Acute Kidney Injury in Neonates: From premature ‘arrested development’ to ‘breaking bad’ outcomes

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Disclosure

• I have no relevant financial relationships to disclose or conflicts of interest to resolve.

Objectives

1. To describe the development of the kidney from gestation through premature birth
2. To define and explain acute kidney injury (AKI) in neonates
3. To analyze the risk of developing chronic kidney disease following premature birth and AKI

Care in the NICU is improving

• Mortality is decreasing
• Limits of viability decreasing
• Youngest survivors just now reaching 30 to 40 years old
  ▪ What chronic health problems are we creating?
    ▪ Chronic Kidney Disease

Chronic kidney disease

• 10% of the population worldwide
  ▫ 20 million patients

• High associated costs
  ▫ > 40 billion/year

• Increased risk of:
  ▫ CV events
  ▫ Stroke
  ▫ Death

When do nephrons develop?

- Nephrogenesis begins at 9 weeks in utero
  - Complete by 36 weeks
  - 60% of nephrons formed in third trimester

Nephrogenesis is complete at 36 weeks in term newborns

Kidney Development: The megalin story

Nephron loss and Renal Function

- 4,500 glomeruli lost per year after age 60

Do premature infants develop nephrons after birth?

- Autopsy study of 56 premature infants < 1000 grams
  - No evidence of new nephrons (defined as ‘basophilic S shaped bodies’) after 40 days of life independent of birth gestational age
  - This may mean that those born < 30 weeks stop forming nephrons prior to 36 weeks leading to ‘oligonephropathy’

Are nephrons normal when they develop after birth?

- Sutherland et al 2011
  - 28 post mortem kidneys from premature neonates
  - 32 kidneys still-birth gestational aged matched controls
  - Findings
    - 13% Morphologically abnormal glomeruli
    - Larger renal corpuscle, increased volume
      - Hyperfiltration?
      - Accelerated maturation?

Premature nephrons are abnormal

Brenner Hypothesis

- A congenital or acquired reduction in nephron number could explain individual’s susceptibility to hypertension and CKD
  - Nephrons can maintain a normal GFR by enlarging, or ‘hyperfiltering’
    - Sodium retention  \rightarrow \text{ Systemic hypertension}
    - Intraglomerular hypertension  \rightarrow \text{ Proteinuria}
    - Sclerosis  \rightarrow \text{ CKD}

Adaptive response of hyperfiltration is harmful


Nephrons do not regenerate

Charlton, J. Pediatr Nephrol, 2014

Prematurity and Proteinuria

- Premature neonates
  - Increased rates of proteinuria
  - 3 times increased risk of chronic kidney disease

Megalin

- Endocytic receptor
  - Proximal tubule
  - Placenta
  - Yolk sack
  - Ciliary epithelium
  - Choroid plexus

- Expression and function unknown in premature neonates
  - Rat studies show developmental regulation

Megalin Ligands

- Albumin
- Angiotensin II
- Cystatin C
- EGF and IGF-I
- Gentamicin
- Insulin
- Lactoferrin
- NGAL
- Prolactin
- Retinol Binding Protein
- Thyroglobulin
- Vitamin D Binding Protein

Retinol Binding Protein

- Binds vitamin A in a 1:1 ratio
  - RBP a surrogate for Vitamin A status

- Critical for development of kidney
  - Animal Studies → Deficiency results in decreased nephron numbers

- Critical for development of lungs
  - Human Studies → Supplementation with Vitamin A results in reduced CLD

Hypothesis

- Premature infants lose RBP in their urine due to a lack of megalin expression/function which is developmentally regulated
**Methods**

**Part A: Urine analysis from NICU patients gestational age 28-40 weeks**

**Urinary RBP**

- **< 33 Weeks**
- **33-35 Weeks**
- **38-40 Weeks**

*36-37 Weeks Excluded*

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**Methods**

**Part B: Post-mortem kidney analysis from fetal and premature neonates**

- **Immunohistochemistry:**
  - Proximal Tubule
  - Megalin
  - Retinol Binding Protein

- **Microscopic Analysis**
  - 24 week fetal kidney stained with megalin
  - Highlighted for counting

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**Megalin by Gestational Age**

