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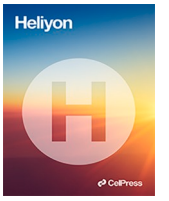

Research article

Protective effect of Danshensu against neurotoxicity induced by monosodium glutamate in adult mice and their offspring

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ABSTRACT

Excessive neuronal excitation by glutamate is a well-established cause of neurotoxicity, leading to severe impairment of brain function. Excitotoxicity is a key factor in numerous neurodegenerative conditions. In this study, we investigated the neuroprotective effects of Danshensu (DSS) against monosodium glutamate (MSG)-induced neurotoxicity in adult mice and their offspring. We randomly divided one hundred 8-week-old Kunming mice (equal number of males and females) into a control group and an experimental group. The experimental group was further subdivided into various treatment groups, including MSG gavage treatment, bwbw DSS treatment group 1 (bwbw DSS treatment group 2, a drug control group, and a normal control group (receiving an equal volume of physiological saline for ten consecutive days). Additionally, another one hundred healthy 8-week-old Kunming mice were similarly divided into groups and treated. These mice were paired randomly (one male and one female) and pregnant females were housed separately to obtain offspring. Subsequently, we conducted histological and behavioral analyses on adult mice and their offspring. MSG treatment induced significant cellular edema and hippocampal damage in both the treated mice and their offspring. However, varying doses of DSS effectively counteracted the neurotoxic effects of MSG, with no adverse impact on brain tissue structure or neural function in either adult mice or their offspring. Behavioral experiments further confirmed that DSS exerted a substantial protective effect against MSG-induced impairment of learning and memory in the treated adult mice and their offspring, in addition to mitigating central nervous system overexcitation and inhibiting exploratory behavior. In conclusion, DSS exerts significant protective effects against MSG-induced neurotoxicity in both adult mice and their offspring.

1. Introduction

Monosodium glutamate (MSG), a sodium salt of a nonessential amino acid, can be dissociated into glutamate (Glu) after entering the body and is one of the main causes of neurotoxicity [1]. Currently, MSG is a food additive used commercially to preserve the freshness, enhance the taste of vegetables, and enhance appetite. It is widely used in countries worldwide, especially Asian countries.

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Although the global consumption of MSG has increased, many studies have shown that excessive intake of MSG may increase the risk of various diseases, such as obesity, diabetes, and Alzheimer's disease [2]. Glu is the main excitatory amino acid in the mammalian brain and is involved in information transmission in the central nervous system [3]. Glutamate receptors, such as N-methyl-D-aspartate (NMDA) receptors, are widely distributed in the central nervous system, including the amygdala, hippocampus, and hypothalamus, and participate in regulating energy metabolism and autonomic function. When excessive MSG is consumed, glutamate enters the brain in large quantities, significantly increasing in concentration, which can produce neurotoxicity and cause neuronal damage or death. MSG-induced neurotoxicity is associated with some specific symptoms and signs. MSG can cause chemical brain damage, cognitive decline, neurodegeneration and mental symptoms (depression, anxiety, sleep disorders and irritability). Exposure to MSG can lead to cognitive impairment in animal models [4–6]. Continuous oral administration of 2 g/kg MSG in 5-week-old male Wistar rats leads to long-term memory impairment and inhibition of Na^+-K^+ ATPase in the hippocampus and cortex [7]. Many studies have shown that because the blood–brain barrier of newborn animals is not yet mature, oral, or subcutaneous injection of MSG can cause neuronal necrosis in some areas of the central nervous system [8–10]. After subcutaneous administration of MSG to pregnant rats, acute necrosis of acetylcholinesterase-positive neurons in the posterior zone was observed in both mother and offspring rats, and fetal neurons were found to be more sensitive to MSG [11].

Salvia miltiorrhiza Bge (Lamiaciae) is a dicotyledonous perennial upright herb. Its medicinal parts are the roots and rhizomes. It tastes bitter, feels slightly cold, and has the functions of removing blood stasis and pain, promoting blood circulation and menstruation, and clearing the heart. *Salvia miltiorrhiza* has a long history as a medicinal plant. According to Shennong Ben Cao Jing (Eastern Han Dynasty, 25-220 AD), the targets of *Salvia miltiorrhiza* include " The main internal evil, intestines rumbling softly like flowing water, accumulation of cold and heat, breaking through diseases and eliminating masses " [12]. Danshensu (DSS) is the main pharmacological component of *Salvia miltiorrhiza*. Its water-soluble component is more stable than its other components and can be quickly absorbed after entering the human body [13]. The chemical name of DSS is D-(+)-β-(3,4-dihydroxyphenyl) lactic acid, and its molecular formula is $C_9H_{10}O_5$. Its physical properties include an elongated needle-like crystal structure and a high degree of solubility in H_2O, CH_3OH, C_2H_6O, $C_4H_{10}O$, etc. DSS is easily oxidized into a quinone-like structure when in contact with water or air and is more easily oxidized in alkaline environments. Therefore, when preparing DSS solution, it is necessary to add certain antioxidants or prepare DSS sodium to improve its stability [14,15]. To date, domestic and foreign scholars have conducted a large amount of basic research on DSS, showing that it has pharmacological effects such as reducing thrombus formation, improving microcirculation, inhibiting bacterial growth, alleviating inflammation, inhibiting cell apoptosis, decreasing oxidative stress, and enhancing the body's immune system. This compound is widely used in clinical and scientific research.

