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determine whether hypoxia per se was an independent and clinically relevant risk factor for thrombosis. We believe that this issue deserves further investigation and that future studies on thrombosis in COVID-19 patients should be designed to assess the independent contribution of hypoxia to the development of thrombosis.

CONFLICTS OF INTEREST

The authors of this article (Angelo Porfidia, Angelo Santoliquido, Giulia Cammá, Enrica Porceddu and Roberto Pola) have no conflicts of interest to disclose.

KEYWORDS

COVID-19, hypoxia, respiratory insufficiency, risk factors, thrombosis

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REFERENCES

1. Thachil J. Hypoxia - an overlooked trigger for thrombosis in COVID-19 and other critically ill patients. *J Thromb Haemost.* 2020.;18(8):3109-3110.
2. Santoliquido A, Porfidia A, Nesci A, et al. Incidence of deep vein thrombosis among non-ICU patients hospitalized for COVID-19 despite pharmacological thromboprophylaxis. *J Thromb Haemost.* 2020; <https://doi.org/https://doi.org/10.1111/jth.14992>

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Response to Maccio et al, “Multifactorial pathogenesis of COVID-19-related coagulopathy: Can defibrotide have a role in the early phases of coagulation disorders?”

We read with interest the letter by Maccio et al, which postulates a role for defibrotide (DF) in the treatment of coagulation disorders observed in COVID-19.¹ Their commentary effectively explores the contribution of macrophages toward the coagulopathy observed in COVID-19, while highlighting the similarities between COVID-19-associated vascular lesions and veno-occlusive disease (VOD), also known as sinusoidal obstruction syndrome (SOS) and commonly referred to as VOD/SOS. DF has indeed been shown to improve overall survival in patients with severe VOD/SOS² and may be beneficial in the treatment of COVID-19, but not exclusively because of the observed similarities to VOD/SOS.

In detailing the role of macrophages in COVID-19-related coagulopathy, the letter overlooks some other etiologies. While

macrophages may drive endothelial dysfunction by production of reactive oxygen species (ROS) and pro-inflammatory cytokines, the endotheliopathy observed in COVID-19 may also be driven by direct infection of endothelial cells by SARS-CoV-2.³

Angiotensin converting enzyme 2 (ACE2) receptors mediate the entry of SARS-CoV-2 into the cell, via direct interaction between ACE2 and primed viral spike proteins. ACE2 is expressed abundantly on lung alveolar epithelial cells and enterocytes of the small intestine but is also present in arterial and venous endothelial cells, pericytes, and vascular smooth muscle cells across numerous organ systems.⁴ Direct infection and apoptotic death of endothelial cells by SARS-CoV-2 has been demonstrated across vascular beds of several organs via post mortem histology.³

The p38 mitogen-activated protein kinase (MAPK) pathway may be a critical factor in COVID-19-related endotheliopathy. Similar to other respiratory RNA viruses, SARS-CoV-2 infection induces upregulation of p38 MAPK, perhaps enhancing viral replication.⁵ More notably, however, the entry mechanism of SARS-CoV-2 disables a key homeostatic process employed by the cell to diminish p38 MAPK

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downstream of angiotensin II (Ang II) signaling. ACE2 on the cell surface converts Ang II into angiotensin 1-7, which binds the Mas receptor and downregulates the vasoconstrictive and proinflammatory effects driven by Ang II through p38 MAPK activation. Binding of ACE2 by SARS-CoV-2 leads to endocytosis and proteolytic cleavage, resulting in decreased ACE2 expression and functionality, thereby diminishing the counterbalancing effect served by ACE2.⁴ The result is upregulation of p38 MAPK, which activates the transcription of proinflammatory cytokines such as interleukin (IL)-6, tissue necrosis factor (TNF)-alpha, and IL-1beta, all of which are detected at high levels in patients with COVID-19. Unchecked p38 MAPK activity may thereby promote endotheliitis, vasoconstriction, and thrombosis while also augmenting the viral life cycle.⁶

DF has been demonstrated to decrease p38 MAPK activation through direct interaction with endothelial cells,⁷ and DF treatment of COVID-19 may serve to target this signaling axis in COVID-19-related endotheliopathy. Studies have demonstrated increased integrin ligand expression, disrupted endothelial cell membranes, and perivascular inflammation in COVID-19, all of which may also contribute to endothelial cell dysfunction and disseminated intravascular coagulation observed clinically.^{7,8} In murine models, DF downregulates the expression of endothelial cell adhesion molecules that are critical to platelet activation and leukocyte migration such as P-selectin, E-selectin, vascular cell adhesion molecule (VCAM), and intercellular adhesion molecule (ICAM)-1, and lowers endothelial cell release of pro-inflammatory cytokines.⁹ In addition, the severity of COVID-19 has been linked to heparanase activity, with significant elevations of heparanase reported in COVID-19 patients.¹⁰ As a key regulator of inflammation at the endothelium, heparanase likely plays a central role in COVID-19-related vasculitis and thrombosis. DF potently inhibits both heparanase expression and enzymatic activity, furthering its significant potential as a treatment for COVID-19.^{2,7,9}

In the ongoing Spanish DEFACOVID phase IIb randomized clinical trial (clinicaltrials.gov: NCT04348383), as well as other international studies either planned or already under way in Italy, the United States, Ireland, and the United Kingdom, patients treated with DF are being comprehensively assessed for serum plasma inflammatory markers, including heparanase, pro-inflammatory and anti-inflammatory cytokines, immune subpopulation cells, and prothrombotic/antifibrinolytic markers such as von Willebrand factor and soluble thrombomodulin. Preliminary results are encouraging to date, and these data will hopefully serve to demonstrate the effects of DF on the proposed targets in COVID-19.

