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Ameliorative Effects of Taurine and Caffeine on Spatial Memory Deficit Associated with Neuropathic Pain in Sciatic Nerve Ligated Rats

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ABSTRACT

Despite several clinical studies which showed that majority of chronic pain sufferers have impaired spatial memory, yet, this remains an area of unmet therapeutic need. In this study, we tested whether effect of concurrent administration of taurine and caffeine could restore the spatial memory impairment induced by neuropathic pain. Rats were rendered neuropathic by unilateral sciatic nerve ligation and treated with taurine and caffeine (taurine (200 mg/kg b.w., i.p) + caffeine (7.5 mg/kg b.w., i.p.) and taurine (200 mg/kg b.w., i.p.) + caffeine (15 mg/kg b.w., i.p.). After 2 weeks of administration, the spatial memory was assessed using Y-maze paradigm. The numbers of entries and percentage alternations over a ten minute period was scored for each animal as a measure of spatial memory. Thereafter the animals were sacrificed and blood samples were collected for the estimation of serum C-reactive protein (CRP), a pro-inflammatory marker. Administration of taurine and caffeine ameliorated spatial memory impairment induced by neuropathic pain as percentage alternation increased significantly ($p < 0.05$) in the groups that received both taurine and caffeine in a dose-dependent manner compared to the ligated control group but there was no significant effect on the locomotor activities as shown by the arm entries. There was also a significant ($p < 0.05$) decrease in the serum CRP level in the groups that were treated with taurine alone as well as taurine and caffeine compared to the ligated control group. It can be concluded that administration of taurine and caffeine ameliorated the spatial memory impairment induced by neuropathic pain.

Keywords: *Caffeine, C-reactive protein, neuropathic pain, memory impairment, rats, taurine*

INTRODUCTION

Chronic pain is a major health concern which accounts for 80% of patients' visit to physicians (Gatchel 2004); clinical studies have shown that about 65% of chronic pain patients exhibit disruption of working memory which is a form of temporary method of information storage (Dick and Rashiq 2007) on which individual achievements in modern life depend (Alloway et al. 2009). In recent years, numerous studies have been carried out to explain why chronic pain patients suffer from cognitive deficit but the results were controversial. Mutso et al. (2012) showed that the cognitive deficit is due to atrophy of hippocampal formation which is essential in learning and memory processing. Eccleston (1995) suggested that memory deficit could result from disruption of attention by pain. While Hart et al. (2000) proposed that the chemical substances produced in chronic pain conditions pain conditions such as pro-inflammatory cytokines may influence function of neural circuitries, leading to the impairment of

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attention and memory.

Caffeine - the most widely consumed psychoactive substance, at non-toxic doses acts as an antagonist of adenosine A1 and A2 receptors (Fredholm et al. 2005). Caffeine consumption has been shown to alleviate cognitive impairment in both humans and animals (Takahashi et al. 2008; Cunha and Agostinho 2010) and affords protection upon central nervous system injury (Cunha 2005). It has been shown that long-term caffeine consumption can prevent both diabetes-associated memory impairments and diabetes-induced astrogliosis and loss of nerve terminal markers in the hippocampus (Duarte et al. 2012).

Taurine on the other hand is a simple sulphur-containing amino acid present in virtually all cells throughout the animal kingdom. In particular, it is enriched in electrically excitable tissues. Neuroprotective effect of taurine has been documented, for example it is effective in a rat hypoxic model, possibly due to its anti-acidotic and membrane stabilizing actions (Mankovskaya et al. 2000). It improves the recovery of neural functions in brain slices after hypoxic conditions (Schurr et al. 1987) and protects neurons from glutamate-induced excitotoxicity (Mutso et al. 2012), probably by preventing or reducing the glutamate-induced elevation of intracellular Ca2+ (Chen et al. 2001).

A previous study from our laboratory showed that, taurine and caffeine when administered together, could attenuate pain related behaviour associated with neuropathic pain in an animal model (Abdulmajeed and Owoyele 2015). This study therefore sought to determine whether or not co-administration of the two substances could also restore the memory impairment that is associated with neuropathic pain.