Researchers have assessedthe effects of DSS on neural cells and SH-SY5Y cells under ischemic and hypoxic conditions. Wei et al. found that DSS can promote the differentiation of neural stem cells and accelerate their migration to the site of brain injury in a study in fetal rats. The experiment suggests that there may be four possible mechanisms through which DSS exerts a protective effect on neurons after cerebral ischemic injury: reducing mitochondrial dysfunction caused by ischemic and hypoxic conditions, inhibiting neuronal apoptosis, regulating cellular energy metabolism function, and reducing mitochondrial phosphorylation impairment caused by cerebral ischemia [16]. In addition, Yu et al. found that DSS can alleviating neuronal functional damage induced by cerebral ischemia by reducing superoxide dismutase activity and malondialdehyde content [17]. Therefore, DSS has a certain protective effect against neuronal damage, alleviating ischemic damage to brain tissue and inhibiting pathological apoptosis of neurons [18]. This article will first analyze the neurotoxicity caused by different doses of MSG in treated mice and their offspring and then study the protective effects of different doses of DSS on the nervous system of mice and their offspring.

2. Materials and methods

2.1. Animals and treatment

All animal experimental procedures were conducted following

protocols approved by the Hainan Technology and Business College Animal Use and Care Committee (HTBC-2023-00000322). One hundred 8-week-old Kunming mice, half male and half female, were randomly divided into a control group and an experimental group. The experimental group was further divided into the MSG gavage group (2.0 g/kg or 4.0 g/kg body weight (bw) MSG for 10 consecutive days) [19], DSS treatment group 1 (2.0 g/kg bw MSG by gavage accompanied by intraperitoneal injection of 5 g/kg, 10 g/kg, or 20 g/kg DSS for 10 days), DSS treatment group 2 (4.0 g/kg MSG by gavage accompanied by intraperitoneal injection of 10 g/kg, 20 g/kg, or 30 g/kg DSS for 10 days), the drug control group (30 g/kg DSS for 10 days) [20], and the normal control group (equal volume of physiological saline for 10 consecutive days). Subsequently, another 100 8-week-old healthy Kunming mice were divided into groups and treated in the manner described above. Mice were housed in randomly assigned male/female pairs. After becoming pregnant, the female mice were housed in separate cages to obtain offspring.

2.2. Hematoxylin&eosin (H&E) staining

Mice were continuously treated with MSG (2.0 g/kg or 4.0 g/kg bw) by gavage and intraperitoneally injected with different doses of DSS. Two mice were selected every two days after drug administration and were anesthetized by intraperitoneal injection of pentobarbital sodium (60 mg/kg). The animals were killed by cardiac perfusion with 10 % formaldehyde solution. The head was cut off, and the brain was fixed in 10 % formaldehyde solution for one week. Paraffin-embedded, 10-μm-thick coronal sections of the whole brain were obtained, and 100-μm-thick sections were prepared from one piece of the brain. After 90 days of drug administration, 4 rats from

each group were selected for histopathological examination, and samples were prepared using the same method. One week after drug administration, when they were 90 days old, four offspring mice were selected from each group for H&E staining. Samples were prepared as described above.

2.3. Y-maze

The Y-maze was used to evaluate the learning and memory abilities of mice. The specific experimental steps are as follows. At the beginning of the experiment, one randomly selected arm of the Y-maze that was illuminated with a light and kept unheated was designated as the safe zone. The other two arms of the maze were kept unlit and heated area at the junction (60–80 °C), and these arms were designated as unsafe zones. After adult mice were treated with DSS on days 1, 2, 3, 4, 5, 6, 36, and 90 or young mice were treated with DSS on days 41, 42, 43, 44, 45, 46, and 76, they were trained in the maze in a dark and quiet environment. Each mouse was placed in the Y-maze and allowed to adapt for 20 s, and then the light in the safe zone was turned on. The mice were placed in the starting arm, and after being stimulated by heat, they were allowed to escape to the safe zone and stay there for 5 s; this procedure was considered one training session. Each mouse was subjected to training sessions until it successfully escaped in $\geq$18 out of 20 consecutive training sessions. Finally, we compared the number of training sessions required and errors among the groups of mice and analyzed whether the learning and memory abilities of adult and young mice were impaired. After each training session, the Y maze was cleaned to eliminate the impact of odor cues on the mice [21].

2.4. Morris water maze

The Morris water maze is used to evaluate the learning and memory abilities of mice toward spatial position and direction. The specific experimental steps were as follows. The pool was filled with clean water in advance, ink was added and mix well to make the pool water opaque black. The safety platform was placed in the middle of the quadrant far from the water entry point, and the position of the safety platform was maintained during the experiment. The water surface was 1 cm higher than the platform, the water depth was 30 cm, and the water temperature was controlled at 24 $\pm$ 0.5 °C. The water entry point of each experimental mouse in other quadrants of the pool was marked. The position of laboratory reference objects such as the water pool, light source, and mouse cage remained unchanged for each experiment, and four different quadrants of water entry points were used for training four times a day. The time was set to 90 s. If the mouse had not yet found the platform within 90 s, it was taken to a safe platform for 15 s. If the mouse found the platform within 90 s, it was also be allowed to stay on the platform for 15 s to end a training session. This process was repeated for 5 days and the mice were tested on the 6th day. During the test, the platform was removed, and each mouse swam for 90 s. The computer software automatically records and processes the incubation period of each mouse's first crossing of the platform [22].

