Original Article

Microcirculation, endothelium and glycocalyx changes associated with the use of milrinone in children with septic shock

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Background: The goal of fluid resuscitation and the use of inotropes in septic shock has traditionally focused on improving blood pressure and cardiac output, without considering the microcirculatory changes. Reaching macrocirculatory goals but with persistent microcirculatory abnormalities (hemodynamic incoherence) in septic shock has been associated with greater organ dysfunction and mortality. The objective of this study was to evaluate the microcirculation (flow and capillary density) and endothelial glycocalyx changes associated with the use of milrinone in children with septic shock, as well as their relationship with clinical variables and organ dysfunction.

Methods: A prospective cohort study from February 2022 to January 2023 at a university hospital (Fundación Cardioinfantil-Instituto de Cardiología). Sublingual video microscopy was used to evaluate capillary density, microvascular flow rates and perfused boundary region (PBR—inverse parameter of glycocalyx thickness—abnormal if >2.0 microns). The primary outcome was the association between microcirculation and endothelial glycocalyx changes related to the use of milrinone.

Results: A total of 140 children with a median age of two years [interquartile range (IQR) 0.58–12.1] were included. About 57.9% (81/140) of the patients received milrinone infusions. Twenty-four hours after receiving milrinone, the patients maintained functional capillary density (P<0.01) and capillary recruitment capacity (P=0.04) with no changes in capillary blood volume versus those who did not receive milrinone. Children under two years old who received milrinone had better 4–6-micron capillary density than older children [odds ratio (OR) 0.33; 95% confidence interval (95% CI): 0.12–0.89; P=0.02] and less endothelial glycocalyx degradation [adjusted OR (aOR) 0.34 95% CI: 0.11–0.99; P=0.04]. These changes persisted despite elevated ferritin (aOR 0.41; 95% CI: 0.18–0.93; P=0.03). Prolonged capillary refill and elevated lactate were correlated with microcirculation changes in both groups. The patients who died had the highest PBR levels (P=0.04).

Conclusions: Children with septic shock who receive milrinone infusions have microcirculation changes compared with those who do not receive them. The group that received milrinone was found to maintain functional capillary density and capillary recruitment capacity and have less endothelial glycocalyx

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degradation 24 hours after administration. These changes were present despite the inflammatory response and were more significant in those under two years of age.

Keywords: Septic shock; endothelium; fluid therapy; mortality; vascular permeability

Submitted Dec 31, 2023. Accepted for publication Apr 11, 2024. Published online May 27, 2024.
doi: 10.21037/tp-23-619
View this article at: https://dx.doi.org/10.21037/tp-23-619

Introduction

Sepsis is an illness with high morbidity and mortality (1). Hemodynamic involvement often determines the outcomes (2,3). Fluid resuscitation and the use of inotropes or vasopressors improve macrocirculation variables like blood pressure or cardiac output. With this goal, one of the medications used for children with septic shock is milrinone (a type III phosphodiesterase inhibitor), which has been considered to increase contractility and improve ventricular diastolic function (1,4). According to the Surviving Sepsis Campaign (SSC), inodilators are used in children with septic shock who have evidence of persistent hypoperfusion and myocardial dysfunction after using other vasoactive drugs (1). However, they also clarify that there are no randomized clinical trials evaluating the efficacy and safety of inodilators in children with septic shock (including milrinone, dobutamine, or levosimendan). The milrinone effect is explained by reduced cyclic adenosine monophosphate degradation and increased intracellular calcium release, which improves the ejection fraction (4). The primary effect of milrinone in children following cardiac surgery may lie more in vasodilation than in optimizing contractility (5). These inodilator effects have been found in children with septic shock, including patients with meningococcal sepsis and severe peripheral vasoconstriction (6). It has recently been suggested that the effects of milrinone in improving cardiac function are mediated by its systemic vasodilating effect, with little inotropic effect (7,8).

In sepsis, inotropes or inodilators are recommended when there is myocardial dysfunction with signs of low cardiac output, increased filling pressures or persistent signs of tissue hypoperfusion (1,7,8). However, the strongest evidence supporting the use of inodilators is mainly based on physiological studies. In experimental models of septic shock, milrinone has been shown to improve serum lactate levels as well as central venous saturation (9). In these studies, tissue perfusion with milrinone infusions has been evaluated indirectly using plasma biomarkers. It has been found to decrease vasoconstriction in small arterioles and improve functional capillary density in experimental models of septic shock (10). These effects could be mediated by a significant reduction in some inflammatory mediators as some *in vitro* studies of sepsis models in cardiomyocytes have suggested (11).

