Acute Kidney Injury After Heart Transplant in Children; Risk Factors and Outcomes

by

Christine MacDonald

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Abstract

Christine MacDonald, for the Master of Nursing

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Title: Acute Kidney Injury After Heart Transplant in Children; Risk Factors and Outcomes

Background: Heart transplant is life-saving for children with end-stage congenital heart disease or acquired heart failure. Critical illness following transplantation can include acute kidney injury (AKI). There is little data on the epidemiology of, risk factors for, or impact on outcomes of AKI after pediatric heart transplant.

Methods: Using secondary analysis of data from an ongoing prospective cohort study, we evaluated 72 children (0- 5 yrs) who had a heart transplant between 2001 and 2012. We evaluated: 1) postoperative AKI rate (defined by pRIFLE); 2) pre-, intra-, and early postoperative AKI risk factors (days on waitlist, inotrope use and ventilation pre-transplant, ECMO / ventricular assist device at transplant, preoperative estimated glomerular filtration rate (eGFR), ABO incompatibility, donor ischemic time, peak intraoperative lactate, tacrolimus level early postoperatively) using stepwise logistic regression; 3) effect of AKI on short-term outcomes (duration of ventilation and length of PICU stay).

Results: AKI occurred in 73% of children. Independent predictors of AKI were pre-transplant ventilation (OR 8.6, p=0.007) and higher eGFR (p=0.032). Following adjustment, preoperative inotrope significantly reduced the risk of AKI (OR 0.13, p=0.016). Sixteen percent of children had a tacrolimus level >15 *ug*/L on day 3 post-transplant and these children had more AKI than children without (OR 7.8, p=0.086). Although not statistically significant, automated model

selection retained tacrolimus level >15 *ug*/L as a predictor (using multiple different modeling strategies). AKI resulted in longer ventilation days and ICU stay (p=0.038 & p=0.004, respectively).

Conclusion: AKI was common after heart transplant and was associated with important outcomes. As in other pediatric cardiac surgery populations, lower preoperative GFR was protective against postoperative AKI; the role of modified immune suppressive strategies in this context needs to be further evaluated. Although not statistically significant, elevated early postoperative tacrolimus is likely biologically important in the prediction of AKI risk and needs further evaluation in a larger cohort.

Preface

This thesis is submitted for the degree of master in nursing at the University of Alberta. This research has been conducted under the supervision of Dr. Colleen Norris and Dr. Gwen Rempel between September 2012 and September 2014.

This work to the best of my knowledge is original, except where referenced to previous work. This work is unpublished, however, the American Society of Nephrology has accepted our request to submit a poster to Kidney Week in Philadelphia November, 2014.

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Abbreviations

Calculated GFR	Globular filtration rate Schartz formula: cGFR(ml/mm/1.73m2) =kl/sCR l is body length (cm), sCR is in milligrams
Cardiac Re-op	Procedure that requires re-opening of the sternum
CAV	Cardiac allograft vasculapathy in advanced coronary disease
CPB	Cardiopulmonary bypass time
CRRT	Continuous renal replacement therapy
CVVH	Continuous venovenous haemofiltration
DHCA	Deep hypothermic circulatory arrest
Dialysis	Hemodialysis or Prisma uses a special type of filter to remove excess waste products and water from the body.
ECMO	Extracorporeal membrane oxygenation: technique of providing both cardiac and respiratory support oxygen to patients whose heart and lungs are so severely diseased or damaged
eCCL	Estimated Creatinine clearance level
Graft Rejection	Graft rejection as grade 2R or higher or if associated with clinical impairment
Listing Status	Listing status prior to transplant; Status 1: at home Status 2: hospitalized for complications of heart disease Status 3: PICU care, inotropes, VAD Status 3.5: high dose inotropes, not a candidate for VAD Status 4S: high PRA Status 4: ventilated, inotropes, ECMO
MMP	Mycophenolate
PD	Peritoneal dialysis uses a fluid that is placed into the patient's stomach cavity through a special plastic tube to remove excess waste products and fluid from the body
PRA	Panel-reactive antibodies

PTLD	Post-transplant lymphoproliferative disorder is the name given to a B-cell proliferation due to therapeutic immunosuppression after organ transplantation.
SCr	Serum Creatinine lab value measurement
VAD	Ventricular assist device: is a mechanical circulatory device that is used to partially or completely replace the function of a failing heart

Chapter 1: Proposal

Among children who undergo heart transplantation little is known about the occurrence and consequences of acute kidney injury (AKI). Pediatric AKI is increasingly acknowledged as a cause of morbidity and mortality for children who undergo lifesaving treatment(Li et al., 2011; Pedersen et al., 2007). Several recent retrospective studies suggest that AKI is associated with longer hospital stay, greater utilization of resources (Basu, Devarajan, Wong, & Wheeler, 2011) and may be associated with future chronic renal failure (Palmieri, Lavrentieva, & Greenhalgh, 2009; Tang, Du, & L'Ecuyer, 2011; Zappitelli et al., 2009) AKI is common after pediatric cardiac surgery, occurring in 30% to 45%, with an associated mortality of 20% to 79% depending on the classification of AKI (Pedersen et al., 2007; Zappitelli et al., 2009). However, in the pediatric heart transplant population, the consequences of AKI are less studied, with most publications focusing on the influence of life-long immunosuppression on renal function (Alonso, 2004; Bharat, Manlhiot, McCrindle, Pollock-BarZiv, & Dipchand, 2009). Additionally, there is a lack of consistency in the clinical evaluation of AKI, with more than 30 definitions over the past 25 years (Basu et al., 2011).

In 2004, the Acute Dialysis Quality Initiative (ADQI) group published a classification system for AKI and failure based on serum creatinine (SCr) and urine output. There are 5 classification categories 1) at risk for renal dysfunction, 2) injury, 3) renal failure, 4) loss of kidney function, and, 5) end-stage renal failure, thus establishing the acronym RIFLE (Bellomo, Ronco, Kellum, Mehta, & Palevsky, 2004). In 2007, a modified RIFLE (pRIFLE) was developed for identifying AKI in pediatrics (Akcan-Arikan et al., 2007). Akcan-Arikan reported pRIFLE identified that AKI occurs in 82% of critically ill children with initial week of PICU admission. In addition, pRIFLE criteria identified that 36% of children post cardiac surgery will develop AKI within 3 days (Zappitelli et al., 2009).

Purpose

The purpose of this retrospective study was to determine prevalence of AKI using the pRIFLE criteria in pediatric cardiac transplant recipients who under went transplantation at the Stollery Children's Hospital between 2002 and 2011 and were followed by The Complex Pediatric Therapies Follow-up Program (CPTFP).

Objectives

The objectives of the study were to describe the epidemiology of AKI postoperatively in a single center cohort of children (age 0-5 years), identify potentially modifiable risk factors for developing AKI, and determine whether AKI is independently associated with clinically important outcomes at 30 days and one year post transplant.

Background AKI

AKI Definition

AKI is defined by a sudden decrease or loss of renal function where the kidney is unable to sufficiently excrete metabolic waste products, maintain fluid and electrolyte homeostasis, and regulate acid-base balance (Basu et al., 2011). Historically, a variety of clinical definitions of AKI were used in clinical investigation but this created difficulty in assessing the incidence of AKI or comparing outcomes (Goldstein & Devarajan, 2010).

In 2004, the Acute Dialysis Quality Initiative (ADQI) group published a classification system for acute kidney injury and failure based on serum creatinine (SCr) and urine output. The condition is classified at risk for renal dysfunction, injury, renal failure, loss of kidney function, or end-stage renal failure, thus establishing the acronym RIFLE (Bellomo et al., 2004). In 2007, a modified RIFLE (pRIFLE) was developed for identifying AKI in pediatrics (see Table 1) (Akcan-Arikan et al., 2007) .

Table 1. RIFLE and pRIFLE

Scheme	Stage	Creatinine Criteria	Urine Output Criteria
RIFLE	R	↑ ≥1.5x or ↓ GFR ≥ 25%	<0.5 ml/kg/hr for 6 hrs
	I	↑ ≥2x or ↓ GFR ≥ 50%	<0.5 ml/kg/hr for 12 hrs
	F	↑ ≥3x or sCR >350µmol/L	<0.3 ml/kg/hr for 24 hrs or anuria for 12 hrs
	L	Persistent failure >4 wks	
	E	Persistent failure >3mths	
pRIFLE	R	eCCL ↓ ≥25%	<0.5 ml/kg/hr for 8 hrs
	I	eCCL ↓ ≥50%	<0.5 ml/kg/hr for 12 hrs
	F	eCCL ↓ ≥75% or eCCL <35ml/min/1.73^2	<0.3 ml/kg/hr for 24 hrs or anuria for 12 hrs
	L	Persistent failure >4 wks	
	E	Persistent failure >3mths	

RIFLE, risk, injury, failure, loss, end stage; pRIFLE, pediatric RIFLE; GFR, glomerular filtration rate; SCr, serum creatinine; eCCL, estimated creatinine clearance.

(Akcan-Arikan et al., 2007; Bellomo et al., 2004).

Causes of AKI

Causes of AKI can be stratified into three categories depending on the location of damage in the kidney. Pre-renal AKI occurs when arteriolar autoregulation cannot compensate for a drop in perfusion pressure and there is a decrease in glomerular filtration rate (GFR). Conditions that can cause a decrease in perfusion pressure are hypovolemia, low cardiac output syndrome and extreme vasodilatation (Basu et al., 2011).

Intra-renal AKI is caused by structural damage to the glomeruli, tubules, and blood vessels. Primarily this occurs when cellular changes are caused by nephotoxic substances or ischemia injury (Askenazi, 2011).

Post-renal AKI refers to an obstruction of the urinary tract which stops the flow and emptying of urine from the kidneys. The renal calculi within the collection tubules may be blocked or compressed which is reflected in abdominal compartment syndrome (Basu et al., 2011)..

AKI in Children

The reported incidence of AKI varies from 10% to 82% dependent on the pediatric population (see Table 2). However, much of the recent literature studying AKI focuses on patients requiring continuous renal replacement therapy (CRRT), indicating severe renal failure (Gulati & Bagga, 2011). Studies report that even a small rise in SCr is a risk for mortality in adults and pediatric patients (Goldstein & Devarajan, 2010; Gude et al., 2010b) Clinical studies report that AKI is evident within seven days post life threatening illness, cardiac surgery, extracorporeal membrane oxygenation (ECMO), or cardiopulmonary bypass (CPB) (Li et al., 2011; Parikh et al., 2011; Smith, Hardison, Worden, Fleming, & Taylor, 2009) .

Table 2. Incidence of acute kidney injury since 2007 using pRIFLE

Authors	Year	Population	n	Mean Age (yrs)	Incidence%	RRT%	Mortality %
Akcan-Arikan et al	2007	PICU	150	6.4±6.4	82%	8.9%	14.6%
Dent et al	2007	CS	120	4.2±0.6	37%		13.3%
Plotz et al	2007	PICU	150	6.1±5.5	58.2%	10%	17%
Palmieri et al	2009	Burns	123		45.5%		8.9%
Smith et al	2009	ECMO	48	134days	71.7%	58%	61%
Zappitelli et al	2009	Post CPB	390	2.8±4.7	35.9%	1.8%	4%
Santiago et al	2010	PICU	174	4.4	40%	42%	35.6%
Schnieder et al	2010	PICU	3396	4.3-7.5mos	10%		30-32%
Chiravuri et al	2011	Post CBP	494	3.5±5.5	52.6%	30%	42%
Krawczeski et al	2011	Post CBP	220	3.3	27%		

PICU, pediatric intensive care unit; CS, cardiac surgery; ECMO, extracorporeal membrane oxygenation; CPB, cardiopulmonary bypass. Selected studies included.

Burden

Pediatric heart transplantation is an established treatment approach for end-stage heart conditions like cardiomyopathies, failed palliation of complex congenital heart defects, and other complex cardiac

malformations that are incompatible with life (de Jonge et al., 2008). However, long wait times to transplantation often cause low cardiac output syndrome and reduced renal function (Alonso, 2004). Several studies report that poor renal function prior to transplantation is associated with early postoperative mortality and CRRT (Jokinen et al., 2010; Tang et al., 2011). Renal dysfunction occurs in 20% of recipients prior to transplant and 3% need dialysis (Gandhi et al., 2011).

Children undergoing heart transplantation often require technologically advanced medical interventions to bridge them to transplant and or recovery. It is not uncommon for children in heart failure to require interventions to support their hemodynamic instability like ECMO or a Ventricular assist device (VAD), mechanical ventilator and inotropes (Bae et al., 2005). Complications of interventions and wait time to transplant result in 1 out of 9 children dying prior to transplant (Gandhi et al., 2011) .

