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Study the Analgesic Activity of Nigella Sativa L. Volatile Oil Against Pain in Mice

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ABSTRACT

Objective: The black seed, Nigella sativa (NS) is a member of the family Ranunculaceae Known commonly as (black cumin). Recently many biological activities have been reported. The present study was carried out to evaluate the in vivo analgesic activity of the seeds volatile oil.

Materials & Methods: The extract was prepared by 50 gram of seeds powder was extracted in soxhlet apparatus. The extract was concentrated under reduced pressure. The concentrated extract was distilled. The distillate was dried over magnesium sulphate. We got 0.5 ml of yellow volatile oil. Writhing test was used as analgesic test by injected each mouse with (0.2 ml) acetic acid (7 %) intraperitonially and we have been measured the number of writhing for 20 minutes to see the result. Aspirin (acetyl salicylic acid) was used as standard drug.

Results: The results shows that volatile oil of Nigella sativa and may be its constituent, thymoquinone, have a good significant effect against pain $p < 0.05$, $p < 0.01$ and $p < 0.001$ to the doses 0.05 ml, 0.1 ml and 0.2 ml respectively in dependent dose.

Conclusion: The volatile oil of Nigella sativa could be considered as a good analgesic activity.

Keywords: analgesic, Nigella sativa, thymoquinone, volatile oil.

1. INTRODUCTION

The black seed, Nigella sativa (NS) is a member of the family Ranunculaceae Known commonly as black cumin (Nickavar B. et al., 2003; Hanafy M.S. and Hatem M.E., 1991). It's an herbaceous plant that grows in Middle East countries (Musa D. et al., 2004). It contain more than 30% of fixed oil and 0.4 – 0.45 % wt/wt of volatile oil. The volatile oil contains 18.4 – 24% thymoquinone (TQ) and 46% many monoterpenes such as p-cymene and α– piene (Nickavar B. et al., 2003). Black cumin seeds can be used as condiment in bread and other food, as additive for spice and flavored and aromatic substances. Recently many biological activities of Nigella sativa seeds (NSE) have been reported, including antifungal, antibacterial antiviral and antihelmintic ones (Hanafy M. S. and Hatem M. E., 1991; Morsi N. M., 2000). Other have reported that the seeds are used for treatment of flatulence and abdominal ailments , decrease fasting plasma glucose concentration in rabbit , increase serum total protein , as diuretic , hepatoprotective and hypotensive (Musa D. et al., 2004) The volatile oil has antioxidant properties (Burits M. and Bucar F., 2000). Antitumor activity of NSE has been recorded by many authors (Salomi N. J. et al., 1992).

In this study we investigated the effect of volatile oil on pain in mice. Pain is defined as an unpleasant sensation that can be either acute or chronic and that is a consequence of complex neurochemical processes in the peripheral and central nervous system. The majority of tissues and organs are innervated by special sensory receptors (nociceptors) connected to primary afferent nerve fibres of different diameters. Small myelinated, as fibres and unmyelinated C fibres are believed to be responsible for the transmission of painful stimuli. These afferent primary fibres terminate in the dorsal horn of the spinal grey matter (Tuboly G. et al., 2009; Park T. J. et al., 2008; Indo Y., 2010; Stone L. S. and Molliver D. C., 2009). Pain transmission onward is far more complex and understood less well.

The most important parts of this process are the wide dynamic range cells that project to the thalamus and beyond in the spinothalamic tract (Serpell M. G. et al., 1998; Rivera-Arconada I. et al., 2009). Modulation or inhibition also occurs at the level of the spinal cord. This process can be activated by stress or certain analgesic drugs. When the pain modulation system is active, noxious stimuli produce less activity in the pain transmission pathway. Various neurotransmitters found in the dorsal horn of the spinal cord may be involved in pain modulation. These include amino acids such as glutamate and γ-amino butyric acid (GABA), monoamines such as noradrenalin and 5–hydroxytryptamine (5HT) and certain peptide molecules of which the upload peptides are the most important (Fields H. L., 2007; Kidd B. L. and Urban L. A., 2001; Guyton A. C. and Hall J. E., 1996).

2. MATERIALS AND METHODS

Plant material and extraction procedure:

The black seeds were purchased from a local herb store and classified according to the family and species. The seeds where finely powdered in a mixer and 50 g of seeds powders were extracted with (300 ml) hexane for 5 hours and a half in soxhlet apparatus on a temperature of 70 ^{o}C. The extract was concentrated under reduced pressure in rotary evaporator to the 1/8 of it volume. Then tokens 25 ml of concentrated extract (which contain hexane) were distilled for 1 hour. The distillate was dried over magnesium sulphate (Kanter M. et al., 2005). We got (0.5 ml) of yellow volatile oil.

