department of Pediatrics, Yan Tai Yu Huang-ding Hospital, Yan Tai, Shan Dong Province, 264001, China

KEYWORDS Anaesthesia. ForkheadBox O1. General Control of Amino Acid. Synthesis 5. Nerve Injury. Newborn. Oxidative Stress

ABSTRACT The researchers of the present study aimed to explore the effects and mechanism of general control of amino acid synthesis 5 like-1 (GCN5L1) in the anaesthesia-induced nerve injury model. In brief, the mRNA and protein of GCN5L1 expression was up-regulated in the anaesthesia-induced nerve injury model. GCN5L1 protein was increased in anaesthesia-induced nerve injury by inducing oxidative stress. Meanwhile, over-expression of GCN5L1 promoted oxidative stress in vitro. Down-regulation of GCN5L1 inhibited oxidative stress in nerve cells. GCN5L1 suppressed the protein expression of forkhead box O1 (FoxO1) in vivo or in vitro. The activation of FoxO1 attenuated the effects of GCN5L1 on oxidative stress in vitro. Finally, FoxO1 protein attenuated the effects of GCN5L protein on induced nerve injury via suppression of nerve apoptosis and oxidative stress in vivo. The researchers concluded that GCN5L1 regulated anaesthesia-induced nerve injury in newborns through FoxO1 via oxidation effects.

INTRODUCTION

In paediatric and obstetric patients, the number and duration of anaesthesia procedures and the complexity of surgical procedures have increased. People have begun to pay attention to the effects or risks of anaesthetics on brain development. A large number of infants and young children undergo anaesthesia every year in China (Goyagi 2019). Thousands of parents are concerned about whether surgical trauma and anaesthetics will affect the growth and development of children (Goyagi 2019). With the development of modern science and technology, continuous progress has also been made in medical technology (Pietraszek 2018). The investigations into the mechanism of anaesthetics have prompted the constant improvement of anaesthetics, improving the effectiveness while reducing the damage to the body (Han et al. 2015). Recent clinical studies and animal experiments have suggested the correlation between the commonly used anaesthetics in current clinical practice and the post-operative cognitive dysfunction in patients undergoing surgery and anaesthesia (Chen et al. 2016b; Han et al. 2015).

Every year, tens of millions of infants and young children worldwide receive general anesthesia because of different diseases or maternal needs during pregnancy (Goyagi 2019). Whether early exposure to general anesthesia impairs children's neurodevelopment and even causes long-term intellectual cognitive impairment has become one of the focuses of attention in the fields of anesthesiology, neuroscience, and pediatrics (Han 2018 et al; Goyagi 2019). As early as 2003, it was reported that early exposure to general anesthetics may lead to brain tissue degeneration. Long-term or repeated exposure to general anesthesia may affect children's neurodevelopment, pushing the research fever to the peak (Goyagi 2019). Later, more studies showed that sevoflurane could lead to abnormal apoptosis of brain neurons in the early stage, concentrated in the hippocampus, and even cause negative effects on long-term learning and cognition (Goyagi 2019). Because most infants and young children are operated under general anesthesia, it is of great significance to reduce and improve the neurotoxicity measures and drug research mediated by general anesthesia drugs (Han et al. 2015). However, due to ethical constraints and the complexity of human trials, clinical research is difficult (Han et al. 2015). At present, some progress has been made in the mechanism of sevoflurane

*Address for correspondence:
Yan-jie Ding,
No.20, Yu Huang-ding East Road,
Yan Tai Yu Huang-ding Hospital, Yan Tai,
Shan Dong Province, 264001, China
Tel: 86-0535669199983159
E-mail: ljbakuno1549@126.com

neurotoxicity in animal models, and a variety of corresponding drug intervention measures have been found. It is of great significance to the neuroprotection of children during perioperative period.

With anesthesiologists' in-depth research on patients' rapid recovery during perioperative period, it is very important to seek a perioperative management plan that can promote patients' faster recovery after operation (Goyagi 2019). At the same time, research shows that postoperative cognitive dysfunction (POCD) is one of the serious complications after general anesthesia, which significantly delays the recovery process after surgery and reduces the quality of life of patients (Goyagi 2019). Therefore, it is very important to select a reasonable and reliable anesthetic method to inhibit the stress response of surgery. At present, total intravenous anesthesia and combined intravenous and inhalation anesthesia are common general anesthesia methods in clinical practice (Han et al. 2015). The former only relies on intravenous anesthetics to maintain the depth of anesthesia, while the latter is combined with intravenous anesthetics and inhalation anesthetics to maintain the anesthesia effect (Han et al. 2015).

Damage to damaged neurons and glial cells will cause the release of structural proteins and metabolic enzymes in brain cells, resulting in oxidative stress damage. In addition, patients with intracranial hematoma and the generation of metabolites in the hematoma will also lead to oxidative stress damage (Li et al. 2016). Oxidative stress is continuously activated, which will lead to neurological function of patients (Li et al. 2016). ROS was initially considered to be a harmful metabolite. The increase of its level can cause DNA damage, protein damage, and ultimately lead to cell apoptosis. However, research has found that ROS is involved in regulating the activation of multiple intracellular signal transduction pathways, such as FoxO1. FoxO1 is not enough to resist the damage caused by oxidative stress.

