Research Article

Pharmacological evaluation of combined neuroprotective effect of Melatonin and Simvastatin against LPS and STZ induced memory impairment in rodents

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Abstract

Objective: To study the synergetic effect of Melatonin and Simvastatin on brain of albino mice of Swiss strain. **Materials and methods:** The neuroprotective activity was studied in LPS and STZ induced learning and memory deficit model. In this combined neuroprotective effect of melatonin and simvastatin against LPS and STZ was investigated the synergistic effect. Neuroprotective activity was evaluated by comparing behavioural (learning and memory) and biochemical (GSH, MDA, Cholinesterases) parameters. **Results and discussion:** There was significant decrease in MDA level and AChE level, as compare to disease group in Melatonin (**p<0.001) and Simvastatin (*p<0.05), Melatonin + Simvastatin (***p<0.001) and significant increase in GSH level respectively, in LPS and STZ memory impairment model. There was significant decrease in escape latency time and no. of crossing behavior in all groups in comparison of session 1-5 except the disease group in LPS and STZ model. **Conclusion:** Data indicates that combined administration of melatonin and simvastatin improve the behavioral and biochemical parameters in comparison to the administration of their individual doses. Biochemical parameters showed significant decrease (**p<0.01) in MDA level, AChE level and increase in (***p<0.001) GSH level which is equivalent to standard drug.

Keywords: Neuroprotective, synergistic, intracerebroventricular, Simvastatin, Melatonin

Introduction

The progressive loss of structure or function of neurons, including death of neurons is collectively termed as neurodegeneration**.** Many neurodegenerative diseases such as amyotrophic lateral sclerosis, Parkinson's, Alzheimer's, and Huntington's occurs due to neurodegenerative processes (Mayeux et al., 2012; Przedborski et al., 2003). Loss of neurons and synapses in the cerebral cortex and certain subcortical regions is the characteristics of Alzheimer's disease. This loss of neurons and synapses developed gross atrophy in the affected regions, which include degeneration in the temporal lobe and parietal lobe, and parts of the frontal cortex and cingulate gyrus (Wenk et al., 2003; Zlokovic et al., 2005)**.** About 24 million of the world's population is affected with alzheimer's type dementia and this figure is predicted to become twice by the year 2040. As the global population continues to age, the number of individuals at risk will moreover increase, particularly among the very old. Alzheimer disease is the leading source of dementia with weak memory (Reitz et al., 2011)**.** An important subset of neurodegenerative disease deal with dementia associated with aging. The most common clinically recognised dementia in aging population is Alzheimer's disease (Blansjaar et al., 2000; Polvikoski et al., 2005). In United States 43% of people with age of 85 or older are thought to suffer from Alzheimer's. Another common nervous system disorder which also associated with the elderly is Parkinson's disease and it affects 1-3 % of the population over 60. After knowing the financial, societal and personal impact of the burden of these diseases, the major focus of basic and clinical research should be to determine their cause, prevention, and treatment. Pathophysiologies of many neurodegenerative diseases are related with genetic mutation and protein misfolding (Thomas et al., 2007)**.** Aggregation of misfolded proteins such as alpha-synuclein, Tau (hyperphosphorylated tau protein is the main component

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of neurofibrillary tangles in Alzheimer's disease), Beta amyloid (the major component of senile plaques in Alzheimer's disease) are well known and central factors in AD pathology (Grundke et al., 1986; Masters et al., 1985; Demattos et al., 2002). Melatonin, which plays an important role in circadian rhythm regulation, is highly potent endogenous free radical scavenger and antioxidant (Wu et al., 2005). On the basis of ability of melatonin (N-acetyl-5-methoxytryptamine) and its metabolites to scavenge a wide variety of free radicals, it is not surprising to consider it as one of its important functions in living organisms leading to protect them from oxidative stress. Acting as a direct scavenger, this neurohormone is able to remove free radicals, such as singlet oxygen, superoxide anion radical, hydroxyl radical and the lipid peroxide radical (Wu et al., 2003; Pappolla et al., 2000; Rosales et al., 2003; Tyagi et al., 2010). Statins are 3-hydroxy-3-methylglutaryl-coenzyme (HMG-CoA) reductase inhibitors, which reduce low-density lipoprotein (LDL) cholesterol levels by blocking the mevalonate pathway and increase LDL cholesterol receptor expression in the liver (Andersson et al., 1995; Moosemann et al., 2004; Kandiah et al., 2009; Solomon et al., 2009). Inhibition of HMG-CoA reductase activity in monocyte and rat mesangial cells treated with lipopolysaccharide (LPS) and granulocyte macrophage-colony stimulating factor reduced the production of IL-8, IL-6, and MCP-1, (monocyte chemotactic protein-1) responsible for leukocyte recruitment at the infection site (Van der Burgh et al., 2013). Although pathogenesis of most of the neurodegenerative diseases are still continuous topics of a common research all over the world, the principal mechanisms linked to these diseases is genetic mutations (Linnane et al., 1989) and intracellular mechanisms in specific brain regions (such as protein degradation pathways, misfolding or occurrence of abnormal protein structure) (Avila et al., 2004). Consequently, neuroprotection is an important treatment option for such neurodegenerative disorders. Ongoing research targeting a number of promising novel compounds has enormous potential to develop new therapeutics with excellent neuroprotective activity. Recent studies have reported that the treatment with statins, reduced the levels of beta-amyloid proteins in hippocampal or mixed cortical neurons from rats (Fassbender et al., 2001). The objective of this research is to evaluate the neuroprotective activity of the combined dose of melatonin and simvastatin in STZ and LPS induced memory impaired mice. In future, the combination therapy may prove to be a major weapon for combating against neurodegenerative disease.

