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Is inflammatory pulpal pain a risk factor for amnesia?

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Abstract

The aim of this study was to investigate the effect of inflammatory pulpal pain on spatial learning and memory in male Wistar rats. Fifty-six adult rats were divided into eight groups as follows (n=7 per group): control, sham-operated group that received normal saline, sham vehicle group that received vehicle of capsaicin, three capsaicin treated groups that received intradental injection of 10, 25 and 100 µg capsaicin, respectively, formalin treated group that received 10 µl formalin 2.5% and ibuprofen treated group that received ibuprofen 20 min before capsaicin (100 µg) injection. After preparing cavities via cutting 2 mm of the distal extremities of the mandibular incisors, the polyethylene crowns were placed on the teeth. Based on the study group, different algesics were administrated under the crowns. After recording the pain scores, spatial learning and memory was assessed using Morris water maze test. Capsaicin 25, 100 µg and formalin 2.5% applications induced significantly more painful stimulation compared with control groups ($p< 0.001$). Capsaicin 25, 100 µg and also formalin-treated groups significantly showed increased escape latency and traveled distance ($p<0.05$). Oral administration of ibuprofen, 20 min before capsaicin injection, caused significant decrease in pain scores, escape latency and traveled distance. Our data suggest that capsaicin- and formalin-induced pulpal pain can impair spatial learning and memory of male rats in Morris water maze task.

Keywords: odontalgia, capsaicin, formalin, spatial learning and memory, Morris water maze

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Introduction

Orofacial pain is one of the most prevalent types of pain suffered by a large portion of the world's population, and odontalgia is the most commonly experienced type (Moure-Leite *et al*., 2011). Odontogenic pain is caused by the release of various inflammatory and pain mediators leading to stimulation of receptors located on terminal endings of nociceptive afferent nerve fibers. In addition, inflammation is recognized as a core process in pulpal pathosis (Bergenholtz, 1981) and may also have a major role in the development of Alzheimer's disease (AD) (Flirski and Sobow, 2005). It has been found that some of the mediators and products of inflammatory reactions, such as cytokines, prostaglandins, complement proteins, adhesion molecules and free radicals, are toxic to neurons in experimental trials (Prasad *et al*., 2002). The products of inflammatory reactions may contribute to neuronal degeneration due to extracellular signal representation (Prasad *et al*., 2002). The inflammatory reaction hypothesis has also been supported by clinical studies in which administration of NSAIDs (non-steroidal anti-inflammatory drugs) effectively reduced the rate of cognitive deficits in moderate to advanced AD patients (Lucca *et al*., 1994, Rich *et al*., 1995).

A large number of studies have demonstrated the relationship between pain and changes in brain anatomy such as cortical thickness and gray matter density (Seminowicz *et al*., 2009). Mechanical hyperalgesia causes a decrease in cortical volume in somatosensory, anterior cingulate, areas 32 and 24, and insular cortices (Seminowicz *et al*., 2009).

Regional gray matter density analyses have shown that fibromyalgia is associated with a significant loss of gray matter in regions associated with stress or pain modulation like as cingulate, insular and medial frontal cortices as well as the thalamus and parahippocampal gyri (Kuchinad *et al*., 2007, Schmidt-Wilcke *et al*., 2007). Furthermore, abnormalities in several gray matter regions of the brain along the central pain network have been shown in patients with migraines (Valfrè *et al*., 2008).

Gray matter atrophy is also known to be involved in cognition (Ceccarelli *et al*., 2009) which is supported by studies conducted by back pain researchers showing lower gray matter volume in the frontal cortex and poorer performance on a task of frontal lobe functioning (Apkarian *et al*., 2004a , Apkarian., *et al* 2004b). Chronic pain stress induced by complete freund's adjuvant also lead to a significant impairment of spatial learning and memory, increasing escape latency, decreasing average percentages of the swimming time and distances in the platform quadrant in Morris water maze task (Li *et al*., 2005). These observed effects might occur through the down-regulation of Bcl-2 and BDNF (Brain-derived neurotrophic factor) mRNA expression in the hippocampus (Li *et al*., 2005).

Furthermore, gene expression analyses have revealed that acute and persistent peripheral nociception evoked a similar down-regulation of both NK-1 receptor and BDNF gene expression in the hippocampus (Duric and McCarson, 2007). It has been shown that BDNF plays a key role in hippocampal function and, subsequently, hippocampal-dependent learning and memory (Hariri *et al*., 2003).

Considering these findings, there is a possibility that pulpitis produces neuronal dysfunction in the central nervous system, leading to deterioration of cognitive function. Since cognitive function commonly includes memory, learning, and attention processes, the present study investigates the effect of inflammatory pulpal pain on spatial learning and memory in rats using the Morris water maze task.

Material and methods

Ethics Statement

All experimental procedures performed on rats were approved by the Animal Research Ethics Committee of Kerman University of Medical Sciences, Kerman, Iran (Permit Number: EC/KNRC/89-39). All efforts were made to minimize suffering.

