Use of defibrotide in COVID-19 pneumonia: comparison of a phase II study and a matched real-world cohort control

Annalisa Ruggeri,[1*] Francesco Corrado,[2,3*] Antonio Voza,[2,4] Lee-Jen Wei,[5] Gloria Catalano,[1] Carmine Liberatore,[1] Rosamaria Nitti,[1] Carlo Fedeli,[4] Alessandro Bruno,[1] Eleonora Calabretta,[2,3] Fabio Giglio,[1] Fabio Sciutti,[6] Francesca Lunghi,[1] Giovanni Landoni,[7,8] Alessio Aghemo,[2,9] Massimo Iacobelli,[10] Patrizia Rovere Querini,[8,11] Paul G. Richardson,[12] Andrea Assanelli,[1] Jacopo Peccatori,[1] Fabio Ciceri[1,9#] and Carmelo Carlo-Stella[2,3#]

[1]Hematology and Bone Marrow Transplantation Unit, IRCCS San Raffaele Scientific Institute, Milan, Italy; [2]Department of Biomedical Sciences, Humanitas University, Pieve Emanuele, Milan, Italy; [3]Department of Oncology and Hematology, IRCCS Humanitas Research Hospital, Milan, Italy; [4]Emergency Department, IRCCS Humanitas Research Hospital, Milan, Italy; [5]Department of Biostatistics, Harvard T.H. Chan School of Public Health, Harvard University, Boston, MA, USA; [6]Emergency Department, IRCCS San Matteo, Pavia, Italy; [7]Anesthesia and Intensive Care Department, IRCCS San Raffaele Scientific Institute, Milan, Italy; [8]School of Medicine, Vita-Salute San Raffaele University, Milan, Italy; [9]Division of Internal Medicine and Hepatology, Department of Gastroenterology, IRCCS Humanitas Research Hospital, Milan, Italy; [10]Life Sciences, Techitra Srl, Milan, Italy; [11]Department of Immunology, Transplantation and Infectious Diseases, IRCCS San Raffaele Scientific Institute, Milan, Italy and [12]Dana-Farber/Brigham and Women's Cancer Center and Harvard Medical School, Boston, MA, USA

*AR and FCo contributed equally as first authors.
#FCi and CC-S contributed equally as senior authors.

Correspondence: C. Carlo-Stella
carmelo.carlostella@hunimed.eu

Received: February 27, 2024.
Accepted: May 13, 2024.
Early view: May 23, 2024.

https://doi.org/10.3324/haematol.2024.285345

Abstract

The coronavirus disease 2019 (COVID-19) pandemic led to an unprecedented burden on healthcare systems around the world and a severe global socioeconomic crisis, with more than 750 million confirmed cases and at least 7 million deaths reported by December 31, 2023. The DEFI-VID19 study (*clinicaltrials gov. Identifier: NCT04335201*), a phase II, single-arm, multicenter, open-label trial was designed in mid-2020 to assess the safety and efficacy of defibrotide in treating patients with COVID-19 pneumonia. Defibrotide was administered at a dose of 25 mg/kg intravenously, divided into four daily doses over a planned 14-day period for patients with COVID-19 pneumonia receiving non-invasive ventilation. The primary endpoint was respiratory failure-free survival (RFFS). Overall survival (OS), the number of post-recovery days, and adverse events were the secondary endpoints. For comparison, a contemporaneous control cohort receiving standard of care only was retrospectively selected by applying the eligibility criteria of the DEFI-VID19 trial. To adjust for the imbalance between the two cohorts in terms of baseline variable distributions, an outcome regression analysis was conducted. In adjusted analysis, patients receiving defibrotide reported a trend towards higher RFFS (hazard ratio [HR]=0.71; 95% confidence interval [CI]: 0.34-1.29; *P*=0.138) and OS (HR=0.78; 95% CI: 0.33-1.53; *P*=0.248]) and showed a significantly increased number of post-recovery days (difference in means =3.61; 95% CI: 0.97-6.26; *P*=0.0037). Despite concomitant thromboprophylaxis with low molecular weight heparin, the safety profile of defibrotide proved to be favorable. Taken together, our findings suggest that defibrotide may represent a valuable addition to the COVID-19 therapeutic options.

Introduction

Coronavirus disease 2019 (COVID-19) pandemic, the global outbreak of a viral illness caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), led to an unprecedented burden on healthcare systems around the world and a severe global socioeconomic crisis. On January 30, 2020 COVID-19 was declared a Public Health Emergency of International Concern (PHEIC) with an official death toll of less than 200. By August 1, 2023, there had been more than 750 million confirmed cases of COVID-19, including at least 7 million deaths reported by the World Health Organization

