National Institute of Standards & Technology

Certificate of Analysis

Standard Reference Material® 1577c

Bovine Liver

Standard Reference Material (SRM) 1577c consists of tissue derived from healthy steers. The material was collected and prepared under strict protocols designed to preserve the original composition, and to minimize contamination. SRM 1577c is intended primarily for use in evaluating the accuracy of analytical methods for selected elements in animal tissues and other biological materials. A unit of the SRM consists of one bottle containing 20 g of freeze-dried liver powder.

Certified Values: Certified values for the mass fraction content of 20 elements are provided in Table 1. The certified values are based on results from either a primary analytical technique carried out at NIST, or the combined results from two or more chemically independent analytical techniques obtained at NIST and collaborating expert laboratories [1]. A NIST certified value is a value for which NIST has the highest confidence in its accuracy in that all known or suspected sources of bias have been investigated or taken into account [1].

Reference Values: Reference values for the mass fraction content of eight additional elements are provided in Table 2. Reference values are non-certified values that are the best estimates of the true values. However, the values do not meet the NIST criteria for certification and are provided with associated uncertainties that may reflect only measurement precision, may not include all sources of uncertainty, or may reflect a lack of sufficient statistical agreement among multiple analytical methods [1].

Information Values: Information values for the mass fraction content of two elements are provided in Table 3. An information value is considered to be a value that will be of interest and use to the SRM user, but for which insufficient information is available to assess adequately the uncertainty associated with the value, or is a value derived from a limited number of analyses [1].

Expiration of Certification: The certification of **SRM 1577c** is valid, within the measurement uncertainties specified, until **01 October 2018**, provided the SRM is handled in accordance with the instructions given in this certificate (see "Instructions for Use"). The certification is nullified if the SRM is damaged, contaminated, or otherwise modified.

Maintenance of SRM Certification: NIST will monitor this SRM over the period of its certification. If substantive technical changes occur that affect the certification before the expiration of this certificate, NIST will notify the purchaser. Return of the attached registration card will facilitate notification.

The coordination of the investigations and technical measurements leading to the certification of this material was under the leadership of R. Zeisler of the NIST Analytical Chemistry Division.

Consultation on the statistical design of the experimental work and evaluation of the data was provided by S.D. Leigh of the NIST Statistical Engineering Division.

Support aspects involved in the preparation of this SRM were coordinated through the NIST Measurement Services Division.

Stephen A. Wise, Chief
Analytical Chemistry Division

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Certificate Issue Date: 15 June 2009

Robert L. Watters, Jr., Chief
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Collection and preparation of SRM 1577c were performed by M.P. Cronise and C.N. Fales of the NIST Measurement Services Division, and E.A. Mackey, R.O. Spatz, and R. Zeisler of the NIST Analytical Chemistry Division. The bovine liver material was collected at Texas A&M University (College Station, TX) with the assistance of W.D. James of the Center for Chemical Characterization and Analysis, and R.R. Riley of the E.M. (Manny) Rosenthal Meat Science and Technology Center.

The technical measurements were performed by S.J. Christopher, R.R. Greenberg, S.E. Long, E.A. Mackey, K.E. Murphy, B.J. Porter, S.A. Rabb, R.O. Spatz, B.E. Tomlin, L.J. Wood, L.L. Yu, and R. Zeisler of the NIST Analytical Chemistry Division, and the following collaborating laboratories and analysts: China Institute of Atomic Energy, Beijing, China: C. Xiao, B. Ni, W. Tian; Massachusetts Institute of Technology, Nuclear Reactor Laboratory, Cambridge, MA: J. Che, L.-W. Hu; Nuclear Physics Institute ASCR, Řež, Czech Republic: J. Kučera; Texas A&M University, College Station, TX, Department of Chemistry: W.D. James and College of Veterinary Medicine: R.J. Taylor; University of São Paulo, Institute of Chemistry, São Paulo, Brazil: C.S. Nomura, P.V. Oliveira; and USDA Beltsville Agricultural Research Center, Human Nutrition Research Center, Beltsville, MD: J. Harnly, E. Greene.

NOTICE AND WARNING TO USERS[1]

Storage: The material should be stored in its original container at room temperature (10 °C to 30 °C). SRM 1577c should not be exposed to intense sources of radiation, including ultraviolet light from lamps or sunlight.

Handling: This material was derived from healthy steers. These animals were inspected by a Veterinary Medical Officer and did not show signs of infectious, contagious, and/or communicable disease. Normal caution and care should be exercised during the material's handling and use. Users should be aware of sources of contamination. To avoid contamination a Class 100 clean-air environment is recommended.

Instructions for Use: Prior to removal of test portions for analysis, the contents of the bottles should be mixed. **The recommended minimum size is 100 mg**; see "Homogeneity Assessment" below. The mass fractions of constituents in SRM 1577c are reported on a dry-mass basis. Desiccator drying over $CaSO_4$ (e.g., Drierite) to stable mass (approximately 10 days) is recommended.

