National Institute of Standards & Technology

Certificate of Analysis

Standard Reference Material® 1588a

Organics in Cod Liver Oil

The concentration values (mass fraction) of SRM 1588a for PCB congeners, chlorinated pesticides, and additional analytes in cod liver oil supercede the corresponding values cited in SRM 1588.

This Standard Reference Material (SRM), a cod liver oil, is intended for use in developing and validating analytical methods for the determination of chlorinated biphenyls and chlorinated pesticides in cod liver oil or in other similar complex lipophilic matrices. SRM 1588a is the same material as SRM 1588 which was originally certified in 1989 for the concentrations of ten chlorinated pesticides, five PCB congeners, and alpha-tocopherol [1]. SRM 1588a updates the original certified values and provides certified and reference values for additional analytes. SRM 1588a consists of five sealed ampoules per unit, each ampoule containing approximately 1.2 mL of the cod liver oil.

Certified Concentration Values: Certified values, expressed as mass fractions [2], for 24 PCB congeners and 14 chlorinated pesticides found in the cod liver oil are provided in Tables 1 and 2. The certified values are based on the agreement of results obtained from two or more independent analytical techniques.

Reference Concentration Values: Reference values, also expressed as mass fractions, for alpha-tocopherol, 34 PCB congeners and 3 chlorinated pesticides found in cod liver oil are provided in Tables 3 and 4. The reference values are noncertified values that do not meet 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 methods. Explanations in support of each reference value are given as a note in each table.

Information Concentration Values: Information concentration values, expressed as mass fractions, for six polychlorodibenzo-*p*-dioxins and octachlorodibenzofuran added to the cod liver oil are provided in Table 5. These are noncertified values with no uncertainties reported as there is insufficient information to make an assessment of the uncertainties. The information values are given to provide additional characterization of the material.

Expiration of Certification: The certification of this SRM lot is valid until **31 October 2007**, within the measurement uncertainties specified, provided the SRM is handled and stored in accordance with the instructions given in this certificate (see Instructions for Use). The certification is nullified if the SRM is damaged, contaminated, or modified. Return of the attached registration card will facilitate notification.

Preparation for the original certificate was performed by R.M. Parris of the NIST Analytical Chemistry Division. The analyses for the update of the certificate were performed by M.M. Schantz and K.E. Sharpless of the NIST Analytical Chemistry Division and by K. Kaczanski, B. Grift, and D.C.G. Muir of the Department of Fisheries and Oceans (DFO) Canada, Freshwater Institute, Winnipeg, Manitoba.

The support aspects involved in the issuance of this SRM were coordinated through the Standard Reference Materials Program by B.S. MacDonald.

Gaithersburg, MD 20899
Certificate Issue Date: 29 May 1998

Thomas E. Gills, Chief
Standard Reference Materials Program

The coordination of the technical measurements leading to the recertification was under the direction of M.M. Schantz and S.A. Wise.

Consultation on the statistical design of the experimental work and evaluation of the data were provided by S.B. Schiller and L.M. Gill of the NIST Statistical Engineering Division.

NOTICE AND WARNING TO USERS

CAUTION: This material contains added polychlorodibenzo-*p*-dioxins and octachlorodibenzofuran. Each is present in this material at a concentration of 1 ng/g or less. Although some of these compounds are reported to be toxic and/or carcinogenic, their concentrations are too low to be considered hazardous under OSHA regulations. However, the material should be handled with care. Use proper disposal methods.

Instructions for Use: Samples of this SRM for analysis should be drawn from ampoules immediately after opening and used without delay for the certified values to be valid within the stated uncertainties. Certified values are not applicable to material in ampoules stored after opening, even if they are resealed. Sealed ampoules, as received, should be stored in the dark at temperatures less than 30 °C.

PREPARATION AND ANALYSIS

Preparation: This SRM is a cod liver oil that has been fortified with selected chlorinated dibenzodioxins and dibenzofurans. It was donated to NIST by the University of Ulm, Ulm, Germany.

