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Technical Report

Analysis of aroma compounds in chocolate formulated from cacaos of different geographical origins using SPME Arrow-GC-MS

Moyu Taniguchi[1], Eiichiro Fukusaki[1,2]

Abstract:

Food flavor and quality are often evaluated by analyzing aroma compounds on a gas chromatography-mass spectrometry (GC-MS) system. A GC-MS system using SPME Arrow was used to analyze the aroma compound profile of single origin chocolate samples, each formulated from cacaos with a different geographical origin. The resulting compound profile data were subjected to principal component analysis (PCA), and the aroma compound characteristics dependent on geographical origin were investigated by comparing four chocolate samples. The characteristic flavor of each chocolate sample seemed to arise from differences in their aroma compound profile.

Keywords: Chocolate, Aroma compounds, SPME Arrow, Gas chromatograph-mass spectrometer, Principal component analysis

1. Introduction

Food flavor and quality are often evaluated by analyzing aroma compounds on a gas chromatograph-mass spectrometry (GC-MS) system. Solid phase micro extraction (SPME) is a preparative technique that can extract aroma compounds simply and without solvent. The use of multifunctional autosamplers facilitates high reproducibility by providing strict control over extraction conditions such as temperature and time and is suited to the qualitative comparative analysis of multiple samples. Compared to conventional SPME, SPME Arrow uses a higher capacity adsorption phase coating, which increases loading capacity and enables higher sensitivity component analysis. SPME Arrow is also more durable due to a larger diameter and more robust construction. For these reasons, SPME Arrow is a component extraction technology expected to find common use in analyzing aroma compounds in food.

In this article, a GC-MS system using SPME Arrow was used to analyze aroma compounds in chocolate. Around 4,700,000 tons of cacao are produced annually worldwide, and chocolate is consumed in all parts of the world. The chocolate market is experiencing a demand for high-quality products, and bean-to-bar chocolate production is gaining in popularity. Bean-to-bar production is a method of producing unique, high-quality chocolate whereby the entire manufacturing process from buying the cacao beans to producing the final product is controlled by the chocolate maker. Single origin chocolate is a product of bean-to-bar production and typically has a high cacao content. Single origin chocolate preserves the flavor profile of cacaos sourced from a particular geographical location and allows people to enjoy their distinctive flavor differences. In this article, four commercially available single origin chocolates (made with cacaos from four different countries) were analyzed. SPME-Arrow-GC-MS analysis and principal component analysis, a type of multivariate analysis, were used to investigate the characteristic features of each aroma compound profile and to examine the effect of geography on the flavors of the chocolate products.

Table 1 SPME Arrow-GC-MS Analysis Conditions

System Configuration	
GCMS	: GCMS-TQ™8050 NX
Autosampler	: AOC-6000
Column	: Supelcowax10 (length 30 m, internal diameter 0.25 mm, film thickness 0.25 µm)
SPME Arrow Conditions	
SPME Arrow	: PDMS (length 20 mm, outer diameter 1.1 mm, film thickness 100 µm)
Conditioning temp.	: 250 °C
Pre Conditioning Time	: 15 min
Incubation Temp.	: 36 °C
Incubation Time	: 30 min
Stirrer Speed	: 250 rpm
Sample Extract Time	: 30 min
Sample Desorb Time	: 2 min
GC Conditions	
Injection Temp.	: 250 °C
Injection Mode	: Splitless
Control Mode	: Constant Linear Velocity (30 cm/sec)
Column Oven Temp.	: 40 °C (5 min), 3 °C/min, 240 °C (20 min)
MS Conditions	
Interface Temp.	: 250 °C
Ion Source Temp.	: 200 °C
Ionization Method	: EI
Measurement Mode	: Scan (35-350 *m/z*)
Event Time	: 0.1 sec

1 Department of Biotechnology, Graduate School of Engineering, Osaka University
2 Osaka University Shimadzu Omics Innovation Research Laboratories

2. Experimental Method

Four single origin chocolate products, each made from cacaos with a different geographical origin (four different countries, n = 3), were analyzed. The cacao content of all chocolate products was 70 %. Each chocolate sample (5 g) was finely chopped with a kitchen knife, placed in a 20-mL vial, and set in the autosampler (room temperature). SPME Arrow polydimethylsiloxane (PMDS) fiber (outer diameter 1.1 mm, film thickness 100 μm, length 20 mm) was then used to capture compounds in the vial headspace at 36 °C or below (human body temperature). The chocolate samples were fully melted during capture. SPME Arrow conditions and GC-MS conditions are shown in detail in Table 1. Data analysis was performed using GCMSsolution™. Compounds were annotated on the basis of mass spectral similarity using a NIST library and FFNSC fragrance library. Multivariate analysis was performed using SIMCA (Umetrics, Sweden).

