Fingerprinting of green and roasted coffee (*Parainema* and *Obata*) volatile organic compounds (VOCs): HS-GC-IMS and GC-MS

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CONTEXT

Once coffee cherries have been picked, they must be processed before being transported and sold. Processing should take place as soon as possible to prevent spoilage, and is typically done in one of two ways: washed or natural. Processing has a significant impact on the final cup profile, affecting everything from acidity and sweetness to body and clarity. The processing method used by producers usually depends on a number of factors, including resources, climate, and cost [**1**]. A coffee cherry is made up of various layers, including skin, fruit, mucilage, and parchment. Once the cherries have been harvested, they need to be processed to remove these layers, so as to be left with only the coffee bean. One of the most widely used processing methods is "***wet processing***". This method involves removing the skin of the cherries before submerging them in a trough of water to break down and remove the mucilage.
This can sometimes take up to 24 hours, allowing time for tiny microorganisms in the beans to create enzymes that break down the sticky outer layer. After fermentation, the beans are washed and dried, either under the sun or using dedicated drying machines (or sometimes a combination of the two). The result tends to be a coffee with high clarity, light body, and prominent acidity.
In contrast, natural processing is a method that has been used for centuries. Also known as "***dry processing***", it involves spreading the harvested cherries out on a large surface to dry for several weeks with the fruit and skin intact. To avoid the build-up of mold and over-fermentation, they are regularly raked and turned. When the cherries reach a moisture level below 11%, the brittle outer layer is removed, and the bean within is kept for milling and sale.
Few data are available for green coffee VOCs analysis by HS-GC-IMS, particularly regarding the evaluation of VOCs related to the processing [**2**]. Main aim of this work was to evaluate the volatile fraction of different wet- or dry-processed coffee beans, directly analyzed (head space) with HS-GC-IMS as well as sampled by headspace-solid phase microextraction (GC-MS).

The aim: VOCs fingerprinting of wet- or dry-processed coffee samples from 10 different Honduran producers was obtained comparing SPME-GC-MS with HS-GC-IMS. Both analytical approaches permitted the clustering of the samples, leading information about aroma precursors. Moreover, HS-GC-IMS permitted the direct rapid analysis of green beans in vials, reducing sample pretreatment.

MATERIALS & METHODS

Samples: Samples: Fifteen different green wet- or dry-processed coffee samples (Parainema and Obata) from ten different Honduran producers were supplied by Andrej Godina. The same set of samples was subsequently roasted under controlled conditions in order to standardize the process.

HS-GC-IMS: Headspace-gas chromatography-ion mobility spectrometry (HS-GC-IMS) (FlavourSpec®, G.A.S., Dortmund, Germany) was used to assess the volatile composition with an untargeted fingerprinting approach. A 20 mL glass vial was filled with 2.0 g of the sample. Then samples were treated for 5 minutes at 50 °C at 500 rpm. Then, in splitless mode, a 300 µL headspace sample was automatically delivered through a 70 °C heated syringe. Using an MXT-5 column (15 m × 0.53 mm i.d., 1 µm film thickness; Restek Corporation, Bellefonte, PA, USA), the volatile chemicals were separated at 40 °C. As the carrier gas, 99.999 percent pure nitrogen was employed, and the flow rate program was configured as follows: 2 mL/min for 3 minutes, followed by a 17-minute rise to 25 mL/min and a 5-minute hold. A 3H ionization source ionized the eluted analytes before driving them to a drift tube, which was run at a constant temperature of 45 °C and voltage of 5 kV.

SPME-GC-MS: the extraction of volatiles was performed with manual 50/30um DVB/CAR/PDMS SPME device (Supelco®, Bellefonte, PA, USA) at 40°C for 10 minutes, after a 10 minutes equilibration time. The analysis was performed on a GCMS-QP2020 NX system (Shimadzu Co., Ltd., Tokyo, Japan), using 99.9995 pure helium as carrier gas. The fibre was desorbed in the injector of the GC in splitless mode at 250 °C. The separation was performed on a SH-I-5MS capillary column (30m x 0.25mm x 0.25um, Shimadzu Co., Ltd., Tokyo, Japan). The GC oven temperature was programmed as follows: 50°C held for 2 minutes, increased to 220 °C at a rate of 10 °C/min, held for 5 minutes. A constant flow of 1,5 ml/min was used. The volatile compounds were tentatively identified by comparing the spectral data obtained with the NIST database.

