SPME Arrow Sampling of Terpenes in Cannabis Plant Material

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Introduction

With the growing legalization of medicinal cannabis worldwide, methods for qualifying and quantifying terpene concentrations have risen to the forefront of the analytical industry. The development of faster and more efficient methods that will produce rapid and accurate results at a low cost is highly desirable.

Since terpenes have high vapor pressures, and are extremely volatile, they are excellent candidates for static headspace gas chromatography (GC) analysis. PAL SPME Arrows can be used for both qualitative and quantitative determination of terpenes in plant material by headspace (HS) sampling combined with GC/MS.

This approach offers several advantages compared to solvent extraction and GC-FID. It does not require the use of organic solvents, does not co-extract matrix (which could potentially interfere with the chromatographic analysis or contaminate the GC system), and provides another means of peak identification using spectral data. PAL SPME Arrows provided the sensitivity and robustness needed to profile the predominant terpenes in an unknown variety of cannabis plant samples.

Terpenes

Cannabis contains more than 100 different terpenes and terpenoids, as well as other miscellaneous compounds of terpenoid origin. Different cannabis strains have been developed that contain distinct aromas and flavors, which is a result of the differing amounts of specific terpenes present.

Terpenes are the naturally occurring combination of carbon and hydrogen, whereas terpenoids are terpenes that have been modified through a drying and curing process (chemical modification), altering the oxygen content of the compound.

References

Analysis of Terpenes in Cannabis Using the Agilent 7697A/7890B/5977B Headspace GC-MSD System Faster Analysis Time = Greater Productivity. Agilent Application Note 5991-8499EN. September 2017.

Stenerson, K. and Halpenny, M. Analysis of Terpenes in Cannabis Using Headspace Solid-Phase Microextraction and GC–MS. LCGC, 1 May 2019. http://www.chromatographyonline.com/analysis-terpenes-cannabis-using-headspace-solid-phase-microextraction-and-gc-ms.

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.

Methodology

Terpene profiling (qualitative)

- **Sample:** 0.02 – 0.03 g of homogenized cannabis plant material was weighed into a 20 mL headspace vial
- **SPME Arrow:** 1.1 mm, 120 µm DVB/CAR WR/PDMS (p/n 5191-5861)

Target terpene analysis (quantitative)

- **Sample:** 0.1 g of homogenized cannabis plant material was weighed into a 20 mL headspace vial
- Calibration: 10 µL of prepared calibration standard (2 – 50 ppm) was added to each sample. Samples were capped and after a 10 min equilibration at room temp, 8 mL of Milli-Q H_2O was added to each sample.
- **SPME Arrow:** 1.1 mm; 100 µm PDMS (p/n 5191-5862)

- PDMS is less prone to overload than the DVB/CAR WR/PDMS phase

Note that the homogenization of the sample and the addition of water increases reproducibility.

SPME-GC/MSD

Terpenes from the cannabis flower were extracted by use of headspace SPME with a PAL RTC rail system. This was combined with the Agilent 7890B GC system, coupled with an Agilent 5977B High Efficiency Source GC/MSD.

Agilent 7890B GC Settings	
Turn top assembly	Agilent 7890 GC turn top assembly enlarged ID – inert (p/n G3452-60930)
Inlet liner	Inlet liner, Ultra Inert, straight, 2 mm id (p/n 5190-6168)
Inj. temp	270 °C
Inj. mode	100:1 split
Control mode	Constant flow (1 mL/min; 1.4 mL/min into MSD)
Column	J&W DB-1ms GC column, 60 m, 0.25 mm, 0.25 µm (p/n 122-0162)
Oven program	60 °C (hold 2 min); 5 °C/min to 140 °C (hold 1 min); 15 °C/min to 250 °C (hold 4 min)

PAL RTC rail system, combined with an Agilent 7890B GC and 5977B High GC/MSD.

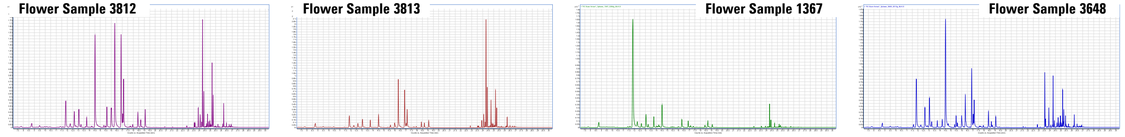
SPME Headspace Parameters	
Incubation time	5 min
Heatex stirrer speed (agitation)	1000 rpm
Heatex stirrer temp (extraction temp)	40 °C
Sample extract time	5 min
Sample desorption time	3 min
Conditioning time	5 min
Conditioning temp	270 °C

Agilent 5977B MS Conditions	
Transfer line	300 °C
Acquisition mode	Scan
Solvent delay	7 min
Tune file	atune.u
Gain	1
MS source temp	280 °C
MS quad temp	150 °C

