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Loss of Hemoglobin Turnover Complexity Predicts Mortality in Septic Shock with Fluid Overload

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Abstract

Background:

The optimal red blood cell transfusion strategy in the context of fluid resuscitation remains controversial. This study aims to investigate the differential dynamics of the hemoglobin turnover rate (HTR) and inflation index between survivors and non-survivors with septic shock complicated by fluid overload, based on a novel circulatory model.

Methods

This study enrolled patients with septic shock diagnosed within 48 hours and collected their sequential fluid data from the first 7 ICU days. Using a novel circulatory model, HTR and inflation index served as markers of erythrocyte metabolism and systemic volume status. Nonlinear trajectories and interactions between HTR and inflation index were modeled with generalized additive models (GAMs), with longitudinal comparisons adjusted via Bonferroni correction. Validation included subgroup analysis, propensity score matching, and external validation with MIMIC-IV. Supplementary analyses assessed inflation index in transfusion-naïve periods and HTR–inflation index dynamics in key cohorts.

Results

Following the ROSE framework, key physiological markers—including HTR, inflation index, fluid balance, and lactate—differed significantly by treatment stage between survivors and non-survivors (all $p < 0.001$). GAM analysis revealed divergent dynamic patterns: survivors exhibited nonlinear HTR trajectories, whereas non-survivors showed linear HTR progression but stronger nonlinear coupling between HTR and the inflation index. These results indicate that non-survivors, despite simpler HTR trends over time, displayed more complex HTR–volume interdependencies. The

findings were robust to sensitivity analyses using propensity score matching and external validation. Additional analyses further showed that clinical factors such as albumin and SOFA score were associated with distinct inflation index trajectories, reinforcing the differential HTR–inflation index relationship between survivors and non-survivors.

Conclusion:

Based on a novel macrocirculation-first model, this study links the HTR to erythrocyte metabolic demand and the inflation index to circulatory volume status. Dynamic decoupling/loss of complexity in HTR trajectories combined with rigid coupling to inflation index was strongly associated with adverse outcomes.

Keywords:

Sepsis; Septic shock; Fluid balance; Red blood cell transfusion; Fluid Therapies; Intensive care unit

1. Introduction

Septic shock, characterized by infection-induced hypotension and tissue hypoperfusion, continues to pose a major challenge in critical care despite therapeutic advances[1]. Mortality remains unacceptably high[2-4], with approximately six million deaths annually from severe sepsis in high-income countries[2]. In China, mortality rates for severe sepsis and septic shock exceed 50% [5,6]. As emphasized by the Surviving Sepsis Campaign[7], effective management hinges on hemodynamic resuscitation[8], infection control, and organ support.

The role of red blood cell transfusion (RBCT) in sepsis management remains debated, particularly regarding the optimal hemoglobin threshold. The landmark Transfusion Requirements in Critical Care (TRICC) trial compared a restrictive strategy (transfusion trigger Hb <7 g/dL, target

7–9 g/dL) with a liberal strategy (trigger 9 g/dL, target 10–12 g/dL), finding no significant difference in 30-day mortality, although a restrictive approach appeared beneficial in younger, less severely ill patients[9]. These findings were later reinforced by a multicenter trial in septic shock patients, which reported no mortality or ischemic event differences between 7 g/dL and 9 g/dL thresholds[10], thereby informing current guideline recommendations[11,12].

However, evidence remains inconsistent. Some studies, including one in septic shock patients with cancer, suggest potential survival benefits with a liberal transfusion strategy[13], while others reaffirm the safety of lower thresholds[10].

Traditional hemodynamic models offer limited insight into the mechanisms underlying transfusion effects in septic shock, especially in the context of concurrent fluid resuscitation. To address this gap, we propose a novel circulatory structural model and examine real-world variability in the hemoglobin turnover rate (HTR) and inflation index dynamics, offering a new framework for understanding transfusion physiology in critical illness.

