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On the Road Back to Normalcy: Following Science Over Noise in SARS-CoV-2

The persistence of the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) pandemic, despite the rapid development of effective vaccines, has led to the slowly emerging acceptance that eradication of COVID-19 is unlikely and that strategies for ongoing management are needed. To that end, ongoing research into disease mechanisms remains vital, and the current article by Dr Parmar reviews a key mechanism of SARS-CoV-2 infectivity with potential therapeutic significance.

The importance of transmembrane protease serine 2 (TMPRSS2) in SARS-CoV-2 was first highlighted in March 2020 by Hoffmann et al.[1] This seemingly instantaneous elucidation of a mechanism at the beginning of the pandemic was made possible because SARS-CoV-2 shares approximately 76% of its amino acid sequence with SARS-CoV, the virus responsible for the severe acute respiratory syndrome outbreak of 2002-2003. Whereas that outbreak was successfully contained, it nevertheless led to research that elucidated the key roles of TMPRSS2 and angiotensin-converting enzyme 2 for viral entry into human cells.[2-4] Hoffmann et al[1] had the insight to quickly replicate these experiments, and they also demonstrated that the TMPRSS2 inhibitor camostat can decrease SARS-CoV-2 infectivity in cell line studies.

As Dr Parmar[5] notes, studies of camostat and related TMPRSS2 inhibitors were rapidly launched around the world, including by our group at Mayo Clinic. Our study is testing camostat mesylate, given 4 times daily, in addition to standard of care to hospitalized patients with COVID-19 (NCT04470544). Other studies span the spectrum of disease states from early, minimally symptomatic infections to the severely ill and given in combination with a variety of different agents (Table).[6-31]

Early results have been mixed. A Danish study randomized 137 hospitalized patients to camostat mesylate at 200 mg 3 times daily vs placebo, with the primary outcome measure being time to discharge or clinical improvement. The investigators reported a median time to clinical improvement of 5 days in both groups (P=.31). The hazard ratio for 30-day mortality in the camostat compared with the placebo group was 0.82 (95% CI, 0.24 to 2.79; P=.75).[6] A similar study in Japan randomized 133 asymptomatic to moderately ill patients to camostat or placebo with a primary end point of time to SARS-CoV-2 test result negativity and saw no difference.[8] A third negative study, ACTIV-2, enrolled 224 participants in a single arm design that examined changes in viral shedding or symptomatic improvement. The announced results state that the camostat arm failed to reach the predetermined criteria for advancement and has thus been discontinued.[10] In contrast, a South Korean study reported thus far only in a press release announced a 40% faster recovery rate for patients treated with camostat. The benefit was greater in patients older than 50 years, with a more than 50% improvement in the recovery rate. In that study, 175 patients were randomized to camostat vs placebo.[9]

Although the hue and cry of public, media, and political demands may deem such data to be unsatisfactory, experienced scientists and investigators are all too familiar with the difficulty of scientific endeavor and discovery. Progress occurs in fits and starts or often too slowly, then all at once. Further analysis is needed to understand whether the existing data point to drug failure or flaws or limitations of study design. The results of the remaining ongoing studies should be instructive, and an international consortium of camostat