<table>
<thead>
<tr>
<th>Gestational Age Range</th>
<th>Percent Megalin</th>
</tr>
</thead>
<tbody>
<tr>
<td>20-23</td>
<td>2</td>
</tr>
<tr>
<td>24-26</td>
<td>4</td>
</tr>
<tr>
<td>26-29</td>
<td>6</td>
</tr>
<tr>
<td>30-34</td>
<td>8</td>
</tr>
<tr>
<td>35-38</td>
<td>10</td>
</tr>
<tr>
<td>39-40</td>
<td>12</td>
</tr>
</tbody>
</table>

R = 0.99, p < 0.01

---

**Megalin vs. Tissue Retinol Binding Protein**

- **R² = 0.8455**
- **20-23 Weeks**
- **24-28 Weeks**
- **29-32 Weeks**
- **33-35 Weeks**
- **36-40 Weeks**
Immunofluorescence

Conclusions on Megalin
- Megalin is present as early as 20 weeks
  - Increases in the proximal tubule with maturation
- Urinary RBP is elevated in the most premature neonates
  - Decreases in the urine with maturation
- Urinary losses of RBP may lead to a compromised ability to deliver vitamin A to the developing kidneys and lungs

Future Directions
- Urinary RBP may serve as a biomarker for proximal tubular maturation and megalin expression in premature human neonates
- Focus on the potential that premature neonates may require supplementation of megalin specific ligands
  - Retinol Binding Protein
  - Vitamin D Binding Protein
  - Other significant ‘NICU’ factors

Acute Kidney Injury: Another role for Caffeine?

Acute Kidney Injury
- Definitions
  - Initially Acute Renal Failure
    - Creatinine > 1.5 mg/dL
    - pRIFLE in 2002
    - AKIN in 2004
    - KDIGO in 2012 – changes over 7 days

<table>
<thead>
<tr>
<th>Stage</th>
<th>Serum Creatinine</th>
<th>Urine Output</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>1.5-1.9 times baseline ≥ 0.3 mg/dL increase (48h)</td>
<td>&lt; 0.5 mL/kg/hr for 6 h</td>
</tr>
<tr>
<td>2</td>
<td>2.0-2.9 times baseline</td>
<td>&lt; 0.5 mL/kg/hr for 12 h</td>
</tr>
<tr>
<td>3</td>
<td>≥ 3 times baseline ≥ 2.5 mg/dL increase</td>
<td>&lt; 0.3 mL/kg/hr for 24 h Anuria for 12h</td>
</tr>
</tbody>
</table>
Acute Kidney Injury

- **Incidence**
  - Viswanathan et al, 2012
    - Definition: Creatinine ≥ 1.5 or UOP < 1 mL/kg/hr
    - 12.7% of ELBW infants (n = 472) developed AKI
  - Koralkar et al, 2011
    - AKIN criteria
      - 12% of VLBW infants (n = 229) developed AKI
      - Increased mortality for AKI group after adjusting for confounders
  - Carmody et al, 2014
    - KDIGO criteria
      - 35% of VLBW infants (n = 455) developed AKI
      - No infants referred for follow-up with a nephrologist
  - Weintraub et al, 2016
    - AKIN criteria
      - 30.3% of infants < 30 weeks
    - Of all AKI, stage 1 represented 72%

Incidence of Neonatal AKI ranges from 12.5-40%

NICU Exposures

- **Nephrotoxic medications**
  - 107 VLBW infants April 2011 to March 2012
  - Exposure to ≥ 1 nephrotoxin in 87% of infants
    - Gentamicin 86%, Indomethacin 43%, Vancomycin 25%
  - Inverse relationship between birth weight and medications received per day
  - Infants with AKI received more nephrotoxins than those who did not have AKI

Nephrotoxin exposure is common in the NICU and is associated with increased rates of AKI

Prevention of AKI

- **Limit Nephrotoxin exposure**
  - Monitor levels at appropriate intervals
  - Antibiotic stewardship
  - Monitor urine output
  - Monitor renal function closely

- **What if there is a medication we already give that can prevent AKI?**
  - Theophylline has been used to prevent AKI in babies with HIE

The benefits of caffeine

- Methyloxanthines have been used in the NICU for decades
  - Caffeine is now the most common
- Caffeine has the potential to protect the kidney
  - Blocks A1 and A2A adenosine receptors
  - Decrease renal vasoconstriction
  - Decrease tubuloglomerular feedback in response to ischemia
- Is Caffeine the ‘silver bullet’ for Neonatology?
  - Improves outcomes of premature neonates
  - Dosing, timing and duration of therapy needs to be further studied

