

## Abstract

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**Title of the diploma thesis:** Supercritical fluids for extraction of biologically active compounds from Eucalyptus leaves

Eucalyptus is nowadays used for its anti-inflammatory, antibacterial, and antioxidant effects in the pharmaceutical and cosmetic industries. Eucalyptus contains various compounds including flavonoids, volatile compounds, terpenoid acids, and phenolic acids.

The aim of this work was to develop a two-step selective extraction method for the extraction of compounds of different physicochemical properties from *Corymbia citriodora* leaves using extraction solvent containing CO<sub>2</sub>.

Supercritical CO<sub>2</sub> is a non-polar solvent with polarity and physicochemical properties, that can be changed by mixing with a suitable polar solvent. In this way, a wide range of compounds with different polarities can be extracted.

In this study, the key parameters, including temperature in the range of 40–80 °C, pressure in the range of 100–300 bar were optimized using Box-Behnken design with heptane and ethanol/H<sub>2</sub>O as a cosolvent. After optimization of the key parameters, the extraction kinetics were tested at flow rates of 2 mL/min and 4 mL/min. The final method based on two individual extraction steps was set up as follows: the volatile compounds were extracted for 20 min at a flow rate of 2 mL/min, temperature of 78 °C, pressure of 133 bar and extraction solvent CO<sub>2</sub>/EtOH (98/2, v/v) with 3 % of H<sub>2</sub>O. In the second step the polar compounds were extracted using CO<sub>2</sub>/EtOH (44/56, v/v) with 1 % of H<sub>2</sub>O for 60 min at flow rate 2 mL/min, temperature 80 °C and pressure 108 bar. The repeatability of the method was verified with RSD lower than 26.2 % for volatile compounds and RSD lower than 23.5 % for polar compounds, including flavonoids and phenolic acids. Subsequently, the optimized method was applied to other plant matrices of *Myrtus communis* and *Eucalyptus gunni* to determine the content of volatiles and polar compounds.