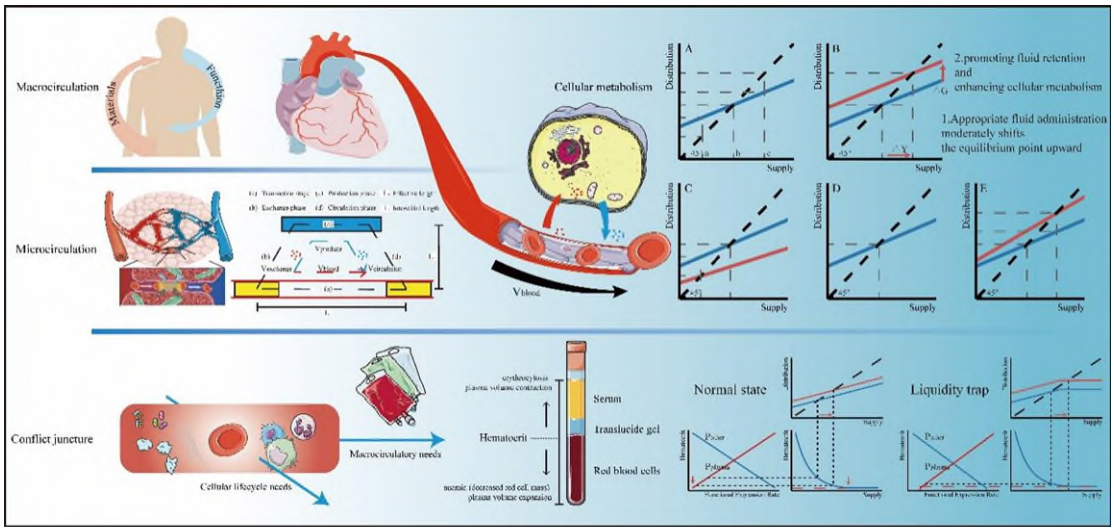


Figure 1. Conceptual framework of the multistage circulatory model and proposed “liquidity-trap” physiology. This model integrates macrocirculation, microcirculation, and cellular metabolism. Oxygen-transport units, as functional entities, interconnect these levels. Body fluid is partitioned into intravascular (circulating) and extravascular (reservoir) compartments. Systemic equilibrium is established when total fluid input equals total distribution volume at point A-b. If input is less than distribution volume (A-a), the reservoir replenishes the circulation, decreasing reserve (C) and indicating fluid need. If input

exceeds distribution volume (A-c), fluid moves to the reservoir, increasing its volume (E). An input increase (ΔY) normally shifts the distribution curve upward by ΔG (where $\Delta Y > \Delta G$) to a new equilibrium (B). Intravenous therapy acts on the circulation, altering oxygen-transport unit composition. Normally, fluid increase dilutes other components, elevating demand to restore balance. In a "liquidity trap," carrier fluid peaks, excess shifts extravascular, and cellular exchange is impaired despite rising demand. Therapy onset shifts P_{plasma} down-left and P_{other} down-right from equilibrium. System compensation drives P_{other} up-left, forming a new equilibrium within the triangular zone. P_{plasma}, functional expression per unit fluid; P_{other}, functional expression of other components. The blue line indicates reference equilibrium; red shows actual change.

2. Hypothesis Development

Building upon our previously proposed circulatory model, we introduce the blood-oxygen unit (BOU) – a conceptualized functional segment of the microcirculation bounded by adjacent erythrocyte centers, comprising one red blood cell and its accompanying plasma volume. This unit forms the basis of the perfusion-metabolism coupling equation (Figure 1; Appendix 1):

$$M \times V = P \times Y$$

where M is the mean number of BOUs exchanged in capillaries, V is mean capillary flow velocity, Y denotes functional output per unit, and P represents the functional expression rate. The erythrocyte-to-plasma ratio within each BOU critically influences microcirculatory efficiency. As direct clinical measurement of microcirculatory and cellular metabolic parameters remains difficult, this study employs macrocirculatory surrogates for validation, with hematocrit (HCT) serving as a key proxy – decreasing during fluid resuscitation and rising after RBCT.

2.1. Intravenous Fluid Therapy and the Liquidity Trap

The modern practice of intravenous fluid therapy traces its origins to the treatment of cholera in the 19th century[14-16]. Rudolph Matas, a pioneer of continuous infusion, also cautioned against the risks of normal saline[17]. Subsequent studies confirmed that 0.9% sodium chloride promotes fluid retention, edema, hemodilution (reduced HCT and albumin)[18], and hyperchloremic metabolic

acidosis[19,20], prompting the shift toward more physiological balanced crystalloids.

Assuming constant physiological activity, body fluid distribution can be categorized into intravascular circulating components (plasma) and extravascular storage components (intracellular fluid, interstitial fluid, etc.). The balance between plasma and interstitial fluid primarily depends on hydrostatic and colloid osmotic pressures. Administered fluids initially enter vascular circulation before redistributing throughout tissues. Under steady-state conditions, input and distribution volumes maintain equilibrium. Insufficient fluid administration triggers mobilization of stored fluids to sustain circulation, while excessive infusion leads to fluid accumulation in storage compartments (Figure 1).

Rapid infusion of crystalloid alters the HCT and dilutes functional components within BOUs, creating a mismatch between cellular metabolic rate and infusion kinetics. This reduces functional output (*Y*) while increasing the functional expression rate (*P*). When vascular integrity and osmotic gradients are compromised, the intravascular compartment reaches its expansion limit, trapping further infused fluid in the extravascular space – a state we term the "liquidity trap", characterized by persistent low HCT despite ongoing fluid administration (Figure 1).

2.2. Red Blood Cell Transfusion

Within our model, erythrocytes occupy a central role, governed by both functional demand and cell-cycle constraints. Hemoglobin is the primary effector molecule for gas transport[21]. Post-fluid resuscitation, blood component dilution reduces *Y* values while elevating *P* values, indicating increased metabolic demand.

Erythropoiesis from hematopoietic precursors requires 3–5 days[22,23], accelerated under stress

[24,25]; reticulocyte maturation adds 1–2 days[26,27]. With limited capacity for acute adaptive increase in production, the body initially relies on mobilizing reserved erythrocytes. Under physiologic conditions, about 1% of circulating red cells are cleared daily[28,29]; this turnover is markedly increased in critical illness[30,31].

Allogeneic RBCT thus serves as a vital intervention, rapidly bridging the temporal gap in endogenous erythropoiesis and sustaining oxygen-carrying capacity during the high-demand state of septic shock.

3. Study Design and Methods

3.1. Data Sources

This study utilized the Hospital Information System (HIS) dataset from Zhejiang Provincial People's Hospital (2018–2025) for model development and internal validation, and the MIMIC-IV database (2008–2022) for external validation.

