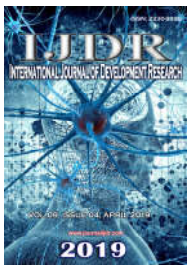
ISSN: 2230-9926

Available online at http://www.journalijdr.com

IJDR

International Journal of Development Research
Vol. 09, Issue, 04, pp. 27099-27106, April, 2019



ORIGINAL RESEARCH ARTICLE OPEN ACCESS

POTENTIAL NEUROPROTECTIVE EFFECTSOF EPIGALLOCATECHIN GALLATEON BEHAVIORAL IMPAIRMENTSIN NORMAL AND ALZHEIMER'S RATS CHEMICALLY INDUCED BY ALUMINUM OXIDE NANOPARTICLES

[1]Mohy eldinAbd el Fattah, [2]Mohamed Ramadan Abdelgawad and *,[1]BasherAbdelghfar El boughdady

[1]Chemistry Department, Faculty of Science, Suez Canal University, Cairo, Egypt
[2]Biological Applications Department, Nuclear Research Centre, Atomic Energy Authority, Cairo, Egypt

ARTICLE INFO

Article History:
Received 14th January, 2019
Received in revised form
26th February, 2019
Accepted 08th March, 2019
Published online 29th April, 2019

Key Words:
Alzheimer's disease; Al2O3NPS;
Oxidative stress,
Epigallocatechin gallate.

Corresponding author:
Basher Abdelghfar El boughdady

ABSTRACT

Alzheimer's disease (AD) is the most common cause of dementia. Epigallocatechin gallate (EGCG), it is most abundant polyphenolin green tea. In addition, it is the most potent neuroprotective compound in green tea. The aim of this study was to evaluate the possible neuroprotective effects of EGCG on behavioral impairments in normal and Alzheimer'srats induced by Aluminum oxide nanoparticles. Forty-eight groups of rats were divided into 8 groups, 6 rats each; Group (1) normal control, received 1ml saline 0.9% orally daily throughout the experiment. Group (2) AI_2O_3NPS-treated rats, received only AI_2O_3NPS. alonein a dose 50 mg/ kg b.wintraperitoneally (i.p), for five weeks. Group (3) received EGCG alone in a dose of 5 mg/kg b.w. i.v every day for five weeks. Group (4) received EGCG alone in a dose of 10 mg/kg b.w. i.v every day for five weeks. Group (5) received Rivastigmine (Exelon) in a dose of 0.3 mg/kg b.w. orally for five weeks. Group (6) received AI_2O_3NPS in a dose of 50 mg/kg (i.p), for five weeks followed by simultaneous administration of EGCG in of a dose 5 mg/kg b.w. i.v every day for five weeks. Group (7) received AI_2O_3NPS in a dose of 50 mg/kg b.w(i.p), for five weeks followed by simultaneous administration of EGCG in a dose of 10 mg/kg b.w. i.v every day for five weeks. Group (8) received AI_2O_3NPS in a dose of 50 mg/kg b.w (i.p), for five weeks followed by simultaneous administration of Rivastigmine (Exelon) in a dose of 0.3 mg/kg b.w. orally for five weeks. The study revealed that, brain neurological damage characterizing induction of AD as indicated by histopathological changes in the brain; in addition to increase in learning time and trials number in the behavioural tests. Hippocampus neuronal degeneration and pyknosis were detected. Our results showed that, EGCG is more effective in minimizing the hazards of Aluminum oxide nanoparticles-induced AD than Rivastigmine (Exelon) on behavioral impairments in Alzheimer's disease rats induced by Aluminum oxide nanoparticles.

Citation: Mohy eldinAbd el Fattah, Mohamed Ramadan Abdelgawad and BasherAbdelghfar El boughdady, 2019. "Potential neuroprotective effectsof epigallocatechin gallateon behavioral impairmentsin normal and alzheimer's rats chemically induced by aluminum oxide nanoparticles", *International Journal of Development Research*, 09, (04), 27099-27106.

INTRODUCTION

Alzheimer's disease (AD) is a chronic neurodegenerative disease, characterized by a slow progressive deterioration of memory function, from an early stage of subtle cognitive symptoms to late-stage dementia, being the most common cause of dementia, accounting for an estimated 60% to 80% of the total number of cases (Alzheimer's Association Society, 2014).The number of AD patients is expected to reach 106.8 million worldwide by the year 2050; therefore, the disease is considered a growing public health concern with major socioeconomic burden (Brookmeyer *et al*., 2007).

Oxidative stress is a condition in which there is an imbalance of reactive oxygen species (ROS), reactive nitrogen species and antioxidant defenses (Dasuri*et al*., 2013; Sies, 1997). Increased oxidative stress is associated with normal aging, but it is further exacerbated in several neurodegenerative disorders including AD (Jomova *et al*., 2010). Nanoparticles (NPs) may be defined as materials that have at least one dimension less than 100 nm (Balasubramanyam *et al*., 2009). Because of their unique chemical, mechanical, and biological propertiesthey are desirable for industrial and health care applications (Oberdörster *et al*., 2005). Nanoparticles can easily reach the brain as their small size, and are taken up by the brain cells (such as neurons and glia). The well-accepted mechanisms of

nanoparticles uptake by cells include endocytosis, pinocytosis reliant on clathrin-dependent endocytosis, caveolae and lipid raft composition and phagocytosis (Asharani *et al*., 2009). NPs have negative health and environmental effects, as they have high surface reactivity. Their small sizes enable cellular uptake and transcytosis across epithelial and endothelial cells into the blood and lymph circulation to reach possibly subtle target sites such as brain, lymph nodes, bone marrow, heart and spleen (Campbell *et al*., 2003). The health-beneficial effects of green tea are mainly attributed to its polyphenol content, particularly flavanols, flavandiols, flavonoids, and phenolic acids; these compounds may account for up to 30% of fresh leaf dry weight (Kuroda and Hara, 1999; Mukhtar and Ahmad, 2000; Riemersma *et al*., 2001). The major flavonoids of green tea are various catechins (Vinson. 2000). There are four types of catechins mainly detected in green tea: epicatechin (EC), epigallo- catechin (EGC), epicatechin-3-gallate (ECG), and epigallocatechin-3- gallate (EGCG) (Sano *et al*., 2001). Epigallocatchin-3-gallate (EGCG) is the most abundant and active compound responsible for most of green tea's role in promoting good health by acting through different pathways; as antioxidant, anti-inflammatory, antiatherogenic and also showing gene expression activity, functioning through growth factor mediated pathways (Shixian *et al*., 2006).

