TEST: LEUKOCYTE ANTIBODY DETECTION

PRINCIPLE:
The cross-match test is an *in vitro* test to determine the presence of anti-lymphocyte antibody to donor cell antigens (lymphocytotoxic antibody) in serum of an individual with preformed antibodies to donor cells. Examples are recipients for an organ transplant or a couple with a history of recurrent spontaneous abortions. The recipient serum is incubated with donor lymphocytes and the binding can be detected by flow cytometry analysis (with fluorescent conjugated reagent). If cytotoxic antibodies are present in maternal serum, they will combine with the surface antigens of donor lymphocytes; the amount of fluorescence on the cells (percentage of positive T or B cells), as measured by flow cytometry, is proportional to the amount of antibody (flow cytometry cross-match).

SPECIMEN REQUIREMENTS:
(Male) 30-40 ml of whole blood collected in green top tubes with heparin from donor. (**Do not refrigerate)**
(Female) One 10ml serum separator tube (gel barrier), from recipient. Separate serum from cells ASAP or within 2 hours of collection by centrifugation.

REJECTION CRITERIA:
(Male) Unacceptable conditions: Cold specimen (due to refrigeration or shipment on ice), extensive clotting, hemolysis or specimens more than 48 hours after collection. If a specimen is more than 48 hours after collection, the lymphocytes will be isolated and viability of the cells will be determined. If viability is greater than 80%, the assay will be performed. If viability is less than 80%, the specimen will be rejected.
(Female) Stability after separation from cells: Ambient: 48 Hours; Refrigerated: 1 week; Frozen: 1 year (avoid repeated freeze/thaw cycles) **Unacceptable conditions:** Plasma or other body fluids. Gross hemolysis

METHOD: Flow Cytometry.

REFERENCES:
1. Cook, D.J., *et al*. Transplant Proc. 20: Suppl. 1:81, 1988.
2. Gilman-Sachs, A., Sung-Ping, L., Beer, A.E., and Beaman, K.D. J. Clin. Lab. Immunol. 30, 53-59, 1989.
3. Inhibition of binding of anti-CD3 antibodies to paternal lymphocytes correlates with failure to immunotherapy for treatment of recurrent spontaneous abortions. Gilman-Sachs,A., Harris, D., Beer, A.E., and Beaman, K.D. J. Reprod. Immunol. 17, 41-51, 1990.

Normal Range: Negative

For specimens positive by flow cytometry, % positive T or B cells will be reported for IgM or IgG antibodies.

Turnaround Time: 2 business days