Original Article

Enhanced Hemolysis in Pediatric Patients Requiring Extracorporeal Membrane Oxygenation and Continuous Renal Replacement Therapy

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Purpose: Hemolysis during extracorporeal membrane oxygenation (ECMO) may be associated with the development of hemoglobinuria (Hb) nephropathy and acute renal failure. For patients requiring ECMO, continuous renal replacement therapy (CRRT) can be simultaneously performed by attaching a hemofilter to the ECMO circuit, thereby shunting part of the ECMO blood flow through the hemofilter. However, the possibility that CRRT may further enhance hemolysis (and the risk of Hb nephropathy) in patients on ECMO has not been previously investigated.

Methods: Medical records of 42 children (1 day–12 years old) who required ECMO (ECMO group, *n*=25) or ECMO and CRRT (ECMO+CRRT group, *n*=17) after cardiac surgery were reviewed.

Results: Forty-one out of 42 patients had elevated plasma-free hemoglobin (FHb) on the first day of ECMO. For all subjects, peak change (mean±SD) in FHb (Peak%C-FHb, 83.6±183%) correlated with serum lactic dehydrogenase (150±324%, *r*=0.49, *p*<0.05) and marginally with ECMO blood flow rate (BFR) (Peak%C-BFR, 36.8±51.0%, *r*=0.29, *p*=0.06). Compared with the ECMO group, the ECMO+CRRT group had a higher Peak%C-FHb (160±259%, *p*<0.05) and Peak%C-BFR (62±64%, *p*<0.05). Also, there was a significant increase in FHb one day after the initiation of CRRT compared with the level prior to CRRT (73.3±49.2 vs. 50.0±30.3 mg/dL, respectively, *p*=0.012). Serum creatinine (but not blood urea nitrogen) was significantly higher in the ECMO+CRRT group compared with the ECMO group. The percent change in serum creatinine during ECMO did not correlate with Peak%C-FHb in the ECMO group.

Conclusion: Our findings suggest that there is enhanced hemolysis during combined ECMO and CRRT compared with ECMO alone. However, the clinical impact of increased hemolysis on renal function in patients receiving ECMO with or without CRRT remains to be determined.

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Key words: continuous renal replacement therapy, hemolysis, extracorporeal membrane oxygenation, cardiac surgery, children

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Introduction

Hemolysis is common during extracorporeal membranous oxygenation (ECMO) support.[1)] Factors such as shear stress,[2,3)] physical properties of the ECMO circuit,[4)] sublethal damage to erythrocytes,[5)] the roller pump,[6)] changes in blood volume,[7)] and pressure changes within the oxygenator[8)] have all been implicated for its development.

As a result of hemolysis, the level of plasma-free hemoglobin (FHb) rises by as much as 10- to 25-fold after 24 h of ECMO support.[9] Hemolysis has also been reported in other types of extracorporeal therapies, such as continuous renal replacement therapy (CRRT)[10] and ventricular assist devices.[11] Under acidic conditions, the excess circulating FHb may precipitate within renal tubules and result in hemoglobinuria nephropathy.[12] This may further impair renal function and delay recovery.

When dialysis is indicated in patients requiring ECMO support, CRRT is the preferred modality because it is generally well tolerated from a hemodynamic perspective. CRRT is usually performed by attaching a hemofilter to the ECMO circuit so that a portion of the heparinized blood in the circuit can be shunted into the hemofilter for dialysis therapy. However, it is not known if the addition of CRRT exacerbates ECMO-induced hemolysis and the possibility for further kidney damage. This may be an important clinical consideration, since these patients are at constant risk of systemic acidosis, intravascular volume depletion, and decreased renal perfusion that may precipitate the development of hemoglobinuria nephropathy.[12] To test the hypothesis that the addition of CRRT to ECMO circuit can aggravate ECMO-induced hemolysis, we reviewed our experience in pediatric patients requiring ECMO and CRRT compared to those requiring ECMO alone after congenital heart disease surgery.

