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BASIC SCIENCE ARTICLE

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Manual single lumen alternating micro-batch dialysis achieves reliable clearance via diffusion

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BACKGROUND: Acute kidney injury is a cause of preventable deaths in low resource settings due to lack of dialysis access and cost. A manual single lumen alternating micro-batch (mSLAMB) dialysis technique performs kidney replacement therapy using single lumen access, low-cost bags/tubing, intravenous fluids, and a filter without electricity, a battery, or a pump. We propose a protocol whereby mSLAMB can perform diffusive clearance simply and efficiently to bring dialysis to underserved populations.
METHODS: Expired packed red blood cells mixed with crystalloid solution were spiked with urea and anticoagulated with heparin. A Static diffusion Technique (with short flushes of fluid before each filter pass) was compared to a Dynamic diffusion Technique (with fluid running through the filter during the forward pass) to assess urea and potassium clearance. Passive ultrafiltration was the difference between the 200 mL batch volume and volume returned to the blood bag per cycle.
RESULTS: Five cycles achieved urea reduction ratios (URR) between 17–67% and potassium clearance of 18–60%, with higher percentages achieved from higher proportions of batch volume dialyzed to patient volume. Dynamic Technique increased clearance over the Static Technique. Passive ultrafiltration volumes were 2.5–10% of batch volume.
CONCLUSION: mSLAMB dialysis performs diffusive clearance and passive ultrafiltration efficiently, while preserving resources and available manpower.

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IMPACT:

- mSLAMB is a dialysis technique that can perform efficient diffusive clearance and passive ultrafiltration without electricity, batteries, or a pump.
- With basic medical supplies and limited manpower, mSLAMB is a cost-effective means of providing emergency dialysis in low resource areas.
- We propose a basic algorithm for safe and cost-effective dialysis for people of different ages and sizes.

INTRODUCTION

Acute kidney injury (AKI) remains a significant cause of preventable deaths in low resource settings due to prohibitive costs and lack of access to basic dialysis.[1] A recent systematic review of outcomes from AKI in children and adults in Sub-Saharan Africa found an overall mortality of 34% in children and 32% in adults needing dialysis, and a striking 73 and 86% when dialysis was needed but not received.[2] Out-of-pocket costs and erratic hospital resources were two of the major barriers to access to care. Even acute peritoneal dialysis, a more economical approach than standard hemodialysis, had an average cost of 420 USD per AKI course after subsidization in a single sub-Saharan African country.[3] In accordance with the International Society of Nephrology's 0by25 initiative, more must be done to prevent death from AKI, especially those related to socio-economic barriers like cost and access to basic medical supplies.[1] Prevention of AKI is laudable, but the reality is that many patients become ill despite preventive measures and present with severe AKI. Thus, low-cost hemodialysis options are needed to support those patients who develop severe AKI.

The single lumen alternating micro-batch (SLAMB) dialysis technique performs kidney replacement therapy (KRT) at a very low cost with widely available equipment, namely premade sterile fluids and a dialysis filter.[4] The manual SLAMB (mSLAMB) set-up works without electricity, a battery, or a pump, can be performed when there are space constraints, and achieves hemofiltration efficiently and accurately with a push-pull system and gravity.[5] In this closed loop system, blood is drawn in small batches from the patient into a sterile tubing circuit, and a hemofilter provides clearance, while syringes can pull fluid off for ultrafiltration.

However, the clearance achieved with the mSLAMB hemofiltration technique was more than would be predicted by convection alone, especially during the first pass through a primed filter. If this extra clearance is from diffusion, we believe equipment, labor, and

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Table 1. mSLAMB diffusion set-up and priming steps.

