ORIGINAL ARTICLE

Eur. J. Anat. 24 (5): 343-356(2020)

Memory state and hippocampal histomorphology in Wistar rat following monosodium glutamate-induced neurotoxicity; role of aqueous extract of Aloe Barbadensis

Olugbenga Ayannuga[1,2], Sinmidele Bamigboye[1], Ayodeji Abijo[1]

[1]Department of Anatomy and Cell Biology, Obafemi Awolowo University, Ile Ife, Nigeria
[2]School of Anatomical Sciences, University of Witwatersrand, South Africa

SUMMARY

This study assessed the effect of varying doses of aqueous extract of Aloe barbadensis on the cellular changes of hippocampal cells, oxidative and memory state of Wistar rats following monosodium glutamate-induced neurotoxicity.

Eighty Wistar rats (8 weeks) were randomly assigned into 4 groups of 20 rats; Group 1 received 3 mL/kg of distilled water. Groups 2, 3 and 4 received 3 g/kg/day of MSG. In addition, groups 3 and 4 received 100 and 200 mg/kg/day of AB extract respectively. Administration was done orally for 28 days in all groups. Five rats per group were sacrificed weekly over a 4-week period. Memory was assessed using radial arm maze on the last day of administration. Following brain harvest, one cerebral hemisphere was homogenized for oxidative state assessment, while the other was fixed in 10% neutral buffered formalin and stained with H&E for hippocampal histomorphology. Data obtained were analyzed using student t-test and p value < 0.05 was considered significant.

Across the 4-week period, group 2 rats showed significant increase in time spent to identify baited arms, significant reduction in density of apparently normal neurons and oligodendrocyte in CA 1-3 regions of hippocampus, and significant increase in reduced glutathione when compared with other groups. However, no significant differences were noted between groups 1, 3 and 4 for the above stated parameters.

The study concluded that MSG caused hippocampal neuronal and oligodendrocytes degeneration and impairment of memory. These anomalies are prevented by 100 and 200 mg/kg of Aloe barbadensis.

Key words: Hippocampus – Monosodium glutamate – Memory – Neuron – Oligodendrocyte – Oxidative stress

INTRODUCTION

Monosodium glutamate (MSG) is a flavor-enhancing food additive commonly found in fast foods as well as commercially packaged food products to improve their palatability (Vindini et al., 2010). It is added to food either as a purified monosodium salt or as a component of a mixture of amino acids and small peptides resulting from the acid or enzymatic hydrolysis of proteins (Schwartz, 2004). MSG is distributed under trade names such as Ajinomoto and Vedan, and it is rapidly absorbed

Corresponding author: Dr. Olugbenga Ayannuga. Obafemi Awolowo University, Dept. of Anatomy and Cell Biology, 228202 Ile-Ife, Nigeria. Phone: +2348080710899.
E-mail: olugbengayannuga@gmail.com

Submitted: 20 January 2020. *Accepted:* 5 May, 2020.

from the gastrointestinal tract resulting in a plasma level spike (Schwartz, 2004). The average daily intake of MSG for an individual in industrialized countries is 0.3-1.0 g. It can be higher occasionally, depending on the MSG content of individual food items and an individual's taste preferences (Geha et al., 2000). Glutamate is the most abundant amino acid in the central nervous system; it is found in high concentrations in regions of the brain that coordinate cognitive processes (Cekic et al., 2005). Prolonged high levels of plasma glutamate have been related to acute neuronal necrosis (Allent et al., 1987). Excitotoxic and oxidative pathways are the two distinct pathways through which glutamate induces neuronal death (Rajagopal et al., 2012).

A sustained high concentration of MSG in the synaptic cleft results in excessive glutamate receptor activation, with persistent depolarization producing metabolic and functional exhaustion of the neurons and subsequent necrosis (Gill, 2000; Mattson, 2008). Increased levels of glutamate results in increased calcium entry, oxidative stress with generation of free radicals, mitochondrial dysfunction, and eventually apoptosis (Goldsmith, 2000; Daniels and Brown, 2001; Jiang et al., 2005), leading to a form of cellular injury known as oxidative glutamate toxicity (Tojo et al., 2002; Audebert et al., 2002).

Monosodium glutamate has been reported to overstimulate nerve cells, resulting in cell damage or death (Bojanic et al., 2004). Neonatal MSG overdose results in disorder of learning and memory, while in older rats it causes neurodegenerative diseases, obesity, infertility, Parkinson's disease and epilepsy (Narayanan et al., 2010).

Aloe barbadensis is a well-known plant with varied medicinal properties. Its phytochemistry revealed the presence of different biologically active substances including vitamins, minerals, enzymes, sugars, anthraquinones or phenolic compounds, lignin, saponins, sterols, amino acids and salicylic acid (Chauhan et al., 2007). It has been proven to have a protective effect on neurons by preventing mitochondrial damage (Wang et al., 2010).

The antioxidant and mitochondrial protecting capacities of Aloe barbadensis makes it a possible neuroprotective agent that might be useful in preventing the neurodegenerating consequences of MSG-induced neurotoxicity. The widespread use of MSG in food preparation will be difficult to curtail, so the possible protective role of Aloe barbadensis in MSG-induced neurotoxicity is the focus of this study.

MATERIALS AND METHODS

Ethical approval for the study was obtained from the Health Research and Ethics Committee of the Institute of Public Health, Obafemi Awolowo Ile-Ife (HREC, IPH OAU). The animals received humane care according to the guidelines of IACUC.

Animal preparation

Wistar rats (Rattus novergicus) were bred in the animal holding of the Department of Anatomy and Cell Biology. Eighty eight-week-old Wistar rats of both sexes were used for this study, with a weight ranging from 60g to 90 g. The rats were randomly assigned into 4 groups (n=20). Rats were housed in clean plastic cages in a clean, well ventilated environment at room temperature. Rats in all groups were fed with standard laboratory rat chow and allowed access to water ad libitum. Natural light and dark cycle was maintained for the period of the study.

Animal grouping

Rats were randomly divided into 4 groups (1, 2, 3 and 4), each containing 20 rats. Group 1 was the control group and was administered 3 mL/kg of distilled water daily. Group 2, 3 and 4 rats were administered 3 g/kg body weight/day of MSG dissolved in distilled water. Groups 3 and 4 rats were simultaneously administered 100 and 200 mg/kg/day of Aloe barbadensis extract respectively. All administration was by oral cannula for a period of 28 days.

Materials

Monosodium Glutamate (trade name Aji-No-Moto) packaged by West African Seasoning Company, Lagos, was procured from a local market in Ile Ife, Osun State with NAFDAC registration number: 01-0298 and Batch No: 0321N. A stock solution of monosodium glutamate was constituted by dissolving 50 g in 100 mL of distilled water.

Aloe barbadensis was obtained from Sammy Agro Horticultural Services, Ile Ife and identified by a taxonomist in the Department of Botany, Obafemi Awolowo University, Ile Ife. A voucher specimen was deposited at Ife herbarium with number IFE 17633 for future reference. Extraction of Aloe barbadensis was done using the modified method of Wabeya et al., 2012. Mature, healthy and fresh Aloe barbadensis leaves were washed with fresh water, and the thick epidermis was removed. Water (50 mL) was added to the gel (500 g) and was homogenized with an electric blender, then concentrated using a vacuum rotary evaporator and freeze dried with a lyophilizer. The thickened paste was stored in a desiccator for further use. It was weighed and dissolved in distilled water for administration to rats.

Neurobehavioral assessment

Memory was assessed using a radial arm maze; this was carried out on rats in the four groups 3 hours before sacrifice. Rats had 2 sessions of training before the final assessment prior to sacrifice. Rats were placed on a central platform from where they accessed hidden baits placed at the

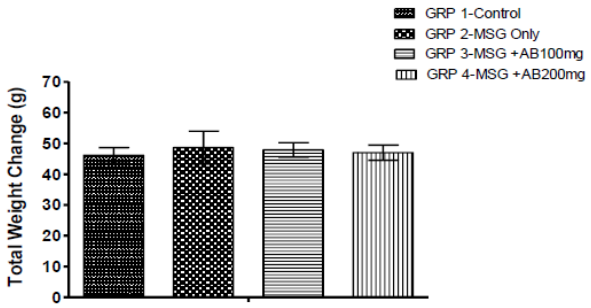

Fig 1. Bar chart showing Total Weight Change across groups.

end of the arms. The time taken for the rat to identify the baited arm was noted, and the maze was cleaned with ethanol in between tests.