Weighed aliquots of gravimetrically prepared 2,2,4-trimethylpentane solutions of six polychlorinated dibenzo-*p*-dioxins and octachlorodibenzofuran were added to a known mass of the cod liver oil. The concentrations of these components, which are included here for information only and are not certified values, were calculated from these gravimetric measurements and are listed in Table 5. The oil was homogenized by stirring for 10 h in a 9-L glass bottle. Each 2-mL amber ampoule was purged with argon just prior to the addition of approximately 1.2 mL of oil to the ampoule which was then flame sealed.

PCBs and Chlorinated Pesticides: SRM 1588a was analyzed for selected PCB congeners and chlorinated pesticides using gas chromatography with electron capture detection (GC-ECD) on two columns with different selectivity and using GC with mass spectrometric detection (GC/MS). This same approach has been used previously for the certification of PCBs and chlorinated pesticides in environmental matrices [3-5]. The majority of the lipid and biogenic material in weighed portions (0.8-1.2 g) of SRM 1588a was removed using size exclusion chromatography (SEC) on a preparative divinylbenzene-polystyrene column. The concentrated eluant was then fractionated on a semi-preparative aminopropylsilane column to isolate two fractions containing: 1) the PCBs and lower polarity pesticides and 2) the more polar pesticides. GC-ECD analyses of the two fractions were performed on two columns of different selectivities for PCB separations: 0.25 mm x 60 m fused silica capillary column with a 5 % phenyl-substituted methylpolysiloxane phase (0.25 µm film thickness) (DB-5, J&W Scientific, Folsom, CA) and a 0.32 mm x 100 m fused silica capillary column with a dimethylpolysiloxane phase containing 50 % (mole fraction) methyl C-18 (0.1 µm film thickness) (CP Sil 5 C18 CB, Chrompack International, Middelburg, The Netherlands). GC/MS analysis of the two fractions were performed on a 0.25 x 60 m fused silica capillary column with a 5 % phenyl-substituted methylpolysiloxane phase (0.25 µm film thickness) (DB-5MS, J&W Scientific, Folsom, CA).

Two PCB congeners (PCB 103 and PCB 198 [6,7]), not significantly present in the native cod liver oil, and perdeuterated (d_8) 4,4'-DDT were added as internal standards for quantification. Response factors for the analytes relative to the internal standards were determined by analyzing aliquots of SRM 2261, Chlorinated Pesticides in Hexane (Nominal Concentration 2 µg/mL); SRM 2262, Chlorinated Biphenyl Congeners in 2,2,4-Trimethylpentane (Nominal Concentration 2 µg/mL), gravimetrically prepared solutions of additional analytes not contained in SRMs 2261 and 2262, and the internal standards.

Additional PCB congeners and pesticides were measured at Department of Fisheries and Oceans (DFO) Canada using the following procedure. Cod liver oil (1.0 g) was dissolved in hexane and spiked with surrogate recovery standards of aldrin and octachloronaphthalene (OCN). A 100 mg lipid equivalent was chromatographed on Florisil (8 g 1.2 % deactivated with H_2O) to separate PCBs, chlorobenzenes, 4,4'-DDE and mirex (hexane elution) from most toxaphene components, chlordane-related compounds and 4,4'-DDT (second elution with dichloromethane:hexane 1:1). A third elution contained heptachlor epoxide and dieldrin. The extracts were analyzed by GC-ECD using a 0.25 mm x 60 m, 0.25 µm film thickness, 5 % phenyl-substituted methylpolysiloxane capillary column with hydrogen carrier gas. A volume correction standard (PCB 30) was added to the samples prior to GC analysis [8-10].

alpha-Tocopherol: The concentration of α-tocopherol was determined using two methods based on normal-phase (NP) and reversed-phase (RP) liquid chromatography (LC). Fluorescence detection (λ_{ex} = 295 nm; λ_{em} = 335 nm) was used for both methods. The NPLC analyses employed a semi-preparative aminopropylsilane column with quantification by the external standard method; aliquots of cod liver oil were diluted in hexane. The RPLC analyses employed a C_{18} column; the cod liver oil was diluted in ethyl acetate and ethanol and was spiked with tocol as an internal standard.