Surgical interventions and organ procurement produce long ischemic and cardiopulmonary bypass (CBP) times (Rylski et al., 2010). The literature reports the incidence of AKI in pediatric cardiac surgery post CBP is 35% to 52% (Chiravuri et al., 2011; Zappitelli et al., 2009). One retrospective study by Tang et al. (2011) reported pediatric heart transplant recipients who required RRT pre and post transplant had a significantly higher mortality in the postoperative cohort. Graft survival for the pre CRRT at one, five, and 10 years is 95%, 80%, and 74%, significantly above the post group (38%, 34%, and 32%; all $p<0.001$).

Long term renal dysfunction caused by immunosuppression therapy is well documented in pediatric heart transplant recipients (Dello Strologo et al., 2006). However, further study is needed to assess what prevalence of AKI exists in pediatric heart transplant recipients. Also, other risk factors perioperatively that might contribute to the AKI in pediatric heart transplant recipients need to be determined. Identification of risk factors could help support development of strategies for improved

timing of interventions. Timing the intervention of dialysis at first clinical signs of AKI may improve morbidity prevalence and mortality.

The necessity of CBP in pediatric heart transplantation establishes risk of AKI. Retrospective studies indicate that AKI is associated with longer hospital stay and increased possibility of chronic renal disease (Sethi et al., 2011; Zappitelli et al., 2009) . The literature indicates that risk factors for AKI following CBP include complexity of congenital heart defect, CBP duration, mechanical ventilation, young age, and small size (Chiravuri et al., 2011; Li et al., 2011).

AKI in pediatric heart transplantation remains a clinical challenge. In adults, pre-operative risk factors are prior cardiac surgery, elevated SCr, and patient age. Additional risk factors include donor ischemic time, CBP, and early postoperative intravenous cyclosporine (Boyle et al., 2006; E. Gude et al., 2010b). Currently, there is limited evidence of AKI in pediatric heart transplant. A small number of retrospective studies report renal insufficiency (RI) perioperatively for pediatric heart transplant recipients (Bharat et al., 2009; Lee et al., 2007; Mah et al., 2009; Phan, West, Stephens, & Hébert, 2003; Tang et al., 2011) . However, these retrospective studies do not yield any data using current standardized definitions available of AKI (Akcan-Arikan et al., 2007).

Although risk factors for AKI have been well identified in pediatric cardiac surgery requiring CBP further research is necessary in the heart transplant population. Understanding the epidemiology of AKI using pRIFLE in association with transplant specific risk factors like immunosuppression therapy could help improve patient outcomes.

Literature Review

Limited information exists about AKI in pediatric heart transplant recipients. The purpose of this literature review is to examine the current knowledge, prevalence, risk factors and outcomes of AKI in pediatric heart transplantation.

Search Strategy

Electronic databases including Medline, CINAHL, Pubmed, and the Cochrane review were searched for relevant articles published between 1990 and June1, 2012 using the following keywords: (Renal Insufficiency+ OR TI renal OR TI kidney OR TI nephr*) AND (heart transplantation OR heart transplant*) AND (risk factors). The Cochrane database yielded no relevant articles. Pubmed contained duplicates of pertinent articles noted in Medline and CINAHL (see Appendix A for search results for Medline and CINAHL). References of the retrieved articles were hand searched for additional relevant papers.

While the search term 'renal insufficiency' when expanded in EBSCO Host included the search term 'AKI', no relevant articles specific to AKI and pediatric heart transplant were identified. Therefore the following inclusion criteria were used: written in English, focusing on renal insufficiency (RI) in pediatric heart transplant recipients (i.e., age 0-18 years); and containing data describing risk factors, characteristics, outcomes, mortality and incidence of RI perioperatively and short term follow-up. Papers describing RI in pediatric non heart organ transplantation regimen were excluded.

Results

The search strategy resulted in 1000 abstracts, of which six publications met the inclusion criteria. Sixty eight papers were not written in English and were excluded; 770 abstracts discussed topics in adult patients and were excluded. Of the remaining 162 articles that referenced pediatrics, 38 were related to perioperative risk factors, and thus included and retrieved. The references of the retrieved papers did not yield additional papers to be included in this review. The Critical Appraisal Skills Programme (CASP, 2011), specifically the cohort study questionnaire was utilized to screen that 38 retrieved articles, resulting in five articles being included in the literature review.

The five studies reviewed (see Appendix B) were retrospective cohort studies conducted in the United States and published between 2003 and 2011. The data source for three of the studies was the Organ Procurement and Transplantation Network (OPTN) (Gandhi et al., 2011; Lee et al., 2007; Mah et al., 2009; Tang et al., 2011). The fourth and fifth studies were single centre studies (n=77 and n=41) ((Phan et al., 2003; Sachdeva et al., 2007)) .

Synthesis of Selected Articles

A synthesis of the five studies will be presented as per following themes: prevalence of RI postoperatively, risk factors and outcomes (see Appendix B).

Postoperative Prevalence. Prevalence of RI postoperatively was reported in the five articles ranging from 7% to 34% (Gandhi et al., 2011; Lee et al., 2007; Phan et al., 2003; Sachdeva et al., 2007; Tang et al., 2011) .

The prevalence of post transplant RI that required dialysis reported by Tang et al (2011) and Gandhi et al. (2011) were 7% and 34%, respectively. Both studies had large samples (Tang et al. n = 3598; Gandhi et al. n = 730) the difference in outcomes may be because Gandhi et al. (2011) included recipients on dialysis who died in their sample. Inclusion of participants who died may appear in statistical analysis as an outlier, skewing the data (Munro, 2005).

Prevalence of long-term RI was noted by three of the articles (Lee et al., 2007; Sachdeva et al., 2007; Tang et al., 2011) . There was inconsistency in the studies reporting length of time of RI from transplantation and in operational definitions. Sachdeva et al. (2007) used a measure of GFR of $<75ml/min/1.73^{m2}$ or dialysis and had a prevalence of RI at one year of 17%, three and five years of 21% and 26%. Similarly, Tang et al. (2011) reported long-term outcomes of RI at one, five, and seven years of 24%, 17% and 26%, respectively. In spite of similar results of RI overtime, Tang et al. (2011)

operational definition was SCr >2.5 mg/dl subsequently diminishing the relevance of this similarity. Conversely, Lee et al. (2007) reported at 10 years a prevalence RI of 16% using SCr of >2.5mg/dl.

Phan et al. (2003) discusses RI as acute renal disease (ARD) or GFR being increased or decreased. ARD was defined as a two-fold increased in baseline SCr. Abnormally low GFR as less than 60ml/min/1.73m^2 for infants less than two months, and less than 80ml/min/1.73m^2 for children older than two months of age. The incidence of the ARD was 29% and POST abnormal GFR of seven% (Phan et al., 2003).

Risk Factors

The literature review yielded risk factors of RI that could help profile pediatric heart transplant recipients postoperatively. Four of the articles (See Appendix B) identified risk factors, however, results were subject to the differences in the research question. For instance, Tang et al. (2011)'s research question concerned RI perioperatively, while, Lee et al. (2007)'s aim was to explore pre-transplant risk factors. Significant statistical data concerning risk factors associated with developing RI in pediatric heart transplant recipients were both similar and contrary across four of the studies. Two of the studies similarly demonstrated that ECMO and mechanical ventilation contributed significantly to the prevalence of RI and mortality ($p = 0.001$) (Lee et al., 2007; Tang et al., 2011). Gandhi et al. (2011) found that RI, ECMO and mechanical ventilation had significance ($p = 0.001$) for mortality but failed to show if ECMO or ventilation was a risk factor for RI. While Sachdeva et al. (2007)'s study found that ECMO and ventilation were not risk factors for RI in pediatric heart recipients ($p = 0.74$).

Equally controversial Sachdeva et al. (2007) found no significant difference in the prevalence of RI who required inotropes or had cardiac diagnosis ($p = 0.8$ and $p = 0.8$, retrospectively). Three studies reported the significance of cardiac diagnoses in prevalence of RI for pediatric transplant recipients; Gandhi et al. (2011) stated there was significant risk factor for RI ($p= <0.001$) in repaired congenital

heart defects (CHD), Tang et al. (2011) for CHD (p = < 0.0001) and Lee et al. (2007) for hypertrophic cardiomyopathy (HCM) (p = 0.001). Admittedly this difference may stem from the research methods and research question. The similarities in risk factors associated with RI in pediatric heart transplant recipients appear in studies, that use the same operational definitions, population sampling, and similar research questions.

Furthermore, Sachdeva et al. (2007) found significance difference in the prevalence of RI for those younger in age at transplant (p= 0.007). Gandhi et al. (2011) did not find age significant, but weight had a significant influence on RI (p=0.001). African-American race had a significance (p = 0.04) in one study (Sachdeva et al., 2007). Similarly, Lee et al. (2007) found significance prevalence of RI in the African-American race (p = 0.0035). Tang et al. (2011) also found risk factors for developing RI included infection (p = 0.0027) and inotropic support (p = <0.0001), which was disputed in Sachdeva et al. (2007) study.

Phan et al. (2003) reported that abnormal GFR preoperatively was a risk factor for abnormal GFR of 7%. This study reported that the number of children with a GFR <80ml/min/1.73m2 at follow-up was small, so statistical analysis to determine risk factors for decreased GFR was not attempted (Phan et al., 2003).

Outcomes

RI in pediatric heart transplant recipients has negative consequences on quality of life and mortality (Bharat et al., 2009). The literature analysis yielded similar or dissimilar outcome measures dependant on methodology and research question. Three studies indicated that RI in pediatric heart transplantation increased over time (Lee et al., 2007; Sachdeva et al., 2007; Tang et al., 2011). Tang et al. (2011) reported that recipients that required RRT postoperatively have an survival rate of 38%, 36%, and 35% at one, five, and 10 years, respectively. As well, Sachdeva et al. (2007) reported RI to be 17%,

21%, and 26% at one, three, and five years with a mortality rate of 22% in their sample. Tang et al. (2011) categorized recipients that required dialysis perioperatively versus no dialysis and demonstrated significance of RI at one-year post transplant ($p = 0.001$), but the difference did not persist at five and seven years. Tang et al. further categorized groups to PRE and POST dialysis that revealed a significant difference between groups ($p = <0.01$): PRE at one, five, and 10 year graft survival was 95%, 80%, and 74% , significantly greater than POST group 28%, 34%, and 32%. Also Sachdeva et al. (2007) reported that dialysis was not associated with later RI ($p = 0.47$), but this disagreement might stem from the Sachdeva's small sample size (n=77). Gandhi et al. (2011) reported that heart transplant recipients had an in-hospital mortality of 11.2%. Phan et al. (2003) reported 29% with ARD and this study excluded 13 children who died in the first year post transplant.

Inconsistent data collection points contributes to a less comprehensive understanding of the prevalence of RI in transplant recipients over time. All five studies had outcome measures at one year (Gandhi et al., 2011; Lee et al., 2007; Mah et al., 2009; Phan et al., 2003; Sachdeva et al., 2007; Tang et al., 2011) . Only two included mortality at 30 days, in which the highest mortality for infants occurs (Gandhi et al., 2011; Tang et al., 2011).

Gaps in the literature review

The literature established that RI is prevalent post transplantation. However, all five studies evaluated RI with different measures; two studies used SCr >2.5mg/dl (Lee et al., 2007; Tang et al., 2011) while Gandi et al. (2011) used creatinine clearance of <40ml/min/1.73m^2 and Sachediva et al. (2003) GFR <75ml/min/1.73 m^2. Lastly, Phan et al. (2003) discussed RI in terms of ARD or GFR being increased or decreased. ARD was defined as two-fold increased in baseline SCr. Abnormally low GFR as less than 60ml/min/1.73m^2 for infants less than two months, and less than 80ml/min/1.73m^2 for children older than two months of age. The inconsistent definitions in variables and operational

measures diminish the strength of research (Wood, 2011). Furthermore, the literature fails to use a current standardized definition for AKI, like pRIFLE (Akcan-Arikan et al., 2007) .

Although the literature reports RI postoperatively it fails to answer if AKI occurred within the time sensitive criteria of pRIFLE; in fact, the earliest reported measure is at 6 months or one year (Gandhi et al., 2011; Lee et al., 2007; Sachdeva et al., 2007; Tang et al., 2011). Although, Phan et al. (2003) reported ARD, this study failed to define ARD beyond using a two-fold increase in SCr from baseline. To determine the prevalence of AKI the literature must include data measures in the acute postoperative as defined by pRIFLE.