The study adhered to the STROBE[32] guidelines and the Declaration of Helsinki. All data were de-identified to protect patient confidentiality. Access to the MIMIC-IV database was granted to certified researcher Huan Jiang (Record ID: 66212733) by Beth Israel Deaconess Medical Center. The Ethics Committee of Zhejiang Provincial People's Hospital waived the requirement for informed consent.

3.2. Study design

This study enrolled adult patients who developed fluid overload (FO) following intravenous fluid resuscitation for severe sepsis or septic shock. Only the first eligible admission was included

for patients with multiple hospitalizations. We excluded patients with insufficient data (<2 records) during admission or within the first 7 days of ICU stay, as well as those undergoing surgical procedures (Appendix2.1; sFigure 1; sTables 1-2).

We collected demographic characteristics (age, sex, height, weight, BMI), SOFA scores, and comorbidities (CCI score, hypertension, diabetes, liver disease, peripheral vascular disease, shock, coronary artery disease). The time of first intravenous therapy in the ICU served as time zero, with each HCT measurement marking corresponding timepoints; other data were carried forward from previous values. Continuous monitoring included MCV, MCHC, HCT, RBC, WBC, Hb, total bilirubin, albumin and lactate until transfer/discharge. Fluid intake and output were categorized by type, and comorbidities were defined using ICD-10 codes (Appendix2.3; sFigure 2).

The primary endpoint was all-cause mortality, defined as survival status from ICU admission until discharge, with specific focus on 7-day mortality.

3.3. Statistical Analysis

This study examines the dynamic balance between macro-circulatory demands and erythrocyte life-cycle requirements, using inflation rate [inflation rate = 1/HCT − 1] and HTR [HTR = total bilirubin/ Hb] as respective biomarkers (Appendix1.5). Patients were stratified according to fluid therapy phases and survival outcomes to assess dynamic physiological responses across different circulatory states.

Aligned with the sepsis ROSE resuscitation strategy[33], Phase I covered Resuscitation and Optimization stages, while Phase II included Stabilization and Evacuation. Critical timepoints comprised initial Phase I measurement, fluid overload occurrence, and Phase II completion. We evaluated fluid balance, HTR, inflation index, per-unit white blood cell count (Y_{WBC}), per-unit

lactate (Y_{lac}), and MCHC, applying Bonferroni correction for multiple comparisons. Longitudinal validation incorporated each patient's initial and final measurements within a 7-day observation period. Generalized additive models (GAMs)[34] delineated nonlinear trajectories of HTR and inflation rate throughout treatment phases, revealing their interactive patterns during fluid loading.

To strengthen the robustness of the study, stratified analyses were conducted across key clinical subgroups, including BMI, baseline albumin, Hb and WBC parameters. Subsequently, all primary analyses were repeated using propensity score matching to balance baseline characteristics (Appendix2.4). To further evaluate the generalizability of our findings, external validation was performed using the MIMIC-IV database.

Supplementary analyses targeted distinct clinical contexts: (1) subgroup-specific trajectories of inflation rate during intravenous therapy in patients without transfusion exposure, and (2) selected patient cohorts analyzed to characterize HTR–inflation rate dynamics throughout intravenous treatment (Figure 2).

Continuous variables are summarized as mean±SD or median (IQR), and categorical variables as frequencies (%). Statistical analyses included t-tests, Wilcoxon rank-sum tests, or chi-square tests as appropriate, all conducted using R 4.1.0.

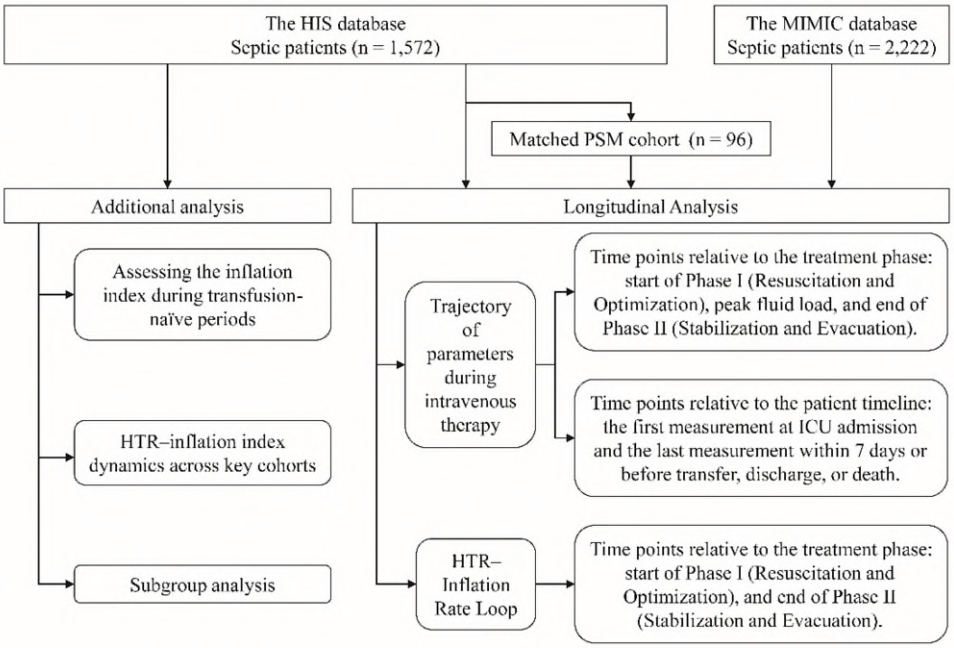


Figure 2. Study cohort selection and dataset partitioning flow chart. This study utilized data from two independent multicenter cohorts: an institutional hospital information system (HIS) database and the publicly available MIMIC-IV database. The HIS dataset served as the primary cohort for hypothesis testing and model development. To evaluate the robustness of the findings, two distinct subsets were derived from the HIS data: a propensity-score matched (PSM) cohort and a sub-cohort comprising only transfusion-naïve periods. The MIMIC database served as an external cohort to assess the generalizability of the model across different institutions and patient populations.