PREPARATION AND ANALYSIS

Sample Collection and Preparation: The liver tissue was collected and processed under observation of principles for "true and representative" sampling as documented in the protocols for human and marine mammal tissues of the National Biomonitoring Specimen Bank [2]. The liver tissue was harvested from 31 steers that were slaughtered at Texas A&M University College of Veterinary Medicine. This material is intended for "in vitro" diagnostic use only. The supplier of this material has reported that this material was produced under sanitary conditions and was derived from clinically healthy animals. The animals were slaughtered for the purpose of teaching bovine anatomy and how to butcher. The meat from these animals was prepared for retail under the supervision of a State of Texas meat inspector to ascertain the health of the animals. The livers were excised whole, placed on a clean Teflon sheet, and inspected. Each liver was rinsed with HPLC-grade water to remove excess blood, bile, and any other extraneous material. The outer membrane and major blood vessels were removed with titanium blade knives, and the tissue was cut into portions of approximately 10 cm^3; 120 kg of fresh tissue was obtained from this process, frozen in clean Teflon bags, and then shipped to NIST. The tissue was thawed and homogenized at NIST with a food processor equipped with titanium blades. The resulting paste was poured into glass trays, frozen, and lyophilized. The dry material was blended again in the food processor before being jet-milled. The resulting fine powder was radiation sterilized and bottled.

Homogeneity Assessment: The homogeneity of SRM 1577c was assessed by analyzing test portions of approximately 100 mg with high-precision instrumental neutron activation analysis (INAA).

Twelve bottles were randomly selected from the lot and two 100 mg test portions were taken for INAA from different locations in each bottle. The results for all elements reported by INAA (see Tables 1 and 2) did not reveal

[1] Certain commercial equipment, instruments, or materials are identified in this report to specify adequately 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.

any significant components of uncertainty due to heterogeneity; therefore **the recommended minimum sample size is 100 mg**. The values and uncertainties reported in this Certificate are valid for a 100 mg minimum sample size. Analysis of smaller amounts may be subject to additional uncertainties due to heterogeneity.

Analyses with solid-sampling graphite furnace atomic absorption spectrometry (SS-GFASS) using test portions in the range of 20 µg to 70 µg showed homogeneity for distribution of Cd, Cu, Pb, and Zn within the uncertainty of the method. For 1 mg test portions, an uncertainty component from heterogeneity of 1 % to 2 % relative was estimated from the experimental data for these elements.

Analytical Approach: All elements for which certified and reference values are provided were determined by using at least one of the following methods carried out at NIST: INAA, radiochemical neutron activation analysis (RNAA), prompt gamma activation analysis (PGAA), pre-concentration and pre-separation neutron activation analysis (PNAA), inductively coupled plasma mass spectrometry (ICP-MS), and ICP optical emission spectrometry (ICP-OES). NIST values for Cd, Pb, and Se were obtained by using isotope dilution (ID) ICP-MS, and for Hg by isotope dilution cold vapor (ID/CV) ICP-MS. The measurements were complemented by results provided by collaborating scientists from research laboratories using ICP-MS, ICP-OES, INAA, RNAA, and SS-GFAAS.

Certified Values and Uncertainties: Certified values were derived from the NIST analytical results and the results provided by collaborating laboratories. The uncertainty listed with each value is an expanded uncertainty, with coverage factor 2 (approximately 95 % confidence). The reporting follows the ISO Guide to the Expression of Uncertainty in Measurements [3,4,5].

For each element, there is a NIST result with an uncertainty that is complete in terms of coverage of recognized sources of uncertainties. Except for the elements measured by a single NIST primary method, these results are combined with results with similarly complete uncertainties from collaborating laboratories, and in certain cases several results without complete uncertainties. The uncertainties of these results were augmented for probable bias on the basis of the differences among the results obtained by different methods [3].

Table 1. Certified Values for Mass Fractions (on a Dry-Mass Basis) of Selected Elements

Element	Unit	Mass Fraction		Element	Unit	Mass Fraction	
Ag [A,B,b,C]	µg/kg	5.9	± 1.6	Mn [A,a,b,D,d]	mg/kg	10.46	± 0.47
As [C]	µg/kg	19.6	± 1.4	Mo [A,a,b,C,D,d]	mg/kg	3.30	± 0.13
Ca [A,a,D,d]	mg/kg	131	± 10	Na [A,a,d]	%	0.2033	± 0.0064
Cd [C,E]	µg/kg	97.0*	± 1.4	Ni [B,b,c]	µg/kg	44.5	± 9.2
Co [A,a,b]	mg/kg	0.300	± 0.018	Pb [E]	µg/kg	62.8	± 1.0
Cr [A]	µg/kg	53	± 14	S [a,D,d,F]	%	0.749	± 0.034
Cu [A,a,C,D,d]	mg/kg	275.2	± 4.6	Se [A,E]	mg/kg	2.031	± 0.045
Fe [A,a,D]	mg/kg	197.94	± 0.65	Sr [B,b,d]	µg/kg	95.3	± 4.2
K [A,a,d,F]	%	1.023	± 0.064	V [c,d,G]	µg/kg	8.17	± 0.66
Mg [A,a,D,d]	mg/kg	620	± 42	Zn [A,a,D,E,h]	mg/kg	181.1	± 1.0

