KJP The Korean Journal of Pain
| Original Article |
Korean J Pain 2014 July; Vol. 27, No. 3: 246-252
pISSN 2005-9159 eISSN 2093-0569
http://dx.doi.org/10.3344/kjp.2014.27.3.246

The Attenuation of Pain Behavior and Serum COX-2 Concentration by Curcumin in a Rat Model of Neuropathic Pain

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Background:

Neuropathic pain is generally defined as a chronic pain state resulting from peripheral and/or central nerve injury. There is a lack of effective treatment for neuropathic pain, which may possibly be related to poor understanding of pathological mechanisms at the molecular level. Curcumin, a therapeutic herbal extract, has shown to be effectively capable of reducing chronic pain induced by peripheral administration of inflammatory agents such as formalin. In this study, we aimed to show the effect of curcumin on pain behavior and serum COX-2 level in a Chronic Constriction Injury (CCI) model of neuropathic pain.

Methods:

Wistar male rats (150-200 g, n = 8) were divided into three groups: CCI vehicle-treated, sham-operated, and CCI drug-treated group. Curcumin (12.5, 25, 50 mg/kg, IP) was injected 24 h before surgery and continued daily for 7 days post-surgery. Behavioral tests were performed once before and following the days 1, 3, 5, 7 after surgery. The serum COX-2 level was measured on day 7 after the surgery.

Results:

Curcumin (50 mg/kg) decreased mechanical and cold allodynia ($P < 0.001$) and produced a decline in serum COX-2 level ($P < 0.001$).

Conclusions:

A considerable decline in pain behavior and serum COX-2 levels was seen in rat following administration of curcumin in CCI model of neuropathic pain. High concentration of Curcumin was able to reduce the chronic neuropathic pain induced by CCI model and the serum level of COX-2. **(Korean J Pain 2014; 27: 246-252)**

Key Words:

allodynia, COX-2, curcumin, neuropathic pain.

Received March 18, 2014. Revised June 7, 2014. Accepted June 8, 2014.
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INTRODUCTION

Neuropathic pain can be produced as a result of damage to the peripheral or central nervous system. Allodynia (pain evoked by normally non-noxious stimuli), and hyperalgesia (an increased response to a noxious stimuli) are routinely observed in human neuropathic pain conditions as well as in relevant animal models [1], and are often resistant to the analgesics and interventional therapeutic methods [2-4]. Many drugs have been tried to reduce neuropathic pain, but since the underlying mechanisms are multiple and complex, therefore treatment and management of this distressing condition requires the use of more than one type of medication [5]. In the recent years, the development of highly targeted biological and synthetic therapies is in progress, although some produce serious side effects. On the other hand, for thousands of years humankind has used certain plants as therapeutic and pain relieving agents. Even in recent years, a renewed public interest in complementary therapies is rose, which include natural treatments with minimal toxicity and diets related to health and disease [6].

Curcumin is a yellow phenolic pigment, and a constituent of the spice turmeric, which is one of the principal ingredients in curry powder. Several clinical trials have found curcumin to have a notable anti-inflammatory and analgesic properties [7,8]. Likewise, the efficacy of curcumin in attenuation of diabetic neuropathic pain and formalin-induced nociceptive behavior has been demonstrated in previous studies [7,9,10]. Safety evaluation studies indicate that curcumin can be used at high doses without any toxic effects [10,11].

Cyclooxygenase is an enzyme that catalyzes the conversion of arachidonic acid to prostaglandins (PGs), which are the key mediators of exaggerated pain sensation [12-14]. Therefore, PGs have possibly an important role in induction of neuropathic pain. Non steroidal anti-inflammatory drugs (NSAIDs) are strong cyclooxygenase inhibitors, which are used in the management of chronic, inflammatory and postoperative pains [15]. However, they have serious side effects (gastro-intestinal complications, and at intense levels, hepatotoxicity and nephrotoxicity) [16-18], which make them not very safe choices for long time managements of the chronic pain.

