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ORIGINAL ARTICLE

MUlticenter STudy of tissue plasminogen activator (alteplase) use in COVID-19 severe respiratory failure (MUST COVID): A retrospective cohort study

Christopher D. Barrett MD[1,2,3] | Hunter B. Moore MD, PhD[4] | Ernest E. Moore MD[4,5] | Dudley Benjamin Christie III MD[6] | Sarah Orfanos MD[7] | Lorenzo Anez-Bustillos MD[3] | Rashi Jhunjhunwala MD, MA[3] | Sabiha Hussain MD[7] | Shahzad Shaefi MD, MPH[8] | Janice Wang MD[9] | Negin Hajizadeh MD, MPH[9] | Elias N. Baedorf-Kassis MD[10] | Ammar Al-Shammaa MSc[8] | Krystal Capers MPH[8] | Valerie Banner-Goodspeed MPH[8] | Franklin L. Wright MD[4] | Todd Bull MD[4] | Peter K. Moore MD[4] | Hannah Nemec MD[6] | John Thomas Buchanan BS[6] | Cory Nonnemacher MD[6] | Natalie Rajcooar BA[9] | Ramona Ramdeo DNP[9] | Mena Yacoub BS[9] | Ana Guevara BA[9] | Aileen Espinal BS[9] | Laith Hattar MD[11] | Andrew Moraco MD[11] | Robert McIntyre MD[4] | Daniel S. Talmor MD, MPH[8] | Angela Sauaia MD, PhD[5,12] | Michael B. Yaffe MD[2,3]

[1]Department of Surgery, Boston University School of Medicine, Boston, Massachusetts, USA

[2]Koch Institute for Integrative Cancer Research, Center for Precision Cancer Medicine, Departments of Biological Engineering and Biology, Massachusetts Institute of Technology, Cambridge, Massachusetts, USA

[3]Department of Surgery, Beth Israel Deaconess Medical Center, Harvard Medical School, Boston, Massachusetts, USA

[4]Department of Surgery, University of Colorado Denver, Aurora, Colorado, USA

[5]Department of Surgery, Ernest E Moore Shock Trauma Center at Denver Health, Denver, Colorado, USA

[6]Department of Trauma and Critical Care, The Medical Center Navicent, Mercer University School of Medicine, Macon, Georgia, USA

[7]Department of Medicine, Rutgers Robert Wood Johnson Medical School, New Brunswick, New Jersey, USA

[8]Department of Anesthesia, Critical Care and Pain Medicine, Beth Israel Deaconess Medical Center, Harvard Medical School, Boston, Massachusetts, USA

[9]Feinstein Institutes for Medical Research, Northwell Health, Manhasset, New York, USA

[10]Division of Pulmonary, Critical Care and Sleep Medicine, Department of Medicine, Beth Israel Deaconess Medical Center, Harvard Medical School, Boston, Massachusetts, USA

[11]Division of Pulmonary and Critical Care, Department of Medicine, St. Elizabeth's Medical Center, Tufts University School of Medicine, Boston, Massachusetts, USA

[12]Colorado School of Public Health and Department of Surgery, University of Colorado Denver, Denver, Colorado, USA

Christopher D. Barrett and Hunter B. Moore are co-first authors.

Correspondence
Ernest E. Moore, Department of Surgery, University of Colorado Denver, 777 Bannock Street, Denver, CO 80204, Aurora, CO, USA.
Email: ernest.moore@dhha.org

Michael B. Yaffe, Koch Institute for Integrative Cancer Research, Center for Precision Cancer Medicine, Departments of Biological Engineering and Biology, Massachusetts Institute of Technology, 500 Main Street, Room 353, Cambridge, MA 02139, USA.
Email: myaffe@mit.edu

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Abstract

Background: Few therapies exist to treat severe COVID-19 respiratory failure once it develops. Given known diffuse pulmonary microthrombi on autopsy studies of COVID-19 patients, we hypothesized that tissue plasminogen activator (tPA) may improve pulmonary function in COVID-19 respiratory failure.

Methods: A multicenter, retrospective, observational study of patients with confirmed COVID-19 and severe respiratory failure who received systemic tPA (alteplase) was performed. Seventy-nine adults from seven medical centers were included in the final analysis after institutional review boards' approval; 23 were excluded from analysis because tPA was administered for pulmonary macroembolism or deep venous thrombosis. The primary outcome was improvement in the PaO_2/FiO_2 ratio from baseline to 48 h after tPA. Linear mixed modeling was used for analysis.

Results: tPA was associated with significant PaO_2/FiO_2 improvement at 48 h (estimated paired difference = 23.1 ± 6.7), which was sustained at 72 h (interaction term $p < 0.00$). tPA administration was also associated with improved National Early Warning Score 2 scores at 24, 48, and 72 h after receiving tPA (interaction term $p = 0.00$). D-dimer was significantly elevated immediately after tPA, consistent with lysis of formed clot. Patients with declining respiratory status preceding tPA administration had more marked improvement in PaO_2/FiO_2 ratios than those who had poor but stable (not declining) respiratory status. There was one intracranial hemorrhage, which occurred within 24 h following tPA administration.

Conclusions: These data suggest tPA is associated with significant improvement in pulmonary function in severe COVID-19 respiratory failure, especially in patients whose pulmonary function is in decline, and has an acceptable safety profile in this patient population.

Essentials

- Few effective therapies exist for critically ill COVID-19 respiratory failure patients.
- A retrospective study of 79 severe COVID-19 respiratory failure patients was performed.
- Systemic alteplase is associated with improved oxygenation in severe COVID-19.
- We found a low risk (1.3%) of intracranial hemorrhage with alteplase use in COVID-19 patients.

1 | INTRODUCTION

The SARS-CoV-2 (COVID-19) global pandemic overwhelmed the capacity of many medical infrastructures to accommodate a large surge of patients with acute respiratory distress syndrome (ARDS), particularly those requiring mechanical ventilation. ARDS currently has little evidence-based treatment other than low tidal volume ventilation to limit mechanical stress on the lung[1] and prone positioning,[2] with additional evidence for benefit of steroids in COVID-related ARDS.[3] Although vaccination and public health measures remain the mainstay of reducing COVID-19 disease burden, new viral variants, inadequate access, and skepticism of vaccines in the broader public exist. Thus, an ongoing effort to develop new therapeutic approaches capable of rapidly treating and attenuating ARDS secondary to COVID-19 is essential.