Immunosuppression is discussed in two of the studies and highlight the changes made to the protocols overtime (Lee et al., 2007; Phan et al., 2003). Lee et al. (2007) omitted immunosuppression as a variable from this study, while Phan et al (2003) did not attempt to correlate drug levels and renal function as that mean trough level of tacroliums was 9 ng/ml and cyclosporine 217 ng/ml. Immunosuppression therapy has been identified as a causative factor of chronic RI (Alonso, 2004; Bharat et al., 2009) . It would be clinically relevant to assess if induction immunosuppression therapy is associated with the risk of AKI.

Research Purpose

The purpose of this retrospective study was to determine prevalence of AKI using the pRIFLE criteria in pediatric cardiac transplant recipients who under went transplantation at the Stollery Children's Hospital between 2002 and 2010 and are followed by The Complex Pediatric Therapies Follow-up Program (CPTFP).

Research Objectives

1. Describe the epidemiology of AKI postoperatively in a in a single center cohort of children who received a heart transplant (0-5 years).

2. Identify potentially modifiable risk factors for developing AKI postoperatively.

3. Determine whether AKI is independently associated with clinically important outcomes at 30 days and one year post transplant.

Research Questions

1. How frequently does AKI occur in pediatric heart transplant recipients?

2. What is the range of severity as per the pRIFLE criteria of risk, injury, renal failure, loss of kidney function, and, end-stage renal failure.

3. What are the risk factors for AKI for pediatric heart transplant recipients?

4. What are the outcomes at 30 days and one year for pediatric heart transplant recipients with AKI?

Methods

Research Design

This is a cohort study with the goal to investigate incidence of AKI occurring postoperatively, describe any potentially modifiable risk factors for AKI, and determine whether outcomes are influenced by AKI. This study will follow a quantitative descriptive exploratory design.

Data Source

Data will be collected through retrospective chart reviews at the Stollery Children's Hospital, Edmonton, Canada. Additional data will be from the Registry of the CPTFP of Western Canada. One purpose of CRTFP, a multi-provincial registry is to provide longitudinal follow-up of infants and children surviving complex cardiac conditions with associated surgeries and treatments and to identify potentially modifiable variables at each level of care.

We will employ purposeful sampling that is frequently used in chart reviews (Hess, 2004). It is acknowledged that purposeful sampling generates sampling bias. However, when researching a specific cohort like pediatric transplant recipients this sample method is acceptable (Wood, 2011).

The sample will include all pediatric heart transplantations for children aged 0-5 years that occurred between January 1, 2002 and Dec 31, 2010. To limit sampling bias, all 74 children in the cohort will be included. Also, maximizing the sample size decreases the risk of a type 2 error (Wood, 2011). Inclusion criteria will require a minimum follow-up of 12 months after transplant.

Consent

To conduct this chart review consent was obtained from Human Ethics Research Online (HERO) at the University of Alberta and Alberta Health Services, Stollery Children's Hospital. In addition operational approval from Alberta Health Services will be obtained to assess health records through Northern Alberta Clinical Trials and Research Centre (NACTRC). The data will be collected within six months of approval.

This study poses no apparent physical, psychological, economical, or social risk to the subjects in the study. There will be no contact between the researcher and the patients. Treatment and care of patients and families will not be influenced by accessing medical records.

Charts will remain on hospital property at all times and will not be reproduced in any manner. There will be no manipulation of data or charts involved in the study. The researcher at all times will take precautions to ensure family and patient anonymity and confidentiality. No names will be recorded, and only the researchers involved will access charts. Data will be de-identified; that is, no protected health information (PHI) will be included in the database (name, age, initials, data of birth, any other unique identifying number, or characteristic).

Since the study is retrospective, patient's involvement in the study did not receive any direct benefit, however, future pediatric heart transplant recipients with AKI may benefit.

Data Collection

Two sources of data will be accessed. The first source is a retrospective review of charts of children who meet the study criteria and who are enrolled in the CPTFP. The second is the selected variables from the CPTFP data base. Any data collected retrospectively from chart reviews will be added to the CPTFP data base.

To ensure reliability of the study a standardized checklist will be created, along with clear operational measures and variable definitions (Wood, 2011). Developing a standardized list will ensure that data collection is consistent (Hess, 2004). The checklist (see Appendix C for standardized checklist) will be predetermined prior to data collection by examining previous studies outlined in the literature review. Collaborating with an expert panel composed of Dr. Ari Joffe (pediatric critical care intensivist, Stollery) and Dr Catherine Morgan (pediatric nephrologist, Stollery) will substantiate the checklist to ensure validity and reliability.

Operational measures and variable definitions for the standardized chart checklist are outlined (see Appendix C and D) to help maintain rigor of the study. Continuously assessing the data collection process and evaluation of possible emerging variables that reoccur may become important in answering the research question. Data collection and all private information will be stored in a secure location and coded to ensure privacy.

Clinical and Laboratory Variables

The data extracted will be relevant pre-, intra- and post-operative variables from the CPTFP and chart review.

Pre-operative variables included: age(years), gender, weight(kg), height(cm), primary diagnosis (CHD or myocarditis), PICU admission, use of pre-operative ventilation, last SCr and lactate drawn prior to surgery, previous cardiac surgery, milrinone, inotrope score ((dopamine + dobutamine) +

(100)(epinephrine +norepinephrine + phynylephrine), micrograms/kg/min, categorized as <10, 10 to 14, 15 to 19, 20 to 24, >25), ECMO, VAD, dialysis (CRRT or PD), days on transplant list, listing status, panel reactive antibodies (PRA), donor specific antibodies (DSA), desensitization, positive crossmatch, dialysis, urine output and percentage fluid overload prior to transplant.

Intra-operative variables included: CPB time (minutes), deep hypothermic circulatory arrest (DHCA), aortic cross clamp time, donor heart ischemic time, and milrinone.

Post-operative variables included: milrinone, peak lactate and inotrope score were measured at 3 times postoperatively: 1) Post-operatively day 1 (Day 1); 2) Post-operative day 2 to 5 (Day 2-5); 3) Post-operative day 6 until hospital discharge (day 6+). Remaining variables collected daily: mechanical ventilation days, ECMO, VAD, peak tacroliums level daily, peak daily SCr, daily urine output, first day negative fluid balance and dialysis (CRRT or PD). SCr measurements are calculated by the Jaffe reaction.

Primary outcome for in-hospital morbidity will be measured by PICU length of stay, ventilator days, and total days in hospital. Secondary outcomes: 1) is mortality as measured by post-operative survival time (days to death) to be evaluated at two time points: 30 days and one year; 2) transplant complication including rejection, re-transplantation, graft vasculapathy (CAV) or post-transplant lymphoproliferative disorder (PTLD) will be collected at one year.

Study Duration and Timeline

Projected start date of study is October 1, 2012. Table 3

Timeline of Study

Consent from Complex Therapies	October 2012
Ethics and Defend Proposal	December 2012
Data collection and data analysis	January 2012
Thesis Completion	September 2014
Submit for Publication	October 2014

Data Analysis

The purpose of data analysis is to provide meaning to the raw data that is collected for the study (Munro, 2005). With the assistance of Dr Catherine Morgan and the use of the STATA 9.2 statistical software the raw data will be manipulated to answer the research questions. To begin the data analysis the answers from the standardized checklist must be coded. For example the categorical data of gender will coded 1 = male and 2 = female (Munro, 2005). The data collected will include specific variables that are measured by nominal, ordinal, and interval scales (Munro, 2005). Once the variables are coded and entered into STATA 9.2 the data can be transformed to meaningful statistics like tables and charts.

Logistic regression will be used to preformed to determine univariate associations between risk factors and AKI. Univariate descriptive statistics (age, wt, ht, sex, primary diagnosis of CHD or HCM) describe the demographic characteristics of the cohort. Frequency distribution will present the specific variables for easy dissemination. Continuous variables, like age, will be expressed by mean $\pm$ standard deviation (s.d.), or median (interquartile range or IQR) when appropriate. (Munro, 2005). The categorical or nominal variables like race and gender can be expressed as percentages. Univariate

associations between risk factors and AKI will be evaluated using Students t-tests or Pearson's chi-squared test (Munro, 2005).

Linear regression models will predict potential AKI risk factors for pediatric heart transplant recipients. The following risk factors were considered for the AKI prediction model: age, gender, weight, height, primary diagnosis, PICU admission, use of pre-operative ventilation, last SCr and lactate drawn prior to surgery, previous cardiac surgery, milrinone, peak inotrope score, ECMO, VAD, days on transplant list, listing status, PRA, DSA, desensitization, positive crossmatch, urine output, fluid balance prior to transplant, CPB time, DHCA, aortic cross clamp time, donor heart ischemic time, and intra-operative milrinone. This will generate odd ratios and 95% confidence intervals. Variables that are significant at the 0.2 level in univariate analysis may be included in a multivariate analysis or possibly stepwise selection modeling (Munro, 2005).

Multiple logistic regression analysis (p-value for entry into model = 0.1; p-value for removal =0.2) will evaluate independent AKI risk factors. Regression will predict outcomes and explain the interrelationships between variables. The limitations to multiple regression is including too many variables for the number of subjects (Munro, 2005). Running so many models can increase the chance that there may be a false significant outcome. During analysis a failure to account for a confounding variables may cause misestimation of the significant association of risk.

To determine AKI as a predictor for short term outcomes, at 30 days, Cox proportional hazards models can be utilized (Munro, 2005). The association between post-operative PICU stay, post-operative hospital stay, and ventilator days will be evaluated. All models potentially will be adjusted for age, gender, weight, height, primary diagnosis, days on transplant list, listing status, PRA, sensitization, DSA, desensitization, positive crossmatch, CBP length, use of DHCA, donor ischemic time, peak post-operative lactate and peak inotrope score, induction therapy, dialysis (CRRT or PD), peak tacrolimus

level, urine output and fluid balance prior to transplant. The association between AKI and in-hospital mortality can be evaluated by multiple logistic regression adjusting for the same covariates.

AKI as a predictor of long term outcomes, at one year, will be evaluated by a multiple regression model. The association between post-operative PICU stay, post-operative hospital stay, rejection, re-transplantation, CAV, PTLD and ventilator days will be evaluated. Adjusting for weight and age at surgery, primary diagnosis, pre and post-operative ventilation and dialysis, post-operative ECMO, peak inotropes, peak tacrolimus level, peak lactate; as these variables are known to be associated with outcomes in this population (Robertson, Charlene M.T. 2004; Sachdeva et al., 2007; Tang et al., 2011; Zappitelli et al., 2009). Cox-multiple regression will be used to evaluate the long-term mortality. Variables to be included in the model are those previously shown to be predictors in this cohort (Akcan-Arikan et al., 2007; Alonso, 2004;Chiravuri et al., 2011): independent effect of AKI on outcome like days ventilated and time in PICU, and hospital discharge. Also the evaluation on the effect of 25% SCr rise (pRIFLE SCr25) on in-hospital, 30 days and one year outcomes. For all analyses, a P-value of <0.05 will be considered statistically significant.

Data Quality

Retrospective studies present challenges for data collection, and consequently, data analysis (Munro, 2005). Researchers must be satisfied with the available data even though often there is missing or incomplete variables noted during the chart review. This challenge to data quality will be assessed determining the quantity or patterns of the missing data. Handling the missing data can be accomplished by listwise deletion, which is the STATA 9.0 default program of automatic deletion of that participant. Another option is deleting the variable from the study. Consideration for any deleting of data will be weighed carefully (Munro, 2005).

Statistical analysis presumes random sampling and generates significance tests or confidence intervals (CI) based on the entire population (Munro, 2005). As purposeful sampling is used for this study attention must be given to the possibility that the CI may be slightly lower. Furthermore, the small sample size may affect the power of the statistical *t*- test which tests the null hypothesis.

Dissemination

Once data analysis is completed dissemination of the study will occur though presentations, both local and peer reviewed venues, both nationally and internationally as well as peer-reviewed publication. All dissemination will be done with the involvement of thesis committee members.

The benefit of dissemination to practitioners is to improve clinical care, decrease AKI and improve outcomes for pediatric heart transplant recipients.

References

Akcan-Arikan, A., Zappitelli, M., Loftis, L. L., Washburn, K. K., Jefferson, L. S., & Goldstein, S. L. (2007). Modified RIFLE criteria in critically ill children with acute kidney injury. *Kidney International, 71*(10), 1028-1035.