We do not know what changes occur directly in the microcirculation of children with septic shock who receive milrinone. The currently available technology (like sublingual video microscopy) allows direct observation of microcirculation changes and indirect observation of endothelial glycocalyx degradation in the pediatric

Highlight box

Key findings

- In children with septic shock, the use of milrinone is associated with direct microcirculatory changes: functional capillary density and capillary recruitment capacity maintenance, and less endothelial glycocalyx degradation.

What is known and what is new?

- Reaching macrocirculatory goals but with persistent microcirculatory abnormalities (hemodynamic incoherence) in septic shock has been associated with greater organ dysfunction and mortality.
- The direct microcirculatory effects of milrinone could be explained by changes in capillary diffusive and convective flow. These changes are more pronounced in children under the age of two and were related to changes in capillary filling, serum lactate and the organ dysfunction score.

What is the implication, and what should change now?

- The goal of fluid resuscitation and the use of inodilators in septic shock has traditionally focused on improving blood pressure and cardiac output, without considering the microcirculatory changes. Understanding the microcirculatory changes that occur with treatment interventions (milrinone) in critically ill children with septic shock allows a more comprehensive approach and potentially helps improve outcomes.

population (12). In children with septic shock, this microvascular technology has found less perfused capillary density and greater perfusion heterogeneity (13). Our hypothesis is that milrinone not only improves macro-hemodynamics, but its direct vasodilating effects can also be seen in the micro-hemodynamics of children with sepsis. The objective of this study was to evaluate the microcirculation (flow and capillary density) and endothelial glycocalyx changes associated with the use of milrinone in children with septic shock, as well as their relationship with clinical variables and organ dysfunction. We present this article in accordance with the STROBE reporting checklist (available at https://tp.amegroups.com/article/view/10.21037/tp-23-619/rc).

Methods

Study design and participants

This was a prospective observational study of all children from one month to 18 years old who were hospitalized for septic shock in the pediatric intensive care unit (PICU) of Fundación Cardioinfantil-Instituto de Cardiología in Bogotá, Colombia. We performed non-probabilistic convenience sampling, consecutively including all patients admitted with septic shock. This study was carried out from February 2022 to January 2023. The study was conducted in accordance with the Declaration of Helsinki (as revised in 2013). It was approved by the Institutional Review Board and the ethics committee of Fundación Cardioinfantil-Instituto de Cardiología (IRB-DDI 4814-2023; Approved January 20, 2022), and all the patients' parents or legal guardians signed informed consent form prior to being included in the study.

Data collection and case definitions

All children who, due to clinical deterioration, were consecutively admitted to the PICU with septic shock were included. Sublingual microcirculation was measured with video microscopy on admission, and 6 and 24 hours later, in the groups with and without milrinone. All patients received fluid resuscitation (10 mL/kg of Ringer's lactate) after admission, and vasoactive medications (noradrenaline, at the discretion of the attending physician) in the event of wide pulse pressure (more than 30% of the systolic pressure) or hypotension after a crystalloid bolus. In line with the SSC recommendation, milrinone was started when there was evidence of persistent hypoperfusion and cardiac dysfunction despite the use of vasoactive agents. Persistent hypoperfusion was identified when patients continued to have a capillary refill time of more than two seconds, an altered state of consciousness, a lactate greater than 2 mmol/L, or an echocardiogram with a cardiac index of less than 3.5 L/min/m^2 after fluid resuscitation. In addition, the entire conventional bundle treatment for septic shock was carried out, including antibiotics, as recommended in the recent sepsis guidelines (1,2). Patients received a milrinone dose ranging from 0.4 to 0.8 mcg/kg/min, according to their kidney function. Children with congenital heart disease or in the postoperative phase of cardiovascular surgery, those with diabetic ketoacidosis, multiple trauma or head trauma, or those who required continuous renal support therapy within the first 12 hours of admission were excluded. Patients who had received fluid resuscitation within the 24 hours prior to PICU admission were not included either. Recently, our group found that unbalanced crystalloids could alter the microcirculation and we wanted to control for this confounding factor from the design (13). The septic shock definition proposed in the SSC and the Latin American Consensus was used (1,2). All the demographic characteristics and macrocirculation variables (blood pressure, heart rate, pulse pressure), as well as standardized capillary refill (pressure exerted on the right index finger for 10 seconds using a glass slide, after which the pressure was released and color recovery was measured with a stopwatch), were recorded. A capillary refill time greater than two seconds was considered abnormal (1,3,11). Disease severity was evaluated within Pediatric Index of Mortality-2 (PIM-2) and the Pediatric Logistic Organ Dysfunction-2 (PELOD-2) scale. A PELOD-2 score higher than 8 was considered to indicate a higher risk of mortality (14). The Vasoactive Inotropic Score (VIS) was considered elevated if greater than 5, in line with the PODIUM recommendation (15). Patients were given oxygen to achieve a saturation above 90%, which included the use of a high-flow nasal cannula. If the saturation was below this level, and the attending physician considered it appropriate, the patients were intubated and started on invasive mechanical ventilation. Hospital protocol stipulates the use of decelerating ventilation modes in children (pressure control or pressure-regulated volume control). Laboratory tests were performed on admission and at 24 hours. Hyperferritinemia was defined as a serum ferritin greater