The dominant pathologic feature of viral-induced ARDS is fibrin accumulation in the microvasculature and airspaces, and multiple autopsy studies have now confirmed that nearly all patients who die of COVID-19 have diffuse pulmonary microthrombi as a prominent feature.[4-6] The high physiologic dead space and relatively preserved lung compliance early in the course of respiratory failure from COVID-19 suggests this histopathologic finding is not incidental, but rather that pulmonary vascular microthrombosis is a significant contributor to the development of these patients'

respiratory compromise,[7] particularly early in their course before the fibroproliferative phase predominates. Therapeutic anticoagulation initiated before precipitous respiratory decline has now been shown to improve clinical outcomes in the large, multicenter ACTIV-4 trial which was halted early for efficacy in the "moderate group".[8] However, in the "severe group" within ACTIV-4 who had severe respiratory failure before initiation of therapeutic anticoagulation, the study was halted early for futility.[9] This is consistent with the concept that anticoagulation before the development of an overwhelming microthrombotic burden is beneficial, but once a significant microthrombotic burden exists it is too late to benefit from anticoagulation because the thrombotic phenomena has already occurred. At this point, when all less aggressive clinical options have been exhausted, our group hypothesized that there may be a role for fibrinolytic therapy with tissue plasminogen activator (tPA) to salvage pulmonary microvascular patency and improve oxygenation in patients who would otherwise die of hypoxemic respiratory failure.[10-12]

The notion that fibrinolytic therapy may have a role in ARDS is not new, with substantial preclinical work suggesting that fibrinolytic therapy can attenuate ARDS provoked from diverse insults (reviewed in Liu et al.[13] and Barrett et al.[10]). Further, in 2001 a small phase 1 clinical trial[14] indicated that urokinase and streptokinase were effective in patients with terminal ARDS, markedly improving oxygenation and reducing an expected mortality from 100% to 70%. A more contemporary approach to thrombolytic therapy uses tPA rather than urokinase or streptokinase because of its higher efficacy of clot lysis with comparable bleeding risk.[15] Several case series and a small retrospective observational study have now been published suggesting a potential benefit of tPA therapy in COVID-19 respiratory failure.[16-21]

To investigate the respiratory changes associated with tPA use in COVID-19 respiratory failure, a retrospective analysis of existing data from multiple centers with experience using tPA in COVID-19 respiratory failure was proposed. To accomplish this, we established a registry of retrospectively collected deidentified clinical data from COVID-19 patients who were treated with tPA for severe acute respiratory failure across multiple centers. We hypothesized that tPA administered to patients with COVID-19-associated acute respiratory failure would be associated with improved pulmonary function within 48 h with a low risk for severe bleeding.

2 | METHODS

2.1 | Study design

The MUST COVID study is a multicenter, retrospective, observational study of patients with confirmed COVID-19 severe respiratory failure (i.e., requiring mechanical ventilation) who received tPA (alteplase, sold under tradename Activase by Genentech, Inc.). Baseline characteristics, comorbidities, and rationale for tPA administration were collected along with tPA dosing information, concomitant anticoagulation, and use of remdesivir and/or dexamethasone. Clinical and laboratory data were obtained at 6-h intervals for the 72 h preceding and the 72 h following administration of tPA in addition to adverse events and hospital mortality data.

2.2 | Setting

Seven academic tertiary care hospitals agreed to participate: Beth Israel Deaconess Medical Center (Boston, MA), Northwell Health/Long Island Jewish Medical Center (Queens, NY), Denver Health Medical Center (Denver, CO), University of Colorado Medical Center (Aurora, CO), Navicent Health Medical Center/Mercer University School of Medicine (Macon, GA), Rutgers Robert Wood Johnson Medical Center (Newark, NJ), and St. Elizabeth's Medical Center/Tufts University School of Medicine (Boston, MA).

2.3 | Participants and ethics approval

All adult patients (≥18 years old) admitted from March 1, 2020, through March 3, 2021, with confirmed COVID19 respiratory failure requiring ventilatory support who were treated with tPA were eligible. Patients for whom tPA was administered specifically for imaging-confirmed pulmonary embolism or deep venous thrombosis were excluded, as were known prisoners. Twenty of the 102 patients were enrolled in the STARS trial (NCT04357730), and 15 patients previously had some of their data included in the Study of Treatment and Outcomes in Critically Ill Patients with COVID-19 database (NCT04343898). All participating trial sites had study approval and oversight from their respective institutional review boards.

2.4 | Variables

2.4.1 | Outcomes

The primary outcome was improvement in PaO_2/FiO_2 from pre-tPA baseline (i.e., 3–6 h before tPA administration) to up to 48 h (within 42–54 h) after the first dose of tPA. Secondary outcomes included improvement in dead-space ventilation,[20] estimated by the ventilatory ratio (calculated as proposed by Sinha et al.[22] and National Early Warning System-2 score [NEWS2][23]),bleeding (defined as any bleeding requiring therapy such as blood product transfusion, operative procedure, tranexamic acid, or resulting in prolonged hospitalization, death, or disability) or thrombotic complications, complications, in-hospital mortality, ventilator-free days, and intensive care unit free days (both up to 28 days since admission). All complications occurring within 72 h of tPA administration were deemed potentially related to tPA. The reported study outcomes were predefined in the

institutional review board application before any data collection or analysis (Appendix S1).

2.4.2 | Covariates

We collected data on demographic characteristics shown in previous studies to affect the prognosis of COVID-19 pulmonary failure such as age, sex, body mass index, comorbidities and complications present before tPA administration, as well as administration of two medications shown in previous studies to improve survival in these patients (dexamethasone and remdesivir).

2.4.3 | Bias minimization

This is a retrospective cohort of patients known to have been treated with tPA, and thus is inherently affected by selection bias because the decision to give tPA was not standardized but instead made by the local health care providers (except for the participants of the STARS randomized controlled trial,[24] in which case the decision was by randomization). Although this generated a heterogenous population, it provides a close to real-world view of the safety profile of the drug and evidence of tPA effect because no patients were excluded resulting from comorbidities. To decrease time effects bias, we used a spline linear regression, a statistical technique to assess pre- and post-tPA time trends in the pulmonary function variables. To further control bias, we excluded patients who received tPA specifically for the treatment of imaging-confirmed pulmonary embolism (PE) or deep venous thrombosis/suspected PE. The relationship of adverse events with tPA was decided based on timing (i.e., all adverse events occurring within 72 h after tPA were deemed potentially associated with the drug to minimize interpretation bias).

2.4.4 | Study size determination

We included all patients who were admitted since the beginning of the US COVID-19 pandemic and who received tPA for respiratory failure up to March 2021, as described previously. As no comparator was selected, we did not calculate power/sample size.

2.5 | Statistical analysis

Analysis was conducted using linear mixed models for the outcomes with pairwise comparisons with the time immediately before tPA was administered. The models allow for missing observations, adjustment for confounders, repeated measures data by subject, and account for the clustering effects by institution. Confounders were chosen based on their univariate association with mortality with $p < 0.25$ or because they were shown to be clinically relevant in COVID-19. Effect modification by trends in outcomes before tPA administration was assessed by testing interactions in the model. A qualitative analysis of nonsurvivors was also conducted to better understand the cause of death and potential association with tPA. Overall significance was set at $p < 0.05$ for the time trends effect, followed by pairwise comparisons between the time right before tPA (baseline) and other times adjusted by false-discovery rate to minimize type I error. All quantitative analyses were carried out with SAS vs 9.4 (SAS Institute, Cary, NC).

