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Understanding Restrictive Versus Liberal Fluid Therapy for Major Abdominal Surgery Trial Results: Did Liberal Fluids Associate With Increased Endothelial Injury Markers?

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OBJECTIVES: Liberal fluid strategies in critically ill patients are associated with harm, thought to be due to endothelial and glycocalyx injury. As the restrictive versus liberal fluid therapy for major abdominal surgery trial not only failed to report survival benefit with restrictive fluids but was associated with a higher rate of acute kidney injury, we hypothesized that factors other than endothelial and glycocalyx injury were likely to account for these findings. Consequently, we measured injury biomarkers in a cohort of the restrictive versus liberal fluid therapy for major abdominal surgery trial.

DESIGN: The restrictive versus liberal fluid therapy for major abdominal surgery trial was an international, randomized, assessor-blinded trial comparing restrictive with liberal IV fluid regimens that represented traditional care in patients undergoing major abdominal surgery.

SETTING AND PATIENTS: Cohort of restrictive versus liberal fluid therapy for major abdominal surgery bloods was collected at a single major site (161 patients) prior to, day 1 and day 3 after surgery.

INTERVENTION: Bloods were blindly and randomly batch analyzed for plasma markers of endothelial/glycocalyx injury–angiopoietin-1, angiopoietin-2, soluble tyrosine-protein kinase-2 receptor, soluble intracellular adhesion molecule-1, syndecan, and tumor necrosis factor-α. Data were examined as restrictive versus liberal enrollment groups and high versus low (± 5,000 mL) fluid groups. Differences were examined by linear mixed modeling.

MEASUREMENT AND MAIN RESULTS: There were no significant differences in any biomarkers between the restrictive ($n = 75$) and liberal ($n = 86$) groups. When examined as low ($n = 81$) and high ($n = 79$) fluid groups, plasma angiopoietin-2 ($p = 0.009$) and soluble intracellular adhesion molecule-1 ($p = 0.01$) were elevated in the high fluid group. There were no differences in other biomarkers.

CONCLUSIONS: Although these results are consistent with previous findings of vascular injury following liberal fluid therapy, they suggest alternative mechanisms underlie the clinical outcomes from restrictive versus liberal fluid therapy for major abdominal surgery study.

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KEY WORDS: abdominal surgery; biomarkers; endothelial injury, glycocalyx injury, inflammation, restrictive fluid strategy

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Liberal fluid administration is associated with adverse clinical and organ-specific outcomes in critically ill patients (1–5). We, and others, have demonstrated an association between fluid administration and positive fluid

balance with endothelial and glycocalyx injury in both small animal models and in healthy volunteers (6–12) providing a mechanistic insights into the benefits of restrictive fluid strategy in critically ill patients (11).

Although these data are consistent with studies of patients undergoing surgery (3), the recent restrictive versus liberal fluid therapy for major abdominal surgery (RELIEF) trial enrolling at-risk patients undergoing major abdominal surgery favored liberal fluids and found no evidence that restrictive fluid translated into better disability-free survival (13). This suggests that other possible mechanisms behind these finding need to be considered and the effect of high-volume/liberal IV fluids on endothelial/ glycocalyx injury should be examined.

Based on previous studies (6–12), we hypothesized that liberal IV fluids will lead to endothelial injury measurable through increase in plasma biomarkers of inflammation, endothelial, and glycocalyx injury. We therefore undertook to examine relevant biomarkers in a single-center cohort of patients enrolled in the RELIEF trial.

METHODS

This preplanned study was conducted in a subgroup of patients enrolled for the RELIEF trial, as described previously (13, 14), at the Royal Adelaide Hospital, SA, Australia. Briefly, RELIEF study was an international, multicenter, pragmatic randomized controlled trial comparing two different fluid regimes over 24 hours in major abdominal surgery. The study included patients undergoing major abdominal surgery, with an expected duration of greater than 2 hour and an expected hospital stay greater than 3 days. The restrictive fluid intervention was designed to achieve a net zero fluid balance, with a 5 mL/kg bolus at induction of anesthesia followed by an intraoperative crystalloid infusion at a rate of 5 mL/kg/hr, continued after surgery at 0.8 mL/kg/hr for 24 hours. In contrast, the liberal group received a 10 mL/kg bolus at induction of anesthesia followed by an intraoperative crystalloid infusion at a rate of 8 mL/kg/hr, continued postoperatively at 1.5 mL/kg/hr for 24 hours (13, 14). Additional ethics approval for this study was obtained (RAH Protocol No: 130409) along with additional informed consent. The study was performed in accordance with the ethical standards as laid down in the 1964 Declaration of Helsinki and its later amendments.