Animal sacrifice

Rats were sacrificed 1, 2, 3 and 4 weeks after the last administration of MSG and Aloe barbadensis extract by cervical dislocation. The brain was harvested and a cerebral hemisphere was homogenised in 10 mL of phosphate buffer solution (0.1 M), using an electric homogenizer. The homogenate was centrifuged at 3000 rpm for 20 minutes, and the supernatant was collected for estimation of the level of glutathione, which is an indicator of oxidative stress. The other hemisphere was fixed by immersion in 10% neutral buffered formalin. One-millimeter-thick coronal brain slice was obtained and processed for routine paraffin embedding.

Estimation of Reduced Glutathione (GSH)

Reduced glutathione (GSH) content of the brain homogenate was assayed according to the procedure that was based on the method of Moron et al. (1979). 100 µl of brain homogenate was pipetted into clean dry sterile test tubes. The brain homogenate was deproteinized with 400 µl of 5% (w/v) trichloroacetic acid TCA, and then centrifuged for 10 min at 4000 rpm. The deproteinized supernatant was collected and the precipitate discarded. To 10 µl of the supernatant was added 90 µl of 0.2 M sodium phosphate buffer (pH 8.0) and 200 µl of 0.6 mM DTNB (freshly prepared in sodium phosphate buffer).

The reaction mixture was incubated for 10 min at room temperature. The absorbance was read at 412nm against the blank (prepared by replacing the brain homogenate with distilled water). The standard GSH curve was constructed by pipetting varying volumes (0, 0.2, 0.4, 0.6, 0.8 and 1 mL) of 10 mg/L standard GSH in triplicates. The volumes were adjusted to 1 mL with distilled water and treated as done for supernatant. The standard calibration curve was prepared by plotting the absorbance of the standard against the concentration of standard GSH. Brain homogenate GSH content was then extrapolated from the glutathione standard curve and the values were expressed in mg/g.

Histopathological assessment

From the brain tissue fixed in 10% neutral buffered formalin a coronal section of 1mm thickness brain slice was obtained at the level of the optic chiasma and processed through routine paraffin. Five (5) µm thick sections were obtained from the brain slice and stained with haematoxylin and eosin for demonstration of the general morphology of the hippocampus. Routine H&E staining was used to demonstrate the general architecture of neurons.

Photomicrography and Image analysis

Stained sections were viewed under a Leica DM 750 microscope, and digital photomicrographs were taken by an attached Leica ICC 50 camera. Image J was used for the analyses of neuronal population in each cortical layer.

Statistical Analysis

Data are presented as mean ± standard error of the mean (mean ± SEM), and the data obtained were analyzed by inferential and descriptive statistics using one-way analysis of variance (ANOVA) and t-test for dual group comparison. P value less than 0.05 ($p<0.05$) was taken to be statistically significant.

RESULTS

There was no significant difference when total weight gained in group 1 was compared with group 2 ($p = 0.6778$), group 3 ($p = 0.6159$), and group 4 ($p = 0.8113$). Similarly, comparison between groups 3 and 4 showed no significant difference ($p = 0.7945$) (Fig. 1).

In the first week post-administration rats, there was significant increase in time spent to identify baited arms in Trial 1 when group 2 was compared with group 1 ($p = 0.0327$). There was significant increase in time spent in Trial 2 when group 2 was compared with group 1 ($p = 0.0346$). Trial 3 also showed significant increase in time spent to identify the baited arms when group 2 was compared with group 1 ($p = 0.0120$). There was significant increase in time spent to identify baited arms in Trial 1 when group 2 was compared with groups 3 and 4 ($p = 0.0497$) ($p = 0.0423$). There was also significant increase in time spent to identify baited arms in Trial 2 when group 2 was compared with groups 3 and 4 ($p = 0.0468$) ($p = 0.0382$). Likewise, when group 3 and 4 were compared with group 2 in trial 3 ($p = 0.0493$) ($p = 0.0443$) respectively.

The result of the second week post-administration rats showed significant increase in time spent to identify baited arms in Group 2 compared with Group 1 in Trial 1 ($p = 0.0450$), Trial 2

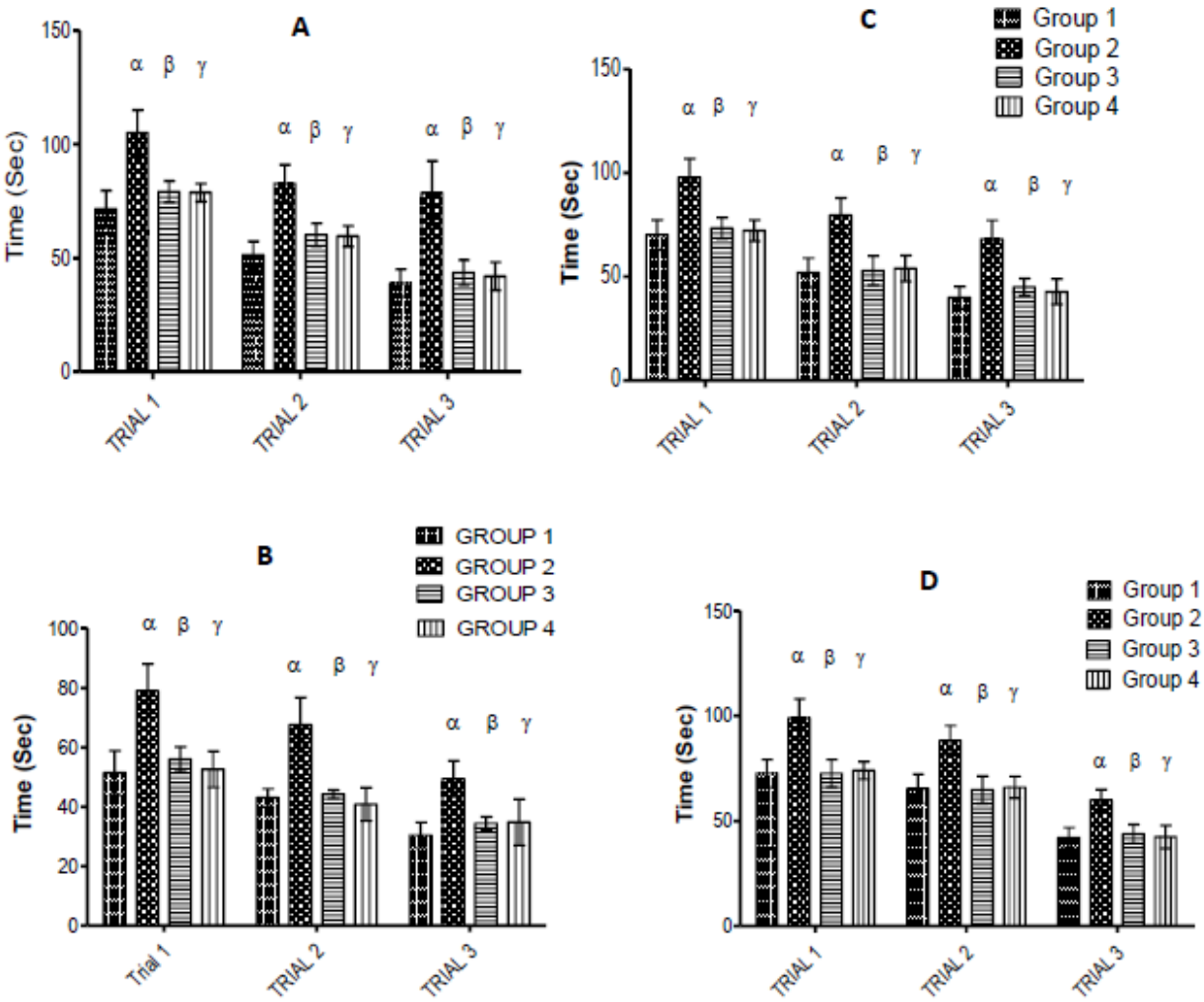

Fig 2. Bar chart showing Time Taken for Rats in Groups 1, 2, 3 and 4 to Identify the Baited Arms after 1 (**A**), 2 (**B**), 3 (**C**) and 4 (**D**) weeks across the three trials. α – significant difference between group 2 and group 1, β - significant difference between group 3 and group 1, γ - significant difference between group 4 and group 1. Data presented as Mean ± SEM.

