Designation: D8399 – 23

Standard Test Method for Multi-residue Analysis of Pesticides in Dried Cannabis and Hemp Raw Materials Using Liquid Chromatography-Tandem Mass Spectrometry (LC-MS/MS)[1]

This standard is issued under the fixed designation D8399; the number immediately following the designation indicates the year of original adoption or, in the case of revision, the year of last revision. A number in parentheses indicates the year of last reapproval. A superscript epsilon (ε) indicates an editorial change since the last revision or reapproval.

1. Scope

1.1 This test method allows for the concentration determination of the pesticides listed in Table 1 and shall apply to any dried raw material from a cannabis plant (Note 1, Note 2) regardless of the type of cannabis plant from which it was derived (**1**, **2**).[2] For the sake of brevity, the term "cannabis" shall be used from now on to refer to any type of cannabis plant including those which can be classified as hemp. The procedure includes sub-sampling a ground, homogenous sample, liquid-solid extraction with acetonitrile:acetic acid (100:1, v:v), solid phase extraction with C18 SPE media, dilution in 3 mM ammonium formate in water with 0.1 % formic acid and 3 mM ammonium formate in methanol with 0.02 % formic acid in a 20:80 ratio (v:v) and analysis by LC-MS/MS. This procedure encompasses the entire process from sample preparation to analyte quantitation encompassing a range of 0.005 µg/g to 0.500 µg/g.

NOTE 1—For this test method, dried raw material from a cannabis plant includes one or more of inflorescence, leaves, or stems.

NOTE 2—Certain jurisdictions or regulations may require specific parts of the plant to be included or excluded for analysis and those regulations will take precedence for the selection of plant parts.

1.2 *Units*—The values stated in SI units are to be regarded as standard. No other units of measurement are included in this standard.

1.3 Table 1 lists the analytes measured by this test method.

1.4 No recommendations found within this test method shall preclude observance of federal, state, or local regulations, which may be more restrictive or have different requirements.

1.5 *This standard does not purport to address all of the safety concerns, if any, associated with its use. It is the responsibility of the user of this standard to establish appropriate safety, health, and environmental practices and determine the applicability of regulatory limitations prior to use.*

1.6 *This international standard was developed in accordance with internationally recognized principles on standardization established in the Decision on Principles for the Development of International Standards, Guides and Recommendations issued by the World Trade Organization Technical Barriers to Trade (TBT) Committee.*

2. Referenced Documents

2.1 *ASTM Standards:*[3]

D1193 Specification for Reagent Water

D8245 Guide for Disposal of Resin-Containing Cannabis Raw Materials and Downstream Products

D8270 Terminology Relating to Cannabis

D8282 Practice for Laboratory Test Method Validation and Method Development

3. Terminology

3.1 *Definitions*—For general terms related to cannabis, refer to Terminology D8270.

3.2 *Definitions of Terms Specific to This Standard:*

3.2.1 *blank, n*—a cannabis-only sample prepared from the same homogenous cannabis blend used to prepare the calibration curve, without the addition of internal standard working solution (ISWS).

3.2.2 *blank-0, n*—a cannabis-only sample prepared from the same homogenous cannabis blend used to prepare the calibration curve, with the addition of ISWS.

3.2.3 *blank-S, n*—a solvent blank prepared from only the mobile phases, without cannabis or analytes.

3.2.4 *growth regulator, n*—a class of chemical compounds used to modulate the growth and development of a crop.

3.2.5 *pesticide, n*—a product or substance used as a means to reduce or mitigate pests such as insects, fungus, bacteria, and weeds.

[1] This test method is under the jurisdiction of ASTM Committee D37 on Cannabis and is the direct responsibility of Subcommittee D37.03 on Laboratory.

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[2] The boldface numbers in parentheses refer to the list of references at the end of this standard.

[3] For referenced ASTM standards, visit the ASTM website, www.astm.org, or contact ASTM Customer Service at service@astm.org. For *Annual Book of ASTM Standards* volume information, refer to the standard's Document Summary page on the ASTM website.