All patients provided venous blood samples (~3 mL) collected into lithium heparin tubes prior to surgery, at day 1 and day 3 thereafter. Samples were stored at 4°C for less than 1 hour before processing. Samples were centrifuged at 2,000 g for 10 minutes, plasma aliquoted, and stored on site at –80°C. Batched plasma samples were transported on dry ice to the Lung Injury Research Laboratory, Flinders University, SA, Australia, and returned to –80°C until analysis.

Plasma samples were blindly and randomly batch analyzed for plasma makers of inflammation, endothelial, and glycocalyx injury (angiopoietin-1, angiopoietin-2, soluble tyrosine-protein kinase (Tie)-2 receptor, soluble intracellular adhesion molecule [ICAM]–1, syndecan, and tumor necrosis factor [TNF]–α) by commercially available enzyme-linked immunosorbent assays (R&D Systems, Minneapolis, MN), according to the manufacturer's instructions. These results were normalized post hoc to plasma volume based on hemoglobin levels, as previously (15, 16).

$$\%\,\text{change in PV} = 100 \times [1 - (\text{HbA} / \text{HbB})]$$

As these samples were a convenience cohort collected from a single center, therefore, sample size was not controlled. However, data from Calfee et al (12) showed a decrease in angiopoietin-2 from 9,460 to 7,216 pg/mL in infective patients managed with conservative fluid balance from day 1 to day 3. Using these data and alpha 0.05 and power of 80% (two-tailed significance), we calculated that 78 patients in each group (conservative and liberal) from the RELIEF study would provide adequate power to discern a difference in angiopoietin-2 from day 1 to day 3.

Statistical analyses were performed using SPSS 23.0 (PASW Inc, Chicago, IL). Variables were tested for normality by Kolmogorov-Smirnov and log transformed where necessary for statistical analyses. Baseline and clinical continuous variables were compared using Mann-Whitney *U* test or two-way analysis of variance, and categorical variables by Pearson's chi-square, as appropriate. Temporal plasma mediator data were examined as liberal versus conservative (intention to treat), high versus low fluid (dichotomized according to actual treatment, based on the median 5,000 mL of fluid administered on day 1), and disability-free survival at 1 year versus not (dichotomized according to World Health Organization Disability Assessment Schedule score ≤ 24). Differences were examined by linear mixed

modeling as an interaction effect. A *p* value of less than or equal to 0.05 was considered statistically significant.

RESULTS

One-hundred sixty-one patients were enrolled in this substudy, constituting 75 patients in the restrictive and 86 patients in the liberal arm of the RELIEF trial. At baseline, demographic and perioperative characteristics of patients were comparable between liberal and restrictive groups with the exception of additional invasive blood pressure monitoring in the liberal arm during perioperative care (**Table 1**). There were differences between the liberal and restrictive groups in the amount of administered fluid during surgery, postanesthesia care, and postoperative day 1 with resultant differences in total administered fluid and fluid balance between the groups (**Table 2**).

There was no difference in any of the examined biomarkers between patients in the restrictive and liberal groups at baseline or over time through day 1 and day 3 post surgery (**Fig. 1**) (**Fig. S1**, Supplemental Digital Content 1, http://links.lww.com/CCX/A482). In this cohort of the RELIEF patients, there was no difference between the restrictive and liberal groups in any of the primary or secondary outcomes (**Table S1**, Supplemental Digital Content 1, http://links.lww.com/CCX/A482).

When the patients were dichotomized as high versus low volume IV fluid groups based on the median of 5,000 mL of fluid administered on day 1, that is, nonrandomized, there were 81 patients in the low fluid and 79 in the high fluid group. More patients in the high fluid group were male, had a greater body weight, and had a longer duration of surgery when compared with the low fluid group (Table 1). There were differences between the groups in the amount of administered fluid during surgery, postanesthesia care, and postoperative day 1 with resultant differences in total administered fluid and fluid balance between the groups (Table 2).