(p = 0.0341) and Trial 3 (p = 0.0310). There was also significant increase in time spent to identify baited arms in group 2 compared with groups 3 and 4 in Trial 1 (p = 0.0488, 0.0410 respectively), Trial 2 (p = 0.0354, 0.0369 respectively) and Trial 3 (p = 0.0482, 0.0487 respectively).

In the third week post-administration rats there was significant increase in time spent to identify baited arms in Group 2 compared with Group 1 in Trials 1, 2 and 3 (p = 0.0431, 0.0359, 0.0266 respectively). There was significant increase in time spent to identify baited arms in group 2 compared with groups 3 and 4 in trial 1 (p = 0.0459, 0.0378 respectively), trial 2 (p = 0.0412, 0.0406, respectively) and trial 3 (p = 0.0469, 0.0474, respectively).

In the fourth week post-administration rats, significant increase in time to identify baited arms was noted in Group 2 compared with Group 1 in trials 1, 2 and 3 (p = 0.0483, 0.0494, 0.0361). There was also significant increase in time spent to identify baited arms in group 2 compared with groups 3 and 4 in trial 1 (p = 0.0481, 0.0383 respectively), trial 2 (p = 0.0414, 0.0362 respectively) and trial 3 (p = 0.0464, 0.0480 respectively) (Fig. 2).

One week after the interventions, there was a significant depletion in reduced Glutathione (GSH) levels (mg/g) when group 2 rats were compared with groups 1, 3 and 4 (p = 0.0177, 0.0482, 0.0393 respectively). In the second week, there was a significant depletion in GSH levels found in group 2 rats when compared with group 1 (p = 0.0213). There was significant depletion when groups 2 was compared with groups 3 and 4 (p = 0.0373, 0.0241 respectively). However, there was no significant difference when groups 3 and 4 was compared with group 1. After three weeks, there was a significant depletion in GSH levels found in group 2 rats when compared with group 1 (p = 0.0482, 0.0116, 0.0372 respectively). In the fourth week, there was a significant depletion in GSH levels found in group 2 rats when compared with groups 1, 3 and 4 (p = 0.0203, 0.0270, 0.0432, respectively) (Fig. 3).

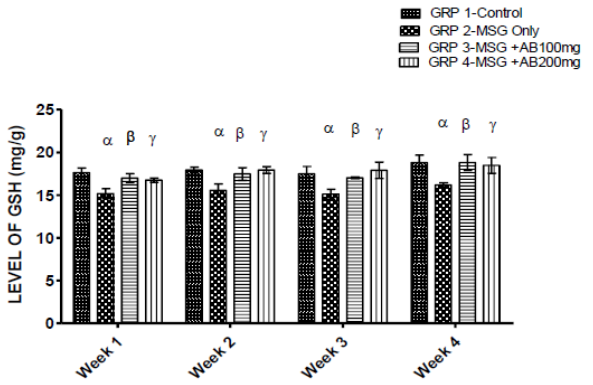

Fig 3. Bar chart showing the levels of GSH across the four groups over the four-week period. α – significant difference between group 2 and group 1, β - significant difference between group 3 and group 1, γ - significant difference between group 4 and group 1.

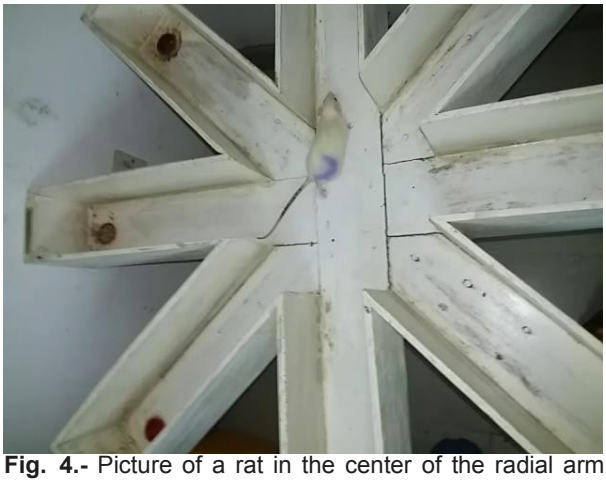

Fig. 4.- Picture of a rat in the center of the radial arm maze.

Hippocampal Histology

In the first week, CA1, CA2 and CA3 regions showed significant decrease in the density of normal neurons in group 2 when compared with group 1 (p = 0.0429, p = 0.0223 and p = 0.0313 respectively). However, there was no significant difference when groups 3 and 4 were compared with group 1. There was significant difference when groups 3 (p = 0.0447, p = 0.0226, p = 0.0305 respectively), and 4 (p = 0.0406, p = 0.0278, p = 0.0227 respectively) were compared with group 2 respectively. There was significant decrease in the density of normal oligodendrocytes in CA1, CA2 and CA3 regions in group 2 when compared with group 1 (p = 0.0438, p = 0.0143 and p = 0.0364 respectively). However, there was no significant difference when groups 3 and 4 were compared with group 1. There was significant difference when groups 3 (p = 0.0370, p = 0.0493, p = 0.0191 respectively), and 4 (p = 0.0431, p = 0.0379, p = 0.0259 respectively) were compared with group 2 respectively.

In the second week, CA1, CA2 and CA3 regions showed significant decrease in the density of normal neurons in group 2 when compared with group 1 (p = 0.0223, p = 0.0192 and p = 0.0437 respectively). However, there was no significant difference when groups 3 and 4 were compared with group 1. There was significant difference when groups 3 (p = 0.0500, p = 0.0059, p = 0.0406 respectively), and 4 (p = 0.047, p = 0.0465, p = 0.0329 respectively) were compared with group 2 respectively. There was significant decrease in the density of normal oligodendrocytes in CA1, CA2 and CA3 regions in group 2 when compared with group 1 (p = 0.0300, p = 0.0405 and p = 0.0324 respectively). However, there was no significant difference when groups 3 and 4 were compared with group 1. There was significant difference when groups 3 (p = 0.0171, p = 0.0485, p = 0.0432 respectively), and 4 (p = 0.0461, p = 0.0271, p = 0.0369 respectively) were compared with group 2 respectively.

In the third week, CA1, CA2 and CA3 regions showed significant decrease in the density of normal neurons in group 2 when compared with group 1 (p = 0.0034, p = 0.0482 and p = 0.0206 respectively). However, there was no significant difference when groups 3 and 4 were compared with group 1. There was significant difference when groups 3 (p = 0.0431, p = 0.0474, p = 0.474 respectively), and 4 (p = 0.0399, p = 0.0495, p = 0.0405 respectively) were compared with group 2 respectively. There was significant decrease in the density of normal oligodendrocytes in CA1, CA2 and CA3 regions in group 2 when compared with group 1 (p = 0.0452, p = 0.0419 and p = 0.0239, respectively). However, there was no significant difference when groups 3 and 4 were compared with group 1. There was significant difference when groups 3 (p = 0.0222, p = 0.0377, p = 0.0175, respectively), and 4 (p = 0.0460, p = 0.0347, p = 0.0178 respectively) were compared with group 2 respectively.

In the fourth week, CA1, CA2 and CA3 regions showed significant decrease in the density of normal neurons in group 2 when compared with group 1 (p = 0.0393, p = 0.062 and p = 0.0146 respectively). However, there was no significant difference when groups 3 and 4 were compared with group 1. There was significant difference when groups 3 (p = 0.0500, p = 0.0388, p = 0.0452, respectively), and 4 (p = 0.0499, p = 0.0123, p = 0.0229, respectively) were compared with group 2 respectively. There was significant decrease in the density of normal oligodendrocytes in CA1, CA2 and CA3 regions in group 2 when compared with group 1 (p = 0.0457, p = 0.0149 and p = 0.0272, respectively). However, there was no significant difference when groups 3 and 4 were compared with group 1. There was significant difference when groups 3 (p = 0.0335, p = 0.0407, p = 0.0369, respectively), and 4 (p = 0.0130, p = 0.0413 and p = 0.0345, respectively) (Figs 5-10).

