

Terpene Identification in Hemp Flower by GC/MS

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Introduction

Terpenes and terpenoids are the chemicals responsible for giving cannabis plants their distinct flavors and aromas. These classes of compounds, while not currently regulated by any governing body, are used to identify and characterize specific strains of both cannabis and hemp plants. Due to the terpenes characteristics, the ability to identify and quantify these compounds has become of great interest to the cannabis industry. Of all the terpenes identified in the *Cannabis sativa* L. plant, there are only a handful of terpenes that are frequently tested for profiling cannabis strains. Agilent offers a suite of cannabis terpene standards to identify and quantitate 39 of these terpenes. Presented here is a brief demonstration of how Agilent chemical standards can be used for identifying terpenes in hemp flower.

Experimental

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

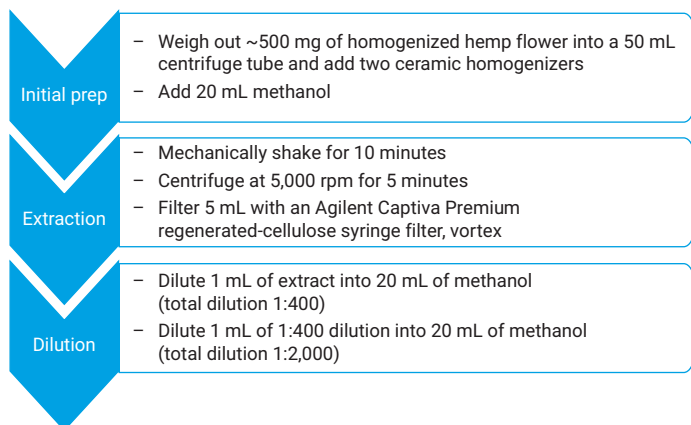


Figure 1. Sample preparation workflow for extraction of hemp flower.

The analysis of the terpenes was carried out on an Agilent 8890 GC with an Agilent 5977B GC/MSD. The method parameters for the instrument are listed in Tables 1 and 2. Agilent MassHunter Workstation software was used for acquisition and analysis.

Table 1. GC method parameters.

GC Parameters	Agilent 8890 GC
Liner	Ultra Inert, split, straight
Injection Mode	Split, 20:1
MMI Inlet Temperature	280 °C
Oven Program	40 °C (hold for 0 min), ramp 10 °C/min to 180 °C (hold for 0 min), ramp 20 °C/min to 280 °C (hold for 1 min)
Equilibrium Time	0.5 min
Column Flow	Constant, 1.3 mL/min
Column	Agilent J&W DB-35ms Ultra Inert, 30 m × 0.25 mm × 0.25 µm
Transfer Line	280 °C

Table 2. MSD method parameters.

MSD Parameters	Agilent 5977B GC/MSD
Acquisition Mode	Scan m/z 65 to 350
Solvent Delay	2 min
Tune File	Atune
Gain	1
MS Source Temperature	230 °C
MS Quadrupole Temperature	150 °C

Results and discussion

Calibration of 39 terpenes was carried out in hemp flower extract matrix, with each Agilent Cannabis Terpenes Mix calibrated individually. A five-point calibration curve was generated for each analyte. A sample calibration curve of a-humulene is shown in Figure 2.

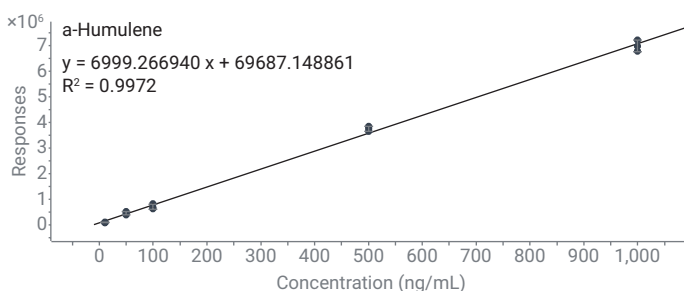


Figure 2. The five-point calibration of a-humulene in hemp flower extract matrix.

A representative chromatogram from each mix was overlaid to show the full separation of all the terpenes that was achieved (Figure 3). This method enables the analysis of all 39 terpenes in a single analytical run. The sample hemp flower bud extract contained the sesquiterpenes a-humulene and b-caryophyllene.

