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Early Increases in Microcirculatory Perfusion During Protocol-Directed Resuscitation are Associated with Reduced Multi-Organ Failure at 24 hours in Patients with Sepsis

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Abstract

OBJECTIVE—Sepsis mortality is closely linked to multi-organ failure, and impaired microcirculatory blood flow is thought to be pivotal in the pathogenesis of sepsis-induced organ failure. We hypothesized that changes in microcirculatory flow during resuscitation are associated with changes in organ failure over the first 24 hours of sepsis therapy.

DESIGN—Prospective observational study

SETTING—Emergency Department and Intensive Care Unit

PARTICIPANTS—Septic patients with systolic blood pressure <90 mmHg despite intravenous fluids or lactate ≥4.0 mM/L treated with early goal-directed therapy (EGDT).

MEASUREMENTS AND RESULTS—We performed Sidestream Dark Field (SDF) videomicroscopy of the sublingual microcirculation <3 hours from EGDT initiation and again within a 3–6 hour time window after initial. We imaged 5 sites and determined the mean microcirculatory flow index (MFI) (0=no flow to 3=normal) blinded to all clinical data. We calculated the Sequential Organ Failure Assessment (SOFA) score at 0 and 24 hours, and defined improved SOFA a priori as a decrease ≥2 points. Of 33 subjects; 48% improved SOFA over 0–24 hours. Age, APACHE II, and global hemodynamics did not differ significantly between organ

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RESPONSE TO REVIEWERS

Please see our point-by-point response in a separate file.

DISCLOSURES

Dr. Trzeciak has previously received industry research support from Biosite and Eli Lilly, and currently receives research support from Novo Nordisk and INO Therapeutics. Dr. Shapiro has previously received industry research support from Biosite and Eli Lilly, and currently receives research support from Cumberland Pharmaceuticals. None of the other authors have potential financial conflicts of interest to disclose.

failure groups. Among SOFA improvers, 88% increased MFI during EGDT, compared to 47% for non-improvers (p=0.03). Median change in MFI was 0.23 for SOFA improvers versus −0.05 for non-improvers (p=0.04).

CONCLUSIONS—Increased microcirculatory flow during resuscitation was associated with reduced organ failure at 24 hours without substantial differences in global hemodynamics. These data support the hypothesis that targeting the microcirculation distinct from the macrocirculation could potentially improve organ failure in sepsis.

Keywords

Microcirculation; resuscitation; sepsis; severe sepsis; septic shock; organ failure

INTRODUCTION

Sepsis is a common and lethal disease.[1] Development of acute multi-organ failure is one of the primary determinants of sepsis mortality.[1–4] Early evidence of multi-organ failure and early changes in organ function, specifically changes over the first 24 hours of severe sepsis presentation, are especially prognostic.[2,4] However, the pathogenic events leading to sepsis-induced organ failure are not entirely understood, and this knowledge gap represents a major challenge to the development of new therapies to ameliorate acute organ failure in septic patients.

The microcirculation, the network of blood vessels <100μm in diameter throughout the body, is an integrated functional system that is the principal site of oxygen transport from blood to underlying tissues and the chief regulator of tissue oxygen delivery to meet cellular oxygen demands. There is abundant data that microcirculatory homeostasis is profoundly disrupted in sepsis, and microcirculatory failure is a hallmark of the septic state.[5,6] Experimental studies show that sepsis is characterized by markedly impaired microcirculatory flow velocity, stopped-flow microvessels, decreased perfused vessel density, and increased spatial heterogeneity of perfused vessels, with a concomitant and marked alteration of tissue oxygen transport.[7–10] Clinical studies using *in vivo* videomicroscopy have confirmed that microcirculatory dysfunction is also a feature of human sepsis, and the severity of microcirculatory derangements predicts mortality.[11–13]

Since early perfusion abnormalities are thought to contribute to sepsis-induced organ failure, we aimed to test the hypothesis that early changes in microcirculatory blood flow during resuscitation are associated with changes in organ failure over the first 24 hours in patients with sepsis. The purpose of this study was to determine if early improvement in microcirculatory blood flow is associated with a reduction in multi-organ failure in septic patients, which would suggest that the microcirculation could be a new therapeutic target for the treatment of sepsis-induced organ failure.

METHODS

Study Design

Single-center prospective observational study.

Setting

Emergency Department (ED) and Intensive Care Unit (ICU) of an urban academic medical center (Cooper University Hospital, Camden, New Jersey, USA).

Human Subjects Protection

The Institutional Review Board approved this study. We obtained informed consent from subjects or next of kin.

