INFLAMMATION AND THROMBOSIS IN COVID-19 PATHOPHYSIOLOGY: PROTEINASE-ACTIVATED AND PURINERGIC RECEPTORS AS DRIVERS AND CANDIDATE THERAPEUTIC TARGETS

COVID-19 causes thromboinflammation, which may be treated via repurposing of available drugs

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CLINICAL HIGHLIGHTS
1. Coronavirus disease 2019 (COVID-19), which is caused by the severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2), has produced a pandemic with clinical features that vary widely among individuals, who can be asymptomatic, mildly or moderately symptomatic (with fever, cough and other symptoms), or have severe disease, which can lead to pneumonia, acute respiratory distress syndrome (ARDS), cytokine storm, multiorgan failure, and death.

2. Clinical observations have revealed a pathobiology resulting from SARS-CoV-2 infection, with multiple phases during the evolution of COVID-19: an upper and then lower respiratory phase followed by a systemic inflammatory phase, which can be excessive and contribute to cellular and tissue damage.

3. Dysregulation in angiotensin signaling, as a consequence of SARS-CoV-2 binding to its cellular receptor, angiotensin-converting enzyme 2 (ACE2), is a key underlying pathophysiological mechanism of cellular and tissue injury and results in increased signaling by thrombin (proteinase-activated) and purinergic receptors on multiple cell types (e.g., platelets, endothelial and epithelial cells, and fibroblasts), leading to thromboinflammation (coagulopathy and thrombosis, accompanying inflammation) and promoting further cell death and tissue injury.

4. Approved drugs that target proteinase-activated or purinergic receptors, as well as other elements of the coagulation cascade, may provide useful therapeutic approaches to blunt multiple aspects of COVID-19 pathobiology, thus reducing morbidity and mortality, but require further preclinical, experimental studies, and randomized controlled trials in patients.
INFLAMMATION AND THROMBOSIS IN COVID-19 PATHOPHYSIOLOGY: PROTEINASE-ACTIVATED AND PURINERGIC RECEPTORS AS DRIVERS AND CANDIDATE THERAPEUTIC TARGETS

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Abstract
Evolving information has identified disease mechanisms and dysregulation of host biology that might be targeted therapeutically in coronavirus disease 2019 (COVID-19). Thrombosis and coagulopathy, associated with pulmonary injury and inflammation, are emerging clinical features of COVID-19. We present a framework for mechanisms of thrombosis in COVID-19 that initially derive from interaction of SARS-CoV-2 with ACE2, resulting in dysregulation of angiotensin signaling and subsequent inflammation and tissue injury. These responses result in increased signaling by thrombin (proteinase-activated) and purinergic receptors, which promote platelet activation and exert pathological effects on other cell types (e.g., endothelial cells, epithelial cells, and fibroblasts), further enhancing inflammation and injury. Inhibitors of thrombin and purinergic receptors may, thus, have therapeutic effects by blunting platelet-mediated thromboinflammation and dysfunction in other cell types. Such inhibitors include agents (e.g., anti-platelet drugs) approved for other indications, and that could be repurposed to treat, and potentially improve the outcome of, COVID-19 patients. COVID-19, caused by the SARS-CoV-2 virus, drives dysregulation of angiotensin signaling, which, in turn, increases thrombin-mediated and purinergic-mediated activation of platelets and increase in inflammation. This thromboinflammation impacts the lungs and can also have systemic effects. Inhibitors of receptors that drive platelet activation or inhibitors of the coagulation cascade provide opportunities to treat COVID-19 thromboinflammation.

1. INTRODUCTION
Evolving clinical and pathological observations implicate a key role for endotheliitis and thrombosis in the pathobiology of infection with SARS-CoV-2 (1–3). Numerous reports have emerged detailing increased risk of thromboembolism (4–6) and its consequences [e.g., stroke (7)] in COVID-19 patients. Increased mortality in COVID-19 has been associated with elevated levels of fibrin/fibrinogen degradation products, D-dimers, and inflammatory markers (8, 9). Platelet-rich thrombi and increased abundance of megakaryocytes (from which platelets are derived) in the microvasculature have been observed with diffuse injury, especially in alveoli in severe cases of COVID-19 (3). Preliminary data indicate increased blood viscosity in severe COVID-19 as a potential contributor to endothelial injury (10). The growing evidence of coagulopathy and thromboinflammation in COVID-19 has led to guidelines that recommend thromboprophylaxis in hospitalized patients [reviewed in (11)].

The pulmonary pathophysiology in COVID-19 has certain features akin to macrophage activation syndrome and disseminated intravascular coagulation (DIC) that occur in other disease settings (albeit with differences in COVID-19 from “classical” DIC, such as normal/elevated fibrinogen levels, less severe or absent anemia, and increased troponin T) (12) and is consistent with the
CLINICAL HIGHLIGHTS

1. Coronavirus disease 2019 (COVID-19), which is caused by the severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2), has produced a pandemic with clinical features that vary widely among individuals, who can be asymptomatic, mildly or moderately symptomatic (with fever, cough and other symptoms), or have severe disease, which can lead to pneumonia, acute respiratory distress syndrome (ARDS), cytokine storm, multiorgan failure, and death.

2. Clinical observations have revealed a pathobiology resulting from SARS-CoV-2 infection, with multiple phases during the evolution of COVID-19: an upper and then lower respiratory phase followed by a systemic inflammatory phase, which can be excessive and contribute to cellular and tissue damage.

3. Dysregulation in angiotensin signaling, as a consequence of SARS-CoV-2 binding to its cellular receptor, angiotensin-converting enzyme 2 (ACE2), is a key underlying pathophysiological mechanism of cellular and tissue injury and results in increased signaling by thrombin (proteinase-activated) and purinergic receptors on multiple cell types (e.g., platelets, endothelial and epithelial cells, and fibroblasts), leading to thrombin-mediated coagulopathy and thrombosis, accompanying inflammation) and promoting further cell death and tissue injury.

4. Approved drugs that target proteinase-activated or purinergic receptors, as well as other elements of the coagulation cascade, may provide useful therapeutic approaches to blunt multiple aspects of COVID-19 pathobiology, thus reducing morbidity and mortality, but require further preclinical, experimental studies, and randomized controlled trials in patients.

