Trigonelline mitigates lipopolysaccharide-induced learning and memory impairment in the rat due to its anti-oxidative and anti-inflammatory effect

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A B S T R A C T

Brain inflammation is associated with cognitive dysfunction, especially in elderly. Trigonelline is a plant alkaloid and a major component of coffee and fenugreek with anti-diabetic, antioxidative, anti-inflammatory, and neuroprotective effects. In this study, the beneficial effect of trigonelline against lipopolysaccharide (LPS)-induced cognitive decline was assessed in the rat. LPS was injected i.p. at a dose of 500 μg/kg to induce inflammation, and trigonelline was administered p.o. at doses of 20, 40, or 80 mg/kg/day 1 h after LPS that continued for one week. Trigonelline-treated LPS-challenged rats showed improved spatial recognition memory in Y maze, discrimination ratio in novel object discrimination test, and retention and recall in passive avoidance paradigm. Additionally, trigonelline lowered hippocampal malondialdehyde (MDA) and acetylcholinesterase (AChE) activity and improved superoxide dismutase (SOD), catalase, and glutathione (GSH). Furthermore, trigonelline depressed hippocampal nuclear factor-kappaB (NF-κB), toll-like receptor 4 (TLR4), and tumor necrosis factor α (TNFα) in LPS-challenged rats. All of the effects of trigonelline followed a dose-dependent pattern and in some aspects, it acted even better than the routinely-used anti-inflammatory drug dexamethasone. Collectively, trigonelline is capable to diminish LPS-induced cognitive decline via suppression of hippocampal oxidative stress and inflammation and appropriate modulation of NF-κB/TLR4 and AChE activity.

1. Introduction

Uncontrolled neuroinflammatory response is noxious to neurons and may lead to disorders like Alzheimer’s disease [1, 2], Parkinson’s disease [3], and multiple sclerosis [4] via mobilization and activation of different types of resident cells within the central nervous system including microglia and astrocytes [5, 6]. On the other hand, immune system undergoes some alterations during aging that leaves the elderly more vulnerable to infections [7, 8] such as bacterial or viral infections of the urinary or respiratory tract [9, 10]. Sepsis following severe infection may lead to lasting cognitive decline, especially in older individuals [11]. Lipopolysaccharide (LPS) is an endotoxin from Gram-negative bacteria that stimulates signaling cascades of pro-inflammatory cytokines [12]. Systemic injection of LPS causes neuroinflammation in brain areas critical for memory function like hippocampus [13], leading to cognitive deficit [14]. Increased oxidative stress burden [15] and disturbed antioxidant system [16], nuclear factor-kappaB (NF-κB) up-regulation and nuclear factor (erythroid-derived 2)-like 2 (Nrf2) down-regulation [17], greater activity of acetylcholinesterase [18], overproduction and release of pro-inflammatory mediators like tumor necrosis factor α (TNFα) [19], and enhanced expression of glial fibrillary acidic protein (GFAP) [16] are prominent subsequent to LPS challenge. Currently, nutraceuticals targeting neuroinflammation have been suggested as novel therapeutic tools for neurodegenerative and neuroinflammatory disorders [20, 21].

Trigonelline is a plant alkaloid that like caffeine is a major component of coffee [22] and is also present in fenugreek (Trigonella foenum-graecum L.) seeds with various medicinal benefits in traditional medicine [23]. Trigonelline has shown several pharmacological activities including anti-hyperglycemic and anti-hyperlipidemic, neuroprotective, anti-migraine, and memory-improving potentials, as reviewed before [24]. Trigonelline is capable to lower lipid peroxidation and enhancing antioxidant defensive system in a model of type 2 diabetes [25], has anti-inflammatory [26, 27] and anti-apoptotic [26, 28] potential, and could protect against development of diabetic peripheral neuropathy [29]. Additionally, trigonelline has exhibited...
acetylcholinesterase inhibitory effect [30] and has been claimed to be able to regenerate dendrites and axons and improve memory functions [31]. Recently, Zhou et al. have shown that trigonelline is able to suppress inflammation and to protect pancreatic β cells during pregnancy in a mouse model of diabetes [32]. However, further studies of its pharmacological activities and exact modes of action are strongly warranted along with its possible application in clinical settings. Since trigonelline may be a potential agent for management of inflammation-related disorders, this study was undertaken to evaluate its efficacy against LPS-induced cognitive deficits in the rat and to explore its related modes of action.