MATERIALS AND METHODS

Drugs and reagents

Taurine and caffeine were products of Sigma chemical Co. (St. Louis MO, U.S.A) while CRP levels was determined using commercially available ELISA kit (Alpha diagnostic, San Antonio, TX, United States).

Animals

A total of thirty six age-matched male Wistar rats (*Rattus norvegicus*) weighing 130±12.5 g were employed in the study. They were placed under standard laboratory condition in cages at the animal holding of the Faculty of Basic Medical Sciences, University of Ilorin, Ilorin, Nigeria. They were fed on standard chow diet and water *ad libitum* and were acclimatized to laboratory conditions for two weeks before the commencement of experiments. The animals were treated in accordance with the ethical guidelines of the University of Ilorin and the guidelines agreed with the internationally accepted principles for animal handling and care.

Experimental protocol

Animals were randomly divided into six groups consisting of six animals each as follows:

Group 1 (vehicle control): Rats were not subjected to any surgical procedure or treatment and were kept for 14 days. Animals in this group received normal saline (2 ml/kg i.p).The behavioural test was employed on day 14. Thereafter, the animals were sacrificed and blood was collected for the estimation of serum CRP level.

Group 2 (ligated control): Rats were subjected to the surgical procedure to expose and ligate the left sciatic nerve. The behavioural test and serum CRP analysis were done as for group 1 animals. Animals in this group also received normal saline (2 ml/kg, i.p.).

Group 3 Rats were subjected to the surgical procedure to expose and ligate the left sciatic nerve and received taurine (200 mg/kg, i.p.) alone once daily for two weeks. Behavioural test and serum CRP estimation were also carried out as described for previous groups.

Group 4: Rats were subjected to the surgical procedure to expose and ligate the left sciatic nerve and received caffeine (15 mg/kg, i.p.) alone for two weeks. Behavioural test and serum CRP estimation were also carried out as described for previous groups.

Group 5: Rats were subjected to sciatic nerve ligation and treated with taurine (200 mg/kg, i.p.) + caffeine (7.5 mg/kg, i.p.) for two weeks. The Behavioural test and serum CRP estimation were also carried out as described for previous groups.

Group 6: Rats were subjected to sciatic nerve ligation and treated with taurine (200 mg/kg i.p.) + caffeine (15 mg/kg, i.p.) for two weeks. The Behavioural test and serum CRP estimation were also carried out as described for previous groups.

Induction of neuropathic pain

Neuropathic pain was induced by chronic constriction injury in line with the modified method of Bennette and Xie (1998). The rats were each anaesthetized with 50 mg/kg ketamine and the skin of the lateral surface of the left thigh was incised. Thereafter, a cut was made directly through the biceps femoris muscle to expose the sciatic nerve. Once exposed, the sciatic nerve was tightly ligated with silk 4-0 thread at two sites with about 1mm gap (Kumar et al. 2010). Extra care was taken to avoid interruption of epineural blood flow while tying the ligature. After performing the ligation, muscular and skin layers were immediately sutured with thread and topical antibiotic was applied. Behavioural test was carried out on day 14 using Y-maze.

Behavioural tests

The behavioural test was conducted in a large quiet room between the hours of 7 and 11 a.m. Spatial memory and locomotor activities were assessed using the Y-maze. All events were observed manually.

Y-maze

The Y-maze is used to assess short term memory, general locomotor activity and stereotypic behaviour (Kokkinidis et al., 1976). Therefore, spontaneous alternation which is a measure of spatial working memory was assessed using a Y- maze composed of three equally spaced arms (120°,41cm long and 15 cm high) and the floor of each arm was made of Perspex that are 5 cm wide (Onaolapo et al. 2012). Each rat was placed at the centre of the maze and was allowed to move freely for a period of 10 min after which the apparatus was cleaned with 5% alcohol and allowed to dry between sessions.