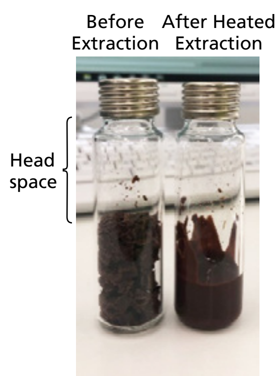


Fig. 1 Before and after compound extraction from chocolate samples after heating and shaking. Compounds in the headspace were extracted with SPME Arrow.

3. Results

SPME Arrow-GC-MS was used to analyze and acquire chromatography data on the aroma compound profile of four single origin chocolates, each made from cacaos with a different geographical origin (Fig. 2). Each chocolate, each with a different geographical origin, resulted in a different chromatogram. Each chocolate displayed different sensory attributes; sensory evaluation revealed the primary sensory attribute of the four chocolate samples as a nutty aroma, fruity aroma, floral aroma, and spicy aroma, respectively. To explore how an aroma compound profile related to a primary sensory attribute generated by cacaos from a single geographical origin, aroma compound chromatogram peak areas were used to perform principal component analysis. The percentage of variance explained by the first principal component was 48.6 % and by the second principal component it was 33.6 %. A score plot (Fig. 3) revealed almost no variation between repeated experiments, showing this experimental system gave highly reproducible results. Sample data were also plotted according to the geographical origin of cacaos and suggested that geographic origin affects the aroma compound profile of chocolate products. The primary sensory attribute of each chocolate sample is also shown on the score plot. The score plot (Fig. 3) and a loading plot (Fig. 4) were used to examine characteristic features of the aroma compound profile of each chocolate sample, and the effect of these characteristic features on the primary aroma attribute of each chocolate was studied. This revealed the chocolate sample with a nutty aroma contained a comparatively large amount of pyrazines, and the chocolate sample with a floral aroma contained a comparatively large amount of aliphatic alcohols. Pyrazines are typically known to produce nutty, cocoa, and roasted aromas[1]. Phenylethyl alcohol and some other alcohols are also known to produce floral aromas[1].

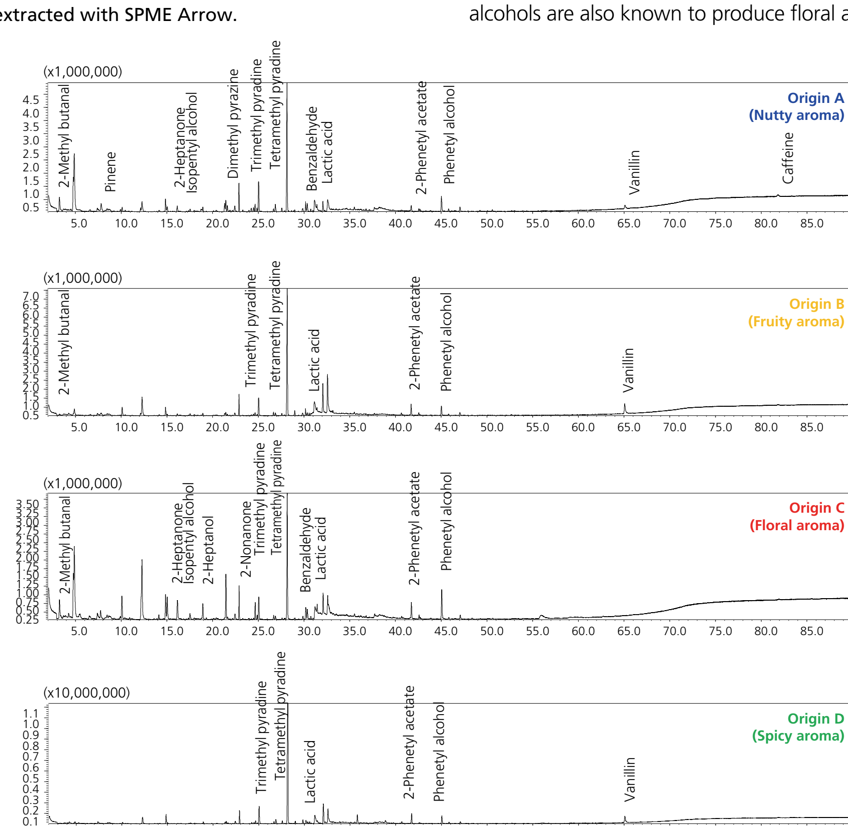


Fig. 2 Total Ion Chromatograms (TIC)
Vertical axis: peak intensity (arbitrary unit) Horizontal axis: elution time (minutes)

