

Analgesic Properties of Methanolic Extract of *Matricaria recutita* in Rats in both Acute and Chronic Pains

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Abstract: *Matricaria recutita* is an important medicinal plant belonging to the family of Asteraceae. It is a well recognized plant in the traditional medicine and is used by people in rural areas. This study is aimed to evaluate the anti-pain effects of methanolic extract of *Matricaria recutita*. The experiments were performed on 40 male wistar rats weighting 190 to 200 g, which were divided into 5 groups, each consisting of 8 rats. To evaluate the analgesic effects, the formalin induced pain-test was used. All animals were pre-treated with an oral dose of extracts (100, 200, 300 mg/kg). The control group received no drug and the witness group at only received distilled water. Results of this study showed that methanolic extract of *Matricaria recutita* can significantly reduce the pain depending on the dose in acute phase. However, in chronic phase, the high dose of the extract could reduce the pain. It seems that flavonoids can probably reduce intracellular calcium through inhibiting of NMDA receptors. It also reduces activities of nitric oxide synthase enzyme and calcium dependent phospholipids A2, which results in antinociceptive effects.

Keywords: Analgesia, *Matricaria recutita*, rat

INTRODUCTION

At present, control of pain is done by using Nonsteroidal Anti-Inflammatory Drugs (NSAIDs) and opioid analgesics. There are many evident that show neurochemical systems, such as opioid system, involvement in pain controlling (Li and Clark, 1999). Opioid analgesics, especially morphine, have a high efficiency in relieving both acute and chronic pain. Repeated use of morphine can cause progressive decrease in its effects such that one would require more dose of morphine to achieve the same effect, which, in turn, is followed by addiction and many side effects too Ekhtiari *et al.* (2002). In this phenomenon, proteins such as protein kinase C, calcium calmodulin, protein kinase A and protein kinases related to CGMP, which play part in phosphorylation of N-Methyl-D-Aspartate (NMDA), are increased quantitatively. Therefore, nitric oxide content increases and NO/CGMP/PKG course is begun (Blednov *et al.*, 2003). A method to attain new analgesics, with more efficiency and less restricting effects, is focusing on and attention to herbs and natural substances. Today, studying herbal species which are traditionally used as analgesics is considered a useful research strategy to prepare new analgesics (Elisabetsky *et al.*, 1995). *Matricaria recutita* belongs to family Asteraceae. It mainly grows many parts of the world. This herb has large history for its therapeutic effects.

Matricaria recutita is well known for its pharmaceutical properties including; anti-inflammatory (Shipochliev *et al.*, 1981; Al-Hindawi *et al.*, 1989), immunomodulatory activity (Uteshev *et al.*, 1999), arcaricadal property (Macchioni *et al.*, 2004), anti-cancer activity (Srivastava and Gupta, 2007), antipruritic effect (Kobayashi *et al.*, 2005), wound healing property (Nayak *et al.*, 2007; Jarrahi, 2008; Jarrahi *et al.*, 2008; Martins *et al.*, 2009), treatment of oral mucositis (Mazokopakis *et al.*, 2005), intracanal irrigant (Sadr *et al.*, 2006), Treatment of infant botulism (Bianco *et al.*, 2008), treatment of gastrointestinal disorders (Mahady *et al.*, 2005), antimicrobial activity (Nogueira *et al.*, 2008), antiulcer activity (Khayyal *et al.*, 2001), treatment of stress and depression (Pinto *et al.*, 2008), anti-allergic activity (Kobayashi *et al.*, 2003), antisolar agent (Ramos *et al.*, 1996), inhibition of poliovirus replication (Vilaginès *et al.*, 1985), anxiolytic agent (Amsterdam *et al.*, 2009), Prevent osteoporosis (Kassi *et al.*, 2004). Due to lack of scientific studies on the effects of *Matricaria recutita* on the pain, we decided to perform this study.

MATERIALS AND METHODS

Plant material: *Matricaria recutita* collected from Agricultural Research institute at University of Zabol, Zabol, Iran, in April 2011 and authenticated at

Department of Biology, University of Zabol. Its leaves, fruits and flowers were dried, under shade and powdered. The extract was prepared by maceration method in methanol and then filtered with filter paper. Ethanol was removed by rotary evaporator. The extract was dissolved in normal saline and administrated orally into rats.

Animals and treatments: In this research, 40 adult male Wistar rats with approximate weight of 190-200 g were selected and were kept under equal conditions regarding weather, light and nutrition for a week before the test. We used formalin test to study antinociceptive effects of the extract. And for this purpose, we used eight adult male rats in each group. An hour after injecting 100, 200 and 300 mg/Kg of methanolic extract of *Matricaria recutita* to experimental groups, 50 μ L formalin 2.5% was injected into rat's right sole. Then the rat was immediately kept within test case. The control group received no treatment while witness group received 1 mL distilled water (solvent of extract), that is, as equivalent as consumed dose by experimental groups. Each rat's behavioral response to algogenic stimulant was rated for an hour at 15 sec intervals. In this observation, rates zero to 3 were given every 15 sec based the rat's behavior in appearing pain in the foot into which formalin has been injected. (Zero, where the rat uses his treated foot like the other foot; 1, where the rat puts his treated foot on the ground but doesn't lean on it; 2 where the rat doesn't put his treated foot on the ground; and 3 where the rat moves, licks and bites his treated foot). Then, we calculated pain intensity of each rat at 5 min intervals and analyzed mean pain intensity statistically. In formalin test, formalin injection into rat's sole would cause 2-step incidence of pain. The first step is quick and acute pain which hits peak during 5 min. Then pain intensity is reduced for 5 to 10 min and 20 min after formalin injection, the second step of pain which is also called chronic pain restarts and lasts 60 min after formalin test.

