Natural Product Research: Formerly Natural Product Letters

Publication details, including instructions for authors and subscription information:
http://www.tandfonline.com/loi/gnpl20

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Version of record first published: 04 Sep 2012

To cite this article: Carlos A. Manassero, Juan R. Girotti, Sergio Mijailovsky, Margarita García de Bravo & Mónica Polo (2012): In vitro comparative analysis of antiproliferative activity of essential oil from mandarin peel and its principal component limonene, Natural Product Research: Formerly Natural Product Letters, DOI:10.1080/14786419.2012.718775

To link to this article: http://dx.doi.org/10.1080/14786419.2012.718775

Natural Product Research
2012, 1–4, iFirst

SHORT COMMUNICATION

In vitro comparative analysis of antiproliferative activity of essential oil from mandarin peel and its principal component limonene

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(*Received 20 December 2011; final version received 3 July 2012*)

The effects of the essential oil of mandarin peel (Corrientes, Argentina) and limonene (its major component) were studied on two human tumour cell lines growth (lung adenocarcinoma A549 and hepatocarcinoma HepG2). The essential oil was obtained by cold press and its composition was investigated by gas chromatography (GC) and GC/mass spectrometry (MS) analysis. The antiproliferative effect was studied using an MTT assay. Both mandarin essential oil and limonene tested showed a strong dose-dependent effect on the growth inhibition of these cell lines. The essential oil was more effective in A549 than in HepG2 cells and more effective than limonene in both the cases. It is likely that minor components and limonene of the oil could exert additive or synergistic effects. Hence, mandarin essential oil could lead to the development of antitumour agent or complementary and alternative medicines for the treatment of diverse cancers.

Keywords: mandarin essential oil; limonene; HepG2; A549

1. Introduction

Essential oils are valuable natural products used as raw materials in many fields, including perfumes, cosmetics, aromatherapy, phytotherapy, spices and nutrition (Buchbauer, 2000). Their potency for treating different pathologies of relevant social impact such as cancer, allergy and diabetes has been reported (Esmonde & Long, 2008; Heinrich & Bremner, 2006; Miller, Binns, & Brickman, 2008). Cytotoxicity has been reported for many essential oils (de Sousa et al., 2004; Lampronti, Saab, & Gambari, 2006; Prashar, Locke, & Evans, 2006). In particular, citrus essential oils have a wide spectrum of biological activities such as antimicrobial (Fisher & Phillips, 2008), antioxidant and anti-inflammatory (Sood et al., 2009) and limonene, a major constituent in several citrus oils (orange, lemon, mandarin, lime and grapefruit), has well-established chemopreventive activity against many types of cancers (Sun, 2007).

The aim of this investigation was to study the effects of the essential oil of mandarin peel (Corrientes, Argentina) on two human tumour cell lines.

The obtained essential oil was investigated by gas chromatography (GC) and GC/mass spectrometry (MS) analysis. The essential oil of mandarin and limonene, which was the

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ISSN 1478–6419 print/ISSN 1478–6427 online

http://dx.doi.org/10.1080/14786419.2012.718775
http://www.tandfonline.com

major component of the oil (>88%), were tested for their antiproliferative activity against human lung adenocarcinoma A549 and human hepatocarcinoma HepG2 because cancers of the respiratory and digestive system continue to be the most common fatal cancers (Jemal, Siegel, Xu, & Ward, 2010).

2. Results and discussion

Limonene was the predominant component of our mandarin essential oil (MO), accounting for over 88% (Figure S1). This was in concordance with many studies on the chemical composition of mandarin oil, as previously reported (Sawamura, Minh Tu, Onishi, Ogawa, & Choi, 2004). Minor components of the volatile organic compounds like linalool, *n*-decanal, perilla aldehyde, citronellol and dodecanal are shown in Figure S2 and Table S1. Mandarin essential oils have been widely used in traditional Chinese medicines for a long time because of their pharmacological activity, low toxicity and costs (Yu, Li, Liu, Xu, & Liang, 2009). The cytotoxic activity of MO and pure limonene were tested *in vitro* against A549 and HepG2 cells. The two tested compounds showed a strong dose-dependent effect on the growth inhibition of these cell lines. Limonene exhibited half-maximal inhibitory concentration (IC_{50}) of 0.150 μL mL^{-1} (0.889 mM) in HepG2 cells and 0.098 μL mL^{-1} (0.586 mM) in A549 cells (Figure 1b and d, respectively). IC_{50} values for limonene were in accordance to data obtained with other cell lines (He, Mo, Hadisusilo, Qureshi, & Elson, 1997; Holstein & Hohl, 2003). In contrast, the crude essential oil showed

