Reference Material Institute for Clinical Chemistry Standards (ReCCS)

Certified Reference Material for Measurement of Total cholesterol, HDL-Cholesterol, LDL-Cholesterol and Triglycerides in Human Serum

JCCRM 223-47

Certificate of Analysis

■ Intended use

This CRM is intended primarily for use in evaluating the accuracy of clinical procedures for the determination of Total cholesterol (TC), HDL-cholesterol(HDL-C), Triglycerides(TG) and LDL-cholesterol.

■ Certified values and expanded uncertainties

①②③④⑤ are same as previous lot, JCCRM 223-46. ⑥ for Triglycerides and ⑦⑧ for LDL-Cholesterol are new.

unit: mg/dL (mmol/L) (25 ℃)

Item	Total Cholesterol (Abell-kendall method)		HDL-Cholesterol (CDC reference Method)		Triglycerides[1)] (ID-GC/MS method)		LDL-Cholesterol (CDC reference Method)	
	Certified value	Uncertainty	Certified value	Uncertainty	Certified value	Uncertainty	Certified value	Uncertainty
①	**140.4 (3.631)**	**1.8 (0.04[illegible])**						
②	**172.0 (4.450)**	**2.[illegible] (0.060)**			**11[illegible].7 (1.318)**	**2.3 (0.026)**		
③	**199.5 (5.161)**	**2.7 (0.071)**						
④			**48.1 (1.244)**	**1.6 (0.041)**				
⑤			**61.7 (1.595)**	**1.7 (0.043)**				
⑥					**176.9 (1.904)**	**3.4 (0.039)**		
⑦							**99.7 (2.579)**	**1.9 (0.050)**
⑧							**131.1 (3.391)**	**2.4 (0.063)**

1) The expanded uncertainty *U* (95 % level of confidence) shown for each certified value in the above table is obtained from the equation $U=ku$, where u is the combined standard uncertainty (the purity of triolein was also combined) calculated according to the ISO Guide 36, and k is a coverage factor [2)].The coverage factor k, determined from the Student's t distribution, is $k=2.0$.
2)The triglycerides concentration is expressed as Tripalmitin concentration.
3) The triglycerides defined in this reference material include triglycerides, most of monoglycerides and most of diglycerides but do not contain free glycerol.

■ Measurement methods for certified values

The certified values were determined by the following reference methods by the ReCCS
・Total Cholesterol was assayed by ID-GC/MS [3), 4), 5)] calculated by Abell-Kendall method.
・HDL-C was assayed by the CDC reference methods and ID/GC-MS [3), 4), 6), 7)]. The measured values were calculated to adjust to Abell-Kendall method.
・LDL cholesterol was assayed by CDC (BQ) reference method and ID/GC-MS [3), 4), 5), 8)]. The measured values were calculated to adjust to Abell-Kendall method.
・Triglycerides were assayed by ID/GC-MS [9), 10)].

■ Instructions for use

A vial is taken out from its case and is thawed at room temperature while placing it cap-side up. Next, the vial is left standing for about 30 minutes to bring the temperature of the serum to room temperature. Once this procedure is completed, hold the cap of the vial; gently rotate the vial in complete circles; several times and tighten the cap; and then mix the content of the vial by turning the vial upside down at least 40 times (the reference material in this manner must be used within three hours. Once thawed, the serum cannot be frozen for reuse.

■ Precautions for use *in vitro* use only

This CRM was prepared from human serum and shown to be negative to HBs antigens, HCV antibodies and HIV antibodies. However, this does not completely deny its infectivity, and take strictly the same caution used for any other infections specimens.

■ Storage and expiration

This CRM shipped in frozen condition on dry ice. Confirm that dry ice remains upon receipt; otherwise the materials could not be used thereafter.
Store this product in a deep freezer immediately after receiving it.
The expiration date is as follows from the shipping (see the label of the outer case).

HDL-C & LDL-C HDL-C	−70 ℃:	1 month	−40 ℃:	1 week
Total cholesterol & Triglycerides (Total glycerides)	−70 ℃:	6 months	−40 ℃:	3 months

■ Product specifications

Configuration: Frozen liquid
A single set of this CRM consists of 8 vials and each vial contains 0.5 mL.

