Advances in Neonatal Acute Kidney Injury

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Abbreviations: AKI: Acute kidney injury, aOR: Adjusted odds ratio, AUC: Area under the curve, AWAKEN: Assessment of Worldwide Acute Kidney Injury Epidemiology in Neonates, CI: confidence interval, CRRT: Continuous renal replacement therapy, CHD: congenital heart disease, CKD: Chronic kidney disease, CysC: Cystatin C, ECMO: extracorporeal membrane oxygenation, ELBW: extremely low birth weight, ELGANS: extremely low gestational age neonates, ESKD: End-Stage Kidney Disease, GA: gestational age, GFR: glomerular filtration rate, HIE: hypoxic ischemic encephalopathy, IQR: Interquartile range, KDIGO: Kidney Diseases: Improving Global Outcomes, NEC: necrotizing enterocolitis, NGAL: Neutrophil gelatinase-associated lipocalin, NICU: Neonatal Intensive Care Unit, NIH: National Institutes of Health, NINJA: Nephrotoxic Injury Negated by Just-in-Time Action, NIDDK: National Institute of Diabetes and Digestive and Kidney Diseases, NIRS: Near InfraRed Spectroscopy, NSAIDs: non-steroidal anti-inflammatory drugs, OR: odds radio, PDA: patent ductus arteriosus, PENUT: Preterm Erythropoietin Neuroprotection Trial, PD: Peritoneal Dialysis, RCT: Randomized clinical trial, $RrSO_2$: Renal tissue oxygenation, RRT: Renal Replacement Therapy, SCr: serum creatinine, UOP: Urine output, VLBW: very low birth weight infants.

Table of Contents Summary: Acute kidney injury in neonates is an evolving field with quickly expanding research. In this review, changes over the past five years are highlighted.

Contributors' Statement Page

Drs Starr and Harer conceptualized and designed this review, drafted the initial manuscript, and reviewed and revised the manuscript.

Drs Charlton and Selewski conceptualized and designed this review, coordinated and provided oversight, and reviewed and revised the manuscript.

Drs Guillet, Reidy, Tipple, Jetton, Kent, Abitbol, Ambalavanan, Mhanna, and Askenazi provided substantial acquisition and assimilation of the data, drafted sections of the manuscript, and critically revised the manuscript.

All authors approved the final manuscript as submitted and agree to be accountable for all aspects of the work.

Abstract

In this state-of-the-art review, we highlight the major advances over the last five years in neonatal acute kidney injury (AKI.) Large multi-center studies show that neonatal AKI is common and independently associated with increased morbidity and mortality. The natural course of neonatal AKI along with the risk factors, mitigation strategies, and the role of AKI on short and long-term outcomes is becoming clearer. Specific progress has been made in identifying potential preventative strategies for AKI like the use of caffeine in premature neonates, theophylline in neonates with hypoxic-ischemic encephalopathy and nephrotoxic medication monitoring programs. New evidence highlights the importance of the kidney in 'crosstalk' between other organs and how AKI likely plays a critical role in other organ development and injury like intraventricular hemorrhage and lung disease. New technology has resulted in advancement in prevention and improvements in the current management in neonates with severe AKI. With specific CRRT machines designed for neonates, this therapy is now available and being used with increasing frequency in NICUs. Moving forward, biomarkers like urinary NGAL and other new technologies such as monitoring of renal tissue oxygenation and nephron counting will likely play an increased role in identification of AKI and those most vulnerable for chronic kidney disease (CKD). Future research needs to focus on determining the optimal follow-up strategy for neonates with a history of AKI to detect CKD.

Introduction

Since the publication of the Neonatal Acute Kidney Injury review in 2015, our understanding of the epidemiology and impact of neonatal acute kidney injury (AKI) has exponentially increased.[1] Single and multicenter work has clearly shown that AKI occurs commonly in critically ill neonates and adversely impacts outcomes.[2-4] In parallel to these advancements, our ability to identify AKI early, mitigate AKI, and provide renal replacement therapy (RRT) with devices designed for neonates have improved.[5, 6] This state-of-the-art review will review neonatal kidney physiology, update neonatal AKI (definitions, prevalence, outcomes, and complications), discuss the current state of research, and appraise cutting edge data on therapeutics and devices that will improve care in the coming decade.

Neonatal Kidney Physiology and Implications for AKI

A basic understanding of kidney structure and function during development is essential to understand neonatal AKI and its consequences.[1] Nephrogenesis begins in the fifth week of gestation and continues until 34-36 weeks of gestation.[7] Nephron number is highly variable at birth, ranging from 200,000 to 2.7 million, and is impacted by a multitude of factors including prematurity.[8, 9]

There are several core principles of neonatal physiology that uniquely impact the diagnosis and management of neonatal AKI. First, both renal blood flow and perfusion pressure increase over the first weeks of life in neonates. The proportion of cardiac output delivered to the kidneys increases from 5% during fetal life to 20% by two years.[10] Much of this increased blood flow occurs following birth, with renal blood flow doubling in the first two postnatal weeks. Following birth, the distribution of blood flow transitions from deeper more mature glomeruli to superficial, cortical glomeruli.[11] This change in blood flow can be altered by medications, such as indomethacin, perinatal asphyxia, and maternal hemorrhage which predispose neonates to AKI. Second, congruent with the increased blood flow,

glomerular filtration rate (GFR) increases dramatically after birth and reaches adult levels by two years of age.[12] GFR is low in infants, both in absolute terms and corrected for body surface area (ml/min/1.73m2). For example, premature infants born at 26 weeks have a GFR as low as 0.7 ml/min/kg on day 1 of age which improves only slightly during the first several weeks of life.[12] In neonates with a physiologically low GFR, additional stressors such as sepsis, hypoxia, hypotension or other clinical conditions common in prematurity may increase the risk AKI. Third, urinary concentrating ability is low at birth and reaches adult levels by 1 year of age.[13] Poor urinary concentrating ability, particularly in neonates with high insensible losses or critical illness, predisposes neonates to volume depletion and subsequent pre-renal azotemia. Finally, neonatal kidneys appear to be particularly susceptible to ischemic injury to the renal tubules – even after a mild and short-term insult. This is further complicated when nephrotoxic medications, such as gentamicin and other aminoglycosides, are commonly prescribed to critically ill neonates and result in tubular injury.

Definitions of Acute Kidney Injury

The neonatal modified Kidney Disease Improving Global Outcomes (KDIGO) definition is the most commonly used definition utilized in clinical practice and most epidemiologic studies (**Table 1**). This empiric definition stages AKI severity based on a rise in serum creatinine (SCr) from a previous trough and/or a decrease in urine output (UOP).[14, 15] The NIH-sponsored Neonatal AKI Workshop in 2013 recommended that researchers and clinicians use the neonatal modified KDIGO definition to define AKI, but emphasized that this definition should be a starting point for an iterative process which is based on clinically meaningful and long-term outcomes.[16]

Since the publication of this definition, there have been several observational studies highlighting potential areas for future refinement of the definition to account for chronologic and

gestational age (GA). For example, while some researchers advocate for excluding SCr from the first 48 hours postnatally when calculating baseline SCr, others believe that this value is a surrogate of nephron number and clinically meaningful.[2, 3, 17-19] Researchers have also pointed out that deviations in the GA appropriate SCr trajectory (which steadily drops from birth in healthy term neonates) could signify AKI, but this not captured in the current KDIGO definition.[20]

The current neonatal modified KDIGO AKI definition incorporates UOP. Despite studies in other older populations that show UOP is critically important to properly identify AKI, few studies in neonatal AKI have included UOP. One study using diaper weights every 3 hours found that UOP <1.5 ml/kg/hr was associated with increased mortality.[21] Furthermore, they found that lower thresholds of UOP (< 1ml/kg/hr) were associated with an even greater mortality rate.[21] Few studies, including the Assessment of Worldwide Acute Kidney Injury Epidemiology in Neonates (AWAKEN) study, included UOP measurement in the assessment of AKI, thus limiting the available data upon which to base thresholds for diagnosis and determination of severity of AKI. Future studies should carefully measure UOP to determine the impact of UOP on AKI diagnosis in neonates.

Given the interest and focus on AKI diagnostic thresholds, it is very likely that a refined definition of neonatal AKI will emerge. Two recent publications, including an analysis of the AWAKEN study, suggest an alternative definition for neonatal AKI.[22, 23] This study proposed different cutoffs for the first postnatal week by GA compared to the subsequent weeks and would provide cutoffs by GA group.[24] For example, in infants ≤29 weeks GA a SCr rise of 0.6 mg/dL confers the highest prediction of mortality while in infants >29 weeks GA a rise of 0.3mg/dL is of highest mortality prediction.[24] Furthermore, we anticipate that novel approaches using urine biomarkers, SCr thresholds, UOP thresholds, and fluid balance metrics will be used to enhance the current neonatal KDIGO definition. In

the interim, we recommend that the neonatal modified KDIGO definition be used as the standard until newer definitions are widely validated in large multi-site trials and correlated with long-term outcomes.

