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**TEST: ANTI- $\beta_2$ -GLYCOPROTEIN I IgG, IgM OR IgA ANTIBODY DETECTION IN HUMAN SERUM BY ELISA**

**PRINCIPLE:**

$\beta_2$  glycoprotein I ( $\beta_2$ GPI) is a 50-kDa glycoprotein that is present in the plasma and binds as a cofactor to anionic phospholipids (such as cardiolipin). It also binds to the cell membrane of activated platelets and endothelial cells which are involved in the antiphospholipid syndrome. During pregnancy,  $\beta_2$ GPI can bind to trophoblast cells. The subsequent binding of anti- $\beta_2$ GPI antibodies may cause the decrease of hCG secretion from trophoblast cells which can lead to defective placentation.  $\beta_2$ GPI also functions as a natural anti-coagulant. Therefore, patients with anti- $\beta_2$ GPI antibodies may be at increased risk for thrombosis. The anti- $\beta_2$ GPI assay may be used in conjunction with the anti-phospholipid antibodies assay for aiding in the diagnosis of antiphospholipid syndrome. The three isotypes of anti- $\beta_2$ GPI (IgG, IgM and IgA) are measured in human serum by ELISA.

Purified  $\beta_2$ GPI antigen is bound to the wells of a polystyrene microwell plate under conditions that will preserve the antigen in its native state. Pre-diluted controls and diluted patient sera are added to separate wells, allowing any  $\beta_2$ GPI IgG antibodies present to bind to the immobilized antigen. Unbound sample is washed away and an enzyme labeled anti-human IgG conjugate is added to each well. A second incubation allows the enzyme labeled anti-human IgG to bind to any patient antibodies, which have become attached to the microwells. After washing away any unbound enzyme labeled anti-human IgG, the remaining enzyme activity is measured by adding a chromogenic substrate and measuring the intensity of the color that develops. The assay can be evaluated spectrophotometrically by measuring and comparing the color intensity that develops in the patient wells with that of a five point calibration curve.

**SPECIMEN REQUIREMENTS:**

Collect one 10 ml red top blood collection tube by standard venipuncture techniques. Do not draw specimen in tubes containing any preservative or anticoagulant. The serum should be separated from the clot; spin clotted blood at 1600 rpm for 10 minutes. Serum can be stored at room temperature for up to 8 hours or can be stored at 2-8°C for up to 48 hours. Store at -20°C or below if testing is delayed further. Send frozen serum on dry ice. Do not freeze in self-defrosting freezers. Avoid repeated freeze-thawing. Microbially contaminated, heat-treated, hemolyzed, lipemic or specimens containing visible particulate should not be used. Minimum volume needed is 100  $\mu$ l of serum.

**METHOD:**

Enzyme Linked Immunoassay (ELISA)

**REFERENCES:**

1. H. Bas de Laat et al. *Clin Immunol* 2004;112:161-168
2. Di Simone et al. *Ann Rheum Dis* 2005;64:462-467

**Normal Range: Negative.**

**Results are reported as negative or positive.**

**Turnaround Time:** 7-10 business days