than 500 mcg/dL (as an inflammatory biomarker) (16) and lactate was elevated if greater than 2 mmol/L.

Microcirculation and glycocalyx degradation measurement

Sublingual video microscopy (Glycocheck System®, Microvascular Health Solutions Inc. 2014, Salt Lake City, UT, USA) was used to measure microcirculation (17). This equipment evaluates 4–25-micron diameter blood vessels using a dark field camera (CapisScope®, HVCS, KK Technology United Kingdom). This information travels to a software program (Glycocheck System®) that analyzes the best images in terms of focus and intensity. This system analyzes data independently from the examiner and reports the perfused boundary region (PBR) which is inversely related to the endothelial glycocalyx dimensions. A normal PBR value in adults and children has been established as less than 2.0 microns (17,18).

Furthermore, the video microscopy device reports the density of 4–6-micron capillaries (CD 4–6) and 4–25-micron capillaries, which indicate the capacity of the capillary network to recruit different-sized blood vessels (in 10^{-2} mm/mm^2). The density of 4–6-micron capillaries determines the functional capillary density (the physical distance between the capillary and the cell and indicator of diffusive flow). In terms of capillary flow, video microscopy measures the relative capillary blood volume (CBV-Rel). The software calculates this by dividing the volume of blood which reaches the larger capillaries (D≥10 µm) by the volume of blood in the smaller capillaries (D≤7 µm). This CBV-Rel corresponds to the blood volume reaching the smallest capillaries for exchange, which in sepsis has been found to depend highly on the larger vessels (18,19). A higher figure indicates better blood volume for exchange flowing from larger to smaller capillaries. This is the main compensatory variable in sepsis in terms of volume, rather than blood flow reduction in small capillaries (19). The capillary blood volume recruitment capacity (CBV-RC) was also measured. This is the capillary's reserve for recruiting additional functional capillary blood vessels when blood flow increases. The Microvascular Health Score (MVHS™) was measured from these values. The MVHS™ evaluates the relationship between these two variables (CBV/PBR) in different capillaries. The MVHS™ has a median of 1.7 points (IQR 1.3–2.5 points) in adults with sepsis and 4.0 points (IQR 2.1–4.3 points) in healthy subjects (19,20). Higher scores are considered to indicate microvascular health recovery. This video microscopy and its software have been found to have very low inter and intra-observer variability and the measured results cannot be modified by the investigators (17).

Outcomes

The primary outcome was the association between microcirculation (capillary flow and density) and endothelial glycocalyx changes associated with the use of milrinone in children with septic shock. The secondary outcomes were the relationship between microcirculation changes and the presence of organ dysfunction, inflammatory response and clinical variables of interest among the groups.