TABLE. Studies Involving Camostat Mesylate

Study, PI, and country	Study design and target population	Randomization, agents, and enrollment	Outcomes analyzed	Results
NCT04321096; EudraCT 2020-001200-42 Gunst et al[6] Denmark and Sweden	Multicenter, double blind, randomized, placebo controlled Hospitalized (mild, moderate, severe, and critical)	Randomization 2:1 N=205 Camostat (n=137) and placebo (n=68) Camostat mesylate 200 mg 3 times a day × 5 days or placebo	Primary outcome: time to discharge or clinical improvement (measured as ≥2 points improvement on 7-point ordinal scale)	Camostat mesylate treatment did not significantly improve clinical outcomes
NCT04451083 Kitagawa et al[7] Japan	Phase 1 study Healthy participants; pharmacokinetics and safety profiles were evaluated	Camostat mesylate orally 600 mg 4 times a day	Time of plasma GBPA concentration exceeding effective concentration was estimated as time above half-maximal effective concentration (EC_{50})	Camostat mesylate was safe and tolerated
Phase 3 study by ONO Pharmaceutical[8] (press release) Japan	Phase 3 study, multicenter, double blind, randomized, placebo controlled Asymptomatic to moderate COVID-19	Camostat (n=77) and placebo (n=76) Camostat mesylate orally 600 mg 4 times daily × 14 days	Primary end point: time to SARS-CoV-2 negative test result	Camostat mesylate did not meet the time to requirement of SARS-CoV-2 negative test result
Daewoong Pharmaceutical[9] (press release) South Korea	Phase 2b Placebo-controlled, randomized, and double-blind multicenter clinical trial Mild COVID-19	N=342 Camostat=86 Placebo=89	Primary end point: time taken to improve clinical symptoms	Improvement of symptoms such as cough and dyspnea in patients >50 years was twice as fast and statistically significant (treatment group, 4 days; placebo group, 9 days)
ACTIV-2 phase 2 study[10]	Placebo-controlled, randomized, and double-blind multicenter clinical trial	N=224 Camostat orally 200 mg every 6 hours × 7 days	Primary end point: early changes in viral shedding or improvement in symptoms	Phase 2 data failed to meet the criteria for graduation to phase 3
NCT04321096; EudraCT 2020-001200-42 Søgaard et al[11] Denmark	Randomized, placebo controlled Mild and severe	Randomization 1:1 (ambulatory) and 2:1 (hospitalized), placebo controlled Ambulatory (2 × 200) and hospitalized (120 + 60) patients Camostat 200 mg 3 times a day × 5 days Analysis completed for the in-hospital cohort	Primary end point: ambulatory: no fever 48 h plus symptom improvement 7-point clinical scale for hospitalized patients	Ongoing
NCT04353284 Vinetz et al[12] Yale University	Randomized, placebo controlled Mild ambulatory cases	Randomization 1:1 N=2 × 57 Camostat 4 × 200 mg daily × 7 days	Primary end point: viral load (analysis in batch, including saliva test) and symptoms	Ongoing

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TABLE. Continued

Study, PI, and country	Study design and target population	Randomization, agents, and enrollment	Outcomes analyzed	Results
NCT04374019 Arnold et al[13] Kentucky University	Randomized multiple arms Ambulatory and hospital (not ventilated)	N=60 patients per arm Comparing with ivermectin Camostat 200 mg 3 times daily × 14 days	Primary end point: 2-point deterioration on 7-point clinical scale	Ongoing
NCT04455815; EudraCT 2020-002110-41 Dhaliwal et al[14] CRUK/Edinburgh University	Randomized open label Ambulatory	1:1 randomized open label N=2 × 195 patients Comparison with standard of care Camostat 200 mg 4 times daily × 14 days	Primary end point: hospitalization requiring supplemental oxygen, time frame days 1-28	Ongoing
NCT04470544 Bryce et al[15] Mayo Clinic Arizona	Randomized placebo controlled Hospitalized patients	1:1 randomized N=2 × 138 patients Camostat 200 mg 4 times daily × 14 days	Primary end point: alive and free from respiratory failure at day 28	Ongoing
NCT04435015 Mani et al[16] Yale University	Randomized placebo controlled Hospitalized patients	1:1 randomized N=2 × 100 patients Camostat 200 mg 3 times daily until discharge	Primary end point: D-dimer	Ongoing planned
NCT04608266 EudraCT 2020-003366-39 (CAMOVID) Boutboul et al[17] France	Randomized, placebo controlled Ambulatory	1:1 randomized N=2 × 298 patients Camostat 200 mg 3 times daily × 14 days	Primary end point: hospitalization	Ongoing recruitment
NCT04521296 Daewoong Pharmaceutical[18] South Korea	Randomized, placebo controlled, phase 2a, phase 2b Mild to moderate COVID	1:1 randomized N=2 × 45 patients in phase 2a, 300 patients in phase 2b Camostat 200 mg 3 times daily × 14 days	Primary end point: time to negative RNA	Ongoing—recruitment complete
NCT04530617 Palazuelos et al[19] Mexico	Randomized, placebo controlled Ambulatory	1:1:1:1 randomized N=4 × 90 patients Camostat 200 mg 3 times daily × 14 days	Primary end point: hospitalization and oxygen use at day 14	Ongoing recruitment
NCT04524663; NCT04662073; NCT04662086 Parsonnet et al[20] Stanford	Randomized, placebo controlled (adaptive design, sharing controls) Ambulatory	1:1 randomized N=2 × 60 patients Camostat 200 mg 4 times daily ×10 days	Primary end point: viral shedding, up to day 28	Ongoing recruitment