3 | RESULTS

Overall, there were 102 patients admitted from March 1, 2020, through March 3, 2021, at the seven participating centers with laboratory confirmed COVID-19 infection resulting in severe acute respiratory failure requiring mechanical ventilation who received tPA. Twenty-three (22.5%) patients were excluded because their principal reason for tPA therapy was an imaging-confirmed diagnosis of PE or deep venous thrombosis/suspected PE.

Table 1 shows the demographics and medical history of the 79 patients included in the analysis, whereas Table 2 shows the pre-tPA physiology, major therapeutic interventions (dexamethasone and remdesivir), tPA dosing and anticoagulation information, and outcomes. White males in the sixth and seventh decades of life predominated, with close to one-half being Latinx and covered by government-issued health insurance. Obesity and comorbidities, especially hypertension and diabetes, were frequent. Most patients were sedated, thus explaining the low Glasgow Coma Scale number. Hemodynamic instability requiring vasopressors was present at the time of tPA administration in about one-half of the patients. The median PaO_2/FiO_2 ratio immediately before tPA administration was low, at 93.0 (interquartile range: 71.0–131.0). Fewer than one-third received (or were receiving) dexamethasone or remdesivir before tPA was given because the study enrollment preceded the discovery of these drugs' benefit in COVID-19. The initial tPA doses varied, but the overwhelming majority ($N = 61$) received 50 mg on their first dose, with the next most common initial dose being 100 mg ($N = 12$). The method of dosing was highly variable, with 20 different variations between how much was "pushed" versus dripped over a short period for bolus patients and how much was pushed/dripped before prolonged infusions versus rates of prolonged infusions with no preceding bolus; this variability did not allow for meaningful analysis between these abundant but minor variations in dosing methods. Overall, 44.0% of patients received a second dose of tPA. Sixty-seven (85%) of patients received concomitant therapeutic anticoagulation with heparin (Table 2), three (4%) received therapeutic enoxaparin and two (2%) received argatroban; the other seven (9%) received either prophylactic doses of heparinoids or no anticoagulants at all. Dosing strategies and amounts for therapeutic heparin regimens

TABLE 1 Demographics and medical history at baseline of the studied patients (baseline is defined as immediately before tPA was administered)

Variables	Total (*N* = 79)	Survivors (*N* = 33)	Nonsurvivors (*N* = 46)	*p* Value
Facility				
6030	13 (17)	5 (15)	8 (17)	0.00
6031	9 (11)	2 (6)	7 (15)	
6032	13 (17)	11 (33)	2 (4)	
6033	16 (20)	10 (30)	6 (13)	
6035	24 (30)	3 (9)	21 (46)	
6438	3 (4)	2 (6)	1 (2)	
Age (years)	61 (51–68)	58 (47–66)	62 (53–68)	0.14
Sex = male	56 (71)	25 (76)	31 (67)	0.42
Race				
Missing	6 (8)	3 (9)	3 (7)	0.56
White	54 (74)	23 (77)	31 (72)	
Black	14 (19)	5 (17)	9 (21)	
Asian	4 (6)	1 (3)	3 (7)	
Two or more races	1 (1)	1 (3)		
Hispanic ethnicity	38 (49)	16 (50)	22 (48)	0.85
Missing	1 (1)	1 (3)		
BMI (kg/m^2)	31.6 (26.0–36.2)	33.8 (26.0–38.0)	29.3 (25.5–35.1)	0.11
Missing	4 (5)	2 (6)	2 (4)	
Health insurance				
Missing	10 (13)	5 (15)	5 (11)	0.04
Government	34 (49)	19 (68)	15 (37)	
Private	19 (28)	5 (18)	14 (34)	
Uninsured	16 (23)	4 (14)	12 (29)	
Diabetes	29 (37)	13 (39)	16 (35)	0.67
Myocardial Infarction	4 (5)	1 (3)	3 (7)	0.49
Cardiac disease	7 (9)	3 (9)	4 (9)	0.95
Stroke	1 (1)	0	1 (2)	0.39
Hypertension	34 (43)	11 (33)	23 (50)	0.14
Chronic Obstructive pulmonary disease	12 (15)	7 (21)	5 (11)	0.21
Cancer	1 (1)	0	1(2)	0.39
Immunosuppression	2 (3)	1 (3)	1 (2)	0.81
Dementia	2 (3)	0	2 (4)	0.23
Hyperlipidemia	28 (35)	11 (33)	17 (37)	0.74
Number of comorbidities	2 (0–3)	2 (1–3)	2 (0–3)	0.82

Note: Categorical variables are expressed as *N* (%); numerical variables are expressed as median (interquartile range).

Abbreviations: BMI, body mass index; tPA, tissue plasminogen activator.

were highly variable between institutions and patients to achieve therapeutic partial thromboplastin time levels or anti-Xa levels. In-hospital mortality for the cohort was high at 58%.

Evaluation of the primary endpoint demonstrated that at 48 and 72 h post-tPA, there was a statistically significant increase in PaO_2/FiO_2 ratio relative to pre-tPA dosing (Figure 1A). Adjustment for confounders (age, sex, body mass index, hospital day when tPA was administered, mode of tPA administration, second tPA dose given, anticoagulation with heparin, remdesivir, and NEWS2, hemoglobin, and creatinine upon tPA administration) and testing of effect modification by the pre-tPA trend in PaO_2/FiO_3 (PaO_2/FiO_2 improving/stable vs PaO_2/FiO_2 declining) are shown