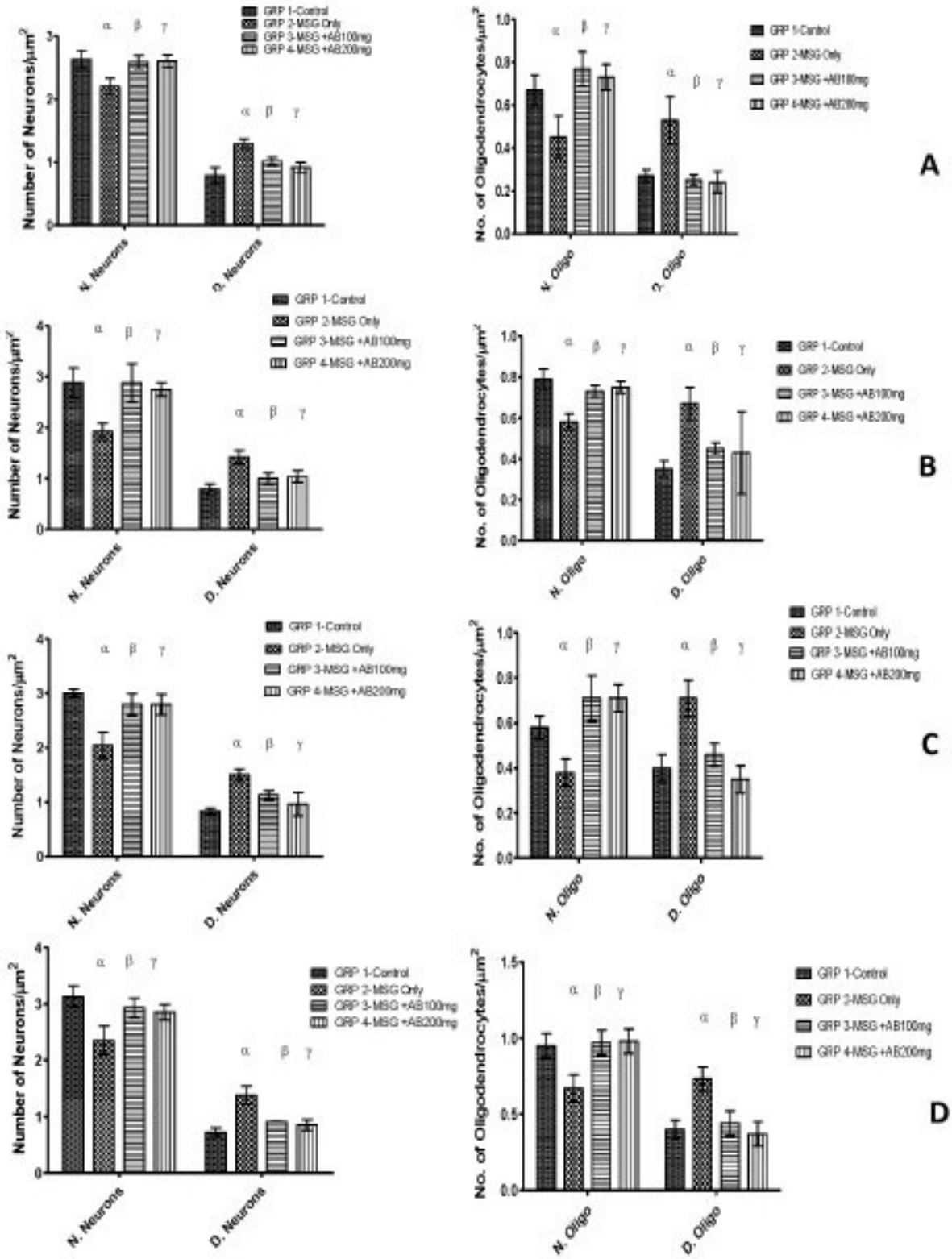

Fig 5. Bar charts showing the density of Neurons (Left Column) and Oligodendrocytes (Right Column) in the Cornus Ammonis1 in weeks 1 (**A**), 2 (**B**), 3 (**C**) and 4 (**D**) post-intervention. Significant differences between group 1 and 2 (α), group 1 and 3 (β), group 1 and 4 (γ) between normal (N) and degenerating (D) are shown. Data presented as Mean ± SEM (x10-4).

DISCUSSION

This study investigated the role of aqueous extract of Aloe barbadensis on the morphological, behavioral and biochemical consequences of monosodium glutamate - induced neurotoxicity in Wistar rats. The reports in literature were conflicting on the effect of MSG on body weight; some

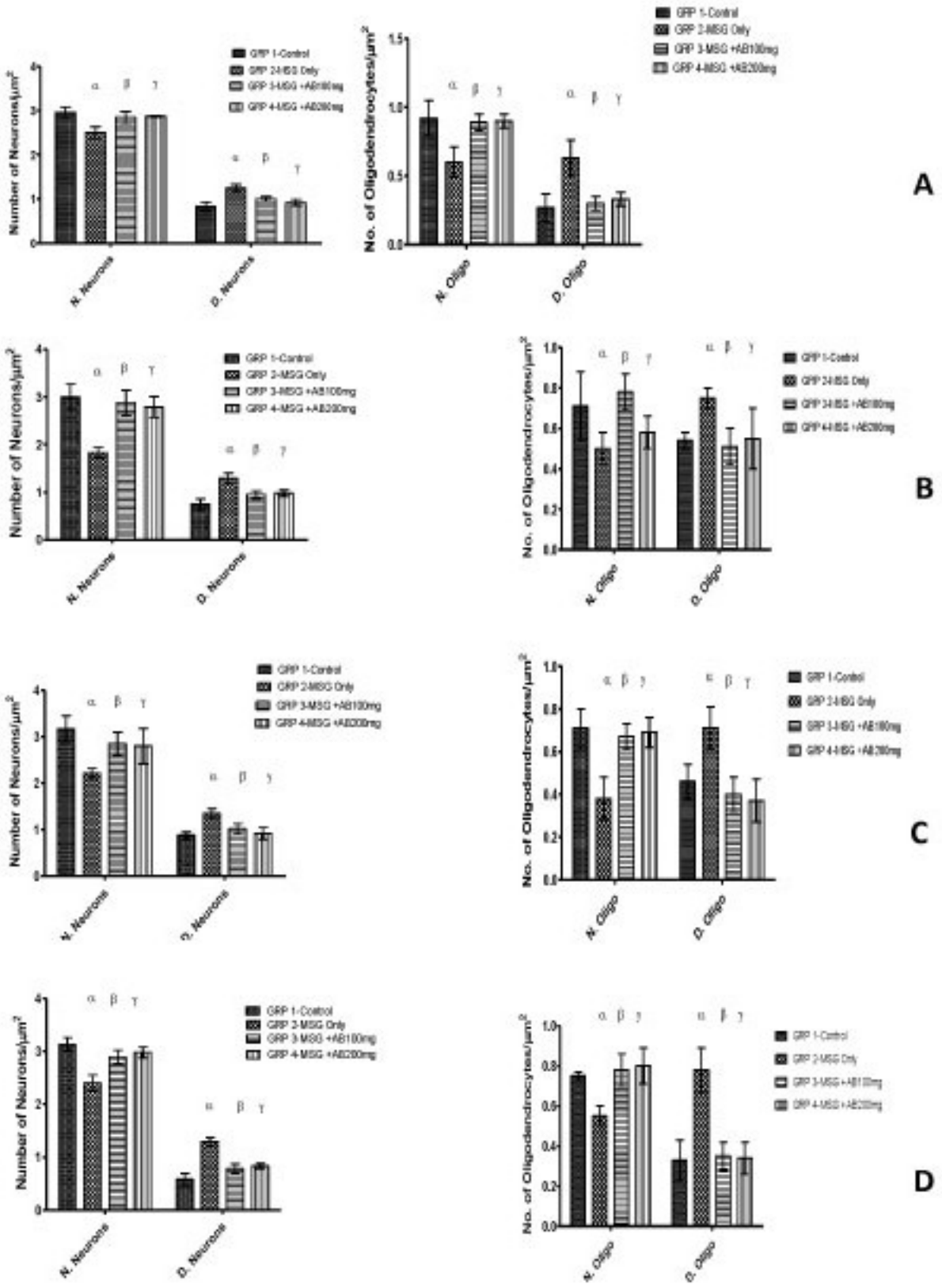

Fig 6. Bar charts showing the density of Neurons (Left Column) and Oligodendrocytes (Right Column) in the Cornus Ammonis2 in weeks 1 (**A**), 2 (**B**), 3 (**C**) and 4 (**D**) post-intervention. Significant differences between group 1 and 2 (α), group 1 and 3 (β), group 1 and 4 (γ) between normal (N) and degenerating (D) are shown. Data presented as Mean ± SEM (x10-4).