There were no differences in biomarkers at baseline; however, there was an increase in plasma angiopoietin-2 ($p = 0.009$) and soluble ICAM-1 ($p = 0.01$) at day 1 and 3 in the high fluid group when compared with the low fluid group (**Fig. 2**). There was no difference between the groups in other examined markers (**Fig. S2**, Supplemental Digital Content 1, http://links.lww.com/CCX/A482). There were statistical differences in the 1-year disability-free survival and duration of hospital stay with worse outcomes in the high fluid group compared with the low fluid group, but other examined outcomes were not statistically different (Table S1, Supplemental Digital Content 1, http://links.lww.com/CCX/A482). The difference in angiopoietin-2 ($p = 0.001$) and soluble ICAM (sICAM)–1 ($p = 0.011$) remained when the total cohort was examined as quartiles based on the fluid administered on day 1, indicating a dose-dependent relationship (**Fig. 3**).

Similar results with angiopoietin-2 and sICAM-1 were seen in the plasma biomarkers when they were not normalized for change in plasma volume based on hemoglobin levels, except for angiopoietin-1 which was high in the high fluid group pre surgery, day 1, and day 3 (**Figs. S3–S6**, Supplemental Digital Content 1, http://links.lww.com/CCX/A482), and there was no statistical difference with change in hemoglobin between the groups (liberal and restrictive group effect, $p = 0.45$; high and low fluid group effect, $p = 0.23$) (**Figs. S7** and **S8**, Supplemental Digital Content 1, http://links.lww.com/CCX/A482). Finally, patients with disability-free survival at 1 year ($n = 134$) had lower plasma angiopoietin-2 but no difference in any of the other examined biomarkers (**Figs. S9** and **S10**, Supplemental Digital Content 1, http://links.lww.com/CCX/A482).

DISCUSSION

The liberal fluid arm of the RELIEF study was not associated with increases in the markers of inflammation, endothelial, or glycocalyx injury examined in this substudy. However, secondary analysis demonstrates an increase in plasma angiopoietin-2 and sICAM-1 suggesting endothelial lung injury with the higher fluid volume (median value > 5 L).

This finding is consistent with previous studies where fluid administration and positive fluid balance have been associated with an increase in angiopoietin-2 levels (7–9, 11, 12, 17). Angiopoietin-2, a marker of endothelial lung injury (18, 19), is increased during liberal fluid therapy which in turn is associated with worse outcomes in patients with acute lung injury (4), whereas the ratio of angiopoietin-1 to angiopoietin-2 predicts mortality in patients with lung injury (20, 21). In our study cohort, although there was a difference in angiopoietin-2 between high and low fluid groups, there was no difference in the angiopoietin-1/-2 ratio indicating alternate factors may affect plasma angiopoietin-1.

Additionally, angiopoietin-2 sensitizes endothelial cells to the action of TNF-α (22) and has a crucial role in induction and perpetuation of inflammation leading

TABLE 1.

Demographic and Perioperative Characteristics of the Patients at Baseline by Enrollment and Total Fluid Administered Groups

Characteristics	Restrictive Fluid (*n* = 75)	Liberal Fluid (*n* = 86)
Age (yr), mean (SD)	65 (13)	67 (14)
Male, *n* (%)	24 (32)	39 (45)
Body weight (kg), median (IQR)	90 (71–109)	95 (77–116)
ASA physical status classification system, *n* (%)		
1	1 (1.3)	1 (1.2)
2	25 (33.3)	22 (25.6)
3	48 (64.0)	60 (69.8)
4	1 (1.3)	3 (3.5)
Preoperative WHODAS score, median (IQR)	17 (14–22)	19 (14–25)
Coexisting medical condition, *n* (%)		
Hypertension	43 (57.3)	55 (64.0)
Coronary artery disease	11 (14.7)	14 (16.3)
Heart failure	2 (2.7)	3 (3.5)
Previous myocardial infarction	9 (12.0)	8 (9.3)
Peripheral vascular disease	2 (2.7)	4 (4.7)
Current smoker	9 (12.0)	7 (8.1)
History of stroke or TIA	4 (5.3)	5 (5.8)
Chronic obstructive pulmonary disease	19 (25.3)	24 (27.9)
Moderate or severe renal disease	2 (2.7)	4 (4.7)
Perioperative care, *n* (%)		
Neuraxial block	16 (21.3)	17 (19.8)
Invasive blood pressure monitoring	42 (56.0)	64 (74.4)[b]
CVP monitoring	3 (4)	8 (9.3)
Type of surgery, *n* (%)		
Esophageal or gastric	10 (13.3)	19 (22.1)
Hepatobiliary	1 (1.3)	2 (2.3)
Colorectal	34 (45.3)	44 (51.2)
Urologic or renal	2 (2.7)	0 (0)
Gynecologic	26 (34.7)	20 (23.3)
Other	2 (2.7)	1 (1.2)