Forkhead transcription factor of class O1 (FoxO1) is an important regulatory factor for metabolism in the body, and its activity is negatively regulated by insulin (Li et al. 2016). Overactivation of FoxO1 can increase the uptake and oxidation of fatty acids, and simultaneously inhibit the utilisation rate of glucose oxidation (Qin et al. 2010). In the case of excessive intake of fatty acid beyond the cellular metabolic capacity, mitochondrial dysfunction would occur, producing excessive reactive oxygen species (ROS) to further cause cell injury (Li et al. 2016; Ciccarone et al. 2019). Thus, FoxO1 can inhibit oxidative stress loss and neuronal apoptosis.

The weakened binding of general control of amino acid synthesis 5 like-1 (GCN5L1) to DNA can increase gene transcription. GCN5L1 can bind to E2FA and CYCLINE1 to subsequently regulate multiple cellular processes, including cell proliferation, differentiation, cell cycle, and repair of nerve injury (Scott et al. 2018; Lv et al. 2019).

Objectives

The researchers of the present study aimed to explore the effects and mechanism of GCN5L1 in anaesthesia-induced nerve injury newborn mice model.

MATERIAL AND METHODS

C57BL/6 newborn mice (postnatal days 7) were maintained at 23 °C ± 2 °C, with a 12-hour-light dark cycle. All mice were treated according to the guidelines of the Guide for the Care and Use of Laboratory Animals. The Laboratory Animal Care Committee of Yan Tai Yu Huang-ding Hospital approved all experimental procedures and protocols.

Management of Experimental Animals

Male C57BL/6 newborn mice were used in this study, and then randomly assigned into groups (n=8 mice/every group). Propofol-induced model group, all mice was administered with 90 mg/kg/every week of propofol for three weeks (Cui et al. 2011). After the 4th week, mice were executed for cognitive function testing.

Model + GCN5L1 group, anaesthesia-induced nerve injury of newborn mice was injected with 100 ng/kg of recombination GCN5L1 protein (IP).

Model + GCN5L1+ FoxO1 group, anaesthesia-induced nerve injury of newborn mice was injected with 100 ng/kg of recombination GCN5L1 protein and 1μg /kg of recombination FoxO1 (IP).

Determination of Cognitive Function

The *open field* test was executed to measure the emotional responses as described previously (Valentim et al. 2010). All mice were released in the centre of the arena (30 cm×30 cm×30 cm). Activity was assessed as the total distance of the rat travelled in 10 minutes. Then, the arena was cleaned with 75 percent alcohol to avoid the presence of olfactory cues.

The *fear conditioning* test was executed to measure as described previously (Li et al. 2014). All mice were exposed in the conditioning chamber for 3 minutes of accommodation and then one tone-foot-shock pairing was handed. Fear conditioning test was executed for 24 hours and then mice travelled by placing mice back to the same test chamber for 5 minutes. After 2 hours, all mice were placed in a novel chamber and the training tone was delivered for 3 minutes to evaluate tone fear conditioning. All mice were narcotised using 35 mg/kg pentobarbital sodium and sacrificed using decollation for another experiment.

Microarray Experiments

Microarray experiments were performed at the Genminix Informatics (China). Gene expression profiles were analysed with the Human Exon 1.0 ST GeneChip (Affymetrix).

Measurement of Oxidative Stress

Serum and tissue samples were collected at 2000 g for 10 minutes at 4 °C and was used to measure GSH, GSH-PX, SOD and MDA levels using GSH, GSH-PX, SOD and MDA kits (Nanjing JianchengBiological Engineering Research Institute Co. Ltd.). Absorbance at 450 nm was measured using a Multiskan Spectrum Microplate Spectrophotometer (ThermoScientific™, USA). ROS levels (S0033, Beyotime) were measured using ROS production levels kit.

Western Blot Analysis

Total proteins werecollected by using RIPA lysis buffer and protease inhibitor cocktail (1:100, Beyotime). 50 μg protein samples were loaded to 10 percent sodium dodecyl sulphate-polyacrylamide gel electrophoresis and then transferred onto polyvinyl difluoride (Thermo Scientific™, USA) membranes. Membranes was incubated with GCN5L1 (ab12188, 1:1000, Abcam), FoxO1 (2880, 1:1000, Santa Cruz Biotechnology) and GAPDH (sc-365062, 1:500, Santa Cruz Biotechnology) at 4°C overnight after blocking with 5 percent non-fat milk in tris-buffered saline with 0.1 percent tween 20 (TBST). The membrane was washed with TBST and then incubated with anti-rabbit secondary antibody (sc-2004, 1:5000, Santa Cruz Biotechnology) for 2 hours at room temperature. Immunoreactive bands were visualised using the ECL kit (Thermo Scientific™, USA) and integrated density of the bands was quantified by Quantity One software (Bio-Rad).

Cell Culture and Transfection

PC-12 cells were obtained from the Shanghai cell bank of Chinese Academy of Sciences and were maintained in RPMI 1640 medium (Hyclone, Logan, UT, USA) containing 10 percent foetal bovine serum (Hyclone) at 37°C with 5 percent CO_2 and 95 percent air. GCN5L1 plasmid, siGCN5L1 and FoxO1 plasmid were purchased from GenePharma Co. (Shanghai, China). These plasmid were transfected into PC-12 cells using Lipofectamine 2000 reagent (Invitrogen, Carlsbad, CA, USA) for 24 hours and Propofol (10 μg/mL) was applied to PC-12 transfection cells for 48 hours at 37 °C with 5 percent CO_2 and 95 percent air.

