



**Colorado Department of Public Health  
and Environment  
Laboratory Services Division**

Colorado Department  
of Public Health  
and Environment

**2012 Newborn Screening  
Program Report**



## Introduction

According to the American Academy of Pediatrics, “Newborn screening is one of the nation's most successful public health programs.”<sup>1</sup> Newborn screening programs test babies for disorders that often have no immediate visible effects on a baby; however, unless detected and treated early, these disorders can cause physical problems, mental retardation and, in some cases, death. “More than 4 million newborns are screened annually in the United States, and thousands of infants are rescued from disability and death,” per the Centers for Disease Control and Prevention.<sup>2</sup>

When a disorder is identified, appropriate medical specialists are consulted for their expertise in metabolic diseases, cystic fibrosis, endocrinology or hematology, depending on the condition and family needs. The March of Dimes stated, “Since most of the conditions included in the newborn screening panel are caused by genetic mutations, families need to be referred for genetic education and counseling to best understand the particular condition, its impact on the child's health and future and the risks in future pregnancies.”<sup>3</sup>

## History of Newborn Screening

Before testing was developed, phenylketonuria (PKU), a metabolic disease that can be addressed by diet, resulted in retardation and often institutionalization for victims. In the 1930s, George Jervis at Letchworth Village State School in Thiells, N.Y., identified 50 clients whose mental retardation was attributed to phenylketonuria (PKU).<sup>4</sup> Pursuing the study in four state institutions, he identified a total of 185 PKU cases among 15,000 patients.

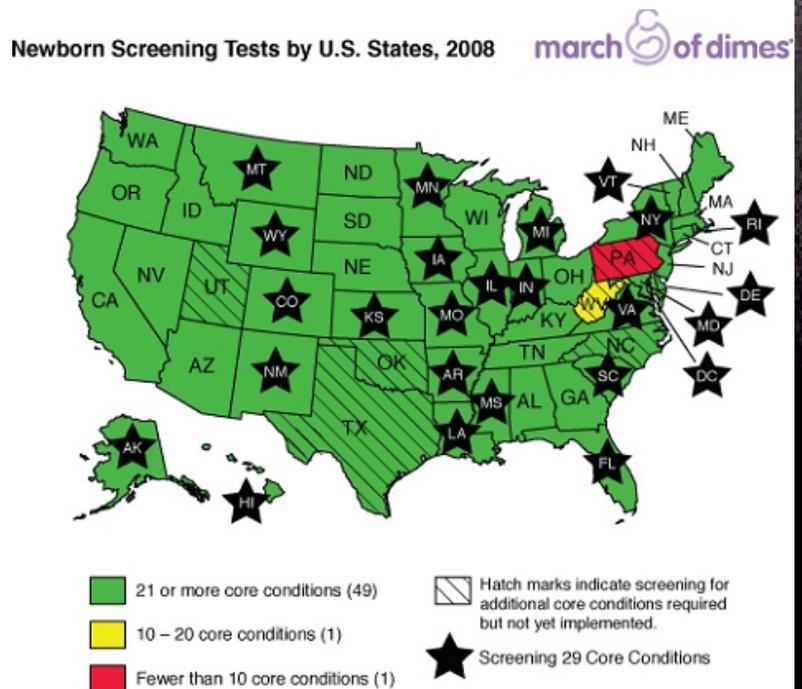
While adults could not be helped, the work of Horst Bickel suggested that early diet therapy could prevent development of the mental retardation usually seen in PKU.<sup>5</sup> Effective treatment depended on early therapy, which required early detection of the affected child before symptoms appeared.

Robert Guthrie, a microbiologist and pediatrician at State University of New York at Buffalo, created a simple, inexpensive PKU screening lab test, to be performed on newborns soon after birth.<sup>6</sup> Following a successful pilot study by Guthrie in the 1960s, which encompassed 29 states and 400,000 newborns, many U.S. states implemented newborn (PKU) screening programs. All 50 states currently provide newborn screening for PKU.<sup>7</sup>

According to the March of Dimes, as of Feb. 18, 2009, “All 50 states and the District of Columbia now require that every baby be screened for 21 or more of the 29 serious or functional disorders on the uniform panel recommended by the American College of Medical Genetics (ACMG) and endorsed by the March of Dimes.”<sup>8</sup>

Currently there is no federal law regulating newborn screening programs. Each state, under its own laws, operates its own newborn screening program and establishes its own policies and procedures.