4. RESULTS

4.1. Study Population

Among 1,572 ICU patients, non-survivors (n = 153) were significantly older than survivors (n = 1,419) and had lower body weight and BMI. Non-survivors also exhibited higher illness severity, reflected by elevated SOFA scores, and greater comorbidity burden, indicated by higher CCI scores. Notably, chronic pulmonary disease and cancer were more prevalent among non-survivors. Laboratory assessment revealed significantly higher lactate levels and Y_{Lac} in non-survivors, whereas most other hematologic and biochemical parameters did not differ between groups (Table 1).

Table 1. Baseline characteristics of patients in the HIS dataset.

	Survival N = 1419	Non- survival N = 153	*p*
Baseline Characteristics			
Gender, Male (%)	927 (65.33)	93 (60.78)	0.3033
Age, year	66.07 (15.08)	71.27 (14.79)	0.0001
Height, cm	164.53 (8.26)	164.12 (8.51)	0.554
Weight, kg	61.68 (13.44)	57.54 (11.20)	0.0002
BMI, kg/m2	22.67 (4.04)	21.25 (3.12)	<0.0001
SOFA score (mean (SD))	5.95 (2.75)	7.25 (3.16)	<0.0001
Comorbidities†			
CCI score (mean (SD))	3.45 (1.90)	4.02 (1.89)	0.0004
Severe liver disease (%)	59 (4.16)	11 (7.19)	0.1283
Chronic pulmonary disease (%)	59 (4.16)	13 (8.50)	0.0254
Congestive heart failure (%)	605 (42.64)	59 (38.56)	0.3772
Diabetes (%)	408 (28.75)	37 (24.18)	0.2724
Renal disease (%)	198 (13.95)	31 (20.26)	0.0476
Cancer (%)	433 (30.51)	61 (39.87)	0.0228
Biochemical parameters			
HCT (median [IQR])	0.31 [0.27, 0.36]	0.30 [0.25, 0.36]	0.3364
Net intake, ml (median [IQR])	246.71 [16.92, 686.71]	150.29 [1.09, 744.97]	0.3231
Total intake, ml (median [IQR])	553.88 [161.69, 2099.70]	604.63 [124.15, 4511.03]	0.5943
Hb, g/dL (median [IQR])	1.11 [0.70, 1.71]	1.05 [0.68, 2.05]	0.5457
RBC, $\times 10^{12}$/L (median [IQR])	3.35 [2.85, 3.88]	3.29 [2.62, 3.89]	0.2389
WBC, $\times 10^{9}$/L (median [IQR])	10.20 [8.60, 12.10]	9.80 [8.30, 12.40]	0.4457
Total bilirubin, mg/dL (median [IQR])	30.40 [27.10, 33.60]	29.70 [26.10, 32.78]	0.1236
Albumin, g/L (median [IQR])	90.80 [87.40, 94.20]	91.75 [87.47, 94.40]	0.2874
MCV, fl (median [IQR])	11.22 [8.03, 15.63]	12.23 [7.29, 18.42]	0.3013
Lactate, mmol/L (median [IQR])	2.00 [1.30, 3.10]	2.70 [1.80, 4.78]	<0.0001
MCHC, g/dL (median [IQR])	32.88 [32.14, 34.19]	32.68 [32.17, 34.04]	0.594
Y_{wbc} (median [IQR])	3.29 [2.48, 4.59]	3.85 [2.24, 5.89]	0.0739
Y_{lac} (median [IQR])	0.60 [0.39, 0.94]	0.87 [0.53, 1.51]	<0.0001
HTR (median [IQR])	0.11 [0.07, 0.18]	0.13 [0.07, 0.23]	0.1122
Inflation rate (median [IQR])	2.24 [1.75, 2.76]	2.34 [1.75, 2.94]	0.3364

BMI, Body Mass Index; CCI, Charlson Comorbidity Index; SOFA, Sequential Organ Failure Assessment; Hb, Hemoglobin; RBC, Red blood cell count; WBC, White blood cell count; MCV, Mean corpuscular volume; MCHC, Mean corpuscular hemoglobin concentration; Lac, Lactate.