2.5. Autonomous activity test

An autonomous activity experiments was used to evaluate the excitability of the central nervous system [23]. On the 1st, 2nd, 3rd, 4th, 5th, 6th, 36th, and 90th days (adult mice) or the 41st, 42nd, 43rd, 44th, 45th, 46th, and 76th days (young mice) after DSS treatment, mice from each group were placed in an ZIL-2 programmable mouse autonomous activity meter. After adaptation for 5 min, testing began, and the number of activities within 5 min was recorded. The mice were tested 3 times a day, and the average number of

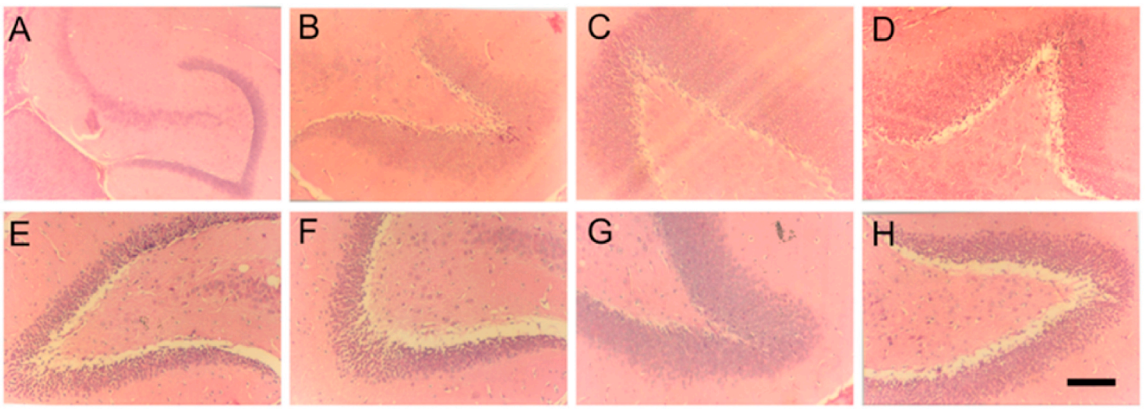

Fig. 1. H&E analysis of the effects of MSG treatment on the hippocampal tissue of adult mice and their offspring **A** Control group: the cells in the hippocampus were arranged neatly and showed no obvious damage. **B–C** MSG (2.0 g/kg bw) group: obvious proliferation or degeneration of cells in the hippocampus could be observed. **D** MSG (4.0 g/kg bw) group: neurocyte proliferation or obvious hyperplasia of cells in the hippocampus could be observed. E-F MSG (4.0 g/kg bw) group: local obvious edema or degeneration of cells in the hippocampus could be observed. **G** MSG (2.0 g/kg bw) group: the cells in the hippocampus of offspring mice also showed significant proliferation. **H** MSG (4.0 g/kg) group: neurocyte proliferation accompanied by mild cell necrosis could be observed in the hippocampus of offspring mice (scale bar, 100 μm).

activities was calculated.

2.6. Hole plate experiment

The hole board experiment was used to evaluate exploratory activity [24]. On the 1st, 2nd, 3rd, 4th, 5th, 6th, 36th, and 90th days (adult mice) or the 41st, 42nd, 43rd, 44th, 45th, 46th, and 76th days (young mice) after DSS treatment, mice from each group were placed in a XDB-2 mouse hole board apparatus, and the number of times the mice explored the target hole over 180 s was recorded.

2.7. Statistical analysis

The experimental data were analyzed by the statistical software SPSS 15.0, and the original data were expressed as the mean ± SEM. Student's *t*-test and chi-squared test were used. $P < 0.05$ indicates that the difference is significant, $P < 0.01$ indicates that the difference is very significant, and $P < 0.001$ indicates that the difference is extremely significant.

3. Results

3.1. Pathological analysis of brain tissue damage induced by MSG gavage in adult mice and their young offspring

To analyze the effect of MSG treatment on the hippocampus of adult mice, we administered different doses of MSG (2 g/kg and 4 g/kg) to mice by gavage for 10 consecutive days. Histopathological analysis showed that after 90 days of MSG treatment, varying degrees of brain tissue damage were observed in the different treatment groups compared to the control group (Fig. 1A). In the group treated with 2.0 g/kg MSG, the cells in the hippocampus of adult mice showed significant proliferation or degeneration (Fig. 1B and C). Adult

