May/June 2011 Newborn Screening Articles of Interest

1. Acta Paediatr. 2011 Aug;100(8):1097-1103.

Ten-year quality assurance of the nationwide hearing screening programme in Dutch neonatal intensive care units.

van Dommelen P, van Straaten H, Verkerk P; the Dutch NICU **Neonatal** Hearing **Screening** Working Group.

Abstract

Aim: To evaluate 10-year quality assurance of **newborn** hearing **screening** (NHS) in Dutch **neonatal** intensive care units (NICU). Methods: Results of the two-stage automated auditory brainstem response (AABR) **screening** and diagnostic examination in NICU graduates were centrally registered between October 1998 and December 2008. This registration facilitates **screening**, tracking and follow-up after abnormal **screening** results. Outcome measures are referral rates, prevalence rate of hearing loss and (trends of) coverage rates and timeliness of follow-up. Results: Thirty-two thousand one hundred and two infants have been screened. Referral rates were 9.2% at the first and 26.3% at the second stage. Hearing loss was diagnosed in 728 infants (2.2%). Coverage rates were 98.7% at the first, 92.1% at the second stage, 92.3% for the diagnostic examination and 97.9% for the complete programme. After correction for gestational age, 95.8% of the infants had their first AABR <1 month, 81.8% of the referred infants had their second AABR <6 weeks and 67.1% were diagnosed <3 months. There was a positive trend in referred infants that had their second AABR <6 weeks ($p = 0.004$) as well as in infants diagnosed <3 months ($p < 0.001$). Conclusion: The NHS in Dutch NICUs is effective. Timely identification of hearing loss is improving over time.

2. Clin Pediatr (Phila). 2011 Aug;50(8):753-6. Epub 2011 May 3.

No more tears? Maternal involvement during the newborn screening examination.

Ganda AJ, Fara Ibrahim L, Natchimutu K, Ryan CA.

Abstract

Background. Babies often show signs of discomfort and distress by crying during the **neonatal screening** examination (NSE). The authors hypothesized that supporting the baby with maternal participation may reduce

infant crying during NSE. The objective of this study was to document incidental infant crying during NSE, before and after training residents, on maternal involvement and infant comfort techniques to help. Methods. A total of 20 NSEs of normal **newborn** babies by pediatric residents were observed (video-recorded) following informed consent of the doctor and the baby's mother. The examining doctors were then taught how to use maternal participation and developmental care (MPDC) comfort techniques to support the baby during NSE. Mothers were shown how to focus on their baby's needs by supporting the baby's head (preventing atonic neck reflexes) and, if necessary, providing nonnutritive sucking to the baby and an encouraging, repetitive low-tone voices to sooth the baby. A further 14 NSEs on different babies were video-recorded using these techniques. The video recordings were analyzed by independent observers for total length of crying and duration of crying during specific components of the NSE. Mothers in both groups were given a questionnaire to assess their opinions of the NSE. Results. The median length of crying was significantly longer in the pre-MPDC group (93.5 seconds; range 0-198 seconds) compared with the post-MPDC infants (0 seconds; range 0-123 seconds; P = .001). Only 1 of 20 infants in the pre-MPDC did not cry during NSE compared with 8 of 14 babies in the post-MPDC group. Conclusion. **Newborn** infants cry less and mothers were more satisfied with NSEs when shown simple support and comfort techniques for their babies.

3. Clin Chim Acta. 2011 Jul 15;412(15-16):1476-9. Epub 2011 May 4.

3-years experience review of neonatal screening for hemoglobin disorders using tandem mass spectrometry.

Boemer F, Cornet Y, Libioulle C, Segers K, Bours V, Schoos R.

Abstract

BACKGROUND:

Neonatal screening programs for sickle cell disease are common in North America and in some European countries. Isoelectric Focusing or High Performance Liquid Chromatography is the main technique used for hemoglobin variant detection.

METHODS:

Since tandem mass spectrometry is being used for **screening** of inherited metabolic disorders and allows protein identification, we had developed an application to identify the most relevant hemoglobin mutations with this technology.

RESULTS:

This approach had been previously validated and has been routinely applied in our laboratory for the last three years. We report here our experience with this new method in the field, applied to our East-Belgian population.

CONCLUSIONS:

To conclude, mass spectrometry provides an efficient alternative approach for laboratories performing **neonatal screening** of hemoglobin disorders.

4. Clin Chim Acta. 2011 Jul 15;412(15-16):1385-90. Epub 2011 Apr 14.

Detection of TPN contamination of dried blood spots used in newborn and metabolic screening and its impact on quantitative measurement of amino acids.

Chace DH, De Jesús VR, Lim TH, Hannon WH, Clark RH, Spitzer AR.

Abstract

BACKGROUND:

Markers derived from dextrose (d-glucose) are observed in the MS/MS-based acylcarnitine profiles from dried-blood spots of some premature infants receiving intravenous nutrition. The presence of these markers at m/z 325, 399 and 473 are thought to arise from contamination of blood by total parenteral nutrition (TPN) solutions during specimen collection from premature infants. These solutions contain high concentrations of amino acids and as a result, false-positive **screening** results for amino acid disorders may occur. This study investigates quantitative parameters of dextrose and amino acids in blood samples enriched with different TPN solutions.

METHODS:

Whole blood collected in heparin was enriched with three different TPN solutions containing 5, 10 or 12.5% dextrose and amino acids that were originally prepared for delivery of 2.5, 3 or 4g/kg/day of Premasol® then spotted onto filter paper cards. Acylcarnitine and amino acid profiles using MS/MS were obtained. Ion ratios of dextrose relative to specific acylcarnitine stable isotope internal standards and amino acid concentrations were obtained.

RESULTS:

The ion ratios for each of the dextrose markers at m/z 325, 399 and 473 exhibit linearity with the concentration of the dextrose component of TPN added to blood. The lowest detectable dextrose concentration added to blood was 7.6mmol/l at 1:80v/v TPN in blood. Furthermore, the concentrations of amino acids were linear with the concentration of the amino acid component of TPN added to blood. At the lowest detectable concentrations of dextrose marker, the amino acid concentrations were at or above the values considered abnormal in **newborn screening** laboratories. The molar ratios of amino acids approached the relative quantity of amino acid in the TPN solution with increasing enrichments in blood.

CONCLUSIONS:

Detection of the combinations of dextrose markers, very high elevations of amino acids and unusual molar ratios can be used to reject a specimen as improperly collected rather than declaring it a false positive and hence reduce false positive rates. This process enhances efficiency, reduces parental anxiety, and improves positive predictive values.

5. Clin Chim Acta. 2011 Jul 15;412(15-16):1376-81. Epub 2011 Apr 14.

Implementation of the first worldwide quality assurance program for cystic fibrosis multiple mutation detection in population-based screening.

Earley MC, Laxova A, Farrell PM, Driscoll-Dunn R, Cordovado S, Mogayzel PJ Jr, Konstan MW, Hannon WH.

Abstract

BACKGROUND:

CDC's **Newborn Screening** Quality Assurance Program collaborated with several U.S. Cystic Fibrosis Care Centers to collect specimens for development of a molecular CFTR proficiency testing program using dried-blood spots for **newborn screening** laboratories.

METHODS:

Adult and adolescent patients or carriers donated whole blood that was aliquoted onto filter paper cards. Five blind-coded specimens were sent to participating **newborn screening** laboratories quarterly. Proficiency testing results were evaluated based on presumptive clinical assessment. Individual evaluations and summary reports were sent to each participating laboratory and technical consultations were offered if incorrect assessments were reported.

RESULTS:

The current CDC repository contains specimens with 39 different CFTR mutations. Up to 45 laboratories have participated in the program. Three years of data showed that correct assessments were reported 97.7% of the time overall when both mutations could be determined. Incorrect assessments that could have lead to a missed case occurred 0.9% of the time, and no information was reported 1.1% of the time due to sample failure.

CONCLUSIONS:

Results show that laboratories using molecular assays to detect CFTR mutations are performing satisfactorily. The programmatic results presented demonstrate the importance and complexity of providing proficiency testing for DNA-based assays.

6. BMC Genet. 2011 Jul 4;12(1):58. [Epub ahead of print]

Robustness of genome-wide scanning using archived dried blood spot samples as a DNA source.

Hollegaard MV, Grove J, Grauholm J, Kreiner-Moller E, Bonnelykke K, Norgaard M, Benfield TL, Norgaard-Pedersen B, Mortensen PB, Mors O, Sorensen HT, Harboe ZB, Borglum AD, Demontis D, Orntoft TF, Bisgaard H, Hougaard DM.

Abstract

ABSTRACT:

BACKGROUND:

The search to identify disease-susceptible genes requires access to biological material from numerous well-characterized subjects. Archived residual dried blood spot (DBS) samples, also known as Guthrie cards, from national **newborn screening** programs may provide a DNA source for entire populations. Combined with clinical information from medical registries, DBS samples could provide a rich source for productive research. However, the amounts of DNA which can be extracted from these precious samples are minute and may be prohibitive for numerous genotypings. Previously, we demonstrated that DBS DNA can be whole-genome amplified and used for reliable genetic analysis on different platforms, including genome-wide scanning arrays. However, it remains unclear whether this approach is workable on a large sample scale. We examined the robustness of using DBS samples for whole-genome amplification following genome-wide scanning, using arrays from Illumina and Affymetrix.