Results and Discussion

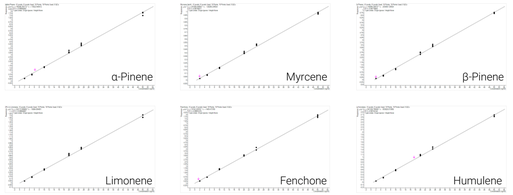
Terpene profiling

Flower samples were profiled with the use of the 120 µm DVB/CAR WR/PDMS (p/n 5191-5861) Arrow:



Targeted terpene analysis

Flower samples were extracted with the use of the 100 µm PDMS (p/n 5191-5862) Arrow:

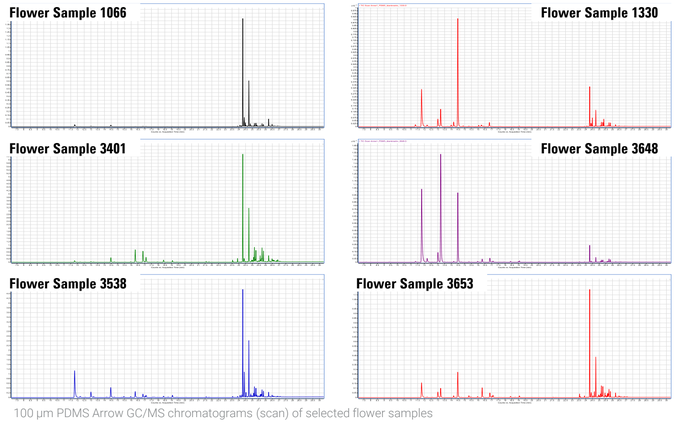


100 µm PDMS Arrow calibration curves (2 – 50 ppm) of selected terpenes

Retention times and linearity of selected terpenes (100 µm PDMS Arrow calibration curves; 2 – 50 ppm)

Terpene	RT	R^2
alpha-Pinene	11.865	0.9956
Camphene	12.274	0.9979
Sabinene	12.876	0.9979
Myrcene (tech)	13.061	0.9974
B-pinene	13.267	0.9972
A-phellandrene	13.782	0.9970
3-carene	14.039	0.9975
(R)-(+)-Limonene	14.526	0.9969
Ocimene	14.926	0.9963
Fenchone	15.985	0.9984

Terpene	RT	R2
Terpinolene (tech grade)	16.26	0.9979
(+)-Fenchol	16.895	0.9981
Camphor	17.598	0.9991
Isoborneol	18.188	0.9964
Menthol	18.606	0.9988
B-caryophyllene	24.35	0.9979
A-humulene	24.76	0.9974
Caryophyllene oxide	26.222	0.9991
(-)-Guaiol	26.307	0.9961
(+)-Cedrol	26.486	0.9968



100 µm PDMS Arrow GC/MS chromatograms (scan) of selected flower samples

Identification and quantitation (ppm) of terpenes in selected flower samples (100 µm PDMS Arrow)

Terpene	Flower 1066	Flower 1330	Flower 3401	Flower 3538	Flower 3648	Flower 3653
alpha-Pinene	6.22	77.70	1.76	28.44	259.56	38.66
Camphene	0.72	3.96	0.73	1.48	14.19	1.88
Sabinene	0.82					0.89
Myrcene (tech)	1.91	12.09	1.25	4.90	24.75	11.43
B-pinene	1.99	30.79	1.84	2.35	376.83	21.38
A-phellandrene	0.16	0.65	0.14	0.17	0.45	0.20
3-carene	0.09	1.79	0.10			
(R)-(+)-Limonene		78.93		4.66	85.49	26.11
Ocimene					1.07	1.05
Fenchone	1.48	1.36	5.31	7.37	24.31	19.53
Terpinolene (tech grade)	0.64	0.93	7.42	2.78	4.06	9.58
(+)-Fenchol			597.15	137.83	536.99	455.93
Camphor		7.62	4.95	5.37	7.15	7.48
Isoborneol			3.69		3.56	2.89
Menthol		19.23				
B-caryophyllene	51.42	13.08	23.19	17.46	10.20	41.23
A-humulene	17.48	4.87	10.04	7.74	3.04	13.21
Caryophyllene oxide	142.20	21.20	63.99	33.62	9.51	71.22
(-)-Guaiol	8.53			1.01	6.88	
(+)-Cedrol	34.87					

Conclusion

PAL SPME Arrows provided the sensitivity and robustness needed to profile the predominant terpenes in an unknown variety of cannabis plant samples. This shows that the SPME Arrows can be used for both qualitative and quantitative determination of terpenes in plant material by headspace (HS) sampling combined with GC/MS.