ACS Infectious Diseases

pubs.acs.org/journal/aidcbc

Perspective

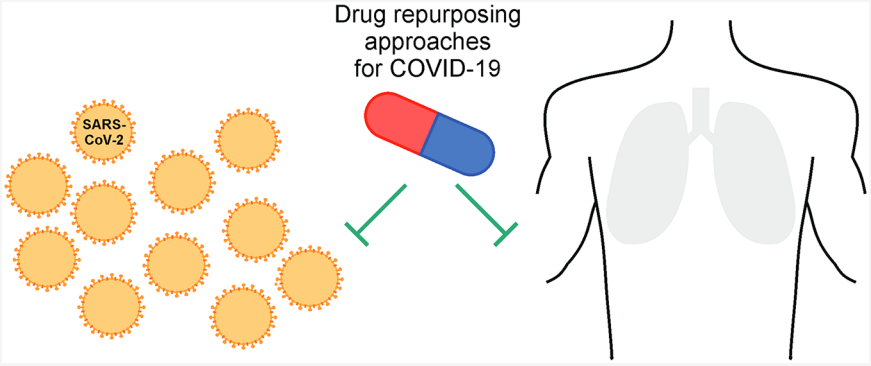
Old Drugs for a New Virus: Repurposed Approaches for Combating COVID-19

Sirle Saul and Shirit Einav*

Cite This: https://dx.doi.org/10.1021/acsinfecdis.0c00343

Read Online

ACCESS | Metrics & More | Article Recommendations



ABSTRACT: There is a large global unmet need for effective countermeasures to combat severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), the causative agent of coronavirus disease 2019 (COVID-19). The development of novel antiviral drugs is expensive and too slow to meet the immediate need. The repurposing of drugs that are approved or are under advanced clinical investigation provides a cost- and time-effective therapeutic solution. This review summarizes the major repurposed approaches that have been proposed or are already being studied in clinical trials for COVID-19. Among these approaches are drugs that aim to reduce SARS-CoV-2 replication by targeting either viral enzymatic functions or cellular factors required for the viral life cycle. Drugs that modulate the host immune response to SARS-CoV-2 infection by boosting it to enhance viral clearance or by suppressing it to prevent excessive inflammation and tissue injury represent another category. Lastly, we discuss means to discover repurposed drugs and the ongoing challenges associated with the off-label use of existing drugs in the context of the COVID-19 outbreak.

KEYWORDS: *antiviral drugs, repurposing, COVID-19, SARS-CoV-2, small molecule inhibitors, anti-inflammatory drugs, direct acting antivirals, host-targeted approaches*

The emergence of the novel, highly pathogenic severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), the causative agent of coronavirus disease 2019 (COVID-19), has evolved into a global pandemic. While the majority of infected individuals remain asymptomatic or experience a mild to moderate, self-limited illness, a fraction of the patients progresses to develop severe infection that is often complicated by respiratory failure, cardiac complications, and sometimes death.[1] Moreover, it appears that COVID-19 may be causing long-lasting lung and heart injury.[2,3] As of the 16th of July 2020, more than 13.9 million COVID-19 cases and over 590,000 deaths have been reported worldwide, and the pandemic has taken a huge toll on the health care system and global economy. Measures such as social distancing and home isolation provide a temporary solution to control viral spread; however, in the absence of an effective approved vaccine, the entire world population continues to be at risk for SARS-CoV-2 infection. Hence, there is an urgent need for approaches to treat COVID-19 patients and prevent the short- and long-term complications associated with this disease.

As with other newly emerging viruses that have caused outbreaks within the past two decades, including the coronaviruses implicated in severe acute respiratory syndrome (SARS) and Middle East respiratory syndrome (MERS) as well as the unrelated Ebola (EBOV, filovirus) and Zika (ZIKV, flavivirus) viruses, no approved, clinically effective therapy is available for treating SARS-CoV-2. Since the development of new drugs typically takes 8 to 12 years,[4] this approach cannot

Received: May 22, 2020
Published: July 20, 2020

provide an immediate solution in the context of such outbreaks. In contrast, repurposing already approved or investigational drugs can significantly accelerate the development and deployment of therapies for such emerging viral infections. Repurposing existing drugs that have already been shown to be safe in humans, albeit in a different disease model(s), reduces the clinical risk and facilitates a faster path into the clinic. Moreover, relying on existing human safety, pharmacology, and toxicology data reduces the substantial cost associated with the development of a new drug (over two billion dollars on average).[4]

Multiple repurposed drugs are currently being studied for COVID-19 in clinical trials, and many more are being investigated in preclinical models. One category of such drugs aims to reduce viral replication and can be classified into direct acting antivirals (DAAs) that target viral enzymes (primarily the viral proteases and polymerase) and host-targeted drugs that inhibit cellular functions required for viral replication. Inhibitors that modulate virus-induced inflammation and tissue injury constitute another category. The major categories of repurposed approaches that are already being studied or have been proposed for COVID-19 treatment with representative examples are discussed below and illustrated in summary tables and figures.

REPURPOSED APPROACHES THAT PRIMARILY INHIBIT VIRAL REPLICATION

Direct Acting Antivirals (DAAs). SARS-CoV-2 is an enveloped, positive-sense, single-stranded RNA virus whose replication and transcription are catalyzed by an RNA-dependent RNA polymerase (RdRp). The two SARS-CoV-2 proteases, papain like protease (PL^{pro}) and the main protease (M^{pro}), play a key role in replication and transcription via processing of the viral polyproteins.[5] As with other viral infections, due to their known biological functions and active enzymatic sites, the SARS-CoV-2 polymerase and proteases are intuitive therapeutic targets.

Polymerase Inhibitors. In the past decade, several chemically distinct nucleoside and nucleotide analogues that inhibit the polymerase of multiple unrelated RNA viruses have been developed. One example is remdesivir, an investigational nucleoside analogue originally developed as a therapy for EBOV disease. Remdesivir gets incorporated into nascent viral RNA chains and results in their premature termination.[6] While not previously approved, intravenous administration of remdesivir was found to be highly effective against EBOV disease in nonhuman primates (NHP).[6] Nevertheless, in a randomized controlled (PALM) clinical trial for Ebola (NCT03719586), mortality rates were higher with remdesivir treatment than with monoclonal antibodies.[7]

Remdesivir has previously demonstrated *in vitro* activity against SARS-CoV and MERS-CoV,[8] and it reduced disease severity, viral replication, and lung injury in mouse (SARS-CoV) and NHP (MERS-CoV) models.[8,9] Recently, remdesivir was shown to inhibit RNA synthesis by a purified recombinant SARS-CoV-2 RdRp[10] and to suppress SAR-CoV-2 infection in both Vero E6 (green monkey kidney) (EC_{50} = 0.77 μM) and Huh7 (human hepatoma) cells.[11] Moreover, remdesivir demonstrated potent inhibition of SARS-CoV-2 replication in human lung cells and primary human airway epithelial cultures (EC_{50} = 0.01 μM).[12] Therapeutic administration of remdesivir in mice infected with SARS-CoV encoding the RdRp of SARS-CoV-2 reduced viral load in the lungs and improved pulmonary function relative to the vehicle.[12] In an NHP SARS-CoV-2 infection model that causes lower respiratory tract disease, remdesivir treatment provided protection from respiratory signs and reduced pulmonary infiltrates on radiographs.[13] Viral titers in bronchoalveolar lavages, lung viral loads, and lung injury were significantly reduced in remdesivir-treated animals relative to controls.[13]

A remdesivir compassionate use study in 53 patients hospitalized for severe COVID-19 demonstrated clinical improvement in 68% of the patients;[14] however, no control arm was included in this study. In a double-blind, placebo-controlled, multicenter trial in patients with severe COVID-19, intravenous remdesivir treatment did not improve the time to clinical improvement; however, the study did not reach its target enrollment and was stopped.[15] In contrast, preliminary data from the double-blind, randomized, placebo-controlled adaptive COVID-19 treatment trial sponsored by the US National Institute of Health (NIH) (NCT04280705) involving 1059 hospitalized adult patients (538 assigned to remdesivir and 521, to placebo) with lower respiratory COVID-19 infection showed that a 10-day remdesivir treatment course shortened the median recovery time relative to the placebo from 15 to 11 days ($p < 0.001$) and reduced the estimates of mortality by 14 days from 11.9% to 7.1% with no increase in serious adverse events.[16] Moreover, a randomized, open-label, phase 3 trial involving 397 hospitalized patients with confirmed SARS-CoV-2 infection with oxygen requirements (but not mechanically ventilated) demonstrated comparable efficacy of 5- and 10-day remdesivir treatment courses with clinical improvement of 2 points or more on the ordinal scale detected in 64% and 54% of patients, respectively. Collectively, these findings suggest that remdesivir is moderately effective and is generally safe and led to the recent emergency use approval of this drug for the treatment of COVID-19 by the US Food and Drug Administration (FDA).

The guanine analogue favipiravir, which is approved in Japan for the treatment of influenza A virus infection, is another example for a broad-spectrum polymerase inhibitor.[17] Favipiravir moderately inhibits replication of several RNA viruses *in vitro*,[18] albeit with high EC_{50} values: 61.88 and 67 μM in the case of SARS-CoV-2 and EBOV, respectively.[11] High drug concentrations are therefore required to maintain levels above these EC_{50} values but appear to be effectively achieved and well tolerated. Moreover, by self-inhibiting its metabolism into an inactive oxidative metabolite, favipiravir increases the plasma parent/inactive metabolite ratio, thereby facilitating increased cellular uptake and compensating for the high EC_{50} values.[19] Indeed, favipiravir protected 100% of mice from an EBOV challenge[20] and was shown to prolong survival and reduce viral load in a retrospective EBOV study during the 2014−2015 outbreak.[21] In a prospective, randomized, multicenter study (ChiCTR200030254), favipiravir demonstrated no effect in critically ill COVID-19 patients; however, it improved clinical recovery at day 7 in moderate cases relative to Arbidol, a fusion inhibitor also used for influenza treatment (71.4% vs 55.9%, p = 0.019). In a clinical trial involving 80 patients in China (ChiCTR2000029600), the combination of favipiravir with interferon-α reduced SARS-CoV-2 clearance time relative to lopinavir/ritonavir plus interferon-α in the control arm (median of 4 vs 11 days).[22] Notably, while the adverse effects attributed to favipiravir treatment were mild and manageable,[23] caution is needed due to its teratogenic risk[18] and potential for drug−drug interactions.[19] Favipiravir is

https://dx.doi.org/10.1021/acsinfecdis.0c00343
ACS Infect. Dis. XXXX, XXX, XXX−XXX

currently being studied in several open label phase 2 clinical trials (NCT04358549, NCT04346628), and it has been recently approved for COVID-19 treatment in China and India.

A third investigational RdRp inhibitor, the adenosine analogue galidesivir, has also shown activity against a wide range of viruses (including coronaviruses) and protected NHP from Marburg virus (filovirus) infection.[24] Galidesivir has recently been shown to tightly bind to SARS-CoV-2 RdRp,[25] and its clinical safety and efficacy are currently being studied in a randomized, double-blind, placebo-controlled clinical COVID-19 trial (NCT03891420).

Viral Protease Inhibitors. Another attractive antiviral target is the viral main protease (M^{pro}). This chymotrypsin-like cysteine protease proteolytically cleaves protein precursors necessary for viral RNA replication and production of infectious viral particles.[26] Several protease inhibitors have been proposed and/or studied for the treatment of COVID-19. One example is the lopinavir/ritonavir combination, which is approved for the treatment of human immunodeficiency virus (HIV-1) infection. While treatment with these drugs individually has demonstrated moderate anti-SARS-CoV-2 activity in Vero E6 cells (EC_{50} = 5.73 μM, lopinavir, and EC_{50} = 8.63 μM, ritonavir),[27] their combination showed no benefit beyond supportive care in a randomized, controlled, open-label clinical trial in hospitalized patients with severe COVID-19.[28] The ability of the lopinavir/ritonavir combination to reduce SARS-CoV-2 load in the respiratory tract in patients with mild COVID-19 is currently being studied (NCT04307693). Nelfinavir, another oral protease inhibitor approved for HIV-1 treatment, showed antiviral activity against SARS-CoV[29] and SARS-CoV-2 (EC_{50} = 0.77 μM, CC_{50} > 20 μM) in Vero E6 expressing TMPRSS2 cells.[27]

A potentially more attractive repurposed option that also targets the viral protease is the organoselenium compound, ebselen. Ebselen mimics the action of glutathione peroxidase and thus has anti-inflammatory, antioxidant, and cytoprotective properties.[30] This compound has demonstrated no side effects when tested at high oral doses in humans in several phase 2/3 clinical studies for various indications including hearing loss,[31] diabetes,[32] and stroke.[33] Ebselen emerged as the top hit in a high-throughput activity screening assay of a library of 10,000 approved and investigational drugs, where it demonstrated the strongest inhibition of SARS-CoV-2 M^{pro} activity (IC_{50} = 0.67 μM) and was shown to covalently bind to cysteine 145 within the catalytic dyad of the viral M^{pro}.[34] Moreover, it showed a moderate anti-SARS-CoV-2 activity in Vero cells, with an EC_{50} of 4.67 μM,[34] which could likely be achieved with the previously tested oral dosing. Notably, since it is nonselective, the likelihood of the emergence of viral resistance to ebselen is predicted to be lower than to other protease inhibitors.

Host-Targeted Antiviral Approaches. Viruses replicate within the host cell while relying on cellular machineries for multiple distinct steps in their life cycle. Repurposing inhibitors that target cellular functions required for SARS-CoV-2 infection is thus an attractive approach that may have the added benefit of a higher genetic barrier to the emergence of resistance.[35]

Inhibitors of Cellular Proteases. Effective SARS-CoV-2 entry requires proteolytic processing for activation of the spike (S) glycoprotein by the cellular endosomal cysteine proteases cathepsin B and L (CatB/L) and the transmembrane serine protease 2 (TMPRSS2).[36] TMPRSS2 is also required for the activation of the S protein of SARS-CoV and MERS-CoV[37,38] and was shown to be essential for viral spread and pathogenesis in animal models of these infections.[39] Pharmacological targeting of TMPRSS2 therefore represents an attractive approach to block the entry of coronaviruses.

Camostat mesylate is an oral serine protease inhibitor that has been clinically used in Japan for the treatment of pancreatitis.[40] Camostat was shown to reduce MERS-CoV, SARS-CoV, and SARS-CoV-2 entry in a lung epithelial cell line (Calu-3) and in primary human lung cells with no apparent cytotoxic effects.[36] In a mouse model, camostat was effective in protecting mice against death due to a lethal SARS-CoV infection, with a survival rate of ∼60% relative to vehicle-treated controls.[41] Several randomized, placebo-controlled clinical trials to determine the effect of camostat on COVID-19 infection have been initiated (NCT04353284, NCT04321096).