2. Materials and methods

2.1. Experimental design

Male albino Wistar rats (procured from Pasteur’s institute of Tehran, 190–230 g) were kept under standard laboratory conditions (a temperature range of 21–23 °C; a humidity of 40–60%; 12:12 h light/dark cycling; free access to food and water). Procedures involving animals and their care were conducted in conformity with NIH guidelines for the care and use of laboratory animals. The experimental design of the study has been shown in Fig. 1.

The rats (n = 56) were randomly assigned to 7 experimental groups, i.e. control, trigonelline80-treated control receiving trigonelline at a dose of 80 mg/kg, LPS, trigonelline20-, trigonelline40- and trigonelline80-treated LPS (receiving trigonelline at doses of 20, 40, or 80 mg/kg), and dexamethasone-treated LPS. To induce systemic inflammation and subsequent neuroinflammation, LPS from Escherichia coli (SigmaAldrich, St Louis, MO, USA; 0111:B4) was freshly dissolved in cold normal saline and i.p. administered at a dose of 500 μg/kg [33, 34]. Systemic LPS administration is a widely-accepted model for neuroinflammation induction in rodents [16, 35, 36]. Animals in the control group received normal saline. Trigonelline-CI (Cayman Chemical, Michigan, USA) was administered p.o. (dissolved in distilled water) at doses of 20, 40, or 80 mg/kg/day, started 1 h after LPS administration and continued for one week. Dexamethasone was administered p.o. at a dose of 0.2 mg/kg with a timetable like that of trigonelline. This dose of dexamethasone has shown to be effective for sepsis treatment and its accompanying memory impairment [37]. Behavioral studies were always conducted 2 h after trigonelline administration on testing days and from 11:00 to 16:00 by a trained experimenter unaware of treatments. Behavioral experiments including Y-maze, novel object recognition, and passive avoidance paradigms were conducted at week 1 post-LPS, as shown in Fig. 1. All rats were euthanized on day-7 after the last behavioral test.

Following LPS challenge, learning and memory deficits of varying degrees and natures develop in rodents [38-40]. Thus, we exploited following tests for assessment of cognitive decline following LPS administration.

2.2. Y-maze task

We used Y-maze test to assess spontaneous alternation behavior as a valid index of short-term spatial recognition memory [41]. In this respect, Y-maze tests are classified as a task for evaluation of spatial learning and memory and for assessment of working memory [42, 43]. The memory component in the Y-maze task is that the mouse must remember which arm was most recently visited in order to alternate the arm choices [41]. It has shown that cognitive impairment related to hippocampal region disturbs performance of rodents in this task [44]. The maze had three arms and a central interconnecting arena. All animals were tested once in a randomized order. After allowance of a 15 min period for adaptation to testing enclosure, animals were individually placed at the end of one arm and allowed to pass through the arms for a period of 8 min. An arm entry was counted when the hind paws were totally within the arm area. Alternation was defined as successive entries into the three arms on overlapping triplet sets (i.e. A, B, C, B, C, A, etc.). The number of maximum spontaneous alternation was the total number of arms entered-2 and the percentage was calculated as the ratio of actual to possible alternations (defined as the total number of arm entries-2). Additionally, total number of arm entrances was used as an index of locomotor activity.

2.3. Novel object discrimination (NOD) test

Novel object discrimination task is dependent on the animal ability to judge earlier occurrence of an event. This test is claimed to be suitable for analysis of episodic memory [45]. It has shown that ER stress associated to hippocampus could disturb cognition and performance of rodents in this task [46]. Our used protocol for this task has described before [47]. In this test, each rat received two consecutive 5 min object exploration trials separated by a 4 h inter-trial interval (ITI). Rats were exposed to two similar objects during the first (familiarization) trial, and one of the objects was randomly selected and replaced with a third, novel object in the second (choice) trial. During the two trials exploration of each object, defined as sniffing, licking, chewing, or having moving vibrissae while directing the nose toward and ≤1 cm from the object, was separately recorded. Sitting on an object in the absence of any directed interest was not regarded as exploratory activity. The objects and test areas were wiped between the trials to lower odor cues. The discrimination (D) ratio was calculated as time spent exploring the novel object compared with the familiar object relative to the total time spent exploring all objects, according to the formula: (t [novel] − t [familiar]) / (t [novel] + t [familiar]) × 100.