Step #	Description
1	Staircase 3 IV poles: the first pole closest to the patient and higher than #2 and #3, the second lower than #1 and higher than #3 and third the lowest and the farthest from the patient
2	Hang the replacement fluid bag (RF bag) on pole #1, blood reservoir #1 (BR #1) on pole #2 at filter height, and dialysis fluid bag (HD bag) on pole #3, 60 cm under the filter. Hang the effluent bag and blood reservoir #2 (BR #2) 100 cm under the filter
3	Attach a 50 mL syringe to the stopcock. Hook the filter to IV pole #2 at the most comfortable height (minimum 100 cm from the ground)
4	Attach the red Luer lock from blood reservoir #1 to the filter inlet and the red Luer lock from blood reservoir #2 to the filter outlet
5	Attach the Hansen connector to one side of the filter, and the effluent bag connector to the other side of the filter
6	Fill the replacement fluid bag with the fluid that has been chosen and begin gravity prime; when finished, discard excess fluid present in blood reservoir #1 and #2, leaving about 10 mL in each bag
7	Fill the dialysis bag with the dialysis fluid and prime the dialysis line, up to the effluent bag
8	Connect the circuit to the patient

time could be conserved by needing fewer cycles, less hemofiltration fluid, and fewer providers to perform the procedure. We sought to test the performance characteristics of the mSLAMB in a diffusion configuration, hypothesizing that mSLAMB could perform effective diffusive clearance, and optimization of technical factors impacting clearance, blood flow rate, and ultrafiltration could be incorporated into a protocol for safe and reliable dialysis in resource limited settings.

METHODS/MATERIALS

mSLAMB diffusion set-up

The mSLAMB set-up has been described previously.[5] Minor adjustments for the diffusion technique are namely that no syringe is necessary between the filter and the effluent bag unless active ultrafiltration is desired, and that instead of replacement fluids being hung before the filter, dialysis fluids are connected to and run through the filter for diffusive exchanges between it and the blood. Briefly, necessary equipment includes 1 disposable kit, 3 IV poles, replacement fluid bags (which also serve as priming solution), any sterile crystalloid for dialysis fluid, 1 dialysis filter, 1 filter holder, 1 50 mL syringe, 1 collection basin/garbage bin, and 1 hemostat. The steps for set-up and priming are listed in Table 1, and the configuration of the set-up is displayed in Fig. 1. The Rexeed 25 S (Asahi Kosei Medical Co, Suffern, NY) and Polyflux 6H (Baxter Healthcare Co, Deerfield, IL) were used for the purpose of these experiments, but any available hemodialyzer, both low and high flux, should work.

In vitro diffusion experiments

A patient's intravascular blood volume was simulated with expired packed red blood cells mixed with crystalloid solution to achieve low and normal hematocrits. Hematocrit determinations were performed using a Stat Profile Prime+ analyzer (Nova Biomedicals, Waltham, MA) per manufacturer protocols. Blood volumes tested were 0.5 L, 0.8L–1.45 L, and 2 L, to simulate infant, toddler, and child patient sizes. Batch volumes were 200 mL regardless of bag size. Each bag was initially spiked with 5 g of urea, and 1–2 g were added to recharge the blood bag before each new experiment. One thousand units of heparin were used per 400 mL of bag volume. The dialysis fluid was any crystalloid available; we used a combination of Lactated Ringers and 0.9% sodium chloride, Plasmalyte, or PrismaSate bags. The blood bag was connected directly to the mSLAMB set-up, without use of a catheter. Baseline potassium and blood urea nitrogen (BUN) was drawn from the blood bag prior to connecting to the mSLAMB set-up, minutes before the first run.

We tested two different techniques to perform diffusive clearance: the Static Technique and Dynamic Technique. For the Static Technique, the filter was refreshed with dialysis fluid for 5 s prior to each run through the filter. For the Dynamic Technique, the dialysis fluid ran freely as blood ran through the filter during the forward pass only. For each cycle, 200 mL of blood was pulled via syringe from the patient blood bag into blood reservoir (BR) #1. This batch volume flowed via gravity through the filter, to BR #2. When all the blood reached BR #2, both lines were clamped between BRs and filter, and BR #2 was switched to the higher position, usually the prior height of BR #1, and BR #1 was moved lower, to the prior height of BR #2. If performing the Static Technique, the filter would then be flushed again with dialysis fluid for 5 s. Thereafter, blood lines were unclamped and blood flowed through the filter to BR #1. Once BR #1 was full, blood was returned to the blood bag via syringe pull. To maintain net neutral volume status in the blood bag, UF was replaced with an equal volume of replacement fluid, which was the same composition as the dialysis fluids, and can be whatever is available.