RESULTS AND DISCUSSION

GC-IMS

The analysis of volatile profile of green coffee is of great interest, particularly regarding the evaluation of the fermentation process. The aim of this work was to establish (and compare) the usefulness of two hyphenated analytical techniques applied to fingerprint green coffee volatile compounds, evaluating the capacity to cluster specific samples subjected to different processing (wet and dry).
Moreover, the direct rapid analysis of green beans by HS-GC-IMS (here proposed as screening rapid method, compared to GC-MS) was functional to clearly recognize clusters of samples. HS-GC-IMS provided 2D chromatogram useful to quickly obtain very clear 2D patterns, avoiding any kind of sample pre-analytical handlings and processing. The main differences are found in the two "dry processed" samples. They present a profile with a higher number of volatile compounds. The remaining 13 samples ("wet processed") generally showed a similar profile (**Figure 1** (*left*)). Substantially, even on the same set of roasted samples, the behavior detected is similar (data not shown for space reasons). The dry processed samples cluster compared to those obtained with wet processing. The latter, however, appeared less similar after the roasting process and their clustering is less evident.

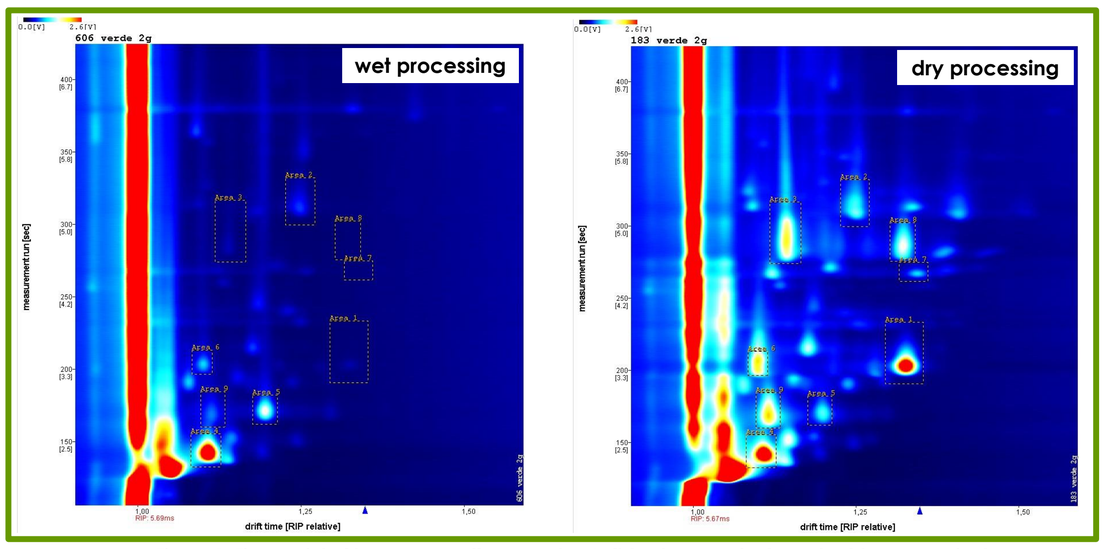


Figure 1. Fingerprint of two green coffee samples. Left (wet processing); center (dry processing).

