LIPC

cobas®

Lipase colorimetric

Order information

REF	CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
07041918190	Lipase colorimetric (580 tests)	System-ID 05 5900 8	**cobas c** 701/702

Materials required (but not provided):

10759350360	Calibrator f.a.s. (12 x 3 mL)	Code 401	
12149435160	Precinorm U plus (10 x 3 mL)	Code 300	
12149443160	Precipath U plus (10 x 3 mL)	Code 301	
05947626160	PreciControl ClinChem Multi 1 (4 x 5 mL)	Code 391	
05947774160	PreciControl ClinChem Multi 2 (4 x 5 mL)	Code 392	
05172152190	Diluent NaCl 9 % (119 mL)	System-ID 08 6869 3	

English

For use in the USA only

System information

LIP: ACN 8789

S-LIP: ACN 8786 (STAT, reaction time: 5)

Intended use

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

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

Lipases are glycoproteins with a molecular weight of 47000 Da. 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 8-14 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-methylresorufin) 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.

1,2-O-dilauryl-rac-glycero-3-glutaric acid-(6-methylresorufin) ester $\xrightarrow{\text{lipase}}$ 1,2-O-dilauryl-rac-glycerol + glutaric acid-(6-methylresorufin) ester

glutaric acid-(6-methylresorufin) ester $\xrightarrow[\text{decomposition}]{\text{spontaneous}}$ glutaric acid + methylresorufin

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
R3 (STAT 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 R3 (STAT R2) is in position C.

Precautions and warnings

For in vitro diagnostic use for healthcare professionals. Exercise the normal precautions required for handling all laboratory reagents.

Infectious or microbial waste:
Warning: handle waste as potentially biohazardous material. Dispose of waste according to accepted laboratory instructions and procedures.

Environmental hazards:
Apply all relevant local disposal regulations to determine the safe disposal.

Safety data sheet available for professional user on request.

This kit contains components classified as follows in accordance with the Regulation (EC) No. 1272/2008:

Warning

H317	May cause an allergic skin reaction.
H319	Causes serious eye irritation.

Prevention:

P261	Avoid breathing mist or vapours.
P280	Wear protective gloves/ eye protection/ face protection.

Response:

P333 + P313	If skin irritation or rash occurs: Get medical advice/attention.
P337 + P313	If eye irritation persists: Get medical advice/attention.
P362 + P364	Take off contaminated clothing and wash it before reuse.

Disposal:

P501	Dispose of contents/container to an approved waste disposal plant.

Product safety labeling follows EU GHS guidance.

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Contact phone: 1-800-428-2336

Reagent handling

Ready for use

Storage and stability

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
On-board on the Reagent Manager:	24 hours

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.

See the limitations and interferences section for details about possible sample interferences.

Sample stability claims were established by experimental data by the manufacturer or based on reference literature and only for the temperatures/time frames as stated in the Method Sheet. It is the responsibility of the individual laboratory to use all available references and/or its own studies to determine specific stability criteria for its laboratory.

Stability in serum:[12]	7 days at 20-25 °C
	7 days at 4-8 °C
	1 year at -20 °C (±5 °C)

Freeze only once.

Stability in plasma:	1 week at 15-25 °C
	1 week at 2-8 °C
	2 months at -20 °C (±5 °C)

Freeze only once.

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 701/702 **test definition**

Assay type	Rate A
Reaction time / Assay points	10 / 22-25 (STAT 5 / 10-13)
Wavelength (sub/main)	700/570 nm
Reaction direction	Increase
Units	U/L (µkat/L)

Reagent pipetting		Diluent (H_2O)
R1	80 µL	20 µL
R3 (STAT R2)	48 µL	–

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

Calibration

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

Calibration interval may be extended based on acceptable verification of calibration by the laboratory.

Traceability: This method has been standardized manually against Roche reagent using 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

cobas c systems automatically calculate the analyte activity 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 60 for conjugated and unconjugated bilirubin (approximate conjugated and unconjugated bilirubin concentration: 1026 µmol/L or 60 mg/dL).

Hemolysis:[13] No significant interference up to an H index of 100 (approximate hemoglobin concentration: 62 µmol/L or 100 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]

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 **cobas c** systems. All special wash programming necessary for avoiding carry-over is available via the **cobas** link, manual input is required in certain cases. The latest version of the carry-over evasion list can be found with the NaOHD/SMS/SmpCln1+2/SCCS Method Sheet and for further instructions refer to the operator’s manual.

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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

Limit of Blank, Limit of Detection and Limit of Quantitation

Limit of Blank = 3 U/L (0.05 µkat/L)

Limit of Detection = 3 U/L (0.05 µkat/L)

Limit of Quantitation = 5 U/L (0.08 µkat/L)

The Limit of Blank, Limit of Detection and Limit of Quantitation were determined in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP17-A2 requirements.

The Limit of Blank is the 95th percentile value from n ≥ 60 measurements of analyte-free samples over several independent series. The Limit of Blank corresponds to the concentration below which analyte-free samples are found with a probability of 95 %.

The Limit of Detection is determined based on the Limit of Blank and the standard deviation of low concentration samples.