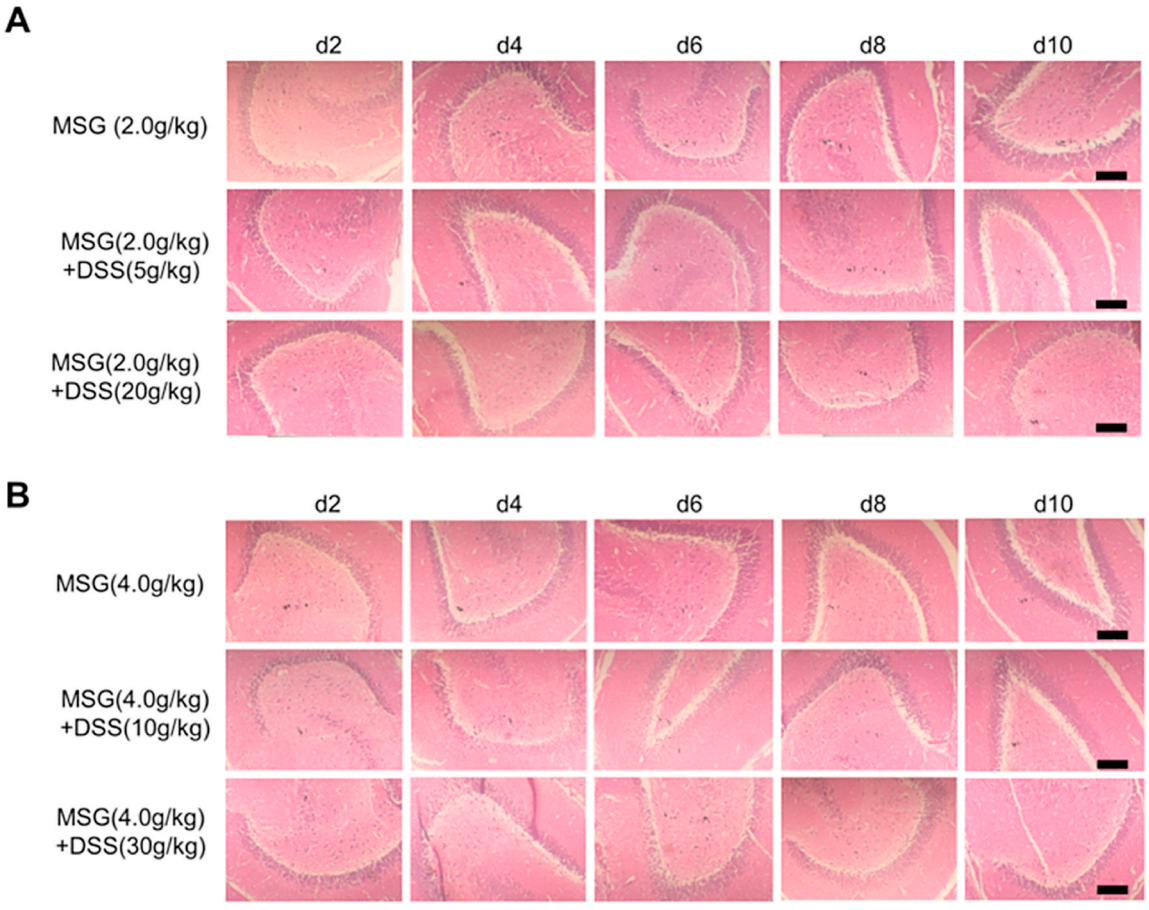

Fig. 2. The protective effect of DSS against hippocampal tissue damage induced by MSG in adult mice **A H&E** analysis of the protective effects of 10 g/kg and 30 g/kg DSS against hippocampal tissue induced by 2 g/kg MSG at 2, 4, 6, 8, and 10 days in adult mice (scale bar, 100 μm). B Histopathological analysis of the protective effects of 10 g/kg and 30 g/kg DSS against hippocampal tissue induced by 2 g/kg bw MSG at 2, 4, 6, 8, and 10 days in adult mice (scale bar, 100 μm).

mice treated with 4.0 g/kg MSG showed more significant proliferation of neural cells in the hippocampus (Fig. 1D) and local edema or degeneration (Fig. 1E and F). To investigate whether MSG treatment affects the offspring of adult mice, we mated male and female mice treated with MSG and obtained F1 offspring after 90 days. Through pathological analysis of the brain tissue of F1 offspring mice, we found significant proliferation of hippocampal cells in the offspring of mice that received 2.0 g/kg MSG (Fig. 1G). Similarly, in the offspring of mice treated with 4.0 g/kg MSG, cell proliferation was more obvious and was accompanied by mild cell necrosis (Fig. 1H). The above results indicate that MSG treatment can dose-dependently damage the hippocampus of adult and offspring mice.

3.2. Protective effect of DSS on brain hippocampal tissue damage induced by MSG in adult mice

We evaluated whether DSS can reduce MSG-induced damage to hippocampal tissue in adult mice. First, we analyzed the protective effect of 5 g/kg and 20 g/kg bw DSS on hippocampal tissue damage caused by 2 g/kg MSG. After oral administration of 2 g/kg MSG, adult mice were treated with 5 g/kg or 10 g/kg DSS. After 2, 4, 6, 8, and 10 days of treatment, H&E staining was used to analyze the histological changes in hippocampal neurons in the mouse brain. As shown in Fig. 2A, both 5 g/kg and 20 g/kg DSS significantly reduced neuronal damage in the hippocampus. More importantly, 20 g/kg DSS significantly reduced nerve cell proliferation and edema, demonstrating a stronger therapeutic effect than the other doses. In addition, to evaluate the reparative effect of DSS on brain tissue damaged by high-dose MSG, we studied the protective effects of 10 g/kg and 30 g/kg DSS against hippocampal tissue damage caused by 4 g/kg MSG. Similarly, after mice were gavaged with 4 g/kg MSG, they were treated with 10 g/kg or 30 g/kg DSS. On the 2nd, 4th, 6th, 8th, and 10th days after treatment, H&E staining was used to analyze the morphological changes in mouse hippocampal tissue. We obtained similar results for 10 g/kg and 30 g/kg DSS, which significantly reduced hippocampal tissue damage caused by high-dose MSG. More importantly, 30 g/kg DSS significantly reduced cell proliferation and edema and exhibited a more significant protective effect against neurotoxicity (Fig. 2B). Overall, the above results indicate that DSS can significantly reduce cellular damage in mouse hippocampal tissue caused by MSG.