Recent studies have shown that curcumin acts mainly by inhibiting the cyclooxygenase 2 pathway. Curcumin has also antirheumatic and antiarthritic effects, which are most likely through down regulation of cyclooxygenase 2, tumor necrosis factor (TNF), and other inflammatory cytokines [19,20].

Based on data supporting the analgesic and anti-inflammatory effects of curcumin in different animal models of inflammatory pain and regarding to its inhibitory effect on cyclooxygenase enzyme, the aim of the present study was to evaluate the effects of curcumin on pain behavior and serum cyclooxygenase 2 level in rat Chronic Constriction Injury (CCI) model of neuropathic pain.

MATERIALS AND METHODS

1. Animals

Wistar male rats, (weight 150-200 g, n = 8) were housed one per cage and placed under 12 hour light/dark cycle in a room with controlled temperature (22 ± 1°C). Animals had free access to food and water. All experiments followed the International Association for the Study of Pain (IASP) guidelines on ethical standards for investigation of experimental pain in animals [21].

2. Surgery

The CCI model of neuropathic pain [22] was used to induce sciatic nerve injury. The surgical procedure was performed under ketamine (60 mg/kg) and xylazine (10 mg/kg) anesthesia. The left sciatic nerve was exposed and 4 loose chromic gut ligatures were placed around the nerve proximal to the trifurcation. The distance between two adjacent ligatures was 1 mm. The wound was irrigated with normal saline (0.9%) and closed in two layers with 4-0 silk sutures (fascial plane) and surgical skin staples. In sham-operated group, rats underwent the same surgical procedure except for the ligation.

3. Drug preparation

Curcumin (Sigma, U.S.A) was suspended in ethyloleate as vehicle in the form of suspension. ketamine hydrochloride (Sigma, U.S.A) and xylazine hydrochloride (Sigma, U.S.A) were used for anesthesia. All drugs were injected by the intra-peritoneal (IP) route.

4. Experimental design

Animals were divided randomly into three experimental groups: 1- CCI vehicle-treated, 2- Sham-operated, 3-

CCI curcumin-treated. Curcumin 12.5, 25 and 50 mg/kg were injected intra-peritonealy [10], 24 hours before ligation continued daily to day 7 post-ligation. All behavioral tests were performed and recorded on days 0 (control day) before ligation and 1, 3, 5, 7 after ligation. The experiment was starting with mechanical test and terminating with cold allodynia. The time between two tests was 30 minutes. On the day 7 after the surgery and termination of the experiments, rats were euthanized by CO_2 asphyxiation, rapidly guillotined and the blood was collected for serum evaluation of cyclooxygenase 2.

5. Behavioral tests

Animals were allowed one week for housing habituation before starting the experiments. Behavioral studies were performed in a quiet room between the hours 9:00 and 11:00 AM. Efforts were made to limit animal distress and use of the minimum number of animals necessary to achieve statistical significance. Animals were acclimated to the testing chambers 30 min before testing. The sensory area of the sciatic nerve (mid-plantar hind paw) was tested for sensitivity to innocuous stimuli.

1) Mechanical allodynia: Mechanical sensitivity to non-noxious stimuli (allodynia) was measured by von Frey filaments. A set of calibrated nylon monofilaments (Stoelting, Wood Dale, Illinois, U.S.A) were used to assess the sensitivity of the skin to tactile stimulation. Rat was placed under a transparent plexiglass cage (Daj, Tehran, Iran), elevated by a metal screen surface with 1 cm mesh openings. Von Frey filaments were applied with increasing strengths (2-60 g) consecutively to the plantar surface of the left hind paw of the animal. The minimum paw withdrawal threshold (PWT) was defined as the minimum gram strength producing two sequential responses at 3 min intervals (withdrawal from pressure) [23].