The sequence of arm entries was manually recorded, the arms being labelled A, B, or C. An alternation is defined as entry into all three arms consecutively, for instance if the animal makes the following arm entries; ACB,CA,B,C,A,CAB,C,A, in this example, the animal made 13 arm entries 8 of which are correct alternations. The number of maximum spontaneous alternations is then the total number of arms entered minus two, and the percentage alternation is calculated as {(actual alternations /maximum alternations) x 100} (Onaolapo et al. 2010).The number of arm entries was used to determine the locomotor activity.

Statistical analysis

The results were expressed as mean ± standard error of the mean (SEM). The results were analyzed using one way Analysis of variance (ANOVA), followed by DUNCAN post-hoc test. P value <0.05 was considered to be statistically significant.

RESULTS

Effect of co-administration of taurine and caffeine on spatial memory

Figure 1 shows the effects of concurrent administration of taurine and caffeine on spatial memory following 10 minutes of exposure in the Y maze. Sciatic nerve ligation resulted into spatial memory impairment as shown by reduction in percentage alternation in the ligated control group compared to the vehicle control group from 75.57 to 56.79% ($p<0.05$).Treatment with caffeine alone did not have a significant effect on the percentage alternation but taurine treated group showed significant ($P<0.05$) increase in percentage alternation from 56.79 to 71.94% compared to the ligated control group ($p<0.05$). However, concurrent administration of taurine and caffeine resulted in an increase in percentage alternation compared to the ligated control group ($p<0.05$).

Effect of co-administration of taurine and caffeine on Y- maze locomotor activity

Figure 2 shows the effects of co-administration of taurine and caffeine on locomotor activity following 10 mins of exposure in the Y maze. Sciatic nerve ligation resulted in reduction in locomotor activity as shown by significant ($p<0.05$) decrease in total arm entries/10mins in the ligated control group from 13 to 8 compared to the vehicle control group ($P<0.05$). Though, treatment with taurine and caffeine alone significantly ($p<0.05$) increased the total arm entries/10min but co-administration of the two substances did not produce a significant effect on locomotor activity.

Effect of co-administration of taurine and caffeine on serum CRP level

Table 1 shows that ligation of sciatic nerve led to increase in serum CRP level. Treatment with caffeine alone did not produce significant effect on the serum CRP level but there was significant ($P<0.05$) reduction in the serum CRP level in the groups that received taurine alone (1.77mg/dl vs 0.91mg/dl, $p<0.05$) and those that received taurine and caffeine together compared to the ligated control group (0.77-1.14 vs 1.77).

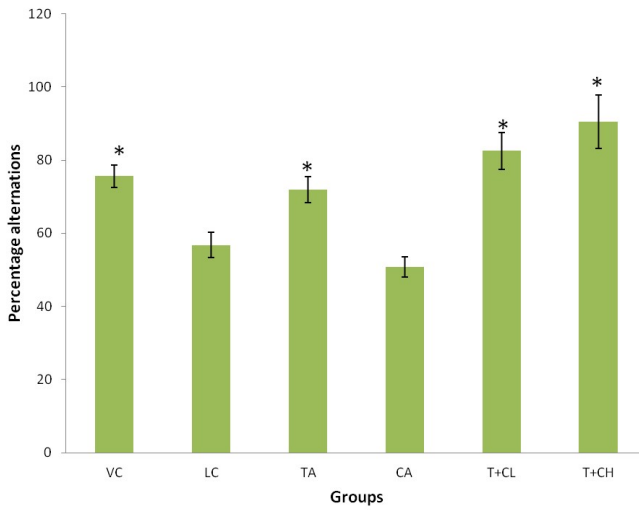

Fig. 1: shows the effects of concurrent-administration of taurine and caffeine on spatial memory following 10minutes of exposure in the Y-maze. * $P < 0.05$ compared with ligated control animals; ANOVA. VC= vehicle control, LC= Ligated control, TA= taurine alone, CA= caffeine alone ,T+CL= taurine (200mg/kg, i.p) + caffeine (7.5mg/kg, i.p), TCH= taurine (200mg/kg, i.p) + caffeine (15mg/kg, i.p). (n=6).