Selection of Participants

The subjects of this study were patients with sepsis meeting criteria for an institutional early goal-directed therapy (EGDT) resuscitation protocol over 18 months. We enrolled sepsis patients non-consecutively, with enrollment dependent on investigators' ability to perform microcirculatory videomicroscopy at early time points (defined below). Inclusion criteria were: (1) age >17 years; (2) confirmed or suspected infection plus two or more signs of the systemic inflammatory response syndrome (definition of sepsis by standardized criteria[14]); (3) one or both of the following triggers of our EGDT protocol[15]: (a) initial systolic blood pressure < 90 mmHg despite a 20 cc/kg intravenous crystalloid fluid challenge, or (b) initial serum lactate ≥ 4 mmol/L; (4) invasive hemodynamic monitoring established for the purpose of EGDT; and (5) ability to perform initial videomicroscopy ≤3 hours from EGDT initiation (defined as the time of catheter insertion for invasive hemodynamic monitoring for the purpose of protocol-directed resuscitation). Exclusion criteria were: (1) inability to perform sublingual videomicroscopy [e.g. inability to place the probe under the tongue due to inability to open the mouth or patient requirement of a high-flow face mask for supplemental oxygen (although SDF imaging could be performed in patients with an endotracheal tube or nasal cannula)]; and (2) patient or next of kin refusal to participate.

Interventions

Cardiovascular support was guided by our institutional protocol for EGDT.[15] Early goal-directed therapy is a resuscitation protocol for patients with evidence of acute sepsis-induced tissue hypoperfusion, and it is recommended by international consensus guidelines for sepsis management.[16,17] Our institution has been using EGDT in routine practice since 2004, and subjects in this study were treated with EGDT as part of standard care.[15] Briefly, our EGDT protocol (an adaptation of the original protocol from Rivers *et al*[18]) uses intravenous volume expansion and (if needed) vasopressors, inotropes, or blood products in a stepwise manner to achieve pre-defined quantitative endpoints of resuscitation derived from invasive hemodynamic monitoring: central venous pressure (CVP) ≥8 mmHg, mean arterial pressure (MAP) ≥65 mmHg, and central venous oxygen saturation ($ScvO_2$) ≥70%. Our EGDT protocol can be utilized for sepsis patients in the ED; alternatively, for inpatients that develop severe sepsis in the hospital, our protocol can be initiated upon arrival to the ICU, where a pulmonary artery catheter can be used in place of a central venous catheter. If a pulmonary artery catheter is used, mixed venous oxygen saturation (SvO_2) ≥65% replaces the $ScvO_2$ target.[13]

Methods of Measurement, Data Collection, and Processing

We directly visualized the sublingual microcirculation with sidestream dark field (SDF) videomicroscopy, a minimally-invasive method of imaging the microcirculation beneath mucosal surfaces. The SDF instrument (Microscan, Microvision Medical, Amsterdam) has a 5× objective giving 326× magnification. The technique consists of a hand-held videomicroscope containing a ring of stroboscopic light emitting diodes.[19] The light is absorbed by hemoglobin so that red blood cells appear dark, yielding high-contrast video of blood flow in submucosal microvessels. The technique [including its predecessor, Orthogonal Polarization Spectral (OPS) imaging] is well validated in both experimental models and human subjects.[19–22] We utilize the sublingual space for imaging because both direct and indirect assessments of tissue perfusion at the sublingual site have previously

been demonstrated to predict mortality in critically ill patients, including those with sepsis.[11–13,23] Real-time examples of healthy and dysfunctional microcirculation are available from the authors for viewing or download at: http://www.cooperhealth.org/content/gme_fellowship_shock.htm.

The timeline for study activities appears in Figure 1. We obtained initial SDF video sequences as soon as possible after EGDT initiation, within the first 3 hours after catheter insertion in all cases. The initial imaging time point is hereafter referred to as **Visit 1**. We obtained the second set of SDF video sequences as early as possible within a time window 3–6 hours after the initial SDF study. We established this as a time window for imaging because of the potential for unavailability of the subject at a single fixed time point due to patient-centered or logistical factors (e.g. immediate surgery, transport of the patient to CT scan, other bedside procedures being simultaneously performed, etc.). The second imaging time point is hereafter referred to as **Visit 2**. Global hemodynamic data (described below) corresponding to Visit 1 and Visit 2 were recorded simultaneously with SDF videomicroscopy.

A detailed description of our standard operating procedure for image acquisition is available in the electronic supplementary material **(ESM1)**. Briefly, we placed the SDF probe in the sublingual space without pressure after removal of secretions. We obtained video sequences of 20 seconds each from five different sublingual sites and recorded them on digital cassette tapes. We stored images by code without source patient identifiers so that the data could be analyzed off-line blinded to all clinical data. We converted videos from tape to audio video interleaved (AVI) file format with video processing software and used a random number generator (1 to 10,000) to assign random number codes to each video clip, so that image analysis was blinded to the identity of the subject as well as the time point that each video was obtained.

We determined the microcirculatory flow index (MFI) with a semi-quantitative methodology originally described by Spronk *et al*[24] (0=absent; 1=intermittent; 2=sluggish; 3=normal); this methodology for MFI has previously shown good inter-rater agreement among multiple raters ($\kappa = 0.77 – 0.85$).[13,25] We calculated the MFI for all 4 quadrants of the image and averaged to yield a single MFI for each sublingual site. We averaged the 5 sites to give a single MFI value for each time point. Our analysis technique for determining MFI is identical to that described in consensus conference recommendations.[26] A single observer (ST) performed all image analysis; a second trained observer (RCA) analyzed 10% of the sample selected at random and we calculated inter-rater agreement for MFI using the κ statistic.

