Prematurity as a Model for Neurodevelopmental Disorders: Development of Biomarkers for Diagnosis and Treatment Response

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Summary

• Neurodevelopmental disorders have many causes and phenotypes

• The causes and substrates of many disorders are poorly understood

• In addition, diagnosis of brain injury in prematures is often late and, therefore, treatments are difficult

• Development of effective treatments will depend on:
  – Identification of causative agents and affected developmental events/pathways
  – Identification of biomarkers that allow early diagnosis and early assessment of response to therapies
Extreme Prematurity as a Model for Many Issues in NICHD

- Pregnancy
- Genetic and Epigenetic Factors in Preterm Labor
- Genetic and Epigenetic Factors Related to Pre, Peri, and Postnatal Brain Injury, resulting in:
  - Sensorimotor Disorders
  - Visual Processing Disorders
  - Cognitive Disorders
  - Behavioral Disorders
- Effects upon Developing Brain
  - Animal Models
  - Direct observation via Imaging or Pathology
- Effects of Plasticity upon Outcome
Long Term Burden of Neurological Morbidity in VLBW Children

• 4.3 million annual births in United States
• 64,500 (1.5%) are very low birthweight (VLBW, less than 1500 gms at birth)
• 58,000 survive
• 29,000 (50%) have neurological, cognitive, or behavioral disorders
• 5,800 (25% of those with neuro-cognitive-behavioral disorders and 10% of survivors) have cerebral palsy
Neurodevelopmental Disorders due to Prematurity

• A surprisingly common disorder about which surprisingly little is known
• Early attention has been (appropriately) focused on pulmonary and cardiovascular support
• CNS issues often not detected until later in infancy or childhood
Delay to Diagnosis of CNS Complications of Premature Birth

- Severe brain damage (1%): minimal delay
  - Gray and White Matter Injury – extensive and grossly obvious
  - Correlates well with Gross Motor/Cognition/Vision/Hearing
  - Better Classified as Neonatal Encephalopathy/HIE

- Cavitary white matter injury leading to Cerebral Palsy or Visual Impairment (10%): months
  - Mainly white matter injury – ± grossly obvious, microscopically abnormal (GM involvement less obvious, may be secondary)
  - Correlates with Motor/Visual/Cognitive testing in infancy
  - Causes are not known, although some mechanisms understood
Evolution of white matter injury of prematurity: Cavitation and resorption
Delay to Diagnosis of Complications of Premature Birth

- Noncavitary cerebral injury or cerebellar hemorrhage or hypoplasia leading to behavioral or cognitive disabilities (50%): years
  - May be microscopic – no overt physical manifestations
  - Need special testing: coordination, perception, attention, communication

- Early diagnosis may allow earlier treatments and may improve results

- Treatments are aimed at symptoms as causes are not known
Mild White Matter Injury and small GMH (found in up to 40% of prematures)
Prevalence of macroscopic brain injuries

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<thead>
<tr>
<th>Condition</th>
<th>Grade 1/2</th>
<th>Grade 3/4</th>
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<tr>
<td>PVL</td>
<td>63%</td>
<td>8%</td>
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<tr>
<td>PVL grade 3/4</td>
<td>8%</td>
<td>3%</td>
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<tr>
<td>IVH</td>
<td>12%</td>
<td>5%</td>
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<tr>
<td>IVH grade 3/4</td>
<td>12%</td>
<td>5%</td>
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<tr>
<td>CH</td>
<td>7%</td>
<td>2%</td>
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<tr>
<td>CH grade 3/4</td>
<td>7%</td>
<td>2%</td>
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No Injury: 63%
## Macroscopic Brain Injuries

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<tr>
<th></th>
<th>No.</th>
<th>MDI Score (SD)</th>
<th>MDI&lt;70 No. (%)</th>
<th>PDI Score (SD)</th>
<th>PDI&lt;70 No. (%)</th>
<th>Cerebral palsy No. (%)</th>
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<td>49.3(18.5)</td>
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<td>82.6(13.5)</td>
<td>3(21.4)</td>
<td>85.1(11.5)</td>
<td>1(7.1)</td>
<td>3(21.4)</td>
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<td>85.7(20.5)</td>
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<td>86.2(18.3)</td>
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<tr>
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<td>95.3(12.2)</td>
<td>0</td>
<td>1(25.0)</td>
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<tr>
<td>Grade 1</td>
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<td>84.3(15.6)</td>
<td>4(26.7)</td>
<td>87.4(17.6)</td>
<td>3(20.0)</td>
<td>2(13.3)</td>
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<tr>
<td>No injury</td>
<td>220</td>
<td>86.4(17.9)</td>
<td>28(13.5)</td>
<td>89.4(15.3)</td>
<td>21(10.1)</td>
<td>10(4.5)</td>
</tr>
</tbody>
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*Courtesy Dr. Terri Inder*
Can Neurodevelopmental Sequela of Prematurity be Prevented/Treated?