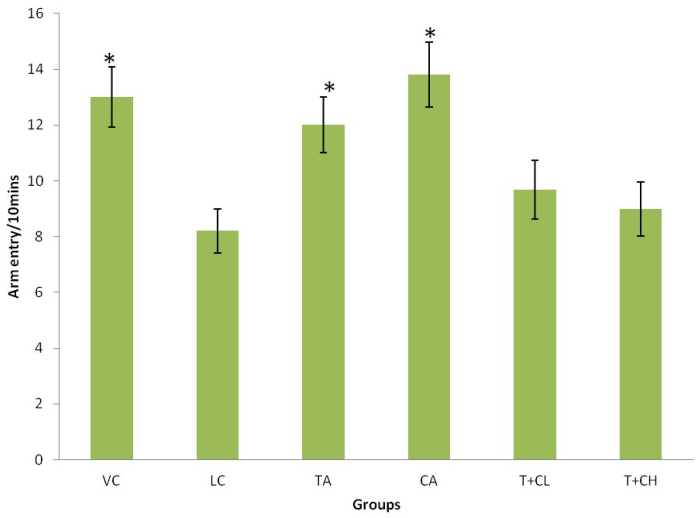

Fig. 2: shows the effects of administration of taurine and caffeine on locomotor activity following 10mins of exposure in the Y maze. * P < 0.05 compared with ligated control animals; ANOVA.VC= vehicle control, LC= Ligated control, TA= taurine alone, CA= caffeine alone ,T+CL= taurine (200mg/kg, i.p) + caffeine (7.5mg/kg, i.p), TCH= taurine (200mg/kg, i.p) + caffeine (15mg/kg, i.p).(n=6).

DISCUSSION

Memory deficit associated with chronic pain has been well documented (Green 2006; Moriarty et al. 2011) yet it remains an area of unmet therapeutic need, hence we sought to investigate whether or not taurine when administered with caffeine could restore the short term memory impairment associated with neuropathic pain in male Wistar rats. The result indicated that neuropathic pain induced by sciatic nerve ligation caused impairment in short term memory as shown by the reduction in percentage alternation (Figure 1) in the ligated control group compared to the vehicle control group ($p<0.05$). But two weeks co-administration of taurine and caffeine restored the memory deficit.

Though, treatment with caffeine alone did not produce a significant effect on the spatial memory impairment in this model as observed in diabetic-induced memory impairment (Duarte et al. (2012). This might be as a result of differences in dosage and duration of administration of the drugs. However, the result showed that caffeine enhanced the restoration effect of taurine on memory impairment induced by chronic pain to the sciatic nerve. Taurine on the other hand, significantly increased the percentage alternation which is an index of spatial memory, this could be due to its ability to partially protect the hippocampal damage (Rodriguez-Martinez et al. 2004) and hippocampus is known to play a major role in learning and memory. This is in line with the finding of Irfan et al (2013) who reported that taurine administration enhanced recognition and spatial memory functions.

Another index that can be observed is the locomotor activity from the total number of arm entries (Lister 1987). Induction of neuropathic pain through sciatic nerve ligation caused reduced locomotor activity as shown by the decrease in number of arm entries in ligated control group compared to the vehicle control group (Figure 2). Treatment with taurine and caffeine alone significantly increased the locomotor activity but the combination of the two substances did not produce any significant effect. Previous studies have shown that caffeine alone increases alertness (Smith and

Table 1: Effect of administration of taurine and caffeine on serum CRP level

Groups	serum CRP (mg/dl)
Vehicle control	0.76±0.05*
Ligated control	1.77±0.19
Taurine alone	0.01±0.10*
Caffeine alone	1.74±0.19
Taurine+ caffeine (Low dose)	1.14±0.10*
Taurine+ caffeine (High dose)	0.77±0.05*

Each value is the mean ± S.E.M. (n=6); * P < 0.05 compared with ligated control animals.

results possibly due to large differences in the dosages used. When the two substances were co-administered, there was no significant effect.

