



ELSEVIER

Contents lists available at ScienceDirect

Thrombosis Research

journal homepage: www.elsevier.com/locate/thromres

Review Article

The hypercoagulable state in COVID-19: Incidence, pathophysiology, and management

Mouhamed Yazan Abou-Ismaïl^{a,b,1}, Akiva Diamond^{a,b,1}, Sargam Kapoor^{c,1}, Yasmin Arafah^{a,b}, Lalitha Nayak^{a,b,*}^a University Hospitals, Cleveland Medical Center, Cleveland, OH, United States of America^b Case Western Reserve University, Cleveland, OH, United States of America^c Alaska Native Medical Center, Anchorage, AK, United States of America

ARTICLE INFO

Keywords:

COVID-19
SARS-CoV-2
coagulopathy
thrombosis
inflammation

ABSTRACT

The 2019 coronavirus disease (COVID-19) presents with a large variety of clinical manifestations ranging from asymptomatic carrier state to severe respiratory distress, multiple organ dysfunction and death. While it was initially considered primarily a respiratory illness, rapidly accumulating data suggests that COVID-19 results in a unique, profoundly prothrombotic milieu leading to both arterial and venous thrombosis. Consistently, elevated D-dimer level has emerged as an independent risk factor for poor outcomes, including death. Several other laboratory markers and blood counts have also been associated with poor prognosis, possibly due to their connection to thrombosis. At present, the pathophysiology underlying the hypercoagulable state is poorly understood. However, a growing body of data suggests that the initial events occur in the lung. A severe inflammatory response, originating in the alveoli, triggers a dysfunctional cascade of inflammatory thrombosis in the pulmonary vasculature, leading to a state of local coagulopathy. This is followed, in patients with more severe disease, by a generalized hypercoagulable state that results in macro- and microvascular thrombosis. Of concern, is the observation that anticoagulation may be inadequate in many circumstances, highlighting the need for alternative or additional therapies. Numerous ongoing studies investigating the pathophysiology of the COVID-19 associated coagulopathy may provide mechanistic insights that can direct appropriate interventional strategies.

1. Introduction

The novel coronavirus, severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), emerged in Wuhan, China at the end of 2019 and is now a pandemic [1]. The disease it causes, coronavirus disease 2019 (COVID-19), has affected more than 7 million people worldwide and claimed more than 400,000 lives as of June 2020 [2,3]. The disease ranges from asymptomatic, or mild to severe illness with multi-organ failure and death [4–6]. Coagulopathy, in the form of venous and arterial thromboembolism, is emerging as one of the most severe sequela of the disease, and has been prognostic of poorer outcomes [7–10]. Reports of high incidence of thrombosis despite prophylactic and therapeutic dose anticoagulation raise question about a pathophysiology unique to COVID-19 [11,12]. Proposed hypotheses include a severely heightened inflammatory response that leads to thrombo-

inflammation, through mechanisms such as cytokine storm, complement activation, and endotheliitis [8,9,13,14]. It has also been suggested that the virus itself can possibly activate the coagulation cascade [15]. Although individual institutions have developed guidelines and protocols to institute prophylactic and therapeutic anticoagulation, the optimal management is rapidly evolving as we continue to gather new insights into the pathophysiology of this disease.

Retrospective studies have identified clinical parameters that predict poor prognosis. In addition to markers of coagulopathy such as D-dimer other hematologic parameters have been studied [9,10,16–19]. Neutrophil count, lymphocyte count, neutrophil/lymphocyte ratio, and platelet count correlate with disease severity [8,20–22]. At present, it is clear that patients with COVID-19 infection have a significantly increased risk of thrombosis that prevails despite anticoagulation. A better understanding of the pathophysiology accompanied by

* Corresponding author at: Department of Medicine, University Hospitals Cleveland Medical Center, Joan and Richard Ainsworth Chair in Hematologic Research Case Western Reserve University, 2103 Cornell Road, WRB2-122, Cleveland, OH 44106-7284, United States of America.

E-mail addresses: yazan.abou-ismail@hsc.utah.edu (M.Y. Abou-Ismaïl), lxn64@case.edu, Nayak@uhhospitals.org (L. Nayak).

¹ These authors have shared first-authorship.

<https://doi.org/10.1016/j.thromres.2020.06.029>

Received 15 May 2020; Received in revised form 13 June 2020; Accepted 17 June 2020

Available online 20 June 2020

0049-3848/© 2020 Published by Elsevier Ltd.

Table 1
Table summarizing global incidence of venous and arterial thromboembolic disease in COVID-19.

Location (first author)	Type of study	Sample size	Use of thromboprophylaxis	Venous thromboembolism incidence	Arterial thrombosis incidence	Key characteristics of patient population/other salient features of the study
Wuhan, China (Cui et al)	Retrospective; hospitalized patients	81	No	VTE 25%; all lower extremity thrombi	None	41% patients had other comorbidity (HTN, DM, CAD) and 43% were smokers
Netherlands (Klok et al)	Retrospective; multicenter; hospitalized patients	184	Yes (nadroparin at different doses)	VTE (n = 28) 27%; of those PE (n = 25) was most common finding in 81%	Ischemic strokes (n = 3) 3.7%	76% were male, 2.7% had active cancer and 9.2% were on therapeutic anticoagulation from prior.
Netherlands (Middelcorp et al)	Retrospective; single center; hospitalized patients	198	Yes (nadroparin 2850 units daily for < 100 kg and 5700 units daily for > 100 kg)	7-day incidence of VTE (15%) and 14-day incidence of VTE (34%)	None	Mean age was 64 and mean weight was 87 kg The 7-day and 14-day incidence of VTE was higher in the ICU (25% and 48% respectively) than the general wards (6.5% and 10% respectively)
Italy (Lodigiani et al)	Retrospective; single center; hospitalized patients	388	Yes (LMWH) Ward: 75% used (41% prophylactic dose, 21% intermediate dose; 23% therapeutic dose) ICU: 100% used	VTE 21% (cumulative rate) ICU 27.6% and general ward 6.6%	Ischemic stroke 2.5% and ACS/MI 1.1%	68% were male, 24.1% had BMI ≥ 30, 47.2% had HTN, 22.7% with DM, 11.6% smokers, 6.4% with active cancer and 3.1% with history of prior VTE.
France (Litjens et al)	Retrospective study; 2 ICUs	26	Yes (31% with prophylactic dose and 69% with therapeutic dose)	VTE 69%	None	77% were male, 85% had HTN, 27% consumed tobacco, median BMI 30.2 kg/m ² , median D-dimer was 1750 ng/mL.
France (Helms et al)	Prospective study; COVID-19 ARDS patients at 4 ICUs in 2 centers	150	Yes (LMWH)	PE 16.7%; DVT 2%	Ischemic stroke 1.3%; limb ischemia 0.7%; mesenteric ischemia 0.7%	56% of patients on therapeutic dose and 100% on prophylactic dose had VTE. Patients with COVID-19 ARDS had significantly higher thrombotic events, especially PE (11.7% vs. 2.1%, OR 6.2, p = 0.008)
France (Poissy et al)	Retrospective case series; ICU	107	Yes	PE (20.6%)	None	59.1% were male, median age was 57, median BMI was 30.
Netherlands (Beun et al)	Retrospective; ICU	75	Unknown	PE (26.6%); 21.3% subsegmental and 5.3% central; DVT 4%	Ischemic stroke 2.7%	Incidence of VTE was 2 × - higher than a historical control period 4 patients had heparin resistance apparent by PTT based methods probably due to elevated factor VIII levels
New York, USA (Oxley et al)	Case series	5	No	None	Ischemic stroke 5 young patients in 2 week period	All patients were < 50 years of age. Historical incidence was 0.73 patients in a 2 week period
Beijing, China (Zhang et al)	Case series	3	Unknown	None	Ischemic strokes in 3 patients	Age 65-70, 2/3 were male, all with cardiovascular comorbidities including 2/3 with history of ischemic stroke. All with anti-phospholipid antibodies
Italy (Bellota et al)	Observational cohort study	20	25% were on anticoagulation at baseline due to atrial fibrillation	None	Acute limb ischemia in 20 patients (16.3%)	90% patients were male, mean age was 75 years, 55% had HTN. Incidence increased at 16.3% compared with a baseline rate of 1.8% in this region

DVT = deep venous thrombosis.
PE = pulmonary embolism.
LMWH = low molecular weight heparin.

Table 2

Summary of ongoing observational trials on incidence of coagulopathic changes or thrombosis in patients with COVID-19.

Clinical trial	Location	Status	Study description
NCT04335162 (CovCardiovasc)	France	Recruiting (100 patients)	Screening of cardiovascular complications in COVID-19
NCT04356950	France	Not yet recruiting (175 patients)	Analysis of coagulopathy developed in COVID-19 patients
NCT04356144 (TGA-TM)	Austria	Not yet recruiting (60 patients)	Diagnostic TGA and TGA-thrombomodulin (TGA-TM) in critically ill patients
NCT04363528	France	Not yet recruiting (50 patients)	Incidence of DVT in COVID patients in the ICU
NCT04366778	France	Not yet recruiting (330 patients)	Thromboelastography with tPA to detect patients with high risk of thrombosis
NCT04373486 (COVID-APE)	France	Not yet recruiting (160 patients)	Assessment of acute PE on CT angiography and relationship to D-dimer
NCT04357847 (COVID-Thelium)	France	Not yet recruiting (100 patients)	Assessment of endothelial and hemostatic changes in Severe SARS-CoV-2 infection
NCT04366752 (THROMBOCOVID)	France	Recruiting (100 patients)	Thrombotic events in critical care patients with acute pneumopathy
NCT04359212 (VTE-COVID)	Italy	Not yet recruiting (90 patients)	Thromboprophylaxis with LMWH or fondaparinux in patients recovered in ICU or medical ward

Observational Clinical Trials presently listed on [ClinicalTrials.gov](https://clinicaltrials.gov).

LMWH = low-molecular-weight heparin; TGA = thrombin generation assay.

identification of biomarkers predictive of disease outcomes are critical to develop appropriate interventional strategies for this devastating disease.

In this review, we summarize results of key studies, and discuss the current understanding of coagulopathy and hematological parameters in COVID-19 patients, as well as the pathophysiology and management of thrombosis.

2. The hypercoagulable state with COVID-19

Previous outbreaks of coronaviruses, including SARS-CoV-1 and Middle-Eastern respiratory syndrome (MERS-CoV) have been associated with increased risk of thrombosis [23]. Similarly, the novel SARS-CoV-2 appears to generate a profoundly prothrombotic milieu as evidenced by a surge in global reports of arterial, venous and catheter-related thrombosis [7,24,25]. We summarize the current literature on the incidence of venous and arterial thrombosis in Table 1, as well as ongoing observational studies on the incidence of thrombotic outcomes in Table 2.

2.1. Venous thromboembolism

Pulmonary embolism is the most common thrombotic manifestation of COVID-19[26]. One of the first substantial datasets on risk of venous thromboembolism (VTE) in critically ill patients with COVID-19, reported a VTE incidence of 25%[16]. In a larger study in the Netherlands, 184 ICU patients with COVID-19 who were all on at least standard thromboprophylaxis had a 27% cumulative incidence of VTE, with pulmonary embolism (PE) being most frequent (81%) [24]. Middeldorp et al. reported a higher incidence of thrombotic complications in their ICU patient population (7-day and 14-day cumulative incidence of 25% and 48% respectively) compared to the patients admitted on the wards. All patients initially received standard of care thromboprophylaxis that was intensified later on[25]. Another data set from France that included 150 patients with COVID-19 associated acute respiratory distress syndrome (ARDS) showed a VTE rate of 18%, with PE being most common. When compared to a historical prospective cohort of non-COVID-19 ARDS after matching, patients with COVID-19 ARDS demonstrated a significantly higher rate of thrombotic events, mainly PEs (11.7% vs 2.1%, OR 6.2, $p = 0.008$) [27]. Current data is too scant to determine the demographic characteristics of patients with COVID-19 that are more likely to develop thrombosis. It has however been suggested that this hypercoagulability may be more pronounced in older age, male

gender, Caucasian and African-American ethnicities[16,17,28].

2.2. Arterial thrombosis

In comparison to venous thrombosis, the incidence of arterial thrombosis in COVID-19 appears to be minor (Table 1). It is nevertheless of significant concern and merits further study.

2.2.1. Myocardial infarction

Myocardial infarction (MI) has not been commonly reported with COVID-19. In the Italian study by Lodigiani et al which included 388 patients with COVID-19, the incidence of MI or acute coronary syndromes was 1.1% [29]. Troponin levels have been noted to be significantly higher in the non-survivors, and may provide prognostic value [7,30]. While there are many explanations for elevated troponins (renal injury, myocarditis), ischemic injury as a result of plaque rupture and consequent infarction or secondary to demand ischemia has been reported, and is postulated as another cause of myocardial injury [14].

