Supplementary Materials Data S1

Characteristic differences in essential oil composition of six *Zanthoxylum bungeanum* Maxim. (Rutaceae) cultivars and their biological significance

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Data S1

Materials and methods

The plants had been cultivated for 11 years in the nursery at Northwest A&F University, Yangling, Shaanxi Province, China. Six cultivars of *Z. bungeanum* Maxim, namely Qin'an (I), Dahongpao A-C (II-IV) and Shizitou (VI), were originally introduced from six typical planting areas in China: the city of Qin'an in Gansu Province, the county of She in Hebei Province, and Fuping, Tongchuan, Feng and Hancheng in Shaanxi Province.

Fresh *Z. bungeanum* Maxim fruits were sampled randomly from each population in mid-July 2014. The air-dried pericarp of *Z. bungeanum* Maxim were ground into powder using a grinder (FW-100, Test Instrument Company, Tianjin, China) and passed through 40 mesh sieves. The powder was prepared directly before undertaking the analysis.

Extraction of essential oils

Essential oils were extracted by hydrodistillation using a Clevenger-type apparatus according to the Chinese Pharmacopoeia (PCPRC, 2010) for 3 h from 100 g ground pericarp. Anhydrous sodium sulfate was used to remove water after extraction. Essential oils were stored at 4 °C for analysis.

Chemicals

All chemical reagents were analytical grade and obtained from Tianjin Chemical Factory (Tianjin, China).

Gas chromatography–mass spectrometry (GC–MS) analysis

GC analysis was performed using a GCMS-QP2010 (SHIMADZU) instrument equipped with an Rxi-5MS capillary column (30 m×0.25 mm; 0.25 μm film thickness). The GC settings were as

follows: the initial oven temperature was held at 50 °C for 1 min and increased at 3 °C min^{-1} to 200 °C for 1 min and then held at 200 °C for 5 min. The injector temperature was maintained at 250 °C. The samples (1 µl) were injected neat, with a split ratio of 1:50. The carrier gas was helium at a flow rate of 1.0 ml·min^{-1}. Spectra were scanned from 30 to 500 *m/z* at 0.5 scans·s^{-1}.

Oil samples were diluted to 1:100 in hexane using internal standards (*n*-octane and *n*-octadecane). Each analysis was repeated three times. A mixture of aliphatic hydrocarbons (C_8–C_{30}, Sigma-Aldrich) in hexane was injected directly into the GC injector under the above temperature program in order to calculate the retention index of each compound.

Identification and quantification of volatile components

Volatile constituents were identified through comparison of the mass spectra with those stored in NIST 08 and NIST 08S. Component relative percentages were calculated based on GC peak areas without using correction factors.Peak assignment was carried out by combining computer matching with the NIST 08 and NIST 08S libraries (Fogang *et al.,* 2012).

Statistical analysis

OriginPro v8.0 was used to process data for TIC of volatile components. SPSS 19.0 was used to carry out Cluster Analysis utilizing squared Euclidean distance with Ward's method.

References

PCPRC (Pharmacopoeia Commission of the People's Republic of China), 2010. Pharmacopoeia of the People's Republic of China, China Medical Science Press, Beijing, Appendix XD (in Chinese).

Fogang, H.P.D., Tapondjou, L.A., Womeni, H.M., Quassinti, L., Bramucci, M., Vitali, L.A., Petrelli, D., Lupidi, G., Maggi, F., Papa, F. et al., 2012. Characterization and biological activity of essential oils from fruits of Zanthoxylum xanthoxyloides Lam. and Z. leprieurii Guill. & Perr., two culinary plants from Cameroon. Flavour and Fragrance Journal, 27(2):171-179. http://dx.doi.org/10.1002/ffj.3083