MATERIALS AND METHODS

Animals: In the present study, adult male albino rats (120±20 g) from the animal house of Faculty of Veterinary Medicine, Suez Canal University, Egypt used as experimental animals. The rats were grouped in special cages with six animals per cage and maintained under our laboratory conditions; temperature (23±2^{o}C), with dark and light cycle (12/12h). Standard pellet diet and water were allowed free access *ad libitum*. The rats were adapted to laboratory conditions for 7 days before starting of experiment. All procedures of experiment were performed between 8-11 a.m.

Chemicals: EGCG (M.W:476.39, CAS Number: 989-51-5, Catalog No.: 4524, Batch No.: 2 B/189017) was purchased from Tocris Bioscience/Clinilab company. Aluminum oxide nanoparticles (AI2O3NPS) were got from Egyptian Atomic Energy Authority, Inshas Science City Rivastigmine (1.5 mg) was purchased from Novartis Co. (Cairo, Egypt).

Experimentaldesign: The rats were randomly divided into 8 groups (6 rats for each group) according to the following design: Group (1); received 1ml saline 0.9% orally daily throughout the experiment and served as normal control group. Group (2); received AI_2O_3NPS alonein a dose of 50 mg/kg b.w*(i.p)*, for five weeks according to (Shah *et al*., 2015) with little modefication. Group (3); received EGCG alone in a dose of 5 mg/kg b.w. *i.v* every day for five weeks. Group (4); received EGCG alone in a dose of 10 mg/kg b.w. *i.v* every day for five weeks (Rasoolijazi *et al*., 2007). Group (5); received Rivastigmine (Exelon) in a dose of 0.3 mg/kg b.w. orally for five weeks (Carageorgiou *et al*., 2008). Group (6); received AI_2O_3NPS in a dose of 50 mg/kg b.w*i.p*, for five weeks followed by simultaneous administration of EGCG in a dose of 5 mg/kg b.w. *i.v* every day for five weeks. Group (7); received AI_2O_3NPS in a dose of 50 mg/kg b.w*i.p*, for five weeks followed by simultaneous administration of EGCG in a dose of 10 mg/kg b.w. *i.v* every day for five weeks. Group (8); received AI_2O_3NPS in a dose of 50 mg/kg b.w*i.p*, for five weeks followed by simultaneous administration of Rivastigmine (Exelon) in a dose of 0.3 mg/kg b.w. orally for five weeks.

Behavioral experiments: The Barnes circular maze was designed to assess visuo-spatial learning and memory in aged rats (Barnes. 1979) . This maze consists of a circular platform (92 cm of diameter) with 20 equally spaced holes (5 cm diameter; 7.5 cm between holes) along the perimeter and is elevated 105 cm above the floor. The color of the maze is black depends on the tested strain. In the Barnes maze, animals receive reinforcement to escape from the open platform surface to a small dark recessed chamber located under the platform called a "target box". Rat can access the target box through an escape tunnel (transparent plastic tube 50 cm long, 5 cm diameter), which is located under the target hole (28×22 x 21 cm). A ramp is placed under the target hole so that rat may reach the escape tunnel easily. From the center of the maze all holes should look identical and the ramp should not be visually discriminated from the other holes from most points on the maze, even if it is difficult to ensure no visual discrimination once the rat is situated adjacent to the escape hole. Several reinforcements have been used as stimulus to complete the task as bright light and noise. Animals will have spatial visual cues endogenous to the room (for example a door or a desk). These cues should not be moved during the whole experiment as these are the animal's reference points for locating the target hole. The rat placed in a start chamber in the middle of the brightly-lit maze. After 10 sec, lifted the chamber and guide the rat gently to the escape box. We directly placed the rat into the escape box, if the rat does not want to get into the target hole. Once the rat is inside the box, the brightly-lit light source should be turned off. The rat should stay in the escape box for 2 min. Before the acquisition phase begins, the maze cleaned with 70% ethanol. In addition to cleaning, we rotated the maze around its central axis to control for possibly remaining odor cues. We placed each rat in the start chamber in the middle of the maze. After 10 s, we were lifted the chamber and allowed the rat to explore the maze for 3 minutes.

During these 3 minutes, the number of errors and timewe measured. The trial ends when: (i) the rat enter the goal box. (ii) The 3 min have elapsed. If the animal does enter the box, the brightly-lite light source should be turned off and the rat allowed staying in the box for 1 min. If the rat does not reach the goal within 3 minutes we should guide the rat gently to the escape box and leave the rat inside for 1 min. The rat placed in its home cage until the next trial. On day 5, short-term retention, the probe trial should be conducted. The target hole must be closed. The maze rotated so that the target hole is closed and re-adjusted the maze so that the holes are in the same position as during the training days. The animal placed in the middle of the maze under the start chamber. After 10 s have elapsed, the chamber lifted and the rat allowed exploring the maze as before. The rat removed after a fixed interval (e.g. 90 s). The probe trial is done in order to determine if the animal remembers where the target goal was located. Number of pokes (errors) in each hole and time to reach the virtually target hole are measured. To assess long-term retention, a second probe trial can be applied on day 12, without any training session between day 5 and day 12.Fig. (1)A scheme of the Barnes maze.

