Cholesterol FS*

Diagnostic reagent for quantitative in vitro determination of cholesterol in serum or plasma on photometric systems

Order Information

Cat. No.	Kit size				
1 1300 99 10 021	R	5 x	25 mL	+ 1 x	3 mL Standard
1 1300 99 10 026	R	6 x	100 mL		
1 1300 99 10 023	R	1 x	1000 mL		
1 1300 99 10 704	R	8 x	50 mL		
1 1300 99 10 717	R	6 x	100 mL		
1 1300 99 10 917	R	10 x	60 mL		
1 1300 99 90 314	R	12 x	25 mL		
1 1300 99 10 030		6 x	3 mL	Standard	

Summary [1,2]

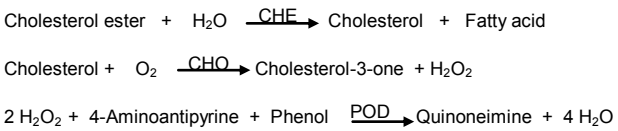
Cholesterol is a component of cell membranes and a precursor for steroid hormones and bile acids synthesized by body cells and absorbed with food. Cholesterol is transported in plasma via lipoproteins, namely complexes between lipids and apolipoproteins. There are four classes of lipoproteins: high density lipoproteins (HDL), low density lipoproteins (LDL), very low density lipoproteins (VLDL) and chylomicrons. While LDL is involved in the cholesterol transport to the peripheral cells, HDL is responsible for the cholesterol uptake from the cells. The four different lipoprotein classes show distinct relationship to coronary atherosclerosis. LDL-cholesterol (LDL-C) contributes to atherosclerotic plaque formation within the arterial intima and is strongly associated with coronary heart disease (CHD) and related mortality. Even with total cholesterol within the normal range an increased concentration of LDL-C indicates high risk. HDL-C has a protective effect impeding plaque formation and shows an inverse relationship to CHD prevalence. In fact, low HDL-C values constitute an independent risk factor. The determination of the individual total cholesterol (TC) level is used for screening purposes while for a better risk assessment it is necessary to measure additionally HDL-C and LDL-C.
In the last few years several controlled clinical trials using diet, life style changes and / or different drugs (especially HMG CoA reductase inhibitors [statins]) have demonstrated that lowering total cholesterol and LDL-C levels reduce drastically CHD risk [2].

Method

"CHOD-PAP": enzymatic photometric test

Principle

Determination of cholesterol after enzymatic hydrolysis and oxidation [3,4]. The colorimetric indicator is quinoneimine which is generated from 4-aminoantipyrine and phenol by hydrogen peroxide under the catalytic action of peroxidase (Trinder's reaction) [3].



Reagents

Components and Concentrations

Reagent:		
Good's buffer	pH 6.7	50 mmol/L
Phenol		5 mmol/L
4-Aminoantipyrine		0.3 mmol/L
Cholesterol esterase	(CHE)	≥ 200 U/L
Cholesterol oxidase	(CHO)	≥ 50 U/L
Peroxidase	(POD)	≥ 3 kU/L
Standard:		200 mg/dL (5.2 mmol/L)

Storage Instructions and Reagent Stability

Reagent and standard are stable up to the end of the indicated month of expiry, if stored at 2 – 8°C, protected from light and contamination is avoided. Do not freeze the reagents!

Note: It has to be mentioned, that the measurement is not influenced by occasionally occurring color changes, as long as the absorbance of the reagent is < 0.3 at 546 nm.

Warnings and Precautions

1. The reagent contains sodium azide (0.95 g/L) as preservative. Do not swallow! Avoid contact with skin and mucous membranes.
2. Standard: Warning. H317 May cause an allergic skin reaction. H319 Causes serious eye irritation. P264 Wash hands and face thoroughly after handling. P280 Wear protective gloves/protective clothing/eye protection/face protection. P302+P352 If on skin: Wash with plenty of soap and water. P337+P313 If eye irritation persists: Get medical advice/attention.
3. In very rare cases, samples of patients with gammopathy might give falsified results [8].
4. N-acetylcysteine (NAC), acetaminophen and metamizole medication leads to falsely low results in patient samples.
5. Please refer to the safety data sheets and take the necessary precautions for the use of laboratory reagents. For diagnostic purposes, the results should always be assessed with the patient's medical history, clinical examinations and other findings.
6. For professional use only!