At each of the imaging time points, all hemodynamic data (CVP, MAP, $ScvO_2$) were recorded simultaneously with SDF imaging. We also calculated the cumulative vasopressor index (CVI), a method for quantifying total amount of vasopressor support at a given time point. The CVI assigns cumulative points for equivalent doses of commonly used vasopressor agents and appears in the electronic supplementary material (**ESM2**).[27–29]

At the time of EGDT initiation and 24 hours later (see timeline, Figure 1), we calculated the sequential organ failure assessment (SOFA) score.[30] As per our previous methodology,[13] we use the SOFA score as modified by Vincent *et al*,[31] which omits the neurologic function component (because of previously reported potential challenges with inter-rater agreement on determination of Glasgow Coma Score). The components of the modified SOFA score appear in the electronic supplementary material (**ESM3)**. If arterial blood gases were not measured at both 0 and 24 hours, we substituted the SaO_2/FiO_2 ratio for PaO_2/FiO_2 ratio according to the conversion technique from Pandharipande *et al*.[32] If serum bilirubin

was not measured, we assumed the value to be normal (0 points) at both 0 and 24 hours. The 0 and 24 hour SOFA calculations were blinded to all microcirculatory data.

Primary Analysis

Subjects were divided into two groups, SOFA improvers and SOFA non-improvers, based on the 0–24 hour ΔSOFA. We defined improved SOFA *a priori* as a decrease of two or more SOFA points over the first 24 hours after EGDT initiation on the basis of our previous data, in which we observed mean SOFA scores at 0 and 24 hours of (mean [95%CI]) 7.3 [6.3–8.5] and 6.3 [4.9–7.7], respectively, mean difference (ΔSOFA) −1.0 [95%CI 0 to −2]. [13,33] A 2-point improvement was used on the grounds that (1) −2 points was the lower limit of the 95% confidence interval that we observed for ΔSOFA in our prior work, and (2) a full point ΔSOFA (rather than a fraction of a point) would be meaningful from a clinical perspective. If the initial (0-hour) SOFA score was 1, we classified the subject as a SOFA improver only if all organ dysfunction resolved at 24 hours (i.e. SOFA=0). If the subject died before the 24 hour mark, we classified the subject as a SOFA non-improver.

The primary covariate of interest used to characterize microcirculatory flow was ΔMFI between Visit 1 and Visit 2. Comparing SOFA improvers versus non-improvers, we analyzed: (1) the difference in proportions of subjects with an increase in MFI using the z-statistic, and (2) the difference in median values for ΔMFI using Mann-Whitney U test. In order to further analyze the relationship between ΔMFI and ΔSOFA, we performed a pre-planned secondary analysis for the entire cohort comparing a normalized raw value for change in microcirculatory flow [percent change in MFI (ΔMFI divided by initial MFI)] versus raw values for ΔSOFA using linear regression with ΔSOFA as the dependent variable. We compared global hemodynamic data at Visit 1 and Visit 2, and the change between Visit 1 and 2, between SOFA improvers and non-improvers by independent samples t-test or Mann-Whitney as appropriate depending on whether or not the data were normally distributed. We used SigmaStat (Systat Software, San Jose, CA) for all analyses.

RESULTS

Characteristics of the Study Subjects

We enrolled 33 subjects. Table 1 displays data for demographics, severity of illness scores, and baseline physiologic data at the time of EGDT initiation, as well as simultaneous measurements of global hemodynamic and microcirculatory indices at Visit 1, for all subjects and both organ failure groups. Sixteen patients (48%) were SOFA improvers, and 17 (52%) were SOFA non-improvers. The mean SOFA scores at 0 and 24 hours were 5.8 [95%CI 4.8–6.8] and 4.7 [3.5–5.9], respectively, mean difference (ΔSOFA) −1.1 [95% CI −0.1 to −2.0]. The in-hospital mortality was 11/33 (33%) for the entire cohort, 3/16 (19%) for SOFA improvers, and 8/17 (47%) for SOFA non-improvers.

Main Results

We performed a total of 66 SDF videomicroscopy studies (330 video sequences) in the 33 subjects. Inter-rater agreement (κ) for image analysis was very good (0.87). Among SOFA improvers, 14/16 (88%) increased MFI, compared to 8/17 (47%) for SOFA non-improvers (p=0.03). Table 2 displays the change in global hemodynamic indices and MFI from Visit 1 to Visit 2 for all subjects and both organ failure groups. Figure 2 displays parallel univariate plots for the change in MFI between Visit 1 and 2 stratified by organ failure group. Median change in MFI was higher for SOFA improvers compared to SOFA non-improvers (0.23 versus −0.05, p=0.04). Median change in MFI was also higher for subjects that survived to hospital discharge compared to subjects who died in the hospital (0.13 versus −0.05, p=0.049). Individual patient data is shown in the electronic supplementary material (**ESM4**).

