Application Brief

Cannabis and Hemp Testing

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A Comparison of Quantitation of Cannabinoids in Hemp Using GC/FID and LC/UV

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Introduction

Total THC is an essential calculation used to classify *Cannabis sativa* L. as cannabis and hemp. Following U.S. federal laws, the total THC must be less than 0.3% total THC (by dry weight) for *Cannabis sativa* L. to be classified as hemp. For quantitating THC and other popular cannabinoids, Agilent offers a comprehensive chemical standards suite for cannabis potency testing. The standards include 11 cannabinoids that can be used as single components mixtures at 1 mg/mL or combined in four mixtures, with each component at 1 mg/mL.

While most testing for total THC and screening for cannabinoids uses LC/UV instrumentation, these cannabinoids can be measured successfully with other popular instrumentation such as GC/MS or GC/FID. Both instruments can analyze samples from the same sample preparation procedure and yield comparable results. This work demonstrates of how the same cannabis potency standards and a single sample preparation procedure were used to measure total THC and CBD in hemp bud extracts using both GC- and LC-based approaches.

Experimental

Hemp flower was purchased from AccuStandard, Inc. and prepared for analysis using the sample preparation procedure outlined in Figure 1.

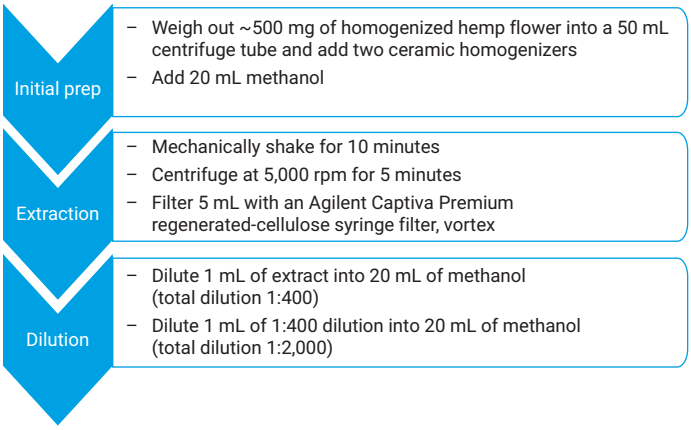

Figure 1. Sample preparation workflow.

The total THC and CBD were measured on two instruments for comparison: an Agilent 8890 GC with FID detection and an Agilent 1290 Infinity II LC with UV detection. Instrument method parameters are detailed in Tables 1 and 2.

Table 1. Instrument parameters for the Agilent 8890 GC with FID detection.

GC Parameters	Agilent 8890 GC
Inlet	Multimode inlet, 300 °C Pulsed split, 5:1
Column	Agilent J&W DB-35ms Ultra Inert, 30 m × 0.25 mm × 0.25 µm
Carrier Gas	He, 1.4 mL/min
Oven	90 °C (hold for 0.5 min), ramp at 40 °C/min to 300 °C (hold for 4.75 min)
FID	**Heater**: 320 °C **H2 flow**: 50 mL/min **Air flow**: 400 mL/min **Make up flow**: 50 mL/min
Injection	1 µL
Software	Agilent OpenLab CDS, version 3.5

Table 2. Instrument parameters for the Agilent 1290 Infinity II LC with UV detection.

LC Parameters	Agilent 1290 Infinity II LC
Flow Rate	0.425 mL/min
Column	Agilent InfinityLab Poroshell 120 SB-C18, 3.0 × 75 mm, 2.7 µm
Mobile Phase A	5 mM ammonium formate + 0.1% formic acid in water
Mobile Phase B	0.1% formic acid in acetonitrile
Column Temperature	35 °C
Injection	5 µL
Elution	Isocratic
% B	72%
Diode Array Detector	228 nm
Software	Agilent MassHunter Workstation software, version 10.0

Results and discussion

The two analytical techniques used to measure total THC and CBD yielded similar results for the same extract of the hemp flower bud. Cannabinoids spiked into the matrix are also detected by both methods. Cannabinolic acids CBDA, THCA, and CBGA are not observed directly via GC as they are decarboxylated into their neutral forms in the GC inlet. Figures 2 and 3 show the chromatograms obtained by the GC/FID method and LC/UV method, respectively.

Table 3. The reported percent total of both THC and CBD is compared with results derived from LC/UV and GC/FID. The acids CBDA and THCA are combined with their neutral counterparts for reporting.