† Defined using International Classification of Diseases, 10th revision codes

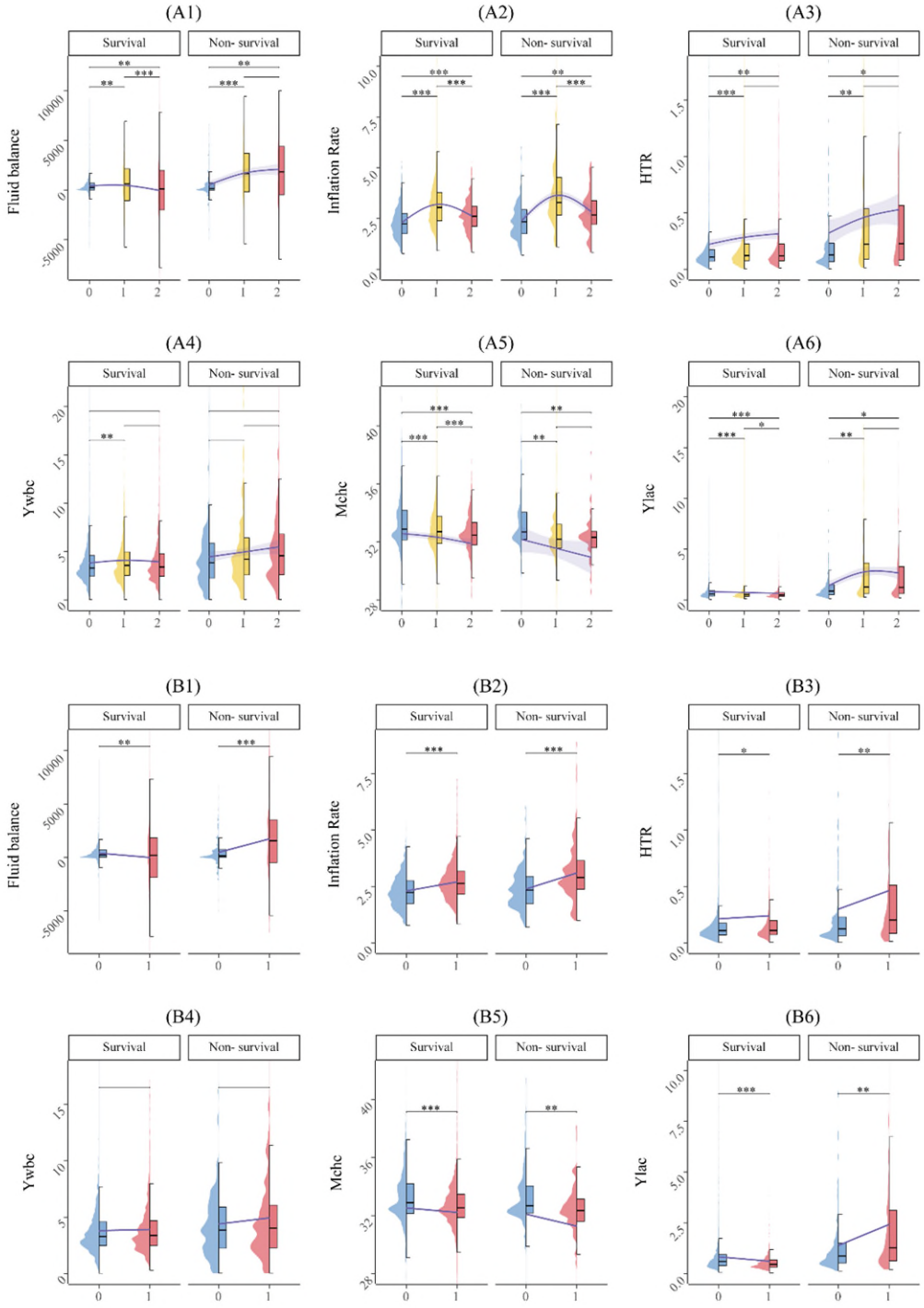


Figure 3. Distribution of Hemodynamic and Metabolic Variables Across Treatment Stages by Survival Status. (A) Time points relative to treatment phase: 0, start of Phase I (Resuscitation and Optimization); 1, peak fluid load; 2, end of Phase II (Stabilization and Evacuation). (B) Time points relative to patient timeline: 0, first measurement at ICU admission; 1, last measurement within 7 days or before transfer/discharge/death. Significance levels: *P < 0.05; **P < 0.001; ***P < 0.0001.