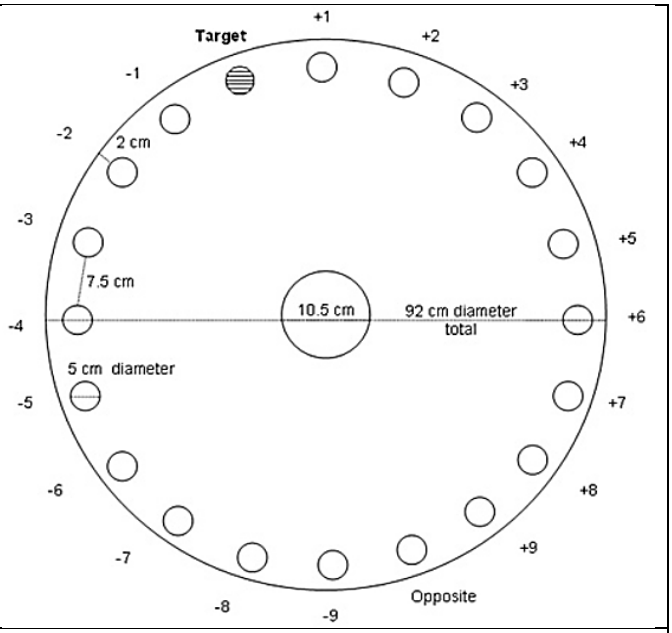


Fig. 1. A scheme of the Barnes maze used is shown. The maze consists of a circular platform (92 cm diameter) elevated 105 cm above the floor with 20 equally spaced holes (5 cm diameter; 7.5 cm between holes). All holes are 2 cm away from the perimeter of the maze. The holes are numbered from +1 to +9 (on the right side of the target hole), from -1 to -9 (on the left side of the target hole) and an opposite hole to the target hole)

Histopathological examination of the brain: The whole brain of each animal was rapidly dissected, thoroughly washed with isotonic saline and dried. Thereafter, each brain divided into two portions. The first portion of each brain about one gram of brain were homogenized immediately with 9.0 ml potassium phosphates buffer solution pH 7.40, then briefly solicited and centrifuged at 3000 rpm for 15 min. the supernatant was separated and used freshly for biochemical assays. The second portion of each brain was fixed in formalin buffer (10%) for histopathological examination.

Statistical Analysis: The result values were expressed as means ±standard error (SE). Data were statistically analyzed using Statistical Package for Social Science (SPSS) version 19, software. One-way analysis of variance (ANOVA) test was performed to statistical analysis for determining the statistical significant differences between means of different groups. Data were considered instatistically significant when the P values were >0.05.

RESULTS

Effect of Exelon and EGCG on behavioural studies by Barnes Maze Test in control rats: As shown in Table (1) there was no variation in the time of short term memory (on 5th day) and Long term memory (on 12th day) to reach rat the target hole, also no variation in the no. of errors for short term memory (on 5th day) and long term memory (on 12th day) in normal rats treated with Exelon (0.3 mg/kg/day for five weeks) and EGCG(5 mg/kg b.w. *i.p* and 10 mg/kg b.w. *i.p.*) daily for five weeks.

Effect of Exelon and EGCGon behavioural studies by Barnes Maze Test in rats treated with Al_2O_3-NPs: As shown in Table (2), time of short term memory and long term memory showed a significant increase in rats treated with Al_2O_3-NPs in a dose 50 mg/kg b.w*i.p*, daily for five weeks to normal rats by 233.9% and 199.6% respectively compared to control.Al_2O_3-NPs treated rates with Exelon (0.3 mg/kg b.w. orally for five weaks) or EGCG (5 mg/kg or10 mg/kg) every day for five weeks were significantly decreased in time of short term memory (on 5th day) and long term memory (on 12th day) as following (69.83 ± 4.26, 74.0 ± 1.46 *vs*. 169.17 ± 1.11, 168.3 ± 3.04) by 58.7% and 56 % respectively compared to Al_2O_3-NPs group. Rats treated with EGCG (5 mg and 10 mg) with Al_2O_3-NPs administration in the previously mentioned dose and period had a significant decrease in time of short term memory and Long term memory compared to Al_2O_3-NPs -treated rats. Time of short term memory of these rats returned nearly to the normal values and more effective than Exelon on rats treated with Al_2O_3-NPs. Treatment of EGCG (5 mg and 10 mg) with Al_2O_3-NPs administration in the previously mentioned dose and period were the same effective as treatment of Exelon with Al_2O_3-NPs administration as following: (65.83 ± 4.31 , 61.17 ± 3.42 *vs*. 50.67 ± 5.3) respectively in time of short term memory (on 5th day) and (75.17 ± 0.70, 64.83 ± 3.86 *vs*. 56.17 ± 4.36) respectively in time of Long term memory (on 12thday). Time of long term memory (on 12th day) in case of EGCG(10 mg) treated rats returned nearly to the normal values and more effective than Exelon and EGCG(10 mg) on rats treated with Al_2O_3-NPs.As shown in Table (2), number of errors for short term memory and long term memory showed a significant increase in rats treated with Al_2O_3-NPs in a dose 50 mg/kg b.w*i.p*, daily for five weeks to normal rats by 331.2% and 231.9% respectively compared to control.Al_2O_3-NPs treated with Exelon (0.3 mg/kg b.w. orally for five weeks) or EGCG(5 mg/kg or10 mg/kg) every day for five weeks were significantly decreased in no. of errors for short term memory and long term memory as following (6.83±0.31, 7.33±0.42 *vs*. 13.67 ± 0.76, 15.5 ± 1.11) by 50% and 52.7 % respectively compared to Al_2O_3-NPs group.

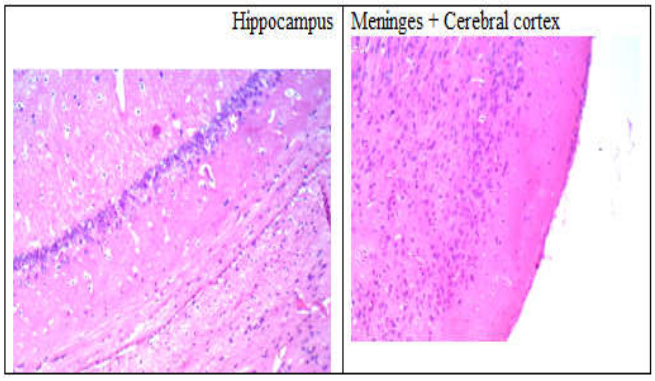


Figure 2. Representative photomicrographs of brain sections stained by Hematoxylin–Eosin stain (magnification 40 X): Sections taken from brain of control group showing normal histological structures of hippocampus, meninges and cerebral cortex

The effect of EGCG (5 mg/kg and 10 mg/kg) with Al_2O_3-NPs is similar to the effect of Exelon on rats treated with Al_2O_3-NPs.Rats treated with EGCG(5 mg and 10 mg) with Al_2O_3-NPs administration in the previously mentioned dose and period had a significant decrease in no. of errors for short term memory and long term memory compared to Al_2O_3-NPs -treated rats. Treatment of EGCG(5 mg) with Al_2O_3-NPs administration in the previously mentioned dose and period were the same effective as treatment with Exelon on rats treated with Al_2O_3-NPs as following: (6.67±0.33*vs*. 6.83± 0.31) respectively.