Blood samples were drawn after the returned blood was mixed in the large patient bag after each cycle for determination of BUN and potassium clearance. The difference between the 200 mL batch volume and the volume of blood returned to the blood bag after each cycle was considered passive ultrafiltration (UF) volume.

Ten different experimental designs were run in duplicate or triplicate, with 5–7 cycles performed per experiment and 21 runs performed in total. Four rounds were excluded from clearance analyses due to technical factors limiting accuracy. Only 2 experiments were performed with the Polyflux HF filter, and the rest used the Rexeed. Three experiments were performed with a normal hematocrit. Five experiments were predominantly Static Technique; five were Dynamic. The amount of dynamic flow time was modulated to achieve desired UF but was predominantly the full time of the forward pass.

Single forward flow experiment

One experiment tested a new design whereby dynamic diffusion was run across the filter in the forward direction, with BR #2 hooked directly back to the patient to perform only a single forward pass. A syringe push-pull maneuver brought the blood back to the patient bag from BR #2, and samples were drawn from the patient blood bag after full blood return.

Clearance determination

Clearance was measured by testing blood urea nitrogen (BUN) and potassium concentrations from 1–3 mL blood samples drawn directly from the blood bag, after each cycle had completed and the 200 mL batch volume had been mixed with the remaining blood. BUN and potassium were analyzed using a Roche c 311 clinical chemistry analyzer (Roche Diagnostics, Indianapolis, IN) per manufacturer protocols in the Cincinnati Children's Hospital Medical Center (CCHMC) Nephrology Research Laboratory. Urea reduction ratio (URR) and potassium clearance were calculated using the formulas: $URR = (Urea_{pre}\text{-}Urea_{post})/Urea_{pre} \times 100$ and $\text{potassium clearance} = (Potassium_{pre}\text{-}Potassium_{post})/Potassium_{pre} \times 100$.

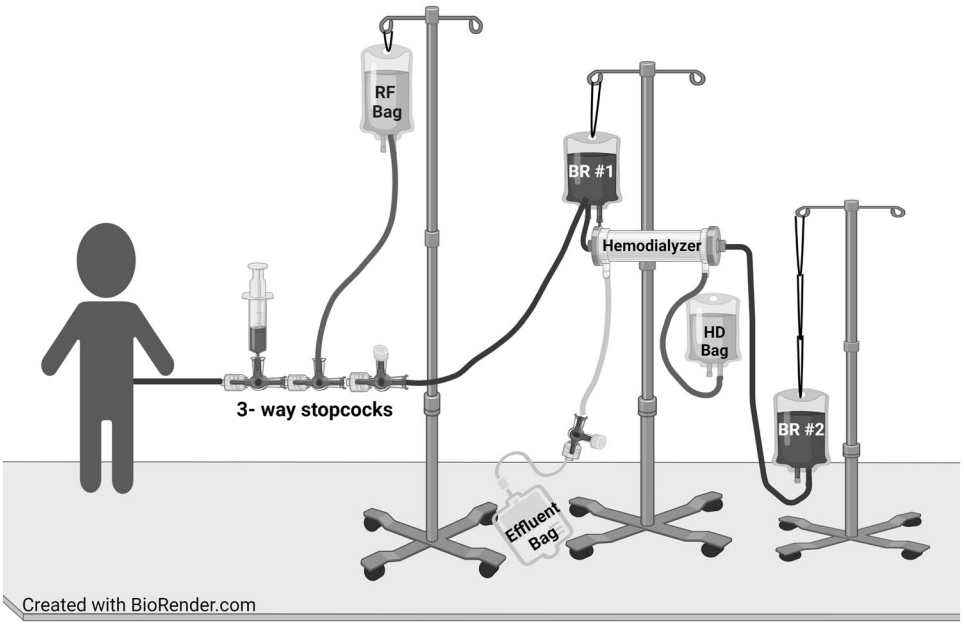

Fig. 1 Suggested mSLAMB dynamic technique configuration. Theoretical time of each pass is about 90 s, theoretical ultrafiltrate volume every cycle is about 20 mL with a low consumption of dialysis fluid. To configure for the simple algorithm explained in Fig. 4, the bottom of BR#1 should be at the height of the filter. RF-replacement fluid. BR-blood reservoir. HD-hemodialysis fluid.

