

Standard Method Performance Requirements (SMPRs®) for Selected Pesticides in Color Additives from Crop-Based Sources

Intended Use: Surveillance and Monitoring by Trained Technicians

1 Purpose

What: AOAC *Standard Method Performance Requirements* (SMPRs®) are voluntary consensus standards developed in accordance with the AOAC policy, “AOAC Due Process for Development of AOAC Non-Method Consensus Standards and Documents.” SMPRs describe a scientific community’s recommended minimum method performance characteristics and analytical requirements for a specific method-related intended use.

Who: Drafted by AOAC working groups, SMPRs are adopted by AOAC by a consensus of stakeholders affiliated with its integrated science programs and projects, which are composed of volunteer subject matter experts representing academia, government, industry, and nonprofit sectors from around the world.

Use: AOAC uses SMPRs in its core science programs in which they are a resource for AOAC method experts, including expert review panels, in the evaluation of validation study data for methods submitted to the AOAC *Official Methods of Analysis*SM and AOAC *Performance Tested Methods*SM programs. Additionally, AOAC SMPRs may be used to provide acceptance criteria for the verification of methods and serve as a resource to guide method development and optimization.

2 Applicability

Quantitative analysis of selected pesticides (Table 1) in natural color additives manufactured from crop-based sources (Tables 2 and 3). Table 1 provides the required list of pesticides. Additional compounds could be included in the method.

3 Analytical Technique

Any analytical technique(s) that measures the analyte of interest and meets the following method performance requirements is/are acceptable. More than one analytical technique may be needed.

4 Definitions

Certified Reference Material (CRM).—A reference material characterized by a metrologically valid procedure for one or more specified properties, accompanied by a reference material certificate issued by an authoritative body that provides the value of the specified property, its associated uncertainty, and a statement of metrological traceability.

Limit of quantitation (LOQ).—Lowest level of analyte in a test sample that can be quantified at a specified level of precision.

Multiresidue method (MRM).—A method able to distinguish, followed by identification and/or quantification of, more than one pesticide residue in one analysis.

Recovery.—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 (in the same laboratory) and repeating during a short time

period. Expressed as the repeatability standard deviation (SD_r); or % repeatability relative standard deviation (%RSD_r).

Reproducibility.—Variation arising when identical test materials are analyzed in different laboratories by different operators on different instruments. 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 Table 4.

6 System Suitability Tests and/or Analytical Quality Control

Suitable methods will include blank check samples and appropriate check standards.

7 Validation Guidance

Validation must be conducted on the final processed color additive (not intermediates or raw materials) using at least one representative matrix from the aqueous extract and dry powder categories and one from each of the two subcategories (oleoresin and other) of the solvent extract category listed in Table 2.

Validation must be conducted at the target LOQ and at 10x the LOQ.

“Appendix D: Guidelines for Collaborative Study Procedures to Validate Characteristics of a Method of Analysis,” (2023) Latimer, Jr., G.W. (Ed.), *Official Methods of Analysis of AOAC INTERNATIONAL*, 22nd Ed., <https://doi.org/10.1093/9780197610145.005.004>

“Appendix F: Guidelines for Standard Method Performance Requirements,” (2023) Latimer, Jr., G.W. (Ed.), *Official Methods of Analysis of AOAC INTERNATIONAL*, 22nd Ed., <https://doi.org/10.1093/9780197610145.005.006>

“Appendix K: Guidelines for Dietary Supplements and Botanicals” (2023) Latimer, Jr., G.W. (Ed.), *Official Methods of Analysis of AOAC INTERNATIONAL*, 22nd Ed., <https://doi.org/10.1093/9780197610145.005.011>

U.S. Food and Drug Administration (May 2018) *Bioanalytical Method Validation Guidance for Industry*

European Commission Guidance Document (2021) *Analytical Quality Control and Method Validation Procedures for Pesticide Residues and Analysis in Food and Feed* (SANTE/11312/2021)

8 Reference Materials

Refer to “Annex F: Development and Use of In-House Reference Materials” in “Appendix F: Guidelines for *Standard Method Performance Requirements*” (2023) Latimer, Jr., G.W. (Ed.), *Official Methods of Analysis of AOAC INTERNATIONAL*, 22nd Ed., <https://doi.org/10.1093/9780197610145.005.006>

Methods submitted for this SMPR should utilize pesticide CRMs for instrument calibration and quality control of method when available from an ISO/IEC 17034:2016 accredited provider.

9 Maximum Time to Results

None.

Approved by AOAC Working Group on Color Additives from Natural Sources (Pesticide Residues Subgroup) on January 12, 2024. Final Version Date: December 5, 2023. Effective Date: January 12, 2024.