Table 1. Barnes Maze Test for Behavioral studies in control, and normal rats treated with Exelon and EGCG (n=6)

Trail / Group	Time (sec.) (term memory)		No. of Errors (term memory)	
	Short (on 5th day)	Long (on 12th day)	Short (on 5th day)	Long (on 12th day)
Normal control	50.67 ±5.31 [a]	56.17± 4.36 [a]	3.17± 0.6 [a]	4.67 ± 0.49 [a]
Range	(39–67)	(42–65)	(1.0–5.0)	(3.0–6.0)
Exelon	52.83± 5.01 [a]	58.67 ± 4.18 [a]	4.17 ± 0.48 [a]	5.33 ± 0.76 [a]
Range	(40–68)	(45–66)	(3.0–6.0)	(2.0–7.0)
%Change compared to control	4.2	4.5	31.5	14.13
EGCG (5 mg)	52.83± 4.4 [a]	57.5± 4.15 [a]	4.0 ± 0.45 [a]	5.33± 0.42 [a]
Range	(41–66)	(44–66)	(2.0–5.0)	(4.0–7.0)
%Change compared to control	4.26	2.37	26.19	14.13
EGCG (10 mg)	49.83± 4.85 [a]	55.67 ± 3.9 [a]	3.67± 0.42 [a]	4.83 ± 0.79 [a]
Range	(39–65)	(43–64)	(2.0–5.0)	(1.0–6.0)
%Change compared to control	-1.65	-0.9	15.8	3.5

Data presented as Mean ± SEM
Means have the same letters considered insignificant (P>0.05).

Table 2. Barnes Maze Test for Behavioral studies in control, AL_2O_3-NPS-treated rats, and AL_2O_3-NPS -treated rats and supplemented with Exelon and EGCG (n=6)

Trail / Group	Time (sec.) (term memory)		No. of Errors (term memory)	
	Short (on 5th day)	Long (on 12th day)	Short (on 5th day)	Long (on 12th day)
Normal control	50.67± 5.3 [c]	56.17± 4.36 [c]	3.17± 0.6 [c]	4.67± 0.49 [c]
Range	(39–67)	(42–65)	(1.0–5.0)	(3.0–6.0)
AL2O3-NPS	169.17± 1.11 [a]	168.3 ± 3.04 [a]	13.67± 0.76 [a]	15.5 ± 1.11 [a]
Range (n=**6**)	(165–172)	(165–177)	(11.0–16.0)	(12.0–19.0)
%Change compared to control	233.9	199.6	331.2	231.9
Exelon	69.83± 4.26 [b]	74.0± 1.46 [b]	6.83± 0.31 [b]	7.33± 0.42 [b]
Range	(50–78)	(68–79)	(6.0–8.0)	(6.0–9.0)
%Change compared to control	37.8	31.7	115.4	56.9
%Change compared to Al_2O_3-NPs	-58.7	-56	-50	-52.7
EGCG (5 mg)	65.83 ± 4.31 [b c]	75.17± 0.70 [b]	6.67± 0.33 [b]	7.67± 0.33 [b]
Range	(48–77)	(73–77)	(6.0–8.0)	(6.5–8.5)
%Change compared to control	29.9	33.8	110	64.2
%Change compared to Al_2O_3-NPs	-61.1	-55.3	-51.2	-50.5
EGCG (10 mg)	61.17± 3.42 [b c]	64.83± 3.86 [b c]	4.5± 0.22 [c]	5.17± 0.48 [b c]
Range	(45–68)	(46–71)	(4.0–5.0)	(4.0–7.0)
%Change compared to control	20.7	15.4	41.9	10.7
%Change compared to Al_2O_3-NPs	-63.9	-61.5	-67	-66.6

Data presented as Mean ± SEM
Means have the same letters considered insignificant (P>0.05).

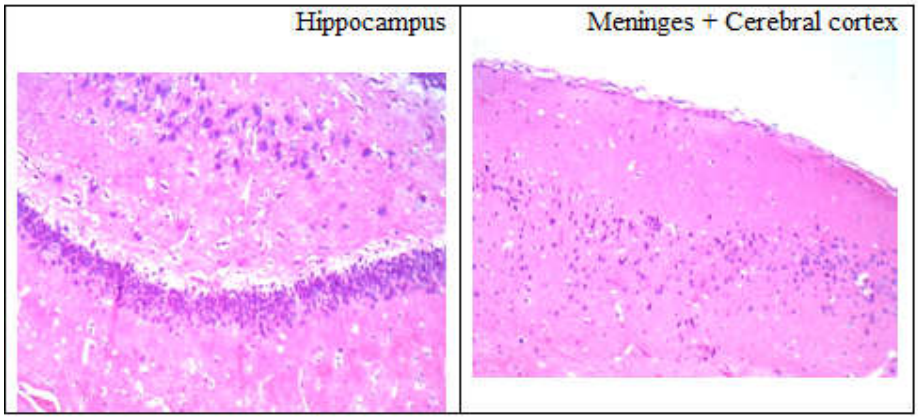


Figure 3. Representative photomicrographs of brain sections stained by Hematoxylin–Eosin stain (magnification 40 X): Section taken from brain of (Exelon 0.3 mg alone) treated group showing no pathological changes hippocampus, meninges and cerebral cortex

No. of errors for short term memory and long term memory in case of EGCG(10 mg) treated rats returned nearly to the normal values and more effective than Exelon on rats treated with Al2O3-NPs as following: (4.5±0.22 vs. 6.83±0.31) and (5.17±0.48 vs. 7.33±0.42), respectively.