Animals

Fifty-six adult male Wistar rats weighing 250-300 grams, purchased from the Neuroscience Research Center (Kerman University of Medical Sciences, Iran), were used in this study. The rats were housed one per cage in a room with a temperature of 23±2ºC where they were subjected to a regimen of 12 day/night cycles and given unlimited access to standard rat chow and water before and during the study.

Dental procedure

Inflammatory pulpal pain induction was constructed as our modified model, representing a modification to the Chidiac *et al*. 2002) described in a previous article (Raoof *et al*., 2011). In brief, 2 mm of the distal of the rats' mandibular incisors were cut off and special polyethylene crowns were fixed on the teeth using a flow composite resin (Tetric Flow, Ivoclar Vivadent). A small space remained between the tooth structure and the internal surface of the crown.

Study drugs

Formalin 2.5%: Formalin solution was freshly prepared from commercially available stock formalin (Sigma-Aldrich) diluted in isotonic saline to 2.5%. Stock formalin is an aqueous solution of 37% formaldehyde.

Capsicin (Sigma-Aldrich): Capsaicin was dissolved in Tween 80-ethanol solution (Merck, Germany) (10% ethanol, 10% Tween 80, 80% distilled water, w/w) at the graded concentrations of 10, 25, and 100 μg and administrated intradentally (i.d.).

Ibuprofen (Kimidaru, Iran): Ibuprofen powder with vehicle (2% Tween 80/distilled water) in a dose of 120 mg/kg was administered by oral gavage.

Study groups

Fifty-six animals were randomly divided into eight groups (n=7) as follows:

1: Control group (CO) included intact animals.

2: Sham operated group (SO) received i.d. injection of normal saline.

3: Sham vehicle group (SV) received i.d. injection of capsaicin vehicle including Tween 80 and ethanol.

4-6: Capsaicin treated groups (C10, C25, and C100) received i.d. injection of 10, 25, and 100 μg capsaicin, respectively.

7: Formalin treated group (F) received i.d. injection of formalin 2.5%.

8: Ibuprofen treated group (I) received ibuprofen 20 min before i.d. capsaicin 100 μg.

After two days of recovery, unanesthetized rats were restrained in plastic holding tubes and the mouth was held open with the use of a small retractor. According to the study group to which the rat was assigned, 10 μL of the specified drug was injected in the hallow chamber through a 27-gauge needle as quickly as possible and cyanoacrylate adhesive was then used to close the crown perforation immediately. Loupes containing a 4x-magnification were utilized.

Nociceptive behavior

Test sessions were carried out during the light phase, between 10:00 and 17:00 h, in a quiet room maintained at 23–24°C. Before the injection, each animal was placed in the test box for a 30-min habituation period to minimize additional stress. The rats did not have access to food or water during the test.

Immediately following the injection, each rat was placed back in the transparent Plexiglass box (25 × 35 × 35) with a transparent floor positioned over a mirror at an angle of 45 degrees to allow for observation of nociceptive behavior. The rats' behavior was observed for 21 minutes, divided into 7 blocks of 3 minutes. A pain score was determined for each block by measuring the number of seconds that the animal presented each of the following responses which represents the same scoring criteria as Chidiac *et al.* study (Chidiac *et al*., 2002): 0 – calm, normal behavior such as grooming; 1 – abnormal head movements such as mild head shaking or continuous placement of the jaw on the floor or the wall of the cage; 2 – abnormal continuous shaking of the lower jaw; 3 – excessive rubbing of the mouth with foreleg movements, such as head grooming, but concentrated consistently and mainly on the lower jaw. A video camera was used to record the behavioral response.

Morris water maze test

Learning and memory was assessed in a water tank as described in a previous study (Morris *et al*., 1982). The water maze was a black circular tank measuring 136 cm in diameter and 60 cm in height. The tank was filled with water at a temperature of 20±1 ºC and to a depth of 25 cm. Cues located outside of the maze consisted of geometric shapes on the walls, posters, shelves, etc.; there were no cues within the maze. The maze was divided into four quadrants [Northeast (NE), Northwest (NW), Southeast (SE), and Southwest (SW)] and four starting positions [North (N), South (S), East (E), West (W)] that were equally spaced around the perimeter of the tank. A hidden circular platform, with a diameter of 10 cm, was located in the center of the SW quadrant, submerged 1.5 cm below the surface of the water. A video camera was mounted directly above the water maze to record the rats' swim path. A tracking system was used to measure the escape latency, each rat's distance traveled, the percent of distance, and the time in each quadrant.

The rats received a block of four trials during five consecutive daily sessions. During the first 4 days, the platform, situated in the center of the southwest quadrant, was submerged 1.5 cm below the surface of water, to make it invisible, for testing of spatial learning. The position of the platform remained stable over 4 days and acquisition of this task was assessed. On the 5th day, every rat was subjected to a probe trial for 60 seconds in the absence of the platform in the pool from the center of the southwest quadrant. For visible test the platform was elevated above the water level, covered with a piece of aluminum foil, and placed in the center of southeast quadrant for assessment of sensory motor towards a visible platform. The trial was terminated either the rat had climbed onto the escape platform or when 90 seconds had elapsed. The rat was allowed to stay on the platform for 20 seconds, after which the next trial was started. If the rat did not find the platform within 90 seconds, it was put on the platform by the experimenter and allowed to stay there for 20 seconds. After the completion of the 4th trial, the rats were gently dried with a towel, kept warm for an hour, and returned to their home cage. All tests were conducted between hours 09:00 and 13:00.

