A clinical and physiological prospective observational study on the MAnagement of Pediatric Shock in the post-FEAST trial era (MAPS study)

Nchafatso G. Obonyo, MB.ChB, PhD[1,2,3,4], Peter Olupot-Olupot, MB.ChB, MPH, PhD[2,3,7], Ayub Mpoya, MSc[1], Julius Nteziyaremye, MB.ChB[3], Martin Chebet, MB.ChB[3,7], Sophie Uyoga PhD[1,2], Rita Muhindo MSc[3], Jonathon P. Fanning MB.ChB, PhD[4,5], Kenji Shiino, MD, PhD[4,6], Jonathan Chan MBBS, PhD[4,6], John F. Fraser, MB.ChB, PhD[4,5,6], Kathryn Maitland, FMedSci, PhD[1,8]*

Author's affiliations:

[1]Kenya Medical Research Institute, Wellcome Trust Research Programme, Kilifi, Kenya

[2]Initiative to Develop African Research Leaders, Kilifi, Kenya

[3]Mbale Clinical Research Institute, Mbale, Uganda

[4]Critical Care Research Group, The Prince Charles Hospital, Brisbane, Australia

[5]Faculty of Medicine, University of Queensland, Brisbane, Australia

[6]School of Medicine, Griffith University, Gold Coast, Australia

[7] Busitema University, Faculty of Health sciences, Mbale, Uganda

[8] Department of Infectious Disease and Institute of Global Health and Innovation, Division of Medicine, Imperial College, London

The study was conducted at Mbale Regional Referral Hospital in Mbale, Uganda and Kilifi County Hospital in Kilifi, Kenya

MAPS study was funded by an Institutional Strategic Support Fund Award, Wellcome Trust Centre for Global Health Research, Imperial College London (Grant code 105603/Z/14/Z).

*Corresponding author: k.maitland@imperial.ac.uk Faculty of Medicine, Medical School Building St Mary's Campus, Imperial College, London, United Kingdom W2 1PG

Keywords: Shock, Pediatrics, Management, Fluid Resuscitation, Cardiac biomarkers, echocardiography

Copyright Form Disclosure: Dr. Obonyo received support for article research from Imperial College London (Institutional Strategic Support Funds, Wellcome Centre for Global Health Research) and the Initiative to Develop African Research Leaders/KEMRI-Wellcome Trust Research Programme. Drs. Olupot-Olupot, Nteziyaremye, Muhindo, and Maitland received support for article research from Research Councils UK. Drs. Nteziyaremye, Chebet, Muhindo, and Maitland received support for article research from Wellcome Trust/COAF. Dr. Chebet received funding from Mbale Clinical Research Institute. The remaining authors have disclosed that they do not have any potential conflicts of interest.

Abstract

Objectives Fluid bolus resuscitation in African children is harmful. Little research has evaluated physiological effects of maintenance-only fluid strategy.

Design We describe the efficacy of fluid-conservative resuscitation of septic shock using case-fatality, hemodynamic and myocardial function endpoints.

Setting Pediatric wards of Mbale Regional Referral Hospital, Uganda and Kilifi County Hospital, Kenya conducted between October 2013-July 2015. Data were analysed from August 2016 to July 2019.

Patients Children (≥60 days to ≤12 years) with severe febrile illness and clinical signs of impaired perfusion.

Interventions Intravenous maintenance fluid (4mL/kg/hr) unless children had World Health Organization (WHO) defined shock (≥3 signs) where they received two fluid boluses (20mls/kg) and transfusion if shock persisted. Clinical, electrocardiographic, echocardiographic and laboratory data were collected at presentation, during resuscitation and on Day-28. Outcome measures were 48-hour mortality, normalisation of hemodynamics and cardiac biomarkers.

Measurement and Main Results Thirty children (70% males) were recruited, 6 had WHO shock, all of whom died (6/6) versus 3/24 deaths in the non-WHO shock. Median fluid volume received by survivors and non-survivors were similar (13 [interquartile range (IQR) 9-32] versus 30 [28-61] mL/kg, z=1.62, p=0.23). By 24 hours, we observed increases median (IQR) stroke volume index (mL/m^2 39 (32-42) to 47 (41-49) and a measure of systolic function: fractional shortening from 30 (27-33) to 34 (31-38) from baseline including children managed with no-bolus. Children with WHO-shock had a higher mean level of cardiac troponin (*t*=3.58; 95%CI 1.24-1.43, p=0.02) and alpha-atrial natriuretic peptide (*t*=16.5; 95%CI 2.80-67.5, p<0.01) at admission compared to non-WHO shock. Elevated troponin (>0.1 µg/mL) and hyperlactemia (>4mmol/L) were putative makerss predicting outcome.

Conclusion Maintenance-only fluid therapy normalised clinical and myocardial perturbations in shock without compromising cardiac or hemodynamic function whereas fluid-bolus management of WHO shock resulted in high fatality. Troponin and lactate biomarkers of cardiac dysfunction could be promising outcome predictors in pediatric septic shock in resource-limited settings.

Article Tweet: MAPS study provides echocardiographic and physiological evidence of normalisation in African children with shock managed with no fluid boluses.

Research in Context

- The 2020 surviving sepsis guidelines recommends that fluid resuscitation in low resource settings should be reserved only for children with hypotension following the publication of the large Phase III trial (FEAST) which demonstrated excess mortality in children with non-hypotensive shock.
- There has been some reticence to adopt a no-bolus strategy, in the absence of physiological studies demonstrating the mechanism of harm and evidence of correction of cardio-haemodynamic measures in children treated with this fluid- conservative strategy.
- We provide a small, detailed observational study evidence demonstrating the normalisation of cardiac and haemodynamic perturbations of shock managed with a no-bolus strategy and evidence of high mortality in children who receiving boluses in accordance with the 2013 World Health Organization pediatric guidelines.

