

# LIPASE

This reagent is intended for the *in-vitro* quantitative determination of Lipase (Triacylglycerolase) in human serum. Determination can be done by Fixed Time method.

## CLINICAL SIGNIFICANCE :

Lipase is most frequently measured to assist with the investigation of pancreatitis. In acute pancreatitis, Lipase increases within 4-8 hours, peaks at approximately 24 hours and decreases within 4-18 days. Lipase may also be elevated in chronic pancreatitis but, if severe destruction of the acinar tissue has occurred, serum levels may be below those normally detected.

Obstruction of the pancreatic duct, caused by a calculus or carcinoma, may also result in increased serum lipase levels. Increased serum lipase may also be observed in chronic or acute renal disease, after endoscopic retrograde pancreatography or treatment with opiates.

## PRINCIPLE :

The Lipase reagent utilises the method of Ziegenhom et al.



Lipase catalyses the hydrolysis of Triolein in the presence of Colipase to form glycerides and fatty acids. The rate of decrease in turbidity, measured at 340 nm, is proportional to the lipase activity.

**WARNING :** Avoid ingestion and contact with skin, mouth and eyes. The toxicity of this reagent has not been determined. Flush with plenty of water when disposing.

## PRESENTATION:

	No. of Bottles/Pouch
All reagents to be stored at 2-8° C.	12 x 1.1 ml
1 Lipase (Powder)	13
2 Lipase (Buffer)	1
Lipase Calibrator (as per stated on vial)	2
Lipase Diluent	1

## PREPARATION OF WORKING REAGENT:

### For 12 x 1.1 ml:

Dissolve the contents of one vial of 1 Lipase with 1.1 ml of 2 Lipase (Buffer). Mix gently to dissolve.

## REAGENT STORAGE AND STABILITY:

When stored refrigerated at 2-8° C, the reagent is stable until the expiry date stated on the bottle and kit box label. Reconstituted reagent is stable for 14 days at 2-8°C.

## PREPARATION OF LIPASE CALIBRATOR & STABILITY :

Add 1 ml of Diluent to the vial of Lipase Calibrator. Mix well to dissolve the contents. The reconstituted control is stable for atleast for 7 days when stored at 2-8°C and for 4 weeks when stored at -20°C

## SPECIMEN COLLECTION AND HANDLING :

- Collection** : No special preparation of the patient serum is required.
- Serum** : Use non-hemolyzed serum.
- Storage** : In serum, lipase is stable for 1 week at room temperature (18-25°C) or for 3 weeks at 2-8°C. If frozen, lipase is stable for several months.

## REACTION PARAMETERS : FOR FIX TIME:

- Type of Reaction : Fix Time (Decreasing O.D)
- Wavelength : 340 nm
- Flowcell Temperature : 37°C
- Delay time : 240 Secs. (4 mins)
- Interval Time : 300 Secs. (5 mins)
- No. of Readings : 2
- Sample Volume : 0.04 ml / 0.02 ml
- Working Reagent Volume : 1.0 ml / 0.5 ml

## PROCEDURE :

PIPETTE INTO TEST TUBES	Calibrator	TEST
• Working Reagent	1.0 ml	1.0 ml
• Sample	-	0.04 ml
• Calibrator	0.04 ml	-

Mix and read 1<sup>st</sup> absorbance of test exactly at 240 seconds (4 min) and 2<sup>nd</sup> absorbance at 540 seconds (9 min) at 340 nm. Determine the mean change in absorbance as per  $\Delta A = (A_1 - A_2)$  Value.

## Test Results:

For Fixed Time:

$$\text{Lipase activity (U/L)} = \frac{\Delta A/\text{min of Sample}}{\Delta A/\text{min of Calibrator}} \times \text{Calibrator Value}$$

Calibrator value obtained as per U/L mentioned on calibrator.

## NOTES:-

**Reagents such as cholesterol/Triglycerides/HDL/LDL contains high concentrations of detergents and hydrolyzing enzymes; cross contamination from such reagents should be avoided. All glassware / tips and cuvette being used for the test should be thoroughly cleaned.**

## LIMITATIONS :

1. Cholesterol and Triglycerides reagents typically contain high levels of activity of lipase and esterase. Care should be taken to avoid contamination with these reagents.
2. Studies to determine the level of interference from haemoglobin and bilirubin were carried out.
3. The following results were obtained :-

**Haemoglobin** : No interference from haemoglobin upto 124 µmol/L.

**Bilirubin** : No interference from bilirubin upto 342 µmol/L.

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**EXPECTED VALUES:**

Using the method described, the expected values were determined to be upto 200 U/L.

The quoted values are representative of the expected range for this method and should serve as a guide only. It is recommended that each laboratory should verify this range or derive a reference interval for the population that it serves.

**LINEARITY :**

When run as recommended, the assay is linear up to 700 U/L.

**REFERENCES :**

1. Tietz N.W., and Shuey D.F., Lipase in serum-the Elusive Enzyme: An Overview. Clin Chem 1993: 39:746-56
2. Ziegenhorn j. et al, Clin. Chem. 1979:25:1067.
3. Moss D.W. and Henderson M.B., "Enzymes" in Tietz Textbook of Clinical Chemistry, Burtis, C.A. and Ahwood, E.R. (Eds). WB Saunder Company 1994.

