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Perspectives: potential therapeutic options for SARS-CoV-2 patients based on feline infectious peritonitis strategies: central nervous system invasion and drug coverage

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Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) infections are continuing to increase globally, and clinicians at hospitals are currently preparing lists of US Food and Drug Administration (FDA)-approved therapies as options for the treatment of SARS-CoV-2. For several years, we have been investigating anti-coronavirus therapies directed at feline infectious peritonitis (FIP) [1,2], a disease caused by a coronavirus with a nearly 100% mortality in felines. Feline enteric coronavirus (FEC), commonly found in many asymptomatic felines, mutates into the virulent and lethal FIP coronavirus [3]. We believe that our experimental observations for the treatment of FIP may be relevant and translational for recent *in vitro* results of SARS-CoV-2 [4] in the absence of extensive laboratory and human clinical trials. A FIP coronavirus protease inhibitor (GC376) was successful in the treatment of a subset of felines with FIP; however, in cases where there was neurological involvement, the protease inhibitor was unable to prevent progression of central nervous system (CNS) disease, resulting in neurological FIP and subsequent euthanasia [5]. The polymerase inhibitor GS-441524 has already demonstrated significant activity in a feline clinical trial against FIP [1], but the treatment of neurological involvement has yet to be demonstrated. Remdesivir, which is a prodrug of GS-441524, shows great promise for the treatment of SARS-CoV-2 [6] but is not currently approved by the FDA and is only available in an intravenous formulation. There is an urgent need for anti-SARS-CoV-2 therapies that are already FDA-approved, orally bioavailable, appropriate for organs that express the SARS-CoV-2 target angiotensin-converting enzyme 2 (ACE2), and may also complement or synergise with remdesivir upon approval. Whilst the detailed experimental results will be communicated elsewhere (unpublished data from BGM Laboratory), we believe our observations could support clinicians regarding treatment options in addition to supportive care.

Severe acute respiratory syndrome coronavirus 1 (SARS-CoV-1) and SARS-CoV-2 both target ACE2 as the receptor [7], which is expressed in the lungs, heart, gastrointestinal tract and CNS [8] in humans. SARS-CoV-1 is known to penetrate the CNS through the olfactory nerve and olfactory bulb route [9], similar to other coronaviruses [10]. Patients with coronavirus disease 2019 (COVID-19), the disease caused by SARS-CoV-2, often experience anosmia (loss of smell), suggesting that this route may also occur following SARS-CoV-2 infection. Almost all Betacoronaviruses penetrate the CNS [10], and SARS-CoV-1 and SARS-CoV-2 share the same ACE2 receptor. It is also reasonable to believe that the massive infection of the brainstem in experimental animals following SARS-CoV-1 nasal exposure [9] may also occur with SARS-CoV-2, which could contribute to sudden respiratory failure as observed with some patients [10]. It is not clear whether SARS-CoV-2 CNS penetration may also occur in patients with recent damage to the blood–brain barrier (BBB) following a stroke or other brain insult. As we have shown in felines, the implications of CNS penetration emphasise the need for a multipronged organ-appropriate strategy that will suppress SARS-CoV-2 both in the periphery and the brain.

We have found that nelfinavir and amodiaquine have anti-FIP activity *in vitro* that is comparable with chloroquine, and superior to ribavirin, penciclovir, favipiravir and nafamostat against SARS-

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CoV-2 [4]. Amodiaquine, like chloroquine and hydroxychloroquine, is a CNS-penetrating 4-aminoquinoline antimalarial drug that inhibits the formation of hemozoin in the parasite, but it has been withdrawn from the US market although it is still available in other countries. Amodiaquine is known to possess some antiviral activity, and derivatives have been explored for inhibition of Ebola virus infection [11]. Pharmacogenomics has revealed that the presence of the CYP450 2C8*2 allele is an important contributor to amodiaquine toxicity [12]. Appropriate monitoring parameters include complete blood counts with differential and liver function tests, as serious adverse events are agranulocytosis and hepatotoxicity with mild adverse events being nausea, emesis and pruritus. Amodiaquine/artesunate is available for the treatment of malaria; it is cost effective and accessible outside of the USA. This is the third observation of a 4-aminoquinoline having activity against a coronavirus and compliments clinical observations from China [13,14]. Second, the 4-aminoquinolines are well known to penetrate the BBB and have been investigated for broad-spectrum antiviral activity against a variety of viral infections, including Zika [15], Dengue [16] and Ebola [17] viruses. It also may have utility for those patients suffering from SARS-CoV-2 in the brainstem. The antiviral mechanisms of action of chloroquine may include altering endosomal RNA release [15], altering autophagy-dependent viral replication [15] and inhibiting ACE2 glycosylation [18].

Nelfinavir is an older anti-human immunodeficiency virus (HIV) protease inhibitor capable of inhibiting HIV-1 and, to a lesser extent, HIV-2 proteases [19], but is no longer the first treatment of choice. However, it has a spectrum of activity that includes both SARS-CoV-1 [20] and FIP coronavirus [21], is orally bioavailable, and can achieve a plasma concentration of 7.3 mg/L at a dose of 3000 mg twice daily [22]. Other protease inhibitors, including the combination of lopinavir and ritonavir, were utilised for the treatment of SARS-CoV-1 [23] and have been used in Singapore [24] and China [25] for the treatment of SARS-CoV-2. However, there have been challenges associated with toxicity at the prescribed doses [24] as well as efficacy as monotherapy [25]. The hypothesis for using older antiretroviral agents with higher toxicity but a potentially broader antiviral spectrum of activity is not novel. However, the experimental observation of nelfinavir suppressing FIP coronavirus [21] provides additional data to consider nelfinavir as an option for SARS-CoV-2. Appropriate monitoring parameters for nelfinavir include echocardiogram for QT interval prolongation and torsades de pointes as well as diarrhoea, fatigue (10–20%), lipodystrophy and hyperglycaemia.

In summary, these observations of *in vitro* activity against FIP coronavirus are not a substitute for clinical data and trials but may provide further guidance for off-label therapeutic strategies. The mutation of FEC into FIP coronavirus may provide a paradigm for considering the relationship between different strains of SARS-CoV-2. Nelfinavir, chloroquine and hydroxychloroquine are FDA-approved, orally bioavailable and commercially available and have at least *in vitro* data against either SARS-CoV-1 or SARS-CoV-2. Nelfinavir may be an alternative to lopinavir/ritonavir. Amodiaquine, hydroxychloroquine and chloroquine all possess CNS penetration ability. Amodiaquine may be an alternative to chloroquine in territories where it is available. These agents can offer clinicians another therapeutic strategy beyond supportive care as monotherapy or in combination.

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Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.ijantimicag.2020.105964.

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