RESULTS:

This study is based on 4,641 DBS samples from the Danish **Newborn Screening** Biobank, extracted for three separate genome-wide association studies. The amount of amplified DNA was significantly ($P<0.05$) affected by the year of storage and storage conditions. Nine (0.2%) DBS samples failed whole-genome amplification. A total of 4,586 (98.8%) samples met our criterion of success of a genetic call-rate above 97%. The three studies used different arrays, with mean genotyping call-rates of 99.385% (Illumina Infinium Human610-Quad), 99.722% (Illumina Infinium HD HumanOmni1-Quad), and 99.206% (Affymetrix Axiom Genome-Wide CEU). We observed a concordance rate of 99.997% in the 38 methodological replications, and 99.999% in the 27 technical replications. Handling variables such as time of storage, storage conditions and type of filter paper were shown too significantly ($P<0.05$) affect the genotype call-rates in some of the arrays, although the effect was minimal.

CONCLUSIONS:

Our study indicates that archived DBS samples from the Danish **Newborn Screening** Biobank represent a reliable resource of DNA for whole-genome amplification and subsequent genome-wide association studies. With call-rates

equivalent to high quality DNA samples, our results point to new opportunities for using the **neonatal** biobanks available worldwide in the hunt for genetic components of disease.

7. Pediatrics. 2011 Jul 4. [Epub ahead of print]

The Need for Vigilance: The Case of a False-Negative Newborn Screen for Cystic Fibrosis.

Dunn CT, Skrypek MM, Powers AL, Laguna TA.

Abstract

Cystic fibrosis (CF) is the most common life-limiting recessive genetic disorder in the white population. CF is caused by abnormalities in the gene that codes for the cystic fibrosis transmembrane conductance regulator protein (CFTR) and may result in severe chronic lung disease, poor growth, and malnutrition. Physicians often do not consider CF in the differential diagnosis of an infant with failure to thrive in the presence of a negative **newborn screening** (NBS) result. In Minnesota, **newborn** infants are screened for CF by immunoreactive trypsinogen (IRT) testing followed by DNA analysis if the IRT screen result is abnormal. All positive NBS results are followed by confirmatory sweat-testing by pilocarpine iontophoresis. We present here the case of a 1-month-old white boy with failure to thrive, chronic diarrhea, and severe malnutrition. Minnesota state CF NBS results were negative at birth (IRT: 43 ng/mL [96% cutoff value: 52 ng/mL]). Clinical symptoms resulted in sweat-testing by Gibson-Cooke pilocarpine iontophoresis at 1 month of age, and the result was positive (102 mmol Cl(-)/L [normal: ≤30 mmol Cl(-)/L]). CFTR mutation analysis confirmed a homozygous f508del genotype, and stool pancreatic elastase testing revealed severe exocrine pancreatic insufficiency. This case represents the first known false-negative result in Minnesota since the initiation of NBS for CF in 2006, which illustrates the importance of considering CF in the evaluation of an infant with failure to thrive and symptoms of malabsorption, regardless of NBS results.

8. Arch Dis Child Fetal **Neonatal** Ed. 2011 Jul;96(4):F312. Epub 2011 Feb 19.

Neonatal pulse oximetry screening: a national survey.

Kang SL, Tobin S, Kelsall W.

9. Arch Pediatr Adolesc Med. 2011 Jul;165(7):589-91.

Hidden in the sixties: newborn screening programs and state authority.

Brosco JP.

10. Chest. 2011 Jul;140(1):170-7. Epub 2010 Nov 24.

Relationships among health-related quality of life, pulmonary health, and newborn screening for cystic fibrosis.

Tluczek A, Becker T, Laxova A, Grieve A, Racine Gilles CN, Rock MJ, Gershan WM, Green CG, Farrell PM.

Abstract

BACKGROUND:

The objective of this study was to examine relationships between pulmonary health and health-related quality of life (HRQOL) in patients with cystic fibrosis (CF) evaluated longitudinally in the Wisconsin **Newborn Screening** Project.

METHODS:

Patients aged 8 to 18 years (mean ± SD, 13.5 ± 2.8) in early diagnosis (n = 45) and control (n = 50) groups completed Cystic Fibrosis Questionnaires (CFQs) to measure HRQOL at three data points over a 2-year period. Pulmonary health was evaluated concurrently by the Wisconsin chest x-ray scoring system (WCXR) and pulmonary function tests (PFTs).

RESULTS:

WCXR showed significant group differences ($P \leq .023$), with the early diagnosis group showing more-severe lung disease. When adjusted for group differences in mucoid Pseudomonas aeruginosa status and pancreatic status, however, WCXR differences and PFT data were not significant. Most patients (74%) had FEV(1) values ≥ 80% predicted (within normal range). For patients aged < 14 years, as WCXR scores worsened CFQ respiratory and physical domain scores decreased (both $P \leq .007$). FEV(1)/FVC showed a positive relationship with the respiratory and physical domains (both $P \leq .006$). WCXR scores for patients aged ≥ 14 years were associated with CFQ weight,

respiratory, and health domains (all P ≤ .011). FEV(1) was associated with CFQ weight, respiratory, health, and physical domains (all P ≤ .003). Changes in pulmonary health were not associated with changes in CFQ over time. Significant group differences on the CFQ-Child social functioning domain favored the control group.

CONCLUSIONS:

To our knowledge, this study is the first to compare pulmonary outcomes with HRQOL indicators assessed by serial, standardized, patient-reported outcome measures for patients with CF identified either through **newborn screening** or diagnosed by use of traditional methods. This study found no benefits of **newborn screening** for pulmonary health or HRQOL after controlling for risk factors. Using WCXR and PFT data collectively helped to identify associations between pulmonary health and HRQOL.

11. Curr Pharm Biotechnol. 2011 Jul 1;12(7):965-75.

Altered Metabolism and Newborn Screening using Tandem Mass Spectrometry: Lessons Learned from the Bench to Bedside.

Chace DH, Spitzer AR.

Abstract

The use of tandem mass spectrometry (MS/MS) for **screening** of inherited metabolic disease in newborns has afforded many unique opportunities in the understanding of the benefits early their early detection, diagnosis and treatment. From the standpoint of the laboratory and modern analytical methods, the use of MS based analysis demonstrated that a multiple metabolite-multiple disease screen-one method approach expanded **screening** significantly. MS/MS and **newborn screening** has served as a model of one type of approach in preventative health care that has shown proven benefits. It has been nearly 20 years since the introduction of MS/MS analysis of dried blood spots from newborns. There have been many lessons learned in both the analytical approach as well as follow-up at the bedside. These lessons can be applied to future applications of MS/MS in **newborn screening** as well as other areas of metabolism and metabolic profiles such as that from acquired disease, environmental disease and other factors such as nutrition and age. The use of a highly specific, sensitive and multiplex platform such as MS/MS will continue to grow and experience in the **newborn screening** application will insure this outcome.

12. Int J Pediatr Otorhinolaryngol. 2011 Jul;75(7):973-5. Epub 2011 May 19.

Auditory neuropathy/dyssynchrony as a cause of failed neonatal hearing screening.

Maris M, Venstermans C, Boudewyns AN.

Abstract

The prevalence of auditory neuropathy/dyssynchrony (AN/AD) is not exactly known. We retrospectively analysed the prevalence of this condition among 135 infants who failed a **neonatal screening**. Hearing **screening** was performed by automated auditory brainstem responses (AABR). Unilateral presence of click-evoked oto-acoustic emissions with absent auditory brainstem responses was found in 4 infants. Magnetic resonance imaging of the posterior fossa showed an aplasia/hypoplasia of the ipsilateral cochlear nerve in these 4 cases. The prevalence of AN/AD was 19% in infants with confirmed hearing loss. Our findings underscore the role of AABR in **neonatal** hearing **screening**.

13. J Cyst Fibros. 2011 Jul;10(4):278-81. Epub 2011 Mar 8.

Optimal DNA tier for the IRT/DNA algorithm determined by CFTR mutation results over 14years of newborn screening.

Baker MW, Groose M, Hoffman G, Rock M, Levy H, Farrell PM.

Abstract

BACKGROUND:

There has been great variation and uncertainty about how many and what CFTR mutations to include in cystic fibrosis (CF) **newborn screening** algorithms, and very little research on this topic using large populations of newborns.

METHODS:

We reviewed Wisconsin **screening** results for 1994-2008 to identify an ideal panel.

RESULTS:

Upon analyzing approximately 1 million **screening** results, we found it optimal to use a 23 CFTR mutation panel as a second tier when an immunoreactive trypsinogen (IRT)/DNA algorithm was applied for CF **screening**. This panel in association with a 96th percentile IRT cutoff gave a sensitivity of 97.3%, but restricting the DNA tier to F508del was associated with 90% (P<.0001).

CONCLUSIONS:

Although CFTR panel selection has been challenging, our data show that a 23 mutation method optimizes sensitivity and is advantageous. The IRT cutoff value, however, is actually more critical than DNA in determining CF **newborn screening** sensitivity.

14. J Pediatr. 2011 Jul;159(1):7-13.e1. Epub 2011 Apr 13.

Newborn bloodspot screening for lysosomal storage disorders.

Zhou H, Fernhoff P, Vogt RF.

15. J Perinatol. 2011 Jul;31(7):507-10. doi: 10.1038/jp.2010.207.

Extremely high phenylalanine levels in a newborn on parenteral nutrition: phenylketonuria in the neonatal intensive care unit.

Lin HJ, Kwong AM, Carter JM, Ferreira BF, Austin MF, Devarajan K, Coleman RJ, Feuchtbaum LB, Lorey F, Jonas AJ.