A closely related compound, nafamostat mesylate, is another safe serine protease inhibitor. It is approved in Japan as an anticoagulant and antipancreatitis agent. Nafamostat was previously shown to inhibit the TMPRSS2-dependent entry of MERS-CoV in Calu-3 cells,[42] and its activity against SARS-CoV-2 entry and overall infection was recently shown to be superior to that of camostat (EC_{50} = 5 nM vs 87 nM, respectively).[43] A randomized, placebo-controlled clinical trial to assess the efficacy of intravenous nafamostat treatment in COVID-19 patients was recently initiated (NCT04352400).

Kinase Inhibitors. Viruses have been shown to hijack a large number of host kinases at distinct stages of their life cycle (reviewed in ref 44). Cellular kinase inhibitors approved as anticancer or anti-inflammatory drugs therefore represent another category of compounds with a great potential to be repurposed into antivirals. Beyond their effect on viral replication, many of these kinase inhibitors have also been shown to reduce inflammation and tissue injury, such as by suppressing the production of cytokines (interleukin 6 (IL-6), tumor necrosis factor α (TNF-α) and transforming growth factor β (TGF-β)) implicated in inflammation and lung fibrosis. These kinase inhibitors may thus potentially achieve a dual role in the treatment of COVID-19 patients.

Members of the Src family of kinases (SFKs) and the Abl family of nonreceptor tyrosine kinases have been implicated in the life cycle of multiple viruses (reviewed in refs 44 and 45). RNAi-mediated depletion of the Src family proteins Lyn and Fyn reduced MERS-CoV infection, suggesting these proteins may be required for coronavirus infection.[46] The approved anticancer drug dasatinib and the investigational (phase 3) compound saracatinib, with activity against both SFKs and c-Abl (cellular Abelson tyrosine kinase), have been shown to inhibit a broad-spectrum of viruses *in vitro*.[47,48] Dasatinib was found to inhibit SARS-CoV and MERS-CoV with EC_{50}'s of 2.1 and 5.4 μM in Vero E6 cells.[49] Saracatinib was shown to inhibit early stages of the MERS-CoV life cycle (EC_{50} of 2.9 μM in Huh7 cells) and was synergistic when combined with gemcitabine, which also inhibits MERS-CoV replication.[46] The inhibition of actin motility was proposed as one mechanism of antiviral action of Src and/or Abl inhibitors in other viral infections;[50] however, the mechanism of antiviral action and the molecular targets (Abl only or Abl plus Src) that mediate the antiviral action of these drugs in coronavirus infections remain to be uncovered.

Similarly, imatinib and/or nilotinib, approved anticancer c-Abl inhibitors lacking anti-Src family kinase activity, also

https://dx.doi.org/10.1021/acsinfecdis.0c00343
ACS Infect. Dis. XXXX, XXX, XXX−XXX

inhibited the replication of coronaviruses[49] and other RNA viruses[51] in cultured cells. Nilotinib and imatinib inhibited SARS-CoV and/or MERS-CoV infections with an EC_{50} at a micromolar range (EC_{50}'s ∼ 10−20 μM).[49] Imatinib was shown to inhibit the fusion of virions at the endosomal membrane by targeting Abl2 kinase,[52] and it was therefore proposed that it may inhibit the function, localization, or activity of TMPRSS2.[52] In a murine model of vaccinia virus, imatinib effectively reduced viral load, viral spread, and mortality. In contrast, dasatinib did not protect mice but rather induced immunosuppression, raising a concern about its utility in controlling viral infections *in vivo*.[50] The activity of these and other kinase inhibitors against SARS-CoV-2 is currently being studied in preclinical models. Moreover, several studies aimed at testing the safety and efficacy of imatinib in COVID-19 patients are ongoing.

Several kinases in the mitogen-activated protein kinase (MAPK)/extracellular signal-regulated kinase (ERK) pathway have been shown to be upregulated and essential for the replication of various viruses (reviewed in ref 44). Infection with MERS-CoV has been shown to activate this pathway and to be suppressed by two anticancer compounds, trametinib, an FDA-approved MEK1/2 inhibitor, and selumetinib, a recently approved MEK1/ERK1/2 inhibitor.[53] The anti-SARS-CoV-2 potential of these compounds is currently being studied in preclinical models.

A recent phosphoproteomics analysis revealed changes in the phosphorylation patterns of cellular and viral proteins during SARS-CoV-2 infection.[54] Among these alterations is the activation of the p38/MAPK signaling pathway, which mediates the response to stress stimuli and was previously shown to be similarly activated in cells infected with SARS-CoV and MERS-CoV.[53,55] Pharmacological inhibition of this pathway by either gilteritinib, an FDA approved anticancer inhibitor of AXL, or ralimetinib, a p38 inhibitor under phase 2 clinical trials for cancer, demonstrated potent anti-SARS-CoV-2 activity (EC_{50} = 0.807 μM and EC_{50} = 0.873 μM, respectively).[54] Prominent activation of PIKfyve (phosphatidylinositol 3-phosphate 5-kinase), a lipid kinase that regulates endosomal trafficking and endomembrane homeostasis, was also detected.[54] Apilimod, an investigational oral anti-inflammatory and anticancer (phase 2), safe drug that was developed to inhibit IL-12/23 and later found to target PIKfyve,[56] was shown to suppress the entry of SARS-CoV-2 pseudovirions[57] and overall replication (EC_{50} < 0.08 μM in Vero E6 cells; EC_{50} = 0.007 μM in A549 cells expressing ACE2).[54] The safety and efficacy of apilimod is being studied in a phase 2 randomized, double-blind, placebo-controlled trial in mild COVID-19 patients in an outpatient setting (NCT04446377). Lastly, SARS-CoV-2 infection was shown to promote the shutdown of mitotic kinases, resulting in cell cycle arrest.[54] Pharmacological inhibition of cyclin-dependent kinases by dinaciclib, an anticancer drug (phase 3), exhibited potent anti-SARS-CoV-2 activity (EC_{50} = 0.127 μM in Vero E6 cells; EC_{50} = 0.032 μM in A549 cells expressing ACE2).[54] However, serious hematological toxicities may limit the use of dinaciclib as an antiviral drug.[58]

Host-Targeted Drugs with Complexed or Unclear Mechanism of Action (MOA). Among the approved drugs that exhibit anti-SARS-CoV-2 activity are agents for which the mechanism of antiviral action is complex and unclear. In general, these drugs have multifunctional activities, and their antiviral effect is unlikely to be mediated by the same mechanism that is relevant to the primary indication for which they are approved.

One such example is niclosamide, an FDA-approved drug used to treat tapeworm infection. Niclosamide has previously shown activity against various viruses including MERS-CoV and SARS-CoV with EC_{50} values of less than 0.1 μM in Vero cells.[59−61] It has recently emerged as a potent inhibitor of SARS-CoV-2 infection via high-throughput screening of a drug library (EC_{50} = 0.28 μM),[62] and it is currently being studied in animal models of SARS-CoV-2. While niclosamide exerts its antihelmintic effect by inhibiting oxidative phosphorylation and stimulating adenosine triphosphatase activity in the mitochondria,[63] it has been shown to regulate multiple signaling pathways and biological processes.[64] The precise mechanism of antiviral action of this multifunctional drug thus remains unknown. A clinical trial to investigate the efficacy of niclosamide in combination with diltiazem (a calcium channel blocker) for the treatment of mild COVID-19 in patients with comorbidities has been initiated (NCT04372082).

Other antiparasitic drugs demonstrating anti-SARS-CoV-2 activity are chloroquine and its less toxic derivative hydroxychloroquine.[65] Chloroquine and hydroxychloroquine are approved for the treatment of malaria and inflammatory diseases, such as lupus and rheumatoid arthritis. These drugs have shown activity against multiple viruses *in vitro* but did not improve virological or clinical outcome in human studies in other viral infections, such as dengue[66−68] and flu.[69] They are thought to exert their antiviral effect by preventing endosomal acidification required for virus/cell fusion and by blocking autophagosome−lysosome fusion.[70] Yet, additional mechanisms, such as interference with the glycosylation of angiotensin-converting enzyme 2 (ACE2), the cellular receptor of coronaviruses, have also been proposed.[71] Although chloroquine demonstrated *in vitro* activity against SARS-CoV and MERS-CoV at a concentration that can be achieved in serum by the approved oral dosing,[72,73] it did not effectively reduce viral replication in SARS-CoV infected mice.[74]

Both drugs were recently shown to moderately inhibit SARS-CoV-2 replication in Vero E6 cells with variable EC_{50} values across different studies (0.72−7.36 μM, chloroquine; 4.5−12.96 μM, hydroxychloroquine).[11,75,76] On the basis of these *in vitro* data and their anti-inflammatory properties, the utility of hydroxychloroquine and chloroquine for the prevention and treatment of COVID-19 has been assessed in several studies (reviewed in 77). The majority of retrospective, observational studies in hospitalized COVID-19 patients have demonstrated no benefit with hydroxychloroquine treatment.[78] A reduction in COVID-19 associated mortality was detected in one study in hospitalized patients who received the drug early during their disease course, but only when controlling for COVID-19 risk factors.[79] Moreover, concerns have been raised about the safety of this approach and, specifically, its association with cardiac toxicity. Indeed, a clinical trial evaluating the safety and efficacy of chloroquine in patients with severe COVID-19 (NCT04323527) was terminated early due to prolongation of the QTc interval leading to increased cardiac toxicity and fatality rates.[80] As a result, prospective COVID-19 clinical trials with hydroxychloroquine and chloroquine have been terminated, and the emergency use authorization for these drugs has been revoked by the FDA.[81] The discussed repurposed approaches that primarily inhibit viral replication are illustrated in Table 1 and Figure 1.

https://dx.doi.org/10.1021/acsinfecdis.0c00343
ACS Infect. Dis. XXXX, XXX, XXX−XXX

Table 1. Repurposed Approaches That Inhibit Viral Replication[a]

		name	type	original indication	cell line	EC50	mouse model	NHP	target for antiviral activity	COVID-19 status
DAAs	viral polymerase inhibitors	remdesivir (GS-5734)	nucleoside analogue	investigational (Ebola)	Vero E6	0.77 μM	SARS-CoV/RdRp of SARS-CoV-2 *(reduced lung viral load)*	SARS-CoV-2 *(reduced viral load and lung injury)*	RdRp	emergency use approval by US FDA
					HAE	0.01 μM				
		favipiravir (T-705)	nucleoside analogue	influenza A virus (approved in Japan)	Vero E6	61.88 μM	ND	ND	RdRp	phase 2/3, approved in China and India
		galidesivir (BCX4430)	nucleoside analogue	investigational	ND	ND	ND	ND	RdRp	phase 1
	viral protease inhibitors	lopinavir (LPV) /ritonavir (RV)	protease inhibitor	HIV-1 infection	Vero E6	5.73 μM (LPV) 8.63 μM (RV)	ND	ND	Mpro	phase 2
		nelfinavir	protease inhibitor	HIV-1 infection	Vero E6	0.77 μM	ND	ND	Mpro	NI
		ebselen	organoselenium compound	investigational (arthritis, stroke etc)	Vero	4.67 μM	ND	ND	Mpro	NI
host-targeted antiviral approaches	cellular protease inhibtors	camostat mesylate (FOY-305)	serine protease inhibitor	pancreatitis (Japan)	Calu-3	0.087 μM	SARS-CoV *(protects mice from death)*	ND	TMPRSS2	phase 2/3
		nafamostat mesylate	serine protease inhibitor	anticoagulant and anti-pancreatitis agent (Japan)	Calu-3	0.005 μM	ND	ND	TMPRSS2	phase 2/3
	cellular kinase inhibitors	dasatinib	anticancer drug	leukemia	Vero E6	2.1 μM (SARS-CoV) 5.4 μM (MERS-CoV)	ND	ND	Src kinase + Abl kinase	NI
		saracatinib	investigational (phase 3)	Alzheimer's disease, cancer	Huh7	2.9 μM (MERS-CoV)	ND	ND	Src kinase + Abl kinase	NI
		imatinib (STI-571)	anticancer drug	leukemia	Vero E6	9.8 μM (SARS-CoV-2) 17.7 μM (MERS-CoV)	ND	ND	Abl kinase	phase 3
		nilotinib (AMN107)	anticancer drug	leukemia	Vero E6	μM range	ND	ND	Abl kinase	NI
		trametinib (GSK1120212)	anticancer drug	melanoma	ND	ND	ND	ND	MEK1/2 inhibition	NI
		selumetinib (AZD6244)	anticancer drug	neurofibromatosis type 1	ND	ND	ND	ND	MEK1/ERK1/1/2 inhibition	NI
		gilteritinib	anticancer drug	acute myeloid leukemia	Vero E6	0.807 μM	ND	ND	AXL kinase	NI
		ralimetinib (LY2228820)	investigantional (phase 2)	glioblastoma, ovarian cancer	Vero E6	0.873 μM	ND	ND	p38 MAPK	NI
		apilimod (STA-5326)	investigational (phase 2)	anti-inflammatory, anti-cancer	Vero E6	< 0.08 μM	ND	ND	PIKfyve	phase 2
					A549-ACE2	0.007 μM				
		dinaciclib (SCH-727965)	investigational (phase 3)	anti-cancer	Vero E6	0.127 μM	ND	ND	cyclin-dependent kinase	NI
					A549-ACE2	0.032 μM				
	complexed or unclear MOA	niclosamide		tapeworm infection	Vero	0.28 μM	SARS-CoV-2 *(being studied)*	ND	ND	phase 3
		chloroquine (CQ)		malaria, inflammatory diseases	Vero E6	0.72 – 7.36 μM (CQ)	SARS-CoV *(did not reduce viral replication)*	ND	endosomal acidification, glycosylation of ACE2	phase 2/3
		hydroxychloroquine (HCQ)				4.5 – 12.96 μM (HCQ)	ND	ND		phase 3/4

[a]Known *in vitro* data for SARS-CoV-2 and *in vivo* data for SARS-CoV, MERS-CoV, and SARS-CoV-2 are included. Alternative names for drugs are mentioned. HAE, human airway epithelial cells; Vero, African green monkey kidney epithelial cells; Calu-3, human lung epithelial cells; Huh7, human hepatoma cells; A549-ACE2, human lung carcinoma cells expressing ACE2 receptor; RdRp, RNA-dependent RNA polymerase; Mpro, viral main protease; ND, not determined; NI, not initiated.

MODULATORS OF INFLAMMATION AND TISSUE INJURY

While the immune response induced by SARS-CoV-2 infection is protective, it also plays an important role in disease pathogenesis.[82] During the incubation period and early stages of COVID-19, the effective immune response facilitates viral clearance. However, when the response is insufficiently protective, SARS-CoV-2 replicates efficiently in tissues expressing ACE2, such as lungs, heart, and intestines, leading to massive tissue destruction, severe inflammation, vascular hyperpermeability, and ultimately, tissue fibrosis and long-term dysfunction. In the lungs, this excessive inflammatory response leads to acute respiratory distress syndrome (ARDS), which is often fatal,[83] or can result in an epidermal growth factor receptor (EGFR)-mediated pulmonary fibrosis, as observed in SARS-CoV infection.[84] Strategies that boost the innate immune response during the early stages of viral infection to enhance viral clearance and modulate the excessive inflammatory response to reduce tissue injury during the later stages of infection may both provide benefit. A number of repurposed approaches that aim to achieve these effects are currently being investigated.