Hemp Composition			
Cannabinoid	Reported % Potency	LC % Potency	GC % Potency
CBD + CBDA	3.47	3.28	3.11
delta9-THC + THCA	0.17	Not detected	Not detected

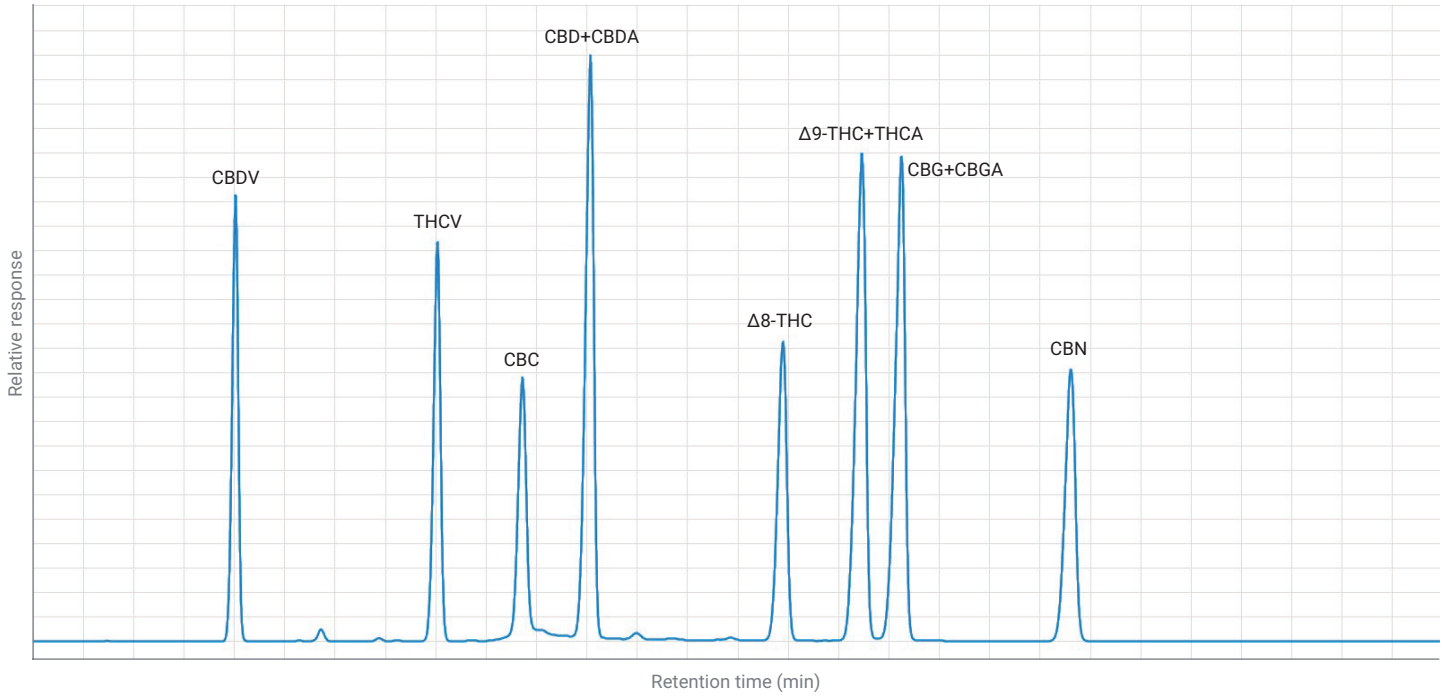

Figure 2. Chromatogram of 11 cannabinoids in hemp bud matrix by GC/FID. The cannabinolic acids CBDA, CBGA, and THCA are all decarboxylated to their neutral forms in the inlet during injection. Total THC and CBD can be derived directly from the measurement.

