SPECTRUM

The Creative Approach to Bioscience

Cholesterol – Liquizyme
CHOD-PAP *(Single Reagent)*

REF: 230 001	(2 x 25 ml)	50 test
REF: 230 002	(4 x 25 ml)	100 test
REF: 230 003	(2 x 50 ml)	100 test
REF: 230 004	(4 x 30 ml)	120 test
REF: 230 005	(10 x 15 ml)	150 test
REF: 230 006	(4 x 50 ml)	200 test
REF: 230 007	(4 x 100 ml)	400 test
REF: 230 008	(5 x 100 ml)	500 test
REF: 230 009	(6 x 100 ml)	600 test
REF: 230 010	(8 x 100 ml)	800 test
REF: 230 011	(4 x 250 ml)	1000 test
REF: 230 012	(2 x 500 ml)	1000 test

Intended Use

Spectrum Diagnostics liquizyme cholesterol reagent is intended for in-vitro quantitative, diagnostic determination of cholesterol in human serum or plasma on both manual and automated systems.

Background

Measurement of serum cholesterol levels is important as an indicator of liver function, intestinal absorption, biliary function and in the diagnosis and classification of hyperlipoproteinemias.
Elevated cholesterol levels may occur with hypothyroidism,diabetes and nephrotic syndrome. Elevated serum cholesterol levels correlate well with the incidence of coronary artery diseases. Stress, age, gender, hormonal balance and pregnancy affect normal cholesterol levels. Depressed levels are associated with hyperthyroidism and severe liver diseases.

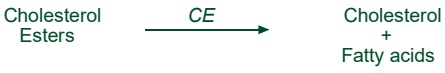
Method

CHOD-PAP-enzymatic colorimetric method.

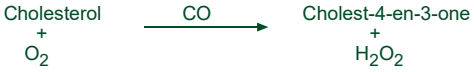
Assay Principle

The series of the reactions involved in the assay system is as follows:

1. Cholesterol esters are enzymatically hydrolyzed by cholesterol esterase (CE) to cholesterol and free fatty acids.



2. Free cholesterol, including that originally present, is then oxidized by cholesterol oxidase (CO) to cholest-4-en-3-one and hydrogen peroxide.



3. The hydrogen peroxide combines with phenol and 4-amino-antipyrine (4AAP) in the presence of peroxidase (POD) to form a chromophore (quinoneimine dye) which may be quantitated at 500 – 550 nm. For bichromatic analyzers the blank wavelength should be set to 600 or 650 nm.

Reagents

Standard cholesterol (ST)

200 mg/dL	5.17	mmol/L

Reagent (R)

Pipes Buffer pH 7.0	50	mmol/L
Phenol	30	mmol/L
Sodium cholate	5.0	mmol/L
Cholesterol esterase	>250	U/L
Cholesterol oxidase	>500	U/L
Peroxidase	>2.0	KU/L
4-amino-antipyrine	1.0	mmol/L
Sodium Azide	8.0	mmol/L

For further information, refer to the Cholesterol reagent material safety data sheet.

SYMBOLS IN PRODUCT LABELLING

EC REP	Authorised Representative		Use by/Expiration Date
IVD	For in-vitro diagnostic use		CAUTION. Consult instructions for use
LOT	Batch Code/Lot number		
REF	Catalogue Number		Manufactured by
	Consult instructions for use		(Xi) - Irritant
	Temperature Limitation		

Precautions and Warnings

Do not ingest or inhale. In case of contact with eyes or skin; rinse immediately with plenty of soap and water. In case of severe injuries; seek medical advice immediately.

Reagent (R) contains sodium azide which may react with copper or lead plumbing.

Reagent Preparation, Storage and Stability

Spectrum cholesterol reagents are supplied ready-to-use and stable up to the expiry date stated on the vial labels when stored at 2- 8^{o}C. Once opened, the reagent and standard are stable for 3 months at the specified temperature.

Deterioration

The reagent is normally clear or pale pink. Do not use liquizyme cholesterol reagent if it is turbid or if the absorbance is greater than 0.15 at 546 nm.

Specimen Collection and Preservation

It is recommended that prior to sample collection, patients should be following their usual diet and be in their usual state of health. Patients who are actually ill, losing weight, pregnant or have had a myocardial infarction in the previous 3 months should be rescheduled. Both fasting and non-fasting samples can be used. Non haemolysed serum or plasma can be stored at 4 ^{o}C up to 7 days prior to analysis, 5-7 days at 20-25^{o}C, stable for 3 months at -20^{o}C, and at -70 ^{o}C for several months. The only acceptable anticoaglulant is heparin.