Interferons (IFN) are one example. These soluble glycoproteins have strong antiviral, antiproliferative, and immunomodulatory effects. In an *ex vivo* model of human lung, SARS-CoV-2 largely failed to induce the expression of any IFNs (type I, II, or III),[85] indicating that the early innate immune response to this virus is impaired and supporting the use of exogenous IFN to stimulate innate immunity as a candidate therapeutic modality. Type I IFNs (IFN-I, e.g., IFN-α and IFN-β) have been approved for the treatment of chronic hepatitis B and C infections and have previously demonstrated *in vitro* activity and protection in various animal models of MERS-CoV[86] and SARS-CoV.[74] In a preliminary, uncontrolled study of patients infected with SARS-CoV, the use of type I IFN plus corticosteroids was associated with a faster resolution of radiographic lung abnormalities and improved oxygen saturation levels than treatment with corticosteroids alone.[87] SARS-CoV-2 appears to be more susceptible to IFN-α treatment *in vitro* than SARS-CoV.[88] In a retrospective study of 77 adults with moderate COVID-19, treatment with nebulized IFN-α2b reduced the duration of both detectable virus in the upper respiratory tract and elevated blood levels of inflammatory markers (IL-6 and C-reactive protein).[89] Several types and routes of administration of IFN-I are currently being studied in COVID-19 patients.

Perhaps a more attractive candidate approach for COVID-19 treatment is, however, type III IFNs (IFN-λ1−4). Although not approved yet, IFN-λ has been shown to be safe in large phase 1/2/3 trials (involving ∼3000 patients) for the treatment of hepatitis B, C, and D virus infections.[90] In

https://dx.doi.org/10.1021/acsinfecdis.0c00343
ACS Infect. Dis. XXXX, XXX, XXX−XXX

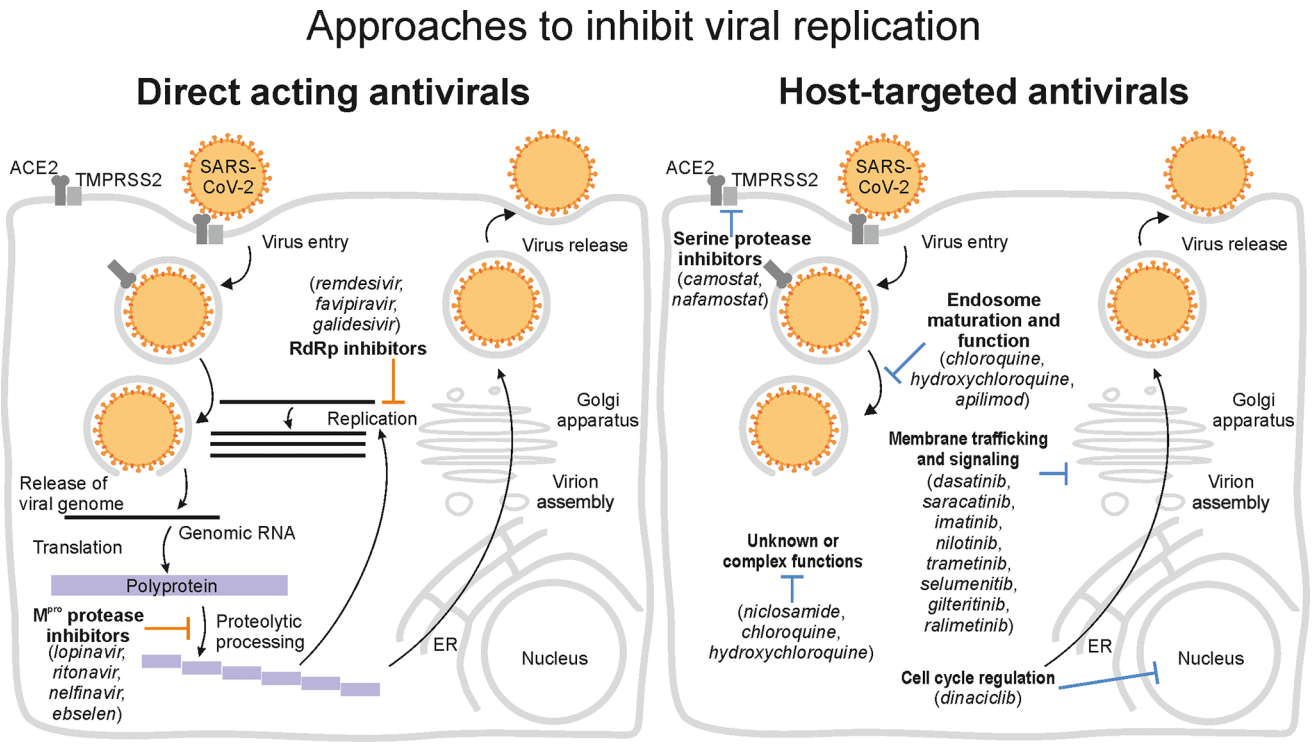


Figure 1. Repurposed drugs that primarily inhibit SARS-CoV-2 replication. The left panel depicts direct acting antivirals (DAAs) targeting SARS-CoV-2 enzymes (RNA-dependent RNA polymerase (RdRp) and viral main protease (M^{pro})). The right panel illustrates host-targeted drugs that inhibit cellular functions required for viral replication. Each panel depicts specific stages of the viral life cycle, some of which are targeted by drugs. Examples of repurposed drugs are connected to the corresponding targeted proteins or pathways by blunt arrows.

contrast to IFN-α/β that signals through a ubiquitously expressed INF-α/β receptor (IFNAR), IFN-λ signals through the INF-λ receptor (IFNLR), whose expression is restricted to epithelial cells (including lung and gut) and a subset of immune cells (reviewed in ref 91). The administration of type III IFN at an early stage of COVID-19 could therefore result in an antiviral response that is localized to epithelial cells, thereby reducing side effects and inflammation associated with the systemic action of type I IFNs.[92] In a human airway epithelial cell culture model, IFN-λ3 and IFN-λ4 exhibited antiviral effects against MERS-CoV.[93] IFN-λ restricted SARS-CoV replication in the respiratory and gastrointestinal tracts in knockout mouse strains lacking receptors for type I IFN, type III IFN, or both.[94] Several phase 2 clinical randomized controlled trials to evaluate the safety and efficacy of pegylated INF-λ compared with the placebo in outpatients with uncomplicated COVID-19 (NCT04331899, NCT04344600), hospitalized patients with noncritical illness (NCT04388709), or both ambulatory and hospitalized patients (NCT04354259) are ongoing.

COVID-19 severity was found to positively correlate with the level of proinflammatory cytokines.[1] Increased serum expression levels of IL-2 and IL-6, in particular, appear to predict the severity and prognosis of patients with COVID-19.[95] Immunomodulatory agents that directly target these key cytokines may therefore help to reduce the inflammatory responses observed in severe cases.

Various approaches are currently being studied for their potential to reduce inflammation and/or tissue injury in moderate and severe forms of COVID-19. The benefit of using corticosteroids for the treatment of COVID-19 was initially debatable.[96,97] A systematic review and meta-analysis of the safety and efficacy of corticosteroids revealed that their use in subjects with SARS-CoV-2, SARS-CoV, and MERS-CoV infections delayed viral clearance and did not convincingly improve survival.[98] Nevertheless, the preliminary results of the RECOVERY trial, a large randomized, controlled, open-label trial, have shown a benefit for dexamethasone treatment in severe COVID-19 patients. Specifically, up to a 10-day dexamethasone treatment course in 2104 patients reduced the 28-day mortality by one-third in patients on mechanical ventilation (29% vs 40.7%) and by one-fifth in patients requiring oxygen (21.5% vs 25%) relative to 4321 patients who received the standard of care, yet it had no effect in patients who did not require oxygen.[99] Additional phase 3 clinical trials testing various dosing regimens and formulations of corticosteroids in severe COVID-19 patients are currently ongoing.

Another class of anti-inflammatory agents that are being studied clinically are JAK inhibitors. Baricitinib is an oral JAK1/JAK2 inhibitor that has been shown to modulate innate and adaptive immune responses by inhibiting the production of inflammatory cytokines including TNF-α, IL-6, IL-17, and IFN-γ[100] and to profoundly reduce inflammation in animal models.[101] It is approved in over 65 countries for the treatment of rheumatoid arthritis. A small pilot study in 12 mild to moderate COVID-19 patients in Italy has suggested that baricitinib in combination with lopinavir/ritonavir may be safe for this study population and can improve clinical and laboratory parameters.[102] In a noncontrolled, retrospective cohort study of 15 patients with moderate to severe COVID-19, a short course of baricitinib in combination with hydroxychloroquine was tolerated and temporally associated with clinical improvement in 11 of 15 patients and recovery in 12 of 15 patients.[103] The safety and efficacy of baricitinib

https://dx.doi.org/10.1021/acsinfecdis.0c00343
ACS Infect. Dis. XXXX, XXX, XXX−XXX

Table 2. Repurposed Approaches Aimed at Modulating Inflammatory Responses and Reducing Tissue Injury[a]

	name	type	status	*in vitro*	mouse model	NHP	mechanism of action	COVID-19 status
innate immune response booster	IFN-I (e.g. IFN-α and INF-β)	glycoprotein (cytokine)	approved for chronic HBV and HCV	SARS-CoV-2 more susceptible to IFN-α treatment than SARS-CoV	SARS-CoV *(inhibits viral replication)*	MERS-CoV *(improves outcome)*	boosts innate immune response	phase 2/3
	IFN-III (IFN-λ1-4)	glycoprotein (cytokine)	investigational (phase 1/2/3)	IFN-λ3 and IFN-λ4 inhibit MERS-CoV	SARS-CoV *(inhibits viral replication)*	ND	boosts innate immune response	phase 2
anti-inflammatory, modulators of tissue injury	dexamethasone	corticosteroid	approved for inflammatory conditions	ND	ND	ND	inhibits inflammatory cytokine production	phase 3
	baricitinib	Janus kinase inhibitor	approved for rheumatoid arthritis	ND	ND	ND	inhibits inflammatory cytokine production	phase 3
	acalabrutinib	Bruton tyrosine kinase inhibitor	approved for lymphoid malignancies	ND	ND	ND	reduces the levels of inflammatory markers and cytokines	phase 2
	tofacitinib	Janus kinase inhibitor	approved for inflammatory diseases	ND	ND	ND	inhibits inflammatory cytokine production	phase 2
	tocilizumab	monoclonal antibody	approved for inflammatory diseases	ND	ND	ND	inhibits IL-6	phase 2/3
	sarilumab	monoclonal antibody	approved for rheumatoid arthritis	ND	ND	ND	inhibits IL-6	failed in phase 3, trial stopped
	ulinastatin	serine protease inhibitor	approved for acute pancreatitis and sepsis (Asia)	ND	ND	ND	inhibits IL-6, modulates RAS system	phase 1/2, recommended in China
	angiotensin 1-7	peptide	investigational	ND	ND	ND	modulates RAS system	phase 2/3
	nintedanib	multikinase inhibitor	approved for pulmonary fibrosis	ND	ND	ND	supresses lung fibrosis	phase 2

[a]Known *in vitro* and *in vivo* data for SARS-CoV, MERS-CoV, and SARS-CoV-2 are included. ND, not determined; IFN, interferon; RAS, renin angiotensin system; IL, interleukin.

individually and/or in combination drug regimens are currently being studied at a larger scale as part of the NIH's multicenter, adaptive, randomized blinded controlled trial for hospitalized COVID-19 patients (NCT04280705) and other trials (NCT04373044). Similarly, the effect of the early administration of tofacitinib, another oral JAK (JAK1/JAK3) inhibitor approved for various inflammatory diseases, on COVID-19 pneumonia, will also be investigated in a phase 2 study (NCT04332042).

Acalabrutinib is a covalent inhibitor of the Bruton tyrosine kinase (BTK), which mediates inflammation via regulation of macrophage signaling and activation. Acalabrutinib is approved in the United States (US) for the treatment of lymphoid malignancies.[104] Its off-label use for treating 19 patients hospitalized with severe COVID-19 was shown to reduce respiratory distress and levels of the inflammatory markers C-reactive protein and IL-6.[105] Additional, phase 2 clinical trials testing the efficacy and safety of acalabrutinib in patients hospitalized with COVID-19 are ongoing in the US (NCT04380688) and Europe (NCT04346199).

Various humanized monoclonal antibodies approved for the treatment of inflammatory diseases or cancer are also being tested for COVID-19 as repurposed candidates. In two retrospective studies, treatment with tocilizumab, an IL-6 receptor (IL-6R) inhibitor approved for rheumatoid arthritis, improved survival of severe COVID-19 patients; however, treatment prolonged their hospital stay and was associated with serious adverse events including transient respiratory worsening and bacterial infections.[106,107] Sarilumab, another IL-6R inhibitor, has recently failed in a US phase 3 trial involving 194 critically ill COVID-19 patients, and the trial has therefore been stopped.[108] The utility of these and other monoclonal antibodies that are currently being studied for COVID-19 treatment, such as bevacizumab that targets vascular endothelial growth factor A (VEGF-A) and nivolumab and camrelizumab that target the programmed cell death protein 1 (PD1), remains uncertain.