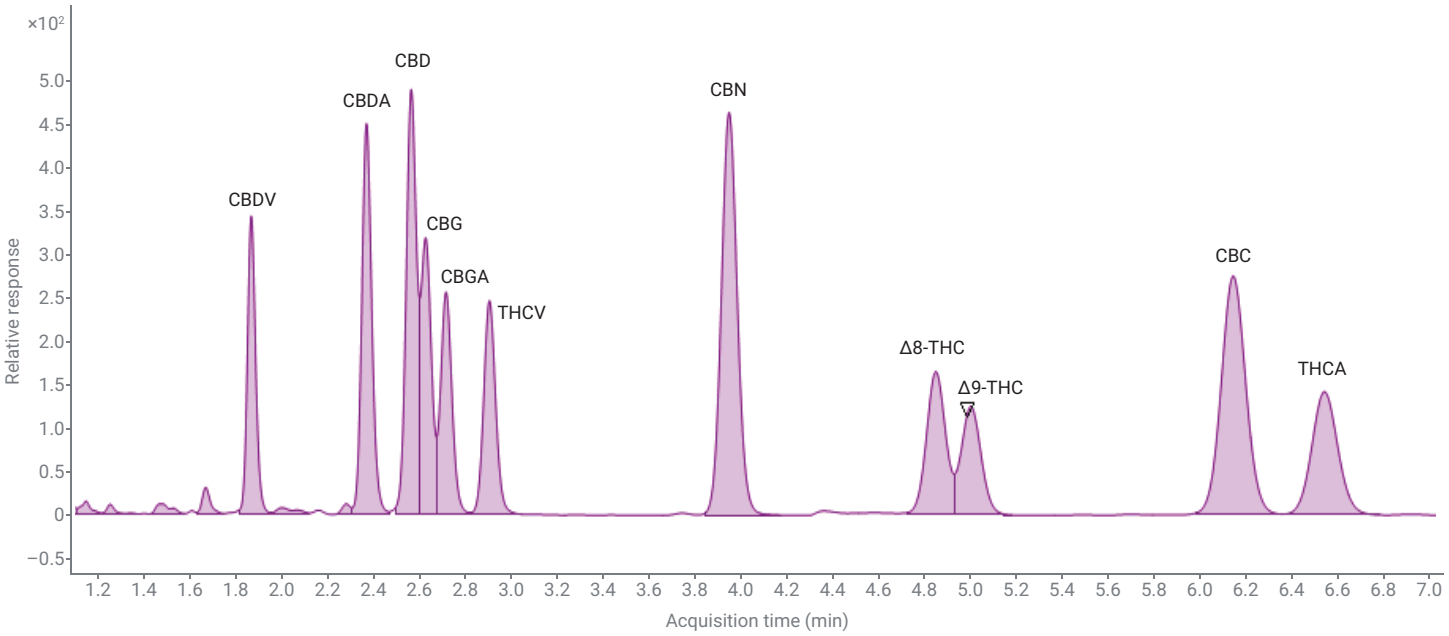

Figure 3. Chromatogram of 11 cannabinoids in hemp bud matrix by LC/UV. Due to ambient temperature analysis, all 11 cannabinoids are observed. To determine total THC, the following equation was used: total THC = Δ9-THC + (0.88 × THCA). Similarly, total CBD is calculated using the equation: total CBD = CBD + (0.88 × CBD).

Conclusion

Agilent Cannabinoid Reference Standards can be used to quantitate total THC and CBD. A single sample preparation process can be used to analyze cannabinoids on multiple Agilent instruments to yield accurate results that are comparable to each other.

Appendix

Table 4. Chemical standards referenced in this document.

Part Number	Agilent Cannabinoid Reference Standards
5190-9430	Cannabinoid Mix A – CBD, CBN, delta9-THC in methanol, 1 mg/mL
5190-9429	Cannabinoid Mix B – CBG, THCA, CBDA in acetonitrile, 1 mg/mL
5190-9428	Cannabinoids Mix C – CBC, CBGA, CBDV in acetonitrile, 1 mg/mL
5190-9427	Cannabinoids Mix D – THCV, delta8-THC in methanol, 1 mg/mL
5191-3920	Cannabidivarin (CBDV) in methanol, 1 mg/mL
5191-3921	Tetrahydrocannabivarin (THCV) in methanol, 1 mg/mL
5191-3922	delta8-Tetrahydrocannabinol (delta8-THC) in methanol, 1 mg/mL
5191-3923	Cannabigerol (CBG) in methanol, 1 mg/mL
5191-3924	Cannabidiol (CBD) in methanol, 1 mg/mL
5191-3925	delta9-Tetrahydrocannabinolic acid (THCA) in acetonitrile, 1 mg/mL
5191-3926	Cannabinol (CBN) in methanol, 1 mg/mL
5191-3927	Cannabigerol acid (CBGA) in acetonitrile, 1 mg/mL
5191-3928	Cannabichromene (CBC) in methanol, 1 mg/mL
5191-3929	delta9-Tetrahydrocannabinol (delta9-THC) in methanol, 1 mg/mL
5191-3930	Cannabidiolic acid (CBDA) in acetonitrile, 1 mg/mL

To see the complete lists of lab supplies for cannabis and hemp workflows, please visit www.agilent.com/chem/cannabis-workflow-ordering-guide.

Agilent products and solutions are intended to be used for cannabis quality control and safety testing in laboratories where such use is permitted under state/country law.

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