2.2.2. Stroke

Ischemic strokes were first reported by Dutch investigators[24]. Oxley et al have reported an alarming seven-fold increase in large vessel strokes in the < 50-year-old age group in New York City, New York (5 patients in a 2-week period during COVID-19 pandemic compared with 0.7 patients pre-COVID) [31]. Another case series reports three patients with COVID-19 presenting with strokes and limb ischemia[32]. Clinical presentation with ischemic strokes was also noted by Klok et al (3.7%) and Lodigiani et al (2.5%) [24,29].

2.2.3. Microvascular thrombosis

Several clinical reports have demonstrated evidence of thrombotic microangiopathy (TMA) in patients with COVID-19, most notably in lung autopsies [33–35]. In a study by Menter et al, five out of eleven patients showed evidence of microthrombi in lung autopsies [34], and another case series by Ackermann et al. showed widespread thrombosis with microangiopathy in the lung autopsies of seven COVID-19 patients [35]. The authors compared these findings to those of severe influenza patients, and found that alveolar microthrombi were 9 times more prevalent in COVID-19 patients ($p < 0.001$) [35]. Interestingly, one report from China described the presence of extensive microvascular thrombosis in extrapulmonary organs where coronavirus was not detected, suggesting that a mechanism beyond viral infection is operative [36]. Tian et al provided evidence of microthrombi in pathological

Table 3
Demographic and clinical characteristics of reported cases of thrombi in unusual sites. An article written in Chinese could not be included in this table but has been cited in the main body of this review.

Location (first author)	Age	Gender	Thromboembolic event	Pathologic findings	D-dimer (institution specific units)	Comorbidities	VTE prophylaxis	Treatment	Outcome
Switzerland (Varga et al)	58	F	Mesenteric ischemia	Endotheliitis in small intestine, lung, heart, kidney and liver	Unknown	Diabetes, hypertension, obesity	Unknown	Surgical removal of necrotic bowel, renal replacement therapy	Death
Switzerland (Varga et al)	69	M	Mesenteric ischemia	Endotheliitis of submucosal vessels in small intestine	Unknown	Hypertension	Unknown	Resection of small intestine, mechanical ventilation	Survived
New York (Magro et al)	32	M	Purpuric rash on buttocks	Thrombogenic vasculopathy with necrosis of epidermis and adnexa; complement deposits	D-dimer 1024 ng/mL that peaked at 2090 ng/mL on day 19 (normal 0-229 ng/mL); INR 1.6-1.9; normal PTT and platelets	Obstructive sleep apnea, anabolic steroid use	Unknown	Hydroxychloroquine, azithromycin, remdesvir, mechanical ventilation	Not mentioned
New York (Magro et al)	66	F	Purpuric rash on palms and soles	Superficial vascular ectasia with occlusive arterial thrombus; complement deposits	D-dimer 7030 ng/mL; low platelets at $128 \times 10^9/L$ on day 10; normal INR and PTT	None	Yes	Hydroxychloroquine, prophylactic anticoagulation, renal replacement therapy and supportive care	Not mentioned
New York (Magro et al)	40	F	Livedo racemosa on chest, arms and legs	perivascular lymphocytic infiltrate in superficial dermis and deep seated small thrombi in rare venules; complement deposits	D-dimer 1187 ng/mL; INR 1.4; normal platelet and PTT	None	Unknown	Mechanical ventilation	Not mentioned
Beijing, China (Zhang et al)	69	M	Lower limb, digital ischemia in hand and stroke	None	D-dimer > 21.0 mg/L; PT 17 s, PTT 43.7 s, fibrinogen 4.15 g/L, FDP 85.5 mg/L all on admission to ICU	Hypertension, diabetes and prior stroke	Unknown	Oseltamivir, intravenous immunoglobulin and mechanical ventilation	Not mentioned
Camden, New Jersey, USA	84	M	Renal infarct in addition to stroke and pulmonary embolus	None	D-dimer 21.6 µg/mL	Hypertension	No (Thromboembolism at presentation)	LMWH infusion, mechanical thrombectomy, mechanical ventilation	Death

LMWH = low-molecular-weight heparin.
VTE = venous thromboembolism.

samples obtained from two asymptomatic patients who underwent lobectomies for lung adenocarcinoma, and were subsequently found to be positive for COVID-19 [37]. Pathological examinations revealed patchy inflammatory cellular infiltrate with focal areas of fibrin deposit, suggesting that a local hypercoagulable state in the pulmonary tissue may be an early occurrence. Furthermore, TMA may contribute to findings of unusual sites and presentations of thrombosis that have been described in several clinical reports (Table 3). COVID-19 induced chilblains eruption has been reported, where histopathology revealed microangiopathy thought to be induced by a robust interferon response to the virus [38]. Chilblain-like lesions have also been reported in otherwise asymptomatic adolescents [39]. Transient livedo reticularis has been reported in two patients and is believed to be secondary to coagulopathy [40]. Several reports have mentioned cases with bowel ischemia [14], limb or acral ischemia [32,41] and cutaneous ischemia [42]. Whether the renal injury seen in COVID-19 is related to direct viral toxicity or endothelial/microvascular injury has not been established. One review of 26 autopsy reports did not find evidence of fibrinous material in the renal vasculature [43], while this was found in 3 out of 18 patients in another autopsy review [34].

3. Pathophysiology of COVID-19 coagulopathy: inflammatory thrombosis

In addition to bedside evidence for a hypercoagulable state in COVID-19, laboratory tests have also been consistent with a prothrombotic milieu such as increased D-dimer, fibrinogen, factor VIII (FVIII), von Willebrand factor (vWF), decreased antithrombin, and TEG results [44]. While critical illness is known to cause a hypercoagulable state due to immobilization, mechanical ventilation, central venous access devices, and nutritional deficiencies, COVID-19 appears to cause a hypercoagulable state through mechanisms unique to SARS-CoV-2 and centers around the cross-talk between thrombosis and inflammation [45,46].

The causal, bi-directional relationship between inflammation and thrombosis is well established [47]. COVID-19 causes a profoundly pro-inflammatory state, as evident from multiple reports of high C-reactive protein, lactate dehydrogenase, ferritin, interleukin-6 and D-dimer levels [48]. IL-6 and fibrinogen levels are shown to correlate with each other in COVID-19 patients, providing credence to the idea of inflammatory thrombosis [46]. Researchers presently believe that the inciting event sparking the cycle of inflammation and thrombosis originates in the pulmonary alveoli, where SARS-CoV-2 enters the alveolar epithelium through the ACE2 receptor. Consequently, a severe inflammatory response is initiated that sets the stage for thrombosis through several mechanisms. We propose the following mechanisms as the central pathophysiological aspects of the inflammatory thrombosis caused by COVID-19, which we visually summarize in Fig. 1.

3.1. Localized intravascular coagulopathy

A report by Tang et al. described a high rate (71.4%) of COVID-19 patients meeting ISTH disseminated intravascular coagulopathy (DIC) criteria [10]. However, clear evidence of overt clinical DIC in COVID-19 is lacking thus far. It is possible that the laboratory abnormalities noted are a reflection of a localized coagulopathy in the pulmonary vasculature, resulting from severe alveolar inflammation [17,49]. Progression of this process to the systemic circulation may explain microthrombotic complications and ensuing multi-organ failure. In fact, Ciceri et al propose to label this entire pathophysiology as microvascular COVID-19 lung vessels obstructive thrombo-inflammatory syndrome or MicroCLOTS. [50] The group postulates that, in individuals predisposed to severe outcomes, initial viral damage occurring in the alveoli generates inflammation and local microvascular pulmonary thrombosis. This is followed by more generalized endothelial dysfunction and thrombo-inflammation in the microvasculature of the brain,

kidneys and other organs leading to a hypercoagulable state and multiple organ failure.

3.2. Inflammatory cytokines

Excessive cytokine release is postulated to cause the severe illness noted in younger patients without pre-existing conditions. Higher serum levels of several inflammatory cytokines and chemokines have been associated with severe illness and death in multiple studies [9,51–54]. The cytokine profiles in patients with severe COVID-19 show increased production of IL-6, IL-7, TNF, and inflammatory chemokines such as CCL2, CCL3, and soluble IL-2 receptor, a profile similar to that seen in cytokine release syndromes, such as macrophage activation syndrome [55]. Excessive cytokine release contributes to thrombosis through multiple mechanisms, including activation of monocytes, neutrophils, and the endothelium, all of which generates a prothrombotic state.

3.3. Endothelial activation & dysfunction

Varga and colleagues first reported endothelial dysfunction in multiple vascular beds on post mortem specimens obtained from three patients [14]. In case series of 7 patients by Ackerman et al, lung autopsies of COVID-19 patients showed severe endothelial injury with presence of intracellular virus, as well as widespread thrombosis with microangiopathy [35]. Furthermore, significantly elevated levels of VWF and FVIII in COVID-19 patients are suggestive of endothelial activation in these patients [27,56]. Endothelial activation or dysfunction with COVID-19 may occur through multiple mechanisms. This includes inflammatory cytokines generated in the pulmonary interstitium, the activation of the complement components in blood, or possibly, as a direct result of SARS-CoV-2 infection of endothelial cells through the ACE2 receptor [57]. Endotheliitis, in turn, is a major forerunner to thrombosis. The observation that male sex, obesity, hypertension, and diabetes are poor prognostic factors for severe disease with COVID-19 further supports this theory due to the presence of endothelial dysregulation at baseline in these patients [14]. Whether anti-phospholipid (aPL) antibodies contribute to endothelial dysfunction and activation in COVID-19 is unclear. Anticardiolipin antibodies, β 2 glycoprotein antibodies and positive lupus anticoagulant have all been reported in a few studies [27,32,56,58]. The presence of aPL antibodies in the general population, especially in states of infection, is common [59]. In addition, the contribution of IgA aPL antibodies, reported by Zhang et al, to thrombosis is controversial. Many lupus anti-coagulant assays are sensitive to C-reactive protein (CRP), and lead to false positive results in states where CRP is markedly elevated such as COVID-19 [59,60]. Thus, the clinical relevance of these findings is yet to be determined.

3.4. Mononuclear phagocytes (MNP)

Monocytes and macrophages are theorized to play a crucial role in the inflammation and thrombosis seen in COVID-19. Liao et al demonstrated that MNPs account for 80% of the total bronchoalveolar fluid from patients with severe COVID-19 illness, compared to 60% and 40% in mild cases and healthy controls, respectively [61]. Furthermore, the composition of the cells was characterized by an abundance of inflammatory monocyte-derived macrophages in patients with severe disease. Bronchoalveolar fluid in severe patients is enriched with chemokines that recruit monocytes [62]. COVID-19 patients requiring ICU hospitalization were noted to have a significant expansion of CD14+, CD16+ monocyte populations producing IL-6 in peripheral blood [55,63–65]. In another study, circulating monocytes were shown to have a sustained production of TNF- α and IL-6, a pattern that differs from bacterial or influenza sepsis [66]. Similarly, post-mortem analyses of COVID-19 positive patients revealed that lymphoid tissue macrophages infected with SARS-CoV-2 viral particles expressed IL-6.

