ANALGESIC EFFECT OF THYMOQUINONE IN RATS WITH EXPERIMENTAL MIGRAINE MODEL AND ITS RELATIONSHIP WITH NEUROGLIAL ACTIVITY AND LEARNING-MEMORY

SUMMARY

Migraine is a neurovascular disorder characterized by unilateral and throbbing headache with recurrent attacks. Headache can be accompanied by cognitive impairments such as attention deficit along with many symptoms. Thymoquinone is the main bioactive ingredient of black cumin and has been used as an herbal remedy for centuries. Recent studies have shown that thymoquinone is a compound with anticancer, antimicrobial, anti-inflammatory, antihistamine, antidiabetic, gastroprotective, hepatoprotective, cardioprotective, and nephroprotective effects. In addition to all these effects, it is also reported to have antinociceptive, neuroprotective and antioxidant activities. In our study, we aimed to investigate the effects of thymoquinone on pain, learning-memory and neuroglial activation in rat model of migraine.

In the study, 27 adults male *Wistar albino* type rats aged 3 months, weighing between 250-350 gram, were used. The migraine model was induced using nitroglycerin (NTG). NTG was prepared by diluting with isotonic saline to be 10 mg/kg/1 ml for each animal. There were 4 groups as; control (K) group (n=6), nitroglycerin (NTG) group (n=7), thymoquinone (TQ) group (n=7) and nitroglycerin and thymoquinone (NTG+TQ) applied group (n=7). The K group and the NTG group were administered 1 ml of corn oil with gastric gavage for 15 days. TQ and NTG+TQ groups were treated with gastric gavage of 10 mg/kg/1 ml of TQ dissolved in corn oil for 15 days. NTG and NTG+TQ groups were administered 10 mg/kg NTG intraperitoneal (i.p.) on days 1, 5, 10 and 15^{th}. K and TQ groups were administered with 0.3 ml saline i.p. on the same days. To determine the pain threshold of the animals, tail flick test was performed just before gastric gavage and 2 hours after i.p. injection administration. On the 16^{th} day of the experiment, the Morris water maze test was administered to assess learning-memory functions. On the 22^{nd} day of the experiment, the animals were decapitated under general anesthesia with 10 mg/kg xylazine i.p. and 100 mg/kg ketamine i.p. and their brains were removed. The hippocampal region of a hemisphere of the brain was used in biochemical analyses to evaluate markers of learning-memory and oxidative stress. The hippocampal region of the other hemisphere of the brain was used in immunohistochemical studies to evaluate neuroglial activation. The data were evaluated by Benforroni's multiple comparison test by applying one-way analysis of variance and the values were given as standard deviation ±. $P<0.05$ value was considered statistically significant.

In the Morris water maze test, thymoquinone significantly reduced the animals' latency to find the hidden platform in the TQ (***$p<0.001$) and NTG+TQ (*$p<0.05$) groups. There was a significant increase (**$p<0.01$) in the latency of the NTG group to find the platform. When BDNF levels in hippocampus tissue were compared to the

K group, it was determined that it was lower in the NTG group. When BDNF levels were compared according to the NTG group, it was found that both TQ and NTG+TQ groups increased, but this increment was only significant in the TQ group (**p<0.01). CREB1 levels were found decreased in the NTG and NTG+TQ groups compared to the K group. But it was statistically significant (*p<0.05) only in the NTG group. When the groups were compared to the NTG group, there was a significant increase in both the TQ (***p<0.001) and NTG+TQ (**p<0.01) groups. SOD levels were evaluated in hippocampus tissue, and it was determined that there was a statistically significant increase (***p<0.001) in the TQ group compared to all other groups. CAT levels similarly increased significantly in the TQ group compared to all other groups (*p<0.05, ***p<0.001). When immunohistochemical staining was evaluated, it was seen that the number of GFAP positive cells increased statistically significantly in all three of the NTG, TQ and NTG+TQ groups compared to the K group (***p<0.001, **p<0.01, *p<0.05, respectively). In the tail flick test, it was found that there was no statistically significant difference within the group and between the groups.

When the results of our study were evaluated, it was seen that the application of thymoquinone in the experimental migraine model; It has been concluded that it does not have an analgesic effect on migraine pain, contributes to the spatial learning and memory mechanism by increasing BDNF and CREB1 levels, has a positive effect on migraine-related oxidative stress by increasing SOD and CAT levels, may have a positive effect on migraine and oxidative stress associated with learning-memory deficit with astrocyte activation, and may have a protective effect on migraine-related astroglial activation.

Keywords: Migraine, Hippocampus, Thymoquinone, Pain, Learning, Memory, Oxidative Stress, Neuroglial Activity, Astrocyte