Original article / *Araştırma*

Efficacy of mirtazapine in neuropathic pain model

Şeyda FİGÜL GÖKÇE,[1] Burhanettin ÇİĞDEM[1]

ABSTRACT

Objective: *Neuropathic pain, which is chronic pain, can affect the quality of life individuals. It can cause psychiatric problems such as depression and anxiety. Mirtazapine is an atypical antidepressant which has both noradrenergic and specific serotonergic receptor antagonism and shown to have anxiolytic, anti-emetic and appetite stimulating effects along with its antidepressant effect. It is generally tolerated better and has a different way of action when compared with other antidepressants used to treat neuropathic pain. Thus, we aimed to investigate its potential preventive effect in neuropathic pain in nerve ligated rat model.* ***Methods:*** *Forty rats were divided into five groups including the sham, the sciatic nerve ligation group and the third, fourth and fifth sciatic nerve ligation+mirtazepine groups which were treated with 5 mg, 10 mg and 15 mg/day mirtazapine, respectively. The tail flick and hot plate tests were performed on all the five groups, and the latency times were measured. Morphine was used as a positive control agent.* ***Results:*** *Administration of mirtazapine, restored both tail flick and hot plate latencies in a dose-dependent manner, meaning that mirtazapine is effective in treating neuropathic pain in this animal model. Mirtazapine also restored the antinociceptive response to morphine significantly.* ***Discussion:*** *Mirtazapine appears to be good candidate for both neuropathic pain treatment and accompanying psychiatric condition such as depression and anxiety with its dual effect on neurotransmitters.* ***(Anatolian Journal of Psychiatry 2020; 21(5):461-467)***

Keywords: *mirtazapine, neuropathic pain*

Nöropatik ağrı modelinde mirtazapinin etkinliği

ÖZ

Amaç: *Kronik ağrı olan nöropatik ağrı bireylerin yaşam kalitesini etkileyeblir ve depresyon, anksiyete bozukluğu gibi psikiyatrik bozukluklara neden olabilir. Mirtazapin hem noradrenerjik, hem de özgül serotonerjik reseptör antagonisti olan atipik bir antidepresandır. Antidepresan etkisi yanında anksiyolitik, antiemetik ve iştah açıcı etkilerinin olduğu da gösterilmiştir. Mirtazapin nöropatik ağrı tedavisinde kullanılan diğer antidepresanlarlardan farklı bir etki yoluna sahiptir ve genel olarak daha iyi tolere edilir. Bu nedenle çalışmamızda sinir ligasyonu yapılan rat modelinde mirtazapinin nöropatik ağrıdaki koruyucu etkinliğini araştırmayı amaçladık.* ***Yöntem:*** *Kırk rat beş gruba ayrıldı. İlk grup sham grubu, ikinci grup sadece siyatik sinir ligasyonu yapılan gruptu. Üçüncü, dördüncü ve beşinci gruplara siyatik sinir ligasyonunun ardından sırasıyla 5 mg, 10 mg ve 15 mg/gün mirtazapin verildi. Grupların hepsine tail flick ve hot plate testleri yapıldı, latans süreleri ölçüldü. Mirtazapinin nöropatik ağrıda analjezik etkinliğinin dışında pozitif kontrol ajanı olarak kullandığımız morfin üzerine etkisi de değerlendirildi.* ***Bulgular:*** *Nöropatik ağrıda mirtazapinin tail flick ve hot plate latanslarında doza bağlı olumlu etkilerinin olduğu ve morfine verilen antinosiseptif yanıtı da önemli ölçüde geri kazandırdığı sonucuna vardık.* ***Tartışma:*** *Mirtazapin, nörotransmitterler üzerinden ikili etkisi ile hem nöropatik ağrı tedavisi, hem de eşlik eden depresyon, anksiyete gibi psikiyatrik bozukluklar için iyi bir aday olarak görünmektedir.* ***(Anadolu Psikiyatri Derg 2020; 21(5):461-467)***