Table 1. Selected pesticides

CAS No.	Name
30560-19-1	Acephate
135410-20-7	Acetamiprid
131860-33-8	Azoxystrobin
149877-41-8	Bifenazate
82657-04-3	Bifenthrin
188425-85-6	Boscalid
69327-76-0	Buprofezin
63-25-2	Carbaryl
133-06-2	Captan ^a
10605-21-7	Carbendazim
500008-45-7	Chlorantraniliprole
122453-73-0	Chlorfenapyr (Alert)
2921-88-2	Chlorpyrifos
210880-92-5	Clothianidin
120116-88-3	Cyazofamid
68359-37-5	Cyfluthrin
52315-07-8	Cypermethrin
121552-61-2	Cyprodinil
333-41-5	Diazinon
119446-68-3	Difenoconazole
35367-38-5	Diflubenzuron
60-51-5	Dimethoate (Cygon)
110488-70-5	Dimethomorph
165252-70-0	Dinotefuran
153233-91-1	Etoxazole
114369-43-6	Fenbuconazole
126833-17-8	Fenhexamid
39515-41-8	Fenpropathrin
120068-37-3	Fipronil
158062-67-0	Fonicamid
272451-65-7	Flubendiamide
131341-86-1	Fludioxonil
239110-15-7	Fluopicolide
658066-35-4	Fluopyram
951659-40-8	Flupyradifurone
76674-21-0	Flutriafol
907204-31-3	Fluxapyroxad
35554-44-0	Imazalil
138261-41-3	Imidacloprid
173584-44-6	Indoxacarb
36734-19-7	Iprodione
91465-08-06	Lambda-cyhalothrin
330-55-2	Linuron
121-75-5	Malathion
374726-62-2	Mandipropamid
57837-19-1	Metalaxyl

Table 1. (continued)

CAS No.	Name
10265-92-6	Methamidophos
16752-77-5	Methomyl (Lannate)
161050-58-4	Methoxyfenozide
88671-89-0	Myclobutanil
116714-46-6	Novaluron
1113-02-6	Omethoate
52645-53-1	Permethrin (<i>cis</i> and <i>trans</i>)
51-03-6	Piperonyl butoxide
41198-08-7	Profenofos
24579-73-5	Propamocarb
60207-90-1	Propiconazole
175013-18-0	Pyraclostrobin
96489-71-3	Pyridaben
53112-28-0	Pyrimethanil
95737-68-1	Pyriproxyfen (F)
124495-18-7	Quinoxifen
187166-40-1	Spinetoram
148477-71-8	Spirodiclofen
203313-25-1	Spirotetramat
946578-00-3	Sulfoxaflor
107534-96-3	Tebuconazole
148-79-8	Thiabendazole
111988-49-9	Thiacloprid
153719-23-4	Thiamethoxam
141517-21-7	Trifloxystrobin
68694-11-1	Triflumizole

^a If the method is not able to reliably determine the parent compound but is able to analyze the corresponding metabolite (THPI), it will be considered by the expert review panel.

Table 2. Matrix categories

Category	Matrix subcategory	Representative matrices
Aqueous extracts		Grape color extract
		Grape skin extract (enocianina)
		Fruit juice
		Vegetable juice
		Spirulina extract
Dry powders		Butterfly pea flower extract
		Annatto extract
		Dehydrated beets (beet powder)
		Paprika
		Turmeric
Solvent extracts		Butterfly pea flower extract
		Fruit juice
		Vegetable juice
	Oleo-resin	Turmeric oleoresin
		Paprika oleoresin
	Other	Tomato lycopene extract; tomato lycopene concentrate
		Carrot oil
		Annatto extract

Table 3. Target matrices

Matrix ^a	21 CFR listing ^b	E No. ^c	Manufacturing process	Source
Annatto extract	73.30	E160b	Alkaline alcoholic or aqueous solvent extraction	Annatto seed
Dehydrated beets (beet powder)	73.40	E162	Dehydration of beets	Beets
Beta-carotene	73.95	E160a	Natural, not synthetic, product	Yellow, orange, and green leafy fruits and vegetables; algae; fungi
Grape color extract	73.169	E163	Aqueous extraction	Concord grapes
Grape skin extract (enocianina)	73.170	E163	Aqueous extraction	Grapes
Fruit juice	73.250		Aqueous extraction or dried powder	Fruit
Vegetable juice	73.260		Aqueous extraction or dried powder	Vegetables
Carrot oil	73.300		Hexane extraction from carrots	Carrots
Paprika	73.340	E160c	Ground dried pod of capsicum	Pod of capsicum
Paprika oleoresin	73.345	E160c	Solvent extraction from paprika	Capsicum
Spirulina extract	73.530		Aqueous extraction	Arthrospira platensis
Tomato lycopene extract; tomato lycopene concentrate	73.585	E160d	Ethyl acetate extraction from tomato pulp	Tomato
Turmeric	73.600	E100	Ground rhizome of <i>Curcuma longa</i> L.	Rhizome of curcuma
Turmeric oleoresin	73.615	Solvent extraction of turmeric	Rhizome of curcuma	Turmeric oleoresin
Butterfly pea flower extract	73.69	Aqueous extraction	Dried butterfly pea flowers	Butterfly pea flower extract

^a Natural source only.

^b U.S. regulations describing the color additive: <https://www.ecfr.gov/current/title-21/chapter-I/subchapter-A/part-73>.

^c E No. = Europe numbers, which are codes for substances used as food additives, including those found naturally in many foods, for use within the European Union (EU) and European Free Trade Association (EFTA).

Table 4. Method performance requirements

LOQ, mg/kg (ppm)	0.01–0.1
Analytical range	0.5 × LOQ to 100 × LOQ
Recovery, %	60–120 ^a
RSD _r , %	≤20
RSD _R , %	≤30

^a In exceptional cases, average recoveries outside the above range can be accepted, but shall lie within 50–130% and only when the precision criteria for RSD_r are met.