BIOLABO
www.biolabo.fr

MANUFACTURER:
BIOLABO SAS,
Les Hautes Rives
02160, Maizy, France

CHOLESTEROL CHOD PAP

Ready-to-use Liquid

Reagent for quantitative determination of Total Cholesterol in human serum and plasma

REF LP80106 **R1** 2 x 100 mL **R2** 1 x 5 mL

TECHNICAL SUPPORT AND ORDERS
Tel: (33) 03 23 25 15 50
support@biolabo.fr
Latest revision: www.biolabo.fr

CE

IVD

Made In France

I: corresponds to significant modifications

INTENDED USE

I This reagent is designated for professional use in laboratory. It may be used with manual procedure on spectrophotometer or with Biochemistry Clinical Analyzer.
This quantitative test is to determine the concentration of Total Cholesterol in human serum or plasma.

GENERALITIES (1) (2)

I Hypercholesterolemia can be observed in case of dietary imbalance, in in hepatic and thyroid disorders, certain cases of diabetes, nephrotic syndrome, pancreatitis, myeloma or familial hypercholesterolemia.
Total cholesterol increased levels may be isolated or associated to other increased lipids (hyperlipidemia).
A decreased level of cholesterol may be due to deficiencies or malnutrition, cancer or hyperthyroidism.

PRINCIPLE (4)

Enzymatic method described by Allain and al., which reaction scheme is as follows:

Cholesterol esters $\xrightarrow{\text{CE}}$ Cholesterol + free fatty acids

Cholesterol + O_2 $\xrightarrow{\text{CO}}$ Cholesten 4 one 3 + H_2O_2

2 H_2O_2 + Phenol + PAP $\xrightarrow{\text{POD}}$ Quinoneimine (pink) + 4 H_2O

REAGENTS

R1 CHOLESTEROL CHOD PAP Reagent

Phosphate buffer	100	mmol/L
Chloro-4-phenol	5	mmol/L
Sodium Cholate	2.3	mmol/L
Triton x 100	1.5	mmol/L
Cholesterol oxydase (CO)	≥ 100	IU/L
Cholesterol esterase (CE)	≥ 170	IU/L
Peroxydase (POD)	≥ 1200	IU/L
4 - Amino – antipyrine (PAP)	0.25	mmol/L
PEG 6000	167	µmol/L
Preservative		

According to 1272/2008 regulation, vial R1 is not classified as dangerous

R2 CHOLESTEROL CHOD PAP Standard

Cholesterol 200 mg/dL (5.17 mmol/L)

Attention Danger

Skin Irrit. 2 : H315 – Causes skin irritation
Eye Dam. 1: H318 – Causes serious eye damage
Flam. Liq. 3: H226 – Flammable liquid and vapor

P210: Keep away from heat/sparks/open flames/hot surfaces – No smoking, P280: Wear protective gloves/protective clothing/eye protection/face protection, P302+P352: IF ON SKIN: Wash with soap and water,

P305+P351+P338: IF IN EYES: Rinse continuously with water for several minutes. Remove contact lenses if present and easy to do – continue rinsing, P501: Dispose of contents/container in accordance with dangerous waste regulations.
I Classification due to N-Propanol and Tergitol 10 - < 25%, For more details, refer to Safety Data Sheet (MSDS)

SAFETY CAUTIONS

- Refer to current Material Safety Data Sheet available on request or on www.biolabo.fr
- Verify the integrity of the contents before use.
- Waste disposal: Respect legislation in force in the country.
- All specimens or reagents of biological origin should be handled as potentially infectious. Respect legislation in force in the country.

I Any serious incident that has occurred in connection with the device is notified to the manufacturer and the competent authority of the Member State in which the user and/or patient is based.

REAGENTS PREPARATION

Ready for use.

STABILITY AND STORAGE

Stored away from light, well cap in the original vial at 2-8°C, reagents are stable when stored and used as described in the insert:

Unopened:
- Until the expiry date stated on the label of the Kit.

Once opened:
- Transfer requested quantity, well recap vials and store at 2-8°C
- Reagent is stable at least 3 months when free from contamination.
- Discard reagent (R1) if cloudy or if reagent blank at 500 nm > 0.400.

SPECIMEN COLLECTION AND HANDLING (2)

Serum or plasma (Heparin or EDTA).
Do not use oxalate, fluoride or citrate. Collect on fasting patient. Separate serum from cells within 2 hours.

Cholesterol is stable:
- 5-7 days at 2-8°C
- 3 months at –20°C
- Many years at –70°C.
- Avoid repeated freezing and thawing

LIMITS (2) (3) (5)

Enzymatic methods increase analytic specificity. Cholesterol oxidase also reacts with 3β-hydroxycholesterols (insignificant quantity in human serum – i.e. DHEA, pregnenolone).

