Supporting Information

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Headspace Analysis of Volatile Compounds Coupled to Chemometrics in Leaves from the Magnoliaceae Family

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S1: Analysis methods

Headspace isolation:

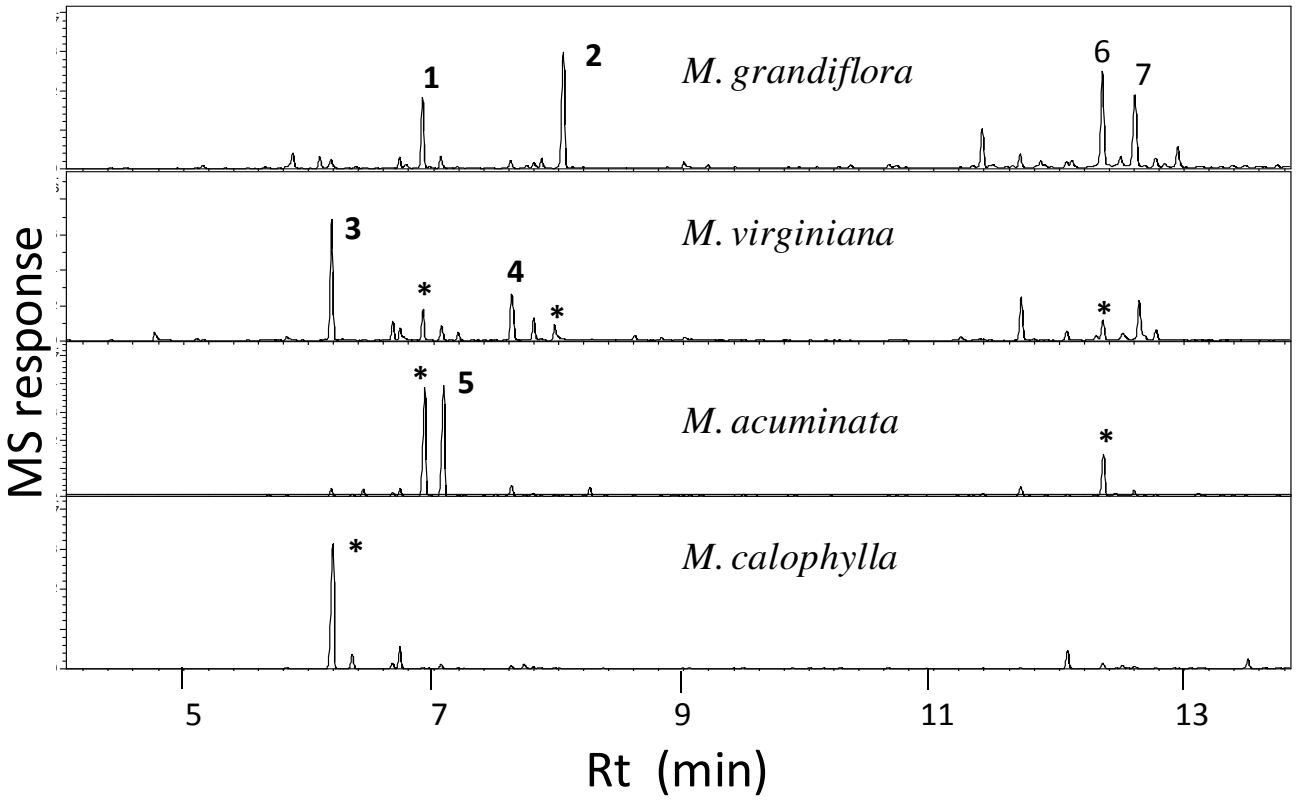
Ten grams of fresh leaves were placed in a side-arm conical flask (250 ml). Charcoal-purified air was passed over the plant from the top and volatiles were collected by pulling vacuum through Tenax (Sigma, St. Louis, MO) traps located at the flask side arm. Volatiles collected on the Tenax adsorbent traps for 8 h were eluted with 500 µL hexane and analyzed by GC/MS & GC/FID. A system blank containing no plant material was run as control.

GC/MS analysis and GC/FID quantification:

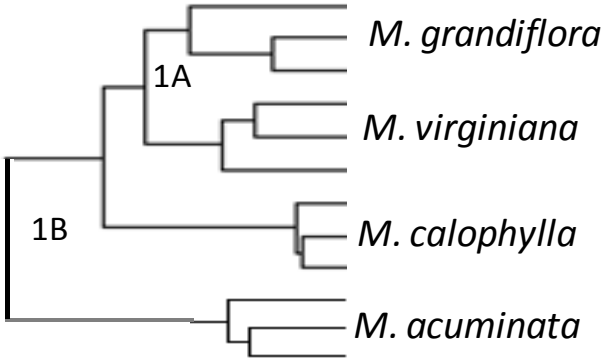
GC/MS analysis was performed on an HP 5890A (Hewlett–Packard, Palo Alto, CA) equipped with an HP-5HS column (Agilent) 30-m x 0.32-mm, 0.25-µm-thick bonded methyl siloxane. Injections were made in the splitless mode for 30 sec, and the gas chromotograph was operated under the following conditions: injector 220 °C, column oven 40 °C for 3 min, then programmed at a rate of 12 °C/min to 180 °C, kept at 180 °C for 5 min and finally ramped at a rate of 40 °C/min to 250 ° C and kept for 2 min, He carrier gas linear flow velocity 50 cm/sec. The transfer line and ion-source temp were adjusted at 230 °C and 190 °C, respectively. The HP quadrupole mass spectrometer was operated in the electron ionization mode at 70 eV and a source temperature of 180 °C. Volatile components were deconvoluted using AMDIS software (www.amdis.net) and identified by its retention indices (relative to n-alkanes), mass spectrum matching to EPA/NIH database and with authentic standards (when available). Similar column type and ramping program was used for GC-FID analysis, with hydrogen used as carrier gas at 2 ml min-1. The relative amounts of the individual volatiles were determined by GC-FID.

HCA and PCA analysis:

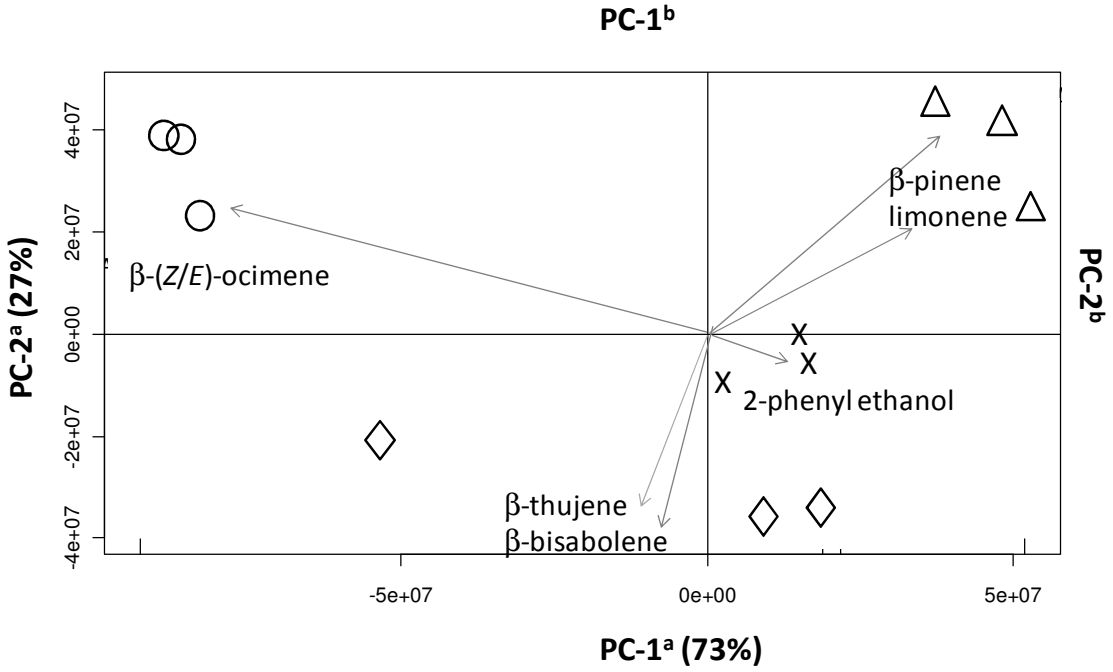
HCA analysis was performed using R packages (Heatplus & PCAmethods) which can be downloaded freely as an R package from the Metlin Metabolite Database (http://137.131.20.83/download/) under R 2.9.2 environment from quantification peak list for a total of 75 volatiles.



S2: GC-MS chromatogram of *M. grandiflora*, *M. virginiana*, *M. acuminata* and *M. calophylla* headspace volatiles with assigned peaks: 1, (*Z*)-β-ocimene; 2, unknown monoterpene; 3, β-pinene; 4, 2-phenyl ethyl alcohol; 5, (*E*)-β-ocimene; 6, germacrene A; 7, β-bisabolene. *, denotes alignment of peaks in other HS extracts.



S3: Hierarchical cluster analysis (HCA) of *Magnolia* species based on group average cluster analysis of volatiles profile as the analytical data, triplicate measurement.



S4: GC/MS based principal component analyses biplot of Magnolia volatile samples. The cluster is denoted as follows: *M. acuminata* (O), *M. calophylla* (Δ), *M. grandiflora* (◊), *M. virginiana* (X), n=3.
[a]Axes refer to scores from the samples.
[b]Axes refer to loadings from volatile constituents (Table 1) with selected variables represented as vectors from the origin. Values in parentheses refer to the variance of each principal component.