researchers reported a weight gain effect (Ashry et al., 2012; Wafaa et al., 2018); others reported a weight loss effect of MSG (Lima et al., 2013; Owoeye and Salami, 2017). In this study there was

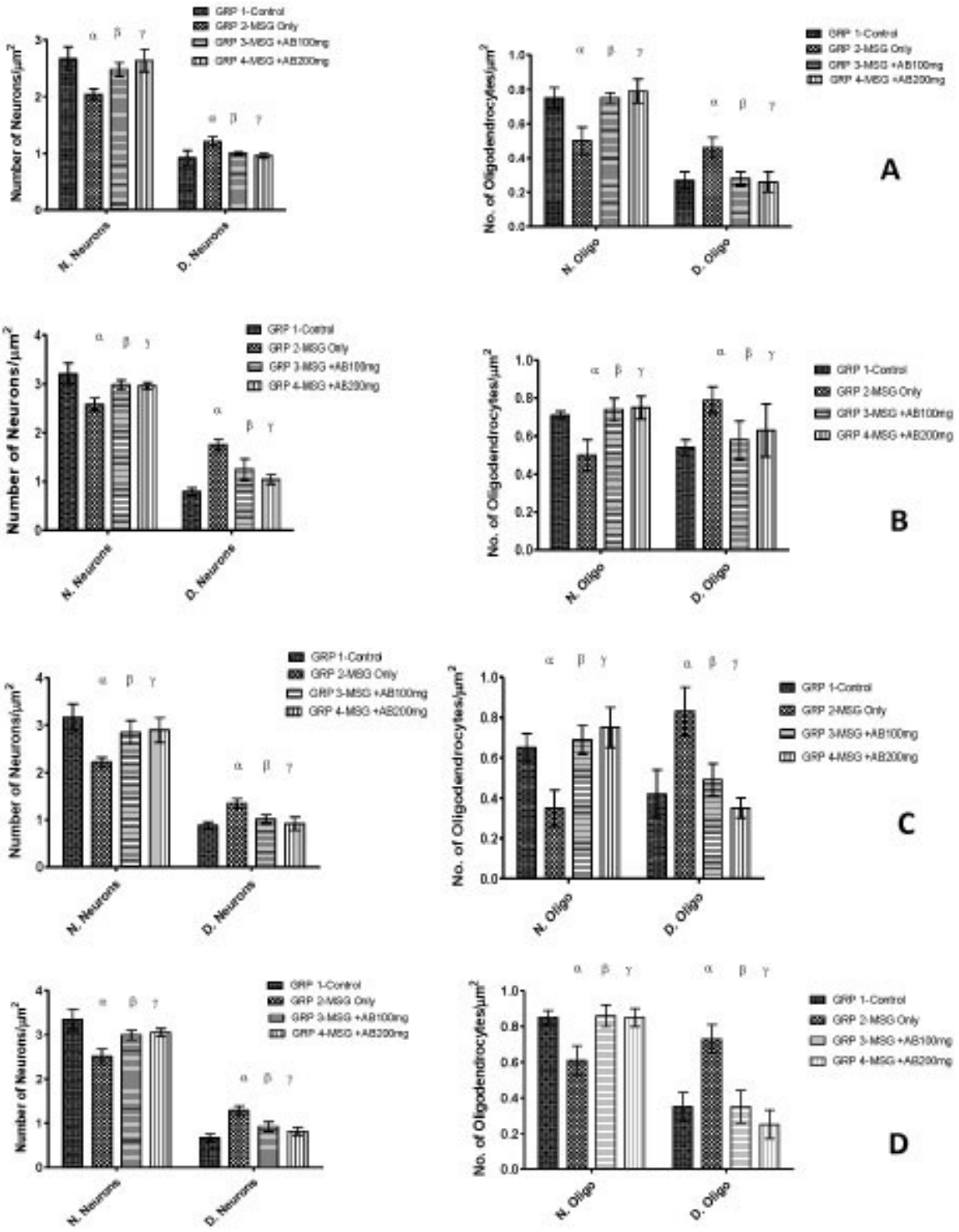

Fig 7. Bar charts showing the density of Neurons (Left Column) and Oligodendrocytes (Right Column) in the Cornus Ammonis3 in weeks 1 (**A**), 2 (**B**), 3 (**C**) and 4 (**D**) post-intervention. Significant differences between group 1 and 2 (α), group 1 and 3 (β), group 1 and 4 (γ) between normal (N) and degenerating (D) are shown. Data presented as Mean ± SEM (x10-4).

no significant consequence of MSG on the body weight of the Wistar rats over the four-week period, this being in consonance with the findings of Onyeama et al. (2012) and Onaolapo (2016). Since degenerative changes in the hypothalamus have been found to cause overweight, it might then explain the reported weight gain effect of MSG in some literature. However, the four-week duration of administration of MSG in this study may not be long enough to cause the degree of

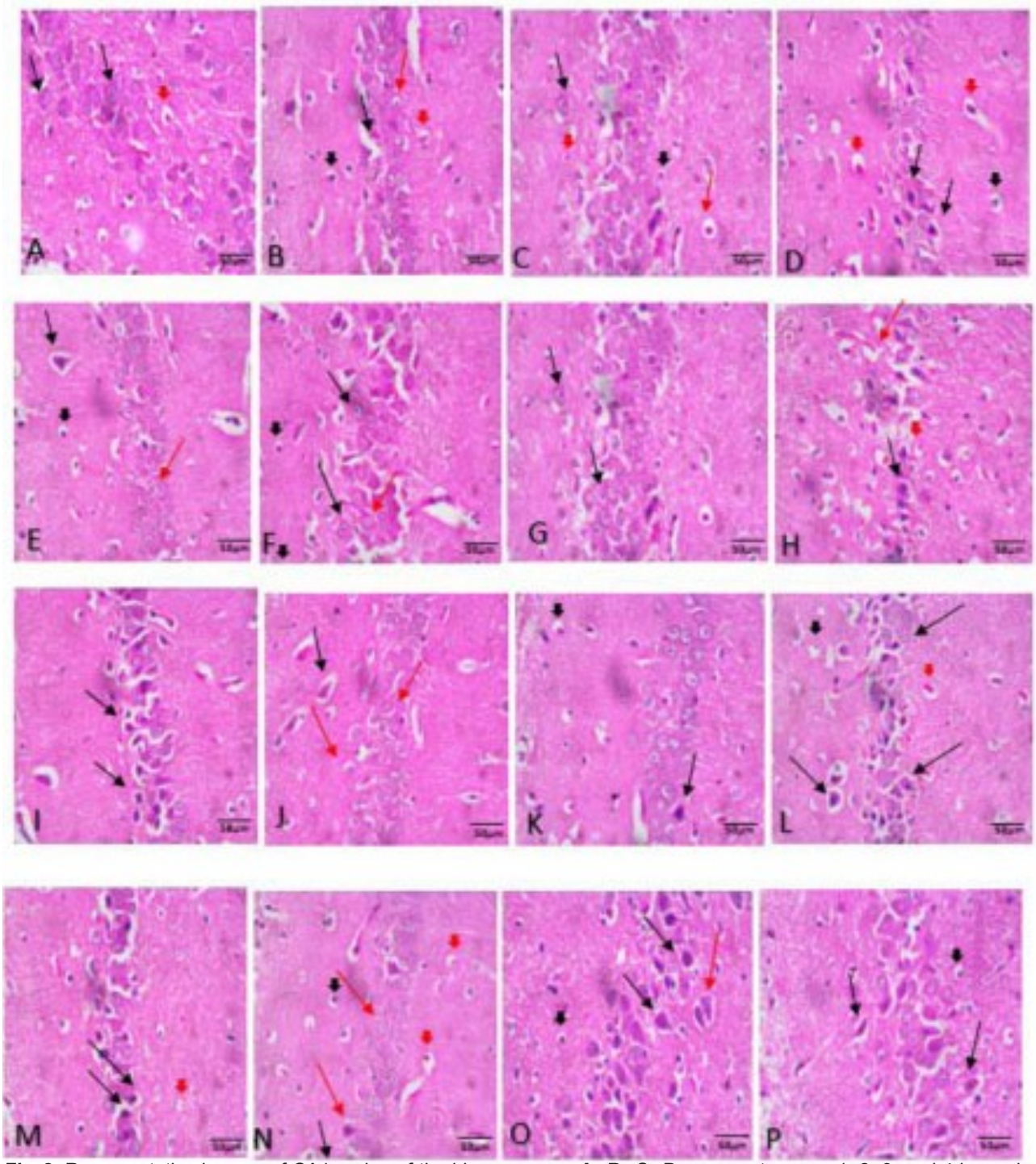

Fig 8. Representative images of CA1 region of the hippocampus. **A, B, C, D** represent groups 1, 2, 3, and 4 in week one. **E, F, G, H** represent the groups in week two, **I, J, K, L** represent week three and **M, N, O, P** represent week four. Red arrow (shrinking eosinophilic neurons), black arrow (normal neuron), black arrowhead (normal oligodendrocyte), red arrowhead (degenerating oligodendrocyte). Stain H&E. Scale bar: 50 µm.

hypothalamic lesion that is needed for weight gain. Though hippocampal cellular degeneration was noted in this study, it is suspected that a wider spread degeneration will be required for weight changes to ensue. In addition, the 100 mg and 200 mg/kg body weight of Aloe barbadensis extract does not appear to enhance weight gain over the 4 -week period.