Introduction

International pediatric septic shock treatment guidelines have recently been updated recommending fluid boluses of between 40-60 mls/kg given over one hour in settings with access to intensive care treatments. Whereas in settings without access to intensive care fluid boluses are restricted to children with hypotension, providing 10-20 mls/kg of isotonic fluids over one hour[1]. The latter recommendation was based on the evidence provided from the multicentre Phase III randomized controlled trial of fluid resuscitation in severe febrile illness (Fluid Expansion As Supportive Therapy (FEAST)) which demonstrated a 45% relative increased risk of mortality with fluid bolus (with albumin or saline) compared to non-bolus controls (95% CI, 1.13 – 1.86; p= 0.003) [2,3]. Subsequent analyses indicated the major excess in mortality in the bolus arms were due to cardiogenic shock as the terminal clinical event rather than fluid overload[4]. Nevertheless, the 2013 updated Wold Health Organization (WHO) pediatric inpatient guidelines for resource-limited settings continue to recommend use of fluid boluses for children with all three signs of impaired perfusion (WHO-shock criteria): a prolonged capillary refilling time (CRT) >3 seconds, cool peripheries and a weak and fast pulse[5]. These criteria were recommended as a proxy for hypotension since blood pressure measurement is not widely available. We have previously demonstrated that the WHO-shock criteria are very rare (~ 0.1%) amongst general pediatric admission cohorts in resource-limited settings. Mortality in this group, however, was very high (80–100%)[6]. These findings question whether the 2013 WHO guidelines for fluid resuscitation are relevant to African children [7].

To date, there have been no clinical physiological studies investigating no-bolus fluid-conservative strategy and the current WHO shock fluid management guideline. We therefore conducted an observational study on non-malnourished children admitted with severe febrile illness and impaired perfusion (FEAST trial criteria [2,3]) managed conservatively with maintenance-only fluid and for children with WHO-shock fluid-boluses as recommended by the 2013 WHO guidelines (the

Management of Paediatric Septic Shock, MAPS study). We also aimed explored baseline clinical indices and biomarkers that could predict mortality.

Materials and Methods

Ethical approval to conduct the study was sought and obtained from the Kenya Medical Research Institute – Scientific and Ethics Review Unit (KEMRI-SERU) (2541: from 10-10-2013 to 09-10-2014) and the Mbale Regional Referral Hospital – Institutional Review Committee (MRRH-IRC) (protocol approval number REIRC 005/2013: from 13-03-2013 to 18-11-2019). When prior written consent from parents or legal guardians could not be obtained, ethics committees approved verbal assent with delayed written informed consent as soon as practicable[8].

Study design

A prospective observational study was conducted in two hospitals (Mbale Regional Referral Hospital, Uganda and Kilifi County Hospital, Kenya) in accordance with STROBE guidelines [9] between October 2013 and July 2015. Data were analysed from August 2016 to July 2019. As in the FEAST trial eligible patients included children aged ≥60 days to ≤12 years old with severe febrile illness (impaired consciousness and/or respiratory distress) and one or more signs of impaired perfusion (CRT > 3 seconds, temperature gradient or weak pulse). Children with severe acute malnutrition, gastroenteritis or non-infectious causes of shock or known heart disease were excluded.

Study procedures, fluid management and endpoints

Consecutive recruitment of the first 30 eligible and consenting children was done with no formal sample size calculation. Consent process followed that of the FEAST trial[8]. All participants had structured clinical assessments, blood/urine samplings, twelve-lead electrocardiograms [10-12] and echocardiograms, (Vivid.i General Electric Medical Systems® with simultaneous ECG display) recordings preformed at prespecified timepoints (baseline), post-fluid resuscitation (at 8 hours), at 24-hours and at 1-month follow-up. (See Additional File: methods). Echocardiographic readings were

reviewed independently by a cardiologist blinded to clinical status. Children with impaired perfusion received maintenance-only intravenous fluids (4mL/kg/hr) as per FEAST control arm followed by oral hydration. Children with WHO shock (≥3 criteria) received an initial two 20ml/kg boluses of saline given over one hour. Children with severe anemia (hemoglobin <5g/dL) or persisting signs of shock at 8 hours received blood transfusion (20mL/kg whole blood or 10mL/kg packed cells) over 3-4 hours as recommended by current WHO guidelines[5].

The endpoints included 48-hours mortality, correction (to normal ranges) of echocardiographic hemodynamic, myocardial performance indices and cardiac bio-markers measuring evidence of myocardial injury (Troponin)[13] and stretch (natriuretic peptides)[14] and adverse events related to the resuscitation.

Statistical analysis

All analysis was performed using STATA®, version 15 (StataCorp LLC©). Mean or mean difference (± standard deviation, SD) and median (inter-quartile range, IQR) were presented for normally and non-normally distributed data respectively. Overall normality of data distribution was assessed using the skewness and kurtosis test (sktest). Test for trend over time was performed on variables recorded at admission and repeated after fluid administration until 1-month follow-up. Baseline parameters in fatalities versus survivors to assess predictors of outcome and to compare number of existing criteria for shock ≥3 (WHO criteria) or ≤2 criteria using a two-sample *t*-test (normally-distributed data) and Wilcoxon rank sum test (continuous non-parametric data). Statistical significance was set at $p<0.05$ for all analyses. Some variables in the univariate analysis had multi-collinearity and Bonferroni's method was used to adjust for multiple comparisons in the multivariate analysis. However, in the final multivariable model, only variables that had shown significance in the univariate analysis were included, none of which were collinear.

Results

Recruitment and baseline data

Of the 54 children screened, 30 were eligible and followed-up for 1-month (**Supplemental Figure S1**)**.** Baseline demographic, anthropometric, clinical and laboratory data of all patients are summarized in **Table 1** comparing survivors versus non-survivors. Median age was 23 months (IQR, 17 – 35 months) and 70% (21/30) of the patients were males. Malaria was present in 27% (8/30), 10% (3/30) had HIV-infection, 40% (12/30) had severe anemia (Hb <5g/dL) and 20% (6/30) had WHO shock. Based on the 2005 age-specific Pediatric Sepsis Consensus Conference Criteria[15], tachycardia was present in 77% (23/30), low systolic blood pressure in 50% (15/30) and leucocytosis in 47% (14/30). **Table 2** compares baseline parameters of all patients with WHO shock versus non-WHO shock. At admission, children with WHO shock criteria were sicker than non-WHO shock, with a higher proportion being hypoxemic (<90%); 67% versus 4%, (χ^2=3.71; p= 0.0002), a higher median potassium (z=2.42, p=0.02), lactate (z=2.05, p=0.04) and troponin (z=3.67, p=0.0002). (Additional File **Table S1**, all fatalities; and **Table S2**, trends from admission to 28-days).