Table 1. Certified Concentrations (Mass Fractions) for Selected PCB Congeners in SRM 1588a[a,b]

		µg/kg
PCB 28	(2,4,4'-Trichlorobiphenyl)	28.32 ± 0.55
PCB 31	(2,4',5-Trichlorobiphenyl)	8.33 ± 0.28
PCB 44	(2,2',3,5'-Tetrachlorobiphenyl)	35.1 ± 1.4
PCB 49	(2,2',4,5'-Tetrachlorobiphenyl)	29.90 ± 0.84
PCB 52	(2,2',5,5'-Tetrachlorobiphenyl)	83.3 ± 2.3
PCB 66	(2,3',4,4'-Tetrachlorobiphenyl)	54.7 ± 1.5
PCB 87	(2,2',3,4,5'-Pentachlorobiphenyl)	56.3 ± 1.1
PCB 95	(2,2',3,5',6-Pentachlorobiphenyl)	36.5 ± 1.1
PCB 101	(2,2',4,5,5'-Pentachlorobiphenyl)	126.5 ± 4.3
PCB 105	(2,3,3',4,4'-Pentachlorobiphenyl)	60.2 ± 2.3
PCB 110	(2,3,3',4',6-Pentachlorobiphenyl)	76.0 ± 2.0
PCB 118	(2,3',4,4',5-Pentachlorobiphenyl)	176.3 ± 3.8
PCB 128	(2,2',3,3',4,4'-Hexachlorobiphenyl)	47.0 ± 2.4
PCB 138	(2,2',3,4,4',5'-Hexachlorobiphenyl)	263.5 ± 9.1
163	(2,3,3',4',5,6-Hexachlorobiphenyl)	
164	(2,3,3',4',5',6-Hexachlorobiphenyl)	
PCB 149	(2,2',3,4',5',6-Hexachlorobiphenyl)	105.7 ± 3.6
PCB 151	(2,2',3,5,5',6-Hexachlorobiphenyl)	54.8 ± 2.1
PCB 153	(2,2',4,4',5,5'-Hexachlorobiphenyl)	273.8 ± 7.7
PCB 156	(2,3,3',4,4',5-Hexachlorobiphenyl)	27.3 ± 1.8
PCB 170	(2,2',3,3',4,4',5-Heptachlorobiphenyl)	46.5 ± 1.1
PCB 180	(2,2',3,4,4',5,5'-Heptachlorobiphenyl)	105.0 ± 5.2
PCB 183	(2,2',3,4,4',5',6-Heptachlorobiphenyl)	31.21 ± 0.62
PCB 187	(2,2',3,4',5,5',6-Heptachlorobiphenyl)	35.23 ± 0.83
159	(2,3,3',4,5,5'-Hexachlorobiphenyl)	
182	(2,2',3',4,4',5,6'-Heptachlorobiphenyl)	
PCB 194	(2,2',3,3',4,4',5,5'-Octachlorobiphenyl)	15.37 ± 0.61
PCB 201	(2,2',3,3',4,5',6,6'-Octachlorobiphenyl)	12.18 ± 0.46

[a] Each certified value is the equally-weighted mean of the means from two or more independent analytical methods. Each uncertainty, computed according to the CIPM approach as described in the ISO Guide [11], is an expanded uncertainty at the 95 % level of confidence, which includes random sources of uncertainty within each analytical method. The expanded uncertainty defines a range of values for the certified value within which the true value is believed to lie, at a level of confidence of approximately 95 %.

[b] PCB congeners are numbered according to the scheme proposed by Ballschmiter and Zell [6] and later revised by Schulte and Malisch [7] to conform with IUPAC rules. When two or more congeners are know to coelute under the conditions used, the PCB congener listed first is the major component and the additional congeners may be present as minor components. The quantitative results are based on the response of the congener listed first.

