CHAPTER VIII
EFFECT OF LASER ACUPUNCTURE AT "HT7" ON MOTOR AND MEMORY DEFICIT IN ANIMAL MODEL OF PARKINSON'S DISEASE

1. Introduction

Parkinson's disease (PD), an important age-related neurodegenerative disorder, disturbs motor and memory impairments of millions of people worldwide. It produces a great impact on the quality of life of patients and caregivers as well as annual healthcare cost (Muslimovic *et al.*, 2005; Svenningsson *et al.*, 2012; Vossius *et al.*, 2011). Parkinson's disease (PD) is primarily considered as a movement disorder defined by the presence of motor symptoms (Alves *et al.*, 2008). Besides motor features, patients attacked by Parkinson's disease also show other neurological symptoms which cause a considerable burden to the patient especially cognitive deficit. Cognitive deficit, a common non-motor feature of Parkinson's disease (PD), has been reported that approximate one-fifth of newly diagnosed PD patients develop the fulfilled clinical criteria for mild cognitive impairment (PD-MCI) (Aarsland *et al.*, 2009) and around one-sixth develop dementia after 5 years (Williams-Gray *et al.*, 2009).

To date, the exact mechanisms of motor and cognitive impairment in PD are still unclearly known. However, recent substantial evidence has demonstrated that cholinergic and dopaminergic systems play the crucial roles on the pathophysiology of motor and cognitive deficit in PD (Dubois *et al.*, 1987; Sriraksa *et al.*, 2011). It has been demonstrated that oxidative stress pay the crucial roles on both memory deficit (Sriraksa *et al*, 2011) and motor impairment in Parkinson's disease (Ebadi *et al*, 1996). The current pharmacological interventions against both motor and non-motor symptoms of Parkinson's disease are still not in satisfaction level and the novel effective therapeutic strategy is still required.

Acupuncture has been long term used for treating various disorders including neuropsychological disorders. HT7, an acupoint located at the ulnar end of the transverse crease of the writs, in the depression on the radial side of the tendon muscle flexor carpi ulnaris, has been long-term used for treating many neuropsychological impairments such as amnesia, insomnia, mania, epilepsy, stupor. In addition, it also regulates the physical response to emotional stimuli such as anxiety, fear and panic (Hecker and Steveling, 2011; MacPherson, 2007). TE5, an acupoint located at the posterior aspect of the forearm, midpoint of the interosseous space between the radius and the ulna, is also claimed for the central nervous system effect. It has been used for treating headache, stroke-related motor, neurological and autonomic nerve problems in clinical practice (Maciocia and Ying, 1994). However, it has been clearly demonstrated that the single acupoint stimulation only at HT7 can produce significant neuroprotective activity against the neuronal impairment and memory dysfunction induced by corticosterone, a stress hormone partly via the improved cholinergic function (Lee *et al.*, 2011) . The stimulation acupoint can occur not only via needle stimulation but also via laser stimulation (Baxter *et al.*, 2008; Gao *et al.*, 2012; Litscher, 2008; Litscher, 2009; Litscher, 2009; Litscher, 2010). Therefore, various wavelengths of laser have been implemented in medicine for various purposes. It is believed that the stimuli must elicit “De Qi” which involves the stimulation and transmission of mechanical signal to connective tissue cells via mechanotransduction (Langevin *et al.*, 2001). Since most of mechanoreceptors are located in the dermis especially at the superficial area of this layer and the skin of the rats did not contain abundant of pigment cells, the stimulation with laser with low penetration power such as blue laser at the wave length of 405 nm is enough to stimulate this group of receptor (Whittaker, 2004). Moreover, it has been reported that the stimulation at HT7 acupoint either via manual or via laser can improve spatial memory impairment in various animal models (Lee *et al.*, 2011; Sutalangka *et al.*, 2013). In addition, laser acupuncture at HT7 acupoint also improves neurodegeneration, and cholinergic function in hippocampus (Ferro *et al.*, 2005). Based on the these pieces of information, the beneficial effect of laser acupuncture at HT7, a non-invasive intervention, on Both motor and non-motor symptoms such as cognitive deficit condition in Parkinson’s disease has been considered. To the best of our knowledge, no scientific evidence concerning this issue is available until now. Thus, this study aimed to determine the effect of laser acupuncture at HT7 acupoint on motor and memory impairment, oxidative stress status and the function of both cholinergic and dopaminergic systems in 6-OHDA lesion rat, a validated animal model of Parkinson’s disease.

2. Materials and Methods

2.1 Animals

Young adult male Wistar rats, 8 weeks old, were used as experimental animals. They were obtained from National Laboratory Animal Center, Salaya, NakornPathom. The weights of the animals on the first day of experiment are 180-220 grams. They were housed 6 per cage and maintained in 12: 12 light: dark cycle and given access to food and water ad libutum. The experiments were performed to minimize animal suffering and the experimental protocols were approved by the Institutional Animal Care and Use Committee KhonKaen University, Thailand (AEKKU 41/2554). All treatments in this study were performed once daily between 8.00 a.m.-5.00 p.m.

2.2 Drugs and Chemicals

6-Hydroxydopamine hydrochloride (6-OHDA) was purchased from Sigma–Aldrich Co., USA. Sodium pentobarbital was obtained from Jagsonpal Pharmaceuticals LTD, Haryana, India. All other chemical substances were analytical grade and purchased from Sigma Chemical Company, St. Louis, MO.

2.3 Experimental Protocol

All rats were randomly assigned to 4 groups of 12 animals each as following:

Group I Control group; rats in this group were exposed to sham operation and received no treatment (no laser treatment).

Group II 6-OHDA: rats were induced partial lesion in substantia nigra by injecting 6-OHDA into the right substantia nigra.

Group III 6-OHDA+sham acupoint+laser: rats received the administration of 6-OHDA into the right substantia nigra and laser acupuncture treatment at non- acupoint.

Group IV 6-OHDA+HT7+laser: rats received the administration of 6-OHDA into the right substantia nigra and laser acupuncture treatment at HT7 acupoint.

Rats had been treated with laser acupuncture once daily for 14 days after the administration of 6-OHDA. Then, they were assessed spatial memory using Morris water maze test at 1-day, 7-day and 14-day periods after 6-OHDA injection.

