AOAC SMPR® 2022.002

Standard Method Performance Requirements (SMPRs®) for Determination of Folic Acid in Dietary Supplements

Intended Use: Reference Method for Compliance with FDA Dietary Supplement cGMPs or Other Relevant Regulatory Agencies

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-based integrated science programs and projects, which are composed of representatives and experts from the academic, government, industry, and nonprofit sectors. AOAC SMPRs are used by AOAC method review experts, including expert review panels, in their evaluation of validation study data for methods being considered for AOAC *Performance Tested Methods*℠, *Reviewed and Recognized*℠, or AOAC *Official Methods of Analysis*℠, and can be used as acceptance criteria for verification at user laboratories.

2 Applicability

Determination of folate in the forms of supplemental folic acid (CAS 59-30-3) and/or 5-methyl-tetrahydrofolate (CAS 68792-52-9), in all forms of dietary supplements (such as tablets, capsules, softgels, gelcaps, liquids, powders, chewable gels, or other forms) intended to be taken by mouth.

3 Analytical Technique

Any analytical technique that meets the following method performance requirements is acceptable.

4 Definitions

Dietary ingredients.—Vitamin, mineral, herb, or other botanical; an amino acid; a dietary substance for use by humans to supplement the diet by increasing total dietary intake; or a concentrate, metabolite, constituent, extract, or combination of any of the above dietary ingredients. {United States Federal Food Drug and Cosmetic Act §201(ff) [U.S.C. 321 (ff)]}

Dietary supplements.—Product for humans to be taken by mouth that contains a "dietary ingredient" intended to supplement the diet. Dietary supplements may be found in many forms [refer to USP <1151> Pharmaceutical Dosage Forms for definitions (usp.org)], such as tablets, capsules, gels, softgels, gelcaps, liquids, or powders.

Limit of detection (LOD).—Minimum concentration or mass of analyte that can be detected in a given matrix with no greater than 5% false-positive risk and 5% false-negative risk.

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

Recovery.—Fraction or percentage of spiked analyte that is 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.—SD or RSD 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 1.

6 System Suitability Tests and 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 Materials

U.S. National Institute of Standards and Technology (NIST) Standard Reference Material (SRM) 3280 and 3289—Multivitamin/Multielement Tablets.

SRM 3252, a protein drink mix, with an assigned folic acid value, is also one of the choices in the lack of the SRM 3280 and 3289.

8 Validation Guidance

"Appendix K: Guidelines for Dietary Supplements and Botanicals," *Official Methods of Analysis* (current edition), AOAC INTERNATIONAL, Rockville, MD, USA (http://www.eoma.aoac.org/app_k.pdf). Also at: *J. AOAC Int*. **95**, 268(2012); DOI: 10.5740/jaoacint.11-447

9 Maximum Time-to-Signal

No maximum time.

Developed by AOAC Working Group on Folic Acid. Approved by Stakeholders for Folic Acid in Botanical and Dietary Supplement Integrity Program. Final version date: June 2, 2022. Effective date: July 15, 2022.

Table 1. Method performance requirements

	Folic acid			5-Methyl-THF		
Analytical range, µg/g		50–1500			50–1500	
LOD, µg/g		15			15	
LOQ, µg/g		50			50	
Range, µg/g	<100	100–1000	>1000	<100	100–1000	>1000
Repeatability (RSD_r, %)[a]	≤10 (≤12)	≤7 (≤9)	≤5 (≤7)	≤10 (≤12)	≤7 (≤9)	≤5 (≤7)
Recovery, %[a]	90–110 (85–115)	95–105 (90–110)	97–103 (92–108)	80–110 (75–120)	85–115 (80–115)	90–110 (85–115)
Reproducibility (RSD_R, %)[a]	≤15 (≤18)	≤10 (≤12)	≤7 (≤10)	≤15 (≤18)	≤10 (≤12)	≤7 (≤10)

[a] Parentheses indicate requirements for difficult matrices (i.e., gummies, chewables).