Statistical analysis: Statistical analysis was done by variance analysis, ANOVA, T. Test. A significant difference was considered between groups ($p < 0.05$). The results are shown as standard deviation \pm average.

RESULTS

As shown in diagram 1, methanolic extract of *Matricaria recutita* (100, 200, 300 mg/Kg) has caused significant decrease in pain rate in acute pain step of formalin test compared to control and witness groups ($p < 0.05$) (Fig. 1). Statistical analysis of results of analgesic effects of methanolic extract of *Matricaria*

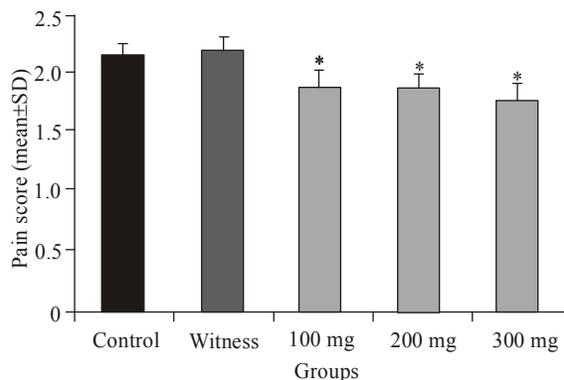


Fig. 1: Comparison mean pain intensity in acute pain step between rats receiving different doses of *Matricaria recutita* extract and control and witness groups, *: ($p < 0.05$)

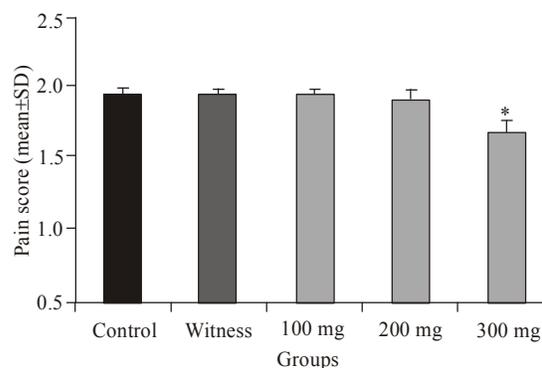


Fig. 2: Comparison mean pain intensity in chronic pain step between rats receiving different doses of extract of *Matricaria recutita* and control and witness groups, *: ($p < 0.05$)

recutita in chronic pain step shows that the extract can relieving pain in chronic step only by maximum quantity ($p < 0.05$) (Fig. 2).

DISCUSSION

According the findings from present study, methanolic extract of *Matricaria recutita* relieves acute pain of formalin test more than chronic pain. It seems that acute pain step is induced by formalin injection, direct stimulation of pain receptors and activities of nervous fibers type C while in chronic pain step, they are a set of inflammatory reactions in injured tissue and functional changes in posterior horn of spinal cord that induce pain (Tjolsen *et al.*, 1991; Chi and Jun, 1990).

These functional changes are induced by stimulations of type C nervous fibers. Substances such as substance P, bradikinin, histamine and

prostaglandines are involved in this stimulation (Mersky, 1986). Studies by other researchers show that extract of *Matricaria recutita* contains flavonoids. Therefore, regarding to presence of flavonoid compounds in this herb, some properties of analgesic effects of *Matricaria recutita* is probably related to these compounds. Flavonoids control phospholipase, lipoxygenase and cyclooxygenase which effect directly on prostaglandines and cause analgesic effects. Flavonoids are considered one of the controllers of nitric oxide synthesizing enzyme and prevent nitric oxide production, which increases following formalin injection (Willis, 2001). Prostaglandins stimulate pain receptors both directly and by promoting their sensitivity to other agents like bradikinin. Therefore, flavonoids improve inflammation by controlling cyclooxygenase in inflamed tissue and prevent formation of prostaglandins (Toker *et al.*, 2004; Longo *et al.*, 2004). Also, studies have shown that flavonoids decrease intracellular calcium by controlling activity of N-Methyl D-aspartate followed by decrease in activity of nitric oxide synthesizing enzyme and phospholipase dependent to calcium. By decrease in nitric oxide and prostaglandins, analgesic effects would appear. The presence of flavonoid compounds in *Matricaria recutita* accounts for its analgesic effects by above mentioned mechanisms. According to the results from present study and studies by other researchers, analgesic effect of methanolic extract of *Matricaria recutita* can be attributed to flavonoids existing in *Matricaria recutita*, although more studies are needed to offer more exact answers on determining its effective substance.

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