The paradigm of “thromboinflammation” (13), i.e., coordinated thrombotic and inflammatory processes. Thrombocytopenia has been noted in certain patients with COVID-19, with lower platelet counts associated with higher mortality (14). Thrombocytopenia may result from increased “platelet consumption” as a consequence of extensive thrombosis in severe cases of COVID-19 (15). The precise mechanisms that evoke thromboinflammation and their impact on clinical features of COVID-19, such as acute respiratory distress syndrome (ARDS) and acute lung injury (ALI), are unclear (16, 17). Although considerable heterogeneity occurs among patients, ARDS in COVID-19 appears to share features with ARDS that occurs in other settings (18, 19). As such, it has been argued that COVID-19 ARDS should be treated in a similar manner as “classical” ARDS (18). This is an active area of research; more data are needed to clarify similarities and differences. Such issues are especially relevant given the potential use of drugs to treat COVID-19 coagulopathy that have been tested in settings such as sepsis, as we discuss in subsequent sections.

Dysregulation of angiotensin signaling induced by interaction of SARS-CoV-2 with ACE2, its cellular receptor, can initiate an increase in cell injury and/or death and inflammation in a range of cell types, providing a mechanistic model for COVID-19 pathobiology, as reviewed in articles proposing this mechanism as a critical factor in COVID-19 pathobiology (20–24). Clinical trials that test the targeting of angiotensin signaling are in progress (e.g., NCT04366050, NCT04335123, NCT04312009, NCT04335136, and NCT04332666). These actions, cytokine production (leading to cytokine storm) and secondary infections, drive tissue injury and organ failure in the lungs and other tissues (25). Platelets do not express ACE2 mRNA or protein (26), implying that enhanced platelet activation in COVID-19 does not result from viral infection of platelets but instead from dysregulation of other cell types that release factors that enhance platelet activation and formation of thrombi. RNA-seq data of megakaryocytes indicate that they also do not express ACE2 (27) (NCBI GEO accession no. GSE131308).

In this review, we identify mechanisms by which the dysregulation of angiotensin signaling in COVID-19 can promote thrombosis and enhance tissue injury by positive feedback mechanisms between inflammation and thrombosis, an idea that extends a previous model (22) (FIGURES 1, 2, and 4). We emphasize the activation of platelets and other cell types relevant to COVID-19 pathobiology, particularly lung endothelial cells, epithelial cells, fibroblasts, and various immune cell types. Existing pharmacological agents that inhibit these mechanisms, i.e., proteinase-activated receptor (PAR) and purinergic (P2Y12) receptor antagonists, are predicted to blunt platelet activation. In addition, we suggest that such antagonists may exert beneficial actions on other cell types affected by COVID-19, highlighting their potential value as candidates for repurposing. The coagulopathy in COVID-19 has led to recommendations for the administration of anticoagulant drugs to mitigate the risk of thrombosis-related complications (11, 28). Most studies regarding treatment of this coagulopathy have emphasized the use of unfractionated or low-molecular-weight heparin (29). The targets/drugs discussed here are complementary approaches and could involve the repurposing of drugs with the potential to address multiple aspects of COVID-19 pathobiology, including coagulopathy.

2. PAR SIGNALING IN COVID-19 THROMBOSIS

Thrombosis-associated processes can occur via angiotensin signaling (and other inflammatory stimuli), which promote the release of the cell surface glycoprotein tissue factor (TF) (22), a receptor for coagulation (serine protease) factor VII (30, 31). In response to injury and inflammation, cell types, including endothelial cells, alveolar epithelial cells, fibroblasts, and innate immune cells (e.g., macrophages and neutrophils) “present” TF (16, 32, 33), which, in concert with factor VII/Vil, initiates the extrinsic coagulation pathway, ultimately leading to the formation of thrombin from circulating prothrombin (31, 34) (FIGURES 1 and
In addition, independent of injury and inflammatory processes, ANG II can induce TF synthesis and expression in various cell types (35, 36). Elevated TF synthesis, in particular, from inflammatory and vascular cells, also occurs in patients with the metabolic syndrome (e.g., hypertension, diabetes, and obesity) (37), contributing to coagulopathy observed in those conditions. Multiple studies show that obese patients are at increased risk of developing severe COVID-19 disease (38–41), underscoring a likely contribution of the metabolic syndrome. Elevation of TF combined with inflammation/injury may, thus, increase the risk of thrombosis in patients with this syndrome.

Thrombin, a proteinase, initiates platelet activation, leading to platelet aggregation, secretion of proinflammatory and additional entities (e.g., thromboxane A2) that further drive thrombosis (42, 43) (FIGURES 1 and 24). These responses of platelets are essential for hemostasis, but in pathological settings, they can promote thromboinflammation (13). Thrombin also exerts effects on endothelial cells, epithelial cells, and fibroblasts (22). All of these cell types express PAR1, a G protein-coupled receptor (GPCR) that is activated by proteinase-mediated cleavage at specific residues on the PAR1 NH2-terminal exodomain (22, 34). Thrombin-mediated agonism of PAR1 on the endothelium is dose-
Cell injury/death releases ADP & ATP, alveolar inflammation via multiple P2Ys & P2Xs

ADP promotes platelet activation; activated platelets secrete ADP

ACE1

Binding to and decreased activity of ACE2

SARS-CoV-2

ANG II↑ ANG 1-7↓
Pathological effects on endothelial cells, epithelial cells, fibroblasts, and innate immune cells in and adjacent to alveoli

Alveolar and neighboring cell types

PAR1 (and PAR4?)

Thrombin

PAR1 and PAR4

Platelets

Prothrombin

Thrombin formation from prothrombin via inhibition of FVa

Protein C

Protein S

Thrombomodulin

→ in diabetes, metabolic syndrome, inflammation

Produced by fibroblasts, endothelial cells, inflammatory cells

Tissue Factor

Coagulation cascade

A

ACE2

ACE1

Conversion to Adenosine

Adenosine Receptors

SARS-CoV-2

ANG II↑ ANG 1-7↓
Pathological effects on endothelial cells, epithelial cells, fibroblasts, and innate immune cells in and adjacent to alveoli