Dynamic UF experiment

We performed separate experiments to determine the factors that modulate UF volume. We performed 49 cycles using the Dynamic Technique with a 2 L blood bag and an initial hematocrit of about 20%, a Polyflux filter, heparin anticoagulation, and 200 mL batch volumes. We changed the bag heights (BRs, dialysis fluid bag, and effluent bag) to test different configurations, measuring the difference between prescribed and actual batch volume at the end of every cycle. Ultrafiltration (UF) accuracy was calculated using the formula: UF accuracy = (Volume Effluent $_{prescribed}$ − Volume Effluent $_{measured}$)/Volume Effluent $_{measured}$.

Ethics statement

This protocol was deemed Not Human Subjects Research by CCHMC institutional guidelines and ethical approval is therefore not required for this study in accordance with local or national guidelines.

Statistics

Statistics were performed using SigmaPlot (Systat Software Inc, San Jose, CA). Descriptive statistics were shown as mean +/− standard deviation (SD).

RESULTS

mSLAMB achieves BUN and potassium clearance

Blood bag hematocrit was either low, 30 + /−2%, or normal, 42 + /− 3%. Mean baseline BUN for experiments 1–10 was 132.4 + /− 75.6 mg/dL (range 81–299 mg/dL), except for 2 experiments where baseline BUNs were 60.1 and 20.6 mg/dL, respectively. Mean baseline potassium was 29.9 + /− 16.2 mmol/L (range 14.3–70.8 mmol/L), except for 2 experiments where starting values were <10 mmol/L. We routinely achieved urea reduction ratios (URR) between 17–67% and potassium clearance of 18–60% with 5 cycles of mSLAMB dialysis (Fig. 2).

The main factor responsible for clearance variability was the proportion of batch volume dialyzed to blood bag volume: 0.5 L bags achieved 40–67% URR, 0.8–1.45 L bags achieved 26–56% URR, and 2 L bags achieved 17–31% URR. Potassium clearance

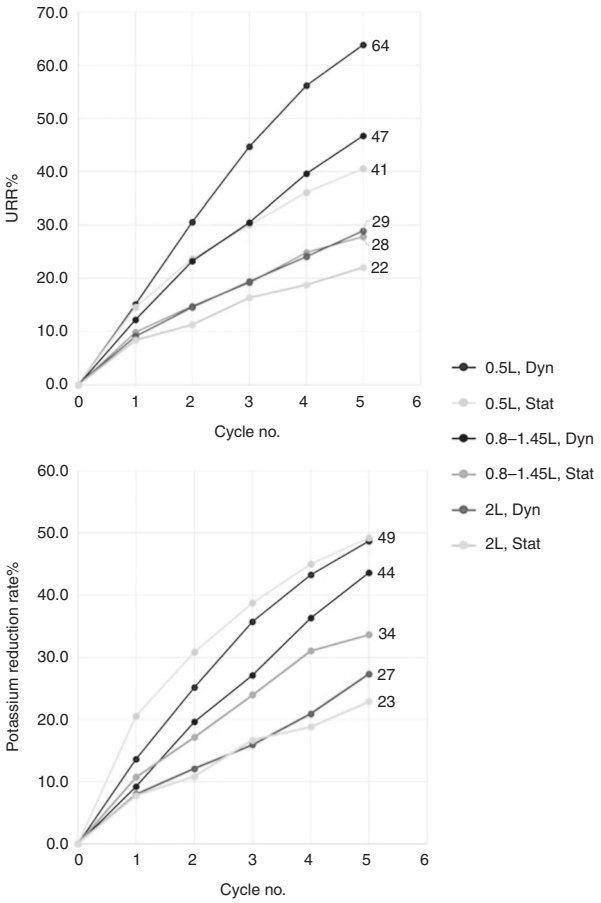

Fig. 2 Mean urea reduction ratio and potassium clearance rate. Experiments are separated by blood bag size and Diffusion Technique. URR-urea reduction ratio. Dyn-dynamic. Stat-static. no.-number.