Alonso, E. M. (2004). Long-term renal function in pediatric liver and heart recipients. *Pediatric Transplantation, 8*(4), 381-385.

Askenazi, D. (2011). Evaluation and management of critically ill children with acute kidney injury. *Current Opinion in Pediatrics, 23*(2), 201-207. doi: 10.1097/MOP.0b013e328342ff37

Bae, J., Frischer, J. S., Waich, M., Addonizio, L. J., Lazar, E. L., & Stolar, C. J. H. (2005). Extracorporeal membrane oxygenation in pediatric cardiac transplantation. *Journal of Pediatric Surgery, 40*(6), 1051-1056.

Basu, R. K., Devarajan, P., Wong, H., & Wheeler, D. S. (2011). An update and review of acute kidney injury in pediatrics. *Pediatric Critical Care Medicine: A Journal of the Society of Critical Care Medicine and the World Federation of Pediatric Intensive and Critical Care Societies, 12*(3), 339-347.

Bellomo, R., Ronco, C., Kellum, J. A., Mehta, R. L., & Palevsky, P. (2004). Acute renal failure - definition, outcome measures, animal models, fluid therapy and information technology needs: The second international consensus conference of the acute dialysis quality initiative (ADQI) group. *Critical Care (London, England), 8*(4), R204-R212.

Bharat, W., Manlhiot, C., McCrindle, B. W., Pollock-BarZiv, S., & Dipchand, A. I. (2009). The profile of renal function over time in a cohort of pediatric heart transplant recipients. *Pediatric Transplantation, 13*(1), 111-118.

Boyle, J. M., Moualla, S., Arrigain, S., Worley, S., Bakri, M. H., Starling, R. C., . . . Thakar, C. V. (2006). Risks and outcomes of acute kidney injury requiring dialysis after cardiac transplantation. *American Journal of Kidney Diseases: The Official Journal of the National Kidney Foundation, 48*(5), 787-796.

Critical Appraisal Skills Programme. (2011). Retrieved from http://www.casp-uk.net/wp-content/uploads/2011/11/CASP_Case-Control_Appraisal_Checklist_14oct10.pdf

Chiravuri, S. D., Riegger, L. Q., Christensen, R., Butler, R. R., Malviya, S., Tait, A. R., & Voepel-Lewis, T. (2011). Factors associated with acute kidney injury or failure in children undergoing cardiopulmonary bypass: A case-controlled study. *Paediatric Anaesthesia, 21*(8), 880-886. doi: 10.1111/j.1460-9592.2011.03532.x

de Jonge, N., Kirkels, J. H., Klöpping, C., Lahpor, J. R., Caliskan, K., Maat, A. P. W. M., . . . Balk, A. H. M. M. (2008). Guidelines for heart transplantation. *Netherlands*

Heart Journal: Monthly Journal of the Netherlands Society of Cardiology and the Netherlands Heart Foundation, 16(3), 79-87.

Dello Strologo, L., Parisi, F., Legato, A., Pontesilli, C., Pastore, A., Ravà, L., . . . Rizzoni, G. (2006). Long-term renal function in heart transplant children on cyclosporine treatment. *Pediatric Nephrology (Berlin, Germany), 21*(4), 561-565.

Gandhi, R., Almond, C., Singh, T. P., Gauvreau, K., Piercey, G., & Thiagarajan, R. R. (2011). Factors associated with in-hospital mortality in infants undergoing heart transplantation in the united states. *Journal of Thoracic & Cardiovascular Surgery, 141*(2), 531; Feb-536.

Goldstein, S. L., & Devarajan, P. (2010). Pediatrics: Acute kidney injury leads to pediatric patient mortality. *Nature Reviews.Nephrology, 6*(7), 393-394.

Gude, E., Andreassen, A. K., Arora, S., Gullestad, L., Grov, I., Hartmann, A., . . . Simonsen, S. (2010a). Acute renal failure early after heart transplantation: Risk factors and clinical consequences. *Clinical Transplantation, 24*(6), E207-13.

Gude, E., Andreassen, A. K., Arora, S., Gullestad, L., Grov, I., Hartmann, A., . . . Simonsen, S. (2010b). Acute renal failure early after heart transplantation: Risk factors and clinical consequences. *Clinical Transplantation, 24*(6), E207-E213. doi: 10.1111/j.1399-0012.2010.01225.x

Gulati, A., & Bagga, A. (2011). Management of acute renal failure in the pediatric intensive care unit. *Indian Journal of Pediatrics, 78*(6), 718-725.

Hess, D. R. (2004). Retrospective studies and chart reviews. *Respiratory Care, 49*(10), 1171-1174.

Jokinen, J. J., Tikkanen, J., Kukkonen, S., Hämmäinen, P., Lommi, J., Sipponen, J., & Lemström, K.,B. (2010). Natural course and risk factors for impaired renal function during the first year after heart transplantation. *The Journal of Heart and Lung Transplantation: The Official Publication of the International Society for Heart Transplantation, 29*(6), 633-640.

Lee, C. K., Christensen, L. L., Magee, J. C., Ojo, A. O., Harmon, W. E., & Bridges, N. D. (2007). Pre-transplant risk factors for chronic renal dysfunction after pediatric heart transplantation: A 10-year national cohort study. *The Journal of Heart and Lung Transplantation: The Official Publication of the International Society for Heart Transplantation, 26*(5), 458-465.

Li, S., Krawczeski, C. D., Zappitelli, M., Devarajan, P., Thiessen-Philbrook, H., Coca, S. G., . . . Parikh, C. R. (2011). Incidence, risk factors, and outcomes of acute kidney injury after pediatric cardiac surgery: A prospective multicenter study. *Critical Care Medicine, 39*(6), 1493-1499.

Mah, D., Singh, T. P., Thiagarajan, R. R., Gauvreau, K., Piercey, G. E., Blume, E. D., . . . Almond, C. S. (2009). Incidence and risk factors for mortality in infants awaiting heart transplantation in the USA. *Journal of Heart & Lung Transplantation, 28*(12), 1292-1298.

Munro, B. H., (2005). Statistical Methods: For health care research (5th ed.). Philadelphia: Lippincott Williams & Wilkins.

Palmieri, T., Lavrentieva, A., & Greenhalgh, D. (2009). An assessment of acute kidney injury with modified RIFLE criteria in pediatric patients with severe burns. *Intensive Care Medicine, 35*(12), 2125-2129.

Parikh, C. R., Devarajan, P., Zappitelli, M., Sint, K., Thiessen-Philbrook, H., Li, S., . . . Krawczeski, C. D. (2011). Postoperative biomarkers predict acute kidney injury and poor outcomes after pediatric cardiac surgery. *Journal of the American Society of Nephrology: JASN, 22*(9), 1737-1747.

Pedersen, K. R., Povlsen, J. V., Christensen, S., Pedersen, J., Hjortholm, K., Larsen, S. H., & Hjortdal, V. E. (2007). Risk factors for acute renal failure requiring dialysis after surgery for congenital heart disease in children. *Acta Anaesthesiologica Scandinavica, 51*(10), 1344-1349.

Phan, V., West, L. J., Stephens, D., & Hébert, D. (2003). Renal complications following heart transplantation in children: A single-center study. *American Journal of Transplantation: Official Journal of the American Society of Transplantation and the American Society of Transplant Surgeons, 3*(2), 214-218.

Robertson, C. M., Joffe A.R., Sauve, R. S., Rebeyka, I. M., Phillipos, E. Z., . . . Harder, J. R. (2004). Outcomes from an interprovincial program of newborn open heart surgery: *The Journal of Pediatrics, 144*(1), 86-92.

Rylski, B., Berchtold-Herz, M., Olschewski, M., Zeh, W., Schlensak, C., Siepe, M., & Beyersdorf, F. (2010). Reducing the ischemic time of donor hearts will decrease morbidity and costs of cardiac transplantations. *Interactive Cardiovascular and Thoracic Surgery, 10*(6), 945-947.

Sachdeva, R., Blaszak, R. T., Ainley, K. A., Parker, J. G., Morrow, W. R., & Frazier, E. A. (2007). Determinants of renal function in pediatric heart transplant recipients: Long-term follow-up study. *The Journal of Heart and Lung Transplantation: The Official Publication of the International Society for Heart Transplantation, 26*(2), 108-113.

Sethi, S. K., Goyal, D., Yadav, D. K., Shukla, U., Kajala, P. L., Gupta, V. K., . . . Juneja, A. (2011). Predictors of acute kidney injury post-cardiopulmonary bypass in children. *Clinical and Experimental Nephrology, 15*(4), 529-534.

Smith, A. H., Hardison, D. C., Worden, C. R., Fleming, G. M., & Taylor, M. B. (2009). Acute renal failure during extracorporeal support in the pediatric cardiac patient. *ASAIO Journal (American Society for Artificial Internal Organs: 1992), 55*(4), 412-416.

Wood, M. J., & Ross-Kerr, J.C. (2011). Basic steps in planning nursing research: From question to proposal (7th ed.). Boston: Jones-Bartlett.

Tang, L., Du, W., & L'Ecuyer, T. J. (2011). Perioperative renal failure in pediatric heart transplant recipients: Outcome and risk factors. *Pediatric Transplantation, 15*(4), 430-436.

Zappitelli, M., Bernier, P., Saczkowski, R. S., Tchervenkov, C. I., Gottesman, R., Dancea, A., . . . Alkandari, O. (2009). A small post-operative rise in serum creatinine predicts acute kidney injury in children undergoing cardiac surgery. *Kidney International, 76*(8), 885-892.

Chapter 2

Acute Kidney Injury After Heart Transplant in Children; Risk Factors and Outcomes

Little is known about the occurrence and consequences of acute kidney injury (AKI) among children who undergo heart transplantation. Pediatric AKI is increasingly acknowledged as a cause of morbidity and mortality for children who undergo lifesaving treatment (Li et al., 2011; Pedersen et al., 2007). Furthermore, several recent retrospective studies suggest that AKI is associated with longer hospital stay, greater utilization of resources,(Basu, Devarajan, Wong, & Wheeler, 2011) and may be associated with future chronic kidney disease (Palmieri, Lavrentieva, & Greenhalgh, 2009; Tang, Du, & L'Ecuyer, 2011; Zappitelli et al., 2009). AKI has been fairly well described after pediatric congenital heart surgery and is common, occurring in 30% to 64%. It carries an associated mortality rate of 20% to 79% depending on the classification of AKI (Morgan et al., 2013; Pedersen et al., 2007; Zappitelli et al., 2009). However, in the pediatric heart transplant population, the consequences of post-operative AKI are less studied, with most publications focusing on the influence of life-long immunosuppression on renal function (Alonso, 2004). Additionally, there is a lack of consistency in the clinical evaluation of AKI, with more than 30 definitions over the past 25 years (Basu et al., 2011).

The purpose of this retrospective study was to determine the prevalence of AKI using the pRIFLE criteria in pediatric cardiac transplant recipients who underwent transplantation at the Stollery Children's Hospital between 2001 and 2012 and were followed by The Complex Pediatric Therapies Follow-up Program (CPTFP).

The objectives of the study were to describe the epidemiology of AKI postoperatively in a single center cohort of children (age 0-5 years); identify potentially modifiable risk factors for developing AKI; and determine whether AKI was independently associated with clinically important outcomes at 30 days and one year post transplant.

Background AKI

AKI Definition

AKI is defined by a sudden decrease or loss of renal function where the kidney is unable to sufficiently excrete metabolic waste products, maintain fluid and electrolyte homeostasis, and regulate acid-base balance (Basu et al., 2011). Historically, a variety of clinical definitions of AKI were used in clinical investigation but this created difficulty in assessing the incidence of AKI or comparing outcomes (Goldstein & Devarajan, 2010).

In 2004, the Acute Dialysis Quality Initiative (ADQI) group published a classification system for AKI and failure based on serum creatinine (SCr) and urine output. There are 5 classification categories 1) at risk for renal dysfunction, 2) injury, 3) renal failure, 4) loss of kidney function, and, 5) end-stage renal failure, thus establishing the acronym RIFLE (Bellomo, Ronco, Kellum, Mehta, & Palevsky, 2004). In 2007, a modified RIFLE (pRIFLE) was developed for identifying AKI in pediatrics (Akcan-Arikan et al., 2007). Akcan-Arikan reported pRIFLE identified that AKI occurs in 82% of critically ill children within the initial week of PICU admission. In addition, pRIFLE criteria identified that 36% of children post cardiac surgery will developed AKI within 3 days (Zappitelli et al., 2009).