3.3. Protective effect of DSS on hippocampal tissue damage induced by MSG in the offspring of treated rats

We analyzed the protective effect of DSS against hippocampal tissue damage caused by MSG in offspring mice. Adult mice were given 2.0 g/kg bw MSG by gavage while receiving multiple doses of DSS. The offspring were obtained 90 days after administration, and histological changes in the histology were observed. Histological analysis showed that the neural cells in the hippocampus of adult mice in the normal control group were neatly arranged and clearly defined (Fig. 3A). However, 2 g/kg MSG caused severe cell necrosis, and after 90 days of treatment with 5 g/kg, 10 g/kg, or 20 g/kg DSS, brain damage in adult mice was significantly reduced, with only mild necrosis or mild hyperplasia of the hippocampus being observed (Fig. 3B–D). Histopathological analysis of the hippocampus of offspring mice treated with 5 g/kg DSS showed slight proliferation, indicating significant amelioration of brain tissue damage in the offspring mice (Fig. 3E). In addition, after 90 days of treatment with 5 g/kg, 10 g/kg, or 30 g/kg DSS, the degree of brain tissue damage

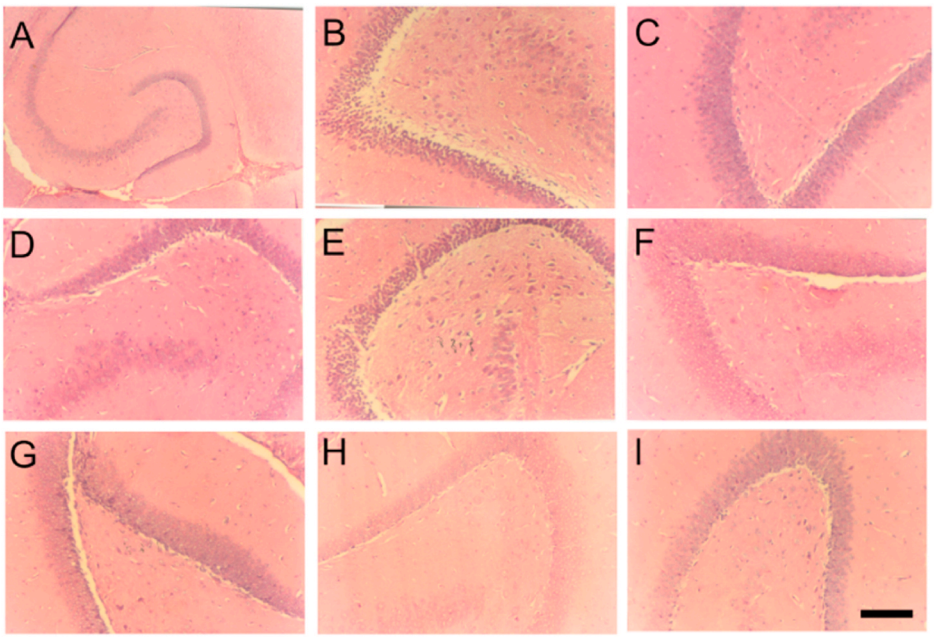

Fig. 3. H&E analysis of the protective effect of DSS against hippocampal tissue damage induced by MSG in offspring mice. **A**: Control group. **B**: 2 g/kg MSG+5 g/kg DSS group. **C**: 2 g/kg MSG+10 g/kg group. **D**: 2 g/kg MSG+20 g/kg group. **E**: F1 (2 g/kg MSG) group. **F**: 4 g/kg MSG+5 g/kg group. **G**: 4 g/kg MSG+10 g/kg group. H: 4 g/kg MSG+30 g/kg group. **I**: F1 (2 g/kg MSG) group. Scale bar, 100 μm.

caused by 4 g/kg MSG treatment in adult mice gradually decreased, with only mild necrosis or cell hyperproliferation being observed in the hippocampus (Fig. 3F–H). Histopathological analysis of the hippocampus of offspring treated with 10 g/kg DSS mice showed slight proliferation, indicating a significant reduction in brain tissue damage in the offspring mice (Fig. 3I). The above results indicate that DSS has a significant protective effect against MSG neurotoxicity in offspring mice, indicating that the protective effect of DSS against MSG neurotoxicity also benefits offspring through some genetic mechanism.

3.4. Protective effect of DSS on MSG-induced learning and memory impairment in adult mice and their offspring

To investigate whether DSS can alleviate learning and memory impairment caused by MSG in adult mice and their offspring, we analyzed the learning and memory abilities of adult mice and their offspring using the Y-maze test. First, as shown in Fig. 4A and B, we found that 30 g/kg DSS itself did not impair learning or memory in adult mice, as measured by the Y-maze test. Second, we found that after 10 consecutive days of oral administration of MSG (2.0 g/kg or 4.0 g/kg bw), adult mice showed significant impairment of learning and memory in the Y-maze test. However, as the dose of DSS increased, it almost completely offset the learning and memory impairment caused by MSG in adult mice, and the therapeutic effect was dose-dependent. Similarly, we observed that 2.0 g/kg or 4.0 g/kg MSG significantly affected the learning and memory of young mice; however, the offspring of mice treated with multiple doses of DSS also showed significant improvements in learning and memory (Fig. 4C and D). These results indicate that DSS has a protective effect against MSG-induced learning and memory impairment in treated adult mice and their offspring.