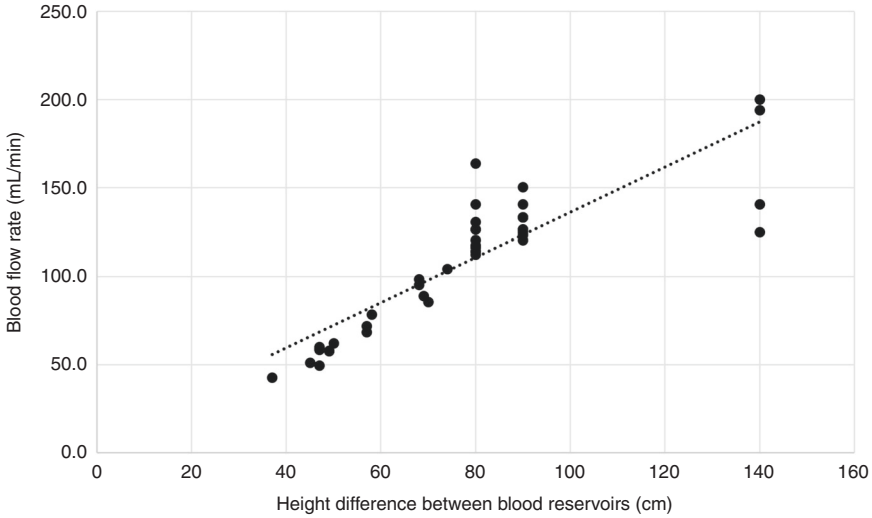

Fig. 3 Blood reservoir height difference determines blood flow rate. Blood flow rate increased as height difference between blood reservoirs increased.

Table 2. Mean "Driving Pressure" as a determinant of mean ultrafiltration volume.

Mean "Driving Pressure" (cm)	Mean UF volume (mL)	SD
15	0.0	3.54
30	5.2	0.55
50	7.4	1.25
65	15.0	0.71
70	14.9	3.36
80	13.2	2.02
130	19.0	0.94
180	32.2	0.96

The difference between the summation of blood reservoirs height and the summation of dialysis fluid bag and effluent bag heights determines "driving pressure."
UF ultrafiltrate, *SD* standard deviation.

mirrored URR: 37–60% in 0.5 L bags, 32–56% in 0.8–1.45 L bags, and 18–29% in 2 L bags. Dynamic Techniques produced consistently increased URR compared to Static Technique, regardless of blood bag volume; however, there was minimal difference in potassium clearance between the two Techniques (Fig. 2). The greatest clearance occurred with a freshly primed filter on the 1st cycle. Filter type did not impact clearance.

Blood flow rate optimized by height difference between blood reservoirs

We achieved blood flow rates (BFR) from 45 mL/min to 200 mL/min per cycle. Greater height differences between BRs resulted in faster BFR (Fig. 3). A height difference of 80 cm reliably produced a BFR of 112–164 mL/min over 11 passes and a difference of 90 cm produced a BFR of 125–150 mL/min over 10 passes. Filter type, patient hematocrit, and dynamic flow during forward vs reverse pass did not impact BFR.

Single forward pass run may achieve superior clearance to a forward/reverse run

A Single Forward Pass Dynamic Technique experiment was attempted to reduce cycle times. This Technique produced a URR of 56% and a potassium clearance of 56% with 5 cycles, as BUN decreased from 201.6 to 88.6 mg/dL and potassium decreased from 14.3 to 6.3 mmol/L (Supplementary Fig. 1).