State programs vary widely in the number and types of conditions for which they test. Some states test for as few as 10 disorders, while others test for 50 or more.<sup>8</sup>



## Colorado Newborn Screening Timeline

1965	Screening for Phenylketonuria (PKU) began in Colorado.
1979	<p>With the aid of a three-year federal grant from the Office of Maternal and Child Health, testing for five other conditions was added: congenital hypothyroidism (CH), homocystinuria (HCY), maple syrup urine disease (MSUD), hemoglobin disease and galactosemia (GALT).</p> <p>The grant funded the establishment of a regional laboratory within the department's Division of Laboratories. Arizona, Wyoming and New Mexico joined Colorado in establishing the Denver-based operation. Over the years, Arizona and New Mexico have established their own state screening programs; Wyoming continues to use the Colorado lab for its newborn screening program.</p> <p>Participation in the first few years of the program was voluntary, and testing was paid for by the grant.</p>
1981	<p>Recognizing the benefit of the screening program to the citizens of the state, the Colorado Legislature passed the "Newborn Screening and Genetic Counseling and Education Act" requiring all infants born in Colorado to be tested for six conditions: phenylketonuria, congenital hypothyroidism, homocystinuria, hemoglobin disease, galactosemia, and MSUD.</p> <p>This law also allowed for cash funding of the Newborn Screening Program, and a fee-for-service program of laboratory testing was instituted in July 1981.</p>
1983	In May 1983, the newborn screening testing fee was increased to cover genetic counseling and education.
1987	On July 1, 1987, a screening test for cystic fibrosis was added to the newborn screening test panel.
1989	On April 1, 1989, a screening test for biotinidase deficiency was added to the panel of tests.
1996	<p>In 1996, the Board of Health discontinued screening for MSUD and HCY. HCY was discontinued because changes in the medical care and feeding of neonates had rendered the existing available testing methodology unreliable. MSUD was discontinued because its incidence in Colorado was not high enough to warrant screening. These criteria and others used to determine the inclusion of tests in Colorado's screening program are defined in the screening statute.</p> <p>Legislation was passed requiring a second specimen (<i>8-14 days of age</i>) to be collected on all babies, which is tested for specific disorders. There was sufficient evidence in the screening literature to justify a second screen on all babies. The concern was that early discharge of infants from the hospital put affected infants at risk to be missed because the screening specimen was collected at too early an age.</p>
1997	Screening of newborns was added to diagnose infants for hearing acuity and to provide prompt and effective early interventions for those infants who are hard of hearing or deaf. Passage of House Bill 97-1095 required the Colorado Infant Hearing Advisory Committee to develop guidelines for reporting and for ensuring that identified children receive referral for appropriate follow-up.
2000	Screening for congenital adrenal hyperplasia (CAH) was added to the newborn screening panel of tests.
2006	On July 1, 2006, expanded newborn screening was added via Tandem Mass Spectrometry (MS/MS) technology, adding testing for 23 additional disorders to include: fatty acid oxidation disorders, aminoacidopathies and organic acidemias. With the introduction of MS/MS screening, MSUD and HCY were reintroduced to the testing panel.
2012	On February 1, 2012, Severe Combined Immunodeficiency (SCID) testing was added using "real time" PCR to determine the levels of T-cell Recombinant Excision Circles (TREC). The first abnormal results reported in the first case identified by newborn screening.

## Newborn Screening Statute: Criteria for Addition of Disorders

The Board of Health uses the following criteria to determine whether or not to test infants for conditions:

1. The condition for which the test is designed presents a significant danger to the health of the infant or his/her family and is amenable to treatment.
2. The incidence of the condition is sufficiently high to warrant screening.
3. The test to detect the condition meets commonly accepted standards of reliability as demonstrated through research or use in another state or jurisdiction.
4. The cost-benefit consequences of screening are acceptable within the context of the total newborn screening program.

### Colorado Newborn Screening Quick Facts

Approximate number of births per year: 66,629

Colorado Statute: Colo. Rev. Stat. §25-4-1001, et seq. - addresses testing and fees.

Section 25-4-1004.5(3) C.R.S. - addresses addition of CAH, and testing responsibilities.

Screening Requirements: Initial and second test required by law on all newborns.

NBS Fee: \$92 covers first and second screen and follow-up.

Aidan is 9 years old and has a metabolic condition known as phenylketonuria, or PKU. If left untreated, PKU usually leads to mental retardation, stunted physical development and emotional problems.

Aidan, however, is the tallest kid in his class, has an IQ well above normal and plays on a competitive soccer team. This is possible only because Aidan's metabolic condition was detected shortly after birth, and he has been on a specialized diet ever since.