On linear regression (n=33), percent change in MFI was associated with the raw score for ΔSOFA, and the variables were inversely related (r=−0.52, p=0.002). Figure 3 displays the mean percent change in MFI stratified by quartile of ΔSOFA score (Quartile I = improved organ function; Quartile IV = worsening organ function).

DISCUSSION

Multiple organ failure is a pivotal event in the pathogenesis of sepsis, a hallmark of the disease, and a critical determinant of outcome. In the United States alone, more than 750,000 persons develop sepsis-associated organ dysfunction every year, and of these, 29% percent (215,000 patients) do not survive.[1] Population-based studies have identified that mortality risk in sepsis increases roughly 20% with each additional organ system failure.[1,3] These data underscore the importance of sepsis and ensuing organ failure from a public health perspective.

Early evidence of organ failure is particularly important from a prognostic standpoint,[4] and early changes in organ failure appear to be the most revealing signs of the trajectory of the disease course.[2] In a multi-center study of 1036 severe sepsis patients, Levy *et al* reported that mortality is closely related to early (i.e. first 24 hours) improvement, or lack thereof, in organ function, and that change in organ function on subsequent days may have little additional impact on the likelihood of survival.[2] Based on these and other data, thought leaders have advocated organ failure assessment as an important acute-phase outcome measure in sepsis clinical trials.[34,35] Because the triggers of acute organ failure in sepsis are incompletely understood, studies aimed at the identification of potential new therapeutic targets in multi-organ failure pathogenesis are high priority for sepsis research.

In this study we performed sublingual *in vivo* videomicroscopy in septic patients to test the hypothesis that augmentation of microcirculatory perfusion during the resuscitation phase of therapy is associated with reduction in organ failure at 24 hours. We found a significantly higher proportion of subjects with increased MFI and a significantly higher change in MFI during the early resuscitation phase in subjects with improvement in organ failure, compared to subjects with persistent or worsening organ failure. In addition, linear regression showed that early percent change in MFI was associated with the total amount of change in organ failure (i.e. the raw value for 0–24 hour ΔSOFA). The associations between microcirculatory changes and organ failure were not accompanied by any major differences in global hemodynamics between organ failure groups, suggesting that conventional bedside cardiovascular monitoring may not provide reliable surrogates for tracking the status of the microcirculation. This suggests that microcirculatory dysfunction in sepsis is a reflection of intrinsic events occurring in the microvasculature (rather than merely a byproduct of global hemodynamic effects), and that microcirculatory indices may yield physiologic and prognostic information that global hemodynamic monitoring cannot.

Our results build upon the findings of other studies. Using sublingual OPS imaging we previously reported a significant inverse linear correlation between MFI and SOFA score at a single early time point in sepsis therapy.[13] In a longitudinal study of septic shock patients, Sakr *et al* performed serial (daily) sublingual OPS imaging and found persistent microcirculatory perfusion impairment in patients who died with multiple organ failure, whereas microcirculatory alterations improved rapidly (i.e. between the first and second daily measurements) in survivors.[12] Our current study is unique in that we visualized the microcirculation at two time points during the early resuscitation phase of therapy and quantified the relationship between early changes in microcirculation and the development of organ failure over the first 24 hours. We also used protocol-directed resuscitation with EGDT to help ensure macrocirculatory homogeneity (CVP, MAP, $ScvO_2$) among study

subjects, which helped us to better isolate (and test hypotheses about) the potential role of the microcirculation. Overall, our study contributes to the available body of evidence that microcirculatory derangements can be an important component of multi-organ failure in sepsis.

We recognize limitations in interpreting our findings. Although the median values for ΔMFI are significantly different, visual inspection of the data (Figure 2) reveals that there is still overlap in ΔMFI values between organ failure groups, so that from a clinical standpoint it could be challenging to use an early ΔMFI value as a single marker of prognosis. In addition, our methodology did not include measurement of vascular density,[11] which may yield different information than assessment of flow velocity alone. To some extent the observed differences in MFI between SOFA improvers and SOFA non-improvers tracked changes in vasopressor utilization, but given that the severity of cardiovascular organ system failure and vasopressor utilization are inextricably linked, we are unable to comment on what role extrinsic application of vasopressor agents played, if any. In addition, there may have been early changes in microcirculatory flow velocity that were missed before the first SDF imaging time point, but using a shorter time window in the inclusion criteria would not have been logistically feasible. As with any study in a sample of this size, it is also possible that individual data points (and in particular individuals with large changes for ΔMFI) had a major impact on the median values for the group, and thus our findings would be bolstered by confirmation in a larger sample. Probably the most important limitation of this study (or any such observational study) is that association does not necessarily indicate causality.