(WHO).[1] The current epidemiological landscape reflects the implementation of extensive policies of vaccination and the approval of a limited number of therapeutic agents. The adoption of vaccination policies is estimated to have prevented nearly 14 million deaths from COVID-19 between December 8, 2020, and December 8, 2021.[2] Baricitinib,[3] interleukin- 6 (IL-6) receptor blockers,[4] and corticosteroids[5] emerged as the main drugs demonstrating a reduction in mortality among patients with severe COVID-19 pneumonia. Conversely, remdesivir has shown the potential to expedite the time to recovery in patients with mild to severe cases of COVID-19 pneumonia, but without a clear survival advantage.[6] The DEFI-VID19 clinical trial was designed in mid-2020, at a time when an effective vaccine was not yet available, and COVID-19 presented as an uncontrolled, devastating, and widespread disease. In the absence of established therapeutic strategies, the mainstay of COVID-19 management was supportive care, with acute respiratory distress syndrome (ARDS) as the primary cause of mortality. COVID-19-related ARDS is uniquely associated with distinct pathobiological pathways, with angiocentric inflammation, microangiopathy, cytokine release syndrome and hypercoagulation emerging as the primary factors influencing the severity of the disease.[7–10] Moreover, specific inflammatory biomarkers, such as ferritin and IL-6 have been found to be independent predictors of fatality in COVID-19[9] further suggesting the central role of endotheliitis in the pathophysiology of the disease. The endothelial damage and cytokine profile associated with severe COVID-19 pneumonia suggested significant parallels with the pathophysiology of veno-occlusive disease (VOD), also referred to as sinusoidal obstruction syndrome (SOS). VOD/SOS is a syndrome characterized by endothelial damage to hepatic sinusoids and subsequent hepatocellular injury with subsequent necrosis, which is most commonly seen after bone marrow transplantation. Both conditions involve reactive endothelial activation, pro-inflammatory responses, cytokine release, progressive endotheliopathy and multi-organ dysfunction. The pathological similarities extend further, with lung lesions in VOD/SOS exhibiting features akin to those observed in COVID-19 patients.[13,14] Defibrotide is a polydisperse mixture of porcine-derived single-stranded oligonucleotides that was approved by the Food and Drug Administration in March 2016 for the treatment of hepatic VOD/SOS with either pulmonary or renal dysfunction after hematopoietic cell transplantation (HCT).[15] Defibrotide has demonstrated profibrinolytic, antithrombotic, anti-inflammatory, and angio-protective properties.[16] Given its potential therapeutic effects and the similarities between VOD/SOS and COVID-19, the DEFI-VID19 study was designed to evaluate the safety and efficacy of defibrotide in treating SARS-CoV-2-related ARDS, as part of several international studies evaluating the role of defibrotide in this setting and the underlying hypotheses of ameliorating endotheliitis, modulating cytokine release and reversing microangiopathy.[17–19] The DEFI-VID19 study was conceived as a phase II, single-arm trial. Subsequently, a comparison group was retrospectively built by selecting an observational cohort of patients who received add abbreviation (SOC) alone.

Methods

DEFI-VID19 was a phase II, prospective, interventional, single-arm, multicenter, open-label trial conducted at IRCCS-San Raffaele Scientific Institute (Milan), IRCCS-Humanitas Research Hospital (Milan), IRCCS-Policlinico San Matteo (Pavia). Approval was provided by the national institutional review board for COVID-19 trials at Institute Spallanzani (Rome) and by the Italian Agency for Drug (AIFA). All patients provided written informed consent. The trial was registered at *clinicaltrials gov. Identifier: NCT04335201*.

DEFI-VID19 eligibility criteria

Inclusion criteria included: documented COVID-19 pneumonia defined as a positive real-time reverse transcription polymerase chain reaction from upper respiratory tract specimens (nasopharyngeal or throat swabs) and/or a computed tomography image suggestive of COVID-19 pneumonia; oxygen saturation (SaO_2) of 92% or less without oxygen support, or reduction of 3% from the basal value of SaO_2, or a ratio of the partial pressure of oxygen (PaO_2) to the fraction of inspired oxygen (FiO_2) (PaO_2/FiO_2) below 300; any sex; age >18 years; written informed consent. Exclusion criteria included onset of COVID-19 pneumonia >14 days; orotracheal intubation; uncontrolled systemic infections (other than COVID-19); concomitant use of thrombolytic therapy; concomitant systemic anticoagulant therapy (e.g., heparin, warfarin, direct thrombin inhibitors, and direct factor Xa inhibitors); hemodynamic instability; hypersensitivity to the active substance or to any of the excipients of the experimental drug; patients who, based on the investigator's clinical judgment, were not able to receive the treatment; pregnancy or breast feeding.

DEFI-VID19 study design

Defibrotide was administered at a dose of 25 mg/kg intravenously fractionated in four doses daily for a planned treatment period of 14 days. Details are provided in the *Online Supplementary Appendix*.

Observational data selection criteria

A contemporary cohort of patients admitted to the IRCCS – Humanitas Research Hospital with COVID-19 pneumonia was retrospectively screened for meeting the eligibility criteria of the DEFI-VID19 trial. After applying the eligibility criteria, 153 patients qualified as controls.

Endpoints

The primary endpoint was the respiratory failure-free survival (RFFS) defined as the time from day 1 until respiratory

failure. Respiratory failure was defined as follows: intensive care unit (ICU) admission or death within day 14; the persistence of respiratory distress despite oxygen therapy at day 14 and for patients with a day 1 P/F ratio >200: a P/F ratio <200 at day 14. Respiratory distress was operationally defined as a respiratory rate >24/min. The number of post-recovery days was defined as the number of days after hospital discharge out of a predefined time window of 28 days from day 1.[22] A value of zero post-recovery days was assigned to patients who were still hospitalized or died before day 28.
Secondary endpoints and biological markers evaluated are indicated in the *Online Supplementary Appendix*.