Volume administered during initial 24 hours

At admission, all patients received initial fluid management with intravenous maintenance-only fluid (4mL/kg/hr) except those with WHO shock who received fluid bolus therapy; 11 children with severe anemia (Hb<5g/dL) at presentation, and 7 children with persistent shock received blood transfusion (10mL/kg packed cells or 20mL/kg whole blood). Overall median intravenous volume given was 14 mL/kg/day (IQR, 10-35 mL/kg/day) since many were able to take oral fluids early in admission. The median volume of fluids administered to non-survivors was 30 mL/kg (IQR 28-61 mL/kg) which compared to median volume received by survivors 13 mL/kg (IQR 9-32 mL/kg), p=0.23. Mean red cell transfusion volume in 11 children with Hb<5g/dL was 9 mL/kg (IQR 7-11 mL/kg); 7/11 patients (64%) required ≥2 transfusions. Over 24 hours, the median volume of blood transfused in non-

survivors was 16 mL/kg (IQR 8-21 mL/kg) compared to medium volume received by survivors 8 mL/kg (IQR 6-12 mL/kg), p=0.24.

Outcome

By 48 hours there were 9/30 (30%) fatalities, all occurring <40-hours post-admission with the majority 7/9 (78%) occurring by 24-hours **(Additional file Figure S2** shows Kaplan-Meier survival at 48-hours and day 28). Six of the fatalities (67%), 5 or who died within 6 hours, had WHO-shock translating to case fatality of 6/6 versus 3/24 in children with <2 shock criteria (χ^2=7.27; p= 0.007). The clinical narratives of all fatalities are summarized in the Supplemental File **Table S3**.

Echocardiography and Electrocardiography

Baseline echocardiographic data are summarized in **Table 3** and **Figure 1** (and Additional File Figure S3) shows trends of selected echocardiographic parameters over the first 24-hours. At admission *Tei* index*, a* measure of global cardiac function, was reduced to ≤0.28 in 24 (80%) children. The *Tei index* improved to the normal range in 11 (52%) but remained low in 10 (48%) of survivors. The baseline echocardiographic parameters among fatalities comparing WHO shock to non-WHO shock (Additional File: **Table S4**). From baseline to 24 hours we observed in volume-sensitive haemoodynamic measures increases median (IQR) stroke volume index (mL/m^2 39 (32-42) to 47 (41-49) and cardiac output 3.0 ml/min (2.4-3.7) to 3.1 ml/min (2.8-3.6), and a measure of systolic function fractional shortening median (IQR) 30 (27-33) to 34 (31-38) (Additional file: **Table S5**) although these were not statistically different. In the 4 fatalities, where we had post-baseline echocardiography, fluid volume received was positively associated with increases in stroke volume index (from a median 36 of 36mls/m^2 [IQR 26, 47] to 41mls/m^2 [IQR 26, 48]) after receiving a median of 30 mL/kg [IQR 28, 61] suggesting fluid responsiveness. Myocardial contractility strain indices measured by speckle tracking in the radial, circumferential and longitudinal axes were within normal reference limits at admission. Amongst survivors, median global radial strain (GRS) and global

longitudinal strain (GLS) at admission were not significantly higher compared to non-survivors (GRS, z=2.88; p=0.05 and GLS, z=3.87; p=0.02). There were no differences in the median GRS and tricuspid annular plane systolic excursion (TAPSE) in patients with WHO-shock versus those with ≤2 shock criteria (z=2.42; p=0.05 and z= 3.14; p=0.02, respectively). Median systemic vascular resistance indices (SVRI) were similar in non-survivors and survivors (z=2.29; p=0.09). Baseline electrocardiographic data are summarized in Additional file **Table S6** comparing survivors versus non-survivors. There were no significant differences in the baseline electrocardiographic parameters at admission between survivors and fatalities or between those with WHO-shock versus ≤2 shock criteria (Additional file: **Table S7 and S8**).

Biomarkers profiles

Figure 2 (and Additional File Figure S4) shows cardiac biomarker trends over the first 24-hours. At admission 16/30 (53%) had a high cardiac troponin I level (cTnI). In survivors, only 2/23 (9%) had a raised baseline cTnI but fatal cases had elevated cTnI levels beyond the upper reference limit (≤0.1 μg/mL). Similarly, elevated levels of β-brain natriuretic peptide (BNP; >300 pg/mL [to convert to nanograms per litre multiply by 1]), α-atrial natriuretic peptide (ANP; >60 pg/mL [to convert to nanograms per litre multiply by 1]) and plasma hyaluronan (>100 ng/mL [to convert to nmol/l multiply by 2.5]) were present in 70%, 36% and 40% respectively at admission with fatalities having higher median levels of ANP and plasma hyaluronan than survivors (Additional file: **Table S9**). At admission, children with WHO-shock had higher median cardiac troponin I (cTnI) levels (*t*=3.58; 95%CI 1.24-1.43 ng/mL, p=0.02) and alpha-atrial natriuretic peptide (ANP) (*t*=16.5; 95%CI 2.80-67.5 pg/mL p<0.01) compared to children ≤2 shock criteria. Comparisons of biomarkers among the fatalities with WHO shock versus non-WHO shock (Additional file: **Table S10**) and summaries of biomarker trends over time (Additional file: **Table S11**).

Predictors of mortality

In the univariate analysis hyperkalemia, hyperlactatemia, hypoglycemia as well as a number of myocardial echocardiographic indices were associated with mortality. The odds ratio (OR) for mortality with an elevated troponin (>0.1 µg/mL) was 4 (95%CI 0.63-25.57), p=0.08 (Additional file: **Table S12**). In the multivariable analysis hyperlactatemia >4 mmol/L was the only variable that showed a significant association with death outcome (OR 5.38; 95%CI 4.49-12.71, p=0.003). WHO shock was not included as the composite scoring variable we created introduced instability in the model and was difficult to interpret due to the small sample size.

Discussion

The MAPS study evaluated feasibility of a fluid conservative strategy in African children with severe febrile illness and shock. Reassuringly, the administration of a maintenance-only fluid management strategy resulted in normalisation of the vital indices (heart rate, respiratory rate, CRT and oxygen saturations) without compromising cardiac function in the cohort of critically ill patients studied. Indeed, it led to a gradual yet significant increase in stroke volume index (SVI) and other volume-sensitive measures of myocardial function. Majority of the deaths 78% (7/9) occurred within the first 24-hours. All six children presenting with WHO-shock (≥3 criteria) and managed as per the current recommended fluid bolus guideline died. Of particular note is that this group had had a markedly higher mean level of cardiac troponin I and alpha-atrial natriuretic peptide compared to those with ≤2 shock criteria. Post-baseline measures were not reported in these children as they died before prespecified data/sampling timepoints. In a pre-clinical trial of ovine endotoxemic shock, fluid bolus resuscitation led to a rise in similar cardiac biomarkers (cTnI and ANP) compared to non-bolus vasopressor support. The rise in ANP was seen in the immediate post-fluid resuscitation period while that of cTnI was temporally distant peaking 12-hours after the fluid bolus [16].