TABLE 1 List of Measureable Analytes

Abamectin	Daminozide	Imidacloprid	Pyraclostrobin
Acephate	Deltamethrin	Iprodione	Pyrethrins
Acequinocyl	Diazinon	Kresoxim Methyl	Pyridaben
Acetamiprid	Dichlorvos	Malathion	Resmethrin
Aldicarb	Dimethoate	Metalaxyl	Spinetoram
Allethrin	Dimethomorph	Methiocarb	Spinosad
Azadirachtin	Dinotefuran	Methomyl	Spirodiclofen
Azoxystrobin	Dodemorph	Methoprene	Spiromesifen
Benzovindiflupyr	Ethoprophos	Mevinphos	Spirotetramat
Bifenazate	Etofenprox	Myclobutanil	Spiroxamine
Bifenthrin	Etoxazole	Naled	Tebuconazole
Boscalid	Fenoxycarb	Novaluron	Tebufenozide
Buprofezin	Fenpyroximate	Oxamyl	Teflubenzuron
Carbaryl	Fensulfothion	Paclobutrazol	Tetrachlorvinphos
Carbofuran	Fenthion	Permethrins	Tetramethrin
Chlorantraniliprole	Fenvalerate	Phenothrin	Thiacloprid
Chlorpyrifos	Fipronil	Phosmet	Thiamethoxam
Clofentezine	Flonicamid	Piperonylbutoxide	Thiophanate Methyl
Clothianidin	Fludioxonil	Pirimicarb	Trifloxystrobin
Coumaphos	Fluopyram	Prallethrin	
Cyantraniliprole	Hexythiazox	Propiconazole	
Cyprodinil	Imazalil	Propoxur	

Pyrethrins = Pyrethrin I and II
Spinetoram = Spinetoram J and L
Spinosad = Spinosyn A and D

3.2.5.1 *Discussion*—May be used in gardens, crops, woodlands or recreational areas, these are fungicides, insecticides, herbicides, and biocontrols approved for use by regulatory authorities to control pests harmful to plants, humans, or animals.

3.3 *Abbreviated Terms, Acronyms, and Initialisms*

3.3.1 *HPLC, n*—high performance liquid chromatography

3.3.2 *ICS, n*—independent check standard

3.3.3 *ISWS, n*—internal standard working solution

3.3.4 *LOD, n*—limit of detection

3.3.5 *LOQ, n*—limit of quantitation

3.3.6 *MCS, n*—master calibration standard

3.3.7 *MS, n*—mass spectrometer

3.3.8 *MS/MS, n*—tandem mass spectrometry

3.3.9 *QMS, n*—quality management system

3.3.10 *SPE, n*—solid phase extraction

3.3.11 *SRM, n*—selective reaction monitoring

4. Summary of Test Method

4.1 The quantitative analysis of pesticides in dried cannabis is performed by liquid-solid extraction into acetonitrile:acetic acid (100:1, v:v) followed by solid phase extraction and dilution prior to quantitative HPLC-MS/MS analysis.

4.2 Pesticides are identified by retention time and by selective reaction monitoring (SRM) transitions. An SRM transition consists of a pseudo-molecular ion, selected in quadrupole one, and a product ion, selected in quadrupole three. Pseudo-molecular ions are fragmented to product ions in quadrupole two (collision cell). The product ion selected in quadrupole three is transmitted to the detector of the mass spectrometer to produce a signal, resulting in a peak for the pesticide in the chromatogram. Pesticides are quantitated using the designated quantitative SRM transition. The final result reported for each sample lists the concentration in cannabis.

5. Significance and Use

5.1 The analysis and reporting of pesticide content in all forms of cannabis raw material that is grown or processed or both, with the intent of ingestion or consumption, is required to address health and safety concerns, satisfy testing and labeling requirements, and meet the regulatory guidelines of various jurisdictions where cannabis has been legalized for ingestion or consumption for medicinal or recreational purposes, or both. This test method is useful in providing quantitative results for the analytes listed in Table 1, which is a subset of the pesticide testing requirements found in regulatory documents for cannabis, not limited to and including Canada, many U.S. states where legalization has occurred, and the European Pharmacopeia (**1-4**). This test method may be appropriate for additional pesticide and growth regulator analytes, which may be added to those of Table 1 provided validation is performed in accordance with Practice D8282. Analytes may be removed from Table 1 without additional validation.

6. Interferences

6.1 Contaminants present in solvents, reagents, glassware, and other apparatus or co-extracted from matrix, producing discrete artifacts or elevated baselines, have the potential to cause method interferences. All of these materials are demonstrated to be free from interferences by analyzing blank matrix samples under the same conditions as samples. A blank sample is used to evaluate potential interferences for the internal standards while a blank-0 sample is used to evaluate potential interferences for the analytes. A blank-S sample is used to evaluate potential interferences caused by LC contamination or analyte carryover.