At the end of experiment, half of the rats were used for the histological study whereas the other half of the rats were used for biochemical assays including the determinations of oxidative stress markers, acetylcholinesterase (AChE) activity and monoamine oxidase type B (MAO-B) activity in both hippocampus and striatum. To determine the oxidative stress, AChE and MAO-B, Hippocampus and striatum of each rat were isolated and prepared as homogenate. Then the determination s of oxidative markers including malondialdehyde (MDA) level and the activities of superoxide dismutase (SOD), catalase (CAT) and glutathione peroxidase (GSH-Px) enzymes were performed. At the end of study, the neuron density in hippocampus and the density of dopaminergic neurons in substantia nigra were also carried out.

2.4 Substantianigra lesion

The animals were anesthetized by intraperitoneal injection of sodium pentobarbital (Jagsonpal Pharmaceuticals LTD, Haryana, India) at dose of 60 $mg.kg^{-1}$ BW. Each animal was mounted on a stereotaxic stand, the skin overlying the skull was cut to expose the skull, and the coordinates for the substantia nigraparcompacta (SNpc) were accurately measured (anteroposterior -0.5 mm from bregma, mediolateral 2.1 mm from midline and dorsoventral -7.7 mm from the skull). Total 6 μg of 6-OHDA was dissolved in 2 μl 0.2% ascorbic acid saline (Ferro *et al.*, 2005) and were perfused into SNpc through a 30 gauge stainless needle. After the surgery, animals were allowed to recover from anesthesia and then placed in their cages.

2.5 Laser acupuncture treatment

Fifteen minutes before laser acupuncture treatment, all rats were anesthetized with sodium pentobarbital (40 $mg.kg^{-1}$, i.p.) to minimize stress. Laser acupuncture treatment via HT7 acupoint was performed once daily for 14 days. The rats were treated with a laser instrument that operated with a continuous blue laser beam at wavelength of 405 nm, output power 100 mW and a spot diameter of 500 μm at HT7 acupoint (the transverse crease of the wrist of the forepaw, radial to the tendon of the muscle flexor carpi ulnaris) or at 2–4 mm lateral to the HT7 acupoint for 10 minutes (Litscher *et al.*, 2010; Wang *et al.*, 2011; Yoon *et al.*, 2010).

2.6 Determination of spatial memory

Spatial memory was evaluated via the Morris water maze. The water maze consists of a metal pool (170 cm in diameter × 58 cm tall) filled with tap water

(25 ºC, 40 cm deep). The pool was divided into 4 quadrants (Northeast, Southeast, Southwest, and Northwest). The water surface was covered with non-toxic milk. The removable platform was placed below the water level at the center of one quadrant. For each animal, the location of the invisible platform was placed at the center of one quadrant and remained there throughout training. The times which animals spent to climb on the hidden platform were recorded as escape latency. In order to determine the capability of the animals to retrieve and retain information, the platform was removed 24 hr later and the rats were released into the quadrant diagonally opposite to that which contained the platform. Time spent in the region that previously contained the platform was recorded as retention time (Brandeis *et al.*, 1989).

2.7 Determination of motor function

After the recovery from the last laser acupuncture treatment, in apomorphine-induced rotation behavior (0.5 mg/kg, s.c.) were assessed using acylindrical container. The diameter of the cylindrical container used for measuring rotational activity was 40 cm. The animals were habituated in the cylindrical container for 10 min and the rotations made by the animals to either side was counted for 45 min. The net number of rotations was counted as follows: the number of contralateral,rotations−the number of ipsilateral rotations with respect to the 6-OHDA injection side. This behavioral test was performed by a blind observer (Ahmad *et al.*, 2005; Yu *et al.*, 2010).

2.8 Histological procedure

After the anesthesia with sodium pentobarbital (60 mg/kg BW), brains were subjected to transcardial perfusion with fixative solution containing 4% paraformaldehyde in 0.1 M phosphate buffer pH 7.3. After the perfusion, brains were removed and stored over a night in a fixative solution that used for perfusion. Then, they were infiltrated with 30% sucrose solution at 4° C. The specimens were frozen rapidly and 10 μM thick sections were cut on cryostat. The selected sections were rinsed in the phosphate buffer and picked up on slides coated with 0.01 % of aqueous solution of a high molecular weight poly L-lysine.

2.9 Immunohistochemical evaluation

Immunohistochemical staining to investigate dopaminergic neuron was performed using the monoclonal anti-tyrosine hydroxylase antibody produced in mouse (Sigma, St Louis, MO, USA) and a modification of a previously described protocol employing the DAKO Strept ABC Complex/HRP duet kit. In brief, the sections were eliminated endogenous peroxidase activity by 0.5% H_2O_2 in methanol. Sections were washed in running tap water and distilled water for 1 min. each, then rinsed in KPBS and KPBS-BT for 5 minutes per each process. Excess buffer was removed, then incubated for 30 min in a blocking solution composed of 5% normal goat serum in KPBS-BT. The sections were then incubated in mouse monoclonal anti-tyrosine hydroxylase antibody diluted 1: 400 in KPBS-BT at room temperature for 2 hours and then incubate at $4^{o}C$ for 48 hours. The tissue was rinsed in KPBS-BT (two washes x 7 min), incubated for 1 hours in biotinylated goat anti-mouse IgG antibody, rinsed in KPBS-BT (two washes x 7 min) and then incubated in Strep ABC Complex/HRP for 4 hours.According to the preparation for visualization step, sections were rinsed inKPBS-BT (1 min), and KPBS (two washes x 10 min). Tyrosine hydroxylaseimmunoreactivity was visualized using 0.025% 3, 3’ diaminobenzidine (DAB, Sigma) and 0.01% H_2O_2 for 48 hours. Finally, sections were rinsed in running tap water, air dried and cover-slipped using permount.

2.10 Morphological analysis

Five coronal sections of each rat in each group were studied quantitatively. Neuronal counts in hippocampus and substantia nigra were performed by eye using a 40x magnification respectively with final field 255 μm^2. The observer was blind to the treatment at the time of analysis. Viable stained neurons were identified on the basis of a stained soma with at least two visible processes. Counts were made in five adjacent fields and the mean number extrapolated to give total number of neurons per 255 μm^2. All data were presented as number of neurons per 255 μm^2.

2.11 Determination of acetylcholinesterase and monoamine oxidase-B activities

The rats were divided into various groups as previously described in the experimental protocol. At the end of experiment, all rats were sacrificed. The hippocampusand striatum were isolated and prepared ashomogenate and used for the determination ofAChE and MAO-B activities. The activities of AChE and MAO-B were determined by using the colorimetric method (Ellman *et al.*, 1961; Holt *et al.*, 1997).

