Quantifying Cannabinoids in Complex Matrices

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BACKGROUND AND SIGNIFICANCE

- The marijuana industry is regulated by the 2018 USDA Farm Bill which defines hemp as cannabis containing less than 0.3% (dry weight) delta-9-tetrahydrocannabinol (Δ9-THC)
- New regulations increase analytical burdens by requiring the quantification of Δ9-THC in complex matrices
- This is creating an unstainable testing environment for publicly funded laboratories
- New innovative solutions are needed to help meet new testing requirements because outsourcing of case work is costly
- Out of the box workflow solutions and well-validated methods enable labs to quickly implement and adopt new testing procedures to improve throughput, data quality and compliance
- This study validates an analytical test kit for high-throughput testing applications in plant, oil, and wax products using gas chromatography-flame ionization detection (GC-FID)
- QuantCaps™ test kits *are injectable-ready matrix-matched reference materials* used in drug analysis applications on GC or LC platforms. QuantCaps™ products are customized to the matrix, analytes, method, and platform of a laboratory's specific application
- QuantCaps™ test kits allow laboratories to standardize their instrument calibration, validation, and methods for routine testing of drugs in complex matrices

GOAL

Validate the QuantCaps™ process for extracting and analyzing Δ9-THC, Δ8-THC and CBD in plant material, waxes and oil using the criteria set forth in ASB 036: Standard Practices for Method Validation in Forensic Toxicology in terms of accuracy, precision, calibration models, sensitivity, carryover, interferences, matrix effects and analyte stability

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WHAT IS QuantCaps™

EXAMPLE QunatCaps™ VALIDATION KIT

CONTENTS	QTY	VOLUME
1 x set of Calibration vials in solution	3	60uL/vial
3 x sets of QC vials in solution	3	60uL/vial
1 x Matrix - Blank (Nettle Extract) in solution with IS	3	60uL/vial
1 x Matrix - Blank (Nettle Extract) in solution without IS	3	60uL/vial
5 x Pos. matrix effect vials (3-analyte mix + IS at 0.3% with nettle)	3	60uL/vial
1 x Bottle of Internal Std (0.5% testosterone) extraction solution	3	100uL/vial
1 Interference study set (10 single-analyte vials; 0.3% level/analyte; see table)	3	30ug/mL/vial
1 Interference solution (10-analyte mix at 0.3% level per analyte)	3	30ug/mL/vial
3 Optimization vials (CBD, delta-9 THC, delta-8 THC)	3	100ug/mL each
3 Solvent blank vials (MeOH:CH_2Cl_2 (9:1)) for carryover study	3	1mL per vial
1 Hemp CRM sample - supplied by Absolute Standards, Inc.	3	Neat Solid

ANALYTICAL PROCEDURE

Step 1: Analysis of pre-manufactured calibrators and second source QCs

Analyze pre-manufactured calibrators and second source QCs

- *Injectable-ready matrix-matched standards*
- *Injectable-ready second source QCs*
- *Both standards and QCs extracted per lab protocol*

Step 2: Unknown sample extraction

- Weigh 50 mg +/- 1mg of unknown sample
- Add 5mL of extraction solvent pre-manufactured with internal standard that is matched to the calibrators and QCs
- Equilibrate for 10 minutes with periodic vortexing (at least 2 times for 15 seconds)
- Decant/filter
- Analyze by GC-FID

GC-FID QUANTITATION METHOD

Instrumentation	Agilent 6890N GC with FID
Column	Agilent DB5-MS 30m 0.25 ID 0.25 film thickness
Gas	N_2 carrier gas split 50:1
Injection	1 uL
Temperature	Inlet 250°C, detector 320°C
Flow	Ramped flow 2.2 mL/min hold 4.3 min 2mL/min per min to 2.5 mL/mim
Temperature Program	Initial temp. 250°C ramp 25°C/min to 300°C hold 3 min

DISCUSSION & CONCLUSIONS

- Results show that validated testing procedures are accurate (% bias < 20%) and precise (% CV < 20%)
- Analytical reportable ranges meet statutory requirements for hemp products made with plant, oil or wax
- Pre-manufactured kits meet laboratory QA/QC requirements
- Kits are equipped with matrix matched, injectable-ready standards and second source quality controls
- Injectable-ready formats allow staff time to focus on unknown sample extractions
- Operational costs are reduced through time and material savings
- One week is required for each matrix to execute validation studies
- This study shows that the commercially available QuantCap™ test kits are suitable for meeting testing requirements associated with the 2018 USDA Farm Bill that regulates hemp products
- Method is easily transferable to GC-MS instrumentation
- QuantCaps™ kit is commercially available by PinPoint Testing

REFERENCES

1. Farm bill. (n.d.). USDA. https://www.usda.gov/farmbill
2. AAFS Standards Board. Standard Practices for Method Validation in Forensic Toxicology, ANSI/ASB Standard 036; Colorado Springs, CO, 2019.

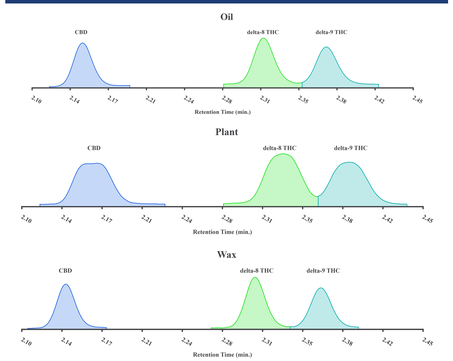
VALIDATION SUMMARY

METHOD PERFORMANCE CHARACTERISTICS

Matrix	Analyte	Average % Bias	Average Between-Run % CV	Average Within-Run % CV	r^2	Model	L.O.D (% by weight)	Carryover	Interference/ Specificity	Extract Stability (h)
OIL										
	CBD	-1.87	1.25	0.86	0.9998	Quadratic	0.10	No	Acceptable*	72
	Delta-8 THC	4.62	2.50	1.49	0.9997	Quadratic	0.10	No	Acceptable*	72
	Delta-9 THC	-7.68	1.83	1.30	0.9999	Quadratic	0.10	No	Acceptable*	72
PLANT									Acceptable*	72
	CBD	-0.91	4.62	4.24	0.9998	Quadratic	0.10	No	Acceptable*	72
	Delta-8 THC	9.52	4.75	4.24	0.9998	Quadratic	0.10	No	Acceptable*	72
	Delta-9 THC	-5.97	4.18	3.71	0.9998	Quadratic	0.10	No	Acceptable*	72
WAX									Acceptable*	72
	CBD	-1.37	2.53	1.33	0.9999	Quadratic	0.10	No	Acceptable*	72
	Delta-8 THC	10.45	1.96	1.30	0.9999	Quadratic	0.10	No	Acceptable*	72
	Delta-9 THC	-6.23	2.28	1.44	0.9999	Quadratic	0.10	No	Acceptable*	72

*Achieved chromatographic separation from CBC, CBDV, CBG, CBN, Delta-10 THC, Delta-8 THC, THCV.

CHROMATOGRAPHIC RESULTS



RESULTS