Policy #:	814 (PLH-814-05)	**Effective Date:**	9/30/2004	**Reviewed Date:**	2/4/2011
Subject:	IMMUNOLOGY ANALYTIC PROTOCOLS				
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IMMUNOLOGY ANALYTIC PROTOCOLS

AUTOIMMUNE DISEASES

The AtheNA Multi-Lyte system detects IgG class antibodies in human sera to ten separate analytes (SS-A, SS-B, Sm, RNP, Scl-70, Jo-a, Centromere B, Histone, dsDNA and qualitative ANA) by a multiplexed bead suspension. The test system is intended to be used as an aid in the diagnosis of various autoimmune disorders.

1. FANA Profile – ANA screen and ANA Titer if screen is positive.
2. Connective Tissue Disease Panel – ANA screen and ANA titer are positive, an anti-dsDNA and an ENA screen are then reflexed. If the ENA screen is positive, eight antibodies (SS-A, SS-B, Sm, RNP, Scl-70, Jo-a, Centromere B, and Histone) are reflexed for analysis.
3. Antibodies to Extractable Nuclear Antigen Panel - If screen is positive, eight antibodies (listed above) are reflexed for analysis.
4. All antibodies may be ordered separately.
5. Vasculitis panel – detects IgG class antibody to 3 separate antigens in human serum: glomerular basement membrane, myeloperoxidase, and proteinase 3.

ELECTROPHORESIS

The Spife 3000 is used for automatic sample application, electrophoresis, staining, and fixing for better resolution and pattern separation for serum and urine proteins, CSF IgG IEF, hemoglobin (normal/abnormal) and immunofixation.

1. Immunoglobulin Quantitation – IgG, IgA, IgM, and protein electrophoresis.
2. Immunofixation (serum and urine) – Protein electrophoresis and immunofixation electrophoresis. MCL now performs an M-spike (a characteristic monoclonal band that is often found in the urine of patients with monoclonal gammopathies) on every 24 hour urine that has orders submitted for quantitative electrophoresis with an immunofixation (IFE). This can be ordered using the mnemonic MONOPRU. This allows the physician to obtain the urine IEP/IFE, M spike calculation, urine volume, urine Total Protein, and urine Creatinine.
3. Hemoglobin Electrophoresis – Alkaline Hemoglobin electrophoresis is used as a screening method for normal to abnormal hemoglobin patterns. Acid hemoglobin electrophoresis is also used for abnormal hemoglobins.
4. Protein Electrophoresis (serum, urine, and CSF) – Protein electrophoresis helps differentiate glomerular, tubular, mixed proteinuria patterns, and monoclonal proteins

in urine. MCL not performs an M-spike (a characteristic monoclonal band that is often found in patients with monoclonal gammopathies) on all tests that contain a serum PEP. If no M-spike is present you will see a "negative" result.

5. L/S Ratio – The determination of lecithin and sphingomyelin levels in amniotic fluid are performed by thin layer chromatography for estimating fetal lung maturity.
6. Oligoclonal profile – aids in diagnosis of inflammatory disease of the CNS by agarose gel isoelectric focusing and immunoblotting CSF and serum.

SHIGATOXIN

EHEC is an immunochromatographic rapid test for the qualitative detection of Shiga toxins 1 and 2 (also called Verotoxins) produced by E. coli. This is used in conjunction with the patient's clinical symptoms and other laboratory tests to aid in the diagnosis of diseases caused by enterohemorrhagic E.coli (EHEC) infections.

***CLOSTRIDIUM DIFFICILE* TESTING**

Early diagnosis and appropriate treatment of *Clostridium difficile* infection are important for preventing complications of infection and decreasing transmission. In order to more reliably identify patients with *C. difficile* infection, the Immunology Laboratory of Medical Center Laboratory has initiate new testing for *C. difficile infection*. The newer test utilizes combined testing for the highly sensitive glutamate dehydrogenase antigen and toxins A and B, and approaches the sensitivity ans specificity of toxogenic culture, which suffers from slow turn around time. However, with more sensitive testing, it is even more important to ensure that only patients with active diarrhea are tested, otherwise the test are more likely to detect asymptomatic patients who are colonized with *C. difficile*. The practice of testing to detect carrier status or "test of cure" does not provide clinically useful information and is discouraged. Despite the adherence to testing criteria to maximize resulting, exceptional cases of patients with *C. difficile* infection who have negative results can show progressive disease.

HELICOBACTER PYLORI

Helicobacter pylori assay is an automated enzyme-linked fluorescent immunoassay for the detection of IgG antibodies to Helicobacter pylori. This assay is intended as an aid in the diagnosis of H. pylori infection in an adult symptomatic population.

ROTAVIRUS

The Rapid Immunocard Rotavirus Assay detects the presence of rotavirus antigen in stool. Rotavirus is a major cause of nonbacterial gastroenteritis especially in the very young and the elderly. Detecting the presence of the virus will facilitate proper patient isolation and eliminate possible unnecessary antibiotic treatment. Co-infection with bacterial pathogens are possible.

FEBRILE AGGLUTININS

Febrile antigens are used in a panel of agglutination tests for the diagnosis of certain febrile diseases such as Salmonellosis, Brucellosis, and Rickettsial diseases.

1. Typhoid H
2. Typhoid O
3. Para A and Para B for Salmonella

4. Rickettsial – Proteus OX19, OXK, and OX2
5. Pasteurella Tularensis
6. Brucella Abortus

LYME SEROLOGY

An automated qualitative enzyme linked fluorescent immunoassay is used for the detection of total antibodies (IgG/IgM) to Borrelia burgdorferi in patients with history, signs, or symptoms suggestive of infection.

HEPARIN ASSOCIATED ANTIBODY

An enzyme linked assay screening test is used in the confirmation of HIT (Heparin Induced Thrombocytopenia) which are antibodies directed against heparin complexes and platelet factor 4. Platelet factor 4 antibodies are present in most patients with HIT.

FUNGAL SEROLOGY

Ouchterlony Double Diffusion is used in determining in vitro precipitating antibodies to four systemic fungal pathogens.

1. Histoplasma capsulatum
2. Blastomyces dermatitidis
3. Coccidiodes immitis
4. Aspergillus fumigatus

HIV –
HIV 1 / 2
A screening test for HIV. If positive, a Western blot will be mailed off.
Analysis performed daily.

INFECTIOUS DISEASE

Epstein Barr Virus IgG (EBV IgG)
Epstein Barr Virus IgM (EBVIg M)
Epstein Barr Nuclear Antigen
Cytomegalovirus IgG (CMV IgG)
Cytomegalovirus IgM (CMV IgM)
Measles (Rubeola) IgG
Mumps IgG
Varicella zoster IgG

SWEAT TEST
Performed on the lotophoresis.
Outpatients are only performed on Tuesdays and must schedule an appointment with Medical Center Laboratory. Inpatients are performed as needed.