AKI in Children

The reported incidence of AKI in children varies from 10% to 82% dependent on the pediatric population. However, many of the recent literature studying AKI focuses on patients

requiring continuous renal replacement therapy (CRRT), indicating severe renal failure (Gulati & Bagga, 2011). Studies report that even a small rise in SCr is a risk for mortality in adults and pediatric patients (Goldstein & Devarajan, 2010; Gude et al., 2010). Clinical studies report that AKI is evident within three days post life threatening illness, cardiac surgery, extracorporeal membrane oxygenation (ECMO), or cardiopulmonary bypass (CPB)(Li et al., 2011)(Parikh et al., 2011)(Smith, Hardison, Worden, Fleming, & Taylor, 2009).

Causes of AKI

AKI can occur when arteriolar autoregulation can not compensate for a drop in perfusion pressure and there is a decrease in glomerular filtration rate (GFR). Conditions that can cause a decrease in perfusion pressure are hypovelemia, low cardiac output syndrome and extreme vasodilatation (Basu et al., 2011). Intra-renal AKI is caused by structural damage to the glomeruli, tubules, and blood vessels. Primarily this occurs when cellular changes are caused by nephotoxic substances or ischemia injury (Askenazi, 2011).

Burden

Pediatric heart transplantation is an established treatment approach for end-stage heart conditions like cardiomyopathies, failed palliation of complex congenital heart defects, and other complex cardiac malformations that are incompatible with life (de Jonge et al., 2008). However, long wait times to transplantation often causes low cardiac output syndrome and reduced renal function (Alonso, 2004). Several studies report that poor renal function prior to transplantation is associated with early postoperative mortality and CRRT (Jokinen et al., 2010; Tang et al., 2011). Renal dysfunction occurs in 20% of recipients prior to transplant and 3% need dialysis (Gandhi et al., 2011). Children undergoing heart transplantation often require technologically advanced medical interventions to bridge them to transplant and or recovery. It is not uncommon for

children in heart failure to require interventions to support their hemodynamic instability like ECMO or a Ventricular assist device (VAD), mechanical ventilator and inotropes (Bae et al., 2005). Complications of interventions and wait time to transplant result in 1 out of 9 children dying prior to transplant (Gandhi et al., 2011).

Surgical interventions and organ procurement produce long ischemic and cardiopulmonary bypass (CBP) times (Rylski et al., 2010). The literature reports the incidence of AKI in pediatric cardiac surgery post CBP is 35% to 52% (Chiravuri et al., 2011; Zappitelli et al., 2009). One retrospective study by Tang et al. (2011) reported pediatric heart transplant recipients who required RRT pre and post transplant had a significantly higher mortality in the postoperative cohort. Graft survival for the pre CRRT at one, five, and 10 years is 95%, 80%, and 74%, significantly above the post group (38%, 34%, and 32%; all $p<0.001$).

Long term renal dysfunction caused by immunosuppression therapy is well documented in pediatric heart transplant recipients (Dello Strologo et al., 2006). However, further study is needed to assess what prevalence of AKI exists in pediatric heart transplant recipients. Also, other risk factors perioperatively that might contribute to the AKI in pediatric heart transplant recipients need to be determined. Identification of risk factors could help support development of strategies for improved timing of interventions. Timing the intervention of dialysis at first clinical signs of AKI may improve morbidity prevalence and mortality.

The necessity of CBP in pediatric heart transplantation establishes risk of AKI. Retrospective studies indicate that AKI is associated with longer hospital stay and increased possibility of chronic renal disease (Sethi et al., 2011)(Zappitelli et al., 2009). The literature indicates that risk factors for AKI following CBP include complexity of congenital heart defect,

CBP duration, mechanical ventilation, young age, and small size (Chiravuri et al., 2011)(Li et al., 2011).

AKI in pediatric heart transplantation remains a clinical challenge. In adults, pre-operative risk factors are prior cardiac surgery, elevated SCr, and patient age. Additional risk factors include donor ischemic time, CBP, and early postoperative intravenous cyclosporine (Gude et al., 2010). Currently, there is limited evidence of AKI in pediatric heart transplant. A small number of retrospective studies report renal insufficiency (RI) perioperatively for pediatric heart transplant recipients (Bharat, Manlhiot, McCrindle, Pollock-BarZiv, & Dipchand, 2009; Lee et al., 2007; Mah et al., 2009; Phan, West, Stephens, & Hébert, 2003; Tang et al., 2011). However, these retrospective studies do not yield any data using current standardized definitions available of AKI (Akcan-Arikan et al., 2007).

Although risk factors for AKI have been well identified in pediatric cardiac surgery requiring CBP further research is necessary in the heart transplant population. Understanding the epidemiology of AKI using pRIFLE in association with transplant specific risk factors like immunosuppression therapy could help improve patient outcomes.

Patients and Methods

We evaluated data collected from an ongoing, muti-provincial (British Colombia, Alberta, Saskatchewan, and Manitoba) prospective cohort study performed by the Registry and Follow-up of Complex Pediatric Therapies Program of Western Canada (CRTFP). For ascertainment of the AKI outcome, hospital charts were retrospectively reviewed to record serum creatinine <24hours pre-transplant and then daily from day 1 to day 5 post-transplant. Data not available in the database was collected by chart review. Children undergoing a heart transplantation prior to 6 years of age between 2001 and 2012 at the Stollery Children's Hospital,

Edmonton, Alberta Canada, were eligible for inclusion. Subjects were excluded if they received a combined heart-lung transplant. Children requiring pre-transplant dialysis were also excluded from the primary analysis. Data was collected on subjects who were re-transplanted, but only data from the first transplant was included in analyses. Ethics board approval was obtained at each site and individual consent collected from a parent or guardian by CRTFP.

Pertinent pre, intra, and post-transplant data were evaluated. Data obtained for pre-transplant included age (0-28 days, 28d-1yr, 1yr-<6yr), sex, length, weight, diagnosis (defined as congenital heart disease (CHD) or cardiomyopathy (CMP), wait list days, ABO incompatibility, intravenous immunoglobin (IVIG), ventilation and inotrope use at the time of transplant, pretransplant ECMO or VAD, and peak lactate. Listing status was determined as outlined by Canadian Cardiac Transplant Network: status 1) at home; status 2) in hospital; status 3) requires intensive care (ICU), inotropes, ventricular assist device (VAD); status 3.5) ICU, high dose inotropes, and not a VAD candidate; status 4) mechanical ventilation, inotropes, and ECMO; status 4S) high panel reactive antibodies (PRA). Intraoperative variables were highest lactate, CPB time, use of deep hyperthermia circulatory arrest (DHCA), x-clamp time, donor ischemic time, IVIG use, and intraoperative milrinone. Postoperative variables included peak SCr, use of dialysis, ECMO, or VAD, peak lactate, and peak inotrope score, calculated by (dopamine + dobutamine) + (100) (epinephrine + norepinephrine + phynylephrine) (Morgan et al., 2013; Wernovsky et al., 1995). Variables were determined at 3 points postoperatively: postoperative day 1 (day 1), postoperative days 2-5 (day 2-5), and postoperative day 6 until hospital discharge (day 6+). Tacrolimus serum levels (*ug*/L) was recorded daily until post-transplant day 5.

AKI was defined based on the Pediatric Modified RIFLE criteria (pRIFLE) decrease in estimated creatinine clearance (eCCl) by > 25% from preoperative value (Akcan-Arikan et al.,

2007) . AKI was further stratified in Risk, Injury, Failure according to pRIFLE: For the purpose of this study, R=eCCL decreased by >25%; I=eCCl decreased by >50%; F=eCCl decreased by >75%. Estimated CCl was calculated using Schwartz formula: eCCl (ml/min/1.73m2) =kl/SCr, where k was a constant value 36.5, l is body length (cm) and SCr is expressed in millimoles per liter (mmol/L) (Schwartz et al., 2009). SCr was measured using Jaffe reaction. We did not use the urine criteria in pRIFLE as the validity of urine output is questionable as a high proportion of this cohort were are diuretics. (Appendix D)

Short-term outcomes were length of post-transplant ventilation days and PICU length of stay.

Statistical Analysis

All analysis were preformed using STATA 9.2 statistical software (StataCorp, College Station, Texas). Statistical significance was set at $P<0.05$. Continuous variables were examined for normal distribution and reported as mean± standard deviation (SD); variables with non-normal distributions were transformed for use in parametric tests. Categorical variables were expressed as percentage (%).

Logistic regression was preformed to determine univariate associations between risk factors and AKI. We used forward stepwise multiple logistic regression analysis (P value for entry into the model = 0.10; P value for removal=0.20) to evaluate independent risk factors for heart transplant associated AKI. The following risk factors were considered for the AKI prediction model: sex, age at transplant, number of days on the wait list, ECMO or VAD at the time of transplant, pre-transplant ventilation, pretransplant inotrope support (including milrinone), ABO incompatibility, baseline eCCl, donor ischemic time, use of DHCA, CPB time, peak OR lactate, day 1 inotrope score, day 1 peak lactate, and tacrolimus level on day 3 >15

*u*g/L. Given that listing status is a risk score consisting of multiple variables for which we had primary data, which were included in modeling, we did not enter status into the predictive models. All potential predictors were assessed for multicolinearity before inclusion in regression.

The association between AKI and short-term outcomes (PICU days and ventilation days) was evaluated by forward stepwise multiple regression. The models were adjusted for donor ischemic time, age at transplant, use of DHCA, CPB time, post-transplant ECMO, post-operative day 1 inotrope score, and peak day 1 lactate. Assumptions of all respective regression analyses were verified.

Results

Group characteristics

A total of 74 primary pediatric heart transplants were performed during the 11-year study period. Seventy-two were eligible for the study, one subject was excluded for missing data and the second received a heart-lung transplant. Six subjects were on dialysis at the time of transplant and not included in the primary analysis. Within the first year three subjects required re-transplant: 2 for poor function post-transplant requiring ECMO, and 1 for CAV.

Data of the remaining 66 subjects were evaluated (38 male; 34 female). Overall for those patients who developed AKI had a mean age of 70.5, median age of 48 days (range 1 day - 490 days). Six patients (9.1%)were recorded as status 1 at transplant; 7.8% (n=5) status 2; 26% (n=17) status 3; 1.5% (n=1) status 3.5; and 65% (43) were status 4. Twenty-five children (38.0%) were supported by ECMO or VAD prior to transplant.

Incidence of AKI

Table 4. Identifies the preoperative and intraoperative characteristics by AKI status. A total of 48 (73%) of the children developed AKI as identified by pRIFLE criteria. When stratified by risk, injury, and failure 16.7% (n=11) were categorized as AKI-R, 34.8% (n=23) were AKI-I, and 21.2% (n=14) were AKI-F. Within the cohort 64% (n=42) were ≤ 1 year of age. Postoperatively, 11 children received dialysis, 14% (n=9) starting on day 1 while an additional 3% (n=2) started between day 2-5. Of the 3 children that died in ICU, 2 had pRIFLE I AKI and 1 had pRIFLE 3 AKI.

Table 5 shows results of multivariate regression modeling of predictors of AKI. Three variables were independently associated with the development of AKI post transplant: 1) pre-transplant use of inotropes reduced AKI risk (adjusted odds ratio [aOR], 0.14; 95% confidence interval [CI], 0.03 - 0.69, $p = 0.02$); 2) ventilation at transplant predicted higher risk (aOR 8.60, CI 1.79 - 41.26, $p = 0.01$) and 3) eCCL, with higher glomerular filtration rate associated with higher risk (aOR 1.02, CI 1.00 - 1.04, $p = 0.03$). Although a day 3 tacrolimus value of > 15 *ug*/L was not statistically significant ($p = 0.09$), it was retained in the automated model as an important predictor. Sex, age at transplant, waitlist days, ABO-incompatibility, DHCA, pretransplant ECMO/VAD, donor ischemic time, OR peak lactate, CPB time, or day 1 peak lactate were not independent predictors.

Outcomes

Table 6 depicts the multivariate prediction model for the outcomes of total postoperative inverse PICU days and total postoperative inverse ventilation days. Acute kidney injury was independently associated with longer PICU stay, as was post-transplant ECMO and younger age. Peak day 1 lactate and day 1 inotrope score were retained in the automated model as important

predictors, with length of ICU stay increasing as lactate and inotrope score increased. Donor ischemic time, DHCA, and CBP time were not predictive of PICU length of stay. AKI did not appear to be an independent predictor of ventilation time although it was retained in modeling as an important variable (see Table 5). Postoperative ECMO, day 1 inotrope score, and younger age at transplant were independently associated with longer ventilation time. Higher peak day 1 lactate, use of DHCA, and longer CPB time were also retained in the automated model as important predictors of longer ventilation time, although not independently significant.