Furthermore, increase in serum CRP level suggests that induction of neuropathic pain led to increased inflammatory activity and this was significantly reduced by treatment with taurine alone. Studies have repeatedly demonstrated that the up regulation of pro inflammatory cytokines following nerve injury have a key role in the development of neuropathic pain (George and Somer 2005), and Lin et al. (2009) showed that CRP mimics the ability of Aβ25–35 to impair memory in Alzheimer's disease.

In conclusion, it has been demonstrated that taurine and caffeine could restore memory impairment associated with neuropathic pain in sciatic nerve ligated rats, but they did not have a significant effect on locomotor activity. However, further studies need to be carried out to elucidate the mechanism(s) by which taurine and caffeine interact to restore the memory impairment associated with neuropathic pain.

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Conflict of Interest
None declared.

REFERENCES

Abdulmajeed, W. and Owoyele, B.V. (2015) Anti-nociceptive effects of taurine and caffeine in sciatic nerve ligated Wistar rats: involvement of autonomic receptors. Journal of African Association for Physiological Sciences. 3 (1): 41-48.

Alloway, T. P., Gathercole, S. E., Kirkwood, H. and Elliott, J. (2009) The cognitive and behavioural characteristics of children with low working memory. Child Development. 80: 606-621.

Bennett, G. J. and Xie, Y. K. (1998) A peripheral mononeuropathy in rat that produces disorders of pain sensation like those seen in man. Pain. 33: 87-107.

Chen, W. Q., Jin, H., Nguyen, M., Carr, J., Lee, Y.J., Hsu, C. C., Faiman, M. D., Schloss, J. V. and Wu, J.Y. (2001) Role of taurine in regulation of intracellular calcium level and neuroprotective function in cultured neurons. Neuroscience Research. 66: 612-619.

Clubley, M., Bye, C. E., Hensen, T. A., Peck, A. W. and Riddinton, C. J. (1979) Effects of caffeine and cyclizine alone and in combination on human performance, subjective effects and EEG activity. British Journal of Clinical Pharmacology. 7: 157-163.

Cowan, N. (2008) What are the differences between long-term, short-term, and working memory? Progress in Brain Research. 169: 323-338.

Cunha, R. A. (2005) Neuroprotection by adenosine in the brain: from A1 receptor activation to A2A receptor blockade. Purinergic Signalling. 1(2): 111-134.

Cunha, R. A. and Agostinho, P. M. (2010) Chronic caffeine consumption prevents memory disturbance in different animal models of memory decline. Journal of Alzheimer's Disease. 20(S1): s95-s116.

Dick, B. D. and Rashiq, S. (2007) Disruption of attention and working memory traces in individuals with chronic pain. Anesthesia and Analgesia. 104: 1223-1229.

Duarte, J. M. N., Agostinho, P. M., Carvalho, R. A. and Cunha, R. A. (2012) Caffeine Consumption prevents diabetes-induced memory impairment and synaptotoxicity in the hippocampus of NONcZNO10/LTJ Mice. PLoS One. 7(4): e21899.

Eccleston, C. (1995) Chronic pain and distraction: an experimental investigation into the role of sustained and shifting attention in the processing of chronic persistent pain. Behaviour Research and Therapy. 33: 391-405.

Fredholm, B. B., Chen, J. F., Cunha, R. A., Svenningsson, P. and Vaugeois, J. M. (2005) Adenosine and brain function. International Review of Neurobiology. 63: 191-270.

Gatchel, R. J. (2004) Comorbidity of chronic pain and mental health disorders: The biopsychosocial perspective. American Psychologist. 59: 795-805.