Methods

The Institutional Review Board of the University of Michigan Health System reviewed and approved the waiver of informed consent for this study. Medical records of pediatric patients undergoing cardiac surgery from 1993 to 2001 who required ECMO support postoperatively were reviewed. Indications for ECMO included systolic and/or diastolic ventricular dysfunction, respiratory failure, or an occluded systemic-to-pulmonary artery shunt. The ECMO circuit consisted of a servo-regulated roller pump, membrane lung, heat exchanger, and 0.25-inch caliber tubing.[13] Patients who required ECMO intraoperatively or in the immediate postoperative period, or who experienced a precipitous circulatory decompensation or cardiac arrest, underwent transthoracic cannulation via the right atrium and ascending aorta. The internal jugular vein and carotid artery were the preferred sites of cannulation among patients who received ECMO support beyond the immediate postoperative period. Prophylactic antibiotics were routinely given to all patients. Blood flow rate (BFR) varied from 80 to 120 mL/min per kg. All patients were anticoagulated with heparin sulfate to achieve an activated clotting time of 180 to 200 s. Platelets were kept greater than 100,000/mm^3 and the hematocrit greater than 35% by transfusion. CRRT was performed when clinically indicated, using an in-line Renaflo II Hemofilter (Minnitech Inc., Minneapolis, MN, USA) for patients greater than 10 kg body weight and a Mini-Plus filter (Millipore Corp., Billerica, MA, USA) for patients weighing ≤ 10 kg. The rate of fluid removal via CRRT was less than 0–3 mL/kg/h, depending on the patient's clinical condition. The countercurrent dialysate flow rate was set at 2,000 mL/1.73 m^2/h. The hemofilter was changed every 48 to 72 h. ECMO was discontinued after a successful weaning trial (with cardiopulmonary stability for several hours) or if it was determined that irreversible organ injury precluded survival. Blood samples for FHb were drawn every morning from the port proximal to the oxygenator. FHb was measured using the catalytic action of hemoglobin on the oxidation of 3,3′,5,5′-Tetramethylbenzidine (Sigma, St. Louis, MO, USA) by hydrogen peroxide. The resulting rate of color formation was measured spectrophotometrically at 550 nm. PRISM III[14] was used to estimate the mortality risk when ECMO was initiated. Because our patients were systemically heparinized, prothrombin time and partial thromboplastin time were not included in the PRISM III scores.

Determination of changes in variables

The peak percent changes in FHb (Peak%C-FHb), serum lactic dehydrogenase (Peak%C-LDH), ECMO flow (Peak%C-BFR), and blood pH (Peak%C-pH) were calculated as follows:

$$\frac{(\text{maximum level during ECMO} - \text{level on initiation of ECMO}) \times 100}{(\text{level on initiation of ECMO})}$$

The change in serum creatinine (End%C-Scr) and blood urea nitrogen (BUN) (End%C-BUN) during ECMO support were calculated as:

$$\frac{(\text{level on the final day of ECMO} - \text{level on initiation of ECMO}) \times 100}{(\text{level on initiation of ECMO})}$$

Statistics

The Mann-Whitney *U* test was used to compare continuous data between the groups. Pearson correlation with Bonferroni correction was used to examine correlations between 2 groups with continuous data. A p value ≤ 0.05

was considered statistically significant. All data are presented as mean ± SD.

Results

From 1993 to 2001, medical records from 42 children who required ECMO support after cardiac surgery for congenital heart disease were available for review. There were 13 males and 29 females with a mean age of 8.29 days (range 1 day to 12 years old) and a weight of 6.00±8.01 kg (range 2.2–51.3 kg) at the time of operation. The cardiac diagnoses included d-transposition of great arteries (*n*=7), total anomalous pulmonary venous connection (*n*=4), truncus arteriosus (*n*=1), hypoplastic left heart syndrome (*n*=13), tetralogy of Fallot (*n*=6), interrupted aortic arch (*n*=4), double outlet right ventricle (*n*=1), hypoplastic aortic arch (*n*=1), tricuspid atresia (*n*=1), atrioventricular septal defect (*n*=2), and pulmonary arteriovenous malformation (*n*=2). The duration of ECMO support was 5.14±2.54 days (range 2–13 days). On the first day of ECMO support, 41 of 42 patients had elevated FHb levels (normal: 1–8 mg/dL). As shown in Table 1, Peak%C-FHb correlated with the peak level of BFR (Peak BFR, mL/min/kg) and Peak%C-LDH during ECMO support. Both Peak%C-FHb and Peak%C-LDH correlated with days on ECMO and weakly with Peak%C-BFR.

There were 25 patients requiring ECMO alone (ECMO group) and 17 requiring CRRT in addition to ECMO (ECMO+CRRT group). On the first day of ECMO support (Table 2), ECMO+CRRT patients had a significantly higher serum creatinine and blood pH compared to the ECMO group. Over the entire ECMO course (Table 3), the ECMO+CRRT group spent more days on ECMO, had a lower blood pH, and a higher peak FHb level, peak BFR, Peak%C-FHb, and Peak%C-BFR compared with the ECMO group. Although the ECMO+CRRT group trended toward a higher Peak%C-LDH level than the ECMO group, this difference was not statistically significant ($p = 0.07$). As shown in Fig. 1, there was a significant increase in the mean level of FHb one day after the initiation of CRRT (73.3±49.2 mg/dL) compared to the level one day prior to CRRT (50.0±30.3 mg/dL) (p=0.012) among patients who received it. This increase was observed in 14 out of 17 patients.