Ulinastatin is another anti-inflammatory agent that inhibits IL-6 but may provide additional benefits for severe COVID-19 patients, beyond those provided by IL-6 inhibition, and would likely be safer than the monoclonal antibodies targeting IL-6R. This urinary serine protease inhibitor has demonstrated significant reduction in the levels of plasma pro-inflammatory cytokines in multiple clinical studies in various inflammatory conditions.[109] Notably, in an animal model of pancreatitis, it was also shown to activate the renin−angiotensin system (RAS) by upregulating the expression of ACE2 as well as the anti-inflammatory factor angiotensin 1−7 (Ang 1−7) and the Mas receptor.[110] This property makes ulinastatin a particularly attractive agent for COVID-19 treatment, since disease severity is thought to be linked in part to an imbalance in the RAS.[111]

The cell surface receptor ACE2 converts angiotensin II (Ang II) into Ang 1−7, which then binds to the Mas receptor to counteract the effects of Ang II on vasoconstriction, apoptosis, and inflammation.[112] An unopposed Ang II effect has been previously linked to increased pulmonary vascular permeability in animal models of ARDS.[113] It was previously shown that SARS-CoV infection and the viral spike protein reduce ACE2 expression and that injection of the spike-Fc protein (spike protein fused to the Fc portion of human IgG1) into mice causes acute lung failure, which could be attenuated by blocking the renin−angiotensin pathway.[113] By reducing ACE2 expression upon its entry into cells, SARS-CoV-2 may also facilitate such imbalance and enhancement of the Ang II effects, thereby contributing to ARDS and possibly myocarditis.[36,114,115] Ulinastatin may reverse this imbalance and reduce tissue injury by directly upregulating ACE2. Moreover, ulinastatin was shown to protect mice from lipopolysaccharide-induced acute lung injury and improve cardiac function in mice with diabetes by downregulating and reducing cytokine expression.[116,117] Collectively, ulinastatin thus has a potential to reduce inflammation and vasodilatation not only by restoring ACE2 and Ang 1−7 levels but also by reducing the activity of pro-inflammatory cytokines. Ulinastatin has been approved for the treatment of acute pancreatitis and sepsis in Asia[118] and, even more relevant, it was studied for the treatment of ARDS. In a meta-analysis of randomized controlled trials including over 2300 patients with ARDS, ulinastatin treatment reduced mortality and ventilator-associated pneumonia by nearly 50% and also reduced ICU and overall hospital stays.[119] On the basis of these findings and its favorable tolerability in patients, ulinastatin has been

https://dx.doi.org/10.1021/acsinfecdis.0c00343
ACS Infect. Dis. XXXX, XXX, XXX−XXX

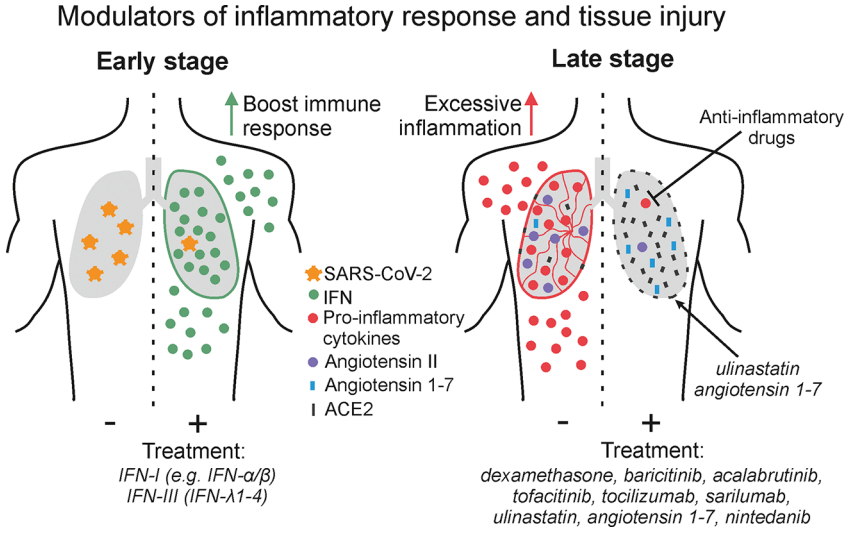


Figure 2. Repurposed drugs that modulate SARS-CoV-2-induced inflammatory responses and tissue injury. Drugs that boost innate immune responses could enhance viral clearance and provide protection during early stages of SARS-CoV-2 infection (left panel), whereas drugs that suppress inflammation by modulating the immune or renin−angiotensin system may reduce tissue injury (right panel).

recommended for the treatment of severe COVID-19 infection in China and a randomized, double-blind, placebo-controlled clinical trial to analyze its safety and efficacy for the treatment of COVID-19 has been initiated (NCT04393311). The utility of an Ang 1−7 peptide,[120] a different approach designed to reverse the RAS imbalance and reduce tissue injury, is also currently being clinically investigated in COVID-19 patients (NCT04332666).

Lastly, another category of agents being considered for its potential to reduce COVID-19-induced tissue damage are kinase inhibitors that suppress lung fibrosis. Several studies have suggested that the inhibition of epidermal growth factor receptor (EGFR) signaling may prevent the fibrotic response associated with various respiratory infections including SARS-CoV.[84] Several kinase inhibitors with anti-EGFR activity including gefitinib and erlotinib have been previously shown to inhibit the induction of fibrosis in various mouse models of tissue injury.[121] Nintedanib, a multikinase inhibitor with activity against fibroblast growth factor receptor (FGFR), platelet-derived growth factor receptor (PDGFR), and vascular endothelial growth factor receptor (VEGFR), which is approved for idiopathic pulmonary fibrosis, is currently being studied in patients with moderate to severe COVID-19 treatment for its potential effect to prevent pulmonary fibrosis (NCT04338802). The modulators of inflammation and tissue injury mentioned above are illustrated in Table 2 and Figure 2.

DISCUSSION

There is an urgent need for countermeasures to combat SARS-CoV-2 infection. It would take many months for a vaccine and much longer for a novel antiviral drug to be developed for effective prevention and treatment of this virus, respectively. Currently, the off-label use of drugs that are approved or are under advanced clinical investigation provides the main solution. Repurposing existing drugs requires significantly less capital and time and diminishes the clinical risks as such drugs have already been rigorously tested (toxicity, pharmacokinetics, pharmacodynamics, dosing, etc.) for their primary indication.

Since the majority of the antiviral drugs approved to date are DAAs that target viral proteins encoded by individual viruses, they typically provide a narrow spectrum of coverage and are therefore unlikely to be attractive candidates for repurposing. One example is provided by the HIV protease inhibitors, lopinavir/ritonavir. In contrast, existing antivirals that have already demonstrated broad-spectrum coverage, such as remdesivir, can facilitate readiness for future outbreaks of emerging pathogens and thus provide an immediate solution.

There are a number of strategies to identify antiviral candidates for repurposing among drugs that have already been approved or studied for other indications. One such approach is high-throughput screening of compound libraries for antiviral activity. Ebselen is one example of a compound that was discovered as having anti-SARS-CoV-2 activity using this approach.[34] The assembly of libraries that include a larger number of compounds that have already been approved or studied in humans, such as the REFRAME library,[122] has provided an important resource to support these efforts. Another approach is to identify existing drugs that target cellular factors known to be required for the life cycle of SARS-CoV-2. The discovery of camostat and nafamostat, inhibitors of cellular serine proteases, including TMPRSS2, exemplifies this strategy. Such host-targeted approaches may also have the added benefit of a higher genetic barrier to the emergence of resistance. At a larger scale, genomic and proteomic screens provide unbiased approaches for the discovery of proviral factors including druggable candidates. Mapping the interaction network of the individual SARS-CoV-2 proteins with the human proteome has recently revealed 66 druggable cellular factors that can be targeted by 69 compounds, many of which are either already approved or currently in clinical trials.[123] Moreover, phosphoproteomics analysis of SARS-COV-2-infected cells has revealed alterations in various cellular pathways during viral infection and facilitated discovery of small molecule inhibitors of these pathways as repurposing candidates.[54]

While repurposing of approved drugs as antivirals can offer an immediate solution, it also poses some challenges and raises important concerns. One challenge is that the antiviral effect

https://dx.doi.org/10.1021/acsinfecdis.0c00343
ACS Infect. Dis. XXXX, XXX, XXX−XXX

observed *in vitro* often cannot be reproduced *in vivo*. In the context of COVID-19, this was evident with lopinavir/ritonavir, which inhibited SARS-CoV-2 in cultured cells but did not show a benefit beyond standard care in patients when used by itself.[28] Furthermore, even a promising effect in animal models cannot guarantee effectiveness in patients, as exemplified by chloroquine for dengue treatment[66,68] and by remdesivir for EBOV disease treatment.[6,7]

Pharmacokinetic factors may contribute in part to the observed lack of correlation between preclinical and clinical models. Factors, such as achievable serum and particularly lung levels, have to be carefully evaluated when specific drugs are being considered for repurposing. For example, the pharmacokinetics of chloroquine and hydroxychloroquine is complex, and it is unclear whether the plasma level achieved with standard dosing is sufficient to effectively inhibit SARS-CoV-2 in patients.[76] Similarly, the plasma concentration of favipiravir in patients in the US has been shown to be 50% of that in Japanese patients, suggesting a possible ethnic or regional difference in its pharmacokinetics, which should be taken into account.[124] Toxicity is another major concern, particularly when targeting host functions. Importantly, drug safety in humans in one disease model cannot guarantee safety in another disease model. Chloroquine and hydroxychloroquine, for example, are generally safe when used to treat malaria and inflammatory diseases, yet in the context of SARS-CoV-2 infection, which affects the heart, they increase the risk of cardiac toxicity.[123] Nevertheless, for some drugs, it may be feasible to find a therapeutic window where the drug level is sufficient to inhibit viral replication with minimal cellular toxicity. Shortening the duration of therapy from months or years required to treat inflammatory diseases or cancer to several days or a few weeks sufficient to treat COVID-19 should further limit toxicity.

The emergence of viral resistance is another potential challenge of any antiviral strategy and particularly DAAs, such as those targeting the SARS-CoV-2 polymerase or protease. Although no phenotypic resistance to the currently used DAAs has been reported to date, there is already genotypic evidence for a high mutation rate in the SARS-CoV-2 RdRp sequence in patients.[125] While viral resistance can also limit treatment of host-targeted approaches,[47,126] in general, targeting cellular factors that are not under the genetic control of viruses is more likely to have a higher barrier to resistance than classical DAAs.[35,127]

Understanding the MOA of repurposed drugs is also often challenging. Cellular proteins function in a complex network of interactions, and their inhibitors are often not selective. The mechanism of antiviral action of host-targeted approaches therefore often remains elusive, and the molecular targets underlying this effect remain unvalidated. For example, erlotinib's effect on hepatitis C virus infection was first attributed solely to its effect on EGFR, its cancer target, yet it was later demonstrated that the inhibition of GAK, another cellular kinase that is inhibited by erlotinib with a comparable potency to EGFR, also plays a role.[127,128] For other drugs, such as chloroquine, the MOA is even less clear and appears to be pathogen specific.[69,129,130]

Access to drugs and cost pose additional challenges to repurposing existing drugs. The demand for specific drugs in the context of an outbreak can far exceed the available stockpiles. Indeed, early on, when used broadly for COVID-19, there was a significant shortage of hydroxychloroquine, which limited its availability for patients with inflammatory diseases who rely on it for their chronic conditions. Similarly, in the face of the upscaled and expedited production, there is an insufficient number of remdesivir doses to meet the global need. The high cost associated with some of the studied repurposed candidates, such as tocilizumab, is another barrier for their wide use in the treatment of COVID-19 patients.

Lastly, there is an urgent need to effectively prioritize among the proposed repurposed approaches and to effectively study them in order to guide clinical practice. The unpredictable nature of outbreaks with respect to size, geographic location, and duration further complicates the design of clinical trials. Consequently, the design of many of the hundreds ongoing clinical trials is somewhat limited, for example, by lacking appropriate controls or by being underpowered to reveal a virological or clinical effect. Moreover, concerns about publication bias are being raised.[14,131,80] This brings up a global concern that, similarly to the 2014 EBOV disease outbreak in West Africa, many approaches will be studied, yet the cumulative knowledge gained will be somewhat limited. One solution to overcome this challenge is offered by the adaptive platform design; a study design approved by the FDA during the 2014 Ebola outbreak[132] (NCT02380625) that is currently being implemented in the clinical trial conducted by the US NIH (NCT04280705). Adapted from clinical studies in cancer,[133] this platform design facilitates the simultaneous investigation of multiple agents individually and in combinations. The interim analysis feature of this platform design enables the entry of new agents as they become available and the removal of failing agents.[132] The incorporation of response adaptive randomization changes the randomization ratio of assigning treatment regimens to gradually favor the arms that perform better.[132] The important advantage is that such a study is designed to determine not only whether specific drugs work or not but also, more importantly, which drugs work the best, thereby guiding clinical practice. The integration of the adaptive platform design with a "core protocol" that involves multiple investigative teams and can extend across multiple infectious disease outbreaks may provide an even better solution to address these challenges yet will require tremendous coordination.[134]

In summary, the discussed examples represent the broad range of repurposed approaches that are already being studied or proposed to combat COVID-19. While a number of these approaches are showing some promise, the clinical utility of the majority of them remains to be determined. Protection against SARS-CoV-2 is more likely to be achieved by a combination of approaches that inhibit viral replication with those that reduce inflammation and tissue injury.

AUTHOR INFORMATION

Corresponding Author

Shirit Einav − *Department of Medicine, Division of Infectious Diseases and Geographic Medicine, and Department of Microbiology and Immunology, Stanford University School of Medicine, Stanford University, Stanford, California 94305, United States;* orcid.org/0000-0001-6441-4171; Email: seinav@stanford.edu

Author

Sirle Saul − *Department of Medicine, Division of Infectious Diseases and Geographic Medicine, and Department of Microbiology and Immunology, Stanford University School of*

https://dx.doi.org/10.1021/acsinfecdis.0c00343
ACS Infect. Dis. XXXX, XXX, XXX−XXX

Medicine, Stanford University, Stanford, California 94305, United States

Complete contact information is available at: https://pubs.acs.org/10.1021/acsinfecdis.0c00343

Notes

The authors declare no competing financial interest.

■ ACKNOWLEDGMENTS

This research was supported by grant HDTRA11810039 from the Defense Threat Reduction Agency and by an Investigator-Initiated Research Award (W81XWH-16-1-0691) from the Department of Defense office of the Congressionally Directed Medical Research Programs (CDMRP), Peer Reviewed Medical Research Program (PRMRP) to S.E. The authors acknowledge all the contributions in the field that could not be included in the review.

■ REFERENCES

(1) Huang, C., Wang, Y., Li, X., Ren, L., Zhao, J., Hu, Y., Zhang, L., Fan, G., Xu, J., Gu, X., Cheng, Z., Yu, T., Xia, J., Wei, Y., Wu, W., Xie, X., Yin, W., Li, H., Liu, M., Xiao, Y., Gao, H., Guo, L., Xie, J., Wang, G., Jiang, R., Gao, Z., Jin, Q., Wang, J., and Cao, B. (2020) Clinical Features of Patients Infected with 2019 Novel Coronavirus in Wuhan, China. *Lancet 395* (10223), 497−506.

(2) Spagnolo, P., Balestro, E., Aliberti, S., Cocconcelli, E., Biondini, D., Casa, G. D., Sverzellati, N., and Maher, T. M. (2020) Pulmonary Fibrosis Secondary to COVID-19: A Call to Arms? *Lancet Respir. Med.*, DOI: 10.1016/S2213-2600(20)30222-8.

(3) Shi, S., Qin, M., Shen, B., Cai, Y., Liu, T., Yang, F., Gong, W., Liu, X., Liang, J., Zhao, Q., Huang, H., Yang, B., and Huang, C. (2020) Association of Cardiac Injury With Mortality in Hospitalized Patients With COVID-19 in Wuhan, China. *JAMA Cardiol. 5*, 802.

(4) DiMasi, J. A., Grabowski, H. G., and Hansen, R. W. (2016) Innovation in the Pharmaceutical Industry: New Estimates of R&D Costs. *J. Health Econ. 47*, 20−33.

(5) Krichel, B., Falke, S., Hilgenfeld, R., Redecke, L., and Uetrecht, C. (2020) Processing of the SARS-CoV Pp1a/Ab Nsp7−10 Region. *Biochem. J. 477* (5), 1009−1019.

