POSTĘPY MIKROBIOLOGII – ADVANCEMENTS OF MICROBIOLOGY
2020, 59, 3, 207–225
DOI: 10.21307/PM-2020.59.3.15

COVID-19 THERAPY: WHAT HAVE WE LEARNED IN 8 MONTHS?

Monika Adamczyk-Popławska*, Agnieszka Kwiatek

University of Warsaw, Faculty of Biology, Institute of Microbiology, Department of Molecular Virology

Received in June, accepted in August 2020

Abstract: SARS-CoV-2, a novel pathogenic human coronavirus, emerged in December of 2019 in Wuhan (Hubei province, China). In most cases, the infection causes a mild to moderate respiratory illness. However, a undefined group of infected may develop a severe or critical illness: Coronavirus disease 2019 (COVID-19) with acute respiratory distress syndrome (ARDS) and many other complications. Current efforts are focused on limiting the spread of the virus in the population. COVID-19 treatments are intensively evaluated, however, 8 months since the start of the pandemic and despite hundreds of clinical trials, our knowledge of effective treatments is still poor. In this review, we present the current status of drugs and treatments used during SARS-CoV-2 infection. Host-directed and virus-directed drugs, as well as new compounds specific for SARS-CoV-2 are presented.

TERAPIA COVID-19: CZEGO NAUCZYLIŚMY SIĘ W CIĄGU 8 MIESIĘCY?

Streszczenie: W grudniu 2019 w mieście Wuhan (Hubei, China) odnotowano pojawienie się nowego, ludzkiego, patogennego koronawirusa SARS-CoV-2. W większości przypadków, wywołana przez niego choroba przebiega bezobjawowo lub łagodnie. Jednakże, istnieje grupa osób u której infekcja powoduje rozwiniecie poważnej lub wręcz krytycznej choroby: *Coronavirus disease* 2019 (COVID-19) z zespołem ostrej niewydolności oddechowej i wieloma innymi powikłaniami. Obecnie walka z infekcją polega na próbach ograniczenia rozprzestrzeniania się wirusa wśród populacji. Intensywnie badane są też sposoby leczenia COVID-19. Jednak, 8 miesięcy po rozpoczęciu pandemii, pomimo setek badań klinicznych, wiedza na temat skutecznego leczenia jest wciąż ograniczona. W niniejszej pracy przedstawiamy obecny stan wiedzy na temat potencjalnych leków i metod leczenia stosowanych podczas zakażenia SARS-CoV-2. Omówione są leki ukierunkowane na komórki gospodarza lub na wirusa, a także nowe, specyficzne dla SARS-CoV-2 rozwiązania.

Key words: antiviral compounds, coronaviruses, COVID-19 treatment, SARS-CoV-2
Słowa kluczowe: leki przeciwwirusowe, koronawirusy, terapia COVID-19, SARS-CoV-2

1. Introduction

Coronaviruses (CoVs) are a group of RNA viruses that infect mammals and birds. In humans, CoVs mainly cause mild diseases, including the common cold. However, in the 21st century three pandemic strains of novel CoVs, highly pathogenic to humans, emerged. These three novel CoVs are zoonosis. In 2002–2003, we dealt with the Severe Acute Respiratory Syndrome coronavirus (currently, SARS-CoV) and in 2012, the Middle East Respiratory Syndrome coronavirus (MERS-CoV). The emergence of SARS-CoV-2 in December 2019 in Wuhan (Hubei province, China) marked the next introduction of a novel, highly pathogenic CoV into the human population [112]. In most cases, SARS-CoV-2 causes mild or moderate respiratory illness and the recovery does not require any special treatment. However, some infected individuals, with associated medical conditions, developed a severe disease called Coronavirus Disease 2019 (COVID-19) with such clinical manifestations as dyspnea, hypoxia, and lung lesions, being an indication for treatment in

Abbreviations:
ARDS: acute respiratory distress syndrome;
COVID-19: Coronavirus disease 2019;
CoVs: coronaviruses;
CP: convalescent plasma;
EUA: Emergency Use Authorization;
MERS: Middle East Respiratory Syndrome;
RdRp: RNA depended RNA polymerase;
SARS: Severe Acute Respiratory Syndrome

* Corresponding author: Monika Adamczyk-Popławska, University of Warsaw, Institute of Microbiology, Department of Molecular Virology, Miecznikowa 1, 02-096 Warsaw, Poland; Phone: 48-22 5541419; Fax: 48-22 5541402; e-mail: poplawa@biol.uw.edu.pl

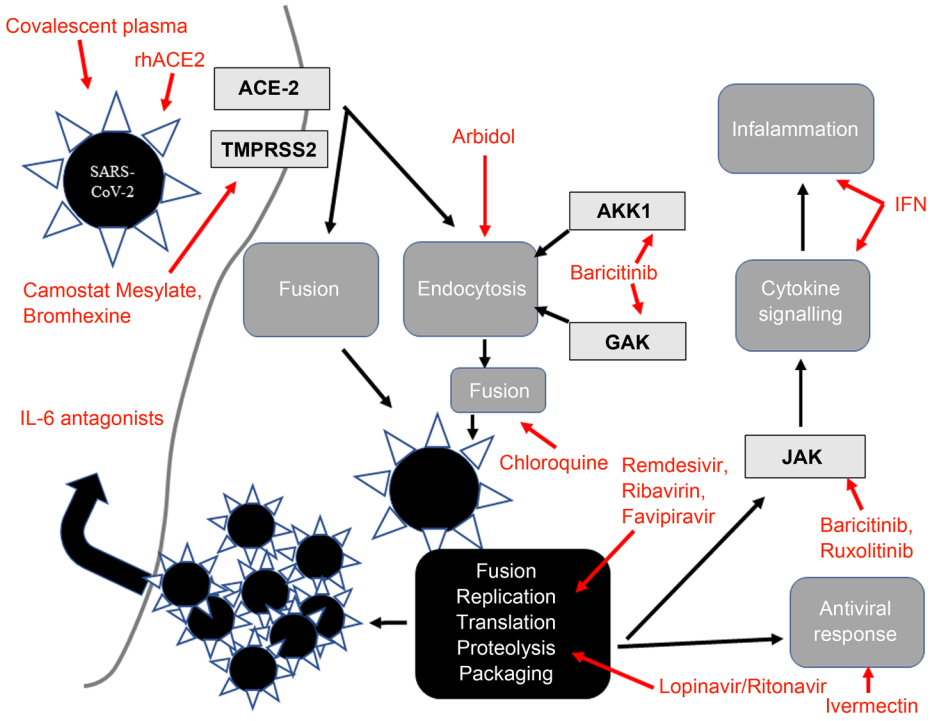

Fig. 1. Schematic view of drugs and compounds currently investigated for COVID-19 treatment

the intensive care unit. The most affected people experienced respiratory failure (ARDS, acute respiratory distress syndrome), shock, or multiorgan system dysfunction associated with cytokine storm. Such critical stage is often fatal [115, 188]. During the pandemic, millions of people around the world are infected at record speed, resulting in 823,687 deaths on August 26, 2020 (https://www.worldometers.info/coronavirus/). Such a situation has created a need for unprecedented urgency. Currently, management of COVID-19 is mainly focused on infection prevention, case detection and monitoring, and supportive care. Since the situation is critical, there is an urgent necessity for elaboration of an effective treatment of symptomatic COVID-19 patients and for the decrease of duration of virus carriage, in order to limit the spread of pathogen between people. Right now, the interest is focused on several known, broad-activity range, antiviral drugs, which are found to have an anti-SARS-CoV-2 potential. The majority of such medicine have been already tested *in vitro*, on animal models, during clinical trials and are approved for use in humans for other conditions than CoVs infections (Fig. 1).

The development of new drugs, specific for human CoVs and especially for SARS-CoV-2 needs time. Such investigations are yet undertaken, since our knowledge about SARS-CoV-2 biology is constantly increasing. In this review, we summarized the targeting strategies, used since December 2020 in COVID-19 treatment and present our current knowledge and describe their mechanism of action.

2. Host-directed drugs

We have recently described the Betacoronavirus family and its cycle of replication and detailed the known functions of SARS-CoV-2 encoded proteins (Kwiatek and Adamczyk-Popławska, *Advancements of Microbiology*, page 197 of this issue).

2.1. Antiparasitic drugs with potential for repurposing

Chloroquine and Hydroxychloroquine

Chloroquine ($C_{18}H_{26}ClN_3$) and its derivative Hydroxychloroquine ($C_{18}H_{26}ClN_3O$) are antimalarial and autoimmune disease drugs. The antiviral activity of Chloroquine was demonstrated against different viruses, including: HIV, Dengue, Hepatitis C, Chikungunya, Influenza, Ebola and novel CoVs (SARS-CoV and MERS-CoV) [150]. Both quinolones inhibit the entry of viruses into host cells. Both compounds were found to block viral infection by increasing endosomal pH and by this impeding viral envelope/endosomal membrane fusion necessary to the liberation of viral capsid into the cytoplasm of infected cell during endocytosis [148]. Studies concerning SARS-CoV demonstrated that Chloroquine, in addition to pH modulation, also interferes with the terminal glycosylation of ACE2, which is the receptor for SARS-CoV, but also for SARS-CoV-2 [74, 201]. ACE2 glycosylation may inhibit the CoV/ACE2 binding and thus abrogate the infection [174]. Chloroquine and Hydroxychloroquine were also

demonstrated to bind with high affinity to sialic acids and gangliosides present on the surface of human cells. During virus endocytosis, such binding may interfere with the attachment of SARS-CoV-2 to lipid rafts and inhibit the contact between SARS-CoV-2 Spike protein and the ACE2 receptor [51].

The *in vitro* inhibitory effect of Chloroquine on the replication of SARS-CoV-2 (i. e. the decrease of viral copy numbers in the cell supernatants) was reported in assays, testing the infection of Vero E6 cells [178].

Quinolones also modulate the innate immune system: Chloroquine and Hydroxychloroquine reduce the activation of the Toll-like receptor (TLR) signaling [97] and Chloroquine reduces synthesis and secretion of several proinflammatory cytokines (TNF-α, IL-1β, IL-6) [86]. This effect may be useful in control of cytokine storm associated with SARS-CoV-2 infection.

Antiviral efficacy of Chloroquine against SARS-CoV-2 was observed by several subsequent clinical trials conducted in China [56]. Hydroxychloroquine, with better clinical safety profile during long-term use, and allowing the administration of high daily dose was also tested against SARS-CoV-2 [196]. A small size pilot study, on 36 patients with different severity of COVID-19 symptoms, was conducted and the antiviral effect of Hydroxychloroquine was demonstrated by the decrease (or even disappearance) of the viral load in nasopharyngeal swabs in patients treated daily with 600 mg of Hydroxychloroquine for 3–6 days [58]. The beneficial effect was reinforced by co-administration of Azithromycin (macrolide antibiotic), [58, 116, 145]. *In vitro* combination of Hydroxychloroquine with Azithromycin led to significant inhibition of SARS-CoV-2 replication in Vero E6 cells [5]. Under an EUA (Emergency Use Authorization) issued on March 28, 2020, U.S. Food and Drugs Administration (FDA) allowed the usage of Chloroquine phosphate and Hydroxychloroquine in certain hospitalized COVID-19 patients outside of clinical trials.

However further studies did not confirm the beneficial effect of Chloroquine or Hydroxychloroquine in COVID-19 treatment. On May 08, 2020, at least 7 clinical trials were completed with published or pre-published results, but the beneficial effect of Chloroquine or its derivative are not conclusive [40]. Some authors even suggest, that these compounds may increase the severity of the illness [152]. Indeed, the progression of the severity of the disease was reported in one Hydroxychloroquine-treated patient and no beneficial effect of Hydroxychloroquine treatment on prognosis of 14 treated patients was observed during a recent study [36]. Another small cohort study with 11 patients also did not proved the protective efficacy of Hydroxychloroquine in COVID-19 patients and even reported one death and one adverse effect (18.1%) after administration of Hydroxychloroquine, comparing to control group [127]. An open label, randomized controlled trial (ChiCTR2000029868) tested long term and high dose (1200 mg for three days, followed by 800 mg for 2–3 weeks) of Hydroxychloroquine, administrated to patients with mild to moderate COVID-19. No effect on virus clearance, compared to standard care (supplemental oxygen, concomitant antiviral medication or antibiotics) was observed at day 28. More adverse events were observed in Hydroxychloroquine-treated group than in control group. The most common adverse event was diarrhea, reported in 10% of patients. Serious adverse events were also reported and included disease progression, upper respiratory infection and kidney injury [164]. Another report described a comparative observational study on data collected from routine care. Patients with pneumonia, who required oxygen were treated with 600 mg/day of Hydroxychloroquine. Treatment did not improved disease progression or patient survival as compared to control group without Hydroxychloroquine at day 21. Almost 10% of Hydroxychloroquine-treated patients had the drug stopped because of changes in electrocardiogram [118].

On May 27, 2020 European governments, following World Health Organization (WHO) decision from May 25, acted to pause the use of quinolones to treat patients suffering from COVID-19 due to safety concerns. The decision was partially taken on the basis studies mentioned above and on a observational study, that is currently retracted due to inconsistencies in the patient data [122]. On June 3, 2020, the WHO decided to continue trials with quinolones, but the FDA retracted the EUA issued to Hydroxychloroquine for use in COVID-19 patients.

Until August 2020, several clinical trials were finished and the conclusions is that the efficacy of Chloroquine or Hydroxychloroquine (alone or in combination with azithromycin) to prevent or treat COVID-19 patients could not be established. The results of RECOVERY trial (NCT04381936) on 4674 patients did not demonstrate any clinical benefit from use of Hydroxychloroquine in hospitalized patients with COVID-19 (preprint [76]). The American College of Physicians recommends against the use of Chloroquine and its derivative [139]. On July 4, 2020, World Health Organization decided to discontinue the research concerning Hydroxychloroquine in "Solidarity" international clinical megatrial, conducted to find an effective treatment for COVID-19 (https://www.who.int/emergencies/diseases/ novel-coronavirus-2019/global-research-on-novel-coronavirus-2019-ncov/solidarity-clinical-trial-for-covid-19-treatments).