2) Cold allodynia: We used acetone test as a model of "cold allodynia". In this method, rats were placed under a transparent plexiglass cage as described previously and the heel of his paw was touched with an acetone bubble (formed at the end of a piece of small polyethylene tubing, connected to a syringe). This test was applied 5 times with one minute intervals. The response was calculated as the percent of paw withdrawal frequency (%PWF) using the following equation: (Number of paw withdrawals/5 trials) ×100 [23].

6. Cyclooxygenase 2 protein analysis by ELISA

Serum cyclooxygenase 2 level was measured by solid phase sandwich ELISA kit specified for cyclooxygenase 2 protein (Cusabio, Biotech, wuhan, Hubei, China) with a lower detection limit of 0.4 ng/ml. Blood sample of different groups was centrifuged at 2500 rpm/20 min and the serum was collected and frozen at −70oC. The analysis of cyclooxygenase 2 protein expression was done according to the manufacturer's instruction, using an antibody specific for rat cyclooxygenase 2 coated onto the wells of the microtiter strips provided, a biotinylated antibody specific for rat cyclooxygenase 2, and Streptavidin-Peroxidase (enzyme) to bind to the biotinylated antibody to complete the fourth member sandwich. The plates are then read by a microplate reader at 450 nm. The cyclooxygenase 2 protein concentration was obtained from a standard curve (Fig. 1).

7. Statistical analysis

Parametric data was analyzed for significance, using Analysis of Variance (ANOVA) followed by a post-hoc Tukey's test. Non parametric data was analyzed using two related samples by Wilcoxon test. In all cases $P < 0.05$ was considered significant.

RESULTS

1. Response to mechanical allodynia

In response to von Frey test, all CCI vehicle-treated animals showed pain behavior on the third day after liga-

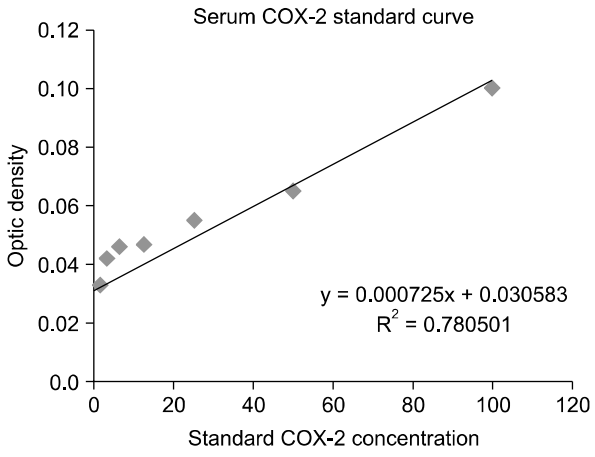

Fig. 1. COX-2 Standard Calibration Curve for serum evaluation of COX-2 in different groups of animals.

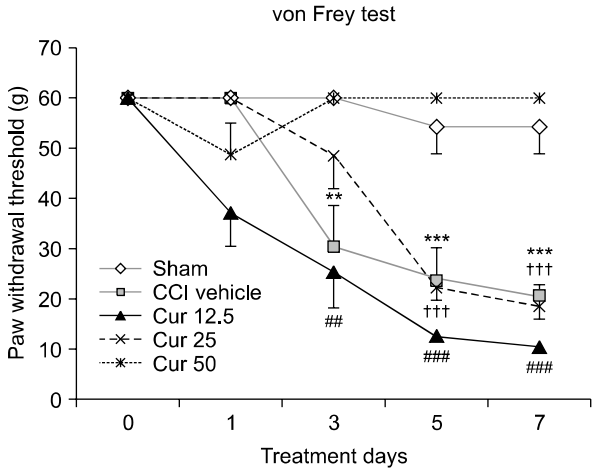

Fig. 2. Paw withdrawal threshold in response to von Frey filaments before and at several time points after surgery in CCI vehicle-treated, sham-operated and CCI curcumin treated-groups. Curcumin (12.5, 25 and 50 mg/kg) was injected i.p. Data are presented as means ± S.E.M. of 8 rats in each group. Asterisks (**$P < 0.01$; ***$P < 0.001$) for CCI vehicle-treated group ($^{\dagger\dagger\dagger}P < 0.01$; $^{\dagger\dagger\dagger}P < 0.001$), for curcumin 25 mg/kg treated group and ($^{\#\#}P < 0.01$; $^{\#\#\#}P < 0.001$) for curcumin 12.5 mg/kg treated group, indicate a statistically significant difference when compared to day 0 paw withdrawal latency value.

