

HDL PRECIPITATING REAGENT

CD 0400 CH

4 x 100 ml

INTENDED USE

Reagent for precipitation of LDL-cholesterol, to be used in quantitative in vitro assay of cholesterol in biological fluids.

SUMMARY OF TEST

Blood total cholesterol levels have long been known to be related to coronary heart disease (CHD). In recent years, in addition to total cholesterol, high density lipoprotein cholesterol (HDL-C) has become an important tool used to assess an individual risk of developing CHD since a strong negative relationship between HDL-C concentration and the incidence of CHD was reported. Thus, there has been substantial interest in HDL-C analysis.

PRINCIPLE OF THE METHOD

Polyethylene glycol, average MW 6000, in aqueous solution, is used to precipitate lipoproteins VLDL and LDL. After centrifugation, the clear supernatant containing HDL fraction is suitable for enzymatic determination of cholesterol.

KIT COMPONENTS

For in vitro diagnostic use only.

The components of the kit are stable until expiration date on the label.

Keep away from direct light sources.

HDL-P R1 4 x 100 ml (liquid) blue cap

Composition: polyethyleneglycol 16 %, non reactive additives and stabilizers.

Store all components at 2-8°C.

MATERIALS REQUIRED BUT NOT SUPPLIED

Current laboratory instrumentation. Spectrophotometer UV/VIS with thermostatic cuvette holder. Automatic micro-pipettes. Glass or high quality polystyrene cuvettes. Saline solution.

The kit CT F400 CH or CT 150F CH CHOLESTEROL FL and your STANDARD are needed to perform the colorimetric assay run.

REAGENT PREPARATION

Use reagent ready to use.

Stability: up to expiration date on labels at 2-8°C.

Stability since first opening of vials: preferably within 60 days at 2-8°C.

PRECAUTIONS

Reagent may contain some non-reactive and preservative components. It is suggested to handle carefully it, avoiding contact with skin and swallow.

Perform the test according to the general "Good Laboratory Practice" (GLP) guidelines.

SPECIMEN

Serum, plasma EDTA.

Sample is stable 3 days at 2-8°C and 1 month at -20°C.

TEST PROCEDURE - PRECIPITATION STEP

Pipet in centrifuge tubes:

sample 500 µl + reagent 500 µl

mix by inversion, incubate 5 minutes, centrifuge at 3000 g/min for 10 minutes.

Separate supernatant and use it as sample into following procedure.

TEST PROCEDURE - QUANTITATIVE STEP

Wavelength: 510 nm (allowed 480 ÷ 520 nm)
Lightpath: 1 cm
Temperature: 37°C

dispense:	blank	standard	sample
reagent	1 ml	1 ml	1 ml
water	25 µl	-	-
standard	-	25 µl	-
sample	-	-	25 µl

Mix, incubate at 37°C for 5 minutes.

Read absorbances of standard (As) and samples (Ax) against reagent blank.

RESULTS CALCULATION

serum/plasma sample:

HDL cholesterol mg/dl = $A_x/A_s \times \text{stand. value} \times 2$
(standard value + dilution factor)

EXPECTED VALUES

	high risk	average	low risk
Men:	< 40 mg/dl	40-50 mg/dl	> 50 mg/dl
Women:	< 45 mg/dl	45-60 mg/dl	> 60 mg/dl

Each laboratory should establish appropriate reference intervals related to its population.

QUALITY CONTROL AND CALIBRATION

It is suggested to perform an internal quality control. For this purpose a suitable human based control sera has to be used.

Please contact Customer Care for further information.

TEST PERFORMANCE

Sample is prepared through precipitation procedures as indicated above, supernatant is analyzed by reagent CT F400 CH - CT 150F CH

Linearity

the method is linear up to 700 mg/dl.

If the limit value is exceeded, it is suggested to dilute sample 1+9 with saline and to repeat the test, multiplying the result by 10.

Sensitivity/limit of detection (LOD)

the limit of detection is 1 mg/dl.

Interferences

no interference was observed by the presence of:

hemoglobin	≤ 500 mg/dl
bilirubin	≤ 15 mg/dl
lipids	≤ 850 mg/dl

Precision

intra-assay (n=10)	mean (mg/dl)	SD (mg/dl)	CV%
sample 1	101.50	1.84	1.80
sample 2	176.20	2.74	1.60

inter-assay (n=20)	mean (mg/dl)	SD (mg/dl)	CV%
sample 1	100.99	2.11	2.10
sample 2	176.51	2.23	1.30

Methods comparison

a comparison between Chema and a commercially available product gave the following results:

Cholesterol FL Chema = x
HDL-direct competitor = y
n = 75

$y = 0.980x + 0.608 \text{ mg/dl}$ $r^2 = 0.957$

WASTE DISPOSAL

This product is made to be used in professional laboratories.

P501: Dispose of contents according to national/international regulations.








REFERENCES

Trinder P., - J. Clin. Path. 22, 158 (1969);
Allain C.C., Poon L.S., Chan C.S., Richmond W., Fu P.C., - Clin. Chem. 20,470 (1974).
National Cholesterol Education Program (NCEP) recommended values for cholesterol
Tietz Textbook of Clinical Chemistry, Second Edition, Burtis-Ashwood (1994).

MANUFACTURER

Chema Diagnostica
Via Campania 2/4
60030 Monsano (AN) - ITALY - EU
phone +39 0731 605064
fax +39 0731 605672
e-mail: mail@chema.com
website: <http://www.chema.com>

SYMBOLS

	in vitro diagnostic medical device
	batch code
	catalogue number
	temperature limit
	use by date
	caution
	consult instructions for use