George, A. and Sommer, C. (2005) Tumor necrosis factor receptor 1 and 2 proteins are differentially regulated during Wallerian degeneration of mouse sciatic nerve. Experimental Neurology. 192: 163-166.

Green, M. F. (2006) Cognitive impairment and functional outcome in schizophrenia and bipolar disorder. Journal of Clinical Psychiatry. 67: 3-9.

Hart, R. P., Martelli, M. F. and Zasler, N. D. (2000) Chronic pain and neuropsychological functioning. Neuropsychology Review.10: 131-149.

Irfan, S., Saima, K., Saiqa, T., Lubna, A., Shoaib, A. and Saida, H. (2013) A dose related study on the effects of taurine administration on recognition and spatial memory functions and depression like symptoms in rats. International Journal of Advanced Research. 1(8): 364-371.

Kokkinidis, L., Walsh, M. D., Lahue, R. and Anisman, H. (1976) Tolerance to d-amphetamine: Behavioral specificity. Life Science. 18: 913-991.

Kumar, N., Laferriere, A., Yu, J. S. C., Leavitt, A. and Coderre, T. J. (2010) Evidence that pregabalin reduces neuropathic pain by inhibiting spinal release of glutamate. Journal of Neurochemistry. 113(2): 552-561.

Lin, H. B., Yang, X. M., Li, T. J., Cheng, Y. F., Zhang, H. T. and Xu, J. P. (2009) Memory deficits and neurochemical changes induced by C-reactive protein in rats: implication in Alzheimer's disease. Psychopharmacology. 204:705-71.

Lister, R. G. (1987) The use of a plus-maze to measure anxiety in the mouse. Psychopharmacology. 92: 180-185.

Mankovskaya, I. N., Serebrovskaya, T. V., Swanson, R. J., Vavilova, G. L. and Kharlamova, O. N. (2000) Mechanisms of taurine antihypoxic and antioxidant action. High Altitude Medicine and Biology. 1: 105-110.

Moriarty, O., McGuire, B. E. and Finn, D. P. (2011). The effect of pain on cognitive function: a review of clinical and preclinical research. Progress in Neurobiology. 93: 385-404.

Mutso, A. A., Radzicki, D., Baliki, M. N., Huang, L., Banisadr, G., Centeno, M. V., Radulovic, J., Martina, M., Miller, R. J. and Apkarian, A. V. (2012) Abnormalities in hippocampal functioning with persistent pain. The Journal of Neuroscience. 32: 5747-5756.

Onaolapo, O. J., Onaolapo, A. Y., Mosaku, T. J., Akanji, O. O. and Abiodun, O. R. (2012) Elevated plus maze and y-maze behavioral effects of subchronic, oral low dose monosodium glutamate in Swiss albino mice. IOSR Journal of Pharmacy and Biological Sciences. 3(4): 21-27.

Rodríguez-Martínez, E., Rugerio-Vargas, C., Rodríguez, A. I., Borgonio-Pérez, G. and RivasArancibia, S. (2004) Antioxidant effect of taurine, vitamin C and Vitamin E on oxidative

damage in hippocampus caused by the administration of 3-nitropropionic acids in rats. International Journal of Neuroscience. 114 (9): 1133-1145.

Schurr, A., Tseng, M. T., West, C. A., and Rigor, B. M. (1987) Taurine improves the recovery of neuronal function following cerebral hypoxia: an *in vitro* study. Life Science. 40: 2059-2066.

Smith, H. J. and Rogers, P. J. (2000) Effect of low doses of caffeine on cognitive performance, mood and thirst in low and high caffeine users. Psychopharmacology. 152: 167-173.

Takahashi, R. N., Pamplona, F. A. and Prediger, R. D. (2008) Adenosine receptor antagonists for cognitive dysfunction: a review of animal studies. Frontiers in Bioscience. 13: 2614-2632.

Tang, X. W., Deupree, D. L., Sun, Y. and Wu, J. Y. (1996) Biphasic effect of taurine on excitatory amino acids. Advances in Experimental Medicine and Biology. 403: 499-505.