Ivermectin (Stromectol)

Ivermectin ($C_{48}H_{74}O_{14}$) is known since almost 40 years, a broad spectrum anti-parasitic drug licensed

in animal and human medicine [99]. In addition to its anti-parasitic effect, Ivermectin has been also demonstrated to limit *in vitro* replication of several RNA viruses [99, 175]. The target of Ivermectin would be the importin (IMP) α/β1, involved in transport of specific viral proteins into the host cell nucleus [125, 175]. Many RNA viruses that replicate in the cytoplasm require the nuclear transport of one or more proteins to inhibit the antiviral response of infected cells.

Ivermectin was described as reducing the presence of NS5 protein, encoded by Zika or Denga viruses, in nucleus of infected cells. The drug blocked the interaction between NS5 and IMP α/β transporter [175]. Studies on novel human CoVs proteins revealed a potential role for IMPα/β1 during infection in signal-dependent nucleocytoplasmic translocation of some viral proteins. For example ORF4b of MERS-CoV inhibits the induction of IFN-β synthesis in both the cytoplasm and nucleus [195]. The N-terminal domain of SARS-CoV N protein was found in nucleus, suggesting that the N may act as a shuttle protein and have several roles during SARS-CoV replication [165]. Recent studies demonstrated that Ivermectin inhibits SARS-CoV-2 replication *in vitro*. A single treatment was able to reduce by almost 5000-fold the virus yield after 48 h of infection [24]. However, the effect was obtained at a very high concentration of Ivermectin. High concentrations may be difficult to obtain *in vivo* and are not approved by FDA in humans [31].

Recently a pilot clinical trial evaluating Ivermectin effect on hospitalized adult patients with mild to moderate COVID-19 is terminated (NCT04343092). Sixteen patients received a single dose of Ivermectin in addition to Hydroxychloroquine and azithromycin treatment. All the patients from Ivermectin group were cured as compared to the control group, in which two person died. The mean time of hospitalization was significantly shorter (7.62 ± 2.75 days) for Ivermectin-treated patients, comparing to controls (13.22 ± 5.90 days). The virus clearance was faster in Ivermectin group [63]. The effectiveness of Ivermectin, as drug or adjuvant in association with other compounds, is currently being evaluated in various randomized clinical trials, but there is still insufficient evidence to draw a conclusion on benefits or harms during treatment of COVID-19 patients [30].

2.2. Host protease inhibitors

TMPRSS2 (Transmembrane Protein Serine 2) protease inhibitors

SARS-CoV-2 exploits ACE2 receptor, recognized by S protein for entry the host cell [74, 201]. Host cell proteases are responsible for coronavirus S protein activation and are essential for viral entry by encompassing S protein cleavage at the S1/S2 and the S2' sites. A recent report suggested that SARS-CoV-2 use the membrane-anchored serine protease TMPRSS2 for S protein priming for plasma membrane fusion [74]. The TMPRSS2 inhibitor might constitute an option for blocking fusion of viral envelope with cellular membrane and by this inhibit virus entry into host cells.

It was previously demonstrated that SARS-CoV and MERS-CoV have exhibited lower spread in TMPRSS2 deficient mice [80]. VeroE6 cells, expressing TMPRSS2, were highly susceptible to these CoVs infection [120]. TMPRSS2 protease participates also in priming of SARS-CoV-2 Spike protein [187]. Thus, inhibition of TMPRSS2 protease might constitute an option for blocking the fusion SARS-CoV-2 envelope with cellular membrane and those lowering the virus infectivity.

Camostat mesylate

Camostat mesylate ($C_{20}H_{22}N_4O_5$) is a drug with clinically proven safety, that is licensed in Japan since 1985, for the suppression of pancreatitis-induced pain due to its ability to inhibit inflammatory proteases [162, 171]. Camostat is serine protease inhibitor and was demonstrated as blocking TMPRSS2 and by this inhibiting the activation of SARS-CoV S protein at the cell surface *in vitro* [87]. Significant inhibition of SARS-CoV entry into TMPRSS2-expressing HeLa cells was also observed in case of simultaneous treatment with Camostat and EST, a cathepsin inhibitor [87]. The inhibition of the spread and pathogenesis of SARS-CoV by Camostat was validated in a murine model. The treatment improved the survival rate of infected animals by 60% [202]. Concerning SARS-CoV-2, Camostat was found to block the virus entry into Caco-2, Vero-TMPRSS2 cells and into the lung cell line Calu-3 *in vitro*, without cytotoxic effect [74]. Some authors pointed its potential as anti SARS-CoV-2 drug [171].

Until August 10, 2020, no clinical trial, evaluating the safety and impact of Camostat mesylate in treatment of COVID-19 was finished but several trials are recruiting.

Nafamostat mesylate

Nafamostat mesylate ($C_{21}H_{25}N_5O_8S_2$) is an FDA-approved drug, used in Asian countries for indications unrelated to coronaviruses. Nafamostat was described to prevent S-mediated membrane fusion between cells expressing Spike protein of MERS-CoV and cells expressing MERS-CoV receptor, CD26, with TMPRSS2 [194]. Moreover Nafamostat blocked MERS-CoV infection of Calu3 lung cells *in vitro* [194]. A short communication reported that Nafamostat inhibited SARS-CoV-2 S-mediated entry into host lung cells with 15-fold higher efficiency than Camostat and with a very low EC_{50} [75]. On August 10, 2020, three trials concerning Nafomostat as COVID-19 treatment are registered at US Clinical Trials Registry (https://clinicaltrials.gov).

Bromhexine

Bromhexine ($C_{14}H_{20}Br_2N_2$) is a widely prescribed medicine used for treatment of many respiratory conditions, associated with a disturbance of mucus secretion, and it is well tolerated and safe. In this context, Bromhexine efficacy is currently evaluated in clinical trial for chest congestion and cough in patients with suspected and mild SARS-CoV-2-associated pneumonia (NCT04273763). In fact, Bromhexine is also a potent and selective inhibitor of the TMPRSS2 [113] and was proposed previously as a candidate drug for treatment of SARS-CoV and MERS-CoV infections [155]. The inhibitory effect of Bromhexine suggest its repurposing either as a treatment or as a prophylactic agent in SARS-CoV-2 infections [117]. To date no results of clinical trial concerning Bromhexine were available (August 10, 2020).

2.3. Endocytosis inhibitors

Baricitinib (Olumiant)

Baricitinib ($C_{16}H_{17}N_7O_2S$) is an inhibitor of the release of cytokines by inhibiting the Janus-associated kinase (JAK) pathway. The drug is presently approved for treatment of rheumatoid arthritis [13]. Using artificial intelligence in a search for candidates that might have both, antiviral and anti-inflammatory activity during SARS-CoV-2 infection, Baricitinib was designed among others as acting on AP2-associated protein kinase 1 (AAK1) and on cyclin G-associated kinase (GAK), two regulators of endocytosis [138, 146]. As SARS-CoV-2 entry into ACE2 expressing cells involves, among others, clathrin-dependent endocytosis [157], Baricitinib was proposed as COVID-19 treatment, basing on prediction that this drug would reduce the ability of the virus to entry lung cells [146]. The inhibitory activity of Baricitinib was recently validated for AAK1, BIKE, GAK, and STK16 kinases [159]. The effect of Baricitinib on SARS-CoV-2 infectivity was also evaluated in 3D primary human liver spheroids. Pretreatment of spheroids with Baricitinib (400 or 800 nM) significantly reduced viral load by 30–40% without injury of liver cells [159].

Moreover, like other JAK inhibitors (Fedratinib or Ruxolitinib) Baricitinib may also reduce the effects of the increased cytokine levels that are frequently seen in patients with COVID-19. *In vitro*, Baricitinib reduces levels of cytokines implicated in COVID-19, including IL-2, IL-6, IL-10, IFN-γ, and G-CSF [159].

The safety of Baricitinib therapy, combined with Lopinavir/Ritonavir, in moderate COVID-19 pneumonia patients was positively evaluated in a pilot study (NCT04358614) [25]. Baricitinib-treatment was well tolerated with no serious adverse events. In the Baricitinib-treated group, clinical and respiratory function parameters significantly improved. Authors confirm that the use of Baricitinib may limit the cytokine-release syndrome associated with COVID-19 [25]. Baricitinib administration was also associated with improvement in clinical, radiologic, and viral load parameters along with a rapid decline in CRP protein and IL-6 levels in a small case study of 4 patients with bilateral COVID-19 pneumonia [159].

However, some authors warn against using this compound. Indeed, Baricitinib may cause lymphocytopenia, neutropenia and enhance the incidence of co-infections that are one of the leading causes of mortality of COVID-19 patients [138]. By inhibiting JAK kinases, Baricitinib may also interfere with antiviral activity of interferon [52]. Baricitinib was also described as re-activating latent viral infections of such viruses as Hepatitis B, so its safety should be evaluated [68].

Arbidol (Umifenovir)

Arbidol ($C_{22}H_{25}BrN_2O_3S$) is a broad-spectrum and well-tolerated antiviral drug, which has been approved in several countries for treatment of influenza infections [19]. This compound was described as active against numerous enveloped and non-enveloped viruses as Influenza A virus, RSV (respiratory syncytial virus), Rhinovirus, Coxsackie virus and Adenovirus *in vitro* and *in vivo* [156] and against the HBV (hepatitis B virus), HCV (Hepatitis C virus), Chikungunya virus, Reovirus and Hantaan virus *in vitro* [18]. The antiviral activity seems mainly to be related to the inhibition of the virus entry into host cell. It was demonstrated on *in vitro* HCV infection model, that Arbidol impeded virus attachment to cell plasma membrane, subsequently impaired the clathrin-dependent endocytosis and caused confinement of viral particles in clathrin-coated vesicles [17]. It has also been reported that Arbidol may exhibit some immunomodulatory activity by decreasing proinflammatory cytokine levels in cell cultures and *in vivo* (on mice and ferret model of influenza infection) and can alleviate lung lesions induced by the Influenza virus [181]. One study describes in vitro inhibition of SARS-CoV replication in GMK-AH-1 cell line by Arbidol and its derivative Arbidol mesylate [90]. Atomistic insights into the Arbidol inhibitory mechanisms on SARS-CoV-2 infection demonstrated that Arbidol binds to both S protein of the virus (receptor-binding-domain: RBD) and ACE2. Arbidol seems to stabilize at the RBD/ACE2 interface with a high affinity and induce structural rigidity, leading to inhibition of the conformational changes in the S-protein that is associated during the virus entry (preprint [134]).

The inhibitory effect of Arbidol on SARS-CoV-2 entry (75% of inhibition) was demonstrated *in vitro* on VeroE6 cells. The drug inhibits not only the virus binding to the studied cells (attachment) but also decrease

the release of SARS-CoV-2 from endosomal vesicles into cytoplasm [179].

Application of Arbidol for treatment of COVID-19 patients took mainly place in China on small number of patients. The reports were very encouraging for the outcome of COVID-19 patients treated with Arbidol [45, 108, 185, 203]. However, a recent evidence has not demonstrated that monotherapy with Arbidol provided any benefits in mild/moderate cases outcome [105]. Another retrospective study also did not found improvement in the prognosis or acceleration of SARS-CoV-2 clearance in 45 hospitalized patients treated with Arbidol versus 36 control patients [106]. Combined administration of Arbidol and IFNα2b improved the COVID-19 associated pneumonia in mild patients, but did not accelerate the virus clearance as compared to monotherapy with IFNα2b [192]. Randomized control clinical trial assessing the efficacy of Arbidol are ongoing.

2.4. Immunomodulating drugs affecting host

Interferons (IFNs)

Type I interferons (IFNs) are a group of cytokines playing an important role in host defense during viral infections [143]. IFNs suppresses viral infection by interfering with replication of the virus and by inducing innate and adaptive immune responses. However, viruses have evolved many mechanisms to evade the IFN activity [143]. Currently, treatment with exogenous type I IFNs is mainly restricted to administration during chronic infections with HBV or HCV [69, 147]. IFNα is also evaluated as drug in HIV infection treatment [59].

Concerning human CoVs, IFNα treatment of VeroE6 cells effectively inhibits the replication of SARS-CoV *in vitro* [161]. Moreover, the beneficial effect of IFNα was also demonstrated *in vivo* in SARS-CoV -infected cynomolgus monkeys, as well as on MERS-CoV-infected rhesus macaques [50, 66, 121]. The therapeutic benefit of IFNα in treatment of patients with SARS-CoV was demonstrated during a preliminary pilot study. Patients, treated with synthetic IFNα and corticosteroids, had improved oxygen saturation, more rapid resolution of radiographic lung abnormalities and lower levels of CRP [111]. Other clinical studies involving type I IFNs to treat MERS-CoV were conducted. The possibility of using IFNα or other IFNs for COVID-19 therapy was recently discussed [149]. Both IFNα and β were described as efficient during *in vitro* studies [73, 161] and in certain animal models, but failed to significantly improve the disease outcome in humans [149]. However, treatment guidelines of COVID-19 patients in China, recommend the administration of IFNα by inhalation in combination with Ribavirin [83].

SNG001, an inhaled formulation of IFNβ showed promise in a recently completed COVID-19 trial (NCT04385095). According to the study, the risk of developing symptoms that required ventilation or cause death was reduced by 79% in group receiving SNG001 compared to patients who received placebo during the treatment period of 16 days (https://www.synairgen.com/wp-content/uploads/2020/07/200720-Synairgen-announces-positive-results-from-trial-of-SNG001-in-hospitalised-COVID-19-patients.pdf).

Janus kinases (JAK) inhibitors

Acute SARS-CoV-2 infection is associated with hypercytokinaemia – the upregulation of pro-inflammatory cytokines and chemokines, also known as Cytokines Release Syndrome (CRS) or cytokine storm. This aspect of COVID-19 outcome was compared to hyperinflammatory syndrome associated with haemophagocytic lymphohistiocytosis (HLH). HLH is potentially fatal disease of normal but overactive T cells and macrophages that excessively produce proinflammatory cytokines, including IFNγ. Cytokine profiles in HLH and acute COVID-19 were described as similar, including the expression of IL-1β, IL-2, IL-6, IL-7, IL-8, TNF and chemokines or its ligands (CXCL10, CCL2) [123, 176]. The management of this cytokine storm is one of the major needs regarding COVID-19 infection.