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Protein C

Protein S

Thrombomodulin

→ in diabetes, metabolic syndrome, inflammation

 Produced by fibroblasts, endothelial cells, inflammatory cells

Tissue Factor

Coagulation cascade

B
**C**

Platelet-derived inflammatory factors and adhesion to immune cells

**D**

ADP promotes platelet activation; activated platelets secrete ADP

**FIGURE 2.** Continued
dependent: elevated levels of thrombin signaling are associated with endothelial disruption, injury, inflammation, and tissue damage (22, 35, 44). PAR1-mediated endothelial disruption also occurs in the context of thromboinflammation in septic shock (45). In addition, PAR1 promotes a profibrotic phenotype in fibroblasts, alveolar inflammation, and apoptosis (35, 46, 47), thus exacerbating tissue damage, especially lung injury in COVID-19 (FIGURE 1) (22). Activation of platelets also promotes their adherence to monocytes and neutrophils, thereby enhancing proinflammatory activity (2, 48, 49). Neutrophil-platelet aggregation (50) and possibly platelet-induced generation (51) of neutrophil extracellular traps (NETs) may contribute to pulmonary pathology and coagulopathy in COVID-19. Further, NETs can activate platelets, inducing coagulation (52, 53). Neutrophils also release other factors, such as cathelicidins, which can activate platelets, underscoring a complex interplay between neutrophils and platelets (49). In addition, platelets secrete proinflammatory factors (e.g., IL-1β, RANTES), prothrombotic entities (e.g., TXA2), and platelet activating factor (PAF) that can further enhance inflammation and platelet activation (FIGURE 1) (42, 43, 48). Thrombin-mediated activation of platelets also results in secretion of ADP from platelet-dense granules (54). Released ADP can initiate purinergic signaling via multiple receptors (described in subsequent sections), further enhancing platelet activation (FIGURES 1 and 2B). Besides stimulating cell-based actions via PAR receptors, thrombin cleaves fibrinogen to generate fibrin and facilitate thrombus formation (31).

In addition to the TF-promoted stimulation of the extrinsic arm of the coagulation cascade, platelet activation contributes to coagulation by engaging the intrinsic coagulation cascade, especially via the release of polyphosphates (polyP) stored in platelet granules (55–57). PolyP is as an activator of factor XII, which initiates the intrinsic coagulation cascade (31, 55, 56). Factor XII activation, which generates factor XIIa, can also occur by other entities (58) [e.g., collagen (59), NETs (60), bacterial contact factors and polyphosphates (61) and extracellular nucleic acids (60)] whose abundance may be increased in COVID-19-induced lung injury and pneumonia. Factor XII activation reportedly occurs rapidly with disease onset in ARDS (62), highlighting a likely role for the intrinsic coagulation cascade in severe COVID-19 pulmonary injury. In addition to its procoagulant role, activated factor XII can contribute to inflammation by its activation of plasma kallikrein, thereby increasing formation of bradykinin (63), a proinflammatory peptide (64). Factor XIIa may also exert direct proinflammatory effects on immune cells, in particular, neutrophils (65, 66), though a clear delineation of the proinflammatory effects of factor XII compared to factor XIIa is still emerging. Thus, activation of coagulation by TF can have multiple thromboinflammatory effects via the diverse effects of factor XIIa.

Hence, the actions of thrombin described in the paragraphs above contribute to a pathological positive feedback loop: angiotensin-promoted inflammation and cell death lead to generation of thrombin, which, via PAR1, activates platelets and other cell types, thereby exacerbating cellular and tissue injury. This mechanism likely contributes to increased thrombosis in COVID-19 (1, 2, 3, 16) and predicts that greater tissue injury and inflammation will be associated with increased thrombosis, consistent with clinical data (12). Inhibiting PAR1 may, thus, have multiple beneficial effects in SARS-CoV-2 infections.

Thrombin-mediated activation of platelets not only occurs via PAR1 [which signals via the heterotrimeric G proteins, Gq/Go (reducing cAMP), Gq/G11 (inducing calcium [Ca2+] signaling) and G12/13 (activating RhoA) (67)] but also by the related GPCR, PAR4 (34, 67, 68), although PAR4 requires higher thrombin concentrations for activation (34). PAR4 signals via Go/G11 and G12/13, suggesting possible synergy with PAR1 signaling (34, 67, 69). In addition, PAR4 forms heterodimers with PAR1 in platelets, leading to long-term Ca2+ response and enhanced activation by thrombin (34, 68, 70). PAR4 also acts on other cell types relevant to COVID-19 pathobiology. PAR4 is expressed by endothelial cells of numerous tissues (71, 72) and enhances adhesion of monocytes to endothelial cells (69, 71). In pulmonary endothelial cells, thrombin, via PAR4 and PAR1, alters actin fiber formation and cytoskeletal remodeling (72). PAR4 also perturbs pulmonary epithelial cell function, enhancing Ca2+ signaling (73), IL-6 and IL-8 secretion (74), and epithelial-to-mesenchymal transition (75). PAR4 may also enhance chemotaxis and proinflammatory signaling in neutrophils and monocytes, including monocyte-endothelial cell interactions (71). Thus, while less extensive data exist for PAR4 than PAR1, the two
PARs have similar actions and may act in concert. Accordingly, inhibition of PAR4 may blunt effects of COVID-19 pathophysiology.

3. REGULATION OF PAR-MEDIATED EFFECTS BY THE PROTEIN C PATHWAY

Activated protein C (a serine protease) modulates thrombin signaling and activation of coagulation by inhibiting thrombin formation from prothrombin (31, 76). (FIGURE 1) Thrombomodulin on the surface of endothelial cells binds to thrombin, forming a complex with the endothelial cell protein C receptor (EPCR), which converts protein C in the circulation to activated protein C (APC). APC, with its cofactor Protein S, inactivates Factor Va (and Factor VIIIa in the intrinsic coagulation cascade) and reduces thrombin formation (76, 77). APC also cleaves PAR1, but at different amino acid residues than does thrombin, and it elicits anti-inflammatory, endothelial-protective and anti-thrombotic responses (44, 77). Decreased protein C, protein S, and thrombomodulin are, thus, risk factors for thrombotic disorders (76, 78, 79). Reduced levels of protein C and protein S occur in diabetic patients, especially those with poorly managed diabetes (80, 81). This raises the possibility that these diabetic patients may have an elevated risk for coagulopathy in COVID-19, given the predisposition for elevated TF in these subjects, as discussed above. Inflammatory stimuli (e.g., certain cytokines) and cleavage resulting from neutrophil-endothelial cell interactions can decrease the expression and availability of thrombomodulin (76, 82). A decrease in endothelial thrombomodulin has been implicated in sepsis and ARDS (76, 77) and may contribute to thrombosis in COVID-19 pathobiology (82). Recombinant thrombomodulin has been suggested as a treatment for ARDS, based on animal studies (77). Activated protein C (83) and thrombomodulin [both administered intravenously (84, 85)] may, thus, be potentially beneficial therapies in COVID-19.