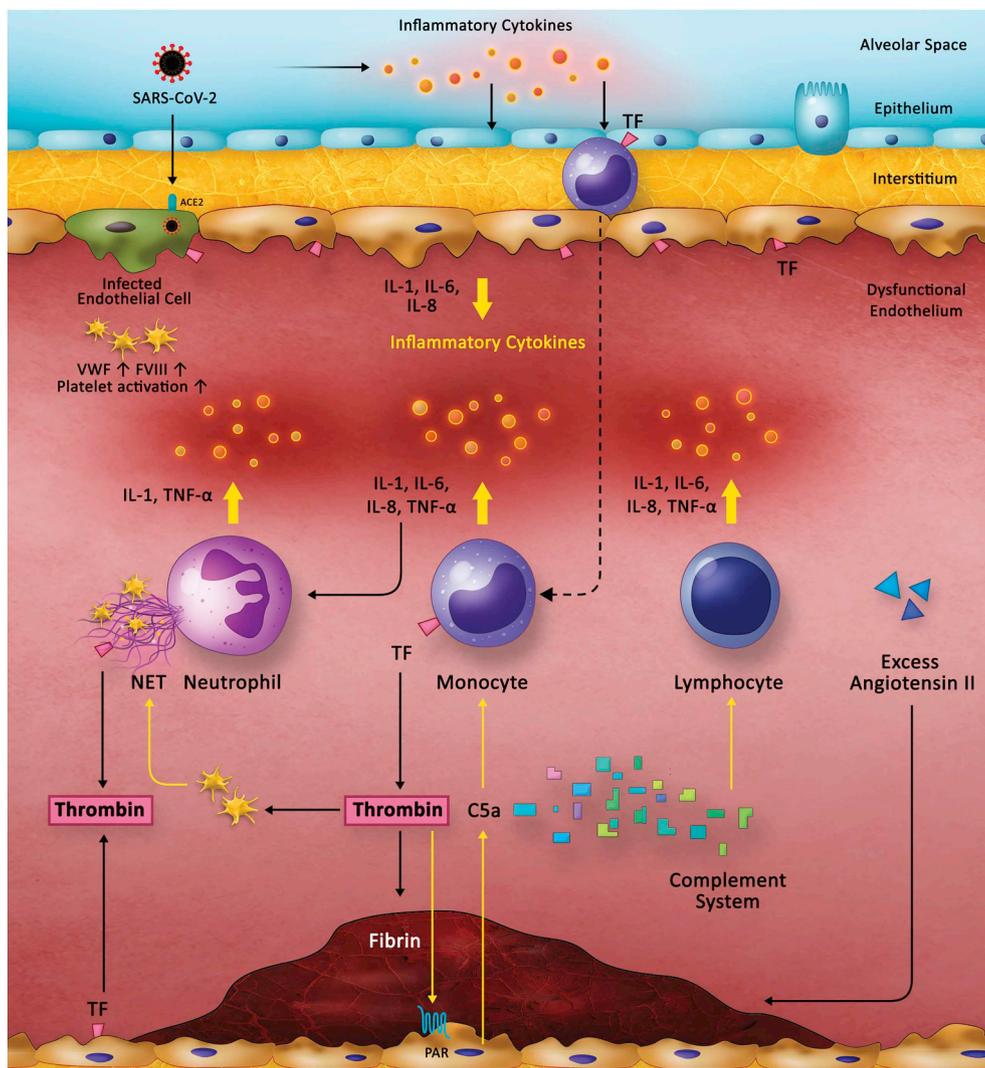


Fig. 1. Pathophysiology of the Hypercoagulable State in COVID-19. The current understanding of the pathophysiology of COVID-19 induced coagulopathy centers around the bidirectional cross-talk between inflammation (yellow arrows) and thrombosis (black arrows). COVID-19 leads to a severe inflammatory response that originates in the alveoli. Release of inflammatory cytokines leads to activation of epithelial cells, monocytes and macrophages. Direct infection of the endothelial cells through the ACE2 receptor also leads to endothelial activation and dysfunction, expression of TF, and platelet activation and increased levels of VWF and FVIII, all of which contribute to thrombin generation and fibrin clot formation. Thrombin, in turn, causes inflammation through its effect on platelets which promote NET formation in neutrophils. It also activates endothelium through the PAR receptor, which leads to release of C5a that further activates monocytes. These mechanisms are currently hypothetical based on existing findings in COVID-19 and previous understanding of the cross-talk between inflammation and thrombosis.
ACE2: Angiotensin-converting enzyme 2.

Further, the presence of IL-6+ macrophages was associated with severe depletion of lymphocytes from lymphoid tissue [67]. These findings suggest that COVID-19 is associated with a clinical and laboratory picture similar to that of macrophage-activation syndrome (MAS). However, there distinguishing features present in COVID-19, such as higher fibrinogen levels and a less pronounced elevation in ferritin and liver dysfunction in comparison to that seen in classical MAS[68]. Presently, the exact mechanisms through which COVID-19 leads to activation of monocytes and macrophages remains unclear. However accumulating data suggests a role for MNPs in the generation of severe illness, including possibly the prothrombotic sequelae. This is not surprising given the understanding that activated monocytes rapidly up-regulate tissue factor (TF) expression. This triggers the coagulation cascade resulting in production of thrombin which in turn leads to thrombus generation, platelet activation, and amplification of pro-inflammatory pathways, primarily through PAR signaling[69].

3.5. Neutrophil extracellular traps (NETs)

While neutrophilia [70] and an elevated neutrophil-lymphocyte ratio (NLR) [71] have been reported by numerous studies now as predictive of worse disease outcomes, the contribution of neutrophil extracellular traps in the pathophysiology of COVID-19 was reported only recently[72]. NETs are implicated to portend pathogenicity in a wide variety of disorders including influenza-associated ARDS [73] and thrombo-inflammation [74,75]. Yu et al report elevated levels of serum

NETs in hospitalized COVID-19 positive patients based on a finding of elevated cell-free DNA, myeloperoxidase-DNA and citrullinated histones in 50 patients with COVID-19. This was especially noted in hospitalized and mechanically ventilated patients [72]. Moreover, they showed that sera obtained from these patients stimulated NET generation in control neutrophils. Taken together with literature linking NETs to pulmonary diseases and thrombo-inflammation, these data begin to implicate NETs as causative in organ damage, widespread thrombosis and mortality that is noted in COVID-19 infection. Finally, a recent manuscript by Barnes et al describes an abundance of neutrophil infiltration in pulmonary capillaries of three patients who succumbed to COVID-19 and suggests that aberrant activation of neutrophils and NET generation may underlie the cytokine storm and severe disease outcomes noted in this disease[76].

3.6. Complement-mediated microangiopathy

Previous studies in animal models provide evidence for complement activation in serum and pulmonary tissue [77,78]. A growing body of evidence suggests a major role for dysregulated complement activation in severe COVID-19[79]. Several reports of post-mortem examinations have demonstrated evidence of TMA, including hyaline thrombi in the small vessels of the lungs and other organs [80,81]. One of those reports demonstrated lung and skin biopsy findings revealing a pauci-inflammatory thrombotic vasculopathy with complement deposit. Researchers in China observed complement hyper-activation in COVID-

19 patients, as well as significantly increased plasma C5a levels in severe cases [82]. Dysregulated complement system activation may be a major contributor to cytokine storm, particularly through the pro-inflammatory effects of anaphylatoxins C3a and C5a [83]. These effects are likely to become more detrimental in patients with a genetic predisposition for decreased complement regulation, and may contribute to findings of TMA and subsequent organ dysfunction.

3.7. Dysregulated renin angiotensin system (RAS)

Although dysfunction of RAS is known to play a significant role in ARDS in general [84,85], this system is specifically important in COVID-19 infections for several reasons. The SARS-CoV2 uses its Spike (S) protein and fuses with the enzyme Angiotensin-Converting Enzyme 2 (ACE2) located on the cell membrane of human cells to gain entry into cells. ACE2 is homologous to ACE, which cleaves angiotensin I (ANGI) to generate ANGI. ANGI binds to the Angiotensin Type 1 Receptor (AT1R) that leads to vasoconstriction and an increase in blood pressure. The inactivation of ANGI by ACE2 results in vasodilation. Conversely, ANGI also negatively regulates ACE2 [86], which is located on lung alveolar epithelial cells, renal tubular epithelial cells, enterocytes of the small intestine, endothelial cells, cardiomyocytes, fibroblasts and pericytes in the heart. SARS-CoV-2 has a high affinity for ACE2, and binding of the SARS-CoV-2 results in loss of ACE2 due to internalization of the virus and ACE2 shedding. This decrease in ACE2 leads to decreased degradation of ANGI resulting in excess ANGI binding to AT1R and increase lung injury [86]. Finally, studies suggest that ANGI binding to AT1R may stimulate IL-6 release, further contributing to the cytokine storm syndrome that is typical of severe COVID-19 infection. Supporting this hypothesis is the evidence showing increased risk of severe disease in patients infected with SARS-CoV or influenza H7N5 that had higher ANGI levels [87,88]. Further, in recent studies on COVID-19 positive patients, viral load and lung injury directly correlated with plasma ANGI levels [89]. In addition, COVID-19 appears to generate worse outcomes in patients with hypertension, cardiovascular disease and diabetes, all of which are associated with reduced baseline levels of ACE2 expression suggesting imbalance in ACE/ACE2 levels [8,90]. It has been shown that ANGI induces TF and plasminogen activator inhibitor 1 (PAI-1) expression by endothelial cells via AT1R, leading to a hypercoagulable state [91,92]. Thus, it is likely these derangements in the RAS pathways, in addition to the multiple pathways described above, likely contribute to the hypercoagulable state of COVID-19, and the ensuing mortality and morbidity of the disease. Presently, rapid investigation into the molecular mechanisms involved is essential in order to gain a better understanding of the pathophysiology of the disease and to direct appropriate, timely therapeutic interventions.

FVIII: Factor VIII.

IL: Interleukin.

NET: Neutrophil extracellular trap.

TF: Tissue factor.

TNF: Tumor necrosis factor.

VWF: von Willebrand factor. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

4. Impact of blood count abnormalities

COVID-19 is associated with a significant effect on the hematological and hemostatic system. In this section we will review the available data on the common hematologic parameters and their prognostic significance. See Table 4 for a summary of the hematologic parameters from various studies.

4.1. Neutrophil count

Although initial studies from Wuhan reported leukopenia in hospitalized COVID-19 positive patients, [9], [8], subsequent reports showed a trend of higher neutrophil count in patients who required ICU admission (ANC 4.2 vs 2.6 $\times 10^9/L$, $p = 0.17$) [20]. Thus, patients requiring ICU care developed neutrophilia during the hospitalization, with a median peak Absolute Neutrophil Count (ANC) of 11.6 $\times 10^9/L$, compared to 3.5 $\times 10^9/L$ in the non-ICU group (P value < 0.001). Further, in a retrospective review of 25 patients who died with COVID-19 in Wuhan the neutrophil count trended up prior to death in 87.5% of patients with evaluable data [93].

4.2. Lymphocyte count

The lymphocyte count has gained much attention in COVID-19 patients and studies consistently report the presence of lymphopenia in this setting. In the report from China Medical Treatment Expert Group for COVID-19 83.2% had lymphocytopenia at hospital admission [8].

Importantly, the lymphopenia is a consistent marker of poor prognosis. Thus, when comparing 109 patients who died in Wuhan versus 116 patients who recovered, the patients who died presented with a decreased lymphocyte count (0.63 vs 1.0 $\times 10^9/L$) and decreased lymphocyte percentage [94]. In a retrospective cohort study of 191 patients with 54 deaths, lymphocyte count was lowest at day 7 of illness onset in survivors and then improved, whereas severe lymphopenia was observed until death in non-survivors [7]. Wang et al. reported that non-survivors developed more severe lymphopenia over time [70]. Flow cytometry on peripheral blood lymphocytes of COVID-19 patients requiring ICU care showed significantly lower CD45+, CD3+, CD4+, CD8+, CD16+, and CD16/56+ counts, without an inversion of the CD4/CD8 ratio [20].

Interestingly, while the number of CD4+ and CD8+ T cells were reduced, both the proportion and number of B cells were not affected or even increased in most patients. Further, the production of IFN- γ by CD4+ T cells and not CD8+ T cells or NK cells tended to be lower in severe cases. Finally, circulating CD8+ T cells contained high concentrations of cytotoxic granules including perforin and granulysin. All these data suggest a dysregulated immune system with overactivation of cytotoxic CD8+ T cells [95].

Potential mechanisms of lymphocytopenia may include direct infection of the lymphocytes by the virus, though the proportion of ACE2-positive lymphocytes is quite small. Lymphocytes express the coronavirus receptor Angiotensin converting enzyme (ACE2) and may be directly targeted [96,97]. In a retrospective comparison of 42 patients with COVID-19 and hypertension, the 17 patients treated with an ACE inhibitor/ARB had absolute numbers of CD3+ and CD8+ T cells that were significantly higher than that in the non-ACEI/ARB group, but no difference in the CD4+ T cells [98].

4.3. Neutrophil to lymphocyte ratio

NLR has been shown to have prognostic significance in septic shock, pancreatitis, pancreatic cancer, and bacteremia, amongst other diseases [99]. An increasing neutrophil count in the setting of a lymphopenia appears to be a sensitive marker of early inflammation and physiologic stress [100–102]. In a study of 413 healthy volunteers the mean NLR was 1.65 (± 1.96 SD: 0.78–3.53) [102].

Several studies have shown a relationship between an elevated NLR and more severe COVID-19 infection. An increased NLR at presentation has a strong association with increased disease severity when compared to patients without severe disease at presentation. Moreover, when stratified by high NLR (> 3.13) and age ≥ 50 , 50% of the patients had severe illness. Similarly, in another analysis of 96 patients, Yang, et al. identified that 46.1% of non-severe patients with an NLR > 3.3 and age > 49.5 would transform into severe cases within a mean of

Table 4
Summary of literature on the impact of common hematologic parameters on disease severity in COVID-19.