Anahtar sözcükler: *mirtazapin, nöropatik ağrı*

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INTRODUCTION

Neuropathic pain (NP), is one of the main types of chronic pain, caused by a defect affecting the afferent nerves carrying the pain signal to the central nerve system. NP has a significant impact on patients' quality of life and increased frequency of psychiatric disorders such as depression and anxiety. Additionally It is connected with a high economic burden on the both individual and society.[1,2]

Although there are many analgesic agents, using different pathways, to reduce pain, there is no effective agent for the treatment of neuropathic pain. Thus, different therapeutic options are currently being considered. Since it has been thought that serotonergic and noradrenergic systems take a big part in the pathophysiology of chronic pain,[3,4] antidepressants and anticonvulsants constitute examples for this novel approach replacing classical analgesics (non-steroidal anti-inflammatory agents) where they are not sufficient.[5]

Antidepressants show their analgesic effect by increasing the levels of monoamines such as noradrenaline and serotonin via presynaptic and postsynaptic reuptake blockage.[6] Many antidepressants present their analgesic effect at peripheral, spinal and supraspinal levels.[7-9]

Mirtazapine is a new generation antidepressant, which has an antagonistic impact on the reuptake of both serotonin and noradrenaline. Mirtazapine activates the descending pain modulatory pathways by increasing the levels of monoamines in the CNS.[10] It enhances noradrenergic transmission through indirectly increasing NA release, instead of by blocking neuronal α2-adrenergic autoreceptors. Release of NA, in turn, intensifies serotonergic neurotransmission on serotonergic cell bodies and presynaptic nerve terminals. Alfa2 adrenergic receptors are responsible for the antinociceptive effect of mirtazapine at spinal level.[11] 5-HT2 receptors, especially 5-HT2C, also mediate the antinociceptive effect at spinal level.[12]

The present study aims to investigate the analgesic effects of mirtazapine in the neuropathic pain model established in rats and compare its potency with morphine in a chronic pain scenario.

METHODS

This prospective and controlled study was conducted in the animal experiments laboratory of Sivas Cumhuriyet University between March and June 2018. The study was duly approved by the Institutional Animal Ethics Committee, Cumhuriyet University Medical Faculty, Sivas, Turkey (Approval no. IAEC/VMMC 02/01/2018 numbered 99 named).

Healthy male Wistar rats weighing 180-250 g were used for the study. Animals were housed in groups of 4-5 per cage with free access to a pellet diet and water in a temperature-controlled facility (temperature: 22±2°C; humidity: 50-55%). All experiments were performed at the daytime between 09^{30} and $15.^{30}$

Forty rats were divided into five groups (eight animals in each), including sham group, nerve ligation group, experiment group 1, experiment group 2 and experiment group 3.

Sham group animals underwent a sham operation. Then, 1 ml physiological serum was administered intraperitoneally (i.p.) per day for 3 weeks.

All other groups underwent nerve ligation operation. In the nerve ligation group, 1 ml physiological serum was administered i.p. per day for 3 weeks. In the experiment group -1, 5 mg/kg/day mirtazapine was administered i.p. for 3 weeks. In the experiment group 2 and 3, 10 mg/kg/day and 15 mg/kg/day mirtazapine was administered i.p. for 3 weeks, respectively. The baseline tail-flick and hot plate reads were repeated in all groups.

After 3 weeks all groups were administrated with 3 mg/kg morphine i.p., and tail flick and hot plate latencies were assessed once again.

The chronic constriction injury of the sciatic nerve was used as a model for the induction of neuropathic pain. Animals were anaesthetized with chloral hydrate (350 mg/kg, i.p.). Thereafter, the right sciatic nerve was exposed at the level of mid-thigh, and two ligatures were tied proximal to the trifurcation of the sciatic nerve, according to the modified method of Bennett and Xie.[8] The ligation was of sufficient strength to produce a slight indentation of the nerve, but not to induce muscle twitch. The wound was closed, and Neosporin powder was sprayed. Sham surgery was performed on the left side, where the nerve was exposed, but no ligature was made. Animals were treated with an antibiotic (gentamicin, 0.4 mg/kg, i.p.) for 3 days and allowed to recover for 21 days, after which they were subjected to the behavioral tests.