As shown by the score plot, compound extraction by SPME Arrow and the AOC-6000 autosampler produced highly reproducible chromatogram data and revealed the differences between the aroma compound profile of different chocolate samples. The above findings show this analytical method is suitable for comparative analysis of multiple samples and can be used to examine the correlation between aroma compounds and the sensory attributes of food.

4. Conclusion

A GC-MS system using SPME Arrow was used to examine the aroma compound profile of single origin chocolate samples, each sample made from cacaos with a different geographical origin. The resulting chromatograms were then analyzed by principal component analysis. This revealed differences between the aroma compound profile of chocolate made from cacaos with different geographical origins and allowed an investigation of which aromatic compounds were potentially responsible for the primary sensory attribute of each chocolate sample.

Reference

[1] Christine Counet, Delphine Callemien, Caroline Ouwerx, and Sonia Collin, Use of Gas chromatography-olfactometry to identify key odorant compounds in dark chocolate. Comparison of samples before and after conching, *Journal of agricultural and food chemistry*, 50, 2385-2391 (2002).

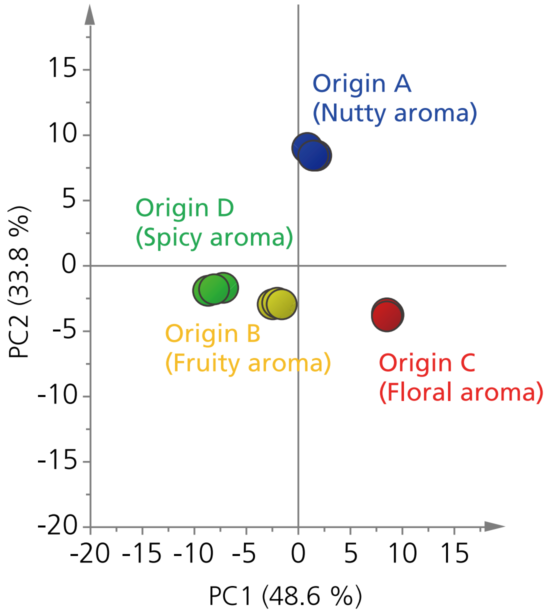


Fig. 3 Principal Component Analysis Score Plot (n = 3)