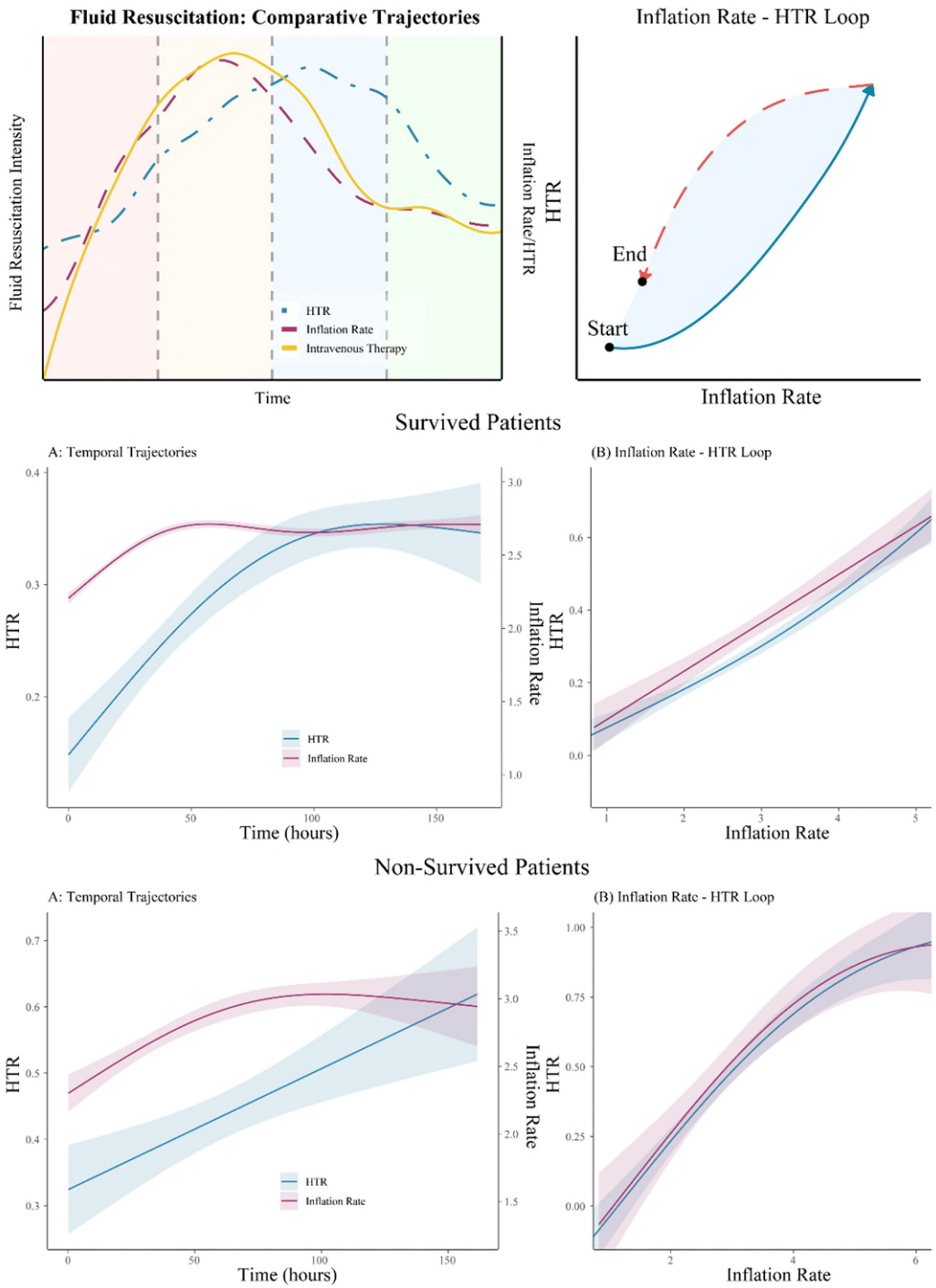


Figure 4. Dynamic Changes in HTR, Inflation Rate, and HTR – Inflation Rate Coupling During Intravenous Therapy. Within the ROSE framework of intravenous therapy, the inflation rate typically peaks first, followed by a subsequent decline in HTR and other biochemical markers as macrocirculatory parameters improve. The coupling between HTR and inflation rate exhibits a largely synchronous trajectory across these phases. In the HTR–inflation rate coupling plot, the blue line represents Phase I (Resuscitation and Optimization), and the red line represents Phase II (Stabilization and Evacuation)

4.2. Main Analyses

Following the ROSE framework, inflation rate demonstrated significant stage-dependent

variations (all $p < 0.001$), with HTR showing a progressive increase in non-survivors (0.13/0.22/0.23; stage 0 vs 1: $p < 0.001$; stage 0 vs 2: $p < 0.05$) but remaining relatively stable in survivors (0.11/0.13/0.12; stage 0 vs 1 and stage 0 vs 2: $p < 0.01$). Fluid balance exhibited markedly different patterns, with non-survivors accumulating substantially higher volumes (150.29/1665.24/1836.93 mL) compared to survivors (246.71/625.66/115.47 mL; all $p < 0.001$). Y_{Lac} levels were persistently elevated in non-survivors (0.87/1.29/1.23) but decreased in survivors (0.60/0.53/0.49; all $p < 0.001$), while Y_{wbc} showed a transient increase in survivors (3.30/3.61/3.42; stage 0 vs 1: $p < 0.05$) versus a steady rise in non-survivors (3.85/4.21/4.58; stage 0 vs 1: $p < 0.05$). Mchc declined gradually in both groups (Table 2; Figure 3). Analysis restricted to first/last timepoints revealed consistent trends (Table 3).

Table 2. Distribution of Hemodynamic and Metabolic Variables Across Treatment Stages by Survival Status