Passive ultrafiltration impacted by technique and bags heights

We achieved ultrafiltration (UF) passively (without active syringe pulling). Thus, we aimed to control UF, with a goal to remove 10% of the batch volume, or 20 mL, per cycle. In the 10 diffusion experiments, UF was more reliably controlled with the Static Technique than the Dynamic Technique, with volumes of 0 to 25 mL removed/cycle during 15 Static Technique runs. UF volumes were reliably around our goal range of 10 to 35 mL removed/cycle when dynamic times were less than 1 min (12 runs) but ranged from 60 mL removed to 40 mL gained/cycle when dynamic times were greater than 1 min (22 runs).

We performed Dynamic UF experiments to better understand and control UF. We achieved UF volumes between 35 mL removed to 10 mL added/cycle (49 cycles), with a mean UF volume of 13.5 mL. We found a consistent relationship between the UF volume and the height difference between the blood system (blood reservoir #1 and #2) and the dialysis system (dialysis fluid bag and effluent bag) (Table 2, Supplementary Fig. 2).

DISCUSSION

In these series of experiments, we demonstrate that the mSLAMB can be effectively configured to perform diffusive KRT. We reliably produced BFRs via gravity comparable to automated dialysis machines, and we showed that clearance can be achieved regardless of different blood volume sizes, patient hematocrits, and filter type. Moreover, we discovered that passive UF occurs, and we were able to control UF volume with bag height manipulation in both Static and Dynamic techniques. Although our URR is an overrepresentation of what is to happen in vivo given urea's volume of distribution, clearance rates should be proportional. As these clearance rates were achieved in 1 hour, we recommend longer sessions for adequate patient stabilization.

In a prior study, we performed manual hemofiltration with mSLAMB that produced effective clearance and accurate ultrafiltration.[5] However, our first cycle achieved clearance greater than predicted; we postulated that diffusive clearance was occurring with the resident priming solution, and that diffusion could be a more efficient clearance technique. Compared to eight hemofiltration cycles, we demonstrated similar clearance rates with five cycles of Static Technique and increased clearance with five cycles of Dynamic Technique. Predicted convective clearance in our Static Technique ranged from 15–37%, while the predicted convective clearance in our Dynamic Technique ranged from

Heights	BR#1	At the filter height
	BR#2	100cm under the filter
	Dialysis fluid bag	60cm under the filter
	EB	100cm under the filter
Dynamic time	Same as the blood run time (about 90s)	
Empty the effluent bag when full		
Batch size	Infant	50mL
	Child	100mL
	Adult	200mL
Dialysis and replacement fluids	First choice	Any balanced IVF
	Second choice	Any IVF
Fluid removal	5–10% of the batch volume with this config.	
	To modify the UF volume lift or lower the BRs (keep the height difference between them 100cm) → Lift BRs to increase the UF → Lower BRs to decrease the UF	

Fig. 4 Simplified mSLAMB Dynamic Technique algorithm. A more detailed explanation of set-up to produce the results presented. We propose using the Dynamic Technique, with batch size dependent on patient size. Fluid removal goals should be dependent on patient's hemodynamic status; thus, we propose ways to modify ultrafiltrate volume achieved. BR blood reservoir, EB effluent bag, IVF intravenous fluid, config configuration, UF ultrafiltrate.

34–45%, making diffusion the predominant source of clearance. In our limited experimentation, the Single Forward Pass Technique provided superior clearance to the forward/reverse experiments. However, we again found that the first cycle had greater clearance, and subsequent cycles had either stable or slowly decreasing clearance. This may be due to microclots that develop in the filter to decrease efficiency, or unequal mixing of batch volume to total bag volume. Regardless, diffusion was the predominant form of clearance, and the techniques required less manual labor and time, achieved ultrafiltration without provider manipulation (syringe push-pulls), and relied less on operator participation for accuracy. The Static Technique requires less dialysis fluid than the replacement fluid required for the hemofiltration experiments, thus conserving resources.