4. PLATELET ACTIVATION BY MATRIX PROTEINS IN COVID-19

Platelets are also activated by adhesion to extracellular matrix (ECM) proteins, in particular, von Willebrand Factor (vWF), collagen (types I, II, and VI), fibronectin, and thrombospondin (86, 87). Multiple platelet membrane glycoproteins (GPs) and integrins are receptors for these ECM proteins, facilitating platelet adhesion to endothelial surfaces as an initial step in thrombus formation (86, 87). The adhesion of platelets by vWF and collagen subsequently triggers signaling events that activate platelets. These ECM proteins are produced in increased amounts in lung fibrosis (88) and endothelial inflammation (89), which are features of the ANG II-mediated COVID-19 pathobiology (FIGURE 1) (22). Elevated vWF levels have been reported in patients with severe COVID-19 (90). Enhanced ECM protein production is, thus, a potentially complementary mechanism to thrombin and ADP-induced platelet activation (discussed below) in COVID-19.

5. PURINERGIC SIGNALING IN COVID-19 PATHOBIOLOGY

In addition to thrombin and ECM proteins, purinergic signaling via ADP is a key mechanism for platelet activation (43, 91, 92). The primary platelet receptors for ADP are the P2Y GPCRs, P2Y12 and to a lesser degree P2Y1, and P2X4, a ligand-gated ion channel. Extracellular ATP can also activate these receptors (93). As with activation by PARs, purinergic activation of platelets is prothrombotic and promotes release of proinflammatory factors (FIGURES 1 and 28), thereby contributing to thromboinflammation (92). Purinergic- and thrombin-mediated mechanisms can synergistically activate platelets (94, 95), and thus, they may be an additional mechanism for thrombosis in COVID-19. Inflammation and cell death are associated with increases in extracellular ADP and ATP, including in ARDS and ALI, resulting in platelet activation, while also exerting potentially pathological effects on other cell types (43, 91, 96). Platelet activation by thrombin also promotes ADP release from platelet dense granules, adding to ADP-driven purinergic signaling and activation of platelets (54). ADP/ATP released with pulmonary inflammation is subsequently converted to adenosine, which can exert a range of effects on cell types relevant to pulmonary inflammation, via adenosine receptors (96) (FIGURE 1). The potential relevance of these receptors in COVID-19 has been suggested (97, 98).

P2Y12-mediated ADP signaling can promote inflammation via actions on innate immune cells, especially dendritic cells and macrophages (99, 100). P2Y1 and P2X2 may also have proinflammatory effects on immune cells (92, 101). In addition, P2Y12 may contribute to endothelial cell pathology (102, 103). P2Y12 inhibition (by clopidogrel or ticagrelor) has potentially beneficial effects that include enhanced nitric oxide production, improved endothelial integrity, and reduced oxidative stress (104). In addition, P2Y1 promotes inflammatory processes in endothelial cells, e.g., adhesion of monocytes, which may contribute to vascular inflammation (92, 105). Hence, as with thrombin, a positive feedback loop exists for
purinergic activation of platelets: tissue injury promotes platelet activation and subsequent thromboinflammation, which, in turn, further exacerbates injury. Purinergic signaling that drives platelet activation can also affect other cell types that increase inflammatory responses and potentially contribute to clinical features of COVID-19. Accordingly, akin to inhibition of PARs, purinergic receptor inhibitors may have therapeutic effects in addition to preventing thrombosis.

Other purinergic receptors, i.e., P2Y2, P2Y6, and P2X7, may also contribute to pathological effects in pulmonary cell types. P2Y2 [activated by ATP but not ADP (93)] regulates endothelial inflammation, most notably by promoting adhesion of inflammatory cells (92, 106). P2Y2 also induces activation and chemotaxis of inflammatory cell types, especially dendritic cells and neutrophils (92, 106, 107) and promotes a profibrotic phenotype in lung fibroblasts (108). Akin to P2Y2, P2X7 [activated by ATP, but not ADP (93)] exerts proinflammatory effects on endothelial cells and dendritic cells (92, 96, 107). P2Y6, which is primarily activated by UDP but also ATP (93), is upregulated in the endothelium and inflammatory cells in settings relevant to lung injury (92, 109), can enhance vascular inflammation and via actions on lung fibroblasts, may contribute to pulmonary fibrosis (110).

6. A ROLE FOR MEGAKARYOCYTES IN COVID-19?

Thrombin and ADP can also affect megakaryocytes, the source of platelets, potentially facilitating megakaryocytopenia (111) (FIGURE 1D). In COVID-19, the number of megakaryocytes increases in the pulmonary circulation proximal to regions with alveolar injury (3). Thrombopoietin (TPO), the principal driver of megakaryocytopenia, is primarily secreted by the liver (111). Hepatic production of TPO, although considered constitutive, can be regulated by inflammatory factors, e.g., increased by IL-6 (112, 113). Elevated systemic inflammation in severe cases of COVID-19 (48) may, therefore, be associated with increased TPO levels and megakaryocytopenia. IL-6, a key inflammatory marker in COVID-19, increased TPO levels in SARS-1 patients (114). Hepatocyte production of TPO is also stimulated by platelet turnover, whereby desialylated platelets activate the hepatocyte Ashwell-Morell receptor (115–117), a mechanism in various pathologies, including sepsis (118). Other factors that can promote megakaryocytopenia include IL-1β and vWF (111, 119), which increase with injury and inflammation, including in ALI and ARDS in COVID-19.

Limited data suggest that thrombin and ADP may enhance megakaryocyte maturation and platelet formation. Megakaryocytes express P2Y12, P2Y1, and P2X1 receptors that can mediate responses to ADP [e.g., generation of Ca2+ currents and binding to fibrinogen (120, 121)] implicated in megakaryocyte maturation and platelet formation (122). Thrombin may also induce megakaryocyte maturation, as suggested by changes in Ca2+ signaling, morphology, activation of pathways associated with platelet formation, secretion of α-granule proteins, and increase in VEGF secretion (123–126). Such effects have not yet been attributed to specific PARs, but RNA-seq data indicate that PAR1 and PAR4 are among the 25% highest expressed genes in megakaryocytes (27) (accession no. GSE131308). However, definitive evidence is lacking for enhancement of megakaryocytopenia or platelet formation by thrombin or purinergic signaling. Megakaryocytes may also have antiviral effects, for example, by secretion of interferon in the context of influenza (127), but such effects have not as-yet been shown in COVID-19.