The WHO-shock group of patients represented the sickest patients in this cohort 5/6 deaths occurred before the 8-hour timepoint for the clinical and physiological assessments. The clinical narratives of the fatalities with WHO shock indicated that 3 died during initial bolus therapy, 2 died awaiting an urgent transfusion and one child had a fatal cardiorespiratory arrest at 16-hours post admission following an initial satisfactory response to fluid boluses. In the FEAST trial only 65 children had WHO- shock (all having prolonged capillary refill time >3 seconds; cold extremities; weak and fast pulse). The 48-hour mortality rate in this subgroup was 48% (24/50) who received fluid boluses compared to 20% (3/15) deaths among the non-bolus controls [2,7]. Nevertheless, the WHO-guidelines which form the basis for most internationally used guidelines in low-and-middle income countries continue to recommend administration of fluid boluses for this group[17]. This was possibly influenced by a systematic reviews of fluid bolus therapy in resource-limited hospitals by Opiyo *et al* that considered those with WHO shock as a separate subgroup[18], concluding that the numbers included in the FEAST trial (n=65) were too small to provide reasonable certainty implying indirectness with respect to evidence. This disregarded the fact that the direction of harm was entirely consistent with the overall analysis thus 'indirectness' of overall effect should not have been inferred[19]. We have previously highlighted that in pediatric hospital admissions in resource-limited hospital, these comprise an exceptionally rare group of children with high fatality (80-100%)[6].

Given the results of the FEAST trial [2,3] and the recommendations of the systematic review by Ford *et al* [20], we are concerned that the current recommendations by WHO risk potential harm to a far wider group of children, since it attempts to implement a set of guidelines separating what it considers as 'true shock' i.e. the WHO shock definition (3 or more signs) from definitions that fewer features of shock which WHO guideline regards as 'impaired perfusion' are bound to lead to slippage[17]. Unlike in adult guidelines, where hypotension is a prerequisite for defining shock, hypotension in pediatric shock, as demonstrated by the FEAST trial, is a rare and serious event. In

the FEAST trial of the 7838 children with severe febrile illness screened for inclusion only 29 (0.9%) fulfilled the definition of severe hypotension (FEAST B definition); all received fluid-bolus therapy and had poor outcomes (62% mortality by 48 hours)[3,6]. Notably, the WHO-shock definition was not informed by relevant physiological evidence and based on expert opinion which was intended to identify children with advanced shock including those with hypotension. In a sub-analysis of the FEAST trial we demonstrated that only 8/29 (27.5%) hypotensive children actually fulfilled the WHO shock definition and all of those children died [6]. In this study, there was no hypotension noted on admission.

A major limitation of the study was the relative small sample size, nevertheless the study represents one of the only paediatric studies to examine myocardial and physiological linked to fluid management. The major strength of this study is that it was conducted in two centres and involved observation of multiple parameters of clinical and haemodynamic function over time. As such the MAPS study provides physiological data which reinforce the observations made in the pre-clinical (RESUS) trial [16] and the FEAST trial [3] that children with WHO-shock have extremely deranged myocardial function and biomarkers and on current recommended management this universally lead to a fatal outcome, including very elevated levels of cTnI, a myocardial regulatory protein with high specificity for cardiac myocyte damage (independent of volume status)[13]. These data therefore corroborate previous findings demonstrating elevated levels of cTnI in septic shock predict left ventricular dysfunction, poor prognosis and risk of mortality [21-24]. Furthermore, we found baseline levels of both ANP and BNP natriuretic peptides were more grossly elevated among patients with WHO-shock (≥3) criteria compared to those with ≤2 shock criteria. Natriuretic peptides classically cause vasodilation, diuresis, natriuresis and are produced in response to volume or pressure loading on the myocardial chambers, to counteract the excessive cardiac wall stress [25-28]. Secretion of ANP also causes increased shedding of the glycocalyx layer lining the vascular endothelia, thus leading to an increase in plasma levels of glycocalyx breakdown products [29]. Hyaluronan, a glycosaminoglycans

component of the glycocalyx-lining microvascular endothelia is a sensitive biomarker for vascular damage[30]. Our findings of elevated levels of hyaluronan in both plasma and urine, which increased slightly post-volume resuscitation also supports our original findings that the excess mortality of fluid boluses in the FEAST was mediated through cardiovascular collapse[4].

It could be suggested that echocardiography be used to identify global radial or longitudinal strain guide fluid resuscitation, however this approach remains technically challenging in young pediatric populations on crowded under-resourced emergency rooms. Having well trained pediatricians with requisite skills to conduct and interpret echocardiographic findings also presents a major hurdle. Alternative parameters such as hyperlactatemia >4 mmol/L which independently predicted outcome (OR 5.38; 95%CI 4.49-12.71, p= 0.003) and elevated troponin (>0.1 µg/mL) could be used in future research studies as potential entry criteria investigating additional strategies to manage cardiovascular compromise .

Box: *At the bedside*

- We provide further physiological and clinical data to support a no-bolus fluid strategy in 34 African children with severe illness and impaired perfusion. Whereas use of the WHO-shock guideline resulted in uniform mortality.
- We provide some evidence that bedside markers of cardiac injury and stretch (troponin and ANP) may be of prognostic value
- Future studies should focus on assessing conservative fluid strategies alongside inotropic support in order to improve outcome in pediatric septic shock

Conclusion

This study showed that an elevation in the biomarkers of cardiac and microvascular dysfunction at admission is associated with outcomes and a fluid-conservative resuscitation strategy using maintenance-only fluid improved clinical signs of shock without compromising cardiac function. We provide physiological evidence that children presenting with WHO-shock (≥3) have strong evidence

of existing myocardial damage which supports extending conservative fluid management to this group. One major challenge in the future is how to support circulatory collapse and myocardial impairment in resource-limited settings which have no access to high dependency care or ventilatory support. Future research to investigate early inotropic support alongside of judicious use of fluid therapy is warranted.

Acknowledgements

We acknowledge the clinical research teams at the KEMRI-Wellcome Trust Research Programme and Mbale Clinical Research Institute as well as the patients and their parents/guardians for participating in the study.

Nchafatso G. Obonyo, Peter Olupot-Olupot and Sophie Uyoga are funded and supported through the Wellcome Trust and the Department for International Development (DFID) funded DELTAS Africa Initiative [DEL-15-003].

Nchafatso G. Obonyo received funding from Imperial College London (Institutional Strategic Support Funds (105603/Z/14/Z), Wellcome Trust Imperial College Centre for Global Health Research 100693/Z/12/Z).