- **Premature Labor: Dr. Hankins’ talk**
  - What initiates preterm labor?
    - Preterm injury
    - External stimuli/events
    - Can it be reversed?

- **Pre- or Perinatal Injury (?)**
  - General assumption: premature brain injury is acquired postnatally. However brain injury can be seen in first few days after birth on early MRI
  - When/why does injury occur?
  - Importance of Inflammation/Inflammatory markers
    - Amniotic fluid inflammation? IL-6-174 genotype?
  - Disruption of axon regeneration by hyaluronan or chondroitin sulfate?
Can Neurodevelopmental Sequelae of Prematurity be Prevented?

• Pre- or Perinatal Injury (?)
  – How early can inflammation or prenatal brain injury be detected?
  – If detected, can it be prevented or treated?
    • Steroids, Melatonin, Caffeine, Intervention in Signaling Pathways, Stem Cells
  – Will treatment cause other problems?

• Postnatal Brain Injury – May be preventable
  – Immaturity of Cardiovascular System → ischemia?
  – Immaturity of Lungs → hypoxia?
  – Immaturity of White Matter?
  – Infection/inflammation?
  – Iatrogenic Factors?
Prevention/Early Rx

• Major difficulty: initiating factors not known
• Cardiovascular instability has impaired early brain imaging; most done at discharge
• Severity of injury difficult to quantify, makes correlation with outcome difficult
  – What brain areas do we assess for correlation?
• Difficulty with assessment of therapies: many signs/symptoms are not detected until early childhood or school age – Need Early Dx
Effects of Pre/Perinatal Events upon Brain Development

• Animal Models
  – Hampered by different embryology/anatomy
    • Different neurogenesis/migration of GABAergic interneurons → larger subventricular zone
    • Different sets of neuronal molecular markers
    • Different gene expression patterns
    • Many structures of the human brain, notably in the prefrontal cortex and the perisylvian areas related to speech and language processing are rudimentary or absent in animal models. Subplate is larger and matures more slowly. Outer cortical layers are thicker.
  – OK for motor deficits, major mechanisms of injury but most animal models cannot explain cognitive/behavioral disorders in humans
### Outcome Assessment

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<tr>
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<th>Overall mental health</th>
<th>ADHD</th>
<th>ASD</th>
<th>Anxiety</th>
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<td>Brown ADD Scales&lt;sup&gt;5,6&lt;/sup&gt; (P/T)</td>
<td>CAST&lt;sup&gt;8,9&lt;/sup&gt; (4-11y)</td>
<td>SCARED&lt;sup&gt;11,12&lt;/sup&gt;</td>
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<td></td>
<td>ASEBA&lt;sup&gt;2&lt;/sup&gt;: CBCL, TRF</td>
<td>Brown ADD Scales (P/T)</td>
<td>SRS&lt;sup&gt;10&lt;/sup&gt;</td>
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<td></td>
<td>ASEBA&lt;sup&gt;3&lt;/sup&gt; (CBCL, TRF) SDQ</td>
<td>Brown ADD Scales (S/P/T)</td>
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<td>ASEBA&lt;sup&gt;3&lt;/sup&gt; (YSR, CBCL, TRF) SDQ</td>
<td>ADHD-Rating Scale IV&lt;sup&gt;7&lt;/sup&gt; (S/P/T)</td>
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<td></td>
<td>KSADS&lt;sup&gt;13&lt;/sup&gt;</td>
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<tr>
<td></td>
<td>Cognitive test (5-18y)</td>
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<tr>
<td><strong>Therapeutics</strong></td>
<td>Scarce research in preterm born children</td>
<td>Medication/structure/school interventions</td>
<td>Coping strategies</td>
<td>Cognitive behavior therapy</td>
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<tr>
<td></td>
<td>Cogmed JM/RM&lt;sup&gt;TM&lt;/sup&gt;??</td>
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<tr>
<td></td>
<td>Genetics?</td>
<td>Other?</td>
<td>Other?</td>
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</tr>
</tbody>
</table>