TABLE 2 Physiology, treatments, and outcomes of the studied patients

Variables	Total (*N* = 79)	Survivors (*N* = 33)	Nonsurvivors (*N* = 46)	*p* Value
Physiology upon tPA administration				
NEWS2	9 (7–11)	8 (6–10)	10 (7–12)	0.02
Systolic blood pressure (mm Hg)	111 (99–125)	115 (101–125)	110 (95–123)	0.25
Diastolic blood pressure (mm Hg)	60 (51–71)	61 (55–71)	59 (49–70)	0.26
Heart rate (beats/min)	93 (74–115)	75 (66–93)	104 (86–120)	0.00
Glasgow Coma Scale	3 (3–8)	3 (3–8)	3 (3–7)	0.33
Temperature (°C)	37.2 (36.6–38.0)	37.3 (36.7–37.8)	37.1 (36.4–38.1)	0.60
Richmond Agitation Sedation Scale	9 (6–10)	8 (7–9)	9 (6–10)	0.60
Vasopressor	40 (51)	13 (39)	27 (59)	0.09
PaO_2/FiO_2	91 (69–136)	105 (82–141)	82 (61–112)	0.02
Ventilatory ratio	1.7 (1.5–2.9)	1.5 (1.0–1.6)	2.5 (1.6–3.8)	<.01
Missing (%)	30 (38)	13 (39)	17 (37)	
Paralytics	29 (37)	12 (36)	17 (37)	0.96
Position				
Prone	25 (32)	8 (24)	17 (37)	0.54
Supine	48 (61)	22 (67)	26 (57)	
Left side	3 (4)	1 (3)	2 (4)	
Right side	3 (4)	2 (6)	1 (2)	
aPTT (s)	34 (30–46)	33 (30–42)	37 (29–48)	0.64
Missing (%)	26 (33)	11 (33)	15 (33)	
INR	1.2 (1.1–1.3)	1.1 (1.1–1.2)	1.2 (1.1–1.3)	0.13
Missing	27 (34)	11 (33)	16 (35)	
D-dimer (ng/ml)	3804 (1920–7565)	2860 (1791–5568)	4270 (2145–11981)	0.06
Missing	12 (15)	5 (15)	7 (15)	
Fibrinogen (mg/dl)	650 (468–760)	654 (536–819)	638 (434–755)	0.37
Missing	15 (19)	6 (18)	9 (20)	
Hemoglobin (g/dl)	11.6 (10.3–13.3)	12.5 (11.2–13.4)	11.4 (9.6–12.9)	0.11
Missing	5 (6)	3 (9)	2 (4)	
Platelet count ($\times 10^9$/L)	274 (171–392)	277 (212–395)	272 (163–368)	0.65
Missing	5 (6)	3 (9)	2 (4)	
Troponin (ng/ml)	0.1 (0.0–5.0)	0.5 (0.0–11.0)	0.1 (0.0–0.6)	0.55
Missing	46 (58)	14 (42)	32 (70)	
C-reactive protein (mg/L)	47.8 (14.9–131.7)	67.5 (19.6–131.6)	39.3 (11.8–131.8)	0.67
Missing	23 (29)	8 (24)	15 (33)	
Bilirubin (mg/dl)	0.7 (0.5–1.0)	0.6 (0.5–0.9)	0.7 (0.4–1.2)	0.52
Missing	19 (24)	8 (24)	11 (24)	
Creatinine (mg/dl)	1.2 (0.8–2.2)	0.9 (0.7–1.3)	1.4 (0.9–2.3)	0.05
Missing	6 (8)	3 (9)	3 (7)	
Remdesivir				
No	52 (68)	18 (55)	34 (79)	0.05
Post-tPA	11 (15)	8 (24)	3 (7)	
Pre-tPA	13 (17)	7 (21)	6 (14)	
Dexamethasone				
No	32 (41)	10 (30)	22 (48)	0.17

TABLE 2 (Continued)

Variables	Total (*N* = 79)	Survivors (*N* = 33)	Nonsurvivors (*N* = 46)	*p* Value
Post-tPA	23 (29)	13 (39)	10 (22)	
Pre-tPA	24 (30)	10 (30)	14 (30)	
tPA administration				
First tPA dose	50 (25–50)	50 (25–50)	50 (50–100)	0.06
Hospital day when first tPA was given	5 (2–8)	3 (2–8)	7 (3–10)	0.04
First tPA mode				
Bolus	30 (38)	14 (42)	16 (35)	0.00
Continuous Infusion	30 (38)	6 (18)	24 (52)	
Bolus + Continuous Infusion	19 (24)	13 (39)	6 (13)	
Second tPA	36 (46)	17 (52)	19 (41)	0.37
Hours between first and second tPA	11 (2–30)	2 (2–26)	25 (2–84)	0.06
Dose of second tPA	50 (25–50)	25 (25–50)	50 (25–50)	0.19
Second tPA mode				
Bolus	19 (53)	10 (59)	9 (47)	0.43
Continuous infusion	10 (28)	3 (18)	7 (37)	
Bolus + continuous infusion	7 (19)	4 (24)	3 (16)	
Required tPA to be stopped	2 (3)	0	2 (4)	0.23
Heparin given with tPA	67 (85)	32 (97)	35 (76)	0.01
Days on heparin	6 (3–10)	7 (5–12)	4 (2–10)	0.04
Ventilation days	11 (5–17)	11 (3–22)	10 (5–16)	1.00
VFD	0 (0–15)	17 (6–25)	0 (0–0)	<.01
Intensive care unit days	15 (9–24)	15 (8–30)	14 (9–21)	0.47
IFD	0 (0–9)	13 (0–20)	0 (0–0)	<.01
Hospital days	20 (13–34)	23 (18–46)	16 (12–28)	0.02

Note: Categorical variables are expressed as *N* (%); numerical variables are expressed as median (interquartile range).

Abbreviations: aPTT, activated partial thromboplastin time; IFD, intensive care unit-free days; INR, international normalized ratio of prothrombin time; NEWS2, National Early Warning Score-2; tPA, tissue plasminogen activator; VFD, ventilation-free days.

in Figure 1B. The pre-tPA trend in oxygenation significantly modified the temporal trend in PaO_2/FiO_2 (interaction term $p < 0.00$), with significant improvements at 24, 48, and 72 h post-tPA being observed in the group experiencing PaO_2/FiO_2 decline before tPA administration. In other words, tPA appeared to reverse the declining trend in pulmonary function and sustained the reversal up to 72 h after infusion. In contrast, patients with stable or upward oxygenation trends experienced nonsignificant peaks in PaO_2/FiO_2.

There were significant decreases in ventilatory ratio (a correlate of dead space ventilation) at 2, 6, 24, 48, and 72 h (Figure 2A). However, these improvements did not persist after adjustment for confounders (Figure 2B). The pre-tPA trends in ventilatory ratios did not significantly modify the temporal trends of this outcome. It should be noted that, distinct from other outcomes, there was a large proportion of missing data for ventilatory ratio (>30%), which may have affected these results.

Figure 3A depicts the significant improvement (decrease) in the NEWS2 at 12, 24, 48, and 72 h in the unadjusted model. Similar to PaO_2/FiO_2, the pretrend in the confounder-adjusted NEWS2 significantly modified (interaction term $p = 0.00$) the temporal trends of the score post-tPA (Figure 3B). Once again, tPA appeared to significantly reverse the downward trend in NEWS2 and sustain the improvement up to 72 h post-tPA. It should be noted, however, that oxygenation (as measured by peripheral pulse oximetry), respiratory rate, and requirement for oxygen support are components of the NEWS2 score (other components are systolic blood pressure, pulse, consciousness, and temperature).

Figure 4A shows that there was a large and statistically significant increase in D-dimer levels at 2 ($p = 0.00$), 6 ($p = 0.00$), and 12 h ($p = 0.00$) post-tPA relative to pre-tPA values, consistent with achieving clot lysis. Fibrinogen levels (Figure 4B) decreased significantly after tPA but remained at levels above 400 mg/dl in most patients. Only four (5%) patients had fibrinogen levels below 300 mg/dl at 72 h (all above 225 mg/dl).