Statistical analysis

All statistical analyses were carried out by an observer blinded to the experimental groups. Differences between groups regarding pain scores were determined by one-way analysis of variance. Data obtained over the first four training days from hidden platform tests were analyzed by two-way analysis of variance (ANOVA) followed by Tukey's test for multiple comparisons. Data from the 5th training day was analyzed by one-way ANOVA. Post-hoc analysis was performed using the Tukey's Honestly Significant Difference (HSD) test and the significance level was set at $p < 0.05$ for all analyses performed. Data are presented as mean ± standard error of mean (S.E.M.).

Results

Effect of intradental administration of

capsaicin and formalin

Fig1 shows that intradental administration of both chemical noxious stimuli, capsaicin (25 and 100 µg /rat) and formalin, significantly affected nociceptive behaviors ($p<0.0001$). Additionally, the data showed that the greatest effect was obtained from capsaicin 100 µg. In ibuprofen-pretreated rats, capsaicin induced algesic effect was prevented.

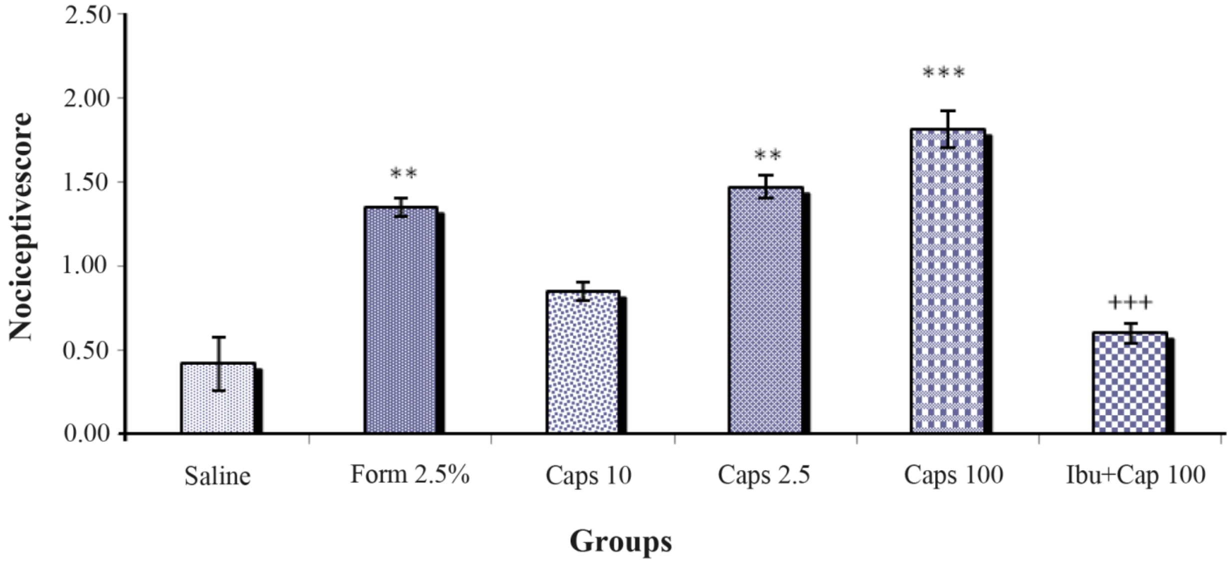

Figure 1. Nociceptive effect of intradental injection of chemical noxious stimuli, capsaicin (10, 25 and 100 µg/rat) and formalin 2.5%. Administration of ibuprofen (120 mg/kg by oral gavage) 20 min before 100 µg capsaicin prevented the nociceptive effect of capsaicin. Ibu, ibuprofen; Caps, capsaicin; Form, formalin. Values are expressed as mean ±SEM. (n=7) . ** $p<0.01$ and *** $p<0.001$ vs saline treated control group. +++ $p<0.001$ vs Capsaicin 100.

Effect of inflammatory tooth pain on spatial learning and memory

Hidden platform Trials (days 1–4)

Figure 2 shows the results obtained from the intradental injection of chemical noxious stimuli, capsaicin and formalin. Capsaicin (100 µg/rat) and formalin 2.5% significantly increased escape latency ($p<0.01$ and $p<0.05$, respectively) and traveled distance ($p<0.05$ and $p<0.01$, respecitvely) (Fig.3).There was no significant change in mean swim speeds during the 4 days of training (p: 0.684) (Fig.4).

Platform Removed Trials (day 5)

There was no significant difference between the capsaicin- and formalin-treated rats and the control group on the 5th day regarding the percentage of distance traveled in the target quadrant with the removed platform (p:0.701) (Fig.5) as well as the percentage of time spent in the target quadrant (p:0.426) (Fig.6).

Visible Test

There was no significant difference between groups on the 5th day regarding the escape latency with the elevated platform (p:0.561) (Fig.7).