2.12 Determination of oxidative stress markers

Tissue samples of each rat were weighed and homogenized with a buffer consisting of 10 mM sucrose, 10 mMTris–HCl and 0.1 mM EDTA (pH 7.4). Then, a hippocampal homogenate was centrifuged at 3000 g at 4 °C for 15 min. The supernatant was separated and used for the determination of oxidative stress markers including malondialdehyde (MDA) level, and the activities of superoxide dismutase (SOD), catalase (CAT) and glutathione peroxidase (GSH-Px). MDA level was assessed via thiobarbituric acid reactive substances (TBARS) (Ohkawa *et al*., 1979). The activity of SOD was determined using a xanthine/xanthine oxidase system for the production of superoxide radical and subsequent measurement of cytochrome *c* as a scavenger of the radicals. Optical density was determined using a spectrometer (UV-1601, Shimadzu) at 550 nm (McCord and Fridovich, 1969). SOD activity was presented as units per milligram of protein (U mg^{-1}protein). One unit of enzyme activity was defined as the quantity of SOD required to inhibit the reduction rate of cytochrome *c* by 50%. CAT activity in the supernatant was measured by recording the reduction rate of H_2O_2absorbance at 240 nm (Goldblith and Proctor, 1950). The activity of CAT was expressed as μmol $H_2O_2.min^{-1}mg^{-1}$ protein. GSH-Px was determined using *t*-butyl hydro peroxide as a substrate. The optical density was spectrophotometrically recorded at 340 nm and expressed as U/mg protein (Eyer and Podhradský, 1986). One unit of the enzyme was defined as micromole (μmol) of reduced nicotinamide adenine dinucleotide phosphate (NADPH) oxidized per minute.

2.13 Statistical analysis

Data were expressed as means ± S.E.M. and analyzed statistically by one-way ANOVA, followed by Post-hoc (LSD) test. The results were considered statistically significant at p-value < 0.05.

3. Results

3.1 Effect of laser acupuncture at HT7 on spatial memory of 6-OHDA lesion rats

Figure 8-1 showed that the administration of 6-OHDA into right substantia nigra significantly enhanced escape latency (p-value<.001 all; compared to control group) but decreased retention time (p-value<.001 and .01 respectively; compared to control group) at 7-day and 14-day period after the 6-OHDA. It was found that sham laser acupuncture failed to improve the alteration of both escape latency and retention time induced by 6-OHDA. Interestingly, laser acupuncture at HT7 acupoint significantly improved the reduction of escape latency induced by 6-OHDA both at 7-day and 14-day intervention period (p-value<.05 and .001 respectively; compared to sham laser acupuncture group) while it showed the significant increase in retention time in 6-OHDA lesion rats only at 14-day day intervention period (p-value<.05; compared to sham laser acupuncture group).

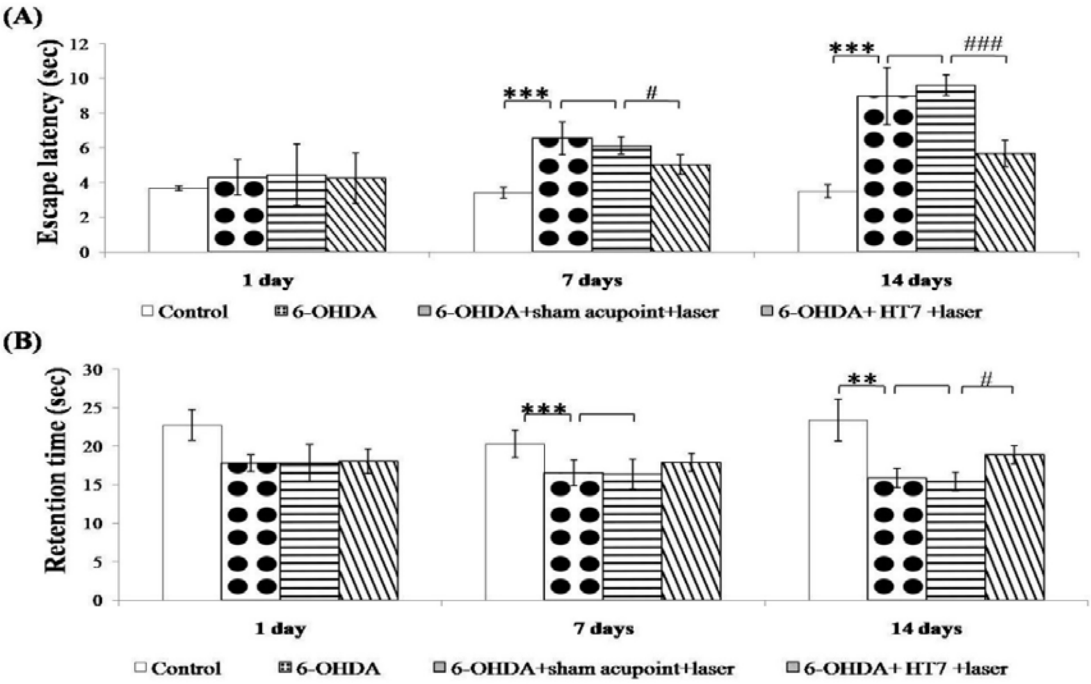

Figure 8-1 Effect of laser acupuncture on spatial memory using the Morris water maze test in Parkinson's disease rats (A) escape latency (B) retention time. Values were expressed as mean ± S.D. (n = 6) *** p-value<.001 as compared with control group and $^{\#}$p-value<.05, $^{\#\#\#}$ p-value<.001 as compared with sham laser acupuncture group

3.2 Effect of laser acupuncture at HT7 on apomorphine-induced rotation behavior

Figure 8-2 showed the effect of lase acupuncture at HT7 on rotational behavior induced by apomorphine. It was found that the administration of 6-OHDA into right substantia nigra significantly increased rotation number (p-value<.001; compared to control group) at 7-day and 14-day periods after the 6-OHDA. Sham laser acupuncture failed to suppress the elevation of rotation number induced by 6-OHDA. However, laser acupuncture at HT7 acupoint significantly suppressed the rotation number induced by 6-OHDA at only 14-day intervention period (p-value<.001; compared to sham laser acupuncture group).