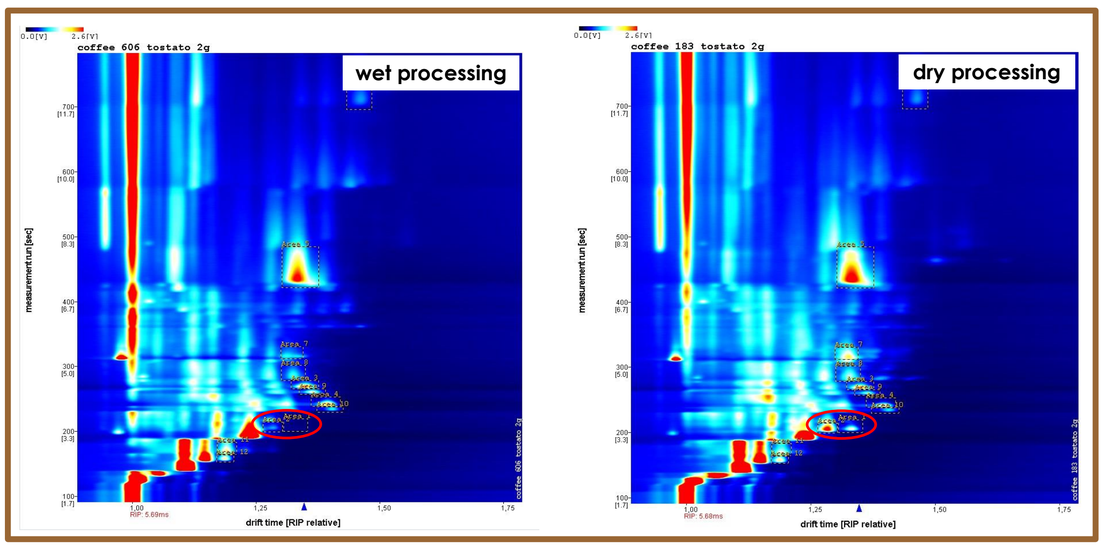
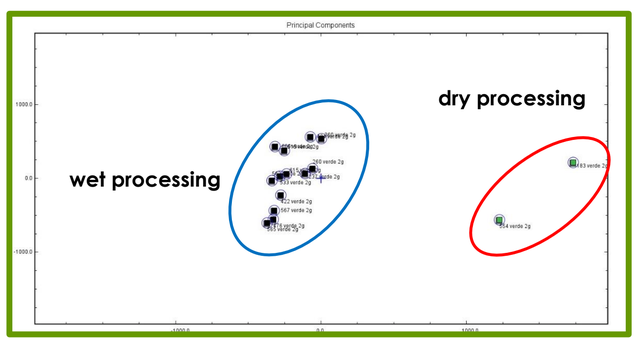
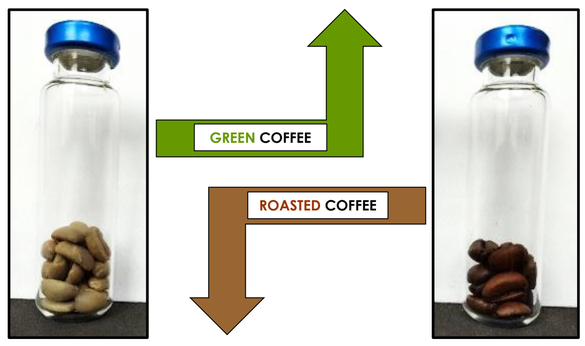


Figure 2. Fingerprint of the corresponding roasted samples. Left (wet processing); center (dry processing).