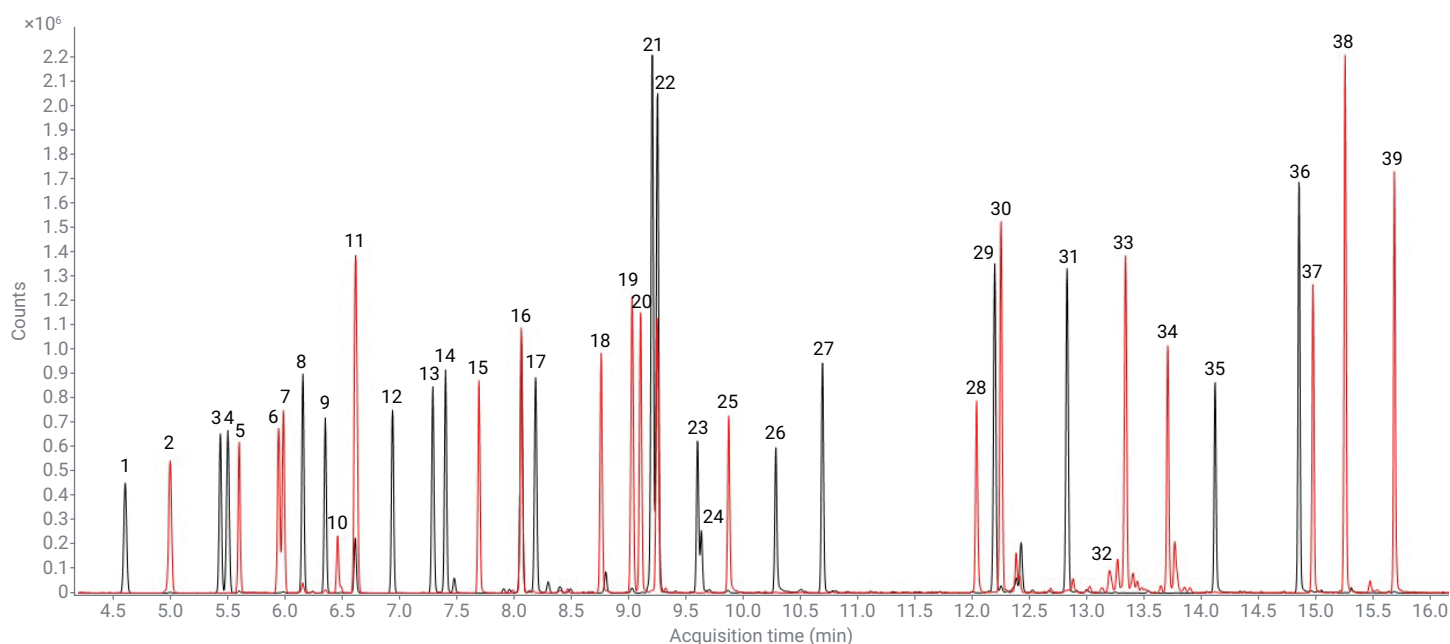


Figure 3. An overlay of two Agilent Cannabis Terpenes Mixes (TPM-100-1 and TPM-105-1), a total of 39 different terpenes fully separated, spiked into hemp flower extract. Analyte 1-borneol coelutes with (–)-borneol. (+)-Fenchone is a mix of isomers that also coelutes as one peak. 4-Thujaanol was spiked separately and is not a part of the mixes. Terpineol is observed as two peaks, the alpha and gamma isomers.

Table 3. List of the 39 terpenes in the combined chromatograms for two Agilent Cannabis Terpenes Mixes (TPM-100-1 and TPM-105-1).

No.	Analyte	No.	Analyte
1	a-Pinene	21	Menthol
2	Camphene	22	(–)-Borneol (sum of enantiomers)
3	Sabinene	23	a-Terpineol
4	Myrcene (tech)	24	g-Terpineol
5	b-Pinene	25	Nerol
6	a-Phellandrene	26	Geraniol
7	(+)-3-Carene	27	Pulegone
8	a-Terpinene	28	Geranyl acetate
9	Ocimene	29	Farnesene (isomeric mix)
10	(R)-(+)-Limonene	30	a-Cedrene
11	Eucalyptol	31	b-Caryophyllene
12	g-Terpinene	32	a-Humulene
13	Terpinolene	33	(+)-Valencene
14	4-Thujaanol (sabinene hydrate)	34	cis-Nerolidol
15	Linalool	35	trans-Nerolidol
16	(+)-Fenchone (sum of enantiomers)	36	(–)-Guaial
17	(+)-Fenchol	37	Caryophyllene oxide
18	(–)-Isopulegol	38	(–)-a-Bisabolol
19	(–)-Camphor	39	(+)-Cedrol
20	Isoborneol		

Conclusion

Using Agilent Cannabis Terpenes Mix standards allows for the detection and quantitation of 39 of the most common terpenes in hemp flower extracts in a single analytical run.

Appendix

Table 4. Chemical standards referred to in this document.

Part Number	Agilent Cannabis Terpenes Standards
TPM-100-1	Cannabis Terpenes Mix – 20 components (in methanol)
TPM-105-1	Cannabis Terpenes Mix – 21 components (in methanol)
SNV-105-1	Cannabis Terpenes Mix – 9 components (in isopropanol)
RSC-CBS-UNI	Universal Cannabis Residual Solvent Mix (in triacetin)

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

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