Waste Management

Please refer to local legal requirements.

Reagent Preparation

The reagent and the standard are ready to use.

Materials required but not provided

NaCl solution 9 g/L
General laboratory equipment

Specimen

Serum, heparin plasma or EDTA plasma

Stability [6]:	7 days	at	20 – 25°C
	7 days	at	4 – 8°C
	3 months	at	–20°C

Discard contaminated specimens! Freeze only once!

Assay Procedure

Application sheets for automated systems are available on request.

Wavelength	500 nm, Hg 546 nm
Optical path	1 cm
Temperature	20 – 25°C/37°C
Measurement	Against reagent blank

	Blank	Sample or standard
Sample or standard	-	10 µL
Dist. water	10 µL	-
Reagent	1000 µL	1000 µL

Mix, incubate for 20 min. at 20 – 25°C or for 10 min. at 37°C.
Read absorbance within 60 min against reagent blank.

* fluid stable

Calculation

With standard or calibrator

$$\text{Cholesterol [mg/dL]} = \frac{\text{A Sample}}{\text{A Std/Cal}} \text{ x Conc. Std/Cal [mg/dL]}$$

Conversion factor

Cholesterol [mg/dL] x 0.02586 = Cholesterol [mmol/L]

Calibrators and Controls

For calibration of automated photometric systems, DiaSys TruCal U calibrator is recommended. The assigned values of the calibrator have been made traceable to the reference method gas chromatography-isotope dilution mass spectrometry (GC-IDMS). For internal quality control, DiaSys TruLab N and P or TruLab L controls should be assayed. Each laboratory should establish corrective action in case of deviations in control recovery.

	Cat. No.	Kit size
TruCal U	5 9100 99 10 063	20 x 3 mL
	5 9100 99 10 064	6 x 3 mL
TruLab N	5 9000 99 10 062	20 x 5 mL
	5 9000 99 10 061	6 x 5 mL
TruLab P	5 9050 99 10 062	20 x 5 mL
	5 9050 99 10 061	6 x 5 mL
TruLab L Level 1	5 9020 99 10 065	3 x 3 mL
TruLab L Level 2	5 9030 99 10 065	3 x 3 mL

Performance Characteristics

Measuring range

The test has been developed to determine cholesterol concentrations within a measuring range from 3 – 750 mg/dL (0.08 – 19.4 mmol/L). When values exceed this range samples should be diluted 1 + 4 with NaCl solution (9 g/L) and the result multiplied by 5.

Specificity/Interferences

No interference was observed by ascorbic acid up to 5 mg/dL, bilirubin up to 20 mg/dL, hemoglobin up to 200 mg/dL and lipemia up to 2,000 mg/dL triglycerides.
For further information on interfering substances refer to Young DS [7].

Sensitivity/Limit of Detection

The lower limit of detection is 3 mg/dL (0.08 mmol/L).

Precision (at 37°C)

Intra-assay precision n = 20	Mean [mg/dL]	SD [mg/dL]	CV [%]
Sample 1	108	1.76	1.62
Sample 2	236	1.45	0.61
Sample 3	254	1.57	0.62

Inter-assay precision n = 20	Mean [mg/dL]	SD [mg/dL]	CV [%]
Sample 1	104	1.19	1.14
Sample 2	211	2.57	1.22
Sample 3	245	2.28	0.93

Method Comparison

A comparison of DiaSys Cholesterol FS (y) with a commercially available test (x) using 78 samples gave following results:
y = 1.00 x – 2.50 mg/dL; r = 0.995

Reference Range [5]

Desirable	≤ 200 mg/dL (5.2 mmol/L)
Borderline high risk	200 – 240 mg/dL (5.2 – 6.2 mmol/L)
High risk	> 240 mg/dL (> 6.2 mmol/L)

Each laboratory should check if the reference ranges are transferable to its own patient population and determine own reference ranges if necessary.

Clinical Interpretation

The European Task Force on Coronary Prevention recommends to lower TC concentration to less than 190 mg/dL (5.0 mmol/L) and LDL-cholesterol to less than 115 mg/dL (3.0 mmol/L) [2].

Literature

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Manufacturer

IVD CE

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