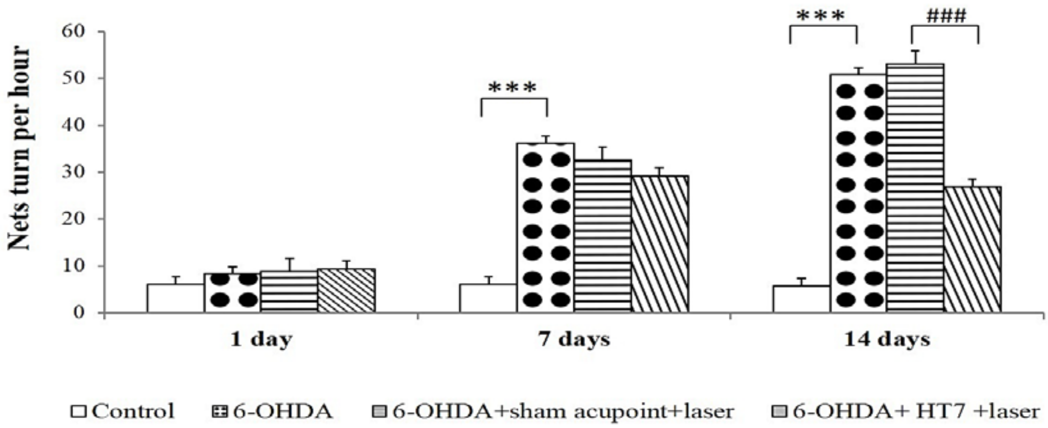

Figure 8-2 Effect of laser acupuncture on apomorphine - induced rotations in Parkinson's disease rats. Values were presented as mean ± S.D. (n = 6) *** p-value < .001 as compared with control group and ### p-value<.001 as compared with sham laser acupuncture group

3.3 Effect of laser acupuncture at HT7 on hippocampal neurons

The administration of 6-OHDA into right substantia nigra induced the decreased survival neuron density in CA1, CA2, CA3 and dentate gyrus of hippocampus (p-value<.001 all; compared to control group). Sham laser acupuncture failed to mitigate the reduction of survival neuron density in all sub-regions of hippocampus mentioned earlier. However, laser acupuncture at HT7 acupoint significantly attenuated the decreased neuron density induced by 6-OHDA in CA3 and dentate gyrus (p-value<.05 all; compared to sham laser acupuncture group) as shown in figure 8-3 to 8-4.

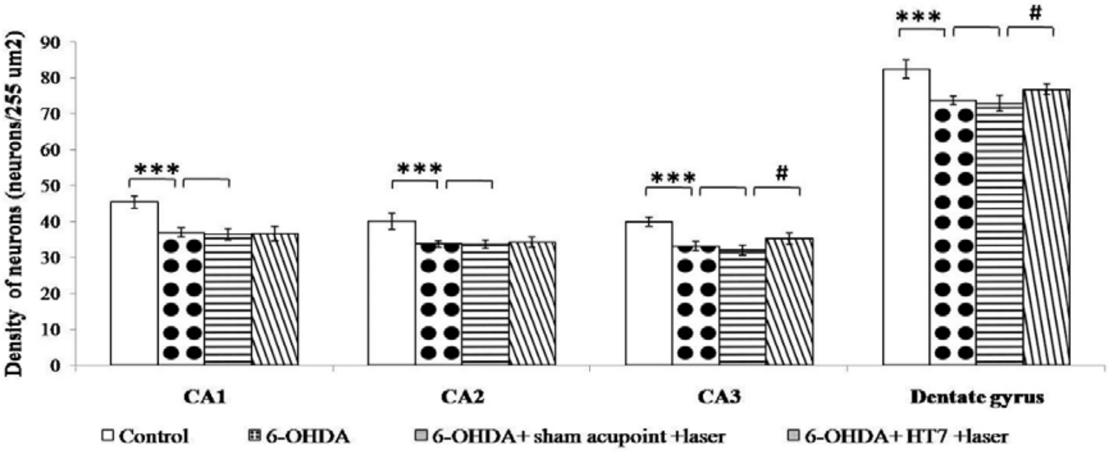

Figure 8-3 Effect of laser acupuncture on the neurons density in various sub-regions of hippocampus after treatments. Values were given as mean ± S.D. (n = 6) *** p-value<.001 as compared with control group and # p-value< .05 as compared with sham laser acupuncture group.

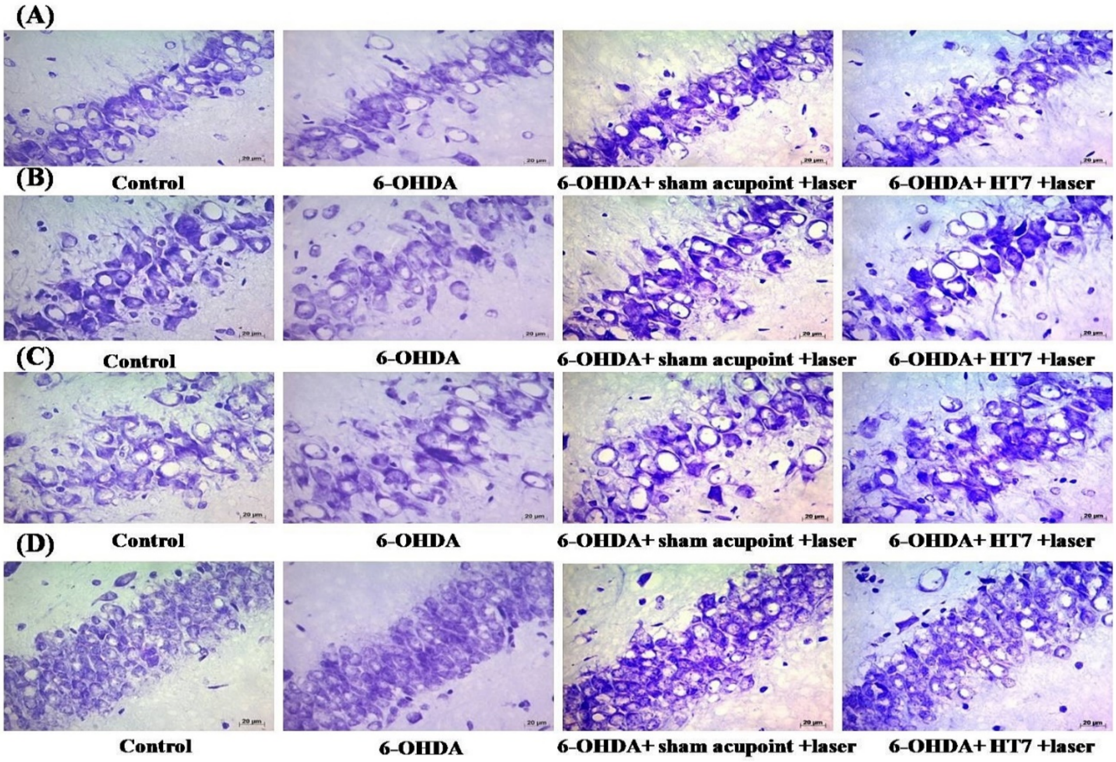

Figure 8-4 Photographic image of neurons with cresyl violet stained in various sub-regions of hippocampus. (A) CA1, (B) CA2, (C) CA3 and (D) dentate gyrus

