



### The effects of different culture conditions on the production of volatiles from *Lippia origanoides* Kunth (Verbenaceae).

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*Lippia origanoides* is popularly known as "salva-de-marajó", occurring mainly in Brazil, northern South America, Colombia and Venezuela (1). This aromatic plant grows in wild habitats, obtained exclusively by extractivism. This species presents five different chemotypes (A-E), so far described: (A) *p*-cymene,  $\alpha$ - and  $\beta$ -phellandrene and limonene, (B) carvacrol, (C) thymol, (D) 1,8-cineole and (E) (*E*)-methyl cinamate and (*E*)-nerolidol (1,2). *In vitro* culture techniques are an alternative to sustainable production and large-scale species. *L. origanoides* was collected in the Amazon region in order to verify the effects of growth regulators on the chemical composition of the volatiles of the *in vitro* plants, compared to the field material collected at different times. Cultures of this plant were maintained *in vitro* in basic medium (3), either without growth regulators or supplemented with different concentrations of cytokinins (kinetin and 6-benzylaminopurine) and auxin (indolylacetic acid) combined with each other. Volatiles were obtained from 5 g of plant material, by simultaneous distillation and extraction (SDE) for 3 h, and collected in dichloromethane. The volatiles were analyzed by GC/FID and GC/MS in Shimadzu GC-2010 systems, both with DB-5MS fused silica capillary columns (30 m X 0.25 mm X 0.25  $\mu$ m). Hydrogen was used as carrier gas for GC/FID and helium for GC/MS, both with a flow rate of 1.0 mL min<sup>-1</sup>. Oven temperature was raised from 60 to 290 °C at 3°C min<sup>-1</sup>. Mass detector was operated in electronic ionization mode at 70 eV. The percentage composition was obtained by normalization from FID. Volatile components were identified by comparison of both mass spectra and linear retention indices with spectral library and literature (4). The major compounds found in volatile extract of *L. origanoides* were: myrcene (trace-5.1 %), *p*-cymene (5.7-20.0 %),  $\gamma$ -terpinene (1.8-10.6 %), linalool (2.5-4.7 %), thymol (3.0- 9.2%), carvacrol (29.7-45.8 %) and  $\beta$ -caryophyllene (2.5-5.3 %). In this study, the carvacrol content was more pronounced in the wild plants (35.1-45.8 %), compared to the *in vitro* material (29.7-45.5 %). Carvacrol has been associated with the aroma of "oregano". Its high content can be considered economically interesting and important for the acceptance of this species as food for culinary purposes. The results showed similar percentage contents of volatile constituents for plants growing in *in vitro* culture media and field plants.

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