Epidemiology, Risk Factors and Associated Findings with AKI

AKI is common in critically ill neonates. We present a summary of the risk factors associated with neonatal AKI (**Table 2**) and the most notable studies evaluating the epidemiology and impact of neonatal AKI in the last 5 years (**Table 3**). Much of this increased knowledge stems from the AWAKEN study, which enrolled neonates at risk of AKI (determined by >48 hours of intravenous fluids). In this cohort, the risk of AKI occurred in a bimodal pattern with extremely low gestational age neonates (ELGANS, <28 weeks) and term infants at the greatest risk.[25] The AWAKEN study identified a clear variation in SCr monitoring practices across centers with less than half of centers checking five or more SCrs during hospital admission. Not surprisingly, the rates of AKI by center were directly correlated with the average number of SCr ascertained per subject.[4] This practice variation is particularly notable in the context of the recent Baby NINJA (Nephrotoxic Injury Negated by Just-in-Time Action) publication showed that by monitoring SCr more frequently in neonates with high nephrotoxic medication exposure, there was an increased awareness of the risk for AKI that, in turn, resulted in a lower rate and duration of AKI.[17] This data suggest that critically ill neonates may benefit from protocolized SCr monitoring during high risk events.

Preterm Neonates

The risk of AKI increases markedly with decreasing GA.[2, 3] In cohorts of very low birth weight (VLBW) neonates, the incidence of AKI is reported between 18-40%.[3, 26] In ELGANS enrolled in the Preterm Erythropoietin Neuroprotection Trial (PENUT), 38% had at least one episode of AKI.[2] In the AWAKEN study, AKI occurred in 45% of <29 weeks GA and in 14% of 29-36 weeks GA neonates. In

AWAKEN, the potentially modifiable risk factors for early AKI in ELGANS were mainly medication exposures.[27] Given immature tubular function and resulting poor urinary concentration ability along with the increased insensible losses common in preterm neonates, volume depletion leading to pre-renal azotemia is a common factor predisposing this population to AKI.

Patent ductus arteriosus (PDA) is an important clinical issue for preterm infants and is associated with a higher risk of AKI.[28, 29] PDA represents a clinical challenge as AKI may result if it is left untreated; however, classic PDA therapies may also be associated with AKI. While NSAID treatment of PDA added an additional risk for mild AKI, severe AKI was less likely when NSAID treatment was effective.[28]

Full-term or Near Term Critically Ill Neonates

In the AWAKEN study the incidence of AKI in neonates born at ≥36 weeks and admitted to a NICU was 37%.[4] The etiology of AKI in term neonates is often multifactorial, and includes risk factors related to their illness and management (**Table 2**).[27, 30] Multi-organ dysfunction is common and occurs in up to 70% of neonates with AKI.[31, 32, 33] Some of the major risk factors for AKI include hypoxic ischemic encephalopathy (HIE), cardiac disease, surgery, and nephrotoxic medications.

Hypoxic-ischemic encephalopathy: There is a general agreement that the presence of AKI in the setting of HIE is associated with poor outcomes (increased mortality, poor neurodevelopmental outcomes, longer hospital stay and longer duration of mechanical ventilation).[32, 34, 35] In addition, there is a correlation between severity of HIE and AKI with 70% of Stage III HIE having AKI compared with 7.4% of those with Stage II HIE.[34]

Cardiac disease and ECMO: Infants who require cardiac surgery and those who need extracorporeal membrane oxygenation (ECMO) are at high risk for AKI. AKI occurs in 30-50% of patients undergoing surgery for congenital heart disease (CHD).[36-38] One single center retrospective

study of neonates with single ventricle physiology undergoing Stage 1 Norwood palliation found that 21% developed AKI.[38] A large Danish registry study showed 33% of neonates had AKI within 5 days of surgery.[37] In a multicenter retrospective cohort study of 832 pediatric patients on ECMO, 74% had AKI.[36] AKI was present at initiation of ECMO in the majority of cases, and was associated with a longer ECMO duration and increased mortality.[36] The risk of AKI in those on ECMO varies by underlying diagnosis, as those with congenital diaphragmatic hernia were more likely to require RRT.[39]

Surgery: The incidence of AKI is high in non-cardiac surgery, as 34% of neonates undergoing abdominal and thoracic surgery have an episode of AKI.[40] Infants with AKI after surgery were more likely to be VLBW. They were also more likely to have sepsis, a longer duration of mechanical ventilation, an operative time >120 minutes, necrotizing enterocolitis (NEC) and a higher risk of mortality.[40] Among infants with surgically managed NEC, almost 60% had severe AKI (Stage 2 or 3).[41]

Nephrotoxic medications: Many neonates are exposed to nephrotoxic medications in the NICU which can contribute to AKI. While there are many nephrotoxic medications, these primarily include antimicrobial agents (e.g. acyclovir, amphotericin B, aminoglycosides, vancomycin).[27, 30] Baby NINJA, a single center quality improvement program focused on reducing nephrotoxic medication associated AKI, recently reported that attention to high risk neonates, including daily SCr monitoring, reduces nephrotoxic medication exposures ($p=0.03$), nephrotoxic medication associated AKI ($p<0.001$), and AKI duration ($p<0.001$).[17] This suggests that identification and monitoring of high risk neonates, with a thoughtful consideration of nephrotoxic medications, may minimize AKI and its consequences in critically ill neonates.

Fluid Balance

The development of fluid overload is an independent predictor of adverse outcomes across pediatric critical care populations.[42-51] The development of fluid overload is often multifactorial resulting from AKI, iatrogenic fluid administration, capillary leak from systemic inflammation, and aberrant homeostatic mechanisms.[43] Additionally, fluid overload can make the diagnosis of AKI more challenging as SCr is diluted in the setting of a positive fluid balance, resulting in a potential under-diagnosis of AKI.[43] Precise definitions are essential to understand the epidemiology and impact of excessive fluid accumulation on outcomes in neonates (**Table 4**).[52] The two most common methods utilized to calculate fluid balance are the cumulative fluid balance and weight-based methods. The weight-based method to describe fluid balance calculates the degree of fluid overload based on a change in weight from a baseline weight (birthweight or determined dry weight):[53]

Daily Fluid Balance = Change in daily weight day to day

Cumulative Fluid Balance = Daily Weight – Baseline Weight

$$\text{Percent FO}^{44} = \frac{\text{Daily Weight} - \text{Baseline Weight}}{\text{Baseline Weight}} * 100$$

While each method has been utilized in older children, the weight-based methods represent the standard in neonates as fluid balances have been shown to be inaccurate.[53-55] It is critical to utilize standardized weight measurement protocols to properly perform the weight-based technique.[54, 56]

Fluid balance in the early postnatal period can be challenging to interpret in the setting of normal post-natal diuresis and expected negative fluid balance. While an average weight loss of 7% from birthweight is described in term neonates, the normal fluid balance for other GA neonates is less clearly defined, particularly in extremely preterm neonates with excessive skin permeability.[57] Early positive postnatal fluid balance is associated with adverse short-term (death, mechanical ventilation on day 7) and long-term outcomes (bronchopulmonary dysplasia) in neonates.[53, 55, 58-61] There remains a paucity of data defining the pathologic state of fluid overload in critically ill neonates, which in older children, this

is commonly defined as a cumulative positive fluid balance of ≥ 10-20%.[50, 62] Multiple critical gaps exist in our understanding of the causes and impact of abnormal fluid balance in neonates. This includes interpreting fluid balance in neonates, especially those that have spent a considerable time in the NICU. Research is greatly needed to understand the optimal threshold to define appropriate fluid balance and the detrimental effects of neonatal fluid overload on extra-renal organ systems (i.e. oxygenation index and cardiac dysfunction due to excessive fluid and reduced contractility). Understanding the role of fluid balance in various neonatal populations (premature vs. term neonates) at critical time-points (perinatal vs. postnatal vs. post-surgical) in the context of underlying disease process (NEC, lung disease, and sepsis) is a critical knowledge gap. Answers to these questions will drive therapeutic interventions designed to prevent and mitigate harm from fluids.

New Advances in AKI Research

Biomarkers of AKI

Proteins and metabolites are two examples of biomarkers that can be measured consistently and correlate with the disease occurrence or progression.[63] Given the diversity of GA and etiologies of AKI in neonates, one biomarker does not appear to reliably predict AKI. However, more reference ranges are becoming available for novel urinary biomarkers by GA and postnatal age.[64] Serum creatinine represents the current standard for diagnosing neonatal AKI. However, it is critical to understand the shortcomings of SCr develop novel biomarkers that fill these gaps. A rise in SCr indicates a loss of kidney function reflecting injury that occurred up to 48-72 hours prior.[65] As a result, biomarker studies such as the ones below have focused on identifying injury and functional changes prior to permanent damage (**Table 5**).

