AOAC SMPR® 2017.019

Standard Method Performance Requirements (SMPRs®) for Quantitation of Cannabinoids in Edible Chocolate

Intended Use: Consensus-Based Reference Method for Use by Trained Technicians in a Laboratory for Routine Quality Assurance Testing

1 Purpose

AOAC SMPRs describe the minimum recommended performance characteristics to be used during the evaluation of a method. The evaluation may be an on-site verification, a single-laboratory validation, or a multi-site collaborative study. SMPRs are written and adopted by AOAC stakeholder panels composed of representatives from industry, regulatory organizations, contract laboratories, test kit manufacturers, and academic institutions. AOAC SMPRs are used by AOAC expert review panels in their evaluation of validation study data for method being considered for *Performance Tested Methods*℠ or AOAC *Official Methods of Analysis*℠, and can be used as acceptance criteria for verification at user laboratories.

2 Applicability

Identification and quantification of individual cannabinoids (as listed in Tables 1 and 2) in finished edible chocolate as listed in Table 3.

3 Analytical Technique

Any analytical technique(s) that measures the analytes of interest and meets the following method performance requirements is/are acceptable.

4 Definitions

Chocolate.—Any edible solid confection substantially consisting of "chocolate" (i.e., dark, milk, or white) without added inclusions.

Limit of quantitation (LOQ).—Minimum concentration or mass of analyte in a given matrix that can be reported as a quantitative result.

Quantitative method.—Method of analysis which response is the amount of the analyte measured either directly (enumeration in a mass or a volume) or indirectly (color, absorbance, impedance, etc.) in a certain amount of sample.

Recovery.—The fraction or percentage of spiked analyte that is recovered when the test sample is analyzed using the entire method.

Repeatability.—Variation arising when all efforts are made to keep conditions constant by using the same instrument and operator and repeating during a short time period. Expressed as the repeatability standard deviation (SD_r); or % repeatability relative standard deviation ($\%RSD_r$).

Reproducibility.—The standard deviation or relative standard deviation calculated from among-laboratory data. Expressed as the reproducibility standard deviation (SD_R); or % reproducibility relative standard deviation ($\%RSD_R$).

5 Method Performance Requirements

See Tables 4 and 5.

6 System Suitability Tests and/or Analytical Quality Control

Suitable methods will include blank check samples, and check standards at the lowest point and midrange point of the analytical range.

7 Reference Material(s)

See Tables 1 and 2 for examples of acceptable reference materials. Also *see:*

AOAC Technical Division on Reference Materials (TDRM) Database at http://www.aoac.org/aoac_prod_imis/AOAC_Member/MEMCF/TDCF/TDRMDbInfo.aspx

Restek Corp. Restek® Reference Standards at http://www.restek.com/Reference-Standards

Shimadzu Corp. Shimadzu Certified Reference Materials at http://www.ssi.shimadzu.com/cannabis_stds

Refer to Annex F: *Development and Use of In-House Reference Materials* in Appendix F: *Guidelines for Standard Method Performance Requirements*, *Official Methods of Analysis of AOAC INTERNATIONAL* (2016) 20th Ed., AOAC INTERNATIONAL, Rockville, MD, USA (http://www.eoma.aoac.org/app_f.pdf)

8 Validation Guidance

See "Clarification on Testing Materials for Cannabinoid SMPRs" (http://www.aoac.org/AOAC_Prod_Imis/AOAC_Docs/SPSFAM/ValidationGuidanceClarificationforSMPR2017_001and_002.pdf)

Method performance does not require demonstration of matrix homogeneity; the chocolate is viewed as the vehicle for cannabinoid delivery. Method performance should be demonstrated using a representative subsample of the matrix which assumes uniform distribution of cannabinoids.

Appendix D: *Guidelines for Collaborative Study Procedures to Validate Characteristics of a Method of Analysis*, *Official Methods of Analysis of AOAC INTERNATIONAL* (2016) 20th Ed., AOAC INTERNATIONAL, Rockville, MD, USA (http://www.eoma.aoac.org/app_d.pdf)

Appendix F: *Guidelines for Standard Method Performance Requirements*, *Official Methods of Analysis of AOAC INTERNATIONAL* (2016) 20th Ed., AOAC INTERNATIONAL, Rockville, MD, USA (http://www.eoma.aoac.org/app_f.pdf)

Appendix K: *Guidelines for Dietary Supplements and Botanicals*, *Official Methods of Analysis of AOAC INTERNATIONAL* (2016) 20th Ed., AOAC INTERNATIONAL, Rockville, MD, USA (http://www.eoma.aoac.org/app_k.pdf)

9 Maximum Time-to-Result

None

Approved by the AOAC Stakeholder Panel on Strategic Food Analytical Methods (SPSFAM) on September 24, 2017. Final Version Date: November 13, 2017.