2.4. Passive avoidance test

This task was used to assess learning and memory process of a conditioned nature. It has demonstrated that learning and memory deficits due to lesioning hippocampal CA1 area could be assessed using

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Fig. 1. Schematic experimental design of the study. Behavioral tests including Y-maze, novel discrimination, and passive avoidance were done 7 day post-LPS.
such task [48]. This test was done according to a previous report with some modifications [49]. It consisted of two compartments, i.e. one lighted and one dark chamber with grid floor connected by a guillotine door. Electric shock was delivered by an isolated stimulator. On the first and second days, each rat was placed into the apparatus and left for 5 min to explore the chambers and to be adapted. During the acquisition trial (third day), animals were placed in the lighted chamber and after a 5 min habituation period, the guillotine door was opened and the latency to enter the dark chamber was recorded. After the rat entering the dark chamber, the door was closed and an electric foot shock (1 mA, 1 s) was applied. On the acquisition day, the initial latency (IL) of entrance into the dark chamber was obtained. One day later, a retention trial was done and the interval between the placement in the lighted chamber and the entrance into the dark chamber was obtained as step-through latency (STL up to a maximum of 300 s as cut-off).

2.5. Measurement of hippocampal oxidative stress

Following lipopolysaccharide challenge, oxidative stress increases in different brain regions like prefrontal cortex and hippocampus [50]. On this basis, we measured some indices of oxidative stress in the hippocampus.

At the end of week 1 post-LPS, rats were deeply anesthetized with an overdose of ketamine-HCl (150 mg/kg), decapitated with a guillotine, brains excavated carefully and quickly, hippocampi from both sides punched out and 10% homogenate (w/v) was made in cold PBS (0.1 M, pH 7.4) in the presence of protease inhibitor cocktail using a rotary homogenizer (IKA, Germany). After centrifuging at 5000 rpm for 10 min at 4 °C, the supernatant was aliquoted and stored at −70 °C for further analysis.

Malondialdehyde (MDA) level as a reliable marker of lipid peroxidation was determined as reported before [51]. For this purpose, a solution of thiobarbituric acid and trichloroacetic acid was mixed with supernatant and incubated at boiling water for 90 min. After cooling, samples were centrifuged and the absorbance was read at 532 nm with tetraethoxypropane as the standard.

For determination of catalase activity as an element of cell defense system, Claiborne’s method was applied [52]. In short, H$_2$O$_2$ was added to a mixture of potassium phosphate buffer (pH 7.0) and supernatant and the rate of H$_2$O$_2$ decomposition was measured at 240 nm.

Activity of superoxide dismutase (SOD) as an enzymatic element of cell defensive system was determined according to earlier reports [53]. Briefly, supernatant was incubated with xanthine and xanthine oxidase in potassium phosphate buffer (pH 7.8, 37 °C) for 40 min, and then nitroblue tetrazolium (NBT) was added. Thereafter, blue formazan was monitored at 550 nm. The amount of protein that inhibited NBT reduction to 50% maximum was defined as 1 nitrite unit (NU) of SOD activity.

Reduced form of glutathione (GSH) as a non-enzymatic element of cell defense system was determined according to earlier reports [54–56]. For this purpose, the supernatant was centrifuged with 5% trichloroacetic acid. To 0.1 ml of homogenate, 2 ml of phosphate buffer (pH 8.4), 0.5 ml of 5% dithiobis (2-nitrobenzoic acid) (DTNB) and 0.4 ml of distilled water was added and the absorbance was read at 412 nm.

Bradford method was applied for measurement of protein with bovine serum albumin as its standard [57].

2.6. Hippocampal acetylcholinesterase (AChE) assessment

Following LPS challenge, acetylcholinesterase level and/or its activity increases in brain regions including hippocampus and cortex and this is related to some cognitive impairments in rodents [58, 59]. The AChE activity was determined on the basis of degradation of acetylthiocholine iodide into a product that binds to 5,5-dithiobis-2-nitrobenzoic acid and turns yellow [60]. The kinetics of the reaction was followed over 5 min at 412 nm. AChE activity was expressed as mmol of substrate hydrolyzed/min/g protein.

2.7. Determination of hippocampal NF-κB, TNFα, and TLR4

Since after systemic administration of lipopolysaccharide, a neuroinflammatory response occurs in brain regions like prefrontal cortex and hippocampus [50, 61], thus we determined some biomarkers related to inflammation. The level of these factors in the hippocampal supernatant was measured using sandwich enzyme-linked immunosorbent assay and commercial kits according to manufacturer’s instructions (assay kits for theses parameters from MyBiosource, Inc., San Diego, USA). The absorbance of samples was read at 450 nm by Synergy HT microplate reader (BioTek, Winooski, Vermont, USA) and values were reported as their final concentration.

