Comparison of Headspace Cannabinoid Profiles Detected from Different Structures of Dried Cannabis Inflorescences

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ABSTRACT

Automated headspace-solid phase microextraction-gas chromatography/mass spectrometry (HS-SPME-GC/MS was adopted to perform headspace chemical analysis for marijuana plant material. Different botanical structures found in standard marijuana samples, such as single structures of stem, leave, flower (bud, or calyx), as well as homogenized samples, and unaltered mixing samples were analyzed separately to evaluate the performance of the system. Relative standard deviation of normalized peak areas of each detected cannabinoid was used to evaluate the reproducibility of headspace chemical profiles. We found the headspace cannabinoids profiles of delta-9-tetrahydrocannabinol (THC) and cannabidiol (CBD) in the same standard marijuana sample were most reproducible when marijuana floral structures were used for HS-SPME-GC/MS analysis.

INTRODUCTION

Currently, liquid-liquid extraction (LLE) is the preferred method to extract cannabinoids from marijuana samples for instrumental analysis. However, we found that HS-SPME coupled with GC/MS can be automated to capture headspace cannabinoid profiles from small amounts of marijuana plant material without any use of organic solvent. HS-SPME is relatively non-destructive, and sensitive enough to detect trace amounts of certain compounds from sample headspace [1]. It has been previously used in the analysis of cannabinoids from the headspace of hair, urine, and blood [3].

Marijuana has a number of botanical structures present: leaves, stems, non-flowering buds, and flowering buds. While the preferred sample for forensic analysis of marijuana is flowering buds, they are not always present in seized samples. Additionally, laboratories will use samples homogenized by mortar and pestle or herb grinders for their analysis [1, 2].

In this study, marijuana samples with known concentrations of THC and CBD were acquired. The main compounds of interest in this experiment were: cannabidiol (CBD), delta-9-tetrahydrocannabinol (THC), and cannabinol (CBN). Different botanical structures were separated from the standard marijuana samples under the microscope. Those samples with the same botanical structures were analyzed by HS-SPME-GC/MS. Headspace cannabinoids profiles were evaluated to determine the variation among different botanical structures in the marijuana sample with the same THC level.

RESULTS AND DISCUSSION

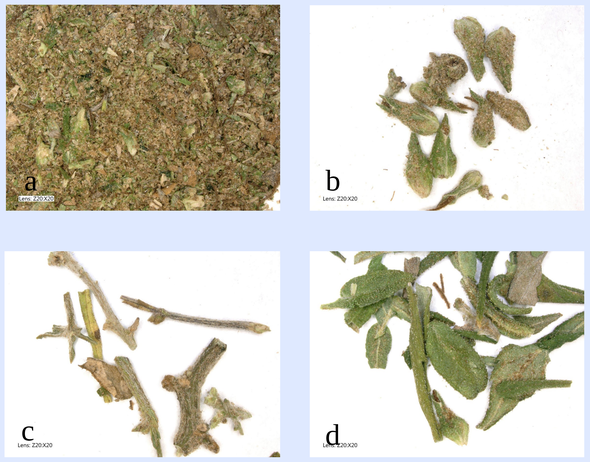

Figure 1: a. Different botanical structures observed in a typical marijuana sample. b. Typical floral structures (Calyx). c. typical stem structures. d. typical leave structures. All images were taken under x20 magnification.

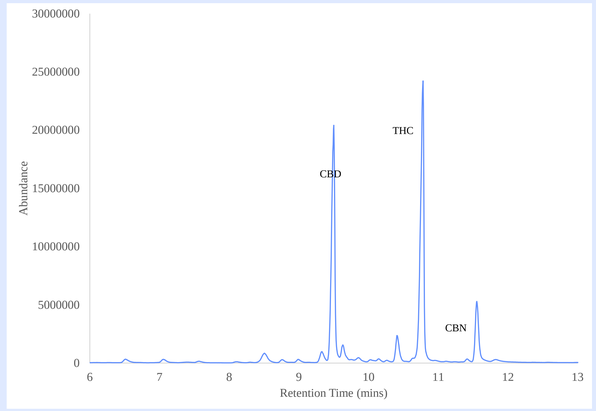

Figure 2: A typical total ion chromatogram (TIC) of headspace cannabinoids profiles from an unaltered (mixed structures) standard marijuana sample analyzed by HS-SPME-GC/MS.