Materials and methods

Experimental animals

Healthy Swiss albino mice (20-25g) were procured from the Laboratory Animal Services Division of Indian institute of toxicology and research centre (IITR), Lucknow, India. They were kept in polyacrylic cage and maintained under standard housing condition (room temperature 25±1 °C and humidity 60–65%) with 12: 12 light/dark cycle (8:00 a.m.–8:00 p.m.). Experiments were performed as per internationally followed ethical standards, after clearance from Institutional Animal Ethics Committee (IAEC) of Central Drug Research Institute which is approved from Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), Government of India on animal experimentation (IAEC Approval No. HIPER/IAEC/03/17/04).

Drugs and Chemicals

Lipopolysaccharides, streptozotocin, donepezil, melatonin, simvastatin were purchased from Sigma Aldrich (USA). Chemicals like TBA, TCA, DTNB, magnesium chloride, butryl cholinesterase potassium chloride were purchased from Merck (India), while magnesium chloride, calcium chloride, phosphate buffer, sodium hydroxide, ethanol, sodium carbonate, sodium tartrate, sodium dihydrogen phosohate were purchased from local commercial suppliers. All the chemicals used were of analytical grade.

Lipopolysaccharides (LPS) Induced Neuroinflammation

Lipopolysachharides induced inflammation model was validated by clinically used agent donepezil, melatonin, simvastatin. LPS was dissolved in vehicle (0.9% normal saline). Mice received LPS (250µg/kg bwt, i.p) consecutively for 7 days to induce chronic neuroinflammation (Lee et al., 2008). The behavioural parameters were assessed on the last 5 days with Morris water maze and T- maze apparatus. Mice were sacrificed at the end of behavioural studies.

Streptozotocin (STZ) Induced Neurodegenration

Neurotoxicity is induced by ICV injection of STZ (0.5 mg/kg) dissolved in 0.9% normal saline in bragma region. The bregma region was identified according to Laursen & Belknap (Laursen et al., 1986; Sharma et al., 2001), by rubbing the point of needle over the skull (approximately 1–3 mm rostral to the line drawn through anterior base of ears), then at 45^0 angle the needle was inserted 2 mm lateral to midline and STZ was injected. Mice received STZ on day 1^{st} and day 3^{rd} (Avinash et al., 2016; Awasthi et al., 2010). The behavioural parameters were assessed on the last 5 days by Morris water maze and T- maze apparatus. Mice were sacrificed at the end of behavioural studies.

Behavioural Assessment

Assessment of learning and memory by Morris water maze

Morris water maze test was used for the evaluation of spatial working memory. In the Morris Water Maze test, the animal learns to swim in a water tank, guided by external cues, and find (and climb up to) a submerged platform . This test was performed for the comparative assessment of the number of crossing over the platform and the escape latency time among the control and the treated groups (Dhingra et al., 2012; Bromley-Brits et al., 2011; Vorhees et al., 2006). Assessment of spatial working memory was made by confirming the number of crossing over the platform. This test was used to assess the combined effect of melatonin and simvastatin using STZ and LPS induced neuroinflammtion in mice. 24 hrs after 6 trials (two times per day for 3 days), mice were given LPS and STZ.

Assessment of Cognitive Function by T-Maze

The purpose of the T-Maze is to evaluate spatial memory, behavioural and cognitive functions of mice in a control vs. disease model/intervention groups, by observing their ability to remember which arm they have previously entered. This test can provide information regarding hippocampus-dependent learning, specifically spatial memory. Assessment of the cognitive functions was done by evaluating the % of alternations and time consumed in T- Maze by mice (Nikiforuk et al., 2018; Dhingra et al., 2012). This test was performed to evaluate the combined effect of melatonin and simvastatin using STZ and LPS induced neuroinflammtion in mice.

Biochemical analysis

Biochemical estimations in the brain homogenate were carried out 24 h after completion of all behavioral assessments.

Brain homogenate preparation

The mice were anaesthesized by urethane. The mice were decapitated and their brains were removed. The isolated brains were washed with chilled normal saline on the ice. 10% (w/v) homogenate of brain samples (0.03M sodium phosphate buffer. pH-7.4) was prepared by using a homogenizer at a speed of 3000 rpm for 15 min. The homogenized tissue preparations were used to measure acetylcholinestrase, malonaldehyde, reduced glutathione as per the following methods.

