

Apo B-48

A Unique Tool for Obesity Drug Development

SAMPLE REQUIREMENTS

Optimum volume: 0.5 mL

Sample Type: Serum or Plasma (Heparin/EDTA)

Method: ELISA

BACKGROUND

Triglyceride-rich lipoproteins (TRL) include chylomicrons (CMs) which are derived from lipids in food, and very low density lipoproteins (VLDL) which are synthesized in the liver.

Apolipoprotein B-48 (Apo B-48) is the protein component of the chylomicrons around which the lipid components of the particle organize. Chylomicrons and chylomicron remnant levels are markedly increased in postprandial hyperlipidemia, and Apo B-48 is specifically distributed in these small intestine-derived lipoproteins. Postprandial hyperlipidemia is associated with:

- Insulin resistance
- Postprandial hyperglycemia
- Metabolic syndrome
- Thickening of the carotid tunica intima and media (atherosclerosis)
- Risk of myocardial infarction

To monitor changes in these chronic conditions, it may be useful to analyze and monitor Apo B-48. Recent research demonstrates that fasting Apo B-48 levels correlate well with an increase in triglyceride AUC after fat loading whereas fasting triglycerides do not correlate well [Kinoshita M. et al., 2005, *Clin. Chim. Acta* 351: 115-120].

This underscores the reality that serum Apo B-48 and triglyceride levels are not always present in parallel.

Apo B-48 contains the same structure (epitopes) present in the N-terminal 48% of the Apo B-100 molecule produced in the liver for secretion in VLDL particles. Because of this structural homology with Apo B-100, it has been difficult to develop a

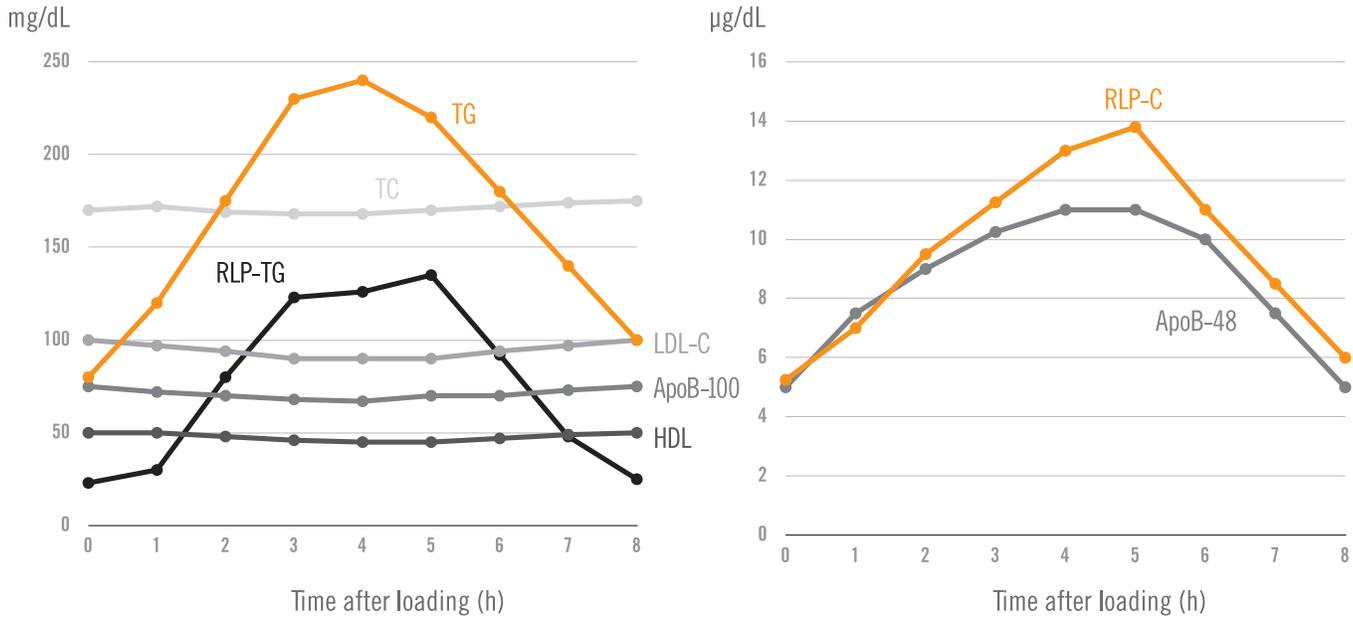
direct measurement specific for Apo B-48. Furthermore, the blood concentration of Apo B-48 of normal individuals during fasting is extremely low, at <2% of the concentration of Apo B-100, adding to the difficulty in developing a simple method to detect and measure Apo B-48 specifically.