(6) Warren, T. K., Jordan, R., Lo, M. K., Ray, A. S., Mackman, R. L., Soloveva, V., Siegel, D., Perron, M., Bannister, R., Hui, H. C., Larson, N., Strickley, R., Wells, J., Stuthman, K. S., Van Tongeren, S. A., Garza, N. L., Donnelly, G., Shurtleff, A. C., Retterer, C. J., Gharaibeh, D., Zamani, R., Kenny, T., Eaton, B. P., Grimes, E., Welch, L. S., Gomba, L., Wilhelmsen, C. L., Nichols, D. K., Nuss, J. E., Nagle, E. R., Kugelman, J. R., Palacios, G., Doerffler, E., Neville, S., Carra, E., Clarke, M. O., Zhang, L., Lew, W., Ross, B., Wang, Q., Chun, K., Wolfe, L., Babusis, D., Park, Y., Stray, K. M., Trancheva, I., Feng, J. Y., Barauskas, O., Xu, Y., Wong, P., Braun, M. R., Flint, M., McMullan, L. K., Chen, S.-S., Fearns, R., Swaminathan, S., Mayers, D. L., Spiropoulou, C. F., Lee, W. A., Nichol, S. T., Cihlar, T., and Bavari, S. (2016) Therapeutic Efficacy of the Small Molecule GS-5734 against Ebola Virus in Rhesus Monkeys. *Nature 531* (7594), 381−385.

(7) Mulangu, S., Dodd, L. E., Davey, R. T., Tshiani Mbaya, O., Proschan, M., Mukadi, D., Lusakibanza Manzo, M., Nzolo, D., Tshomba Oloma, A., Ibanda, A., Ali, R., Coulibaly, S., Levine, A. C., Grais, R., Diaz, J., Lane, H. C., and Muyembe-Tamfum, J.-J. (2019) A Randomized, Controlled Trial of Ebola Virus Disease Therapeutics. *N. Engl. J. Med. 381* (24), 2293−2303.

(8) Sheahan, T. P., Sims, A. C., Graham, R. L., Menachery, V. D., Gralinski, L. E., Case, J. B., Leist, S. R., Pyrc, K., Feng, J. Y., Trantcheva, I., Bannister, R., Park, Y., Babusis, D., Clarke, M. O., Mackman, R. L., Spahn, J. E., Palmiotti, C. A., Siegel, D., Ray, A. S., Cihlar, T., Jordan, R., Denison, M. R., and Baric, R. S. (2017) Broad-Spectrum Antiviral GS-5734 Inhibits Both Epidemic and Zoonotic Coronaviruses. *Sci. Transl. Med. 9* (396), No. eaal3653.

(9) de Wit, E., Feldmann, F., Cronin, J., Jordan, R., Okumura, A., Thomas, T., Scott, D., Cihlar, T., and Feldmann, H. (2020) Prophylactic and Therapeutic Remdesivir (GS-5734) Treatment in the Rhesus Macaque Model of MERS-CoV Infection. *Proc. Natl. Acad. Sci. U. S. A. 117* (12), 6771.

(10) Gordon, C. J., Tchesnokov, E. P., Woolner, E., Perry, J. K., Feng, J. Y., Porter, D. P., and Gotte, M. (2020) Remdesivir Is a Direct-Acting Antiviral That Inhibits RNA-Dependent RNA Polymerase from Severe Acute Respiratory Syndrome Coronavirus 2 with High Potency. *J. Biol. Chem. 295*, 6785.

(11) Wang, M., Cao, R., Zhang, L., Yang, X., Liu, J., Xu, M., Shi, Z., Hu, Z., Zhong, W., and Xiao, G. (2020) Remdesivir and Chloroquine Effectively Inhibit the Recently Emerged Novel Coronavirus (2019-NCoV) in Vitro. *Cell Res. 30* (3), 269−271.

(12) Sheahan, T. P., Sims, A. C., Zhou, S., Graham, R. L., Pruijssers, A. J., Agostini, M. L., Leist, S. R., Schäfer, A., Dinnon, K. H., Stevens, L. J., Chappell, J. D., Lu, X., Hughes, T. M., George, A. S., Hill, C. S., Montgomery, S. A., Brown, A. J., Bluemling, G. R., Natchus, M. G., Saindane, M., Kolykhalov, A. A., Painter, G., Harcourt, J., Tamin, A., Thornburg, N. J., Swanstrom, R., Denison, M. R., and Baric, R. S. (2020) An Orally Bioavailable Broad-Spectrum Antiviral Inhibits SARS-CoV-2 in Human Airway Epithelial Cell Cultures and Multiple Coronaviruses in Mice. *Sci. Transl. Med. 12* (541), No. eabb5883.

(13) Williamson, B. N., Feldmann, F., Schwarz, B., Meade-White, K., Porter, D. P., Schulz, J., van Doremalen, N., Leighton, I., Yinda, C. K., Pérez-Pérez, L., Okumura, A., Lovaglio, J., Hanley, P. W., Saturday, G., Bosio, C. M., Anzick, S., Barbian, K., Cihlar, T., Martens, C., Scott, D. P., Munster, V. J., and de Wit, E. (2020) Clinical Benefit of Remdesivir in Rhesus Macaques Infected with SARS-CoV-2. *bioRxiv*, DOI: 10.1101/2020.04.15.043166.

(14) Grein, J., Ohmagari, N., Shin, D., Diaz, G., Asperges, E., Castagna, A., Feldt, T., Green, G., Green, M. L., Lescure, F.-X., Nicastri, E., Oda, R., Yo, K., Quiros-Roldan, E., Studemeister, A., Redinski, J., Ahmed, S., Bernett, J., Chelliah, D., Chen, D., Chihara, S., Cohen, S. H., Cunningham, J., D'Arminio Monforte, A., Ismail, S., Kato, H., Lapadula, G., L'Her, E., Maeno, T., Majumder, S., Massari, M., Mora-Rillo, M., Mutoh, Y., Nguyen, D., Verweij, E., Zoufaly, A., Osinusi, A. O., DeZure, A., Zhao, Y., Zhong, L., Chokkalingam, A., Elboudwarej, E., Telep, L., Timbs, L., Henne, I., Sellers, S., Cao, H., Tan, S. K., Winterbourne, L., Desai, P., Mera, R., Gaggar, A., Myers, R. P., Brainard, D. M., Childs, R., and Flanigan, T. (2020) Compassionate Use of Remdesivir for Patients with Severe Covid-19. *N. Engl. J. Med. 382*, 2327.

(15) Wang, Y., Zhang, D., Du, G., Du, R., Zhao, J., Jin, Y., Fu, S., Gao, L., Cheng, Z., Lu, Q., Hu, Y., Luo, G., Wang, K., Lu, Y., Li, H., Wang, S., Ruan, S., Yang, C., Mei, C., Wang, Y., Ding, D., Wu, F., Tang, X., Ye, X., Ye, Y., Liu, B., Yang, J., Yin, W., Wang, A., Fan, G., Zhou, F., Liu, Z., Gu, X., Xu, J., Shang, L., Zhang, Y., Cao, L., Guo, T., Wan, Y., Qin, H., Jiang, Y., Jaki, T., Hayden, F. G., Horby, P. W., Cao, B., and Wang, C. (2020) Remdesivir in Adults with Severe COVID-19: A Randomised, Double-Blind, Placebo-Controlled, Multicentre Trial. *Lancet 395* (10236), 1569−1578.

(16) Beigel, J. H., Tomashek, K. M., Dodd, L. E., Mehta, A. K., Zingman, B. S., Kalil, A. C., Hohmann, E., Chu, H. Y., Luetkemeyer, A., Kline, S., Lopez de Castilla, D., Finberg, R. W., Dierberg, K., Tapson, V., Hsieh, L., Patterson, T. F., Paredes, R., Sweeney, D. A., Short, W. R., Touloumi, G., Lye, D. C., Ohmagari, N., Oh, M., Ruiz-Palacios, G. M., Benfield, T., Fätkenheuer, G., Kortepeter, M. G., Atmar, R. L., Creech, C. B., Lundgren, J., Babiker, A. G., Pett, S., Neaton, J. D., Burgess, T. H., Bonnett, T., Green, M., Makowski, M., Osinusi, A., Nayak, S., and Lane, H. C. (2020) Remdesivir for the Treatment of Covid-19 — Preliminary Report. *N. Engl. J. Med.*, DOI: 10.1056/NEJMoa2007764.

(17) De Clercq, E. (2015) Ebola Virus (EBOV) Infection: Therapeutic Strategies. *Biochem. Pharmacol. 93* (1), 1−10.

(18) Furuta, Y., Komeno, T., and Nakamura, T. (2017) Favipiravir (T-705), a Broad Spectrum Inhibitor of Viral RNA Polymerase. *Proc. Jpn. Acad., Ser. B 93* (7), 449−463.

(19) Du, Y.-X., and Chen, X.-P. (2020) Favipiravir: Pharmacokinetics and Concerns About Clinical Trials for 2019-NCoV Infection. *Clin. Pharmacol. Ther. 108* (2), 242−247.
(20) Oestereich, L., Lüdtke, A., Wurr, S., Rieger, T., Muñoz-Fontela, C., and Günther, S. (2014) Successful Treatment of Advanced Ebola Virus Infection with T-705 (Favipiravir) in a Small Animal Model. *Antiviral Res. 105*, 17−21.
(21) Bai, C.-Q., Mu, J.-S., Kargbo, D., Song, Y.-B., Niu, W.-K., Nie, W.-M., Kanu, A., Liu, W.-W., Wang, Y.-P., Dafae, F., Yan, T., Hu, Y., Deng, Y.-Q., Lu, H.-J., Yang, F., Zhang, X.-G., Sun, Y., Cao, Y.-X., Su, H.-X., Sun, Y., Liu, W.-S., Wang, C.-Y., Qian, J., Liu, L., Wang, H., Tong, Y.-G., Liu, Z.-Y., Chen, Y.-S., Wang, H.-Q., Kargbo, B., Gao, G. F., and Jiang, J.-F. (2016) Clinical and Virological Characteristics of Ebola Virus Disease Patients Treated With Favipiravir (T-705)—Sierra Leone, 2014. *Clin. Infect. Dis. 63* (10), 1288−1294.
(22) Cai, Q., Yang, M., Liu, D., Chen, J., Shu, D., Xia, J., Liao, X., Gu, Y., Cai, Q., Yang, Y., Shen, C., Li, X., Peng, L., Huang, D., Zhang, J., Zhang, S., Wang, F., Liu, J., Chen, L., Chen, S., Wang, Z., Zhang, Z., Cao, R., Zhong, W., Liu, Y., and Liu, L. (2020) Experimental Treatment with Favipiravir for COVID-19: An Open-Label Control Study. *Engineering*, DOI: 10.1016/j.eng.2020.03.007.
(23) Chen, C., Zhang, Y., Huang, J., Yin, P., Cheng, Z., Wu, J., Chen, S., Zhang, Y., Chen, B., Lu, M., Luo, Y., Ju, L., Zhang, J., and Wang, X. (2020) Favipiravir versus Arbidol for COVID-19: A Randomized Clinical Trial. *medRxiv*, DOI: 10.1101/2020.03.17.20037432.
(24) Warren, T. K., Wells, J., Panchal, R. G., Stuthman, K. S., Garza, N. L., Van Tongeren, S. A., Dong, L., Retterer, C. J., Eaton, B. P., Pegoraro, G., Honnold, S., Bantia, S., Kotian, P., Chen, X., Taubenheim, B. R., Welch, L. S., Minning, D. M., Babu, Y. S., Sheridan, W. P., and Bavari, S. (2014) Protection against Filovirus Diseases by a Novel Broad-Spectrum Nucleoside Analogue BCX4430. *Nature 508* (7496), 402−405.
(25) Elfiky, A. A. (2020) Ribavirin, Remdesivir, Sofosbuvir, Galidesivir, and Tenofovir against SARS-CoV-2 RNA Dependent RNA Polymerase (RdRp): A Molecular Docking Study. *Life Sci. 253*, 117592.
(26) Hegyi, A., and Ziebuhr, J. (2002) Conservation of Substrate Specificities among Coronavirus Main Proteases. *J. Gen. Virol. 83*, 595−599.
(27) Ohashi, H., Watashi, K., Saso, W., Shionoya, K., Iwanami, S., Hirokawa, T., Shirai, T., Kanaya, S., Ito, Y., Kim, K. S., Nishioka, K., Ando, S., Ejima, K., Koizumi, Y., Tanaka, T., Aoki, S., Kuramochi, K., Suzuki, T., Maenaka, K., Matano, T., Muramatsu, M., Saijo, M., Aihara, K., Iwami, S., Takeda, M., McKeating, J. A., and Wakita, T. (2020) Multidrug Treatment with Nelfinavir and Cepharanthine against COVID-19. *bioRxiv*, DOI: 10.1101/2020.04.14.039925.
(28) Cao, B., Wang, Y., Wen, D., Liu, W., Wang, J., Fan, G., Ruan, L., Song, B., Cai, Y., Wei, M., Li, X., Xia, J., Chen, N., Xiang, J., Yu, T., Bai, T., Xie, X., Zhang, L., Li, C., Yuan, Y., Chen, H., Li, H., Huang, H., Tu, S., Gong, F., Liu, Y., Wei, Y., Dong, C., Zhou, F., Gu, X., Xu, J., Liu, Z., Zhang, Y., Li, H., Shang, L., Wang, K., Li, K., Zhou, X., Dong, X., Qu, Z., Lu, S., Hu, X., Ruan, S., Luo, S., Wu, J., Peng, L., Cheng, F., Pan, L., Zou, J., Jia, C., Wang, J., Liu, X., Wang, S., Wu, X., Ge, Q., He, J., Zhan, H., Qiu, F., Guo, L., Huang, C., Jaki, T., Hayden, F. G., Horby, P. W., Zhang, D., and Wang, C. (2020) A Trial of Lopinavir−Ritonavir in Adults Hospitalized with Severe Covid-19. *N. Engl. J. Med. 382* (19), 1787−1799.
(29) Yamamoto, N., Yang, R., Yoshinaka, Y., Amari, S., Nakano, T., Cinatl, J., Rabenau, H., Doerr, H. W., Hunsmann, G., Otaka, A., Tamamura, H., Fujii, N., and Yamamoto, N. (2004) HIV Protease Inhibitor Nelfinavir Inhibits Replication of SARS-Associated Coronavirus. *Biochem. Biophys. Res. Commun. 318* (3), 719−725.
(30) Azad, G. K., and Tomar, R. S. (2014) Ebselen, a Promising Antioxidant Drug: Mechanisms of Action and Targets of Biological Pathways. *Mol. Biol. Rep. 41* (8), 4865−4879.
(31) Kil, J., Lobarinas, E., Spankovich, C., Griffiths, S. K., Antonelli, P. J., Lynch, E. D., and Le Prell, C. G. (2017) Safety and Efficacy of Ebselen for the Prevention of Noise-Induced Hearing Loss: A Randomised, Double-Blind, Placebo-Controlled, Phase 2 Trial. *Lancet 390* (10098), 969−979.
(32) Beckman, J. A., Goldfine, A. B., Leopold, J. A., and Creager, M. A. (2016) Ebselen Does Not Improve Oxidative Stress and Vascular Function in Patients with Diabetes: A Randomized, Crossover Trial. *Am. J. Physiol.-Heart Circ. Physiol. 311* (6), H1431−H1436.
(33) Yamaguchi, T., Sano, K., Takakura, K., Saito, I., Shinohara, Y., Asano, T., and Yasuhara, H. (1998) Ebselen in Acute Ischemic Stroke. *Stroke 29* (1), 12−17.
(34) Jin, Z., Du, X., Xu, Y., Deng, Y., Liu, M., Zhao, Y., Zhang, B., Li, X., Zhang, L., Peng, C., Duan, Y., Yu, J., Wang, L., Yang, K., Liu, F., Jiang, R., Yang, X., You, T., Liu, X., Yang, X., Bai, F., Liu, H., Liu, X., Guddat, L. W., Xu, W., Xiao, G., Qin, C., Shi, Z., Jiang, H., Rao, Z., and Yang, H. (2020) Structure of Mpro from SARS-CoV-2 and Discovery of Its Inhibitors. *Nature 582*, 289.
(35) Bekerman, E., and Einav, S. (2015) Combating Emerging Viral Threats. *Science 348* (6232), 282.
(36) Hoffmann, M., Kleine-Weber, H., Schroeder, S., Krüger, N., Herrler, T., Erichsen, S., Schiergens, T. S., Herrler, G., Wu, N.-H., Nitsche, A., Müller, M. A., Drosten, C., and Pöhlmann, S. (2020) SARS-CoV-2 Cell Entry Depends on ACE2 and TMPRSS2 and Is Blocked by a Clinically Proven Protease Inhibitor. *Cell 181* (2), 271−280.E8.
(37) Matsuyama, S., Nagata, N., Shirato, K., Kawase, M., Takeda, M., and Taguchi, F. (2010) Efficient Activation of the Severe Acute Respiratory Syndrome Coronavirus Spike Protein by the Transmembrane Protease TMPRSS2. *J. Virol. 84* (24), 12658.
(38) Shirato, K., Kawase, M., and Matsuyama, S. (2013) Middle East Respiratory Syndrome Coronavirus Infection Mediated by the Transmembrane Serine Protease TMPRSS2. *J. Virol. 87* (23), 12552.
(39) Iwata-Yoshikawa, N., Okamura, T., Shimizu, Y., Hasegawa, H., Takeda, M., and Nagata, N. (2019) TMPRSS2 Contributes to Virus Spread and Immunopathology in the Airways of Murine Models after Coronavirus Infection. *J. Virol. 93* (6), No. e01815-18.
(40) Gibo, J., Ito, T., Kawabe, K., Hisano, T., Inoue, M., Fujimori, N., Oono, T., Arita, Y., and Nawata, H. (2005) Camostat Mesilate Attenuates Pancreatic Fibrosis via Inhibition of Monocytes and Pancreatic Stellate Cells Activity. *Lab. Invest. 85* (1), 75−89.
(41) Zhou, Y., Vedantham, P., Lu, K., Agudelo, J., Carrion, R., Nunneley, J. W., Barnard, D., Pöhlmann, S., McKerrow, J. H., Renslo, A. R., and Simmons, G. (2015) Protease Inhibitors Targeting Coronavirus and Filovirus Entry. *Antiviral Res. 116*, 76−84.
(42) Yamamoto, M., Matsuyama, S., Li, X., Takeda, M., Kawaguchi, Y., Inoue, J., and Matsuda, Z. (2016) Identification of Nafamostat as a Potent Inhibitor of Middle East Respiratory Syndrome Coronavirus S Protein-Mediated Membrane Fusion Using the Split-Protein-Based Cell-Cell Fusion Assay. *Antimicrob. Agents Chemother. 60* (11), 6532.
(43) Hoffmann, M., Schroeder, S., Kleine-Weber, H., Müller, M. A., Drosten, C., and Pöhlmann, S. (2020) Nafamostat Mesylate Blocks Activation of SARS-CoV-2: New Treatment Option for COVID-19. *Antimicrob. Agents Chemother.*, DOI: 10.1128/AAC.00754-20.
(44) Schor, S., and Einav, S. (2018) Repurposing of Kinase Inhibitors as Broad-Spectrum Antiviral Drugs. *DNA Cell Biol. 37* (2), 63−69.
(45) Pagano, M. A., Tibaldi, E., Palù, G., and Brunati, A. M. (2013) Viral Proteins and Src Family Kinases: Mechanisms of Pathogenicity from a "Liaison Dangereuse. *World J. Virol. 2* (2), 71−78.
(46) Shin, S. J., Jung, E., Kim, M., Baric, S. R., and Go, Y. Y. (2018) Saracatinib Inhibits Middle East Respiratory Syndrome-Coronavirus Replication In Vitro. *Viruses 10* (6), 283.
(47) de Wispelaere, M., LaCroix, A. J., and Yang, P. L. (2013) The Small Molecules AZD0530 and Dasatinib Inhibit Dengue Virus RNA Replication via Fyn Kinase. *J. Virol. 87* (13), 7367.
(48) Chu, J. J. H., and Yang, P. L. (2007) C-Src Protein Kinase Inhibitors Block Assembly and Maturation of Dengue Virus. *Proc. Natl. Acad. Sci. U. S. A. 104* (9), 3520.
(49) Dyall, J., Coleman, C. M., Hart, B. J., Venkataraman, T., Holbrook, M. R., Kindrachuk, J., Johnson, R. F., Olinger, G. G., Jahrling, P. B., Laidlaw, M., Johansen, L. M., Lear-Rooney, C. M.,