JAK are a family of intracellular tyrosine kinases involved, among others, in cytokine (including IFNs) signaling. Several cytokines employ the intracellular signaling pathway mediated by JAKs. JAK inhibitors found application in a broad spectrum of diseases concerning immune system such as autoimmune and autoinflammatory disorders [22]. Among JAK inhibitors tested in COVID-19 treatment Ruxolitinib and, previously mentioned, Barinicitib are currently evaluated.

Ruxolitinib (Jakavi)

Ruxolitinib ($C_{17}H_{18}N_6$) is a JAK inhibitor approved for patient treatment by the FDA, though not for immunological disorders, but for treatment of intermediate or high-risk myelofibrosis [111]. The beneficial effect of Ruxolitinib on HLH outcome was demonstrated on both, preclinical models and in clinical practice [204]. Ruxolitinib has been shown to reduce cytokine levels and improve outcomes in different conditions. Ruxolitinib has attenuated T-cell activation and decreased inflammation in murine model of HLH infected with LCMV [4]. Several case reports and a pilot study (on five patients) determined the activity and safety of Ruxolitinib in adults with secondary HLH [3, 158]. Cytopenia improvement was observed, allowing transfusion independence, discontinuation of corticosteroids, and hospital discharge of Ruxolitinib-treated patients [3]. Other studies also confirmed the therapeutic effect of Ruxolitinib in HLH patients [21, 177].

Recently a bioinformatic approach, involving artificial intelligence, has identified Ruxolitinib among the

potential therapeutics for combining antiviral and anti-inflammatory treatments [160]. The safety of such treatment is of concern: among 5 HLH patients treated with Ruxolitinib a serious adverse event (grade 4 febrile neutropenia) was reported [3]. Another study mentioned leukopenia, thrombocytopenia, elevated transaminases, elevated bilirubin and hypertriglyceridemia in Ruxolitinib-treated HLH patients. However none of patients stopped the therapy due to its toxicity [177].

Ruxolitinib has been also used for treatment of COVID19 patients. The appearance of purpuric lesions on the skin and an erythrodermic rash were described in two COVID19 patients after administration of Ruxolitinib. However, these persons have several different treatments against SARS-CoV-2 before receiving Ruxolitinib and the effect may be the result of accumulated drugs [57]. A multicenter, single-blind, randomized controlled phase II trial, involving patients with severe coronavirus disease 2019 confirmed that levels of 7 cytokines (IL-6, nerve growth factor β, IL-12 (p40), migration inhibitory factor, MIP-1α, MIP-1β, and VEGF) were significantly decreased in the Ruxolitinib group. Ruxolitinib patients had a faster clinical improvement, chest computed tomography improvement and a faster recovery from lymphopenia [27]. A monocentric retrospective chart analysis on a subgroup of patients with severe COVID-19 that suffered from acute respiratory distress syndrome and multi organ failure also confirm the beneficial outcome of Ruxolitinib administration and the decrease of markers indicating the hyper inflammation. Side effects of short term treatment with Ruxolitinib were manageable (mild anemia and liver enzyme elevation) [98]. Data from RESPIRE Protocol (NCT04361903) have been published. This retrospective multicenter observational study concerned case series of 18 critically ill patients with COVID-19 and Acute respiratory distress syndrome (ARDS). Data collection demonstrated a rapid clinical response without evolution from non-invasive ventilation to mechanical ventilation in 16 of 18 patients treated with Ruxolitinib. After 14 days, 16 patients showed complete recovery of respiratory function. No adverse effect were observed [28]. Taken together, these data suggest that Ruxolitinib may lower the hyperinflammatory state observed in patients experiencing COVID-19-associated cytokine storm and ARDS.

Interleukin-6 inhibitors

In COVID-19 patients, a large number of T lymphocytes and mononuclear macrophages are activated, producing pro-inflammatory cytokines such as multifunctional interleukin-6 (IL-6). IL-6 is the key factor in acute inflammation and CRS [199]. IL-6 binds to its receptor on the target cells, causing the cytokine storm and severe inflammatory responses in lungs and other tissues and organs. Its inhibition may be of great value in reducing COVID-19-associated mortality [198, 199].

Tocilizumab (Actemra)

Tocilizumab is a recombinant humanized monoclonal antibody specific for IL-6 receptor. By binding IL-6 receptor Tocilizumab prevents IL-6 itself from binding and by thus blocks IL-6 transduction pathway. These IL-6 antagonist is currently administrated for the treatment of rheumatoid arthritis and systemic juvenile idiopathic arthritis [129]. Many reports described the beneficial effect of Tocilizumab administration to severe or critical COVID-19 patients. A small sample clinical trial (ChiCTR2000029765), on 21 patients with severe or critical COVID-19, has shown good efficacy of Tocilizumab administration: body temperature returned to normal, CRP (C reactive protein) decreased, oxygen intake was reduced and pulmonary lesions were absorbed [193]. Similar observation were made during treatment with Tocilizumab of 26-year-old patient in critical stage: 2 days after drug administration ventilation conditions improved and after 5 days laboratory parameters decreased (CRP returned to normal). Complete resolution of lung abnormalities was noticed on day 7 [172]. However, a recent report, describing two patients with COVID-19 and CRS complications reported progression to HLH despite treatment with Tocilizumab. One patient develop viral myocarditis [140]. So, the safety and clinical usefulness of Tocilizumab in the treatment of COVID-19-induced CRS has to be verified. Another study on 15 patients with different COVID-19 severity indicated that treatment failed in four patients but the solution may be a repeated dose of the Tocilizumab [114]. However, the more recent preliminary results from SMACORE (SMAtteo COvid19 REgistry) indicated that Tocilizumab administration did not reduce intensive care unit admission or mortality rate in a cohort of 21 patients, even with repeated treatment [42].

Similar results were obtained during a global randomized, double-blind, placebo-controlled phase III clinical trial concerning Tocilizumab administration to hospitalized patients with severe COVID-19 pneumonia (COVACTA- NCT04320615): the results did not confirm beneficial effect of treatment (https:// www. gene. com/media/press-releases/14867/2020-07-28/genentech-provides-an-update-on-the-phas). On contrary, a non-controlled, prospective clinical trial support that Tocilizumab may be a promising for patients with severe or critical SARS-CoV-2 infection. However, the analysis of its results is confounding. During this study 42 patients in severe or critical stage received a single dose of 400 mg Tocilizumab *via* intravenous infusion. After Tocilizumab treatment, only 6 patients required mechanical ventilation and 35 patients showed

clinical improvement. However, by day 28, 7 patients died. Neurological adverse effects were also observed in 3 patients [44].

Other IL-6 inhibitors, Sarilumab [141] and Siltuximab [173], were also investigated as potential treatment of COVID-19 patients. In a recent review, 352 articles concerning IL-6 antagonists and COVID-19 were reported, but only 11 study were further analyzed [7]. The conclusion is that use of Tocilizumab may be beneficial and currently the use of Tocilizumab and other IL-6 inhibitors is intensively investigated in several clinical trials [8].

Convalescent plasma therapy

Convalescent plasma (CP) therapy is a passive immunotherapy, already applied to the prevention and treatment of several infectious diseases. CP therapy was successfully used in the treatment of different viral conditions and more recently during Ebola, SARS-CoV, MERS-CoV, and Influenza virus H1N1 pandemics with satisfactory efficacy and safety [29, 119]. Administration of convalescent plasma may be also of clinical benefit for treatment of severe acute respiratory infections of SARS-CoV-2 etiology [29, 37]. Such possibility is reinforced by several experimental studies. A case report describes CP treatment of a centenarian with laboratory confirmed SARS-CoV-2, not suitable for antiviral treatment. Patient received two CP doses (200 and 100 ml). Significant improvement of laboratory indicators and clinical symptoms was observed. SARS-CoV-2 viral load decreased after the first transfusion (from 2.55×10^4 to 1.39×10^3 copies RNA/ml) and became undetectable (13 days of hospitalization) after the second one [94]. Another study reported treatment of 10 patients, with confirmed by RT-PCR infection, treated with CP transfusion (ChiCTR2000030046) [46]. Administration of one dose of 200 ml CP was well tolerated, the clinical symptoms significantly improved with the increase of oxyhemoglobin saturation within 3 days and CRP decrease. Radiological examination showed varying degrees of absorption of lung lesions within 7 days. The neutralization of viremia was also accelerated: the viral load was undetectable after transfusion in 7 patients with previous viremia. No severe adverse effects were observed [46]. A preliminary communication about 5 patients in critical stage of COVID-19 (with acute respiratory distress syndrome) reinforces the possibility of improvement of clinical status by CP treatment. One of effects of CP administration was the decrease of viral loads: virus became undetectable within 12 days after the transfusion. 3 patients has been discharged from hospital and 2 were stabilized. No adverse events were reported [154]. The results of a randomized trial (NCT04342182) concerning CP treatment of COVID19 patients are announced as preprint [60]. The study was discontinued due to the concerns about very high antibodies titers in COVID19 patients (comparable to the levels observed in donors). No difference in mortality, hospitalization time or disease severity at day-15 was observed between CP treated patients and patients on standard care [60]. Many other trials are ongoing and results are expected.

Recently, recommendations on biological characteristics of a CP preparation were published [135]. The CP must be collected from donor who had recovered from COVID-19 for more than two weeks. CP should be tested for HIV, HCV, HBV and syphilis (nucleic acids and serology tests). A negative result of RT-PCR testing for SARS-CoV-2 is also clearly expected [135]. CP should also contain sufficient amount of SARS-CoV-2 -specific antibodies [94]. Recommendations of FDA for investigational CP treatment are also available at FDA site (https://www.fda.gov/vaccines-blood-biologics/investigational-new-drug-ind-or-device-exemption-ide-process-cber/recommendations-investigational-covid-19-convalescent-plasma).

3. Virus-directed drugs

3.1. Broad-range-antiviral drugs

Viral protease inhibitors

Lopinavir/ritonavir (Kaletra, Aluvia)

Lopinavir ($C_{37}H_{48}N_4O_5$)/Ritonavir ($C_{37}H_{48}N_6O_5S_2$) treatment is used since 2000 in HIV infections in adults and children. Lopinavir is the HIV protease inhibitor, blocking HIV polyproteins maturation [33]. Molecular modeling evaluated that Lopinavir may also bind to the $3CL^{pro}$ protease encoded by SARS-CoV-2 and by thus inhibit the processing of viral polyproteins [107, 130]. Recent study suggested that Lopinavir was active against SARS-CoV-2 *in vitro* [41], but there is currently no much evidence for the efficacy of Lopinavir/ritonavir in the treatment of COVID-19 patients. Jomah *et al.*, listed 19 completed clinical trials or case series concerning Lopinavir/Ritonavir in COVID-19 treatment [84]. Earlier administration of Lopinavir/Ritonavir treatment could shorten viral shedding duration in hospitalized non-critically ill patients [122], but several clinical trials do not confirm the beneficial effect of this type of treatment on COVID-19 progression. One study (ChiCTR2000029308) compared the outcome of a group of Lopinavir/Ritonavir 99 adult patients to control group with standard-care (100 patients), both groups with laboratory-confirmed SARS-CoV-2 infection. No benefits, associated with Lopinavir/Ritonavir treatment, were observed concerning the mortality or the time of illness [26]. In a second clinical trial (NCT04252885), it was also demonstrated that mono-

therapy with Lopinavir/Ritonavir provided no benefit for improving the clinical outcome of patients hospitalized with mild/moderate COVID-19 over supportive care [105]. Another clinical trial (ChiCTR2000029387) had negatively evaluated Lopinavir/Ritonavir with association with IFNα as compared to Ribavirin treatment [78]. A case study of a patient, treated with Lopinavir/Ritonavir for chronic HIV demonstrate that the treatment failed not only to prevent SARS-CoV-2 infection, but also failed to prevent rapid progression to severe pneumonia [85]. Also a retrospective study did not confirm the efficacy of Lopinavir/Ritonavir in COVID19 treatment compared to standard care [100]. Also the comparison of effectiveness of Lopinavir/Ritonavir to Arbidol demonstrate the superiority of Arbidol in COVID-19 treatment [203].

The concern is also about Lopinavir/Ritonavir safety. ACE2 receptor is also expressed by kidney and post-mortem biopsies confirmed viral inclusions in tubular epithelial cells and podocytes. Lopinavir/Ritonavir treatment may enhance acute kidney injury in SARS-CoV-2 infected patients [16]. On July 6, 2020, WHO decided to discontinue the Lopinavir/Ritonavir arms of SOLIDARITY international trial.

Other clinical trials are completed, but results are missing and several ongoing trials of Lopinavir/Ritonavir, essentially in combination with other treatments, are currently recruiting. The lack of efficacy of Lopinavir may be due to the fact that $3CL^{pro}$ is a cysteine protease while HIV protease is an aspartic protease and moreover $3CL^{pro}$, does not contain a C2-symmetric pocket in the catalytic site- the target of these HIV protease inhibitors [9, 92].