Lab	Location (reference)	N (total, non-severe/severe)	Non-severe	Severe	P value
White blood cell count ($\times 10^9/L$) Median (IQR) or [SD]	China (Guan, et.al NEJM)	1099, 926/174	4.9 (3.8–6.0)	3.7 (3.0–6.2)	NR
	Wuhan, China (Qin, et al. Clin Inf Disease)	452, 166/286	4.9 (3.7–6.1)	5.6 (4.3–8.4)	< 0.001
	China (Wang et al. JAMA)	138, 102/36	4.3 (3.3–5.4) ^a	6.6 (3.6–9.8) ^a	0.003
	Shanghai, China (Wu, et al. Jama)	201, 117/84	5.02 (3.37–7.18) ^b	8.32 (5.07–11.20) ^b	< 0.001
	Wuhan, China (Chen, et al. BMJ)	247, 161/113	5.0 (3.7–6.3) ^c	10.2 (6.2–13.6) ^c	NR
Absolute neutrophil count ($\times 10^9/L$) Median (IQR) or [SD]	Wuhan, China (Zhou, et.al)	191, 137/54	5.2 (4.3–7.7) ^c	9.8 (6.9–13.9) ^c	< 0.0001
	Wuhan, China (Qin, et al. Clin Inf Disease)	452, 166/286	3.2 (2.1–4.4)	4.3 (2.9–7.0)	< 0.001
	China (Wang et al. JAMA)	138, 102/36	2.7 (1.9–3.9) ^a	4.6 (2.6–7.9) ^a	< 0.001
	Shanghai, China (Wu, et al. Jama)	201, 117/84	3.06 (2.03–5.56) ^b	7.04 (3.98–10.12) ^b	< 0.001
	Wuhan, China (Chen, et al. BMJ)	247, 161/113	3.2 (2.4–4.5) ^c	9.0 (5.4–12.7) ^c	NR
Absolute lymphocyte count ($\times 10^9/L$) Median (IQR) or [SD]	China (Guan, et.al NEJM)	1099, 926/174	1.0 (0.8–1.4)	0.8 (0.6–1.0)	NR
	Wuhan, China (Qin, et al. Clin Inf Disease)	452, 166/286	1.0 (0.7–1.3)	0.8 (0.6–1.1)	< 0.001
	China (Wang et al. JAMA)	138, 102/36	0.9 (0.6–1.2) ^a	0.8 (0.5–0.9) ^a	0.03
	Shanghai, China (Wu, et al. Jama)	201, 117/84	1.08 (0.72–1.45) ^b	0.67 (0.49–0.99) ^b	< 0.001
	Wuhan, China (Chen, et al. BMJ)	247, 161/113	1.0 (0.7–1.4) ^c	0.6 (0.4–0.7) ^c	NR
Neutrophil/lymphocyte ratio Median (IQR) or [SD]	Wuhan, China (Zhou, et.al)	191, 137/54	1.1 (0.8–1.5) ^c	0.6 (0.5–0.8) ^c	< 0.0001
	Beijing, China (Liu, et al. preprint)	61, 44/17	2.2 (1.4–3.1)	3.6 (2.5–5.4)	0.003
	Wuhan, China (Qin, et al. Clin Inf Disease)	452, 166/286	3.2 (1.8–4.9)	5.5 (3.3–10.0)	< 0.001
	China (Yang, et al. Int Immun)	93, 69/24	4.8 [\pm 3.5]	20.7 [\pm 24.1]	< 0.001
	Wuhan, China (Ma, et al.)	37, 17/20	2.6 (1.8–3.5) ^c	5.5 (3.6–6.5) ^c	0.022
Platelet count ($\times 10^9/L$) Median (IQR) or [SD]	China (Guan, et.al NEJM)	1099, 926/174	172 (139–212)	137 (99–179.5)	NR
	China (Wang et al. JAMA)	138, 102/36	165 (125–188) ^a	142 (119–202) ^a	0.78
	Shanghai, China (Wu, et al. Jama)	201, 117/84	178 (140.0–239.5) ^b	187 (124.5–252.5) ^b	0.73
	Wuhan, China (Chen, et al. BMJ)	247, 161/113	198 (160–256) ^c	156 (111.8–219.3) ^c	NR
	Wuhan, China (Zhou, et.al)	191, 137/54	220 (168–271) ^c	165.5 (107–229) ^c	< 0.0001
Hemoglobin (g/dL) Median (IQR) or [SD]	China (Guan, et.al NEJM)	1099, 926/174	13.5 (12.0–14.8)	12.8 (11.2–14.1)	NR
	Wuhan, China (Chen, et al. BMJ)	247, 161/113	12.8 (11.8–13.8) ^c	12.8 (11.4–14.5) ^c	NR
	Wuhan, China (Zhou, et.al)	191, 137/54	12.8 (12.0–14.0) ^c	12.6 (11.5–13.8) ^c	0.3
	New York, USA (Goyal et al. NEJM)	393, 263/130	13.5 (12.4–14.8) ^d	13.7 (12.3–15.3) ^d	NR

NR = not reached.

^a ICU vs non-ICU.

^b Without ARDS vs with ARDS.

^c Survivors vs Non-Survivors.

^d Non-invasive vs invasive ventilation.

^e Cancer patients.

6.3 days.[103] In a study of 301 patients, an NLR of 2.973 (AUC 0.7338, sensitivity 75.8%, specificity 66.8%) was associated with progression of disease [104]. Finally, a meta-analysis of 5 studies from China with 828 patients, NLR was found to increase significantly in patients with severe disease (standardized mean difference = 2.404, 95% CI - 0.98-3.82) [21].

Increased NLR was also associated with VTE with a mean NLR of 9.5 (5.9-13) in 33 patients who developed VTE versus 5 (3.5-7.9) in 165 patients without VTE [25].

4.4. Platelets

Although a significant drop in platelet counts has not been a prominent feature of the disease, there are certain situations when the presence of severity thrombocytopenia is being recognized as a marker of worse outcomes. Thrombocytopenia was more pronounced in patients with severe infection with a mean platelet count of $137 \times 10^9/L$ vs $172 \times 10^9/L$ in non-severe patients. See Table 4 for the difference in platelet counts between patients with mild versus severe disease. In a retrospective review specifically investigating the relationship between thrombocytopenia and mortality of 1476 consecutive patients in Jinyintan Hospital, Wuhan, thrombocytopenia was reported in 20.7% of patients, using a cutoff of $125 \times 10^9/L$. 72.7% of non-survivors had $< 125 \times 10^9/L$ platelets vs only 10.7% in survivors, $p < 0.001$ [22]. 76 patients (5.1%) had a platelet nadir $< 50 \times 10^9/L$ with a mortality rate of 92.1%. The mortality rate was 61.2% in the group of patients with a nadir platelet count between 50 and $100 \times 10^9/L$. Overall, the majority of patients appear to have a mild thrombocytopenia, which is more pronounced with severe infection.

Presently, it is unclear if a decreased platelet count reflects a more severe DIC and increased consumption or a direct platelet-viral

interaction [105]. Multiple possible mechanisms are possible for viral infection induced thrombocytopenia. These include the development of autoantibodies and immune complexes mediating clearance; direct infection of hematopoietic stem/progenitor cells and the megakaryocytic lineage via CD13 or CD66a resulting in decreased production of platelets; and pathologic activation of the coagulation pathway and consumption of platelets[105,106].

4.5. Hemoglobin

No significant abnormalities have been described regarding red blood cells and anemia. There has been a trend of worse anemia in patients with more severe disease, [20] with a median hemoglobin (Hgb) of 13.2 g/dL in patients requiring ICU versus 14.2 g/dL in non ICU patients ($p = 0.07$), and the majority of patients presented with a normal Hgb count. Of 1099 patients with confirmed COVID-19 in China, the median Hgb was 13.5 g/dL in non-severe patients and 12.8 g/dL in severe patients[8]. However, this trend was not appreciated in a review of 393 patients who required or did not require invasive ventilation in New York City[107]. See Table 4 for a review of the median Hgb levels reported in the various studies.

5. Management

The first general rule in the management of coagulopathy is the treatment of the underlying cause. However, with COVID-19, treatments of the viral infection remain experimental at the current time. As such, until an effective treatment option is available, it is crucial to be able to appropriately manage the sequela of COVID-19-associated coagulopathy.

Table 5
Summary of current literature evidence on the prognostic value of elevated D-dimer in COVID-19 across the globe.

Location (first author)	Sample size	Clinical setting	D-dimer assay (reference range)	D-dimer cut-off for risk assessment	Outcome of interest	Statistics (sensitivity/specificity/odds ratio with p-value)	Salient findings
Wuhan, China (Zhou et al)	191	Hospitalized	Unknown	> 1 µg/mL	Mortality	OR 18.42, 95% CI: 2.64–128.55; p = 0.0033 Se 88.2%/Sp 71.3%	D-dimer > 1 µg/mL indicative of higher odds of death
Wuhan, China (Yao et al)	248	Hospitalized	Immunoturbidimetric assay (0–0.50 mg/L)	> 2.14 mg/L	Mortality		D-dimer elevated in 74.6% of inpatients. Median D-dimer 6.21 mg/L and 1.02 mg/L in non-survivors and survivors respectively, p = 0.000
Wuhan, China (Zhang et al)	343	Hospitalized	CS5100 automatic coagulation analyzer (0–0.5 µg/mL)	> 2 µg/mL	Mortality	HR 51.5, p < 0.001; adjusted HR 22.4 (for age, gender and comorbidity), p = 0.003 N/A	D-dimer > 2.0 µg/mL had higher incidence of mortality when compared to < 2 (12/67 vs 1/267, p < 0.001)
Wuhan, China (Tang et al)	183	Hospitalized	STA-R MAX coagulation analyzer	N/A (continuous variable)	Mortality	N/A	Median D-dimer values were 2.12 µg/mL vs 0.61 µg/mL in the non-survivors and survivors respectively, p < 0.001. 71.4% of non-survivors had DIC per ISTH criteria.
Mainland China (Guan et al)	1099	Hospitalized	Not mentioned	N/A (continuous variable)	Severe disease; Primary composite endpoint was admission to ICU/mechanical ventilation or death	N/A	1) 59.6% of the severe cases presented with elevated D-dimer vs 43.2% of non-severe cases (p = 0.002). 2) 69.4% of patients with the composite primary endpoint had elevated D-dimer vs. 44.2% of those without (p = 0.001).
Wuhan, China (Huang et al)	41	Hospitalized	Not mentioned	N/A (continuous variable)	ICU admission	N/A	Median D-dimer values were 2.4 vs 0.5 in the ICU patients and non-ICU patients respectively, p = 0.0042.
Wuhan, China (Wang et al)	138	Hospitalized	Not mentioned (0–500 mg/L)	N/A (continuous variable)	ICU admission	N/A	Median D-dimer values were 414 mg/L vs 166 mg/L, p < 0.001 in ICU cases and non-ICU cases respectively.
Wuhan, China (Wu et al)	201	Hospitalized	Not mentioned	N/A (continuous variable)	ARDS; mortality	ARDS HR = 1.03, p < 0.001; mortality HR = 1.02, p = 0.002 N/A	Higher D-dimer associated with progress to ARDS and mortality
Milan, Italy (Lodigiani et al)	388	Hospitalized	Not mentioned	N/A (continuous variable)	ICU; mortality	N/A	Table 2 in this published study highlights the higher D-dimer values in non-survivors vs survivors and also in ICU patients vs general ward patients.
Beijing, China (Cui et al)	81	ICU	Succeeder-SF8200 automatic coagulation analyzer	> 1.5 µg/mL	VTE	Se 85%/Sp 88.5%/NPV 94.7%	20/81 (25%) patients had VTE. 8/20 patients with VTE died. D-dimer values were 5.2 ± 3.0 vs 0.8 ± 1.2 µg/ml in the VTE group and non-VTE group respectively, p < 0.001.
Strasbourg, France (Leonard-Lorant et al)	106	Hospitalized	Unknown	> 2660 µg/L	Pulmonary embolism	Se 100%/Sp 67%	32/106 (30%) patients had a PE. Median D-dimer values were IQR 6110 ± 4905 versus 1920 ± 3674 µg/L in the PE and non-PE group respectively, p < 0.001

5.1. Monitoring of laboratory parameters

As a result of the crosstalk between inflammatory and thrombotic pathways, infections are almost always associated with a concomitant activation of the coagulation system, evidenced by elevation in the markers of an activated coagulation system. The D-dimer has been shown to be frequently elevated in patients with COVID-19 [7,16,19,28,108,109]. Increased fibrinogen, fibrin degradation products, prothrombin time (PT), activated partial thromboplastin time (aPTT), and shortened thrombin time (TT) have been described in patients with COVID-19 compared to healthy controls [26,110].