(*Continued*)

TABLE 1. (*Continued*).

Demographic and Perioperative Characteristics of the Patients at Baseline by Enrollment and Total Fluid Administered Groups

Characteristics	Restrictive Fluid ($n = 75$)	Liberal Fluid ($n = 86$)
Surgical method, *n* (%)		
Open	46 (61.3)	46 (53.5)
Laparoscopic	20 (26.7)	35 (40.7)
Laparoscopic assisted	9 (12.0)	5 (5.8)
Duration of surgery (hr), median (IQR)	2.2 (1.5–2.8)	2.5 (1.7–3.4)
Planned postoperative care in HDU or ICU, *n* (%)	17 (22.6)	16 (18.6)
Characteristics	Low Fluid (< 5,000 mL) ($n = 81$)	High Fluid (> 5,000 mL) ($n = 79$)
Age (years), mean (SD)	67 (14)	65 (13)
Male, *n* (%)	25 (31)	37 (47)[b]
Body weight (kg), median (IQR)	82 (67–106)	100 (84–122)[a]
ASA physical status classification system, *n* (%)		
1	2 (2.5)	0 (0)
2	28 (34.6)	18 (22.8)
3	50 (61.7)	58 (73.4)
4	1 (1.2)	3 (3.8)
Preoperative WHODAS score, median (IQR)	17 (14–23)	19 (14–24)
Coexisting medical condition, *n* (%)		
Hypertension	47 (58.0)	51 (64.5)
Coronary artery disease	16 (19.8)	8 (10.1)
Heart failure	2 (2.5)	3 (3.8)
Previous myocardial infarction	12 (14.8)	5 (6.3)
Peripheral vascular disease	2 (2.5)	4 (5.1)
Current smoker	11 (13.6)	5 (6.3)
History of stroke or TIA	5 (6.2)	4 (5.1)
Chronic obstructive pulmonary disease	22 (27.2)	21 (26.6)
Moderate or severe renal disease	4 (4.9)	2 (2.5)
Perioperative care, *n* (%)		
Neuraxial block	18 (22.2)	15 (19.0)
Invasive blood pressure monitoring	44 (54.3)	61 (77.2)
CVP monitoring	2 (2.5)	9 (11.4)

(*Continued*)

TABLE 1. (*Continued*).
Demographic and Perioperative Characteristics of the Patients at Baseline by Enrollment and Total Fluid Administered Groups

Characteristics	Restrictive Fluid (*n* = 75)	Liberal Fluid (*n* = 86)
Type of surgery, *n* (%)		
Esophageal or gastric	10 (12.3)	19 (24.1)
Hepatobiliary	1 (1.2)	2 (2.5)
Colorectal	37 (45.7)	40 (50.6)
Urologic or renal	1 (1.2)	1 (1.3)
Gynecologic	31 (38.3)	15 (19.0)
Other	1 (1.2)	2 (2.5)
Surgical method, *n* (%)		
Open	48 (59.3)	44 (55.7)
Laparoscopic	23 (28.4)	31 (39.2)
Laparoscopic assisted	10 (12.3)	4 (5.1)
Duration of surgery (hr), median (IQR)	2.0 (1.3–2.6)	2.9 (2.0–3.5)[a]
Planned postoperative care in HDU or ICU, *n* (%)	13 (16.0)	20 (25.4)

CVP = central venous pressure, HDU = high dependency unit, IQR = interquartile range, TIA = transient ischemic attack, WHODAS = World Health Organization Disability Assessment Schedule.
[a]Significant difference $p \leq 0.001$ by Mann-Whitney *U* test.
[b]Significantly different $p < 0.05$ by Pearson χ^2.