Pain response to thermal stimuli was assessed by the method, as described by D'Amour and

Smith,[13] using a standard tail-flick apparatus. In this, radiant heat was applied to the distal end of the tail. The intensity of the thermal stimulus was adjusted to provide an average baseline tail-flick latency of 6-7 seconds. A cut-off time of 15 seconds was kept to avoid any injury to the tail. Animals not responding within the cut-off time were removed and assigned a latency of 15 seconds.

Responses to thermal stimuli were assessed using a hot-plate test.[14] Rats were placed on a hot plate (Kommat Ltd, Turkey), for which the temperature was set at 55°C. The latency response to either a hindpaw lick or a jump was recorded. A cut-off time of 30 seconds was observed to avoid any tissue damage.

The percentage of maximum possible effect (% MPE) was calculated for each rat at each dose and time point according to the following formula: %MPE=(post-treatment latency (s) – baseline latency (s)) / (cut-off latency (s) – baseline latency (s)) x 100.

The results obtained are expressed as mean± SEM (standard error of the mean). The effect of antinociception was measured, and the mean of %MPEs in all groups was calculated. The data were analyzed by analysis of variance followed by the Tukey test. $p<0.05$ value was considered significant.

RESULTS

Induction of neuropathic pain

Tail-flick and hot plate latencies were re-measured 3 weeks after the sciatic nerve ligation. Significant differences between sham and sciatic nerve ligation groups in both tail flick and hot plate tests were detected, that is, 4.4±0.4 vs. 2.3±0.2 and 13.4±0.4 vs. 7.3±0.6, respectively ($p=0.001$) (Figure 1).

Effect of mirtazapine on neuropathic pain

In order to see if mirtazapine has a preventive effect on the neuropathic pain, mirtazapine was

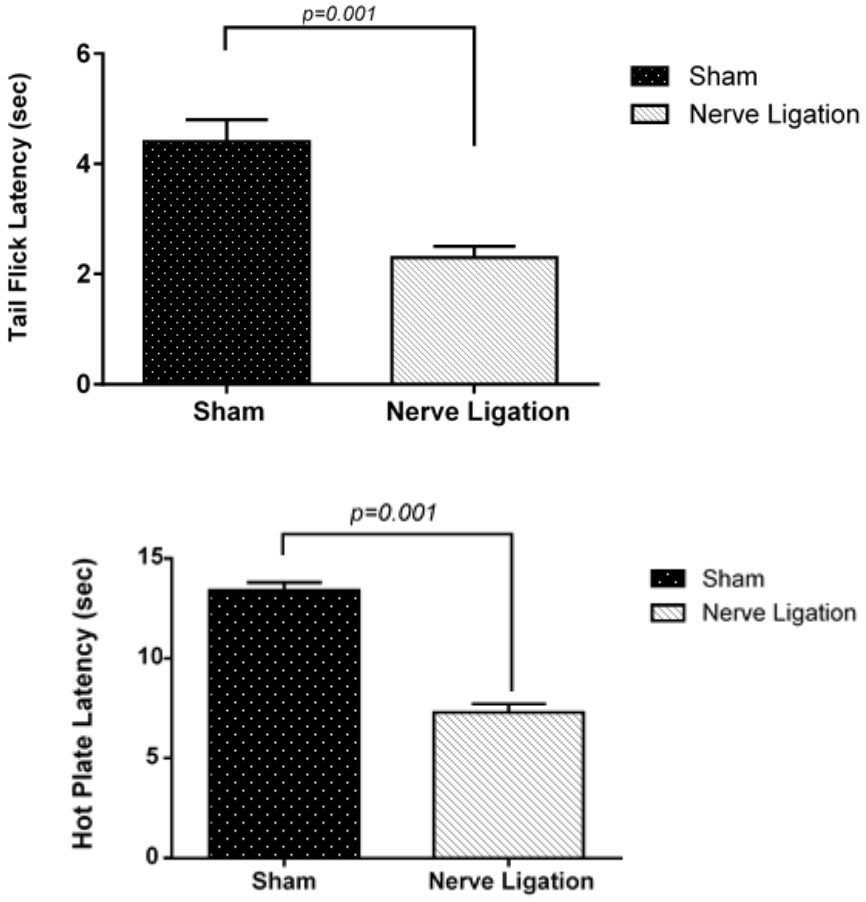

Figure 1. Tail flick (A) and hot plate (B) latencies of sham and nerve ligation groups