Statistical Analysis

Data were expressed as the mean ± standard deviations (SD). Comparisons between groups of independent samples were assessed by Student's t-test or one-way analysis of variance (ANOVA) and Tukey's post test. p values < 0.05 were considered to indicate statistical significance.

RESULTS

The Expression of GCN5L1 in Newborns in Anaesthesia-induced Nerve Injury Mouse Model

Firstly, to analyse expression changes of GCN5L1 in newborns of anaesthesia-induced cognitive dysfunction of mouse model, the researchers found that there was an induced escape la-

tency and increased mean path length after five days of training in mice from the model group (36±3.2 vs 65±3.5; 410±70 vs 1810 ± 110; all p <0.01, Fig. 1A-1C). Compared to sham control, the time in the target quadrant and the number of times of crossing the former platform location were both decreased in anaesthesia-induced mice (76±2.8 vs22±2.2;11.3±0.8vs2.26±0.5; all p <0.01, Fig. 1D-1E). MDA level was increased, while the levels of anti-oxidant factors (SOD, GSH and GSH-px) were decreased in newborns of anesthesia-induced nerve injury mouse model (17.66±0.72 vs 53.31±2.72; 11.37±0.57 vs 3.18±0.42; 72.67±5.67 vs 21.76±3.56; 29.89±1.35 vs 8.86±1.59; all p <0.01, Fig. 1F-1J). In addition, the mRNA and protein expression of GCN5L1 was expanded in mice with anaesthesia-induced nerve injury, in comparison with sham control group (1.00±0.68 vs 3.56±0.086; 1.00±0.074 vs 2.25±0.052; all p <0.01, Fig. 1K-1L).

GCN5L1 Regulated Oxidative Stress in Newborns of Anaesthesia-induced Nerve Injury Mouse Model

In order to explain the function of GCN5L1 in newborns of anaesthesia-induced nerve injury mouse model, the researchers analysed the effects of GCN5L1 on newborns of anaesthesia-induced nerve injury mouse model. As a result, GCN5L1 protein reduced escape latency and mean path length after five days of training in mice from the model group (68±3.2 vs 33±3.5; 1920±110 vs 690±150; all p <0.01, Fig. 2A-2C). In addition, GCN5L1 protein caused less time in the target quadrant and decreased the number of times of crossing the former platform location (25±2.8 vs 72±2.2; 2.1 ±0.8 vs 14.96± 0.9; all p <0.01, Fig. 2D-2E). GCN5L1 protein induced MDA levels, and reduced the levels of anti-oxidant factors in mice from the model group (11.66±0.52 vs

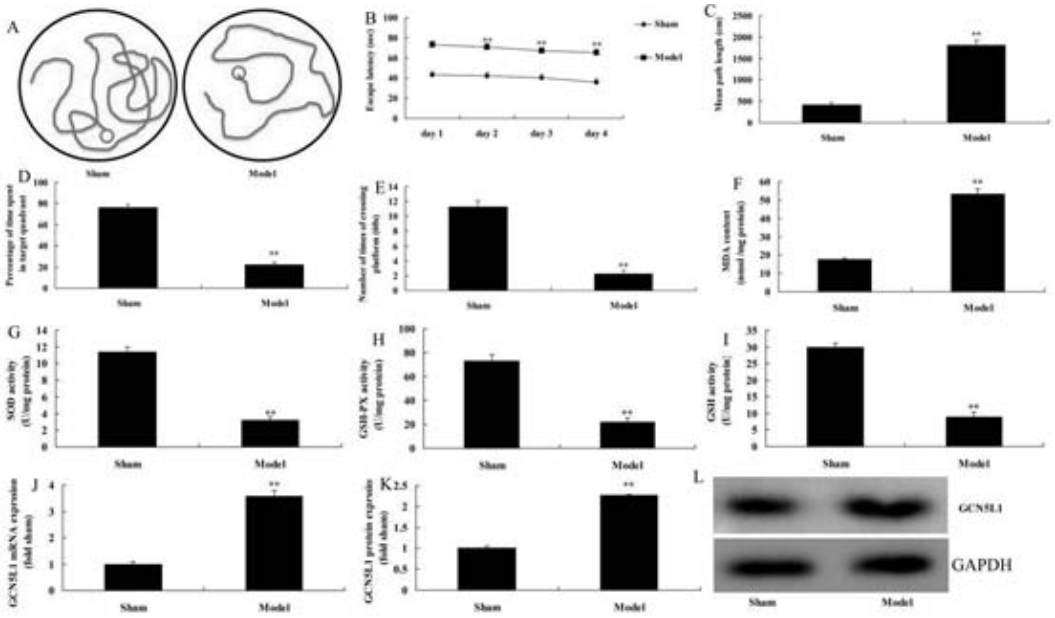

Fig. 1. The expression of GCN5L1 in anaesthesia-induced nerve injury in newborn of mice model
Record chart of determination of cognitive function (A), escape latency (B) and mean path length (C), spent less time in the target quadrant (D) the number of times the animals crossed the former platform location (E), MDA level (F), SOD level (G), GSH-px level (H), GSH level (I), GCN5L1 mRNA expression (J), GCN5L1 protein expression (K and L)
Sham, control sham group; Model, anaesthesia-induced Nerve injury of newborn mice model group
Data were expressed as the mean ± standard deviations (SD)
****p<0.01 versus sham group**