Abstract

A 1890-g **newborn** on total parenteral nutrition (TPN) had phenylalanine levels reaching 4164 μM indicating phenylketonuria (PKU). Review of 64 PKU cases from the California **Newborn Screening** Program disclosed another **newborn** diagnosed while on TPN. Phenylalanine levels rose five times faster with TPN, as estimated from rates in these infants. Thus, TPN use is associated with very high phenylalanine levels in newborns with PKU. When

starting TPN soon after birth (for example, on day 1), early detection of PKU-by **newborn screening** 12 to 24 h after infusions are begun-should be helpful in limiting exposures to toxic levels of phenylalanine.

16. Pediatr Pulmonol. 2011 Jul;46(7):696-700. doi: 10.1002/ppul.21434. Epub 2011 Mar 1.

Bronchoscopy in cystic fibrosis infants diagnosed by newborn screening.

Stafler P, Davies JC, Balfour-Lynn IM, Rosenthal M, Bush A.

Abstract

BACKGROUND:

There is evidence of early functional and structural changes in babies with cystic fibrosis (CF) diagnosed on **newborn screening** (NBS). The aim of the present study was to determine the yield of bronchoalveolar lavage (BAL) microbiology and cytology, and 24 hr pH monitoring in a group of CF infants diagnosed on NBS.

METHODS:

Infants referred to a tertiary pediatric respiratory center between July 2007 and November 2009 underwent surveillance fiber-optic bronchoscopy (FOB), BAL, and insertion of a 24 hr dual pH probe under a single general anesthetic.

RESULTS:

We studied 33 infants, median age of 100 days (47-215 days) at the time of FOB. In 9 of 33 (27%) bacterial organisms were identified. Seven of the nine patients (78%) were asymptomatic and only one had had a positive cough swab prior to FOB. Neutrophilia was identified in 18/27 (67%) cases with a median of 11% (6-73%). 13/31 (42%) had an abnormal pH study with a pH index >12%.

CONCLUSIONS:

The high yield of microbiology, cytology, and pH probe investigations in NBS infants justifies invasive surveillance. Longitudinal studies to determine if early aggressive treatment results in improved outcome are awaited.

17. Pediatrics. 2011 Jul;128(1):53-61. Epub 2011 Jun 27.

Parents' experiences of expanded newborn screening evaluations.

Deluca JM, Kearney MH, Norton SA, Arnold GL.

Abstract

OBJECTIVE:

Abnormal results of **newborn screening** for common metabolic diseases are known to create substantial distress for parents. We explored parents' perceptions during diagnostic evaluations for newer disorders that are less well understood.

METHODS:

Thirty families completed 48 open-ended interviews before and/or after parents received confirmatory test results for their infants. Qualitative content analysis was used to analyze the data.

RESULTS:

Parents were shocked by the notification of the abnormal test result. Their urgent and often frustrating searches for information dominated the early phase of the **screening** process. Treatment center personnel were mainly informative and reassuring, but waiting for results exacerbated parents' distress. Equivocal results from diagnostic testing created uncertainties for parents regarding their infants' long-term health. After counseling, some parents reported inaccurate ideas about the disorders despite exposure to large amounts of information. Regardless of the challenges and anxieties of the evaluation, nearly every parent thought **newborn screening** was an important program for infant health.

CONCLUSIONS:

The evaluation of a **newborn** for an abnormal **screening** result was highly stressful for parents. To help reduce parents' distress, improvements in communications and clinical services are needed. Recommendations of useful Internet sites and discussions of this information may benefit parents. Tailoring counseling to meet the needs of culturally and educationally diverse families is needed. Families and infants with equivocal results are a new group of patients who merit comprehensive clinical follow-up.

18. Pediatrics. 2011 Jul;128(1):e246-50. Epub 2011 Jun 13.

Siblings With Mitochondrial Acetoacetyl-CoA Thiolase Deficiency Not Identified by Newborn Screening.

Sarafoglou K, Matern D, Redlinger-Grosse K, Bentler K, Gaviglio A, Harding CO, Rinaldo P.

Abstract

Screened for by all state **newborn screening** (NBS) programs in the United States, mitochondrial acetoacetyl-coenzyme A thiolase (T2), or β-ketothiolase, deficiency is a rare autosomal recessive disorder that causes ketoacidosis and hypoglycemia/hyperglycemia. Outcomes vary from normal development to severe cognitive impairment or even death after an acute episode of ketoacidosis. The classical biochemical profile of T2 deficiency is a result of null mutations in both alleles of the ACAT1 gene and consists of persistently increased urinary excretion of ketones, characteristic organic acids, and tiglylglycine as well as abnormal blood or plasma acylcarnitine profiles in acute and stable conditions. Early diagnosis and aggressive management can prevent further episodes of ketoacidosis and lead to normal development. We report the cases of 3 children, all subsequently found to have mutations predicted to be associated with no residual T2 enzymatic activity, but only 1 was identified by NBS in Minnesota since 2001. To our knowledge, this is the first description of compound heterozygotes for null mutations associated with no enzymatic activity exhibiting normal urinary organic acid, blood, and plasma acylcarnitine profiles when clinically well, thereby explaining the false-negative NBS results. We suggest that T2 deficiency may be underrecognized, because the incidence of T2 deficiency in Minnesota, on the basis of these 3 cases, is 1 in 232 000, higher than the reported <1 in 1 million incidence. Our cases emphasize that T2 deficiency must be considered in patients who present with ketoacidosis disproportionately severe to the triggering illness despite normal NBS results or nonspecific biochemical findings in blood and urine during asymptomatic periods.

19. Genet Med. 2011 Jun 28. [Epub ahead of print]

What questions should newborn screening long-term follow-up be able to answer? A statement of the US Secretary for Health and Human Services' Advisory Committee on Heritable Disorders in Newborns and Children.

Hinton CF, Feuchtbaum L, Kus CA, Kemper AR, Berry SA, Levy-Fisch J, Luedtke J, Kaye C, Boyle CA.

Abstract

The US Secretary of Health and Human Services' Advisory Committee on Heritable Disorders in Newborns and Children provides guidance on reducing the morbidity and mortality associated with heritable disorders detectable through **newborn screening**. Efforts to systematically evaluate health outcomes, beyond long-term survival, with a few exceptions, are just beginning. To facilitate these nascent efforts, the US Secretary of Health and Human Services' Advisory Committee on Heritable Disorders in Newborns and Children initiated a project to define the major overarching questions to be answered to assure that **newborn screening** is meeting its goal of achieving the best quality outcome for the affected children and their families. The questions identified follow the central components of long-term follow-up-care coordination, evidence-based treatment, continuous quality improvement, and new knowledge discovery-and are framed from the perspectives of the state and nation, primary and specialty healthcare providers, and the impacted families. These overarching questions should be used to guide the development of long-term follow-up data systems, quality health indicators, and specific data elements for evaluating the **newborn screening** system.

20. Eur Arch Otorhinolaryngol. 2011 Jun 23. [Epub ahead of print]

Universal newborn hearing screening, a revolutionary diagnosis of deafness: real benefits and limitations.

Papacharalampous GX, Nikolopoulos TP, Davilis DI, Xenellis IE, Korres SG.

Abstract

The finding that early detection of permanent congenital childhood hearing loss produces worthwhile benefit in terms of improved speech and language provides the rationale for the universal **screening** of newborns. The aim of the present study is to collect the current evidence with regard to the efficacy, the results and outcomes of universal hearing **screening** programs. An extensive search of the literature was performed in Medline and other available database sources. Study selection was based on the evaluation of the protocols used and the assessment of their

efficacy in the early diagnosis of congenital hearing impairment. The initial referral rate and the rate of false positives were also evaluated. A total of 676,043 screened children have been identified in 20 studies. The average initial referral rate in these studies was 3.89%. The initial referral rate varied from 0.6 to 16.7%. The lost-to-follow-up rates varied from 3.7 to 65%. Although universal hearing **screening** is now widely adopted, there are still some serious drawbacks and limitations. False positives rates remain considerably high when newborns are screened with TEOAE's. The combination of TEOAE's and a-ABR provides a significantly reduced referral rate. Close cooperation between audiological centres and maternity units and a dedicated secretariat team are of paramount importance with regard to the reliability and efficacy of universal hearing **screening**.

21. Orphanet J Rare Dis. 2011 Jun 20;6(1):44. [Epub ahead of print]

Efficacy and Outcome of Expanded Newborn Screening for Metabolic Diseases - Report of 10 years from South-West Germany.

Lindner M, Gramer G, Haege G, Fang-Hoffmann J, Schwab KO, Tacke U, Trefz FK, Mengel E, Wendel U, Leichsenring M, Burgard P, Hoffmann GF.

ABSTRACT:

BACKGROUND:

National **newborn screening** programmes based on tandem-mass spectrometry (MS/MS) and other **newborn screening** (NBS) technologies show a substantial variation in number and types of disorders included in the **screening** panel. Once established, these methods offer the opportunity to extend **newborn screening** panels without significant investment and cost. However, systematic evaluations of **newborn screening** programmes are rare, most often only describing parts of the whole process from taking blood samples to long-term evaluation of outcome.

METHOD:

S In a prospective single **screening** centre observational study 373 cases with confirmed diagnosis of a metabolic disorder from a total cohort of 1,084,195 neonates screened in one **newborn screening** laboratory between January 1, 1999, and June 30, 2009 and subsequently treated and monitored in five specialised centres for inborn errors of

metabolism were examined. Process times for taking **screening** samples, obtaining results, initiating diagnostic confirmation and starting treatment as well as the outcome variables metabolic decompensations, clinical status, and intellectual development at a mean age of 3.3 years were evaluated.