Discussion

Following treatment with capsaicin at doses of 25 and 100 µg, animals spent more time and distance to find the hidden platform compared with animals in the other groups. This data indicates that capsaicin-induced pulpal pain impairs spatial learning of male rats in MWM. Oral administration of ibuprofen 20 min before capsaicin injection caused a significant decrease in escape latency and distance traveled. However, there was no significant effect of the 10 µg dose of capsaicin on any of the learning and memory indices in rats. Intradental application of 2.5% formalin was shown to increase distance traveled and time's period in the target quadrant while having a decreasing effect on escape latency.The probe test showed that there was no significant difference in traveled distance and time's period in target quaderant. between the two groups. A positive correlation was observed between the pain scores and the time and distance to reach the platform in the 25 and 100 µg capsaicin and 2.5% formalin groups.

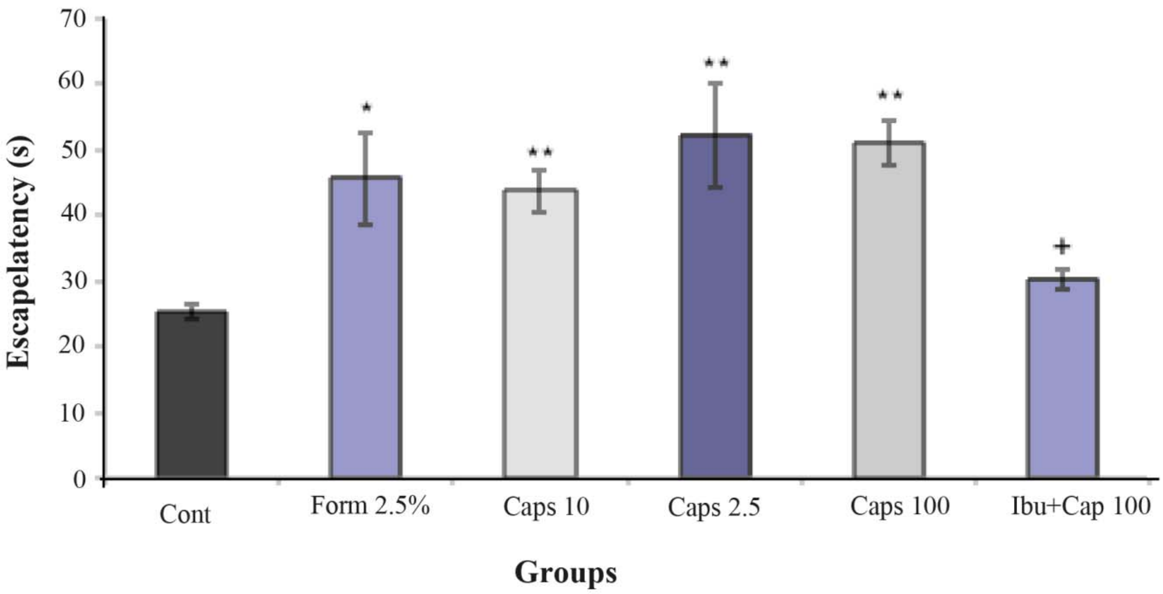

Figure 2. The effects of intradental injection of chemical noxious stimuli, capsaicin (10, 25 and 100 µg/rat) and formalin 2.5% during water maze training; Mean escape latency during 4 days of training in water maze with the hidden platform. Ibu, ibuprofen; Caps, capsaicin; Form, formalin.Values are expressed as mean ±SEM .(n=7). ****p*<0.05 and ** *p*<0.01 vs Control; + *p*<0.05 vs Capsaicin 100.