CONFLICTS OF INTEREST

Edward Richardson has no actual or potential conflict of interest to declare. Dr Carmelo Carlo-Stella has received research support from ADC Therapeutics and Rhizen Pharmaceuticals; has served as consultant or advisor for Servier, Novartis, Genenta Science srl, ADC Therapeutics, Roche, Sanofi, Karyopharm, and Jazz Pharmaceuticals; and has received honoraria for speaker engagements from Bristol-Myers Squibb, Merck Sharp & Dohme, Janssen Oncology, and Astra-Zeneca. Dr Ruben Jara declares honorarium

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AUTHOR CONTRIBUTIONS

Edward Richardson and Jose M. Moraleda contributed to the composition, scholarship, and revision of the manuscript. Carmelo Carlo-Stella, Ruben Jara, Israel Vlodavsky, Massimo Iacobelli, Jawed Fareed, Clifton Mo, Peter O'Gorman, Gregory Yanik, Marta Palomo, and Maribel Diaz-Ricart all contributed to the scholarship and revision of the manuscript.

KEYWORDS

COVID-19, defibrotide, heparanase, p38 mitogen-activated protein kinases, thrombosis, Vascular Endothelium

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REFERENCES

1. Macciò A, Madeddu C, Caocci G, La Nasa G. 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):3106-3107.
2. Richardson PG, Aggarwal S, Topaloglu O, et al. Systematic review of defibrotide studies in the treatment of veno-occlusive disease/sinusoidal obstruction syndrome (VOD/SOS). *Bone Marrow Transplant.* 2019;54:1951-1962.
3. Varga Z, Flammer AJ, Steiger P, et al. Endothelial cell infection and endotheliitis in COVID-19. *Lancet.* 2020;395(10234):1417-1418.
4. Gheblawi M, Wang K, Viveiros A, et al. Angiotensin-converting enzyme 2: SARS-CoV-2 receptor and regulator of the renin-angiotensin system: celebrating the 20th anniversary of the discovery of ACE2. *Circulation Res.* 2020;126(10):1456-1474.
5. Marchant D, Singhera GK, Utokaparch S, et al. Toll-like receptor 4-mediated activation of p38 mitogen-activated protein kinase is a determinant of respiratory virus entry and tropism. *J Virol.* 2010;84(21):11359-11373.
6. Grimes JM, Grimes KV. p38 MAPK inhibition: a promising therapeutic approach for COVID-19. *J Mol Cell Cardiol.* 2020;144:63-65.
7. Palomo M, Mir E, Rovira M, Escolar G, Carreras E, Diaz-Ricart M. What is going on between defibrotide and endothelial cells? Snapshots reveal the hot spots of their romance. *Blood.* 2016;127(13):1719-1727.
8. Ackermann M, Verleden SE, Kuehnel M, et al. Pulmonary vascular endotheliitis, thrombosis, and angiogenesis in Covid-19. *N Engl J Med.* 2020;383:120-128.
9. García-Bernal D, Palomo M, Martínez CM, et al. Defibrotide inhibits donor leukocyte endothelial interactions and protects against acute graft-versus-host disease. *J Cell Mol Med.* 2020. First published: 10 June 2020. <https://doi.org/10.1111/jcmm.15434>
10. Buijsers B, Yanginlar C, Grondman I, et al. Increased plasma heparanase activity in COVID-19 patients. *MedRxiv.* 2020. <https://doi.org/10.1101/2020.06.12.2012930>

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Defibrotide in the COVID-19 coagulopathy: What is the timing?

Dear Editor,

We appreciate the insightful comments by Richardson et al¹ about our letter,² which contributed to better explore the fundamental concepts of our study by providing important, additional information for reflection.

The identification of additional mechanisms involved in the endothelial damage as that described by Richardson et al¹ mediated by the p38 MAPK pathway, which is upregulated as a result of the binding of SARSCoV2 on ACE2 receptors on the surface of endothelial cells and, in turn, activates the transcription of the proinflammatory

cytokines, poses an important problem regarding the timing of the onset of coagulopathy in patients with COVID-19. Consequently it is fundamental to understand the timing of the start of anticoagulant therapy based on the drugs available to date³ to establish a targeted etiopathogenesis approach (ie, based on the etiopathogenesis mechanisms).

The subdivision of COVID-19 into different stages (paucisymptomatic, mild, and critically severe) would reinforce this exigency, and strongly requires us to understand when the coagulopathy begins. Therefore, it is essential to pay attention to the specific phases of the defense mechanism of our body against the pathogen in order to improve our understanding of what happens during the course of COVID-19. SARSCoV2 infection, as previously described, involves