We analyzed the effect of DSS on spatial learning and memory impairment induced by MSG in adult mice and their offspring using the water maze test. As shown in Fig. 5A and B, DSS alone did not affect the spatial learning and memory abilities of adult mice. Compared to the control group of adult mice, the latency of adult mice treated with 2.0 g/kg or 4.0 g/kg MSG to reach the platform was significantly increased. However, adult mice treated with different doses of DSS showed a significant decrease in latency, indicating that DSS treatment improved the ability of mice to quickly locate the original platform area. Similarly, two doses of MSG caused spatial learning and memory impairment in offspring mice; however, after DSS treatment, the damaging effect of MSG on the learning and memory abilities of offspring mice in the water maze test was almost completely offset, resulting in a shortened latency and a reduced number of errors (Fig. 5C and D). The above results indicate that DSS can dose-dependently alleviate learning and memory impairment induced by MSG in adult mice and their offspring.

3.5. Protective effect of DSS against MSG-induced central nervous system hyperexcitability in adult mice and their offspring

In addition, we analyzed the effect of DSS on central nervous system hyperexcitability caused by MSG. After 10 days of continuous oral administration of MSG (2.0 g/kg or 4.0 g/kg bw) to adult mice, hyperexcitability of the central nervous system was observed, which was manifested as a significant increase in the frequency of spontaneous activity compared to the control group mice. However, after multiple doses of DSS, the frequency of spontaneous activity in adult mice was significantly reduced, indicating that DSS can significantly prevent MSG-induced hyperexcitability of the central nervous system in adult mice (Fig. 6A and B). Similarly, we analyzed the effect of DSS on the excitability of the central nervous system in the offspring of treated mice. The experimental results showed that 2.0 g/kg and 4.0 g/kg MSG treatment could significantly increase the frequency of spontaneous activity in offspring mice;

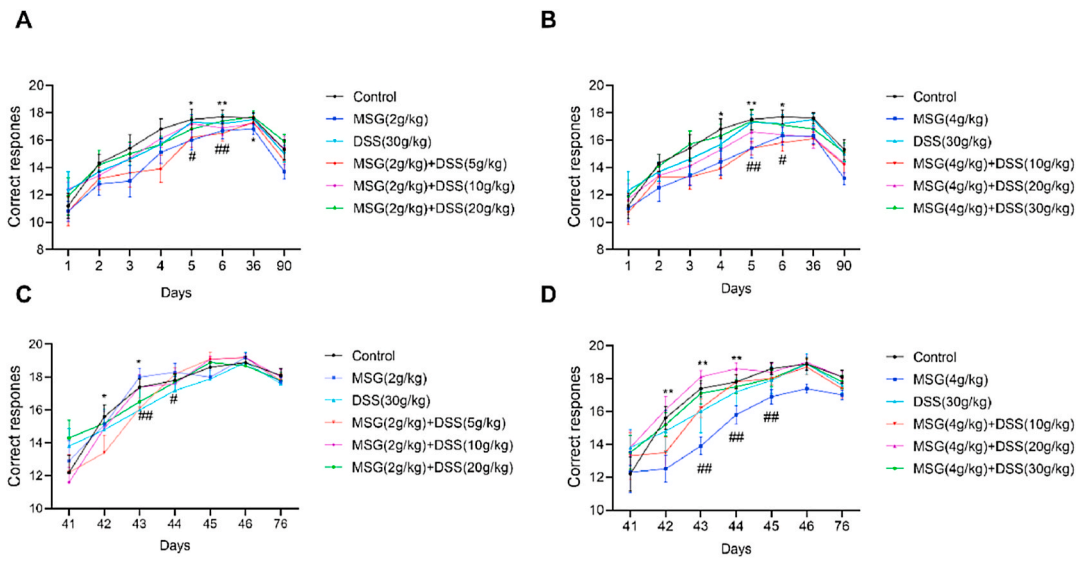

Fig. 4. Protective effects of DSS against MSG-induced impairment of the discrimination, learning and memory abilities of adult mice (**A-B**) and offspring mice (**C-D**) in the Y-maze test. The results are expressed as the mean ± SEM (#P < 0.05, # #P < 0.01, vs. the control group; *P < 0.05, **P < 0.01, vs. the MSG-treated group).