We next examined if the changes in renal function during ECMO support were associated with the degree of hemolysis in the ECMO group. As shown in Table 4, End%C-Scr correlated with Peak%C-LDH, but not with Peak%C-FHb or Peak%C-BFR. End%C-BUN correlated with peak BFR, Peak%C-LDH, and Peak%C-BFR, but not with Peak%C-FHb. As expected, End%C-Scr was associated with End%C-BUN (r=0.62, p=0.001). Neither End%C-Scr nor End%C-BUN correlated with the observed changes in blood pH (data not shown).

Discussion

To the best of our knowledge, this study is the first to demonstrate that hemolysis is enhanced when a hemofilter is added to an ECMO circuit in children with congenital heart disease following cardiac surgery. We found that 41 out of a total of 42 patients in this study had elevated FHb levels within one day of ECMO support, and that relative changes in their peak FHb levels correlated with those in the peak serum LDH levels. These findings are consistent with reports from previous studies that hemolysis is very common during ECMO support.[1] Among patients who required dialysis, there was a further significant increase in FHb level within 24 h after the initiation of CRRT. The peak FHb levels were higher among patients requiring CRRT than among those not requiring it. These results suggest that the addition of an in-line hemofilter to the ECMO circuit enhances hemolysis. The etiology of this finding is most likely multifactorial.

The fragmentation of erythrocytes can be induced by a combination of shear stress, positive pressure, wall impact forces, and properties of nonendothelialized surfaces.[15] The variety of etiologies may explain why we observed only a marginal correlation between the peak change in ECMO blood flow and the peak changes in FHb and serum LDH levels (Table 1). In our system, the caliber of the CRRT circuit where part of the ECMO blood is diverted is 50% of the caliber of the remainder of the ECMO circuit. De Wacher et al. demonstrated in vitro that at a blood flow rate of 500 mL/min, FHb levels varied inversely with the sizes of the cannulae connecting to a blood line with a large bore.[16] Therefore the increased shear stress and positive pressure through the blood-nonendothelial surface at the junction of the hemofilter and ECMO circuits are quite likely a significant contributing factor to an elevated FHb. We have observed that whenever a hemofilter is attached to the ECMO circuit, the speed of the ECMO roller pump needs to be increased to maintain adequate oxygenation. This increased pump speed is very likely to be another contributing factor to elevated FHb, as reported previously by other investigators.[6]

Table 1. Correlation coefficients between variables in all patients (*n*=42)

	Peak % change in FHb 83.6±183%	Peak % change in LDH 150±324%
Peak BFR (131±42 mL/min/kg)	0.33*	0.15
Peak % change in LDH (%)	0.49*	–
Days in ECMO (5.1(2.5)	0.39*	0.53*
Peak % change in BFR (36.8±51.0%)	0.29[#]	0.28[§]

FHb, plasma free hemoglobin; LDH, lactic dehydrogenase; BFR, ECMO blood flow rate.
*p<0.05; [#]p=0.063; [§]p=0.08.

Table 2. Comparisons between patients with and without CRRT on the first day of ECMO

	ECMO (*n*=25)	ECMO+CRRT (*n*=17)
Age (days)	3.85±8.15	14.8±36.5
Weight (kg)	4.81±3.27	7.74±12.1
CVP (cmH_2O)	8.32±3.41	12.6±6.4
BUN (mg/dL)	15.9±13.3	13.3±9.5
Serum creatinine (mg/dL)	0.69±0.26	0.88±0.36*
Serum LDH (IU/L)	1,180±1,390	1,269±1,354
Blood pH	7.33±0.07	7.35±0.10*
FHb (mg/dL)	41.4±32.2	51.7±28.0
PRISM III	8.12±4.1	10.4±4.6
BFR (mL/min per kg)	118±38	141±61

ECMO, patients requiring ECMO alone; ECMO+CRRT, patients requiring both ECMO and CRRT; LDH, lactate dehydrogenase; BFR, ECMO blood flow rate; CVP, central venous pressure; BUN, blood urea nitrogen. *p<0.05.

Table 3. Comparisons between patients with and without CRRT during entire ECMO course

	ECMO (*n*=25)	ECMO+CRRT (*n*=17)
Days on ECMO	4.08±8.15	6.71±2.85*
Lowest blood pH	7.33±0.07	7.20±0.12*
Peak LDH level (IU/L)	1,527±2,051	2,396±1,301*
Peak FHb level (mg/dL)	49.3±37.6	99.5±58.5*
Peak BFR (mL/min per kg)	138±41	204±56*
Peak % change in BFR (%)	20.0±31.5	61.5±64.0*
Peak % change in FHb (%)	31.4±72.9	160±259*
Peak % change in LDH (%)	38.3±74.4	277±436

ECMO, patients requiring ECMO alone; ECMO+CRRT, patients requiring both ECMO and CRRT; FHb, plasma-free hemoglobin; BFR, ECMO blood flow rate; LDH, lactic dehydrogenase. *p<0.05.

