EFFECTS OF BERGAMOT ESSENTIAL OIL BERGAPTENE-FREE IN *IN VIVO* EXPERIMENTAL MODELS OF INFLAMMATION AND PAIN

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Introduction: The renewed interest in natural products as potential source of drugs, led us to investigate on both the anti-inflammatory and antinociceptive activity of bergamot essential oil (BEO). Citrus bergamia Risso & Poiteau (bergamot) is a small tree belonging to the Rutaceae family, endemic of the southern coasts of the Calabria region (Italy), whose fruit is mostly used to retrieve its essential oil, commonly employed in perfume industry and in aromatherapy. Since the toxicity of bergapten is well known, our study has been performed using the BEO fraction deprived of bergapten (BEO-BF).

Materials and methods: The antioxidant capability of BEO-BF was assessed by the 2,2-diphenyl-1-picrylhydrazyl (DPPH) test, the ferric reducing power and the chelating activity assays, while carrageenan-induced oedema in rats was used as an *in vivo* model of inflammation. To gain a better insight into BEO-BF's mechanism(s) of action, we evaluated some mediators of the early phase of inflammation, such as interleukin (IL)-6, IL-1β and tumor necrosis factor (TNF)-α, in the paw homogenates by enzyme-linked immunosorbent assay (ELISA) technique. In addition, nitrite/nitrate levels and prostaglandin E_2(PGE_2) content in paw exudates were measured respectively by Griess reaction and ELISA assay. Moreover, histological and immunohistochemical analysis of paws biopsies were performed. The antinociceptive activity of BEO-BF was examined in two different pain models in mice: the writhing test, a model of inflammatory pain, and the hot plate test, a model of supraspinal response.

Results: BEO-BF possesses antioxidant properties, as determined by cell-free assays. As assessed by plethysmometer, treatment with BEO-BF (86,24mg/Kg and 431,2mg/Kg) led to a significant inhibition of paw oedema induced by a subplantar injection of carrageenan. Moreover, histological examination of paw biopsies showed a reduction of pathological changes typical of oedema in BEO-BF treated rats. Pre-treatment with 100 μl (86,24mg/kg) of BEO-BF led to a significant reduction of TNF-α, IL-6and IL-1β levels in the paw homogenate, compared to control. Furthermore, in BEO-BF-treated rats we observed a reduction of nitrite/nitrate and PGE_2content in exudates. Results of the writhing test showed that BEO-BF elicited a pronounced analgesic response, as demonstrated by a significant inhibition of constrictions in mice receiving acetic acid, with respect to control animals. On the other hand, administration of BEO-BF did not produce any significant analgesic effect in the hot-plate test.

Discussion and conclusion: In the continuous search for new bioactive natural products against inflammation, essential oils are increasingly being referred to as a rich source of such products. According to our results, this study indicates that BEO exerts protective effects in carrageenan-induced paw oedema, as well as inhibits both inflammatory and central pain *in vivo*, suggesting its potential role as anti-inflammatory drug.