Estimation of lipid peroxidation levels

The MDA in the sample is reacted with thiobarbituric acid (TBA) to produce MDA-TBA_2 adduct (appear as a pink chromogen) is a frequently used assay used for MDA estimation. The MDA levels in the brain were quantified spectrophotometrically at 532 nm (Beckman DU-640 Spectrophotometer). The values were expressed as nmol/mg protein (Garcia et al., 2005; Gaweł et al., 2004; Colado et al., 1997).

Estimation of reduced glutathione level

The reduced glutathione (GSH) content in brain was analyzed according to the method described by Ellman. To 1 mL of tissue homogenate, 1ml of 4% sulfosalicylic acid and cold digested at 4°C for 1 hour. The samples were later centrifuged at 1200g for 15 min at 4°C. To 1 ml of this supernatant, 2.7 ml of phosphate buffer (0.1 mol/l, pH 8) and 0.2 ml of DTNB were added. The colour developed was measured immediately at 412 nm (Beckman DU-640 Spectrophotometer). The enzymatic activity in supernatant was expressed as µmol per mg protein (Bhatt et al., 2014; Singh et al., 2013; Beutler et al., 1963).

Estimation of AChE level

The quantitative estimation of acetylcholinesterase levels in brain was performed according to the method described by Ellman. To 0.05 mL of tissue homogenate, 3 mL of 0.01M sodium phosphate buffer (pH 8), 0.10 ml of acetylcholine iodide and 0.10 ml of DTNB were added. The change in absorbance was measured at 412 nm (Beckman DU-640 Spectrophotometer).The enzymatic activity in supernatant was expressed as µmol/mg protein (Singh et al., 2013; Voss et al., 1970; Ellman et al., 1961).

Randomization, grouping and dosing of animals in LPS induced neuroinflammation model

Animals were divided into six groups containing 6 animals each. All the animals were pretrained on days 6, 7, 8, 9, 10 and the behavioural parameters were assessed on days 18, 19, 20, 21, 22.

Normal control group (Group I): Mice in this group received vehicle (0.9% normal saline, i.p.) for 17 days.

Vehicle + LPS (Group II): Mice were treated with vehicle for 10 days. On the days 11, 12, 13, 14, 15, 16, 17 the mice were treated with consecutive doses of vehicle (0.9% normal saline) and LPS (250 µg/kg bwt, i.p.).

LPS + Donepezil (Group III): Mice were treated with vehicle for 10 days. On the days 11, 12, 13, 14, 15, 16, 17 the mice were treated with consecutive doses of LPS (250 µg/kg bwt, i.p.) and donepezil (5 mg/kg bwt, p.o.).

LPS+ Melatonin (Group IV): Mice were treated with vehicle for 10 days. On the days 11, 12, 13, 14, 15, 16, 17 the mice were treated with consecutive doses of LPS (250 µg/kg bwt, i.p.) and melatonin (10 mg/kg bwt, p.o.).

LPS+ Simvastatin (Group V): Mice were treated with vehicle for 10 days. On the days 11, 12, 13, 14, 15, 16, 17 the mice were treated with consecutive doses of LPS (250 µg/kg bwt, i.p.) and simvastatin (10 mg/kg bwt, p.o.). The behavioural parameters were assessed on days 18, 19, 20, 21, 22.

LPS+ Melatonin+ Simvastatin (Group VI): Mice were treated with vehicle for 10 days. On the days 11, 12, 13, 14, 15, 16, 17 the mice were treated with consecutive doses of LPS (250 µg/kg bwt, i.p.), melatonin (10 mg/kg bwt, p.o.) and simvastatin (10 mg/kg bwt, p.o.).

Randomization, grouping and dosing of animals in STZ induced memory impairment model

Animals were divided into six groups containing 6 animals each. The behavioural parameters were assessed consecutively on days 18, 19, 20, 21, 22.

Normal control group (Group I): Mice in this group received vehicle (0.9% normal saline, i.c.v) for 22 days.

Vehicle + STZ (Group II): On 1st and 3rd day, mice were infused with streptozotocin (0.5mg/kg bwt, i.c.v.) dissolved in 5µl of vehicle (0.9% normal saline) in bragma region (4th ventricle). STZ mice were administered vehicle (0.9% normal saline, i.p.).

STZ + Donepezil (Group III): On 1st and 3rd day, mice were infused with streptozotocin (0.5mg/kg bwt, i.c.v.) dissolved in 5µl of vehicle (0.9% normal saline) in bragma region (4th ventricle). STZ mice were administered donepezil (5 mg/kg bwt, p.o) for a period of 19 days.

STZ+ Melatonin (Group IV): On 1st and 3rd day, mice were infused with streptozotocin (0.5mg/kg bwt, i.c.v.) dissolved in 5µl of vehicle (0.9% normal saline) in bragma region (4th ventricle). STZ mice were administered Melatonin (10 mg/kg p.o) for a period of 19 days.

STZ+ Simvastatin (Group V): On 1st and 3rd day, mice were infused with streptozotocin (0.5mg/kg bwt, i.c.v.) dissolved in 5µl of vehicle (0.9% normal saline) in bragma region (4th ventricle). STZ mice were administered Simvastatin (10 mg/kg p.o) for a period of 19 days.