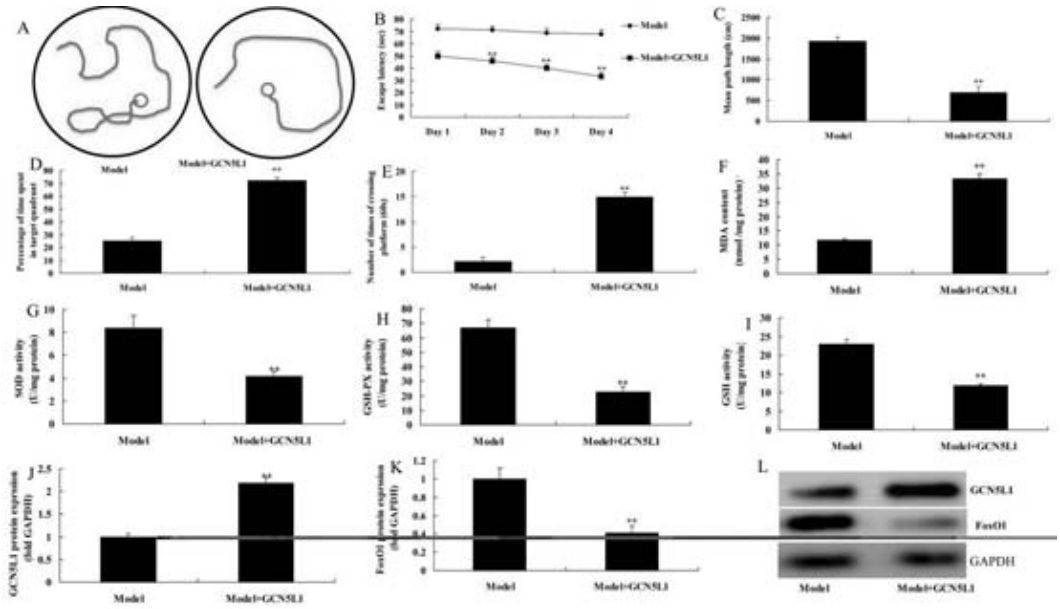

Fig. 2. GCN5L1 regulated oxidative stress in anaesthesia -induced nerve injury in newborn of mice model
Record chart of determination of cognitive function (A), escape latency (B) and mean path length (C), spent less time in the target quadrant (D) the number of times the animals crossed the former platform location (E), MDA level (F), SOD level (G), GSH-px level (H), GSH level (I), GCN5L1 and FoxO1 protein expression (J, K and L)
Model, anaesthesia-induced nerve injury of newborn mice model group; Model+ GCN5L1, anaesthesia-induced nerve injury of newborn mice with 100 ng of recombination GCN5L1 protein group. Data were expressed as the mean ± standard deviations (SD)
**p<0.01 versus anaesthesia-induced nerve injury of newborn mice model group

33.31±1.72; 8.37±1.07 vs 4.18±0.42; 66.67±5.67 vs 22.76±3.56; 22.89±1.35 vs 11.86±0.59; all p <0.01, Fig. 2F-2I). Meanwhile, the protein expression of GCN5L1 was induced while that of FoxO1 was suppressed in brain tissue of mice from GCN5L1 group (1.00±0.042 vs 2.32±0.074; 1.00± 0.68 vs 0.43±0.51; all p <0.01, Fig. 2J-2L).

GCN5L1 Regulated Oxidative Stress in Anaesthesia-induced Nerve Injury Model In Vitro

To identify the effects of GCN5L1 in oxidative stress in anaesthesia-induced nerve injury, GCN5L1 plasmid or si-GCN5L1 mimics was transfected to increase or decrease GCN5L1 mRNA expression in vitro, respectively (1.00±0.058 vs 11.85±0.52; 1.00±0.061 vs 0.39±0.071; all p <0.01, Fig.3A, 3H). Up-regulation of GCN5L1 enhanced ROS production levels and MDA levels, and reduced the levels of anti-oxidant factors in vitro, in comparison with negative group (1±0.06 vs 2.27±0.22; 16.66±1.12 vs 63.81±2.82; 7.37±0.57 vs 2.68±0.42; 49.67±2.67 vs 10.76±2.56; 39.89±7.86 vs 2.35±1.59; all p <0.01, Fig.3B-3G). Down-regulation of GCN5L1 weakened ROS production levels and MDA levels, and enhanced anti-oxidant factors in vitro (1±0.05 vs 0.42±0.07; 15.66±5.81 vs 1.12±0.82; 10.37±1.67 vs 60.18±4.12; 41.67±5.67 vs 252.76±18.56; 55.89±8.35 vs 194.86±12.59; all p <0.01, Fig. 3I-3N).