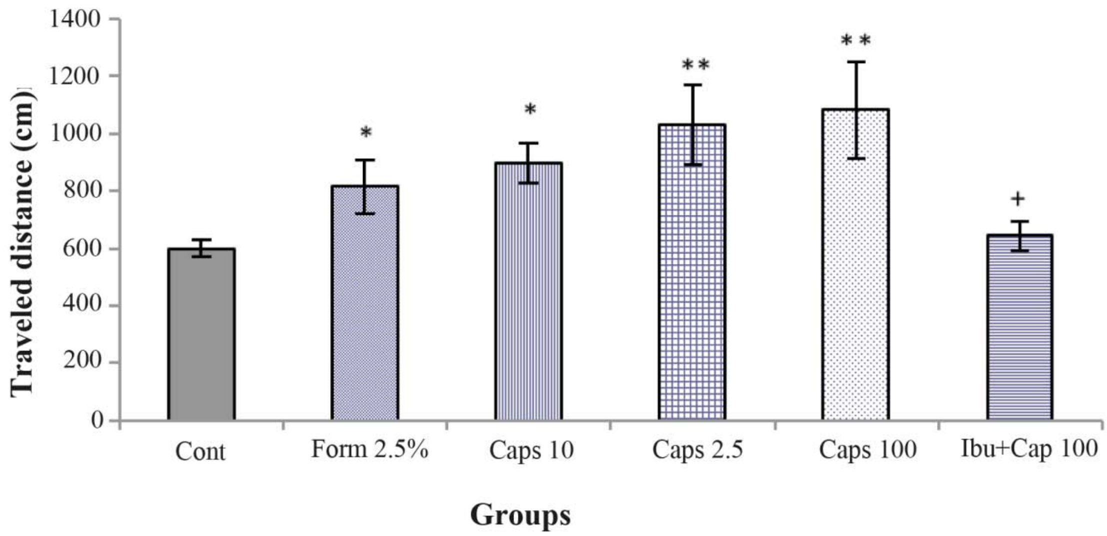

Figure 3. The effects of intradental injection of chemical noxious stimuli capsaicin (10, 25 and 100 µg/rat) and formalin 2.5% during water maze training; Mean traveled distance during 4 days of training in water maze with the hidden platform. Ibu, ibuprofen; Caps, capsaicin; Form, formalin. Values are expressed as mean ± SEM. (n=7). *p<0.05, **p<0.01 vs Control; +p<0.05 vs Capsaicin100.

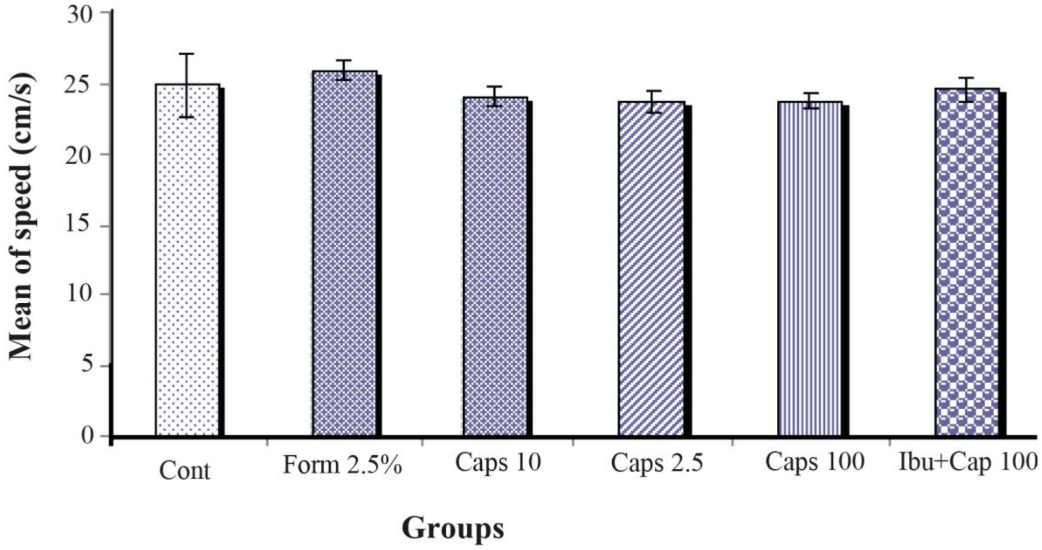

Figure 4. The effects of intradental injection of chemical noxious stimuli, capsaicin (10, 25 and 100 µg/rat) and formalin 2.5% on the mean of swim speed in water maze tank. Ibu, ibuprofen; Caps, capsaicin; Form, formalin. Values are expressed as mean ± SEM. (n=7). Ibu,ibuprofen; cap=capsaicin.

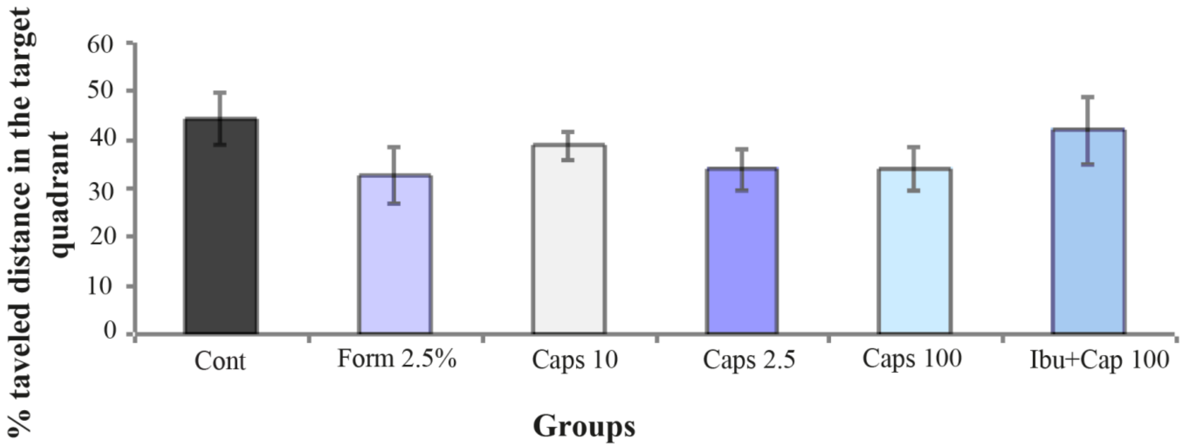

Figure 5. The effects of intradental injection of chemical noxious stimuli, capsaicin (10, 25 and 100 µg/rat) formalin 2.5% on the percentage of traveled distance in the target quadrant during the day 5 of training in a water maze with the removed platform. Ibu, ibuprofen; Caps, capsaicin; Form, formalin.Values are expressed as mean ± SEM. (n=7).