Spatial memory was evaluated across the groups over the four-week period of the study, a significant increase in the time spent to locate the baited arm was noted in group 2 rats consistently in the 4 -week period showing an impairment of memory in the rats. Though a decline in time to locate the

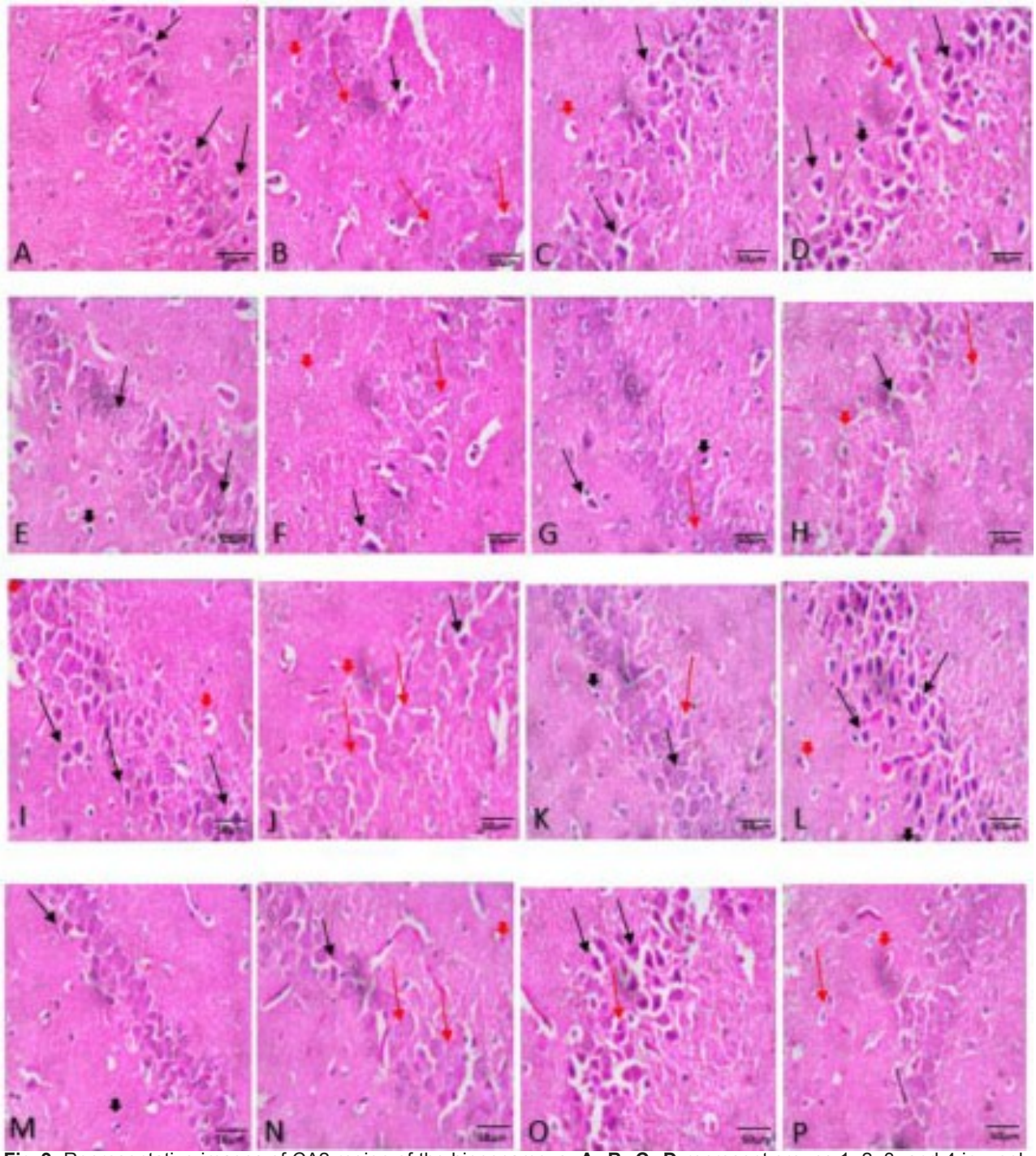

Fig 9. Representative images of CA2 region of the hippocampus. **A, B, C, D** represent groups 1, 2, 3, and 4 in week one. **E, F, G, H** represent the groups in week two, **I, J, K, L** represent week three and **M, N, O, P** represent week four. Red arrow (shrinking eosinophilic neurons), black arrow (normal neuron), black arrowhead (normal oligodendrocyte), red arrowhead (degenerating oligodendrocyte). Stain H&E. Scale bar: 50 µm.

baited arm was noted in all groups over the 3 trials, time taken by group 2 rats was significantly higher than other rats weekly. In addition, groups 3 and 4 that had different doses of Aloe barbadensis extract spent a time that was not significantly higher than that of the group 1 rats across the 4-week period. Therefore, while oral administration of MSG impaired memory in rats as early as one week, the memory impairment was prevented by concurrent administration of 100-200 mg/kg body weight of Aloe barbadensis extract. The hippocampus is known to play a critical role in the formation and preservation of memory (Sutula, 2003; Compton, 2004); the memory impairment effect of MSG has been attributed to its neurodegenerating role on the different parts of the hippocampus. Such neurodegeneration is known to be secondary to overstimulation of the neurons due to increased