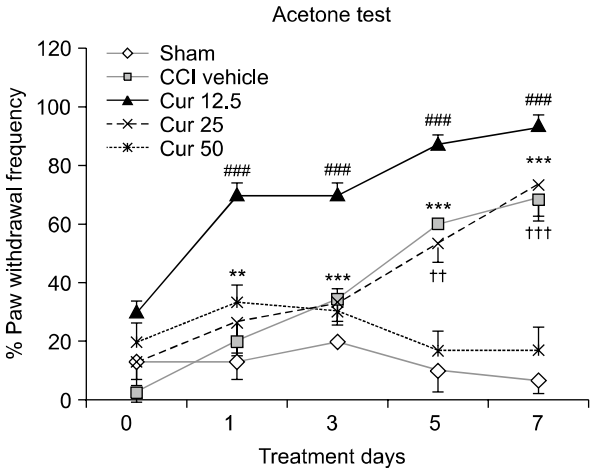

Fig. 3. The frequency of paw withdrawal in response to acetone before and at several time points after surgery in CCI vehicle-treated, sham-operated and CCI curcumin treated-groups. Curcumin (12.5, 25 and 50 mg/kg) was injected i.p. Data are presented as means ± S.E.M. of 8 rats in each group. Asterisks (**$P < 0.01$; ***$P < 0.001$) for CCI vehicle-treated group ($^{\dagger\dagger}P < 0.01$; $^{\dagger\dagger\dagger}P < 0.001$), for curcumin 25 mg/kg treated group and ($^{\#\#\#}P < 0.001$) for curcumin 12.5 mg/kg treated group, indicate a statistically significant difference when compared to day 0 paw withdrawal frequency value.

tion compared to day 0 ($P < 0.01$), this effect was remained until the end of the study ($P < 0.001$). On the contrary, sham-operated animals and curcumin (50 mg/kg) treated rats did not produce any significant paw withdrawal reaction during the experimental days compared to day 0. However, allodynia was produced in curcumin (12.5 and 25 mg/kg) treated rats when compared to day 0 ($P < 0.01$, $P < 0.001$) (Fig. 2).

2. Response to cold allodynia

In CCI vehicle-treated rats, a significant difference in pain behavior ($P < 0.01$) was seen on the first day post-injury compared to day 0, which continued until the end of the study ($P < 0.001$). However, cold allodynia was not observed in any of post-ligation days in sham-operated group. In CCI curcumin-treated group, allodynic effect was significantly reduced only by using 50 mg/kg curcumin ($P < 0.001$) (Fig. 3).

3. Cyclooxygenase 2 protein analysis

Protein analysis by ELISA revealed a reduction in serum cyclooxygenase 2 level in the presence of curcumin.