The newborn screening test administered to Aidan within hours of his birth detected this inherited metabolic disease at such an early stage that we were able to implement a customized diet and completely avoid any of the negative developmental consequences traditionally associated with PKU. Our gratitude to the CDPHE Newborn Screening Program and the staff at Denver Children's Hospital cannot be measured.

As an employee of the Colorado Department of Public Health and Environment, I am aware of many of the preventative programs that are implemented at the state level. My son's tremendous vitality reminds me about the far-reaching impact that these programs have.



Aidan Bell

*Korey Bell  
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## Current Disorders Tested for at the State Laboratory

CATEGORY	DESCRIPTION	DISORDERS
<b>AMINO ACID DISORDERS</b>	Some babies lack enzymes that are needed to break down the building blocks (amino acids) of protein, while others have deficiencies in enzymes that help the body rid itself of the nitrogen incorporated in amino acid molecules. Toxic levels of amino acids or ammonia build up in the body causing a variety of signs and symptoms, or death.	Arginase deficiency Argininosuccinic acidemia Citrullinemia Homocystinuria Hypermethioninemia Maple syrup urine disease Phenylketonuria (PKU)
<b>ENDOCRINE DISORDERS</b>	CAH is a group of disorders in which there is a deficiency of certain hormones, sometimes affecting genital development. In severe cases, CAH also can cause life-threatening salt loss from the body. Lifelong treatment with the missing hormones suppresses this disease.	Congenital adrenal hyperplasia
	Congenital hypothyroidism is a thyroid hormone deficiency that retards growth and brain development. If it is detected in time, a baby can be treated with oral doses of thyroid hormone to permit normal development.	Congenital hypothyroidism
<b>Fatty acid oxidation disorders</b>	This group of disorders is characterized by inherited defects of enzymes needed to convert fat into energy. When the body runs out of glucose (sugar), it normally breaks down fat to support production of alternate fuels (ketones) in the liver. Because individuals with these disorders have a block in this pathway, their cells suffer an energy crisis when they run out of glucose. This most often occurs when an individual is ill or skips meals. Without treatment, the brain and many organs can be affected, sometimes progressing to coma and death.	Carnitine acylcarnitine translocase deficiency Carnitine palmitoyltransferase II deficiency Carnitine palmitoyltransferase deficiency 1a Carnitine uptake defect Long-chain L-3-hydroxyacyl-CoA dehydrogenase deficiency Medium-chain acyl-CoA dehydrogenase deficiency Short-chain acyl-CoA dehydrogenase deficiency Trifunctional protein deficiency Very long-chain acyl-CoA dehydrogenase deficiency
<b>Hemoglobinopathies</b>	These inherited diseases of the red blood cells result in varying degrees of anemia (shortage of red blood cells) and other health problems. The severity of these disorders varies greatly from one person to the next. Babies have an increased risk of infections,	Beta-thalassemia Sickle cell anemia Hemoglobin SC disease
<b>Organic acid disorders</b>	Organic acid disorders are a group of inherited metabolic conditions. Each disorder is associated with a specific enzyme deficiency that causes the accumulation of organic acids or their metabolites in the blood and urine, resulting in the clinical features of these disorders. Typically, newborns appear normal for the first days of life, but then may develop vomiting, poor feeding, failure to thrive, hypoglycemia, hyperammonemia, seizures, hypotonia and lethargy, progressing to coma. Common findings include ketosis, metabolic acidosis and, in some cases, an unusual odor. Many individuals affected with organic acid disorders have a significant risk of death during infancy. <sup>9</sup>	3-Hydroxy-3-Methylglutaryl-CoA Lyase deficiency 3-Methylcrotonyl-CoA carboxylase deficiency 3-Methylglutaconic aciduria (3-MGA) Beta-ketothiolase deficiency Biotinidase deficiency Glutaric acidemia type I Glutaric acidemia type II Isovaleric acidemia Malonic acidemia Methylmalonic acidemia Multiple carboxylase deficiency Propionic acidemia
<b>Cystic fibrosis</b>	CF is an inherited disease that affects the normal movement of salt (sodium chloride) and water into and out of certain cells, including those that line the lungs and pancreas. This results in thick, sticky mucus and other secretions, which can clog and damage lungs, cause lung infections and lead to early death. Thick, digestive fluids also prevent digestive enzymes from reaching the small intestine, causing digestive problems, slow growth and malnutrition.	
<b>Galactosemia</b>	Galactosemia is a rare hereditary condition caused by the body's inability to breakdown galactose (a sugar found in milk and milk products). The high levels of galactose poison the body causing serious damage such as a swollen and inflamed liver, kidney failure, stunted physical and mental growth, and cataracts in the eyes.	
<b>Severe Combined Immunodeficiency</b>	Severe combined immunodeficiency (SCID) represents a group of rare, sometimes fatal, congenital disorders characterized by little or no immune response. The defining feature of SCID, commonly known as "bubble boy" disease, is a defect in the specialized white blood cells (B- and T-lymphocytes) that defend the body from infection by viruses, bacteria and fungi. Without a functional immune system, SCID patients are susceptible to recurrent infections such as pneumonia, meningitis and chicken pox, and can die before the first year of life.	