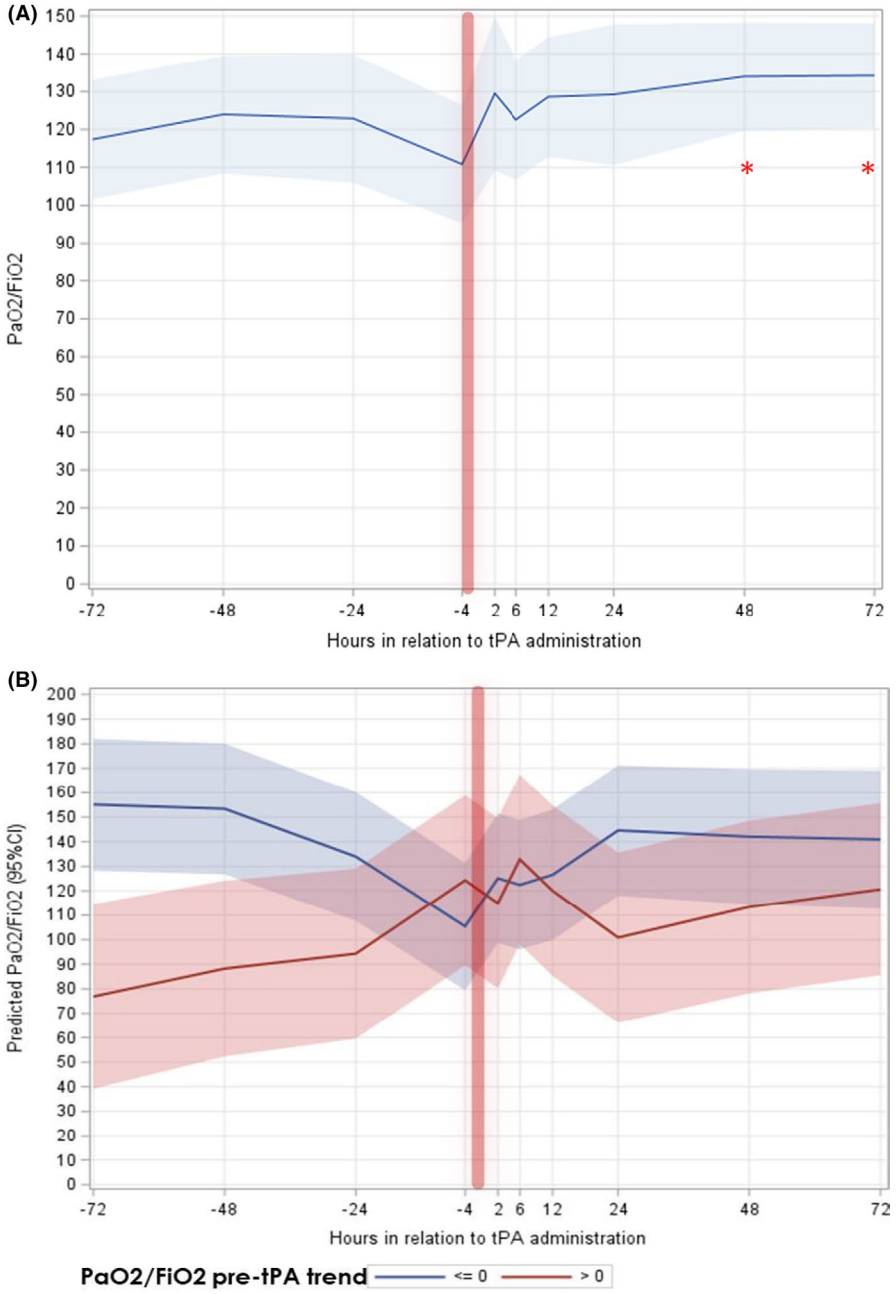

FIGURE 1 PaO_2/FiO_2 estimates over time. The value "-4" in the x-axis indicates the baseline PaO_2/FiO_2, collected 3–6 h before tPA was administered. The red bar marks the administration of tPA. (A) Unadjusted PaO_2/FiO_2; overall time effect $p = 0.02$, asterisks indicate significant differences compared with baseline. (B) PaO_2/FiO_2 estimates, adjusted for significant covariates (see text), stratified by the trend in pre-tPA PaO_2/FiO_2 (significant effect modifier interaction time × pre-tPA trend $p < 0.00$). Color-concordant asterisks indicate significant differences compared with baseline. Only the group with declining PaO_2/FiO_2 showed significant differences compared with baseline. tPA, tissue plasminogen activator

3.1 | Complications

Complications are described in Table 3, stratified by overall (over the entire admission) and those that occurred within 72 h of tPA administration. Overall, 47 (59.5%) patients presented at least one complication, whereas 25 (31.6%) patients presented complications within 72 h of receiving tPA. Bleeding complications were documented in 13 (16.5%) patients, and in nine patients, these bleeding episodes occurred within 72 h of tPA administration. There was one intracranial hemorrhage (ICH; 1.27%), which resulted in death. Of note, this patient received tPA on hospital day 47, much later than all other patients, and had no head imaging before the administration of tPA, raising concern that this event was a hemorrhagic conversion of an undiagnosed thrombotic stroke.

3.2 | Causes of death and death prediction

Table 4 depicts the causes of death as documented in the medical record. Most patients succumbed to lung failure, in isolation or accompanied by other organ dysfunctions.

4 | DISCUSSION

Fibrinolytic therapy for COVID-19 respiratory failure has been hypothesized to improve respiratory function in critically ill patients with tenuous respiratory status and a poor prognosis.[10,25] The results of this study provide evidence that fibrinolytic therapy (1) improves respiratory function and oxygenation in COVID-19 patients,