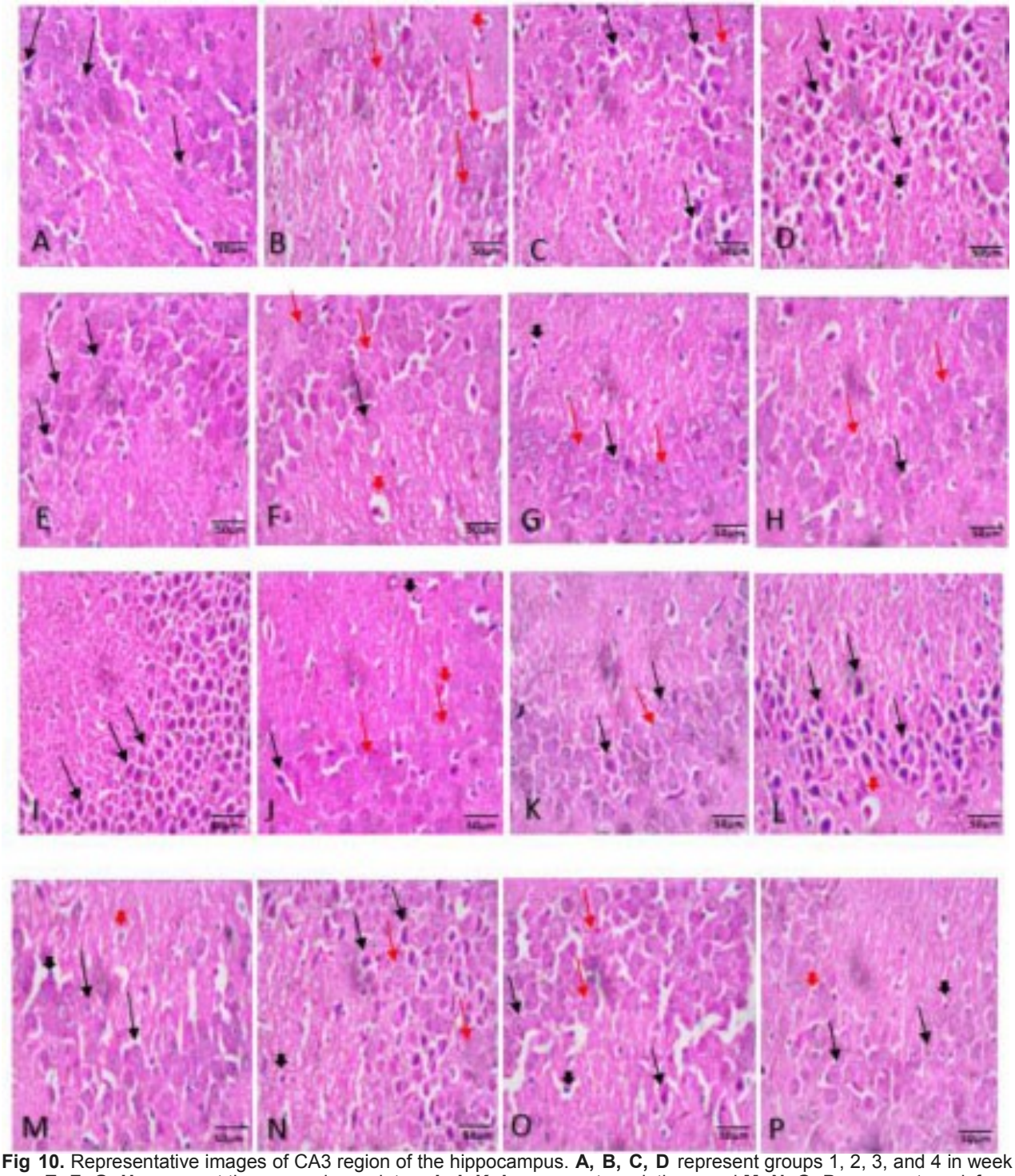

Fig 10. Representative images of CA3 region of the hippocampus. **A, B, C, D** represent groups 1, 2, 3, and 4 in week one. **E, F, G, H** represent the groups in week two, **I, J, K, L** represent week three and **M, N, O, P** represent week four. Red arrow (shrinking eosinophilic neurons), black arrow (normal neuron), black arrowhead (normal oligodendrocyte), red arrowhead (degenerating oligodendrocyte). Stain H&E. Scale bar: 50 µm.

concentration of glutamate in the synaptic cleft following MSG toxicity (Aysen and Eyüp, 2017). The counteracting effect of Aloe barbadensis could be as a result of enhanced metabolism of glutamate or attenuation of the glutamate receptors at the synaptic cleft.

In consonance with the findings on memory, a consistent significant decrease in neuronal density was observed in the group 2 rats when compared with the control rats. In addition, there was no significant difference in neuronal density of groups 1, 3 and 4. A consistent reverse finding was noted for degenerating neuron. These findings showed that the state of the hippocampal neurons is directly related to the memory status in MSG toxicity. While MSG was causing neurodegeneration as

explained above, Aloe barbadensis was preventing the adverse consequences of MSG on the neuron thereby leading to the preservation of memory. Phytochemical evaluation of Aloe barbadensis revealed an abundance of phenolic compounds which have been suspected to have strong antioxidant capacity (Priyanka, 2013).

Oligodendrocyte density in the CA1, 2 and 3 was consistent over the 4-week period which revealed that MSG does not only have cytotoxic effect on neurons but also on oligodendrocytes. However, the deleterious effect on oligodendrocyte was reversed significantly by 100 and 200 mg/kg body weight of Aloe barbadensis in a dose-dependent version. The supporting role of oligodendrocyte to the well-being of neurons is a possible link between the two cell types in the hippocampus. In addition, almost all cells of the brain have been found to have a type of glutamate receptor (Bergles et al., 2000), thereby making them susceptible glutamate induced excitotoxicity. However, whether the depletion of the oligodendrocyte is causal to or consequence of neuronal degeneration or whether the receptor types that facilitated excitotoxicity in neurons and oligodendrocyte are the same will be a subject of further studies. The depletion of oligodendrocyte might also result in a diminished hippocampal myelination which will subsequently result in delayed and energy demanding transmission of axonal impulses. This might make the process of memory formation and or recall more energy sapping or slower.

The significant decrease in GSH in group 2 rats across the 4-week period (Fig. 3) confirms the role of oxidative stress in MSG-induced neurotoxicity. The level of GSH in groups 3 and 4 rats which were not significantly different from that of the group 1 rats attested to the antioxidative capacity of Aloe barbadensis. Monosodium glutamate-induced neurotoxicity have been linked with oxidative stress, therefore, the antioxidant capacity of Aloe barbadensis must have been employed in preventing neurotoxicity. This shows that Aloe barbadensis functions as a naturally occurring antioxidant capable of mopping the reactive oxygen species generated by MSG. The high metabolic activities in the brain and the subsequent increased utilization of oxygen in the process make the brain vulnerable to oxidative stress. GSH antioxidative function is both intracellular and extracellular, making it a sensitive maker of oxidative stress in the brain. Aloe barbadensis at 100 mg/kg and 200 mg/kg body weight showed promises in mopping the ravaging reactive oxygen species thereby sparing the GHS content of the brain.

Oxidative-stress-related neurodegeneration is associated with structural disruption of the neuronal mitochondria, however, Aloe barbadensis have been reported to have a protective effect on the mitochondria of neurons (Wang et al., 2010), further enhancing its ability to prevent neurodegeneration following MSG toxicity. The abundance of flavonoids in Aloe barbadensis will also enhance its ability to maintain neuronal health as reported by Ofusori et al., (2007). It is known that the different subfields of hippocampus serve different purposes in the formation and consolidation of spatial memory (Andersen et al., 2007), protective effect of Aloe barbadensis on the architecture, morphology and density of pyramidal neurons and oligodendrocytes cuts across the Cornus ammonis 1, 2 and 3 of the hippocampus (Tables 1 to 4). These show that the ability of Aloe barbadensis to protect the hippocampus against MSG neurotoxicity is multifaceted, making it a candidate for intense research in the prevention of hippocampal neuronal degeneration from different disease conditions.

CONCLUSION

This study concludes that oral administration of 3 g/kg body weight of MSG for 28 days resulted in degeneration of hippocampal neurons and oligodendrocytes as well as oxidative stress and memory impairment. In addition, 100 mg and 200 mg/kg body weight of Aloe barbadensis is capable of preserving spatial memory, protecting hippocampal neurons and oligodendrocyte and preventing oxidative stress in MSG-induced neurotoxicity in Wistar rats. The protective effects of Aloe barbadensis on the cells of hippocampus, memory and brain oxidative state is not dose dependent but appeared to involve several mechanism making Aloe barbadensis a potential pharmacological candidate for protection against monosodium glutamate-induced neurotoxicity.

REFERENCES

ALLENT ZH, DELOHERY J, BAKER G (1987) Monosodium glutamate induced asthma. J Allergy Clin Immunol, 80: 530-537.

ANDERSEN JK (2004) Oxidative stress in neurodegeneration: cause or consequences. Nat Med, 10: S18-25.

ASHRY MA, ABD.ELLAH HF, GHETH EMM (2012) The possible ameliorative effect of propolis in rat's liver treated with monosodium glutamate (MSG). Nature Sci, 10(12).

AUDEBERT M, CHARBONNIER JB, BOITEUX S, RADICELLA JP (2002) Mitochondrial targeting of human 8-oxoguanine DNA glycosylase hOGG1 is impaired by a somatic mutation found in kidney cancer. DNA Repair (Amst), 17: 497-505.

AYŞEN ÇK, EYÜP B (2017) Investigation of the behavioral and neurochemical effects of monosodium glutamate on neonatal rats. Turkish J Med Sci Turk J Med Sci, 47: 1002-1011.

BERGLES DE, ROBERTS JD, SOMOGYI P, JAHR CE (2000) Glutamatergic synapses on oligodendrocyte precursor cells in the hippocampus. Nature, 405 (6783): 187-191.

BIRBEN E, SAHINER UM, SACKESEN C, ERZURUM S, OMER K (2012) Oxidative stress and antioxidant defense. World Allergy Organ J, 9-19.

BLAYLOCK RL (1994) Excitotoxin: The taste that kills. Santa Fe, NM. Health Press.

BOJANIC VV, BOJANIC Z, NAJMAN S, IVANOV-EURLIS J, TOMIN J, DINOIC B, SAVIC T (2004) Diltiazem prevention of monosodium glutamate toxicity on hypothalamus in Wistar rats. Arch Oncol, 12: 19-20.