STZ+ Melatonin+ Simvastatin (Group VI): On 1st and 3rd day, mice were infused with streptozotocin (0.5mg/kg bwt, i.c.v.) dissolved in 5µl of vehicle (0.9% normal saline) in bragma region (4th ventricle). STZ mice were administered Melatonin (10 mg/kg p.o.) and Simvastatin (10 mg/kg p.o.) for a period of 19 days.

Statistical analysis

Results are expressed as mean ± S.E.M. Statistical analysis was done by one-way analysis of variance (ANOVA), The P value < 0.05 was considered statistically significant.

Results

Combined effect of melatonin and simvastatin on hippocampus-dependent learning and memory using Morris water maze and T- Maze apparatus

Morris water maze test was utilized to assess the combined

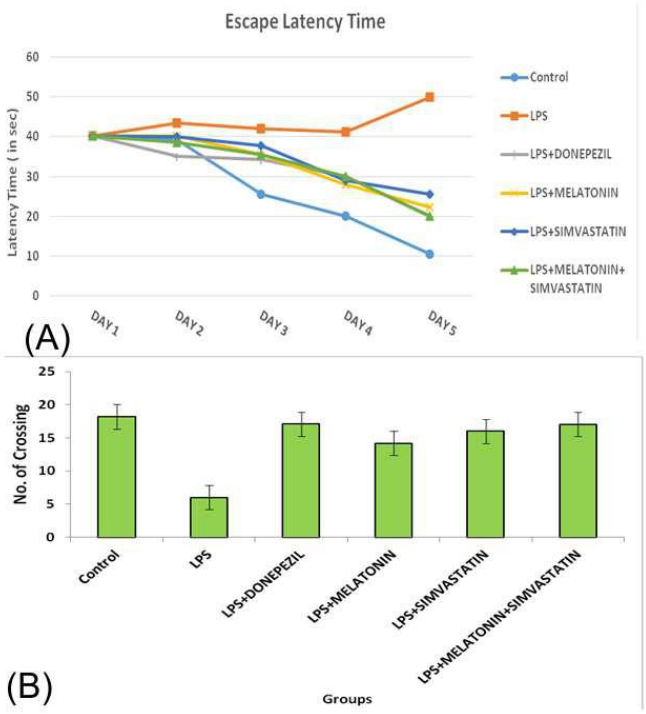

Figure 1. Effect of melatonin and simvastatinon (A) escape latency in training trial and probe trial session and (B) the number of crossing platform area for LPS induced memory deficit mice. Values are expressed as mean±SEM. (n=6) significant values were compared with ***p<0.001 vs LPS treated group, ### p<0.001 vs. control group.

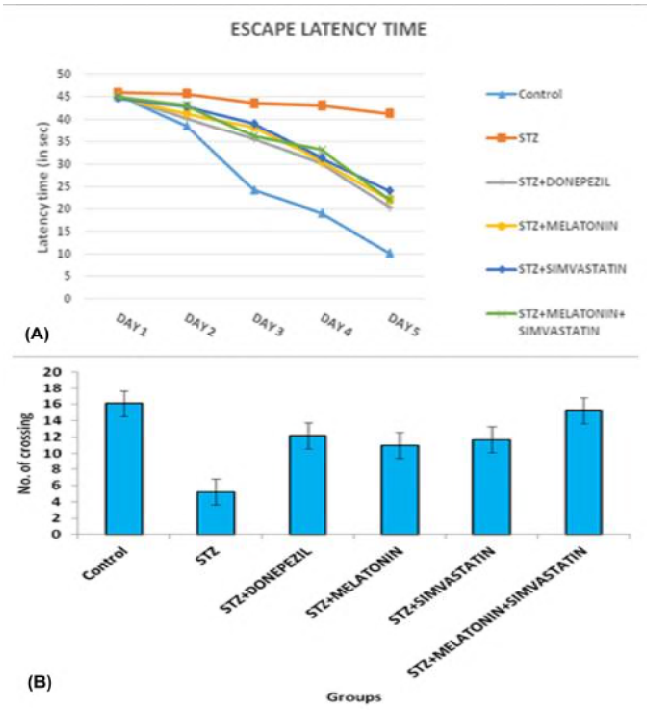

Figure 2. Effect of melatonin and simvastatin on (A) escape latency in training trial and probe trial session and (B) the number of crossing platform area for STZ induced memory deficit mice. Values are expressed as mean±SEM. (n=6) significant values were compared with ***p<0.001 vs. STZ treated group, ### p<0.001 vs control group.

effect of melatonin and simvastatin using STZ and LPS induced neuroinflammtion in mice. The escape latency was significantly increased in STZ (Figure 1A) and LPS (Figure 2A) treated groups as compared to control groups in all trial sessions. This confirmed that STZ and LPS treatment impaired the memory in mice. The escape latency was significantly decreased in donepezil (5mg/kg) and melatonin (10 mg/kg) + simvastatin (10 mg/kg) treated groups as compared to STZ and LPS group in the probe trail and final sessions. Assessment of spatial working memory was made by confirming the number of crossing over the platform. The number of crossing was significantly decreased in STZ (Figure 1B) and LPS (Figure 2B) treated groups while melatonin + simvastatin (10 mg/kg + 10 mg/kg) significantly increased the number of crossing over the platform. donepezil also significantly increased the number of crossing over the platform. Melatonin and simvastatin alone also showed the significant neuroprotection but were found less effective as compared to melatonin and simvastatin treated group. Animals were further evaluated for neurochemical analysis.

