

STANDARD METHOD PERFORMANCE REQUIREMENTS

Standard Method Performance Requirements (SMPRs®)

2018.001: Sugars in Animal Feed, Pet Food, and Human Food

Stakeholder Panel on Strategic Food Analytical Methods

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Intended Use: Reference/regulatory method to measure individual mono- and di-saccharides in animal feed, pet food, and human food

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 the 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 methods being considered for *Performance Tested Methods*SM or AOAC *Official Methods of Analysis*SM, and can be used as acceptance criteria for verification at user laboratories.

2 Applicability

Individually measure free nutritional sugars, minimally: fructose, galactose, glucose, sucrose, maltose, and lactose (*see* Table 1 for additional information on analytes) found in selected ingredients and foods consumed by animals, pets, and humans. The analytical method must account for potential interferences (*see* Table 2) in these matrices. Methods that target a narrower matrix scope will also be considered when appropriate.

3 Analytical Technique

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

4 Definitions

Animal and pet foods.—Material consumed or intended to be consumed by animals other than humans that contributes nutrition, taste, or aroma or has a technical effect on the consumed material. This includes raw materials, ingredients, and finished product. (AAFCO)

Human food.—Material consumed or intended to be consumed by adult humans. For the purpose of this SMPR, this does not include infant formulas nor adult nutritionals.

Ingredients.—The base materials used in the formulation of animal food and human food.

Recovery.—The fraction or percentage of analyte that is measured 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 laboratory 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).

Sugars.—The sugars in the analytical scope are minimally: fructose, galactose, glucose, sucrose, maltose, and lactose.

5 Method Performance Requirements

See Table 3.

6 System Suitability Tests and/or Analytical Quality Control

Suitable methods will include blanks, and appropriate check standards.

7 Validation Guidance

Method data packages must include performance data covering the method claimed matrixes. A useful tool can be found in food pyramid concept described in Annex F of Appendix F the *Official Methods of Analysis of AOAC INTERNATIONAL*. Matrix variations in non-sugar carbohydrate and moisture content must be investigated and data provided. For example, using the food pyramid in sector 7, a matrix may be either high in sugar or high in non-sugar carbohydrates (e.g., starch, sugar alcohols), each of which may affect method performance.

Method data packages must include relevant data regarding interferences and instabilities, such as listed in Table 2. Data packages must include data to demonstrate that the sugar composition in the extract is a representation of the true sugar composition in the food product and assess the stability of extracts over the duration of the test.

8 Reference Materials

Potential reference materials, depending on method scope, may include the following:

National Institute of Standards and Technology (NIST):

3233: Fortified breakfast cereal

2383A: Baby food composite

3282: Low-calorie cranberry juice cocktail

1849a: Infant/adult nutritional formula

Institute for Reference Materials and Measurements (IRMM):

BCR644: Artificial foodstuff

Millipore Sigma (formerly Sigma-Aldrich):

1181302: Dextrose, United States Pharmacopeia (USP) Reference Standard

286504: Fructose, USP Reference Standard

1356701: Lactose monohydrate, USP Reference Standard

1623637: Sucrose, USP Reference Standard

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

9 Maximum Time-to-Results

None.

Table 1. Additional information on analytes			
Common name	IUPAC name	CAS No.	Molecular structure
Fructose	(3 <i>S</i> ,4 <i>R</i> ,5 <i>R</i>)-1,3,4,5,6-pentahydroxyhexan-2-one	57-48-7	
Galactose	(3 <i>R</i> ,4 <i>S</i> ,5 <i>R</i> ,6 <i>R</i>)-6-(hydroxymethyl)oxane-2,3,4,5-tetrol	59-23-4	
Glucose	(2 <i>R</i> ,3 <i>S</i> ,4 <i>R</i> ,5 <i>R</i>)-2,3,4,5,6-pentahydroxyhexanal	50-99-7	
Sucrose	(2 <i>R</i> ,3 <i>R</i> ,4 <i>S</i> ,5 <i>S</i> ,6 <i>R</i>)-2-[(2 <i>S</i> ,3 <i>S</i> ,4 <i>S</i> ,5 <i>R</i>)-3,4-dihydroxy-2,5-bis(hydroxymethyl)oxolan-2-yl]oxy-6-(hydroxymethyl)oxane-3,4,5-triol	57-50-1	
Maltose	2-(Hydroxymethyl)-6-[4,5,6-trihydroxy-2-(hydroxymethyl)oxan-3-yl]oxyoxane-3,4,5-triol	69-79-4	
Lactose	β -D-galactopyranosyl-(1 \rightarrow 4)-D-glucose	63-42-3	

Table 2. Potential interferants

Nontarget mono, di- and tri- saccharides (e.g., maltotriose interfering with maltose)
Enzymatic activity (amylase, invertase)
Hydrolytic activity (e.g., acidic)
Organic acids
Glucose with higher degrees of polymerization
Sugar alcohols, such as glycerol, erythritol, xylitol, sorbitol, mannitol, maltitol, lactitol, isomalt
Hydroxylated compounds (nontargeted carbohydrates, sugar alcohols, sugar acids, sucralose, etc.)
Salts, such as sodium chloride
Amine containing compounds (glucosamine HCl, amino acids, peptides, glycoproteins, etc.)

Table 3. Recovery, repeatability, and reproducibility parameters^a

Analytical range, %	0.1–5	>5–50	>50–100
Recovery, %	90–110	95–105	97–103
RSD _r , %	≤7	≤5	≤3
RSD _R , %	≤10	≤8	≤4

^a Reported as the individual sugars (fructose, glucose, sucrose, maltose, lactose, and galactose).