was administrated in three different doses, including 5, 10 and 15 mg/kg/day for 3 weeks after nerve ligation. Tail flick latencies of three doses of mirtazapine administration were 3.2±0.2, 3.5±0.3 and 4.1±0.2, respectively. Hotplate latencies were also measured after the administration of three different doses of mirtazapine. Latencies were 8.3+0.5, 10.5+0.6 and 12.8+0.8, respectively. Mirtazapine created a dose-dependent antinociceptive effect and increased latencies in both tail flick and hot plate tests. There was no significant difference between the sham group and experiment group 3 (mirtazapine 15 mg/kg/day), which indicated that mirtazapine restored neuropathic pain and it was increased to the sham group levels (Figure 2).

Effect of morphine on nerve ligation

In order to see the effect of the neuropathic pain on analgesic processes, the analgesic effect of

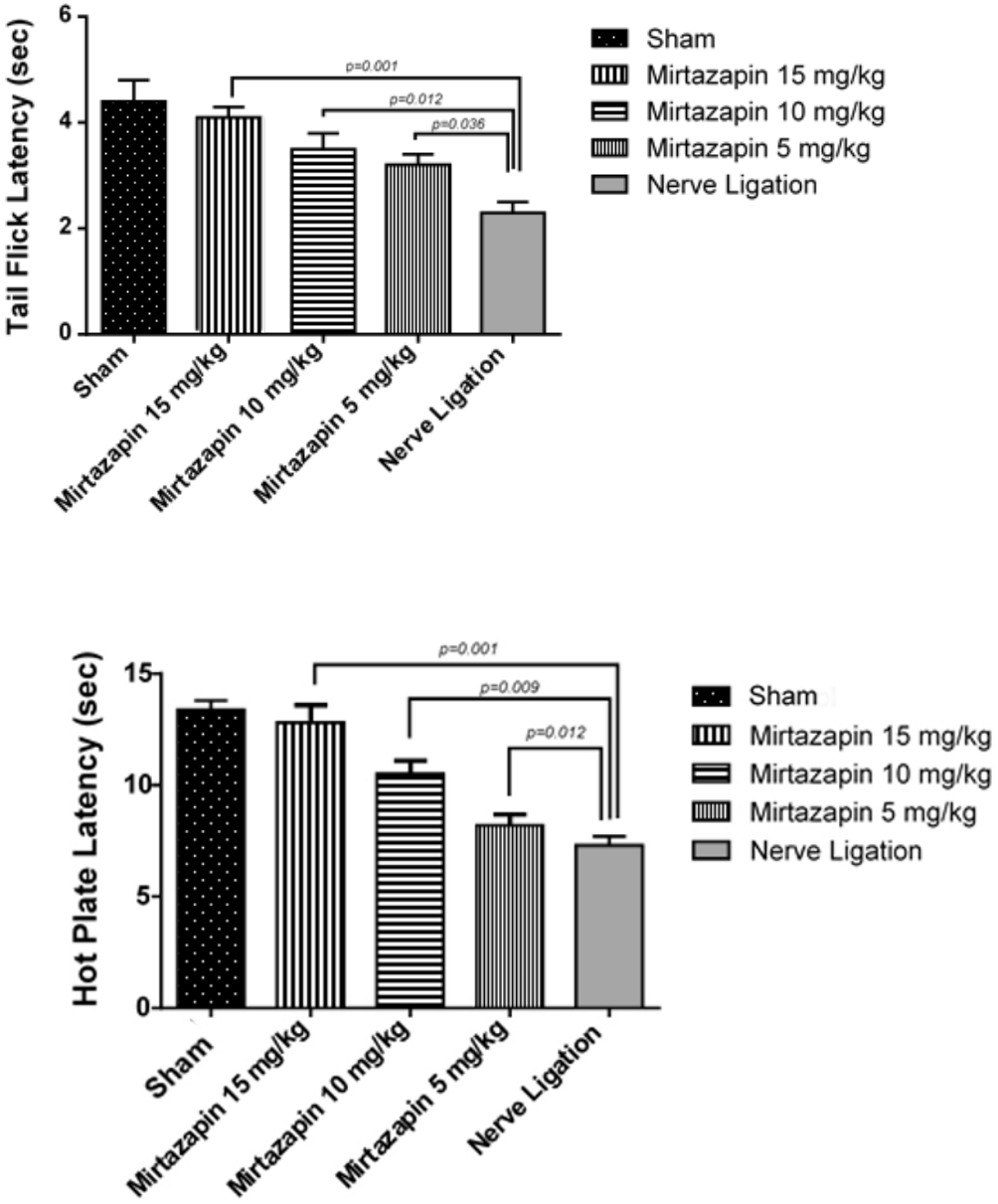

Figure 2. Tail flick (A) and hot plate (B) latencies of sham and nerve ligation and experiment 1, 2 and 3 groups

morphine has been evaluated on all study groups. Morphine (3 mg/kg) created a time-dependent strong antinociceptive effect. The same dose could not manage to create the same amount of analgesia in nerve ligation group. In experiment 1, 2 and 3 groups, administration of mirtazapine also prevented this attenuation and restored both tail flick and hot plate responses (Figure 3).