The Limit of Detection corresponds to the lowest analyte concentration which can be detected (value above the Limit of Blank with a probability of 95 %).

The Limit of Quantitation is the lowest analyte concentration that can be reproducibly measured with a precision coefficient of variation of 20 %. It has been determined using low concentration of lipase samples.

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

Repeatability and intermediate precision were determined using human samples and controls in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP5 requirements (2 aliquots per run, 2 runs per day, 21 days). The following results were obtained on the **cobas c** 701 analyzer:

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Repeatability	*Mean* *U/L (µkat/L)*	*SD* *U/L (µkat/L)*	*CV* *%*
PCCC Multi 1	45.4 (0.76)	0.49 (0.01)	1.1
PCCC Multi 2	100 (1.67)	0.79 (0.01)	0.8
Human serum 1	12.0 (0.20)	0.28 (0.005)	2.3
Human serum 2	47.1 (0.79)	0.48 (0.01)	1.0
Human serum 3	74.5 (1.24)	1.12 (0.02)	1.5
Human serum 4	139 (2.32)	2.10 (0.04)	1.5
Human serum 5	286 (4.78)	3.23 (0.05)	1.1

Intermediate precision	*Mean* *U/L (µkat/L)*	*SD* *U/L (µkat/L)*	*CV* *%*
PCCC Multi 1	45.4 (0.76)	0.91 (0.02)	2.0
PCCC Multi 2	100 (1.67)	2.10 (0.04)	2.1
Human serum 1	12.0 (0.20)	0.41 (0.01)	3.5
Human serum 2	47.1 (0.79)	0.92 (0.02)	1.9
Human serum 3	74.5 (1.24)	1.71 (0.03)	2.3
Human serum 4	139 (2.32)	3.04 (0.05)	2.2
Human serum 5	286 (4.78)	5.85 (0.10)	2.0

S-LIP

Repeatability	*Mean* *U/L (µkat/L)*	*SD* *U/L (µkat/L)*	*CV* *%*
PCCC Multi 1	46.2 (0.77)	0.47 (0.01)	1.0
PCCC Multi 2	102 (1.70)	0.89 (0.01)	0.9
Human serum 1	12.3 (0.21)	0.29 (0.005)	2.3
Human serum 2	47.8 (0.80)	0.50 (0.01)	1.1
Human serum 3	75.4 (1.26)	0.96 (0.02)	1.3
Human serum 4	141 (2.35)	1.56 (0.03)	1.1
Human serum 5	290 (4.84)	2.46 (0.04)	0.8

Intermediate precision	*Mean* *U/L (µkat/L)*	*SD* *U/L (µkat/L)*	*CV* *%*
PCCC Multi 1	46.2 (0.77)	0.94 (0.02)	2.0
PCCC Multi 2	102 (1.70)	1.94 (0.03)	1.9
Human serum 1	12.3 (0.21)	0.37 (0.01)	3.0
Human serum 2	47.8 (0.80)	0.94 (0.02)	2.0
Human serum 3	75.4 (1.26)	1.55 (0.03)	2.1
Human serum 4	141 (2.35)	2.51 (0.04)	1.8
Human serum 5	290 (4.84)	4.93 (0.08)	1.7

PCCC = PreciControl ClinChem

Method comparison

Lipase values for human serum and plasma samples obtained on a **cobas c** 701 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 501 analyzer (x).

LIP:

Sample size (n) = 102

Passing/Bablok[18]	Linear regression
y = 1.00x + 0.482 U/L	y = 0.997x + 0.792 U/L
τ = 0.985	r = 1.000

The sample activities were between 4.2 and 294 U/L (0.07 and 4.91 µkat/L).

S-LIP:

Sample size (n) = 99

Passing/Bablok[18]	Linear regression
y = 1.00x + 0.153 U/L	y = 1.00x + 0.304 U/L
τ = 0.984	r = 0.999

The sample activities were between 4.2 and 294 U/L (0.07 and 4.91 µkat/L).

References

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Any serious incident that has occurred in relation to the device shall be reported to the manufacturer and the competent authority of the Member State in which the user and/or the patient is established.

Symbols

Roche Diagnostics uses the following symbols and signs in addition to those listed in the ISO 15223-1 standard (for USA: see navifyportal.roche.com for definition of symbols used):

CONTENT	Contents of kit
→	Volume for reconstitution
GTIN	Global Trade Item Number
Rx only	For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

FOR US CUSTOMERS ONLY: LIMITED WARRANTY

Roche Diagnostics warrants that this product will meet the specifications stated in the labeling when used in accordance with such labeling and will be free from defects in material and workmanship until the expiration date printed on the label. THIS LIMITED WARRANTY IS IN LIEU OF ANY OTHER WARRANTY, EXPRESS OR IMPLIED, INCLUDING ANY IMPLIED WARRANTY OF MERCHANTABILITY OR FITNESS FOR PARTICULAR PURPOSE. IN NO EVENT SHALL ROCHE DIAGNOSTICS BE LIABLE FOR INCIDENTAL, INDIRECT, SPECIAL OR CONSEQUENTIAL DAMAGES.

For USA: Rx only

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