Nucleotide analogues: inhibitors of RNA depended RNA polymerases (RdRp)

Remdesivir

The nucleoside analogue, Remdesivir (GS-5734) ($C_{27}H_{35}N_6O_8P$) is a well-known broad-spectrum antiviral drug, that inhibits viral RdRp, causing termination of RNA synthesis [62]. This drug was developed in response to the Ebola outbreak in West Africa from 2014 [81]. Remdesivir had as well showed its antiviral efficacy against SARS-CoV-2 *in vitro* [41, 178] by inhibiting novel human coronaviruses RdRp [62, 197]. Its efficacy was recently evaluated against MERS-CoV *in vitro* and *in vivo* on murine model [153]. Remdesivir was also applied intravenously to 53 patients with severe COVID-19 as compassionate use and clinical improvement was observed in 36 of treated patients (68%) [64]. However this study was controversial since several scientists expressed concern about the interpretation of the data [64]. Moreover, 60% of treated patients reported adverse events, the serious one include renal or organ failure [64]. Remdesivir was tested as COVID-19 treatment in a rhesus macaque model of SARS-CoV-2 infection [186]. Virus titers in bronchoalveolar lavages of Remdesivir-treated animals were significantly reduced and the lung tissue presented less damage than in control, infected, animals. Macaques treated with Remdesivir did not show signs of respiratory disease and had reduced pulmonary infiltrates [186]. A case report described the clinical improvement by treatment of severe COVID-19 patient with Remdesivir in association of anakinra (IL-1 receptor antagonist) [53]. First randomized, double-blind, placebo-controlled phase 3 clinical trial (NCT04257656) evaluating Remdesivir efficacy in COVID-19 treatment has just terminated [184]. 200 mg Remdesivir was administrated intravenously on day 1, followed by 100 mg once-daily maintenance doses for 9 days. Only a non-statistical improvement of time to clinical recovery was observed in Remdesivir-treated group, with symptom duration of 10 days or less [184]. However the same protocol, administrated to four critical COVID-19 patients resulted in negative nasal swab for SARS-CoV-2 RNA in 3 patients after 3 days of therapy [47]. Another study on 35 patients with SARS-CoV-2 associated pneumonia suggest that administration of Remdesivir to critical patients was associated with frequent adverse events (hepatotoxicity, acute kidney injury), but may benefit to patients with non-severe form of illness [6]. Compassionate use of Remdesivir to children and adolescents with severe infection, admitted he pediatric intensive care unit was recently described [34]. During a randomized, double-blind, placebo-controlled, multicentre trial (NCT04257656) at ten hospitals in Hubei, China no association between Remdesivir and statistically significant clinical benefits was observed [183]. Similar conclusions on lack of significant differences between a 5-day or 10-day course of Remdesivir administrated to patients not requiring mechanical ventilation were recently published. However, these study (NCT04292899) had no placebo control and the overall benefit cannot be determined [61]. On contrary a double-blind, randomized, placebo-controlled trial (NCT04280705) of intravenous Remdesivir in adults hospitalized with Covid-19 with evidence of lower respiratory tract involvement suggest a significantly shorter time to recovery in treated group (11 days for Remdesivir and 15 for placebo) [15].

FDA has approved a EUA of Remdesivir to treat COVID-19 patients. Remdesivir should be administered intravenously. However, as pointed by FDA "Remdesivir is not yet licensed or approved anywhere globally and has not been demonstrated to be safe or effective for the treatment of COVID-19" (May 06, 2020). The use Remdesivir in patients with nephrologic or with hepatic impairment is not recommended. On 7th May 2020, Remdesivir (Veklury®) was approved for

COVID-19 treatment in Japan. Remdesivir is the first medicine authorized at European Union level for treatment of COVID-19 since July 3, 2020. Several clinical trials evaluating its efficacy and safety were ongoing through USA, Europe and China and the results are expected within weeks.

Ribavirin

Ribavirin ($C_8H_{12}N_4O_5$) is a nucleoside analogue with broad antiviral activity, approved in antiviral treatment. In Canada, for example, Ribavirin is licensed for the treatment of RSV-associated bronchiolitis and pneumonia in infants for treatment of hepatitis C infections [95, 96]. Known mechanisms of Ribavirin efficacy against RNA and DNA viruses involve polymerase inhibition, interference with RNA capping, lethal mutagenesis and inhibition of GTP synthesis [131]. Another study pointed out the boost of immune system, especially Th1 cells by Ribavirin. So, the antiviral effect may be not only due to direct inhibition of virus replication but also involves immune system activation [93, 163].

Ribavirin was extensively studied as anti- SARS-CoV and MERS-CoV drug. Although the drug activity against these CoVs was demonstrated *in vitro* [49], no inhibitory effect of monotherapy with Ribavirin was found in SARS-CoV animal models [126]. Studies on a mouse model even showed that Ribavirin may prolong or enhance viral replication [10]. During MERS-CoV infection of Vero cells, Ribavirin was inhibitory only at very high concentrations [49]. Ribavirin was used to treat SARS-CoV patients (with or without concomitant use of steroids) in Hong Kong [95] and other countries, as well as treatment of MERS-CoV infections [102, 132, 151]. The conclusion was that dose required to treat patients may be difficult to estimate and to reach [67]. Moreover, Ribavirin-associated toxicity was noticed (reviewed in [126]). More effective was the combinational therapy with Ribavirin and IFN-β. Such combination was proposed as SARS-CoV and MERS-CoV treatment on the basis of positive results *in vitro* [49], on nonhuman primate models [32, 50] and *in vivo* studies [132, 151].

Ribavirin tightly binds to SARS-CoV-2 RNA-dependent RNA polymerase (RdRp) *in silico* [48] and thus may be useful in COVID-19 treatment. A case report of combination of IFN-α, Ribavirin and Lopinavir/Ritonavir on COVID-19 patient demonstrated its efficacy as evaluated by patient recovery [200]. Currently several clinical evaluations are ongoing in China [88]. One phase II clinical trial (NCT04276688) has been terminated at the University of Hong Kong. This study evaluated the benefit of triple combination of Lopinavir/Ritonavir, Ribavirin and IFN-β in treatment of COVID-19. Authors observed a significantly shorter median time from study start to virus clearance (nasopharyngeal swab) in combination group (7 days) comparing to control group (12 days). The administration of combination drugs did not increase adverse events occurrence (mainly nausea and diarrhea) [79]. Another study compared the use of Ribavirin, associated or not with IFN-α or Lopinavir/Ritonavir or both. No significant difference in outcome of mild to moderate COVID-19 patients was observed. Combination of Ribavirin with Lopinavir/Ritonavir induced a significant increase in gastrointestinal adverse events, suggesting these drugs should not be administered simultaneously [78]. Also a retrospective cohort study did not found any improvement in virus clearance or decrease in mortality rate in Ribavirin-treated patients with severe COVID-19 [167].

Favipiravir (Avigan)

Favipiravir ($C_5H_4FN_3O_2$) acts as a purine nucleoside analogue and is a competitive substrate inhibitor of the viral RdRp [55]. Unlike Ribavirin, this compound does not influence host cell nucleic acids synthesis. Favipiravir has been approved for the treatment of Influenza virus infections in Japan and is also licensed in China, but not in Europe or USA [55]. It has a broad spectrum of activity towards RNA viruses like Influenza virus, Rhinovirus or RSV [89]. Its efficacy against SARS-CoV-2 has been tested *in vitro* and *in vivo*. Favipiravir inhibited SARS-CoV-2 infection of Vero E6 cells, but much weaker than other tested antiviral drugs [178]. An experimental treatment, comparing the efficacy of Favipiravir to Lopinavir/Ritonavir administration, was conducted in China. The viral clearance was faster in the Favipiravir group (4 days) infected with SARS-CoV-2, than in Lopinavir/Ritonavir group (11 days). Moreover the clinical outcome of patients treated with Favipiravir was improved concerning the disease progression. However the study was performed on small number of patients with mild or moderate COVID-19 [23]. Another study compared the treatment of 116 patients with Favipiravir to 120 patients with Arbidol [35]. Favipiravir significantly improved the decrease of fever and cough. Adverse effects are mild and manageable [35]. Also a retrospective observational study conducted in Thailand reported the promising efficacy of Favipiravir in SARS-CoV-2 patients. Administration of Favipiravir (≥45 mg/kg/day) was identified as a good prognostic factor for clinical improvement (preprint [144]).

The evidence of the clinical safety of short term use of Favipiravir seems to be well documented [137]. Generally, the drug was well tolerated, but liver toxicity, hyperuricemia and diarrhea were reported in some patients [23, 35]. Recently, a Phase I clinical trial did not revealed adverse effects of this drug in healthy persons (NCT04400682).

One clinical trials did not confirm beneficial effect of Favipiravir administration to COVID19 patients (ChiCTR 2000029544) (preprint [110]).

In March 2020, Favipiravir was approved by the National Medical Products Administration of China as the first anti-COVID-19 drug [169]. In April 2020 FDA approves clinical trials using Favipiravir in treatment of COVID-19 patients in USA. On August 12, 2020 5 trials were completed as mentioned at USA Clinical trials registry.

3.2. Inhibitors of viral S glycoprotein

rhACE2

The predominant physiological function of SARS-CoV-2 cellular receptor-ACE2 is the maintenance of cardiovascular homeostasis: ACE2 increase blood pressure by cleaving the angiotensin I to angiotensin II (potent vasoconstrictor) and by inactivation of bradykinin (vasodilator). A possibility of blockage of virus-ACE2 interaction is the use of truncated, soluble recombinant human ACE2 (rhACE2) to block S protein, without affecting ACE2 itself [166]. Intravenous administration of active soluble ACE2 protein significantly improved the outcome of respiratory failure, induced by lipopolysaccharide, by its ability to increase the oxygenation in pigs [168]. Administration of a rhACE2 protein alleviated the severity of RSV or H5N1 virus-induced lung injury in ACE2 deficient mice [65, 205].

Soluble rhACE2 has been shown to block binding of the SARS-CoV spike S1 protein to its receptor *in vitro* [104]. Ongoing SARS-CoV-2 pandemic increases the interest for rhACE2 as COVID-19 treatment. The hypothesis is that soluble receptor rhACE2 may act as a trap for SARS-CoV-2 by intercepting viral particles, preventing virus binding to cell membrane-associated ACE2 and by inhibiting the virus entry to host cells [11, 103]. SARS-CoV-2 infection of Vero cells or engineered human organoids (blood vessel and kidney) was significantly limited by administration of soluble clinical grade rhACE2 [128]. A fusion protein, generated by connecting the extracellular domain of human ACE2 to the Fc region of the human immunoglobulin IgG1 has high binding affinity to the S proteins of SARS-CoV and SARS-CoV-2 and neutralized studied CoVs *in vitro* [103]. The safety of administration of rhACE2 was previously demonstrated during clinical studies on healthy volunteers (NCT00886353) or patients with acute lung injury (NCT01597635) and on patients with pulmonary arterial hypertension (NCT01884051). This form of rhACE2 was well-tolerated and an improvement was observed concerning pulmonary haemodynamics [70, 72, 91]. Nonetheless, the evidence for beneficial effect of rhACE2 on COVID-19 patients is lacking. For non-declared reasons, the first planned pilot study has been withdrawn (NCT number: NCT04287686) and the other planned trail is recruiting patients.

Other soluble form of ACE2 receptor, promising as COVID-19 treatment, is the recombinant bacterial ACE2-like soluble enzyme (rbACE2). B38-CAP obtained from *Paenibacillus* sp. B38 has a beneficial activity in murine model as measured by hypertension modulation, cardiac hypertrophy, and fibrosis [124]. Currently, two clinical trials investigating the effect of rbACE2 on COVID-19 patients are planned in US clinical trials registry (August 13, 2020). This form of treatment are not investigated in China.

3.3. New potential virus-directed drugs against SARS-CoV-2

Pan-CoV fusion inhibitors: EK1, EK1C4

As mentioned above, SARS-CoV-2 mainly enter to host cells by endocytosis pathway and fusion occurs between viral envelope and endosomal membrane [133]. Both mechanisms involved S protein and the recognition of specific receptor present on cellular membrane: ACE2. During viral maturation, S protein is post-translationally cleaved into a S1 receptor binding unit and a S2 membrane fusion unit. The S2 subunit contain the fusion peptide and two 4,3 hydrophobic (heptad) repeat regions designated HR1 and HR2. Receptor binding induces conformational changes in the S2 subunit. HR1 and HR2 regions interact with each other to form a six-helix bundle (6-HB) fusion core, which in turn induces fusion by insertion of fusion peptide into the host target cell membrane, bringing viral and cellular membranes into close proximity. It was demonstrated that HR2-dervided peptide prevented cell entry (fusion entry model) of SARS-CoV, probably by interfering with the 6-HB formation. However, the endosomal pathway entry was not inhibited [170].

In studies, concerning SARS-CoV and MERS-CoV, a pan-coronavirus fusion inhibitor, named EK1, was obtained [190]. This soluble peptide, derived from the HR2 domain of HCoV-OC43, exhibited broad fusion inhibitory activity against multiple human CoVs, including SARS-CoV and MERS-CoV, *in vitro*, as well as in a murine model of CoV infection. It was demonstrated that EK1 efficiently inhibited S protein-mediated cell fusion. Intranasal administration of EK1, pre- or post-challenge with CoVs, protected the treated mice from HCoV-OC43 or MERS-CoV [190]. It was also demonstrated that infection by SARS-CoV-2 of a T lymphocyte cell line (MT-2) was inhibited by EK1 peptide [180]. EK1 mechanism seems to involve the targeting of the HR1 domain of S protein: EK1 forms a stable 6-HB structure with HR1 and prevents the HR1-HR2 interaction and formation of 6-HB fusion core, which

is an indispensable step during host-viral membrane fusion [190]. Safety tests did not reveal any pathological abnormality in mice treated with EK1 [190].

Recently, a lipopeptide derived from EK1 (EK1C4) was developed [189]. The addition of cholesterol to C-terminus of EK1 peptide improves its inhibitory activity on pseudo CoVs infection. Moreover EK1C4 blocked *in vitro* the infection by different human CoVs, including SARS-CoV-2 [189]. Intranasal application of EK1C4 to mice, short before or short after challenge with HCoV-OC43, protected animals from infection. Authors concluded that EK1C4 may have both, prophylactic and therapeutic potential against SARS-CoV-2 and could be used in an intranasal or inhalation administration [189]. Further studies are needed to evaluate this compound efficacy in COVID-19 treatment or prophylaxis.

Inhibitors of CoV mRNA capping

Capping of 5' termini of mRNA (addition of a 7-methyl-Gppp) is essential for efficient mRNA translation in eukaryotic cells and is also involved in pre-mRNA splicing, mRNA export from the nucleus and protection from mRNA degradation [142]. Capping is also supposed to facilitate evasion from the host's immune response. CoVs protect their RNA with a cap moiety. However, as they replicate in the cytoplasm, CoVs don't have access the host capping machinery, present in the nucleus, and encode their own capping and methylation apparatus. The mechanism of capping of CoVs RNA was recently described [38]. Like for CoVs that cause SARS or MERS, the mechanism of RNA capping may also be a the target in treatment of SARS-CoV-2 infection.