Analgesic test (the writhing test)

In this study, we have been used Albino mice which they weighed (20 – 25 g) and aged (2 months). They were housed in an animal house with temperature (21 – 25 ^{o}C) and excess of food and water. Each group contains 6 mice. As the volume of extracted oil was (0.5 ml) yield (1 %), the volatile oil diluted by adding (5 ml) of olive oil, stomach tube was used to injection the groups (n= 6) (0.05 ml), (0.1 ml) and (0.2 ml) given orally. After 1 hour, we injected each mouse with (0.2 ml) acetic acid (7 %) intraperitonially and we have been measured the number of writhing for 20 minutes to see the result. Aspirin (acetyl salicylic acid) was used as standard drug 50 mg/kg/p.o. Control group resaved 0.1 ml olive oil by oral only (Alam M. A. et al., 2011; Shanmugasundaram P. and Venkataraman S., 2005).

Statistical analysis

All data are expressed as means ± SD. The data were analyzed using the repeated measures of variance (ANOVA) Student t-test; differences were considered significant when $p < 0.05$, $p < 0.01$ and $p < 0.001$ (Giorgi R. et al., 1998).

3. RESULT:

The volatile oil of Nigella sativa and may be its constituent, thymoquinone, has a good effect against pain caused by 7% acetic acid, so it could be considered as a good analgesic. The results as show in table 1 and figure 1. The volatile oil as well as aspirin gives significant ($p< 0.001$) to reduce the number of writhing and then reduce the pain in dependent dose. The pain considers induced by chemical effect (chemical method).

Sample	Control	Aspirin	Volatile oil (VO)		
			0.05 ml	0.1 ml	0.2 ml
Number of Writhing	30±2	8^{***}±3.1	19^{*}±3.3	15^{**} ± 2.4	12^{***}±1.6
Statistical analysis	$^{*} = p < 0.05$		$^{**} = p < 0.01$		$^{***} = p < 0.001$

Table 1. The results of writhing test of volatile oil.

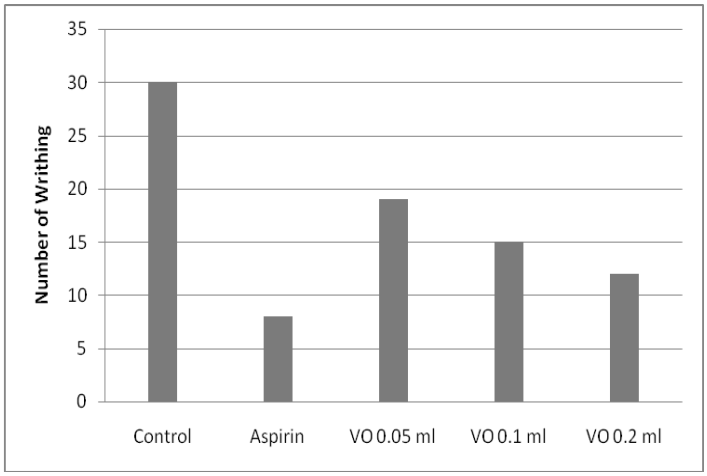

Figure 1. Result of the writhing test of volatile oil.

4. DISCUSSION

Nigella sativa seeds are orally ingested by people as condiment or additive in food dishes. Patients with gastrointestinal disorders ingest seeds mixed with honey. Our study was undertaken to demonstrate the effect of volatile oil against pain.

In the extraction procedure, we crushed and powdered the black seeds in order to increase the surface area of them and to allow to the volatile oil to liberate and secrete completely from the seeds during extraction. The extraction procedure should be done directly after powdering the black seeds to prevent losing of the most liberating active constituents (volatile oil).

After distillation process, we dried the resulted volatile oil by using of magnesium sulphate to get rid of any moisture found within the oil, because the presence of water with the oil may cause chemical changes to the oil and it may loss its properties.

We gave the mice firstly the oil then after 1 hour, we injected them with 7% acetic acid (which causes the required pain), in order to allow good absorption and distribution of the oil inside mice's body and so they will be ready to act against injected acetic acid. We have been waiting for 20 minutes from the acetic acid injection to count the resulted contraction of mice. Usually the ill mouse contracts for about 30 contractions in 20 minutes, If less than 30 contractions after giving the specific drug, it means that the drug is good acting. Which means the volatile oil of Nigella sativa is very good acting as analgesic against pain (Shafiee A. et al., 2003).

We have seen in our study that the female mouse had more defence mechanism against acetic acid than the male mouse As a result, the volatile oil of Nigella sativa and may be its constituent, thymoquinone, can reduce the pain and may be used as an analgesic drug. The chemical structure of thymoquinone shows in figure 2.

