Newborn screening of Rare Diseases in Iceland and other Nordic Countries

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Causes of Inborn Errors of Metabolism

- Lack of enzyme
- Loss of enzyme activity
- Disturbance in enzyme-cofactor

Cell Membrane

Outside

Inside

Loss of transport protein

Enzyme activity

Disturbance in other Metabolic reactions

Carnitine -> Acyl Carnitine

Diurnal Variation
Inborn Errors of Metabolism (IEM)

- Symptoms can show up at birth, month, year, decades after birth or never
- Symptoms can be from heart, liver, neurological, acid base disturbances and abnormal mental and physical development
- Symptoms can be triggered by medications, infections, vaccination, fasting, accidents (trauma), surgery
- Some diseases can cause sudden death at any age
- For some diseases “easy” cure by change of diet, vitamins etc
Methods:
MRM function: Measures one analyte only
Scan functions: Measures all analytes (acylcarnitines, aminoacids) with certain characteristics
Iceland uses the same setup for NBS as for Clinical Testing
**Tandem Massspectrometer (MsMs)**

- MsMs measures abnormal concentration of an analyte, which can be a marker for one or more IEM
- Nearly all IEM show elevation in concentration of markers
- Elevation of one marker can cause elevation/decrease in concentration of other markers – spectral pattern (scan method)
- Different sensitivity of markers to detect diseases
- With MsMs 36 different acylcarnitines are being measured
- Measure 20 aminoacids

- Preparation of samples 2 hours
- All measurements 2.20 minutes.
Selection of Diseases
- The 10 commandments of screening –
  Wilson & Jungner Criteria (WHO, 1968)

- The condition should be an important health problem
- There should be an accepted treatment for patients with recognised disease
- Facilities for diagnosis and treatment should be available
- There should be a recognised latent or early symptomatic stage
- There should be a suitable and simple test or examination
- The test should be acceptable to the population
- The natural history of the condition, including development from latent to declared disease should be adequately understood
- There should be an agreed policy on whom to treat as patients
- The cost of case-finding (including diagnosis and treatment of patients diagnosed) should be economically balanced in relation to possible expenditure on medical care as a whole
- Case-finding should be a continuous process and not a “a once and for all” project
Selection of Diseases

- **Denmark:** Pilot project 2002-2005. Report 2008
- **Iceland:** Committee (Pediatrics, Lab and the Director of Health) 2007. NBS Started in 2008
- **Sweden:** Committee (Pediatrics and Lab 2011). NBS started 2011.
- **Finland:** No NBS except for Congenital Hypothyroidism from cord blood.
# NBS - Core conditions

<table>
<thead>
<tr>
<th>Core Conditions</th>
<th>Abbreviation</th>
<th>Full Name of Condition</th>
<th>Markers L=Low H=High</th>
<th>US</th>
<th>Iceland</th>
<th>Denmark</th>
<th>Sweden</th>
<th>Norway</th>
<th>Germany</th>
<th>Netherlands</th>
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<td>Yes</td>
<td>Yes</td>
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<td>CAH</td>
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<td>Congenital Adrenal Hyperplasia</td>
<td>HO-Progesteron</td>
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<td>Yes</td>
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<td>Hemoglobin</td>
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<td>Biotinidase</td>
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<td>Yes</td>
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<td>Galactocemia</td>
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<td></td>
<td></td>
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<td>No</td>
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<td>Cystic Fibrosis</td>
<td>CF</td>
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<td></td>
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<td>No (Exp)</td>
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<td>Yes</td>
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<td>Severe Combine Immune Deficiency</td>
<td>SCID</td>
<td>Severe Combine Immune Deficiency</td>
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<td>No (Exp)</td>
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<td>No (exp)</td>
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<td>Lysosomal Storage diseases</td>
<td>LSD</td>
<td>Pompe, Krabbe, Niemand-Pick, Fabry, Gaucher, MPS I,II</td>
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</table>
# MsMs-Core Metabolic Conditions (20)

