The role of HSCCC to separate volatile metabolites from Brazilian native species.

André M. Marques, Maria Auxiliadora C. Kaplan

Instituto de Pesquisas de Produtos Naturais - Universidade Federal do Rio de Janeiro Rio de Janeiro, Brazil.
andrefarmaciarj@yahoo.com.br

Keywords: Separation process, *Piper, Eugenia,* terpenes, HSCCC.

Essential oils (EOs) are hydrophobic liquids, usually extracted by hydrodistillation, containing volatile aromatic compounds. The preparative separation of volatile compounds from these complex mixtures is always a challenge due to their structural similarity, strongly hydrophobic properties and poor stability. High Speed Counter Current Chromatography (HSCCC), a support-free liquid–liquid partition chromatographic technique, is the chromatographic method that permits a wide range polarity phase operation, and a fast running time separation process. It is as saving solvent process useful in the isolation of compounds, with a very limited thermal and chemical stability. In this work, we present the successful separation process strategy of major volatile compounds from native Brazilian plant species in preparative scale using HSCCC. The method uses a solvent system composed by hexane/acetonitrile (1:1) to isolate many terpenes in isocratic elution (normal and reversed modes) in run time average of 3h recovering up to 94 % of the mass applied. In this process, we separate the major constituents of four Brazilian aromatic species: *Piper clausseniánum*, *Manekia obtusa*, *Pectis brevipedunculata* and *Eugenia uniflora*. *P. clausseniánum* showed to produce a very active leaf EO against *Leishmania* species. The HSCCC separation process provided the isolation of the major metabolites nerolidol (75 mg, 93.0 %) and linalool (230 mg, 98.2 %) as well as camphor (46 mg, 97.4 %), bornyl acetate (7 mg, 87.8 %) and camphene (35 mg, 81.8 %). Safrole represents roughly 8-10 % of the crude EO of *M. obtusa*. This compound has an important economic value since it is used as derivative in many chemical synthesis processes. Through HSCCC, we obtained 200 mg of safrole in 95 % of purity in 2 h of running separation process. The EO from the native grass *P. brevipedunculata* has a citric fragrancy due to the presence of citral as its major component. In a 2 h running time, it was possible to separate the isomers pair neral/geranial (780 mg, 98.7 %), as well as geraniol (8 mg, 86.0 %), geranial (12 mg, 91.0 %) and neral (7 mg, 81.0 %) with high percentage mass recovery and at high purity level. *Eugenia uniflora*, a popular Myrtaceae species, is known as Brazilian cherry due to its edible fruits. Using HSCCC it was also possible to isolate in high percentage their major constituents:(-)-oxidoseline-1,3,7-(11)-trien-8-one (375 mg, 97.0 %) and (364 mg, 92.0%) of (-)-seline-1,3,7(11)-trien-8-one in just one separation step. HSCCC is a useful tool to separate terpenes and bioactive non-polar compounds in the essential oils from different Brazilian native plant species.

Acknowledgements: We thank to CAPES and CNPq.

ISBN: 978-85-66836-11-0