RESULTS:

Optimal outcome is achieved especially for the large subgroup of patients with medium-chain acyl-CoA dehydrogenase deficiency. Kaplan-Meier-analysis revealed disorder related patterns of decompensation. Urea cycle disorders, organic acid disorders, and amino acid disorders show an early high and continuous risk, medium-chain acyl-CoA dehydrogenase deficiency a continuous but much lower risk for decompensation, other fatty acid oxidation disorders an intermediate risk increasing towards the end of the first year. Clinical symptoms seem inevitable in a small subgroup of patients with very early disease onset. Later decompensation can not be completely prevented despite pre-symptomatic start of treatment. Metabolic decompensation does not necessarily result in impairment of intellectual development, but there is a definite association between the two.

CONCLUSIONS:

Physical and cognitive outcome in patients with presymptomatic diagnosis of metabolic disorders included in the current German **screening** panel is equally good as in phenylketonuria, used as a gold standard for NBS. Extended NBS entails many different interrelated variables which need to be carefully evaluated and optimized. More reports from different parts of the world are needed to allow a comprehensive assessment of the likely benefits, harms and costs in different populations.

22. Anal Chem. 2011 Jun 15;83(12):4822-4828. Epub 2011 May 17.

Comparative Triplex Tandem Mass Spectrometry Assays of Lysosomal Enzyme Activities in Dried Blood Spots Using Fast Liquid Chromatography: Application to Newborn Screening of Pompe, Fabry, and Hurler Diseases.

Spáčil Z, Elliott S, Reeber SL, Gelb MH, Scott CR, Tureček F.

Abstract

We report a comparative study of triplex tandem mass spectrometry (MS/MS) based assays of lysosomal enzymes in dried blood spots for the early detection of Pompe, Fabry, and Hurler diseases in newborns. Four methods have been evaluated that differed in sample handling and the equipment used. A newly developed method uses assay quenching with acetonitrile to precipitate blood proteins followed by analysis on an LC-electrospray/MS/MS system capable of multiple consecutive sample injections on two parallel chromatographic columns. This method requires 1.5 min per a triplex analysis of enzyme products and internal standards, which matches the throughput of the previously reported flow injection method. LC separation reduces matrix effects and allows for more facile sample workup. The new LC-based method showed figures of merit that were superior to those of the currently used method based on liquid-liquid extraction into ethyl acetate and flow injection into the mass spectrometer. The other methods we investigated for comprehensive comparison involved liquid-liquid extraction into ethyl acetate followed by LC-ESI-MS/MS and acetonitrile quenching followed by direct flow injection. Both methods using acetonitrile quenching were found to be robust and provide good quality data while requiring fewer liquid transfer steps and less disposable material and labor than did the extraction methods. The individual merits of the new methods are discussed to present an evaluated alternative approach to high-throughput analysis in **newborn screening** laboratories.

23. Genet Med. 2011 Jun 15. [Epub ahead of print]

Optimizing Bob Guthrie's legacy-Storage and use of residual newborn screening specimens.

Webster D.

24. Genet Med. 2011 Jun 13. [Epub ahead of print]

Emergency preparedness for genetics centers, laboratories, and patients: The Southeast Region Genetics Collaborative strategic plan.

Andersson HC, Perry W, Bowdish B, Floyd-Browning P.

Abstract

Emergencies occur unpredictably and interrupt routine genetic care. The events after hurricanes Katrina and Rita have led to the recognition that a coherent plan is necessary to ensure continuity of operations for genetic centers

and laboratories, including **newborn screening**. No geographic region is protected from the effects of a variety of potential emergencies. Regional and national efforts have begun to address the need for such preparedness, but a plan for ensuring continuity of operations by creating an emergency preparedness plan must be developed for each genetic center and laboratory, with attention to the interests of patients. This article describes the first steps in development of an emergency preparedness plan for individual centers.

25. Genet Med. 2011 Jun 13. [Epub ahead of print]

Storage and use of residual newborn screening blood spots: A public policy emergency.

Tarini BA.

26. Indian J Pediatr. 2011 Jun 10. [Epub ahead of print]

Impact of Inborn Errors of Metabolism on Admission in a Neonatal Intensive Care Unit-A Prospective Cohort Study.

Tu W, He J, Dai F, Wang X, Li Y.

Abstract

OBJECTIVE:

To estimate the incidence of Inborn errors of metobolism (IEM) in **Neonatal** intensive care unit (NICU) using tandem mass spectrometry and to determine the impact that these disorders have on NICU resources.

METHODS:

During the period of study, 724 (81% eligible cases) dried blood filter-paper samples were collected from a NICU. The samples were analysed using tandem mass spectrometry. The diagnosis was further confirmed through clinical symptoms and by gas chromatography-mass spectrometry. The results were also confirmed by clinical follow-up of all positive patients in an overall interval of 1 year. The mean observation period was 11 months per neonate.

RESULTS:

In total, 22 cases were screen positive and 8 cases of inborn errors of metabolism were detected. The incidence of IEM in the population of patients admitted to the authors' NICU was 1.1%. The most common inborn error found was methylmalonic acidemia (3 cases, 37.5%), and all of the cases needed aggressive treatment and invasive mechanical ventilation. There were two cases of Tyrosinemia type 1, one case each of Maple Syrup Urine Disease, Propionic Acidemia, and Multiple Acyl-CoA dehydrogenase deficiency (MADD). Five of the eight patients required invasive mechanical ventilation. The median length of NICU stay was 3 days (1~7 days) and early therapeutic intervention was effective for four of them and other four patients (50%) died.

CONCLUSIONS:

The incidence of IEM in NICU was 1.1%, indicating an underestimation of the incidence of metabolic disorders prior to implementing **screening**. Most patients with IEM in the NICU required invasive mechanical ventilation and the mortality was increased due to underlying IEM.

27. J Pediatr. 2011 Jun 3. [Epub ahead of print]

Birth Prevalence Rates of Newborn Screening Disorders in Relation to Screening Practices in the United States.

Hertzberg VS, Hinton CF, Therrell BL, Shapira SK.

Abstract

OBJECTIVE:

To examine the associations between the first-tier-**screening** laboratory methods and criteria and the birth prevalence of congenital adrenal hyperplasia (CAH), phenylketonuria (PKU), and the sickle hemoglobinopathies occurring in the United States between 1991 and 2000.

STUDY DESIGN:

By using validated data from the National **Newborn Screening** and Genetics Resource Center, we fit Poisson regression models with laboratory methods and criteria used in every year for each state for each disorder. We also

examined whether there was an overall change in birth prevalence over the decade and whether there was an effect resulting from obligatory second screenings.

RESULTS:

There were no associations among any of the factors and the birth prevalence of PKU in this decade. Use of the enzyme-linked immunosorbent assay was more likely than any other laboratory method to identify cases of CAH (OR 1.16; 95% CI 1.04-1.30), but no other factors were associated with this disorder. None of the factors examined were associated with the birth prevalence rates of any of the sickle hemoglobinopathies.

CONCLUSION:

There were no substantial changes in the birth prevalence rates of PKU, CAH, or the sickle hemoglobinopathies over the study period despite rapid changes in technology.

28. N Engl J Med. 2011 Jun 2;364(22):2111-8.

Saliva polymerase-chain-reaction assay for cytomegalovirus screening in newborns.

Boppana SB, Ross SA, Shimamura M, Palmer AL, Ahmed A, Michaels MG, Sánchez PJ, Bernstein DI, Tolan RW Jr, Novak Z, Chowdhury N, Britt WJ, Fowler KB; National Institute on Deafness and Other Communication Disorders CHIMES Study.

Abstract

BACKGROUND:

Congenital cytomegalovirus (CMV) infection is an important cause of hearing loss, and most infants at risk for CMV-associated hearing loss are not identified early in life because of failure to test for the infection. The standard assay for **newborn** CMV **screening** is rapid culture performed on saliva specimens obtained at birth, but this assay cannot be automated. Two alternatives--real-time polymerase-chain-reaction (PCR)-based testing of a liquid-saliva or dried-saliva specimen obtained at birth--have been developed.

METHODS:

In our prospective, multicenter **screening** study of newborns, we compared real-time PCR assays of liquid-saliva and dried-saliva specimens with rapid culture of saliva specimens obtained at birth.

RESULTS:

A total of 177 of 34,989 infants (0.5%; 95% confidence interval [CI], 0.4 to 0.6) were positive for CMV, according to at least one of the three methods. Of 17,662 newborns screened with the use of the liquid-saliva PCR assay, 17,569 were negative for CMV, and the remaining 85 infants (0.5%; 95% CI, 0.4 to 0.6) had positive results on both culture and PCR assay. The sensitivity and specificity of the liquid-saliva PCR assay were 100% (95% CI, 95.8 to 100) and 99.9% (95% CI, 99.9 to 100), respectively, and the positive and negative predictive values were 91.4% (95% CI, 83.8 to 96.2) and 100% (95% CI, 99.9 to 100), respectively. Of 17,327 newborns screened by means of the dried-saliva PCR assay, 74 were positive for CMV, whereas 76 (0.4%; 95% CI, 0.3 to 0.5) were found to be CMV-positive on rapid culture. Sensitivity and specificity of the dried-saliva PCR assay were 97.4% (95% CI, 90.8 to 99.7) and 99.9% (95% CI, 99.9 to 100), respectively. The positive and negative predictive values were 90.2% (95% CI, 81.7 to 95.7) and 99.9% (95% CI, 99.9 to 100), respectively.