Histopathological findings: Figures (2, 3, 4 and 5) showing normal histological structures of hippocampus, meninges, cerebral cortex, medulla oblongata and striatum in the cerebrum of control and normal rats with different treatment. As shown in figure (6), sections taken from brain of AD model rats (AL_2O_3-NPs 50 mg alone) showing marked congestion and hemorrhage in meningeal vessels and in cerebral cortex vessels, There is neuronal vacuolar degeneration and pyknosis at cerebrum and medulla, hippocampus showed enlargement of nuclei and vacuolation of cytoplasm. The histopathological examination of rats treated with Exelon alone in a dose (0.3 mg/kg b.w. orally) for five weeks showing no pathological changes hippocampus, meninges, cerebral cortex and medulla oblongata (Figure 7). However, in rats treated with Al2O3-NPs treated with Exelon showing mild neuronal degeneration, mild congestion of meningeal vessels, mild thickening, moderate vacuolation of neurons at hippocampus and medulla was found as shown in (Figure 7).

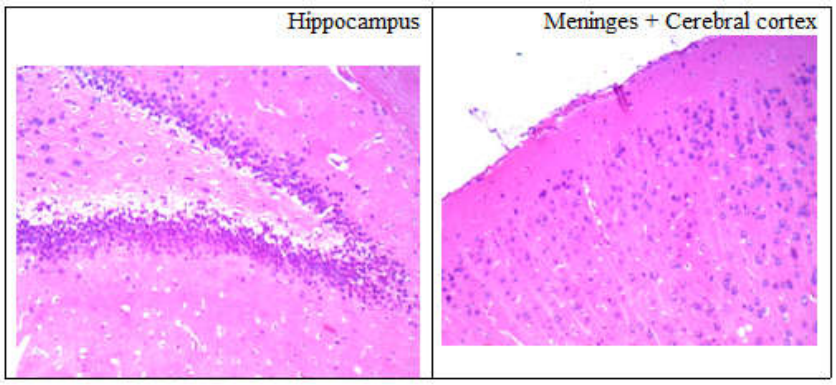


Figure 4. Representative photomicrographs of brain sections stained by Hematoxylin –Eosin stain (magnification 40 X): Section taken from brain of (EGCG 5 mg alone) treated group showing no pathological changes hippocampus, meninges and cerebral cortex

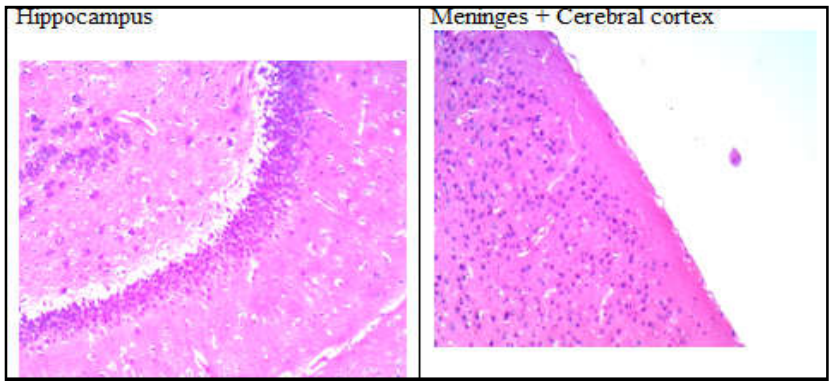


Figure 5. Representative photomicrographs of brain sections stained by Hematoxylin–Eosin stain (magnification 40 X): Section taken from brain of (EGCG 10 mg alone) treated group showing no pathological changes hippocampus, meninges and cerebral cortex

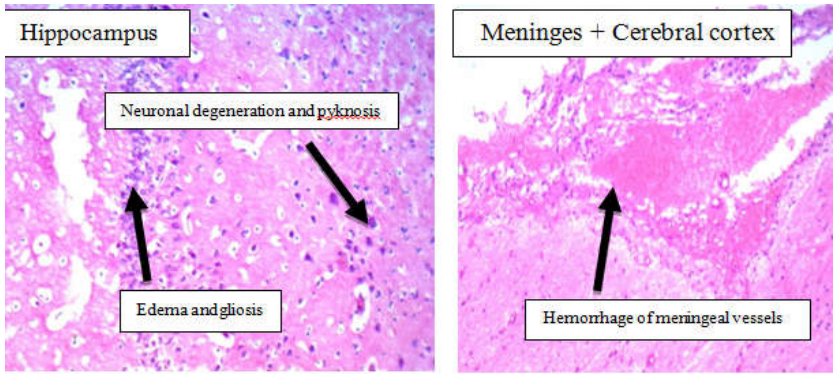


Figure 6. Representative photomicrographs of brain sections stained by Hematoxylin–Eosin stain (magnification 40 X): Sections taken from brain of AD model rats (AL_2O_3-NPS 50 mg alone) treated group showing marked congestion and hemorrhage in meningeal vessels and in cerebral cortex vessels, hippocampus showed enlargement of nuclei and vacuolation of cytoplasm

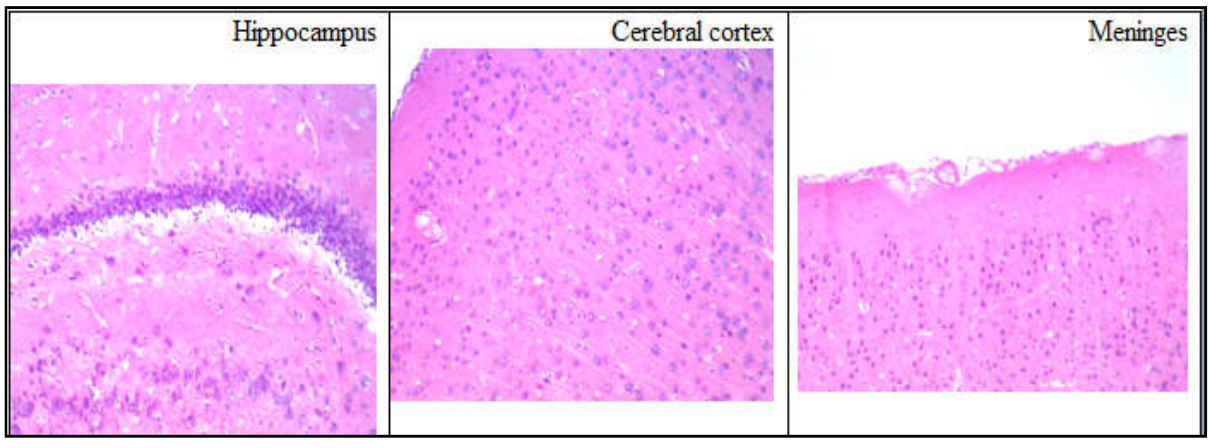


Figure 7. Representative photomicrographs of brain sections stained by Hematoxylin–Eosin stain (magnification 40 X): Sections taken from brain of (AL_2O_3-NPS + Exelon 0.3 mg) treated group showing mild neuronal degeneration, mild congestion of meningeal vessels, mild thickening, moderate vacuolation of neurons at hippocampus and medulla