## Specimen Collection

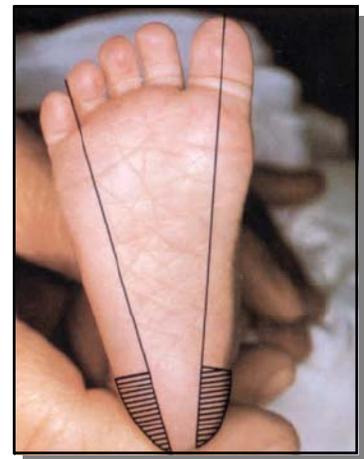
Testing is performed on capillary blood collected by a heel stick from newborns before they leave the hospital or other birthing facility. The timing of collection and specimen quality are important factors. The receipt of a good quality specimen in the laboratory in a timely fashion permits early identification of infants at risk for one of these diseases. Since symptoms are nonspecific or absent in the newborn, irreversible damage to the infant may occur if laboratory diagnosis is delayed.

For healthy, full term infants, the specimen should be collected as late as possible before discharge, but no later than 24-48 hours. For sick or premature infants, the specimen should be collected no later than 24-48 hours of age unless one of the diseases is suspected. The blood is applied to a special filter paper and sent within 24 hours, by first class mail or other expedient means, to the laboratory. The newborn screening laboratory supplies the collection forms for newborn screens, pre-addressed envelopes for mailing and informational brochures explaining the program to parents.

Approximately 1.5% of all specimens (over 2,000 per year) are rejected because they are unsatisfactory for testing.

Reasons for specimen rejection include:

1. Damage of the blotter and/or uneven application of the blood by the use of capillary tubes, especially micro hematocrit tubes
2. Contamination of the blotter or blood spots with alcohol, urine, powder from gloves or other substances
3. Improper drying of the specimen
4. Over-application or layering of the blood
5. Quantity of blood insufficient to perform tests
6. Serum or tissue fluids evident in sample
7. Specimens too old (>14 days after collection)
8. Application of blood to both sides of filter paper
9. Use of cord blood to saturate the blotter

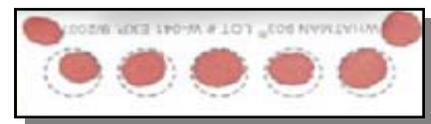
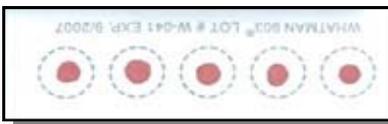


Above: Proper heel stick region (hatched area) on newborn foot for obtaining a newborn screening sample<sup>10</sup>

Unsatisfactory newborn specimens rejected by the lab are shown below.

Left: Quantity insufficient

Right: Over application



Satisfactory specimen for newborn screening testing is shown below.

Specimens are not rejected when the infant has been transfused or has undergone dialysis. However, a transfusion may interfere with the results of the testing, especially for galactosemia and hemoglobin screening, where the possibility of a false negative may result. The screening specimen should be drawn before a transfusion or dialysis if the condition of the infant permits.

When an unsatisfactory specimen is received, the submitting health care provider is notified by telephone immediately. This procedure allows for collection of a new specimen from the infant as soon as possible. The rapid progression of some of the diseases, such as, galactosemia and congenital adrenal hyperplasia, makes it critical that a repeat specimen can be submitted to the lab quickly. Training on the proper collection of specimens occurs via on-site demonstrations, and online classes are available. Approximately 110 people have taken the online training.