Statistical methods

Statistical analyses were conducted using GraphPad Prism (version7.5) and R version 4.2.2. To account for the potential imbalance between the two cohorts in terms of baseline variable distributions, an outcome regression analysis was conducted.[23] Specifically, as previously described by Richardson *et al.*,[24] a survival Cox prediction model was developed using the data from the observational cohort. A set of clinically relevant baseline covariates was selected.[25]

Results

Patient characteristics

Overall, 52 patients were enrolled in the DEFI-VID19 trial from September 2020 to April 2021; 48 were evaluated for efficacy and safety; four patients were excluded due to screen failure (N=2) or withdrawal of informed consent at day 2 after defibrotide was initiated (N=2). The distribution of patient baseline characteristics according to defibrotide exposure is shown in Table 1. DEFI-VID19 patients were younger than patients who did not receive defibrotide (median, 60.5 *vs.* 72.7 years old), had fewer cardiovascular diseases (6.2% *vs.* 21.6%), hypertension (33.3% *vs.* 54.9%), and higher $PaO_2:FiO_2$ ratio at baseline (210.5 *vs.* 164). Median values of C-reactive protein (mg/dL), D-dimer (ng/mL), Ferritin (ng/mL), IL-6 (pg/mL), lactate dehydrogenase (UI/L) and platelet counts ($x10^9/L$) for the DEFI-VID19 cohort are reported in Table 1. At study entry, all treated and untreated patients received oxygen support either through a high-flow nasal cannula (HFNC) or continuous positive air pressure (CPAP) and were assigned a score of 5 on the WHO ordinal scale for clinical activity (Table 2). The median time from the diagnosis to the start of defibrotide was 4 days (range, 1-11). Therapy duration ranged from 1 to 14 days (median 8 days). Fourty-one

Table 1. Patient characteristics.

Characteristics	DEFI-VID 19 cohort	Observational cohort	*P*
Patient N	48	153	-
Age in years, median (Q1-Q3)	60.5 (53.75-70.25)	72.7 (62.6-79.7)	<0.001
Male sex, N (%)	35 (72.9)	109 (71.2)	0.97
BMI median (Q1-Q3)	26.9 (25.00-29.06)	27 (24.6-29.7)	0.79
Comorbidities, median (Q1-Q3]	0.5 (0.0-1.0)	1.0 (0.0-2.0)	0.04
Cardiovascular diseases, N (%)	3 (6.2)	33 (21.6)	0.03
Hypertension, N (%)	16 (33.3)	84 (54.9)	0.015
Diabetes, N (%)	7 (14.6)	32 (20.9)	0.3
Malignancies, N (%)	2 (4.2)	14 (9.2)	0.42
Respiratory diseases, N (%)	5 (10.4)	18 (11.8)	1.0
WHO score day 1, score 5, N (%)	48 (100)	153 (100.0)	1.0
P/F ratio day 1, median (Q1-Q3)	210.5 (135.2-272.5)	164.0 (125.0-231.0)	0.08
C-reactive protein mg/dL, median (Q1-Q3)	4.9 (2.5-11.0)	-	-
D-Dimer ng/mL, median (Q1-Q3)	450.0 (285.5-1,337.7)	-	-
Ferritin ng/mL, median (Q1-Q3)	854.5 (496.7-1,173.5)	-	-
IL-6 pg/mL, median (Q1-Q3)	21.3 (11.0-33.2)	-	-
Platelets count $x10^9/L$, median (Q1-Q3)	231 (191-336)	-	-
Lactate dehydrogenase IU/L, median (Q1-Q3)	415.0 (294.0-509.0)	-	-

WHO: World Health Organization; BMI: body mass index; IL-6: interleukin-6; Q1-Q3: interquartile range; N: number; P/F ratio: ratio of the partial pressure of oxygen to the fraction of inspired oxygen.

patients received less than 14 days of treatment because of the independence from oxygen therapy or discharge before day 14 (N=29), disease progression(death [N=3] or the need for invasive ventilation [N=5]) or the requirement of full anticoagulant therapy (N=4). No other COVID-19 experimental therapy, including monoclonal antibodies, was given to patients while they received defibrotide. Every patient in the study group received dexamethasone (6 mg/dose) and antiviral therapy according to the current local guidelines. All DEFI-VID19 patients also received thromboprophylaxis with low molecular weight heparin (LMWH) according to each institution's standards of care.

DEFI-VID19 safety

Forty-eight patients were evaluated for safety. The infusions were generally well tolerated, with no infusion-related reactions reported. No significant hemorrhagic or bleeding episodes occurred during study therapy. Thirteen patients (27.08%; 95% CI: 16.6-41) developed a serious adverse event (SAE). Twelve (18.7%; 95% CI: 10.2-32) developed respiratory failure, which was associated with death for seven patients. Four patients (8.3%; 95% CI: 3.3-19.55) developed thrombotic events that lead to the initiation of a full anticoagulant therapy and withdrawal of defibrotide treatment per protocol. One thrombotic event, a pulmonary embolism, proved to be fatal, despite prophylactic LMWH. However, no serious adverse event was deemed to be associated with or related to the administration of defibrotide, and importantly no bleeding with concomitant LMWH was reported. The median time of onset to SAE was 6 days (range, 2-12).

Respiratory failure-free survival

RFFS rate at day 14 was 0.75 (95% CI: 0.68-0.88) in the DEFI-VID19 cohort and 0.50 (95% CI: 0.43-0.59) in the observational cohort. In unadjusted analysis, the RFFS was significantly higher in DEFI-VID19 patients (HR=0.43; 95% CI: 0.23-0.79; *P*=0.005) (Figure 1).