In low resource settings, priorities are safety, reliability, access to equipment, and availability of capable providers. We propose a basic protocol in Fig. 4 utilizing the Dynamic Technique, given its greater clearance efficacy and ability to control UF volume, at the expense of increased consumption of dialysis fluid. Using the heights described in Fig. 1, we believe this set-up will produce a fluid removal rate that will be safe for the patient and less clotting risk for the dialysis filter. Based largely on our findings that the batch volume in relation to the total patient volume is the greatest determinant of clearance, we proposed batch volumes based on patient size to optimize clearance while ensuring hemodynamic stability. Should hemodynamics not allow for volume removal, basic replacement fluids can be returned to the patient to further lower potassium and neutralize acidosis. The Static Technique requires less dialysis fluid than the Dynamic Technique, and less time and labor making, hanging, and draining dialysis/effluent fluid bags, so if fluid and human resources are limited, the Static Technique should be employed.

Should advanced providers (eg nurses, physicians) and sophisticated testing be available for intensive diagnostic testing and monitoring, we propose a more complex algorithm with greater control and clearance optimization (Supplementary Fig. 3). In cases of emergent hyperkalemia or severe uremia, if the patient's hemodynamics and fluid status can be monitored, we recommend the single pass Dynamic Technique, given improved clearance and less time, at the expense of more dialysis fluid requirement and bag manipulation. If uremia is the sole problem, we recommend balanced dialysis/replacement fluids containing potassium to prevent hypokalemia. We have performed calculations based on our own cycle times and clearance rates to approximate the number of cycles necessary to achieve clearance rates for normal patient blood volumes (Supplementary Fig. 3).

A strength of these experiments was the discovery that UF occurs without planned, active fluid removal; however, control of UF is necessary to ensure hemodynamic stability. UF is generated by a driving pressure between "blood side" pressures and "dialysis fluid side" pressures. A positive pressure is generated by the column of fluid above the filter (by gravity), and a negative pressure is generated if the fluid bag is under the filter. These pressures are proportional to their heights from the filter. The difference of the pressure between the filter sides (BR#1 + BR#2)-(dialysis fluid bag+ effluent bag) generates the convective movement of the fluid from the higher pressure side to the lower pressure side that produces the UF. The strong correlation between this "driving pressure" and UF volume is shown in Supplementary Fig. 2, and a suggested mSLAMB dynamic set-up configured to give a reliable UF (about 20 mL per cycle) with an acceptable BFR, adequate clearance, and minimal dialysis fluid consumption is shown in Fig. 1.

Further work will address the limitations of this study. Although the Diffusion Techniques provide clearance, the proportion of batch volume to total blood volume we used doesn't represent the in vivo ratios. As urea is removed from total body water, usually approximately 10 times that of blood volume, URR in a real patient will likely be significantly less than what we report, albeit proportional. There was no difference in potassium clearance between the Dynamic and Static Technique experiments, which we attributed to increased hemolysis during the Dynamic Technique. We believe this to be a product of the fragility of the expired red blood cells, but this has not been proven. Additionally, clearance may have been overestimated by replacement fluid addition to the blood bag. Other differences in clearance will result from the filtration fraction of smaller filters used for smaller patients and the BFR possible with smaller access points, as children's veins tolerate smaller intravenous gauge sizes. Lastly, we have not tested different types or doses of anticoagulation. We used heparin with citrated packed red blood cells without any monitoring. It is possible that different techniques have different anticoagulation needs, and ultimately anticoagulation may depend on the stability of the patient's clinical condition.

We conclude that mSLAMB dialysis can effectively perform diffusive clearance and achieve passive ultrafiltration, and that resource and provider preservation can be prioritized without loss of efficiency with our proposed algorithms. Next steps involve bringing and teaching mSLAMB to low resource areas in need.

DATA AVAILABILITY

All data generated or analyzed during this study are available from the corresponding author who will gladly make available all data on reasonable request.

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AUTHOR CONTRIBUTIONS

G.C., A.K.C., J.M., and D.C.H. all conceived, designed, and performed experiments and data acquisition. G.C. and D.C.H. performed data analysis, interpretation, and were primary manuscript authors. J.R. performed laboratory techniques that allowed for data acquisition. S.L.G. provided critical article drafting and editing.

COMPETING INTERESTS

S.L.G. reports receiving personal fees from Baxter Healthcare, BioPorto Inc., Nuwellis, Fresenius, MediBeacon, and Medtronic. J.M. is a consultant for Medtronic.

ADDITIONAL INFORMATION

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