Cystatin C (CysC) is a cysteine protease inhibitor that is freely filtered by glomeruli and reabsorbed in the proximal tubule. While both serum and urinary CysC have been assessed as early

markers of AKI, a rise in serum CysC reflects a change in kidney function, while elevated urinary CysC is considered reflective of tubular injury.[66-71] A recent systematic review of neonates across all GA groups suggested that serum CysC may be superior to SCr for assessing GFR.[67] Furthermore, elevated urinary CysC has been shown to have an area under the curve (AUC) of 0.85 (95% CI 0.81-0.88) in predicting a rise in SCr 24-96 hours later in neonates post-surgery or following perinatal asphyxia.[69]

There are several other biomarkers which may help identify injury and functional changes prior to permanent damage in neonates with AKI. Neutrophil gelatinase-associated lipocalin (NGAL) is a protein bound to neutrophil granules, filtered by the glomerulus, and reabsorbed by the proximal tubules.[72] NGAL is highly sensitive (87-93%) and specific (87-93%) for AKI in neonates with perinatal asphyxia.[73] The combination of G1 cell cycle arrest urinary tissue inhibitor of metalloproteinase-1 (TIMP-2) and insulin-like growth factor binding protein (IGFBP-7) is a promising biomarker for detecting early AKI. It has been shown to have sensitivity of 89% and specificity of 51% in critically ill neonates for developing AKI.[74] This combination has been evaluated in pediatric and adult populations and have been shown to perform well in predicting subsequent severe AKI.[65] Both NGAL and TIMP-2/IGFBP-7 are currently used off-label in multiple pediatric and adult ICUs to evaluate for AKI and there are several ongoing studies assessing their utility in pediatric AKI.[75] Kidney Injury Molecule 1 (KIM-1) is a transmembrane protein that is upregulated in kidney injury, and elevated levels have been shown to predict AKI.[76] The AUC of several of these biomarkers – in particular NGAL and TIMP2-IGFBP7 are highly predictive for AKI, using SCr as the gold standard.[76] It is imperative that we begin to incorporate novel biomarkers into future definitions of neonatal AKI and clinical care.

Nephron Number

In humans, there is wide variability in nephron number present even in the neonatal period as the completion of nephrogenesis occurs at 34-26 weeks.[8, 9, 77] The mechanisms (*in utero* and *ex utero*) that

influence nephrogenesis, and ultimately nephron endowment, and the processes regulating nephron loss are poorly understood.[7] Preclinical advancements have been made in measuring whole kidney functional nephron number *in vivo* using a novel contrast agent. Cationic ferritin enhanced-MRI has been used to measure glomerular number and size in health and disease.[78, 79] Radial glomerular count, a surrogate marker of glomerulogenesis, suggests that nephron number is decreased in both premature neonates and in those with AKI.[80] Further work is necessary to translate this technique to humans, including neonates, to further understand those at risk for future CKD.[81]

Tissue oxygenation

Non-invasive continuous monitoring of renal oxygen saturation with Near InfraRed Spectroscopy (NIRS) is a new diagnostic tool that may lead to earlier diagnosis of AKI.[82] Renal tissue oxygenation ($RrSO_2$) monitoring is a surrogate for local tissue oxygen utilization. In neonates with CHD, NIRS monitoring of the kidney post-operatively can predict AKI.[83] In premature neonates, those that subsequently develop AKI have lower $RrSO_2$ in the first postnatal day or week.[84, 85] As in postoperative cardiac patients, NIRS detected a decline in $RrSO_2$ prior to AKI defined by SCr or UOP.[86] In those undergoing therapeutic hypothermia for HIE, neonates with AKI had higher $RrSO_2$ values, likely pointing to a different cause or type of injury than in preterm or infants with CHD.[87] Further work is needed to establish normative $RrSO_2$ values in neonatal populations and treatment guidelines incorporating $RrSO_2$ values.

Evaluation of Neonatal AKI

Evaluating a neonate that develops AKI requires a systematic approach, which includes consideration of common factors contributing to AKI. A detailed history should be obtained to assess for risk factors for AKI, including birth weight and GA, antenatal events (including prenatal ultrasounds),

pregnancy complications, birth history (interventions required at delivery), nephrotoxic medications exposure, and post-natal events. Physical examination should include an assessment of volume status, which should also include weight, daily fluid balance and cumulative fluid balance. Fluid balance assessment is essential, as volume depletion is a common cause of AKI and volume overload is a common complication of AKI. Positive fluid balance still may mean poor renal perfusion if neonates have ongoing third spacing due to capillary leak. Maintaining euvolemia is a challenging but essential management strategy in infants both to prevent AKI and to mitigate severe volume overload and complications. Focused laboratory evaluation should be performed including serum electrolytes, blood urea nitrogen, and SCr and/or CystC for GFR assessment.[67] At this time, there is not a definitive role for urine biomarker assessment in all neonates, but growing data suggests that it may be useful in certain clinical settings to predict AKI.[74] Fractional excretion of sodium (FeNa) may be helpful in some infants in differentiating volume depletion from intrinsic causes of AKI, but can be challenging to interpret in premature infants due to tubular immaturity.[12] We recommend an ultrasound be obtained to evaluate for evidence of obstruction as well as congenital abnormalities of the kidney. An ultrasound can also determine kidney size. However, more studies are needed to know if kidney size is helpful in understanding clinically kidney-related meaningful outcomes.[88]

Management of Neonatal AKI

While the search for treatments or interventions for established neonatal AKI have remained elusive, medications have been evaluated in high-risk neonatal cohorts to prevent AKI. There have been multiple therapeutics evaluated for AKI prevention in neonates without positive results including erythropoietin, therapeutic hypothermia, remote ischemic preconditioning and corticosteroids.[89-91]

Methylxanthines have been evaluated in multiple neonatal populations and shown promise as a preventative treatment for AKI in high-risk populations.[92, 93]

After diagnosis of AKI, careful management of fluid balance and medications are essential to prevent the development of complications. Strict documentation of all fluid input and output, along with daily weights, are essential to optimize fluid balance. Nephrotoxic medications should be assessed daily and reduced or eliminated whenever possible.[94] Cumulative fluid balance should be carefully monitored to assess overall volume status. Infants with volume depletion may require additional fluid either in the form of enteral feeding, intravenous boluses or drips. In infants with volume overload, diuretics can be trialed to maintain urine output.[95] Response to furosemide (furosemide stress test) has been used as a functional biomarker for predicting severe AKI.[96] While not evaluated in all neonatal populations, term infants with CHD with a lower response to furosemide (median UOP at 2 hours after furosemide 1.2 versus 3.4 mL/kg/hour, $p = 0.01$) have an increased risk for persistent AKI. While furosemide responsiveness is a potential functional marker of kidney status, more studies in neonates are needed to standardize the dose and definitions used.

Theophylline and its related salt, aminophylline, have shown success in increasing UOP and may prevent AKI in infants with HIE. Theophylline is an adenosine receptor antagonist that prevents AKI by inhibiting adenosine-induced renal vasoconstriction. A recent meta-analysis of seven randomized controlled trials (458 asphyxiated neonates not receiving therapeutic hypothermia) found that theophylline administration was associated with a significantly lower incidence of AKI (OR: 0.24, 95% CI: [0.16, 0.36]).[93] Based on this evidence, a single dose of theophylline within the first six postnatal hours in newborns with HIE is endorsed in the 2012 KDIGO guidelines to prevent AKI.[15] Aminophylline has also shown promise as a rescue therapy in neonates with AKI treated with therapeutic hypothermia.[97]

Caffeine is also an adenosine receptor antagonist that has been evaluated for reno-protective effects in preterm cohorts. Two studies have shown that AKI occurred less frequently in VLBW and preterm infants <33 weeks GA who received caffeine within the first postnatal week. In a retrospective study of 140 VLBW neonates[98], AKI occurred less frequently in those who received caffeine (17.8% vs. 43.6% ; p=0.002). In a secondary analysis of the AWAKEN study, AKI occurred less frequently in neonates <33 weeks GA who received caffeine in the first postnatal week (11.2% vs. 31.6%, p<0.01).[92] Based on these data, the number needed to be exposed to caffeine to prevent one episode of AKI is 4.3.[92]

Renal Replacement Therapy

The primary therapy for the complications of severe AKI remains RRT. The indications for RRT in neonates include acidosis, fluid overload, electrolyte abnormalities, and uremia refractory to medical management. The two most common modalities for RRT in neonates are peritoneal dialysis (PD) and continuous renal replacement therapy (CRRT). CRRT can be added to the extra-corporeal circuit in infants receiving Extracorporeal Membrane Oxygenation (ECMO) therapy. Between PD and CRRT, the choice often depends on the available resources, center experience, and patient characteristics.[99] These therapies are complementary in that some neonates or situations will have a higher chance of success using PD while others would benefit from CRRT. PD remains a common first choice for RRT in most institutions as it does not require vascular access, is more available, and is often technically easier in the smallest patients.[99] PD can be performed using a temporary catheter if the RRT requirement is thought to be short-term. Depending on the catheter utilized, PD can be successful in neonates as small as 830 grams.[100]