to endothelial barrier dysfunction (23). Angiopoietin-2 appears to be an essential and central component of cytokine induced vascular leakage (24). Its inhibitory action on Tie-2 receptors blocks the regulation/stabilization function of angiopoietin-1, leading to a greater induction of permeability by TNF-α on blood vessels and further perpetuation of inflammation through release of soluble adhesion molecules such as ICAM and vascular cell adhesion molecule (25). ICAM-1 mediates lung leukocyte recruitment, activates lung macrophages, enhances lung injury, and its levels are increased in patients with acute lung injury (26–29). Although we did not find any change in the soluble Tie-2 receptor with increased fluid administration, there was an increase in soluble ICAM in the high fluid group. The lack of increase in soluble Tie-2 receptor might not only represent a true finding but also may signify that the soluble marker of Tie-2 and the up-regulation/down-regulation of these receptors may not be easily measurable in clinical samples. In the main RELIEF study, the rate of pulmonary edema was lower with the restrictive fluid group (risk ratio [95% CI], 0.63 [0.36–1.09]; $p = 0.10$), and an increase in angiopoietin-2 and sICAM levels with higher fluids may provide some rationale to these findings.

Similarly, the volume of IV fluid administered during sepsis resuscitation is independently associated with the degree of glycocalyx degradation (6, 10). However, we did not find any difference in the syndecan levels between the different fluid strategies. This suggests that patients in the restrictive fluid or low fluid group may have an additional insult leading to similar glycocalyx injury, or there might have been minimal injury in these patients. Furthermore, renal function was worse in the restrictive fluid group, potentially effecting the systemic levels of biomarkers not specific to the lung such as syndecan (30), which is renally cleared, and may have altered the levels at day 3.

The difference in the results examined as restrictive versus liberal and high versus low fluid group seems worth exploring. The additional 500 mL of cumulative fluid in the 24 hours after surgery, 300 mL of additional

TABLE 2.
Blood Loss and Administered IV Fluid Volumes by Enrollment and Total Fluid Administered Groups

Variables	Restrictive Fluid ($n = 75$)	Liberal Fluid ($n = 86$)	p
During surgery			
Intraoperative blood loss (mL), median (IQR)	200 (100–300)	200 (100–325)	0.999
Intraoperative fluid administration (mL), median (IQR)			
Crystalloid	1,400 (1,100–2,000)	2,800 (2,200–3,725)	≤ 0.001
Colloid	0 (0–0)	0 (0–0)	0.616
Infusion rate (mL/kg/hr), median (IQR)	7.6 (5.6–9.3)	12.7 (9.6–16.1)	≤ 0.001
In PACU			
Administration of fluid (mL), median (IQR)			
Crystalloid	228 (160–365)	390 (300–615)	≤ 0.001
Colloid	0 (0)	0 (0)	0.498
Postoperative day 1, post PACU			
Administration of fluid (mL), median (IQR)			
Crystalloid	1,515 (1,260–1,800)	2,845 (2,295–3,370)	≤ 0.001
Colloid	0 (0)	0 (0)	0.425
Infusion rate (mL/kg/hr), median (IQR)	0.9 (0.8–1.0)	1.5 (1.3–1.6)	≤ 0.001
At 24 hr after surgery			
Cumulative total for IV fluids (mL), median (IQR)	3,350 (2,720–4,000)	6,053 (5,382–7,425)	≤0.001
Fluid balance (mL), median (IQR)	1,339 (789–2,294)	3,685 (2,691–4,637)	≤ 0.001
Weight gain (kg), median (IQR)	−0.3 (− 1.9 to 1.1)	1.2 (−1.0 to 2.2)	0.006
Variables	Low Fluid (< 5,000 mL) ($n = 81$)	High Fluid (> 5,000 mL) ($n = 79$)	p
During surgery			
Intraoperative blood loss (mL), median (IQR)	150 (100–300)	200 (100–400)	0.071
Intraoperative fluid administration (mL), median (IQR)			
Crystalloid	1,400 (1,060–2,000)	3,000 (2,500–3,800)	≤ 0.001
Colloid	0 (0)	0 (0)	0.051
Infusion rate (mL/kg/hr), median (IQR)	8.1 (5.7–11.4)	11.7 (8.7–14.7)	≤ 0.001