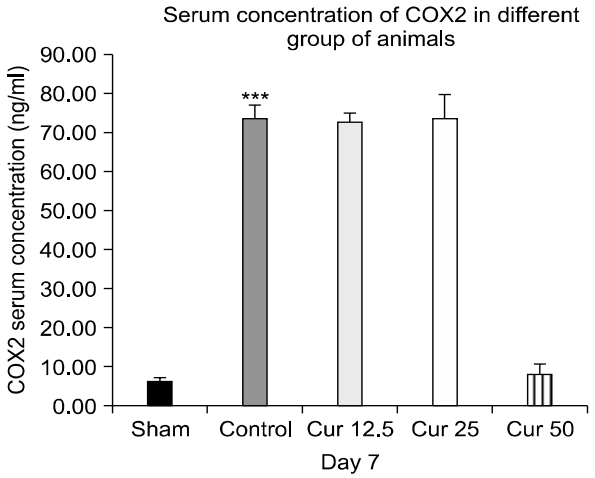

Fig. 4. Serum concentration of COX-2 in CCI vehicle-treated, sham-operated and CCI curcumintreated rats on day 7 post-ligation. Data are presented as means ± S.E.M. of 8 rats in each group. Asterisks (***$P < 0.001$) indicate a statistically significant difference when compared to CCI vehicle-treated rats. Cur 12.5 = curcumin 12.5 mg/kg, Cur 25 = curcumin 25 mg/kg, Cur 50 = curcumin 50 mg/kg.

Curcumin 50 mg/kg ($P < 0.001$) but not 25 and 12.5 mg/kg, attenuated the serum concentration of cyclooxygenase 2 compared to CCI vehicle-treated animals. In sham-operated animals there was no increase in serum cyclooxygenase 2 level ($P < 0.001$) compared to CCI vehicle-treated animals (Fig. 4).

DISCUSSION

In this study we investigated the analgesic effects of curcumin on CCI model of neuropathic pain and in relation to that, the serum level of cyclooxygenase 2 enzyme was measured, during the experimental period no overt signs of sedation were observed in any of the animals dosed with curcumin. CCI model of nerve injury has an inflammatory and a nerve injury component, which mimics the type of neuropathic pain found in human [22]. The peripheral nociceptor is an important target of pain therapy because many pathological conditions such as inflammation excite and sensitize peripheral nociceptors [24]. Acute and chronic peripheral inflammation, interleukins and spinal cord injury increase the expression of cyclooxygenase 2 and release of PGE_2 and PGI_2. The spinal cord is one of the sites where non-steroidal anti-inflammatory drugs (NSAIDs) act to produce analgesia. Expression of cyclooxygenase 1 and cyclooxygenase 2 in the spinal cord and primary afferents suggests that NSAIDs act by inhibiting the synthesis of PGs. However adverse reactions complicate therapy with NSAIDs. A large number of studies have revealed that curcumin has wide therapeutic actions. The most interesting feature of curcumin is lack of gastrointestinal side effects despite being an anti-inflammatory agent [25]. Curcumin may be considered as a natural alternative to non-steroidal agents for the treatment of inflammatory pain [26].

In our study, we demonstrated the effective reduction of mechanical and cold allodynic pain behaviors in CCI model of neuropathic pain in rat treated by curcumin. These findings are consistent with the previous studies showing the ability of curcumin to reduce pain behavior [27]. Evidence has shown the antihyperalgesic effect of curcumin on inflammatory and neuropathic pain. Curcumin significantly attenuated mechanical allodynia and successfully prevented the development of neuropathic pain [28]. In another study, chronic treatment with curcumin significantly reversed thermal hyperalgesia and cold allodynia in sciatic nerve ligated animals, indicating its therapeutic potentials against neuropathic pain [29,30]. Moreover, it is well established that the anti inflammatory activity might be one of the underlying mechanisms for this effect [31,32]. In a mouse model of diabetic neuropathic pain, using tail immersion and hot plate assays, curcumin produced antinociceptive effect to hyperalgesic pain [33]. It is also assumed that curcumin plays a defensive role in alleviating the formalin-induced nociceptive behavior [7]. Accordingly, the effectiveness of curcumin in formalin test model has been reported, in which intraperitoneal administration of curcumin decreased both acute and tonic phases of formalin test [10].

The medicinal properties of curcumin could be mediated by numerous molecular targets. The anti inflammatory activity of curcumin is mainly due to inhibition of arachidonic acid metabolism, cyclooxygenase, Lipooxygenase, cytokines (Interleukins and Tumor necrosis factor) and inducible nitric oxide synthase (iNOS) most likely through the down regulation of NF-κB activation, which is involved in regulation of cyclooxygenase 2 expression [19,25,34,35].