Results of cognitive function tests in T-maze in LPS and STZ induced neuroinflammation

Working memory evaluated by measuring the rate of spontaneous alternations. % of alternations was significantly reduced in LPS (Figure 3A) and STZ (Figure 4A) induced mice as compared with the control ($p<0.001$). Melatonin and simvastatin treatment significantly improved this task to be

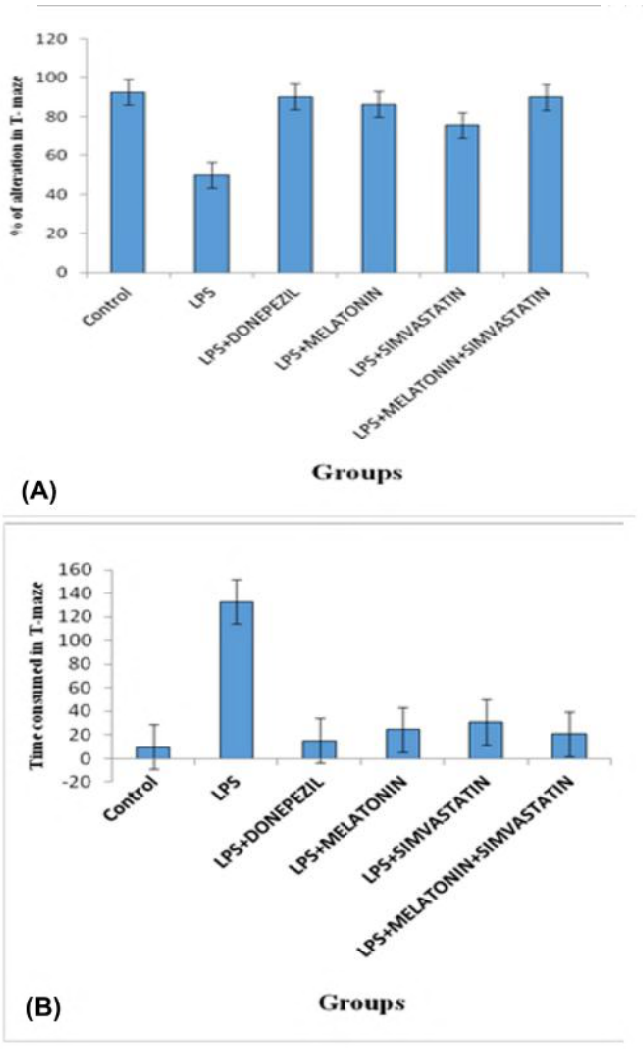

Figure 3. Effect of melatonin and simvastatin treatment on the % of alternations in T-maze (A) and time consumed in T- Maze (B) in mice of LPS induced neuroinflammation. Values are expressed as mean±SEM. (n=6) significant values were compared with ***p<0.001 vs LPS treated group, ### p<0.001 vs control group.