As early development of organ failure is a critical determinant of sepsis survival, the identification of new therapeutic targets to ameliorate multi-organ failure in sepsis is of paramount importance. Microcirculatory dysfunction represents one potential therapeutic target. Going forward, the link between microcirculatory dysfunction and multi-organ failure in sepsis should be further tested with randomized clinical trials of novel therapies or therapeutic strategies to counteract microcirculatory failure, examining the impact of these strategies on both microcirculatory indices and organ function.

CONCLUSIONS

Early increases in microcirculatory blood flow during protocol-directed resuscitation were associated with reduced organ failure at 24 hours in patients with sepsis. These data support the hypothesis that targeting the microcirculation distinct from the macrocirculation could potentially improve organ failure in sepsis.

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APPENDIX

Electronic Supplementary Material

ESM1

Standard Operating Procedure for Sidestream Dark Field (SDF) sublingual microcirculatory image acquisition.[13]

1. Remove secretions from the surface of the sublingual mucosa with a suction catheter or gauze.
2. Place a sterile disposable cap on the videomicroscope probe.
3. Position the probe on the sublingual mucosal surface.
4. The following steps are to be followed for proper image collection and must be repeated prior to the recording of images at each sublingual site:
    a. Position the probe so that the microcirculation comes into view.
    b. Gently advance the probe into the sublingual area until the flow is partially or completely occluded.
    c. Retract the probe from the sublingual mucosal surface until contact with the tissue is lost.
    d. Just before contact is lost, you will see what the flow looks like with no pressure. This represents an acceptable image quality for recording data.
    e. Advance the probe again slowly until contact is regained and the microcirculation comes into view (as in 4d above).
    f. Focus the image.
5. Record 5 video clips of 20 seconds each.
    a. Obtain two of the clips from the left side of the frenulum of the tongue and two from the right side. The fifth clip can be obtained from either side depending on where the clearest images can be obtained (operator discretion).

ESM2

The cumulative vasopressor index (CVI).[27–29]

VASOPRESSOR	Dose range 1 Point	Dose range 2 Points	Dose range 3 Points	Dose range 4 Points
Dopamine (mcg/kg/min)	0 < dose ≤ 5	5 < dose ≤ 10	10 < dose ≤ 15	>15
Epinephrine (mcg/kg/min)	---	0 < dose ≤ 0.05	0.05 < dose ≤ 0.1	>0.1
Norepinephrine (mcg/kg/min)	---	0 < dose ≤ 0.05	0.05 < dose ≤ 0.1	>0.1
Phenylephrine (mcg/kg/min)	---	0 < dose ≤ 0.4	0.4 < dose ≤ 0.8	>0.8
Vasopressin (units/min)	---	---	---	any dose

ESM3

The sequential organ failure assessment (SOFA) score as modified by Vincent *et al*.[31] The total SOFA score is a cumulative value of the points from each organ system.

SOFA points†	1	2	3	4
Respiration PaO_2/FiO_2 ratio	<400	<300	<200	<100
Coagulation Platelets × $10^3/mm^3$	<150	<100	<50	<20
Liver Bilirubin, mg/dl	1.2–1.9	2.0–5.9	6.0–11.9	≥12.0
Cardiovascular Hypotension*	MAP <70	Dopamine ≤ 5 or Dobutamine (any)	Dopamine >5 or norepinephrine ≤ 0.1	Dopamine >15 or norepinephrine >0.1
Renal Creatinine, mg/dl	1.2–1.9	2.0–3.4	3.5–4.9	≥5.0

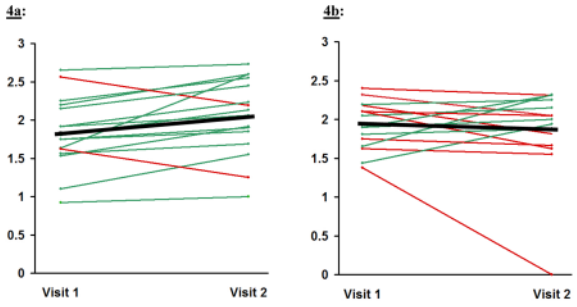
[MAP = mean arterial pressure; PaO_2 = partial pressure of arterial oxygen; FiO_2 = fraction of inspired oxygen]

† If parameters are normal assign a score of 0 for that organ system.

* Units for vasoactive mediations: dopamine mcg/kg/min; norepinephrine mcg/min.

ESM4

Individual patient data for MFI values at Visit 1 and Visit 2 for SOFA improvers (4a) and SOFA non-improvers (4b). Green lines represent subjects with any increase in MFI from Visit 1 to Visit 2. Red lines represent subjects with any decrease in MFI from Visit 1 to Visit 2. The black line represents the mean values.