7. A PUTATIVE SEQUENCE OF PHASES IN COVID-19 THROMBOINFLAMMATION

On the basis of the discussion above of the pathobiology of thromboinflammation, FIGURES 2, A–D show a putative sequence of signaling events and cell-cell interactions that drive phases of pulmonary (and potentially systemic) thromboinflammation in COVID-19. Inflammation and tissue injury occur as a consequence of dysregulated angiotensin signaling caused by SARS-CoV-2 binding to ACE2 (47). As a result, TF production increases from cell types in and near the alveoli. Thereby, the extrinsic coagulation cascade is activated, promoting thrombin formation and signaling, which exerts effects on platelets and alveolar cells (FIGURE 2A). As tissue inflammation/injury and thrombin-promoted platelet activation continue, ADP release is increased, further enhancing platelet activation (FIGURE 2B). The increase in platelet activation enhances their participation in inflammatory processes, including by the release of inflammatory entities and adhesion to inflammatory cells, further enhancing tissue injury (FIGURE 2C). Platelet activation also increases the activation of Factor XII and the intrinsic coagulation cascade (FIGURE 2C). As systemic inflammation increases (especially in severe COVID-19), and perhaps also as a consequence of increased thrombin and purinergic signaling, megakaryocytopenia and platelet number may increase, further promoting thromboinflammation (FIGURE 1D). FIGURES 1 and 2, thus, highlight temporal events in COVID-19 induced
thromboinflammation (e.g., enhanced thrombin signaling likely precedes enhanced platelet activation by purinergic signaling) and identifies potential targets for treating and preventing this aspect of COVID-19 pathophysiology.

8. CROSS TALK BETWEEN COMPLEMENT SIGNALING AND THROMBOINFLAMMATION

The complement cascade is a key immune mechanism that shows a high degree of crosstalk with the coagulation cascade and inflammatory signaling (128–132). The complement system likely contributes to thromboinflammation in COVID-19 (31, 40, 82, 137) and other settings with ARDS (137). Numerous studies have shown activation of the complement cascade in coronavirus infections, including SARS-CoV-1, MERS, and SARS-CoV-2, often early in the course of the disease (133, 138, 139).

The mechanisms by which SARS-CoV-2 induces complement activation are somewhat unclear (134–136). Three main mechanisms exist for complement activation (FIGURE 3): 1) the classical pathway, via complexes of antibody, such as “natural” IgM and IgG antibodies and antigen (131, 135); 2) the lectin pathway, driven by mannose-binding lectin, whose expression on inflammatory and other cells promotes complement activation by binding to mannose on pathogen membranes (131, 135, 139); and 3) the alternative pathway, a constitutive mechanism, whereby baseline hydrolysis and cleavage of C3 complement component occurs and increases with tissue injury and inflammation by properdin and other inflammatory factors (131, 132). It is unknown whether SARS-CoV-2 infection activates the classical or lectin pathway. Data do not exist for the targeting of SARS-CoV-2.

FIGURE 3. Interactions between the components of the complement and coagulation cascades in driving thromboinflammation.
CoV-2 by natural IgM and IgG antibodies. Limited evidence from SARS-CoV-1 infection indicates suppression of mannose binding lectin by the virus as a means of immune evasion (139). It has been suggested (133, 134–136) that inflammation, tissue injury and coagulation associated with serious cases of COVID-19 may induce complement activation, in particular, via the alternative pathway. In addition, platelet activation can induce both the classical pathway (even without antibody-antigen interactions) and the alternative pathway (140) (FIGURE 3).

FIGURE 3 shows a schema of interactions between the coagulation pathway (activated initially by tissue inflammation, FIGURE 1) and complement pathways, adapted from previous models (128–132, 136, 137, 141, 142). In brief, the complement cascade results in cleavage of C3 to C3a and C3b. C3a, an immunity-stimulating ligand, activates inflammatory cells via C3a receptors (which are GPCRs). C3b drives cleavage of the C5 complement component into C5a and C5b. C5a acts via C5a receptors (also GPCRs), while C5b further drives the complement cascade, leading to generation of the membrane attack complex (MAC), which has cytotoxic effects on pathogens and damaged host cells and/or cells undergoing apoptosis (143). Proinflammatory effects of complement signaling are associated with TF production (128, 130, 132) and, hence, the extrinsic coagulation cascade. Thus, in hyperinflammation, proinflammatory effects of the complement cascade may exacerbate pathology. In addition, C3a, C5a, and MACs activate platelets (130, 132, 142, 144). Platelets can also form MACs locally via upstream complement cascade components (142, 144, 145), which can further drive platelet activation. Thus, complement activation and signaling may worsen coagulopathy and thromboinflammation.

In parallel, the coagulation cascade can increase complement activation (FIGURE 3). In addition to the induction of the classical and alternative complement cascade pathways by activated platelets, thrombin, and other components/products of the coagulation cascade (e.g., F, XII, and kallikrein) cleave C3 and C5, thereby activating the complement cascade and increasing levels of C3a, C5a, and MACs, all of which can promote thromboinflammation (FIGURE 3) (128–132). Plasmin also cleaves C3 and C5 (130–132). Platelet-activated thrombus formation, along with kinin-kallikrein pathway activation, enhance conversion of plasminogen to plasmin in the vicinity of thrombi (130, 132, 146). These mechanisms likely result in localized increase in complement activity in regions of the lung with high levels of immune infiltration and platelet activation, with MACs likely targeting dead/dying cells [such as large numbers of immune cells in severe COVID-19 cases (147, 148)]. This provides a possible explanation as to how SARS-CoV-2 results in increased complement action, but it is able to evade this response: complement actions become “overloaded” by the high degree of cell death and tissue injury and are unable to selectively target the pathogen. Secondary infection may be a further source of complement activation, wherein complement response to bacterial infection further enhances complement action and thromboinflammation.

FIGURE 4 shows a schema of thromboinflammation in COVID-19 with three main driver mechanisms: 1) ANG II-mediated pulmonary injury and inflammation; 2) activation of the coagulation cascade, contributing to thromboinflammation; 3) complement signaling, which synergizes with increased coagulation signaling and indirectly couples with ANG II-mediated effects, as each mechanism further contributes to inflammation and injury. In addition, as reviewed in Ref. 22, inflammatory effects can increase the expression and activity of ANG II-signaling components in fibroblasts and alveolar epithelial cells, further emphasizing the connection between the different mechanisms. On the basis of this framework, pharmacological agents that target thromboinflammation (TABLE 1) may reduce the contribution to inflammation in COVID-19 by multiple mechanisms, including the complement cascade. FIGURE 4 also shows that it is unclear whether SARS-CoV-2 directly activates the classical or lectin pathways and how the virus evades the complement response.