Statistical analysis

Descriptive statistics were calculated for the groups with and without milrinone. According to the nature of the variable, proportions (with percentages) were reported for qualitative variables. Means or medians were reported for quantitative variables, according to their distribution, along with their measures of dispersion. Then, a bivariate analysis was run evaluating the exposure variable (receiving or not receiving milrinone) against the outcome variables using a Student's *t* or Mann-Whitney *U* test for variables measured on admission and at 24 hours (laboratory tests). A repeated measures ANOVA or Friedman test for non-normal variables or non-homogeneous variance was used for the microcirculation variables that were measured at three points. Spearman's rank correlation coefficient or Pearson's correlation coefficient (depending on the distribution of the continuous variable) was calculated between the vital signs and microcirculation measurements. Confounding was controlled for from the design, considering the inclusion and exclusion criteria, particularly for disease severity (evaluated with PIM-2). Multivariate analysis was performed, including in the model the variables which met the Hosmer-Lemeshow criteria in the bivariate analysis and those considered to have biological plausibility. In addition, since the patients who received milrinone were younger, age was included in the logistic regression as a potentially confounding co-variable (21). The dependent variables were the microcirculation abnormalities measured by video microscopy: functional capillary density, percent blood flow and glycocalyx degradation (PBR). The model was constructed using the forward method and adjusted

Table 1 Patient characteristics on Pediatric Intensive Care Unit admission

Characteristic	Total, n=140	With Milrinone, n=81	Without Milrinone, n=59	P value
Age (years), median (IQR)	2.0 (0.58–12.1)	1.1 (0.5–8.1)	6 (0.75–14)	0.01
Female sex (%)	68 (48.6)	43 (53.1)	25 (42.4)	0.21
Focus of infection (%)				0.09
Respiratory	43 (30.7)	27 (33.3)	16 (27.1)	
Gastrointestinal tract	38 (27.1)	25 (30.9)	13 (22.1)	
Genitourinary	1 (0.7)	0 (0)	1 (1.7)	
Central nervous system	9 (6.4)	2 (2.5)	7 (11.9)	
Other	49 (35.1)	27 (33.3)	22 (37.3)	
PELOD-2 score, median (IQR)	7 (3–10)	8 (5–10)	7 (2–9)	0.12
Lactate (mmol/L), median (IQR)	1.21(0.82–1.78)	1.30 (0.9–1.9)	1.2 (0.7–1.6)	0.06
Glucose (mg/dL), median (IQR)	109.4 (91.8–138.8)	114.3 (90.9–143.5)	107 (92.1–132.8)	0.54
Ferritin (mg/dL), median (IQR)	409.6 (173.9–1,678.8)	557.7 (222.4–2,459.1)	248.1 (119.1–585.3)	<0.01
CRP (mg/dL), median (IQR)	5.1 (2.1–9.9)	5.1 (2.6–12.8)	4.4 (1.6–8.8)	0.20
D-dimer (mg/L), median (IQR)	3.17 (1.17–5.84)	3.21 (1.21–9.24)	2.61 (1.21–3.71)	0.27
Procalcitonin (g/dL), median (IQR)	1.4 (0.41–5.21)	2.1 (0.54–6.1)	1.13 (0.33–3.14)	0.05
Creatinine (mg/dL), median (IQR)	0.41 (0.39–0.61)	0.41 (0.31–0.61)	0.51 (0.41–0.61)	0.23

IQR, interquartile range; PELOD-2, Pediatric Logistic Organ Dysfunction-2; CRP, C-reactive protein.

with the omnibus test. A P<0.05 was considered statistically significant. We used the IBM® SPSS version 26.0 statistical package.

Results

Of the 1,123 patients admitted to the PICU during the study period, 140 had septic shock and met the inclusion criteria (*Table 1*; Figure S1). The median age was 2 years (IQR 0.58–12.1 years). The participants were similarly distributed by sex (48.6%; 68/140 females). Eighty-four percent (118/140) of the patients received invasive mechanical ventilation. Of the total number of patients included, 57.9% (81/140) received milrinone infusions. We found no differences in the Pediatric Logistic Organ Dysfunction (PELOD-2) scale on PICU admission (P=0.12).