Table 4. Correlation coefficients between variables in ECMO-alone group (*n*=25)

	End%C-Scr 38.7±50.4%	End%C-BUN 123±169%
Peak FHb (49.3±37.6 mg/dL)	0.31	0.55*
Peak % change in FHb (31.4±72.9%)	–0.07	0.17
Peak BFR (138±41 mL/min per kg)	0.26	0.53*
Peak % change in BFR (20.0±31.5%)	0.08	0.50*
Peak % change in LDH (38.3±74.4%)	0.46[*]	0.45*

End%C-Scr, the relative change in serum creatinine at the last day of ECMO from the initial level; End%C-BUN, the relative change in blood urea nitrogen at the last day of ECMO from the initial level; FHb, plasma-free hemoglobin; LDH, lactic dehydrogenase; BFR, ECMO blood flow rate. *p<0.05.

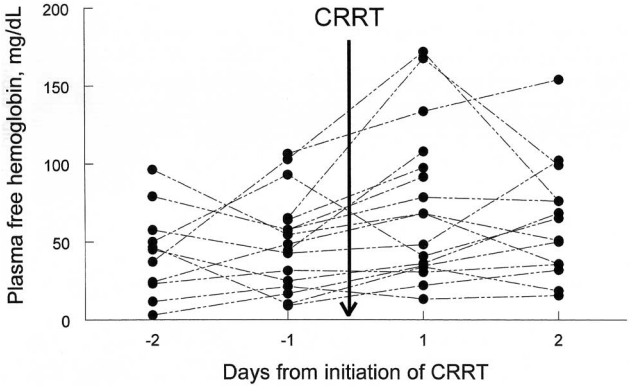

Fig. 1. Plasma-free hemoglobin levels within 2 days from the initiation of CRRT.
Horizontal bars stand for the mean values at days –1 and 1, respectively. *p=0.012 between days –1 and 1.

The hemofilter itself may also induce hemolysis. Bierer et al. measured FHb from a postdialyzer port in patients receiving CRRT with a 200 mL/min blood flow rate. FHb levels were significantly increased to more than 15 mg/dL if the hemofilter was used for greater than 100 h.[17] This prolonged filter use was associated with an increased venous pressure, suggesting clot formation within the circuit. While hemofilter thrombosis could contribute to enhanced hemolysis in our patients, the hemofilters were routinely changed every 48–72 h. Moreover, the increase in FHb levels in our patients averaged 23 mg/dL after CRRT was initiated, which exceeded the range of 0–15 mg/dL induced by hemofilters reported in the previous study [17]. Therefore, though hemolysis within the hemofilter may be a contributing factor, other etiologies must also be considered.

Hemolysis has been observed in 51% of patients requiring ventricular assist devices (VAD).[11] Luckraz et al. examined hemolysis in 9 VAD adult patients who required CRRT, which was performed through separate catheters from those for the VAD.[10] FHb was significantly increased in 6 patients within 2 days after the initiation of CRRT. The authors speculated that sublethal damage to erythrocytes[5] might have occurred through the VAD circuit, which developed into frank hemolysis of the damaged erythrocytes when they passed through the CRRT circuit. Similarly, we speculate that in our patients some erythrocytes might have suffered sublethal damage through the ECMO circuit and hemolyzed when they passed through the CRRT hemofilter and/or circuit.

The impact of hemolysis on kidney function in patients on ECMO is not known. In our ECMO patients who did not require CRRT, changes in serum creatinine and BUN at the end of ECMO did not correlate with the degree of peak changes in FHb level during ECMO. Although the percent change in BUN at the end of ECMO correlated with the peak FHb level and the degree of peak changes in LDH during ECMO, these observations could simply reflect hemolysis rather than renal dysfunction. Another possibility is that our small sample size prohibited us from observing a significant impact of hemolysis on renal function at the end of ECMO support. Further studies with more subjects are needed to clarify the association between hemolysis and renal function in patients on ECMO support, as well as the recovery of renal function in those patients who develop acute renal failure during ECMO.

In conclusion, our study suggests that there is enhanced hemolysis during combined ECMO and CRRT support compared with ECMO alone in children after cardiac surgery. Increased shear stress, physical properties of the bypass circuit, the roller pump, the hemofilter, and sublethal damage to erythrocytes are all potentially contributing factors to the development of hemolysis. Further studies are needed to clarify the clinical effects of excessive hemolysis on renal function in patients requiring ECMO support.

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