



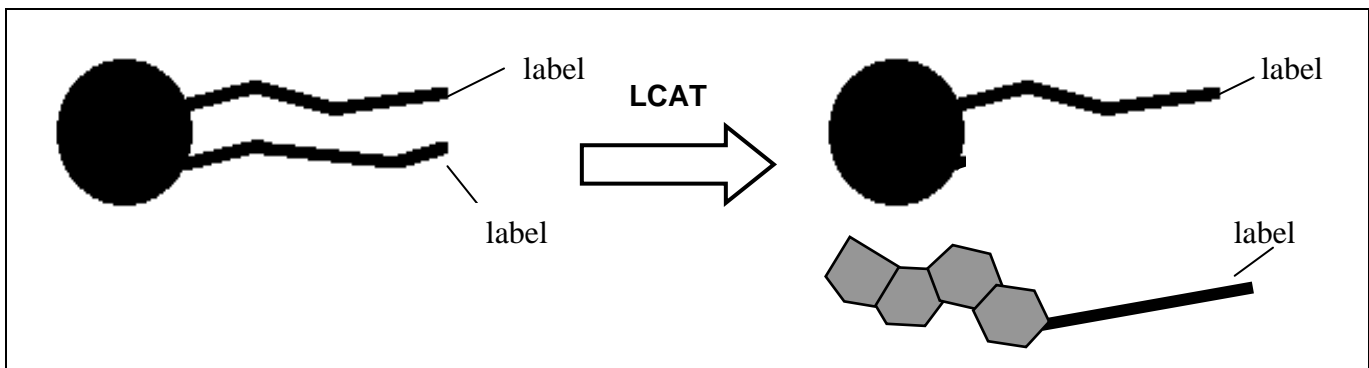
Product Information

Roar LCAT Activity Assay Kit, 240 assays

Catalog No. RB-LCAT

Lecithin:cholesterol acyltransferase (LCAT) mediates the formation of cholesteryl esters in human plasma. LCAT transfers an acyl chain from the sn-2 position of phosphatidylcholine to cholesterol.

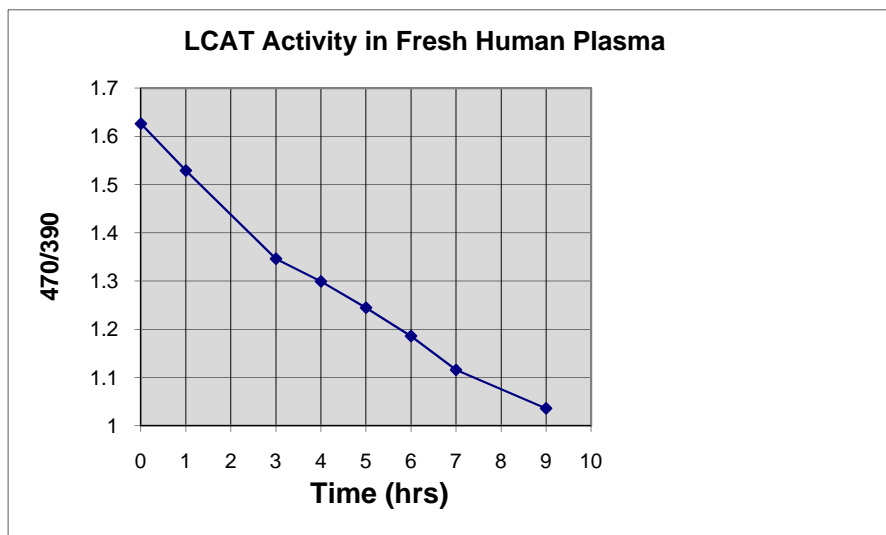
Combine:



Incubate: 37°C

Read: Activity is assessed as a change in 470/390 emission intensity

LCAT is incubated with a fluorescently labeled substrate. The intact substrate fluoresces at 470 nm. During hydrolysis by LCAT, the monomer emission at 390 nm increases. Intra- and interassay coefficients of variation: <5%.^{1,11}



Kit Components

LCAT substrate reagent (240 µl) - sufficient for 240 assays

READ reagent (1 ml) - supplied in concentrated form

Materials Required, But Not Supplied

LCAT assay buffer (150 mM NaCl, 10 mM tris, 1 mM EDTA, 4 mM 2-mercaptoethanol, pH 7.4)

READ reagent buffer (150 mM NaCl, 10 mM tris, 1 mM EDTA, pH 7.4)

Storage

LCAT substrate reagent: Store at -20° C

READ reagent: Refrigerate

Method

1. Prepare LCAT assay buffer: 150 mM NaCl / 10 mM tris / 1mM EDTA / 4 mM 2-mercaptoethanol. Adjust pH to 7.4.
2. Reconstitute READ reagent with 99 ml 150 mM NaCl / 10 mM tris / 1mM EDTA, pH 7.4. The kit is supplied with 1 ml READ reagent.
3. Mix 1 µl LCAT substrate reagent and 200 µl assay buffer per assay with protein source.
4. Incubate for 4 to 8 hours at 37 °C.
5. Add 100 µl of the incubated mixture to 300 µl of READ reagent and then vortex. Read the fluorescent label at 340 nm excitation and emission at 390 nm and at 470 nm. Do not incubate assay in READ reagent: this will inactivate LCAT.
6. The emission intensities of 390 nm and 470 nm represent the emission of the substrate hydrolyzed (390 nm) and not hydrolyzed (470 nm). A ratio of the two emission intensities (470/390) will indicate an increase in concentration of 390 nm emitter and simultaneous decrease in concentration of 470 nm emitter in the presence of LCAT.

Related Products

Cat no.	Description	Quantity
RB-PRHL	Active, purified recombinant human LCAT	100 µl
RB-LIAG	LCAT inhibitory antiserum, from goat	100 µl

For Research Use Only. Not for Diagnostic or Therapeutic Purposes.

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RB-LCAT Cited References

Roar LCAT Activity Assay Kit

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