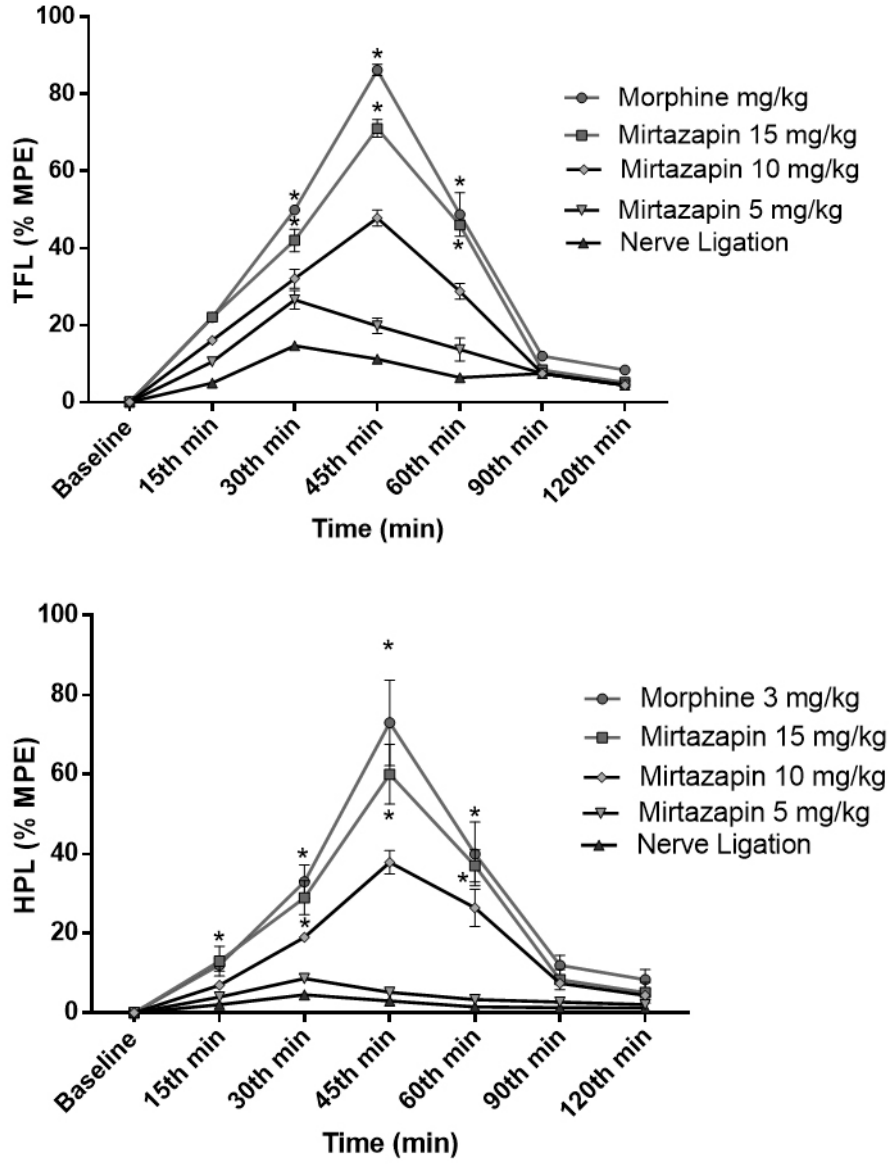

Figure 3. Tail flick (A) and hot plate (B) latencies of 3 mg/kg morphine in the presence and absence of mirtazapine.

DISCUSSION

Neuropathic pain is defined as pain secondary to a lesion or disease process involving the somatosensory system. It can be secondary to a variety of central processes, such as post-stroke thalamic pain or peripheral disorders, such as diabetic neuropathy. Recent reports on NP have suggested its prevalence in the general population ranging from 5 to 8%.[15,16] This pain is considered particularly difficult to treat. Numerous therapeutic recommendations for NP have been proposed over the past decade.[17] While nonsteroidal anti-inflammatory drugs (NSAIDs) and opioids are considered principal medications for somatic pain, adjuvant medications, such as

antidepressants, anti-epileptics, anesthetics, and adrenergic agents, are often useful.[15] In certain patients with moderate to severe NP, opiate agents, such as tramadol, methadone or morphine may also be appropriate considerations.[18] Nevertheless, none of the treatment options could achieve a considerable amount of success probably due to the complexity of the underlying process. With that regard, antidepressants are getting special attention because of their established association with pain pathways and neurotransmitters, which play an important role in pain transmission and perception.[19] Mirtazapine, which is a new generation antidepressant, seems to be a preferable in NP since it affects both noradrenaline and serotonin and has a strong analgesic effect along with a low side effect profile. Having said that, there is limited information regarding the analgesic effect of mirtazapine especially on NP. In a study conducted by Sikka et al.,[20] the analgesic effects of mirtazapine have been evaluated in mice at the doses of 5 and 7 mg/kg. They have managed to show the analgesic effect of mirtazapine but only at high doses. This study is consistent with our study in many ways. In the present study, we have shown that, both in control and nerve ligation groups, mirtazapine