Goal

- We sought to determine whether early exposure to caffeine within the first 7 days after birth reduced the incidence of AKI among VLBW infants in our NICU
Methods

- Retrospective Chart Review
- Inclusion criteria
  - Birth weight ≤1500 grams
  - Admitted March-December 2011
- Exclusion criteria
  - Those admitted >2 days old
  - Death during hospitalization
- Caffeine exposure was defined as having received caffeine in the first 7 days after birth

AKI Staging

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<th>Serum Creatinine</th>
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Demographics

<table>
<thead>
<tr>
<th></th>
<th>Caffeine</th>
<th>No Caffeine</th>
<th>p-Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patients (#)</td>
<td>63</td>
<td>20</td>
<td></td>
</tr>
<tr>
<td>Gestational Age (Wks)</td>
<td>27.4</td>
<td>28.7</td>
<td>0.226</td>
</tr>
<tr>
<td>Birth Weight (kg)</td>
<td>1.05</td>
<td>0.95</td>
<td>0.248</td>
</tr>
<tr>
<td>SGA (#)</td>
<td>5</td>
<td>12</td>
<td>0.001</td>
</tr>
<tr>
<td>Length of stay (days)</td>
<td>67</td>
<td>70</td>
<td>0.808</td>
</tr>
</tbody>
</table>

Caffeine vs AKI

<table>
<thead>
<tr>
<th></th>
<th>No AKI</th>
<th>AKI</th>
<th>Total</th>
</tr>
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<tbody>
<tr>
<td>No Caffeine</td>
<td>10</td>
<td>10</td>
<td>20</td>
</tr>
<tr>
<td>Caffeine</td>
<td>51</td>
<td>12</td>
<td>63</td>
</tr>
<tr>
<td>Total</td>
<td>61</td>
<td>22</td>
<td>83</td>
</tr>
</tbody>
</table>

- No caffeine = 10/20 (50%) experienced AKI
- Caffeine = 12/63 (19%) experienced AKI
- Caffeine is associated with less frequent AKI
  (p = 0.006, chi square)

Logistic Regression Models

<table>
<thead>
<tr>
<th>Variables</th>
<th>AKI Odds Ratio</th>
<th>95% CI</th>
<th>P-Value</th>
</tr>
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<tbody>
<tr>
<td>Gestational Age + Birth Weight</td>
<td>7.66</td>
<td>1.3 - 46.7</td>
<td>0.027</td>
</tr>
<tr>
<td>Gestational Age + Birthweight + SGA</td>
<td>8.44</td>
<td>1.1 - 62.6</td>
<td>0.037</td>
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Illness Severity Score

- CRIB II
  - Gestational age, birth weight, gender
  - Base excess and admission temperature

<table>
<thead>
<tr>
<th>Variable</th>
<th>Number of Patients</th>
<th>Mean CRIB II Score</th>
<th>Median CRIB II Score</th>
</tr>
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<tbody>
<tr>
<td>Caffeine + AKI</td>
<td>10</td>
<td>11.08</td>
<td>10</td>
</tr>
<tr>
<td>No Caffeine + AKI</td>
<td>12</td>
<td>13</td>
<td>14.5</td>
</tr>
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- Mann Whitney Test, P-value of 0.171
Limitations

- Retrospective
- Limited numbers
- Confounding

Conclusions

- There is an association between early caffeine and prevention of AKI in VLBW infants
  - Not receiving caffeine is associated with a 7.6 times increase in the odds of developing AKI
  - Only need to treat 2.7 infants with caffeine to prevent one case of AKI

Next Steps

- Analysis of a large multicenter study
- PAS abstract

<table>
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<th>No MX (n=684)</th>
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<td>AKI 133 (%)</td>
<td>47 (9.5%)</td>
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Preterm AKI → CKD: the FANCY study

Linking prematurity to CKD

- Animal Studies
  - Mice born premature develop a CKD phenotype of 5 weeks of age - hypertension, albuminuria and reduced nephron number
  - Baboons born premature continue to develop nephrons postnatally, but similar to humans have histologic abnormalities

Animal studies suggest early development of CKD following premature birth

- Reduced nephron number (Oligonephropathy)
  - Hyperfiltration
  - Systemic Hypertension
  - Proteinuria
  - Glomerulosclerosis
  - Nephron loss
  - Chronic Kidney Disease

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Evidence of chronic injury

• Human Studies
  – LBW infants have 70% increase in rates of adult CKD
  – Series of 6 former premature patients (gestational age 22-30) with secondary FSGS at an average age of 32
  – CKID (Chronic Kidney Disease in Children study)
    • 426 participants
      – 17% LBW
      – 14% SGA
      – 12% Premature
      – 40% NICU after delivery
  
  LBW, SGA and Prematurity are all CKD risk factors

Does AKI affect rates of CKD in premature neonates?