Through stimulating vanilloid receptor 1 (VR1), capsaicin causes tonic activation of polymodal C fibers and some lightly myelinated A-delta fibers in the pulp (Chidiac *et al*., 2002). The nociceptive C fiber participates in the wound by releasing calcitonin gene-related peptide (CGRP), nitric oxide (NO), Substance P (SP) and its closely related neuropeptide neurokinin A (NKA), all of which contribute to neurogenic inflammation and hyperalgesia (Henry and Hargreaves, 2007).

Other studies found that formalin activates primary afferent sensory neurons through a specific and direct action on TRPA1 (Transient receptor potential cation channel) (McNamara *et al*., 2007). Formalin also evokes short-lived activity in large myelinated fibers as well as continual stimulation of C and Aδ fibers (Heapy *et al*., 1987). The similar action of formalin and capsaicin in rat incisor pulp may be due to unmyelinated fiber activation.

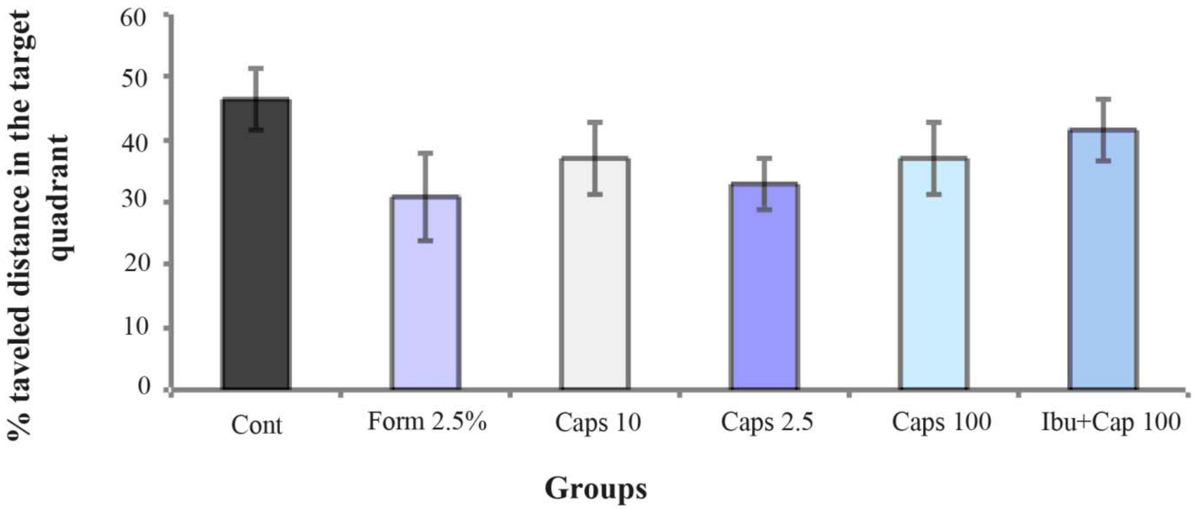

Figure 6. The effects of intradental injection of chemical noxious stimuli, capsaicin (10, 25 and 100 µg/rat) and formalin 2.5% on the percentage of time spent in the target quadrant during the day 5 of training in water maze with the removed platform. Ibu, ibuprofen; Caps, capsaicin; Form, formalin.Values are expressed as mean ± SEM. (n=7).

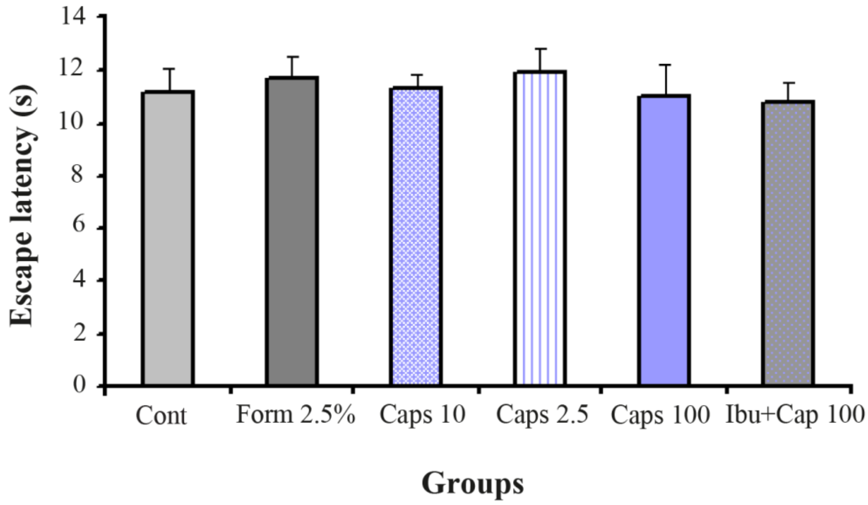

Figure 7. The effects of intradental injection of chemical noxious stimuli, capsaicin (10, 25 and 100 µg/rat) and formalin 2.5% during water maze training; Mean escape latency during visible test of training in water maze with the visible platform. Ibu, ibuprofen; Caps, capsaicin; Form, formalin.Values are expressed as mean ±SEM .(n=7).