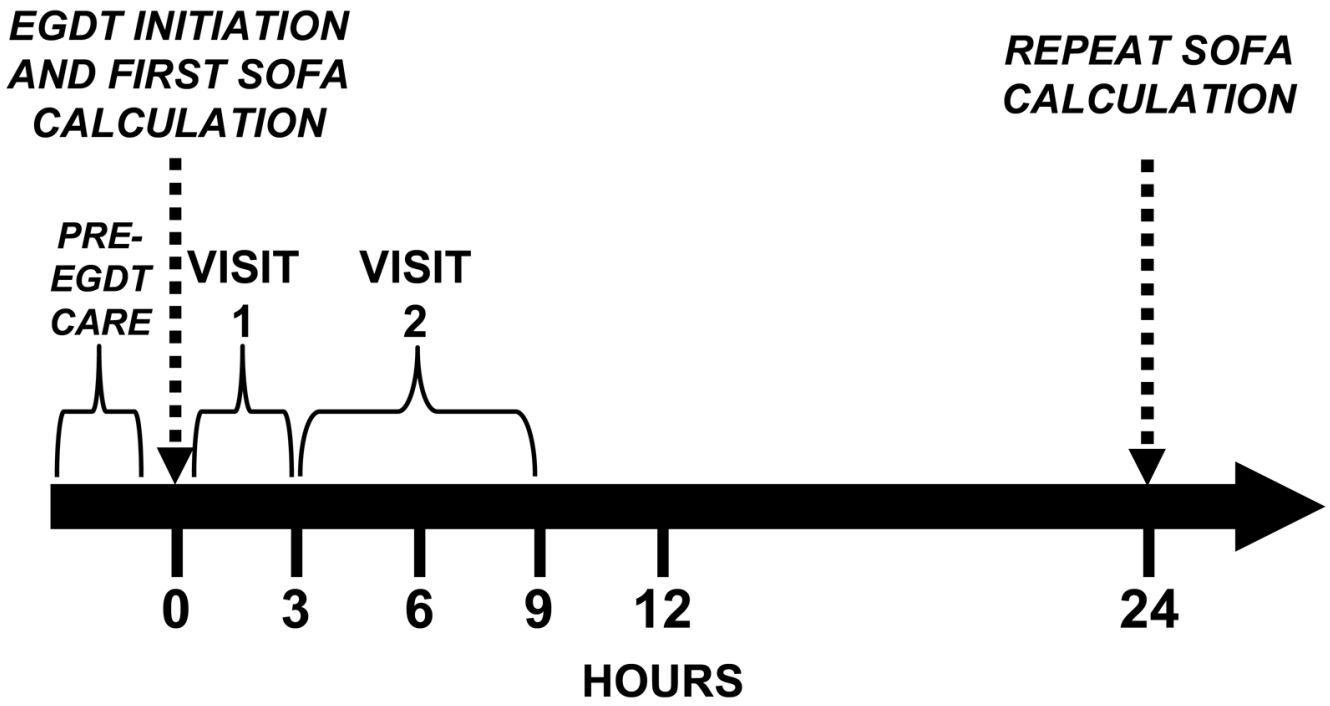


Figure 1.

Timeline of study activities. Time zero was the time of early goal-directed therapy (EGDT) initiation, defined as the time of insertion of a central venous catheter for invasive monitoring for protocol-directed resuscitation. Sequential organ failure assessment (SOFA) scores were calculated at 0 and 24 hours. We performed initial SDF videomicroscopy as soon as possible after EGDT initiation, within 3 hours of catheter insertion in all cases (Visit 1), and we repeated SDF (Visit 2) as soon as possible within a 3–6 hour window after initial the SDF study.