CRRT may be preferred in hemodynamically unstable infants, those with history of abdominal surgery or NEC which make PD technically challenging, or those in whom tight control of volume

status is necessary. In the past, the sole availability of CRRT equipment designed for adults and larger children presented a challenge to perform CRRT in neonates. This necessitated the use for larger catheters, tubing, and filters, resulting in high extracorporeal volumes and greater hemodynamic instability often requiring either blood transfusions or blood priming with each circuit change. Recent advances have made CRRT increasingly accessible and successful for neonates. Introduction of smaller filters, such as the HF-20 (total extracorporeal volume of 60 mL) that was recently approved by the FDA for use in the United States, has improved both the availability and acceptance of therapies by decreasing the extracorporeal volume and improving fluid removal precision.[101]

In recent years, industry has made significant innovations in neonatal RRT by developing neonatal specific CRRT machines and repurposing machines for neonatal use.[102] CARPEDIEM (Cardio-Renal Pediatric Dialysis Emergency Machine, Medtronic, Minneapolis, MN) was approved for use in children between 2.5 and 8 kg the United States in 2020 and has been used in multiple countries outside of the United States. This machine was developed specifically for neonates and small children.[5] In contrast, Aquadex (Aquadex FlexFlow, CHF Solutions, Eden Prairie, MN) is an ultrafiltration device designed for adults, but with an extracorporeal volume of 33mL making it ideal to adapt to safely provide CRRT to infants.[5] Successful CRRT using Aquadex has been reported in infants as small as 1.4 kg.[5] These machines, and others such as NIDUS (Newcastle Infant Dialysis and Ultrafiltration System, The Newcastle NHS Foundation Trust, UK), have begun to revolutionize the field of CRRT for neonates by lowering the associated risks, and will change the conversation about when and in whom to initiate CRRT. [6, 103]

Complications of AKI

Cross talk between AKI and Other Organs

AKI has been shown to adversely impact other organs.[104] Initially thought to be only an association, studies suggest a causal relationship in which AKI appears to drive other organ dysfunction, and vice-versa, referred to as "crosstalk".[105] Experimental models describe a lung focused inflammatory process, driven in part by cytokines such as IL-6 and 8, following AKI which is deleterious to the lungs.[106] Both preterm and full-term infants with AKI have worse lung outcomes than their peers without AKI including longer durations of mechanical ventilation and higher rates of bronchopulmonary dysplasia.[107, 108] Neonatal AKI has been shown to be an independent risk factor for neurologic complications like IVH, poor long-term neurocognitive outcomes, and cardiovascular disease (hypertension).[109, 110]

Risk of CKD following AKI

The risk of developing chronic kidney disease (CKD) or end stage kidney disease (ESKD) after AKI is well detailed in adults.[111] The evidence of progression from AKI to CKD is less established in children with AKI. In a systematic review of 346 children (mean follow-up 6.5 years), the incidence of abnormal GFR <90 mL/min/1.73m^2 was 6.3% (CI 5.1-7.5).[112] The mechanisms for progression to CKD are incompletely understood, but likely are secondary to maladaptive repair, ongoing inflammation, and disordered regeneration.[113, 114] Histologic findings of preterm neonates show abnormal glomeruli likely to develop sclerosis later which could be the explanation for later CKD in those with AKI.[115] These changes may be superimposed upon a decreased nephron number and reduction in future development of nephrons due to prematurity.[116]

The evidence for progression from AKI to CKD in neonates is less clear. Several studies have identified evidence of kidney abnormalities in preterm infants with a history of AKI.[117, 118] In contrast, other studies failed to identify differences in CKD or GFR in follow-up of preterm infants who had AKI as neonates.[89, 119, 120] The lack of appropriately powered studies, consensus definitions for AKI and

CKD, and a consistent follow-up period are barriers to clearly defining the relationship between neonatal AKI and subsequent CKD. While the Chronic Kidney Disease in Children (CKiD) study follows children with CKD and includes information on birth weight, it does not include detailed data on neonatal course and AKI. Large multi-center long-term follow-up studies of neonates following AKI are needed in order to completely understand the future risk of CKD.

Conclusion

Dramatic advances in the diagnosis and epidemiology of neonatal AKI and our ability to care for neonates with kidney disease have occurred in the last decade.[121] New technologies and therapies designed to prevent and treat neonatal AKI augment these findings. Future work, including interventional trials of therapeutics to treat AKI (methylxanthines, RRT with novel devices), prospective long-term follow-up studies to understand risk factors for CKD development, and improved definitions of fluid overload are needed. Additionally, continuing to integrate biomarkers into routine clinical use, more widespread availability and use of neonatal specific extracorporeal devices for kidney support therapy and standardization of monitoring and follow-up of neonates with AKI will continue to advance the field of neonatal AKI.[122, 123] Ongoing collaboration between neonatologists, pediatricians and nephrologists (including the Neonatal Kidney Collaborative www.babykidney.org) will help drive these research initiatives, mentor young faculty, educate clinicians, inform families, and advocate for neonates at risk for short and long-term kidney-related disease.[102, 124]

Table 1. Neonatal AKI KDIGO Classification

Stage	**SCr**	**Urine Output**
0	No change in SCr or rise <0.3 mg/dL	≥ 0.5 mL/kg/h
1	Increase in SCr of ≥0.3 mg/dL within 48 h or SCr rise ≥1.5–1.9 X reference SCr[a] within 7 d	< 0.5 ml/kg/h for 6 to 12 h
2	SCr rise ≥ 2–2.9 X reference SCr[a] within 7 d	< 0.5 ml/kg/hour for ≥12 h
3	SCr ≥ 3 X reference SCr[a] or SCr > 2.5 mg/dL[b] or Receipt of RRT	< 0.3 ml/kg/hour for ≥24 hours or anuria for ≥12 hours

Differences between the neonatal AKI definition and KDIGO include the following:
a Reference SCr defined as the lowest previous SCr value
b SCr value of 2.5mg/dL represents <10ml/min/1.73m2

Table 2. Epidemiology, Risk Factors and Associated Findings with neonatal AKI

Prenatal	**Perinatal**	**Postnatal**
Factors that increase risk of preterm or LBW neonate Placental insufficiency	Exposure to nephrotoxic medications (ACE inhibitors, NSAIDs) Delivery complications, resulting in hypoxia and/or asphyxia Hypoxic ischemic encephalopathy	Prematurity Low birth weight Congenital Heart Disease Inborn errors of metabolism Sepsis Nephrotoxin exposure PDA Extracorporeal therapies

Table 3. Summary of Neonatal AKI Studies Published between 2015 – 2020.

Author, Year (N)	**Study design / Population Details**	**Main Findings**
Jetton (2017) [4] (n= 2162)	Neonates requiring >48h of intravenous fluids in the NICU	AKI occurred in 45% of the infants 28 w GA or less, and in 14% of infants at 29-36 w GA.
Charlton (2019) [27, 30] (n= 2162)	Secondary analysis of infants from the AWAKEN study	Antimicrobial agents, methylxanthines, diuretics, NSAIDs, hypertensive disorders of pregnancy, and hypoglycemia were associated with lower odds of early AKI. In 29-25 w GA infants, outborn status, saline bolus during resuscitation, and more frequent SCr monitoring associated with higher odds of early AKI
Askenazi (2000) [2] (n= 923)	Prospective study of ELGANS in the PENUT trial	38% had at least one episode of stage 1 or higher AKI and 18.2% had one episode of stage 2 or higher AKI.
Selewski (2019) [53] (n= 645)	Retrospective analysis of infants ≥36 w GA from the AWAKEN study	Median peak fluid balance was 1.0% (IQR: -0.5, 4.6) over the first postnatal week and occurred on postnatal day 3 (IQR: 1, 5). Mechanical Ventilation on postnatal day 7 was associated independently with the following measures of fluid balance over the first postnatal week: • Peak fluid balance (aOR 1.12, 95% CI [1.08-1.17]) • Lowest fluid balance in 1st postnatal week (aOR 1.14, 95% CI [1.07-1.22]) • Fluid balance on postnatal day 7 (aOR 1.12, 95% CI 1.07-1.17) • Negative fluid balance at postnatal day 7 (aOR 0.3, 95%CI 0.16-0.67)
Selewski (2020) [55] (n=1007)	Retrospective analysis of infants <36 w GA from the AWAKEN study	Median peak FB was 0% (IQR: -2.9, 2) and occurred on postnatal day 2 (IQR: 1,5). Mechanical Ventilation on postnatal day 7 was associated independently with the following measures of fluid balance over the first postnatal week: • Peak fluid balance (aOR 1.14, 95% CI [1.10-1.19]) • Lowest fluid balance (aOR 1.12, 95% CI [1.07-1.16]) • Fluid balance on postnatal day 7 (aOR 1.10, 95% CI [1.06-1.13]) • Negative FB at postnatal day 7 protected against the need for MV at postnatal day 7 (aOR 0.21, 95% CI [0.12-0.35])
Nour (2020) [125] (n=30)	Retrospective analysis in neonates with HIE who underwent cooling using selective head cooling	No difference in AKI between the those receiving selective head cooling and those not. SCr and UOP were significantly improved on day-4 and day-10 samples compared to base-line samples in both groups regardless of cooling. There was a difference in NGAL, but not cystatin C on days 4 and 10.
Bellos (2019)[93] (n=458)	Meta-analysis of effectiveness of	Incidence of AKI significantly lower in neonates receiving theophylline (OR: 0.24, 95% CI: [0.16, 0.36]), while mortality

