LIPC

cobas®

Lipase colorimetric assay

Order information

REF	CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
03029590 322	Lipase colorimetric assay 200 tests	System-ID 07 5900 7	Roche/Hitachi **cobas c** 311, **cobas c** 501/502
10759350 190	Calibrator f.a.s. (12 x 3 mL)	Code 401	
10759350 360	Calibrator f.a.s. (12 x 3 mL, for USA)	Code 401	
12149435 122	Precinorm U plus (10 x 3 mL)	Code 300	
12149435 160	Precinorm U plus (10 x 3 mL, for USA)	Code 300	
12149443 122	Precipath U plus (10 x 3 mL)	Code 301	
12149443 160	Precipath U plus (10 x 3 mL, for USA)	Code 301	
10171743 122	Precinorm U (20 x 5 mL)	Code 300	
10171735 122	Precinorm U (4 x 5 mL)	Code 300	
10171778 122	Precipath U (20 x 5 mL)	Code 301	
10171760 122	Precipath U (4 x 5 mL)	Code 301	
05117003 190	PreciControl ClinChem Multi 1 (20 x 5 mL)	Code 391	
05947626 190	PreciControl ClinChem Multi 1 (4 x 5 mL)	Code 391	
05947626 160	PreciControl ClinChem Multi 1 (4 x 5 mL, for USA)	Code 391	
05117216 190	PreciControl ClinChem Multi 2 (20 x 5 mL)	Code 392	
05947774 190	PreciControl ClinChem Multi 2 (4 x 5 mL)	Code 392	
05947774 160	PreciControl ClinChem Multi 2 (4 x 5 mL, for USA)	Code 392	
04489357 190	Diluent NaCl 9 % (50 mL)	System-ID 07 6869 3	

English

System information

For **cobas c** 311/501 analyzers:

LIPC: ACN 731

SLIPC: ACN 733 (STAT, reaction time: 5)

For **cobas c** 502 analyzer:

LIPC: ACN 8731

SLIPC: ACN 8733 (STAT, reaction time: 5)

Intended use

Enzymatic in vitro test for the quantitative determination of lipase in human serum and plasma on Roche/Hitachi **cobas c** systems.

Summary[1,2,3,4,5,6,7]

Lipases are glycoproteins with a molecular weight of 47000 daltons. They are defined as triglyceride hydrolases which catalyze the cleavage of triglycerides to diglycerides with subsequent formation of monoglycerides and fatty acids. In addition to α-amylase, pancreatic lipases have for many years been undeniably the most important clinical chemistry parameters for the differential diagnosis of diseases of the pancreas. The lipase activity determination has gained increasing international recognition because of its high specificity and rapid response. After acute pancreatitis the lipase activity increases within 4-8 hours, reaches a peak after 24 hours and decreases after 8 to 14 days. However, there is no correlation between the lipase activity determined in serum and the extent of damage to the pancreas.

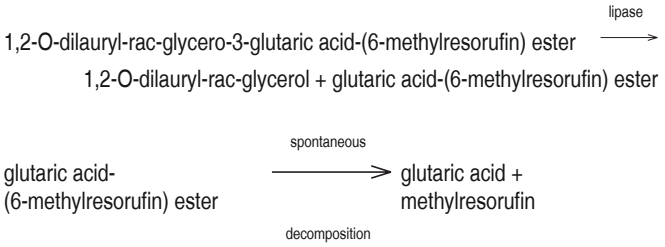
Numerous methods have been described for the determination of lipase which determine the decrease in substrate turbidimetrically or nephelometrically or determine degradation products.

This method is based on the cleavage of a specific chromogenic lipase substrate 1,2-O-dilauryl-rac-glycero-3-glutaric acid-(6-methylresorufin) ester emulsified with bile acids. The pancreatic enzyme activity is determined specifically by the combination of bile acid and colipase used in this assay. Virtually no lipase activity is detected in the absence of colipase. Colipase only activates pancreatic lipase, but not other lipolytic enzymes found in serum. The high amount of cholates ensures that the esterases present in the serum do not react with the chromogenic substrate due to the highly negative surface charge.

Test principle[8,9,10,11]

Enzymatic colorimetric assay with 1,2-O-dilauryl-rac-glycero-3-glutaric acid-(6-methyl-resorufin) ester as substrate.

The chromogenic lipase substrate 1,2-O-dilauryl-rac-glycero-3-glutaric acid-(6-methylresorufin) ester is cleaved by the catalytic action of alkaline lipase solution to form 1,2-O-dilauryl-rac-glycerol and an unstable intermediate, glutaric acid-(6-methylresorufin) ester. This decomposes spontaneously in alkaline solution to form glutaric acid and methylresorufin. Addition of detergent and colipase increases the specificity of the assay for pancreatic lipase.