For a more comprehensive review of factors affecting this assay refer to the publication of Young D.S or N. W. Tietz.

MATERIALS REQUIRED BUT NOT PROVIDED

1. Basic medical analysis laboratory equipment.
2. Spectrophotometer or Biochemistry Clinical Analyzer

QUALITY CONTROL

- REF 95010 EXATROL-N Level I.
- REF 95011 EXATROL-P Level II.
- External quality control program.

It is recommended to control in the following cases:

- At least once a run.
- At least once within 24 hours.
- When changing vial of reagent.
- After maintenance operations on the instrument.

If control is out of range, apply following actions:

1. Prepare a fresh control serum and repeat the test.
2. If control is still out of range, use a new vial of calibrator or a fresh calibrator and repeat the test.
3. If control is still out of range, repeat with a new vial of reagent.

If control is still out of range, please contact BIOLABO technical support or your local Agent.

EXPECTED VALUES (2)

Values for adults, estimated in term of risk for atherosclerotic diseases:

Total cholesterol	**mg/dL**	**[mmol/L]**
Recommended values	< 200	[< 5.18]
Low risk	200-239	[5.18-6.19]
High risk	≥ 240	[≥ 6.22]

Each laboratory should establish its own normal ranges for the population that it serves.

PERFORMANCES

on KENZA 240TX Analyser, 37°C, 505 nm

Linearity Range: between 9 and 500 mg/dL

Detection limit: approx. 2 mg/dL

Precision:

Within-run N = 20	*Low level*	*Normal level*	*High level*
Mean (mg/dL)	119	208	299
S.D. mg/dL	2.5	5.0	7.7
C.V. %	2.1	2.4	2.6

Between run N = 20	*Low level*	*Normal level*	*High level*
Mean (mg/dL)	123	201	299
S.D. mg/dL	2.1	4.2	5.6
C.V. %	1.7	2.3	1.9

Analytical Sensitivity: approx. 0.3246 abs for 100 mg/dL

Interferences:

Turbidity	No interference up to 0.288 OD
Total bilirubin	Negative interference from 295 µmol/L
Direct bilirubin	Negative interference from 190 µmol/L
Ascorbic acid	Negative interference from 998 mg/dL
Glucose	No interference up to 1089 mg/dL
Haemoglobin	No interference up to 405 µmol/L

Other substances may interfere (see § Limits)

On the board stability: 2 months

Calibration Stability: 2 months

Make a new calibration when changing reagent batch, if quality control results are found out of the established range and after maintenance operations.

Performances and stability data on Kenza 450TX/ISE and Kenza ONE are available on request

I On Kenza 450TX, clinical comparison study with commercially available reagent using serum specimens between 29 and 320 mg/dL (n=138):

y = 0.9108 x + 9.8254, R = 0.9815

CALIBRATION (6)

- REF 95015 Multicalibrator traceable *to SRM 1951c*
- Standard (vial R2)

The calibration frequency depends on proper instrument functions and on the preservation of the reagent.

PROCEDURE

Detailed Kenza 240TX procedure is available on request

Wavelength: 505 nm

Temperature: 37°C

	Automated analyzer	Manual procedure
Reagent	300 µL	1000 µL
Standard, Controls, Specimen	3 µL	10 µL
Mix. Let stands for 5 minutes at 37°C or 10 minutes at room temperature. Record absorbance at 500 nm (480-520) against reagent blank. Colour is stable for 1 hour.		

1- Performances with manual procedure should be validated by user.

2- Kenza applications and other applications proposal are available on request

CALCULATION

Calculate the result as follows:

$$\text{Result} = \frac{\text{Abs (Assay)}}{\text{Abs (Standard)}} \times \text{Standard concentration}$$

Automatic Biochemistry analyzer:

The analyzer provides directly result.
For more details about calibration and calculation of results, refer to User's manual and specific application.

REFERENCES

(1) TIETZ N.W. Text book of clinical chemistry, 3rd Ed. C.A. Burtis, E.R. Ashwood, W.B. Saunders (1999) p. 826-835.
(2) Clinical Guide to Laboratory Test, 3rd Ed., N.W. TIETZ (1995) p. 130-131.
(3) YOUNG D.S., Effect of Drugs on Clinical laboratory Tests, 4th Ed. (1995) p. 3-143 to 3-164
(4) Allain C. C. et al., Clin. Chem. (1974), 20/4, p.470-475
(5) Allan C., Deacon et Peter J. G. Dawson, Clin. Chem. (1979) 25/6, p.976-984
(6) SRM: Standard Reference material ®

Manufacturer	Expiry date	In vitro diagnostic	Storage temperature	Dematerialized water	Biological risk
Product Reference	See Insert	Batch number	Store away from light	Sufficient for	Dilute with