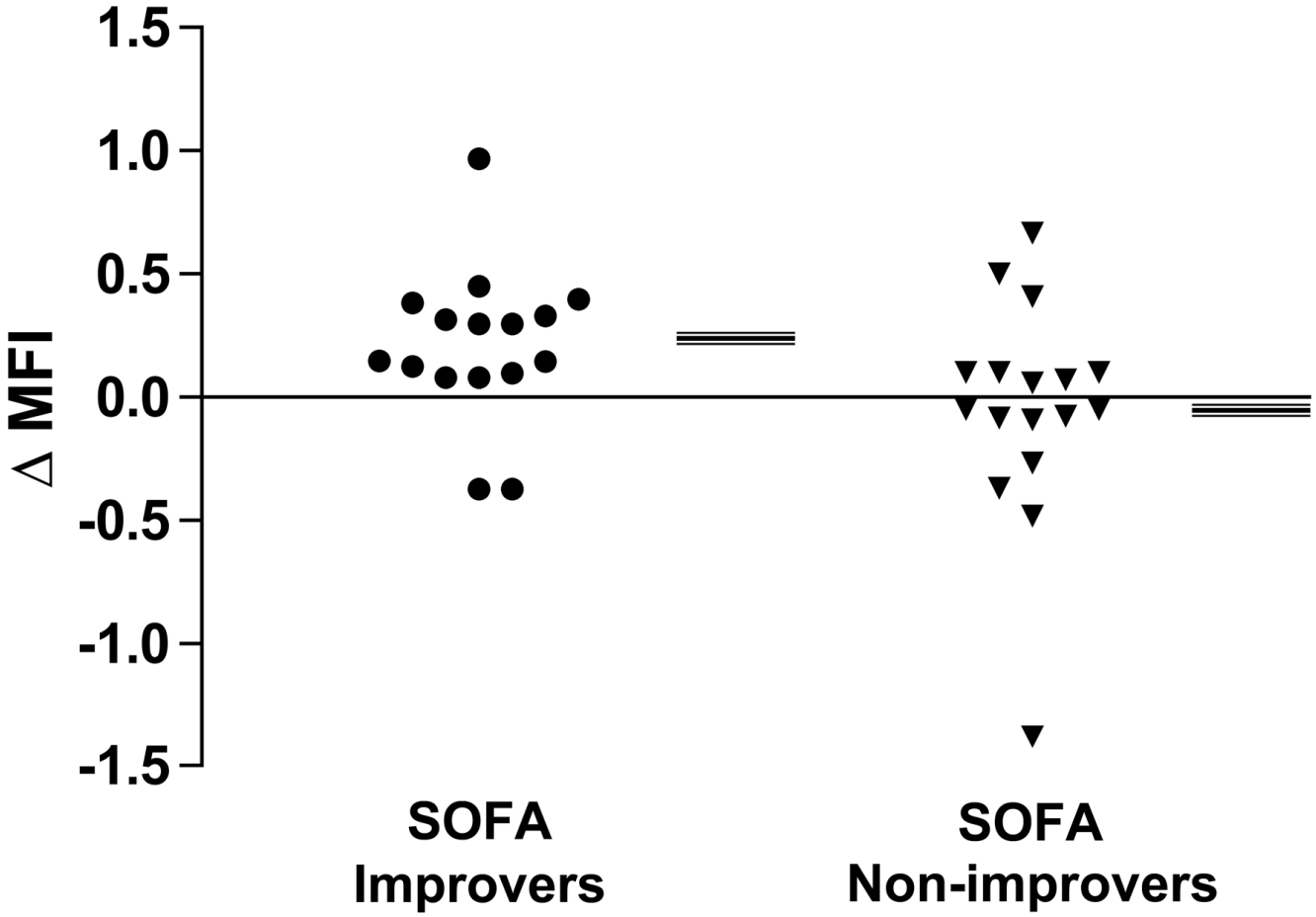


Figure 2.
Parallel univariate plots of the change in microcirculatory flow index (ΔMFI) between early time points (Visit 1 and 2) in both organ failure groups. The horizontal lines to the right of each plot represent median values for ΔMFI for each organ failure group. The median ΔMFI for organ failure improvers was a rise of 0.23, and for non-improvers was −0.05 (p=0.04). The proportion of subjects that increased MFI was higher in SOFA improvers compared to non-improvers (88% vs. 47%, p=0.03). [SOFA = Sequential Organ Failure Assessment score]

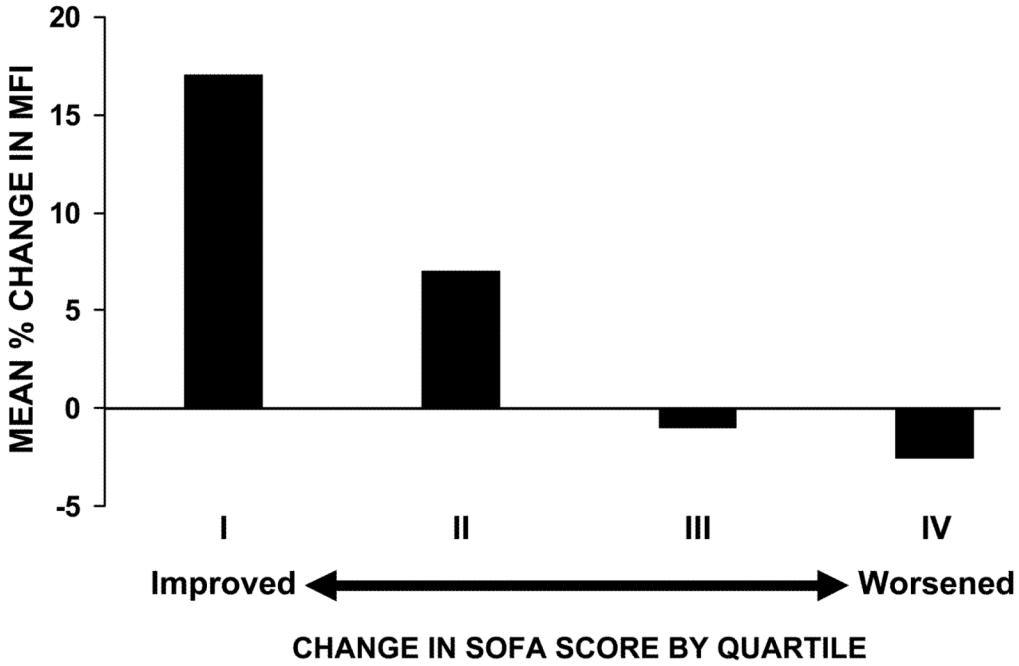


Figure 3.
Mean values for percent change in microcirculatory flow index (MFI) stratified by quartile of raw values for change in organ function (ΔSOFA). [Quartile I = improved organ function; Quartile IV = worsening organ function]

Table 1

Baseline characteristics of all study subjects and both organ failure subgroups. Physiologic parameters were measured at the time of EGDT initiation, except where noted.