(*Continued*)

TABLE 2. (*Continued*).
Blood Loss and Administered IV Fluid Volumes by Enrollment and Total Fluid Administered Groups

Variables	Restrictive Fluid (*n* = 75)	Liberal Fluid (*n* = 86)	*p*
In PACU			
Administration of fluid (mL), median (IQR)			
Crystalloid	234 (155–375)	437 (300–673)	≤0.001
Colloid	0 (0)	0 (0)	0.252
Postoperative day 1, post PACU			
Administration of fluid (mL), median (IQR)			
Crystalloid	1,625 (1,347–1,895)	2,922 (2,306–3,400)	≤ 0.001
Colloid	0 (0)	0 (0)	0.083
Infusion rate (mL/kg/hr), median (IQR)	0.9 (0.8–1.1)	1.4 (1.1–1.6)	≤ 0.001
At 24 hr after surgery			
Cumulative total for IV fluids (mL), median (IQR)	3,576 (2,912–4,095)	6,505 (5,760–7,670)	≤ 0.001
Fluid balance (mL), median (IQR)	1,422 (852–2,319)	3,915 (2,967–5,033)	≤ 0.001
Weight gain (kg), median (IQR)	−0.2 (−1.6 to 1.5)	1.1 (−1.1 to 2.3)	0.025

IQR = interquartile range, PACU = postanesthesia care unit.
Mann-Whitney *U* test.

cumulative fluid balance in the high fluid group when compared with the liberal group, is unlikely to explain these different results. Factors such as the use of vasopressors for treating hypotension in the presence of hypovolemia or the effects of different types of anesthesia (31) on kidney function may have been responsible and should be explored in the future. Finally, the effect of fluids may be different in patients developing lung injury with different phenotypes (32) and could have contributed to the observed findings.

Although the difference in clinical outcomes in the high and low fluid volume groups, with decrease in length of hospital stay and increase in the disability-free survival at 1 year in the low fluid group, is hypothetically plausible, the examined numbers are small and because of multiple testing, risk type I error.

Our study has highlighted the necessity for caution when considering extrapolation of the results of the RELIEF trial, which were conducted in a defined cohort of patients undergoing major abdominal surgery, to the use of liberal fluids to a nonspecific critical care population, as often these patients have pre-existing endothelial and lung injury which may be worsened by a liberal fluid approach.

Our study had some limitations, we conducted post hoc alternate analyses of the randomized patient data examining patients based on their actual administered fluid volumes, which may introduce multiple imbalance in the study groups. Patients in the high fluid group were heavier in body weight and had longer duration of surgery and may represent a sicker cohort of patients, and this may have biased our findings even though they have similar 1) ASA status, and (2) type of surgery, 3) weight-based fluid administration was capped at the maximum weight of 100 kg, 4) the markers have been corrected to the estimated change in plasma volume based on hemoglobin levels as previously described (15, 16, 33); however, this might be not be accurate in patients undergoing