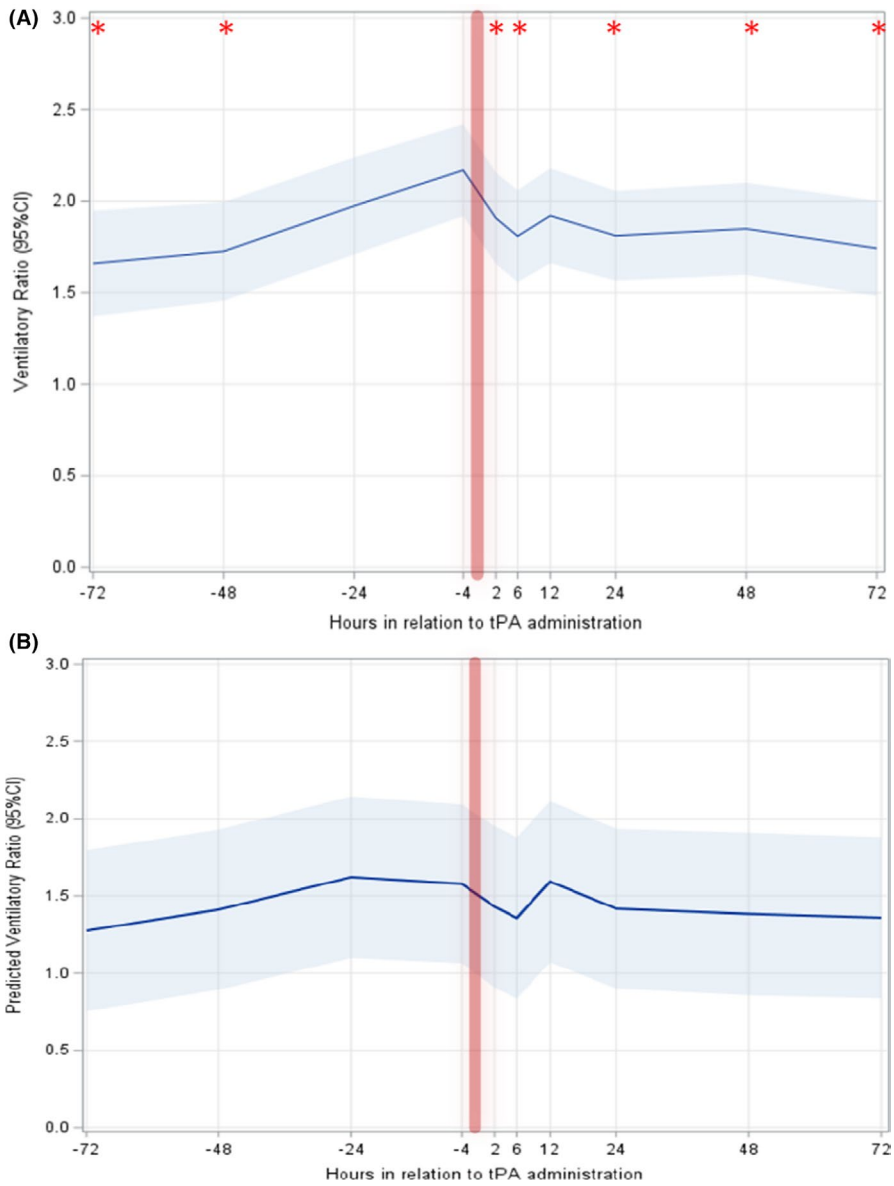

FIGURE 2 Ventilatory ratio (correlate of dead space) estimates over time. The value "-4" in the x-axis indicates the baseline ventilatory ratio, collected 3–6 h before tPA was administered. The red bar marks the administration of tPA. (A) Unadjusted ventilatory ratio; overall time effect $p = 0.00$, asterisks indicate significant differences compared with baseline. (B) Ventilatory ratio estimates after adjustment for significant covariates (see text) did not significantly change over time ($p = 0.16$) and the temporal trend was not modified by the pre-tPA trend in ventilatory ratio (interaction time × pre-tPA trend $p = 0.15$). tPA, tissue plasminogen activator

particularly those with rapidly declining respiratory status; and (2) that the safety profile is acceptable, particularly when considering the potential benefit in such a critically ill cohort with high mortality. The risk of ICH in this multi-institutional series was 1.3%, much smaller than those reported in recent reviews of the ICH risk associated with extracorporeal membrane oxygenation (ECMO), the next likely treatment intervention for these patients if the hospital has such extensive resources. Recent reports of ICH in ECMO for the treatment of COVID-19-associated respiratory failure varied from 6% in the Extracorporeal Life Support Organization registry (36 countries) to 9% in a New York, US, institution, and 12% in the report of the ECMO network for Greater Paris (17 intensive care units) to 33% in two US academic centers.[26-29] Further, it is unclear whether this complication could have been mitigated with pre-tPA neurologic examination or cross-sectional imaging of the brain (e.g., computed tomography; Table 4).

At the time this study was collecting data, there were only small case series[16,18,21] and two small retrospective studies in the published literature evaluating fibrinolytic therapy in COVID-19 respiratory failure, one found an apparent benefit but consisted of just 15 patients[20] whereas the other study by Douin et al. included 59 patients and found no benefit.[30] The latter study, the largest to date until the present report, assembled arterial blood gas data from patients by chronologic order (arterial blood gas 1, 2, 3) post-tPA rather than by synchronous times after tPA (e.g., 12, 24, 48 h), introducing substantial heterogeneity and margins of error that rendered to results difficult to interpret given the time dependency of thrombolysis and its results. Additionally, the Douin et al. study included patients who were being treated for known macroscopic PE and patients in the peri-arrest setting, which was not the study question. Our study was larger, contained highly granular data in 6-h increments for 72 h before and after tPA administration