5.1.1. D-Dimer

Numerous studies in COVID-19 patients highlight the prognostic value of increased D-dimer (Table 5) [9,10,16–19]. Accumulating data clearly suggests that an elevated D-dimer, and presence of coagulopathy, serve as prognostic indicators of worse morbidity and mortality in hospitalized patients with COVID-19. In general, the D-dimer is a marker of fibrin formation and degradation, and specifically of plasmin-catalyzed degradation of fibrin polymers and thus, in the case of COVID-19 infection, reflective of pathological activation of the hemostatic pathways. In this review, we describe D-dimer units as reported in the original studies. In the study by Lodigiani et al, the median D-dimer of survivors on admission was 353 ng/mL ($\mu\text{g/L}$), and trended to 529 ng/mL a week later, in comparison to 869 ng/mL and 1494 ng/mL for non-survivors, respectively [111]. Consistently, data by Tang et al shows that non-survivors of COVID-19 infection demonstrated higher D-dimer levels and platelet counts, lower fibrinogen and antithrombin levels, all highly suggestive of ongoing DIC [10]. Monitoring hemostatic parameters such as platelet counts, coagulation assays, D-dimer and fibrinogen is common practice in critical care, especially in DIC [112–115]. These data now provide evidence for the importance of these assessments especially in COVID-19 patients and offer crucial prognostic insights which will likely guide alterations in management.

The ASH expert panel recommends serial monitoring of platelet count, PT, aPTT, D-dimer, and fibrinogen in hospitalized COVID-19 patients. The panel suggests that since worsening of these parameters, specifically the D-dimer, indicates worsening illness, this predicts the need for aggressive critical care or experimental therapies. Likewise, improvement of these parameters along with stable or improving clinical condition may support the decision to step down on aggressive anticoagulation therapy [116]. The ISTH provides similar recommendations [117]. On the other hand, blood product support can be considered in bleeding scenarios as with bleeding in DIC, although clinically overt bleeding is reportedly uncommon in the setting of COVID-19 [112,118].

For therapeutic monitoring of patients who are on anticoagulation with heparin products, anti-Xa monitoring is recommended over aPTT since the latter may be elevated in COVID-19. Furthermore, this allows for less frequent visits to the patient room.

5.2. Anticoagulation

5.2.1. Use of prophylactic or therapeutic dose anticoagulants

As our understanding of the coagulopathy associated with COVID-19 evolves, the best approach to management continues to be explored. Given the paucity of data in the pathophysiology of this disorder, physicians globally are compelled to prepare guidelines for management of this hypercoagulable state based on the established understanding of crosstalk between inflammation and thrombosis. Thus, clinicians are using prophylactic, intermediate, or therapeutic doses of anticoagulation, based on coagulation parameters and the clinical scenario.

Although the optimal dosing remains unclear the benefit of anticoagulation with heparin products (mostly LMWH at prophylactic doses) in COVID-19 patients was demonstrated by a study in China. [119]

Importantly, in the sub-group analysis, those with a sepsis-induced coagulopathy (SIC) score of greater than 3 ($n = 97$) had decreased 28-day mortality (40.0% vs 64.2%, $P = 0.029$), as did the 161 patients with D-dimer greater than 6 times the upper limit of normal (32.8% vs 52.4%, $P = 0.017$). Of concern, a Dutch study by Klok et al reported a 31% incidence of thrombotic complications that occurred despite the presence of at least prophylactic anticoagulation (with LMWH) in patients hospitalized for COVID-19 pneumonia [11]. Consistently, another study, albeit smaller, done in France demonstrated the significant hypercoagulability that exists in COVID-19 patients. Here, Llitjos et al showed that all patients with severe COVID-19 who received prophylactic anticoagulation ($n = 8$) developed VTE, as did 56% of those who received therapeutic dose anticoagulation ($n = 18$) [12].

These findings make a strong argument for considering higher doses (intermediate or therapeutic) of anticoagulation in the management of patients with severe COVID-19, especially in the absence of obvious contraindications such as ongoing bleeding. However, they also bring to mind, the concern that anticoagulation in itself may be insufficient to prevent thrombotic complications. While we battle with these dilemmas, hemostasis experts outline and revise guidelines based on rapidly accumulating data, bearing in mind that there may exist pathophysiologic differences in COVID-19 induced coagulopathy. Below, we summarize the current guidelines and recommendations from different societies, as of the date this article is written, on the role of prophylactic and therapeutic anticoagulation in Table 6.

While all guidelines presently recommend intravenous or subcutaneous low-molecular-weight heparin (LMWH) or unfractionated heparin (UFH) for anticoagulation, another proposed approach has been the use of nebulized heparin for an enhanced localized anticoagulant effect in the pulmonary vasculature. Prior data demonstrated that nebulized heparin significantly reduces coagulation activation in the lungs of critically ill patients [120]. However, as with so many other therapies, this approach is yet to be studied in the context of COVID-19.

5.2.2. Drug Interactions with anticoagulants and antiplatelets

Attention should be given to potential drug interactions between anticoagulants and experimental drugs for COVID-19. ISTH guidelines advise caution (although not avoidance) in patients on DOACs who are admitted with COVID-19 illness due to interactions with antiviral or any other investigational drugs. In a small study by Testa et al, DOAC patients treated with antiviral drugs showed an alarming increase in DOAC plasma levels [124]. The effect of direct oral anticoagulants appears to be potentiated by atazanavir, lopinavir/ritonavir, hydroxychloroquine, and decreased by tocilizumab. Furthermore, apixaban may confer increased risk for QT prolongation when used with hydroxychloroquine. Atazanavir and lopinavir/ritonavir may decrease the active metabolite of clopidogrel and prasugrel. Among atazanavir, lopinavir/ritonavir, remdesivir, hydroxychloroquine, tocilizumab, and interferon beta, there has not been shown to be interactions with heparin products, fondaparinux, or argatroban. A list of drug interactions (collated by the University of Liverpool) can be found at <http://covid19-druginteractions.org> [125].

5.2.3. Duration of Anticoagulation

Data on the optimal duration and method of anticoagulation for COVID-19 patients post-discharge is currently not available. Aspirin has been studied for extended VTE prophylaxis in low-risk orthopedic patients, and could be considered for COVID-19 VTE prophylaxis if the post-discharge criteria are met [126]. The ASH expert panel recommends that any decision to use extended post-discharge thromboprophylaxis with anticoagulation or aspirin should consider the individual patient's VTE risk factors, such as reduced mobility, coagulopathy, and bleeding risk [121]. The International Medical Prevention Registry on Venous Thromboembolism (IMPROVE) VTE risk score has been used as a tool to identify patients who would benefit from extended-use prophylaxis with LMWH [127]. Protocols suggest

Table 6
Current guidelines and recommendations on prophylactic and therapeutic anticoagulation from different societies and institutions.

Recommending source	When to consider prophylactic dose anticoagulation	When to consider therapeutic dose anticoagulation
International Society of Thrombosis & Hemostasis	In all patients with COVID-19 who are hospitalized, including non-critically ill, in the absence of contraindications (active bleeding and platelet count $< 25 \times 10^9/L$). PT and PTT abnormalities are not considered a contraindication [117].	
American Society of Hematology (Expert Panel)	All hospitalized patients with COVID-19. LMWH or fondaparinux (suggested over UFH to reduce contact) in the absence of increased bleeding risk[121].	<ul style="list-style-type: none"> • Intubated patients who develop sudden clinical and laboratory findings consistent with PE, especially when chest X-ray and/or markers of inflammation are stable or improving • Patients with physical findings consistent with thrombosis, such as superficial thrombophlebitis, peripheral ischemia or cyanosis, thrombosis of dialysis filters, tubing or catheters, or retiform purpura • Patients with respiratory failure, particularly when D-dimer and/or fibrinogen levels are very high, in whom other causes are not identified (e.g., ARDS, fluid overload) [122]
Thrombosis UK	<ul style="list-style-type: none"> • For CrCl > 30 mL/min: Give LMWH or fondaparinux • For CrCl < 30 mL/min or acute kidney injury: UFH 5000 units SC BD or TDS or dose-reduced LMWH • All completely immobilized patients would benefit from intermittent pneumatic compression in addition to pharmacological thromboprophylaxis • Mechanical thromboprophylaxis should be used alone if platelets $< 30 \times 10^9/L$ or bleeding [123]. 	
National Institute for Public Health of the Netherlands	All patients with (suspected) COVID-19 admitted to the hospital, irrespective of risk scores.	<ul style="list-style-type: none"> • In patients with a D-dimer $< 1,000$ $\mu\text{g/L}$ on admission but a significant increase during hospital stay to levels above 2,000–4,000 $\mu\text{g/L}$, when imaging is not feasible, therapeutic-dose LMWH can be considered when the risk of bleeding is acceptable. • In patients with a strongly increased D-dimer on admission (e.g. 2,000–4,000 $\mu\text{g/L}$), D-dimer testing should be repeated within 24–48 h to detect further increases in which case imaging for DVT or PE, or empiric anticoagulation, should be considered [15].

that patients hospitalized with COVID-19, especially those with an IMPROVE VTE score of > 3 , an elevated D-dimer level ($> 2 \times$ upper limit of normal), and 2 or more of the following characteristics: age > 60 , previous VTE, known thrombophilia, current cancer, should be strongly considered for extended thromboprophylaxis up to 39–45 days post-discharge either with prophylactic dose LMWH or rivaroxaban [127–129]. For patients who have been empirically started on therapeutic anticoagulation for suspected PE, the ASH panel recommends that they should remain anticoagulated for at least 3 months, regardless of results of future investigation studies. Furthermore, cases of confirmed VTE should be considered as “provoked” and treated for 3–6 months duration [121].

5.3. Antifibrinolytics

There has been interest in the use of anti-fibrinolytics in the management of thrombosis and ARDS in the setting of COVID-19. Fibrin deposition in the alveolar spaces and lung parenchyma is a known observation in ARDS leading to worse respiratory outcomes [130]. Although inhibiting thrombin generation with heparin agents may prevent further fibrin deposition, unlike the use of anti-fibrinolytics, it is not effective against pre-existing fibrin deposits. The use of tPA to treat ARDS in COVID-19 has been proposed, following a case series of 3 COVID-19 patients in whom tPA was associated with temporary improvement in respiratory parameters [131,132]. However, bleeding complications remain a major concern, and given the paucity of data, the use of anti-fibrinolytics is not yet a strong recommendation. An alternative, safer approach that may confer benefit in COVID-19 induced ARDS is the use of nebulized fibrinolytics. In 2019, a study on 60 patients with ARDS showed that use of nebulized streptokinase in patients with severe ARDS resulted in improvements in oxygenation and lung mechanics more rapidly than nebulized heparin [133]. This approach would need further investigation in the COVID-19 setting.

Another agent with fibrinolytic properties that has been considered is Nafamostat. Nafamostat is a synthetic serine protease inhibitor that has been used in Japan for treatment of DIC in pancreatitis for decades. Nafamostat possesses both anti-fibrinolytic activity as well as anti-viral activity, and has thus generated interest in being repurposed as a potential therapeutic agent for COVID-19 in ongoing studies [134].

5.4. Future therapeutic targets and areas of research

As previously highlighted, SARS-CoV-2 presents unique mechanisms of inducing coagulopathy. The limited data revealing a high incidence of thrombosis despite anticoagulation underscores the necessity for novel therapeutic approaches to prevent thrombotic complications and mortality [11,12]. We enlist below the main areas of research that are either being pursued or warrant further study, and provide a summary of ongoing observation and interventional trials in Table 6 and Table 7. These approaches remain theoretical or experimental, but offer insight into potential future strategies that bear promise.

5.4.1. Inflammatory thrombosis

As previously highlighted, the bidirectional cross-talk between inflammation and thrombosis, or “immuno-thrombosis”, is emerging as a major cornerstone of the prothrombotic complications of COVID-19. Experimental use of immunosuppressive agents can potentially set the brakes on this inflammatory process in COVID-19 patients and has been of interest [117].

5.4.1.1. Complement pathway. As previously mentioned, a growing body of evidence supports the role of complement hyper-activation contributing to severe COVID-19 [79]. These studies provide the basis for the suggestion that severe and fatal cases of COVID-19, that have findings consistent with excessive complement activation, may have an increased susceptibility to TMA due to a genetic predisposition to a

Table 7
Summary of ongoing interventional trials using different anti-thrombotic agents in COVID-19 patients.