	All subjects (n = 33)	SOFA improvers (n = 16)	SOFA non-improvers (n = 17)	p-value*
Age, years [mean (SD)]	63 (14)	62 (16)	64 (13)	0.64
Gender, female [n, (%)]	13 (39)	7 (44)	6 (35)	0.73
Origin [n, (%)]				
Emergency Department	17 (52)	9 (56)	8 (47)	0.73
Inpatient	16 (48)	7 (44)	9 (53)	0.73
Site of infection [n, (%)]				
Lung	10 (31)	4 (25)	6 (35)	0.71
Urinary tract	11 (33)	6 (38)	5 (29)	0.72
Abdomen	5 (15)	1 (6)	4 (24)	0.17
Skin/soft tissue	2 (6)	2 (13)	0 (0)	0.13
Undetermined primary source	5 (15)	3 (18)	2 (12)	0.58
APACHE II score [mean (SD)]	21 (9)	19 (9)	23 (9)	0.19
SOFA score† [mean (SD)]	5.8 (2.9)	5.8 (3.2)	5.8 (2.7)	0.94
Vasopressor support [n, (%)]	16 (48)	10 (63)	6 (35)	0.17
PaO_2/FiO_2 ratio [n, (%)]				
≥400	11 (33)	6 (38)	5 (29)	0.72
300–399	10 (31)	5 (31)	5 (29)	0.99
200–299	1 (3)	0 (0)	1 (6)	0.99
100–199	8 (24)	3 (18)	5 (29)	0.69
<100	3 (9)	2 (13)	1 (6)	0.60
Platelets, 10^3/mL [mean (SD)]	287 (167)	304 (181)	272 (156)	0.60
Bilirubin, mg/dL [mean (SD)]	1.6 (1.6)	1.6 (1.8)	1.7 (1.7)	0.88
Creatinine, mg/dL [mean (SD)]	2.8 (2.1)	2.2 (1.8)	3.3 (2.3)	0.16
Serum lactate, mM/L [mean (SD)]	3.5 (2.9)	2.9 (2.4)	4.1 (3.3)	0.24
Global Hemodynamics†† [mean (SD)]				
Central venous pressure, mmHg	12 (6)	11 (5)	12 (8)	0.79
Mean arterial pressure, mmHg	73 (14)	74 (15)	72 (13)	0.59
Central venous oxygen saturation§, %	67 (13)	64 (14)	69 (13)	0.36
Cumulative vasopressor index	1.5 (2.0)	2.1 (2.3)	0.9 (1.5)	0.12
Microcirculation†† [mean (SD)]				
Microcirculatory flow index	1.89 (0.38)	1.83 (0.47)	1.93 (0.29)	0.47

[SOFA = sequential organ failure assessment; APACHE = Acute Physiology and Chronic Health Evaluation]

*Comparing SOFA improvers versus SOFA non-improvers. Unpaired t-test was used in all cases except for cumulative vasopressor index in which Mann-Whitney test was used (data not normally distributed).

†Sequential Organ Failure Assessment score modified according to Vincent *et al*[31]

††Measured at Visit 1 (simultaneous global hemodynamic and microcirculatory assessment)

[§]Measured in the superior vena cava [central venous oxygen saturation ($ScvO_2$)] for subjects with a central venous catheter and measured in the pulmonary artery [mixed venous oxygen saturation (SvO_2)] for subjects with a pulmonary artery catheter

Table 2

Change in global hemodynamic and microcirculatory indices between Visit 1 and 2 for all subjects and both organ failure subgroups.

	All subjects (n = 33)	SOFA improvers (n = 16)	SOFA non-improvers (n = 17)	p-value*
Global Hemodynamics [mean (SD)]:				
ΔCentral venous pressure, mmHg	0 (4)	−2 (3)	3 (5)	<0.01
ΔMean arterial pressure, mmHg	2 (12)	5 (13)	−1 (10)	0.13
ΔCentral venous oxygen saturation§, %	3 (13)	7 (10)	0 (15)	0.57
ΔCumulative vasopressor index	0.3 (1.3)	−0.13 (1.3)	0.65 (1.3)	0.20
Microcirculation [mean (SD)]:				
ΔMicrocirculatory flow index	0.08 (0.41)	0.21 (0.31)	−0.05 (0.45)	0.04

*Comparing SOFA improvers versus SOFA non-improvers. Unpaired t-test was used, except for central venous oxygen saturation and microcirculatory flow index in which Mann-Whitney test was used (data not normally distributed).

§Measured in the superior vena cava [central venous oxygen saturation ($ScvO_2$)] for subjects with a central venous catheter and measured in the pulmonary artery [mixed venous oxygen saturation (SvO_2)] for subjects with a pulmonary artery catheter.