	theophylline administration in neonates with perinatal asphyxia	rates were similar between the two groups (OR: 0.86, 95% CI: [0.46, 1.62]). Theophylline administration was associated with significantly decreased SCr (MD: -0.57 mg/dl, 95% CI: [-0.68, -0.46]) in the 3rd day of life.
Harer (2018) [92] (n=675)	Retrospective analysis of premature infants <33 w GA from the AWAKEN study.	AKI occurred less frequently in neonates who received caffeine in the first week of life than in those who were not treated with caffeine (11.2% vs. 31.6%, P< .01). Neonates who received caffeine had more AKI risk factors including lower average GA, lower birth weight, and high severity of illness scores and were still less likely to develop stage 2 or 3 AKI. Number needed to treat with caffeine to prevent one episode of AKI is 4.3.
Stoops (2019)[17]	Prospective quality improvement effort to reduce Nephrotoxic Medication-Associated Acute Kidney Injury in the Neonatal Intensive Care Unit	Reduced high nephrotoxic medication exposures from 16.4 to 9.6 per 1000 patient-days (P = .03), reduced nephrotoxic medication-AKI from 30.9% to 11.0% (P < .001), and reduced AKI severity from 9.1 to 2.9 per 100 susceptible patient-days (P < .001). This prevented 100 AKI episodes during the 18-month sustainability era.
Starr (2019)[107] (n=546)	Retrospective analysis of premature infants <32 w GA from the AWAKEN study	Infants born between 29 and 32 w GA with AKI had four-fold higher odds of moderate or severe BPD/death that remained after controlling for multiple factors (aOR: 4.21, 95% CI: [2.07–8.61]).

Table 4. Definitions of excessive fluid accumulation in neonates

Term	**Definition**
Daily fluid balance	Daily difference between input and output or change in weight over a 24-hour period.
Cumulative fluid balance	Change in fluid balance over a given duration.
Fluid overload	Cumulative fluid balance expressed as a percent of body weight. Utilized to refer to the pathologic state of excessive fluid accumulation associated with the development of sequelae attributable to fluid accumulation and adverse outcomes.

Table 5. Biomarkers in use or under study to assist with AKI prediction and diagnosis

Biomarker	**Location of Injury**	**Notes**
Serum Creatinine, SCr	Reflects kidney function, not injury Metabolic product of skeletal muscle creatine	Delayed marker (48-72 hours) of kidney function
Cystatin C, CysC	Reflects kidney function, not injury (serum) Proximal tubule injury (urine)	Increase in serum CysC is thought to reflect a change in GFR or kidney function, while elevated urinary CysC is considered reflective of tubular injury.
Neutrophil gelatinase-associated lipocalin, NGAL	Distal tubule and collecting duct	Highly sensitive for ischemia, and nephrotoxins
Kidney Injury Molecule, KIM-1	Proximal tubule injury	Regulates apoptosis, promotes epithelial regeneration
Tissue inhibitor of Metalloproteinases-2 (TIMP-2) and insulin-like growth factor binding protein (IGFBP-7)	Tubule cells	Limits proliferation of damaged tubule cells Marker of early AKI

References

1. Selewski DT, Charlton JR, Jetton JG, Guillet R, Mhanna MJ, Askenazi DJ, et al. Neonatal Acute Kidney Injury. *Pediatrics*. 2015;136(2):e463-473.
2. Askenazi DJ, Heagerty PJ, Schmicker RH, Griffin R, Brophy P, Juul SE, et al. Prevalence of acute kidney injury (AKI) in extremely low gestational age neonates (ELGAN). *Pediatr Nephrol*. 2020;35(9):1737-1748.
3. Carmody JB, Swanson JR, Rhone ET, Charlton JR. Recognition and reporting of AKI in very low birth weight infants. *Clin J Am Soc Nephrol*. 2014;9(12):2036-2043.
4. Jetton JG, Boohaker LJ, Sethi SK, Wazir S, Rohatgi S, Soranno DE, et al. Incidence and outcomes of neonatal acute kidney injury (AWAKEN): a multicentre, multinational, observational cohort study. *Lancet Child Adolesc Health*. 2017;1(3):184-194.
5. Menon S, Broderick J, Munshi R, Dill L, DePaoli B, Fathallah-Shaykh S, et al. Kidney Support in Children using an Ultrafiltration Device: A Multicenter, Retrospective Study. *Clin J Am Soc Nephrol*. 2019;14(10):1432-1440.
6. Vidal E, Garzotto F, Parolin M, Manenti C, Zanin A, Bellettato M, et al. Therapeutic Plasma Exchange in Neonates and Infants: Successful Use of a Miniaturized Machine. *Blood Purif*. 2017;44(2):100-105.
7. Chambers JM, Wingert RA. Advances in understanding vertebrate nephrogenesis. *Tissue Barriers*. 2020;8(4):1832844.
8. Chevalier RL. Evolutionary Nephrology. *Kidney Int Rep*. 2017;2(3):302-317.
9. Straub RH, Schradin C. Chronic inflammatory systemic diseases: An evolutionary trade-off between acutely beneficial but chronically harmful programs. *Evol Med Public Health*. 2016;2016(1):37-51.
10. Iacobelli S, Guignard JP. Maturation of glomerular filtration rate in neonates and infants: an overview. *Pediatr Nephrol*. 2020.
11. Hyink DP, Abrahamson DR. Origin of the glomerular vasculature in the developing kidney. *Semin Nephrol*. 1995;15(4):300-314.
12. Abitbol CL, Seeherunvong W, Galarza MG, Katsoufis C, Francoeur D, Defreitas M, et al. Neonatal kidney size and function in preterm infants: what is a true estimate of glomerular filtration rate? *J Pediatr*. 2014;164(5):1026-1031.e1022.
13. Su S, Stonestreet B. Core concepts: neonatal glomerular filtration. rate. *Neoreviews*. 2010;11:e714e721.
14. Jetton JG, Guillet R, Askenazi DJ, Dill L, Jacobs J, Kent AL, et al. Assessment of Worldwide Acute Kidney Injury Epidemiology in Neonates: Design of a Retrospective Cohort Study. *Front Pediatr*. 2016;4:68.
15. Kellum JA, Lameire N, Group KAGW. Diagnosis, evaluation, and management of acute kidney injury: a KDIGO summary (Part 1). *Crit Care*. 2013;17(1):204.
16. Zappitelli M, Ambalavanan N, Askenazi DJ, Moxey-Mims MM, Kimmel PL, Star RA, et al. Developing a neonatal acute kidney injury research definition: a report from the NIDDK neonatal AKI workshop. *Pediatr Res*. 2017;82(4):569-573.
17. Stoops C, Stone S, Evans E, Dill L, Henderson T, Griffin R, et al. Baby NINJA (Nephrotoxic Injury Negated by Just-in-Time Action): Reduction of Nephrotoxic Medication-Associated Acute Kidney Injury in the Neonatal Intensive Care Unit. *J Pediatr*. 2019;215:223-228 e226.
18. Thayyil S, Sheik S, Kempley ST, Sinha A. A gestation- and postnatal age-based reference chart for assessing renal function in extremely premature infants. *J Perinatol*. 2008;28(3):226-229.