The color intensity of the red dye formed is directly proportional to the lipase activity and can be determined photometrically.

Reagents - working solutions

R1 BICIN[a] buffer: 50 mmol/L, pH 8.0; colipase (porcine pancreas): ≥ 0.9 mg/L; Na-deoxycholate: 1.6 mmol/L; calcium chloride: 10 mmol/L; detergent; preservative

R2 Tartrate buffer: 10 mmol/L, pH 4.16; 1,2-O-dilauryl-rac-glycero-3-glutaric acid-(6-methylresorufin) ester: 0.27 mmol/L; taurodeoxycholate: 8.8 mmol/L; detergent; preservative

a) BICIN = N,N-bis(2-hydroxyethyl)glycine

R1 is in position B and R2 is in position C.

Precautions and warnings

For in vitro diagnostic use.

Exercise the normal precautions required for handling all laboratory

reagents.
Disposal of all waste material should be in accordance with local guidelines.
Safety data sheet available for professional user on request.

Reagent handling
Ready for use

Storage and stability

LIPC

Shelf life at 2-8 °C:	See expiration date on **cobas c** pack label.
On-board in use and refrigerated on the analyzer:	4 weeks

Diluent NaCl 9 %

Shelf life at 2-8 °C:	See expiration date on **cobas c** pack label.
On-board in use and refrigerated on the analyzer:	12 weeks

Specimen collection and preparation
For specimen collection and preparation only use suitable tubes or collection containers.
Only the specimens listed below were tested and found acceptable.
Serum.
Plasma: Li-heparin plasma
The sample types listed were tested with a selection of sample collection tubes that were commercially available at the time of testing, i.e. not all available tubes of all manufacturers were tested. Sample collection systems from various manufacturers may contain differing materials which could affect the test results in some cases. When processing samples in primary tubes (sample collection systems), follow the instructions of the tube manufacturer.
Centrifuge samples containing precipitates before performing the assay.

Stability:[12]	1 week at 15-25 °C
	1 week at 2-8 °C
	1 year at (-15)-(-25) °C

Materials provided
See “Reagents – working solutions” section for reagents.

Materials required (but not provided)
See “Order information” section
General laboratory equipment

Assay
For optimum performance of the assay follow the directions given in this document for the analyzer concerned. Refer to the appropriate operator’s manual for analyzer-specific assay instructions.
The performance of applications not validated by Roche is not warranted and must be defined by the user.

Application for serum and plasma

cobas c 311 **test definition**

Assay type	Rate A	
Reaction time / Assay points	10 / 15-22 (STAT 5 / 15-22)	
Wavelength (sub/main)	700/570 nm	
Reaction direction	Increase	
Units	U/L (µkat/L)	
Reagent pipetting		Diluent (H_2O)
R1	80 µL	20 µL
R2	48 µL	–

Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	2 µL	–	–
Decreased	2 µL	15	135
Increased	2 µL	–	–

cobas c 501 **test definition**

Assay type	Rate A	
Reaction time / Assay points	10 / 22-31 (STAT 5 / 22-31)	
Wavelength (sub/main)	700/570 nm	
Reaction direction	Increase	
Units	U/L (µkat/L)	
Reagent pipetting		Diluent (H_2O)
R1	80 µL	20 µL
R2	48 µL	–

Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	2 µL	–	–
Decreased	2 µL	15	135
Increased	2 µL	–	–

cobas c 502 **test definition**

Assay type	Rate A	
Reaction time / Assay points	10 / 22-31 (STAT 5 / 22-31)	
Wavelength (sub/main)	700/570 nm	
Reaction direction	Increase	
Units	U/L (µkat/L)	
Reagent pipetting		Diluent (H_2O)
R1	80 µL	20 µL
R2	48 µL	–

Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	2 µL	–	–
Decreased	2 µL	15	135
Increased	4 µL	–	–

Calibration

Calibrators	S1: H_2O S2: C.f.a.s.
Calibration mode	Linear
Calibration frequency	2-point calibration - after reagent lot change - and as required following quality control procedures

Traceability: This method has been standardized manually against Roche reagent using calibrated pipettes together with a manual photometer providing absolute values and the substrate-specific absorptivity, ε.

Quality control
For quality control, use control materials as listed in the "Order information" section.
In addition, other suitable control material can be used.
The control intervals and limits should be adapted to each laboratory’s individual requirements. Values obtained should fall within the defined

limits. Each laboratory should establish corrective measures to be taken if values fall outside the defined limits.