The nsp14 protein encoded by SARS-CoV was identified as a cap (guanine-N7)-methyltransferase (N7-MTase) and was designated as target for new antiviral drugs [38, 82]. The second viral methyltransferase involved in mRNA capping is ribose-2'-O-methyltransferase (2'-O-MTase) encoded by *nsp16* gene. Studies of SARS-CoV showed that the 2'-O-MTase activity of nsp16 needs to be activated by nsp10 [20]. Mutations in viral methyltransferases nsp14 or nsp16 render CoVs unable to efficiently replicate [12, 39]. Comparing SARS-CoV and SARS-CoV-2 encoded proteins, nsp14, nsp16 and nsp10 share 95.07%, 93.60% and 97.35% identities, respectively [191]. So, targeting RNAs capping may allow to develop an anti SARS-CoV-2 drug.

In vitro assays of N7-MTase or 2'-O-MTase activity showed the inhibitory effect of aurintricarboxylic acid on both SARS-CoV methyltransferases [20]. This compound has been also shown to inhibit SARS-CoV replication in Vero cell culture by decreasing viral production by 1000-fold [71]. The 2'-O-MTase activity may also be inhibited by nsp10-derived peptide *in vitro* and *in vivo* on a murine model and in consequence SARS-CoV replication was reduced as well as overall infectivity and pathogenesis [182]. These observations suggested the possibility for development of a broad-spectrum peptide inhibitors by targeting 2'-O-MTase encoded by CoVs,

A recent bioinformatic report describe the screening 123 antiviral drugs for select SARS-CoV-2 targeted inhibitors. Dolutegravir (HIV integrase inhibitor) and its derivative Bictegravir were selected as able of inhibition of 2'-O-MTase encoded by SARS-CoV-2, based on their estimated free energy of binding, the orientation of drug molecules in the active site and the interacting residues [91]. Dolutegravir was also proposed for COVID-19 treatment through a drug-target interaction deep learning model (artificial intelligence) but not as inhibitor of 2'-O-MTase but other RNA processing enzymes [14]. Currently, no studies evaluate Dolutegravir efficacy in treatment of COVID-19 patients (May 30, 2020).

CRISPR (Clustered Regularly Interspaced Short Palindromic Repeats)

CRISPR-Cas is the bacterial antiviral system that use sequence-specific crRNAs (CRISPR RNA) to inhibit bacteriophage replication by sequence-specific destruction of target viral DNA. Among different application of CRISPR, this system may allow the development of antiviral therapy targeting incurable chronic viral infections [2] such as HIV, HPV, HBV or herpesvirus or other infections as JCV (polyoma JC virus), ASF (african swine fever virus) or pseudorabies virus [101].

Such alternative antiviral approach, which relies on a CRISPR-Cas13 system was currently proposed for recognizing and specifically degrading the intracellular SARS-CoV-2 RNA genome and its mRNA [1]. The Cas13 ribonuclease could be an effective antiviral drug for single-stranded RNA viruses because it cleaves RNAs complementary to crRNA. Cas13 activity against LCV (lymphocytic choriomeningitis virus), Influenza A virus and VSV (vesicular stomatitis virus) was recently demonstrated in cell culture [54]. Currently, by testing the degradation of synthesized fragments of SARS-CoV-2 in lung epithelial cell cultures, a pool of crRNA able to direct Cas13 to viral RNA was designed. Half of these crRNAs targeted the conserved sequences of the *RdRp* and N genes of SARS-CoV-2 [1]. Such approach should be however validated *in vitro* and *in vivo*-on animal models, before clinical trials.

4. Conclusions

Our knowledge about novel CoVs and SARS-CoV-2 biology is constantly increasing, giving opportunities for the rational design of therapeutic drugs, targeting

CoVs replication, as well as for elaboration of potential therapies of COVID-19 patients. Currently tested drugs against SARS-CoV-2 are shown on Figure 1. A lot of data are provided during precedent outbreaks of SARS-CoV and MERS-CoV, but many issues remains unresolved. The lack of serious preclinical studies encourage the use of different drugs, that efficacy is ambiguous even during *in vitro* assays: several drugs, including Ritonavir, Litonavir, Lopinavir, Favipiravir were tested to protect Vero E6 cells from cytopathic effect induced by SARS-CoV-2. None of them was found to have an antiviral effect [109]

Unfortunately, the huge number of currently conducted case studies and clinical trials does not always turn in real know-how. In general a combination of several drugs are combined during trials. Many research was performed chaotically, trials have not always been rigorously designed and their main aim remains the assistance to hospitalized patients to save their lives. Some papers have been retracted due to methodological concern. Due to, the results are not always reliable and conclusive.

The use of Chloroquine and Hydroxychloroquine was halted in USA and Europe, due to safety issues and lack of efficacy against SARS-CoV-2. Also Lopinavir/Ritonavir and Ribavirin were removed from the COVID-19 institutional protocol. The National Institutes of Health publishes, constantly upgraded, guidelines for the medical management of COVID-19. Experts recommend against high-dose Chloroquine, against the combination of Hydroxychloroquine and Azithromycin (safety issues) and against Lopinavir/Ritonavir, or other HIV protease inhibitors (negative clinical trial data) (https://www.covid19treatmentguidelines.nih.gov/). Also the use of Baricitinib rises safety questions. On the other hand, Ruxolitinib treatment seems to lower the inflammation, cytokine storm and ARDS and its use is intensively investigated. The CP treatment seems to be very promising and is intensively investigated. On August 23, 2020, FDA issued an new EUA for CP for the treatment of hospitalized patients with COVID-19. Nonetheless, well-controlled randomized trials remain crucial for a demonstration of COVID-19 CP efficacy in COVID-19 treatment.

In China, the management of COVID-19 included antivirals (Hydroxychloroquine, Tocilizumab, IFN), high flow oxygen, mechanical ventilation, corticosteroids, intravenous immunoglobulin and CP administration [136]. Treatments based on unproven traditional medicines were also promoted in China and in Africa, but clinical evidences for their efficacy are lacking [43, 77].

Based on available data, only the administration of Remdesivir use is accepted through an EUA issued by FDA for certain hospitalized patients requiring supplemental oxygen. Remdesivir is also accepted in European Union and Japan for treatment of COVID-19. Another inhibitor of viral polymerase, Favipiravir, also displayed encouraging results as treatment.

Bibliography

1. Abbott T.R., Qi L.S. *et al.*: Development of CRISPR as an Antiviral Strategy to Combat SARS-CoV-2 and Influenza. *Cell*, **181**, 865–876.e12 (2020)
2. Abudayyeh O.O., Zhang F. *et al.*: RNA targeting with CRISPR--Cas13. *Nature*, **550**, 280–284 (2017)
3. Ahmed A., Wilcox R.A. *et al.*: Ruxolitinib in adult patients with secondary haemophagocytic lymphohistiocytosis: an open-label, single-centre, pilot trial. *Lancet Haematol,* **6**, e630–e637 (2019)
4. Albeituni S., Verbist K.C., Tedrick P.E., Tillman H., Picarsic J., Bassett R., Nichols K.E.: Mechanisms of action of ruxolitinib in murine models of hemophagocytic lymphohistiocytosis. *Blood*, **134**, 147–159 (2019)
5. Andreani J., Raoult D. *et al.*: In vitro testing of combined hydroxychloroquine and azithromycin on SARS-CoV-2 shows synergistic effect. *Microb. Pathog.* **145**, 104228 (2020)
6. Antinori S., Galli M. *et al.*: Compassionate remdesivir treatment of severe Covid-19 pneumonia in intensive care unit (ICU) and Non-ICU patients: Clinical outcome and differences in post_ treatment hospitalisation status. *Pharmacol. Res*, **158**, 104899–104899 (2020)
7. Antwi-Amoabeng D., Kanji Z., Ford B., Beutler B.D., Riddle M.S., Siddiqui F.: Clinical Outcomes in COVID-19 Patients Treated with Tocilizumab: An Individual Patient Data Systematic Review. *J. Med. Virol.* (2020)
8. Atal S., Fatima Z.: IL-6 Inhibitors in the Treatment of Serious COVID-19: A Promising Therapy? *Pharmaceut. Med.* 1–9 (2020)
9. Báez-Santos Y.M., St John S.E., Mesecar A.D.: The SARS-coronavirus papain-like protease: structure, function and inhibition by designed antiviral compounds. *Antiviral Res.* **115**, 21–38 (2015)
10. Barnard D.L., Sidwell R.W. *et al.*: Enhancement of the infectivity of SARS-CoV in BALB/c mice by IMP dehydrogenase inhibitors, including ribavirin. *Antiviral Res.* **71**, 53–63 (2006)
11. Batlle D., Wysocki J., Satchell K.: Soluble angiotensin-converting enzyme 2: a potential approach for coronavirus infection therapy? *Clini. Sci.* **134**, 543–545 (2020)
12. Becares M., Pascual-Iglesias A., Nogales A., Sola I., Enjuanes L., Zuñiga S.: Mutagenesis of coronavirus nsp14 reveals its potential role in modulation of the innate immune response. *J. Virol,* **90**, 5399–5414 (2016)
13. Bechman K., Subesinghe S., Norton S., Atzeni F., Galli M., Cope A.P., Winthrop K.L., Galloway J.B.: A systematic review and meta-analysis of infection risk with small molecule JAK inhibitors in rheumatoid arthritis. *Rheumatology*, **58**, 1755–1766 (2019)
14. Beck B.R., Shin B., Choi Y., Park S., Kang K.: Predicting commercially available antiviral drugs that may act on the novel coronavirus (SARS-CoV-2) through a drug-target interaction deep learning model. *Comput. Struct. Biotechnol. J.* **18**, 784–790 (2020)
15. Beigel J.H., Tomashek K.M., Dodd L.E.: Remdesivir for the Treatment of Covid-19 – Preliminary Report. *N. Engl. J. Med.* (2020)
16. Binois Y., Hachad H., Salem J.-E., Charpentier J., Lebrun--Vignes B., Pène F., Cariou A., Chiche J.-D., Mira J.-P., Nguyen L.S.: Acute kidney injury associated with lopinavir/ritonavir combined therapy in patients with Covid-19. *Kidney Int. Rep.* (2020)