When we repeated the modeling in exploratory analysis, including those children on pretransplant dialysis, we found that AKI was an independent predictor of both PICU days and ventilation days (coefficient (inverse ICU days), -0.05 [CI], -0.09 - -0.02, $p = 0.00$; coefficient (inverse ventilation days), -0.06 [CI], -0.12 - -0.00, $p = 0.04$. When we evaluated those children with pRIFLE-R only, AKI remained a significant predictor of postoperative PICU days (coefficient (inverse ICU days), -0.06 [CI], -0.11 - - 0.02, $p = 0.01$, but not ventilation days. When adjusting for ventilation in forward stepwise regression model for PICU inverse days only AKI was retained in model, -0.02 [CI], -0.04 - - 0.00, p = 0.02.

In our cohort only 2 children had grade 1 rejection within 30 days post-transplant and 3 children who survived to 1 year had grade 1 rejection in the first year. Of the children with any AKI (n=48), 6% of the children (n=4) died at 30 days post-transplant. Of the total cohort, nine children did not survive to 1 year post-transplant. Of the children who died, 4.2% (n=3) had AKI-I and 8.3% (n=6) had AKI-F by pRIFLE.

Discussion

Limited evidence exists regarding the post-operative AKI epidemiology in pediatric heart transplantation. Our single-centered study demonstrated that AKI is common in children after heart transplant; 73% of pediatric heart transplant recipients developed AKI post-operatively, as identified by

pRIFLE criteria. Importantly, AKI is also an independent predictor of longer PICU admission and post-transplant ventilation time, independent of a number of other predictors and measures of illness severity.

The incidence of AKI in the critical care pediatric population varies depending on the cohort evaluated and ranges between 10% to 82% (Akcan-Arikan et al., 2007; Schneider, Khemani, Grushkin, & Bart, 2010). (Aydin et al., 2012; Morgan, Gill, Lam, & Joffe, 2013). The incidence of AKI in our study is in keeping with previously reported incidence in very sick children, as well as in adult heart transplant patients recipients (Türker, Zeyneloglu, Sezgin, Pirat, & Arslan, 2013), and demonstrates that children undergoing heart transplantation should be considered at high risk of experiencing this clinically important complication.

Lower preoperative eCCl appeared to be protective against AKI in our study. This is in keeping with observations from a number of other pediatric studies of children with congenital heart disease (Li et al., 2011; Naik et al., 2014; Zappitelli et al., 2008; Morgan et al., 2013). Children with end stage heart failure requiring transplant are likely to have changes in renal perfusion leading to varying degrees of renal ischemia, reflected by a lower eCCl. A period of nonlethal ischemia has been shown to protect the kidneys against ischemic AKI (Kaur et al., 2011; Munshi, Hsu, & Himmelfarb, 2011). Increased glomerular filtration could also result in higher exposure of the renal tubules to potential nephrotoxins in the intra-operative and early post-operative period, including free hemoglobin or toxic free iron generated during CPB. More simplistically, children with lower SCr (and hence higher eCCl) may have lower muscle mass and less optimal nutritional status (Kaur et al., 2011; Parikh et al., 2011), which might increase AKI risk. In addition, pre-operative fluid overload may lead to artifactual decrease in pre-operative SCr and also be associated with higher post-operative AKI risk. Further studies are needed to evaluate why lower pre-operative eGFR appears to reduce the risk of AKI

Being on inotropes preoperatively also was associated with lower risk of AKI, with children not on inotropes having 7 times the odds of sustaining AKI than those on inotropes. To our knowledge this is the only study to identify pre-transplant inotropes as a ‘protective’ factor in the development of AKI post transplant. Tang et al. (2011) revealed that inotropes at the time of listing for transplant was a risk factor

for renal failure perioperatively. Two other pediatric cardiac surgery studies identified that milrinone preoperatively or intraoperatively was a significant risk factor for the development of AKI (Aydin et al., 2012; Chiravuri et al., 2011). Although the small sample size of our cohort could have contributed to these differences, explanation may be contributed to our center's early in-hospital care and cardiovascular interventions. Studying pre-transplant inotropic levels could be important in ascertaining if low-dose inotropes provide improved hemodynamic stability and are protective against AKI.

In our study, donor ischemic time was not a significant risk factor for AKI in multivariate analysis. Two adult studies have evaluated donor ischemic time for AKI by RIFLE criteria and report conflicting results. (De Santo et al., 2011; Türker et al., 2013). We did demonstrate that children with AKI had longer mean donor ischemia time (see Table 1); the lack of statistical significance could be related to small sample size. Similarly, children with severe AKI (pRIFLE F) had longer CPB time (see Table 1) in keeping with previous studies of cardiac surgery associated AKI but statistical analysis did not identify it as an independent predictor; CPB time in the current cohort likely has less variability than in other cardiac surgery populations

Consistent with previous studies (Aydin et al., 2012; Chiravuri et al., 2011; Tang et al., 2011;Morgan et al., 2013;Naik et al., 2014), mechanical ventilation at time of transplant was a predictor for AKI in our cohort. The association of pulmonary-renal syndrome may explain these findings; pulmonary-renal syndrome is associated with a decrease in cardiac output, renal perfusion, GFR, and urine output (Koyner & Murray, 2008). The physiologic impact of positive pressure ventilation and its effects on kidney perfusion and function have also been well documented (Pannu & Mehta, 2004).

Although not statistically significant, day 3 postoperative tacrolimus greater than 15 *u*g/l was retained in this model and is likely biologically important in the prediction of AKI risk and needs further evaluation in a larger cohort. Using RIFLE criteria an adult study determined that cyclosporine was an independent risk factor for AKI following heart transplant (Türker et al., 2013). In

addition, calcineurin inhibitors have been identified as a risk factor for AKI post heart transplant (Gude et al. , 2010; Robinson, Shroff, & Spencer, 2013). Calcineurin inhibitors, particularly, CyA cause acute renal vasoconstriction 2-4 hours after peak levels, which decreases renal blood flow and GFR (Robinson, Shroff, & Spencer, 2013). Also acute fluctuation in absorption due to hemoinstability and poor kidney perfusion may precipitate toxicity levels and intermittent trough levels of tacrolimus may not be capturing true levels (Bloom,R D. 2006).

Similar to other studies, (Morgan et al., 2013; Pagowska-Klimek, Pychynska-Pokorska, Krajewski, & Moll, 2011; Tang et al., 2011; Zappitelli et al., 2009) our study identified that AKI was associated with longer length of PICU stay after heart transplant, independent of multiple other risk factors and markers of severity of illness. When we evaluated mild AKI alone (pRIFLE R), it also independently predicted this outcome. This is in keeping with other studies demonstrating that even small changes in SCr are associated with worse outcome (Morgan et al., 2013;Akcan-Arikan et al., 2007).

We did not include duration of ventilation in our primary model of length or PICU stay, as prolonged ventilation could be a causative relationship and negate the AKI effect. However, in exploratory analysis, AKI remained an important predictor after including ventilation time in the analysis; AKI was the only variable in addition to ventilation time that remained independently predictive of PICU length of stay. These finding are important as they support the development of strategies to avoid AKI in order to improve outcomes in this population. .

This study had several limitations. First, it is a relatively small cohort, so although we were able to identify some important predictors of AKI in this population, it is possible that other measured variables might improve our prediction in a larger cohort. Second, using data from a single center cohort may limit the generalizability of our results, and as with other cohort studies there may be unknown confounders that may be associated with the results. Third, fluid status is a significant factor to consider when interpreting AKI incidence and outcomes in critical care populations and merits further prospective evaluation in children having heart transplant. Variability in volume status either pre or postoperatively

might possibly have led to a misclassification of kidney injury due to intravascular volume depletion or fluid overload with its associated dilution of SCr. However, the changes in eCCL (and its associated definition of AKI) in this study were associated with important outcomes after cardiac transplant. Further prospective studies are needed to clarify the implications of volume balance in children undergoing heart transplant at risk of AKI.

This study has several strengths. It was an inception cohort study, with all data recorded prospectively except preoperative SCr. Preoperative SCr was measured very close to the time of transplant and was not estimated. None of the data for covariates in the prediction of CS-AKI were missing and all children had complete outcome and covariate data for assessing the association between AKI and short-term outcomes. We were able to perform multivariate analysis on a moderately sized cohort, and almost the entire population of young children undergoing cardiac transplant in Canada's 4 western provinces over a recent 12 year period were included.

Conclusion

AKI was common after pediatric heart transplant and was associated with important outcomes. As in other pediatric cardiac surgery populations, lower preoperative GFR was protective against postoperative AKI; the role of modified immune suppressive strategies in this context needs to be further evaluated. Although not statistically significant, elevated early postoperative tacrolimus is likely biologically important in the prediction of AKI risk and needs further evaluation in a larger cohort.

TABLE 4. Preoperative and intraoperative characteristics of "11-Year Cohort

		Mean±s.d.			*P* value
Characteristics	No AKI (N=18)	AKI-R (N=11)	AKI-I (N=23)	AKI-F (N-14)	
Continuous					
Weight (kg)	7.5 ± 4.0	7.5 ± 3.6	7.9 ± 3.5	9.1 ± 6.0	0.53
Length (cm)	68.0 ± 15.1	64 ± 23.2	70.2 ± 16.2	75.9 ± 66.0	0.85
Days on wait list	59.9 ± 43.7	45.7 ± 47.4	70.3 ± 85.0	123.0 ± 133.6	0.68
Pre eCCL	69.1 ± 33.2	68.8 ± 29.0	105.0 ± 32.6	81.7 ± 47.5	0.12
Donor ischemic time (mins)	286.5 ± 83.9	332.7 ±111.7	331.5 ± 99.8	311.0 ± 108.1	0.77
CPB (mins)	182.5 ± 66.8	155.9 ± 73.7	182.0 ± 81.2	234.1 ± 116.6	0.63
X-clamp (mins)	68.7 ± 60.8	54.6 ± 31.2	80.6 ± 79.9	75.9 ± 66.0	0.32
OR lactate	3. 2± 1.3	3.2 ± 1.8	3.0 ± 1.3	5.7 ± 3.2	0.78
D1 inotropes	17.9 ± 8.9	20.5 ± 10.9	17.5 ± 6.7	30.5 ± 15.1	0.03
D1 lactate	2.9 ± 1.8	2.7 ± 2.1	3.4 ± 2.2	7.5 ± 4.7	0.36
		N (%)			
Categorical					
Sex					
Male	8 (44.4)	7 (63.6)	12 (52.2)	7 (50.0)	0.05
Primary diagnosis					
CHD	10 (56.0)	5 (45.0)	12 (52.0)	13 (93.0)	0.69
CMP	8 (44.0)	6 (55.0)	11 (48.0)	1(7.0)	
Status at transplant					0.10
1	1 (5.6)	2 (18.2)	1 (4.3)	2 (14.3)	
2	2 (11.1)		3 (13.0)		
3	6 (33.3)	2 (18.2)	5 (22.0)	4 (28.6)	
4	0	0	1 (1.5)	0	
5	9 (50.0)	7(63.6)	13 (56.5)	8 (57.1)	
Age at transplant					0.85
0-28d	2 (11.1)	1 (9.1)	1 (4.3)	3 (21.4)	
28d-1y	10 (55.6)	7 (63.6)	13 (56.5)	5 (35.7)	
1y-5y	6 (33.3)	3 (27.3)	9 (39.1)	6 (42.9)	
ABO-incompatible	4 (22.2)	3 (27.3)	9 (39.1)	6 (42.9)	0.43
Vented at Transplant	6 (50.0)	8 (72.7)	9 (39.1)	8 (57.1)	0.09
Pre inotropes	14 (21.1)	8 (12.1)	10 (15.2)	6 (9.1)	0.49
Pre ECMO/VAD	5 (27.8)	4 (36.4)	9 (39.1)	7 (50.0)	0.05
DHCA	6 (33.3)	1 (9.1)	2 (8.7)	6 (42.9)	0.02

CMP, cardiomyopathy; *CHD*, congenital heart disease; *ECMO*, extracorporeal membrane oxygenation; *VAD*, ventricular assist device; *eCCL*, estimated creatinine clearance; *CPB*, cardiopulmonary bypass; *DHCA*, deep hypothermic circulatory arrest; *72 heart transplants and 6 on dialysis pretransplant.