K Maitland received funding from Wellcome East African Overseas Programme Award from the Wellcome Trust 203077/Z/16/Z).

This research was funded in whole, or in part, by the Wellcome Trust [105603/Z/14/Z, 100693/Z/12/Z and 203077/Z/16/Z]. For the purpose of open access, the author has applied a CC BY public copyright licence to any Author Accepted Manuscript version arising from this submission.

The funding body did not play any role in the design of the study, data collection or analysis or manuscript preparation.

Authors' contributions

NGO, AM, KM – study conception and study design.

NGO, AM, JN, MC – study conduct and data collection.

NGO, SU, RM – sample analyses

NGO, KS – data analyses

NGO, POO, SU, RM, JPF, KS, JC, JFF, KM – review and interpretation of analysed data

NGO, KM – first draft of manuscript

All authors read and approved the final manuscript

Availability of data and materials

The datasets used and/or analysed during the current study are available from the corresponding author on reasonable request.

References

1. Cruz AT, Lane RD, Balamuth F, et al. Updates on pediatric sepsis. *J Am Coll Emerg Physicians Open* 2020; **1**(5): 981-93.
2. Moisi JC, Gatakaa H, Berkley JA, et al. Excess child mortality after discharge from hospital in Kilifi, Kenya: a retrospective cohort analysis. *Bulletin of the World Health Organization* 2011; **89**(10): 725-32, 32A.
3. Maitland K, Kiguli S, Opoka RO, et al. Mortality after fluid bolus in African children with severe infection. *N Engl J Med* 2011; **364**(26): 2483-95.
4. Maitland K, George EC, Evans JA, et al. Exploring mechanisms of excess mortality with early fluid resuscitation: insights from the FEAST trial. *BMC Med* 2013; **11**: 68.
5. In: nd, ed. Pocket Book of Hospital Care for Children: Guidelines for the Management of Common Childhood Illnesses. Geneva; 2013.
6. Houston KA, George EC, Maitland K. Implications for paediatric shock management in resource-limited settings: a perspective from the FEAST trial. *Crit Care* 2018; **22**(1): 119.
7. Kiguli S, Akech SO, Mtove G, et al. WHO guidelines on fluid resuscitation in children: missing the FEAST data. *Bmj* 2014; **348**: f7003.
8. Maitland K, Molyneux S, Boga M, Kiguli S, Lang T. Use of deferred consent for severely ill children in a multi-centre phase III trial. *Trials* 2011; **12**: 90.
9. von Elm E, Altman DG, Egger M, et al. The Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) statement: guidelines for reporting observational studies. *Ann Intern Med* 2007; **147**(8): 573-7.
10. Kligfield P, Gettes LS, Bailey JJ, et al. Recommendations for the standardization and interpretation of the electrocardiogram: part I: The electrocardiogram and its technology: a scientific statement from the American Heart Association Electrocardiography and Arrhythmias Committee, Council on Clinical Cardiology; the American College of Cardiology Foundation; and the Heart Rhythm Society: endorsed by the International Society for Computerized Electrocardiology. *Circulation* 2007; **115**(10): 1306-24.
11. Bazett HC. An analysis of the time-relations of electrocardiograms. *Heart-J Stud Circ* 1920; **7**(4): 353-70.
12. Tutar HE, Ocal B, Imamoglu A, Atalay S. Dispersion of QT and QTc interval in healthy children, and effects of sinus arrhythmia on QT dispersion. *Heart* 1998; **80**(1): 77-9.
13. Towbin JA, Gajarski RJ. Cardiac troponin I: a new diagnostic gold standard of cardiac injury in children? *J Pediatr* 1997; **130**(6): 853-5.
14. Levin ER, Gardner DG, Samson WK. Natriuretic peptides. *N Engl J Med* 1998; **339**(5): 321-8.
15. Goldstein B, Giroir B, Randolph A, International Consensus Conference on Pediatric S. International pediatric sepsis consensus conference: definitions for sepsis and organ dysfunction in pediatrics. *Pediatr Crit Care Med* 2005; **6**(1): 2-8.
16. Byrne L, Obonyo NG, Diab SD, et al. Unintended Consequences: Fluid Resuscitation Worsens Shock in an Ovine Model of Endotoxemia. *Am J Respir Crit Care Med* 2018; **198**(8): 1043-54.
17. Guideline:updates on paediatric emergency triage, assessment and treatment: care of critically-ill children. In: Organization WH, editor. Geneva: World Heatlh Organization; 2016.
18. Opiyo N, Molyneux E, Sinclair D, Garner P, English M. Immediate fluid management of children with severe febrile illness and signs of impaired circulation in low-income settings: a contextualised systematic review. *BMJ Open* 2014; **4**(4): e004934.
19. Guyatt GH, Oxman AD, Kunz R, et al. GRADE guidelines: 8. Rating the quality of evidence--indirectness. *Journal of clinical epidemiology* 2011; **64**(12): 1303-10.
20. Ford N, Hargreaves S, Shanks L. Mortality after fluid bolus in children with shock due to sepsis or severe infection: a systematic review and meta-analysis. *PloS one* 2012; **7**(8): e43953.
21. Mehta NJ, Khan IA, Gupta V, Jani K, Gowda RM, Smith PR. Cardiac troponin I predicts myocardial dysfunction and adverse outcome in septic shock. *Int J Cardiol* 2004; **95**(1): 13-7.
22. ver Elst KM, Spapen HD, Nguyen DN, Garbar C, Huyghens LP, Gorus FK. Cardiac troponins I and T are biological markers of left ventricular dysfunction in septic shock. *Clin Chem* 2000; **46**(5): 650-7.