Given these findings, it is not surprising that inflammatory pulpal pain, the most prevalent orofacial pain type, impairs spatial learning in rats. To our knowledge, there is no study exploring memory deficits resulting from odontalgia in humans or animals. However, in a study by Kuhajda and associates, headache adversely affected memory (Kuhajda *et al*., 2002). Similarly, Boyette-Davis *et al*., 2008 concluded that formalin-induced pain behaviors negatively impacts attention in a 5-choice serial reaction time task (Boyette-Davis *et al*., 2008). Chronic pain stress induced by complete freund's adjuvant also resulted in a significant impairment of spatial learning and memory function in neonatal rats (Li *et al*., 2005). Li *et al*., 2005 reported that these effects might occur through the down-regulation of Bcl-2 and BDNF mRNA expression in the hippocampus (Li *et al*., 2005). Pain signals are transmitted in a more non-specific and scattered way than other modalities. There is also remarkable modulation where pain messages are relayed (Julius and Basbaum, 2001). The interaction between pain and other modalities may act via release of neuronal mediators (Woolf and Salter, 2000). This interaction from the dorsal horn of the spinal cord to the cortical surface may be associated with higher-level cognitive processes. Different sensory modalities are involved in formation, development and stabilization of cognitive messages (Woolf and Salter, 2000).

The hippocampus, as the most important part of the brain related to learning and memory, may indirectly receive nociceptive inputs from the periphery, primarily via trigeminothalamic and parabrachial ascending pathways (Duric and McCarson, 2006). Although there are differences between the synaptic plasticity contributing to memory and pain, some of the similarities are striking (Ji *et al*., 2003).

Central sensitization underlies a mechanism of transition from acute to chronic pain. Pain-induced synaptic plasticity has also been shown to occur in higher brain regions with known roles in cognitive function (Zhao *et al*., 2009; Zhuo, 2007).

It is unclear whether supraspinal pain-induced plasticity is an extension of central sensitization that occurs in the spinal cord and subnucleus caudalis, though the critical involvement of NMDA, AMPA (Zhao *et al*., 2009) and metabotropic glutamate receptors (Ji *et al*., 2010) indicate similar mechanisms.

Co-occurrence of pain-induced synaptic plasticity and learning/memory related LTP raise the possibility of cognitive impairment at the molecular level (Gravius *et al*., 2010). Electrophysiological studies demonstrate that blockade of metabotropic glutamate receptors (mGluR1) significantly reduce evoked firing in spinal wide dynamic range neurons and disruption of motor and cognitive performances in the Y-maze and the Water Maze tests (El-Kouhen *et al*., 2006).

L5 spinal nerve transection has been shown to induce mechanical allodynia and decrease the function of learning and memory as well as the expression of brain-derived neurotrophic factor (BDNF) in rats. Hu *et al*.,2010 concluded that neuropathic pain may impair cognitive function via downregulation of BDNF expression of the hippocampus, while amitriptyline can reverse cognitive impairment via upregulation of brain-derived neurotrophic factor of the hippocampus (Hu *et al*., 2010).

Hoot *et al*.,2010 showed alterations in cannabinoid receptor function within the rostral anterior cingulate cortex in response to a model of neuropathic pain (Hoot *et al*., 2010). While cannabinoid receptor agonists impair memory formation, antagonists ameliorate impaired recognition memory. These results are consistent with those obtained from electrophysiological recordings which reveal reduction in neural plasticity after cannabinoid treatment and increased plasticity following antagonist administration. The exogenous selective CB1 agonists may, therefore, facilitate the extinction of hippocampus-dependent learning and memory by 'increasing the noise' rather than 'decreasing the signal' at potentiated inputs (Riedel and Davies, 2005).

Cannabinoid CB1 receptors, are up-regulated following a chronic constriction injury of the sciatic nerve, possibly to inhibit pain (petrosino *et al*., 2007). Cognitive deficits have been attributed to cannabinoids via interaction with neurochemical processes in the prefrontal cortex and hippocampus (Egerton *et al*., 2006).

Altered stress-induced hypothalamus-pituitary-adrenal (HPA) axis responses have been associated with pain. This is a stimulus to hippocampal plasticity and increased hippocampal volume (Barha *et al*., 2010). Activation of HPA axis induces an increased glucocorticoid release (Webster, 2004).

Reports have demonstrated possible crosstalk between glucocorticoid and receptor tyrosine kinase for BDNF (TrkB). Decreased BDNF function results in reduced neurogenesis in pyramidal cells and dentate gyrus within the hippocampus and impaired hippocampal-dependent spatial cognition (Kunugi *et al*., 2010).