GCN5L1 Regulated FoxO1 in Anaesthesia-induced Nerve Injury

To further explore the reliable biological mechanism of GCN5L1 on oxidative stress in anaesthesia-induced nerve injury, a gene chip was used to analyse gene expression. FoxO1 may be a target for the effects of GCN5L1 in anaesthesia-

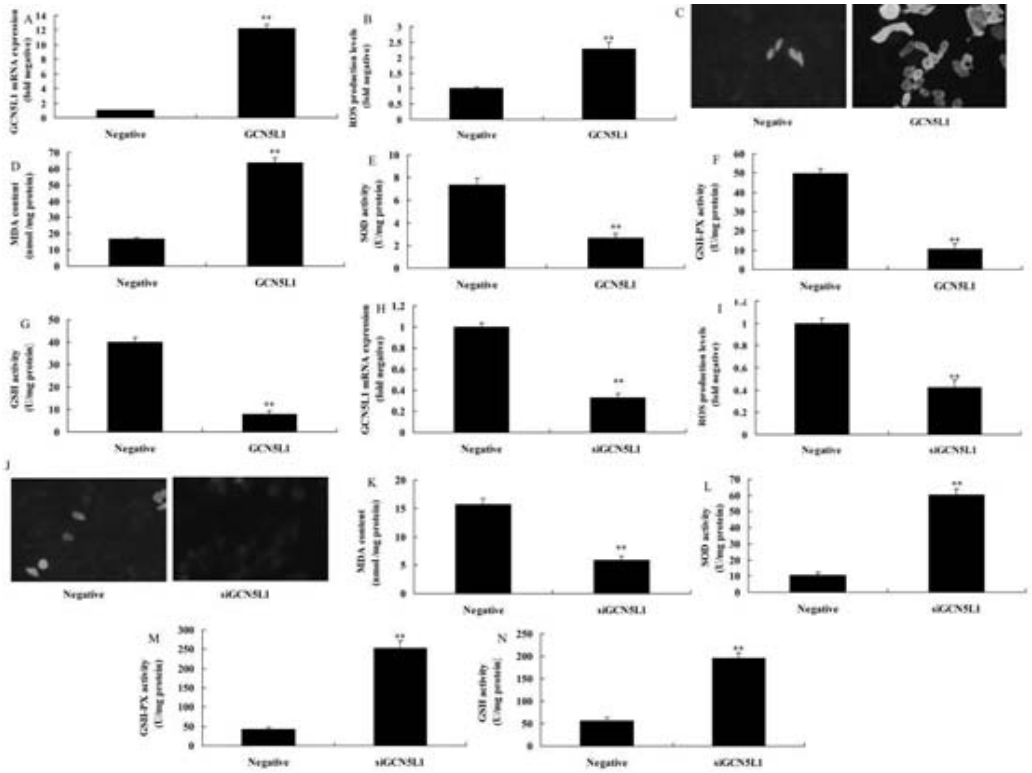

Fig. 3. GCN5L1 regulated oxidative stress in anaesthesia -induced nerve injury of in vitro model
GCN5L1 mRNA expression (A), ROS production (B and C), MDA level (D), SOD level (E), GSH-PX level (F), GSH level (G) by up-regulation of GCN5L1; GCN5L1 mRNA expression (H), ROS production (I and J), MDA level (K), SOD level (L), GSH-PX level (M), GSH level (N) by down-regulation of GCN5L1
Negative, negative mimics group; GCN5L1, up-regulation of GCN5L1 group; si GCN5L1, down-regulation of GCN5L1 group
Data were expressed as the mean ± standard deviations (SD)
****p<0.01 versus negative mimics group**

induced nerve injury (Fig. 4A). GCN5L1 sensitized GCN5L1 protein expression, and abatemented FoxO1 protein expression in vitro (1.00± 0.41 vs 3.02±0.087; 1.00±0.048 vs 0.28± 0.032; all p <0.01, Fig. 4B-4D). By contrast, down-regulation of GCN5L1 abatemented GCN5L1 protein expression, and sensitized FoxO1 protein expression in vitro (1.00±0.025 vs 0.39±0.036; 1.00±0. 034 vs 3.48±0.064; all p <0.01, Fig. 4E-4G).

FoxO1 lighten the effects of GCN5L1 on newborns of anaesthesia-induced nerve injury mouse model.

To confirm the role of FoxO1 on the effects of GCN5L1 on newborns of anaesthesia-induced nerve injury mouse model, FoxO1 recombinant protein (1 μg/kg, Sangon Biotech) was treated in newborns of anaesthesia-induced nerve injury mouse model by GCN5L1 protein. As a result, FoxO1 recombinant protein induced escape latency and increased mean path length after five days of training in model group treated with GCN5L1 protein, compared with those only treated with GCN5L1 protein (68±3.2 vs 36±3.5 vs 62±3.1; 1803±127 vs 623±88 vs 1151±55; all p <0.01, Fig. 5A-5C). FoxO1 recombinant protein reduced time spent in the target quadrant and decreased the number of times crossing the former platform location (24±2.1 vs 682.1±2.2 vs 42±2.7; 4.8±0.3 vs 12.6±0.6 vs 7.2±0.6; all p <0.01, Fig. 5D-5E). FoxO1 recombinant protein reduced MDA levels,