23. Sheyin O, Davies O, Duan W, Perez X. The prognostic significance of troponin elevation in patients with sepsis: a meta-analysis. *Heart Lung* 2015; **44**(1): 75-81.
24. Soldin SJ, Murthy JN, Agarwalla PK, Ojeifo O, Chea J. Pediatric reference ranges for creatine kinase, CKMB, Troponin I, iron, and cortisol. *Clin Biochem* 1999; **32**(1): 77-80.
25. Maack T. The broad homeostatic role of natriuretic peptides. *Arq Bras Endocrinol Metabol* 2006; **50**(2): 198-207.
26. Price JF, Thomas AK, Grenier M, et al. B-type natriuretic peptide predicts adverse cardiovascular events in pediatric outpatients with chronic left ventricular systolic dysfunction. *Circulation* 2006; **114**(10): 1063-9.
27. Gangnus T, Burckhardt BB. Potential and Limitations of Atrial Natriuretic Peptide as Biomarker in Pediatric Heart Failure-A Comparative Review. *Front Pediatr* 2018; **6**: 420.
28. Kotby AA, Taman KH, Sedky HT, Habeeb NM, El-Hadidi ES, Yosseif HS. Atrial natriuretic peptide as a marker of heart failure in children with left ventricular volume overload. *J Paediatr Child Health* 2013; **49**(1): 43-7.
29. Chappell D, Bruegger D, Potzel J, et al. Hypervolemia increases release of atrial natriuretic peptide and shedding of the endothelial glycocalyx. *Crit Care* 2014; **18**(5): 538.
30. Cowman MK, Lee HG, Schwertfeger KL, McCarthy JB, Turley EA. The Content and Size of Hyaluronan in Biological Fluids and Tissues. *Front Immunol* 2015; **6**: 261.

Tables:

Table 1: Baseline characteristics for all patients; survivors and fatalities.

Test statistic: χ^2-test for categorical variables and Mann Whitney, U, for continuous variables
* p-value comparing medians of baseline characteristics of survivors versus non-survivors
** Definitions include:
Tachypnoea: >34 brpm if <12 months of age; >22 brpm if >1 to 5 yrs and >18 brpm if >5 yrs
ǂTachycardia: >180 bpm if <12 months of age; >160 bpm if 1 to 5 yrs of age; and >140 bpm >5 yrs
#Hypotension: Systolic blood pressure of <50 mmHg if <12 months; <60 mmHg if 1-5yrs; and <70 mmHg if > 5yrs
+Leucocytosis: White cell count: >17.5 X 10^9/L if <12 months; >15.5 X 10^9/L if 1-5 yrs and >13.5 X 10^9/L if > 5yrs
MUAC, mid-upper arm circumference; CRT, capillary refill time

Table 2: Baseline characteristic for WHO shock (≥3) and non-WHO shock (≤2) criteria.

Test statistic: χ^2-test for categorical variables and Mann Whitney, U, for continuous variables
* p-value comparing medians of baseline characteristics of patients with WHO shock *versus* non-WHO shock criteria
Tachypnoea: >34 brpm if <12 months of age; >22 brpm if >1 to 5yrs and >18 brpm if > 5yrs
ǂTachycardia: >180 bpm if <12 months of age; >160 bpm if 1 to 5yrs of age; and >140 bpm > 5yrs
#Hypotension: Systolic blood pressure of <50 mmHg if <12 months; <60 mmHg if 1-5 yrs; and <70 mmHg if > 5yrs
+Leukocytosis: White cell count: >17.5 X 10^9/L if <12 months; >15.5 X 10^9/L if 1-5 yrs and >13.5 X 10^9/L if > 5yrs

Table 3: Baseline echocardiography echocardiography variables at admission for all patients; survivors and fatalities

*Test-statistic: Mann Whitney *U*-test and p-value comparing baseline characteristics of survivors versus fatalities
[1]IVCCI, inferior vena cava collapsibility index; [2]TAPSE, tricuspid annular plane systolic excursion; [3]MAPSE, mitral annular plane systolic excursion

Figures:

Figure 1: Echocardiographic parameters at admission, post-resuscitation and at 24-hours. Box (displaying the data distribution through their upper and lower quartiles) and Whiskers (outliers) plots of ejection fraction, cardiac index, systemic vascular resistance index, and global radial, circumferential and longitudinal cardiac strain comparing survivors versus fatalities. The red lines indicate published pediatric reference values

Figure 2: Biomarker profiles at admission, post-resuscitation and at 24-hours.

Box and Whisker Plots of cardiac troponin I (cTnI), alpha-atrial natriuretic peptide (ANP), beta-brain natriuretic peptide (BNP) and hyaluronan comparing survivors versus fatalities. The red lines indicate published reference values.

Table 1: Baseline characteristics for all patients; survivors and fatalities.

% otherwise median (IQR)	All (n, 30)	Survivors (n, 21)	Fatalities (n, 9)	Statistic	p*
Sex (% males), n (%)	21 (70%)	14 (67%)	7 (78%)	-0.61	0.54
Age (months),	23 (17, 35)	23 (17, 37)	26 (18, 27)	0.01	0.99
Weight (kg),	10 (10, 12)	10 (10, 12)	10 (9, 11)	0.25	0.81
Height (cm),	83 (79, 86)	81 (79, 87)	85 (82, 85)	0.59	0.56
MUAC (cm),	15 (14, 16)	15 (14, 16)	14 (13, 15)	0.22	0.37
Clinical assessment					
Temperature (^{0}C),	37.9 (37.2, 38.4)	37.9 (37.2, 38.9)	37.6 (37.0, 38.1)	0.16	0.69
Fever (>39.0^{0}C), n (%)	5 (17)	5 (24)	0 (0)	1.61	0.11
Hypothermia (<36.0^{0}C), n (%)	2 (7)	1 (5)	1 (11)	-0.60	0.55
Respiratory rate (breaths/min),	53 (46, 60)	54 (46, 60)	50 (49, 58)	0.16	0.69
Tachypnoea**, n (%)	30 (100)	21 (100)	9 (100)	N/A	N/A
Oxygen saturation (%),	97 (91, 98)	97 (94, 98)	95 (83, 98)	0.72	0.40
Hypoxemia (<90%), n (%)	5 (17)	1 (5)	4 (44)	-2.64	0.008
Heart rate (beats/min),	166 (155, 172)	166 (163, 173)	160 (129, 169)	0.91	0.34
Tachycardia‡, n (%)	20 (67)	16 (76)	4 (44)	1.69	0.09
Systolic blood pressure (mmHg),	95 (83, 103)	93 (83, 102)	103 (83, 109)	1.43	0.23
Hypotension#, n (%)	0 (0)	0 (0)	0 (0)	N/A	N/A
Diastolic blood pressure (mmHg),	53 (51, 68)	53 (52, 68)	53 (50, 66)	0.06	0.81
Mean arterial pressure (mmHg),	69 (62, 79)	68 (62, 79)	70 (61, 78)	1.43	0.23
CRT>3 sec, n (%)	9 (30)	5 (24)	4 (44)	-1.13	0.26
WHO Shock, n (%)	6 (20)	-	6 (67)	-	-
Laboratory assessment					
Sodium (mmol/L),	133 (132, 136)	133 (132, 136)	133 (123, 136)	0.36	0.55
Potassium (mmol/L),	4.8 (4.2, 6.2)	4.7 (4.0, 6.1)	5.5 (4.8, 7.6)	4.89	0.03
Hyperkalemia (>5.5mmol/L), n (%)	11 (37)	7 (33)	4 (44)	-0.58	0.56
Creatinine (µmol/L),	53 (35, 67)	46 (22, 67)	63 (35, 77)	1.76	0.19
Lactate (mmol/L),	3.7 (2.0, 11.9)	3.5 (1.9, 11.4)	7.9 (6.4, 12.3)	4.89	0.03
Hyperlactatemia (>4mmol/L), n (%)	12 (40)	6 (29)	6 (67)	-1.95	0.05
Glucose (mmol/L),	8.5 (5.8, 9.7)	9.2 (6.6, 10.0)	5.2 (1.4, 6.2)	8.00	0.005
Hypoglycemia (<3mmol/L), n (%)	2 (7)	0 (0)	2 (22)	-2.24	0.03
Hemoglobin (g/dL),	5.4 (4.0, 9.6)	5.4 (3.0, 12.0)	5.9 (4.0, 9.0)	0.22	0.64
Severe anemia (<5g/dL), n (%)	12 (40)	9 (43)	3 (33)	0.49	0.63
White blood cell count X 10^9/L,	15.4 (10.9, 23.5)	13.1 (9.0, 19.5)	23.2 (15.6, 47.9)	4.89	0.03
Leucocytosis+ n (%)	16 (53)	10 (48)	6 (67)	-0.96	0.34
Platelets X 10^9/L,	162 (119, 238)	162 (118, 264)	142 (119, 223)	0.20	0.65
Malaria positive, n (%)	8 (27%)	5 (24%)	3 (33%)	-0.54	0.59
HIV positive, n (%)	3 (10%)	2 (9.5%)	1 (11%)	-0.13	0.89

