

## INTRODUCTION

- Infinite** Liquid Lipase is a reagent set for determination of lipase activity in human serum and plasma based on **colorimetric method**.
- Infinite** Liquid Lipase is a **ready-to-use, two liquid reagent system** stable till expiry.
- Infinite** Liquid Lipase is **linear** upto **300 IU/l**.
- Infinite** Liquid Lipase can be determined in just **9 minutes** at **37°C**.

## PRINCIPLE

The chromogenic lipase substrate 1, 2-o-dilauryl-rac-glycero-3-glutaric acid-(6-methylresorufin) ester is cleaved by the catalytic action of lipase to form 1,2-o-dilauryl-rac-glycerol and an unstable intermediate glutaric acid-(6-methyl resorufin) ester. This decomposes spontaneously in alkaline medium to form glutaric acid and methylresorufin.

The lipase activity in the specimen is proportional to the rate of formation of methylresorufin in the reaction and can be determined photometrically at 578 nm.

1,2-o-dilauryl-rac-glycero-3-glutaric acid-(6-methylresorufin) ester



1,2-o-dilauryl-rac-glycerol + glutaric acid-(6-methylresorufin) ester



glutaric acid + methylresorufin

## REAGENT STORAGE, STABILITY & HANDLING

The reagents R1 & R2 are ready-to-use.

The reagent kit should be stored at 2 - 8°C and is stable till the expiry date indicated on the label.

## COMPONENTS & CONCENTRATION OF WORKING SOLUTION

Component	Concentration
<b>R1</b>	
• BICIN buffer, pH 8.0	50 mmol/l
• Colipase	≥1 mg/l
• Sodium deoxycholate	1.6 mmol/l
• Calcium chloride	10 mmol/l
<b>R2</b>	
• Tartarate buffer, pH 4.0	10 mmol/l
• Taurodeoxycholate	8.8 mmol/l
• DGGMR*	0.27 mmol/l

\* Abbreviations:

DGGMR = 1,2-o-dilauryl-rac-glycero-3-glutaric acid-(6-methylresorufin) ester

## SPECIMEN COLLECTION & PRESERVATION

Blood should be collected in a clean dry container. Although serum is preferred, plasma with lithium heparin can be used. Samples with any visible haemolysis are not acceptable.

Lipase activity in serum/plasma is stable for 5 days at 2 - 8°C or for 24 hours at 20 - 25°C. Do not use specimens that are repeatedly subjected to freezing and thawing.

## PROCEDURE

<input type="checkbox"/> Reaction type	Kinetic
<input type="checkbox"/> Reaction direction	Increasing
<input type="checkbox"/> Wavelength	578 nm.
<input type="checkbox"/> Flowcell temperature	37°C
<input type="checkbox"/> Zero setting with	Reagent Blank
<input type="checkbox"/> Delay time	120 seconds
<input type="checkbox"/> No. of readings	3
<input type="checkbox"/> Interval	60 seconds
<input type="checkbox"/> Sample volume	0.01 ml (10 µl)
<input type="checkbox"/> R1 volume	0.8 ml (800 µl)
<input type="checkbox"/> R2 volume	0.2 ml (200 µl)
<input type="checkbox"/> Linearity	300 IU/l

### Manual assay procedure

Perform the assay as given below:

	Reagent Blank	Calibrator	Serum/Plasma
R1	0.8 ml	0.8 ml	0.8 ml
	Distilled H <sub>2</sub> O (0.01 ml)	0.01 ml	0.01 ml

Mix and incubate for 5 minutes at 37°C.

R2	0.2 ml	0.2 ml	0.2 ml
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Mix and aspirate the assay mixture. The first reading should be recorded at 120<sup>th</sup> second followed by two more readings with 60 second intervals at 578 nm.

Measure activity of lipase against reagent blank.

### Calculation :

$$\text{Factor} = \frac{\text{Activity of Calibrator}}{\Delta \text{ Abs./min. of Calibrator}}$$

$$\text{Lipase (IU/l)} = \text{Factor} \times \Delta \text{ Abs./min. of Sample}$$

**Note:** For convenience, the reagent/sample ratio can be altered to R1 : 800 µl ; R2 : 200 µl and Sample : 20 µl. In this case, the laboratory would have to establish a fresh factor. The linearity of this assay would be 220 IU/l.

## EXPECTED VALUES

13 - 60 IU/l

Expected range varies from population to population. It is therefore recommended that each laboratory should establish its own normal range.

## PROCEDURE LIMITATIONS

If the lipase activity exceeds 300 IU/l, dilute the specimen with normal saline and repeat the assay. The result obtained should then be multiplied with the dilution factor to obtain correct lipase activity.

## QUALITY CONTROL

To ensure adequate quality control, it is recommended that each batch should include a normal and an abnormal commercial reference control serum. It should be realized that the use of quality control material checks both instrument and reagent functions together. Factors which might affect the performance of this test include instrument function, temperature control, cleanliness of glassware and accuracy of pipetting.

