

Selective and Sensitive Analysis of 20 Category I and II Residual Solvents for Cannabis Application using Headspace-GCMS

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1. Introduction

In the cannabis industry, many different solvents are used in the extraction process, each one classified based on the level of toxicity. Also, sample preparation is difficult with cannabis matrices due to their lack of solubility, and the limitation of available non-interferent diluent solvents. The ideal way for testing is direct analysis of the sample in a headspace vial without any sample preparation involved. However, this can be challenging if high sample amounts are required. Furthermore, other method development challenges exist, such as co-elution of several residual solvent analytes or mass-to-charge interferences from the environment. Finding an acceptable balance between proper separation for unequivocal identification and quantitation, and reasonable analysis throughput time can be complicated. The work described herein demonstrates the viability of headspace GC-MS with SIM mode for the analysis of twenty Category I and II residual solvents, including three soluble gases, which may be present in cannabis products.

2. Experimental Methods

A 6-point calibration curve was created from CPI Category I and II standards. An aliquot of 150 µL was placed in a 20 mL headspace vial and capped. Butyl acetate was used as diluent solvent. Concentration ranges and calculated sample amounts are listed in Table 1. Quality Control standards were prepared by weighing a minimum of 250 mg of methyl cellulose (substitute matrix), and spiked with residual solvents standards to obtain a Level 5 concentration in the QC samples tested.

Table 1. Target Residual Solvents (left table) and Calculated Amounts for Calibration Curve Standards (right table).

Category I (action limit = 1 µg/g)	Category II (action limit > 1 µg/g)	Calibration Curve Standards	Concentration (µg/mL)	Volume in vial (mL)	Amount (µg)
1,2-Dichloroethane	Acetone	Category I Residual Solvent			
Benzene	Acetonitrile	Calibrator Level 1	0.781	0.150	0.117
Chloroform	Butane	Calibrator Level 2	1.563	0.150	0.234
Ethylene oxide	Ethanol	Calibrator Level 3	3.125	0.150	0.469
Methylene chloride	Ethyl Acetate	Calibrator Level 4	6.25	0.150	0.938
Trichloroethylene	Ethyl ether	Calibrator Level 5	12.5	0.150	1.875
	Heptane	Calibrator Level 6	25.0	0.150	3.75
	Hexane	Category II Residual Solvent			
	Isopropyl alcohol	Calibrator Level 1	312.5	0.150	46.875
	Methanol	Calibrator Level 2	625	0.150	93.75
	Pentane	Calibrator Level 3	1250	0.150	187.5
	Propane	Calibrator Level 4	2500	0.150	375
	Toluene	Calibrator Level 5	5000	0.150	750
	Total xylenes (ortho-, meta-, para-)	Calibrator Level 6	10000	0.150	1500

3. Analytical Conditions

Table 2. System Configuration and Instrument Parameters.

HeadSpace	HS-20 Loop Model
Operation Mode	Static headspace with loop
Sample	150µL sample volume
Equilibration	20 mL headspace vial 15.00 minutes at 120°C
Sample Loop	0.2-mL Loop Val pressure 300kPa Pressurizing Time 1.50 min Loop hold time 0.20 min, equilibration 0.20 min Injection time 0.20min
Sample Line Temperature	150°C
Transfer Line Temperature	150°C
Gas Chromatograph	GC-2016 Plus or 2020 NX
Injection	Split injection from HS-20, with 50:1 split ratio Rui 624 SI MS 30.0m x 0.25 mm x 1.40 um Helium carrier gas
Column	Constant linear velocity, 38.8cm/sec Column Flow 1.24mL/min Purge flow 0.2mL/min 50°C, hold 3.0 min 10°C/min to 140°C, hold 0.0 min 45°C/min to 200°C, hold 1.0 min Total GC run time 16.33 min Total GC cycle time: 25.00 mins
Oven Program	
Detector	GCMS-QP2010 SE or 2020 NX
Operation Mode	Selected Ion Monitoring Mode (SIM)
Ion Source	200°C, EI mode, 70eV
Solvent Cut Time	0.1 min
MS Interface	300°C



Figure 1. Shimadzu GCMS-QP2020 NX with HS-20 autosampler.

4. Calibration Curve and QC Results

Figure 3. Top Left Panel – Representative image of Total Ion Chromatogram (TIC) showing the 20 Residual Solvent peaks analyzed in a standard sample. Bottom Left Panel – Zoomed-in version to show smaller solvent peaks. Right Panel – Representative images of selective ion monitoring (SIM) m/z ion peaks for each residual solvent analyzed in the standard sample and its associated calibration curve.