9. PROGRESSION OF THROMBOINFLAMMATION IN COVID-19 DISEASE

On the basis of published literature (as of 8/26/20) on COVID-19 pathobiology discussed above (25, 149–153) and additional sources reviewed in Ref. 22, we propose two models of COVID-19 disease progression (FIGURE 5). FIGURE 5, bottom, relates disease severity with thromboinflammation, (adaptive) immune response, and viral clearance in patients with severe disease, in which SARS-CoV-2 infection results in extensive pulmonary and systemic effects (154), perhaps leading to death or necessitating prolonged recovery in patients who survive (151, 155). Patients with mild to moderate disease (FIGURE 5, top) may have symptoms and signs that extend well beyond the ~14-day time frame during which most acute features typically resolve (151, 155). On the basis of multiple reports describing the kinetics of COVID-19 disease progression (149, 151–153), a typical timeframe involves a gradual onset of symptoms following exposure to the virus, typically taking ~5–7 days for symptoms to manifest. In mild cases, disease resolves.
within the next ~7 days, whereas in more severe cases, this “late” stage (~14–21 days), is marked by continued increase in disease severity and inflammation alongside progression to a hypercoagulable phase (149, 151), with processes, such as fibrosis occurring subsequently, over several weeks (151).

The course of disease involves initial exposure and infection in the upper respiratory tract (156). Infection subsequently spreads to the lungs, impacting the pulmonary alveoli, especially type II pneumocytes, which express ACE2 (22, 157). Involvement of the gastrointestinal tract is a relatively common feature of COVID-19, with gastrointestinal manifestations observed in both mild and severe cases, often occurring early in disease, including as a first symptom (158, 159). In severe COVID-19, the pulmonary infection is progressive and associated with widespread inflammation and cell injury and death, producing a vicious cycle of elevated inflammation, alveolar disruption, pulmonary edema/exudates, and pneumonia, as the alveolar epithelial barrier and surfactant production are compromised (22, 150, 151, 160). This hyperinflammatory phase can be associated with cytokine storm, leading to high levels of systemic inflammation, causing multiple organ failure, along with pulmonary injury that leads to ARDS (22, 25, 154, 161). Organs most severely impacted include the heart (162, 163), kidneys (164), and liver (165), among others (154). In addition, viremia can occur, leading to direct infection of these and other organs (149).

Coagulopathy increases with elevated inflammation and tissue injury that drive platelet activation, which, in turn, contributes to inflammation and thrombosis. As described above and in Figure 1, thrombosis and coagulopathy are followed by increased inflammation, in severe disease resulting in a phenotype akin to aspects of DIC (12). Reports indicate an early increase in coagulopathy in hospitalized patients, including at time of admission (6, 9), implying the need for rapid thromboprophylaxis, as per society guidelines (11, 28). Systemic
Coagulopathy has been reported in later stages of disease progression in patients with severe disease (3, 166, 167), but more data are needed to define the extent and frequency of this feature in such patients. By contrast, those with mild-moderate disease have less increase in coagulation, which likely resolves as inflammation and viral load subside. However, even in less severe cases, residual coagulopathy may occur, resulting, for example, in increased risk of stroke (168). Long-term monitoring of recovering COVID-19 patients is needed to clarify the frequency and severity of residual disease manifestations and the length of time needed for their resolution, as subsets of patient struggle with long-term complications (169, 170).

Comorbidities are a critical contributing factor in disease severity and outcome in COVID-19 patients (171–173). Several of these comorbidities (e.g., hypertension and cardiac disease, diabetes, obesity, and chronic lung injury) are associated with underlying inflammatory processes, and in many cases, elevated ANG II signaling (e.g., in cardiac remodeling and lung injury) [reviewed in (22)]. Many of these conditions and comorbidities are age-associated (174) and may accelerate or intensify positive feedback loops that involve inflammation, cell death, tissue injury, and thrombosis, leading to increased pathobiology (22). The resulting hyperinflammation likely inhibits adaptive immune response, thus blunting viral clearance and resolution of disease (22, 153, 174–176). Severe COVID-19 cases are associated with lymphopenia and, in particular, T-cell elimination and functional exhaustion, accompanying hyperinflammation (147, 148, 177), a likely mechanism through which

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**Table 1. Candidate drugs for repurposing, that target thromboinflammation in COVID-19 pathophysiology**

| Drug | Target/Mechanism | Cells Types in COVID-19 Pathobiology Potentially Impacted | Regulatory Status |
|------|------------------|----------------------------------------------------------|------------------|
| **Drugs Targeting Activation of Platelets by PAR and P2Y12 Receptors** | | | |
| Vorapaxar and atopaxar | PAR1 inhibitors | Platelets, endothelial cells, fibroblasts, epithelial cells, Megakaryocytes? Certain immune cells types? | Vorapaxar is FDA-approved for patients with myocardial infarction and peripheral arterial disease. Atopaxar is an investigational drug tested in Phase 2 trials. |
| Clopidogrel, prasugrel, canagrelor, ticagrelor | P2Y12 inhibitors | Platelets, endothelial cells, Dendritic cells, macrophages | FDA-approved for treatment and prevention of thromboembolic disorders |
| BMS-986120 | PAR4 inhibitor | Platelets, endothelial cells, epithelial cells, monocytes? Other immune cells? | Clinical trials in progress as an anti-platelet drug |
| **Drugs Targeting Thrombin Activity/Synthesis** | | | |
| Dagibatran, argatroban, bivalirudin, desirudin (and others) | Thrombin inhibitors | Cellular targets of thrombin (Platelets, endothelial cells, fibroblasts, epithelial cells, certain immune cells?) | FDA-approved for prophylaxis of thromboembolism |
| Rivaroxaban, apixaban, edoxaban (and others) | Factor Xa inhibitors | Cellular targets of thrombin | FDA-approved for prophylaxis of thromboembolism |
| Recombinant thrombomodulin | Restoration of protein C activation; inhibition of thrombin formation | Cellular targets of thrombin | Approved in Japan for sepsis-induced disseminated intravascular coagulation |
| Drotrecogin alfa (activated) | Recombinant protein C; inhibition of thrombin formation | Cellular targets of thrombin | FDA-approved for treatment of sepsis, but approval was subsequently withdrawn |
| Tifacogin (recombinant TFPI) | Inhibition of factor Xa, tissue factor | Cellular targets of thrombin | Tested in Phase 3 trials for sepsis, not currently approved |
**Mild/Moderate COVID-19 disease**