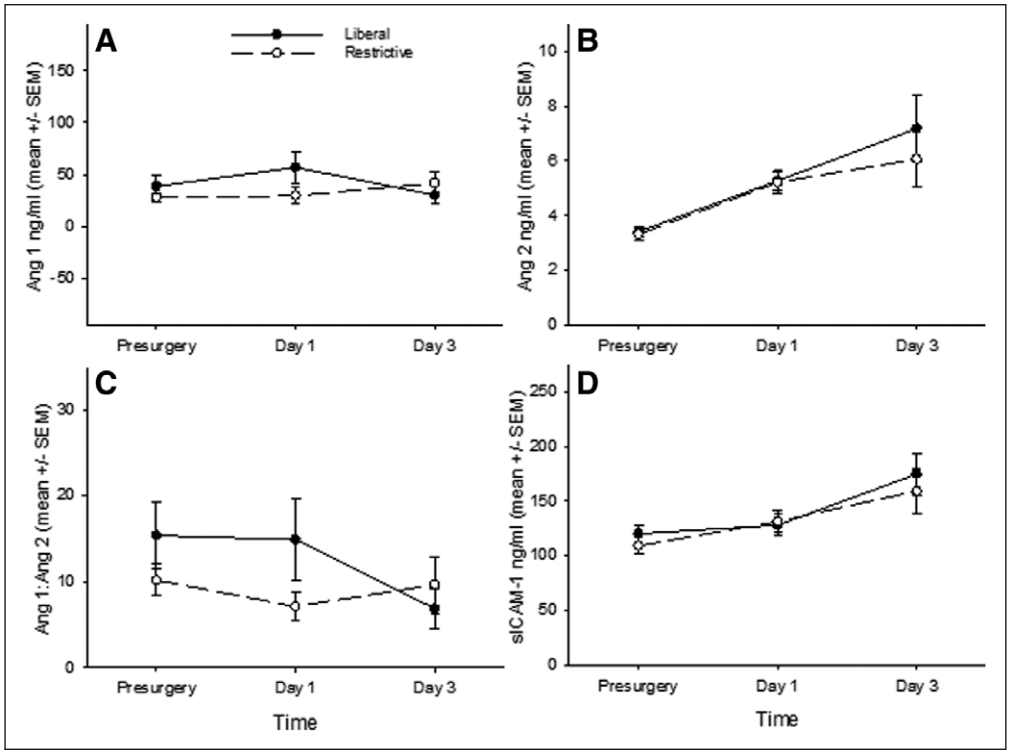

Figure 1. Plasma biomarkers, normalized for change in plasma volume, by treatment groups during the first 3 d from surgery. Ang = angiopoietin, sICAM = soluble intracellular adhesion molecule.

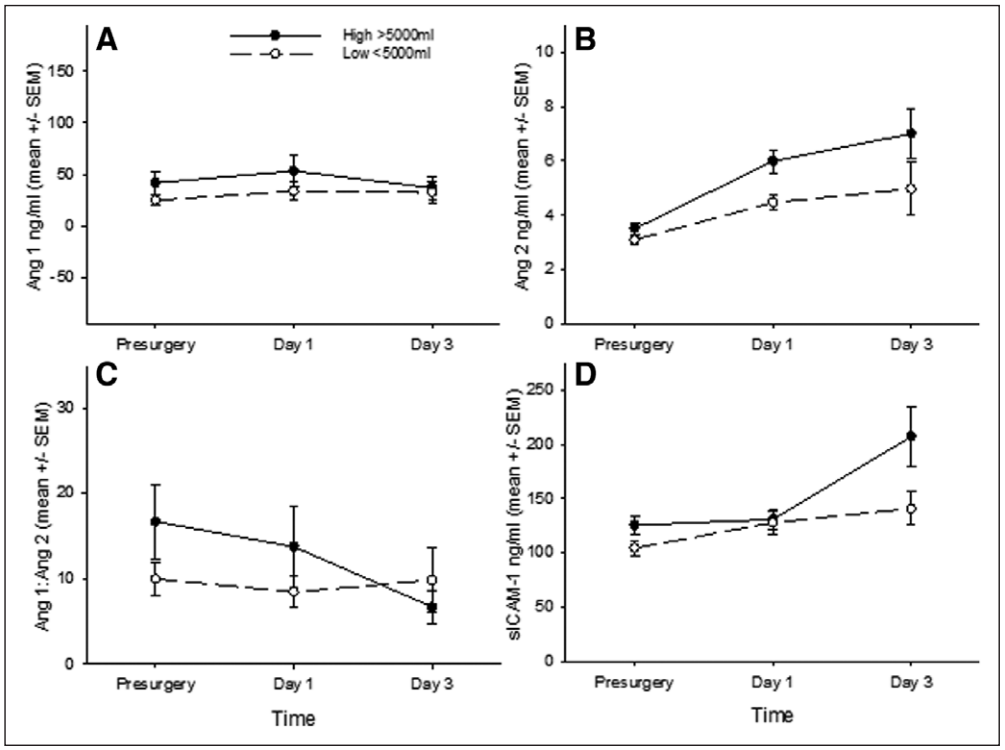

Figure 2. Plasma biomarkers, normalized for change in plasma volume, by total IV fluid administered, dichotomized into High greater than 5,000 mL and Low less than 5,000 mL groups, during the first 3 d from surgery. Ang = angiopoietin, sICAM = soluble intracellular adhesion molecule.