CONCLUSIONS:

Real-time PCR assays of both liquid- and dried-saliva specimens showed high sensitivity and specificity for detecting CMV infection and should be considered potential **screening** tools for CMV in newborns. (Funded by the National Institute on Deafness and Other Communication Disorders.).

29. Acta Paediatr. 2011 Jun;100(6):935. doi: 10.1111/j.1651-2227.2011.02314.x.

Retraction. Neonatal pulse oximetry screening: a national survey.

[No authors listed]

Retraction of

- Kang S, Tobin S, Kelsall W. Acta Paediatr. 2011 Feb 15;. doi: 10.1111/j.1651-2227.2011.02200.x.

30. Arch Dis Child. 2011 Jun;96(6):565-6. Epub 2010 Jun 28.

The TSH threshold in neonatal screening for congenital hypothyroidism: a variable solution.

Colón C, Alonso-Fernández JR.

Abstract

An algorithm is described whereby the threshold for thyroid-stimulating hormone used in **neonatal screening** for congenital hypothyroidism is re-set for each run on the basis of the variation and values of measurements of certified samples

31. Genet Med. 2011 Jun 1. [Epub ahead of print]

Long-term follow-up of children with confirmed newborn screening disorders using record linkage.

Wang Y, Caggana M, Sango-Jordan M, Sun M, Druschel CM.

Abstract

BACKGROUND:

Long-term follow-up of children identified through **newborn screening** is a critical process of data collection and analysis for advancing the public health understanding of the health outcomes and service uptake of the affected children. This article describes first steps toward the long-term follow-up of **newborn screening** children with confirmed disorders through records linkage using population-based administrative data.

METHODS:

The study cohort consisted of children born in 2006-2007 with confirmed disorders identified through **newborn screening**. Deterministic data linkage methods were used for record matching.

RESULTS:

The cohort was followed up to 2 years after birth by matching to data sources including vital records, hospital discharges, the Congenital Malformations Registry, and Early Intervention to monitor service utilization,

comorbidities, and mortality of the affected children. Of 1215 children with confirmed conditions identified through **newborn screening**, 25 deaths (2.1%) were identified, 86.1% used hospital (in- or outpatient) services, 36.1% were enrolled in the Congenital Malformations Registry, and 19.9% used the services of the Early Intervention program during the 2-year follow-up period.

CONCLUSIONS:

Long-term follow-up of children with disorders identified through **newborn screening** can be initiated by using existing administrative data. This method is an inexpensive, cost-effective. and efficient approach for periodical assessment of services utilization, the efficiency of service delivery, and health outcomes for affected individuals.

32. Int J Pediatr Otorhinolaryngol. 2011 Jun;75(6):745-8.

Highlights of the new WHO Report on Newborn and Infant Hearing Screening and implications for developing countries.

Olusanya BO.

Abstract

The Report summarizes the outcome of a recent informal consultation convened by the World Health Organization (WHO) in 2009 pursuant to the 1995 resolution of the World Health Assembly (WHA) urging Member States to promote programs for early hearing detection in babies and infants. The consultation was geared towards reaching global consensus on key principles on this subject based on the experiences and contributions of leading experts from various world regions and across relevant disciplines. After reviewing the current evidence on early hearing detection in babies and infants the Report outlined guiding principles for action by Member States covering issues such as etiology, case definition of hearing impairment, options for **screening**, program implementation, cost-effectiveness as well as policy and legislation. The need for context-specific adaptations of current practices in the developed world to facilitate the development of effective and culturally appropriate early hearing detection programs in developing countries was emphasized. The potential role of private-public partnerships including non-governmental organizations in designing and implementing hearing **screening** programs was highlighted while recognizing the necessity to develop requisite support services for infants detected with hearing impairment. Overall, the Report is likely to

stimulate greater interest and progress towards early hearing detection initiatives particularly in countries where necessary actions are yet to be taken to implement the WHA resolution. However, any effort in this direction must be backed by greater professional engagement, appropriate national policies and strong involvement of WHO regional offices in developing countries.

33. J Allergy Clin Immunol. 2011 Jun;127(6):1394-9.

Neonatal screening for severe combined immunodeficiency caused by an adenosine deaminase defect: A reliable and inexpensive method using tandem mass spectrometry.

Azzari C, la Marca G, Resti M.

Abstract

BACKGROUND:

Adenosine deaminase (ADA)-severe combined immunodeficiency (SCID) is an SCID caused by a defect in the enzyme adenosine deaminase. It is usually fatal in infancy because of severe recurrent infections. When diagnosis is made, permanent damage caused by infections or by metabolites is often present. Gene therapy, bone marrow transplantation, or enzyme therapy might be effective if performed early. ADA-SCID complies with all the criteria for inclusion in a **newborn screening** program. However, **screening** methods are still expensive or provide a non-negligible number of indeterminate results.

OBJECTIVE:

The aim of the present study was to develop a simple, reliable, and inexpensive method for diagnosis of ADA-SCID by using dried blood spot (DBS) samples taken at birth. Cost per test was calculated, including the cost for reagents, equipment, and operators.

METHODS:

DBS samples from 4 patients with genetically confirmed ADA-SCID and 12,020 DBS samples from healthy newborns were examined. Adenosine and 2'-deoxyadenosine were tested by using tandem mass spectrometry (PCT EP2010/070517).

RESULTS:

The mean levels of adenosine and 2'-deoxyadenosine were 7.8 ± 3.1 and 8.5 ± 6.0 μmol/L, respectively, in affected children; adenosine was found at 0.23 ± 0.09 μmol/L, whereas 2'-deoxyadenosine was never detected in healthy control subjects (adenosine: P < 10(-6) [95% confidence limit, 7.59-7.78] and 2'-deoxyadenosine: P < 10(-6) [95% confidence limit, 8.65-8.82] for control subjects vs patients with ADA-SCID). No indeterminate or false-positive results were found. Cost per test was €0.01 ($0.013). A pilot population-based **newborn screening** for ADA-SCID has started in Tuscany, Italy.

CONCLUSION:

Tandem mass spectrometry can be used for diagnosis of one of the most frequent form of SCID at a negligible cost.

34. J Chromatogr B Analyt Technol Biomed Life Sci. 2011 Jun 1;879(19):1565-72. Epub 2011 Apr 4.

Simultaneous quantification of 17α-OH progesterone, 11-deoxycortisol, Δ4-androstenedione, cortisol and cortisone in newborn blood spots using liquid chromatography-tandem mass spectrometry.

Magnisali P, Chalioti MB, Livadara T, Mataragas M, Paliatsiou S, Malamitsi-Puchner A, Moutsatsou P.

Abstract

Adrenal steroid profiling, including 17α-OH progesterone (17OHP), 11-deoxycortisol (S), Δ4-androstenedione (Δ4-A) and cortisol (F) in blood spots by tandem mass spectrometry, is used for **newborn screening** to detect congenital adrenal hyperplasia (CAH). Pre-analytical sample processing is critical for assay specificity and accuracy; however, it is laborious and time-consuming. This study describes the development and validation of a new Liquid Chromatography-Tandem Mass Spectrometry (LC-MS/MS) method for the simultaneous quantification of five steroids: 17OHP, S, Δ4-A, F and cortisone (E) in blood spots from newborns. Whole blood was eluted from a 5.00 mm dried blood spot by an aqueous solution containing the deuterium-labeled internal standards d8-17OHP and d4-

cortisol. The steroids extracted from blood spot into aqueous solution were subsequently purified via Extelut mini NT1 column using diethylether. The extracts were evaporated and quantified using LC-MS/MS. The detection limit was 0.25 ng/mL for 17OHP and S, 0.4 ng/mL for Δ4-A and 0.5 ng/mL for F and E. The limit of quantification was 0.5 ng/mL for 17OHP, S and Δ4-A and 1 ng/mL for F and E. Precision for 17OHP, S, Δ4-A at concentrations of 0.5, 2, and 8 ng/mL (n=5) in fortified steroid free serum samples was 1.3-3.5% (intra-assay CV) and 7-14.8% (inter-assay CV). Precision for F and E at concentrations of 5 and 20 ng/mL was 1.5-4.8% (intra-assay, CV%) and 6-15% (inter-assay, CV%). Accuracy was calculated at concentrations of 0.5, 2, and 8 ng/mL for 17OHP, S and Δ4-A and ranged from -0.3 to 0.2%, while for F and E it ranged from -3.2 to 0.2%. Relative recoveries at concentration 2 ng/mL and 8 ng/mL for 17OHP, S, Δ4-A and at 5 ng/mL and 20 ng/mL for F and E ranged from 55% to 80%. Reference intervals were estimated for all steroids in newborns (on day 3). The steroid profile assay herein described is sensitive, specific and accurate and involves a simple pre-analytical sample manipulation; it is therefore suitable for routine analysis and provides data for samples within normal range as well as those with elevated levels. For the first time to our knowledge, cortisone levels are reported in dried blood spots from newborns.

35. J Clin Endocrinol Metab. 2011 Jun 1. [Epub ahead of print]

Is the Incidence of Congenital Hypothyroidism Really Increasing? A 20-Year Retrospective Population-Based Study in Quebec.

Deladoëy J, Ruel J, Giguère Y, Van Vliet G.