2.8. Statistical analysis

All results were presented as mean ± S.E.M. For assessment of data distribution, we applied Kolmogorov–Smirnov statistical test. Then, two-way ANOVA and one-way ANOVA tests were used for statistical analysis of data and if a significant difference was found out, pair-wise comparison was done using the Tukey post-hoc test. In addition, Pearson correlation test was used to assess possible association between IL and STL in passive avoidance test in different groups. In all calculations, significance level was set at p < 0.05.

3. Results

3.1. Assessment of spatial recognition memory in Y-maze test

Related data showing performance of animals in Y-maze task as an index of short-term spatial recognition memory ability has been shown in Fig. 2A. Two-way ANOVA excluding groups receiving trigonelline at doses of 20 and 40 mg/kg and dexamethasone-treated LPS group revealed a significant effect of LPS [F(1,30) = 17.74, p < 0.001] and a non-significant effect of treatment [F(1,30) = 3.74, p > 0.05]. There was also a significant LPS-treatment interaction [F(3,28) = 6.27, p < 0.05]. Our further one-way ANOVA analysis showed that alternation score in LPS (p < 0.01) (a decrease of 41.9%), LPS + Trigonelline20 (p < 0.01) (a decrease of 32.4%), and LPS + Trigonelline40 (p < 0.05) (a decrease of 25.8%) groups was significantly lower when compared to control and trigonelline administration at a dose of 80 mg/kg to LPS group significantly improved alternation index versus LPS-challenged group (p < 0.05) (an increase of 46.7%). In addition, dexamethasone administration to LPS group also significantly and to a lower degree improved alternation in LPS group (p < 0.05) (an increase of 33.4%). With respect to locomotor activity of rats and the possible effect of LPS injection and trigonelline treatments, total number of entrances into the arms in this task was counted. In this respect, although the total number of entrances was lower in various LPS-challenged groups, but the existing difference was statistically non-significant.

3.2. Novel object discrimination findings

Two-way ANOVA showed a significant effect of LPS [F(1,30) = 16.85, p < 0.001] and a non-significant effect of treatment [F(1,30) = 2.98, p > 0.05]. In addition, there was also a significant LPS-treatment interaction [F(3,28) = 6.18, p < 0.05]. One-way ANOVA showed a significant drop of discrimination ratio in LPS-challenged group (a decrease of 78.1%) as compared to control (p < 0.005) and this significant drop also existed for trigonelline-treated LPS group at a dose of 20 (p < 0.01) (a decrease of 44.5%) versus LPS group. In contrast, discrimination ratio in LPS + Trigonelline40 (p < 0.05) (an increase of 83.8%) and LPS + Trigonelline80 (p < 0.01) (an increase
of 99.6%) groups was significantly greater as compared to LPS-challenged rats (Fig. 2B). Meanwhile, dexamethasone did not exert a significant effect in this respect.

### 3.3. Passive avoidance paradigm findings

Fig. 2C exhibited the performance of animals in passive avoidance test as noted by initial (IL) and step-through (STL) latencies. Regarding IL, performance of two-way ANOVA showed no significant effect of LPS and treatment. In addition, there was also no significant LPS-treatment interaction. Furthermore, one-way ANOVA also showed no statistically significant difference amongst the groups. On the other hand, with respect to STL, performance of two-way ANOVA showed a significant effect of LPS \([F(1,30) = 24.51, p < 0.001]\) and a non-significant effect of treatment \([F(1,30) = 2.34, p > 0.05]\). In addition, there was also a significant LPS-treatment interaction \([F(3,28) = 6.67, p < 0.05]\). Further one-way ANOVA indicated that LPS \((p < 0.01)\) (a decrease of 55.6%) and LPS + Trigonelline20 \((p < 0.05)\) (a decrease of 41.3%) groups have a significant deficit of retention and recall in comparison with control group, as it was shown by a lower STL, and trigonelline administration at doses of 40 (an increase of 78.6%) or 80 mg/kg (an increase of 71.8%) to LPS-challenges groups significantly elevated STL when compared to LPS group \((p < 0.05)\). In addition, dexamethasone did not exert a significant effect in this respect.

Performance of Pearson correlation test for data of IL and STL for each group showed no statistically significant relationship.