major abdominal surgery and the hemoglobin levels might not represent a true reflection of plasma volume and 5) the increase in angiopoietin-2 was seen with all the quartiles of administered fluid volume. Our sample size was small and represents only a small cohort from a single center of the original study, and this may have introduced a selection bias. However, the patient characteristics and administered fluid volumes in the restrictive and liberal groups were similar to the main study. In addition, there are other markers of endothelial and glycocalyx injury such as heparin sulphate (6) and endocans which we did not examine and should be examined in future studies to enable a more complete picture of the potential mechanisms. Finally, it is important to emphasize that, despite our use of mixed modeling to account for measured confounders, our cohorts were underpowered to address additional pertinent variables that could affect these biomarkers, such as underlying comorbidities. Despite analysis limitations, the subset data are consistent with prior work, albeit without an associated clinical endpoint. This suggests that additional mechanisms need to be considered and that clinical approaches to fluid therapy (liberal vs restrictive) need to be

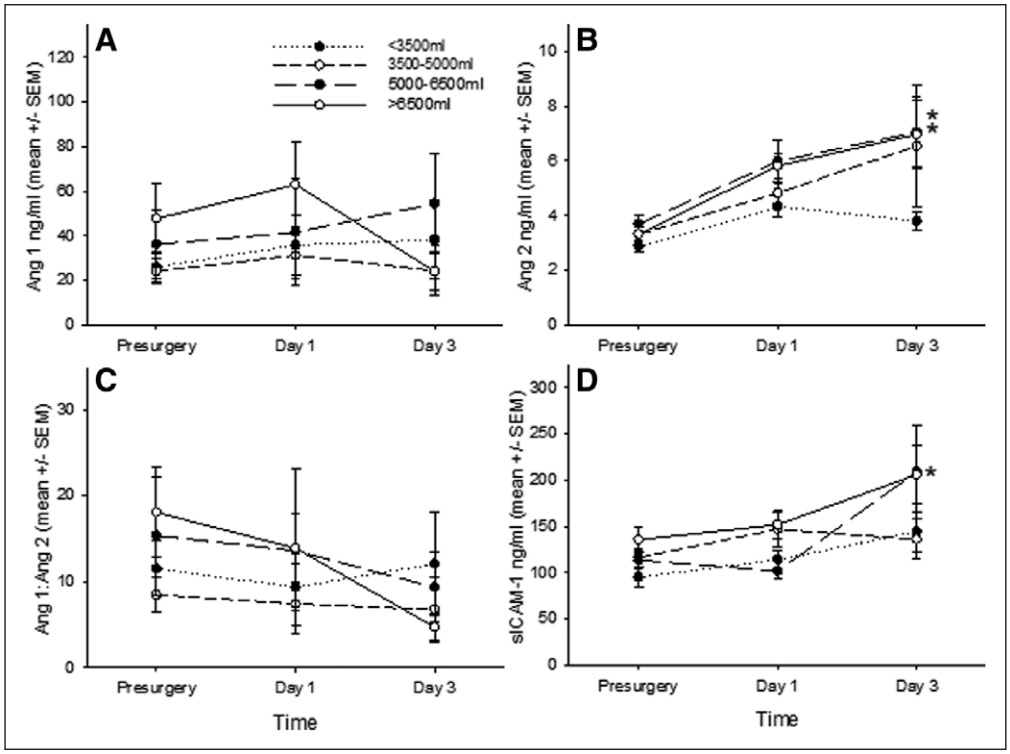

Figure 3. Plasma biomarkers, normalized for change in plasma volume, by total IV fluid administered, analyzed as quartiles, during the first 3 d from surgery. *Significantly different from less than 3,500 mL group; $p \leq 0.011$. Ang = angiopoietin, sICAM = soluble intracellular adhesion molecule.

contextual and that additional data are key to assisting with study design (34) that will elucidate this issue.

CONCLUSIONS

The liberal fluid arm of the RELIEF study was not associated with an increase in the markers of endothelial or glycocalyx injury investigated in this substudy; however, greater fluid administration was associated with increased plasma angiopoietin-2 and soluble ICAM-1 suggesting endothelial injury. These results indicate alternative factors to endothelial and glycocalyx injury may have been responsible for the clinical results of the RELIEF study.

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