Microcirculation changes associated with milrinone infusion

Capillary density (4–6 CD) at baseline in the group that received milrinone was 22.5 (IQR 15.4–43.8) and 36.8 (IQR 20.3–51.7) mm/mm^2 in the group without milrinone (P=0.17). There were no differences in these findings related to the different age groups (P=0.32) nor at 6 hours in the groups with or without milrinone (P=0.21). At 24 hours, these small capillaries (4–6 CD) were still recruited in the milrinone group, with a value similar to that on admission (25.1; IQR 14.1–38.7 mm/mm^2) (*Figure 1*). However, we saw a 28% reduction in the recruitment of these vessels in the group that did not receive milrinone (26.3; IQR 18.1–44.6 mm/mm^2; P<0.01), compared with the value on admission. The 4–6 CD value at 24 hours in the milrinone group was higher in those under two years old than in those older than two [28.4 (IQR 17.2–42.2) *vs.* 21.5 (IQR 13.6–28.9) mm/mm^2; P<0.01, respectively].

We found no differences in CBV-Rel between the groups on admission (P=0.37) and at 24 hours (P=0.27). Likewise, we found that the CBV-RC in the group which received milrinone was maintained at both assessment times (68% baseline *vs.* 65.9% 24 h). The group that did not receive milrinone showed a reduction in the CBV-RC at 24 hours, compared with the admission measurement (57.6% *vs.* 80.1%; P=0.04).

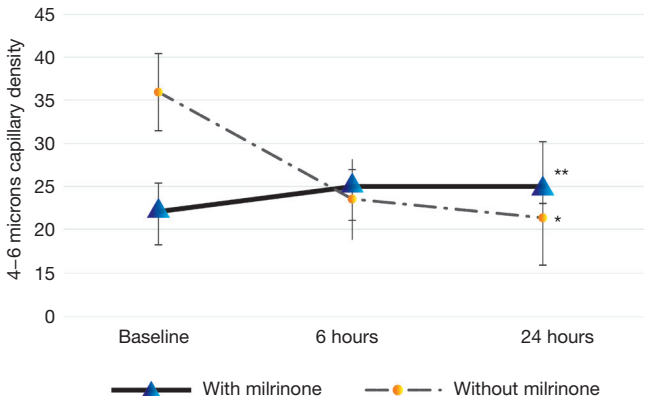

Figure 1 Changes in functional capillary density in the groups with and without milrinone infusion. Recruitment and functional capillary density diffusion in circulatory shock tend to worsen in sepsis without intervention. Increasing or sustaining adequate functional capillary density is important as a compensatory mechanism to increase oxygen supply to tissues in patients with circulatory shock (22). A comparison between the baseline and 24 hours with and without milrinone. Error bars represent 95% confidence interval. Mann-Whitney *U* test *, P<0.05; **, P>0.1.

Table 2 A multivariate analysis of the factors associated with microcirculation abnormalities measured with sublingual video microscopy in children with septic shock who received milrinone infusions*

Variable	aOR (95% CI)	P value†
Under two years old	0.33 (0.12–0.89)	0.02
Risk of ferritin >500 mgr/dL	0.41 (0.18–0.93)	0.03
Central venous oxygen saturation <70%	7.70 (1.58–37.61)	0.01
Venous to arterial CO_2 difference >6 mmHg	1.31 (0.52–3.31)	0.56

*, the microcirculation variables considered were: functional capillary density (4–6 microns), percentage of blood flow and perfused boundary region; †, adjusted for Pediatric Index mortality-2, age and vasoactive score. aOR, adjusted odds ratio; 95% CI, 95% confidence interval.

We evaluated changes in microcirculation according to the median age in the studied population. After 24 hours, children under two years old who received milrinone had fewer PBR abnormalities regardless of the disease severity and vasoactive score [63.8% 30/47 *vs.* 84.0% 21/25; adjusted odds ratio (aOR) 0.33, 95% confidence interval (95% CI): 0.12–0.89]. Patients with high ferritin were found to have a lower risk of glycocalyx degradation when they received milrinone (aOR 0.41; 95% CI: 0.18–0.93) (*Table 2*).

The MVHS™ score was abnormal in both groups (with and without milrinone) at baseline in all children with septic shock. We found that when the MVHS™ score was evaluated on admission, the group receiving milrinone had a lower level (2.5 points, IQR: 1.5–6.1) than the group without milrinone (4.0 points, IQR: 2.6–5.7; P=0.02) (*Figure 2*). At 24 hours, this value worsened 28% in the group which did not receive milrinone (2.9 points, IQR: 1.8–5.2), while the group with milrinone remained at levels similar to those at admission (2.7 points, IQR: 1.9–4.9) P=0.03.