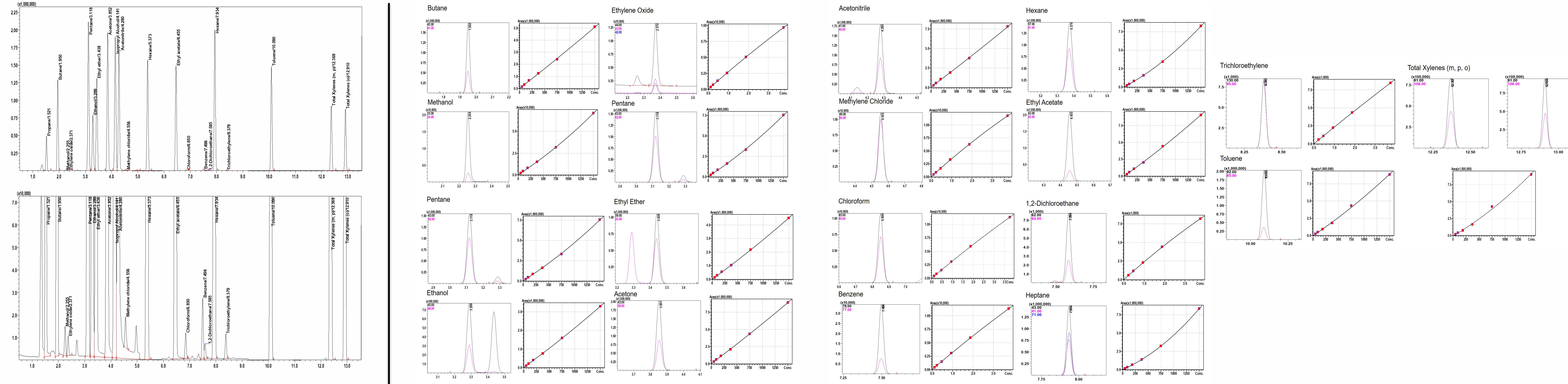


Table 3. Accuracy % and R² results for calibration curves.

Residual Solvents Calibration Curve Standards	Accuracy % (Criteria: 80-120% for Standards)				R ² (Criteria: ≥ 0.99)
	Lowest Level in Cal Curve (ug)	Result (%)	Highest Level in Cal Curve (ug)	Result (%)	
Propane	46.875	97.46	1500	100.40	0.9991
Butane	46.875	93.54	1500	100.61	0.9989
Methanol	46.875	92.07	1500	100.24	0.9996
Ethylene Oxide	0.117	93.56	3.75	100.15	0.9996
Pentane	46.875	91.19	1500	100.43	0.9993
Ethanol	46.875	94.71	1500	100.03	0.9997
Ethyl Ether	46.875	96.21	1500	99.95	0.9997
Acetone	46.875	94.54	1500	100.03	0.9996
Isopropanol	46.875	96.44	1500	99.85	0.9996
Acetonitrile	46.875	89.69	1500	100.47	0.9990
Methylene Chloride	0.117	99.04	3.75	100.07	0.9998
Hexane	46.875	91.67	1500	100.27	0.9996
Ethyl Acetate	46.875	97.59	1500	99.73	0.9994
Chloroform	0.117	100.12	3.75	100.00	1.0000
Benzene	0.117	99.58	3.75	99.99	0.9999
1,2-Dichloroethane	0.117	98.64	3.75	99.98	1.0000
Heptane	46.875	94.04	1500	100.10	0.9999
Trichloroethylene	0.117	100.67	3.75	99.93	0.9997
Toluene	46.875	104.01	1500	99.32	0.9984
Total Xylenes (m, p, and o)	46.875	109.97	1500	99.02	0.9964

Table 4. Recovery % results for spiked QC samples.