Abstract

Context: Congenital hypothyroidism (CH) is reportedly increasing in the United States, possibly reflecting changes in **screening** methods. In Québec, the same initial TSH cutoff (15 mU/liter) has been used for the last 20 yr, but in 2001, the cutoff was decreased from 15 to 5 mU/liter for the second test, which is requested when TSH is intermediate (15-30 mU/liter) on the first. Objectives: Our objective was to assess the incidence of CH over the last 20 yr in Québec. Design, Setting, Patients, and Main Outcome Measure: This is a population-based retrospective study. Incidences by etiology based on thyroid scintigraphy with technetium were compared between 1990-2000 and 2001-2009. Results: Of 1,660,857 newborns over 20 yr, 620 had CH (incidence 1:2679). Etiology was dysgenesis (n = 389, 1:4270), either ectopy (n = 290) or athyreosis (n = 99), goiter (n = 52, 1:31,940), normal-size gland in situ (n = 115, 1:14,442), and unknown (n = 64, 1:25,950). The new **screening** algorithm identified 49 additional cases (i.e. 25 normal-size gland in

situ, 12 unknown etiology, 10 ectopies, and two goiters). Consequently, the incidence of normal-size gland in situ or of unknown etiology more than doubled (1:22,222 to 1:9,836, P = 0.0015; and 1:43,824 to 1:17,143, P = 0.0018, respectively) but that of dysgenesis and goiter remained stable. Had the 1990-2000 algorithm been applied in 2001-2009, no change in incidence would have been observed in any category. Conclusion: Estimating the incidence of CH is influenced by minimal changes in TSH **screening** cutoffs. Lower cutoffs identify additional cases that have predominantly functional disorders whose impact on intellectual disability, if left untreated, remains to be determined.

36. J Clin Endocrinol Metab. 2011 Jun;96(6):1671-3.

The continuing health burden of congenital hypothyroidism in the era of neonatal screening.

Van Vliet G, Grosse SD.

37. J Inherit Metab Dis. 2011 Jun;34(3):569-74. Epub 2011 Apr 16.

Newborn screening: how are we travelling, and where should we be going?

Wilcken B.

Abstract

In general, **newborn screening** is now a highly successful enterprise. The introduction of tandem mass spectrometry in the mid-1990s changed the pace of **screening**, raising its profile and increasing its relevance to a wider range of health professionals. The clinical effectiveness is not in doubt for some conditions, but is lacking for others. Evaluation has major difficulties for the rarer disorders and has been sadly neglected. Partly because clinical effectiveness has not been enthusiastically addressed, but also because of undue caution on the part of regulators, who often seem to ignore available evidence, there are huge differences in the adoption of **screening** programmes in different jurisdictions. New treatments, especially mutation-specific treatments, and technological advances in diagnostic testing are being rapidly developed, and this will further change the face of **newborn screening** and probably magnify these differences. The challenges will be considerable, especially with the increasing availability of DNA testing at modest cost. It is likely that there will be pressure to change the aims of **newborn screening** to encompass "personalised medicine". We must all prepare in a thoughtful way for these future challenges.

38. J Trop Pediatr. 2011 Jun;57(3):232-4. Epub 2010 Jul 7.

Congenital Adrenal Hyperplasia in Alexandria, Egypt: A High Prevalence Justifying the Need for a Community-based Newborn Screening Program.

Tayel SM, Ismael H, Kandil H, Abd Rabuh AR, Sallam H.

39. Pediatrics. 2011 Jun;127(6):e1455-63. Epub 2011 May 29.

Parents' decisions to screen newborns for FMR1 gene expansions in a pilot research project.

Skinner D, Choudhury S, Sideris J, Guarda S, Buansi A, Roche M, Powell C, Bailey DB Jr.

Abstract

OBJECTIVE:

The goal of this study was to document rates of parental consent in a pilot study of **newborn screening** for FMR1 gene expansions, examine demographic characteristics of mothers who consented or declined, describe the reasons for their decision, and discuss ethical and social aspects of the consent process.

METHODS:

A brief survey was used to record basic demographic data from mothers and an open-ended question was used to elicit parents' reasons for accepting or declining **screening**. A descriptive analysis was conducted on the number of mothers who consented to or declined **screening**, and a logistic regression model predicted mothers' likelihood to agree to **screening** based on demographic characteristics. Reasons for decisions were analyzed using content analysis. The study was conducted at University of North Carolina Hospitals. A total of 2137 mothers were approached.

RESULTS:

The uptake rate for couples was 63%. Acceptance rates varied by race/ethnicity, with black respondents being less likely to accept **screening**. Primary reasons for accepting were "to know," "belief in research," and "the test was

minimal/no risk." Reasons for declining included not wanting to know or worry, not being a good time, and issues with testing children or with genetic tests.

CONCLUSIONS:

Findings demonstrate that a majority of parents accepted **newborn screening** for FMR1 gene expansions, but decision rates and reasons for accepting or declining varied in part as a function of race/ethnicity and in part as a function of what parents most valued or feared in their assessment of risks and benefits.

40. Pediatrics. 2011 Jun;127(6):e1593-4. Epub 2011 May 29.

Newborn screening for fragile x syndrome: do we care what parents think?

Botkin JR.

41. Semin Perinatol. 2011 Jun;35(3):155-61.

Neonatal screening for glucose-6-phosphate dehydrogenase deficiency: biochemical versus genetic technologies.

Kaplan M, Hammerman C.

Abstract

Glucose-6-phosphate dehydrogenase (G-6-PD) deficiency, a commonly occurring genetic condition, is associated in neonates with severe hemolytic episodes, extreme hyperbilirubinemia, and bilirubin encephalopathy. **Neonatal screening** programs for the condition should increase parental and caretaker awareness, thereby facilitating early access to treatment with resultant diminished mortality and morbidity. However, **screening** for G-6-PD deficiency is not widely performed. Although G-6-PD-deficient males may be accurately identified, females are more difficult to categorize because many in this group may be heterozygotes with phenotype overlap between normal homozygotes, heterozygotes, and deficient homozygotes. **Screening** methodologies include biochemical qualitative assays, quantitative enzymatic activity measurements and DNA-based polymerase chain reaction molecular **screening**. The appropriateness of any of these technologies for any particular population group or geographic area must be assessed before setting up a **screening** program. The pros and cons of each method, including ease of testing, cost, need for

sophisticated laboratory equipment and degree of personnel training, as well as the ability to identify females, are discussed.

42. World J Pediatr. 2011 Jun 1. [Epub ahead of print]

Diagnosis and treatment of subclinical hypothyroidism detected by neonatal screening.

Chen XX, Qin YF, Zhou XL, Yang RL, Shi YH, Mao HQ, Qu YP, Wang X, Zhao ZY.

Abstract

BACKGROUND:

This study was undertaken to explore the clinical outcome and prognosis of subclinical hypothyroidism detected by **newborn screening**.

METHODS:

Newborn screening was conducted at 1156 health care institutions in Zhejiang Province from October 1999 to September 2006. Included were (1) infants who had thyroid-stimulating hormone (TSH) ≥20 mU/L, and normal or lower normal levels of triiodothyronine (T(3)) and thyroxine (T(4)) and (2) infants with TSH between 5.6 mU/L and 20 mU/L at a confirmatory examination and follow-up showing TSH levels ≥20 mU/L or delayed reduction in T4 levels. These infants were considered as having subclinical hypothyroidism and levothyroxine (LT(4)) at an initial dose of 3-5 μg/kg per day was administered. The levels of TSH and T(4), developmental quotient (DQ), and index of growth were evaluated.

RESULTS:

A total of 204 infants met our criteria for subclinical hypothyroidism, with an incidence of 1/8809. After 2-4 weeks of standard therapy, serum TSH level dropped to normal and T4 reached a higher normal level in all the 204 infants. Evaluations of 60 patients after 2 years of therapy showed that their average DQ was 101±14.61, and body weight and height were within the normal ranges. Bone age test for 54 patients revealed normal development in 44, slightly retarded development in 7, and advanced development in 3.

CONCLUSIONS:

Newborns with high TSH levels should be given particular attention to ensure early diagnosis. A L-T(4) dose of 3-5 μg/kg per day was effective in the initial treatment of subclinical hypothyroidism.

43. Clin Endocrinol (Oxf). 2011 May 27. doi: 10.1111/j.1365-2265.2011.04128.x. [Epub ahead of print]

THE INCREASED INCIDENCE OF CONGENITAL HYPOTHYROIDISM: FACT OR FANCY?

Mitchell ML, Hsu HW, Sahai I; and the Massachusetts Pediatric Endocrine Work Group.

Abstract

Objective: The incidence of congenital hypothyroidism (CH) detected by **newborn screening** in the US has increased significantly since the early 1990s. We defined the characteristics associated with the increased incidence. Patients: A cohort of children with CH born during an earlier period of low incidence (1991-94) was compared with a cohort born during a later period when the incidence of CH had doubled (2001-04). Measurements: **Screening** was performed with T4 as the primary marker and TSH on selected specimens. Follow-up on hypothyroid children determined whether they had permanent or transient hypothyroidism. Cases were classified based on laboratory results: initial TSH ≥100mU/L was "severe," initial TSH <100mU/L but ≥20mU/L was "mild," and initial TSH <20mU/L with subsequent abnormal TSH was "delayed. Results: The overall incidence of CH almost doubled between the two time periods, from 1:3010 to 1:1660. Excess cases were found in the mild and delayed categories, with no increase in severe cases. The proportion of transient cases was <5% in severe cases, 40% in mild cases and 70% among delayed cases. There was no difference in the proportion of transient case between the two time periods. Modifications to the T4/TSH testing protocol between the two time periods resulted in substantially increased numbers of specimens in the younger cohort being selected for TSH testing in both initial and repeat specimens. Conclusion: The rising incidence of CH in Massachusetts is confined to mild and delayed cases. Our findings suggest that this rise is attributable to enhanced detection rather than an absolute increase in numbers.