Follow the applicable government regulations and local guidelines for quality control.

Calculation

Roche/Hitachi **cobas c** systems automatically calculate the analyte concentration of each sample.

Conversion factor: U/L x 0.0167 = µkat/L

Limitations - interference

Criterion: Recovery within ± 10 % of initial values at a lipase activity of 60 U/L (1.00 µkat/L).

Icterus:[13] No significant interference up to an I index of 50 for conjugated and unconjugated bilirubin (approximate conjugated and unconjugated bilirubin concentration: 855 µmol/L (50 mg/dL)).

Hemolysis:[13] No significant interference up to an H index of 1000 (approximate hemoglobin concentration: 620 µmol/L (1000 mg/dL)).

Lipemia (Intralipid):[13] No significant interference up to an L index of 2000. There is poor correlation between the L index (corresponds to turbidity) and triglycerides concentration.

Drugs: No interference was found at therapeutic concentrations using common drug panels.[14,15]

Exception: Calcium dobesilate causes artificially low lipase results.

In very rare cases, gammopathy, in particular type IgM (Waldenström's macroglobulinemia), may cause unreliable results.[16]

For diagnostic purposes, the results should always be assessed in conjunction with the patient's medical history, clinical examination and other findings.

ACTION REQUIRED

Special Wash Programming: The use of special wash steps is mandatory when certain test combinations are run together on Roche/Hitachi **cobas c** systems. The latest version of the carry-over evasion list can be found with the NaOHD/SMS/Multiclean/SCCS or the NaOHD/SMS/SmpCln1+2/SCCS Method Sheets. For further instructions refer to the operator's manual. **cobas c** 502 analyzer: All special wash programming necessary for avoiding carry-over is available via the **cobas** link, manual input is not required.

Where required, special wash/carry-over evasion programming must be implemented prior to reporting results with this test.

Limits and ranges

Measuring range

3-300 U/L (0.05-5.01 µkat/L)

Determine samples having higher activities via the rerun function. Dilution of samples via the rerun function is a 1:10 dilution. Results from samples diluted using the rerun function are automatically multiplied by a factor of 10.

Lower limits of measurement

Lower detection limit of the test

3 U/L (0.05 µkat/L)

The lower detection limit represents the lowest measurable analyte level that can be distinguished from zero. It is calculated as the value lying 3 standard deviations above that of the lowest standard (standard 1 + 3 SD, repeatability, n = 21).

Expected values[17]

Adults: 13-60 U/L (0.22-1.00 µkat/L)

Each laboratory should investigate the transferability of the expected values to its own patient population and if necessary determine its own reference ranges.

Specific performance data

Representative performance data on the analyzers are given below. Results obtained in individual laboratories may differ.

Precision

Precision was determined using human samples and controls in an internal protocol with repeatability (n = 21) and intermediate precision (3 aliquots per run, 1 run per day, 21 days). The following results were obtained:

Repeatability	*Mean* *U/L (µkat/L)*	*SD* *U/L (µkat/L)*	*CV* *%*
Precinorm U	62.2 (1.04)	0.4 (0.01)	0.7
Precipath U	102 (1.70)	1 (0.01)	0.7
Human serum 1	30.1 (0.50)	0.3 (0.01)	1.0
Human serum 2	231 (3.86)	2 (0.03)	0.9
Intermediate precision	*Mean* *U/L (µkat/L)*	*SD* *U/L (µkat/L)*	*CV* *%*
Precinorm U	61.0 (1.02)	0.9 (0.02)	1.5
Precipath U	99.3 (1.66)	1.9 (0.03)	1.9
Human serum 3	28.8 (0.48)	0.6 (0.01)	2.1
Human serum 4	320 (5.34)	6 (0.09)	1.7

Method comparison

Lipase values for human serum and plasma samples obtained on a Roche/Hitachi **cobas c** 501 analyzer (y) were compared with those determined using the same reagent on a Roche/Hitachi 917 analyzer (x).

Sample size (n) = 185

Passing/Bablok[18]	Linear regression
y = 0.982x - 0.249 U/L	y = 0.962x + 1.32 U/L
τ = 0.935	r = 0.998

The sample activities were between 9.40 and 299 U/L (0.157 and 4.99 µkat/L).

References

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A point (period/stop) is always used in this Method Sheet as the decimal separator to mark the border between the integral and the fractional parts of a decimal numeral. Separators for thousands are not used.

Symbols

Roche Diagnostics uses the following symbols and signs in addition to those listed in the ISO 15223-1 standard.

CONTENT	Contents of kit
→	Volume after reconstitution or mixing

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Significant additions or changes are indicated by a change bar in the margin.

CE

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