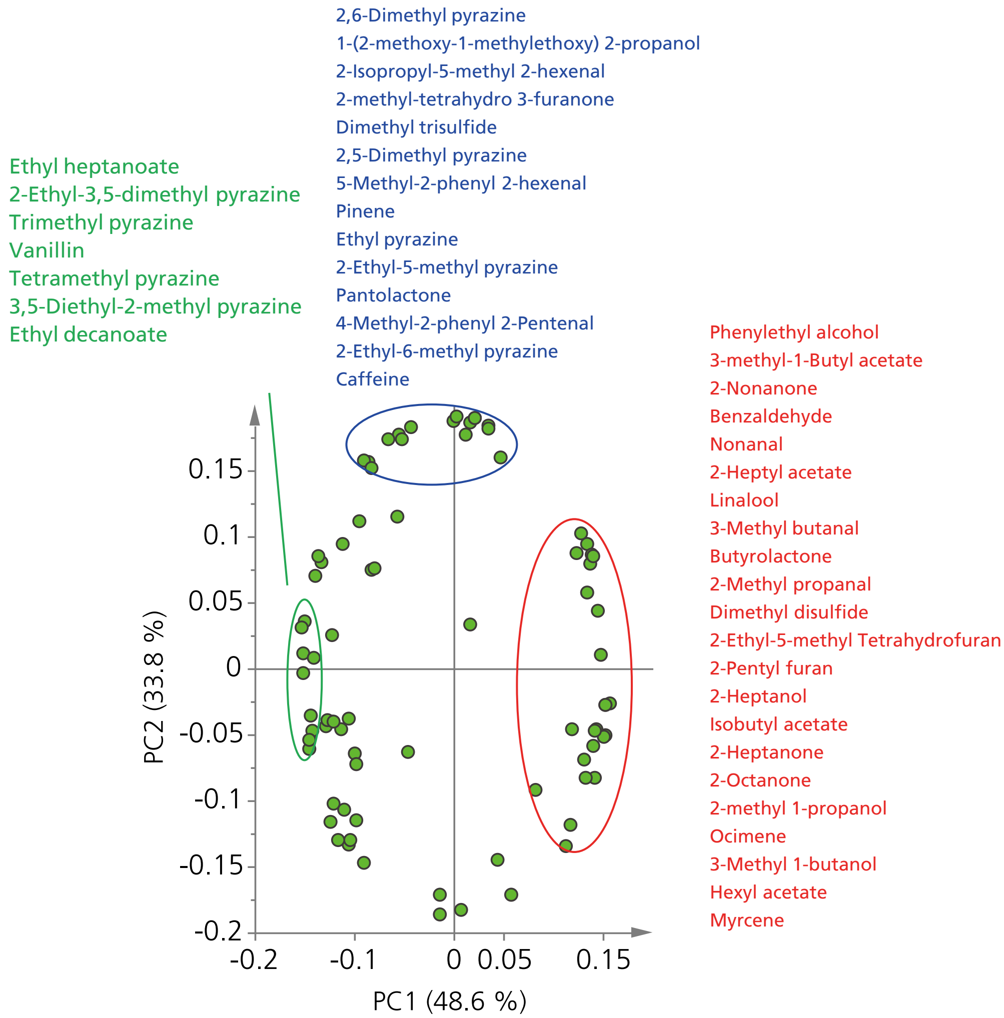


Fig. 4 Principal Component Analysis Loading Plot
(Data conversion: none, scaling: auto)

Multifunctional Autosampler

AOC-6000 Plus

Multifunctional Autosampler Dramatically Improves GC/MS Analysis Productivity

Multiple GCMS Sample Injection Methods in One Device

Perform liquid sample injection, headspace injection, solid phase micro extraction (SPME) and more, all in one device. In addition, by using the tool-switching feature, all the syringe tools for various injection methods installed on the park station can be swapped automatically.

Increased Data Reliability

Recording of syringe and fiber usage history on a chip improves the reliability of acquired data. Automated sample adjustment also reduces the burden on the operator and contributes to analysis accuracy.

High-Sensitivity Analysis Achieved by Latest Concentration Technology

Compared to previous SPME methods, the SPME Arrow achieves enhanced sensitivity and durability, and the ITEX DHS (In-tube Extraction Dynamic Headspace) offers higher sensitivity compared with previous HS, which makes analyses that employ the latest concentration technology possible.

Accommodates a Wide Range of Sample Forms

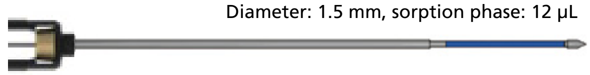
By using the AOC-6000 Plus with the OPTIC-4 multimode inlet, with its wealth of injection modes, pyrolysis analysis of solid samples, thermal desorption analysis of gaseous components, and a wide variety of other samples and analyses can be handled.

SPME Arrow Provides High Sensitivity and High Durability

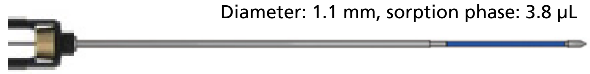
SPME Arrow, cutting-edge SPME technology, is coated with more adsorbent than conventional SPME fibers, enabling high-sensitivity analysis. Further, the thick and sturdy construction provides high durability.

SPME Arrow

Diameter: 1.5 mm, sorption phase: 12 µL

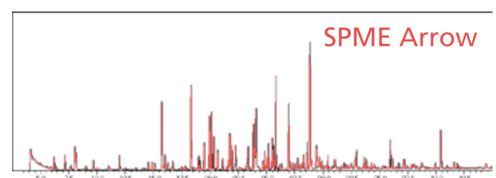


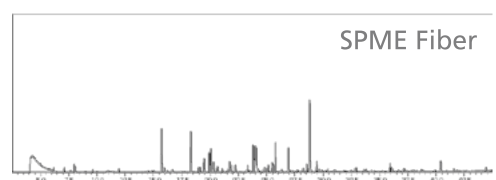
Diameter: 1.1 mm, sorption phase: 3.8 µL



SPME Fiber

Diameter: 0.6 mm, sorption phase: 0.6 µL





Analysis of Aroma Compounds in Coffee
(The PDMS 100 µm type SPME Arrow and SPME fibers are used.)

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