TABLE. Continued

Study, PI, and country	Study design and target population	Randomization, agents, and enrollment	Outcomes analyzed	Results
NCT04583592 (CAMELOT) Sagent Pharmaceuticals[21]	Randomized, placebo controlled Ambulatory	2:1 randomized N=200 + 100 patients Camostat 200 mg 4 times daily × 14 days	Primary end point: hospitalization or death before day 28	Ongoing—recruitment complete
NCT04625114 De Scheerder et al[22] Belgium	Randomized, placebo controlled Mild symptoms or no symptoms with high viral load Ambulatory	1:1 randomized N=2 × 75 patients Camostat 300 mg 3 times daily × 5-10 days	Primary end point: viral load change from day 0 to day 5	Ongoing recruitment
NCT04355052 Levi et al[23] Israel	Randomized, open label (amended) Hospital	2:1 randomized N=160 + 80 patients Comparison: standard of care Camostat 200 mg 3 times daily × 10 days	Primary end point: NEWS and PCR	Ongoing recruitment
NCT04338906; EudraCT 2020-004695-18 Feldt et al[24] Germany	Randomized, placebo controlled (amended) Early treatment of ambulatory/hospital	2:2:1:1 randomized N=332 + 332 + 166 + 166 patients Comparison: (convalescent plasma); standard of care; placebo Camostat 200 mg 3 times daily × 7 days	Primary end point: progression to clinical status ≥4b WHO	Ongoing recruitment
JPRN-jRCTs031200113 Yasuriro et al[25] Tokyo	Randomized, placebo controlled Preventive use Ambulatory	1:1 randomized N=2 × 300 patients Camostat 5 mg in 100 mL orally 4 times daily × 56 days	Primary end point: positive antibody or PCR test	Ongoing recruitment
NCT04730206; EudraCT 2020-005911-27 Van den Bruel et al[26] Belgium	1:1 randomized, placebo controlled Symptomatic >50 years Ambulatory	1:1 randomized N=2 × 653 patients Compare to placebo Camostat 200 mg 4 times daily × 7 days	Primary end point: hospitalization >24 h or death before day 30	Ongoing, not yet recruiting
NCT04652765 Marshall et al[27] Johns Hopkins, Baltimore	Randomized open label Ambulatory	1:1:1 randomized Comparison: standard of care Camostat 600 mg 4 times daily × 7 days + bicalutamide 150 mg daily for 7 days	Primary end point: hospitalization before day 28	Ongoing recruitment
NCT04518410 (ACTIV-2) Smith et al[28] UC San Diego	Randomized, placebo-controlled, adaptive platform trial Symptomatic, higher risk of progression to severe disease Ambulatory	1:1 randomized N=2000 patients Compare: IV bamlanivimab, BRII-196/BRII-198, or AZD7442; inhaled SNG001; intramuscular AZD7442;	Primary end point: duration of symptoms	Ongoing recruitment

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TABLE. Continued				
Study, PI, and country	Study design and target population	Randomization, agents, and enrollment	Outcomes analyzed	Results
		subcutaneous C135-LS + C144-LS Camostat 200 mg 4 times daily × 7 days		
jRCTs031200196 Kenji et al[29] Japan	Randomized open label Mild severity Hospital	1:1 randomized N=2 × 50 patients Compare to standard of care Camostat + favipiravir + inhaled ciclesonide for 10 days	Primary end point: length of hospital stay	Ongoing recruitment
NCT04713176 Daewoong Pharmaceutical[30]	Randomized open label Severe disease Hospital	1:1 randomized N=2 × 560 patients Compare: placebo + IV remdesivir up to 5 days Camostat 200 mg 3 times daily × 14 days + IV remdesivir up to 5 days	Primary end point: mortality or ECMO up to 29 days	Ongoing recruitment
EudraCT 2020-002233-15 Schultz-Heienbrok et al[31] Germany	Randomized placebo controlled Mild severity, ambulatory	N=40 patients Comparison: placebo Camostat 600 mg 4 times daily + niclosamide 2 g × 7 days	Primary end point: viral load	Ongoing recruitment

