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Professor of Pediatrics and Genetics
2014 APHL Newborn Screening and Genetic Testing Symposium
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Conflicts of Interest

• None
Next Generation Sequencing

Pipeline Overview

DNA sample collected

Library and Sample preparation

NGS platform DNA sequencing

Alignment, mapping

variant calling

Annotations

Filter VCF

Raw VCF

Bioinformatics computing facility

Final consolidated clinical report

Specialist validation and conclusions

Visualization and Analysis

Patient

Next Gen Sequencing

• Can search for mutations in all genes (20,000)
• Whole exome: just coding parts of genes (exons)
• Whole genome: everything (exons and introns)
• Analysis is complex – our understanding of what is a significant mutation and what is a benign polymorphism is incomplete
Sanger vs. Whole-Exome Sequencing: Technical Considerations

- **Sanger**
  - 100-800+ bp
  - Targeted mutation analysis
  - Complete coverage
  - “Gold standard”

- **WES**
  - 30 Mb in exome (3 billion in entire genome)
  - Mutation fishing in many targets
  - Interpretation difficulties
  - Not considered reliable enough to use without confirmation
• **Question A)** For **disorders currently screened for in newborns**, how can genomic sequencing replicate or augment (e.g., make more accurate, comprehensive or inexpensive) known newborn screening results?

• **Question B)** What knowledge about **conditions not currently screened for in newborns** could genomic sequencing of newborns provide?

• **Question C)** What **additional clinical information** could be learned from genomic sequencing relevant to the clinical care of newborns?

In order to be considered responsive to the FOA, each applicant must also propose a research plan that includes **each of the following three component projects**:

• **Research Component 1)** acquisition and analysis of **genomic datasets** that expand considerably the scale of data available for analysis in the newborn period;

• **Research Component 2)** **clinical research** that will advance understanding of specific disorders identifiable via newborn screening through promising new DNA-based analysis; and

• **Research Component 3)** research related to the **ethical, legal and social implications (ELSI)** of the possible implementation of genomic sequencing of newborns.

The methods and scope of the research in all three of these component projects should be tailored to **focus on the newborn period** and the **research context** in which the sequencing is performed.
Four Centers Funded – U19/NICHD and NHGRI

University of North Carolina at Chapel Hill
Brigham and Women’s Boston
Children’s Mercy Hospital in Kansas City, MO
University of California San Francisco

**NSIGHT**: Newborn Sequencing In Genomic medicine and public Health
Recommended Uniform Screening Panel

- 2005 Task Force funded by HRSA through contract to ACMG recommended core panel of 29 conditions that all states should screen for and 29 additional secondary conditions that would be detected as part of screening for core conditions
- New conditions can be “nominated”
- Limitations include no screening tool, screening tool too expensive, no treatment
- Currently 31 core disorders and 26 secondary disorders
Fig. 7. Scores for all conditions distinguished by screening panel category
Fig. 8. Distribution of conditions into screening panel categories
Next Generation Sequencing in Newborn Screening

• Barriers to adding any disorder to NBS panel may now be overcome if there is a genetic etiology established for a condition
NC Newborn Exome Sequencing for Universal Screening (NC NEXUS) Overarching Aims

1. Evaluate how Next Generation Sequencing (NGS)-Newborn Screening (NBS) can extend the utility of current NBS.

2. Devise and evaluate a clinically oriented framework for analysis of NGS-NBS.

3. Develop best practices for incorporating NGS-NBS into clinical care.
A Semi-Quantitative Metric Approach to Scoring Genes

Severity
- 3 = Sudden death
- 2 = Possible death
- 1 = Serious morbidity
- 0 = Modest or no morbidity

Likelihood
- 3 = >50%
- 2 = 5-49%
- 1 = 1-5%
- 0 = <1%

Efficacy
- 3 = Highly
- 2 = Moderately
- 1 = Minimally
- 0 = Ineffective

Acceptability
- 3 = Highly
- 2 = Moderately
- 1 = Minimally
- 0 = Ineffective

Knowledge
- 3 = Substantial
- 2 = Moderate
- 1 = Minimal
- 0 = Controversial or poor

TOTAL SCORE RANGE
0 – 15
A “medical actionability” score

J. Berg
Example: PAH (PKU)

– Severity of disease (ID) = 1
– Likelihood of a severe outcome = 3
– Effectiveness of interventions = 3
– Acceptability of interventions = 2
– Knowledge base = 3