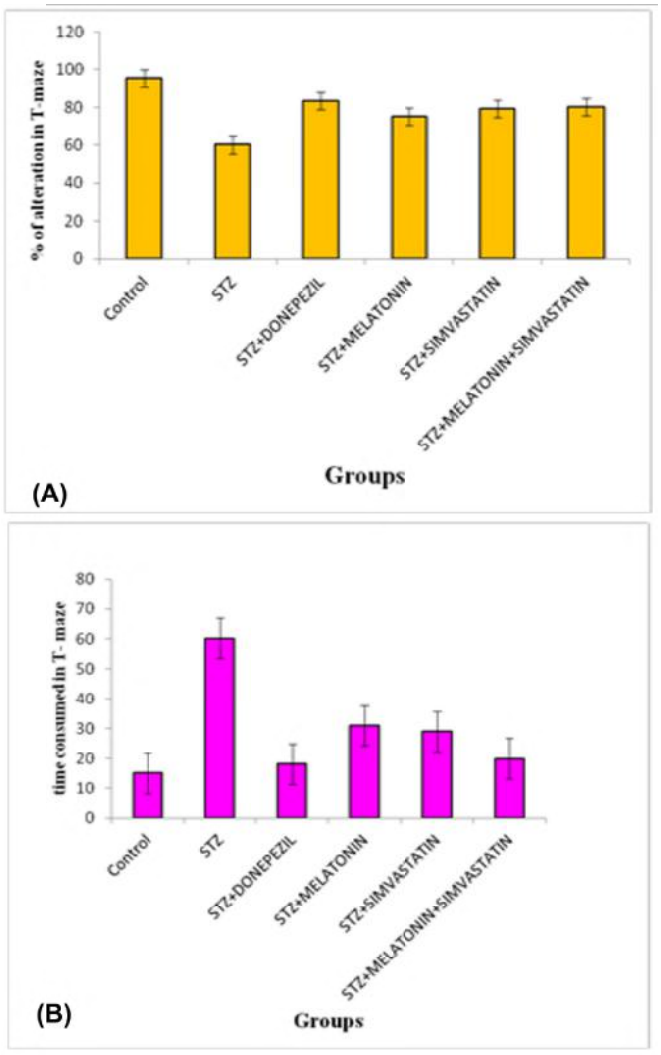

Figure 4. Effect of melatonin and simvastatin treatment on the % of alternations in T-maze (A) and time consumed in T-Maze (B) in mice of STZ induced neuroinflammation. Values are expressed as mean±SEM. (n=6) significant values were compared with ***p<0.001 vs. STZ treated group, ### p<0.001 vs control group.

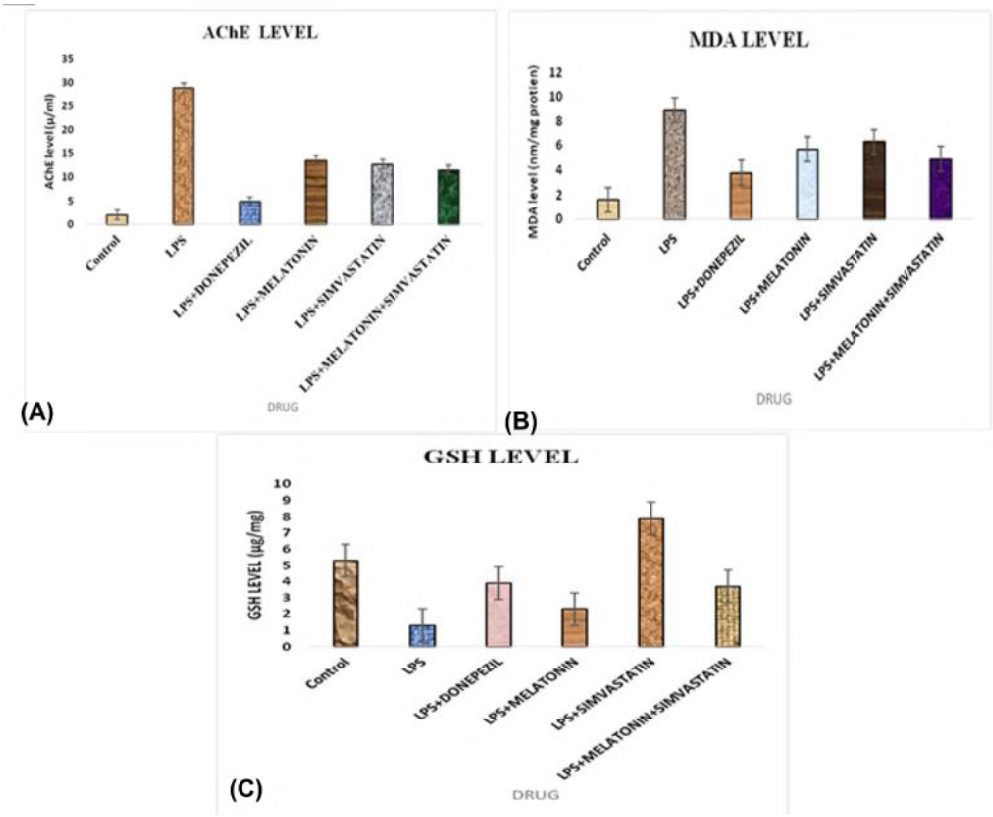

Figure 5. Ex vivo anticholinesterase and antioxidant properties of melatonin and simvastatin. AChE and MDA level significantly increased and GSH level was significantly reduced in LPS treated group (A, B, C,). Combined treatment with melatonin and simvastatin significantly decreased acetylcholinesterase and MDA and increased GSH level (A, B, C) as compared to LPS treated group. Values are expressed as mean ± SEM. (n=6) significant values were compared with ***p<0.001 vs. LPS treated group, **p<0.01 vs. LPS treated group, *p<0.05 vs. LPS treated group, ###p<0.001 vs. control group.

comparable with that of the control. This indicated that melatonin and simvastatin can relatively improve working memory. However, time consumed in T- maze was shown to be significantly increased in LPS (Figure 3B) and STZ (Figure 4B) induced mice compared with the control group (p<0.001). This indicating prolonged latency in performance, which might be caused by generally altered cognition. Although it was significantly decreased in the melatonin and simvastatin treated mice (p<0.001), but it was still significantly more than that of the control animals (p<0.001).