In the DEFI-VID19 cohort, all respiratory failure events were represented by death or ICU admission before day 14 regardless of baseline P/F ratio. Within the observational cohort, 68 of 76 cases of respiratory failure were characterized by death or ICU admission before day 14, or by a deterioration of the P/F ratio from over 200 on day 1 to below 200 by day 14. The remaining eight cases were marked by persistent respiratory distress at day 14 despite oxygen therapy. Notably, among these eight cases, seven had a baseline P/F ratio below 200, which persisted below 200 after 14 days. In all these instances, respiratory rates were confirmed to be above 24 in at least two measurements throughout the day.

An outcome regression model was built with data from the observational cohort using a standard statistical variable selection process. For the overall analysis, the variables selected to be adjusted in the prediction model were age (*P*=0.0004) baseline P/F ratio (*P*=0.078) and the presence of malignancies (*P*=0.079), and respiratory disease (*P*=0.074) (*Online Supplementary Table S1A*). The C-index for this fitted model was C=0.67 (95% CI: 0.59-0.75). Adjusted HR for RFFS between the DEFI-VID19 population and SOC cohorts was HR=0.71; 95% CI: 0.34-1.29; *P*=0.138. Therefore, after adjusting for potentially confounding baseline covariates, we observed a tendency towards a higher RFFS in the DEFI-VID19 group (Figure 2).

In the DEFI-VID 19 cohort the median day 1 D-dimer value was of 450 ng/mL (Q1-Q3, 285.5-1,33.7), with no significant difference between patients who developed respiratory failure and those who did not (median 515 *vs.* 441 ng/mL; *P*=0.26). However, at the end of treatment, patients in respiratory failure showed significantly higher values of D-dimer compared to the responding patients (median 2,632.5 *vs* 380 ng/mL; *P*=0.0005).

Overall survival

The overall survival (OS) of patients enrolled in the DEFI-VID19 trial was 0.90 (95% CI: 0.81-0.98) at day 28 and 0.83 (95% CI: 0.73-0.95) at day 60. OS of patients receiving only SOC was 0.60 (95% CI: 0.53-0.68) at day 28 and 0.59 (95% CI: 0.52-0.68) at day 60. In unadjusted analysis, OS was significantly higher in patients treated with defibrot-

Table 2. World Health Organization ordinal scale for clinical activity.

Clinical status	Activity and respiratory support	Ordinal score
Ambulatory	No limitation of activity Activities limited	1 2
Hospitalized	No oxygen therapy Oxygen by mask or nasal cannula Non-invasive ventilation or high-flow oxygen Intubation or mechanical ventilation Mechanical ventilation plus one of the following: pressors, ECMO, or dialysis	3 4 5 6 7
Deceased	Death	8

ECMO: extracorporeal membrane oxygenation.

ide (HR=0.33; 95% CI: 0.16-0.69; *P*=0.003) (Figure 1). The variables selected to be adjusted in the survival prediction model were age (*P*<0.0001), baseline P/F ratio (*P*=0.034), the presence of malignancies (*P*=0.009), and respiratory diseases (*P*=0.0007) (*Online Supplementary Figure S1B*). The C-index for this fitted model was 0.81 (95% CI: 0.76-0.87). Adjusted HR for OS between the DEFI-VID19 population and SOC cohorts was HR=0.78; 95% CI: 0.33-0.53; *P*=0.248.