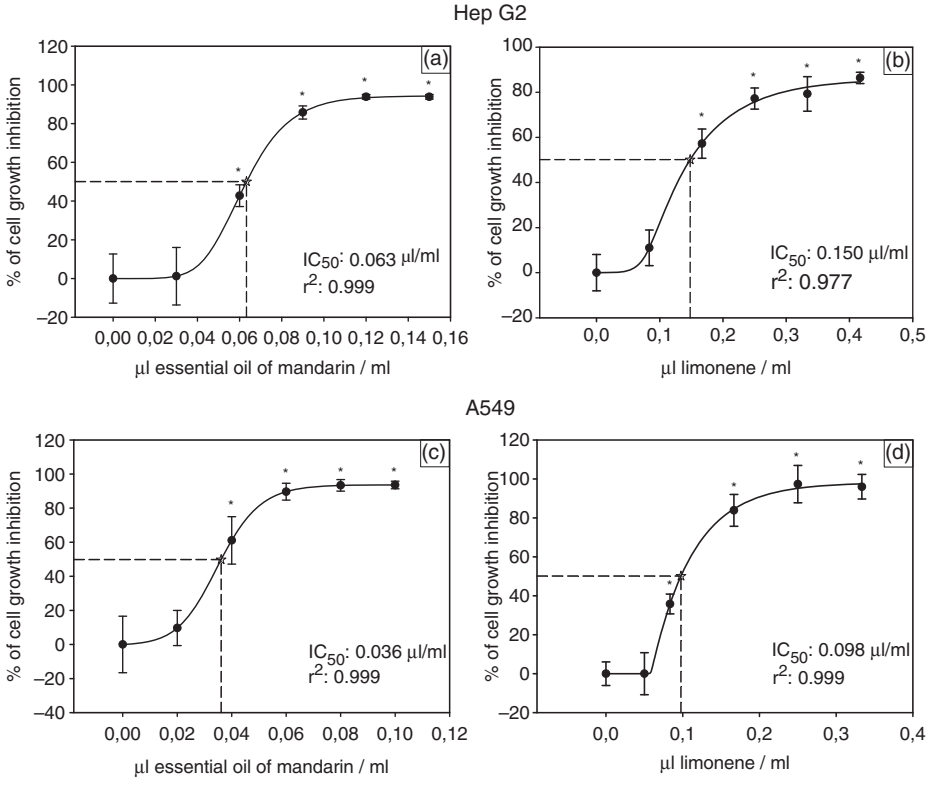

Figure 1. Cytotoxic activity of the mandarin essential oil (MO) and limonene (LI) against HepG2 (a and b) and A549 (c and d) cell lines.

an IC_{50} of 0.063 μL mL^{-1} (over 0.329 mM LI) and 0.036 μL mL^{-1} (over 0.188 mM LI), respectively (Figure 1a and c). The essential oil was then more effective in A549 than in HepG2 cells and more effective than limonene in both cases. It is likely that minor components and limonene of MO could exert additive or synergistic effects on cell proliferation. Itani et al. (2008) described that the combination of three bioactive components of Lebanese sage (*Salvia libanotica*) caused synergistic inhibition on the growth of two human colon cancer cell lines. We have already described that the combination of two monoterpenes (cineole and linalool) inhibited synergistically the proliferation of A549 and HepG2 cells (Rodenak Kladniew, Manassero, Polo, & García de Bravo, 2010).

3. Conclusion

Mandarin essential oil exhibits antiproliferative activity against human hepatocarcinoma (HepG2) and human lung adenocarcinoma (A549) cells; it is more effective than limonene, its principal component. Hence, our results suggest that MO could lead to the development of anti-tumour agent or complementary and alternative medicines for the treatment of diverse cancers.

Supplementary material

Experimental details relating to this article are available online, alongside Table S1 and Figures S1 and S2.

Acknowledgements

This study was supported by research grants from CONICET, ANPCyT and UNLP, Argentina. The authors are grateful to Dr F. Speroni and Dr G. Rinaldi for their help in obtaining the essential oil and to N. Tedesco for language revision.

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