Clinical Trial	Location	Status	Intervention
NCT04345848	Switzerland	Recruiting (200 patients)	Low and high dose anticoagulation with UFH or LMWH
NCT04354155 (COVAC-TP)	USA	Not yet recruiting (38 patients)	Thromboprophylaxis with enoxaparin
NCT04344756 (CORIMUNO-COAG)	France	Not yet recruiting (808 patients)	Tinzaparin or UFH for 14 days
NCT 04357730	USA	Not yet recruiting (60 patients)	Fibrinolytic therapy to treat ARDS in COVID-19 patients
NCT04362085	Canada	Not yet recruiting (462 patients)	Therapeutic LMWH or UFH versus standard of care
NCT04360824	USA	Not yet recruiting (170 patients)	Standard versus intermediate dose enoxaparin
NCT04367831 (IMPROVE)	USA	Not yet recruiting (100 patients)	Intermediate to prophylactic dose anticoagulation
NCT04363840 (LEAD COVID-19)	USA	Not yet recruiting (1080 patients)	Aspirin 81 mg plus vitamin D
NCT04373707 (COVI-DOSE)	France	Not yet recruiting (602 patients)	Weight adjusted versus fixed low dose LMWH for VTE
NCT04354155 (COVAC-TP)	USA	Not yet recruiting (38 patients)	Thromboprophylaxis with LMWH in children

Interventional Clinical Trials presently listed on [ClinicalTrials.gov](#).

LMWH = low molecular weight heparin; UFH = unfractionated heparin.

pathogenic complement activation. Previous data showed that treatment with humanized anti-C5a antibody greatly ameliorated lung injury and inflammation in a monkey model of virus-induced acute lung injury (H7N9 virus) [135]. In fact, in the same study in China where elevated C5a levels were demonstrated in severe COVID-19, researchers also treated two severe cases with a recombinant C5a monoclonal antibody. Both patients showed significant improvement with resolved fever and increased oxygenation [82]. Thus, complement inhibition, with agents such as eculizumab, may also be a promising treatment for severe COVID-19 [13].

5.4.1.2. Neutrophil extracellular traps. Accumulating data show that neutrophils extracellular traps or NETs have the potential to propagate both inflammation and thrombosis. As highlighted above, NETs are postulated to play a role in COVID-19 associated immunothrombosis as well. This suggests a role for potentially targeting NETs to reduce the clinical severity of COVID-19. Barnes et al propose several mechanisms of either directly targeting the molecules that form NETs, or the process that leads to their formation [76]. Anakinra targets IL1 β , which enhances NET formation, and is currently being investigated in COVID-19 trials ([ClinicalTrials.gov](#) identifiers: [NCT04324021](#), [NCT04330638](#), [NCT02735707](#)) [76].

5.4.1.3. Inflammatory cytokines. Tocilizumab, an interleukin-6 inhibitor, has been used in the setting of cytokine release syndrome in COVID-19, and recent pilot prospective data suggests a survival benefit when given early in the course of the disease [136]. While the impact of tocilizumab on thrombotic outcomes is unclear, it is possible that the survival benefit is secondary to reduced endothelial inflammation and microvascular thrombosis. Regardless, this warrants further investigation. In a small case series in China, the use of plasma exchange for the similar purpose of reducing cytokine storm was attempted with the suggestion of potential benefit [137]. Additionally, the use of non-anticoagulant heparin-like molecules has been proposed as a mechanism to attenuate the release of inflammatory cytokines. LMWH is known to possess anti-inflammatory properties, which could provide additional benefit in COVID-19 patients subject to pro-inflammatory cytokines [8,9,138]. Specifically designed heparan sulfate oligosaccharides with neutralizing proinflammatory proteins and potential anti-viral effect are being investigated in managing COVID-19 [139].

5.4.2. Natural anticoagulant pathways

Since the pathogenesis of inflammation and sepsis-induced coagulopathy involves the generation of thrombin and the reduction of natural anticoagulant proteins, numerous studies have explored the benefit of using coagulation inhibitors such as antithrombin in these situations. Hayakawa et al demonstrated that AT treatment in sepsis-induced DIC may be associated with a reduced in-hospital all-cause mortality [140]. Similarly a study by Kato et al suggested possible mortality benefit in septic patients with coagulopathy treated with recombinant thrombomodulin [141]. Although none of the studies with anticoagulants provide data as robust as the PROWESS trial studying activated protein C (APC), the benefit of using APC is offset with the increased risk for bleeding [142]. In general, these data provide fuel for considering experimental therapies that address physiologic anticoagulant pathways that are most likely inhibited or inactivated in COVID-19. To this end, a group in Austria has already registered a study with [ClinicalTrials.gov](#) to examine the thrombin generation potential of COVID-19 positive patients, with and without the addition of thrombomodulin ([NCT04356144](#)). The results from such studies may provide the foundation for interventional studies exploring anticoagulants such as recombinant thrombomodulin or antithrombin for the management of COVID-19 coagulopathy.

5.5. COVID-19 in patients with bleeding disorders

While we still do not understand how COVID-19 infection may affect people with bleeding disorders, we are aware that severe illness is usually associated with a coagulopathy that resembles DIC. Since some of the treatments used presently for management of bleeding disorders may also interfere with clotting assays, this information is vital to the management of the disease in patients who may receive these therapies.

This is especially true of patients with hemophilia who are treated with emicizumab. The pharmaceutical company manufacturing emicizumab, Genetech, wasted no time in distributing guidelines and information to the physicians at hemophilia treatment centers and other healthcare professionals. Given in [Table 8](#) is a summary of information they provided, which enlists the common coagulation assays that may be used to monitor patients with COVID-19 associated coagulopathy and if these assays are affected by emicizumab [143].

Table 8
Coagulation assays used to monitor COVID-19 positive patients that may be affected by emicizumab (adapted from Genetech).

Analyte/assay	Assay with interference with emicizumab?	Alternatives
aPTT	Yes (overestimate coagulation potential of emicizumab)	For heparin monitoring: anti Xa assay
PT	Yes (weak effect)	No mitigation required (small effect)
D-dimer	No	
Fibrinogen: Clauss method	No	
Fibrinogen: derived	Yes (weak effect)	No mitigation required (small effect); or use Clauss method
Protein C: chromogenic	No	
Protein C: aPTT-based	Yes (overestimate coagulation potential of emicizumab)	Chromogenic protein C assay
Antithrombin activity	No	
Anti-Xa activity	No	
FVIII activity: aPTT based	Yes (overestimate coagulation potential of emicizumab)	Chromogenic FVIII assay (see below guidance)
FVIII activity: chromogenic bovine reagents	No	Does not detect emicizumab, but allows measurement of endogenous or infused FVIII activity
FVIII activity: chromogenic human reagents	No	Responsive to emicizumab, but may overestimate clinical hemostatic potential of emicizumab

For additional information on effects and interferences of emicizumab on coagulation assays, please refer to Adamkewicz et al. *Thromb Haemost* 2019;119:1084-1093.

6. Conclusion

The hypercoagulable state in COVID-19 is emerging as a major pathological occurrence with serious consequences in mortality and morbidity. It is clear that this derangement in the hemostatic pathways is unlike the one seen in other kinds of infection, sepsis, or ARDS. Strikingly, current reports indicate clinical manifestations of both widespread microvascular as well as large vessel thrombosis. Of concern are the data suggesting that anticoagulation in itself may not be adequate in preventing these thrombotic events. Accumulating evidence supports the notion that the hypercoagulability of SARS-CoV-2 involves a unique mechanism of thrombo-inflammation triggered by viral infection, originating in the pulmonary vasculature. A better understanding of this pathophysiology will allow for the development of appropriate therapeutic modalities. Furthermore, the identification of biomarkers of thrombosis and severe illness can guide clinicians on early interventional strategies and focus healthcare resources towards the group of patients at risk for worse outcomes. The significant number of studies listed on [ClinicalTrials.gov](https://www.clinicaltrials.gov) is very reassuring and physicians eagerly await the results of these investigations.

Declaration of competing interest

The authors report no conflicts of interest.

Author contributions

MA, AD, SK, YA, LN all contributed to the writing of the manuscript. MA illustrated the pathophysiology illustration. LN mentored and supervised the manuscript.

Acknowledgements

LN's work is supported by National Heart, Lung, and Blood Institute grants HL142647-01, HL121131-01, and U01 HL143402.

References

- [1] H. Zhu, L. Wei, P. Niu, The novel coronavirus outbreak in Wuhan, China, *Glob Health Res Policy*, 2020.
- [2] World Health Organization, Situation Reports. [Webpage], Available from: <https://www.who.int/emergencies/diseases/novel-coronavirus-2019/situation-reports/>.
- [3] Coronavirus Disease (COVID-2019): Cases in the U.S. 2020 5/14/2020 5/14/2020; Available from: <https://www.cdc.gov/coronavirus/2019-ncov/cases-updates/cases-in-us.html>.
- [4] A. Emami, et al., Prevalence of underlying diseases in hospitalized patients with COVID-19: a systematic review and meta-analysis, *Arch Acad Emerg Med*. 8 (1) (2020) e35, <https://doi.org/10.22037/aaem.v8i1.600>.
- [5] Y. Yi, et al., COVID-19: what has been learned and to be learned about the novel coronavirus disease, *Int J Biol Sci*. (2020) 1753–1766.
- [6] Severe Outcomes Among Patients with Coronavirus Disease 2019, (COVID-19) - United States, February 12-March 16, 2020, *MMWR Morb Mortal Wkly Rep*. 69 (12) (2020) 343–346.
- [7] F. Zhou, et al., Clinical course and risk factors for mortality of adult inpatients with COVID-19 in Wuhan, China: a retrospective cohort study, *Lancet* 395 (10229) (2020) 1054–1062.
- [8] W.J. Guan, et al., Clinical characteristics of coronavirus disease 2019 in China, *N. Engl. J. Med*. 382 (18) (2020) 1708–1720, <https://doi.org/10.1056/NEJMoa2002032>.
- [9] C. Huang, et al., Clinical features of patients infected with 2019 novel coronavirus in Wuhan, *China*. *Lancet* 395 (10223) (2020) 497–506.
- [10] N. Tang, et al., Abnormal coagulation parameters are associated with poor prognosis in patients with novel coronavirus pneumonia, *J. Thromb. Haemost.* 18 (4) (2020) 844–847.
- [11] Klok, F., et al., Incidence of thrombotic complications in critically ill ICU patients with COVID-19, (*in Thromb Res*).
- [12] J.F. Llitjos, et al., High incidence of venous thromboembolic events in anticoagulated severe COVID-19 patients, *J. Thromb. Haemost.* (2020), <https://doi.org/10.1111/jth.14869> In press.
- [13] C.M. Campbell, R. Kahwash, Will Complement Inhibition be the New Target in Treating COVID-19 Related Systemic Thrombosis? *Circulation* 141 (22) (2020) 1739–1741.
- [14] Z. Varga, et al., Endothelial cell infection and endotheliitis in COVID-19, *Lancet* 395 (10234) (2020) 1417–1418.
- [15] Oudkerk, M., et al., Diagnosis, prevention, and treatment of thromboembolic complications in covid-19: report of the National Institute for Public Health of the Netherlands. *Radiology*. 0(0): p. 201629.
- [16] S. Cui, et al., Prevalence of venous thromboembolism in patients with severe novel coronavirus pneumonia, *J. Thromb. Haemost.* 18 (6) (2020) 1421–1424.
- [17] H. Fogarty, et al., COVID-19 coagulopathy in Caucasian patients, *Br. J. Haematol*. 189 (6) (2020) 1044–1049.
- [18] L. Zhang, et al., D-dimer levels on admission to predict in-hospital mortality in patients with covid-19, *J. Thromb. Haemost.* 18 (6) (2020) 1324–1329.
- [19] W.J. Guan, et al., Clinical characteristics of coronavirus disease 2019 in China, *N. Engl. J. Med*. 382 (18) (2020) 1708–1720.
- [20] B.E. Fan, et al., Hematologic parameters in patients with COVID-19 infection, *Am. J. Hematol*. 95 (6) (2020) E131–E134.
- [21] F.A. Lagunas-Rangel, Neutrophil-to-lymphocyte ratio and lymphocyte-to-C-reactive protein ratio in patients with severe coronavirus disease 2019 (COVID-19): A meta-analysis, *J. Med. Virol.* (2020), <https://doi.org/10.1002/jmv.25819> In press.
- [22] X. Yang, et al., Thrombocytopenia and its association with mortality in patients with COVID-19, *J. Thromb. Haemost.* 18 (6) (2020) 1469–1472.
- [23] D. Giannis, I.A. Ziogas, P. Gianni, Coagulation disorders in coronavirus infected patients: COVID-19, SARS-CoV-1, MERS-CoV and lessons from the past, *J Clin Virol* 127 (2020) 104362.
- [24] F.A. Klok, et al., Incidence of thrombotic complications in critically ill ICU patients with COVID-19, *Thromb. Res*. 191 (2020) 145–147.
- [25] S. Middeldorp, et al., Incidence of venous thromboembolism in hospitalized patients with COVID-19, *Journal of Thrombosis and Haemostasis* (2020), <https://doi.org/10.1111/jth.14888> In press.
- [26] H. Han, et al., Prominent changes in blood coagulation of patients with SARS-CoV-

