

# Reference Material 8666

## Ginger (*Zingiber officinale*) Extract

### REFERENCE MATERIAL INFORMATION SHEET

**Purpose:** This Reference Material (RM) is intended primarily for use in evaluating analytical methods for the determination of elements, gingerols, and shogaols in ginger extracts and similar matrices. RM 8666 provides a common matrix to those in the botanical supplements and natural products communities who wish to conduct testing or research studies. This RM is extracted from a portion of the authenticated ground rhizome material used in the preparation of SRM 3398. A unit of RM 8666 consists of five packets, each containing approximately 3 g of *Zingiber officinale* (ginger rhizome) extract.

**Non-Certified Values:** NIST non-certified values do not meet the NIST criteria for certification [1] and are the best estimates of the true values based on available data. The values are provided with an uncertainty that may reflect only measurement reproducibility, may not include all sources of uncertainty, and/or may reflect a lack of sufficient statistical agreement among multiple analytical methods.

Non-certified mass fraction values for analytes in RM 8666, reported on a dry-mass basis, are provided in Table 1. Values are expressed as  $x \pm U_{95\%}(x)$ , where  $x$  is the non-certified value and  $U_{95\%}(x)$  is the expanded uncertainty of the non-certified value. The method-specific value of the analyte lies within the interval  $x \pm U_{95\%}(x)$  with 95 % confidence. To propagate this uncertainty, the non-certified value should be treated as a normally distributed random variable with mean  $x$  and standard deviation  $U_{95\%}(x)/2$  [2–4]. The measurands are the total mass fraction of each analyte in Table 1. Metrological traceability is to the International System of Units (SI) derived unit for mass fraction (expressed as micrograms per kilogram (elements) or milligrams per gram (gingerols and shogaols)) as realized by the methods used [2].

**Table 1. Non-Certified Mass Fraction Values for Gingerols, Shogaols, and Elements in RM 8666**

	Mass Fraction (mg/g)
6-gingerol	23.90 ± 0.39
8-gingerol	3.806 ± 0.082
10-gingerol	4.751 ± 0.056
6-shogaol	5.554 ± 0.075
8-shogaol	0.980 ± 0.033
10-shogaol	1.645 ± 0.038
	Mass Fraction (µg/kg)
Arsenic (As)	43.7 ± 5.4
Mercury (Hg)	8.3 ± 2.6
Lead (Pb)	99.8 ± 7.0

**Period of Validity:** The non-certified values are valid within the measurement uncertainty specified until **30 January 2030**. The value assignments are nullified if the material is stored or used improperly, damaged, contaminated, or otherwise modified.

**Maintenance of Non-Certified Value:** NIST will monitor this material to the end of its period of validity. If substantive technical changes occur that affect the certification before the expiration of this certificate, NIST will notify the purchaser. Registration (see attached sheet or register online) will facilitate notification.

## NOTICE AND WARNING TO USERS

RM 8666 IS INTENDED FOR RESEARCH USE; NOT FOR HUMAN CONSUMPTION.

**Storage and Handling:** RM 8666 should be stored at controlled room temperature (20 °C to 25 °C) in the original unopened packet until required for use. For elemental analyses the packet can be opened for removal of test portions and then resealed until the material reaches its expiration date. For gingerol and shogaol analysis, the packet can be opened for removal of test portions, resealed, and stored under refrigeration for one week after the packet was initially opened.

**Use:** Before use, the contents of a packet of material should be mixed thoroughly. To relate analytical determinations to the values in this Reference Material Information Sheet, the following mass used for NIST analyses should be used as the minimum sample size to ensure valid results: 250 mg for gingerols and shogaols (see “Source, Preparation, and Analysis” in Appendix B). Test portions should be analyzed as received and results converted to a dry-mass basis. The moisture conversion factor given below (see “Determination of Moisture”) can be used for the sample(s) when using an unopened packet for the first time. If using a previously opened and resealed packet, moisture must be determined using one of the recommended techniques described below. Analytical results should include their own estimates of uncertainty and can be compared to the reference values using procedures described in reference 5.

**Determination of Moisture:** Moisture content of RM 8666 was determined at NIST by (1) drying over magnesium perchlorate in a desiccator at room temperature for 42 d and (2) drying for 3 h in a forced-air oven at 90 °C. The means from both techniques were averaged to determine a dry-mass proportion of (0.9329 ± 0.0028) gram dry-mass per gram as-received mass; the uncertainty shown on this value is an expanded uncertainty to represent a 95 % level of confidence. The conversion factor used to convert data from an as-received to a dry-mass basis is the inverse of the dry-mass proportion. A relative uncertainty component of 0.15 % for the conversion factor obtained from the moisture measurements is incorporated in the uncertainties of the assigned values, reported on a dry-mass basis, that are provided in this report.