Table 5. Independently predictive variables for any AKI by pRIFLE following adjustment*

	Adjusted odds ratio (95% CI)	*P* value
Pretransplant inotropes	0.14 (0.03, 0.69)	0.02
Pretransplant ventilation	8.59 (1.79, 41.26)	0.01
Pretransplant eCCL	1.02 (1.00, 1.04)	0.03
D3 tacrolimus >15	7.88 (0.75, 83.00)	0.09

eCCL, estimated creatinine clearance. * Adjusted Sex, age at transplant, waitlist days, abo-incompatibility, DHCA, pretransplant ECMO/VAD, pretransplant ventilation, pretransplant inotropes, pretransplant eCCL, donor ischemic time, or peak lactate, cardiopulmonary bypass; day 1 peak lactate. model excluded pretransplant dialysis children (n=66).

Table 6: Multivariate regression model for predicting inverse PICU days and inverse ventilation days

PICU days	Coefficient	95% CI	P value
AKI pRIFLE	-0.04	(-0.08 - - 0.00)	0.04
Post-transplant ECMO	-0.05	(-0.10 - -0.01)	0.03
Age at Transplant (1 to 6yr vs <28 do)	0.04	(0.00 - 0.07)	0.04
Peak day 1 lactate	-0.00	(-0.01 – 0.00)	0.11
Day 1 inotrope score	-0.00	(-0.00 – 0.00)	0.08

Ventilation days	Coefficient	95% CI	P value
AKI pRIFLE	-0.04	(-0.10 – 0.02)	0.18
Post-transplant ECMO	-0.09	(-0.16 - -0.01)	0.03
Day 1 inotrope score	-0.00	(-0.01 - -0.00)	0.01
Age at Transplant (1 to 6yr vs <28 day)	-0.09	(-0.03 - - 0.15)	0.00
Peak day 1 lactate	-0.01	(-0.01 - 0.00)	0.18
DHCA	-0.05	(-0.12 – 0.01)	0.12
CPB time	-0.00	(-0.00 – 0.00)	0.18

ECMO, extracorporeal membrane oxygenation; *DHCA*, deep hypothermic circulatory arrest; *CPB, cardiopulmonary bypass time Excluded pre-transplant dialysis (n=66*) ICU days and vent days inverse transformed for non-normal distribution

References

Akcan-Arikan, A., Zappitelli, M., Loftis, L. L., Washburn, K. K., Jefferson, L. S., & Goldstein, S. L. (2007). Modified RIFLE criteria in critically ill children with acute kidney injury. *Kidney International, 71*(10), 1028-1035.

Alonso, E. M. (2004). Long-term renal function in pediatric liver and heart recipients. *Pediatric Transplantation, 8*(4), 381-385.

Askenazi, D. (2011). Evaluation and management of critically ill children with acute kidney injury. *Current Opinion in Pediatrics, 23*(2), 201-207. doi:10.1097/MOP.0b013e328342ff37

Aydin, S. I., Seiden, H. S., Blaufox, A. D., Parnell, V. A., Choudhury, T., Punnoose, A., & Schneider, J. (2012). Acute kidney injury after surgery for congenital heart disease. *The Annals of Thoracic Surgery, 94*(5), 1589-1595. doi:10.1016/j.athoracsur.2012.06.050

Bae, J., Frischer, J. S., Waich, M., Addonizio, L. J., Lazar, E. L., & Stolar, C. J. H. (2005). Extracorporeal membrane oxygenation in pediatric cardiac transplantation. *Journal of Pediatric Surgery, 40*(6), 1051-1056.

Basu, R. K., Devarajan, P., Wong, H., & Wheeler, D. S. (2011). An update and review of acute kidney injury in pediatrics. *Pediatric Critical Care Medicine: A Journal of the Society of Critical Care Medicine and the World Federation of Pediatric Intensive and Critical Care Societies, 12*(3), 339-347.

Bharat, W., Manlhiot, C., McCrindle, B. W., Pollock-BarZiv, S., & Dipchand, A. I. (2009). The profile of renal function over time in a cohort of pediatric heart transplant recipients. *Pediatric Transplantation, 13*(1), 111-118.

Bloom, R. D., & Doyle, A. M. (2006). Kidney disease after heart and lung transplantation. *American Journal of Transplantation: Official Journal of the American Society of Transplantation and the American Society of Transplant Surgeons, 6*(4), 671-679.

Network, C.C.T. Cardiac Transplantation: Eligibility and Listing Criteria in Canada 2012. in CCTN Meeting. 2012. Toronto: CCTN.

Chiravuri, S. D., Riegger, L. Q., Christensen, R., Butler, R. R., Malviya, S., Tait, A. R., & Voepel-Lewis, T. (2011). Factors associated with acute kidney injury or failure in children undergoing cardiopulmonary bypass: A case-controlled study. *Paediatric Anaesthesia, 21*(8), 880-886. doi:10.1111/j.1460-9592.2011.03532.x

de Jonge, N., Kirkels, J. H., Klöpping, C., Lahpor, J. R., Caliskan, K., Maat, A. P. W. M., . . . Balk, A. H. M. M. (2008). Guidelines for heart transplantation. *Netherlands Heart Journal: Monthly Journal of the Netherlands Society of Cardiology and the Netherlands Heart Foundation, 16*(3), 79-87.

De Santo, L. S., Romano, G., Amarelli, C., Maiello, C., Baldascino, F., Bancone, C., . . . Nappi, G. (2011). Implications of acute kidney injury after heart transplantation: What a surgeon should know. *European Journal of Cardio-Thoracic Surgery: Official*

Journal of the European Association for Cardio-Thoracic Surgery, 40(6), 1355-1361. doi:10.1016/j.ejcts.2011.02.068

Dello Strologo, L., Parisi, F., Legato, A., Pontesilli, C., Pastore, A., Ravà, L., . . . Rizzoni, G. (2006). Long-term renal function in heart transplant children on cyclosporine treatment. *Pediatric Nephrology (Berlin, Germany), 21*(4), 561-565.

Dent, C. L., Ma, Q., Dastrala, S., Bennett, M., Mitsnefes, M. M., Barasch, J., & Devarajan, P. (2007). Plasma neutrophil gelatinase-associated lipocalin predicts acute kidney injury, morbidity and mortality after pediatric cardiac surgery: A prospective uncontrolled cohort study. *Critical Care (London, England), 11*(6), R127-R127.

Gandhi, R., Almond, C., Singh, T. P., Gauvreau, K., Piercey, G., & Thiagarajan, R. R. (2011). Factors associated with in-hospital mortality in infants undergoing heart transplantation in the united states. *Journal of Thoracic & Cardiovascular Surgery, 141*(2), 531; Feb-536.

Goldstein, S. L., & Devarajan, P. (2010). Pediatrics: Acute kidney injury leads to pediatric patient mortality. *Nature Reviews.Nephrology, 6*(7), 393-394.

Gude, E., Andreassen, A. K., Arora, S., Gullestad, L., Grov, I., Hartmann, A., . . . Simonsen, S. (2010). Acute renal failure early after heart transplantation: Risk factors and clinical consequences. *Clinical Transplantation, 24*(6), E207-13.

Gulati, A., & Bagga, A. (2011). Management of acute renal failure in the pediatric intensive care unit. *Indian Journal of Pediatrics, 78*(6), 718-725.

Jokinen, J. J., Tikkanen, J., Kukkonen, S., Hammainen, P., Lommi, J., Sipponen, J., & Lemstrom, K. B. (2010). Natural course and risk factors for impaired renal function during the first year after heart transplantation. *Journal of Heart & Lung Transplantation, 29*(6), 633-640.

Kaur, S., Jain, S., Saha, A., Chawla, D., Parmar, V. R., Basu, S., & Kaur, J. (2011). Evaluation of glomerular and tubular renal function in neonates with birth asphyxia. *Annals of Tropical Paediatrics, 31*(2), 129-134. doi:10.1179/146532811X12925735813922

Krawczeski, C. D., Goldstein, S. L., Woo, J. G., Wang, Y., Piyaphanee, N., Ma, Q., . . . Devarajan, P. (2011). Temporal relationship and predictive value of urinary acute kidney injury biomarkers after pediatric cardiopulmonary bypass. *Journal of the American College of Cardiology, 58*(22), 2301-2309.

Koyner, J. L., & Murray, P. T. (2008). Mechanical ventilation and lung-kidney interactions. *Clinical Journal of the American Society of Nephrology: CJASN, 3*(2), 562-570. doi:10.2215/CJN.03090707

Lee, C. K., Christensen, L. L., Magee, J. C., Ojo, A. O., Harmon, W. E., & Bridges, N. D. (2007). Pre-transplant risk factors for chronic renal dysfunction after pediatric heart transplantation: A 10-year national cohort study. *Journal of Heart & Lung Transplantation, 26*(5), 458-465.

Li, S., Krawczeski, C. D., Zappitelli, M., Devarajan, P., Thiessen-Philbrook, H., Coca, S. G., . . . Parikh, C. R. (2011). Incidence, risk factors, and outcomes of acute kidney

injury after pediatric cardiac surgery: A prospective multicenter study. *Critical Care Medicine, 39*(6), 1493-1499.

Mah, D., Singh, T. P., Thiagarajan, R. R., Gauvreau, K., Piercey, G. E., Blume, E. D., . . . Almond, C. S. (2009). Incidence and risk factors for mortality in infants awaiting heart transplantation in the USA. *Journal of Heart & Lung Transplantation, 28*(12), 1292-1298.

Mehta, R. L., Kellum, J. A., Shah, S. V., Molitoris, B. A., Ronco, C., Warnock, D. G., & Levin, A. (2007). Acute kidney injury network: Report of an initiative to improve outcomes in acute kidney injury. *Critical Care (London, England), 11*(2), R31-R31.

Morgan, C. J., Gill, P. J., Lam, S., & Joffe, A. R. (2013). Peri-operative interventions, but not inflammatory mediators, increase risk of acute kidney injury after cardiac surgery: A prospective cohort study. *Intensive Care Medicine, 39*(5), 934-941. doi:10.1007/s00134-013-2849-4

Morgan, C. J., Zappitelli, M., Robertson, C. M. T., Alton, G. Y., Sauve, R. S., Joffe, A. R., . . . Rebeyka, I. M. (2013). Risk factors for and outcomes of acute kidney injury in neonates undergoing complex cardiac surgery. *The Journal of Pediatrics, 162*(1), 120-7.e1. doi:10.1016/j.jpeds.2012.06.054

Munshi, R., Hsu, C., & Himmelfarb, J. (2011). Advances in understanding ischemic acute kidney injury. *BMC Medicine, 9*, 11-11. doi:10.1186/1741-7015-9-11

Naik, S., Sharma, J., Yengkom, R., Kalrao, V., & Mulay, A. (2014). *Acute kidney injury in critically ill children: Risk factors and outcomes*. India: Indian Society of Critical Care Medicine. doi:10.4103/0972-5229.128701

Pagowska-Klimek, I., Pychynska-Pokorska, M., Krajewski, W., & Moll, J. J. (2011). Predictors of long intensive care unit stay following cardiac surgery in children. *European Journal of Cardio-Thoracic Surgery: Official Journal of the European Association for Cardio-Thoracic Surgery, 40*(1), 179-184.

Palmieri, T., Lavrentieva, A., & Greenhalgh, D. (2009). An assessment of acute kidney injury with modified RIFLE criteria in pediatric patients with severe burns. *Intensive Care Medicine, 35*(12), 2125-2129.

Parikh, C. R., Devarajan, P., Zappitelli, M., Sint, K., Thiessen-Philbrook, H., Li, S., . . . Krawczeski, C. D. (2011). Postoperative biomarkers predict acute kidney injury and poor outcomes after pediatric cardiac surgery. *Journal of the American Society of Nephrology: JASN, 22*(9), 1737-1747.

Phan, V., West, L. J., Stephens, D., & Hébert, D. (2003). Renal complications following heart transplantation in children: A single-center study. *American Journal of Transplantation: Official Journal of the American Society of Transplantation and the American Society of Transplant Surgeons, 3*(2), 214-218.

Plotz, F. (2010). RIFLE criteria in the pediatric intensive care unit. *Critical Care Medicine, 38*(11), 2270-2271.

Pannu, N., & Mehta, R. L. (2004). Effect of mechanical ventilation on the kidney. *Best Practice & Research.Clinical Anaesthesiology, 18*(1), 189-203.