The inducible cyclooxygenase 2 is an important enzyme that mediates inflammatory responses. Following nociceptive stimulation, PGs are released at the site of injury and in the dorsal horn of the spinal cord and stimulate the "sleeping" nociceptors, which are associated with the induction of hyperalgesia and allodynia [36]. It has been shown that direct administration of PGs to the spinal cord causes hyperalgesia and allodynia, and some studies have shown an association between induction of cyclooxygenase 2, increased PG release and enhanced nociception. Also cyclooxygenase 2 is expressed in activated microglial cells and appears to be an important source of PGs during inflammatory conditions [36-38]. Cyclooxygenase 2 inhibitors act primarily in the dorsal horn to cause analgesia. Cyclooxygenase 2, which is expressed constitutively in the dorsal horn of spinal cord, up-regulates briefly after a trauma and therefore, facilitates transmission of the nociceptive input [13,39].

It has also been demonstrated that cyclooxygenase 2 is dramatically up-regulated in infiltrating macrophages in injured nerve following partial sciatic nerve ligation (PSNL). The highest cyclooxygenase 2 expression, in terms of distribution region and the number of cells expressing cyclooxygenase 2 was observed in injured sciatic nerve of CCI

rats. CCI results in a much wider injury region due to four loose ligatures. Thus, injured nerve of CCI rat likely produces the greatest amount of PGs [40].

In our study, low dose of curcumin was not effective in reducing pain behavior, however, regarding to the pain duration, it seems that, different doses of curcumin could be effective in different models of chronic pain [7,9,10]. Therefore, considering that curcumin even with high dose do not exhibit toxic effects [10,11], in this research, higher dose of curcumin was used to control pain behavior.

In this regard, we found that in animal treated with curcumin, serum cyclooxygenase 2 level was markedly declined. This finding could probably highlight the fact that curcumin can reduce pain by reducing the inflammatory effects of increased cyclooxygenase 2 in rats. A down regulation of the expression of the NF-kB-regulated gene products such as cyclooxygenase 2, TNF, IL-1, IL-6 and other molecules has been described in previous studies [41]. These findings suggest that curcumin treatment can play an important role in pain regulations at central and peripheral levels [2-4,26].

In conclusion, curcumin could be an effective and a useful drug in treatment of inflammatory and chronic pain, in comparison to other synthetic analgesic agents. However, further investigation is needed to explore the new areas of therapeutic applications and the precise mechanism of action of curcumin in the neuropathic pain.

ACKNOWLEDGEMENTS

This research was supported by the grant offered by the Neuroscience Research Center of Shahid Beheshti University of Medical Sciences. The authors thank the staff of the Department of Pharmacology specially Dr Ziai for their kind collaboration.