has analgesic effects starting from 5 mg/kg. In another study in which the analgesic effect of mirtazapine (from 1 mg/kg to 12.5 mg/kg) was evaluated, the authors have shown an analgesic effect of mirtazapine especially starting from 10 mg/kg which is also consistent with our study. The only small difference that mirtazapine has shown its antinociceptive effect at lower doses, may be related to experiment protocols. The only study close to the current one is the one conducted by McCormick et al..[21] In this study, they conducted a systematic review demonstrating the effect of mirtazapine on phantom-limb pain which also is a kind of NP.

In the present study, administration of mirtazapine during the process of NP development alleviated tail flick and hot plate latencies, which implicate a positive effect. Furthermore, we administrated mirtazapine after the development of neuropathic pain, creating a robust antinociceptive effect. These findings show that mirtazapine has analgesic effects along with its antidepressant effect, and it can both prevent and treat NP.

In the present study, we not only presented the direct effect of mirtazapine on NP but also tried to show the analgesic effect of morphine in nerve ligated rats pretreated with mirtazapine to investigate whether mirtazapine can increase the efficacy of classical opioid analgesics. The findings demonstrated that mirtazapine might modulate neurotransmitters, which play an important role in the regulation of nociception at the spinal level.

As a result, neuropathic pain, which is a chronic pain, can seriously affect the quality of life of patients and cause problems such as depression, anxiety and sleep disturbance. When evaluated from this point of view, mirtazapine is seen as both a good option in neuropathic pain treatment and a preference for the comorbid conditions mentioned.

Authors' contributions: *Ş.F.G.: finding the subject, planning, conducting the study, statistics, writing articles; B.Ç: planning, conducting the study, article writing, field literature review.*

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