Table 2. Certified Concentrations (Mass Fractions) for Selected Chlorinated Pesticides in SRM 1588a[a]

	µg/kg
2,4'-DDE	22.0 ± 1.0
4,4'-DDE	651 ± 11
2,4'-DDD	36.3 ± 1.4
4,4'-DDD	254 ± 11
2,4'-DDT	156.0 ± 4.4
4,4'-DDT	524 ± 12
Hexachlorobenzene	157.8 ± 5.0
γ-hexachlorohexane (HCH)	24.9 ± 1.7
α-HCH	85.3 ± 3.4
Heptachlor epoxide	31.6 ± 1.5
cis-Chlordane	167.0 ± 5.0
cis-Nonachlor	94.8 ± 2.8
trans-Nonachlor	214.6 ± 7.9
Dieldrin	155.9 ± 4.5

[a] Each certified value is the equally-weighted mean of the means from two or more independent analytical methods. Each uncertainty, computed according to the CIPM approach as described in the ISO Guide [11], is an expanded uncertainty at the 95 % level of confidence, which includes random sources of uncertainty within each analytical method. The expanded uncertainty defines a range of values for the certified value within which the true value is believed to lie, at a level of confidence of approximately 95 %.

Table 3. Reference Concentrations (Mass Fractions in mg/kg) for alpha-Tocopherol in SRM 1588a[a]

NOTE: This concentration is provided as a reference value because the results have not been confirmed by an independent analytical technique as required for certification. The associated uncertainty, which incorporates variability from within and between the measurement techniques used, may reflect only measurement precision and may not include all sources of uncertainty. This reference value should be useful for comparison with results obtained using similar procedures (see Preparation and Analysis, alpha-Tocopherol).

alpha-Tocopherol	134.2 ± 2.5

[a] The uncertainty is based on 95 % confidence interval for the mean. It incorporates variability within and between the measurement techniques used.

Table 4. Reference Concentrations (Mass Fractions) for Additional PCB Congeners and Chlorinated Pesticides in SRM 1588a[a,b]

NOTE: These concentrations are provided as reference values because the results have not been confirmed by an independent analytical technique as required for certification. The associated uncertainties may reflect only measurement precision and may not include all sources of uncertainty.

		µg/kg
PCB 16	(2,2',3-Trichlorobiphenyl)	2.6 ± 0.8
32	(2,4',6-Trichlorobiphenyl)	
PCB 17	(2,2',4-Trichlorobiphenyl)	6.5 ± 1.1
PCB 18	(2,2',5-Trichlorobiphenyl)	8.1 ± 2.2
PCB 22	(2,3,4'-Trichlorobiphenyl)	3.0 ± 0.6
PCB 33	(2',3,4-Trichlorobiphenyl)	3.3 ± 1.4
PCB 41	(2,2',3,4-Tetrachlorobiphenyl)	5.3 ± 2.2
71	(2,3',4',6-Tetrachlorobiphenyl)	
PCB 42	(2,2',3,4'-Tetrachlorobiphenyl)	14 ± 3
PCB 46	(2,2',3,6'-Tetrachlorobiphenyl)	11 ± 2
PCB 55	(2,3,3',4-Tetrachlorobiphenyl)	18 ± 11
60	(2,3,4,4'-Tetrachlorobiphenyl)	
PCB 64	(2,3,4',6-Tetrachlorobiphenyl)	14 ± 1
PCB 70	(2,3',4',5-Tetrachlorobiphenyl)	27 ± 4
76	(2',3,4,5-Tetrachlorbiphenyl)	
PCB 74	(2,4,4',5-Tetrachlorobiphenyl)	40 ± 4
PCB 83	(2,2',3,3',5-Pentachlorobiphenyl)	21 ± 5
PCB 85	(2,2',3,4,4'-Pentachlorobiphenyl)	27 ± 8
PCB 91	(2,2',3,4',6-Pentachlorobiphenyl)	21 ± 8
PCB 97	(2,2',3',4,5-Pentachlorobiphenyl)	42 ± 4
PCB 136	(2,2',3,3',6,6'-Hexachlorobiphenyl)	14 ± 2
PCB 137	(2,2',3,4,4',5-Hexachlorobiphenyl)	16 ± 2
PCB 141	(2,2',3,4,5,5'-Hexachlorobiphenyl)	24 ± 4
PCB 146	(2,2',3,4',5,5'-Hexachlorobiphenyl)	39 ± 6
PCB 158	(2,3,3',4,4',6-Hexachlorobiphenyl)	21 ± 2
PCB 172	(2,2',3,3',4,5,5'-Heptachlorobiphenyl)	17 ± 4
197	(2,2',3,3',4,4',6,6'-Octachlorobiphenyl)	
PCB 174	(2,2',3,3',4,5,6'-Heptachlorobiphenyl)	41 ± 10
PCB 177	(2,2',3,3',4',5,6-Heptachlorobiphenyl)	4.9 ± 0.8
PCB 178	(2,2',3,3',5,5',6-Heptachlorobiphenyl)	28 ± 1
129	(2,2',3,3',4,5-Hexachlorobiphenyl)	
PCB 179	(2,2',3,3',5,6,6'-Heptachlorobiphenyl)	4.4 ± 1.2
PCB 189	(2,3,3',4,4',5,5'-Heptachlorobiphenyl)	2.9 ± 0.6
PCB 191	(2,3,3',4,4',5',6-Heptachlorobiphenyl)	4.5 ± 0.7
PCB 193	(2,3,3',4',5,5',6-Heptachlorobiphenyl)	14 ± 3
PCB 195	(2,2',3,3',4,4',5,6-Octachlorobiphenyl)	4.6 ± 0.6
PCB 196	(2,2',3,3',4,4',5',6-Octachlorobiphenyl)	24 ± 3
203	(2,2',3,4,4',5,5',6-Octachlorobiphenyl)	
PCB 199	(2,2',3,3',4,5,6,6'-Octachlorobiphenyl)	17 ± 2
PCB 206	(2,2',3,3',4,4',5,5',6-Nonachlorobiphenyl)	3.4 ± 1.6
PCB 209	(2,2',3,3',4,4',5,5',6,6'-Decachlorobiphenyl)	3.5 ± 1.0
Oxychlordane		38 ± 4
trans-Chlordane		52 ± 7
Mirex		16 ± 3