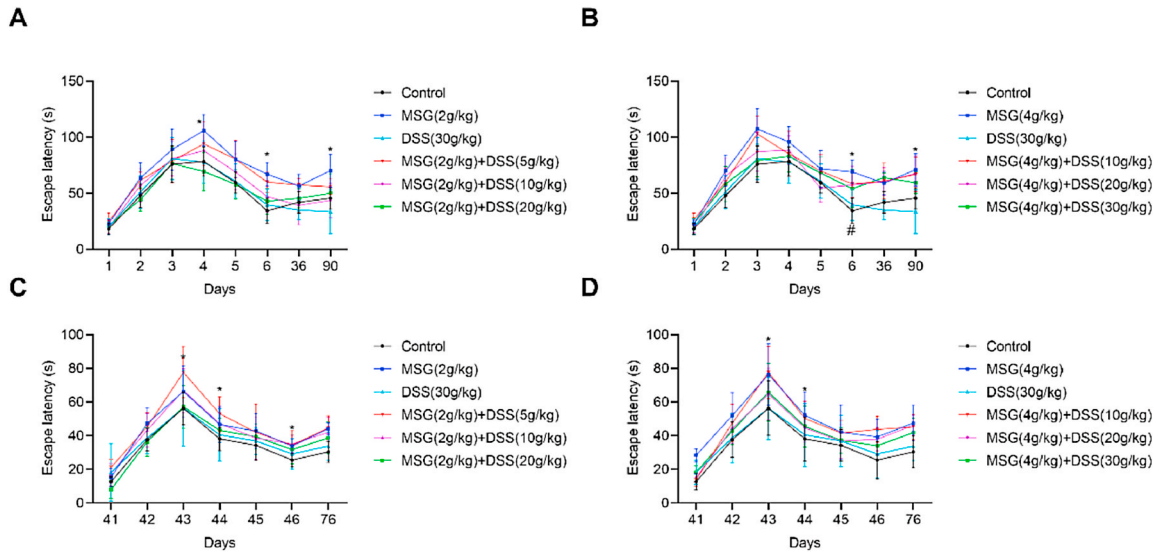

Fig. 5. Protective effects of DSS against MSG-induced impairment the spatial learning and memory abilities of adult mice (**A-B**) and offspring mice (**C-D**) in the water maze test. The results are expressed as the mean ± SEM. #P < 0.05, # #P < 0.01, vs. the control group; *P < 0.05, **P < 0.01, vs. the MSG-treated group.

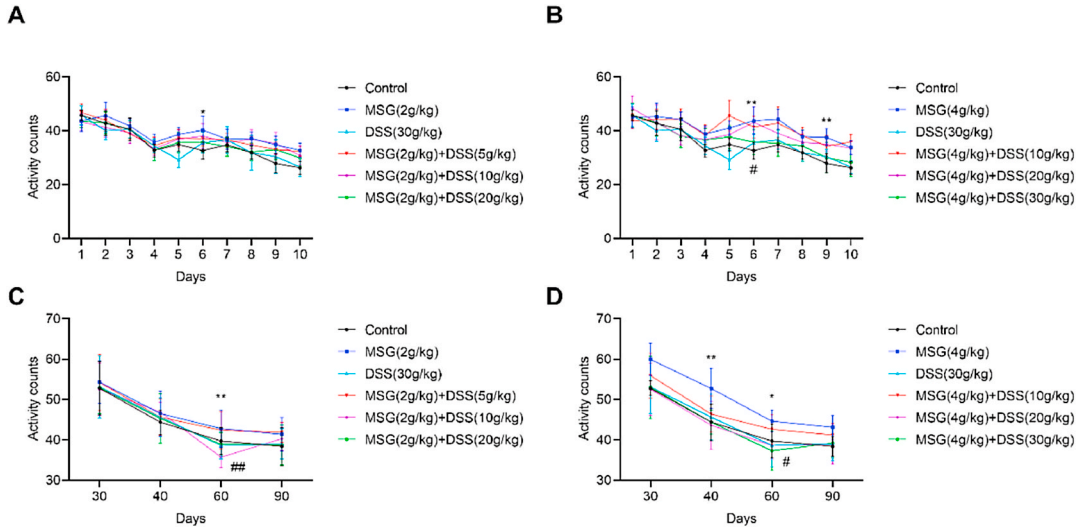

Fig. 6. Protective effects of DSS against MSG-induced CNS hyperexcitability in adult mice (**A-B**) and offspring mice (**C-D**). The results are expressed as the mean ± SEM. #P < 0.05, # #P < 0.01, vs. the control group; *P < 0.05, **P < 0.01, vs. the MSG-treated group.

however, after simultaneous administration of DSS (5 g/kg, 10 g/kg, 20 g/kg or 10 g/kg, 20 g/kg, and 30 g/kg), the frequency of spontaneous activity in young mice significantly decreased (Fig. 6C and D). These results indicate that DSS can also significantly inhibit the excitotoxic effect of MSG on the central nervous system in offspring mice.

3.6. The protective effect of DSS against the inhibition of exploratory behavior by MSG in adult mice and their offspring

We analyzed whether DSS can prevent the inhibition of exploratory behavior in mice induced by MSG. After 10 continuous days of oral administration of MSG (2.0 g/kg or 4.0 g/kg bw) to adult mice, the number of burrowing attempts by mice was significantly reduced. However, after simultaneous administration of DSS (5 g/kg, 10 g/kg, and 20 g/kg) or (10 g/kg, 20 g/kg, and 30 g/kg), the number of exploratory attempts by adult mice significantly increased, and DSS itself did not affect the number of exploratory attempts by adult mice (Fig. 7A and B). Similarly, we also analyzed whether DSS could affect the ability of young mice to explore the holes in the hole board test. The results showed that 2.0 g/kg and 4.0 g/kg MSG could significantly inhibit the exploratory behavior of young mice, manifested by a significant decrease in the number of burrowing attempts. However, for offspring mice treated with multiple doses of