COVID-19, coronavirus disease 2019; ECMO, extracorporeal membrane oxygenation; GBPA, 4-(4-guanidinobenzoyloxy)phenylacetic acid; IV, intravenous; NEWS, National Early Warning Score; PCR, polymerase chain reaction; PI, principal investigator; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2; WHO, World Health Organization.

investigators has already been convened to analyze pooled data.

One possibility is that multidrug inhibition of SARS-CoV-2 entry will ultimately be needed to provide clinically meaningful levels of benefit. Evidence supporting combinations of entry inhibitors over monotherapy already exists with various compounds, including the cathepsin inhibitors EST [(23,25) *trans*-epoxysuccinyl-L-leucylamindo- 3-methylbutane ethyl ester][32] and E64d,[1,33] a proprotein convertase inhibitor,[33] and the protease furin.[33] It may also be that the timing, dosing, or delivery of camostat or other TMPRSS2 inhibitors remains to be optimized, and variations on these parameters, such as higher dosing and aerosolized nasal delivery, are being pursued.

Ultimately, there is every reason to believe that scientific discovery such as that reviewed here by Dr Parmar will stop the endless waves of outbreaks that strain health care systems across multitudinous communities. Whether targeting of TMPRSS2 is part of that solution remains to be seen, but insights such as these into disease mechanisms are the vital signposts guiding us back to normalcy.

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REFERENCES

1. Hoffmann M, Kleine-Weber H, Schroeder S, et al. SARS-CoV-2 cell entry depends on ACE2 and TMPRSS2 and is blocked by a clinically proven protease inhibitor. *Cell*. 2020;181(2):271-280.e8.
2. Glowacka I, Bertram S, Müller MA, et al. Evidence that TMPRSS2 activates the severe acute respiratory syndrome coronavirus spike protein for membrane fusion and reduces viral control by the humoral immune response. *J Virol*. 2011; 85(9):4122-4134.
3. Matsuyama S, Nagata N, Shirato K, Kawase M, Takeda M, Taguchi F. Efficient activation of the severe acute respiratory syndrome coronavirus spike protein by the transmembrane protease TMPRSS2. *J Virol*. 2010;84(24):12658-12664.
4. Shulla A, Heald-Sargent T, Subramanya G, Zhao J, Perlman S, Gallagher T. A transmembrane serine protease is linked to the severe acute respiratory syndrome coronavirus receptor and activates virus entry. *J Virol*. 2011;85(2):873-882.
5. Parmar MS. TMPRSS2: an equally important protease as ACE2 in the pathogenicity of SARS-CoV-2 infection. *Mayo Clin Proc*. 2021. https://doi.org/10.1016/j.mayocp.2021.07.005.
6. Gunst JD, Staerke NB, Pahus MH, et al. Efficacy of the TMPRSS2 inhibitor camostat mesilate in patients hospitalized with Covid-19—a double-blind randomized controlled trial. *EClinicalMedicine*. 2021;35:100849.
7. Kitagawa J, Arai H, Iida H, et al. A phase I study of high dose camostat mesylate in healthy adults provides a rationale to repurpose the TMPRSS2 inhibitor for the treatment of COVID-19. *Clin Transl Sci*. 2021 May 13. https://doi.org/10.1111/cts.13052.
8. Phase III study result of Foipan tablets, a protease enzyme inhibitor. in Japan in patients with novel coronavirus infection (COVID-19), https://www.ono-pharma.com/sites/default/files/en/news/press/enews20210611.pdf.
9. Daewoong Pharmaceutical announces camostat achieving 50% faster recovery time for mild COVID-19 patients over age of 50 in topline results from phase 2B clinical trial. https://www.prnewswire.com/news-releases/daewoong-pharmaceutical-announces-camostat-achieving-50-faster-recovery-time-for-mild-covid-19-patients-over-age-of-50-in-topline-results-from-phase-2b-clinical-trial-301343800.html.
10. ACTG announces Camostat will not advance to phase 3 in outpatient treatment study for COVID-19. https://www.eurekalert.org/news-releases/723362.
11. The impact of camostat mesilate on COVID-19 infection (CamoCO-19). https://clinicaltrials.gov/ct2/show/NCT04321096.
12. Camostat mesylate in COVID-19 outpatients. https://clinicaltrials.gov/ct2/show/NCT04353284.
13. Randomized, multi-arm phase II trial of novel agents for treatment of high-risk COVID-19 positive patients. https://fdaaa.trialstracker.net/trial/NCT04374019/.
14. A trial looking at the use of camostat to reduce progression of symptoms of coronavirus (COVID-19) in people who have tested positive (SPIKE-1). https://clinicaltrials.gov/ct2/show/NCT04455815.
15. Camostat mesilate treating patients with hospitalized patients with COVID-19 (RECOVER). https://clinicaltrials.gov/ct2/show/NCT04470544.
16. The utility of camostat mesylate in patients with COVID-19 associated coagulopathy (CAC) and cardiovascular complications. https://clinicaltrials.gov/ct2/show/NCT04435015.
17. CAMOVID: evaluation of efficacy and safety of camostat mesylate for the treatment of SARS-CoV-2 infection—COVID-19 in ambulatory adult patients (CAMOVID). https://clinicaltrials.gov/ct2/show/NCT04608266.
18. Efficacy and safety of DWJ1248 in patients with mild to moderate COVID-19 compared to the placebo. https://clinicaltrials.gov/ct2/show/NCT04521296.
19. Camostat and artemisia annua vs placebo in COVID-19 outpatients. https://clinicaltrials.gov/ct2/show/NCT04530617.
20. Stanford Medicine. Camostat mesilate (drugs). Development lead. https://racetoacure.stanford.edu/development-leads/10.
21. Camostat efficacy vs. placebo for outpatient treatment of COVID-19 (CAMELOT). https://clinicaltrials.gov/ct2/show/NCT04583592.
22. The potential of oral camostat in early COVID-19 disease in an ambulatory setting to reduce viral load and disease burden. https://clinicaltrials.gov/ct2/show/NCT04625114.
23. Open label study to compare efficacy, safety and tolerability of hydroxychloroquine combined with azithromycin compared to

