World Academy of Science, Engineering and Technology
International Journal of Chemical and Molecular Engineering
Vol:12, No:01, 2018

Approach to Honey Volatiles' Profiling by Gas Chromatography and Mass Spectrometry

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Abstract : Biodiversity of flora provides many different nectar sources for the bees. Unifloral honeys possess distinctive flavours, mainly derived from their nectar sources (characteristic volatile organic components (VOCs)). Specific or nonspecific VOCs (chemical markers) could be used for unifloral honey characterisation as addition to the melissopalynologycal analysis. The main honey volatiles belong, in general, to three principal categories: terpenes, norisoprenoids, and benzene derivatives. Some of these substances have been described as characteristics of the floral source, and other compounds, like several alcohols, branched aldehydes, and furan derivatives, may be related to the microbial purity of honey processing and storage conditions. Selection of the extraction method for the honey volatiles profiling should consider that heating of the honey produce different artefacts and therefore conventional methods of VOCs isolation (such as hydrodistillation) cannot be applied for the honey. Two-way approach for the isolation of the honey VOCs was applied using headspace solid-phase microextraction (HS-SPME) and ultrasonic solvent extraction (USE). The extracts were analysed by gas chromatography and mass spectrometry (GC-MS). HS-SPME (with the fibers of different polarity such as polydimethylsiloxane/ divinylbenzene (PDMS/DVB) or divinylbenzene/carboxene/ polydimethylsiloxane (DVB/CAR/PDMS)) enabled isolation of high volatile headspace VOCs of the honey samples. Among them, some characteristic or specific compounds can be found such as 3,4-dihydro-3-oxoedulan (in Centaurea cyanus L. honey) or 1H-indole, methyl anthranilate, and cis-jasmone (in Citrus unshiu Marc. honey). USE with different solvents (mainly dichloromethane or the mixture pentane : diethyl ether 1 : 2 v/v) enabled isolation of less volatile and semi-volatile VOCs of the honey samples. Characteristic compounds from C. unshiu honey extracts were caffeine, 1H-indole, 1,3-dihydro-2H-indol-2-one, methyl anthranilate, and phenylacetonitrile. Sometimes, the selection of solvent sequence was useful for more complete profiling such as sequence I: pentane → diethyl ether or sequence II: pentane → pentane/diethyl ether (1:2, v/v) → dichloromethane). The extracts with diethyl ether contained hydroquinone and 4-hydroxybenzoic acid as the major compounds, while (E)-4-(r-1’,t-2’,c-4’-trihydroxy-2’,6’,6’-trimethylcyclo-hexyl)but-3-en-2-one predominated in dichloromethane extracts of Allium ursinum L. honey. With this two-way approach, it was possible to obtain a more detailed insight into the honey volatile and semi-volatile compounds and to minimize the risks of compound discrimination due to their partial extraction that is of significant importance for the complete honey profiling and identification of the chemical biomarkers that can complement the pollen analysis.

Keywords : honey chemical biomarkers, honey volatile compounds profiling, headspace solid-phase microextraction (HS-SPME), ultrasonic solvent extraction (USE)
Conference Title : ICFCA 2018 : International Conference on Food Chemistry and Analysis
Conference Location : Amsterdam, Netherlands
Conference Dates : January 22-23, 2018

Open Science Index, Chemical and Molecular Engineering Vol:12, No:01, 2018 publications.waset.org/abstracts/81756.pdf