17. Blaising J., Lévy P.L., Polyak S.J., Stanifer M., Boulant S., Pécheur E.-I.: Arbidol inhibits viral entry by interfering with| clathrin-dependent trafficking. *Antiviral Res.* **100**, 215–219 (2013)
18. Blaising J., Polyak S.J., Pécheur E.-I.: Arbidol as a broad-spectrum antiviral: An update. *Antiviral Res.* **107**, 84–94 (2014)
19. Boriskin Y.S., Leneva I.A., Pécheur E.I., Polyak S.: Arbidol: A Broad-Spectrum Antiviral Compound that Blocks Viral Fusion. *Curr. Med. Chem,* **15**, 997–1005 (2008)
20. Bouvet M., Debarnot C., Imbert I., Selisko B., Snijder E.J., Canard B., Decroly E.: In vitro reconstitution of SARS-coronavirus mRNA cap methylation. *PLoS Path,* **6**, e1000863-e1000863 (2010)
21. Broglie L., Pommert L., Rao S., Thakar M., Phelan R., Margolis D., Talano J.: Ruxolitinib for treatment of refractory hemophagocytic lymphohistiocytosis. *Blood advances*, **1**, 1533–1536 (2017)
22. Bryan M.C., Rajapaksa N.S.: Kinase Inhibitors for the Treatment of Immunological Disorders: Recent Advances. *J. Med. Chem.* **61**, 9030–9058 (2018)
23. Cai Q., Liu L. *et al.*: Experimental Treatment with Favipiravir for COVID-19: An Open-Label Control Study. *Engineering*, (2020)
24. Caly L., Druce J.D., Catton M.G., Jans D.A., Wagstaff K.M.: The FDA-approved Drug Ivermectin inhibits the replication of SARS-CoV-2 *in vitro*. *Antiviral Res.* **178**, 104787 (2020)
25. Cantini F., Niccoli L., Matarrese D., Nicastri E., Stobbione P., Goletti D.: Baricitinib therapy in COVID-19: A pilot study on safety and clinical impact. *J. Infec.* **81**, 318–356 (2020)
26. Cao B., Wang C. *et al.*: A Trial of Lopinavir-Ritonavir in Adults Hospitalized with Severe Covid-19. *N. Engl. J. Med.* **382**, 1787–179 (2020)
27. Cao Y., Zhou J. *et al.*: Ruxolitinib in treatment of severe coronavirus disease 2019 (COVID-19): A multicenter, single-blind, randomized controlled trial. *J. Allergy Clin. Immunol.* **146**, 137–146.e133 (2020)
28. Capochiani E., Bocchia M. *et al.*: Ruxolitinib Rapidly Reduces Acute Respiratory Distress Syndrome in COVID-19 Disease. Analysis of Data Collection From RESPIRE Protocol. *Front. Med,* **7**, 466 (2020)
29. Casadevall A., Pirofski L.-A.: The convalescent sera option for containing COVID-19. *J. Clin. Invest.* **130**, 1545–1548 (2020)
30. Chaccour C., Hammann F., Ramón-García S., Rabinovich N.R.: Ivermectin and COVID-19: Keeping Rigor in Times of Urgency. *Am. J. Trop. Med. Hyg.* **102**, 1156–1157 (2020)
31. Chaccour C., Hammann F., Ramón-García S., Rabinovich N.R.: Ivermectin and Novel Coronavirus Disease (COVID-19): Keeping Rigor in Times of Urgency. *Am. J. Trop. Med. Hyg.*(2020)
32. Chan J.F.-W., Yuen K.-Y. *et al.*: Treatment With Lopinavir/Ritonavir or Interferon-β1b Improves Outcome of MERS-CoV Infection in a Nonhuman Primate Model of Common Marmoset. *J. Inf. Dis.* **212**, 1904–1913 (2015)
33. Chandwani A., Shuter J.: Lopinavir/ritonavir in the treatment of HIV-1 infection: a review. *Ther. Clin. Risk Manag.* **4**, 1023–1033 (2008)
34. Chao J.Y., Derespina K.R., Herold B.C., Goldman D.L., Aldrich M., Weingarten J., Ushay H.M., Cabana M.D., Medar S.S.: Clinical Characteristics and Outcomes of Hospitalized and Critically Ill Children and Adolescents with Coronavirus Disease 2019 (COVID-19) at a Tertiary Care Medical Center in New York City. *J. Ped.* **223**, 14–19.e2 (2020)
35. Chen C., Wang X. *et al.*: Favipiravir versus Arbidol for COVID-19: A Randomized Clinical Trial. *medRxiv*, 2020.2003.2017.20037432 (2020)
36. Chen J., Lu H. *et al.*: A pilot study of hydroxychloroquine in treatment of patients with common coronavirus disease-19 (COVID-19). *J. Zhejiang University* (*Med. Sci.*), **49** (2020)
37. Chen L., Xiong J., Bao L., Shi Y.: Convalescent plasma as a potential therapy for COVID-19. *Lancet Infect. Dis.* **20**, 398–400 (2020)
38. Chen Y., Guo D.: Molecular mechanisms of coronavirus RNA capping and methylation. *Virologica Sinica*, **31**, 3–11 (2016)
39. Chen Y., Guo D. *et al.*: Biochemical and structural insights into the mechanisms of SARS coronavirus RNA ribose 2'-O-methylation by nsp16/nsp10 protein complex. *PLoS Path.* **7**, e1002294–e1002294 (2011)
40. Chowdhury M.D.S., Rathod J., Gernsheimer J.: A Rapid Systematic Review of Clinical Trials Utilizing Chloroquine and Hydroxychloroquine as a Treatment for COVID-19. *Acad. Emerg. Med.* **27**, 493–504 (2020)
41. Choy K.-T., Yen H.-L. *et al.*: Remdesivir, lopinavir, emetine, and homoharringtonine inhibit SARS-CoV-2 replication *in vitro*. *Antiviral Res.* **178**, 104786 (2020)
42. Colaneri M., Bogliolo L., Valsecchi P., Sacchi P., Zuccaro V., Brandolino F., Montecucco C., Mojoli F., Giusti E.M., Bruno R.: Tocilizumab for treatment of severe COVID-19 patients: preliminary results from SMAtteo COvid19 REgistry (SMACORE). *Microorganisms*, **8**, (2020)
43. Cyranoski D.: China is promoting coronavirus treatments based on unproven traditional medicines. *Nature*, (2020).
44. Dastan F., Tabarsi P. *et al.*: Promising Effects of Tocilizumab in COVID-19: A Non-Controlled, Prospective Clinical Trial. *Int. Immunopharmacol.* **88**, 106869 (2020)
45. Deng L., Li C., Zeng Q., Liu X., Li X., Zhang H., Hong Z., Xia J.: Arbidol combined with LPV/r versus LPV/r alone against Corona Virus Disease 2019: A retrospective cohort study. *J. Infect.* **81**, e1–e5 (2020)
46. Duan K., Yang X. *et al.*: Effectiveness of convalescent plasma therapy in severe COVID-19 patients. *Proc. Nat. Acad. Sci. USA*, **117**, 9490–9496 (2020)
47. Durante-Mangoni E., Andini R., Bertolino L., Mele F., Florio L.L., Murino P., Corcione A., Zampino R.: Early experience with remdesivir in SARS-CoV-2 pneumonia. *Infection*, 1–4 (2020)
48. Elfiky A.A.: Ribavirin, Remdesivir, Sofosbuvir, Galidesivir,)and Tenofovir against SARS-CoV-2 RNA dependent RNA polymerase (RdRp): A molecular docking study. *Life Sci.* **253**, 117592–117592 (2020)
49. Falzarano D., de Wit E., Martellaro C., Callison J., Munster V.J., Feldmann H.: Inhibition of novel β coronavirus replication by a combination of interferon-α2b and ribavirin. *Sci. Rep.* **3**, 1686–1686 (2013)
50. Falzarano D., Feldmann H. *et al.*: Treatment with interferon--alpha 2b and ribavirin improves outcome in MERS-CoV-infected rhesus macaques. *Nat. Med.* **10**, 1313–7 (2013)
51. Fantini J., Di Scala C., Chahinian H., Yahi N.: Structural and molecular modelling studies reveal a new mechanism of action of chloroquine and hydroxychloroquine against SARS-CoV-2 infection. *Int. J. Antimicrob. Agents*, **55**, 105960 (2020)
52. Favalli E.G., Biggioggero M., Maioli G., Caporali R.: Baricitinib for COVID-19: a suitable treatment? *Lancet Infect. Dis.* **9**, 1012–1013 (2020)
53. Franzetti M., Piconi S. *et al.*: Interleukin-1 receptor antagonist anakinra in association with remdesivir in severe Coronavirus disease 2019: A case report. *Int. J. Infect.Dis.* **97**, 215–218 (2020)
54. Freije C.A., Sabeti P.C. *et al.*: Programmable Inhibition and Detection of RNA Viruses Using Cas13. *Mol. Cell*, **76**, 826–837. e811 (2019)
55. Furuta Y., Komeno T., Nakamura T.: Favipiravir (T-705), a broad spectrum inhibitor of viral RNA polymerase. *Proc. Jpn. Acad. Ser. B. Phys. Biol. Sci.* **93**, 449–463 (2017)

56. Gao J., Tian Z., Yang X.: Breakthrough: Chloroquine phosphate has shown apparent efficacy in treatment of COVID-19 associated pneumonia in clinical studies. *BioSci. Trends*, **14**, 72–73 (2020)
57. Gaspari V., Zengarini C., Greco S., Vangeli V., Mastroianni A.: Side effects of ruxolitinib in patients with SARS-CoV-2 infection: Two case reports. *Int. J. Antimicrob. Agents*, **56**, 106023–106023 (2020)
58. Gautret P., Raoult D. *et al.*: Hydroxychloroquine and azithromycin as a treatment of COVID-19: results of an open-label non-randomized clinical trial. *Int. J. Antimicrob. Agents*, **56**, 105949 (2020)
59. George J., Mattapallil J.J.: Interferon-α Subtypes As an Adjunct Therapeutic Approach for Human Immunodeficiency Virus Functional Cure. *Front. Immun.* **9**, 299–299 (2018)
60. Gharbharan A., Rijnders B. *et al.*: Convalescent Plasma for COVID-19. A randomized clinical trial. *medRxiv*, 2020.2007.2001.20139857 (2020)
61. Goldman J.D., Subramanian A. *et al.*: Remdesivir for 5 or 10 Days in Patients with Severe Covid-19. *N. Engl. J. Med.* (2020)
62. Gordon C.J., Tchesnokov E.P., Woolner E., Perry J.K., Feng J.Y., Porter D.P., Gotte M.: 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**, 6785–6797. (2020)
63. Gorial F.I., Mashhadani S., Sayaly H.M., Dakhil B.D., AlMashhadani M.M., Aljabory A.M., Abbas H.M., Ghanim M., Rasheed J.I.: Effectiveness of Ivermectin as add-on Therapy in COVID-19 Management (Pilot Trial). *medRxiv*, 2020.2007.2007.20145979 (2020)
64. Grein J., Flanigan T. *et al.*: Compassionate Use of Remdesivir for Patients with Severe Covid-19. *N. Engl. J. Med.* **382**, 2327–2336 (2020)
65. Gu H., Yang P. *et al.*: Angiotensin-converting enzyme 2 inhibits lung injury induced by respiratory syncytial virus. *Sci. Rep.* **6**, 19840 (2016)
66. Haagmans B.L., Osterhaus A.D.M.E. *et al.*: Pegylated interferon-alpha protects type 1 pneumocytes against SARS coronavirus infection in macaques. *Nat. Med.* **10**, 290–293 (2004)
67. Habib A.M.G., Ali M.A.E., Zouaoui B.R., Taha M.A.H., Mohammed B.S., Saquib N.: Clinical outcomes among hospital patients with Middle East respiratory syndrome coronavirus (MERS-CoV) infection. *BMC Infect. Dis.* **19**, 870–870 (2019)
68. Harigai M., Genovese M.C. *et al.*: FRI0077 Hepatitis b virus reactivation in patients with rheumatoid arthritis treated with baricitinib: post-hoc analysis from clinical trials. *Ann. Rheumatic Dis.* **77** (2018)
69. Hartwell D., Jones J., Baxter L., Shepherd J.: Peginterferon alfa and ribavirin for chronic hepatitis C in patients eligible for shortened treatment, re-treatment or in HCV/HIV co-infection: a systematic review and economic evaluation. *Health Technol. Assess.* **17**, i-xii, 1–210 (2011)
70. Haschke M., Schuster M., Poglitsch M., Loibner H., Salzberg M., Bruggisser M., Penninger J., Krähenbühl S.: Pharmacokinetics and pharmacodynamics of recombinant human angiotensin-converting enzyme 2 in healthy human subjects. *Clin. Pharmacokinet.* **52**, 783–792 (2013)
71. He R., Adonov A., Traykova-Adonova M., Cao J., Cutts T., Grudesky E., Deschambaul Y., Berry J., Drebot M., Li X.: Potent and selective inhibition of SARS coronavirus replication by aurintricarboxylic acid. *Biochem. Biophys. Res. Commun.* **320**, 1199–1203 (2004)
72. Hemnes A.R., West J. *et al.*: A potential therapeutic role for angiotensin-converting enzyme 2 in human pulmonary arterial hypertension. *Europ. Respir. J.* **51**, 1702638 (2018)
73. Hensley L.E., Fritz L.E., Jahrling P.B., Karp C.L., Huggins J.W., Geisbert T.W.: Interferon-beta 1a and SARS coronavirus replication. *Emerg. Infect. Dis.* **10**, 317–319 (2004)
74. Hoffmann M., Pöhlmann S. *et al.*: SARS-CoV-2 Cell Entry Depends on ACE2 and TMPRSS2 and Is Blocked by a Clinically Proven Protease Inhibitor. *Cell*, **181**, 271–280.e278 (2020)
75. Hoffmann M., Schroeder S., Kleine-Weber H., Müller M.A., Drosten C., Pöhlmann S.: Nafamostat Mesylate Blocks Activation of SARS-CoV-2: New Treatment Option for COVID-19. *Antimicrob. Agents Chemother.* **64**, e00754–00720 (2020)
76. Horby P., Landry M. *et al.*: Effect of hydroxychloroquine in hospitalized patients with COVID-19: Preliminary results from a multi-centre, randomized, controlled trial. *medRxiv*, 2020.2007.2015.20151852 (2020)
77. Huang F., Luo L. *et al.*: A review of therapeutic agents and Chinese herbal medicines against SARS-COV-2 (COVID-19). *Pharmacol. Res.* 104929 (2020)
78. Huang Y.-Q., Chen Y.-K. *et al.*: No Statistically Apparent Difference in Antiviral Effectiveness Observed Among Ribavirin Plus Interferon-Alpha, Lopinavir/Ritonavir Plus Interferon-Alpha, and Ribavirin Plus Lopinavir/Ritonavir Plus Interferon-Alpha in Patients With Mild to Moderate Coronavirus Disease 2019: Results of a Randomized, Open-Labeled Prospective Study. *Front. Pharmacol.* **11**, 1071–1071 (2020)
79. Hung I.F.-N., Yuen K.-Y. *et al.*: Triple combination of interferon beta-1b, lopinavir-ritonavir, and ribavirin in the treatment of patients admitted to hospital with COVID-19: an open-label, randomised, phase 2 trial. *Lancet*, **395**,1695–1704 (2020)
80. Iwata-Yoshikawa N., Okamura T., Shimizu Y., Hasegawa H., Takeda M., Nagata N.: TMPRSS2 Contributes to Virus Spread and Immunopathology in the Airways of Murine Models after Coronavirus Infection. *J. Vir.* **93**, e01815–01818 (2019)
81. Jacobs M., Thomson E.C. *et al:* Late Ebola virus relapse causing meningoencephalitis: a case report. *Lancet*, **388**, 498–503 (2016)
82. Jin X., Guo D. *et al.*: Characterization of the guanine-N7 methyltransferase activity of coronavirus nsp14 on nucleotide GTP. *Virus Res.* **176**, 45–52 (2013)
83. Jin Y.-H., Wang X.-H. *et al.*: A rapid advice guideline for the diagnosis and treatment of 2019 novel coronavirus (2019-nCoV) infected pneumonia (standard version). *Mil. Med. Res.* **7**, 4 (2020)
84. Jomah S., Asdaq S.M.B., Al-Yamani M.J.: Clinical efficacy of antivirals against novel coronavirus (COVID-19): A review. *J. Infect. Public Health*, **9**, 1187–1195 (2020)
85. Joumaa H., Regard L., Carlier N., Chassagnon G., Alabadan E., Canouï E., L'Honneur A., Rozenberg F., Burgel P.R., Roche N.: A severe COVID-19 despite ongoing treatment with Lopinavir-Ritonavir. *Respi. Med. Res.* **78**, 100780 (2020)
86. Karres I., Kremer J.P., Dietl I., Steckholzer U., Jochum M., Ertel W.: Chloroquine inhibits proinflammatory cytokine release into human whole blood. *Am. J. Physiol.* **274**, R1058–1064 (1998)
87. Kawase M., Shirato K., van der Hoek L., Taguchi F., Matsuyama S.: Simultaneous treatment of human bronchial epithelial cells with serine and cysteine protease inhibitors prevents severe acute respiratory syndrome coronavirus entry. *J. Vir.* **86**, 6537–6545 (2012)
88. Khalili J., Zhu H., Mak A., Yan Y., Zhu Y.: Novel coronavirus treatment with ribavirin: Groundwork for evaluation concerning COVID-19. *J. Med. Vir.* (2020)
89. Khambholja K., Asudani D.: Potential repurposing of Favipiravir in COVID-19 outbreak based on current evidence. *Travel Med. Infecti. Dis.* **35**,101710 (2020)
90. Khamitov R.A., Loginova S., Shchukina V.N., Borisevich S.V., Maksimov V.A., Shuster A.M.: Antiviral activity of arbidol and