19. Askenazi DJ, Ambalavanan N, Goldstein SL. Acute kidney injury in critically ill newborns: what do we know? What do we need to learn? *Pediatr Nephrol*. 2009;24(2):265-274.
20. Gupta C, Massaro AN, Ray PE. A new approach to define acute kidney injury in term newborns with hypoxic ischemic encephalopathy. *Pediatr Nephrol*. 2016;31(7):1167-1178.
21. Bezerra CT, Vaz Cunha LC, Liborio AB. Defining reduced urine output in neonatal ICU: importance for mortality and acute kidney injury classification. *Nephrol Dial Transplant*. 2013;28(4):901-909.
22. Bruel A, Rozé JC, Flamant C, Simeoni U, Roussey-Kesler G, Allain-Launay E. Critical serum creatinine values in very preterm newborns. *PLoS One*. 2013;8(12):e84892.
23. Askenazi D, Abitbol C, Boohaker L, Griffin R, Raina R, Dower J, et al. Optimizing the AKI definition during first postnatal week using Assessment of Worldwide Acute Kidney Injury Epidemiology in Neonates (AWAKEN) cohort. *Pediatr Res*. 2019;85(3):329-338.
24. Askenazi D, Abitbol C, Boohaker L, Griffin R, Raina R, Dower J, et al. Optimizing the AKI definition during first postnatal week using Assessment of Worldwide Acute Kidney Injury Epidemiology in Neonates (AWAKEN) cohort. *Pediatr Res*. 2019;85(3):329-338.
25. Jetton JG, Boohaker LJ, Sethi SK, Wazir S, Rohatgi S, Soranno DE, et al. Incidence and outcomes of neonatal acute kidney injury (AWAKEN): a multicentre, multinational, observational cohort study. *Lancet Child Adolesc Health*. 2017;1(3):184-194.
26. Koralkar R, Ambalavanan N, Levitan EB, McGwin G, Goldstein S, Askenazi D. Acute kidney injury reduces survival in very low birth weight infants. *Pediatr Res*. 2011;69(4):354-358.
27. Charlton JR, Boohaker L, Askenazi D, Brophy PD, D'Angio C, Fuloria M, et al. Incidence and Risk Factors of Early Onset Neonatal AKI. *Clin J Am Soc Nephrol*. 2019;14(2):184-195.
28. Majed B, Bateman DA, Uy N, Lin F. Patent ductus arteriosus is associated with acute kidney injury in the preterm infant. *Pediatr Nephrol*. 2019;34(6):1129-1139.
29. Guillet R, Selewski DT, Griffin R, Rastogi S, Askenazi DJ, D'Angio CT, et al. Relationship of patent ductus arteriosus management with neonatal AKI. *J Perinatol*. 2021.
30. Charlton JR, Boohaker L, Askenazi D, Brophy PD, Fuloria M, Gien J, et al. Late onset neonatal acute kidney injury: results from the AWAKEN Study. *Pediatr Res*. 2019;85(3):339-348.
31. Polglase GR, Ong T, Hillman NH. Cardiovascular Alterations and Multiorgan Dysfunction After Birth Asphyxia. *Clin Perinatol*. 2016;43(3):469-483.
32. Bozkurt O, Yucesoy E. Acute Kidney Injury in Neonates with Perinatal Asphyxia Receiving Therapeutic Hypothermia. *Am J Perinatol*. 2020.
33. Kirkley MJ, Boohaker L, Griffin R, Soranno DE, Gien J, Askenazi D, et al. Acute kidney injury in neonatal encephalopathy: an evaluation of the AWAKEN database. *Pediatr Nephrol*. 2019;34(1):169-176.
34. Michniewicz B, Al Saad SR, Karbowski LM, Gadzinowski J, Szymankiewicz M, Szpecht D. Organ Complications of Infants with Hypoxic Ischemic Encephalopathy Before Therapeutic Hypothermia. *Ther Hypothermia Temp Manag*. 2021;11(1):58-63.
35. Selewski DT, Jordan BK, Askenazi DJ, Dechert RE, Sarkar S. Acute kidney injury in asphyxiated newborns treated with therapeutic hypothermia. *J Pediatr*. 2013;162(4):725-729 e721.
36. Fleming GM, Sahay R, Zappitelli M, King E, Askenazi DJ, Bridges BC, et al. The Incidence of Acute Kidney Injury and Its Effect on Neonatal and Pediatric Extracorporeal Membrane Oxygenation Outcomes: A Multicenter Report From the Kidney Intervention During Extracorporeal Membrane Oxygenation Study Group. *Pediatr Crit Care Med*. 2016;17(12):1157-1169.

37. Madsen NL, Goldstein SL, Frøslev T, Christiansen CF, Olsen M. Cardiac surgery in patients with congenital heart disease is associated with acute kidney injury and the risk of chronic kidney disease. *Kidney Int*. 2017;92(3):751-756.
38. Wong JH, Selewski DT, Yu S, Leopold KE, Roberts KH, Donohue JE, et al. Severe Acute Kidney Injury Following Stage 1 Norwood Palliation: Effect on Outcomes and Risk of Severe Acute Kidney Injury at Subsequent Surgical Stages. *Pediatr Crit Care Med*. 2016;17(7):615-623.
39. Murphy HJ, Gien J, Sahay R, King E, Selewski DT, Bridges BC, et al. Acute Kidney Injury, Fluid Overload, and Renal Replacement Therapy Differ by Underlying Diagnosis in Neonatal Extracorporeal Support and Impact Mortality Disparately. *Blood Purif*. 2021:1-10.
40. Wu Y, Hua X, Yang G, Xiang B, Jiang X. Incidence, risk factors, and outcomes of acute kidney injury in neonates after surgical procedures. *Pediatr Nephrol*. 2020;35(7):1341-1346.
41. Garg PM, Britt AB, Ansari MAY, Sobisek S, Block DK, Paschal JL, et al. Severe acute kidney injury in neonates with necrotizing enterocolitis: risk factors and outcomes. *Pediatr Res*. 2021.
42. Flori HR, Church G, Liu KD, Gildengorin G, Matthay MA. Positive fluid balance is associated with higher mortality and prolonged mechanical ventilation in pediatric patients with acute lung injury. *Crit Care Res Pract*. 2011;2011:854142.
43. Gist KM, Selewski DT, Brinton J, Menon S, Goldstein SL, Basu RK. Assessment of the Independent and Synergistic Effects of Fluid Overload and Acute Kidney Injury on Outcomes of Critically Ill Children. *Pediatr Crit Care Med*. 2020;21(2):170-177.
44. Goldstein SL, Currier H, Graf C, Cosio CC, Brewer ED, Sachdeva R. Outcome in children receiving continuous venovenous hemofiltration. *Pediatrics*. 2001;107(6):1309-1312.
45. Gorga SM, Sahay RD, Askenazi DJ, Bridges BC, Cooper DS, Paden ML, et al. Fluid overload and fluid removal in pediatric patients on extracorporeal membrane oxygenation requiring continuous renal replacement therapy: a multicenter retrospective cohort study. *Pediatr Nephrol*. 2020;35(5):871-882.
46. Hassinger AB, Wald EL, Goodman DM. Early postoperative fluid overload precedes acute kidney injury and is associated with higher morbidity in pediatric cardiac surgery patients. *Pediatr Crit Care Med*. 2014;15(2):131-138.
47. Li Y, Wang J, Bai Z, Chen J, Wang X, Pan J, et al. Early fluid overload is associated with acute kidney injury and PICU mortality in critically ill children. *European journal of pediatrics*. 2016;175(1):39-48.
48. Mah KE, Hao S, Sutherland SM, Kwiatkowski DM, Axelrod DM, Almond CS, et al. Fluid overload independent of acute kidney injury predicts poor outcomes in neonates following congenital heart surgery. *Pediatr Nephrol*. 2018;33(3):511-520.
49. Selewski DT, Cornell TT, Lombel RM, Blatt NB, Han YY, Mottes T, et al. Weight-based determination of fluid overload status and mortality in pediatric intensive care unit patients requiring continuous renal replacement therapy. *Intensive Care Med*. 2011;37(7):1166-1173.
50. Sutherland SM, Zappitelli M, Alexander SR, Chua AN, Brophy PD, Bunchman TE, et al. Fluid overload and mortality in children receiving continuous renal replacement therapy: the prospective pediatric continuous renal replacement therapy registry. *American journal of kidney diseases : the official journal of the National Kidney Foundation*. 2010;55(2):316-325.
51. Valentine SL, Sapru A, Higgerson RA, Spinella PC, Flori HR, Graham DA, et al. Fluid balance in critically ill children with acute lung injury. *Critical care medicine*. 2012;40(10):2883-2889.
52. Hoste EA, Maitland K, Brudney CS, Mehta R, Vincent JL, Yates D, et al. Four phases of intravenous fluid therapy: a conceptual model. *Br J Anaesth*. 2014;113(5):740-747.