https://dx.doi.org/10.1021/acsinfecdis.0c00343
ACS Infect. Dis. XXXX, XXX, XXX−XXX

Glass, P. J., Hensley, L. E., and Frieman, M. B. (2014) Repurposing of Clinically Developed Drugs for Treatment of Middle East Respiratory Syndrome Coronavirus Infection. *Antimicrob. Agents Chemother. 58* (8), 4885.
(50) Reeves, P. M., Smith, S. K., Olson, V. A., Thorne, S. H., Bornmann, W., Damon, I. K., and Kalman, D. (2011) Variola and Monkeypox Viruses Utilize Conserved Mechanisms of Virion Motility and Release That Depend on Abl and Src Family Tyrosine Kinases. *J. Virol. 85* (1), 21.
(51) Clark, M. J., Miduturu, C., Schmidt, A. G., Zhu, X., Pitts, J. D., Wang, J., Potisopon, S., Zhang, J., Wojciechowski, A., Hann Chu, J. J., Gray, N. S., and Yang, P. L. (2016) GNF-2 Inhibits Dengue Virus by Targeting Abl Kinases and the Viral E Protein. *Cell Chem. Biol. 23* (4), 443−452.
(52) Coleman, C. M., Sisk, J. M., Mingo, R. M., Nelson, E. A., White, J. M., and Frieman, M. B. (2016) Abelson Kinase Inhibitors Are Potent Inhibitors of Severe Acute Respiratory Syndrome Coronavirus and Middle East Respiratory Syndrome Coronavirus Fusion. *J. Virol. 90* (19), 8924.
(53) Kindrachuk, J., Ork, B., Hart, B. J., Mazur, S., Holbrook, M. R., Frieman, M. B., Traynor, D., Johnson, R. F., Dyall, J., Kuhn, J. H., Olinger, G. G., Hensley, L. E., and Jahrling, P. B. (2015) Antiviral Potential of ERK/MAPK and PI3K/AKT/MTOR Signaling Modulation for Middle East Respiratory Syndrome Coronavirus Infection as Identified by Temporal Kinome Analysis. *Antimicrob. Agents Chemother. 59* (2), 1088.
(54) Bouhaddou, M., Memon, D., Meyer, B., White, K. M., Rezelj, V. V., Correa Marrero, M., Polacco, B. J., Melnyk, J. E., Ulferts, S., Kaake, R. M., Batra, J., Richards, A. L., Stevenson, E., Gordon, D. E., Rojc, A., Obernier, K., Fabius, J. M., Soucheray, M., Miorin, L., Moreno, E., Koh, C., Tran, Q. D., Hardy, A., Robinot, R., Vallet, T., Nilsson-Payant, B. E., Hernandez-Armenta, C., Dunham, A., Weigang, S., Knerr, J., Modak, M., Quintero, D., Zhou, Y., Dugourd, A., Valdeolivas, A., Patil, T., Li, Q., Hüttenhain, R., Cakir, M., Muralidharan, M., Kim, M., Jang, G., Tutuncuoglu, B., Hiatt, J., Guo, J. Z., Xu, J., Bouhaddou, S., Mathy, C. J. P., Gaulton, A., Manners, E. J., Félix, E., Shi, Y., Goff, M., Lim, J. K., McBride, T., O'Neal, M. C., Cai, Y., Chang, J. C. J., Broadhurst, D. J., Klippsten, S., De Wit, E., Leach, A. R., Kortemme, T., Shoichet, B., Ott, M., Saez-Rodriguez, J., tenOever, B. R., Mullins, R. D., Fischer, E. R., Kochs, G., Grosse, R., García-Sastre, A., Vignuzzi, M., Johnson, J. R., Shokat, K. M., Swaney, D. L., Beltrao, P., and Krogan, N. J. (2020) The Global Phosphorylation Landscape of SARS-CoV-2 Infection. *Cell*, DOI: 10.1016/j.cell.2020.06.034.
(55) Mizutani, T., Fukushi, S., Saijo, M., Kurane, I., and Morikawa, S. (2004) Phosphorylation of P38 MAPK and Its Downstream Targets in SARS Coronavirus-Infected Cells. *Biochem. Biophys. Res. Commun. 319* (4), 1228−1234.
(56) Cai, X., Xu, Y., Cheung, A. K., Tomlinson, R. C., Alcázar-Román, A., Murphy, L., Billich, A., Zhang, B., Feng, Y., Klumpp, M., Rondeau, J.-M., Fazal, A. N., Wilson, C. J., Myer, V., Joberty, G., Bouwmeester, T., Labow, M. A., Finan, P. M., Porter, J. A., Ploegh, H. L., Baird, D., De Camilli, P., Tallarico, J. A., and Huang, Q. (2013) PIKfyve, a Class III PI Kinase, Is the Target of the Small Molecular IL-12/IL-23 Inhibitor Apilimod and a Player in Toll-like Receptor Signaling. *Chem. Biol. 20* (7), 912−921.
(57) Ou, X., Liu, Y., Lei, X., Li, P., Mi, D., Ren, L., Guo, L., Guo, R., Chen, T., Hu, J., Xiang, Z., Mu, Z., Chen, X., Chen, J., Hu, K., Jin, Q., Wang, J., and Qian, Z. (2020) Characterization of Spike Glycoprotein of SARS-CoV-2 on Virus Entry and Its Immune Cross-Reactivity with SARS-CoV. *Nat. Commun. 11* (1), 1620.
(58) Mita, M. M., Mita, A. C., Moseley, J. L., Poon, J., Small, K. A., Jou, Y.-M., Kirschmeier, P., Zhang, D., Zhu, Y., Statkevich, P., Sankhala, K. K., Sarantopoulos, J., Cleary, J. M., Chirieac, L. R., Rodig, S. J., Bannerji, R., and Shapiro, G. I. (2017) Phase 1 Safety, Pharmacokinetic and Pharmacodynamic Study of the Cyclin-Dependent Kinase Inhibitor Dinaciclib Administered Every Three Weeks in Patients with Advanced Malignancies. *Br. J. Cancer 117* (9), 1258−1268.
(59) Xu, J., Shi, P.-Y., Li, H., and Zhou, J. (2020) Broad Spectrum Antiviral Agent Niclosamide and Its Therapeutic Potential. *ACS Infect. Dis. 6* (5), 909−915.
(60) Wu, C.-J., Jan, J.-T., Chen, C.-M., Hsieh, H.-P., Hwang, D.-R., Liu, H.-W., Liu, C.-Y., Huang, H.-W., Chen, S.-C., Hong, C.-F., Lin, R.-K., Chao, Y.-S., and Hsu, J. T. A. (2004) Inhibition of Severe Acute Respiratory Syndrome Coronavirus Replication by Niclosamide. *Antimicrob. Agents Chemother. 48* (7), 2693.
(61) Gassen, N. C., Niemeyer, D., Muth, D., Corman, V. M., Martinelli, S., Gassen, A., Hafner, K., Papies, J., Mösbauer, K., Zellner, A., Zannas, A. S., Herrmann, A., Holsboer, F., Brack-Werner, R., Boshart, M., Müller-Myhsok, B., Drosten, C., Müller, M. A., and Rein, T. (2019) SKP2 Attenuates Autophagy through Beclin1-Ubiquitination and Its Inhibition Reduces MERS-Coronavirus Infection. *Nat. Commun. 10* (1), 5770.
(62) Jeon, S., Ko, M., Lee, J., Choi, I., Byun, S. Y., Park, S., Shum, D., and Kim, S. (2020) Identification of Antiviral Drug Candidates against SARS-CoV-2 from FDA-Approved Drugs. *Antimicrob. Agents Chemother. 64*, e00819-20.
(63) Frayha, G. J., Smyth, J. D., Gobert, J. G., and Savel, J. (1997) The Mechanisms of Action of Antiprotozoal and Anthelmintic Drugs in Man. *Gen. Pharmacol. 28* (2), 273−299.
(64) Chen, W., Mook, R. A., Premont, R. T., and Wang, J. (2018) Niclosamide: Beyond an Antihelminthic Drug. *Cell. Signalling 41*, 89−96.
(65) McChesney, E. W. (1983) Animal Toxicity and Pharmacokinetics of Hydroxychloroquine Sulfate. *Am. J. Med. 75* (1), 11−18.
(66) Farias, K. J. S., Machado, P. R. L., Muniz, J. A. P. C., Imbeloni, A. A., and da Fonseca, B. A. L. (2015) Antiviral Activity of Chloroquine Against Dengue Virus Type 2 Replication in Aotus Monkeys. *Viral Immunol. 28* (3), 161−169.
(67) Borges, M. C., Castro, L. A., and Fonseca, B. A. L. de. (2013) Chloroquine Use Improves Dengue-Related Symptoms. *Mem. Inst. Oswaldo Cruz 108* (5), 596−599.
(68) Tricou, V., Minh, N. N., Van, T. P., Lee, S. J., Farrar, J., Wills, B., Tran, H. T., and Simmons, C. P. (2010) A Randomized Controlled Trial of Chloroquine for the Treatment of Dengue in Vietnamese Adults. *PLoS Neglected Trop. Dis. 4* (8), No. e785.
(69) Yan, Y., Zou, Z., Sun, Y., Li, X., Xu, K.-F., Wei, Y., Jin, N., and Jiang, C. (2013) Anti-Malaria Drug Chloroquine Is Highly Effective in Treating Avian Influenza A H5N1 Virus Infection in an Animal Model. *Cell Res. 23* (2), 300−302.
(70) Mauthe, M., Orhon, I., Rocchi, C., Zhou, X., Luhr, M., Hijlkema, K.-J., Coppes, R. P., Engedal, N., Mari, M., and Reggiori, F. (2018) Chloroquine Inhibits Autophagic Flux by Decreasing Autophagosome-Lysosome Fusion. *Autophagy 14* (8), 1435−1455.
(71) Vincent, M. J., Bergeron, E., Benjannet, S., Erickson, B. R., Rollin, P. E., Ksiazek, T. G., Seidah, N. G., and Nichol, S. T. (2005) Chloroquine Is a Potent Inhibitor of SARS Coronavirus Infection and Spread. *Virol. J. 2* (1), 69.
(72) Keyaerts, E., Vijgen, L., Maes, P., Neyts, J., and Ranst, M. V. (2004) In Vitro Inhibition of Severe Acute Respiratory Syndrome Coronavirus by Chloroquine. *Biochem. Biophys. Res. Commun. 323* (1), 264−268.
(73) de Wilde, A. H., Jochmans, D., Posthuma, C. C., Zevenhoven-Dobbe, J. C., van Nieuwkoop, S., Bestebroer, T. M., van den Hoogen, B. G., Neyts, J., and Snijder, E. J. (2014) Screening of an FDA-Approved Compound Library Identifies Four Small-Molecule Inhibitors of Middle East Respiratory Syndrome Coronavirus Replication in Cell Culture. *Antimicrob. Agents Chemother. 58* (8), 4875.
(74) Barnard, D. L., Day, C. W., Bailey, K., Heiner, M., Montgomery, R., Lauridsen, L., Chan, P. K., and Sidwell, R. W. (2006) Evaluation of Immunomodulators, Interferons and Known in Vitro SARS-CoV Inhibitors for Inhibition of SARS-Cov Replication in BALB/c Mice. *Antivir. Chem. Chemother. 17* (5), 275−284.
(75) Liu, J., Cao, R., Xu, M., Wang, X., Zhang, H., Hu, H., Li, Y., Hu, Z., Zhong, W., and Wang, M. (2020) Hydroxychloroquine, a Less

https://dx.doi.org/10.1021/acsinfecdis.0c00343
ACS Infect. Dis. XXXX, XXX, XXX−XXX