3.4 Effect of laser acupuncture at HT7 on tyrosine hydroxylase positive neuron

Since dopaminergic neurons play the pivotal role on the pathophysiology of PD, the density of tyrosine hydroxylase positive neuron in substantia nigra was investigated by using immunohistochemistry technique. The rats subjected to the unilateral lesion of substantia nigra induced by 6-OHDA showed the reduction of dopaminergic neuron in the substantia nigra (p-value<.001; compared to control group). This change was attenuated by laser acupuncture at HT7 acupoint (p-value<.001; compared to sham laser acupuncture group) while no significant change was observed in sham laser acupuncture treated group as shown in figure 8-5.

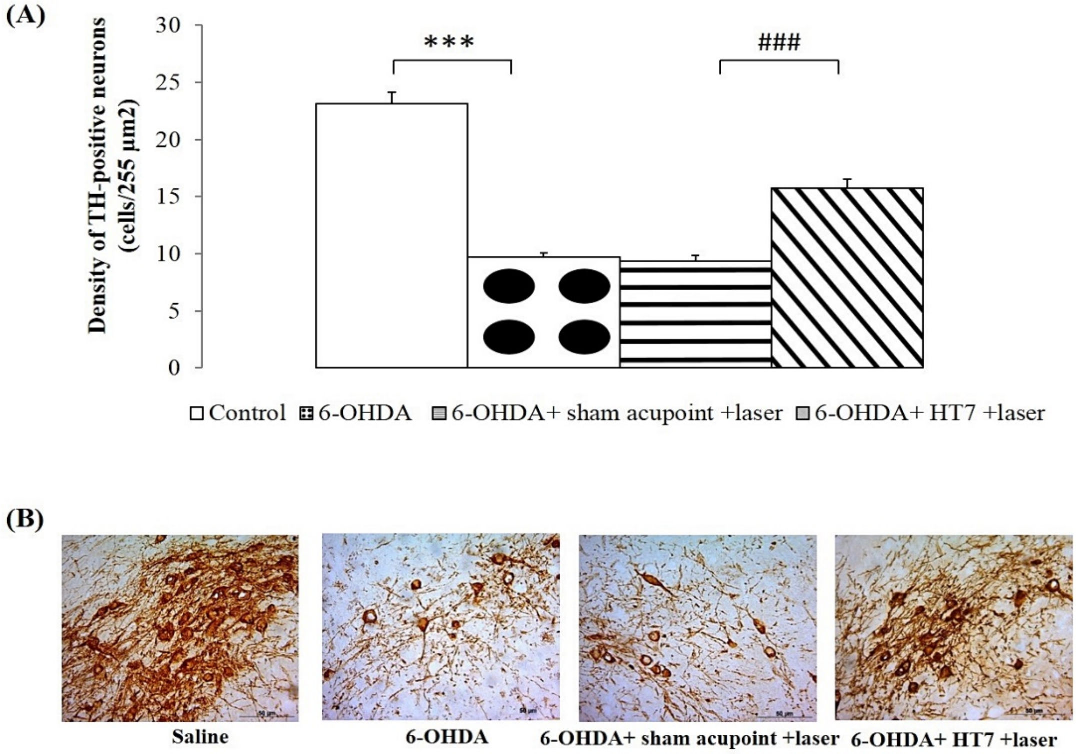

Figure 8-5 The effect of laser acupuncture on tyrosine hydroxylase positive neuron density in substantia nigra. Values werepresented as mean ± S.D. (n = 6) *** p-value<.001 as compared with control group and # p-value<.001 as compared with sham laser acupuncture group

3.5 Effect of laser acupuncture at HT7 on AChE and MAO-B activities

In this study, the activity of AChE was used as indirect index to reflect the function of cholinergic whereas the activity of MAO-B was used as indirect index to reflect the function of monoaminergic especially dopaminergic system. The rats subjected to the unilateral lesion of substantia nigra induced by 6-OHDA showed the elevation of AChE in hippocampus and striatum (p-value<.001 all; compared to control group). This change was mitigated by laser acupuncture at HT7 acupoint (p-value<.05 all; compared to sham laser acupuncture group) while no significant change was observed in sham laser acupuncture treated group as shown in figure 8-6.

Figure 8-7 showed the effect of laser acupuncture on MAO-B activity in hippocampus. Rats with the unilateral lesion of substantia nigra induced by 6-OHDA demonstrated the significant reduction of MAO-B in the hippocampus and striatum (p-value<.001 and .01 respectively; compared to control group). Sham laser acupuncture failed to mitigate the elevation of MAO-B activity whereas laser acupuncture at HT7 significantly decreased the elevation of MAO-B activity in hippocampus and striatum (p-value<.05 all; compared to sham laser acupuncture group).