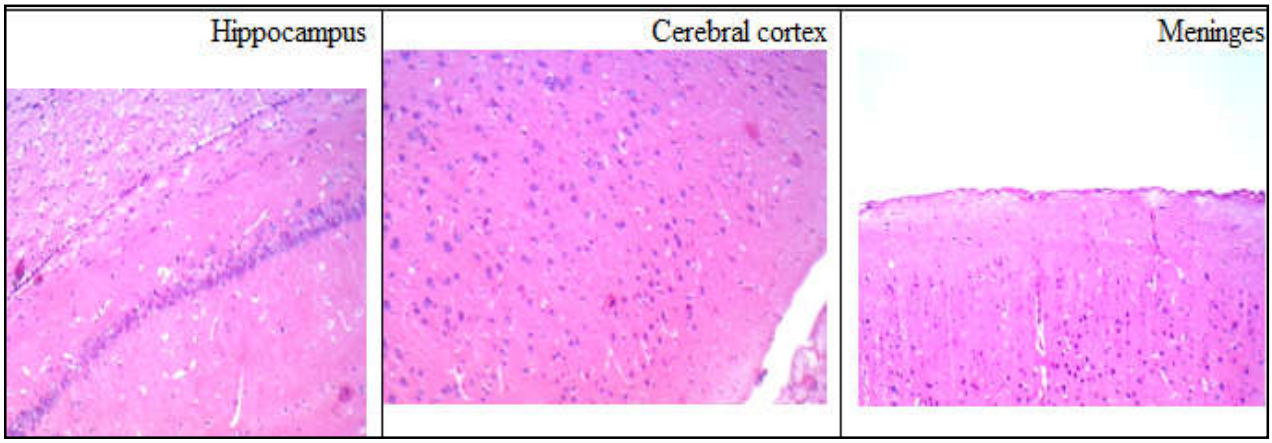


Figure 8. Representative photomicrographs of brain sections stained by Hematoxylin–Eosin stain (magnification 40 X): Sections taken from brain of (AL_2O_3-NPS +EGCG 5 mg) treated group showing minimal neuronal degeneration, minimal congestion of meningeal vessels, minimal meningeal thickening, mild vacuolation of neurons at hippocampus and medulla

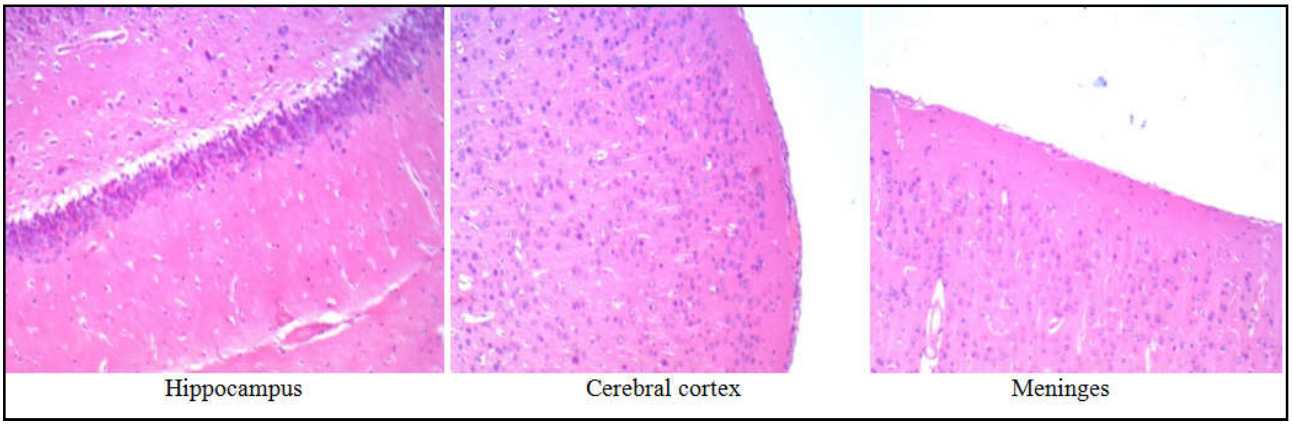


Figure 9. Representative photomicrographs of brain sections stained by Hematoxylin–Eosin stain (magnification 40 X): Sections taken from brain of (AL_2O_3-NPS +EGCG 10 mg) treated group showing minimal neuronal degeneration, No congestion of meningeal vessels, no thickening, minimal vacuolation of neurons at hippocampus and medulla

Brain sections in treatment rats with EGCG alone in a dose (5 mg/kg b.w. i.v.) every day for five weeks showing no pathological changes hippocampus, meninges, cerebral cortex and medulla oblongata.

DISCUSSION

The present study was designed to investigate the protective role of EGCG on Behavioral studies in normal and Alzheimer's diseaseby aluminum oxide nanoparticles-induced neurotoxicity and brain damage in adult male albino rats. It is important that the protective agent is present in brain tissues after administration of aluminum oxide nanoparticles and damage occurs. Alzheimer's disease is the most common cause of dementia and is a degenerative brain disease. Dementia is a syndrome that has a number of causes-a group of symptoms. Dementia's characteristic symptoms are memory, language, problem solving and other cognitive skills that affect the ability of a person to carry out everyday activities. These problems are caused by damage or destruction of nerve cells (neurons) in parts of the brain involved in cognitive function. Neurons in other parts of the brain are eventually damaged or destroyed in Alzheimer's disease, including those that allow a person to perform basic body functions such as walking and swallowing. People in the final stages of the disease are in bed and need 24-hour care (Alzheimer Association Society, 2018).
In the present study, AD was induced in rats by injection of Al_2O_3NP (50 mg/kg, i.p) three times a week for five weeks.