REFERENCES

1. Blackburn-Munro G. Pain-like behaviours in animals - how human are they? Trends Pharmacol Sci 2004; 25: 299-305.
2. Zimmermann M. Pathobiology of neuropathic pain. Eur J Pharmacol 2001; 429: 23-37.
3. Campbell JN, Meyer RA. Mechanisms of neuropathic pain. Neuron 2006; 52: 77-92.
4. Dray A. Neuropathic pain: emerging treatments. Br J Anaesth 2008; 101: 48-58.
5. Suzuki R, Dickenson AH. Differential pharmacological modulation of the spontaneous stimulus-independent activity in the rat spinal cord following peripheral nerve injury. Exp Neurol 2006; 198: 72-80.
6. Epstein J, Sanderson IR, Macdonald TT. Curcumin as a therapeutic agent: the evidence from in vitro, animal and human studies. Br J Nutr 2010; 103: 1545-57.
7. Sharma S, Kulkarni SK, Agrewala JN, Chopra K. Curcumin attenuates thermal hyperalgesia in a diabetic mouse model of neuropathic pain. Eur J Pharmacol 2006; 536: 256-61.
8. Oh SW, Cha JY, Jung JE, Chang BC, Kwon HJ, Lee BR, et al. Curcumin attenuates allergic airway inflammation and hyper-responsiveness in mice through NF-κB inhibition. J Ethnopharmacol 2011; 136: 414-21.
9. Sharma S, Chopra K, Kulkarni SK. Effect of insulin and its combination with resveratrol or curcumin in attenuation of diabetic neuropathic pain: participation of nitric oxide and TNF-alpha. Phytother Res 2007; 21: 278-83.
10. Mittal N, Joshi R, Hota D, Chakrabarti A. Evaluation of antihyperalgesic effect of curcumin on formalin-induced orofacial pain in rat. Phytother Res 2009; 23: 507-12.
11. Chattopadhyay I, Biswas K, Bandyopadhyay U, Banerjee RK. Turmeric and curcumin: biological actions and medicinal applications. Curr Sci 2004; 87: 44-53.
12. Stock JL, Shinjo K, Burkhardt J, Roach M, Taniguchi K, Ishikawa T, et al. The prostaglandin E2 EP1 receptor mediates pain perception and regulates blood pressure. J Clin Invest 2001; 107: 325-31.
13. Luo C, He ML, Bohlin L. Is COX-2 a perpetrator or a protector? Selective COX-2 inhibitors remain controversial. Acta Pharmacol Sin 2005; 26: 926-33.
14. Neuss H, Huang X, Hetfeld BK, Deva R, Henklein P, Nigam S, et al. The ubiquitin- and proteasome-dependent degradation of COX-2 is regulated by the COP9 signalosome and differentially influenced by coxibs. J Mol Med (Berl) 2007; 85: 961-70.
15. Rainsford KD. Current status of the therapeutic uses and actions of the preferential cyclo-oxygenase-2 NSAID, nimesulide. Inflammopharmacology 2006; 14: 120-37.
16. Kimura T, Iwase M, Kondo G, Watanabe H, Ohashi M, Ito D, et al. Suppressive effect of selective cyclooxygenase-2 inhibitor on cytokine release in human neutrophils. Int Immunopharmacol 2003; 3: 1519-28.
17. Rang HP, Dale MM, Ritter JM, Flower RJ. Anti-inflammatory and immunosuppressant drugs. In: Rang and Dale pharmacology. 6th ed. Edinburgh, Churchill Livingston. 2007, p 232.
18. Bryant DM, Alldred A. Rheumatoid arthritis and osteoarthritis. In: Clinical pharmacy and therapeutics. 4th ed. Edited by Walker R, Whittlesea C. Edinburgh, Churchill Livingstone. 2007, pp 791-812.
19. Shishodia S, Sethi G, Aggarwal BB. Curcumin: getting back