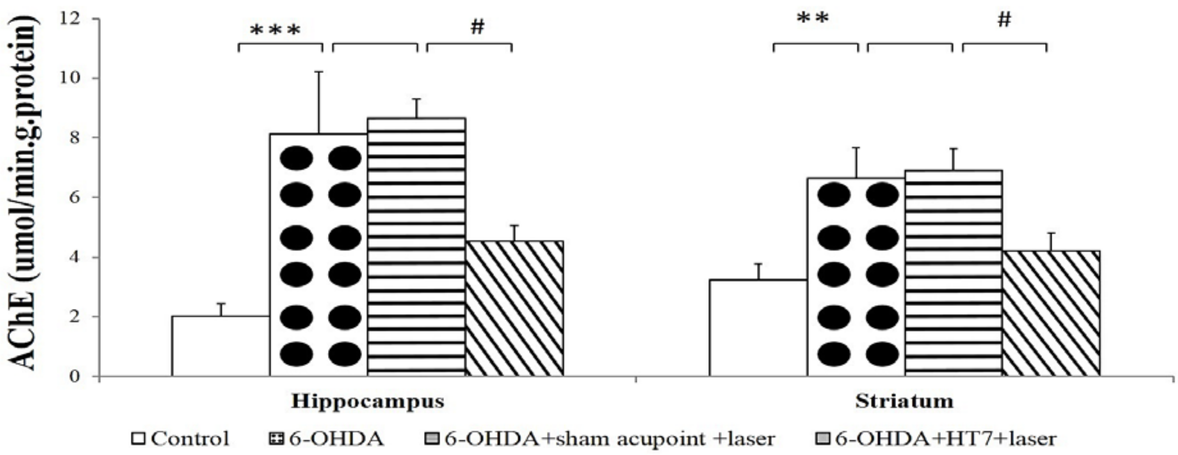

Figure 8-6 Effect of laser acupuncture on the activity of acetylcholinesterase (AChE) in the hippocampus and striatum. Given values were the mean ± S.D. (n=6) *** p-value<.001 as compared with control group and # p-value<.05 as compared with sham laser acupuncture group

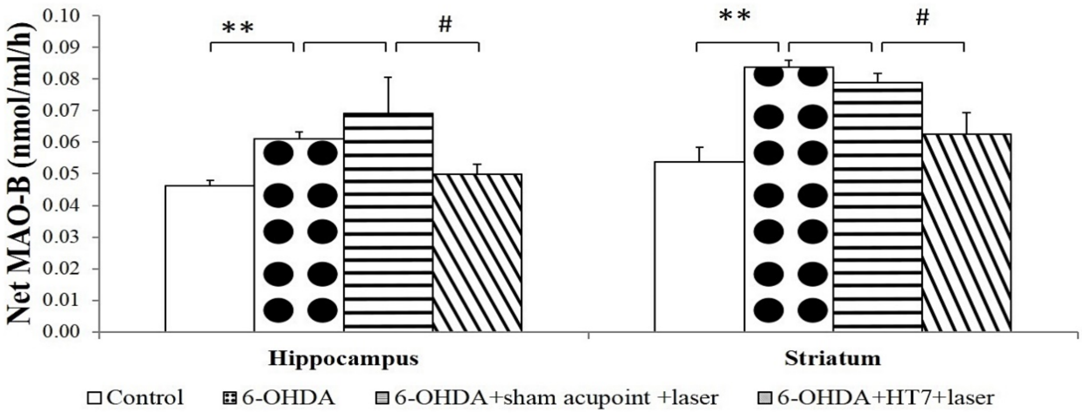

Figure 8-7 Effect of laser acupuncture on the activity of monoamine oxidase-B (MAO-B) in the hippocampus and striatum. Given values were the mean ± S.D. (n = 6) ** p-value<.01 as compared with control group and # p-value< .05 as compared with sham laser acupuncture group

3.6 Effect of laser acupuncture at HT7 on oxidative stress markers

It was found that the administration of 6-OHDA into right substantia nigra significantly decreased CAT and GSH-Px activities but increased MDA level in hippocampusand striatum (p-value<.05, .001 and .001 respectively; compared to control group). Sham laser acupuncture failed to produce significant changes of CAT and GSH-Px activities and MDA level induced by 6-OHDA in hippocampus and striatum. However, laser acupuncture at HT7 could significantly mitigate the decreased GSH-Px activity (p-value<.05 all; compared to sham laser acupuncture group) and the elevation of MDA level in hippocampus and striatum (p-value<.01 and .05 respectively; compared to sham laser acupuncture group) as shown in Fig 8-8 to 8-11.

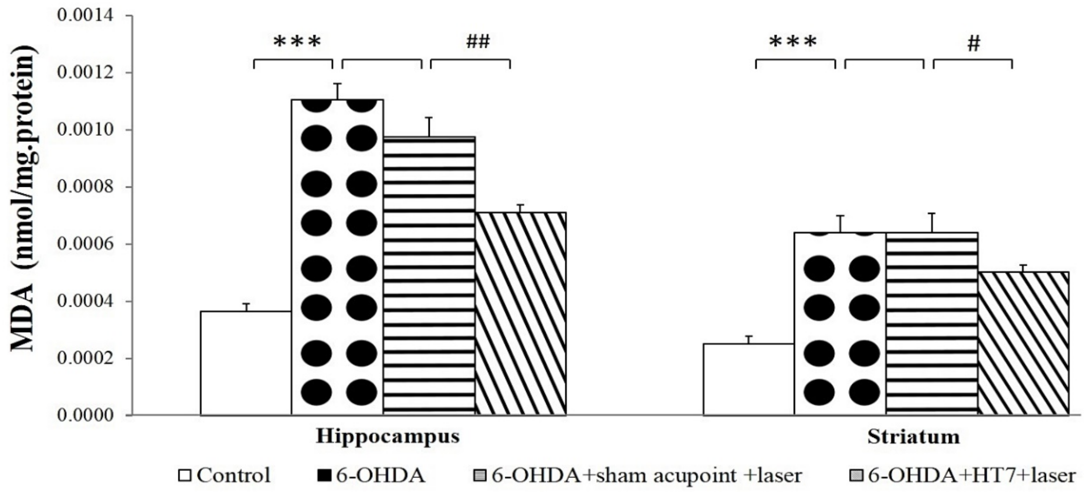

Figure 8-8 Effect of laser acupuncture on the malondialdehyde (MDA) level in the hippocampus and striatum. Values were expressed as mean ± S.D. (n = 6) *** p-value<.001 as compared with control group and $^{\#}$ p-value< .05, $^{\#\#}$ p-value<.01 as compared with sham laser acupuncture group