*univariate analysis with no correction made for multiple comparisons.

Table 2: Baseline characteristic for WHO shock (≥3) and non-WHO shock (≤2) criteria

n % otherwise medians (IQR)	WHO shock (n, 6)	Non-WHO shock (n, 24)	Statistic	p*
Sex (% males), n (%)	5 (83)	16 (67)		
Age (months)	22 (14 – 27)	23 (17 – 38)	-0.60	0.55
MUAC (cm)	13 (13 – 15)	15 (14 – 16)	-2.17	0.03
Temperature (^{0}C),	37.8 (37.4, 38.1)	37.9 (37.1 – 38.9)	-0.57	0.57
Fever (>39.0^{0}C), n (%)	0 (0)	5 (21)	-1.23	0.22
Hypothermia (<36.0^{0}C), n (%)	1 (17)	1 (4)	1.15	0.25
Respiratory rate (breaths/min)	50 (49, 64)	54 (44, 60)	0.05	0.96
Oxygen saturation (%),	84 (57, 97)	98 (95, 98)	-2.10	0.04
Hypoxemia (<90%), n (%)	4 (67)	1 (4)	3.71	<0.001
Heart rate (beats/min),	152 (43, 181)	166 (160, 172)	-0.32	0.75
Tachycardia[ǂ], n (%)	3 (50)	17 (71)	-0.98	0.33
Systolic blood pressure (mmHg),	103 (82, 109)	93 (83, 103)	0.44	0.66
Hypotension[#], n (%)	0 (0)	0 (0)	N/A	N/A
Diastolic blood pressure (mmHg),	53 (50, 66)	57 (52, 68)	-0.55	0.58
Mean arterial pressure (mmHg),	70 (61, 80)	68 (62, 79)	-0.08	0.94
CRT>3 sec, n (%)	3 (50)	6 (25)	1.20	0.23
Sodium (mmol/L),	130 (123, 138)	133 (132, 136)	-0.36	0.72
Potassium (mmol/L),	**7.0 (5.9, 8.3)**	**4.7 (4.1, 6)**	2.42	0.02
Hyperkalemia (>5.5mmol/L), n (%)	4 (67)	7 (29)	1.73	0.08
Creatinine (µmol/L),	50 (23, 71)	53 (38, 67)	-0.39	0.70
Lactate (mmol/L),	**12.1 (9.9, 13.1)**	**3.6 (1.9, 10.8)**	2.05	0.04
Hyperlactatemia (>4mmol/L), n (%)	4 (67)	8 (33)	1.52	0.13
Glucose (mmol/L),	5.3 (1.3, 8.2)	8.9 (6.2, 9.7)	-1.67	0.09
Hypoglycemia (<3mmol/L), n (%)	1 (17)	1 (4)	1.15	0.25
Hemoglobin (g/dL),	4.2 (2.5, 7.6)	6.5 (4.0, 9.6)	-1.02	0.31
Severe anemia (<5g/dL), n (%)	2 (33)	10 (42)	-0.40	0.69
White blood cell count x 10^9/L	23.4 (20.2, 37.8)	14.0 (9.0, 19.5)	1.78	0.08
Leukocytosis[+] n (%)	4 (67)	12 (50)	0.75	0.46
Platelets x 10^9/L,	131 (80, 254)	167 (126, 238)	-0.78	0.43
Malaria positive, n (%)	2 (33)	6 (25)	0.40	0.69
HIV positive, n (%)	1 (17)	2 (8)	0.66	0.51
Biomarkers				
Troponin (ng/mL)	**0.67 (0.61 – 0.74)**	**0.09 (0.04 – 0.26)**	3.67	<0.001
BNP (pg/mL)	798 (563 – 1,033)	512 (328 – 1,004)	0.24	0.81
ANP (pg/mL)	111 (91 – 130)	67 (62 – 72)	1.08	0.28
Plasma hyaluronan (ng/mL)	168 (168 – 168)	75 (40 – 168)	1.78	0.07
Urine hyaluronan (ng/mL)	168 (168 – 168)	158 (129 – 168)	2.07	0.04

*univariate analysis with no correction made for multiple comparisons

Table 3: Baseline echocardiography variables at admission: all patients, survivors and fatalities