Because total Apo B is generally more than 98% Apo B-100, the terms Apo B and Apo B-100 are often used interchangeably. The easiest way to specifically determine Apo B-100 concentration is to subtract Apo B-48 from total serum Apo B. Although the contribution of Apo B-48 is usually low, it is higher in postprandial samples but could be even lower in special circumstances, such as during treatment with drugs that inhibit chylomicron formation, such as microsomal triglyceride transport protein (MTP) and diacylglycerol acyl transferase (DGAT) inhibitors.

Traditional Apo B-48 measurements are difficult and cumbersome (e.g. immuno-affinity chromatography, immunoblotting and protein staining). Large VLDL and small chylomicrons may overlap in density and gel mobility. At Pacific Biomarkers (PacBio), Apo B-48 is measured by a recently available sandwich ELISA assay. This method is specific for Apo B-48 with less than 0.001% cross-reactivity with Apo B-100. The assay has been optimized at PacBio to improve performance. Serum and plasma (EDTA or heparin) specimens are very stable (5 days at 4°C; 2 years at -70°C).

Table: Apolipoprotein Distribution in Lipoproteins

	CM	CM (REMNANTS)	VLDL	VLDL (REMNANTS)	IDL	LDL	HDL
A1	•						•
B-48	•	•					
B-100			•	•	•	•	
C	•		•				•

FIGURE 1: Lipid Markers After Fat Loading

Changes of Apo B-48 concentration correspond to the expected transient accumulation of Chylomicron (CM) particles in circulation after fat loading. These observations are in line with the notion that Apo B-48 is a reliable marker of CM and CM remnants.

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	CM	CM (REMNANTS)	VLDL	VLDL (REMNANTS)	IDL	LDL	HDL
E	•	•	•	•	•		•

Apo B-48 is the most appropriate marker for the measurement of postprandial lipoproteins because it is associated exclusively with intestinally derived chylomicrons and their remnants. Whereas Apo B-100, which is of hepatic origin, associates with VLDL, IDL and LDL.

APO B-48 AT A GLANCE

- Apo B-48 is highly specific for chylomicrons and chylomicron remnants, and is produced only in the intestine after fat ingestion.
- Apo B in lipoproteins are the only apolipoproteins that do not transfer between lipoproteins, making them excellent markers for tracking kinetics of TRL.
- Until the development of the Apo B-48-specific antibodies for use in this ELISA, no really satisfactory method for measurement of serum Apo B48 was available.

- Direct measurement of Apo B-48 by ELISA can be done without prior separation from Apo B-100 containing lipoproteins.
- This Apo B-48 assay is technically more feasible than older methods.
- Apo B-48 by ELISA has improved precision and sensitivity.
- The Apo B-48 ELISA can be a high throughput method.
- This method offers lower cost.

RELATED INFORMATION**APO B-48 IS FREQUENTLY REQUESTED IN CONJUNCTION WITH:**

- Free Fatty Acids (NEFA)
- RLP-C
- RLP-TG
- Lipoprotein Fraction assays
 - e.g., Triglycerides, Cholesterol, Apolipoproteins
- Apolipoprotein B-100
- Apolipoprotein C-II
- Apolipoprotein C-III

APO B-48 IS A USEFUL EFFICACY BIOMARKER FOR SOME DRUG CLASSES:

- Microsomal Transfer Protein Inhibitors (MTPi)
- Diacylglycerol Acyl Transferase Inhibitors (DGATI)
- Fibrates + Statins
- Fat Absorption Blockers
- Cholesterol Absorption Inhibitors

FREQUENTLY REQUESTED TESTING**CARDIOVASCULAR RISK – DIABETES, DYSLIPIDEMIA, METABOLIC SYNDROME & OBESITY**

- Adiponectin
- Apolipoproteins (i.e. AI, B48, CIII, etc)
- CETP (Cholesterol Ester Transfer Protein: Activity and Mass)
- Cholesterol (Total, Esterified, and Free)
- E-Selectin
- Fibrinogen
- Fructosamine
- Glycerol, Free
- Glycomark™
- HDL Cholesterol Subclasses by Precipitation
- HDL Cholesterol
 - Chemical Precipitation (Dextran Sulfate or Hep Mn)
 - Direct Homogeneous Method
- HDL Particle Assay, LpAI:All
- Homocysteine
- ICAM and VCAM
- Leptin
- LDL Cholesterol
 - β -Quantification
 - Direct Homogeneous Method
 - Friedewald Estimation
- Lp (a) [Lipoprotein (a)]
- LpPLA2
- Myeloperoxidase
- Non-Esterified Fatty Acids (NEFA or FFA)
- Oxidized LDL (antigen and autoantibody)
- PAI-1 (plasminogen activator inhibitor)
- Paraoxonase
- RLP-C & RLP-Tg
- sd LDL
- Triglycerides
- VLDL