hydroxychloroquine combined with camostat mesylate and to "no treatment" in SARS CoV 2 virus (COSTA). https://clinicaltrials.gov/ct2/show/NCT04355052.

24. Combination therapy with camostat mesilate + hydroxychloroquine for COVID-19 (CLOCC). https://clinicaltrials.gov/ct2/show/NCT04338906.

25. EUnetHTA. "Rolling Collaborative Review" of Covid-19 treatment. Camostat for the treatment of COVID-19. https://www.eunethta.eu/wp-content/uploads/2021/07/EUnetHTA-COVID-19-RollingCR04-v10.0.pdf.

26. The DAWN camostat trial for ambulatory COVID-19 patients (DAWN). https://clinicaltrials.gov/ct2/show/NCT04730206.

27. Camostat with bicalutamide for COVID-19 (COMBO). https://clinicaltrials.gov/ct2/show/NCT04652765.

28. ACTIV-2: a study for outpatients with COVID-19. https://clinicaltrials.gov/ct2/show/NCT04518410.

29. Combination therapy of favipiravir, camostat mesilate and inhaled ciclesonide for COVID-19 pneumonia. https://covid-19.cochrane.org/studies/crs-16897364.

30. Efficacy and safety of DWJ1248 with remdesivir in severe COVID-19 patients. https://clinicaltrials.gov/ct2/show/NCT04713176.

31. Clinical trials for eudract_number:2020-002233-15. https://www.clinicaltrialsregister.eu/ctr-search/search?query=eudract_number:2020-002233-15.

32. 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 Virol. 2012;86(12):6537-6545.

33. Shang J, Wan Y, Luo C, et al. Cell entry mechanisms of SARS-CoV-2. Proc Natl Acad Sci U S A. 2020;117(21):11727-11734.