**Pre-school age 0-5 y**  
**School age 5-10 y**  
**Adolescence 11/12-18 y**

Courtesy Dr. Marit Indredavik
Behavior/Cognition - Autism

• Problems with definitions of ASD
  – Medical co-morbidities common (epilepsy, Tourette)
  – ASD is heterogeneous in expression (wide range of phenotypes) and, probably, in cause. Therefore, signs in DSM show poor specificity.
  – Psychopharmacology helps some symptoms (anxiety, irritability, impulsivity) but not the core sx of social reciprocity or communication

• Gold Standards: ADI-R and ADOS

• The diagnosis and subsequent Rx would be helped by development of objective biomarkers
Autism Spectrum Disorders

• Too large a topic, too heterogeneous to seriously address (Ecker et al, J Neurosci 2010, Kumar et al, Cereb Cortex 2010)

• New Approach: Atypical language lateralization in premies in later life (Ment et al, Lancet Neurol 2009) suggests framing the question in terms of sensory input and processing

• Investigations using functional and anatomic connectivity, deformation based morphometry, and MEG are currently underway to investigate these
Modifiers of Outcome

• Plasticity
  – Usually assessed by neurodevelopmental testing; how has function recovered?
  – Direct assessment by MRI (functional and connectivity measurements).


  • **Cortical thickness** (Taubert et al, J Neurosci 2010)

  • **Connectivity** measures increase with increasing FA
Mild White Matter Injury and small GMH Quantifiable and potentially treatable?
To date, nearly all imaging research of brain injury has been qualitative.

It has not been demonstrated whether quantification of brain structural and microstructural injury parameters, serving as biomakers, can allow early diagnosis and assess effects of therapy.
Does Preterm Birth by itself cause Abnormal White Matter Development?
Bonifacio et al, J Peds (2010)

• Gestational Age as a linear variable had no effect on diffusion parameters
  – FA increased by 0.008 per week
  – Dav decreased by 0.021 mm²/sec per week
• Birth at <26 weeks associated with lower FA, but effect eliminated when co-morbid conditions (NEC, infection, etc) were considered
• Mod-Severe brain injury was associated with decreasing WM FA (-0.012/wk) p=0.002
• Brain maturation was not disturbed by prematurity; injury and co-morbid conditions appeared to be the important determinants of microstructure maturation.
Concept

- MRI can provide biomarkers that can be used for early detection and quantification of WM injury in preterm neonates.
- Early detection may facilitate identification of causative factors.
Fetal Imaging

• Subtle developmental anomalies can now be detected and quantified by fetal imaging

• Real time fetal imaging → fetal neurological exams, possibly fetal behavior? (Prayer et al, 2009)

• Can better assessment of fetus give clues to prenatal processes – behavior, genetic, destructive?
Early Neonatal Assessment

• New Imaging Methods may help to better quantify brain injury and responses to therapy
  – Diffusion Tractography to identify specific pathways
  – Proton MR Spectroscopy
  – Functional Connectivity MR imaging
  – Structural Connectivity MR imaging

• Without knowledge of causes or of mechanisms, therapy is not likely to be successful

• Early diagnosis of brain damage and the effects on brain function, however, is necessary to determine need for and assess response to therapy

• This will require early, safe and powerful neonatal brain assessments.
Findings for Normative Preterm Infants

Neonatal 3D-MRSI - Premature Infant

T2-weighted MRI of premature infant with suspected subependymal hemorrhage.
Functional Connectivity MRI (fcMRI)

Increasing connectivity with maturation

26 wks  30 wks  34 wks  38 wks  Term Control

Motor Hand

Post Cingulate Cortex

Cerebellum

Courtesy Dr. R. McKinstry
Functional Connectivity MRI

Premature birth impairs connectivity

Courtesy Dr. R. McKinstry
Summary

• Neurodevelopmental disorders have many causes and phenotypes

• The causes and substrates of many disorders are poorly understood

• In prematures, diagnosis of brain injury is often late and, therefore, treatments are difficult

• Development of effective treatments will depend on:
  – Identification of affected metabolic and signaling pathways
  – Identification of biomarkers that allow early diagnosis and early assessment of response to therapies