44. J Inherit Metab Dis. 2011 May 27. [Epub ahead of print]

Putting a value on the avoidance of false positive results when screening for inherited metabolic disease in the newborn.

Dixon S, Shackley P, Bonham J, Ibbotson R.

Abstract

Despite the increase in the number of inherited metabolic diseases that can be detected at birth using a single dried blood spot sample, the impact of false positive results on parents remains a concern. We used an economic approach - the contingent valuation method - which asks parents to give their maximum willingness to pay for an extension in a **screening** programme and the degree to which the potential for false positive results diminishes their valuations. 160 parents of a child or children under the age of 16 years were surveyed and given descriptions of the current **screening** programme in the UK, an extended programme and an extended programme with no false positives. 148 (92.5%) respondents said they would accept the screen for the five extra conditions in an expanded **screening** programme whilst 10 (6.3%) said they would not and two were unsure. When asked to indicate if they would choose to be screened under an expanded **screening** programme with no false positive results, 152 (95%) said they would, five (3.1%) said they would not, two were unsure, and there was one non-response. 151 (94.4%) said they preferred the hypothetical test with no false-positives. The mean willingness to pay for the expanded programme was £178 compared to £219 for the hypothetical expanded programme without false positives ($p > 0.05$). The results suggest that there is widespread parental support for extended **screening** in the UK and that the number of false-positives is a relatively small issue.

45. Ital J Pediatr. 2011 May 21;37:25.

Storage and use of residual newborn screening dot blood samples in Italy.

Petrini C, Olivieri A, Corbetta C, Cerone R, D'Agnolo G, Bompiani A.

46. Genet Med. 2011 May 19. [Epub ahead of print]

Committee Report: Considerations and Recommendations for National Guidance Regarding the Retention and Use of Residual Dried Blood Spot Specimens After Newborn Screening.

Therrell BL Jr, Hannon WH, Bailey DB Jr, Goldman EB, Monaco J, Norgaard-Pedersen B, Terry SF, Harris A, Vasquez LM, Johnson A, Lloyd-Puryear MA, Howell RR.

Abstract

Newborn screening programs are state based with variable policies. Guidance regarding the retention, storage, and use of portions of **newborn screening** dried blood spots that remain after **screening** (residual specimens) was first published in 1996. Since then, **newborn screening** programs have paid increased attention to specimen storage and usage issues. Standard residual specimen uses include quality assurance and program evaluation, treatment efficacy, test refinement, and result verification. In all cases, privacy and security are primary concerns. In general, two distinct state practices regarding the storage and use of residual **newborn screening** specimens exist: (1) short-term storage (<3 years), primarily for standard program uses and (2) long-term storage (>18 years), for standard program uses and possible important public health research uses. Recently, there have been concerns in some consumer communities regarding both the potential uses of residual specimens and patient (**newborn** and family) privacy. To assist in policy improvements that can protect the individual's privacy and allow for important public health uses of residual **newborn screening** specimens, the Secretary of Health and Human Services' Advisory Committee on Heritable Disorders in Newborns and Children has developed recommendations (with requested action by the Secretary where applicable). This report presents the Committee's recommendations and reviews the pertinent associated issues.

47. Genet Test Mol Biomarkers. 2011 May 16. [Epub ahead of print]

Providing Genetic Risk Information to Parents of Newborns with Sickle Cell Trait: Role of the General Practitioner in Neonatal Screening.

Vansenne F, de Borgie CA, Legdeur M, Spauwen MO, Peters M.

Abstract

Purpose: In 2007, the **neonatal screening** program in the Netherlands was expanded to include hemoglobinopathies. Newborns with sickle cell disease (SCD), as well as SCD carriers are identified. The benefit of reporting SCD carriers includes detection of more couples at risk (both parents are carriers) who can be informed about future reproductive choices, a responsibility of their general practitioner (GP). We evaluated knowledge, ideas, and actions

of GPs after reporting SCD carriers and explored and analyzed potential barriers. Methods: A questionnaire study. Results: A total of 139 GPs responded to our questionnaire (49%). Ninety GPs (90%) stated they informed parents of the test result. In only 23 cases (23%) both parents had themselves tested for hemoglobinopathies. Eighty-one GPs (64%) stated that they did not have enough clinical experience with SCD. Almost half of the GPs indicated that they did not experience any barriers in counseling patients (n=60, 48%). Conclusion: At the moment, the goal of the **neonatal screening** for SCD carriers has not been achieved as the majority of parents were not tested for hemoglobinopathies after disclosure of carrier status in their **newborn**. With GPs reporting few barriers in counseling parents and only indicating a lack of knowledge and clinical experience, more effort is required to provide better information to GPs to help facilitate their work.

48. Pediatr Blood Cancer. 2011 May 16.

Increased prevalence of false positive hemoglobinopathy newborn screening in premature infants.

Hustace T, Fleisher JM, Sanchez Varela AM, Podda A, Alvarez O.

Abstract

BACKGROUND:

The objective was to investigate the specificity of the hemoglobinopathy **newborn screening** in premature neonates as compared to term neonates.

PROCEDURE:

The **screening** results from infants suspected to have hemoglobinopathy disease identified by the Florida **Newborn Screening** Program for years 2002-2007 were compared to the corresponding confirmatory testing. The risks for false positives for preterm and full term newborns were calculated by Chi-square or the Cochran-Armitage test for trend. Isoelectric focusing and HPLC were the methods of hemoglobin **screening**.

RESULTS:

Over 2,300 neonates (1/576 neonates born in Florida) were suspected to have hemoglobinopathy. The most common abnormal pattern in term and preterm infants (gestational age 22-36 weeks) suggesting disease at **screening** was FS. Overall, 93% of the children who screened positive for FCA and 64% of infants identified with FSA were later confirmed with trait. FSC was confirmed in 96% of the cases in both preterm and term infants. Compared to term newborns, preterm newborns were more likely to have a false positive result for FS or FC at **screening**. Twenty-three percent of preterms with FS and 59% of preterms with FC were diagnosed as traits by confirmatory testing, compared to only 2% and 6% for term infants ($P < 0.001$).

CONCLUSIONS:

As compared to term newborns, more preterm newborns with trait were misidentified as having sickle cell anemia or hemoglobin C at **screening**. We speculate that abnormal hemoglobins may precede the development of hemoglobin A during fetal life.

49. Scand J Clin Lab Invest. 2011 May 5. [Epub ahead of print]

Neonatal plasma TSH - estimated upper reference intervals for diagnosis and follow up of congenital hypothyroidism.

Evans C, Neale S, Geen J, Jones G, Mannings L, Trow S, Brain A, Nix B, Ellis R, Hancock S, Shine B, Warner J, Gregory JW, Moat SJ.

Abstract

Abstract Age- and method-dependent plasma TSH reference intervals are essential for the diagnosis and management of congenital hypothyroidism. However, accurate reference intervals for plasma TSH have not been adequately defined due to the difficulties in obtaining samples from a healthy paediatric population. To overcome the difficulties in generating such intervals we estimated method-dependent plasma TSH upper-reference intervals by determining the blood spot TSH upper-reference interval from **newborn** blood spot TSH **screening** data (N = 10,697) and then derived method-dependent conversion factors for blood spot TSH to plasma TSH concentration from paired-blood spot and plasma TSH measurements. The upper reference interval for blood spot TSH of 3.04 mU/L was obtained from the 97.5th centile of the selected data. Using experimentally-derived conversion factors, estimates of plasma TSH upper reference intervals of 7.6, 6.3, 7.3, 8.3 and 6.5 mU/L were obtained for the Siemens

Centaur, Abbott Architect, Roche Elecsys E170, Siemens Immulite 2000 and Beckman access HYPERsensitive TSH assays respectively. These estimated method-dependent plasma TSH upper reference intervals will be of great practical use to clinicians to diagnose and to follow up infants found to have increased blood spot TSH concentrations identified by **Newborn Screening** programmes.

50. Genet Med. 2011 May 4. [Epub ahead of print]

Genetic counseling following the detection of hemoglobinopathy trait on the newborn screen is well received, improves knowledge, and relieves anxiety.

Kladny B, Williams A, Gettig EA, Krishnamurti L.

Abstract

PURPOSE:

The primary purpose of **newborn screening** for hemoglobinopathies is the presymptomatic diagnosis and early treatment of sickle cell disease. Hemoglobinopathy traits detected on the **newborn screening** provide an opportunity for genetic counseling of families regarding the trait and information that may impact reproductive decisions of the parents. We describe the results of a study to determine the impact of **newborn screening** and genetic counseling on the lives of families in which an abnormal hemoglobin trait had been identified.

METHODS:

From June 2003 to December 2009, families of children with trait attending a clinic visit and receiving professional genetic counseling were asked to participate in a semistructured follow-up survey regarding their experience and the impact of genetic counseling on their families.

RESULTS:

Of the 300 patients seen in clinic during the specified time period, 209 consented to be recontacted and 114 have completed the survey. Eighty-five percent of responders reported knowing that the **newborn** screen had been performed, but only 55% understood the purpose of **newborn screening**. When asked about the effect of finding out that trait was present in their baby, 19% reported feeling guilty or upset, whereas 4% believed that their partner

blamed them for the child's results. That genetic counseling was found to be beneficial was indicated by the fact that 99% reported that their questions were answered, 82% reported feeling less anxious, and 78% discussed the trait with their partner after the appointment.