Medians (IQR)	All (n, 30)	Survivors (n, 21)	Fatalities (n, 9)	U*	p-value
Tei index	0.15 (0.10-0.23)	0.15 (0.11-0.21)	0.12 (0.10-0.36)	1.67	0.11
Ejection Fraction (%)	60 (53 – 64)	61 (55-66)	59 (47-63)	0.86	0.40
Fractional Shortening (%),	30 (27 – 33)	32 (28-35)	30 (23-32)	1.50	0.19
Stroke Volume (SV) (mls),	19 (16 – 24)	19 (17-23)	19 (14-24)	0.36	0.97
SV Index (mls/m^2)	39 (31 – 46)	39 (35-41)	36 (26-47)	0.40	0.76
End Diastolic Volume (EDV) (mls),	31 (26 – 42)	31 (27-37)	33 (22-45)	0.39	0.76
EDV Index (mls/m^2)	65 (52 – 74)	63 (55-69)	68 (41-93)	1.09	0.63
Systemic vascular resistance (SVR) (dynes.sec/cm^5)	433 (394 – 578)	416 (381-520)	586 (430-791)	2.36	0.08
SVR Index (dynes.sec/cm^5/m^2)	893 (756- 1,271)	865 (720-1,018)	1,231 (811-2,077)	2.29	0.09
Cardiac Output (mls/min)	3.0 (2.4 – 3.7)	3.0 (2.5-3.6)	2.9 (1.8-4.1)	0.25	0.96
Cardiac Index (mls/min/m^2)	6.0 (4.9 – 7.2)	6.0 (5.4-7.1)	5.4 (4.9-8.4)	0.27	0.96
IVCCI[1] (%)	28 (18 – 40)	27 (18-39)	35 (21-40)	0.44	0.67
TAPSE[2] (mm)	16 (14 – 18)	16 (15-18)	14 (12-15)	2.53	0.06
MAPSE[3] (mm)	11 (10 – 12)	11 (10-12)	10 (8-10)	1.97	0.06
Mitral inflow (E')	0.15 (0.12-0.18)	0.16 (0.13-0.18)	0.11 (0.09-0.15)	2.46	0.06
Early diastolic mitral inflow (MDF)	7.95 (5.86-9.36)	8.11 (6.10-9.14)	7.91 (5.67-9.36)	0.25	0.96
Early to late MDF ratio (E/A)	1.24 (1.11-1.63)	1.22 (1.14-1.35)	1.63 (1.04-1.69)	0.95	0.73
Systolic left ventric. peak velocity	0.08 (0.06 -0.09)	0.08 (0.07-0.09)	0.07 (0.05-0.07)	2.34	0.08
Global radial strain (%)	26 (22 – 32)	41 (29-50)	23 (23-40)	2.88	**0.05**
Global circumferential strain (%)	-21 [(-26) - (18)]	-19 [(-21)- (-16)]	-21 [(-24) - (-13)]	1.57	0.06
Global longitudinal strain (%)	-24 [(-27)-(-21)]	-22 [(-24)-(-19)]	-13 [(-17) - (-13)]	3.87	**0.02**

Figure 1: Echocardiographic parameters at admission, post-resuscitation and at 24-hours

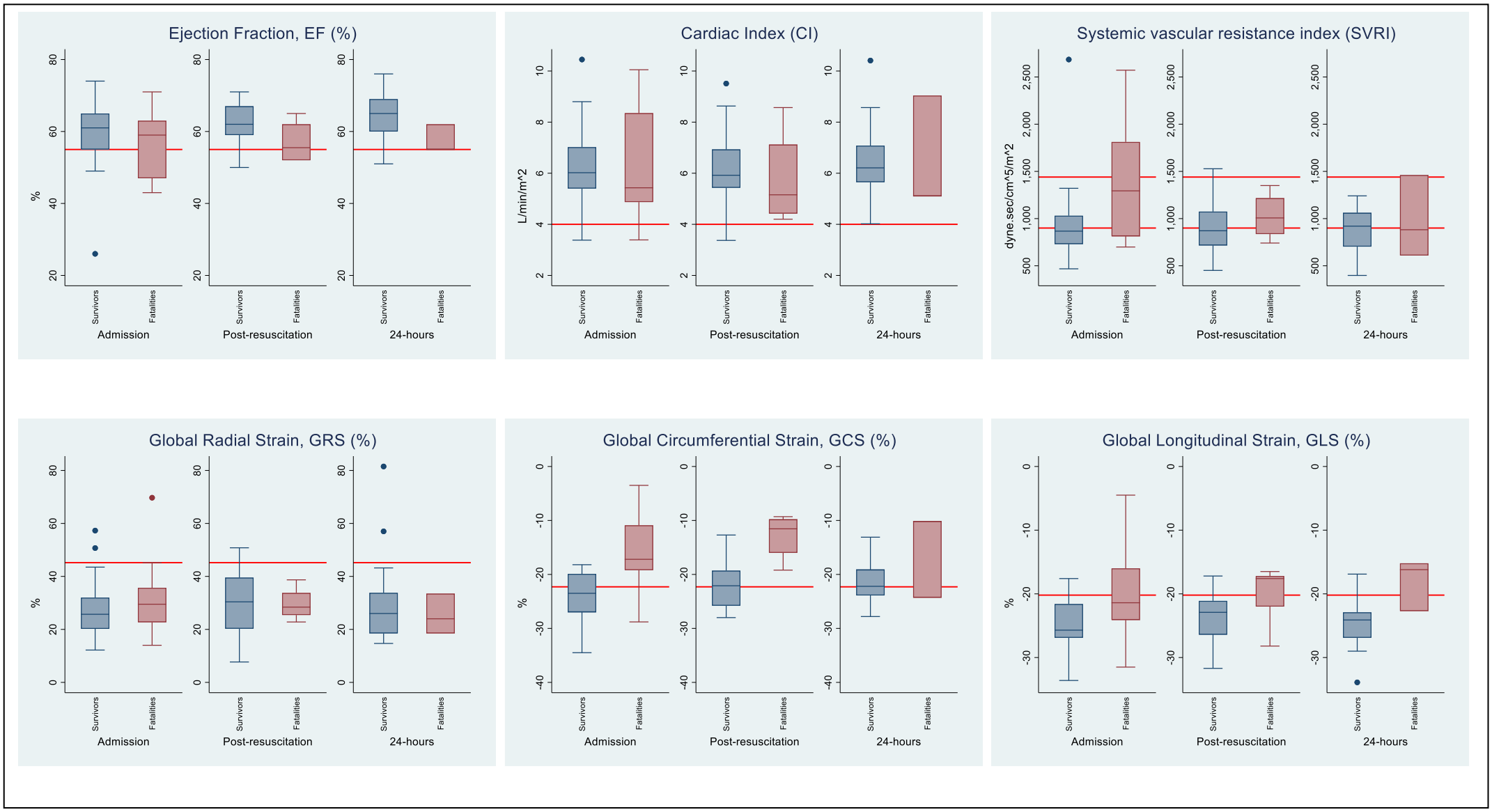

Figure 2: Biomarker profiles at admission, post-resuscitation and at 24-hours