to the roots. Ann N Y Acad Sci 2005; 1056: 206–17.
20. Menon VP, Sudheer AR. Antioxidant and anti-inflammatory properties of curcumin. Adv Exp Med Biol 2007; 595: 105–25.
21. Zimmermann M. Ethical guidelines for investigations of experimental pain in conscious animals. Pain 1983; 16: 109–10.
22. Bennett GJ, Xie YK. A peripheral mononeuropathy in rat that produces disorders of pain sensation like those seen in man. Pain 1988; 33: 87–107.
23. Stuesse SL, Crisp T, McBurney DL, Schechter JB, Lovell JA, Cruce WL. Neuropathic pain in aged rats: behavioral responses and astrocytic activation. Exp Brain Res 2001; 137: 219–27.
24. Schaible HG, Ebersberger A, Natura G. Update on peripheral mechanisms of pain: beyond prostaglandins and cytokines. Arthritis Res Ther 2011; 13: 210.
25. Kohli K, Ali J, Ansari MJ, Raheman Z. Curcumin: a natural antiinflammatory agent. Indian J Pharmacol 2005; 37: 141–7.
26. Lim GP, Chu T, Yang F, Beech W, Frautschy SA, Cole GM. The curry spice curcumin reduces oxidative damage and amyloid pathology in an Alzheimer transgenic mouse. J Neurosci 2001; 21: 8370–7.
27. Li Y, Zhang Y, Liu DB, Liu HY, Hou WG, Dong YS. Curcumin attenuates diabetic neuropathic pain by downregulating TNF-α in a rat model. Int J Med Sci 2013; 10: 377–81.
28. Jeon Y, Kim CE, Jung D, Kwak K, Park S, Lim D, et al. Curcumin could prevent the development of chronic neuropathic pain in rats with peripheral nerve injury. Curr Ther Res Clin Exp 2013; 74: 1–4.
29. Kim CE, Park ES, Jeon YH. Curcumin attenuates chronic constriction nerve injury-induced neuropathic pain in rats. Korean J Med Crop Sci 2008; 16: 183–7.
30. Han YK, Lee SH, Jeong HJ, Kim MS, Yoon MH, Kim WM. Analgesic effects of intrathecal curcumin in the rat formalin test. Korean J Pain 2012; 25: 1–6.
31. Bengmark S. Curcumin, an atoxic antioxidant and natural NFkappaB, cyclooxygenase-2, lipooxygenase, and inducible nitric oxide synthase inhibitor: a shield against acute and chronic diseases. JPEN J Parenter Enteral Nutr 2006; 30: 45–51.
32. Yeon KY, Kim SA, Kim YH, Lee MK, Ahn DK, Kim HJ, et al. Curcumin produces an antihyperalgesic effect via antagonism of TRPV1. J Dent Res 2010; 89: 170–4.
33. Tajik H, Tamaddonfard E, Hamzeh-Gooshchi N. Interaction between curcumin and opioid system in the formalin test of rats. Pak J Biol Sci 2007; 10: 2583–6.
34. Chainani-Wu N. Safety and anti-inflammatory activity of curcumin: a component of tumeric (Curcuma longa). J Altern Complement Med 2003; 9: 161–8.
35. Orlando RA, Gonzales AM, Hunsaker LA, Franco CR, Royer RE, Vander Jagt DL, et al. Inhibition of nuclear factor kappaB activation and cyclooxygenase-2 expression by aqueous extracts of Hispanic medicinal herbs. J Med Food 2010; 13: 888–95.
36. Tegeder I, Niederberger E, Vetter G, Bräutigam L, Geisslinger G. Effects of selective COX-1 and -2 inhibition on formalin-evoked nociceptive behaviour and prostaglandin E(2) release in the spinal cord. J Neurochem 2001; 79: 777–86.
37. Vanegas H, Schaible HG. Prostaglandins and cyclooxygenases [correction of cycloxygenases] in the spinal cord. Prog Neurobiol 2001; 64: 327–63.
38. Kang G, Kong PJ, Yuh YJ, Lim SY, Yim SV, Chun W, et al. Curcumin suppresses lipopolysaccharide-induced cyclooxygenase-2 expression by inhibiting activator protein 1 and nuclear factor kappab bindings in BV2 microglial cells. J Pharmacol Sci 2004; 94: 325–8.
39. Hinz B, Brune K. Cyclooxygenase-2--10 years later. J Pharmacol Exp Ther 2002; 300: 367–75.
40. Ma W, Eisenach JC. Cyclooxygenase 2 in infiltrating inflammatory cells in injured nerve is universally up-regulated following various types of peripheral nerve injury. Neuroscience 2003; 121: 691–704.
41. Aggarwal BB, Harikumar KB. Potential therapeutic effects of curcumin, the anti-inflammatory agent, against neurodegenerative, cardiovascular, pulmonary, metabolic, autoimmune and neoplastic diseases. Int J Biochem Cell Biol 2009; 41: 40–59.