Robinson, P. D., Shroff, R. C., & Spencer, H. (2013). Renal complications following lung and heart-lung transplantation. *Pediatric Nephrology (Berlin, Germany), 28*(3), 375-386. doi:10.1007/s00467-012-2200-2

Rylski, B., Berchtold-Herz, M., Olschewski, M., Zeh, W., Schlensak, C., Siepe, M., & Beyersdorf, F. (2010). Reducing the ischemic time of donor hearts will decrease morbidity and costs of cardiac transplantations. *Interactive Cardiovascular & Thoracic Surgery, 10*(6), 945-947.

Schneider, J., Khemani, R., Grushkin, C., & Bart, R. (2010). Serum creatinine as stratified in the RIFLE score for acute kidney injury is associated with mortality and length of stay for children in the pediatric intensive care unit. *Critical Care Medicine, 38*(3), 933-939. doi:10.1097/CCM.0b013e3181cd12e1

Schwartz, G. J., Muñoz, A., Schneider, M. F., Mak, R. H., Kaskel, F., Warady, B. A., & Furth, S. L. (2009). New equations to estimate GFR in children with CKD. *Journal of the American Society of Nephrology: JASN, 20*(3), 629-637. doi:10.1681/ASN.2008030287

Sethi, S. K., Goyal, D., Yadav, D. K., Shukla, U., Kajala, P. L., Gupta, V. K., . . . Juneja, A. (2011). Predictors of acute kidney injury post-cardiopulmonary bypass in children. *Clinical and Experimental Nephrology, 15*(4), 529-534.

Smith, A. H., Hardison, D. C., Worden, C. R., Fleming, G. M., & Taylor, M. B. (2009). Acute renal failure during extracorporeal support in the pediatric cardiac patient. *ASAIO Journal (American Society for Artificial Internal Organs: 1992), 55*(4), 412-416.

Tang, L., Du, W., & L'Ecuyer, T. J. (2011). Perioperative renal failure in pediatric heart transplant recipients: Outcome and risk factors. *Pediatric Transplantation, 15*(4), 430-436.

Türker, M., Zeyneloglu, P., Sezgin, A., Pirat, A., & Arslan, G. (2013). RIFLE criteria for acute kidney dysfunction following heart transplantation: Incidence and risk factors. *Transplantation Proceedings, 45*(10), 3534-3537. doi:10.1016/j.transproceed.2013.08.100

Washburn, K. K., Zappitelli, M., Arikan, A. A., Loftis, L., Yalavarthy, R., Parikh, C. R., . . . Goldstein, S. L. (2008). Urinary interleukin-18 is an acute kidney injury biomarker in critically ill children. *Nephrology, Dialysis, Transplantation: Official Publication of the European Dialysis and Transplant Association - European Renal Association, 23*(2), 566-572.

Wernovsky, G., Wypij, D., Jonas, R. A., Mayer, J. E., J., Hanley, F. L., Hickey, P. R., . . . Wessel, D. L. (1995). Postoperative course and hemodynamic profile after the arterial switch operation in neonates and infants. A comparison of low-flow cardiopulmonary bypass and circulatory arrest. *Circulation, 92*(8), 2226-2235.

Zappitelli, M., Bernier, P., Saczkowski, R. S., Tchervenkov, C. I., Gottesman, R., Dancea, A., . . . Alkandari, O. (2009). A small post-operative rise in serum creatinine predicts acute kidney injury in children undergoing cardiac surgery. *Kidney International, 76*(8), 885-892.

Zappitelli, M., Moffett, B. S., Hyder, A., & Goldstein, S. L. (2011). Acute kidney injury in non-critically ill children treated with aminoglycoside antibiotics in a tertiary healthcare centre: A retrospective cohort study. *Nephrology, Dialysis, Transplantation: Official Publication of the European Dialysis and Transplant Association - European Renal Association, 26*(1), 144-150. doi:10.1093/ndt/gfq375

Zappitelli, M., Parikh, C. R., Akcan-Arikan, A., Washburn, K. K., Moffett, B. S., & Goldstein, S. L. (2008). Ascertainment and epidemiology of acute kidney injury varies with definition interpretation. *Clinical Journal of the American Society of Nephrology: CJASN, 3*(4), 948-954. doi:10.2215/CJN.05431207

Appendix A

Literature Search

Search Terms	Search Options	Action	
S5	("heart transplantation OR heart transplant*" OR (MH "Heart Transplantation+")) AND (S1 and S2)	**Limiters** - English Language **Narrow by SubjectAge:** - All Child: 0-18 years **Search modes** - Find all my search terms	View Results (162) View Details Edit
S4	("heart transplantation OR heart transplant*" OR (MH "Heart Transplantation+")) AND (S1 and S2)	**Limiters** - English Language **Search modes** - Find all my search terms	View Results (932) View Details Edit
S3	("heart transplantation OR heart transplant*" OR (MH "Heart Transplantation+")) AND (S1 and S2)	**Search modes** - Find all my search terms	View Results (1000) View Details Edit
S2	"heart transplantation OR heart transplant*" OR (MH "Heart Transplantation+")	**Search modes** - Find all my search terms	View Results (30620) View Details Edit
S1	((MH "Acute Kidney Injury+") OR (MH "Renal Insufficiency+")) OR TI renal OR TI nephr* OR TI kidney	**Search modes** - Find all my search terms	View Results (435414) View Details Edit

Appendix B

Literature review summery

Author	Year	Population	Renal Insufficiency	Incidence of Renal Insufficiency	Risk Factors	Mortality	Conclusion
Tang et al Perioperative Renal Failure	2011	OPTN ≤18 yrs 2 groups n=3595	Dialysis or SCR >2.5mg/dl	Pre TX 31(1%) Post TX 223(7%) Post TX of the 223 1,5,7 yrs 54(45%),7(16%),10(28%)	ECMO Ventilation CHD Infection Inotropes	1,5 , 10 years PRE 3%, 10% 16% Both 62%, 72%,72% POST 62%,64%,65%	Post dialysis patients had decreased survival rate at 30 days & 1,5,10 yr
Gandhi et al In-hospital mortality	2011	OPTN Infants <12mths n=730	CC <40ml/min/1.73^{m2}	Pre TX 161(23%) Post TX 251(34%)	CHD ECMO Ventilation Renal Function	In hospital 82(11.2%)	1 out 9 will die before TX. Mortality increased with ECMO, renal failure, CHD
Lee et al Pre TX risk for CRD post TX	2007	OPTN ≤18 yrs 2 subsets n =2032 & n=1055	SCr >2.5mg/dl	Post TX 10years 327(16%)	Pre TX dialysis HCM African race ECMO	Not reported	Post Tx patients with chronic kidney injury 9 times higher mortality
Sachdeva et al Determinants of renal function	2003	Single center ≤18 yrs n=77	Dialysis or GFR <75ml/min/1.73 ^{m2}	Pre TX 25(33%) Post TX 1,3,5 yrs 13(17%),16(21%), 20(26%)	African race Younger age RI at 6 mths Time before TX	Post TX 17(22%)	Risk factors for renal injury for younger age at TX, renal injury at 6mths & African -American
Phan et al Post TX renal complications	2003	Single center < 18 yrs n=41	GFR for <2mths <60ml/min/1.73 ^{m2} >2mths <80ml/min/1.73 ^{m2} ARD 2-fold ↑ in baseline Cr	Post ARD 12(29%) Pre GFR ↓ 42% Post GFR ↓ 7%	Abnormal GFR	Excluded from study children that died in first year n=13	GFR did not decrease in 1st year, but increased, then stabilized. ARD episodes are frequent in post TX

Abbreviations: OPTN, Organ Procurement and Transplantation Network; mths, months; PFR; perioperative renal failure; TX, transplant; CHD, congenital heart disease; ECMO, extracorporeal membrane oxygenation; RI, renal insufficiency; ICU, intensive care unit; CRD, chronic renal disease; SCr, serum creatinine; GFR, globular filtration rate; CHD, Congenital heart disease; HCM, hypertrophy cardiomyopathy; ARD, acute renal dysfunction; CC, creatinine clearance:

Appendix C

*generated through chart review; all others through CPTP Registry

I. Risk Factors

1.1 Pre-transplant Clinical and Demographic Characteristics

Age (years)		
Weight (kg)		
Height (cm)		
Gender		F M
Primary Diagnosis	Congenital Heart Disease	Yes No
	Cardiomyopathy/ Myocarditis	Yes No

1.2 Pre-operative AKI (pRIFLE)

	0 (No AKI)	1 (Risk)	2 (Injury)	3 (Failure)
Collected within 24hours to Transplant				

1.3 Pre-operative Variables

*Last SCr drawn prior to surgery (µmol/l)	
Last lactate drawn prior to surgery (mmol/l)	
Ventilator	Yes No
ECMO	Yes No
PICU	Yes No
Previous Cardiac Surgery	Yes No
*Milrinone	Yes No
*Inotrope Score	
Dopamine	
Dobutamine	
Epinephrine	
Norepinephrine	
Phenylephrine	
How long on waitlist to transplant (days)	
Listing Status	1 2 3 3.5 4S 4
PRA	Yes No
DSA	Yes No

Desensitization		Yes	No
X-match at listing			
*Fluid balance over the 24hr prior to transplant		Positive	Negative
*Urine output in the day prior to transplant (ml/kg/hr over 6hrs)			
Dialysis	None	Yes	No
	CRRT	Yes	No
	PD	Yes	No

Intra-operative Variables

CPB time (minutes)	
DHCA (minutes)	
Aortic cross clamp time (minutes)	
Donor heart ischemic time (minutes)	
*Milrinone (during CBP)	Yes No

Post-operative Variables (up to day 5):

	Day 1	Day 2	Day 3	Day 4	Day 5
*Milrinone	Y N	Y N	Y N	Y N	Y N
*Inotrope Score (mg/kg/min)					
Dopamine					
Dobutamine					
Epinephrine					
Norepinephrine					
Phenylephrine					
Tacrolimus Trough level (PO)					
Mean level (IV)					
Other immunosuppression *MMF *steroid	Y N	Y N	Y N	Y N	Y N
*Peak Lactate (mmol/l)					
*Peak daily SCr (measured using the Jaffe reaction)					
*Urine output					

mls/kg/hr (collected every 24hr at 0659)					

Post-operative Variables (Number of days and/or 1st day)

Dialysis (first day)	Y N
PD	Y N
CRRT	Y N
*First day of negative fluid balance	
Daily Fluid balance	
Chest open (number of days)	
ECMO (number of days & first day)	
VAD (number of days & first day)	
Ventilation (number of days & first day)	

2. Outcomes

2.1 AKI (pRIFLE) Day 1-5

	0 (No AKI)	1 (Risk)	2 (Injury)	3 (Failure)
Postoperative day 1				
Postoperative day 2				
Postoperative day 3				
Postoperative day 4				
Postoperative day 5				

2.2 In-hospital outcomes:

Days in PICU (postoperative)	
Days in Hospital (postoperative)	
Ventilator days (postoperative)	
Number of postoperative days at death	

2.3 Outcomes at 30 Days

Death	Y	N
Graft rejection grade 2R or higher or if associated with clinical impairment	Y	N
Re-transplant	Y	N

2.4 Outcomes at 1 Year

Death	Y	N
Re-transplant	Y	N
Graft vasculapathy (CAV)	Y	N
PTLD	Y	N
Graft rejection grade 2R or higher or if associated with clinical impairment	Y	N

Appendix D

RIFLE and pRIFLE

Scheme	Stage	Creatinine Criteria	Urine Output Criteria
RIFLE	R	↑ ≥1.5x or ↓ GFR ≥ 25%	<0.5 ml/kg/hr for 6 hrs
	I	↑ ≥2x or ↓ GFR ≥ 50%	<0.5 ml/kg/hr for 12 hrs
	F	↑ ≥3x or sCR >350µmol/L	<0.3 ml/kg/hr for 24 hrs or anuria for 12 hrs
	L	Persistent failure >4 wks	
	E	Persistent failure >3mths	
pRIFLE	R	eCCL ↓ ≥25%	<0.5 ml/kg/hr for 8 hrs
	I	eCCL ↓ ≥50%	<0.5 ml/kg/hr for 12 hrs
	F	eCCL ↓ ≥75% or eCCL <35ml/min/1.73^2	<0.3 ml/kg/hr for 24 hrs or anuria for 12 hrs
	L	Persistent failure >4 wks	
	E	Persistent failure >3mths	

RIFLE, risk, injury, failure, loss, end stage; pRIFLE, pediatric RIFLE; GFR, glomerular filtration rate; SCr, serum creatinine; eCCL, estimated creatinine clearance.

(Akcan-Arikan et al., 2007; Bellomo et al., 2004).