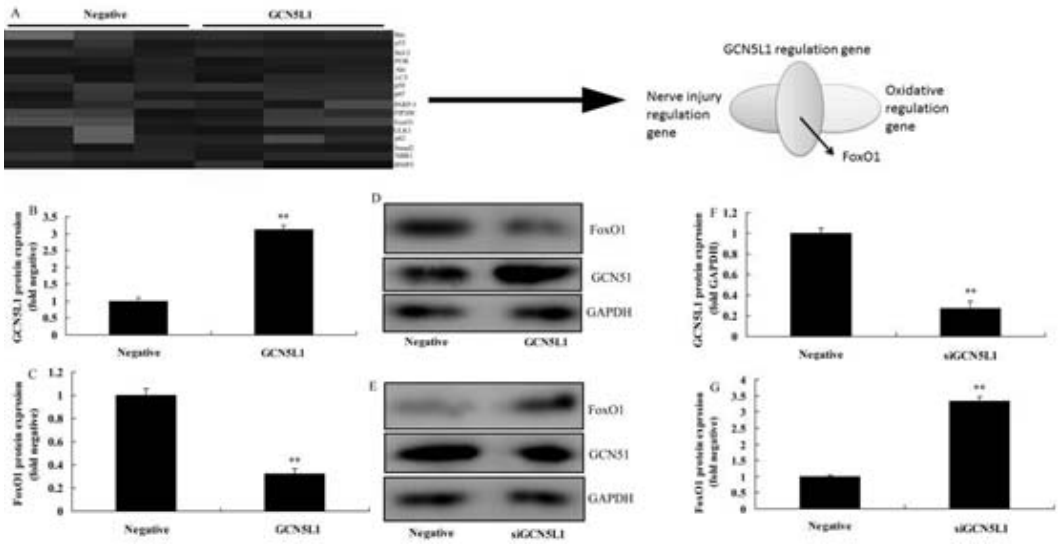

Fig. 4. GCN5L1 regulated FoxO1 in anaesthesia -induced nerve injury
Heat map (A), GCN5L1 and FoxO1 protein expression (B, C and D) by up-regulation of GCN5L1; GCN5L1 and FoxO1 protein expression (E, F and G) by down-regulation of GCN5L1
Negative, negative mimics group; GCN5L1, up-regulation of GCN5L1 group; si GCN5L1, down-regulation of GCN5L1 group. Data were expressed as the mean ± standard deviations (SD)
****p<0.01 versus negative mimics group**

and increased the levels of anti-oxidant factors in model group treated with GCN5L1 protein, compared with those only treated with GCN5L1 protein (13.66±1.12 vs 56.81±2.52 vs 27.52±2.52; 12.37±0.67 vs 3.18±0.52 vs 7.56±0.71; 37.67±1.67 vs 6.76±1.56 vs 22.99±1.68; 51.89±2.35 vs 12.86± 2.59 vs 30.95±2.36; all p <0.01, Fig.5F-5J). FoxO1 recombinant protein induced the protein expression of FoxO1 in brain tissue of model group treated with GCN5L1 protein, compared with those only treated with GCN5L1 protein (9.66± 0.12 vs 2.51±0.52 vs 5.52±0.32, p <0.01, Fig.5K-5L).

FoxO1 lighten the effects of GCN5L1 on anaesthesia-induced nerve injury in vitro.

FoxO1 plasmid also enhancedthe protein expression of FoxO1 in vitro by GCN5L1 (1±0.07vs 0.18±0.12 vs 0.56±0.11; p <0.01, Fig. 6A-6B). FoxO1 reduced ROS-induced oxidative stress in vitro by GCN5L1 (1±0.02 vs 2.4±0.1 vs 1.35±0.13; 13.66± 1.12 vs 56.81±2.52 vs 27.52±2.52 ; 12.37±0.67 vs 3.18±0.52 vs 7.56±0.71 ; 37.67±1.67 vs 6.76±1.56 vs 22.99±1.68; 51.89±2.35 vs 12.86±2.59 vs 30.95±2.36; all p <0.01, Fig. 6C-6H).

DISCUSSION

In consideration of poor tolerance and poor coordination during surgery in young children, general anaesthesia is required with effective and complete analgesia, fewer adverse reactions to attenuate the disturbance and impact on the paediatric physiological function and rapid postoperative wake up (Chen et al. 2016a). Clinical observations have revealed that exposure to propofol would cause anterograde amnesia within a certain period of consciousness recovery in patients (Chen et al. 2016a). Recent studies reported that anterograde amnesia is defined as the process of forgetting after the event that has caused the amnesia (Chen et al. 2016a; Huang et al. 2016). Total intravenous anesthesia is one of the commonly used general anesthesia methods for tracheal intubation in clinical practice. It is mainly used to maintain the depth of anesthesia required for surgery through intravenous injection or target-controlled infusion of anesthetic drugs (Chen et al. 2016a). It has the advantages of rapid onset, less irritation in