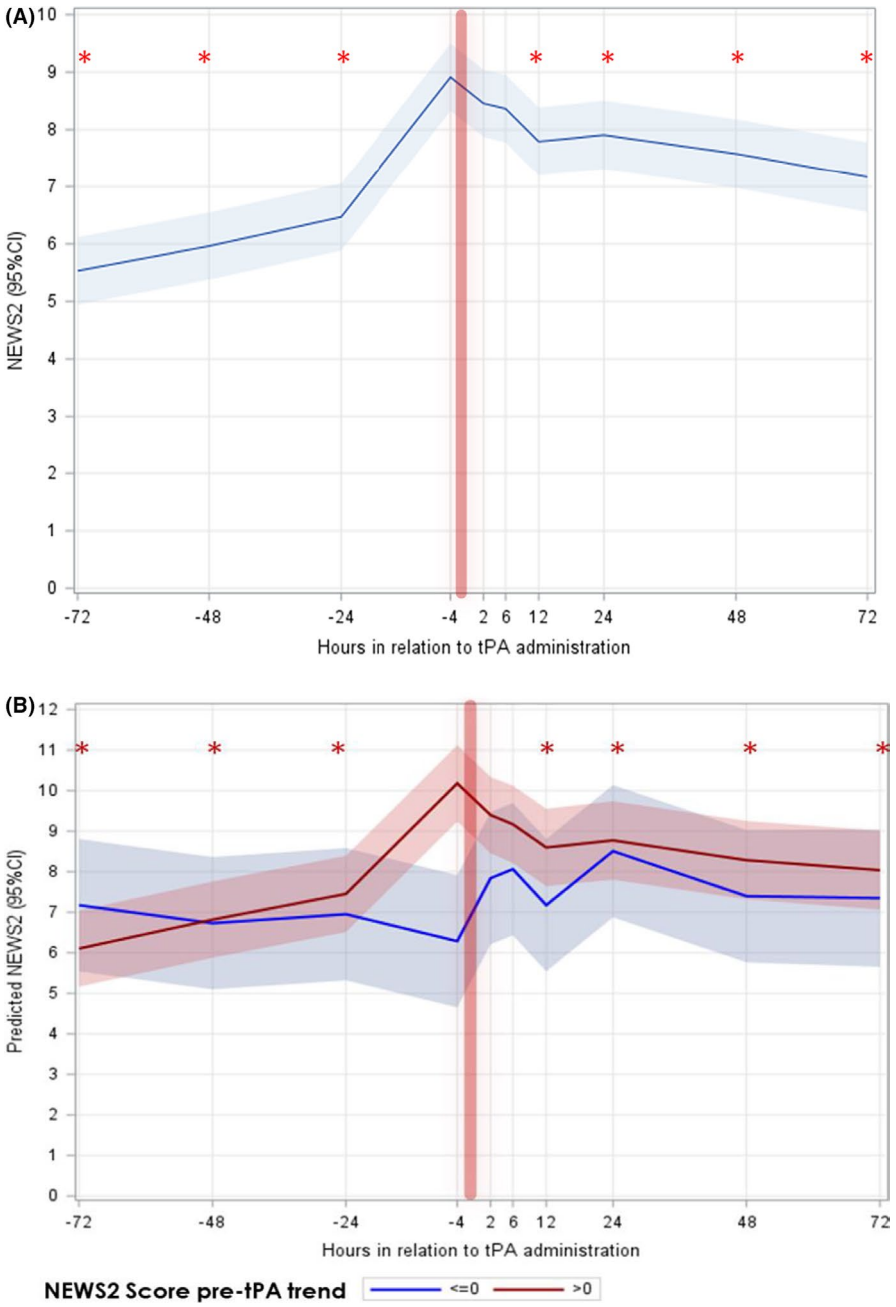

FIGURE 3 NEWS2 score estimates over time. The value "-4" in the x-axis indicates the baseline NEWS2 score, collected 3 to 6 hours before tPA was administered. The red bar marks the administration of tPA. (A) Unadjusted NEWS2 score; overall time effect $p < 0.00$, asterisks indicate significant differences compared with baseline. (B) NEWS2 score estimates after adjustment for significant covariates (see text), stratified by the trend in pre-tPA NEWS2 score (significant effect modifier interaction time × pre-tPA trend $p = 0.001$). Color-concordant asterisks indicate significant differences compared with baseline. Only the group with increasing NEWS2 score showed significant differences compared with baseline. NEWS2, National Early Warning Score 2; tPA, tissue plasminogen activator

collected with the explicit intent of testing our hypothesis, and used prespecified outcomes defined before data collection. Since then, the STARS multicenter randomized controlled trial has been published, which was a phase 2a study looking at different doses of tPA and heparin versus control for severe COVID-19 respiratory failure.[31] The STARS study showed significant improvements in oxygenation and a trend toward 12 more ventilator-free days and one-half the mortality in the tPA bolus with immediate therapeutic heparin group relative to controls. STARS was not powered for the latter outcomes and, importantly, had no severe or intracranial bleeding events in the tPA groups. The present study, with more than three times as many tPA patients, found similar and significant results in oxygenation improvements, further validating this earlier finding by the STARS study.

Our study has a number of limitations. First, it is retrospective, and therefore cannot be used in isolation to establish a causal relationship between tPA therapy and improved respiratory function with COVID-19 respiratory failure. Second, there was no control group, so analysis was limited to a pre-/post-tPA analysis. Third, although it is the largest study to date to address the topic of fibrinolytic therapy in COVID-19 respiratory failure, larger randomized controlled studies are needed to mitigate the effect of confounders. Additionally, there were multiple different tPA dosing strategies and anticoagulation strategies used, some of which may have influenced

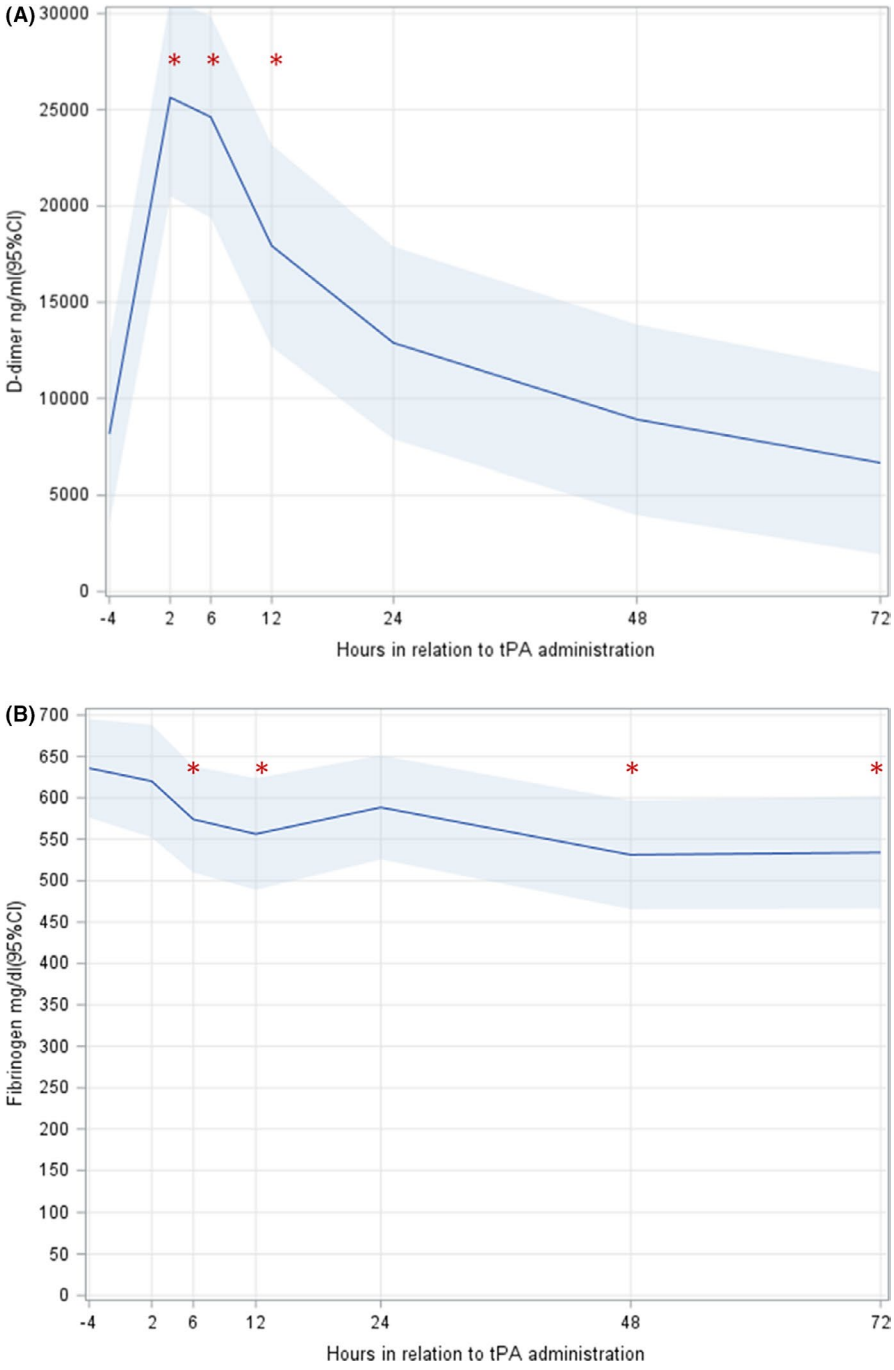

FIGURE 4 D-dimer and fibrinogen levels before and after tPA administration. (A) Unadjusted D-dimer values over time (time effect $p < 0.00$). The value "-4" in the x-axis indicates the baseline D-dimer, collected 3–6 h before tPA was administered. The red bar indicates the tPA administration. Asterisks denote significant differences compared with baseline. (B) Fibrinogen values over time (time effect $p = 0.02$). The value "-4" in the x-axis indicates the baseline D-dimer, collected 3–6 h before tPA was administered. The red bar indicates the tPA administration. Asterisks denote significant differences compared with baseline. tPA, tissue plasminogen activator