Combined anticholinestrase and antioxidative effects of melatonin and simvastatin in STZ and LPS induced memory impaired mice

AChE level was significantly reduced as well as oxidative parameters were significantly modified (decreased MDA level and increased GSH level) by melatonin and simvastatin. To evaluate whether melatonin and simvastatin exerts better inhibitory effect on AChE activity than alone, Brain tissue of mice treated with melatonin + simvastatin were subjected to colorimetric estimation to establish the AChE activity in the brain. Melatonin + simvastatin reduced AChE level to 9.05±0.25 µmol/ml (p<0.01) as compared to STZ (Figure 5A) and LPS (Figure 6A) treated groups (16.41±0.48 and 28.48±0.93) µmol /ml. This data authenticated the fact that oral dose of melatonin + simvastatin significantly reverted AChE level which was increased by STZ and LPS respectively, thus confirming its anticholinestrase activity. The combined effect of melatonin and simvastatin were analysed on oxidative parameters. The result showed that STZ and LPS treatment significantly (p<0.001) increased the brain MDA level (8.69±0.13 and 8.95±0.10) nmol/mg proteins compared to the control group (1.59±0.32 and 1.60±0.31) nmol/mg proteins. Treatment with standard drug (donepezil) and melatonin + simvastatin significantly (p<0.001) reduced the brain MDA level. Donepezil (3.55±0.38 and 3.80±0.13) and melatonin+ simvastatin (4.95±0.38 and 4.90±0.38) nmol/mg proteins] compared to corresponding STZ (Figure 5B) and LPS (Figure 6B) treated groups. Further STZ and LPS treatment significantly (p<0.001) decreased the brain GSH levels (1.45±0.28 and 1.30±0.26) µmol/mg protein compared to the respective control drugs (5.84±0.31 and 5.30±0.04) µmol/mg proteins. Treatment with standard drug donepezil and the melatonin + simvastatin significantly (p<0.001) elevated brain GSH [donepezil (4.02±0.06 and 3.90±0.12)