| Fatty Acid Oxidation disorder | Abbreviation | Full Name of Condition | Markers L=Low H=High | US | Iceland | Denmark | Sweden | Norway | Germany | Netherlands | Icel. Found |
|--------------------------------|--------------|------------------------|----------------------|----|---------|---------|--------|--------|---------|------------|-------------|-------------|
| CTD/CUD | Carnitin Transport Defect/Carnitin Uptake Defect | L: C0, (C2, C3, C16, C18, C18:1) | Yes | Yes | Yes | Yes | Yes | No | No | 3 cases |
| LCHAD | Long-Chain L-3-hydroxyacyl-CoA dehydrogenase deficiency | H: C16:1-OH, C16-OH, C18:1-OH, C18-OH | Yes | Yes | Yes | Yes | Yes | Yes | Yes |
| MCAD | Medium-chain-acyl-CoA dehydrogenase deficiency | H: C8, C6, C10:1, C10 | Yes | Yes | Yes | Yes | Yes | Yes | Yes |
| TFP | Trifunctional protein deficiency | H: C16:1-OH, C16-OH, C18:1-OH, C18-OH | Yes | Yes | Yes | Yes | Yes | Yes | Yes |
| VLCAD | Very long-chain acyl-CoA dehydrogenase deficiency | H: C14:1, C14:2, C14, (C16) | Yes | Yes | Yes | Yes | Yes | Yes | Yes |
| DERED | Dienoyl reductase deficiency | C10:2 | Some (Yes) | No | No | No | No | No | No |
| Organic Acid disorders | Abbri-vation | Full Name of Condition | Markers L=Low H=High | US | Iceland | Denmark | Sweden | Norway | Germany | Netherl | Icel. Found |
|------------------------|-------------|------------------------|-----------------------|----|--------|---------|--------|--------|---------|---------|----------|-----------|
| GA-1                   | Glutaric acidemia type 1 | H: C5DC | Yes | Yes | Yes | Yes | Yes | Yes | Yes | Yes | Yes | 1 case |
| IVA                    | Isovaleric Acidemia (Isovaleryl-CoA dehydrogenase deficiency) | H: C5 | Yes | Yes | No | No | No | No | Yes | Yes | Yes |
| 3-MCC                  | 3-Methylcrotonyl-CoA carboxylase deficiency | H: C5-OH | Yes | Yes | No | No | No | No | Yes | Yes | Yes |
| HMG                    | 3-Hydroxy3-methylglutaric acidemia (3-Hydroxy 3-methylglutaryl-CoA lyase deficiency) | H: C5-OH, C6DC | Yes | Yes | No | No | No | No | Yes | Yes | Yes |
| BKT                    | Beta ketothiolase def; short-chain ketoacyl thiolase deficiency | H: C5:1, C5-OH | Yes | Yes | No | Yes | Yes | Yes | No | No | No |
| MCD HLCS               | Multiple carboxylase deficiency (Holocarboxylase synthetase deficiency) | H: C5-OH C3* | Yes | Yes | Yes* | Yes* | Yes* | Yes* | Yes | Yes | 1 mat case |
| Cbl-A,B                | Methylmalonic acidemia (Vitamin B12 disorders) | H: C3 C16:1-OH | Yes | Yes | No | Yes | Yes | Yes | No | No | No |
| MUT                    | Methylmalonic Acidemia (methylmalonyl-CoA mutase deficiency) | H: C3 | Yes | Yes | Yes | Yes | Yes | Yes | No | No | No (exp) |
| PROP                   | Propionic acidemia (Propionyl-CoA carboxylase deficiency) | H: C3 | Yes | Yes | Yes | Yes | Yes | Yes | No | No | No (exp) |
| Amino Acid Disorders | Abbreviation | Full Name of Condition                                                                 | Markers L=Low H=High | US | Iceland | Denmark | Sweden | Norway | Germany | Netherl. | Icel. Found |
|---------------------|--------------|----------------------------------------------------------------------------------------|----------------------|----|---------|---------|--------|--------|---------|----------|-----------|----------------|
| ASA                 | Argininosuccinate acidemia                                                              | H: ASA, Citrullin    | Yes | Yes | Yes     | Yes     | No     | No     | No      |          |           |                |
| CIT                 | Citrullinemia type I (Argininosuccinate synthetase deficiency)                          | H: Citrullin         | Yes | Yes | No      | Yes     | No     | No     | No      |          |           |                |
| HCY                 | Homocysteinuria (cystathione beta synthase deficiency)                                  | H: Methionin         | Yes | Yes | No      | Yes     | Yes    | Yes    | No      | Yes      | 1 case     |                |
| MSUD                | Maple syrup urine disease (branched-chain ketoacid dehydrogenase deficiency)           | H: XLeucine (Leucine+Isoleucine+hydroxyprline), valine. | Yes | Yes | Yes     | Yes     | Yes    | Yes    | Yes     | Yes      |           |                |
| PKU                 | Phenylketonuria (hyperphenylalanemia)                                                   | H: Phenylalanine     | Yes | Yes | Yes     | Yes     | Yes    | Yes    | Yes     | Yes      |           |                |
| TYR-1               | Tyrosinemia Type 1 (Fumarylacetate acetate hydrolasi)                                   | SUAC (Succinyl Acetone), Tyr* | Yes | Yes | (Yes)*  | Yes     | Yes    | Yes    | No      | Yes      |           |                |
| NKHG                | Non ketotic hyperglycinemia                                                            | Glycine              | Some | (Yes) | No      | No      | No     | No     | No      | No       | No         |                |
| PC                  | Pyruvate carboxylase deficiency                                                        | Citrullin            | (Yes) | (Yes) | No      | (No)    | No     | No     | No      | No       | No         |                |
Secondary Diseases (22)

Core Metabolic Diseases vs Secondary Diseases

One acylcarnitine can be a marker for several diseases

- Propionyl Carnitine (C3) - 10 different diseases
- 3 Hydroxyisovaleric Carnitine (C5OH) - 7 different diseases
- Phenylalanine (Phe) - 6 different diseases.

Differential diagnosis necessary

Sometimes variable what secondary diseases different NBS programs report even after differential diagnosis.
Question!