the observed results in ways we were unable to capture with the present data set. Finally, the practice patterns between institutions and over time as the pandemic evolved were inherently variable and may have had unknown effects on the observed outcomes, although we did control for both remdesivir and dexamethasone administration in our analysis. Despite these limitations, the results of our study can help enrich cohort selection for tPA administration in patients with worsening trajectories of hypoxemia. Further, it is likely that given the multiorgan involvement of COVID-19, survival benefit will require early administration before the onset of multiorgan involvement and the potential compounding negative effects of treatments such as deep sedation, paralytics and oxygen trauma, barotrauma, or volutrauma.

In summary, fibrinolytic therapy with tPA is associated with a significant improvement in respiratory function and oxygenation in severe COVID-19 respiratory failure. These findings were most pronounced in patients with ongoing decline in their respiratory status, rather than in a plateau or improving phase of poor respiratory status. The incidence of intracranial hemorrhage was low with only one occurrence in 79 patients (1.27%), lower than published rates of ICH in ECMO for COVID-19 ARDS. Further study is urgently needed.

TABLE 3 Complications: overall and complications occurring within 72 h of tPA administration

Overall complications	No. of events	No. of Patients	Patients with the event
Bleeding events	17	14	17.7%
Hematuria	4	1	1.3%
Vascular access	3	3	3.8%
Intracranial hemorrhage	1	1	1.3%
Gastrointestinal hemorrhage	2	2	2.5%
Oral bleeding	2	2	2.5%
Hemoptysis	1	1	1.3%
Intramuscular hematoma	1	1	1.3%
Perisplenic hematoma	1	1	1.3%
Other bleeding event	2	2	2.5%
Bacterial pneumonia or empyema	8	8	10.1%
Sepsis	6	6	7.6%
Renal failure	5	5	6.3%
Bacteremia	4	4	5.1%
Cardiac arrhythmia requiring treatment	4	4	5.1%
Hypotension	4	4	5.1%
Thrombocytopenia	4	4	5.1%
Anemia	3	1	1.3%
Cardiac arrest not resulting in death	2	1	1.3%
Heparin-induced thrombocytopenia	2	2	2.5%
Pulmonary embolism	2	2	2.5%
Acute worsening of lung function	2	2	2.5%
Deep venous thrombosis (includes vascular access thrombosis)	1	1	1.3%
Metabolic alkalosis	1	1	1.3%
Multiple organ failure	1	1	1.3%
Transaminitis	1	1	1.3%
Other complications	25	25	31.6%
Total number of events	93		
Total number of patients	47	59.5%	
Total number of bleeding events	18		
Total number of patients with bleeding events	13	16.5%	
Complications occurring within 72 h of the first dose of tPA	**No. of events**	**No. of Patients**	**Patients with the event**
Bleeding events	11	9	11.4%
Hematuria	3	1	1.3%
Vascular access	3	3	3.8%
Oral bleeding	2	2	2.5%
Gastrointestinal hemorrhage	1	1	1.3%
Hemoptysis	1	1	1.3%
Intracranial hemorrhage	1	1	1.3%
Renal failure	3	3	3.8%
Bacteremia	2	2	2.5%
Bacterial pneumonia or empyema	2	2	2.5%
Cardiac arrhythmia requiring treatment	2	1	1.3%
Sepsis	2	2	2.5%
Thrombocytopenia	2	2	2.5%

TABLE 3 (Continued)

Complications occurring within 72 h of the first dose of tPA	No. of events	No. of Patients	Patients with the event
Anemia	1	1	1.3%
Pulmonary embolism	1	1	1.3%
Transaminitis	1	1	1.3%
Other complications	6	6	7.6%
Total number of events	33		
Total number of patients	25	31.6%	
Total number of bleeding events	11		
Total number of patients with bleeding events	9	11.4%	

Abbreviation: tPA, tissue plasminogen activator.

TABLE 4 Causes of death as documented in the medical record

Documented causes of death	Frequency
Cardiac arrest	13
Multiple organ failure	9
Acute respiratory distress syndrome	8
Septic shock	6
Withdrawal of care for post-ARDS fibrosis	3
Intracranial hemorrhage	2
Complete heart block	1
Diffuse thromboembolic strokes with hemorrhagic conversion	1
Hypoxic brain injury after cardiac arrest and brain hemorrhage	1
Unknown	1
Withdrawal of care because of multiple organ failure	1

Abbreviation: ARDS, acute respiratory distress syndrome.

RELATIONSHIP DISCLOSURE

C.D.B., H.B.M., E.E.M., and M.B.Y. have patents pending related to both coagulation/fibrinolysis diagnostics and therapeutic fibrinolytics and are passive cofounders and hold stock options in Thrombo Therapeutics, Inc. H.B.M. and E.E.M. have received grant support from Haemonetics and Instrumentation Laboratories. M.B.Y. has previously received a gift of alteplase (tPA) from Genentech, and owns stock options as a cofounder of Merrimack Pharmaceuticals. C.D.B., H.B.M., E.E.M., J.W., N.H., D.S.T., A.S., and M.B.Y. have received research grant funding from Genentech. All other authors have nothing to disclose. Salary support for M.B.Y. was provided by National Institutes of Health grant ES028374 and an anonymous donation for COVID-related research to MIT.

AUTHOR CONTRIBUTIONS

Christopher D. Barrett, Hunter B. Moore, Ernest E. Moore, Ammar Al-Shammaa, and Michael B. Yaffe had access to all data and contributed to all components of the study and manuscript generation. All other authors were involved in data collection, project administration, supervision, conceptualization, investigation, review, and/or editing.

ORCID

Christopher D. Barrett https://orcid.org/0000-0001-9720-8155
Valerie Banner-Goodspeed https://orcid.org/0000-0002-7644-2521

TWITTER

Christopher D. Barrett @ChrisBarrettMD
Michael B. Yaffe @mbyaffe

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SUPPORTING INFORMATION

Additional supporting information may be found in the online version of the article at the publisher's website.

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