Antinociceptive effect of essential oil from the leaves of *Zanthoxylum piperitum* (L.) DC. (Rutaceae).

Nikki Wong[1], Graciela R. Donald[2], Fabio Boylan[1], Patrícia D. Fernandes[2]

[1] Trinity College Dublin, Ireland
[2] Federal University of Rio de Janeiro, Institute of Biomedical Science, Laboratory of Pharmacology of Pain and Inflammation. Rio de Janeiro, Brazil
wongn@tcd.ie

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The species *Zanthoxylum piperitum* (ZP) has been traditionally used in Asia for treating the symptoms of diarrhoea and abdominal pain. Previous studies reported the antibacterial and anti-inflammatory effects of ZP, and chemical studies previously published showed that ZP essential oil (ZPEO) contains monoterpenes, such as phellandrene, as its main component although some authors have shown that ZPEO components can vary according to the season. Despite the traditional use for pain, there is no pharmacological study reporting this property for this species. Therefore, the aim of this study was to evaluate the possible antinociceptive and/or anti-inflammatory properties of ZPEO. Fresh leaves (150 g) of ZP were collected in Dublin May 2015 and they were exposed to hydrodistillation in a Clevenger-type apparatus for 2 hours. The oil was analyzed by gas chromatography and its components were identified by comparison of both mass spectra and linear retention indices with spectral library and literature. ZPEO was given (p. o.) to Swiss Webster mice (25-30 g) at doses of 10 µl, 30 µl or 100 µl/kg, and these mice were investigated using the formalin test, which consists of injecting formaldehyde (2.5%) into the hind paw (plantar). The time that each animal spent licking its hind paw was recorded. The response to formalin (paw licking) involves an early phase (0-5 min after injection) and a late phase (15-30 min after injection). The early phase is known to involve the C-fibre suggesting a peripheral pain stimulus, whereas the late phase seems to engage in an inflammatory process and affects the activity in the dorsal horn of the spinal cord. Acetylsalicylic acid (ASA) and morphine were used as reference drugs. Both drugs show significant effect in both phases, however morphine has a stronger effect in the first phase whereas ASA is more potent in the second phase. All ZPEO doses tested presented significant inhibition in the first phase of the formalin test with a reduction rate of 36, 41 and 49% of the licking time, respectively, when compared to the vehicle group (cooking soybean oil). No significant effect was observed in the second phase for ZPEO. The drugs morphine and ASA presented a 54% and 36% licking inhibition in the first phase and 39% and 58% in the second phase, respectively. Chemical analysis of ZPEO showed the presence of 29 compounds, among them 14 compounds have already been reported as having some antinociceptive effects. The major components of ZPEO are beta-phellandrene (29.39%), (*E,E*)-farnesyl acetate (14.55%), beta-citronellol (10.32%), alpha-pinene (9.75%) and beta-citronellal (6.83%). The fact that ZPEO was only able to inhibit licking in the first phase suggests that its components have effect on peripheral pain but they were inactive against inflammatory pain. A literature survey shows that the essential oil of ZP has been previously reported to being used as an anti-inflammatory treatment, however when comparing the chemical composition of the essential oil obtained in this current study with that reported in the literature, they have distinctly different composition with few common compounds.

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