[a] Concentrations based on seven analyses done at the Department of Fisheries and Oceans, Winnipeg, Manitoba Canada over a period of two years. Uncertainties are expressed as 95 % confidence intervals of the mean.

[b] PCB congeners are numbered according to the scheme proposed by Ballschmiter and Zell [6] and later revised by Schulte and Malisch [7] to conform with IUPAC rules. When two or more congeners are know to coelute under the conditions used, the PCB congener listed first is the major component and the additional congeners may be present as minor components. The quantitative results are based on the response of the congener listed first.

Table 5. Information Concentrations (Mass Fractions) for Dibenzo-*p*-dioxins and Octachlorodibenzofuran

NOTE: These concentrations are provided as information values because the results, which are based only on gravimetry, have not been confirmed by an independent analytical technique as required for certification. These values are provided for information only.

		µg/kg
Dioxins and Furans		
1,2,7-Trichlorodibenzo-*p*-dioxin		0.32
1,2,3,4-Tetrachlorodibenzo-*p*-dioxin		0.38
2,3,7,8-Tetrachlorodibenzo-*p*-dioxin		0.21
1,2,3,6,7,8-Hexachlorodibenzo-*p*-dioxin	0.39	
1,2,3,7,8,9-Hexachlorodibenzo-*p*-dioxin	0.22	
Octachlorodibenzo-*p*-dioxin		1.01
Octachlorodibenzofuran		1.00

REFERENCES

[1] Certificate of Analysis, Standard Reference Material (SRM) 1588, Organics in Cod Liver Oil, National Institute of Standards and Technology, Gaithersburg, MD, (January 27, 1989).

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[11] *Guide to the Expression of Uncertainty in Measurement*, ISBN 92-67-10188-9, 1st Ed. ISO, Geneva, Switzerland, (1993): see also Taylor, B.N. and 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).

It is the responsibility of users of this SRM to assure that the certificate in their possession is current. This can be accomplished by contacting the SRM Program at: Phone (301) 975-6776 (select "Certificates"), Fax (301) 926-4751, e-mail srminfo@nist.gov, or via the Internet http://ts.nist.gov/srm.