Studies have shown that peripheral injury activates glial components of the peripheral and central cellular circuitry. Glial proinflammatory cytokines such as interleukin-1 beta may facilitate pain via interaction with glutamate receptors (Abbott *et al*., 2006; Watkins *et al*., 2001). Glial D-serine, as a gliotransmitter, controls the activity of NMDARs and, as such, has important impacts on cognitive processes such as learning, memory, and spatial orientation (Fossat *et al*., 2012). As the results showed, oral administration of ibuprofen 20 min before capsaicin injection caused a significant decrease in escape latency and distance traveled. The primary mechanism of ibuprofen action is cyclooxygenase (COX) inhibition. In addition, capsaicin increases the expression of cyclooxygenase-2 (COX-2) and the release of inflammatory mediators such as interleukin-8 and prostaglandin E2 by activation of VR1 receptor (Shetty *et al*.,2013). It has been documented that the pain and inflammation reducing effects of NSAIDs such as ibuprofen are mediated through the inhibition of COX-2. Surprisingly, ibuprofen can significantly decrease gingival crevicular fluid prostaglandin E2 levels during orthodontic tooth movement in human subjects (Shetty *et al*., 2013).

A possible link between increased pro-inflammatory molecules and memory dysfunction has been proposed. During the induced pulpal inflammation in rat incisors, a 9.3-fold increase in PGE2 levels was observed (Okiji *et al*., 1989). PGE2 signaling, likely through the EP3R, can reduce BDNF mRNA induction, a molecule necessary for normal hippocampal-dependent memory. Moreover, when applied to human pulp cells in vitro, LPS from Porphyromonas endodontalis led to the release of interleukin-1β (IL-1β) in a dose-dependent manner. Several studies have found a causative relationship between memory deficits and elevated IL-1β levels (Barrientos *et al*., 2002; Hein *et al*., 2007). IL-1β-induced attenuation of LTP in CA1 (Bellinger *et al*., 1993), CA3 (Katsuki *et al*., 1990) and dentate gyrus (Cunningham *et al*., 1996) has been expressed. Peripheral nerve injury increased TNF-α in the CSF, plasma, and hippocampus. The increase in TNF-α was closely correlated with LTP inhibition and impairment of synaptic plasticity and memory (Ren *et al*., 2011). Although the precise mechanisms of pain-related cognitive impairment have not yet been elucidated, a number of possible mechanisms have been proposed (Kozlovsky *et al*., 2007; Khairova *et al*., 2009). Damage to the hippocampal structures has also been associated with learning and memory impairments (Squire *et al*., 1992).

In conclusion, we demonstrated that formalin and capsaicin-induced pulpal pain impairs spatial learning and memory ability of male rats in the Morris water maze. A number of possible mechanisms for this phenomenon may include synaptic plasticity, downregulation of BDNF, elevated pro-inflammatory cytokines, and alterations in cannabinoid receptor function as well as pain induced stress signaling. In a different study, we are exploring the possible underlying mechanisms. However, further studies are necessary to elucidate the exact molecular mechanisms of this disorder.

Acknowledgements

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The authors deny any conflicts of interest.

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چکیده

این مطالعه به منظور بررسی تاثیرات درد التهابی پالپ دندان بر یادگیری وحافظه فضایی موشهای صحرایی نر ویستار انجام گردید. ۵۶ سر موش صحرایی نر بالغ بصورت تصادفی به هشت گروه هفت تایی شامل موارد زیر تقسیم شدند. گروه های کنترل، شاهد جراحی که نرمال سالین دریافت کرد، دریافت کننده حلال کاپسایسین که حلال کاپسایسین دریافت کرد، سه گروه دریافت کننده دوزهای متفاوت کاپسایسین (µg ۱۰۰ و ۲۵ و ۱۰) و گروه دریافت کننده ۱۰ میکرولیتر فرمالین ۵/۲ درصد، و گروه دریافت کننده ایبوپروفن بیست دقیقه قبل از تزریق کپسایسین µg ۱۰۰. پس از تراش ۲mm از نواحی دیستال دندانهای انسیزور فک پایین روکش پلی اتیلنی روی دندانها قرار گرفت. بسته به گروه مورد آزمایش مواد مختلف در حفره تعبیه شده زیر روکش تزریق شد. پس از ثبت درجه درد، یادگیری و حافظه فضایی در ماز آبی موریس مورد ارزیابی قرار گرفت. کپسایسین ۲۵ و ۱۰۰ میکروگرم و فرمالین ۲/۵ درصد نسبت به گروه کنترل باعث افزایش معنی دار درد شدند (p<۰/۰۰۱). همچنین کپسایسین ۲۵ و ۱۰۰ میکروگرم و فرمالین ۲/۵ درصد مسافت طی شده و زمان سپری شده تا رسیدن به سکوی پنهان را افزایش داد. تجویز دهانی ایبوپروفن ۲۰ دقیقه قبل از کاپسایسین نمره درد، تاخیر در پیدایش سکو و مسافت پیموده شده را به طور معنی داری کاهش داد. درد القا شده توسط کپسایسین و فرمالین منجر به اختلال در یادگیری و حافظه فضایی موشهای صحرایی نر در ماز آبی موریس شد.

واژگان کلیدی: درد التهابی دندان، کپسایسین، فرمالین، یادگیری و حافظه فضایی، ماز آبی موریس