• How is it possible to screen for rare diseases one might NEVER find in a population with a birth rate of 4800 per year?
- Solid Backup -

- Dr Piero Rinaldo Mayo Clinic Roch. MN
- Dr. Zoltan Lukacs Hamburg University Medical Center Hamburg
- Excellent cooperation between NBS Laboratories
- Sample exchange, 2nd tier testing (Mayo)
- External Quality control schemes (CDC)
- For discussing Inborn Errors of Metabolism metab-l-bounces@lists.franken.de
- The STORK - project
Aims of STORK
1. To coordinate Newborn Screening Methods for IEM and confirmatory measures.
2. Prevent false positives
3. 150 International NBS laboratories participate by sharing data.
   - Normal distribution (percentiles) of normal newborns
   - Results from confirmed IEM are reported and stored in a databank for calculation of Disease Ranges
   - Results from “dubious” NBS sample are inserted in a table via the STORK homepage and an instant report is given about the possibility of a disease.
4. Have training sessions with laboratories (at Mayo Clinic Rochester)
5. Good contact with participants with monthly updates, case of the month etc.
MS/MS COLLABORATIVE PROJECT

Number of Cases: 13,291

May 2012
ACT SHEETS

All materials were approved by the Board of Directors of the American College of Medical Genetics (ACMG) on March 22, 2006. The materials will be maintained by ACMG over time and additional materials added as new conditions are introduced into newborn screening programs. The project was partially funded through grant U22MC03957 from the Maternal and Child Health Bureau (MCHB) of the Health Resources and Services Administration (HRSA) of the U.S. Department of Health and Human Services.

http://www.acmg.net/resources/policies/ACT/condition-analyte-links.htm
Newborn Screening ACT Sheet  
[Decreased C0 and other acylcarnitines]  
Carnitine Uptake Defect (CUD)

**Differential Diagnosis:** Carnitine uptake defect (CUD).

**Condition Description:** CUD is caused by a defect in the carnitine transporter that moves carnitine across the plasma membrane. Reduced carnitine limits acylcarnitine formation preventing transport of fatty acids into mitochondria, thereby limiting energy production. Tissues with high energy needs (skeletal and heart muscle) are particularly affected.

**MEDICAL EMERGENCY - TAKE THE FOLLOWING IMMEDIATE ACTIONS:**
- Contact family to inform them of the newborn screening result and ascertain clinical status (poor feeding, lethargy, tachypnea).
- Consult with pediatric metabolic specialist.
- Evaluate the newborn (tachycardia, hepatomegaly, reduced muscle tone); initiate emergency treatment as indicated by metabolic specialist.
- Initiate timely confirmatory/diagnostic testing as recommended by specialist.
- Educate family about signs, symptoms and need for urgent treatment if infant becomes ill.
- Report findings to newborn screening program.

**Diagnostic Evaluation:** Plasma and urine carnitine analysis will reveal decreased free and total carnitine (C0) in plasma and overexcretion of carnitine in urine. The newborns mother should be investigated as well because several cases of maternal CUD have been identified following an abnormal newborn screening result in their offspring. Transporter assays and OCTN2 gene sequencing establish the diagnosis.

**Clinical Considerations:** Carnitine transporter defect has a variable expression and variable age of onset. Characteristic manifestations include lethargy, hypotonia, hepatomegaly, and cardiac decompensation due to cardiomyopathy. Hypoglycemia is typical in acute episodes.

**Additional Information:**  
(Click on the name to take you to the website. Complete URLs are listed in the Appendix)

- **OMIM**
- Genetics Home Reference
- **STAR-G/HRSA**

**Referral (local, state, regional and national):**
- Testing
- Clinical

Disclaimer: These standards and guidelines are designed primarily as an educational resource for physicians to help them provide quality medical services. Adherence to these standards and guidelines does not necessarily ensure a successful medical outcome. These standards and guidelines should not be considered inclusive of all proper procedures and tests or exclusive of other procedures and tests that are reasonably directed to obtaining the same result. In determining the proper use of any specific procedure or test, the healthcare provider should apply his or her own professional judgment to the specific clinical circumstances presented by the individual patient or specimen. It may be prudent, however, to document in the patient's record the rationale for any significant deviation from these standards and guidelines.

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(Funded in part through MCHB-RESA/HRSA grant U22MC03957)
CO (Free Carnitine) Low

Decreased Free Carnitine (CO); Other ACs relatively low.

+ Routine Labs
  Glucose, electrolytes, blood gas, ammonia, LFT, CPK

Assay free and total carnitine in plasma and urine

Plasma CO – Low
  Plasma Total Carnitine – Normal/Low
  Urine CO – High/Normal
  Urine Total Carnitine – High/Normal

  Carritine Uptake Defect (CUD)

Plasma CO – Normal
  Plasma Total Carnitine – Normal
  Urine CO – Normal
  Urine Total Carnitine - Normal

  Infant healthy. Rule out maternal CUD

OPTIONAL CONFIRMATORY TESTS.
  Transporter Assay, OCTN2 gene analysis
If you don't believe in miracles perhaps you've forgotten you are one.