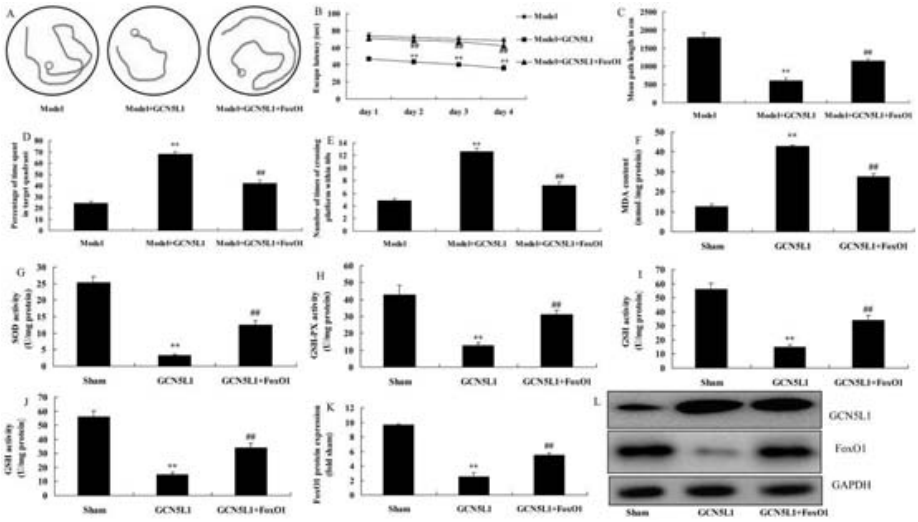

Fig. 5. The activation of FoxO1 reduced the effects of GCN5L1 on anaesthesia-induced nerve injury in newborn of mice model

Record chart of determination of cognitive function (A), escape latency (B) and mean path length (C), spent less time in the target quadrant (D) the number of times the animals crossed the former platform location (E), MDA level (F), SOD level (G), GSH-px level (H), GSH level (I), GCN5L1 and FoxO1 protein expression (J, K and L) Model, anaesthesia-induced nerve injury of newborn mice model group; GCN5L1, anaesthesia-induced nerve injury of newborn mice with GCN5L1 protein group; GCN5L1+ FoxO1, anaesthesia-induced nerve injury of newborn mice with GCN5L1 protein and FoxO1 protein group; Data were expressed as the mean ± standard deviations (SD)

**p<0.01 versus anaesthesia-induced nerve injury of newborn mice model group; $^{\#\#}$p<0.01 versus anaesthesia-induced nerve injury of newborn mice with GCN5L1 protein group

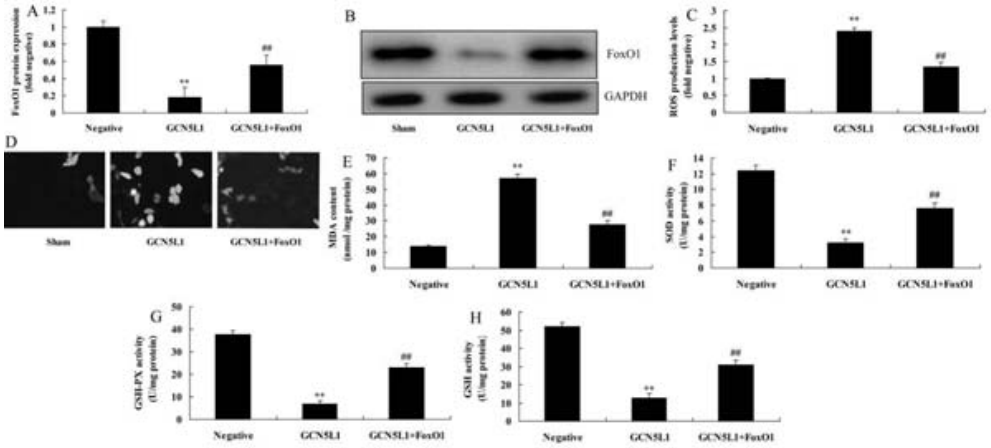

Fig. 6. The activation of FoxO1 reduced the effects of GCN5L1 on anaesthesia-induced nerve injury in in vitro model FoxO1 protein expression (A and B), ROS production (C and D), MDA level (E), SOD level (F), GSH-PX level (G), GSH level (H)

Negative, negative mimics group; GCN5L1, up-regulation of GCN5L1 group; GCN5L1+ FoxO1, up-regulation of GCN5L1 and FoxO1 group; Data were expressed as the mean ± standard deviations (SD)

**p<0.01 versus negative mimics group

the induction process, simple use method, and can achieve good anesthesia effect. However, some literatures have pointed out that only taking total intravenous anesthesia can increase the dosage of narcotic drugs, resulting in prolonged postoperative recovery time and increased risk of complications (Chen et al. 2016a). The apoptosis of neurons induced by sevoflurane is related to the apoptosis pathway of mitochondria and endoplasmic reticulum induced by stimulation. In addition, inhalation of anesthetics will break the calcium homeostasis of endoplasmic reticulum, increase intracellular and mitochondrial calcium ions, and ultimately lead to neuronal apoptosis. It can be seen that the decrease of neuron number at the peak stage of neural development will inevitably have a negative impact on long-term cognitive function. Oxidative stress causes different mechanisms of inflammation by activating various transcription factors, and destroys the integrity of cell membrane and cell function. Some studies have found that subclinical sevoflurane exposure will lead to the imbalance of free radical metabolism in newborn mice, resulting in the accumulation of reactive oxygen species and malondialdehyde, a by-product of oxidative stress in the plasma. 2.6 percent sevoflurane exposure will significantly inhibit the activity of antioxidant enzyme superoxide dismutase (SOD), resulting in a surge of malondialdehyde in the hippocampus of mice. Timely removal of these reactive oxygen species can reduce neuronal death. The inhibition of ERK phosphorylation caused by oxidative stress will affect the normal development of the nervous system. The above studies show that oxidative stress may be involved in sevoflurane-mediated neurotoxicity, and there is a certain concentration dependence. Timely removal of excessive oxygen free radicals is a potential treatment strategy.