53. Selewski DT, Akcan-Arikan A, Bonachea EM, Gist KM, Goldstein SL, Hanna M, et al. The impact of fluid balance on outcomes in critically ill near-term/term neonates: a report from the AWAKEN study group. *Pediatr Res*. 2019;85(1):79-85.
54. van Asperen Y, Brand PL, Bekhof J. Reliability of the fluid balance in neonates. *Acta Paediatr*. 2012;101(5):479-483.
55. Selewski DT, Gist KM, Nathan AT, Goldstein SL, Boohaker LJ, Akcan-Arikan A, et al. The impact of fluid balance on outcomes in premature neonates: a report from the AWAKEN study group. *Pediatr Res*. 2020;87(3):550-557.
56. Bontant T, Matrot B, Abdoul H, Aizenfisz S, Naudin J, Jones P, et al. Assessing fluid balance in critically ill pediatric patients. *European journal of pediatrics*. 2015;174(1):133-137.
57. Paul IM, Schaefer EW, Miller JR, Kuzniewicz MW, Li SX, Walsh EM, et al. Weight Change Nomograms for the First Month After Birth. *Pediatrics*. United States: © 2016 by the American Academy of Pediatrics.; 2016.
58. Wadhawan R, Oh W, Perritt R, Laptook AR, Poole K, Wright LL, et al. Association between early postnatal weight loss and death or BPD in small and appropriate for gestational age extremely low-birth-weight infants. *J Perinatol*. 2007;27(6):359-364.
59. Schmidt B, Roberts RS, Fanaroff A, Davis P, Kirpalani HM, Nwaesei C, et al. Indomethacin prophylaxis, patent ductus arteriosus, and the risk of bronchopulmonary dysplasia: further analyses from the Trial of Indomethacin Prophylaxis in Preterms (TIPP). *J Pediatr*. 2006;148(6):730-734.
60. Oh W, Poindexter BB, Perritt R, Lemons JA, Bauer CR, Ehrenkranz RA, et al. Association between fluid intake and weight loss during the first ten days of life and risk of bronchopulmonary dysplasia in extremely low birth weight infants. *J Pediatr*. 2005;147(6):786-790.
61. Askenazi DJ, Koralkar R, Hundley HE, Montesanti A, Patil N, Ambalavanan N. Fluid overload and mortality are associated with acute kidney injury in sick near-term/term neonate. *Pediatr Nephrol*. 2013;28(4):661-666.
62. Selewski DT, Goldstein SL. The role of fluid overload in the prediction of outcome in acute kidney injury. *Pediatr Nephrol*. 2018;33(1):13-24.
63. Askenazi DJ, Koralkar R, Patil N, Halloran B, Ambalavanan N, Griffin R. Acute Kidney Injury Urine Biomarkers in Very Low-Birth-Weight Infants. *Clin J Am Soc Nephrol*. 2016;11(9):1527-1535.
64. DeFreitas MJ, Seeherunvong W, Katsoufis CP, RamachandraRao S, Duara S, Yasin S, et al. Longitudinal patterns of urine biomarkers in infants across gestational ages. *Pediatr Nephrol*. 2016;31(7):1179-1188.
65. Wasung ME, Chawla LS, Madero M. Biomarkers of renal function, which and when? *Clin Chim Acta*. 2015;438:350-357.
66. Chowdhary V, Vajpeyajula R, Jain M, Maqsood S, Raina R, Kumar D, et al. Comparison of different definitions of acute kidney injury in extremely low birth weight infants. *Clin Exp Nephrol*. 2018;22(1):117-125.
67. Renganathan A, Warner BB, Tarr PI, Dharnidharka VR. The progression of serum cystatin C concentrations within the first month of life after preterm birth-a worldwide systematic review. *Pediatr Nephrol*. 2020.
68. Yang Y, Li SJ, Pan JJ, Hu YH, Zhou XY, Cheng R, et al. Reference values for serum cystatin C in very low-birthweight infants: From two centres of China. *J Paediatr Child Health*. 2018;54(3):284-288.

69. Nakashima T, Inoue H, Fujiyoshi J, Matsumoto N. Longitudinal analysis of serum cystatin C for estimating the glomerular filtration rate in preterm infants. *Pediatr Nephrol*. 2016;31(6):983-989.
70. Treiber M, Pečovnik Balon B, Gorenjak M. A new serum cystatin C formula for estimating glomerular filtration rate in newborns. *Pediatr Nephrol*. 2015;30(8):1297-1305.
71. Abitbol CL, Seeherunvong W, Galarza MG, Katsoufis C, Francoeur D, Defreitas M, et al. Neonatal kidney size and function in preterm infants: what is a true estimate of glomerular filtration rate? *J Pediatr*. 2014;164(5):1026-1031 e1022.
72. Devarajan P. Neutrophil gelatinase-associated lipocalin: a promising biomarker for human acute kidney injury. *Biomark Med*. 2010;4(2):265-280.
73. Tanigasalam V, Bhat V, Adhisivam B, Sridhar MG. Does therapeutic hypothermia reduce acute kidney injury among term neonates with perinatal asphyxia?--a randomized controlled trial. *J Matern Fetal Neonatal Med*. 2016;29(15):2545-2548.
74. Chen J, Sun Y, Wang S, Dai X, Huang H, Bai Z, et al. The effectiveness of urinary TIMP-2 and IGFBP-7 in predicting acute kidney injury in critically ill neonates. *Pediatr Res*. 2020;87(6):1052-1059.
75. Fan W, Ankawi G, Zhang J, Digvijay K, Giavarina D, Yin Y, et al. Current understanding and future directions in the application of TIMP-2 and IGFBP7 in AKI clinical practice. *Clin Chem Lab Med*. 2019;57(5):567-576.
76. Stojanovic VD, Barisic NA, Vuckovic NM, Doronjski AD, Peco Antic AE. Urinary kidney injury molecule-1 rapid test predicts acute kidney injury in extremely low-birth-weight neonates. *Pediatr Res*. 2015;78(4):430-435.
77. Charlton JR, Baldelomar EJ, Hyatt DM, Bennett KM. Nephron number and its determinants: a 2020 update. *Pediatr Nephrol*. 2021;36(4):797-807.
78. Baldelomar EJ, Charlton JR, deRonde KA, Bennett KM. In vivo measurements of kidney glomerular number and size in healthy and Os(/+) mice using MRI. *Am J Physiol Renal Physiol*. 2019;317(4):F865-f873.
79. Baldelomar EJ, Charlton JR, Beeman SC, Bennett KM. Measuring rat kidney glomerular number and size in vivo with MRI. *Am J Physiol Renal Physiol*. 2018;314(3):F399-f406.
80. Rodríguez MM, Gómez AH, Abitbol CL, Chandar JJ, Duara S, Zilleruelo GE. Histomorphometric analysis of postnatal glomerulogenesis in extremely preterm infants. *Pediatr Dev Pathol*. 2004;7(1):17-25.
81. Charlton JR, Baldelomar EJ, deRonde KA, Cathro HP, Charlton NP, Criswell SJ, et al. Nephron loss detected by MRI following neonatal acute kidney injury in rabbits. *Pediatr Res*. 2020;87(7):1185-1192.
82. Harer MW, Chock VY. Renal Tissue Oxygenation Monitoring-An Opportunity to Improve Kidney Outcomes in the Vulnerable Neonatal Population. *Front Pediatr*. 2020;8(241):241.
83. Hazle MA, Gajarski RJ, Aiyagari R, Yu S, Abraham A, Donohue J, et al. Urinary biomarkers and renal near-infrared spectroscopy predict intensive care unit outcomes after cardiac surgery in infants younger than 6 months of age. *J Thorac Cardiovasc Surg*. 2013;146(4):861-867 e861.
84. Bonsante F, Ramful D, Binquet C, Samperiz S, Daniel S, Gouyon JB, et al. Low Renal Oxygen Saturation at Near-Infrared Spectroscopy on the First Day of Life Is Associated with Developing Acute Kidney Injury in Very Preterm Infants. *Neonatology*. 2019;115(3):198-204.
85. Dorum BA, Ozkan H, Cetinkaya M, Koksal N. Regional oxygen saturation and acute kidney injury in premature infants. *Pediatr Int*. 2021;63(3):290-294.