- **Initial exposure**
- **Upper respiratory infection**
- **Gastrointestinal effects?**
- **Mild pulmonary infection**
- **Abatement of illness, with viral clearance**
- **Resolution of lingering effects?**

**Disease severity**

**Inflammation + Thrombosis**

**Protective Immune Response**

- **Minimal underlying inflammation; absence of comorbidities**
- **Small/no increase in pulmonary coagulopathy and thrombosis**
- **Reduced coagulopathy with viral clearance and resolution of inflammation**

**Early Stages (~5-7 days)**

**Late Stages (~14-21 days)**

**Severe COVID-19 disease**

- **Initial exposure**
- **Upper respiratory infection**
- **Gastrointestinal effects**
- **Pulmonary infection increases in severity**
- **Systemic effects: multiple organ failure**

**Disease severity**

**Inflammation + Thrombosis**

**Protective Immune Response**

- **Inflammation from comorbidities**
- **Seroconversion and T cell infiltration**
- **Increased inflammation impedes immune response**

**Early Stages (~5-7 days)**

**Late Stages (~14-21 days)**

**FIGURE 5.** Mild-moderate and severe COVID-19: The balance between immune response, inflammation, and thrombosis (thromboinflammation). Disease course of COVID-19 as a function of time, with putative progression of inflammation, adaptive immune response, and viral clearance in parallel with coagulopathy and thrombosis for (top) more mild/moderate disease and (bottom) severe disease. This general framework is adapted from that presented by Romagnoli et al. (17) and highlights specific aspects of disease pathophysiology discussed in the text.
protective immune responses are blunted by excess inflammation in COVID-19. In milder cases, especially in patients without underlying comorbidities, slower, less extensive disease progression allows the mounting of an effective adaptive immune response, resulting in viral clearance and disease resolution before substantial tissue injury occurs. In addition to increased inflammation, comorbidities (e.g., diabetes), can also be associated with increased coagulopathy, as discussed above, thus giving thrombin inflammation a “head start”. Hence, in the “competition” between progressive infection/disease pathobiology versus (beneficial) immune response, comorbidities shift the “balance” toward more severe inflammation, coagulopathy, and disease progression.

These pathophysiological mechanisms also are consistent with the concept of “inflamming” (178, 179): the association of aging with elevated systemic inflammation and concurrently, weakened adaptive immune response, thereby, compromising the response to infection. This age-associated immune dysfunction/dysregulation is also predicted to increase the risk of cytokine storm and the likelihood of complications in COVID-19 (174). The framework that we propose predicts that immune-compromised individuals would likely be at elevated risk for thrombin inflammation and severe disease, because their weakened immune response is insufficient to impede the feedback loops that drive COVID-19 pathobiology (FIGURE 1) and lead to a hyperinflammatory phenotype before an effective adaptive immune response can be mounted (FIGURE 5, bottom). Cancer, another comorbidity that affects the immune system, enhances inflammatory signaling and likely increases susceptibility to COVID-19 (180), consistent with the framework presented in FIGURE 5.

An important implication of this model is that early therapeutic interventions that inhibit thrombosis and the feedback loops of inflammation (FIGURE 1) have the potential to reduce disease severity. This idea underscores the need for rapid testing and early diagnosis, providing a window for early administration of potentially beneficial therapies. Drugs that might inhibit both thrombosis and inflammation are listed in TABLE 1 and are discussed in the following section.

10. OPPORTUNITIES FOR THERAPEUTIC INTERVENTION VIA REPURPOSING EXISTING AGENTS

TABLE 1 and FIGURE 6 highlight drugs tested in human trials [some that are Food and Drug Administration (FDA)-approved] that may inhibit thrombin inflammation. These include antiplatelet drugs that inhibit GPCRs for ADP and thrombin on platelets and other cells relevant to COVID-19 pathobiology, as well as inhibitors of coagulation cascade components.

PAR1 and P2Y receptors and their contribution to platelet activation, thrombosis, and cell/tissue injury can be inhibited pharmacologically (TABLE 1). Vorapaxar, an FDA-approved PAR1 antagonist, is prescribed to patients with peripheral arterial disease and myocardial infarction. Inhibiting PAR1 has the potential to reduce thrombosis in COVID-19, through actions on platelets and other cell types. PAR1 inhibition by vorapaxar increases bleeding risk (181), an important concern if patients are also treated with anticoagulants, as noted in guidelines on thromboprophylaxis in COVID-19 (11, 28). However, vorapaxar could provide a novel approach to treat and perhaps lessen the severity of COVID-19. Results showing efficacy of vorapaxar in animal studies could lead to the initiation of clinical trials in COVID-19 patients. Atolexapar, another PAR1 inhibitor, has been tested in Phase 2 trials for coronary artery disease and acute coronary syndrome and appears to have similar efficacy but fewer bleeding-associated adverse effects than vorapaxar (34). Moreover, multiple FDA-approved P2Y12 antagonists (e.g., clopidogrel, prasugrel, cangrelor, and ticagrelor) inhibit platelet activation and are used to treat and/or prevent a variety of cardiovascular disorders (182). Such drugs may also be useful for treating COVID-19 patients.

In addition to inhibition of platelet receptors, other therapeutic opportunities are suggested by the ideas discussed above. A recombinant form of APC [drotrecogin alfa (activated)] was FDA-approved for treating sepsis. However, the increased risk of bleeding with protein C therapy (183) and the lack of improvement in mortality in patients with severe sepsis in subsequent trials (84) resulted in the drug’s withdrawal from the market. Thus, the use of drotrecogin alfa is challenging, but its blunting of thrombin-mediated effects provides a mechanistic rationale to consider this possibility for COVID-19. A similar rationale exists for use of recombinant thrombomodulin (ART-123), which is approved in Japan for sepsis-related DIC (184). However, a recent multinational trial of ART-123 in sepsis failed to show significant reduction in mortality (85). PAR4 inhibition is another possible means to treat thrombosis in COVID-19 with potential therapeutic effects on platelets and other cell types. PAR4 has a more selective tissue expression profile than PAR1; a PAR4 antagonist with antithrombotic effects is under investigation (BMS-986120) and has been tested in phase-1 trials (68).