• Pediatric ICU study
  – Prospective cohort study evaluating long term renal outcomes of AKI in the PICU
  – 13 of 126 patients developed CKD within 1-3 years
  – 59 of 126 at risk for CKD
    • At risk defined as:
      – Mildly decreased GFR, 60-90 mL/min/1.73 m²
      – Hyperfiltration

  CKD occurs within 1-3 years after AKI

CKD rates in former preterm infants

• 64 children born with ELBW, 36 term
  – Renal ultrasound showed significantly smaller renal volume at 7 and 11 years of age
  – Serum cystatin C values higher in ELBW children

  CKD in former preterm infants presents in childhood with decreased GFR and small kidneys

CKD rates in former preterm infants with history of AKI

• 222 total ELBW infants included
  – 49% AKI rate (Stage 1 = 39%, Stage 2/3 = 10%)
  – Findings:
    • More diastolic hypertension at discharge for stage 2/3
    • At >2 years of age
      – 4% had eGFR < 90
      – 5% had HTN

  CKD in former preterm infants with history of AKI presents in childhood with HTN, decreased GFR or proteinuria

Masquod et al. Outcome of extremely low birth weight infants with a history of neonatal acute kidney injury. 2017
The FANCY Study

Follow-up of AKI in Neonates during Childhood Years

- **Hypothesis**
  - Infants born less than 1500 grams (VLBW) who experience any stage AKI in the NICU will signs of chronic kidney disease at the ages of 2-6

Outcomes

- **Primary Outcomes**
  - Renal Dysfunction
    - Estimated GFR
    - Serum creatinine
    - Serum cystatin C
  - Proteinuria
  - High blood pressure

Cystatin C is 100% filtered and is not secreted like creatinine and it is less affected by age, gender, body size, and smoking making it a better marker.

Study Criteria

- **Inclusion criteria**
  - Premature birth: gestational age of 23 1/7 to 35 6/7 weeks
  - NICU stay at the University of Virginia
  - Birth weight less than 1500 grams
  - Signed and witnessed consent
  - Willing to return for a follow-up visit

- **Exclusion criteria**
  - Patients with Congenital Anomalies of the Kidney and Urinary Tract

Methods

- **Recruitment phone call**
  - Background for study
  - Explain clinic visit — “Kidney Screening”

- **Goal patients = 25-40 patients**
  - Power analysis
    - Using the standard deviations from the preliminary data
    - Mann-Whitney test has 80% power, with a 2-sided significance level of 5% to detect a GFR difference of 23

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Results

- Median age of 5 years
- Overall, 26% of the cohort had an eGFR less than 90 mL/min/1.73m² using cystatin C
  - 65% of AKI group had ‘renal dysfunction’
  - 14% of group without AKI had ‘renal dysfunction’
  - Relative risk of 4.5 (p<0.01)

Next Steps

- Analysis of Urine for biomarker identification
- See poster outside!

Conclusions

- Megalin Project
  - Premature birth leads to reduced and abnormal nephron development
- Caffeine Project
  - NICU exposures are significant and can lead to increased rates of AKI
- FANCY Project
  - AKI is not reversible and can lead to the early development of CKD
Neonatal Kidney Collaborative (NKC)

25 International Medical Centers
– Team of Neonatologists and Nephrologists from each Center

Goals:
1. Develop a standardized evidenced based definition of neonatal AKI
2. Improve understanding of risk factors for neonatal AKI
3. Evaluate how fluid and electrolytes affect clinical outcomes
4. Enhance understanding of the association between neonatal AKI and long term renal outcomes

Thank You

• University of Virginia
• Dr. Charlton and her lab
• Mentors: Dr. Sinkin, Dr. Swanson, Dr. Fairchild
• Dr. Ryan McAdams, Neonatology Division Chief at UW

References