- 2 infection, *Clin. Chem. Lab. Med.* 58 (7) (2020) 1116–1120.
- [27] J. Helms, et al., High risk of thrombosis in patients with severe SARS-CoV-2 infection: a multicenter prospective cohort study, *Intensive Care Med.* 46 (6) (2020) 1089–1098.
- [28] I. Leonard-Lorant, et al., Acute pulmonary embolism in COVID-19 patients on CT angiography and relationship to D-dimer levels, *Radiology* (2020) 201561.
- [29] C. Lodigiani, et al., Venous and arterial thromboembolic complications in COVID-19 patients admitted to an academic hospital in Milan, *Italy. Thromb Res* 191 (2020) 9–14.
- [30] T. Guo, et al., Cardiovascular Implications of Fatal Outcomes of Patients With Coronavirus Disease 2019 (COVID-19), *JAMA Cardiology* (2020).
- [31] T.J. Oxley, et al., Large-vessel stroke as a presenting feature of Covid-19 in the young, *N. Engl. J. Med.* 382 (20) (2020) e60.
- [32] Y. Zhang, et al., Coagulopathy and antiphospholipid antibodies in patients with Covid-19, *N. Engl. J. Med.* 382 (17) (2020) e38.
- [33] D. Wichmann, et al., Autopsy findings and venous thromboembolism in patients with COVID-19, *Ann. Intern. Med.* M20-2003 (2020), <https://doi.org/10.7326/M20-2003>.
- [34] T. Menter, et al., Post-mortem examination of COVID19 patients reveals diffuse alveolar damage with severe capillary congestion and variegated findings of lungs and other organs suggesting vascular dysfunction, *Histopathology* (2020), <https://doi.org/10.1111/his.14134> In press.
- [35] M. Ackermann, et al., Pulmonary vascular endothelialitis, thrombosis, and angiogenesis in Covid-19, *N. Engl. J. Med.* (2020), <https://doi.org/10.1056/NEJMoa2015432> In press.
- [36] T. Zhang, L.X. Sun, R.E. Feng, Comparison of clinical and pathological features between severe acute respiratory syndrome and coronavirus disease 2019, *Zhonghua Jie He He Hu Xi Za Zhi* 43 (0) (2020) E040.
- [37] S. Tian, et al., Pulmonary pathology of early-phase 2019 novel coronavirus (COVID-19) pneumonia in two patients with lung cancer, *J. Thorac. Oncol.* 15 (5) (2020) 700–704.
- [38] A. Kolivras, et al., Coronavirus (COVID-19) infection-induced chilblains: a case report with histopathologic findings, *JAAD Case Rep.* 2020 United States.
- [39] C. Guarnieri, et al., Silent COVID-19: what your skin can reveal, *Lancet Infect. Dis.* (2020), [https://doi.org/10.1016/S1473-3099\(20\)30402-3](https://doi.org/10.1016/S1473-3099(20)30402-3) In press.
- [40] I.F. Manalo, et al., A dermatologic manifestation of COVID-19: transient livedo reticularis, *J. Am. Acad. Dermatol.* (2020), <https://doi.org/10.1016/j.jaad.2020.04.018> In press.
- [41] Y. Zhang, et al., Clinical and coagulation characteristics of 7 patients with critical COVID-2019 pneumonia and acro-ischemia, *Zhonghua Xue Ye Xue Za Zhi* 41 (0) (2020) E006.
- [42] C. Magro, et al., Complement associated microvascular injury and thrombosis in the pathogenesis of severe COVID-19 infection: a report of five cases, *Transl. Res.* 220 (2020) 1–13, <https://doi.org/10.1016/j.trsl.2020.04.007>.
- [43] H. Su, et al., Renal histopathological analysis of 26 postmortem findings of patients with COVID-19 in China, *Kidney Int.* 98 (1) (2020) 219–227.
- [44] M. Panigada, et al., Hypercoagulability of COVID-19 patients in intensive care unit. A report of thromboelastography findings and other parameters of hemostasis, *J. Thromb. Haemost.* (2020), <https://doi.org/10.1111/jth.14850> In press.
- [45] T. Wang, et al., Attention should be paid to venous thromboembolism prophylaxis in the management of COVID-19, *Lancet Haematol* 7 (5) (2020) e362–e363.
- [46] M. Ranucci, et al., The procoagulant pattern of patients with COVID-19 acute respiratory distress syndrome, *J. Thromb. Haemost.* (2020), <https://doi.org/10.1111/jth.14854> In press.
- [47] S.P. Jackson, R. Darbousset, S.M. Schoenwaelder, Thromboinflammation: challenges of therapeutically targeting coagulation and other host defense mechanisms, *Blood* 133 (9) (2019) 906–918.
- [48] C.T. Esmon, Inflammation and thrombosis, *J. Thromb. Haemost.* 1 (7) (2003) 1343–1348.
- [49] F. Marongiu, E. Grandone, D. Barcellona, Pulmonary thrombosis in 2019-nCoV pneumonia? *Journal of Thrombosis and Haemostasis* 18 (6) (2020) 1511–1513.
- [50] F. Ciceri, et al., Microvascular COVID-19 lung vessels obstructive thromboinflammatory syndrome (MicroCLOTS): an atypical acute respiratory distress syndrome working hypothesis, *Crit Care Resusc* (2020) In press.
- [51] G. Chen, et al., Clinical and immunological features of severe and moderate coronavirus disease 2019, *J. Clin. Invest.* 130 (5) (2020) 2620–2629.
- [52] C. Qin, et al., Dysregulation of immune response in patients with COVID-19 in Wuhan, China, *Clin Infect Dis.* 2020.
- [53] Y. Yang, et al., Exuberant elevation of IP-10, MCP-3 and IL-1ra during SARS-CoV-2 infection is associated with disease severity and fatal outcome, *medRxiv* (2020), <https://doi.org/10.1101/2020.03.02.20029975> Submitted for publication.
- [54] J. Gong, et al., Correlation analysis between disease severity and inflammation-related parameters in patients with COVID-19 pneumonia, *medRxiv* (2020), <https://doi.org/10.1101/2020.02.25.20025643> Submitted for publication.
- [55] M. Merad, J.C. Martin, Pathological inflammation in patients with COVID-19: a key role for monocytes and macrophages, *Nat. Rev. Immunol.* 20 (2020) 355–362.
- [56] R. Escher, N. Breakey, B. Lammle, Severe COVID-19 infection associated with endothelial activation, *Thromb. Res.* 190 (2020) 62.
- [57] V. Monteil, et al., Inhibition of SARS-CoV-2 infections in engineered human tissues using clinical-grade soluble human ACE2, *Cell* 181 (4) (2020) 905–913.e7.
- [58] Harzallah, I., A. Deblquis, and B. Drénou, Lupus anticoagulant is frequent in patients with covid-19, (in *J Thromb Haemost*).
- [59] N.T. Connell, E.M. Battinelli, J.M. Connors, Coagulopathy of COVID-19 and antiphospholipid antibodies, *Journal of Thrombosis and Haemostasis* (2020), <https://doi.org/10.1111/jth.14893> In press.
- [60] S.M. Schouwers, J.R. Delanghe, K.M. Devreese, Lupus anticoagulant (LAC) testing in patients with inflammatory status: does C-reactive protein interfere with LAC test results? *Thromb. Res.* 125 (1) (2010) 102–104.
- [61] M. Liao, et al., Single-cell landscape of bronchoalveolar immune cells in patients with COVID-19, *Nat Med* 26 (2020) 842–844 p. 2020.02.23.20026690.
- [62] Zhou, Z.A.R., Lili and Zhang, Li and Zhong, Jiaxin and Xiao, Yan and Jia, Zhilong and Guo, Li and Yang, Jing and Wang, Chun and Jiang, Shuai and Yang, Donghong and Zhang, Guoliang and Li, Hongru and Chen, Fuhui and Xu, Yu and Chen, Mingwei and Gao, Overly exuberant innate immune response to SARS-CoV-2 infection. *Cell Host & Microbe* D-20-00205.
- [63] Y. Zhou, et al., Pathogenic T cells and inflammatory monocytes incite inflammation storm in severe COVID-19 patients, *National Science Review* (2020), <https://doi.org/10.1093/nsr/nwaa041> In press.
- [64] D. Zhang, et al., COVID-19 infection induces readily detectable morphological and inflammation-related phenotypic changes in peripheral blood monocytes, the severity of which correlate with patient outcome, *medRxiv* (2020), <https://doi.org/10.1101/2020.03.24.20042655> Submitted for publication.
- [65] W. Wen, et al., Immune cell profiling of COVID-19 patients in the recovery stage by single-cell sequencing, *Cell Discov.* 6 (2020) 31.
- [66] E.J. Giamarellos-Bourboulis, et al., Complex Immune dysregulation in COVID-19 patients with severe respiratory failure, *Cell Host Microbe* 27 (6) (2020) 992–1000.e3.
- [67] Y. Chen, et al., The novel severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) directly decimates human spleens and lymph nodes, *medRxiv* (2020), <https://doi.org/10.1101/2020.03.27.20045427> (Preprint).
- [68] D. McGonagle, et al., The role of cytokines including interleukin-6 in COVID-19 induced pneumonia and macrophage activation syndrome-like disease, *Autoimmun. Rev.* 19 (6) (2020) 102537.
- [69] J.H. Foley, E.M. Conway, Cross talk pathways between coagulation and inflammation, *Circ. Res.* 118 (9) (2016) 1392–1408.
- [70] D. Wang, et al., Clinical characteristics of 138 hospitalized patients with 2019 novel coronavirus-infected pneumonia in Wuhan, China, *JAMA* 323 (11) (2020) 1061–1069.
- [71] J. Liu, et al., Neutrophil-to-lymphocyte ratio predicts severe illness patients with 2019 novel coronavirus in the early stage, *medRxiv* (2020), <https://doi.org/10.1101/2020.02.10.20021584> (Preprint).
- [72] Y. Zuo, et al., Neutrophil extracellular traps in COVID-19, *JCI Insight* 5 (11) (2020) e138999.
- [73] B.N. Porto, R.T. Stein, Neutrophil extracellular traps in pulmonary diseases: too much of a good thing? *Front. Immunol.* 7 (2016) 311.
- [74] E. Frangou, et al., REDD1/autophagy pathway promotes thromboinflammation and fibrosis in human systemic lupus erythematosus (SLE) through NETs decorated with tissue factor (TF) and interleukin-17A (IL-17A), *Ann. Rheum. Dis.* 78 (2) (2019) 238–248.
- [75] T.A. Fuchs, et al., Extracellular DNA traps promote thrombosis, *Proc. Natl. Acad. Sci. U. S. A.* 107 (36) (2010) 15880–15885.
- [76] B.J. Barnes, et al., Targeting potential drivers of COVID-19: neutrophil extracellular traps, *J. Exp. Med.* (2020) 217(6).
- [77] Y. Jiang, et al., Blockade of the C5a-C5aR axis alleviates lung damage in hDPP4-transgenic mice infected with MERS-CoV, *Emerg Microbes Infect* 7 (1) (2018) 77.
- [78] S. Sun, et al., Inhibition of complement activation alleviates acute lung injury induced by highly pathogenic avian influenza H5N1 virus infection, *Am. J. Respir. Cell Mol. Biol.* 49 (2) (2013) 221–230.
- [79] W.-C. Song, G.A. FitzGerald, COVID-19, microangiopathy, hemostatic activation, and complement, *The Journal of Clinical Investigation* (2020), <https://doi.org/10.1172/JCI140183> In press.
- [80] X.H. Yao, et al., A pathological report of three COVID-19 cases by minimally invasive autopsies, *Zhonghua Bing Li Xue Za Zhi* 49 (0) (2020) E009.
- [81] S.E. Fox, et al., Pulmonary and cardiac pathology in covid-19: the first autopsy series from New Orleans, *medRxiv* (2020), <https://doi.org/10.1101/2020.04.06.20050575> (Preprint).
- [82] T. Gao, et al., Highly pathogenic coronavirus N protein aggravates lung injury by MASP-2-mediated complement over-activation, *medRxiv* (2020), <https://doi.org/10.1101/2020.03.29.20041962> (Preprint).
- [83] X. Zhang, et al., Regulation of Toll-like receptor-mediated inflammatory response by complement in vivo, *Blood* (2007) 228–236.
- [84] E. Vrigkou, et al., The evolving role of the renin-angiotensin system in ARDS, *Crit. Care* 21 (1) (2017) 329.
- [85] H. Zhang, A. Baker, Recombinant human ACE2: acting out angiotensin II in ARDS therapy, *Crit. Care* 21 (1) (2017) 305.
- [86] F. Zores, M.E. Rebeaud, COVID and the Renin-Angiotensin System: Are Hypertension or Its Treatments Deleterious? *Frontiers in Cardiovascular Medicine* 7 (2020) 71.
- [87] S. Itoyama, et al., ACE1 polymorphism and progression of SARS, *Biochem Biophys Res Commun.* (2004) 1124–1129.
- [88] F. Huang, et al., Angiotensin II plasma levels are linked to disease severity and predict fatal outcomes in H7N9-infected patients, *Nat Commun.* 5 (2014) 3595.
- [89] Y. Liu, et al., Clinical and biochemical indexes from 2019-nCoV infected patients linked to viral loads and lung injury, *Sci China Life Sci.* (2020) 364–374.
- [90] C. Tikellis, M.C. Thomas, Angiotensin-converting enzyme 2 (ACE2) is a key modulator of the renin angiotensin system in health and disease, *Int. J. Pept.* 102 (2012).
- [91] D.E. Vaughan, S.A. Lazos, K. Tong, Angiotensin II regulates the expression of plasminogen activator inhibitor-1 in cultured endothelial cells. A potential link between the renin-angiotensin system and thrombosis, *J. Clin. Invest.* 95 (3) (1995) 995–1001.
- [92] S. Nakamura, et al., Plasminogen activator inhibitor-1 expression is regulated by