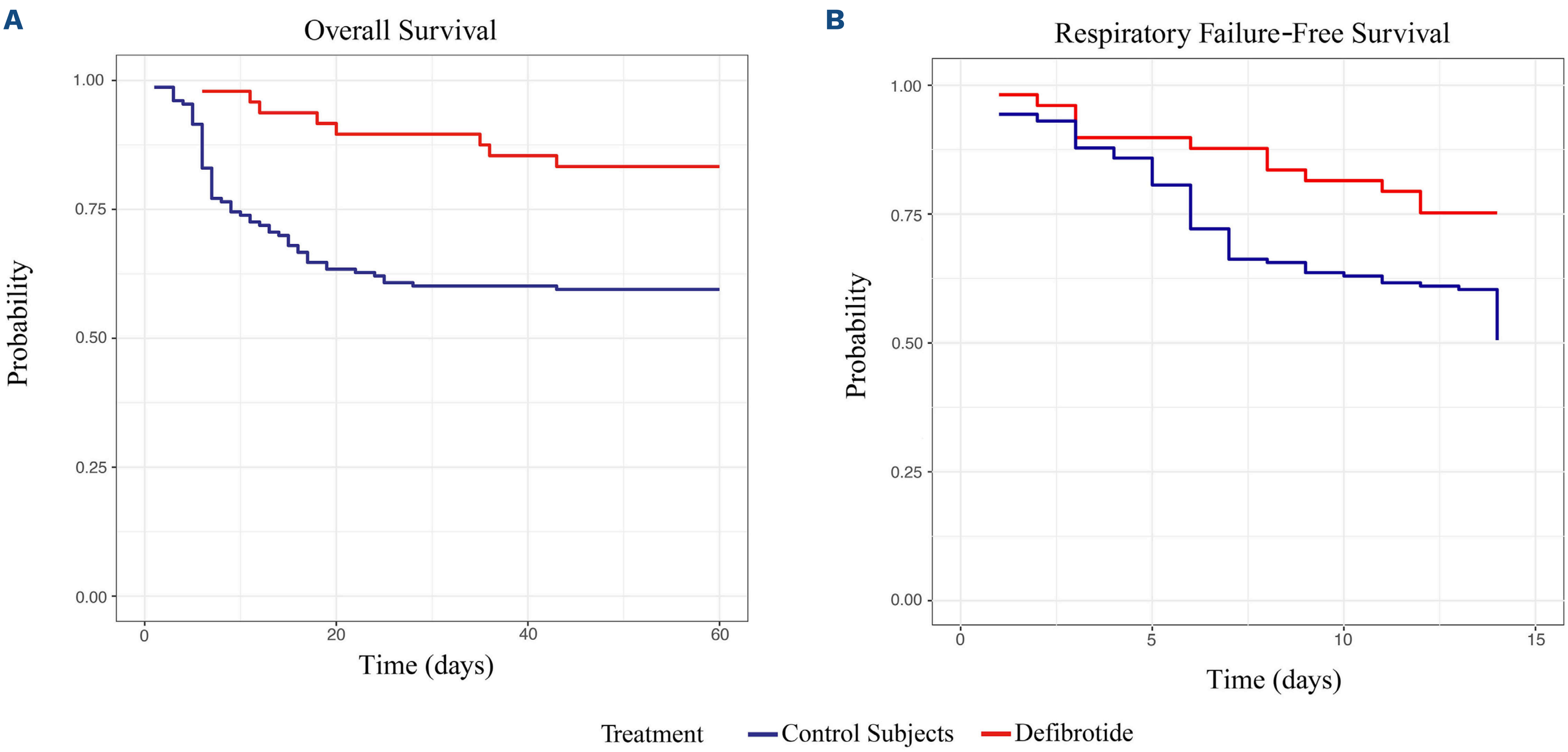

Figure 1. Probability of survival of patients receiving defibrotide. (A) Probability of overall survival (OS) and (B) unadjusted respiratory failure-free survival (RFFS) of patients receiving defibrotide and control group. Unadjusted Kaplan-Meier curves for RFFS and OS show significantly better outcomes for patients receiving defibrotide in addition to standard of care.