### 3.4. Assessment of oxidative stress

For MDA, two-way ANOVA showed a significant effect of LPS \([F(1,30) = 19.76, p < 0.001]\) and a significant effect of treatment \([F(1,30) = 4.86, p < 0.05]\). In addition, there was also a significant LPS-treatment interaction \([F(3,28) = 4.13, p < 0.05]\). For SOD, two-way ANOVA showed a significant effect of LPS \([F(1,30) = 14.69, p < 0.001]\) and a significant effect of treatment \([F(1,30) = 7.43, p < 0.05]\). In addition, there was also no significant LPS-treatment interaction \([F(3,28) = 0.58, p > 0.05]\). Regarding catalase activity, two-way ANOVA showed a significant effect of LPS \([F(1,30) = 15.73, p < 0.001]\) and a significant effect of treatment \([F(1,30) = 7.85, p < 0.05]\). In addition, there was also no significant LPS-treatment interaction \([F(3,28) = 0.76, p > 0.05]\). With respect to GSH, two-way ANOVA indicated a significant effect of LPS \([F(1,30) = 15.81, p < 0.001]\) and a significant effect of treatment \([F(1,30) = 7.41, p < 0.05]\). In addition, there was also no significant LPS-treatment interaction \([F(3,28) = 0.81, p > 0.05]\). Performance of one-way ANOVA showed that LPS-challenges group has a significantly greater level of MDA (an increase of 42.1%) (Fig. 3A) \((p < 0.05)\) and significantly lower activity of SOD (a decrease of 40.7%) (Fig. 3B) \((p < 0.01)\) and catalase (a decrease of 46.3%) (Fig. 3C) \((p < 0.01)\) and lower level of GSH (a decrease of 47.2%) (Fig. 3D) \((p < 0.01)\) as compared to control group and administration of trigonelline at a dose of 80 mg/kg to LPS group significantly restored MDA (a decrease of 25.5%) \((p < 0.05)\), SOD activity (an increase of 46.7%) \((p < 0.01)\), catalase activity (an increase of 50.7%) \((p < 0.05)\), and GSH (an increase of 51.1%) \((p < 0.05)\) relative to LPS group and trigonelline at a dose of 20 mg/kg did not have such a significant effect. Additionally, dexamethasone did exert a significant effect regarding MDA and SOD activity in this respect \((p < 0.05)\).

### 3.5. Findings of hippocampal level of NF-κB, TLR4, and TNFαs

For NF-κB, two-way ANOVA showed a significant effect of LPS \([F(1,30) = 15.67, p < 0.001]\) and a significant effect of treatment \([F(1,30) = 5.81, p < 0.05]\). In addition, there was no significant LPS-treatment interaction \([F(3,28) = 2.69, p > 0.05]\). For TLR4, two-way ANOVA indicated a significant effect of LPS \([F(1,29) = 15.67, p < 0.001]\) and a significant effect of treatment \([F(1,29) = 5.81, p < 0.05]\). In addition, there was also no significant LPS-treatment interaction \([F(3,27) = 2.47, p > 0.05]\). For TNFα, two-way ANOVA showed a significant effect of LPS \([F(1,28) = 22.38, p < 0.001]\) and a significant effect of treatment \([F(1,28) = 7.65, p < 0.05]\). In addition, there was no significant LPS-treatment interaction \([F(3,26) = 3.75, p > 0.05]\). Performance of one-way ANOVA showed that hippocampal...
level of NF-κB (an increase of 119.8%) (Fig. 4A) (p < 0.01), TLR4 (an increase of 124.5%) (Fig. 4B) (p < 0.01), and TNFα (an increase of 117.9%) (Fig. 4C) significantly raises in LPS-challenged group when compared to control group. In addition, treatment of LPS group with trigonelline at a dose of 80 mg/kg significantly reversed NF-κB (a decrease of 41.4%) (p < 0.05), TLR4 (a decrease of 44.2%) (p < 0.01), and TNFα (a decrease of 36.4%) (p < 0.05) in comparison with LPS group. Meanwhile, dexamethasone also significantly reduced hippocampal level of these parameters versus LPS group (p < 0.05).

3.6. Assessment of hippocampal activity of AChE

Fig. 5 displays hippocampal AChE activity in different experimental groups. For this parameter, two-way ANOVA showed a significant effect of LPS [F(1,30) = 16.81, p < 0.001] and a significant effect of treatment [F(1,30) = 5.97, p < 0.05]. In addition, there was no significant LPS-treatment interaction [F(3,28) = 2.09, p > 0.05]. Performance of one-way ANOVA showed that AChE activity is significantly greater in LPS group (an increase of 132.6%) (p < 0.01) and LPS + Trigonelline40 (a decrease of 27.2%) (p < 0.05) and LPS + Trigonelline80 (a decrease of 31.1%) (p < 0.05) groups had a lower AChE activity as compared to LPS group. In addition, dexamethasone did not exert a significant impact in this regard.