## REFERENCES

- [1] Beauchamp, C.R.; Camara, J.E.; Carney, J.; Choquette, S.J.; Cole, K.D.; DeRose, P.C.; Duewer, D.L.; Epstein, M.S.; Kline, M.C.; Lippa, K.A.; Lucon, E.; Phinney, K.W.; Polakoski, M.; Possolo, A.; Sharpless, K.E.; Sieber, J.R.; Toman, B.; Winchester, M.R.; Windover, D.; *Metrological Tools for the Reference Materials and Reference Instruments of the NIST Material Measurement Laboratory*; NIST Special Publication 260-136; U.S. Government Printing Office: Washington, DC (2020); available at <https://nvlpubs.nist.gov/nistpubs/SpecialPublications/NIST.SP.260-136-2020.pdf> (accessed Jan 2021).
- [2] Thompson, A.; Taylor, B.N.; *Guide for the Use of the International System of Units (SI)*; NIST Special Publication 811; U.S. Government Printing Office: Washington, DC (2008); available at <https://www.nist.gov/pml/special-publication-811> (accessed Jan 2021).
- [3] JCGM 100:2008; *Evaluation of Measurement Data — Guide to the Expression of Uncertainty in Measurement (GUM 1995 with Minor Corrections)*; Joint Committee for Guides in Metrology (JCGM) (2008); available at [https://www.bipm.org/utls/common/documents/jcgm/JCGM\\_100\\_2008\\_E.pdf](https://www.bipm.org/utls/common/documents/jcgm/JCGM_100_2008_E.pdf) (accessed Jan 2021); see also Taylor, B.N.; Kuyatt, C.E.; *Guidelines for Evaluating and Expressing the Uncertainty of NIST Measurement Results*; NIST Technical Note 1297; U.S. Government Printing Office: Washington, DC (1994); available at <https://www.nist.gov/pml/nist-technical-note-1297> (accessed Jan 2021).
- [4] JCGM 101:2008; *Evaluation of Measurement Data – Supplement 1 to the “Guide to the Expression of Uncertainty in Measurement” — Propagation of Distributions using a Monte Carlo Method*; JCGM (2008); available at [https://www.bipm.org/utls/common/documents/jcgm/JCGM\\_101\\_2008\\_E.pdf](https://www.bipm.org/utls/common/documents/jcgm/JCGM_101_2008_E.pdf) (accessed Jan 2021).
- [5] Sharpless, K.E.; Lippa, K.A.; Duewer, D.L.; Rukhin, A.L.; *The ABCs of Using Standard Reference Materials in the Analysis of Foods and Dietary Supplements: A Practical Guide*; NIST Special Publication 260-181; U.S. Government Printing Office: Washington, DC (2014); available at <https://www.nist.gov/sites/default/files/documents/srm/SP260-181.pdf> (accessed Jan 2021).
- [6] Efron, B.; Tibshirani, R. J.; *An Introduction to the Bootstrap*, Chapman & Hall UK (1993).
- [7] Searle, S.; Casella, G.; McCulloch, C.; *Variance Components*; John Wiley: Hoboken, NJ (1992).
- [8] Rukhin, A.L.; Possolo, A.; *Laplace Random Effects Models for Interlaboratory Studies*; Computational Statistics and Data Analysis, Vol. 55, pp. 1815–1827 (2011).

*Certain commercial equipment, instruments or materials may be identified in this Reference Material Information Sheet to adequately specify the experimental procedure. Such identification does not imply recommendation or endorsement by the National Institute of Standards and Technology, nor does it imply that the materials or equipment identified are necessarily the best available for the purpose.*

*Users of this RM should ensure that the Reference Material Information Sheet in their possession is current. This can be accomplished by contacting the Office of Reference Materials 100 Bureau Drive, Stop 2300, Gaithersburg, Maryland 20899-2300; telephone (301) 975-2200; e mail [srminfo@nist.gov](mailto:srminfo@nist.gov); or the Internet at <https://www.nist.gov/srm>.*

**\*\*\*\*\* End of Reference Information Sheet\*\*\*\*\***

# Appendix A

## RESPONSIBILITIES

**Coordination:** C.A. Rimmer and L.J. Wood of the NIST Chemical Sciences Division.

The development of RM 8666 was a collaboration among the National Institute of Standards and Technology (NIST), the National Institutes of Health Office of Dietary Supplements (NIH-ODS), and the Food and Drug Administration Center for Drug Evaluation and Research (FDA CDER).