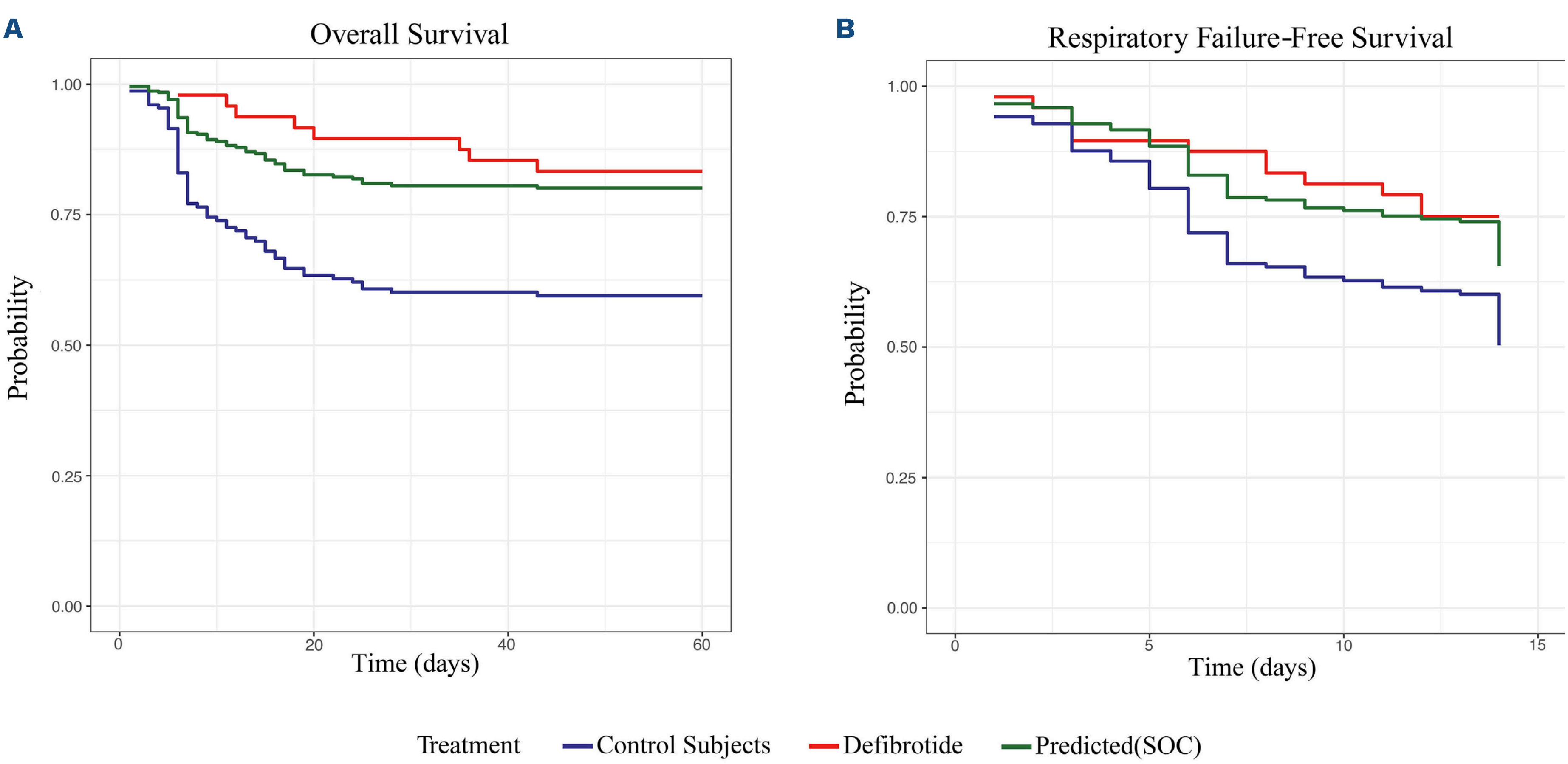

Figure 2. Probability of survival of patients receiving defibrotide. (A) Probability of overall survival and (B) unadjusted respiratory failure-free survival (RFFS) of patients receiving defibrotide and control group. After adjusting for potentially confounding baseline covariates, DEFI-VID19 patients exhibited a trend towards an increased RFFS (hazard ratio [HR]=0.71; 95% confidence interval [CI]: 0.34-1.29; *P*=0.138) and OS (HR=0.78; 95% CI: 0.33-1.53; *P*=0.248). SOC: standard of care.

Therefore, in the adjusted analysis we noted a trend towards an increased OS in the DEFI-VID19 cohort (Figure2).

Post-recovery days

DEFI-VID19 patients showed a mean number of post-recovery days of 11.60 (95% CI: 9.25-13.96) while patients from the observational cohort had a mean of 5.29 post-recovery days (95% CI: 4.19-6.39). In unadjusted analysis, the difference in means was statistically significant (6.32; 95% CI: 3,94-8,69; *P*<0.0001) (Figure 3). A linear prediction model was built with data from the observational cohort and the variables selected to be adjusted in the prediction model were age (*P*<0.0001), the presence of malignancies (*P*=0.026), and respiratory diseases (*P*=0.002) (*Online Supplementary Figure S1C*). The mean absolute error (MAE) for this fitted model was 4.32. By entering these covariates values for each DEFI-VID19 patient we obtained individual predicted values of post-recovery days. The mean value of predicted post-recovery days was 7.99 (95% CI: 6.94-9.05). The adjusted analysis confirmed that DEFI-VID19 patients had a significantly higher number of post-recovery days compared to patients receiving SOC, with a difference in means of 3.61 (95% CI: 0.97-6.26; *P*=0.0037] (Figure 3). Of note, at the time of discharge, patients treated with defibrotide showed no limitation in activities and did not require home oxygen therapy (WHO score of 1).

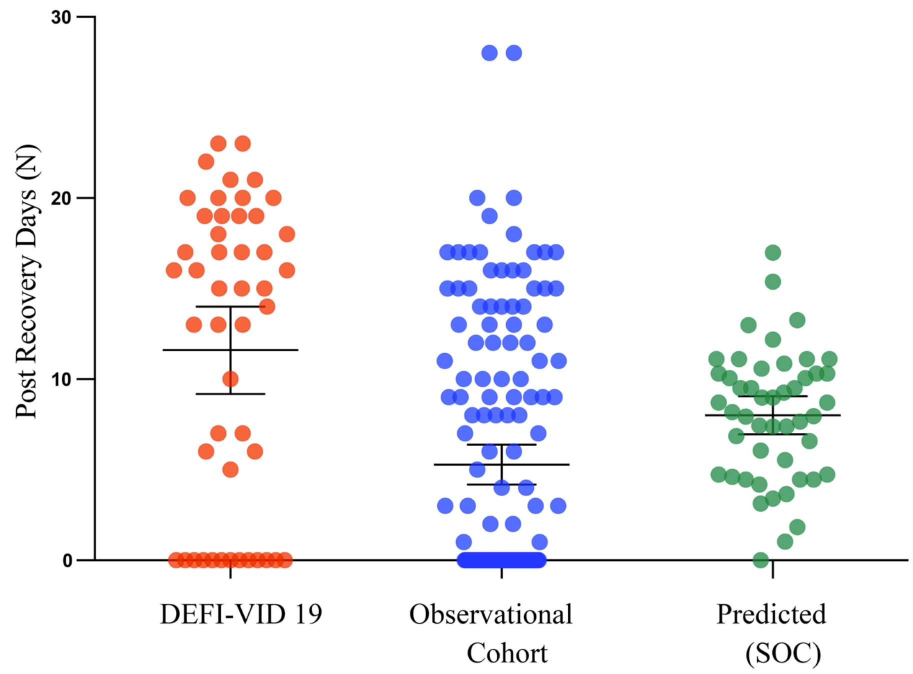

Figure 3. Observed and predicted numbers of post-recovery days. Mean values and 95% confidence intervals (CI) are shown in the plot. DEFI-VID19 patients (red dots) showed a significantly higher number of post-recovery days after adjusting for potentially confounding baseline covariates (green dots; difference in means =3.61; 95% CI: 0.97-6.26; *P*=0.0037); SOC: standard of care.