its derivatives against the pathogen of severe acute respiratory syndrome in the cell cultures. *Vopr. Virusol.* **53**, 9–13 (2008)
91. Khan R.J., Jha R.K., Amera G.M., Jain M., Singh E., Pathak A., Singh R.P., Muthukumaran J., Singh A.K.: Targeting SARS--CoV-2: a systematic drug repurposing approach to identify promising inhibitors against 3C-like proteinase and 2'-O-ribose methyltransferase. *J. Biomol. Struct. Dyn.* **0**, 1–14 (2020)
92. Ko G.M., Reddy A.S., Kumar S., Bailey B.A., Garg R.: Computational analysis of HIV-1 protease protein binding pockets. *J. Chem. Inform. Model.* **50**, 1759–1771 (2010)
93. Kobayashi T., Nakatsuka K., Shimizu M., Tamura H., Shinya E., Atsukawa M., Harimoto H., Takahashi H., Sakamoto C.: Ribavirin modulates the conversion of human CD4(+) CD25(−) T cell to CD4(+) CD25(+) FOXP3(+) T cell via suppressing interleukin-10-producing regulatory T cell. *Immunol.* **137**, 259–270 (2012)
94. Kong Y., Cai C., Ling L., Zeng L., Wu M., Wu Y., Zhang W., Liu Z.: Successful treatment of a centenarian with coronavirus disease 2019 (COVID-19) using convalescent plasma. *Transf. Apheres. Sci.* 102820 (2020)
95. Koren G., King S., Knowles S., Phillips E.: Ribavirin in the treatment of SARS: A new trick for an old drug? *CMAJ*, **168**, 1289–1292 (2003)
96. Krilov L.: Safety issues related to the administration of ribavirin. *Pediatr. Infec. Dis. J.* **21**, 479–481 (2002)
97. Kuznik A., Bencina M., Svajger U., Jeras M., Rozman B., Jerala R.: Mechanism of endosomal TLR inhibition by antimalarial drugs and imidazoquinolines. *J. Immunol.* **186**, 4794–4804 (2011)
98. La Rosée F., La Rosée P. *et al.*: The Janus kinase 1/2 inhibitor ruxolitinib in COVID-19 with severe systemic hyperinflammation. *Leukemia*, **34**, 1805–1815 (2020)
99. Laing R., Gillan V., Devaney E.: Ivermectin – Old Drug, New Tricks? *Trends Parasitol.* **33**, 463–472 (2017)
100. Lecronier M., Dres M. *et al.*: Comparison of hydroxychloroquine, lopinavir/ritonavir, and standard of care in critically ill patients with SARS-CoV-2 pneumonia: an opportunistic retrospective analysis. *Crit. Care*, **24**, 418–418 (2020)
101. Lee C.: CRISPR/Cas9-Based Antiviral Strategy: Current Status and the Potential Challenge. *Molecules*, **24**, 1349 (2019)
102. Lee J.Y., Kim Y.-J., Chung E.H., Kim D.-W., Jeong I., Kim Y., Yun M.-R., Kim S.S., Kim G., Joh J.-S.: The clinical and virological features of the first imported case causing MERS-CoV outbreak in South Korea, 2015. *BMC Infect. Dis.* **17**, 498–498 (2017)
103. Lei C., Fu W., Qian K., Li T., Zhang S., Fu W., Ding M., Hu S.: Neutralization of SARS-CoV-2 spike pseudotyped virus by recombinant ACE2-Ig. *Nat. Commun.* **11,** 2070 (2020)
104. Li W., Moore M.J., Vasilieva N., Sui J., Wong S.K., Berne M.A., Somasundaran M., Sullivan J.L., Luzuriaga K., Greenough T.C. *et al*: Angiotensin-converting enzyme 2 is a functional receptor for the SARS coronavirus. *Nature*, **426**, 450–454 (2003)
105. Li Y., Li L. *et al.*: An exploratory randomized controlled study on the efficacy and safety of lopinavir/ritonavir or arbidol treating adult patients hospitalized with mild/moderate COVID-19 (ELACOI). *medRxiv*, 2020.2003.2019.20038984 (2020)
106. Lian N., Xie H., Lin S., Huang J., Zhao J., Lin Q.: Umifenovir treatment is not associated with improved outcomes in patients with coronavirus disease 2019: a retrospective study. *Clin. Microbiol. Infect.* **26**, 917–921 (2020)
107. Lin S., Shen R., He J., Li X., Guo X.: Molecular modeling evaluation of the binding effect of ritonavir, lopinavir and darunavir to severe acute respiratory syndrome Coronavirus 2 proteases. *bioRxiv*, 2020.2001.2031.929695 (2020)
108. Liu Q., Peng P. *et al.*: The effect of Arbidol Hydrochloride on reducing mortality of Covid-19 patients: a retrospective study of real world date from three hospitals in Wuhan. *medRxiv*, 2020.2004.2011.20056523 (2020)
109. Liu S., Lien C.Z., Selvaraj P., Wang T.T.: Evaluation of 19 antiviral drugs against SARS-CoV-2 Infection. *bioRxiv*, 2020.2004.2029.067983 (2020)
110. Lou Y., Liu L., Qiu Y.: Clinical outcomes and plasma concentrations of baloxavir marboxil and favipiravir in COVID-19 patients: an exploratory randomized, controlled trial. *medRxiv*, 2020.2004.2029.20085761 (2020)
111. Loutfy M.R., Fish E. N. *et al.*: Interferon Alfacon-1 Plus Corticosteroids in Severe Acute Respiratory Syndrome A Preliminary Study. *JAMA*, **290**, 3222–3228 (2003)
112. Lu H., Stratton C.W., Tang Y.-W.: Outbreak of pneumonia of unknown etiology in Wuhan, China: The mystery and the miracle. *J. Med. Vir.* **92**, 401–402 (2020)
113. Lucas J.M., Nelson P.S. *et al.*: The androgen-regulated protease TMPRSS2 activates a proteolytic cascade involving components of the tumor microenvironment and promotes prostate cancer metastasis. *Cancer Discov.* **4**, 1310–1325 (2014)
114. Luo P., Liu Y., Qiu L., Liu X., Liu D., Li J.: Tocilizumab treatment in COVID-19: A single center experience. *J. Med. Virol.* **92**, 814–818 (2020)
115. Kowalik M.M., Trzonkowski P., Łasińska-Kowara M., Mital A., Smiatacz T., Jaguszewski M.: COVID-19 – Toward a comprehensive understanding of the disease. *Cardiol. J.* **27**, 99–114 (2020)
116. Madrid P.B., Tanga J. *et al.*: Evaluation of Ebola Virus Inhibitors for Drug Repurposing. *ACS Infect. Dis.* **1**, 317–326 (2015)
117. Maggio R., Corsini G.U.: Repurposing the mucolytic cough suppressant and TMPRSS2 protease inhibitor bromhexine for the prevention and management of SARS-CoV-2 infection. *Pharmacolog. Res.* 104837 (2020)
118. Mahévas M., Costedoat-Chalumeau N. *et al*: Clinical efficacy of hydroxychloroquine in patients with covid-19 pneumonia who require oxygen: observational comparative study using routine care data. *BMJ*, **369**, m1844 (2020)
119. Mair-Jenkins J., Beck C.R. *et al.*: The effectiveness of convalescent plasma and hyperimmune immunoglobulin for the treatment of severe acute respiratory infections of viral etiology: a systematic review and exploratory meta-analysis. *J. Infect. Dis.* **211**, 80–90 (2015)
120. Matsuyama S., Takeda M. *et al.*: Enhanced isolation of SARS--CoV-2 by TMPRSS2-expressing cells. *Proc. Nat. Acad. Sci. USA*, **117**, 7001 (2020)
121. McAuliffe J., Subbarao K. *et al.*: Replication of SARS coronavirus administered into the respiratory tract of African Green, rhesus and cynomolgus monkeys. *Virology*, **330**, 8–15 (2004)
122. Mehra M.R., Desai S.S., Ruschitzka F., Patel A.N.: Hydroxychloroquine or chloroquine with or without a macrolide for treatment of COVID-19: a multinational registry analysis. *Lancet*, **RETRACTED** (2020)
123. Mehta P., McAuley D.F., Brown M., Sanchez E., Tattersall R.S., Manson J.J.: COVID-19: consider cytokine storm syndromes and immunosuppression. *Lancet*, **395**, 1033–1034 (2020)
124. Minato T., Kuba K. *et al.*: B38-CAP is a bacteria-derived ACE2--like enzyme that suppresses hypertension and cardiac dysfunction. *Nat. Commun.* **11**, 1058–1058 (2020)
125. Miyamoto Y., Yamada K., Yoneda Y.: Importin α: a key molecule in nuclear transport and non-transport functions. *J. Biochem.* **160**, 69–75 (2016)
126. Mo Y., Fisher D.: A review of treatment modalities for Middle East Respiratory Syndrome. *J. Antimicrob. Chemother.* **71**, 3340–3350 (2016)

127. Molina J.M., Delaugerre C., Le Goff J., Mela-Lima B., Ponscarme D., Goldwirt L., de Castro N.: No evidence of rapid antiviral clearance or clinical benefit with the combination of hydroxychloroquine and azithromycin in patients with severe COVID-19 infection. *Méd. Mal. Infect.* **50**, 384 (2020)
128. Monteil V., Penninger J.M. *et al.*: Inhibition of SARS-CoV-2 Infections in Engineered Human Tissues Using Clinical-Grade Soluble Human ACE2. *Cell*, **181**, 905–913.e7 (2020)
129. Navarro G., Taroumian S., Barroso N., Duan L., Furst D.: Tocilizumab in rheumatoid arthritis: a meta-analysis of efficacy and selected clinical conundrums. *Semin. Arthritis Rheum.* **43**, 458–469 (2014)
130. Nutho B., Mahalapbutr P., Hengphasatporn K., Pattaranggoon N.C., Simanon N., Shigeta Y., Hannongbua S., Rungrotmongkol T.: Why are lopinavir and ritonavir effective against the newly emerged coronavirus 2019? atomistic insights into the inhibitory mechanisms. *Biochemistry*, (2020)
131. Nyström K., Waldenström J., Tang K.-W., Lagging M.: Ribavirin: pharmacology, multiple modes of action and possible future perspectives. *Fut. Virol.* **14**, 153–160 (2019)
132. Omrani A.S., Saad M.M., Baig K., Bahloul A., Abdul-Matin M., Alaidaroos A.Y., Almakhlafi G.A., Albarrak M.M., Memish Z.A., Albarrak A.M.: Ribavirin and interferon alfa-2a for severe Middle East respiratory syndrome coronavirus infection: a retrospective cohort study. *Lancet Infect. Dis.* **14**, 1090–1095 (2014)
133. Ou X., Qian Z. *et al.*: Characterization of spike glycoprotein of SARS-CoV-2 on virus entry and its immune cross-reactivity with SARS-CoV. *Nat. Commun.* **11**, 1620 (2020)
134. Padhi A., Seal A., Tripathi T.: How does Arbidol Inhibit the Novel Coronavirus SARS-CoV-2? Atomistic Insights from Molecular Dynamics Simulations; ChemRxiv, **PREPRINT** (2020)
135. Patrizia A., Pierluigi B., Vincenzo d.A., Giustina D.S., Luca M., Angelo O.: Position paper on the preparation of immune plasma to be used in the treatment of patients with COVID-19. *Transfus. Apher. Sci.* **59**, 102817 (2020)
136. Peng F., Tu L., Yang Y., Hu P., Wang R., Hu Q., Cao F., Jiang T., Sun J., Xu G. *et al*: Management and Treatment of COVID-19: The Chinese Experience. *Can. J. Cardiol.* **36**, 915–930 (2020)
137. Pilkington V., Pepperrell T., Hill A.: A review of the safety of favipiravir – a potential treatment in the COVID-19 pandemic? *J. Virus Erad.* **6**, 45–51 (2020)
138. Praveen D., Puvvada R.C., M V.A.: Janus kinase inhibitor baricitinib is not an ideal option for management of COVID-19. *Int. J. Antimicrob. Agents*, **55**, 105967–105967 (2020)
139. Qaseem A., Yost J., Etxeandia-Ikobaltzeta I., Miller M.C., Abraham G.M., Obley A.J., Forciea M.A., Jokela J.A., Humphrey L.L.: Should clinicians use chloroquine or hydroxychloroquine alone or in combination with azithromycin for the prophylaxis or treatment of COVID-19? Living practice points from the american college of physicians (Version 1). *Ann. Intern. Medi.* **173**, 137–142 (2020)
140. Radbel J., Narayanan N., Bhatt P.J.: Use of Tocilizumab for COVID-19-Induced Cytokine Release Syndrome: A Cautionary Case Report. *Chest*, **158**, e15–e19 (2020)
141. Raimondo M.G., Biggioggero M., Crotti C., Becciolini A., Favalli E.G.: Profile of sarilumab and its potential in the treatment of rheumatoid arthritis. *Drug Des. Devel. Ther.* **11**, 1593–1603 (2017)
142. Ramanathan A., Robb G.B., Chan S.-H.: mRNA capping: biological functions and applications. *Nucleic Acids Res.* **44**, 7511–7526 (2016)
143. Randall R., Goodbourn S.: Interferons and viruses: An interplay between induction, signalling, antiviral responses and virus countermeasures. *J. Gen. Virol.* **89**, 1–47 (2008)
144. Rattanaumpawan P., Jirajariyavej S., Lerdlamyong K., Palavutitotai N., Saiyarin J.: Real-world experience with favipiravir for treatment of COVID-19 in Thailand: Results from a multi-center observational study. *medRxiv*, 2020.2006.2024.20133249 (2020)
145. Retallack H., DeRisi J.L. *et al.*: Zika virus cell tropism in the developing human brain and inhibition by azithromycin. *Proc. Nat. Acad. Sci. USA*, **113**, 14408 (2016)
146. Richardson P., Griffin I., Tucker C., Smith D., Oechsle O., Phelan A., Stebbing J.: Baricitinib as potential treatment for 2019-nCoV acute respiratory disease. *Lancet*, **395**, e30–e31 (2020)
147. Rijckborst V., Janssen H.L.A.: The Role of Interferon in Hepatitis B Therapy. *Curr. Hepat. Rep.* **9**, 231–238 (2010)
148. Rolain J.-M., Colson P., Raoult D.: Recycling of chloroquine and its hydroxyl analogue to face bacterial, fungal and viral infections in the 21st century. *Int. J. Antimicrob. Agents*, **30**, 297–308 (2007)
149. Sallard E., Lescure F.-X., Yazdanpanah Y., Mentre F., Peiffer-Smadja N.: Type 1 interferons as a potential treatment against COVID-19. *Antiviral Res.* **178**, 104791–104791 (2020)
150. Savarino A., Boelaert J.R., Cassone A., Majori G., Cauda R.: Effects of chloroquine on viral infections: an old drug against today's diseases? *Lancet Infect. Dis.* **3**, 722–727 (2003)
151. Shalhoub S., Farahat F., Al-Jiffri A., Simhairi R., Shamma O., Siddiqi N., Mushtaq A.: IFN-α2a or IFN-β1a in combination with ribavirin to treat Middle East respiratory syndrome coronavirus pneumonia: a retrospective study. *J. Antimicrob. Chemother.* **70**, 2129–2132 (2015)
152. Sharma A.: Chloroquine Paradox May Cause More Damage Than Help Fight COVID-19. *Microbes Infect.* **22**, 154–156 (2020)
153. Sheahan T.P., Baric R.S. *et al.*: Comparative therapeutic efficacy of remdesivir and combination lopinavir, ritonavir, and interferon beta against MERS-CoV. *Nat. Commun.* **11**, 222–222 (2020)
154. Shen C., Liu L. *et al.*: Treatment of 5 Critically Ill Patients With COVID-19 With Convalescent Plasma. *JAMA*, **323**, 1582–1589 (2020)
155. Shen L.W., Mao H.J., Wu Y.L., Tanaka Y., Zhang W.: TMPRSS2: A potential target for treatment of influenza virus and coronavirus infections. *Biochimie*, **142**, 1–10 (2017)
156. Shi L., Xiong H., He J., Deng H., Li Q., Zhong Q., Hou W., Cheng L., Xiao H., Yang Z.: Antiviral activity of arbidol against influenza A virus, respiratory syncytial virus, rhinovirus, coxsackie virus and adenovirus in vitro and in vivo. *Arch. Virol.* **152**, 1447–1455 (2007)
157. Shirato K., Kawase M., Matsuyama S.: Wild-type human coronaviruses prefer cell-surface TMPRSS2 to endosomal cathepsins for cell entry. *Virology*, **517**, 9–15 (2018)
158. Sin J.H., Zangardi M.L.: Ruxolitinib for secondary hemophagocytic lymphohistiocytosis: First case report. *Hematol. Oncol. Stem Cell Ther.* **12**, 166–170 (2019)
159. Stebbing J., Corebellino M. *et al.*: Mechanism of baricitinib supports artificial intelligence-predicted testing in COVID-19 patients. *EMBO Mol. Med.* **12**, e12697 (2020)
160. Stebbing J., Phelan A., Griffin I., Tucker C., Oechsle O., Smith D., Richardson P.: COVID-19: combining antiviral and anti-inflammatory treatments. *Lancet Infect. Dis.* **20**, 400–402 (2020)
161. Ströher U., DiCaro A., Li Y., Strong J.E., Aoki F., Plummer F., Jones S.M., Feldmann H.: Severe Acute Respiratory Syndrome-Related Coronavirus Is Inhibited by Interferon-α. *J. Infect. Dis.* **189**, 1164–1167 (2004)
162. Talukdar R., Tandon R.K.: Pancreatic stellate cells: New target in the treatment of chronic pancreatitis. *J. Gastroenterol. Hepatol.* **23**, 34–41 (2008)