Table 1. Required cannabinoids

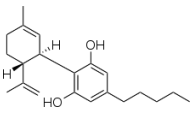
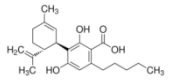
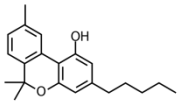
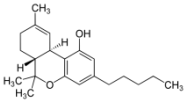
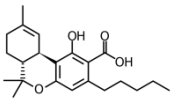
Common name	Abbreviation	IUPAC name	CAS No.	Molecular structure	Reference material
Cannabidiol	CBD	2-[(1*R*,6*R*)-6-isopropenyl-3-methylcyclohex-2-en-1-yl]-5-pentylbenzene-1,3-diol	13956-29-1		Restek Cerilliant Sigma-Aldrich API Standards Echo Pharm Lipomed AG
Cannabidiolic acid	CBDA	2,4-Dihydroxy-3-[(1*R*,6*R*)-3-methyl-6-prop-1-en-2-ylcyclohex-2-en-1-yl]-6-pentylbenzoic acid	1244-58-2		Cerilliant USP Restek Lipomed AG Echo Pharmaceutical
Cannabinol	CBN	6,6,9-Trimethyl-3-pentyl-benzo[*c*]chromen-1-ol	521-35-7		Cerilliant Restek
$^{\Delta 9}$Tetrahydro-cannabinol	THC	(−)-(6a*R*,10a*R*)-6,6,9-Trimethyl-3-pentyl-6a,7,8,10a-tetrahydro-6*H*-benzo[*c*]chromen-1-ol	1972-08-3		Cerilliant USP Echo Pharmaceuticals
Tetrahydro-cannabinolic acid	THCA	(6a*R*,10a*R*)-1-hydroxy-6,6,9-trimethyl-3-pentyl-6a,7,8,10a-tetrahydro-6*H*-benzo[*c*]chromene-2-carboxylic acid	23978-85-0		Cerilliant USP Echo Pharmaceuticals

Table 2. Additional, desirable cannabinoids

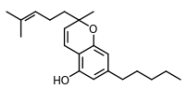
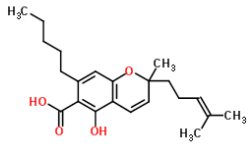
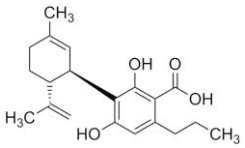
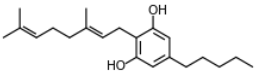
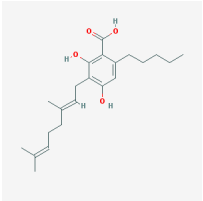
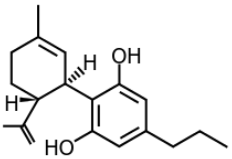
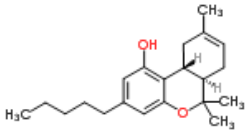
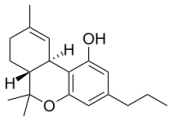
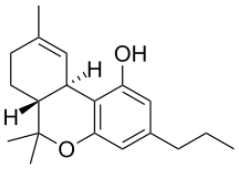
Name	Abbreviation	IUPAC name	CAS No.	Molecular structure	Reference material
Cannabichromene	CBC	2-Methyl-2-(4-methylpent-3-enyl)-7-pentyl-5-chromenol	20675-51-8		Cerilliant Sigma-Aldrich Echo Pharmaceuticals
Cannabichromenicacid	CBCA	5-Hydroxy-2-methyl-2-(4-methyl-3-penten-1-yl)-7-pentyl-2H-chromene-6-carboxylic acid	20408-52-0		Cerilliant
Cannabidivarinic acid	CBDVA	2,4-Dihydroxy-3-[(1*R*,6*R*)-3-methyl-6-prop-1-en-2-ylcyclohex-2-en-1-yl]-6-propylbenzoic acid	31932-13-5		Cerilliant
Cannabigerol	CBG	2-[(2*E*)-3,7-dimethylocta-2,6-dienyl]-5-pentyl-benzene-1,3-diol NIST: 1,3-Benzenediol, 2-(3,7-dimethyl-2,6-octadienyl)-5-pentyl-	25654-31-3 NIST: 2808-33-5		Cerilliant Lipomed AG Echo Pharmaceuticals SPEX Certiprep Tocris (UK)
Cannabigerolic acid	CBGA	3-[(2*E*)-3,7-dimethylocta-2,6-dienyl]-2,4-dihydroxy-6-pentylbenzoic acid	25555-57-1		Cerilliant Echo Pharmaceuticals SPEX Certiprep
Cannabidivarin	CBDV	2-((1*S*,6*S*)-3-methyl-6-(prop-1-en-2-yl) cyclohex-2-enyl)-5-propylbenzene-1,3-diol	24274-48-4		Cerilliant SPEX Certiprep
$^{\Delta 8}$Tetrahydro-cannabinol	$^{\Delta 8}$THC	6,6,9-Trimethyl-3-pentyl-6a,7,10,10a-tetrahydrobenzo[*c*] chromen-1-ol	5957-75-5		Cerilliant SPEX Certiprep
Tetrahydro-cannabivarin	THCV	6,6,9-Trimethyl-3-propyl-6a,7,8,10a-tetrahydro-6*H*-benzo[*c*]chromen-1-ol	28172-17-0		Cerilliant USP
Tetrahydrocannabivarin acid	THCVA		28172-17-0		Cerilliant

Table 3. Matrices

Chocolate chips
Chocolate bars
Chocolate truffles
White chocolate
Milk chocolate
Dark chocolate

Table 4. Method performance requirements (part 1) for individual cannabinoids

Parameter	Requirement
Limit of quantitation (LOQ) (% by weight)	≤0.008
Minimum analytical range (% by weight)	0.008–5[a]

[a] Lower concentrations may be acceptable as applicable for cannabinoids listed in Table 2.

Table 5. Method performance requirements (part 2) for individual cannabinoids[a]

	Range (% by weight)	
Parameter	≤0.008–1	>1
Recovery, %	90–110	95–105
RSD_r, %	≤5	≤4
RSD_R, %	≤8	≤5

[a] Analytical range based on following assumptions:
Smallest amount of cannabinoid in a serving: 10 mg
Largest amount of cannabinoid in a serving: 100 mg
Smallest serving size: 2.5 g
Largest serving size: 120 g
Lowest concentration: 10 mg/120 g = 0.008%
Highest concentration: 100 mg/2.5 g = 4%