Results indicated that, it caused progressive deterioration of learning ability and spatial memory as evidenced by Barnez maze test. There is significant increase in time of short term memory (on 5^{th} day) and long term memory (on 12^{th} day) to reach rates the target hole in rats treated with Al_2O_3-NPs in a dose 50 mg/ kg b.w (i.p), daily for five weeks to normal rats compared to control. It seems that these impairments of behaviors in relation to learning and memory are due to the disturbance of the hippocampal circuit and its vast connections (through cortical and subcortical pathway) (Skutella and Nitsch, 2001). The hippocampus is the site in the brain responsible for memory and learning. Some authors have indicated that Al_2O_3-NPs can cross the BBB regardless of the route of administration (Shah *et al*., 2015); however, there is insufficient evidence to confirm that. Some authors indicated that Al_2O_3-NPs could cross the BBB irrespective of the administration route (Shah *et al*., 2015); However, insufficient evidence is available to confirm this. We believe that the ionic form of NPs is absorbed in the brain. Al is a neurotoxin involved in certain neurodegenerative diseases like Alzheimer's disease (Abdel-Aal *et al*., 2011), Where patients experience deterioration of certain skills such as attention, concentration, visual memory, vocabulary scores and frontal lobe function (Kumar *et al*., 2011). Treatment of AD group of rats with EGCG (5 mg or10 mg) had a significant decrease in time and no. of errors of short term memory (on 5^{th} day) and Long term memory (on 12^{th} day) to reach the target hole compared to Al_2O_3-NPs -treated rats. Many investigators have reported that hippocampus is one of the most vulnerable

regions in the AD brain (Lu *et al*., 2003) andhippocampal lesions in general produce changes in rat's activity levels and impairment in spatial memory (Deacon *et al*., 2002). Therefore, behavioral improvement in this study may be attributed to the effect of EGCG on the power of memory in these animals (Skrzypczak-Jankun *et al*., 2003). Although, the mechanism of EGCG action is still unclear, EGCG has been found to inhibit cancer angiogenesis by suppressing vascular endothelial growth factor production (Basini *et al*., 2005). EGCG have phenol rings that act as electron traps to scavenge peroxy radicals, superoxide anions, and hydroxyl radicals and prevent oxidation of iron. Therefore, we suggest that in addition to the reduction of iNOS expression, these compounds may block peroxynitrite and nitrite production through inhibition of oxidative reactions (Kim *et al*., 2004). In addition, it seems that, EGCG may act as an antioxidant and anti-inflammatory agent (Skrzypczak-Jankun *et al*., 2003) against β-amyloid aggregation in hippocampus and in this way EGCG may have a neuroprotective effect. Therefore, EGCG were effective in reducing the cognitive deficit observed in Barnes maze performance.

Treatment of AD group of rats with Exelon (Rivastigmine) had a significant decrease in time and no. of errors of short term memory (on 5^{th} day) and Long term memory (on 12^{th} day) to reach the target hole compared to Al_2O_3-NPs-treated rats. Rivastigmine protects behavioral changes, restores antioxidant defense enzyme in brain and improves mitochondrial enzyme level induced neurotocixity (Kumar & Kumar, 2009). In the present study, AD was induced in rats by injection of Al_2O_3-NP (50 mg/kg, IP daily for five weeks. Results indicated that, Administration of Exelon or EGCG ameliorated or reversed the changes induced by Al_2O_3-NPs in part. The lesions in the brain of rats that received Al2O3-NPs with EGCG (5mg and 10 mg) or with Exelon were conspicuously less than those in the rats that received Al_2O_3-NPs only, in rats treated with Al_2O_3-NPs together with EGCG in a dose (5 mg/kg b.w. i.v.) showing minimal neuronal degeneration, minimal congestion of meningeal vessels, minimal meningeal thickening, mild vacuolation of neurons at hippocampus and medulla was observed. Also, in rats treated with Al_2O_3-NPs together with EGCG in a dose (10 mg/kg b.w. i.v.) showing minimal neuronal degeneration, No congestion of meningeal vessels, no thickening, minimal vacuolation of neurons at hippocampus and medulla was observed. Besides, the histopathological examination to treatment of Al_2O_3-NPs treated with Exelon for 5 weeks, 3 times weekly showed mild neuronal degeneration, mild congestion of meningeal vessels, mild thickening, moderate vacuolation of neurons at hippocampus and medulla. Our results are in agreement with the observation of (Levites *et al*., 2003; Chow *et al*., 2005) who observed improvement in histopathological with received EGCG green tea. So, histopathological findings EGCG 10 mg is more effective than EGCG 5mg and Exelon 0.3 mg.

Conclusion

EGCG is more effective in minimizing the hazards of Al_2O_3-NPs during induction and progression of AD than Exelon (Rivastigmine). This may be attributed to its additional anti-inflammatory and antioxidant effects as well as to its ability to antagonize Aβ aggregation in the hippocampus. In this way EGCG have a neuroprotective effect which confirmed by behavioral and histopathological examination.

REFERANCES

Abdel-Aal, R. A., Assi, A.-A.A. and Kostandy, B. B. 2011.Rivastigmine reverses aluminum-induced behavioral changes in rats. *European journal of pharmacology*, 659(2-3), 169-176.

Asharani, P., Hande, M. P. and Valiyaveettil, S. 2009. Anti-proliferative activity of silver nanoparticles.*BMC cell biology,* 10(1), 65.

Alzheimer's Association, A. S. 2014.2014 Alzheimer's disease facts and figures.Alzheimer's and dementia, 10(2), e47-e92.

Alzheimer's ssociation, A. S. 2018.2018 Alzheimer's disease facts and figures. Alzheimer's and Dementia, 14(3), 367-429.

Balasubramanyam, A., Sailaja, N., Mahboob, M., Rahman, M., Hussain, S. M. and Grover, P. 2009. In vivo genotoxicity assessment of aluminium oxide nanomaterials in rat peripheral blood cells using the comet assay and micronucleus test.*Mutagenesis*, 24(3), 245-251.

Barnes, C. A. 1979. Memory deficits associated with senescence: a neurophysiological and behavioral study in the rat. *Journal of comparative and physiological psychology*, 93(1), 74.