Residual Solvents	Recovery % (Criteria: 70-130% for spiked QCs)			
	ICV (level 5, %)	CCV (level 5, %)	LCS (level 5, %)	MS (level 5, %)
Propane	109.27	106.68	105.23	101.03
Butane	109.68	106.53	106.83	101.22
Methanol	109.11	105.78	88.00	83.33
Ethylene Oxide	106.81	101.09	94.18	87.46
Pentane	107.83	104.79	104.84	100.93
Ethanol	107.19	104.95	89.56	83.77
Ethyl Ether	104.80	101.74	100.72	97.28
Acetone	104.63	103.22	97.54	93.28
Isopropanol	104.34	102.80	92.49	88.08
Acetonitrile	108.30	107.54	93.25	88.30
Methylene Chloride	102.65	99.70	94.46	90.08
Hexane	106.91	104.14	106.12	102.74
Ethyl Acetate	100.91	100.51	100.27	96.96
Chloroform	100.92	99.77	89.86	83.86
Benzene	100.25	99.77	98.73	95.10
1,2-Dichloroethane	98.80	97.66	96.95	92.56
Heptane	102.68	101.70	108.45	105.40
Trichloroethylene	101.75	99.05	100.95	97.05
Toluene	95.80	96.28	99.92	96.64
Total Xylenes (m, p, and o)	91.29	92.71	98.08	95.36

5. Cannabis Sample Application*

The images shown on Figure 4 provide a visual example of the many varying types of concentrate and pre-roll samples that are received in the laboratory for the state-required residual solvent analysis. Headspace analysis is ideal for these types of samples because of the high volatility of residual solvents. These sample types can be weighed and analyzed directly in a headspace vial. Therefore, minimizing the amount of manual sample handling required and time spent in sample preparation.

Figure 4. Picture of some of the variety of concentrate and pre-roll samples received in cannabis testing laboratories for the analysis of residual solvents.



A representative portion of at least 250mg in sample size taken from the pre-roll sample shown in Figure 4 was weighed in a 20mL headspace vial for residual solvent headspace analysis using GC-MS. The results from the analysis are shown in Figure 5. Ethanol, methanol, acetone, isopropanol, and acetonitrile were identified in the sample, but with a quantitation result below the State regulated action level for each of these residual solvents. Therefore the sample passed the residual solvents test.

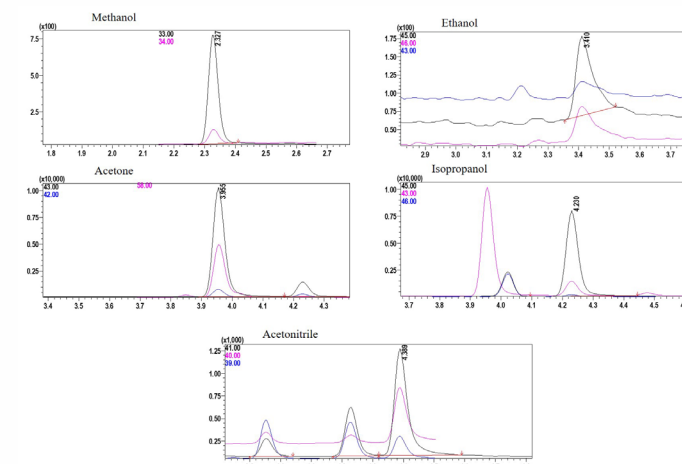


Figure 5. Representative GC-MS SIM chromatograms for the Category II residual solvents methanol, ethanol, acetone, isopropanol, and acetonitrile found in an aliquot sample from the pre-roll pictured in Fig. 4.

6. Conclusions

- In this poster, a GC-MS method using Selected Ion Monitoring (SIM) with headspace injection was developed for the analysis of cannabis concentrates. This includes the identification and use of appropriate standard mixes and diluent solvents for simple preparation of calibration curves, as well as method optimization to eliminate or minimize co-elution and ambient interferences for maximum sensitivity and accuracy in quantitation, all without compromising run time. Moreover, method parameters and hardware configuration changes were performed to accommodate high sample size requirements for compliant testing. These were conducted to avoid analytical column overloading and MS detector saturation, while directly testing sample without preparation steps.
- Results showed good coefficient of determination and accuracy % for calibrator standards. The curve fit type used is Quadratic with a 1/C weighing regression. All QC samples (Initial Calibration Verification, Continuing Calibration Verification, Laboratory Control Sample, and Laboratory Replicate Sample) showed good recovery %.

7. References

- "Analysis of Residual Solvents – Class 1, Class 2A, Class 2B in Pharmaceuticals using Headspace-GC/MS", Shimadzu's application note.
- "A Fast, Simple FET Headspace GC-FID Technique for Determining Residual Solvents in Cannabis Concentrates", Restek's application note.
- "Protocol for Quantitative Determination of Residual Solvents in Cannabis Concentrates", Restek's protocol.
- Current BCC Cannabis Regulations (January 2019), https://www.bcc.ca.gov/law_regs/cannabis_order_of_adoption.pdf
- Photos shown in Figure 4, and GC-MS data shown in Figure 5 are courtesy of Caligreen Cannabis Testing Laboratory in South California.