Additional agents to blunt the pathological effects of thrombin in COVID-19 (FIGURES 1, 2A, and 6, and TABLE 1) include inhibitors of thrombin (e.g., dabigatran and argatroban) and Factor Xa (e.g., rivaroxaban and apixaban). These orally administered drugs generally do not require the close monitoring needed with administration of heparin (185). On the basis of recent guidelines
indicating the need for prophylaxis of thromboembolism in patients with severe COVID-19, even after hospital discharge (28), clinical trials are needed to evaluate these drugs. In addition to approved inhibitors of the coagulation cascade, tifacogin, a recombinant TF pathway inhibitor (TFPI) has been tested in Phase 3 clinical trials for sepsis (186) but has not been approved. TFPI inhibits Factor Xa and actions of TF in driving Factor X activation (inhibiting TF–Factor VIIa and prothrombinase complexes), thereby reducing thrombin formation (187, 188).

One might also consider other approved agents that blunt platelet activity, e.g., GPIIb/IIIa inhibitors and aspirin. However, we focus here on approaches more likely to blunt multiple aspects of COVID-19 pathobiology, i.e., those targeting purinergic and thrombin signaling (FIGURE 6; TABLE 1). Factor Xlla inhibitors have shown promise in preclinical studies (189) but are still investigational compounds and not included in TABLE 1. Tranexamic acid, an analog of lysine and an antifibrinolytic agent, has been suggested as a therapeutic candidate for COVID-19 (190), possibly mitigating activation of SARS-CoV-2 by plasmin (9), preventing excessive fibrinolysis and reducing bleeding risk, potentially in combination with anticoagulants, so as to also blunt coagulopathy.

Recent articles on complement signaling in SARS-CoV-2 (134–136) have suggested the potential of inhibitors of the complement cascade as candidate therapeutics. Given the many unknowns discussed above regarding mechanisms of complement activation in SARS-CoV-2 and the extent/mechanisms of viral evasion of complement response, this aspect of COVID-19 pathophysiology requires further study.

Two randomized control trials (RCTs) are under way for the treatment of patients with COVID-19 using the P2Y12 inhibitor clopidogrel (NCT04409834 [not yet recruiting], NCT04333407 [recruitment ongoing]). Those open-label studies compare clopidogrel with other agents, such as aspirin or heparin. In addition, four randomized trials are recruiting patients, to compare rivaroxaban with other anticoagulants (NCT04351724, NCT04324463, NCT04394377, and NCT04333407). Therefore, only two drugs listed in TABLE 1 are currently being tested in patients with COVID-19. By contrast, >30 trials are under way that involve the use of forms of heparin and/or aspirin or other anticoagulants. Because even within a drug class, one can observe differences in responses, e.g., with different P2Y12 inhibitors (191), we propose that trials are needed to evaluate multiple platelet-targeted (and related) drugs, especially to identify the most effective drugs and therapeutic regimens. On the basis of the mechanistic discussions in earlier sections, possible disadvantages of P2Y12 antagonists in COVID-19 include activation of platelets by ADP is largely a secondary effect to activation by thrombin and
thrombin is likely to mediate a wider range of pathological effects in cell types besides platelets. Thus, drugs that target the formation or actions of thrombin are potentially preferable for mitigating COVID-19 thromboinflammation. Moreover, antiplatelet agents discussed in this text may not address other features of COVID-19 pathobiology, such as additional sources of inflammation, implying the potential value of combining antiplatelet drugs with anti-inflammatory and other medications. Perhaps, combination therapies using multiple anticoagulant/antiplatelet agents to inhibit platelet activation and coagulopathy might also be useful, such as combining purinergic receptor and PAR inhibitors, as has been proposed in other settings (192). Careful monitoring of bleeding risk will be critical, especially because of intrapulmonary hemorrhage and features of DIC in severe cases of COVID-19 (12). Pending completion of RCTs, retrospective studies of COVID-19 patient records may be of value, to indicate whether patients already being treated with antiplatelet or anticoagulant medications experience different outcomes compared with those not on such medications.

Preclinical studies using animal models have the potential to shed light on the utility of the different treatment options listed in Table 1, in particular, to explore various strategies for mitigating thromboinflammation. Use of animal models to provide preclinical guidance for clinical studies in COVID-19 has been challenging (reviewed in Ref. 193). Models in rhesus monkeys (194), Syrian golden hamsters (195), and mice expressing human ACE2 (196) replicate aspects of COVID-19 in humans, including fever, weight loss, and pulmonary injury. However, data are not available regarding thrombosis and related phenomena in most of these models. Recent data (197) in a mouse model with transgenic expression of human ACE2 provide the first evidence of thrombosis in an animal model of COVID-19 (along with severe pneumonia), suggesting the value of this model in preclinical studies of thromboinflammation. A key priority is to clarify which animal models best recapitulate aspects of coagulopathy and thromboinflammation observed in patients. Data from such studies would not only help validate these models but also provide a means to test a variety of potential regimens with candidate drugs.

11. CONCLUSIONS

The COVID-19 pandemic has created an urgent need for therapies that target the SARS-CoV-2 virus, such as antiviral therapies and vaccines, along with approaches that mitigate the pathological effects of the disease on host cells and tissues. In this review, we focus on mechanisms that mediate the pathophysiology of SARS-CoV-2 infection in hosts, in particular, as related to thromboinflammation, mediated by signaling via thrombin and ADP/ATP. The model of COVID-19 pathobiology that we discuss (Figures 1 and 2) describes how pulmonary inflammation/injury leads to coagulopathy, which, in turn, further exacerbates injury in the lungs and other tissues, including through actions by and coupling with other inflammatory mechanisms, in particular, the complement cascade. Our analysis highlights the therapeutic potential of a range of approved and experimental drugs (Table 1, Figure 6), which inhibit purinergic signaling, thrombin signaling, or the coagulation cascade, as candidates for repurposing to address multiple aspects of COVID-19 pathophysiology. Such drugs have the potential to inhibit platelet-associated thrombotic processes, as well to blunt signaling in cells that contribute to pathophysiology. These approaches provide an opportunity to target key contributors to COVID-19 morbidity and mortality and, thereby, to improve the clinical course and outcome of patients with SARS-CoV-2 infection.
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## DISCLOSURES

P.A.I. is not currently, but within the past 3 years, has served as a consultant or received research support from Merck, Pfizer, and Bristol Myers Squibb. K.S. has no conflicts of interest.

## AUTHOR CONTRIBUTIONS

K.S. prepared figures; K.S. and P.A.I. drafted manuscript; K.S. and P.A.I. edited and revised manuscript; K.S. and P.A.I. approved final version of manuscript.

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