Group	Variable	N (stage0/stage1/stage2)	Value (stage0/stage1/stage2)	*P* (0vs1)	*P* (1vs2)	*P* (0vs2)
Survival	HTR	1233 / 1418 / 943	0.11 / 0.12 / 0.12	<0.0001	1	0.001
Survival	Inflation rate	1419 / 1419 / 945	2.24 / 3.05 / 2.61	<0.0001	<0.0001	<0.0001
Survival	MCHC	1419 / 1419 / 945	32.88 / 32.70 / 32.46	<0.0001	<0.0001	<0.0001
Survival	Fluid balance	1419 / 1419 / 945	246.71 / 625.66 / 115.47	0.003	<0.0001	0.002
Survival	Y_{lac}	1374 / 1419 / 945	0.60 / 0.52 / 0.49	<0.0001	0.02	<0.0001
Survival	Y_{wbc}	1417 / 1419 / 945	3.29 / 3.57 / 3.40	0.003	0.369	0.444
Non-survival	HTR	125 / 153 / 78	0.13 / 0.22 / 0.23	0.0004	1	0.027
Non-survival	Inflation rate	153 / 153 / 78	2.34 / 3.29 / 2.68	<0.0001	0.0001	0.002
Non-survival	MCHC	153 / 153 / 78	32.68 / 32.19 / 32.31	0.0007	1	0.0009
Non-survival	Fluid balance	153 / 153 / 78	150.29 / 1665.24 / 1836.93	<0.0001	1	0.0004
Non-survival	Y_{lac}	147 / 153 / 78	0.87 / 1.29 / 1.23	0.0004	1	0.014
Non-survival	Y_{wbc}	152 / 153 / 78	3.85 / 4.21 / 4.58	0.603	1	0.202

Stages 0, 1, and 2 correspond to the initial Phase I measurement, fluid overload occurrence, and Phase II completion.

† P values were Bonferroni-adjusted for multiple comparisons; values smaller than 0.0001 are reported as <0.0001.

Table 3. Distribution of Hemodynamic and Metabolic Parameters Across the First and Last points by Survival Status

Group	Variable	N (First / Last)	Value (First / Last)	*P* (First / Last)
Survival	HTR	1233 / 1302	0.11 / 0.11	0.012
Survival	Inflation rate	1419 / 1304	2.24 / 2.64	<0.0001

Survival	MCHC	1419 / 1304	32.88 / 32.54	<0.0001
Survival	Fluid balance	1419 / 1304	246.71 / 186.89	0.006
Survival	Y_{lac}	1374 / 1304	0.60 / 0.46	<0.0001
Survival	Y_{wbc}	1417 / 1304	3.29 / 3.39	0.1
Non-survival	HTR	125 / 137	0.13 / 0.20	0.004
Non-survival	Inflation rate	153 / 137	2.34 / 2.91	<0.0001
Non-survival	MCHC	153 / 137	32.68 / 32.35	0.0007
Non-survival	Fluid balance	153 / 137	150.29 / 1561.47	<0.0001
Non-survival	Y_{lac}	147 / 137	0.87 / 1.26	0.0007
Non-survival	Y_{wbc}	152 / 137	3.85 / 4.04	0.532

GAM analysis revealed fundamentally distinct physiological dynamics between survivors and non-survivors. Non-survivors exhibited predominantly linear trajectories in HTR over time (edf $\approx$ 1.0), whereas survivors showed significantly nonlinear patterns (edf 1.94–2.51). Although both groups demonstrated nonlinear inflation index dynamics, survivors displayed greater model complexity (edf 1.99–3.94) compared to non-survivors (edf 1.93–2.33). Notably, non-survivors exhibited stronger nonlinear coupling between HTR and the inflation index during both resuscitation-optimization and stabilization-evacuation phases, accompanied by better model fit (higher F-statistics, all $p < 2 \times 10^{-16}$). Increasing model flexibility enhanced nonlinearity more prominently in non-survivors. These findings indicate that although HTR progressed more simply over time in non-survivors, its interdependence with volume status was more rigid and complex, suggesting a maladaptive shift toward a volume-dependent physiological state (sTables 3-4; Figure 4).

4.3. Sensitivity Analyses

After defining the baseline cohort through PSM, the analyses were repeated, yielding comparable results (sTables 5-8; sFigure 3). External validation in the MIMIC database revealed some differences—most notably, a more concentrated distribution of the inflation rate compared with the HIS database—yet the primary conclusions remained robust (sTables 9-12; sFigure 4). In

subgroup analyses, very small subgroups (age≤60 years, BMI≥24 kg/m^2, HB<6 g/dL) exhibited notable instability, with highly variable HTR values and irregular inflation rate patterns, but the overall conclusions remained unchanged (sTables 13-14; sFigures 5-8).

4.4. Additional Analyses

Analysis of the selected treatment-phase windows showed that inflation rate trajectories varied across patient groups (sFigure 9). Higher albumin, lower SOFA scores, and anemia were associated with lower baselines but steeper increases, whereas elevated WBC counts were associated with lower baselines and markedly steeper rises. Regarding HTR–inflation rate dynamics (sFigures 10-11), survivors exhibited parallel increases followed by joint declines, while non-survivors typically showed a rapid peak in inflation rate despite a continued rise in HTR.

5. Discussion

In this large real-world cohort of 1,572 patients with septic shock complicated by fluid overload, we identified two divergent dynamical phenotypes of hemoglobin turnover that strongly discriminated 7-day mortality. Survivors exhibited nonlinear HTR trajectories over the treatment course, whereas non-survivors displayed linear HTR progression yet stronger nonlinear coupling between HTR and the inflation index. These phenotypes remained consistent across resuscitation phases, were robust to propensity score matching and extensive sensitivity analyses, and were externally validated in the MIMIC-IV cohort.

The model conceptualizes the BOU as an integrated functional entity, whose properties are shaped by internal composition and the stability of cellular boundaries, particularly the integrity of the erythrocyte and capillary membranes.

5.1. Alterations in Blood Components

Organismal homeostasis relies on dynamic, multi-level coordination. The "liquidity trap" following large-volume resuscitation results not from fluid administration per se, but from a short-term relative reduction in colloid osmotic pressure.