Basini, G., Bianco, F., and Grasselli, F. (2005). Epigallocatechin-3-gallate from green tea negatively affects swine granulosa cell function. *Domestic animal endocrinology,* 28(3), 243-256.

Brookmeyer, R., Johnson, E., Ziegler-Graham, K. and Arrighi, H. M. 2007.Forecasting the global burden of Alzheimer's disease.Alzheimer's and dementia, 3(3), 186-191.

Campbell, A., Becaria, A., Lahiri, D., Sharman, K. and Bondy, S. 2004. Chronic exposure to aluminum in drinking water increases inflammatory parameters selectively in the brain. *Journal of neuroscience research*, 75(4), 565-572.

Carageorgiou, H., Sideris, A. C., Messari, I., Liakou, C. I. and Tsakiris, S. 2008. The effects of rivastigmine plus selegiline on brain acetylcholinesterase,(Na+, K+)-, Mg2+-ATPase activities, antioxidant status, and learning performance of aged rats. *Neuropsychiatric disease and treatment*, 4(4), 687.

Chow, H. S., Hakim, I. A., Vining, D. R., Crowell, J. A., Ranger-Moore, J., Chew, W. M. and Alberts, D. S. 2005. Effects of dosing condition on the oral bioavailability of green tea catechins after single-dose administration of Polyphenon E in healthy individuals.*Clinical cancer research*, 11(12), 4627-4633.

Dasuri, K., Zhang, L. and Keller, J. N. 2013.Oxidative stress, neurodegeneration, and the balance of protein degradation and protein synthesis.*Free Radical Biology and Medicine*, 62, 170-185.

Deacon, R. M., Croucher, A. and Rawlins, J. N. P. 2002. Hippocampal cytotoxic lesion effects on species-typical behaviours in mice.*Behavioural brain research*, 132(2), 203-213.

Jomova, K., Vondrakova, D., Lawson, M. and Valko, M. 2010.Metals, oxidative stress and neurodegenerative disorders.*Molecular and cellular biochemistry,* 345(1-2), 91-104.

Kim, S. Y., Ahn, B. H., Kim, J., Bae, Y. S., Kwak, J. Y., Min, G. andYoon, S. H. 2004. Phospholipase C, protein kinase C, Ca2+/calmodulin-dependent protein kinase II, and redox state are involved in epigallocatechin gallate-induced phospholipase D activation in human astroglioma cells. *European journal of biochemistry*, 271(17), 3470-3480.

Kumar, A., Prakash, A. and Dogra, S. 2011. Neuroprotective effect of carvedilol against aluminium induced toxicity: possible behavioral and biochemical alterations in rats. *Pharmacological Reports,* 63(4), 915-923.

Kumar, P. and Kumar, A. 2009. Protective effect of rivastigmine against 3-nitropropionic acid-induced Huntington's disease like symptoms: possible behavioural, biochemical and cellular alterations. *European journal of pharmacology*, 615(1), 91-101.

Kuroda, Y. and Hara, Y. 1999. Antimutagenic and anticarcinogenic activity of tea polyphenols.*Mutation Research/Reviews in Mutation Research*, 436(1), 69-97.

Levites, Y., Amit, T., Mandel, S. and Youdim, M. B. 2003.Neuroprotection and neurorescue against Aβ toxicity and PKC-dependent release of nonamyloidogenic soluble precursor protein by green tea polyphenol (-)-epigallocatechin-3-gallate. *The FASEB Journal*, 17(8), 952-954.

Lu, Y.-P., Zeng, M., Hu, X.-Y., Xu, H., Swaab, D. F., Ravid, R. and Zhou, J.-N. 2003. Estrogen receptor α-immunoreactive astrocytes are increased in the hippocampus in Alzheimer's disease. *Experimental neurology*, 183(2), 482-488.

Mukhtar, H. and Ahmad, N. 2000. Tea polyphenols: prevention of cancer and optimizing health–. The *American journal of clinical nutrition*, 71(6), 1698S-1702S.

Oberdörster, G., Oberdörster, E., and Oberdörster, J. (2005).Nanotoxicology: an emerging discipline evolving from studies of ultrafine particles. *Environmental Health Perspectives,* 113(7), 823-839.

Rasoolijazi, H., Joghataie, M. T., Roghani, M., and Nobakht, M. 2007. The beneficial effect of (-)-epigallocatechin-3-gallate in an experimental model of Alzheimer's disease in rat: A behavioral analysis. *Iranian Biomedical Journal*, 11(4), 237-243.

Riemersma, R., Rice-Evans, C., Tyrrell, R., Clifford, M. and Lean, M. 2001.Tea flavonoids and cardiovascular health.Qjm, 94(5), 277-282.

Sano, M., Tabata, M., Suzuki, M., Degawa, M., Miyase, T. and Maeda-Yamamoto, M. 2001.Simultaneous determination of twelve tea catechins by high-performance liquid chromatography with electrochemical detection.*Analyst,* 126(6), 816-820.

Shah, S. A., Yoon, G. H., Ahmad, A., Ullah, F., Amin, F. U. and Kim, M. O. 2015. Nanoscale-alumina induces oxidative stress and accelerates amyloid beta (Aβ) production in ICR female mice. *Nanoscale*, 7(37), 15225-15237.

Shixian, Q., VanCrey, B., Shi, J., Kakuda, Y. and Jiang, Y. 2006. Green tea extract thermogenesis-induced weight loss by epigallocatechin gallate inhibition of catechol-O-methyltransferase. *Journal of medicinal food*, 9(4), 451-458.

Sies, H. 1997. Oxidative stress: oxidants and antioxidants. Experimental Physiology: Translation and Integration, 82(2), 291-295.

Skrzypczak-Jankun, E., Zhou, K. and Jankun, J. 2003. Inhibition of lipoxygenase by (-)-epigallocatechin gallate: X-ray analysis at 2.1 Å reveals degradation of EGCG and shows soybean LOX-3 complex with EGC instead. *International journal of molecular medicine*, 12(4), 415-420.

Skutella, T. and Nitsch, R. 2001.New molecules for hippocampal development.*Trends in neurosciences*, 24(2), 107-113.

Vinson, J. A. 2000. Black and green tea and heart disease: a review. *Biofactors*, 13(1-4), 127-132.

****