System Parameters

Wavelength	546 nm (500 – 550 nm)
Optical path	1 cm
Assay type	End-point
Direction	Increase
Sample : Reagent Ratio	1 : 100
e.g .: Reagent volume	1 ml
Sample volume	10 µl
Temperature	15 – 25 ^{o}C or 37 ^{o}C
Zero adjustment	Reagent blank
Incubation time	5 minutes at 37 ^{o}C or 10 minutes at 15 – 25 ^{o}C
Reagent Blank Limits	Low 0.00 AU High 0.15 AU
Sensitivity	5 mg/dL (0.13mmol/L)
Linearity	750 mg/dL (19.5 mmol/L)

Procedure

	Blank	Standard	Sample
Reagent (**R**)	1.0 ml	1.0 ml	1.0 ml
Standard	-----	10 µl	-----
Sample	-----	-----	10 µl

Mix and incubate for 5 minutes at 37 ^{o}C or 10 minutes at 15 – 25^{o}C. Measure absorbance of specimen ($A_{specimen}$) and standard ($A_{standard}$) against reagent blank within 30 minutes.

Calculation

$$\text{Serum cholesterol conc. (mg/dL)} = \frac{(A_{specimen})}{(A_{standard})} \times 200$$

Quality Control

Normal and abnormal commercial control serum of known concentrations should be analyzed with each run.

Performance Characteristics

Precision

Within run (Repeatability)

	Level 1	Level 2
n	20	20
Mean (mg/dL)	149.8	252
SD	1.69	1.91
CV%	1.13	0.76

Run to run (Reproducibility)

	Level 1	Level 2
n	20	20
Mean (mg/dL)	157	259
SD	1.77	2.12
CV%	1.13	0.82

Methods Comparison

A comparison between Spectrum Diagnostics Cholesterol reagent and a commercial reagent of the same methodology was performed on 20 human sera. A correlation of 0.988 was obtained.

Sensitivity

When run as recommended, the minimum detection limit of the assay is 5 mg/dL (0.13 mmol/L).

Linearity

The reaction is linear up to a cholesterol concentration of 750 mg/dl; specimens showing higher concentration should be diluted 1+1 using physiological saline and repeat the assay (result × 2).

Interfering Substances

Haemolysis
No significant interference up to a level of 500 mg/dl.

Icterus
No interference from free bilirubin up to a level of 15 mg /dL (260 mmol/L) and conjugated bilirubin up to a level of 7 mg/dL (116 mmol/L).

Lipemia
No significant interference up to 1.7 AU.

Drugs
Of the drugs tested in vitro, methyldopa causes artificially low total cholesterol values at the tested drug level.

Others
Physiological ascorbic acid concentration does not interfere with the test. Ascorbic Acid levels higher than 425 mmol/l (7.5 mg/dl) decrease the apparent total cholesterol concentration significantly.

Expected Values

The following guidelines may be used for clinical interpretation:

Risk classification	Total cholesterol	
Desirable	<200 mg/dl	<5.2 mmol/L
Borderline high	200-239 mg/dl	5.2-6.2 mmol/L
High	⩾ 240 mg/dl	⩾ 6.2 mmol/L

Spectrum Diagnostics does not interpret the results of a clinical laboratory procedure; interpretation of the reults is considered the responsibility of qualified medical personnel. All indications of clinical significance are supported by literature references.

Analytical Range

5 - 750 mg/dl (0.13 - 19.5 mmol/L).

Waste Disposal

This product is made to be used in professional laboratories.
Please consult local regulations for a correct waste disposal.
S56: dispose of this material and its container at hazardous or special waste collection point.
S57: use appropriate container to avoid environmental contamination.
S61: avoid release in environment. refer to special instructions/safety data sheets.

References

1. Ellefson RD and Caraway WT : Fundamentals of clinical chemistry. Ed Tietz NW 1976; p506.
2. Flegg HM : Ann Clin Biochem 1963 ; 10 : 79.
3. NCEP expert panel, Arch Intern Med 1988; 148: 36–69
4. Richmond. N., Clin. Chem. 1973; 19: 1350-1356.
5. Roeschlau,P., Bernt. E. and Gruber. W.J., Clin. Chem Clin. Biochem. 1974; 12:403.
6. Trinder, P, Ann. Clin. Biochem. 1969; 6: 24.
7. Young DS .et al. Clin Chem. 1975 ; 21.

ORDERING INFORMATION	
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230 011	4 x 250 ml
230 012	2 x 500 ml

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