Fluid management is fundamental in septic shock for hemodynamic stability and tissue perfusion[35-37]. Early resuscitation lowers mortality[38-41] by reactivating hypoperfused cells and promoting long-term recovery. However, excessive resuscitation disrupts homeostasis when reserves are inadequate, and FO is strongly linked to adverse outcomes[42-46].

Anemia is nearly universal in sepsis[47]. FO causes proportional hemodilution, often necessitating RBC transfusion to improve oxygen delivery[48,49]. While transfusion can increase venous oxygen saturation and microvascular density, it does not consistently improve capillary flow velocity[50-52], with benefits primarily observed in patients with baseline microcirculatory impairment[53]. Impaired RBC rheology in sepsis—characterized by increased aggregation and decreased deformability[54,55] —likely contributes to microcirculatory dysfunction[54]. We propose that focusing on MCHC may offer greater insight than the traditional debate on transfusion thresholds (Appendix1.5.3).

5.2. Membrane Stability

The boundaries of BOUs are defined by capillary walls and erythrocyte membranes. We propose HTR as an indirect marker of erythrocyte membrane stability, acknowledging its dual influence by circulatory and extra-circulatory factors.

Capillary integrity is more central for fluid and composition maintenance. Sepsis primarily

injures the vasculature[56]. Endothelial damage, glycocalyx degradation, and disrupted pericyte interactions increase capillary permeability[57-59], leading to protein extravasation, reduced plasma colloid pressure, increased interstitial pressure, and tissue edema—a distinct redistribution pattern with a flatter circulating slope and a rapidly rising storage intercept.

5.3. Special Blood Components and Metabolic Decoupling

Immune cells occupy a critical niche within the BOUs. As these units traverse from the heart to functional capillaries for material exchange, nucleated components such as immune cells participate in active metabolic processes. Building on this circulatory model, we propose a new perspective to explain the potential mechanisms of macro- and micro-circulatory decoupling[60-62].

Sepsis involves coexisting hyperinflammation and immunosuppression: excessive inflammatory mediators and neutrophil extracellular traps cause direct injury[63], while immune effector cells exhibit progressive dysfunction[64-67]. Metabolically, a shift to aerobic glycolysis occurs early[68], but repeated cytokine exposure can induce an "immune paralysis" phenotype with broad metabolic defects[68]. A substrate allocation "squeezing effect" privileges immune cells, depriving parenchymal tissues and exacerbating organ dysfunction

Fluid resuscitation is essential for circulatory function in shock[11,42,69] , but its metabolic benefit is uncertain. Hemodilution dilutes both substrates and metabolites. While transient circulatory activation may aid clearance, persistent dilution suppresses exchange and—combined with prevalent mitochondrial dysfunction[70,71] — can precipitate earlier metabolic failure, exacerbating macro-microcirculatory decoupling.

5.4. Limitations

This hypothesis-driven study, based on a novel circulatory model and validated with real-world data, has several limitations. First, the observational design precludes causal inference regarding the association between HTR/inflation rate trajectories and clinical outcomes. Second, within-individual *P*-values for concurrent metabolic or perfusion-related variables cannot be directly compared across patients, as they may be influenced by dynamic perfusion changes. Moreover, despite statistical adjustments, residual confounding cannot be ruled out. Thus, our findings require validation in larger, ideally prospective, studies.

6. Conclusions

Grounded in a novel macrocirculation-first model, this study establishes HTR as a marker of erythrocyte demand and the inflation index of volume status. Their decoupling often signals clinical deterioration. Conversely, an increasing inflation index generally entails a parallel rise in metabolic demands, underscoring the integrated nature of circulatory and metabolic adaptation.

Abbreviations

RBCT, Red blood cell transfusion; TRICC, Transfusion Requirements in Critical Care; BMI, Body Mass Index; ICU, intensive care unit; BOU, blood-oxygen unit; SOFA, Sequential Organ Failure Assessment; CCI, Charlson Comorbidity Index; FB, fluid balance; FO, fluid overload; GAMs, generalized additive models; Hb, hemoglobin; HCT, hematocrit; HTR, hemoglobin turnover rate; WBC, white blood cell count; Lac, lactate; MCHC, mean corpuscular hemoglobin concentration; MCV, mean corpuscular volume; RBC, red blood cell count; IQR, interquartile range; SD, standard deviations; CI, confidence interval.

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Authors' contributions

Siyu Tang: Conceptualization, Methodology, Software, Validation, Formal analysis, Investigation, Data curation, Writing – original draft, Project administration. Huan Jiang: Data curation, Formal analysis, Writing – review & editing. Bai Xu and Yifan Xu: Supervision, Validation, Writing – review & editing. Jingquan Liu and Xianghong Yang: Resources, Supervision, Validation, Writing – review & editing

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Availability of data and materials

The datasets supporting the conclusions of this article are included within the article.

Clinical trial number

Not applicable

Ethics approval and consent to participate

This study was approved by the Institutional Review Committee of the People's Hospital of Zhejiang Province，which complies with the Declaration of Helsinki(ethics approval number: QT2022383, date: November 21, 2022). And individual consent for this retrospective analysis was waived.

Consent for publication

Not Applicable.

Competing interests

All the authors declare that there is no conflict of interest.

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