163. Tam R.C., Pai B., Bard J., Lim C., Averett D.R., Phan U.T., Milovanovic T.: Ribavirin polarizes human T cell responses towards a Type 1 cytokine profile. *J. Hepatol.* **30**, 376–382 (1999)
164. Tang W., Xie Q. *et al.*: Hydroxychloroquine in patients with mainly mild to moderate coronavirus disease 2019: open label, randomised controlled trial. *BMJ*, **369**, m1849 (2020)
165. Timani K.A., Zhu Y. *et al.*: Nuclear/nucleolar localization properties of C-terminal nucleocapsid protein of SARS coronavirus. *Virus Res.* **114**, 23–34 (2005)
166. Tipnis S., Hooper N., Hyde R., Karran E.H., Christie G., Turner A.J.: A human homolog of angiotensin-converting enzyme. Cloning and functional expression as a captopril-insensitive carboxypeptidase. *J. Biol. Chem.* **275**, 33238–33243 (2000)
167. Tong S., Su Y., Yu Y., Wu C., Chen J., Wang S., Jiang J.: Ribavirin therapy for severe COVID-19: a retrospective cohort study. *Int. J. Antimicrob. Agents*, **56**, 106114–106114 (2020)
168. Treml B., Loeckinger A. *et al.*: Recombinant angiotensin-converting enzyme 2 improves pulmonary blood flow and oxygenation in lipopolysaccharide-induced lung injury in piglets. *Crit. Care Med.* **38**, 596–601 (2010)
169. Tu Y.-F., Chien C.-S., Yarmishyn A., Lin Y.-Y., Luo Y.-H., Lin Y.-T., Lai W.-Y., Yang D.-M., Chou S.-J., Yang Y.-P. *et al.*: A Review of SARS-CoV-2 and the ongoing clinical trials. *Int. J. Mol. Sci.* **21**, 2657 (2020)
170. Ujike M., Nishikawa H., Otaka A., Yamamoto N., Yamamoto N., Matsuoka M., Kodama E., Fujii N., Taguchi F.: Heptad repeat-derived peptides block protease-mediated direct entry from the cell surface of severe acute respiratory syndrome coronavirus but not entry via the endosomal pathway. *J. Virol.* **82**, 588 (2008)
171. Uno Y.: Camostat mesilate therapy for COVID-19. *Intern. Emerg. Med.* 1–2 (2020)
172. van Kraaij T.D.A., Mostard R.L., Ramiro S., Magro Checa C., van Dongen C.M., van Haren E.H., Buijs J., Landewé R.B.: Tocilizumab in Severe COVID-19 Pneumonia and Concomitant Cytokine Release Syndrome. *Eur. J. Case Rep. Int. Med.* **7**, 001675–001675 (2020)
173. van Rhee F., Kurzrock R. *et al.*: Siltuximab, a novel anti-interleukin-6 monoclonal antibody, for Castleman's disease. *J. Clin. Oncol.* **28**, 3701–3708 (2010)
174. Vincent M.J., Bergeron E., Benjannet S., Erickson B.R., Rollin P.E., Ksiazek T.G., Seidah N.G., Nichol S.T.: Chloroquine is a potent inhibitor of SARS coronavirus infection and spread. *Virol. J.* **2**, 69–69 (2005)
175. Wagstaff K.M., Sivakumaran H., Heaton S.M., Harrich D., Jans D.A.: Ivermectin is a specific inhibitor of importin α/β-mediated nuclear import able to inhibit replication of HIV-1 and dengue virus. *Biochem. J.* **443**, 851–856 (2012)
176. Wan S., Chen Y. *et al.*: Characteristics of lymphocyte subsets and cytokines in peripheral blood of 123 hospitalized patients with 2019 novel coronavirus pneumonia (NCP). *medRxiv*, 2020.2002.2010.20021832 (2020)
177. Wang J., Wang Y., Wu L., Wang X., Jin Z., Gao Z., Wang Z.: Ruxolitinib for refractory/relapsed hemophagocytic lymphohistiocytosis. *Haematologica*, **105**, e210–e212 (2019)
178. Wang M., Cao R., Zhang L., Yang X., Liu J., Xu M., Shi Z., Hu Z., Zhong W., Xiao G.: Remdesivir and chloroquine effectively inhibit the recently emerged novel coronavirus (2019-nCoV) in vitro. *Cell Res.* **30**, 269–271 (2020)
179. Wang X., Cao R., Zhang H., Liu J., Xu M., Hu H., Li Y., Zhao L., Li W., Sun X. *et al*: The anti-influenza virus drug, arbidol is an efficient inhibitor of SARS-CoV-2 *in vitro*. *Cell Discov.* **6**, 28 (2020)
180. Wang X., Xu W., Hu G., Xia S., Sun Z., Liu Z., Xie Y., Zhang R., Jiang S., Lu L.: SARS-CoV-2 infects T lymphocytes through its spike protein-mediated membrane fusion. *Cell. Mol. Immunol.* **RETRACTED** (2020)
181. Wang Y., He Z. *et al.*: Inhibition of the infectivity and inflammatory response of influenza virus by Arbidol hydrochloride in vitro and in vivo (mice and ferret). *Biomed. Pharmacother.* **91**, 393–401 (2017)
182. Wang Y., Guo D. *et al.*: Coronavirus nsp10/nsp16 methyltransferase can be targeted by nsp10-derived peptide *in vitro* and *in vivo* to reduce replication and pathogenesis. *J. Virol.* **89**, 8416–8427 (2015)
183. Wang Y., Wang C. *et al.*: Remdesivir in adults with severe COVID-19: a randomised, double-blind, placebo-controlled, multicentre trial. *Lancet*, **395**, 1569–1578 (2020)
184. Wang Y., Wang C. *et al.*: Remdesivir in adults with severe COVID-19: a randomised, double-blind, placebo-controlled, multicentre trial. *Lancet*, **395**, 1569–1578 (2020)
185. Wang Z., Chen X., Lu Y., Chen F., Zhang W.: Clinical characteristics and therapeutic procedure for four cases with 2019 novel coronavirus pneumonia receiving combined Chinese and Western medicine treatment. *BioScience Trends*, **14**, 64–68 (2020)
186. Williamson B.N., de Wit E. *et al.*: Clinical benefit of remdesivir in rhesus macaques infected with SARS-CoV-2. *bioRxiv*, 2020.2004.2015.043166 (2020)
187. Wrapp D., Wang N., Corbett K.S., Goldsmith J.A., Hsieh C.-L., Abiona O., Graham B.S., McLellan J.S.: Cryo-EM Structure of the 2019-nCoV Spike in the Prefusion Conformation. *bioRxiv*, 2020.2002.2011.944462 (2020)
188. Wujtewicz M., Dylczyk-Sommer A., Aszkiełowicz A., Zdanowski S., Piwowarczyk S., Owczuk R.: COVID-19 – what should anaethesiologists and intensivists know about it? *Anaesthesiol. Intensive Ther.* **52**, 34–41 (2020)
189. Xia S., Lu L. *et al.*: Inhibition of SARS-CoV-2 (previously 2019-nCoV) infection by a highly potent pan-coronavirus fusion inhibitor targeting its spike protein that harbors a high capacity to mediate membrane fusion. *Cell Res.* **30**, 343–355 (2020)
190. Xia S., Lu L. *et al.*: A pan-coronavirus fusion inhibitor targeting the HR1 domain of human coronavirus spike. *Sci. Adv.* **5**, eaav4580 (2019)
191. Xu J., Zhao S., Teng T., Abdalla A.E., Zhu W., Xie L., Wang Y., Guo X.: Systematic Comparison of Two Animal-to-Human Transmitted Human Coronaviruses: SARS-CoV-2 and SARS-CoV. *Viruses*, **12**, 244 (2020)
192. Xu P., Huang J., Fan Z., Huang W., Qi M., Lin X., Song W., Yi L.: Arbidol/IFN-α2b therapy for patients with corona virus disease 2019: a retrospective multicenter cohort study. *Microbes Infect.* **22**, 200–205 (2020)
193. Xu X., Wei H. *et al.*: Effective treatment of severe COVID-19 patients with tocilizumab. *Proc. Nat Acad. Sci. USA*, **117**, 10970 (2020)
194. Yamamoto M., Matsuyama S., Li X., Takeda M., Kawaguchi Y., Inoue J.-I., Matsuda Z.: 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**, 6532–6539 (2016)
195. Yang Y., Ye F., Zhu N., Wang W., Deng Y., Zhao Z., Tan W.: Middle East respiratory syndrome coronavirus ORF4b protein inhibits type I interferon production through both cytoplasmic and nuclear targets. *Sci. Rep.* **5**, 17554 (2015)
196. Yao X., Liu D. *et al.*: 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.* ciaa237 (2020)
197. Yin W., Xu H.E. *et al.*: Structural basis for inhibition of the RNA-dependent RNA polymerase from SARS-CoV-2 by remdesivir. *Science*, **368**, 1499–1504 (2020)

198. Yokota S., Miyamae T., Imagawa T., Iwata N., Katakura S., Mori M., Woo P., Nishimoto N., Yoshizaki K., Kishimoto T.: Therapeutic efficacy of humanized recombinant anti-interleukin-6 receptor antibody in children with systemic-onset juvenile idiopathic arthritis. *Arthritis Rheum.* **52**, 818–825 (2005)
199. Zhang C., Wu Z., Li J.-W., Zhao H., Wang G.-Q.: The cytokine release syndrome (CRS) of severe COVID-19 and Interleukin-6 receptor (IL-6R) antagonist Tocilizumab may be the key to reduce the mortality. *Int. J. Antimicrob. Agents*, **55**, 105954–105954 (2020)
200. Zhang P., Wang F. *et al.*: The novel coronavirus (COVID-19) pneumonia with negative detection of viral ribonucleic acid from nasopharyngeal swabs: a case report. *BMC Infect. Dis.* **20**, 317–317 (2020)
201. Zhou P., Yang X.-L., Wang X.-G., Hu B., Zhang L., Zhang W., Si H.-R., Zhu Y., Li B., Huang C.-L. *et al.*: A pneumonia outbreak associated with a new coronavirus of probable bat origin. *Nature*, **579**, 270–273 (2020)
202. Zhou Y., Simmons G. *et al.*: Protease inhibitors targeting coronavirus and filovirus entry. *Antiviral Res.* **116**, 76–84 (2015)
203. Zhu Z., Lu Z., Xu T., Chen C., Yang G., Zha T., Lu J., Xue Y.: Arbidol monotherapy is superior to lopinavir/ritonavir in treating COVID-19. *J. Infect.* **81**, e21–e23 (2020)
204. Zinter M.S., Hermiston M.L.: Calming the storm in HLH. *Blood*, **134**, 103–104 (2019)
205. Zou Z., Jiang C. *et al.*: Angiotensin-converting enzyme 2 protects from lethal avian influenza A H5N1 infections. *Nat. Commun.* **5**, 3594–3594 (2014)