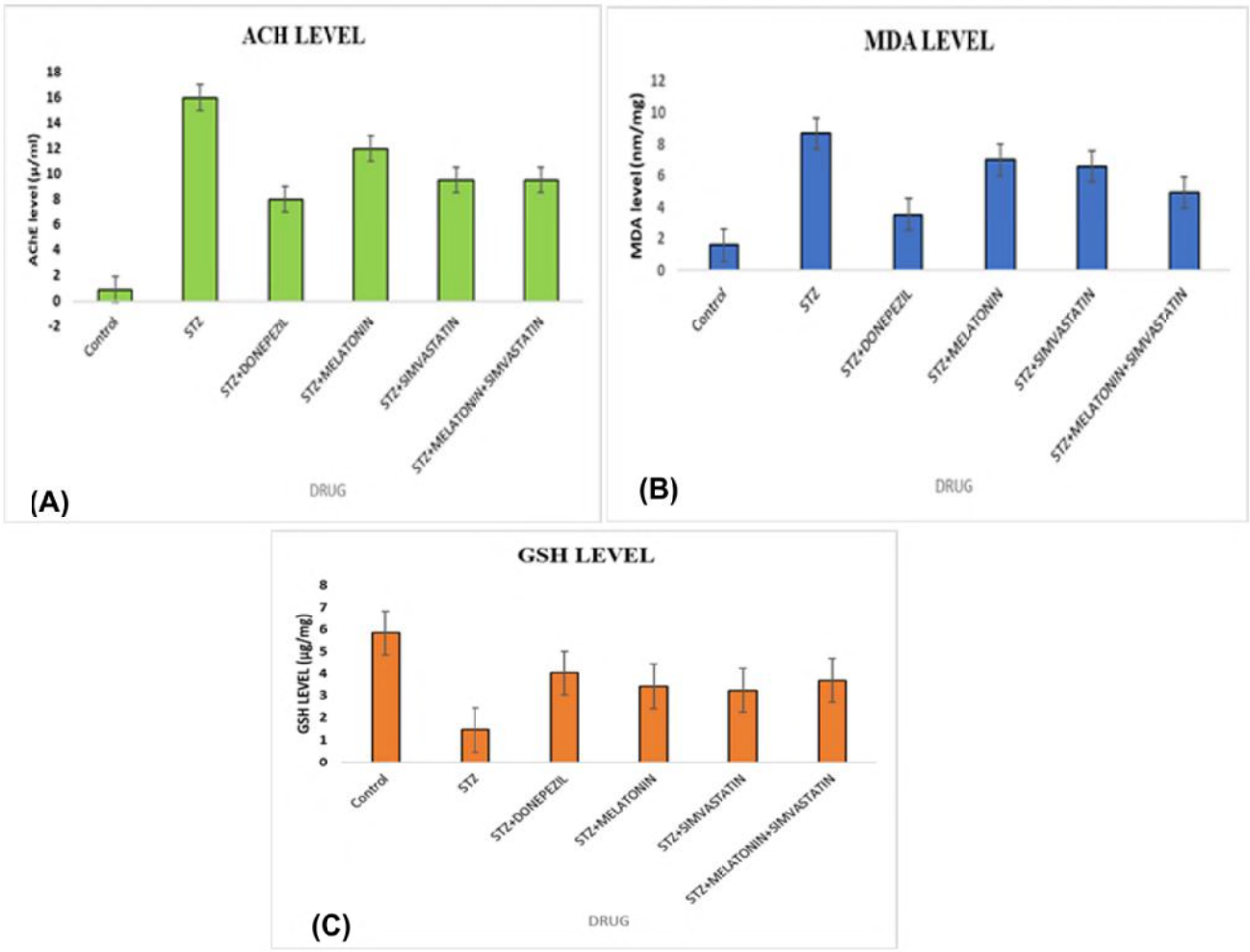

Figure 6. Ex vivo anticholinesterase and antioxidant properties of melatonin and simvastatin. AChE and MDA level significantly increased and GSH level was significantly reduced in STZ treated group (A, B, C,). Combined treatment with melatonin and simvastatin significantly decreased acetylcholinesterase and MDA and increased GSH level (A, B, C,) as compared to STZ treated group. Values are expressed as mean ± SEM. (n=6) significant values were compared with ***p<0.001 vs STZ treated group, **p<0.01 vs. STZ treated group, *p<0.05 vs STZ treated group ###p<0.001 vs. control group.

and melatonin + simvastatin (3.69± 0.12 and 3.70± 0.13) µmol/mg proteins] compared to corresponding STZ (Figure 5C) and LPS (Figure 6C) treated groups respectively. Melatonin and simavastatin alone also showed the significant anticholinestrase and antioxidative mechanism but were found less effective as compared to melatonin + simvastatin treated group. These results revealed the combined anticholinestrase and antioxidant properties of melatonin and simvastatin.

Discussion

The present study observed the combined effect of melatonin + simvastatin on memory impairment, oxidative stress, cholinergic dysfunction and protein levels in intracerebral (i.c) STZ and intraperitoneal LPS injected mice. Pre-treatment with melatonin + simvastatin in STZ and LPS administered mice, improved spatial memory. Furthermore, melatonin + simvastatin also improve the decrease of antioxidant (GSH), increase of lipid peroxide (MDA) and the cholinergic dysfunction in the mice brain following STZ and LPS injection.Sporadic Alzheimer type dementia can he examined by using an appropriate animal model viz. the intracerebroventricular (i.c.v.) STZ model. Present study showed that administration of STZ in mice caused a persistent memory deficit as evidenced by significant increase in escape latency time in Morris water maze test. This finding authenticated the previous experiments reporting memory impairment by STZ in rodents. Administration of melatonin + simvastatin significantly attenuated STZ induced memory impairment as demonstrated by reduced latency to reach platform and increased number of crossings across the platform. There was significant correlation in between latency time and number of crossing of all the groups in all sessions demonstrating direct alteration in both the parameters. It is familiar that mitochondria prompt oxidative stress generates free radicals which have been associated in neuronal damage

during AD. Free radical facilitated injury leads to lipid peroxidation that produces a number of secondary products like MDA by damaging the membranes. This elevated free radical production additionally prompts diminished antioxidant enzymes such as GSH. In the current study, STZ produced oxidative stress as demonstrated by significant rise in MDA level and reduction in GSH level signifying that STZ prompted learning and Memory impairment is linked with oxidative stress in rodents. An essential part has been played by cholinergic neuronal framework in the cognitive deficits connected with aging and neurodegenerative disorders. Previous report showed that there was a significant decline in the quantity of muscarinic binding sites acetylcholine (Ach) level in the brains of AD patients. Acetylcholinesterase (AchE) degrades Ach level which is essential for appropriate working of cholinergic transmission to control learning and memory processes. Symptomatic treatment of AD has been achieved by inhibiting AChE through there inhibitors. In the present study, we established that STZ treatment significantly increased AChE level in mice. This result is similar with the precious reports demonstrating significant increase in AChE activity and expression following treatment with STZ. Furthermore, administration of melatonin + simvastatin significantly attenuated STZ induced memory impairment as demonstrated by reduced AChE level similar to the standard drug donepezil. Besides it was additionally stated that LPS induces memory impairment when administered intraperitoneally in mice. However, the underlying mechanism behind LPS induced memory impairment is still unclear. In our findings LPS significantly induces memory impairment as evidenced by increased latency time and decreased number of crossings across the platform. Furthermore, pre-treatment with melatonin + simvastatin significantly attenuated LPS induced memory impairment as demonstrated by significantly decreased latency time and increased number of crossing across the platform. This suggested the combined neuroprotective role of melatonin and simvastatin in LPS induced memory impairment. Previous reports also showed that LPS treatment significantly increased MDA and decreased GSH in rodents. Similarly, in the present study LPS significantly increased brain MDA and decreased GSH in mice. However, pre-treatment with melatonin and simvastatin significantly attenuated LPS induced oxidative stress parameters as demonstrated by significant decrease in MDA and increase in GSH levels in LPS treated mice. This validated the neuroprotective and antioxidant properties of melatonin and simvastatin.

Conclusion

The data obtained from the current research clearly indicates that the combination therapy involving melatonin and simvastatin may be used as a potential therapeutic approach that may block this disease pathway and improve pathology in alzheimer's type dementia. However, further studies can be done to understand the mechanisms which can enhance the synergistic action and therapeutic efficacy associated with the combination therapy.

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Conflict of Interest

None

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