4. Discussion

The findings of this study showed that trigonelline could dose-dependently ameliorate LPS-induced cognitive dysfunctions via suppression of hippocampal oxidative stress and neuroinflammation and appropriate modulation of NF-κB/TLR4 and AChE activity.

Oxidative stress is as a result of an imbalance in pro-oxidant/antioxidant homeostasis and is considered as a pivotal pathogenic process for a variety of human disorders including AD [62, 63]. This imbalance may be increased free radicals production or decreased activity of antioxidant defensive system. In our research study, following LPS challenge, hippocampal level of MDA as a reliable indicator of lipid peroxidation elevated, clearly denoting raised oxidative stress in the hippocampus. This finding was in agreement with previous reports [64]. In addition, we observed depressed activity of catalase and SOD and a lower level of GSH in the hippocampus in LPS group. These findings are also consistent with previous studies [16]. Part of cognitive deficits in different behavioral tasks in our study in LPS-challenged groups could be attributed to enhanced oxidative stress in those regions of the brain related to learning and memory processes. In this respect, it has been shown that oxidative stress burden at different regions of the brain including hippocampus, frontal cortex, and cerebellum could inappropriately affect learning and memory processes in overweight mice [65]. Also, it has been suggested that enhanced oxidative stress interferes with the functional roles of the hippocampus and leads to cognitive decline and antioxidant supplementation could improve cognitive abilities through protection of the hippocampus against destructive effects of reactive oxygen species [66]. In our study, trigonelline was able to enhance antioxidant defense system in LPS-challenged group. Consistent with our findings, it has been shown that trigonelline could decrease oxidative stress biomarkers in high-fat high-fructose diet-induced insulin resistance in rats [67] and in type 2 diabetic Goto-Kakizaki rats [68]. Of related significance, Mirzaie et al. in 2016 have also reported that trigonelline could ameliorate oxidative stress in 6-
hydroxydopamine model of Parkinson's disease in the rat [69]. Since trigonelline was capable to ameliorate hippocampal oxidative stress and to augment antioxidant defense system elements, part of its beneficial effect in prevention of cognitive decline in LPS-challenged groups could be explained in this way.

Development of neuroinflammation is associated with cognitive deficits [70, 71]. LPS administration in experimental animals leads to development of neuroinflammation and in this way produces learning and memory decline [16, 34]. Cognitive decline is also noticeable in neurodegenerative disorders such as Alzheimer's disease and some diseases associated with brain inflammation [72]. LPS challenge elevates inflammatory markers including TNF-α [73]. NF-κB is also over-expressed after LPS challenge, TNFα, and elevated level of free radicals [74–76]. LPS is also reported as an effective activator of macrophages and microglial cells via TLR4 cascade, finally leading to inflammation [77]. TLR4 signaling negatively affects hippocampus-dependent cognition [78] with subsequent learning and memory decline, as was also observed in our research study. Suppression of neuroinflammation using natural products could alleviate cognitive deficits following LPS challenge [16]. It has recently been shown that trigonelline pretreatment could lower inflammatory biomarkers like TNF-α and IL-6 in both hippocampal and cortical regions following LPS in mice [59]. No evidence is still available on the potential impact of trigonelline on TLR4 signaling. However, trigonelline could have exerted its anti-inflammatory effect by modulating NF-κB and TLR4 signaling in our study.

Brain cholinergic system also plays a key role in learning and memory processes [79]. Part of cognitive dysfunction in inflammatory diseases is related to deranged cholinergic transmission [80, 81]. There are conflicting evidences on the impact of LPS-induced inflammation on AChE activity, some researches demonstrating its increase [80, 82] and some showing its depression [83, 84]. According to our data, we had an elevated activity of AChE in the hippocampus subsequent to LPS and trigonelline ameliorated AChE activity. In support of our finding, AChE inhibitory potential of trigonelline has already been reported [30].

To conclude, trigonelline could attenuate LPS-induced cognitive decline via suppression of hippocampal oxidative stress and inflammation and appropriate modulation of NF-κB/TLR4 and AChE activity and this may be put forward as an ancillary therapeutic agent to lower cognitive decline in inflammation-related dysfunctions.

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Conflict of interest

The authors declare that they have no competing interests.

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