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## Expanded Newborn Screening

Inborn errors of metabolism include a vast array of disorders that are caused by deficiencies of specific enzymes or transport proteins. The goal of newborn screening is the presymptomatic diagnosis of disorders for which early treatment can reduce morbidity and mortality. State mandated newborn screening was initiated in the early 1960s to identify infants affected with phenylketonuria (PKU). Screening programs have gradually expanded to include additional inborn errors, but state mandated programs are not uniform across the United States. Until recently, the Missouri Department of Health has screened for five disorders including phenylketonuria, congenital hypothyroidism, congenital adrenal hyperplasia, galactosemia and hemoglobinopathy. On July 1, Missouri expanded its newborn screening panel to test for 20 additional inborn errors, which are categorized into Fatty Acid Oxidation, Organic Acid and Amino Acid Disorders. The complete list includes:

- ◆ Congenital hypothyroidism
- ◆ Congenital adrenal hyperplasia
- ◆ Hemoglobinopathy
- ◆ Galactosemia
- ◆ Fatty Acid Disorders
  - Carnitine/acylcarnitine translocase deficiency (CAT)
  - Carnitine palmitoyl transferase deficiency (CPT)
  - Long-chain hydroxy acyl-CoA dehydrogenase deficiency (LCHAD)
  - Medium-chain acyl-CoA dehydrogenase deficiency (MCAD)
  - Multiple acyl-CoA dehydrogenase deficiency (MAD)
  - Short-chain acyl-CoA dehydrogenase deficiency (SCAD)
  - Trifunctional protein deficiency (TFP)
  - Very long-chain acyl-CoA dehydrogenase deficiency (VLCAD)
- ◆ Organic Acid Disorders
  - Glutaric acidemia, type 1 (GA-1)
  - Isovaleric acidemia (IVA)

- Methylmalonic acidemia (MMA)
- Propionic acidemia (PA)
- 3-hydroxy-3-methylglutaryl CoA lyase deficiency (HMG)
- 3-methylcrotonyl CoA carboxylase deficiency (3MCC)
- ◆ Amino Acid Disorders
  - Argininosuccinic aciduria
  - Citrullinemia
  - Homocystinuria
  - Hypermethioninemia
  - Maple Syrup Urine Disease (MSUD)
  - Phenylketonuria (PKU)
  - Tyrosinemia, type II

This expanded screen is estimated to detect an additional 15 to 20 inborn errors out of the 75,000 newborns tested each year. Results of the screening tests will be reported as normal or abnormal. If the result is abnormal, a quantitative result will be reported along with the expected ranges. The expected ranges, or cutoff levels, were established during a pilot program that involved screening nearly 18,000 samples. The State lab will reevaluate these cutoff levels in six months and modify them as needed. Each abnormal result will also be accompanied by an interpretive comment indicating whether it is considered to be slightly elevated, elevated, or highly elevated and the suggested follow-up. In general, slightly elevated results require prompt repeat newborn screening, or if clinically indicated, diagnostic testing. For elevated and highly elevated results, the State recommends immediate diagnostic testing and consultation with a referral center. Dr. Majed Dasouki, Biochemical and Clinical Geneticist, is available for consultation in the Saint Luke's Health System. His telephone number is 816-932-1785.

No additional blood spots are required for the expanded screening; the State lab will continue to use the 5-spot collection cards. The laboratories at Saint Luke's Hospital and Saint Luke's Northland Hospital will continue their policy of sending letters to parents and contacting the physician of record

when abnormal results are obtained from the State laboratory.

### **New Assay for Activated Protein C Resistance**

Resistance to activated protein C (APC) is a relatively frequent finding in patients with unexplained or familial venous thromboembolism (VTE), detected by a simple clotting test, the APC resistance assay. This abnormality is associated in over 95% of cases with a single point mutation in the factor V gene, termed factor V Leiden. This defect is highly prevalent, occurring in the heterozygous form in 5% of healthy Caucasians, and in 30-50% of patients with recurrent VTE. It is the most common hereditary cause of thrombosis, imparting an 8-fold increased risk of VTE in heterozygotes and an 80-fold increased risk of VTE in homozygotes. The APC resistance clotting test is an important component of laboratory panels for venous thrombosis. A normal result rules out factor V Leiden, while an abnormal result should be followed up with a PCR assay to confirm the presence of factor V Leiden.

Up until recently, most APC resistance assays were based on the APTT. While the newer versions of the APTT-based assays have offered good sensitivity (98-100%) and specificity (98-100%) for factor V Leiden diagnosis, they have had a number of disadvantages. Most significantly, lupus anticoagulants interfere with these assays, which is a problem in view of the high prevalence of both APC resistance and lupus anticoagulants in thrombophilic populations. High factor VIII levels may also affect the APTT-based assays. Furthermore, the APTT-based assays do not always provide perfect discrimination between normal individuals and those with factor V Leiden.

A new APC resistance assay has recently become available, using snake venoms to activate factor V and prothrombin, thus involving the lower portion of the clotting pathway and eliminating interference by factor VIII, and in the absence of phospholipid-based complexes, eliminating interference by lupus anticoagulants. Furthermore, the new assay has so far been shown to be 100% sensitive and specific for factor V Leiden, with excellent and wide discrimination between normal individuals and carriers of the factor V Leiden mutation. The assay is not affected by anticoagulation with heparin (standard or low molecular weight) or warfarin.

Saint Luke's Regional Laboratories switched to the new improved APC resistance assay in early July, 2005. As previously, the APC resistance assay is available as part of the venous thrombosis panels, or as an individual test. The sample requirement remains the same – one light blue-top (sodium citrate) tube, and the test is run on Monday, Wednesday and Friday. The reference range has changed to  $\geq 2.5$ . If an abnormal result is obtained ( $< 2.5$ ), the laboratory will automatically follow up with a confirmatory PCR assay for factor V Leiden

### **Cortisol Testing Available 24 x 7**

In order to support the increasing use of steroid replacement therapy for critically ill patients at Saint Luke's Hospital, the laboratory has begun offering stat cortisol testing on a 24 hours per day, 7 days per week basis.

The Cortrosyn, or ACTH, Stimulation test consists of a baseline specimen collected prior to the administration of Cortrosyn and two post-stimulation specimens collected at 30 and 60 minutes after dosing. Because of the continuing confusion that has existed concerning the ordering of this test, the "Cortrosyn" mnemonic that was previously used to order Cortrosyn stimulation has been removed from STAR. From this point forward, a cortisol should be ordered for each specimen with either Stat or Timed draw status. Each result will be reported with the collection time. Specimen requirement is a red top gel tube.

### **Reflex Testing**

Reflex testing refers to those situations where an initial test result is abnormal and the laboratory automatically performs follow-up testing. Federal regulations require that laboratories inform physicians of their reflex test policy. Saint Luke's Regional Laboratories offers the following reflex tests.

<b>Initial Test</b>	<b>Reflex Test</b>
Activated Protein C Resistance	Factor V Leiden
CBC only	CBC w/ Diff
Protein electrophoresis	Immunofixation
Thyroid cascade	ft4 & Total T3
HIV antibody	Western Blot
Lyme antibody	Western Blot