*Alternate statistical method [6]

Analytical techniques used for assignment of certified values; capital letters indicate that the method was used by NIST.

[A,a] Instrumental neutron activation analysis (INAA)
[B,b] Inductively coupled plasma mass spectrometry (ICP-MS)
[C,c] Radiochemical neutron activation analysis (RNAA)
[D,d] Inductively coupled plasma optical emission spectrometry (ICP-OES)
[E] Isotope dilution inductively coupled plasma mass spectrometry (ID ICP-MS)
[F] Prompt gamma activation analysis (PGAA)
[G] Pre-concentration pre-separation neutron activation analysis (PNAA)
[h] Solid-sampling graphite furnace atomic absorption spectrometry (SS-GFAAS)
[I] Isotope dilution cold vapor inductively coupled plasma mass spectrometry (ID/CV ICP-MS)

Reference Values and Uncertainties: Reference values are based on results from one method carried out at NIST or at NIST and in several collaborating laboratories. The methods of combining the results of different methods from different laboratories were applied as above. These results do not fulfill the criteria for certification because they lack a full estimate of method bias. The reporting follows the ISO Guide to the Expression of Uncertainty in Measurement [5].

Table 2. Reference Values for Mass Fractions (on a Dry-Mass Basis) of Selected Elements

Element	Unit	Mass Fraction			Element	Unit	Mass Fraction		
Cl [A,a]	%	0.287	±	0.013	N [F]	%	10.30	±	0.34
Cs [A,a]	µg/kg	21.7	±	1.4	P [D,d]	%	1.175	±	0.027
H [F]	%	7.35	±	0.24	Rb [A,a]	mg/kg	35.3	±	1.1
Hg [I]	µg/kg	5.36	±	0.17	Sb [a,C]	µg/kg	3.13	±	0.31

Note: Analytical techniques used for assignment of reference values are provided following Table 1.

Information Values: Information values are given to assist users in the assays of two non-certified elements that may be of interest in method development and other investigations. These information values are based on results that did not allow complete assessment of all sources of uncertainty.

Table 3. Information Values for Mass Fractions (on a Dry-Mass Basis) of Selected Elements

Element	Unit	Mass Fraction
Li [d]	µg/kg	12
Si [d]	mg/kg	6

Note: Analytical techniques used for assignment of information values are provided following Table 1.

SUPPLEMENTAL INFORMATION

Particle size:

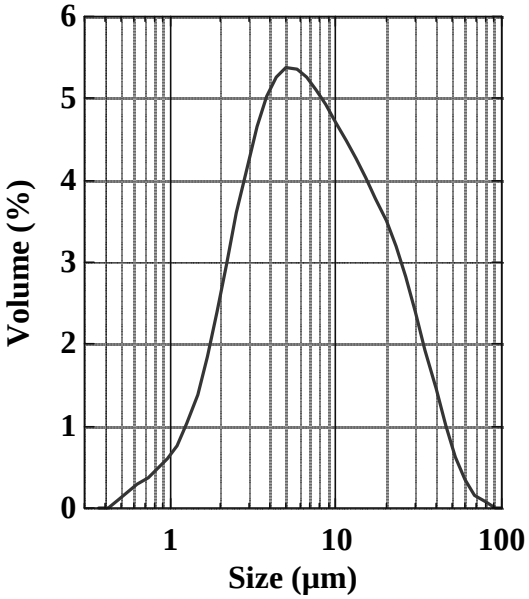

Figure 1. Particle size distributions in SRM 1577c determined in aqueous suspension via laser light scattering instrumentation (Malvern Mastersizer 2000). Calculated 10, 50, and 90 percentile particle sizes (percent volume of particles smaller than the value) for SRM 1577c are: $d_{0.1}$ = 2.31 µm, $d_{0.5}$ = 7.57 µm, $d_{0.9}$ = 28.5 µm. Uncertainties in these values are estimated at ± 10% relative (2s).

REFERENCES

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Users of this SRM should ensure that the certificate in their possession is current. This can be accomplished by contacting the SRM Program at: telephone (301) 975-2200; fax (301) 926-4751; e-mail srminfo@nist.gov; or via the Internet at http://www.nist.gov/srm.