CHOLESTEROL (2-8°C)

(CHOD PAP)

CATALOGUE NUMBER	KIT SIZE (ML)
CHO1201	3 x 40ml
CHO2401	3 x 40ml
CHO4201	4 x 75ml

Intended Use:
For *In Vitro* diagnostic use by trained professionals only.
This reagent is intended for the quantitative determination of cholesterol in human serum and plasma.

Clinical Significance:
Elevated levels of cholesterol are primarily considered as an indication of increased risk of cardiovascular disease and should be taken into consideration combined with the overall lipid profile.

Test Principle:
Cholesterol is present in serum as cholesterol esters and free cholesterol. The Cholesterol esters present in serum are hydrolysed by cholesterol esterase and the cholesterol is then measured by oxidizing with cholesterol oxidase to form hydrogen peroxide. The hydrogen peroxide in turn reacts with phenol and 4-aminoantipyrine present to form the red quinoneimine dye. The intensity of the dye formed can be measured photometrically and is directly proportional to the level of cholesterol present in the sample.

Reagent Composition

REAGENT	COMPONENT	CONCENTRATION
Cholesterol Reagent R1	Pipes Buffer	90 mmol/l
	Phenol	26 mmol/l
	Choleserol Oxidase	200 U/l
	Cholesterol Esterase	300 U/l
	Peroxidase	1250 U/l
	4-Aminoantipyrine	0.4 mmol/l
Cholesterol Standard	Cholesterol	5.17 mmol/l (200mg/dl)

Reagent Preparation and Stability:
R1: Liquid, ready to use
Standard: Liquid, ready to use
R1 and Standard are stable up to the expiry date stated when stored tightly capped at 2 - 8°C. Once opened the reagents are stable for a period of 12 weeks, when stored without contamination at 2 - 8°C.
Dispose of reagents carefully in line with local guidelines.

Sample Collection, Preparation and Stability:
Collect serum and Li-Heparin or EDTA plasma by standard venepuncture technique. Do not use citrate, oxalate or fluoride as anticoagulants.
Cholesterol is stable in serum and plasma for 5 days at 2 - 8°C and up to 3 months when frozen at -20°C.

Assay Procedure:

WAVELENGTH	546nm (500 – 550nm)
TEMPERATURE	37°C
CUVETTE	1cm Path Length
BLANK	Reagent Blank

	Blank	Standard	Sample
Sample	-	-	10µl
Standard	-	10µl	-
Reagent	1000µl	1000µl	1000µl
Mix and incubate for 5 minutes at assay temperature. Read the absorbance (Δ Abs) of Sample/Standard against the Reagent Blank.			

Calculation:

Concentration (mmol/l) = $\frac{\Delta Abs\ Sample}{\Delta Abs\ Standard}$ x Concentration of Standard (5.17mmol/l)

To convert mmol/l to mg/dl multiply the result by 38.66

Performance Characteristics:

Measuring range:
800 mg/dl (20.7 mmol/l)
Dilute samples with higher concentrations using Normal saline 1+2 and rerun the assay. Multiply the result by the dilution factor (for 1+2 dilution, the dilution factor is 3)

Analytical Sensitivity: (Lowest detection limit):
3 mg/dl (0.08 mmol/l)

Imprecision
Intra-Assay Precision:

Sample	Mean (mg/dl)	SD (mg/dl)	CV %
Pool 1	103	1.35	1.32
Pool 2	158	0.98	0.62
Pool 3	180	0.99	0.55

Inter-Assay Precision:

Sample	Mean (mg/dl)	SD (mg/dl)	CV %
Pool 1	124	1.71	1.38
Pool 2	162	3.13	1.93
Pool 3	186	1.97	1.06

Method Comparison:
A comparison of the Prestige Diagnostics Cholesterol Reagent (y) with an alternative commercial assay (x) gave the following result:
y = 1.006x + 0.258, r = 0.999

Interferences:
Criterion: Recovery within +/- 10% of initial value.
Icterus: No significant interference up to 8 mg/dl of Bilirubin
Haemolysis: No significant interference up to a concentration of 450 mg/dl haemoglobin.

Reference Range:

Normal	< 5.2 mmol/l (200mg/dl)
Borderline Risk	5.2 – 6.2 mmol/l (200-239 mg/dl)
High Risk	>6.2 mmol/l (239mg/dl)

These values are in line with the recommendations of the European atherosclerosis Society, however we recommend that each laboratory should establish its own mean reference range according to the population.

Limitations:
The Cholesterol result from this test should not be used as the sole criteria for the diagnosis of lipid disorders, a confirmed diagnosis should only be made by a physician after all clinical and laboratory findings have been evaluated.

Automated systems:
Contact Prestige Diagnostics Technical Department for applications on a wide range of automated analysers.
For automation we recommend the use of a serum based calibrator.

Quality Control and Calibration Material:
Calibration Serum: **QCCCAL1 / QCCCAL2**
Human Assayed Control Normal: **QCCHAN1 / QCCHAN2**
Human Assayed Control Elevated: **QCCHAE1 / QCCHAE2**

References:

1. Weibe D.A., Bernert J.T., Clin Chem 1984; 30:352
2. Richmond N Clin. Chem 1973; 19: 1350-1356
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4. Trinder P *Annals Clin Biochem. 1969; 6: 24*
5. Abell L. et al. Standard Methods in Clinical Chemistry 1958; 26:2
6. Allain C.C. et al. Clin Chem 1974; 20: 470
7. Bablock W et al. A General Regression Procedure for Method Transformation. J Clin Chem Clin Biochem 1988 26: 783 – 790
8. Tietz NW *Clinical Guide to Laboratory Tests* 3rd Edition
9. Glick M. R., Ryder K.E., Jackson SA. Graphical Comparisons of Interferences in Clinical Chemistry Instrumentation. Clin Chem 1986; 32: 470-474

REF	Catalog number		Temperature limitation
	Consult instructions for use	LOT	Batch code
IVD	*In vitro* diagnostic medical device		Use by
	Manufacturer		Keep away from sunlight

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