## Testing Detection Methods used by the Newborn Screening Lab

Disease category	Test method
<b>Amino acid disorders</b>	Tandem Mass Spectrometry (MS/MS)
<b>Endocrine disorders</b>	Perkin-Elmer Auto-DELFI <sup>®</sup> (time resolve fluoro-immuno assay)
<b>Fatty acid oxidation disorders</b>	Tandem Mass Spectrometry
<b>Hemoglobinopathies</b>	Isoelectric focusing
<b>Organic acid disorders</b>	Tandem Mass Spectrometry (Except biotinidase deficiency which is a colorimetric assay)
<b>Cystic fibrosis</b>	Perkin-Elmer Auto-DELFI <sup>®</sup> (time resolved fluoro-immuno assay)
<b>Galactosemia</b>	Fluorometric assay
<b>Severe Combined Immunodeficiency</b>	Real Time Polymerase Chain Reaction (PCR)



(Above) Scientist, Mark Dymerski, performs newborn screening testing using the MS/MS instrument.



(Above) Lab technician, Kay Reilly, uses Perkin-Elmer dried blood spot multi-puncher to distribute 3 millimeter blood disks, which are used for newborn screening testing.

## Reporting and Follow-Up

All positive screening results requiring immediate follow-up are telephoned to the physician of record and/or appropriate specialist. This telephone report is followed by a written report, sent by certified mail, containing information about the results and recommending procedures for follow-up. There are agencies available in each state that provide consultation, clinical evaluation, follow-up laboratory testing and genetic counseling.

Numbers of Cases Identified Through the Newborn Screening Program in the Last Three Years (Three-year period consists of calendar years 2009, 2010 and 2011.)					
Abbreviation	Condition name	2009	2010	2011	TOTAL
CAH	Congenital adrenal hyperplasia	4	6	8	18
CH	Congenital hypothyroidism	30	32	30	92
CF	Cystic fibrosis	18	16	11	45
BIOT	Biotinidase deficiency (all "partial" except for two "profound" cases in 2009 and 2010)	4 1 before MS/MS 1 after MS/MS	4 1 before MS/MS 1 after MS/MS		8
3-MCC	3-methylcrotonyl-CoA carboxylase deficiency	4	3	1	8
CACT	Carnitine-acyl-carnitine translocase deficiency		1		1
CIT	Citrullinemia	2	1		3
CPT1	Carnitine palmitoyltransferase deficiency	1			1
GA1	Glutaric acidemia type 1		2	5	7
GAL	Galactosemia	2	3	2	7
IVA	Isovaleric acidemia				
MCADD	Medium chain acyl-CoA dehydrogenase deficiency	3	2	3	8
MGA	3-methylglutaconic aciduria				
MMA	Methylmalonic acidemia	1 (mutase)		1 (mutase)	2
MCD	Multiple carboxylase deficiency				
PKU	Phenylketonuria	2	7	7	16
VLCADD	Very long chain acyl-CoA dehydrogenase deficiency	1	3	2	6
TOTALS		72	80	70	222

### Not provided in the table above:

During the same three-year period, 2009 through 2011, the Sickle Cell Treatment and Research Center at University of Colorado-Denver and The Children's Hospital diagnosed the following disorders through newborn screening (*right*):

17 cases of sickle cell anemia	2 D beta plus thalassemia
8 sickle C disease	1 C beta plus thalassemia
2 sickle beta zero thalassemia	1 beta thalassemia major

## Incidence of Disease

The following data represent occurrent births, or babies born in Colorado hospitals that are screened at the Colorado Department of Public Health and Environment's Newborn Screening laboratory during the period of 2009 through 2011. The incidence rates below do not reflect the out-of-state residents delivering babies in Colorado.

Out-of-state residents delivering babies in Colorado are primarily from Wyoming, which has a very low birthrate.

Disease	Number of cases	Incidence Rate**
Congenital hypothyroidism	92	1:2,172
Congenital adrenal hyperplasia (classic only)	18	1:11,105
Cystic fibrosis	45	1:4,441
All hemoglobinopathies (including sickle cell anemia)	32	1:6,246

Number of live births (3-year period 2009-2011): 199,887.



*(Above) A nursing professional collects a newborn screening specimen from an infant's heel.*

## False Positives and False Negatives

A test result reported erroneously as positive in the absence of true disease is designated as a false positive and a test result reported as negative, failing to reveal a disease, is designated as a false negative. Both false positive and false negative test results are a consequence of a test's sensitivity, specificity and normal range values, which are established based on test methods used.

The reported missed-case rate for cystic fibrosis in Colorado is 5.4 percent, 95 percent confidence interval, 3.8-10.0, due to infants with normal levels of immunoreactive trypsinogen, the target analyte of the cystic fibrosis newborn screening. To improve the detection rate of infants with cystic fibrosis, the algorithm was changed on June 1, 2008, with an expected missed case rate of <1 percent. All other newborn screening tests have a sensitivity of greater than 99 percent and false negatives are rare.