Toxic Derivative of Chloroquine, Is Effective in Inhibiting SARS-CoV-2 Infection in Vitro. *Cell Discovery 6* (1), 16.
(76) Yao, X., Ye, F., Zhang, M., Cui, C., Huang, B., Niu, P., Liu, X., Zhao, L., Dong, E., Song, C., Zhan, S., Lu, R., Li, H., Tan, W., and Liu, D. (2020) In Vitro Antiviral Activity and Projection of Optimized Dosing Design of Hydroxychloroquine for the Treatment of Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2). *Clin. Infect. Dis.*, No. ciaa237.
(77) Pastick, K. A., Okafor, E. C., Wang, F., Lofgren, S. M., Skipper, C. P., Nicol, M. R., Pullen, M. F., Rajasingham, R., McDonald, E. G., Lee, T. C., Schwartz, I. S., Kelly, L. E., Lother, S. A., Mitjà, O., Letang, E., Abassi, M., and Boulware, D. R. (2020) Review: Hydroxychloroquine and Chloroquine for Treatment of SARS-CoV-2 (COVID-19). *Open Forum Infect. Dis. 7*, ofaa130.
(78) Geleris, J., Sun, Y., Platt, J., Zucker, J., Baldwin, M., Hripcsak, G., Labella, A., Manson, D. K., Kubin, C., Barr, R. G., Sobieszczyk, M. E., and Schluger, N. W. (2020) Observational Study of Hydroxychloroquine in Hospitalized Patients with Covid-19. *N. Engl. J. Med. 382* (25), 2411−2418.
(79) Arshad, S., Kilgore, P., Chaudhry, Z. S., Jacobsen, G., Wang, D. D., Huitsing, K., Brar, I., Alangaden, G. J., Ramesh, M. S., McKinnon, J. E., O'Neill, W., Zervos, M., Nauriyal, V., Hamed, A. A., Nadeem, O., Swiderek, J., Godfrey, A., Jennings, J., Gardner-Gray, J., Ackerman, A. M., Lezotte, J., Ruhala, J., Fadel, R., Vahia, A., Gudipati, S., Parraga, T., Shallal, A., Maki, G., Tariq, Z., Suleyman, G., Yared, N., Herc, E., Williams, J., Lanfranco, O. A., Bhargava, P., and Reyes, K. (2020) Treatment with Hydroxychloroquine, Azithromycin, and Combination in Patients Hospitalized with COVID-19. *Int. J. Infect. Dis. 97*, 396−403.
(80) Borba, M. G. S., Val, F. F. A., Sampaio, V. S., Alexandre, M. A. A., Melo, G. C., Brito, M., Mourão, M. P. G., Brito-Sousa, J. D., Baía-Da-Silva, D., Guerra, M. V. F., Hajjar, L. A., Pinto, R. C., Balieiro, A. A. S., Pacheco, A. G. F., Santos, J. D. O., Jr, Naveca, F. G., Xavier, M. S., Siqueira, A. M., Schwarzbold, A., Croda, J., Nogueira, M. L., Romero, G. A. S., Bassat, Q., Fontes, C. J., Albuquerque, B. C., Daniel-Ribeiro, C.-T., Monteiro, W. M., Lacerda, M. V. G., and for the CloroCovid-19 Team (2020) Effect of High vs Low Doses of Chloroquine Diphosphate as Adjunctive Therapy for Patients Hospitalized With Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) Infection: A Randomized Clinical Trial. *JAMA Netw. Open 3* (4), No. e208857.
(81) *Coronavirus (COVID-19) Update: FDA Revokes Emergency Use Authorization for Chloroquine and Hydroxychloroquine*, https://www.fda.gov/news-events/press-announcements/coronavirus-covid-19-update-fda-revokes-emergency-use-authorization-chloroquine-and.
(82) Tay, M. Z., Poh, C. M., Rénia, L., MacAry, P. A., and Ng, L. F. P. (2020) The Trinity of COVID-19: Immunity, Inflammation and Intervention. *Nat. Rev. Immunol. 20*, 363.
(83) Shi, Y., Wang, Y., Shao, C., Huang, J., Gan, J., Huang, X., Bucci, E., Piacentini, M., Ippolito, G., and Melino, G. (2020) COVID-19 Infection: The Perspectives on Immune Responses. *Cell Death Differ. 27* (5), 1451−1454.
(84) Venkataraman, T., Coleman, C. M., and Frieman, M. B. (2017) Overactive Epidermal Growth Factor Receptor Signaling Leads to Increased Fibrosis after Severe Acute Respiratory Syndrome Coronavirus Infection. *J. Virol. 91* (12), No. e00182-17.
(85) Chu, H., Chan, J. F.-W., Wang, Y., Yuen, T. T.-T., Chai, Y., Hou, Y., Shuai, H., Yang, D., Hu, B., Huang, X., Zhang, X., Cai, J.-P., Zhou, J., Yuan, S., Kok, K.-H., To, K. K.-W., Chan, I. H.-Y., Zhang, A. J., Sit, K.-Y., Au, W.-K., and Yuen, K.-Y. (2020) Comparative Replication and Immune Activation Profiles of SARS-CoV-2 and SARS-CoV in Human Lungs: An Ex Vivo Study with Implications for the Pathogenesis of COVID-19. *Clin. Infect. Dis.*, No. ciaa410.
(86) Chan, J. F.-W., Yao, Y., Yeung, M.-L., Deng, W., Bao, L., Jia, L., Li, F., Xiao, C., Gao, H., Yu, P., Cai, J.-P., Chu, H., Zhou, J., Chen, H., Qin, C., and Yuen, K.-Y. (2015) Treatment With Lopinavir/Ritonavir or Interferon-B1b Improves Outcome of MERS-CoV Infection in a Nonhuman Primate Model of Common Marmoset. *J. Infect. Dis. 212* (12), 1904−1913.
(87) Loutfy, M. R., Blatt, L. M., Siminovitch, K. A., Ward, S., Wolff, B., Lho, H., Pham, D. H., Deif, H., LaMere, E. A., Chang, M., Kain, K. C., Farcas, G. A., Ferguson, P., Latchford, M., Levy, G., Dennis, J. W., Lai, E. K. Y., and Fish, E. N. (2003) Interferon Alfacon-1 Plus Corticosteroids in Severe Acute Respiratory SyndromeA Preliminary Study. *JAMA 290* (24), 3222−3228.
(88) Lokugamage, K. G., Schindewolf, C., and Menachery, V. D. (2020) SARS-CoV-2 Sensitive to Type I Interferon Pretreatment. *bioRxiv*, DOI: 10.1101/2020.03.07.982264.
(89) Zhou, Q., Chen, V., Shannon, C. P., Wei, X.-S., Xiang, X., Wang, X., Wang, Z.-H., Tebbutt, S. J., Kollmann, T. R., and Fish, E. N. (2020) Interferon-A2b Treatment for COVID-19. *Front. Immunol. 11*, 1061.
(90) Muir, A. J., Arora, S., Everson, G., Flisiak, R., George, J., Ghalib, R., Gordon, S. C., Gray, T., Greenbloom, S., Hassanein, T., Hillson, J., Horga, M. A., Jacobson, I. M., Jeffers, L., Kowdley, K. V., Lawitz, E., Lueth, S., Rodriguez-Torres, M., Rustgi, V., Shemanski, L., Shiffman, M. L., Srinivasan, S., Vargas, H. E., Vierling, J. M., Xu, D., Lopez-Talavera, J. C., and Zeuzem, S. (2014) A Randomized Phase 2b Study of Peginterferon Lambda-1a for the Treatment of Chronic HCV Infection. *J. Hepatol. 61* (6), 1238−1246.
(91) Broggi, A., Granucci, F., and Zanoni, I. (2020) Type III Interferons: Balancing Tissue Tolerance and Resistance to Pathogen Invasion. *J. Exp. Med. 217*, No. e20190295.
(92) Prokunina-Olsson, L., Alphonse, N., Dickenson, R. E., Durbin, J. E., Glenn, J. S., Hartmann, R., Kotenko, S. V., Lazear, H. M., O'Brien, T. R., Odendall, C., Onabajo, O. O., Piontkivska, H., Santer, D. M., Reich, N. C., Wack, A., and Zanoni, I. (2020) COVID-19 and Emerging Viral Infections: The Case for Interferon Lambda. *J. Exp. Med. 217*, No. e20200653.
(93) Hamming, O. J., Terczyńska-Dyla, E., Vieyres, G., Dijkman, R., Jørgensen, S. E., Akhtar, H., Siupka, P., Pietschmann, T., Thiel, V., and Hartmann, R. (2013) Interferon Lambda 4 Signals via the IFNλ Receptor to Regulate Antiviral Activity against HCV and Coronaviruses. *EMBO J. 32* (23), 3055−3065.
(94) Mordstein, M., Neugebauer, E., Ditt, V., Jessen, B., Rieger, T., Falcone, V., Sorgeloos, F., Ehl, S., Mayer, D., Kochs, G., Schwemmle, M., Günther, S., Drosten, C., Michiels, T., and Staeheli, P. (2010) Lambda Interferon Renders Epithelial Cells of the Respiratory and Gastrointestinal Tracts Resistant to Viral Infections. *J. Virol. 84* (11), 5670.
(95) Chen, L., Liu, H. G., Liu, W., Liu, J., Liu, K., Shang, J., Deng, Y., and Wei, S. (2020) [Analysis of Clinical Features of 29 Patients with 2019 Novel Coronavirus Pneumonia]. *Zhonghua Jie He He Hu Xi Za Zhi Zhonghua Jiehe He Huxi Zazhi Chin. J. Tuberc. Respir. Dis. 43* (3), 203−208.
(96) Russell, C. D., Millar, J. E., and Baillie, J. K. (2020) Clinical Evidence Does Not Support Corticosteroid Treatment for 2019-NCoV Lung Injury. *Lancet 395* (10223), 473−475.
(97) Shang, L., Zhao, J., Hu, Y., Du, R., and Cao, B. (2020) On the Use of Corticosteroids for 2019-NCoV Pneumonia. *Lancet 395* (10225), 683−684.
(98) Li, H., Chen, C., Hu, F., Wang, J., Zhao, Q., Gale, R. P., and Liang, Y. (2020) Impact of Corticosteroid Therapy on Outcomes of Persons with SARS-CoV-2, SARS-CoV, or MERS-CoV Infection: A Systematic Review and Meta-Analysis. *Leukemia 34* (6), 1503−1511.
(99) Horby, P., Lim, W. S., Emberson, J., Mafham, M., Bell, J., Linsell, L., Staplin, N., Brightling, C., Ustianowski, A., Elmahi, E., Prudon, B., Green, C., Felton, T., Chadwick, D., Rege, K., Fegan, C., Chappell, L. C., Faust, S. N., Jaki, T., Jeffery, K., Montgomery, A., Rowan, K., Juszczak, E., Baillie, J. K., Haynes, R., and Landray, M. J. (2020) Effect of Dexamethasone in Hospitalized Patients with COVID-19: Preliminary Report. *medRxiv*, DOI: 10.1101/2020.06.22.20137273.
(100) Kubo, S., Nakayamada, S., Sakata, K., Kitanaga, Y., Ma, X., Lee, S., Ishii, A., Yamagata, K., Nakano, K., and Tanaka, Y. (2018) Janus Kinase Inhibitor Baricitinib Modulates Human Innate and Adaptive Immune System. *Front. Immunol. 9*, 1510.

https://dx.doi.org/10.1021/acsinfecdis.0c00343
ACS Infect. Dis. XXXX, XXX, XXX−XXX