CONCLUSIONS:

Genetic counseling after **newborn screening** relieves anxiety, provides knowledge, facilitates dialog within families and between partners about hemoglobinopathy trait, and was seen as a positive experience for the majority of responders.

51. Int J Lab Hematol. 2011 May 3. doi: 10.1111/j.1751-553X.2011.01315.x. [Epub ahead of print]

An evaluation of the Sebia capillarys Neonat Haemoglobin FAST™ system for routine newborn screening for sickle cell disease.

Murray C, Hall SK, Griffiths P.

Abstract

The West Midlands **Newborn Screening** Laboratory (NBSL) at Birmingham Children's Hospital (BCH), UK, screens approximately 71 000 babies per annum using the Bio-Rad automated VARIANT™ nbs (Vnbs) high-pressure liquid chromatograph (HPLC). Any abnormal haemoglobins detected, including S, C, D-Punjab, E and O-Arab as directed by the NHS Sickle Cell and Thalassaemia **Screening** Programme (NHS Sickle Cell and Thalassaemia **Screening** Programme Website, http://sct.**screening**.nhs.uk), are then confirmed using Resolve(®) isoelectric electric focusing (IEF) kits supplied by Perkin-Elmer. The Sebia capillarys Neonat Haemoglobin FAST™ system was evaluated as a possible replacement for the first- or second-line methods used. Both the Sebia and Bio-Rad methods were compared using anonymized blood spots with known haemoglobin patterns. These results were then confirmed when necessary by IEF. The Sebia-recommended sample preparation was also modified to enable testing to be more comparable with our current processes. Percentages of haemoglobins calculated from integration of areas under the peaks were compared between the Bio-Rad Vnbs HPLC and Sebia capillarys Neonat Haemoglobin FAST™ system. Of the 347 blood spots tested by both HPLC and capillary electrophoresis, there were no significant differences. The Sebia capillarys Neonat Haemoglobin FAST™ system can be used to

successfully screen newborns for sickle cell disease in blood spots collected for **newborn screening** with full positive sample identification and traceability.

52. Am J Med Genet A. 2011 May;155A(5):

Late onset Pompe disease revealed by newborn screening.

Levenson D

53. Ann Saudi Med. 2011 May-Jun;31(3):316-7.

Comment on: Outcome of a newborn hearing screening program in a tertiary hospital in Malaysia: The first five years. Ann Saudi Med 2011; 31: 24-8.

Al-Mendalawi MD.

54. J Inherit Metab Dis. 2011 May;34 Suppl 1:S1-16.

Screening newborns: current state and future challenges. Abstracts of the International Congress on Prevention of Congenital Diseases. Vienna, Austria. May 13-14, 2011.

55. Med Health R I. 2011 May;94(5):121-3.

Rhode Island metabolic newborn screening: the effect of early identification. A case report of argininosuccinic aciduria (ASA).

Beck NM, Johnston JP, Lemke KS, Pogacar P, Phornphutkul C.

56. J Cyst Fibros. 2011 Apr 30. [Epub ahead of print]

Cystic fibrosis newborn screening does not delay the identification of cystic fibrosis in children with negative results.

Maclean JE, Solomon M, Corey M, Selvadurai H.

Abstract

BACKGROUND:

Several studies have demonstrated the benefit of Cystic Fibrosis **Newborn Screening** (CFNBS) for early diagnosis and, hence, intervention but the impact of CFNBS on those children not detected on CFNBS is not known. CFNBS may provide false reassurance that all CF has been detected and, therefore, lead to a delay in the diagnosis of children with CF which is not detected on CFNBS. The aim of this study was to determine the impact of CFNBS on the presenting features of children with CF where CF was not detected on CFNBS.

METHODS:

Subjects at the CFNBS center were selected if CF was identified subsequent to a negative CFNBS with subjects at the No CFNBS selected based on the absence of ΔF508 mutations. Children presenting with features that would lead to investigation for CF independent of clinical status were excluded. Presenting features at diagnosis and pulmonary function at 6years of age were extracted from medical records.

RESULTS:

Twelve children from the CFNBS site and 19 from the No CFNBS site were included in the analysis. The only significant difference between the two in features at diagnosis was lower mean weight z-scores at the No CFNBS site (-2.9±1.8) compared to the CFNBS center (-1.4±1.3, $p<0.05$). Age at diagnosis, presenting complaint and nutritional status did not differ by site. Growth parameters and pulmonary function at 6years of age showed no differences between sites.

CONCLUSIONS:

This study demonstrates that access to CFNBS does not result in delay in diagnosis or poorer outcomes in those children for whom CF was not detected on CFNBS. In addition, children with CF not detected on CFNBS present with typical features of CF and sweat chloride results that are diagnostic of CF.

57. Clin Genet. 2011 Apr 29. doi: 10.1111/j.1399-0004.2011.01694.x. [Epub ahead of print]

Molecular testing in congenital adrenal hyperplasia due to 21α-hydroxylase deficiency in the era of newborn screening.

Sarafoglou K, Lorentz C, Otten N, Oetting W, Grebe S.

Abstract

Sarafoglou K, Lorentz CP, Otten N, Oetting WS, Grebe SKG. Molecular testing in congenital adrenal hyperplasia due to 21α-hydroxylase deficiency in the era of **newborn screening**. **Newborn screening** (NBS) identifies the majority of classical [salt-wasting (SW) and simple-virilizing (SV)] cases of congenital adrenal hyperplasia (CAH) due to 21α-hydroxylase (21α-OHase) during the first days of life. Diagnosis of classical CAH is confirmed by follow-up serum 17-hydroxyprogesterone and/or the adrenocorticotropin stimulation test; however, neither test definitively distinguishes between the classical subtypes. After confirmation, all newborns are started on hydrocortisone (glucocorticoid) and fludrocortisone (mineralocorticoid) treatment. While initiating fludrocortisone treatment in classical CAH patients, independent of subtype and before SW signs or symptoms occur, prevents a life-threatening SW crisis, it may later complicate distinguishing between the classical subtypes. Genotype-phenotype correlations in 21α-OHase deficiency are excellent; however, molecular testing is not a regular part of the diagnostic workup. Molecular testing on 39 patients (25 identified by NBS) with an already established diagnosis of CAH identified 11 SW patients (8 identified by NBS) whose mutations suggested further biochemical and clinical reassessment of their subtype. Overall, SW accounted for 57.6% of our classical CAH patients, below the generally accepted figure that >75% of classical CAH are comprised of the SW form. In the era of NBS, molecular testing is a valuable supplemental tool identifying patients who may benefit from reassessment of their salt-retaining ability.

58. Clin Endocrinol (Oxf). 2011 Apr 25.

Neonatal thyroid screening results are related to gestational maternal thyroid function.

Kuppens SM, Kooistra L, Wijnen HA, Vader HL, Hasaart TH, Oei SG, Vulsma T, Pop VJ.

Abstract

Objective: To study the relationship between maternal thyroid function at each pregnancy trimester and **neonatal screening** results. Background: Overt maternal thyroid dysfunction during gestation is associated with poor **neonatal** thyroid function. However, research on the relationship between suboptimal maternal thyroid function (assessed at 3 trimesters) and **neonatal** thyroid **screening** outcome is scarce. Design/Patients: Prospective follow-up study during three trimesters of gestation in 886 Dutch Caucasian healthy pregnant women followed from 12 weeks gestation until term delivery (> 37 weeks) and their neonates. Measurements: The relation between

neonatal data from the Congenital Hypothyroidism (CH) **screening** and maternal thyroid determinants (TSH, FT4 and TPO-Ab) assessed at 12, 24 and 36 weeks gestation. Results: Boys have lower **screening** TT4 levels and their mothers have higher TSH levels at 24 and 36 weeks gestation. Higher maternal TSH levels (> 97.5th percentile, as defined in 810 women without TPO-Ab at 12 weeks) at one or more times during pregnancy (O.R: 2.26, 95% CI: 1.20 - 4.29). and lower gestational age (O.R : 1.22, 95% CI: 1.05 - 1.41) are independently related to lower **screening** TT4 levels Conclusions: Maternal thyroid function during gestation is related to **neonatal** TT4 at **screening**. The finding of both lower **neonatal** TT4 levels in boys and higher TSH levels in mothers carrying boys is worthy of further investigation, as both observations may be meaningfully related.

59. Amino Acids. 2011 Apr 21. [Epub ahead of print]

A simple method for the analysis by MS/MS of underivatized amino acids on dry blood spots from newborn screening.

Wang C, Zhang W, Song F, Liu Z, Liu S.

Abstract

The analysis by electrospray-ionization tandem mass spectrometry of amino acids with butyl esterification and isotopically labeled internal standard is routine in **newborn screening** laboratories worldwide. In the present study, we established a direct analysis method of higher accuracy that uses a non-deuterated internal standard. The automatic sampler and the pump of an LC apparatus were used to inject sample and mobile phase to MS, but no LC column was needed. The dry blood spot (DBS) material was prepared at levels of low, medium and high concentration; the running time was 1 min. In parallel to the new procedure, we applied the established method to analyze nine amino acids on DBS of healthy newborns and phenylketonuria newborns. The newly proposed method of product ion confirmation scan along with multiple reaction monitoring resulted in a very accurate identification of each amino acid. Our innovative protocol had high sensitivity and specificity in the analysis of cases of suspected metabolic diseases.