Microcirculation changes and clinical variables

Nor was an association found between elevated VIS and changes in CBV-Rel (P=0.59), 4–6 CD (P=0.56) or the CBV-RC (P=0.08). An association was found between a high VIS and higher PBR levels in both groups (P=0.02). We also found a moderate correlation between pulse pressure and 7–25-micron capillary density (rho 0.55; P=0.03), but not with 4–6-micron capillary density (rho 0.02; P=0.82) in both groups. Prolonged capillary refill (PCR) on admission was correlated with an abnormal PBR (rho 0.20; P=0.01), and elevated lactate with higher PBR levels (rho 0.24; P=0.01), CBV-CR (rho –0.20; P=0.04) and MVHS™ scores (rho –0.24; P=0.01) in the groups. We found that patients with normal blood pressure and prolonged capillary refill (hemodynamic incoherence) who did not receive milrinone had higher PELOD-2 scores associated with microcirculation dysfunction (*Figure 3*). An association was found between higher PBR levels and greater mortality in the groups with and without milrinone (2.99±0.39 *vs.* 3.40±0.69 microns; P=0.04).

Discussion

In this study, we found that children with septic shock have microcirculation changes associated with the use of milrinone infusions. Specifically, we found that 24 hours after receiving this medication, the 4–6-micron capillary density (functional) was stable, while it decreased in the group that did not receive it. Furthermore, we found that the group that received milrinone was able to maintain capillary recruitment capacity and had less endothelial glycocalyx degradation, while these two variables worsened in the group without milrinone. These findings occurred principally in children under two years old and in patients

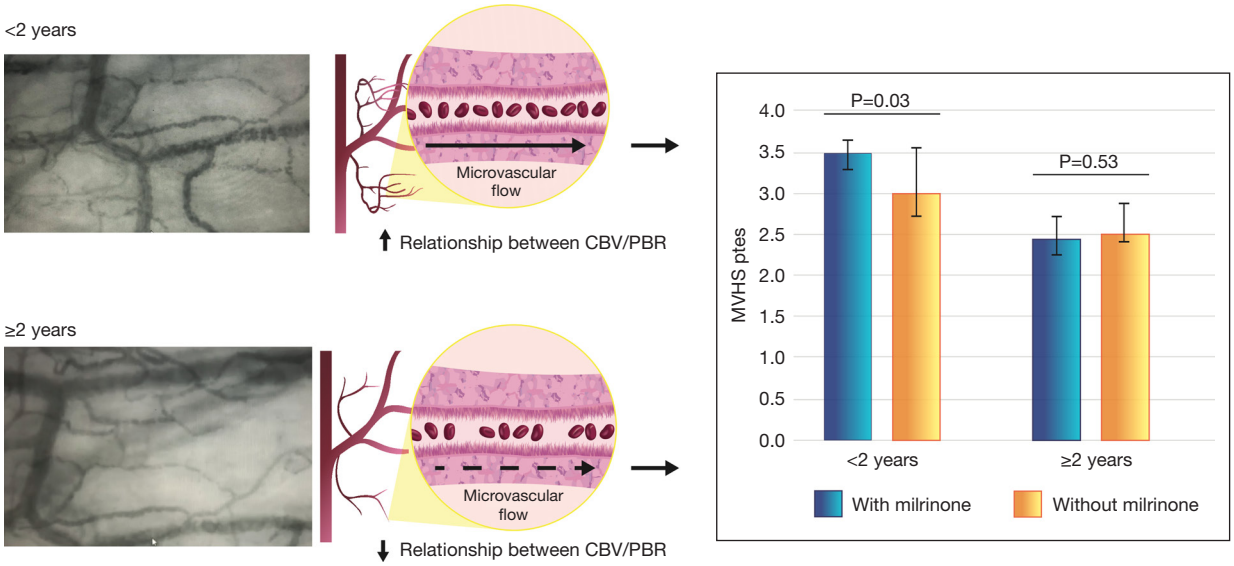

Figure 2 Microvascular blood flow and glycocalyx degradation in patients treated with milrinone according to age. The MVHS measures the relationship between CBV and the PBR. In healthy adults, a value greater than 4 points has been found to be normal (17). Error bars represent 95% confidence interval (Student's *t*-test). CBV, capillary blood volume; PBR, perfused boundary region; MVHS ptes, microvascular health score points.