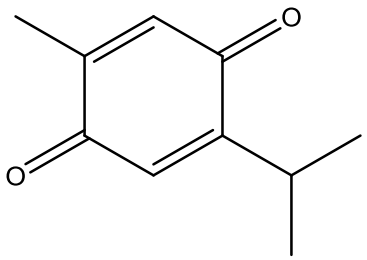

Figure 2. The chemical structure of thymoquinone.

5. CONCLUSION

The active constituent of the volatile oil may be thymoquinone, has an analgesic effect to reduce pain, the volatile oil was dose dependent, but we should give a small and accurate dose of it to prevent toxicity.

6. REFERENCES

Alam MA, Jahan R, Rahman S, Das AK, Rahmatullah M. Antinociceptive and anti-hyperglycemic activity of methanol leaf extract of Cyperus scariosus. Pak J. Pharm. Sci. 2011; 24(1): 53-56.

Burits M, Bucar F. Antioxidant activity of Nigella sativa essential oil. Phytotherapy Res. 2000; 14: 323-328.

Fields HL. Understanding How Opioids Contribute to Reward and Analgesia. Regional Anesthesia and Pain Medicine. 2007; 32(3): 242–246.

Giorgi R, Pagano RL, Amorim Dias MA, Aguiar-Passeti T, Sorg C, Mariano M. Antinociceptive effect of the calcium-binding protein MRP-14 and the role played by neutrophils on the control of inflammatory pain. Journal of Leukocyte Biology. 1998; 64: 214-220.

Guyton AC, Hall JE. Textbook of Medical Physiology. 9th ed. W B sanders company, Philadelphia 1996.

Hanafy MS, Hatem ME. Studies on the antimicrobial activity of Nigella sativa seed (Black cumin). J Ethnopharmacol. 1991; 34: 275-278.

Indo Y. Nerve growth factor, pain, itch and inflammation: lessons from congenital insensitivity to pain with anhidrosis. Expert Review of Neurotherapeutics. 2010; 10(11): 1707-1724.

Kanter M, Demir H, Karakaya C, Ozbek H. Gastroprotective activity of Nigella sativa oil and its constituent, thymoquinone against acute alcohol– induced gastric mucosal injury in rats. World J Gastroenterol. 2005; 11(42): 6662–6666.

Kidd BL, Urban LA. Mechanisms of inflammatory pain. Br J Anaesth. 2001; 87(1): 3-11.

Morsi NM. Antimicrobial effect of crude extracts of Nigella sativa on multiple antibiotics resistant bacteria. Acta Microbiol Polonica. 2000; 49: 63-67.

Musa D, Dilsiz N, Gumushan H, Ulakoglu G, Bitiren M. Antitumor activity of an ethanol extract of Nigella sativa seeds. Biologia Bratislava. 2004; 59(6): 735–740.

Nickavar B, Mojab F, Javidnia K, Amoli MA. Chemical composition of the fixed and volatile oils of Nigella sativa L. from Iran. Z. Naturforsch. 2003; 58: 629–631.

Park TJ, Lu Y, Juttner R, Smith EJ, Hu J, Brand A, Wetzel C, Milenkovic N, Erdmann B, Heppenstall PA, Laurito CE, Wilson SP, Lewin GR. Selective Inflammatory Pain Insensitivity in the African Naked Mole-Rat (Heterocephalus glaber). Plos Biol. 2008; 6(1): 156-170.

Rivera-Arconada, I, Roza, C, Lopez-Garcia, JA. Enhancing M currents a way out for neuropathic pain. Frontiers in Molecular Neuroscience. 2009; 2: 3389-3399.

Salomi NJ, Nair SC, Jayawardhanan, KK, Verghese CD, Panikkar, KR. Antitumor principles from Nigella sativa seeds. Cancer Lett. 1992; 63: 41-46.

Serpell MG, Makin A, Harvey A. Acute pain physiology and pharmacological targets the present and future. Acute Pain. 1998; 1(3): 31-47.

Shafiee A, Bagheri M, Shekarchi M, Abdollahi M. The antinociceptive activities of 1-(4-aryl-2-thiazolyl)-3,5-disubstituted-2 pyrazolines in mouse writhing test. J Pharm Pharmaceut Sci. 2003; 6(3): 360-362.

Shanmugasundaram P, Venkataraman S. Anti-nociceptive activity of Hygrophila auriculata (Schum) heine. Afr. J. Trad CAM. 2005; 2(1): 62-69.

Stone LS, Molliver DC. In Search of Analgesia: Emerging Poles of GPCRs in Pain. Mol Interv. 2009; 9(5): 234–251.

Tuboly G, Mecs L, Benedek G, Horvath G. Antinociceptive interactions between anandamide and endomorphin-1 at the spinal level. Clin Exp Pharmacol Physiol. 2009; 36(4): 400-405.