Discussion

We report the first study on the safety and efficacy of defibrotide therapy in patients with severe COVID-19 pneumonia who were receiving non-invasive ventilation. The results from the phase-II DEFI-VID19 trial were promising, demonstrating a favorable safety profile, a day-14 RFFS of 75%, a 60-day OS of 83%, and a mean number of post-recovery days of 12. Acknowledging that comparison between studies is challenging because of potential differences in patient characteristics and across different hospital systems, the comparison of DEFI-VID 19 results with a carefully selected observational cohort of patients who received SOC proved informative. Specifically, using data from patients who were treated for COVID-19 pneumonia during the same period as those enrolled in the DEFI-VID19 study allowed us to minimize various confounders and biases that could have been introduced because of changes in patient management over time. Additionally, we screened healthcare records excluding patients who did not receive CPAP and/or HFNO and then selected potential control patients based on the DEFI-VID19 eligibility criteria. Nevertheless, despite these measures, the 153 control patients were older, had more cardiovascular comorbidities and a lower PaO_2:FiO_2 ratio at baseline. One may argue that the controls were selected from one center, however more than 95% of patients enrolled into the study came from two centers in the same city, with common strategies in patients management and care. In order to correct for additional confounding factors, a tailored, model-based approach was used, as previously described by Torbicki *et al.*[23] Other novel statistical methods employed for making adjustments of baseline covariates, such as propensity score matching, inverse probability of treatment weighting (IPTW), and the use of double robustness were considered.[26] However, the outcome regression employed in our study appeared to be the most intuitive procedure based on the fundamental regression analysis prediction principle.[23,24]
We are aware of some limitation of our study namely that the ICU transfer could be considered as a subjective parameter, however due to the unprecedent situation during the COVID pandemic ICU admission was considered clinically relevant according to Italian policy.
Moreover, the propensity score and IPTW methods proved ineffective, as the score sets from the two groups of patients varied significantly. Overall, after adjusting for potential confounding baseline variables, we found that the DEFI-VID19 cohort exhibited a trend towards increased RFFS (HR=0.71; *P*=0.138) and OS (HR=0.78; *P*=0.248) when compared to patients receiving SOC only. Furthermore, there was a clear advantage in terms of time to recovery, as indicated by a difference in means of post-recovery days of 3.61 (95% CI: 0.97-6.26; *P*=0.0037). Notably, at the time of discharge, patients treated with defibrotide showed no limitation in activities and did not require home oxygen therapy (i.e., a WHO score of 1). In another study involving 13 critically ill patients with COVID-19-associated ARDS who were receiving invasive ventilation, the administration of defibrotide was found to be safe,

and no instances of hemorrhagic events were recorded,[27] with improved survival over that expected in such a sick population. It is worth noting that only three patients in this US study received prophylactic low molecular weight heparin (LMWH), whereas all patients in the DEFI-VID19 trial received prophylaxis with LMWH. Despite this difference in LMWH administration, no bleeding was observed in our DEFI-VID19 patient group, supporting the observed safety of defibrotide in this setting. In addition, due to its favorable pharmacokinetic profile, defibrotide is not contraindicated in patients with end-stage renal disease[28] or liver dysfunction,[29] in contrast to other approved drugs for severe COVID-19 pneumonia, such as baricitinib, remdesivir, and tocilizumab.

Accumulating evidence supports the central role of lung microvascular endothelial damage in the pathophysiology of COVID-19 related ARDS.[7,8,10] As a result of its angio-protective properties,[16,17-19] the early administration of defibrotide has the potential to mitigate Sars-Cov-2-induced endothelial injury and improve clinical outcomes, as illustrated in this study. Specifically, defibrotide may represent a valuable addition to the COVID-19 therapeutic options based upon our results and others.[27] Clinical benefit was reflected in our trial by its positive effect on reducing the time to discharge in patients with COVID-19 pneumonia receiving non-invasive ventilation, an improved OS using a novel case-control methodology, and a manageable safety profile. Further studies are therefore warranted to confirm these findings, both in the setting of COVID-19 and potentially in other related viral and post infectious syndromes characterized by endothelial injury.[18,19]

Disclosures

PR receives research funding from Jazz Pharmaceuticals. The remaining authors have no conflicts of interest to disclose.

Contributions

AR, JP, FC and CCS designed the study and wrote the manuscript. AR and FCo collected data. FCo and LJW performed statistical analyses and wrote the manuscript. AR, FCo, LJW, FC and CCS analyzed and interpreted data. AV, GC, CL, RN, CF, AB, EC, FG, FA, FL, GL, AA, PRQ and AAs were involved in patient recruitment, MI and PR provided significant advice throughout the study. All authors had access to the primary data and participated in editing the manuscript.

Acknowledgments

The authors gratefully acknowledge all the patients who participated in the study and their families, as well as the medical and nursing staff, and research personnel, at each of the participating centers. Hajime Uno PhD is especially acknowledged for his contribution to the statistical analysis and Luigi di Filippo MD from IRCCS San Raffaele, for helping in patients recruitment.

Funding

The study was partially funded by Jazz Pharmaceuticals, and the Paula and Rodger Riney Foundation.

Data-sharing statement

Original data may be available upon specific request to the corresponding author.