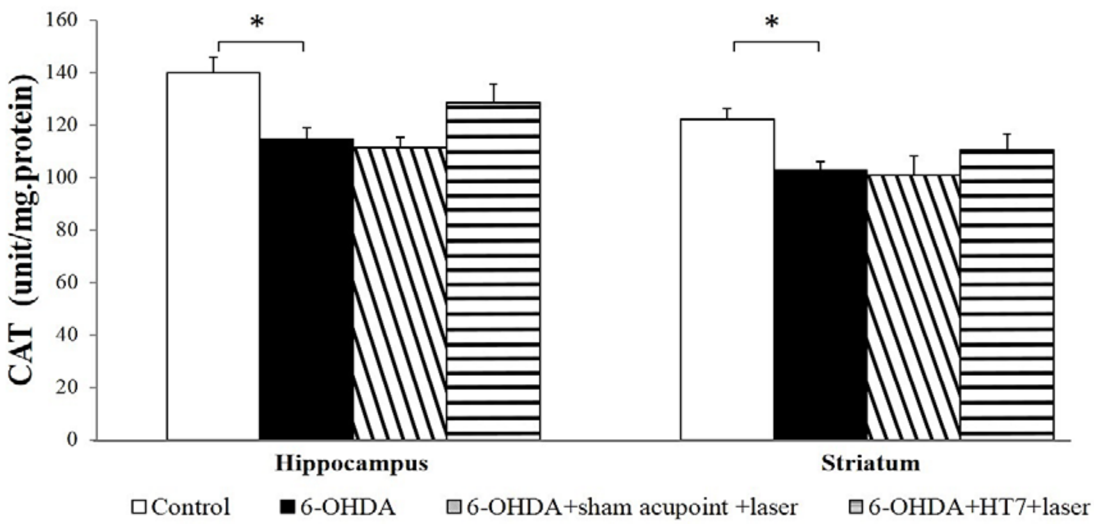

Figure 8-9 Effect of laser acupuncture on the activity of catalase (CAT) in the hippocampus and striatum. Values were presented as mean ± S.D. (n = 6) * p-value<.05 as compared with control

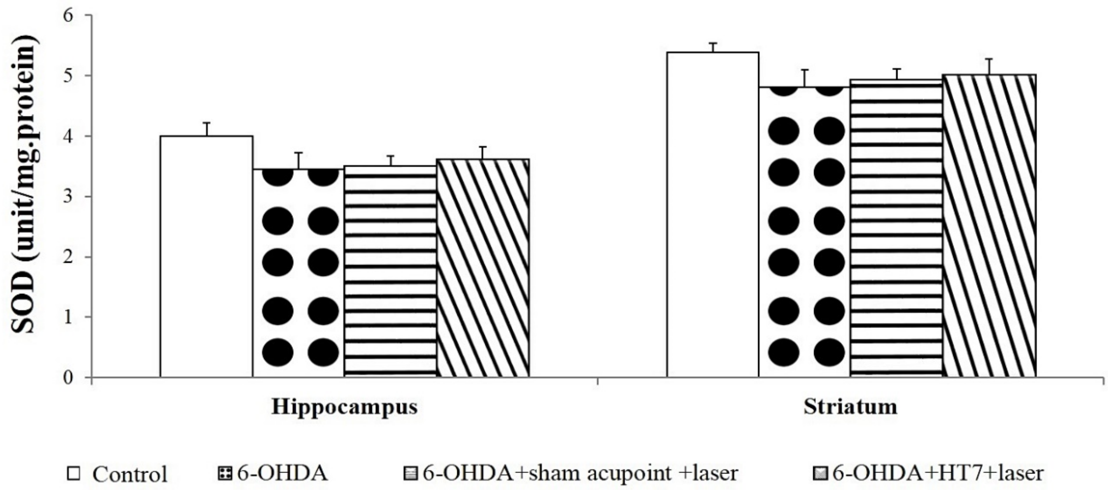

Figure 8-10 Effect of laser acupuncture on the activity of superoxide dismutase (SOD) in the hippocampus and striatum. Values were presented as mean ± S.D. (n = 6)

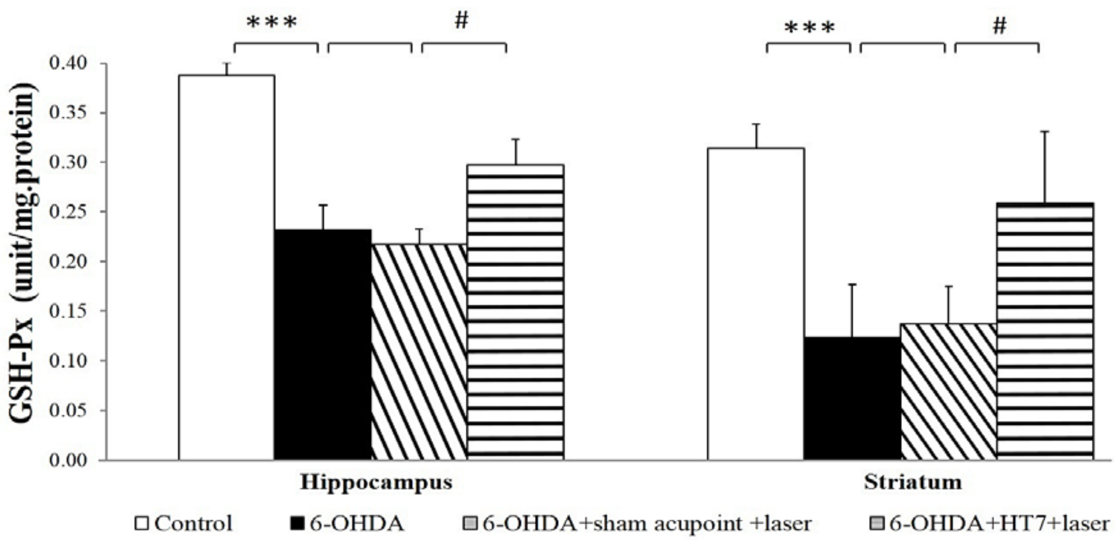

Figure 8-11 Effect of laser acupuncture on the activity of glutathione peroxidase (GSH-Px) in the hippocampus and striatum. Values were presented as mean ± S.D. (n = 6) *** p-value<.001 as compared with control group and # p-value<.05 as compared with sham laser acupuncture group

4. Discussion

The current study has clearly demonstrated that the administration of 6-OHDA into substantia nigra induced the elevation of MAO-B, AChE and oxidative stress in hippocampus and striatum. The possible explanation for this phenomenon might be attributed to the disturbance of dopaminergic function in substantia nigra induced by 6-OHDA produced the disturbance in function of striatum via nigrostriatal pathway and striatum in turn induced the functional disturbance of hippocampus via the connection between ventral striatum and hippocampal pathway which plays a critical role on the association contextual-position information (Pennartz *et al.,* 2011). In addition, several lines of evidence have demonstrated that dentate gyrus of the hippocampus received the dopaminergic projection form substantia nigra (A9) (Sutalangka *et al.*, 2013; Swanson, 1982) and ventral tegmental area (A10), a structure nearby substantia nigra, via mesolimbic pathway. Therefore, the injected 6-OHDA might be transported into the nerve terminals in the area of both substantia nigra and ventral tegmental area and induced the disturbance of hippocampus via the mesolimbic connection mentioned earlier (Sriraksa *et al.*, 2011). However, the precise underlying mechanism is still unclearly known and required further investigation.