**Analytical Measurements:** S.H. Coskun, C.A. Rimmer, L.J. Wood of the NIST Chemical Sciences Division.

Analysts at the following laboratories performed measurements that contributed to the value assignment of analytes in RM 8666 as part of a Grocery Manufacturers Association (GMA) Food Industry Analytical Chemists Committee (FIACC) interlaboratory comparison exercise: Con Agra Foods (Omaha, NE, USA); Covance (Asia) Pte. Ltd. (Singapore); Covance Laboratories (Battle Creek, MI, USA); Covance Laboratories (Greenfield, IN, USA); Del Monte Foods (Walnut Creek, CA, USA); Eurofins Frontier Global Sciences (Bothell, WA, USA); Eurofins Nutrition Analysis Center (Des Moines, IA, USA); Eurofins Steins Vitamin Competence Center (Vejen, Denmark); Eurofins WEJ Contaminants GmbH (Hamburg, Germany), Krueger Food Labs (Chelmsford, MA, USA); Land O'Lakes (Arden Hills, MN, USA); Mérieux Nutrisciences Brasil (Sao Paulo, Brazil); Mérieux Nutrisciences China (Beijing, China); Microchem Silliker (Mumbai, India); NSF International (Ann Arbor, MI, USA); Silliker Canada Co., (Markham, ON, Canada); and The Coca-Cola Company (Shanghai, China).

**Statistical Analysis:** J.H. Yen of the NIST Statistical Engineering Division.

\* \* \* \* \* End of Appendix A \* \* \* \* \*

# Appendix B

## SOURCE, PREPARATION AND ANALYSIS

**Source and Preparation:** RM 8666 is a spray dried alcohol/aqueous extract of a portion of the same single harvest of *Zingiber officinale* (ginger rhizome) used in the preparation of SRM 3398. The powdered extract was transferred to High-Purity Standards (Charleston, SC) where it was blended, aliquoted, and heat-sealed inside nitrogen-flushed 4 mil polyethylene bags, which were then sealed inside nitrogen-flushed aluminized plastic bags along with two packets of silica gel each. Following packaging, RM 8666 was irradiated by Neutron Products, Inc. (Dickerson, MD) to an absorbed dose of 6.3 kGy to 8.4 kGy.

**Homogeneity Assessment:** The homogeneity of gingerols and shogaols was assessed at NIST using the methods and test portion sizes described below. No inhomogeneity was found by analyses of variance with 5 % significance level run on NIST data where box information was available.

**Analytical Approach for Determination of Gingerols and Shogaols:** Value assignment of the mass fractions of gingerols and shogaols in RM 8666 was based on the measurements provided by NIST using liquid chromatography with ultraviolet absorbance detection (LC/UV-absorbance).

*NIST Analyses for Gingerols and Shogaols using LC/UV-Absorbance:* The mass fractions of 6-gingerol, 8-gingerol, 10-gingerol, 6-shogaol, 8-shogaol, and 10-shogaol were measured by LC/UV-absorbance in duplicate 0.25 g test portions taken from each of ten packets of RM 8666. Formononetin was added to each test portion as an internal standard and gingerols and shogaols were extracted in 5 mL of methanol by ultrasonication for 60 min followed by 15 min of centrifugation. The supernatant was saved and the extraction process using only methanol was repeated with the supernatant added to the previous portion. LC separation followed by UV absorption was used as the detection method with analytes being monitored at 281 nm and 310 nm. A gradient mobile phase was used to separate the gingerols, shogaols, and formononetin. Four stock calibration solutions were prepared gravimetrically at levels intended to approximate the levels of the gingerols and shogaols in the RM following extraction. The purity of the gingerol and shogaol calibrant materials was evaluated at NIST using liquid chromatography with absorbance detection and by the manufacturers.

**Analytical Approach for Determination of Elements:** Value assignment of the mass fractions of toxic elements in RM 8666 was based on results from collaborating laboratories.

**Collaborating Laboratories' Analyses:** The GMA FIACC collaborating laboratories were asked to use their usual methods to make measurements on single test portions taken from each of two packets of RM 8666. Because of variability among data provided by laboratories participating in an interlaboratory comparison exercise the weighted median based on a Laplace random effects model [8] is used; the uncertainty is estimated using a bootstrap procedure based on a Laplace random effects model for the between-laboratory and within-laboratory effects [3,4,6–8].

\* \* \* \* \* End of Appendix B \* \* \* \* \*