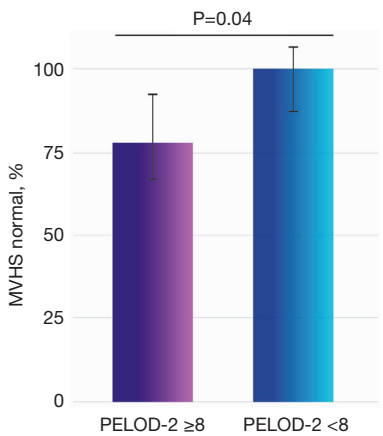

Figure 3 Changes in the microvascular health score in patients with hemodynamic incoherence and organ dysfunction measured 24 hours after admission. Error bars represent 95% confidence interval (Student's *t*-test). MVHS, microvascular health score.

with a greater inflammatory response. We believe there are two explanations for the effects of milrinone on the natural history of microvascular injury in sepsis. First, an indirect effect related to systemic macrocirculation vasodilation, and second, a direct effect on the microcirculation, improving tissue oxygen delivery, optimizing the capacity for capillary recruitment and reducing endothelial structure damage (less glycocalyx degradation).

Microcirculation involvement in sepsis and septic shock has been extensively described in adults (23-25). In our study, we found that the 4–6 CD was low but sustainable at 24 hours, when patients received milrinone infusions. In the group that did not receive milrinone, the 4–6 CD followed its natural course, with a tendency to worsen. De Backer *et al.* found that the main microcirculation characteristic in adult patients with septic shock is a reduced density of capillary blood vessels coupled with high heterogeneity (the presence of capillaries with good and poor perfusion) (26). This same author found that low doses of dobutamine in patients with septic shock recovered microvascular flow, apart from its systemic effects (27). We found that, in patients who received milrinone, 4–6 CD was maintained despite having a greater inflammatory response and disease severity.

Likewise, children under two years old were able to maintain a better functional capillary density. This superior capillary density in those who received milrinone (especially those under two years old) could be explained by changes in diffusive and convective flow associated with this medication. Diffusive flow is one of the most important determinants of oxygen transport to the tissues (16,28,29).

It depends not only on the opening or closing of capillaries (Krogh's classical theory) but also on the diffusion distance between tissues (28). In septic shock, blood vessel density tends to decrease, which increases the oxygen diffusion distance (25). In this same regard, convective flow depends on red blood cell rheology and the capillary driving pressure (the difference between capillary and venule pressure). In children, milrinone can decrease arterial and central venous pressure (CVP) by increasing vessel distensibility (5). Therefore, it has been suggested that its predominant effect is more systemic vasodilatory than inotropic (8). At the microcirculation level, milrinone's effects could be related to the reduction of venule pressure (due to a decrease in CVP), thereby optimizing the capillary driving pressure and thus convective flow.

The microcirculation changes as children grow older (30). The functional capillary density in premature infants' skin decreases significantly in the first month of life (31). These changes are related to decreased hemoglobin concentration and environmental temperature changes. The temperature modifications of functional capillary density may be related to autonomic factors. The adult microcirculation pattern is believed to be reached at the age of three months (29). In this study, we found an age-related difference in the microcirculation's response to milrinone. Children under the age of two who received milrinone had a higher functional capillary density and less endothelial glycocalyx degradation. These changes cannot be explained by the fact that they were younger. Recent sublingual videomicroscopy study findings have shown that there are no age-related microcirculation changes in healthy children (32). Our group recently found that the glycocalyx worsens in children with sepsis and septic shock when unbalanced solutions are used for fluid resuscitation (12). Maintaining capillary density and not worsening vascular flow while receiving milrinone infusions could explain why one of its consequences is less glycocalyx degradation and a modified inflammatory response. In fact, we found that patients who received milrinone had less glycocalyx degradation despite having high ferritin. The PBR elevation in both groups in our study was associated with greater mortality, as observed in studies in adults with septic shock (19,20). The effects of milrinone on the inflammatory response and its outcomes have also been found in animal models (33). At therapeutic doses, this medication has been found to interfere with the inflammatory cascade and platelet aggregation, thus contributing to the restoration of microvascular flow (9,34,35).