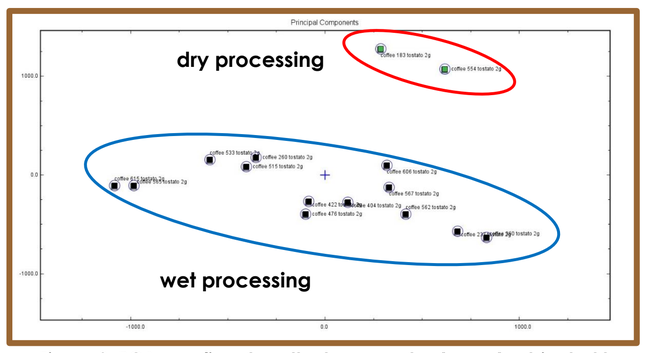
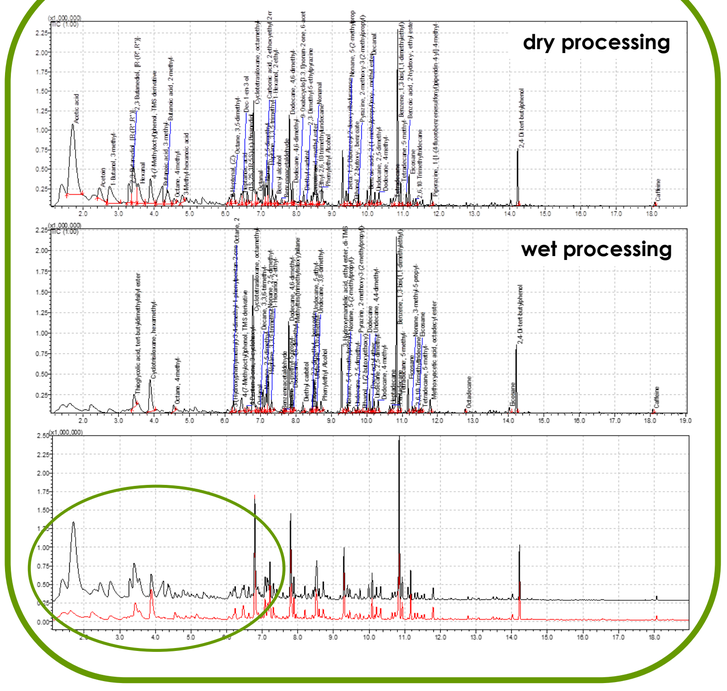
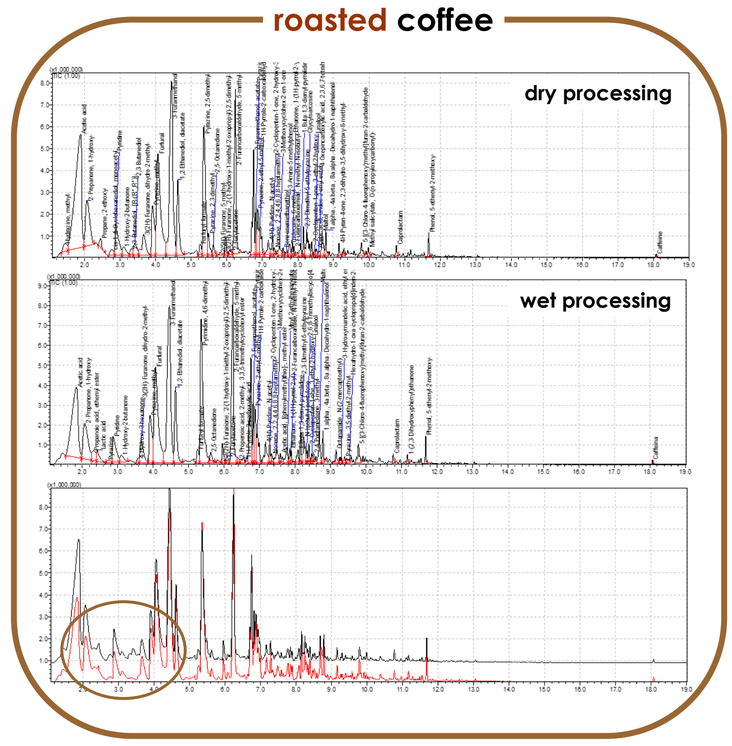


Figure 3. PCAs confirms how the two samples (green) subjected to the "dry" process cluster together.

GC-MS





These approaches emphasize the usefulness of the hyphenated multi-platform approach as analytical tool, preliminary to data mining [**3, 4**]. Both HS-GC-IMS and GC-MS analysis allowed to identify specific clusters of samples of known geographical origin, also permitting some considerations regarding the precursor of aroma. More particularly, rapid analysis using HS-GC-IMS reduced the time of the analysis, permitting the direct analysis of beans in vials without preliminary SPME extraction, opening new perspectives for green and/or roasted coffee quality control.

References
[1] B. Bertrand *et al*. Food Chemistry, 135(4) (**2012**) pp 2575-2583.
[2] C. Min, M. Bivi, L. Jianneng, L. Yimin, L. Yijun and C. Long, RSC advances, 12(24) (**2022**) pp 15534-15542.
[3] M. Bordiga *et al*., Food Bioscience, 55 (**2023**) 102987.
[4] L. Cecchi et al., Food Chemistry, 404 (**2023**) 134696