Laser acupuncture at HT7 is demonstrated to suppress AChE in hippocampustogether with the improved memory impairment in animal model of Alzheimer's disease (Sutalangka *et al.*, 2013). Our findings are also in agreement with this study, the stimulation of HT7 acupoint can produce significant suppression of AChE and memory improvement even in animal model of PD. It has been reported that the enhanced neurons density in hippocampus induced by the improved oxidative stress status in the mentioned area (Chonpathompikunlert *et al*., 2010), and the dopaminergic function in hippocampus play the important role on the persistence of spatial memory (McNamara *et al.*, 2014). Therefore, the suppression of MAO-B activity in hippocampus induced by laser acupuncture at HT7 observed in this study might increase dopaminergiv function and the persistence of memory. In addition, the increased GSH-Px in hippocampus might in turn decrease oxidative stress status and MDA level in hippocampus giving rise to the increased neuron density in hippocampus especially in CA3 and dentate gyrus leading to the improved spatial memory.

The improved dopaminergic function has been reported to play an important role on the improved motor behaviors disorder such as rotational behavior (Fox *et al*., 2011). In addition, recent findings show that oxidative stress (Jenner, 2003; Ebadi *et al*, 1996) and apoptosis (Adams, 2012) also play the crucial role on the degeneration of dopaminergic neurons in substantia nigra. Therefore, the enhanced dopaminergic neuron density in substnatianigramight involve the improved oxidative stress and apoptosis in this area leading to the improved functions of nigrostriatal pathway and striatum and finally resulting in the improved motor movement disorders. In addition, it is also well known that the hypodopaminergic function in striatum also induces movement disorder. Based on these findings, it is suggested that laser acupuncture might suppress MAO-B activity in striatum giving rise to the enhanced dopaminergic function in this area which in turn improved motor disorders induced by laser acupuncture at HT7 in animal mode l of Parkinson's disease observed in this study.

In conclusion, laser acupuncture at HT7 is the potential non-invasive therapy to improve both motor and non-motor symptoms of Parkinson's disease. The mechanism to improve memory may possibly occur via the suppression of AChE and MAO-B in hippocampus which in turn enhance the functions of both cholinergic and dopaminergic systems in the area just mentioned and via the improved oxidative stress status in hippocampus resulting in the increased neurons density in the mentioned area and finally leading to the enhanced spatial memory.

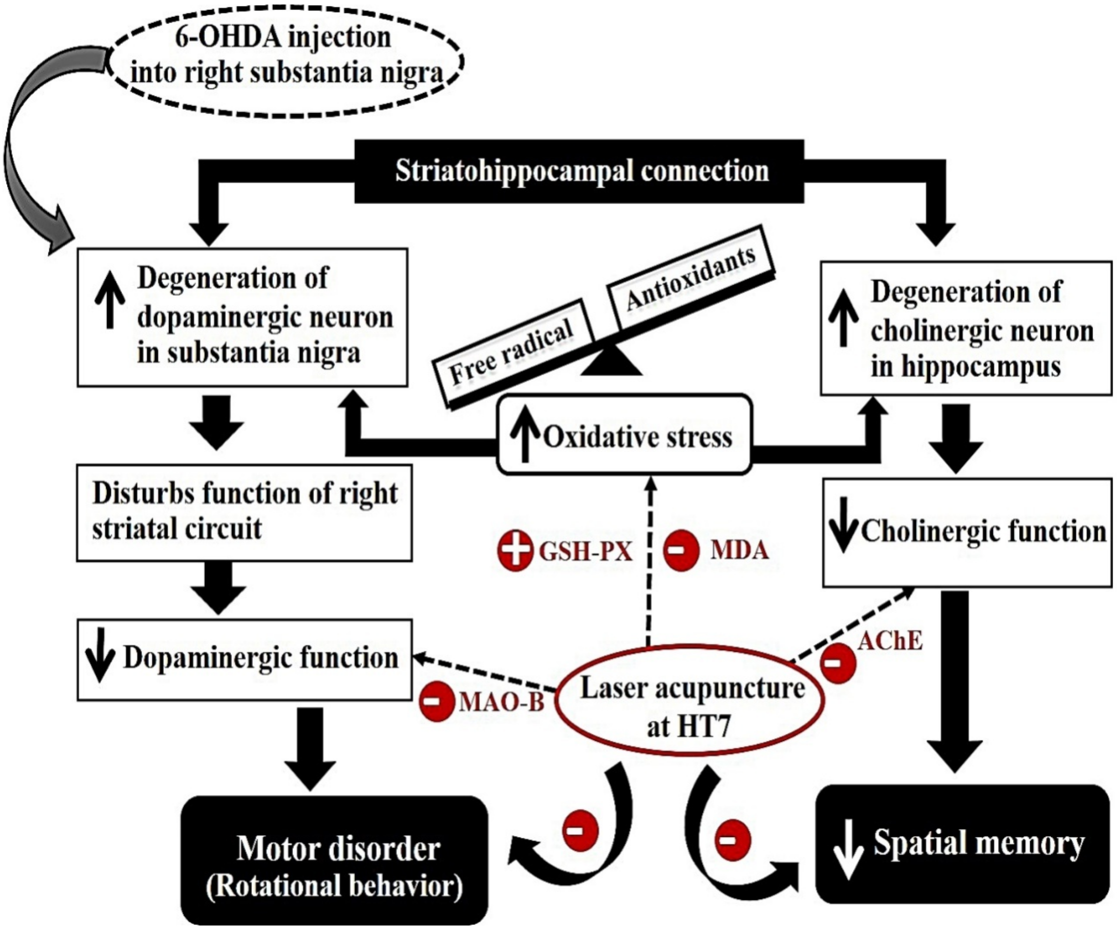

Figure 8-12 Schematic diagram concerning the possible mechanisms to improve memory and motor disorders of laser acupuncture at HT7 in Parkinson's disease rats induced by the unilateral injection of 6-OHDA