COMPTON DM (2004) Behavior strategy learning in rat: effects of lesions of the dorsal striatum or dorsal hippocampus. Behav Process, 67: 335-342.

CEKIC S, FILIPOVIC M, PAVLOVIC V, CIRIC M, NESIC M, JOVIC Z, BRANKOVIC S (2005) Histopathologic changes at the hypothalamic, adrenal and thymic nucleus arcuatus in rats treated with monosodium. Acta Medica Medianae, 44: 35-42.

CHAUHAN OP, RAJU PS, KHANUM F, BAWA AS (2007) Aloe vera - pharmaceutical and food applications. Ind Food Ind, 26: 43-51.

DANIELS M, BROWN DR (2001) Astrocytes regulate N-methyl-D-aspartate receptor subunit composition increasing neuronal sensitivity to excitotoxicity. J Biol Chem, 276: 22446-22452.

GAO W, XIAO P (1997) Peroxidase and soluble protein in the leaves of Aloe vera L. var. Chinensis (Haw.) Berger. China J Chin Mat Med, 22(11): 653-654.

GARATTINI S (2000) Glutamic acid, twenty years later. J Nutr, 130: 901S-909S.

GEHA RS, BEISER A, REN C, PATTERSON R, PAUL A, GREENBERGER PA, LESLIE C, GRAMMER LC, DITTO AM, HARRIS KE, SHAUGHNESSY MA, YARNOLD PR, CORREN J, SAXON A (2000) Review of alleged reaction to monosodium glutamate and outcome of a multicenter double-blind placebo-controlled study. J Nutr, 130: 1058S-1062.

GILL SS (2000) Potential target sites in peripheral tissues for excitatory neurotransmission and excitotoxicity. Toxicol Pathol, 28: 277-284.

GOLDSMITH PC (2000) Neuroglial responses to elevated glutamate in the medial basal hypothalamus of the infant mouse. J Nutr, 130: 1032S-1038S.

JIANG SX, LERTVORACHON J, HOU ST, KONISHI Y, WEBSTER J, MEALING G, BRUNETTE E, TAUSKELA J, PRESTON E (2005) Chlortetracycline and demeclocycline inhibit calpains and protect mouse neurons against glutamate toxicity and cerebral ischemia. J Biol Chem, 280: 33811-33818.

JOSEPH B, RAJ SJ (2010) Pharmacognostic and phytochemical properties of Aloe vera Linn−An overview. Int J Pharmac Sci Rev Res, 4: 106-110.

KULJEET SA, VIKAS D (2012) Hippocampus in health and disease: An overview. Annals of Indian Academy Neurology.

LIMA CB, SOARES GDE S, VITOR SM, CASTELLANO B, ANDRADE DA COSTA BL, GUEDES RC (2013) Neonatal treatment with monosodium glutamate lastingly facilitates spreading depression in the rat cortex. Life Sci, 293: 388-392.

LOO BV, BACHSCHMID M, SPITZER V, BREY L, ULLRICH V, LUSCHER TF (2003) Decreased plasma and tissue levels of vitamin C in a rat model of aging: implications for antioxidative defense. Biochem Biophys Res Commun, 303: 483-487.

MARWA A, MANAL R, EL-HALEEM A (2011) Evaluation of monosodium glutamate-induced neurotoxicity and nephrotoxicity in adult male albino rats. Am J Sci, 7(8): 264-276.

MATTSON MP (2008) Glutamate and neurotrophic factors in neuronal plasticity and disease. Ann N Y Acad Sci, 1144: 97-112.

NALINI G, CHIDAMBARANATHAN N, SANTHANA-KUMAR M, JEYASUNDARI K, NIRUBAN C (2016) Neuroprotective effect of hydroalcoholic extract of Boerhaavia diffusa linn against MPTP-induced neurodegeneration in rats. Int J Pharmacol Toxicol, 6(2): 50-57.

NARAYANAN SN, KUMAR RS, PAVAL J, NAYAK S (2010) Effect of ascorbic acid on the monosodium glutamate-induced neurobehavioral changes in periadolescent rats. Bratisl Lek Listy, 111(5): 247-252.

PRIYANKA D, ALOK KS (2013) Phytochemical extraction and characterization of the leaves of aloe vera barbadensis for its antibacterial and anti-oxidant activity. Int J Sci Res (IJSR), 2319-7064.

OFUSORI D, AYOKA A, ADELAKUN A, FALANA B, ADEEYO O, AJEIGBE K, YUSUF U (2007) Microanatomical effect of ethanolic extract of Garcinia kola on the lung of Swiss albino mice. Internet J Pulmonary Med, 10(1).

ONAOLAPO OJ, ONAOLAPO AY, AKANMU MA, OLAYIWOLA G (2016) Evidence of in brain structure and antioxidant status following 'low-dose' monosodium glutamate ingestion. Pathophysiology, 23: 147-156.

ONYEMA OO, ALISI CS, IHETUGE AP (2012) Monosodium glutamate induces oxidative stress and affects glucose metabolism in the kidney of rats. Int J Biochem Res Rev, 2: 1-11.

OWOEYE O, SALAMI OA (2017) Monosodium glutamate toxicity: sida acuta leaf extract ameliorated brain histological alterations, biochemical and haematological changes in Wistar rats. Afr J Biomed Res, 20: 173-182.

RAJAGOPAL SS, LAKSHMINARAYANAN G, RAJESH R, DHARMALINGAM SR, RAMAMURTHY S, CHIDAMBARAM K, SRINIVASAN R (2013) Neuroprotective potential of Ocimum sanctum (Linn) leaf extract in monosodium glutamate induced excitotoxicity. Afr J Pharm Pharmacol, 7(27): 1894-1906.

SCHUBERT D, PIASECKI D (2001) Oxidative glutamate toxicity can be a component of the excitotoxicity cascade. J Neurosci, 21: 7455-7462.

SCHWARTZ JR (2004) In bad taste, the MSG "syndrome" MSG. The 5th Annual Conference of the Weston: A. Price Foundation.

SHIH AY, ERB H, SUN X, TODA S, KALIVAS PW, MURPHY TH (2006) Cystine/glutamate exchange modulates glutathione supply for neuroprotection from oxidative stress and cell proliferation. J Neurosci, 26: 10514-10523.

SHIVASHARAN BD, NAGAKANNAN P, THIPPESWAMY BS, VEERAPUR VP (2013) Protective effect of Calendula officinalis L. flowers against monosodium glutamate- induced oxidative stress and excitotoxic brain damage in rats. Indian J Clin Biochem, 28(3): 292-298.

SULTANA R, PERLUIGI M, ALLAN DB (2013) Lipid peroxidation triggers neurodegeneration: A redox proteomics view into the Alzheimer disease brain. Free Radic Biol Med, 62: 157-169.

SUTULA TP, HAGEN J, PITKANEN A (2003) Do epileptic seizures damage the brain? Curr Opin Neurol, 16: 189-195.

TOJO A, ONOZATO ML, KOBAYASHI N, GOTO A, MATSUOKA H, FUJITA T (2002) Angiotensin II and oxidative stress in Dahl salt-sensitive rat with heart failure. Hypertension, 40: 834-839.

VINDINI NA, NAYANTARA AK, RAMASWAMY C, GOWDA D, AHMED B, BHAT R (2010) Study on evaluation of monosodium glutamate-induced oxidative damage on renal tissue on adult Wistar rats. J Chin Clin Med, 3: 112-115.

WABEYA B, DAVIDSON B, ERLWANGER K (2012) The effects of crude aqueous and alcohol extracts of aloe vera on growth and abdominal viscera of suckling rats. African J Tradit Compl Altern Med AJTCAM / African Networks on Ethnomedicines, 9: 553-560.

WAFAA M, ABDEL MONEIM HA, YASSA RA, NADA AM (2018) Monosodium glutamate affects cognitive functions in male albino rats. Egypt J Forensic Sci, 2018:89.

WANG H, LI F, WANG T, LI J, LI J, YANG X, LI J (2004) Determination of aloin content in callus of Aloe vera var. chinensis. Zhong Yao Cai, 27(9): 627-628.

WANG Y, CAO L, DU G (2010) Protective effects of Aloe vera extract on mitochondria of neuronal cells and rat brain. Zhongguo Zhong Yao Za Zhi, 35(3): 364 -368.