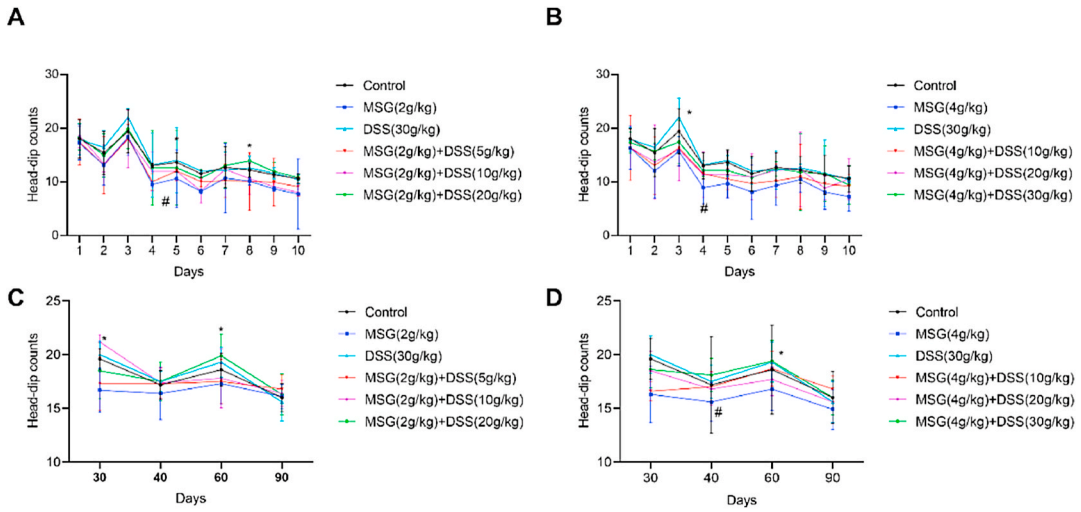

Fig. 7. Preventive effect of DSS against the inhibition of exploratory behavior induced by MSG in adult mice (**A-B**) and offspring mice (**C-D**). The results are expressed as the mean ± SEM. $^{\#}P < 0.05$, vs. the control group; $^{*}P < 0.05$, vs. the MSG-treated group.

DSS, there was a significant increase in the number of burrowing attempts (Fig. 7C and D). These results indicate that DSS can significantly prevent the inhibition of exploratory behavior in adult mice and their offspring caused by MSG.

4. Discussion

Previous studies have shown that DSS can expand cerebral blood vessels, reduce vasoconstrictive force, and thus improve neural function. Here, in this study, we investigated the effect of DSS on excitatory neurotoxicity induced by MSG in the adult mice and their offspring. We found that DSS exerted a dose-dependent protective effect against excitotoxicity caused by MSG in treated adult mice and their offspring. Interestingly, we also found that MSG caused an increase in physical activity and a decrease in the luster of the fur in mice. The reason for this increased activity is that Glu is the main excitatory amino acid in the mammalian brain and is involved in the transmission of information in the central nervous system; excessive Glu can lead to increased neuronal activity, resulting in increased physical activity [25]. The reason for the dullness of the coat is that a large amount of MSG converts zinc in the body into zinc glutamate and causes it to be eliminated from the body, leading to zinc deficiency in mammals. Zinc is an important trace element in mammals, helping to maintain normal development and growth. Zinc deficiency may cause dry skin and hair and other symptoms.

Histopathological examination showed that different doses of MSG caused varying degrees of necrosis or proliferation in the hippocampus of adult mice and their offspring. As the dose of MSG increased, the brain damage in adult mice and their offspring worsened. MSG treated resulted in a decrease in the number of normal cells in the hippocampus of adult mice and their offspring. The above results are consistent with the findings of Saiqa Tabassum et al. on the effects of MSG regarding learning and memory abilities and hippocampal cell counts in mice [26]. As the dose of DSS increased, adult mice and their offspring showed significantly reduced brain damage compared to the control group, indicating that different doses of DSS have varying degrees of protective effects against brain damage caused by MSG in adult mice and their offspring. The observed protective effect of DSS against brain injury is consistent with previously published research results [27–31]. DSS has a significant protective effect against MSG neurotoxicity, and this protective effect also benefits offspring through a genetic mechanism.

Different doses of MSG lead to varying degrees of learning and memory impairment, central nervous system hyperexcitability, inhibition of exploratory behavior, and impairment of high-altitude coordination ability in adult mice and their offspring. As the MSG dose increases, the decline in learning and memory abilities of adult mice and their offspring becomes more pronounced, the central nervous system became more overexcited, the inhibition of exploratory curiosity behavior becomes more obvious, and the impairment of high-altitude coordination ability becomes more obvious. This is consistent with previously published research results [32–34]. As the dose of DSS increased, the protective effect of DSS against MSG-induced impairment of learning and memory abilities, exploratory behavior, and high-altitude coordination and MSG-induced damage to the central nervous system became more apparent in treated adult mice and their offspring. This indicates that different doses of DSS have varying degrees of protective effects against these adverse effects of MSG in adult mice and their offspring. This suggests that the protective effect of DSS against the adverse effects of MSG, as illustrated, for example, by climbing ability, can also benefit offspring through some genetic mechanism.

Conclusion: DSS has a significant protective effect against the neurotoxic damage caused by MSG in adult mice and their offspring. With a rising standard of living, the public is becoming increasingly demanding regarding the taste of their food, leading to the excessive consumption of MSG, a widely used food additive. This study has practical significance as a resource to help guide the use of DSS to treat MSG-induced neurotoxic damage.

Data availability statement

We confirm that the data used in this study are available upon reasonable request from the corresponding authors (Yuanzhu Li and Yuxia Zhang).

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Qiong Liang, Yanan Zou, and Yuxia Zhang designed the experiments and interpreted the data. Qiong Liang, Dingguo Li. carried out most of the experiments with technical assistance from Jianli Li and Yuanzhu Li. Yanan Zou and Yuxia Zhang provided critical input regarding the overall research direction. Qiong Liang wrote the paper with input from all coauthors. All the authors have read and approved the final manuscript.

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