- the angiotensin type 1 receptor in vivo, *Kidney Int.* 58 (1) (2000) 251–259.
- [93] X. Li, et al., Clinical characteristics of 25 death cases with COVID-19: a retrospective review of medical records in a single medical center, Wuhan, China, *Int J Infect Dis* 94 (2020) 128–132.
- [94] Y. Deng, et al., Clinical characteristics of fatal and recovered cases of coronavirus disease 2019 (COVID-19) in Wuhan, China: a retrospective study, *Chin. Med. J.* 133 (11) (2020) 1261–1267.
- [95] Z. Xu, et al., Pathological findings of COVID-19 associated with acute respiratory distress syndrome, *Lancet Respir. Med.* 8 (4) (2020) 420–422.
- [96] L. Tan, et al., Lymphopenia predicts disease severity of COVID-19: a descriptive and predictive study, *Signal Transduct Target Ther* 5 (1) (2020) 33.
- [97] H. Xu, et al., High expression of ACE2 receptor of 2019-nCoV on the epithelial cells of oral mucosa, *Int J Oral Sci* 12 (1) (2020) 8.
- [98] J. Meng, et al., Renin-angiotensin system inhibitors improve the clinical outcomes of COVID-19 patients with hypertension, *Emerg Microbes Infect* 9 (1) (2020) 757–760.
- [99] Y. Zhou, et al., Prognostic role of the neutrophil-to-lymphocyte ratio in pancreatic cancer: A meta-analysis containing 8252 patients, *Clin. Chim. Acta* 479 (2018) 181–189.
- [100] R.J. Benschop, M. Rodriguez-Feuerhahn, M. Schedlowski, Catecholamine-induced leukocytosis: Early observations, current research, and future directions, *Brain Behav. Immun.* 10 (2) (1996) 77–91.
- [101] M. Onsrud, E. Thorsby, Influence of in vivo hydrocortisone on some human blood lymphocyte subpopulations. I. Effect on natural killer cell activity, *Scand. J. Immunol.* 13 (6) (1981) 573–579.
- [102] R. Zahorec, Ratio of neutrophil to lymphocyte counts—rapid and simple parameter of systemic inflammation and stress in critically ill, *Bratisl. Lek. Listy* 102 (1) (2001) 5–14.
- [103] A.-P. Yang, et al., The diagnostic and predictive role of NLR, d-NLR and PLR in COVID-19 patients, *Int. Immunopharmacol.* 84 (2020) 106504.
- [104] L. Long, et al., Short-term Outcomes of Coronavirus Disease 2019 and Risk Factors for Progression, *Eur. Respir. J.* 55 (5) (2020) 2000990.
- [105] A. Amgalan, M. Othman, Exploring possible mechanisms for COVID-19 induced thrombocytopenia: Unanswered questions, *J. Thromb. Haemost.* 18 (6) (2020) 1514–1516.
- [106] M. Goeijenbier, et al., Review: viral infections and mechanisms of thrombosis and bleeding, *J. Med. Virol.* (2012) 1680–1696.
- [107] P. Goyal, et al., Clinical characteristics of covid-19 in New York City, *N. Engl. J. Med.* 382 (24) (2020) 2372–2374.
- [108] T. Li, H. Lu, W. Zhang, Clinical observation and management of COVID-19 patients, *Emerg Microbes Infect* 9 (1) (2020) 687–690.
- [109] N. Chen, et al., Epidemiological and clinical characteristics of 99 cases of 2019 novel coronavirus pneumonia in Wuhan, China: a descriptive study, *Lancet* 395 (10223) (2020) 507–513.
- [110] E. Terpos, et al., Hematological findings and complications of COVID-19, *American Journal of Hematology* 95 (7) (2020) 834–847.
- [111] C. Lodigiani, et al., Venous and arterial thromboembolic complications in COVID-19 patients admitted to an academic hospital in Milan, Italy, *Thromb Res.* (2020) 9–14.
- [112] H. Wada, et al., Guidance for diagnosis and treatment of DIC from harmonization of the recommendations from three guidelines, *J. Thromb. Haemost.* 11 (4) (2013) 761–767, <https://doi.org/10.1111/jth.12155>.
- [113] F.B. Taylor Jr. et al., Towards definition, clinical and laboratory criteria, and a scoring system for disseminated intravascular coagulation, *Thromb. Haemost.* 86 (5) (2001) 1327–1330.
- [114] M. Levi, et al., Guidelines for the diagnosis and management of disseminated intravascular coagulation. British Committee for Standards in Haematology, *Br. J. Haematol.* 145 (1) (2009) 24–33.
- [115] M. Di Nisio, et al., Diagnosis and treatment of disseminated intravascular coagulation: guidelines of the Italian Society for Haemostasis and Thrombosis (SISET), *Thromb. Res.* 129 (5) (2012) e177–e184.
- [116] Y.Y. Agnes, J.M.C. Lee, Lisa Baumann Kreuziger, Mike Murphy, Terry Gernsheimer, Yulia Lin, COVID-19 and Coagulopathy: Frequently Asked Questions. [Webpage], April 14th, 2020; Version 2.0: Available from: <https://www.hematology.org/covid-19/covid-19-and-coagulopathy>.
- [117] Thachil, J., et al., ISTH interim guidance on recognition and management of coagulopathy in COVID-19. *Journal of Thrombosis and Haemostasis.* n/a(n/a).
- [118] B. Bikdeli, et al., COVID-19 and thrombotic or thromboembolic disease: implications for prevention, antithrombotic therapy, and follow-up, *J. Am. Coll. Cardiol.* (2020) 27284.
- [119] N. Tang, et al., Anticoagulant treatment is associated with decreased mortality in severe coronavirus disease 2019 patients with coagulopathy, *J. Thromb. Haemost.* 18 (5) (2020) 1094–1099.
- [120] B. Dixon, et al., Nebulized heparin reduces levels of pulmonary coagulation activation in acute lung injury, *Crit Care.* (2010) 445.
- [121] L.B. Kreuziger, et al., COVID-19 and VTE/Anticoagulation: Frequently Asked Questions. [Webpage], (2020) April 17, 2020 [cited 2020; Version 2.1].
- [122] A. Lee, et al., COVID-19 and Pulmonary Embolism: Frequently Asked Questions. [Webpage], April 9, 2020; Version 1.0: Available from: <https://www.hematology.org/covid-19/covid-19-and-pulmonary-embolism>.
- [123] B. Hunt, A. Retter, C. McClintock, Practical guidance for the prevention of thrombosis and management of coagulopathy and disseminated intravascular coagulation of patients infected with COVID-19, *Thrombosis UK* (2020), <https://b-s-h.org.uk/media/18171/th-and-covid-25-march-2020-final.pdf>, (2020) , Accessed date: 19 June 2020.
- [124] Testa, S., et al., Direct oral anticoagulant plasma levels striking increase in severe COVID-19 respiratory syndrome patients treated with antiviral agents. The Cremona experience. *Journal of Thrombosis and Haemostasis.* n/a(n/a).
- [125] Liverpool Drug Interactions Group, Interactions With Experimental COVID-19 Therapies, April 9th 2020; Available from: <https://www.covid19-druginteractions.org/>.
- [126] D.R. Anderson, et al., Aspirin or rivaroxaban for VTE prophylaxis after hip or knee arthroplasty, *N. Engl. J. Med.* 378 (8) (2018) 699–707.
- [127] A.C. Spyropoulos, et al., Modified IMPROVE VTE risk score and elevated D-dimer identify a high venous thromboembolism risk in acutely ill medical population for extended thromboprophylaxis, *TH Open* 4 (1) (2020) e59–e65.
- [128] Cohoon, K.P., et al., Emergence of institutional antithrombotic protocols for coronavirus 2019. *Research and Practice in Thrombosis and Haemostasis.* n/a(n/a).
- [129] R.D. Hull, et al., Extended-duration venous thromboembolism prophylaxis in acutely ill medical patients with recently reduced mobility: a randomized trial, *Ann. Intern. Med.* 153 (1) (2010) 8–18.
- [130] Whyte, C.S., et al., Fibrinolytic abnormalities in acute respiratory distress syndrome (ARDS) and versatility of thrombolytic drugs to treat COVID-19. *Journal of Thrombosis and Haemostasis.* n/a(n/a).
- [131] H.B. Moore, et al., Is there a role for tissue plasminogen activator (tPA) as a novel treatment for refractory COVID-19 associated acute respiratory distress syndrome (ARDS)? *J Trauma Acute Care Surg* 88 (6) (2020) 713–714.
- [132] J. Wang, et al., Tissue plasminogen activator (tPA) treatment for COVID-19 associated acute respiratory distress syndrome (ARDS): a case series, *J. Thromb. Haemost.* (2020), <https://doi.org/10.1111/jth.14828> In press.
- [133] A. Abdelaal Ahmed Mahmoud, et al., Streptokinase versus unfractionated heparin nebulization in patients with severe acute respiratory distress syndrome (ARDS): a randomized controlled trial with observational controls, *J Cardiothorac Vasc Anesth* 34 (2) (2020) 436–443.
- [134] H. Asakura, H. Ogawa, Potential of heparin and nafamostat combination therapy for COVID-19, *J. Thromb. Haemost.* 18 (6) (2020) 1521–1522.
- [135] R. Wang, et al., The role of C5a in acute lung injury induced by highly pathogenic viral infections, *Emerg Microbes Infect* 4 (5) (2015) e28.
- [136] S. Sciascia, et al., Pilot prospective open, single-arm multicentre study on off-label use of tocilizumab in severe patients with COVID-19, *Clin. Exp. Rheumatol.* 38 (3) (2020) 529–532.
- [137] J. Ma, et al., Potential effect of blood purification therapy in reducing cytokine storm as a late complication of critically ill COVID-19, *Clin Immunol.* (2020) 108408.
- [138] T.J. Poterucha, P. Libby, S.Z. Goldhaber, More than an anticoagulant: do heparins have direct anti-inflammatory effects? *Thromb. Haemost.* 117 (3) (2017) 437–444.
- [139] J. Liu, et al., Using heparin molecules to manage COVID-2019, *Research and Practice in Thrombosis and Haemostasis* 4 (4) (2020) 518–523.
- [140] M. Hayakawa, et al., Antithrombin supplementation and mortality in sepsis-induced disseminated intravascular coagulation: a multicenter retrospective observational study, *Shock* 46 (6) (2016) 623–631.
- [141] T. Kato, K. Matsuura, Recombinant human soluble thrombomodulin improves mortality in patients with sepsis especially for severe coagulopathy: a retrospective study, *Thromb. J.* 16 (1) (2018) 19.
- [142] G.R. Bernard, et al., Efficacy and safety of recombinant human activated protein C for severe sepsis, *N. Engl. J. Med.* 344 (10) (2001) 699–709.
- [143] J. Thachil, et al., DOACs and 'newer' haemophilia therapies in COVID-19, *J. Thromb. Haemost.* (2020), <https://doi.org/10.1111/jth.14841> In press.