<b>Expanded Newborn Screening Summary</b>			
<i>Data provided by the Inherited Metabolic Diseases (IMD) clinic at The Children's Hospital-Denver</i>			
<b>Date Range</b>	<b>2009</b>	<b>2010</b>	<b>2011</b>
<b>Total number of first screens Colorado only</b>	68,548	65,778	64,832
<b>Organic acidemias</b>	108	148	113
<b>Fatty acid oxidation disorders</b>	133	144	121
<b>Amino acidemias</b>	124	159	238
<b>Biotinidase deficiency</b>	8	12	1
<b>Galactosemia</b>	2	7	2
<b>Total abnormal screens reported</b>	375	470	475
<b>Percentage of abnormal screens from infants in neonatal ICU</b>	183 (49%)	246 (52%)	320 (67%)
<b>True positives</b>	24	37	25
<b>Maternal true positives</b>	4	8	3
<b>False Negatives (missed cases)</b>	0	0	1

The Colorado Department of Public Health and Environment's Newborn Screening Laboratory continues to effectively identify newborns at risk for more than thirty disorders. The early identification and intervention for infants have prevented or reduced the morbidity and mortality associated with these disorders. The test protocols used in the laboratory are in common use in all newborn screening laboratories throughout the country. They exhibit an appropriate level of sensitivity and specificity for screening large numbers of newborns. Recognizing the rare instance of any screening test giving a false negative, it is important to keep in mind that a "normal" newborn screening result alone cannot be used to rule out one of these diseases in a symptomatic child.

### POST LABORATORY FOLLOW-UP

Newborn screening does not end with laboratory testing and the reporting of results. Follow-up steps subsequent to testing are crucial to a healthy infant outcome. The Colorado Department of Public Health and Environment's Newborn Screening Follow-up Program, which is housed in another division of the health department (Prevention Services Division-Children, Youth and Families Branch) tracks children with abnormal newborn screening results, whose true disease risk is unknown until the potential diagnosis is confirmed or ruled out through subsequent diagnostic testing not performed at the state lab.

Staff employed by, or under contract to, the state health department follows up on different categories of abnormal screening results. Follow up activities include contacting primary care providers, medical specialists, families and others to confirm test results and clinical findings and verify initiation of treatment when applicable.

## References

<sup>1</sup> American Academy of Pediatrics, Newborn Screening Task Force. *Newborn Screening: A Blueprint for the Future*. *Pediatrics*. 2000;106(2)S:383-427.

<sup>2</sup> U.S. Department of Health and Human Services Centers for Disease Control and Prevention National Center for Environmental Health Division of Laboratory Sciences. *Newborn Screening Laboratory Bulletin, October 2008*. <http://www.cdc.gov/nbslabbulletin/bulletin.html#1>

<sup>3-6</sup> March of Dimes ([http://www.marchofdimes.com/professionals/24279\\_9606.asp](http://www.marchofdimes.com/professionals/24279_9606.asp))

<sup>4</sup> Jervis G. Phenylpyruvic oligophrenia: introductory study of 50 cases of mental deficiency associated with excretion of phenylpyruvic acid. *Archives of Neurology and Psychiatry* 1937;38:944.

<sup>5</sup> Bickel, H., Gerrard, J., Hickmans, EM. *Influence of phenylalanine intake on phenylketonuria*. *Lancet* 1953;2:812.

<sup>6</sup> Guthrie, R., Susi, A. *A simple phenylalanine method for detecting phenylketonuria in large populations of newborn infants*. *Pediatrics* 1963;32:338-43.

<sup>7</sup> Guthrie, R., Whitney, S. *Phenylketonuria detection in the newborn infant as a routine hospital procedure: a trial of a phenylalanine screening method in 400,000 infants*. Children's Bureau Publication 419. Washington (DC): U.S. Department of Health, Education and Welfare; 1964.

<sup>8</sup> March of Dimes, [http://www.marchofdimes.com/professionals/24279\\_9606.asp](http://www.marchofdimes.com/professionals/24279_9606.asp)

<sup>9</sup> Illinois Department of Health - Genetics and Newborn Screening, <http://www.idph.state.il.us/HealthWellness/fs/organic.htm>

<sup>10</sup> Whatman Neonatal Screening: Blood Specimen Collection and Handling Procedure