In sepsis, especially in advanced stages, there may be a state in which macrocirculation variables (blood pressure, cardiac output) have normalized but microcirculation abnormalities persist. This phenomenon has been termed "hemodynamic incoherence" and is associated with worse outcomes (36). Patients with a normal blood pressure and PCR who did not receive milrinone were more often found to have microcirculation abnormalities. We believe that these abnormalities can be explained by a loss of this coherence between the macro and microcirculation. In fact, we found no correlation between microcirculation changes and high VIS or normal blood pressure. This hemodynamic incoherence phenomenon should be studied more in children. It could appear in advanced stages of the disease or even be a specific phenotype of sepsis or septic shock. The lack of response to fluid resuscitation in advanced stages of septic shock and its association with worse outcomes in places where pediatric ICUs are not available could also be explained by the presence of hemodynamic incoherence. These hypotheses should be further corroborated through prospective studies (2,37).

We believe that our study has several limitations. First, it is the experience of a single center and an observational study which found associations which are not necessarily causal. However, we sought to control for confounding variables and our findings of microcirculation abnormalities are consistent with studies performed in animals and adults with septic shock. In addition, our evaluation was limited to the first 24 hours after the PICU admission of children with septic shock. We did not include children who had received a crystalloid bolus in the emergency room, to avoid this confounding variable. We do not know if a crystalloid bolus prior to PICU admission would affect our study's results, because patients who received such a bolus were excluded in the study design. We are also unaware of what happens in the microcirculation more than 24 hours after receiving milrinone, because we did not conduct follow up after that time. Another limitation of our study is that we did not measure endothelial glycocalyx injury biomarkers. Measurements were taken using videomicroscopy (an indirect measure) which, while having high inter-observer concordance (17), is not a direct measure of glycocalyx damage. Finally, our objective was to explore and learn a bit more about the microcirculation, and endothelial glycocalyx changes in children with septic shock. We did not attempt to determine if milrinone specifically modifies

the inflammatory response in sepsis. Another type of design and measurement of cytokines and other biomarkers are needed to study these effects in more detail.

Conclusions

In this study of children with septic shock, we found that the use of milrinone infusions was associated with microcirculation changes compared with the group of patients who did not receive milrinone. The children who received milrinone were found to maintain functional capillary density and capillary recruitment capacity and have less endothelial glycocalyx degradation 24 hours after its administration. These microcirculation changes associated with the use of milrinone were more significant in children under the age of two and were related to changes in capillary refill, serum lactate and less organ dysfunction. We believe that these findings are explained by a direct effect on the microcirculation and an indirect effect through systemic vasodilation. More studies are needed to deepen the knowledge of microcirculation changes in children with septic shock and their relationship with the use of fluids and medications.

Acknowledgments

Funding: This study was supported by the Clinical Research of Universidad de la Sabana, Faculty of Medicine and Fundacion Cardioinfantil-Instituto de Cardiologia, Research Department, Bogotá, Colombia (No. MED 256-2019).

Footnote

Reporting Checklist: The authors have completed the STROBE reporting checklist. Available at https://tp.amegroups.com/article/view/10.21037/tp-23-619/rc

Data Sharing Statement: Available at https://tp.amegroups.com/article/view/10.21037/tp-23-619/dss

Peer Review File: Available at https://tp.amegroups.com/article/view/10.21037/tp-23-619/prf

Conflicts of Interest: All authors have completed the ICMJE uniform disclosure form (available at https://tp.amegroups.com/article/view/10.21037/tp-23-619/coif). The authors have no conflicts of interest to declare.

Ethical Statement: The authors are accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved. The study was conducted in accordance with the Declaration of Helsinki (as revised in 2013). It was approved by the Institutional Review Board and the ethics committee of Fundación Cardioinfantil-Instituto de Cardiología (IRB-DDI 4814-2023; approved January 20, 2022), and all the patients' parents or legal guardians signed informed consent form prior to being included in the study.

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Cite this article as: Sarta-Mantilla M, Fernández-Sarmiento J, Acevedo L, Mulett H, Nieto A, Lucena N, Lancheros J, Duque C. Microcirculation, endothelium and glycocalyx changes associated with the use of milrinone in children with septic shock. Transl Pediatr 2024;13(5):727-737. doi: 10.21037/tp-23-619