(101) Yaekura, A., Yoshida, K., Morii, K., Oketani, Y., Okumura, I., Kaneshiro, K., Shibanuma, N., Sakai, Y., and Hashiramoto, A. (2020) Chronotherapy Targeting Cytokine Secretion Attenuates Collagen-Induced Arthritis in Mice. *Int. Immunopharmacol. 84*, 106549.
(102) Cantini, F., Niccoli, L., Matarrese, D., Nicastri, E., Stobbione, P., and Goletti, D. (2020) Baricitinib Therapy in COVID-19: A Pilot Study on Safety and Clinical Impact. *J. Infect. 81*, P318−356.
(103) Titanji, B. K., Farley, M. M., Mehta, A., Connor-Schuler, R., Moanna, A., Cribbs, S. K., O'Shea, J., DeSilva, K., Chan, B., Edwards, A., Gavegnano, C., Schinazi, R. F., and Marconi, V. C. (2020) Use of Baricitinib in Patients with Moderate and Severe COVID-19. *Clin. Infect. Dis.*, No. ciaa879.
(104) Wang, M., Rule, S., Zinzani, P. L., Goy, A., Casasnovas, O., Smith, S. D., Damaj, G., Doorduijn, J., Lamy, T., Morschhauser, F., Panizo, C., Shah, B., Davies, A., Eek, R., Dupuis, J., Jacobsen, E., Kater, A. P., Le Gouill, S., Oberic, L., Robak, T., Covey, T., Dua, R., Hamdy, A., Huang, X., Izumi, R., Patel, P., Rothbaum, W., Slatter, J. G., and Jurczak, W. (2018) Acalabrutinib in Relapsed or Refractory Mantle Cell Lymphoma (ACE-LY-004): A Single-Arm, Multicentre, Phase 2 Trial. *Lancet 391* (10121), 659−667.
(105) Roschewski, M., Lionakis, M. S., Sharman, J. P., Roswarski, J., Goy, A., Monticelli, M. A., Roshon, M., Wrzesinski, S. H., Desai, J. V., Zarakas, M. A., Collen, J., Rose, K. M., Hamdy, A., Izumi, R., Wright, G. W., Chung, K. K., Baselga, J., Staudt, L. M., and Wilson, W. H. (2020) Inhibition of Bruton Tyrosine Kinase in Patients with Severe COVID-19. *Sci. Immunol. 5* (48), No. eabd0110.
(106) Somers, E. C., Eschenauer, G. A., Troost, J. P., Golob, J. L., Gandhi, T. N., Wang, L., Zhou, N., Petty, L. A., Baang, J. H., Dillman, N. O., Frame, D., Gregg, K. S., Kaul, D. R., Nagel, J., Patel, T. S., Zhou, S., Lauring, A. S., Hanauer, D. A., Martin, E., Sharma, P., Fung, C. M., and Pogue, J. M. (2020) Tocilizumab for Treatment of Mechanically Ventilated Patients with COVID-19. *Clin. Infect. Dis.*, No. ciaa954.
(107) Rossotti, R., Travi, G., Ughi, N., Corradin, M., Baiguera, C., Fumagalli, R., Bottiroli, M., Mondino, M., Merli, M., Bellone, A., Basile, A., Ruggeri, R., Colombo, F., Moreno, M., Pastori, S., Perno, C. F., Tarsia, P., Epis, O. M., and Puoti, M. (2020) SAFETY AND EFFICACY OF ANTI-IL6-RECEPTOR TOCILIZUMAB USE IN SEVERE AND CRITICAL PATIENTS AFFECTED BY CORONAVIRUS DISEASE 2019: A COMPARATIVE ANALYSIS. *J. Infect.*, DOI: 10.1016/j.jinf.2020.07.008.
(108) *Sanofi and Regeneron provide update on Kevzara (sarilumab) Phase 3 U.S. Trial in COVID-19 patients*, https://www.sanofi.com/en/media-room/press-releases/2020/2020-07-02-22-30-00.
(109) Wang, H., Liu, B., Tang, Y., Chang, P., Yao, L., Huang, B., Lodato, R. F., and Liu, Z. (2019) Improvement of Sepsis Prognosis by Ulinastatin: A Systematic Review and Meta-Analysis of Randomized Controlled Trials. *Front. Pharmacol. 10*, 1370.
(110) Liu, R., Qi, H., Wang, J., Wang, Y., Cui, L., Wen, Y., and Yin, C. (2014) Ulinastatin Activates the Renin−Angiotensin System to Ameliorate the Pathophysiology of Severe Acute Pancreatitis. *J. Gastroenterol. Hepatol. 29* (6), 1328−1337.
(111) South, A. M., Tomlinson, L., Edmonston, D., Hiremath, S., and Sparks, M. A. (2020) Controversies of Renin−Angiotensin System Inhibition during the COVID-19 Pandemic. *Nat. Rev. Nephrol. 16* (6), 305−307.
(112) Simões e Silva, A., Silveira, K., Ferreira, A., and Teixeira, M. (2013) ACE2, Angiotensin-(1−7) and Mas Receptor Axis in Inflammation and Fibrosis. *Br. J. Pharmacol. 169* (3), 477−492.
(113) Kuba, K., Imai, Y., Rao, S., Gao, H., Guo, F., Guan, B., Huan, Y., Yang, P., Zhang, Y., Deng, W., Bao, L., Zhang, B., Liu, G., Wang, Z., Chappell, M., Liu, Y., Zheng, D., Leibbrandt, A., Wada, T., Slutsky, A. S., Liu, D., Qin, C., Jiang, C., and Penninger, J. M. (2005) A Crucial Role of Angiotensin Converting Enzyme 2 (ACE2) in SARS Coronavirus−Induced Lung Injury. *Nat. Med. 11* (8), 875−879.
(114) Xu, J., Zhao, S., Teng, T., Abdalla, E. A., Zhu, W., Xie, L., Wang, Y., and Guo, X. (2020) Systematic Comparison of Two Animal-to-Human Transmitted Human Coronaviruses: SARS-CoV-2 and SARS-CoV. *Viruses 12* (2), 244.
(115) Kassiri, Z., Zhong, J., Guo, D., Basu, R., Wang, X., Liu, P. P., Scholey, J. W., Penninger, J. M., and Oudit, G. Y. (2009) Loss of Angiotensin-Converting Enzyme 2 Accelerates Maladaptive Left Ventricular Remodeling in Response to Myocardial Infarction. *Circ.: Heart Failure 2* (5), 446−455.
(116) Ju, M., He, H., Chen, S., Liu, Y., Liu, Y., Pan, S., Zheng, Y., Xuan, L., Zhu, D., and Luo, Z. (2019) Ulinastatin Ameliorates LPS-induced Pulmonary Inflammation and Injury by Blocking the MAPK/NF-κB Signaling Pathways in Rats. *Mol. Med. Rep.*, 3347−3354.
(117) Wang, W.-K., Lu, Q.-H., Wang, X., Wang, B., Wang, J., Gong, H.-P., Wang, L., Li, H., and Du, Y.-M. (2017) Ulinastatin Attenuates Diabetes-Induced Cardiac Dysfunction by the Inhibition of Inflammation and Apoptosis. *Exp. Ther. Med. 14* (3), 2497−2504.
(118) He, G., Li, Q., Li, W., Ruan, Y., Xiong, X., Song, X., and Zeng, F. (2020) Effect of Ulinastatin on Interleukins and Pulmonary Function in Bypass Patients: A Meta-Analysis of Randomized Controlled Trials. *Herz 45*, 335.
(119) Zhang, X., Zhu, Z., Jiao, W., Liu, W., Liu, F., and Zhu, X. (2019) Ulinastatin Treatment for Acute Respiratory Distress Syndrome in China: A Meta-Analysis of Randomized Controlled Trials. *BMC Pulm. Med. 19* (1), 196.
(120) Khajah, M. A., Fateel, M. M., Ananthalakshmi, K. V., and Luqmani, Y. A. (2016) Anti-Inflammatory Action of Angiotensin 1−7 in Experimental Colitis. *PLoS One 11* (3), No. e0150861.
(121) Hardie, W. D., Davidson, C., Ikegami, M., Leikauf, G. D., Le Cras, T. D., Prestridge, A., Whitsett, J. A., and Korfhagen, T. R. (2008) EGF Receptor Tyrosine Kinase Inhibitors Diminish Transforming Growth Factor-α-Induced Pulmonary Fibrosis. *Am. J. Physiol.-Lung Cell. Mol. Physiol. 294* (6), L1217−L1225.
(122) Kim, Y.-J., Cubitt, B., Chen, E., Hull, M. V., Chatterjee, A. K., Cai, Y., Kuhn, J. H., and de la Torre, J. C. (2019) The ReFRAME Library as a Comprehensive Drug Repurposing Library to Identify Mammarenavirus Inhibitors. *Antiviral Res. 169*, 104558.
(123) Gordon, D. E., Jang, G. M., Bouhaddou, M., Xu, J., Obernier, K., White, K. M., O'Meara, M. J., Rezelj, V. V., Guo, J. Z., Swaney, D. L., Tummino, T. A., Huettenhain, R., Kaake, R. M., Richards, A. L., Tutuncuoglu, B., Foussard, H., Batra, J., Haas, K., Modak, M., Kim, M., Haas, P., Polacco, B. J., Braberg, H., Fabius, J. M., Eckhardt, M., Soucheray, M., Bennett, M. J., Cakir, M., McGregor, M. J., Li, Q., Meyer, B., Roesch, F., Vallet, T., Mac Kain, A., Miorin, L., Moreno, E., Naing, Z. Z. C., Zhou, Y., Peng, S., Shi, Y., Zhang, Z., Shen, W., Kirby, I. T., Melnyk, J. E., Chorba, J. S., Lou, K., Dai, S. A., Barrio-Hernandez, I., Memon, D., Hernandez-Armenta, C., Lyu, J., Mathy, C. J. P., Perica, T., Pilla, K. B., Ganesan, S. J., Saltzberg, D. J., Rakesh, R., Liu, X., Rosenthal, S. B., Calviello, L., Venkataramanan, S., Liboy-Lugo, J., Lin, Y., Huang, X.-P., Liu, Y., Wankowicz, S. A., Bohn, M., Safari, M., Ugur, F. S., Koh, C., Savar, N. S., Tran, Q. D., Shengjuler, D., Fletcher, S. J., O'Neal, M. C., Cai, Y., Chang, J. C. J., Broadhurst, D. J., Klippsten, S., Sharp, P. P., Wenzell, N. A., Kuzuoglu, D., Wang, H.-Y., Trenker, R., Young, J. M., Cavero, D. A., Hiatt, J., Roth, T. L., Rathore, U., Subramanian, A., Noack, J., Hubert, M., Stroud, R. M., Frankel, A. D., Rosenberg, O. S., Verba, K. A., Agard, D. A., Ott, M., Emerman, M., Jura, N., von Zastrow, M., Verdin, E., Ashworth, A., Schwartz, O., d'Enfert, C., Mukherjee, S., Jacobson, M., Malik, H. S., Fujimori, D. G., Ideker, T., Craik, C. S., Floor, S. N., Fraser, J. S., Gross, J. D., Sali, A., Roth, B. L., Ruggero, D., Taunton, J., Kortemme, T., Beltrao, P., Vignuzzi, M., García-Sastre, A., Shokat, K. M., Shoichet, B. K., and Krogan, N. J. (2020) A SARS-CoV-2 Protein Interaction Map Reveals Targets for Drug Repurposing. *Nature 583*, 459.
(124) Madelain, V., Nguyen, T. H. T., Olivo, A., de Lamballerie, X., Guedj, J., Taburet, A.-M., and Mentré, F. (2016) Ebola Virus Infection: Review of the Pharmacokinetic and Pharmacodynamic Properties of Drugs Considered for Testing in Human Efficacy Trials. *Clin. Pharmacokinet. 55* (8), 907−923.
(125) Pachetti, M., Marini, B., Benedetti, F., Giudici, F., Mauro, E., Storici, P., Masciovecchio, C., Angeletti, S., Ciccozzi, M., Gallo, R. C., Zella, D., and Ippodrino, R. (2020) Emerging SARS-CoV-2 Mutation

https://dx.doi.org/10.1021/acsinfecdis.0c00343
ACS Infect. Dis. XXXX, XXX, XXX−XXX

Hot Spots Include a Novel RNA-Dependent-RNA Polymerase Variant. *J. Transl. Med. 18* (1), 179.
(126) Haqqani, A. A., and Tilton, J. C. (2013) Entry Inhibitors and Their Use in the Treatment of HIV-1 Infection. *Antiviral Res. 98* (2), 158−170.
(127) Bekerman, E., Neveu, G., Shulla, A., Brannan, J., Pu, S.-Y., Wang, S., Xiao, F., Barouch-Bentov, R., Bakken, R. R., Mateo, R., Govero, J., Nagamine, C. M., Diamond, M. S., De Jonghe, S., Herdewijn, P., Dye, J. M., Randall, G., and Einav, S. (2017) Anticancer Kinase Inhibitors Impair Intracellular Viral Trafficking and Exert Broad-Spectrum Antiviral Effects. *J. Clin. Invest. 127* (4), 1338−1352.
(128) Lupberger, J., Zeisel, M. B., Xiao, F., Thumann, C., Fofana, I., Zona, L., Davis, C., Mee, C. J., Turek, M., Gorke, S., Royer, C., Fischer, B., Zahid, M. N., Lavillette, D., Fresquet, J., Cosset, F.-L., Rothenberg, S. M., Pietschmann, T., Patel, A. H., Pessaux, P., Doffoël, M., Raffelsberger, W., Poch, O., McKeating, J. A., Brino, L., and Baumert, T. F. (2011) EGFR and EphA2 Are Host Factors for Hepatitis C Virus Entry and Possible Targets for Antiviral Therapy. *Nat. Med. 17* (5), 589−595.
(129) Shiryaev, S. A., Mesci, P., Pinto, A., Fernandes, I., Sheets, N., Shresta, S., Farhy, C., Huang, C.-T., Strongin, A. Y., Muotri, A. R., and Terskikh, A. V. (2017) Repurposing of the Anti-Malaria Drug Chloroquine for Zika Virus Treatment and Prophylaxis. *Sci. Rep. 7* (1), 15771.
(130) Matt, U., Selchow, P., Dal Molin, M., Strommer, S., Sharif, O., Schilcher, K., Andreoni, F., Stenzinger, A., Zinkernagel, A. S., Zeitlinger, M., Sander, P., and Nemeth, J. (2017) Chloroquine Enhances the Antimycobacterial Activity of Isoniazid and Pyrazinamide by Reversing Inflammation-Induced Macrophage Efflux. *Int. J. Antimicrob. Agents 50* (1), 55−62.
(131) Gautret, P., Lagier, J.-C., Parola, P., Hoang, V. T., Meddeb, L., Mailhe, M., Doudier, B., Courjon, J., Giordanengo, V., Vieira, V. E., Dupont, H. T., Honoré, S., Colson, P., Chabrière, E., La Scola, B., Rolain, J.-M., Brouqui, P., and Raoult, D. (2020) Hydroxychloroquine and Azithromycin as a Treatment of COVID-19: Results of an Open-Label Non-Randomized Clinical Trial. *Int. J. Antimicrob. Agents 56*, 105949.
(132) Berry, S. M., Petzold, E. A., Dull, P., Thielman, N. M., Cunningham, C. K., Corey, G. R., McClain, M. T., Hoover, D. L., Russell, J., Griffiss, J. M., and Woods, C. W. (2016) A Response Adaptive Randomization Platform Trial for Efficient Evaluation of Ebola Virus Treatments: A Model for Pandemic Response. *Clin. Trials 13* (1), 22−30.
(133) Barker, A., Sigman, C., Kelloff, G., Hylton, N., Berry, D., and Esserman, L. (2009) I-SPY 2: An Adaptive Breast Cancer Trial Design in the Setting of Neoadjuvant Chemotherapy. *Clin. Pharmacol. Ther. 86* (1), 97−100.
(134) Dean, N. E., Gsell, P.-S., Brookmeyer, R., Crawford, F. W., Donnelly, C. A., Ellenberg, S. S., Fleming, T. R., Halloran, M. E., Horby, P., Jaki, T., Krause, P. R., Longini, I. M., Mulangu, S., Muyembe-Tamfum, J.-J., Nason, M. C., Smith, P. G., Wang, R., Henao-Restrepo, A. M., and De Gruttola, V. (2020) Creating a Framework for Conducting Randomized Clinical Trials during Disease Outbreaks. *N. Engl. J. Med. 382* (14), 1366−1369.

https://dx.doi.org/10.1021/acsinfecdis.0c00343
ACS Infect. Dis. XXXX, XXX, XXX−XXX