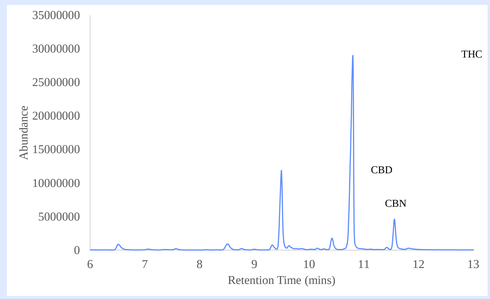

Figure 3: A typical total ion chromatogram (TIC) of headspace cannabinoids detected from 10 mg of calyx by HS-SPME-GC/MS.

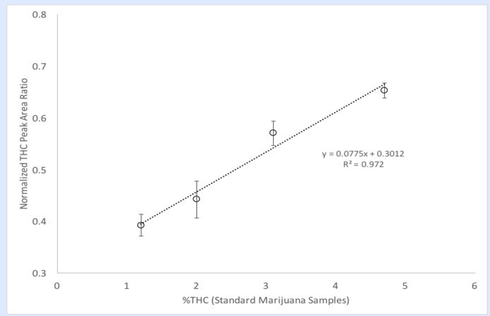

Figure 4: THC analysis with HS-SPME-GC/MS, 10 mg of unaltered mixed samples were used. Normalized THC peak areas calculated from total ion chromatogram (TIC) were used to construct the calibration curve (N=3 for each THC level).

Table 1. Percent standard deviations of peak areas of each cannabinoids detected from sample headspace from different botanical structures.

Sample Type	Δ9-THC	CBN	CBD
Unaltered mix	19%	12%	40%
Crushed	25%	22%	28%
Leaf	26%	23%	48%
Stem	33%	25%	44%
Calyx	11%	11%	19%

REFERENCES

1. Lai, H., Corbin, I., Almirall, J. Headspace sampling and detection of cocaine, MDMA, and marijuana via volatile markers in the presence of potential interferences by solid phase microextraction-ion mobility spectrometry (SPME-IMS). *Analytical and Bioanalytical Chemistry*, **2008**, 392(1-2), 105-113.
2. Ilias, Y., Rudaz, S., Mathieu, P., Christen, P., Veuthey, J. Extraction and analysis of different Cannabis samples by headspace solid-phase microextraction combined with gas chromatography-mass spectrometry. *Journal of Separation Science*, **2005**, 28(17), 2293-2300.
3. de Oliveira, C., Yonamine, M., Moreau, R. Headspace solid-phase microextraction of cannabinoids in human head hair samples. *Journal of Separation Science*, **2007**, 30(1), 128-134.

MATERIALS AND METHODS

Materials

Fourteen marijuana standard samples with different levels of THC and CBD were provided by the National Institute on Drug Abuse (NIDA) Drug Supply Program (DSP) and were analyzed at the Southwest Regional Science Center (U.S Customs and Border Protection). Different botanical structures in each standard marijuana were first separated under a microscope. Each sample (10 mg) was weighed and placed in a 20 mL vial for HS-SPME.

Instrumentation

An Agilent 7890B system coupled to dual detectors, 5977A mass selective detector and flame ionization detector (MSD/FID), was used for GC/MS analysis. The column was Restek Rxi 35Sil-M3 [Length: 15m, Inner Diameter: 0.25 mm, Film Thickness: 0.25 µm].

HS-SPME Sampling

Polydimethylsiloxane (PDMS) SPME fiber (23 gauge, 100 µm coating) was installed on an Agilent GC Sampler 120 for HS-SPME automation. Vials were placed in the GC Sampler 120 and heated to 150°C for 5 minutes with agitation before HS-SPME. After 5 minutes of heating, HS-SPME was performed by inserting the SPME fiber into the headspace of vial. After HS-SPME, the extracts from the sample headspace was then injected to the GC/MS by thermal desorption at 250°C for 30 seconds.

CONCLUSIONS

This study showed that sampling floral structure (calyx) from marijuana samples yielded the most reproducible profiles for THC, CBN, and CBD from sample headspace. However, calyces were not present in all standard marijuana samples. When calyces are missing from marijuana sample, sampling a mix of all botanical structures for HS-SPME should be considered to alleviate variations among different botanical structures.

The HS-SPME-GC/MS approach is also capable of quantifying marijuana plant materials at a linear range between 1% to 5 % of THC. When THC concentration in marijuana is higher than 5%, the peak area of THC calculated from TIC started to deviate from linear range, indicating potential saturation of THC on the SPME fiber during HS-SPME.

HS-SPME-GC/MS is a promising analytical technique to obtain cannabinoid profiles from the headspace of small amount (10 mg) of marijuana samples. No sample preparation was needed. For our future work, we will apply this analytical platform for the classification of marijuana samples.

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