86. Harer MW, Adegboro CO, Richard LJ, McAdams RM. Non-invasive continuous renal tissue oxygenation monitoring to identify preterm neonates at risk for acute kidney injury. *Pediatr Nephrol*. 2021.
87. Chock VY, Frymoyer A, Yeh CG, Van Meurs KP. Renal Saturation and Acute Kidney Injury in Neonates with Hypoxic Ischemic Encephalopathy Undergoing Therapeutic Hypothermia. *J Pediatr*. 2018;200:232-239.e231.
88. Sanderson K, Chang E, Bjornstad E, Hogan S, Hu Y, Askenazi D, et al. Albuminuria, Hypertension, and Reduced Kidney Volumes in Adolescents Born Extremely Premature. *Front Pediatr*. 2020.
89. Askenazi DJ, Heagerty PJ, Schmicker RH, Brophy P, Juul SE, Goldstein SL, et al. The Impact of Erythropoietin on Short and Long-term Kidney-Related Outcomes in Extremely Low Gestational Age Neonates. Results of a Multi-center Double-Blind Placebo-Controlled Randomized Clinical Trial. *J Pediatr*. 2021.
90. Pedersen KR, Ravn HB, Povlsen JV, Schmidt MR, Erlandsen EJ, Hjortdal VE. Failure of remote ischemic preconditioning to reduce the risk of postoperative acute kidney injury in children undergoing operation for complex congenital heart disease: a randomized single-center study. *J Thorac Cardiovasc Surg*. 2012;143(3):576-583.
91. Lomivorotov V, Kornilov I, Boboshko V, Shmyrev V, Bondarenko I, Soynov I, et al. Effect of Intraoperative Dexamethasone on Major Complications and Mortality Among Infants Undergoing Cardiac Surgery: The DECISION Randomized Clinical Trial. *Jama*. 2020;323(24):2485-2492.
92. Harer MW, Askenazi DJ, Boohaker LJ, Carmody JB, Griffin RL, Guillet R, et al. Association Between Early Caffeine Citrate Administration and Risk of Acute Kidney Injury in Preterm Neonates: Results From the AWAKEN Study. *JAMA Pediatr*. 2018;172(6):e180322.
93. Bellos I, Pandita A, Yachha M. Effectiveness of theophylline administration in neonates with perinatal asphyxia: a meta-analysis. *J Matern Fetal Neonatal Med*. 2019:1-9.
94. Stoops C, Stone S, Evans E, Dill L, Henderson T, Griffin R, et al. Baby NINJA (Nephrotoxic Injury Negated by Just-in-Time Action): Reduction of Nephrotoxic Medication-Associated Acute Kidney Injury in the Neonatal Intensive Care Unit. *J Pediatr*. 2019;215:223-228.e226.
95. Mohamed TH, Klamer B, Mahan JD, Spencer JD, Slaughter JL. Diuretic therapy and acute kidney injury in preterm neonates and infants. *Pediatr Nephrol*. 2021.
96. Kakajiwala A, Kim JY, Hughes JZ, Costarino A, Ferguson J, Gaynor JW, et al. Lack of Furosemide Responsiveness Predicts Acute Kidney Injury in Infants After Cardiac Surgery. *Ann Thorac Surg*. 2017;104(4):1388-1394.
97. Chock VY, Cho SH, Frymoyer A. Aminophylline for renal protection in neonatal hypoxic-ischemic encephalopathy in the era of therapeutic hypothermia. *Pediatr Res*. 2021;89(4):974-980.
98. Carmody JB, Harer MW, Denotti AR, Swanson JR, Charlton JR. Caffeine Exposure and Risk of Acute Kidney Injury in a Retrospective Cohort of Very Low Birth Weight Neonates. *J Pediatr*. 2016;172:63-68 e61.
99. Kaddourah A, Goldstein SL. Renal replacement therapy in neonates. *Clin Perinatol*. 2014;41(3):517-527.
100. Harshman LA, Muff-Luett M, Neuberger ML, Dagle JM, Shilyansky J, Nester CM, et al. Peritonal Dialysis in an extremely low-birth weight infant with acute kidney injury. *Clin Kidney J*. 2014;7:582-585.

101. Munshi R, Lee-Son K, Hackbarth RM, Quigley R, Sutherland SM, Echeverri J, et al. Clinical evaluation of the Prismaflex HF 20 set and Prismaflex system 7.10 for acute continuous kidney replacement therapy (CKRT) in children. *Pediatr Nephrol*. 2020;35(12):2345-2352.
102. Askenazi DJ. AWAKEN-Ing a New Frontier in Neonatal Nephrology. *Front Pediatr*. 2020;8:21.
103. Garzotto F, Vidal E, Ricci Z, Paglialonga F, Giordano M, Laforgia N, et al. Continuous kidney replacement therapy in critically ill neonates and infants: a retrospective analysis of clinical results with a dedicated device. *Pediatr Nephrol*. 2020;35(9):1699-1705.
104. Faubel S, Shah PB. Immediate Consequences of Acute Kidney Injury: The Impact of Traditional and Nontraditional Complications on Mortality in Acute Kidney Injury. *Adv Chronic Kidney Dis*. 2016;23(3):179-185.
105. Basu RK, Wheeler DS. Kidney-lung cross-talk and acute kidney injury. *Pediatr Nephrol*. 2013;28(12):2239-2248.
106. Grigoryev DN, Liu M, Hassoun HT, Cheadle C, Barnes KC, Rabb H. The local and systemic inflammatory transcriptome after acute kidney injury. *J Am Soc Nephrol*. 2008;19(3):547-558.
107. Starr MC, Boohaker L, Eldredge LC, Menon S, Griffin R, Mayock DE, et al. Acute Kidney Injury and Bronchopulmonary Dysplasia in Premature Neonates Born Less than 32 Weeks' Gestation. *Am J Perinatol*. 2020;37(3):341-348.
108. Starr MC, Boohaker L, Eldredge LC, Menon S, Griffin R, Mayock D, et al. Acute Kidney Injury is Associated with Poor Lung Outcomes in Infants Born >/=32 Weeks of Gestational Age. *Am J Perinatol*. 2020;37(2):231-240.
109. Stoops C, Sims B, Griffin R, Askenazi DJ. Neonatal Acute Kidney Injury and the Risk of Intraventricular Hemorrhage in the Very Low Birth Weight Infant. *Neonatology*. 2016;110(4):307-312.
110. Kraut EJ, Boohaker LJ, Askenazi DJ, Fletcher J, Kent AL, Neonatal Kidney C. Incidence of neonatal hypertension from a large multicenter study [Assessment of Worldwide Acute Kidney Injury Epidemiology in Neonates-AWAKEN]. *Pediatr Res*. 2018;84(2):279-289.
111. Coca SG, Singanamala S, Parikh CR. Chronic kidney disease after acute kidney injury: a systematic review and meta-analysis. *Kidney Int*. 2012;81(5):442-448.
112. Greenberg JH, Coca S, Parikh CR. Long-term risk of chronic kidney disease and mortality in children after acute kidney injury: a systematic review. *BMC Nephrol*. 2014;15:184.
113. Chawla LS, Eggers PW, Star RA, Kimmel PL. Acute kidney injury and chronic kidney disease as interconnected syndromes. *N Engl J Med*. 2014;371(1):58-66.
114. Basile DP, Bonventre JV, Mehta R, Nangaku M, Unwin R, Rosner MH, et al. Progression after AKI: Understanding Maladaptive Repair Processes to Predict and Identify Therapeutic Treatments. *J Am Soc Nephrol*. 2016;27(3):687-697.
115. Sutherland MR, Gubhaju L, Moore L, Kent AL, Dahlstrom JE, Horne RS, et al. Accelerated maturation and abnormal morphology in the preterm neonatal kidney. *J Am Soc Nephrol*. 2011;22(7):1365-1374.
116. Charlton JR, Baldelomar EJ, Hyatt DM, Bennett KM. Nephron number and its determinants: a 2020 update. *Pediatr Nephrol*. 2020.
117. Abitbol CL, Bauer CR, Montane B, Chandar J, Duara S, Zilleruelo G. Long-term follow-up of extremely low birth weight infants with neonatal renal failure. *Pediatr Nephrol*. 2003;18(9):887-893.
118. Harer MW, Pope CF, Conaway MR, Charlton JR. Follow-up of Acute kidney injury in Neonates during Childhood Years (FANCY): a prospective cohort study. *Pediatr Nephrol*. 2017;32(6):1067-1076.

119. Bruel A, Roze JC, Quere MP, Flamant C, Boivin M, Roussey-Kesler G, et al. Renal outcome in children born preterm with neonatal acute renal failure: IRENEO-a prospective controlled study. *Pediatr Nephrol*. 2016;31(12):2365-2373.
120. Maqsood S, Fung N, Chowdhary V, Raina R, Mhanna MJ. Outcome of extremely low birth weight infants with a history of neonatal acute kidney injury. *Pediatr Nephrol*. 2017;32(6):1035-1043.
121. Askenazi DJ, Morgan C, Goldstein SL, Selewski DT, Moxey-Mims MM, Kimmel PL, et al. Strategies to improve the understanding of long-term renal consequences after neonatal acute kidney injury. *Pediatr Res*. 2016;79(3):502-508.
122. Starr MC, Kula A, Lieberman J, Menon S, Perkins AJ, Lam T, et al. The impact of increased awareness of acute kidney injury in the Neonatal Intensive Care Unit on acute kidney injury incidence and reporting: results of a retrospective cohort study. *J Perinatol*. 2020;40(9):1301-1307.
123. Vincent K, Murphy HJ, Ross JR, Twombley KE. Acute Kidney Injury Guidelines Are Associated With Improved Recognition and Follow-up for Neonatal Patients. *Adv Neonatal Care*. 2020;20(4):269-275.
124. Kent AL, Charlton JR, Guillet R, Gist KM, Hanna M, El Samra A, et al. Neonatal Acute Kidney Injury: A Survey of Neonatologists' and Nephrologists' Perceptions and Practice Management. *Am J Perinatol*. 2018;35(1):1-9.
125. Nour I, Elmaghraby R, Shehata R, El-Refaey A, Aldomiaty H, Mosbah A, et al. Selective head cooling and acute kidney injury in neonates with hypoxic ischemic encephalopathy. *J Neonatal Perinatal Med*. 2020;13(1):21-30.