This phenomenon suggests that propofol has a significant interference effect on the uptake and transmission of brain memory information (Yang et al. 2014). The findings demonstrated that GCN5L1 mRNA and protein expression increased anaesthesia-induced nerve injury in mice. Manning et al. (2019) showed that GCN5L1 restricts recovery from ischemia-reperfusion injury. Meanwhile, Wang et al. (2017) showed that oxidative stress is increased in GCN5L1 KO mice, which indicated these results existed some opposite, and it needs further research whether GCN5L1 regulates inflammation and its activity as an acetyltransferase. The researchers will further study whether GCN5L1 regulates inflammation.

Hypoxia is defined as a pathological process characterised by abnormal alterations in the metabolism, function and morphology in cells and tissues, caused by inadequate supply of oxygen and disordered use of oxygen (Lu et al. 2017; Guo et al. 2019). Severe or long-term hypoxia can cause serious harm to the body (Kalimeris et al. 2013). Short-term cerebral hypoxia can cause symptoms such as headache and restlessness, while severe hypoxia can induce brain edema, coma, convulsions, and even death (Kalimeris et al. 2013; Wang et al. 2019). Severe hypoxia can attenuate judgement, while the brain is highly sensitive to hypoxia, mainly in the hippocampus (Lu et al. 2017). The cognitive functions include learning ability and memory ability. On the other hand, the findings suggest that upregulation of GCN5L1 enhanced ROS -induced oxidative stress in vitro model. Manning et al. (2019) conclude that loss of GCN5L1 promotes cell death via promotion of oxidative stress. This study showed that GCN5L1 revealed antioxidant activity by regulation of FoxO1.

Generally, the antioxidant and oxidation systems in the organism maintain a dynamic balance (Liu et al. 2018). The imbalance between antioxidation and oxidation system in cells will cause oxidative stress. There are also many peroxides and antioxidants in the body. Such as MDA, GSH and CAT. MDA is the final product of lipid peroxidation reaction, which will lead to cross-linking reaction of protein and DNA, resulting in cell death, and also affect the damage of mitochondrial membrane and cell membrane (Liu et al. 2018). Oxidative stress can eliminate ROS by activating the expression of antioxidant stress kinase downstream of FoxO1(Liu et al. 2018). FoxO1 critically involved in neuronal apoptosis and survival (Chen et al. 2019; Ji et al. 2019). However, it requires further investigation on the specific mechanism underlying the effects of oxidative stress (Chen et al. 2019; Liu et al. 2019). GCN5L1 suppressed FoxO1 protein expression in vivo of newborn mice and vitro model. FoxO1 reduced the effects of GCN5L1 on anaesthesia-induced nerve injury in vivo of newborn mice and vitro model. Wang et al. (2017) reported that GCN5L1 controls glucose levels through FoxO1

levels. These results showed that GCN5L1 suppressed Fox O1 protein to promote oxidative stress, however, the mechanism of GCN5L1 regulated Fox O1 was unclear and it needs further research.

CONCLUSION

The results indicate that GCN5L1 mRNA and protein expression increased anaesthesia-induced nerve injury. GCN5L1 enhanced ROS production levels and oxidative stress in anaesthesia-induced nerve injury of newborn mice via suppression of FoxO1 by oxidation effects, which may have the potential for clinical applications of anaesthesia-induced nerve injury in newborn mice.

RECOMMENDATIONS

Therefore, the researchers speculate that GCN5L1 may be one clinical significance for anaesthesia-induced nerve injury and other neurological diseases.

ETHICAL APPROVAL

All rats were treated according to the guidelines of the Guide for the Care and Use of Laboratory Animals. The Laboratory Animal Care Committee of Yan Tai Yu Huang-ding Hospital approved all experimental procedures and protocols.

AVAILABILITY OF DATA AND MATERIAL

The datasets used or analysed during the current study are available from the corresponding author on reasonable request.

CONFLICTS OF INTEREST

None

FUNDING

None

AUTHOR CONTRIBUTION

- Guarantor of integrity of the entire study: Hui Li
- Study concepts: Hui Li
- Study design: Hui Li
- Literature research: Yan-jie Ding
- Clinical studies: Xiao-ling Jiao, Jing-jing Ge
- Experimental studies: Xiao-ling Jiao, Jing-jing Ge
- Data acquisition: Xiao-ling Jiao, Jing-jing Ge
- Data analysis: Hui Li, Xiao-ling Jiao, Jing-jing Ge
- Statistical analysis: Xiao-ling Jiao, Jing-jing Ge
- Manuscript preparation: Hui Li
- Manuscript editing: Hui Li
- Manuscript review: Yan-jie Ding

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