References

1. WHO COVID-19 Dashboard. Geneva: World Health Organization, 2020. Accessed August 1, 2023. https://covid19.who.int/.
2. Watson OJ, Barnsley G, Toor J, Hogan AB, Winskill P, Ghani AC. Global impact of the first year of COVID-19 vaccination: a mathematical modelling study. Lancet Infect Dis. 2022;22(9):1293-1302.
3. Selvaraj V, Finn A, Lal A, Khan MS, Dapaah-Afriyie K, Carino GP. Baricitinib in hospitalised patients with COVID-19: a meta-analysis of randomised controlled trials. E Clinical Medicine. 2022;49:101489.
4. Abani O, Abbas A, Abbas F, et al. Tocilizumab in patients admitted to hospital with COVID-19 (RECOVERY): a randomised, controlled, open-label, platform trial. Lancet. 2021;397(10285):1637-1645.
5. The RECOVERY Collaborative Group. Dexamethasone in hospitalized patients with Covid-19. N Engl J Med. 2021;384(8):693-704.
6. Beigel JH, Tomashek KM, Dodd LE, et al. Remdesivir for the treatment of Covid-19 - final report. N Engl J Med. 2020;383(19):1813-1826.
7. Ackermann M, Verleden SE, Kuehnel M, et al. Pulmonary vascular endothelialitis, thrombosis, and angiogenesis in Covid-19. N Engl J Med. 2020;383(2):120-128.
8. Varga Z, Flammer AJ, Steiger P, et al. Endothelial cell infection and endotheliitis in COVID-19. Lancet. 2020;395(10234):1417-1418.
9. Ciceri F, Beretta L, Scandroglio AM, Colombo S, et al. Microvascular COVID-19 lung vessels obstructive thromboinflammatory syndrome (MicroCLOTS): an atypical acute respiratory distress syndrome working hypothesis. Critic Care Resusc. 2020;22(2):95-97.
10. Grasselli G, Tonetti T, Protti A, et al. Pathophysiology of COVID-19-associated acute respiratory distress syndrome: a multicentre prospective observational study. Lancet Respir Med. 2020;8(12):1201-1208.
11. Valla DC, Cazals-Hatem D. Sinusoidal obstruction syndrome. Clinics Res Hepatol Gastroenterol. 2016;40(4):378-385.
12. Ruan Q, Yang K, Wang W, Jiang L, Song J. Clinical predictors of mortality due to COVID-19 based on an analysis of data of 150 patients from Wuhan, China. Intensive Care Med. 2020;46(5):846-848.
13. Zhang H, Zhou P, Wei Y, et al. Histopathologic changes and SARS-CoV-2 immunostaining in the lung of a patient with COVID-19. Ann Int Med. 2020;172(9):629-632.
14. Bunte MC, Patnaik MM, Pritzker MR, Burns LJ. Pulmonary veno-occlusive disease following hematopoietic stem cell transplantation: a rare model of endothelial dysfunction. Bone

Marrow Transplant. 2008;41(8):677-686.
15. Richardson PG, Riches ML, Kernan NA, et al. Phase 3 trial of defibrotide for the treatment of severe veno-occlusive disease and multi-organ failure. Blood. 2016;127(13):1656-1665.
16. Richardson PG, Carreras E, Iacobelli M, Nejadnik B. The use of defibrotide in blood and marrow transplantation. Blood Adv. 2018;2(12):1495-1509.
17. Calabretta E, Moraleda JM, Iacobelli M, et al. COVID-19-induced endotheliitis: emerging evidence and possible therapeutic strategies. Br J Haematol. 2021;193(1):43-51.
18. Richardson E, García-Bernal D, Calabretta E, et al. Defibrotide: potential for treating endothelial dysfunction related to viral and post-infectious syndromes. Expert Opin Ther Targets. 2021;25(6):423-433.
19. Richardson E, Carlo-Stella C, Jara R, et al. Response to Maccio et al, "Multifactorial pathogenesis of COVID-19-related coagulopathy: Can defibrotide have a role in the early phases of coagulation disorders?" J Thromb Haemost. 2020;18(11):3111-3113.
20. Rubio-Rivas M, Mora-Luján JM, Formiga F, et al. WHO ordinal scale and inflammation risk categories in COVID-19. Comparative study of the severity scales. J Gen Intern Med. 2022;37(8):1980-1987.
21. Hernán MA. Methods of public health research - strengthening causal inference from observational data. N Engl J Med. 2021;385(15):1345-1348.
22. McCaw ZR, Tian L, Vassy JL, et al. How to quantify and interpret treatment effects in comparative clinical studies of COVID-19. Ann Int Med. 2020;173(8):632-637.
23. Torbicki A, Bacchi M, Delcroix M, et al. Integrating data from randomized controlled trials and observational studies to assess survival in rare diseases: insights from pulmonary arterial hypertension. Circ Cardiovasc Qual Outcomes. 2019;12(5):e005095.
24. Richardson PG, Jagannath S, Chari A, et al. Overall survival with oral selinexor plus low-dose dexamethasone versus real-world therapy in triple-class-refractory multiple myeloma. EJHaem. 2020;2(1):48-55.
25. Ciceri F, Castagna A, Rovere-Querini P, et al. Early predictors of clinical outcomes of COVID-19 outbreak in Milan, Italy. Clin Immunol. 2020;217:108509.
26. Stuart EA. Matching methods for causal inference: a review and a look forward. Stat Sci. 2010;25(1)1-21.
27. Frame D, Scappaticci GB, Braun TM, et al. Defibrotide therapy for SARS-CoV-2 ARDS. Chest. 2022;162(2):346-355.
28. Tocchetti P, Tudone E, Marier JF, Marbury T, Zomorodi K, Eller M. Pharmacokinetic profile of defibrotide in patients with renal impairment. Drug Des Devel Ther. 2016;10:2631-2641.
29. Defibrotide. In: LiverTox: clinical and research information on drug-induced liver injury. Bethesda (MD): National Institute of Diabetes and Digestive and Kidney Diseases; 2012. Accessed June 11, 2023.