■ Preparations

This CRM was prepared from pooled human serum as fresh as possible according to CLSI (Clinical and Laboratory Standards Institute) C37-A [1)].

■ Traceability

The ID/GC-MS for total cholesterol, HDL-cholesterol and LDL-cholesterol was conducted using NMIJ CRM 6001-a (purity 99.9±0.1 %) as a calibrator.
The ID/GC-MS method for triglycerides was conducted using NIST SRM 1595 (purity 99.5 % ± 0.2 %) as a calibrator.

■ Date of CertificationCertification

February 13, 2023

■Characteristics

The characteristics of this CRM are as follows:

	JCCRM 223-47								Unit	Methods
	①	②	③	④	⑤	⑥	⑦	⑧		
	TC/L	TC/M, TG/M	TC/H	HDL-C /L	HDL-C /H	TG/H	LDL-C/M	LDL-C/H		
TP	6.9	6.9	6.9	7.2	6.8	7.0	7.4	7.4	g/dL	Biurette method
Alb	4.2	4.1	4.2	4.1	3.8	4.2	4.2	4.1	g/dL	BCG colorimetric method
Lp(a)	9.9	16.1	17.0	9.5	13.0	3.9	12.0	16.9	mg/dL	LA
apo E	3.0	3.6	21.2	3.0	3.5	4.5	2.8	3.7	mg/dL	TIA
HDL	39	34	36	35	37	30	—	—	%	PAGE
LDL	40	44	40	44	43	35	—	—	%	
IDL	5	4	12	-	-	15	—	—	%	
VLDL	17	18	13	21	20	21	—	—	%	
UA	4.9	5.4	5.2	4.6	3.9	5.0	5.2	6.5	mg/dL	Enzymatic method
Bil	0.4	0.5	0.4	0.4	0.3	0.4	0.3	0.3	mg/dL	Vanadic acid oxidation method
Hb	Less than 4	Less than 4	Less than 4	Less than 4	Less than 4	Less than 4	Less than 4	Less than 4	mg/dL	Colorimetry

■References

1) CLSI Publication C37-A. Preparation and validation of commutable frozen human serum pools as secondary reference materials for cholesterol measurement procedures; approved guideline: CLSI, Wayne, PA, 1999.
2) Evaluation of measurement data - Guide to the expression of uncertainty in measurement. ISO/IEC Guide 98-3 (JCGM 100:2008).
3) Cohen A, et al. Total serum cholesterol by isotope dilution/mass spectrometry; a candidate definitive method, Clin Chem 1980; 26:854-860.
4) Ellerbe P, Meiselman S, Sniegoski LT, Welch MJ, White VE. Determination of serum cholesterol by a modification of the isotope dilution mass spectrometric definitive method. Anal Chem 1989; 61:1718-1723.
5) Edwards SH, Kimberly MM, Pyatt SD, et al. Proposed serum cholesterol reference measurement procedure by gas chromatography-isotope dilution mass spectrometry. Clin Chem 2001; 47:614-621.
6) N Rifai, GR Warnick, MH Daminiczak. Handbook of lipoprotein testing, 2nd Ed. pp.227-233, AACC Press, 2000, Washington DC.
7) Warnick GR, Albers JJ. Heparin-Mn^{2+} quantitation of high-density-lipoprotein cholesterol with ultrafiltration procedure for lipemic samples. Clin Chem 1978; 24:900-904.
8) Bachorik PS, Ross JW. National Cholesterol Education Program recommendations for measurement of low-density lipoprotein cholesterol. Clin Chem 1995; 41:1414-1420.
9) Ellerbe P, Sniegoski LT, Welch MJ. Isotope dilution mass spectrometry as a candidate definitive method for determining total glycerides and triglycerides in serum. Clin Chem, 1995; 41:397-404.
10) Bernert JT Jr, Bell CJ, McGuffey JE, Waymack PP. Determination of "free" glycerol in human serum reference materials by isotope-dilution gas chromatography-mass spectrometry. J Chromatogr 1992; 578:1-7.

■Provider of JCCRM 223-47

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