• TOTAL SCORE = 12
Example: APC (Familial adenomatous polyposis)

- Severity: possible death due to cancer = 2
- Likelihood: high penetrance = 3
- Effectiveness of intervention: colonoscopy = 3
- Acceptability of intervention: colonoscopy = 2
- Knowledge base: high = 3

• TOTAL SCORE = 13
An age-based modified metric system

From Dr. Jonathan Berg
An age-based modified metric system

<table>
<thead>
<tr>
<th>Actionability</th>
<th>Onset</th>
</tr>
</thead>
<tbody>
<tr>
<td>NGS-NBS</td>
<td>Adult-onset medically actionable</td>
</tr>
<tr>
<td>PKU</td>
<td>Adult-onset medically actionable</td>
</tr>
<tr>
<td>FAP</td>
<td>Adult-onset non-medically actionable</td>
</tr>
<tr>
<td>MEN2B/RET</td>
<td>Adult-onset non-medically actionable</td>
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<tr>
<td>MCAD</td>
<td>Childhood onset non-medically actionable</td>
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<td>Duchenne MD</td>
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<td>Tay-Sachs</td>
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<td>Rett syndrome</td>
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<td>Retinitis pigmentosa</td>
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<td>Lynch syndrome</td>
<td>Childhood onset medically actionable</td>
</tr>
<tr>
<td>BRCA1</td>
<td>Childhood onset medically actionable</td>
</tr>
</tbody>
</table>

Infancy  Childhood  Adolescence  Adulthood  30+
NGS-NBS
Childhood medically actionable conditions

- Conditions currently on the recommended uniform screening panel (RUSP)
- Conditions that fit a similar profile to RUSP

ALGORITHM
- Severity of outcome
- Likelihood of severe outcome
- Efficacy of intervention
- Acceptability/burden of intervention
- Knowledge base
An age-based modified metric system
Affected cohorts (200)
Diagnosed Conditions
PKU, MCADD, CF, HL, LSD, ALD, PCD

Healthy newborn cohort (200)

Diagnostic results
Pathogenic variants and VUS

NGS-NBS Results: RUSP conditions and those determined by scoring process to meet criteria (childhood onset/medically actionable)
Pathogenic variants only

Control Group
(no additional results)

Decision Group

randomization

Using decision aid tool parents decide which additional categories of information to receive
Childhood-onset non-medically actionable, Adult-onset medically actionable, Carrier status
Pathogenic variants only

NCNeXus
North Carolina Newborn Exome Sequencing for Universal Screening
NGS-NBS
Childhood medically actionable conditions

Optional reporting based on parental decision-making

Reported to all participants

Findings that do not meet NGS-NBS criteria but may be of interest to some parents

Additional information

Excluded information
Adult onset non-medically actionable conditions

Not reported to any participants

Subject of randomized trial to assess parental preferences and potential psychosocial implications

Childhood onset NON-medically actionable

Adult onset medically actionable

Carrier status for recessive disorders
Next Gen-Newborn Screening?

Not as a stand-alone test
If genetic sequence information is not returned should it be stored? Where? Whose responsibility is it?
Child’s autonomy versus parental rights to child’s DNA sequence
How to recontact if conditions become treatable?
New gene/variant discoveries?
Commercial testing
Mandatory/voluntary? Health disparities
Demands on public health and health care systems
Genetic discrimination (employment, insurance,...)
Can Next-Gen Sequencing Expand the Utility of Newborn Screening?

• Test for additional conditions
• Improve specificity and sensitivity of standard screening
  – Cystic fibrosis
  – Hemoglobinopathies
  – Severe combined immunodeficiency
  – PKU
  – Fatty acid oxidation disorders
  – Urea cycle disorders
  – Hearing loss
NC NEXUS TEAM

Principal Investigators
• Cynthia Powell – PI and Project 2 PI
• Jonathan Berg – PI and Project 1 PI
• Don Bailey – Project 3 PI

Project Coordinator
• Laura Milko

Investigators
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• James Evans – Projects 1 and 3
• Megan Lewis – Project 3
• Piotr Mieczkowski – Project 1 (HTSF)
• George Retsch-Bogart – Project 2
• Christine Rini – Project 3/Aim 3
• Myra Roche – Projects 2 and 3
• Pat Roush – Project 2
• Neeta Vora – Project 2
• Karen Weck-Taylor – Project 1
• Kirk Wilhelmsen – Project 1
• Phillips Owen - RENCI
NC NEXUS TEAM

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