VIEWPOINTS ON HIV RESEARCH

COVID-19 research: an opinion piece

L Waters [ID][1] and JK Rockstroh [ID][2]

[1]*Department of HIV & Sexual Health, Mortimer Market Centre, Central & North West London NHS Trust, London, UK and* [2]*Department of Medicine I, Universitätsklinikum Bonn, Bonn, Germany*

The unprecedented global scale of COVID-19 globally has triggered a race to discover interventions to reduce associated morbidity and mortality and rapid release of research findings prior to any degree of critical review. As with previous novel infection outbreaks, antiretrovirals are just one drug class that has been held up as a potential strategy for prophylaxis and treatment with scant evidence and risk of harm. Here we summarize the evidence for antiretrovirals to treat COVID-19 and, as a drug that has also been studied in HIV, hydroxychloroquine, and flag some of the pitfalls of using therapies that have not been evaluated robustly.

Keywords: antiretroviral, COVID, COVID-19, HIV, hydroxychloroquine

Accepted 11 June 2020

Introduction

December 2019 in China saw the start of an outbreak of a novel coronavirus, SARS-CoV-2. The associated illness, COVID-19, emerged as a global pandemic with almost 6.5 million cases and exceeding 380 000 deaths worldwide at the time of writing [1].

The speed at which COVID-19 has taken hold is mirrored by the speed at which related research has been undertaken, and published. While the drivers for rapid assessment of possible interventions are obvious, we have seen widespread promotion, and implementation, of poor-quality findings and consequent harm that was potentially avoidable.

An outpouring of case reports and case series supporting the use of repurposed drugs for serious outbreaks is nothing new, as described for the Ebola epidemic in 2015/2016 [2]. Drugs may yield benefits, of course, though few have yet done so convincingly for COVID-19; they may also cause harm to individuals and to broader populations, for example, where surges in use create shortages for people with an evidence-based indication for the agent in question. Drug repurposing offers many advantages, including bypassing earlier stages of drug development and opportunities to utilize off-patent medications [3]. Drug repurposing does not negate the need for some preclinical validation, yet despite this, phase 3 trials are underway for drugs that have not yet demonstrated any *in vitro* or animal model activity against SARS-CoV-2 [4-6].

The balance of research speed and research quality is delicate, and though rapid and open publication is laudable, we must include the impact of less rigorous, or even absent, peer review when appraising that evidence. Prepublication access [7,8], rapid online and social media dissemination of conclusions (valid and otherwise), and the breakneck speed at which national and international bodies include new findings in their guidance create an environment rife with myth propagation.

Here we highlight some of the proposed antiretroviral (ARV)-based treatment and prevention strategies for COVID-19, where hysteria may have trumped objectivity.

Obviously, this is a rapidly evolving field so we direct readers to the 'living mapping and living systematic review' of COVID-19 studies accessible here: https://covid-nma.com/.

HIV antiretrovirals for COVID-19 treatment

A recent systematic review summarized the efficacy of ARVs against three serious coronavirus-associated diseases: COVID-19, severe acute respiratory syndrome (SARS) and Middle East respiratory syndrome (MERS) [9]. The most studied ARV is the HIV protease inhibitor, ritonavir-boosted lopinavir (LPV/r) but, of the 10 published SARS and MERS 'trials', six were single case reports. The 14 COVID-19 trials comprised three single case reports, a case series ($n = 4$), eight retrospective cohorts (totalling 408 patients, many of whom received additional treatments, including interferon, steroids, oseltamivir and ganciclovir); no clear benefits were reported.

Correspondence: Laura Waters, Department of HIV & Sexual Health, 5th Floor Mortimer Market Centre, Central & North West London NHS Trust, Off Tottenham Court Road, London, WC1E 6JB, UK. Tel: +44 20 3317 5241; e-mail: lwaters@nhs.net

To date there have been two published randomized controlled trials (RCTs) of LPV/r for COVID-19: one randomized 86 people with mild/moderate COVID-19 to LPV/r ($n = 34$), umifenovir ($n = 35$) or no antivirals ($n = 17$) [10]. Numerically more individuals in the LPV/r arm experienced clinical deterioration and the authors concluded that neither LPV/r nor umifenovir monotherapy provided clinical benefit over supportive care for hospitalized patients with mild/moderate COVID-19. A larger open-label RCT in 199 people hospitalized with severe COVID-19 demonstrated no benefit of LPV/r over standard of care [11] in terms of clinical improvement or viral clearance. However, a trend to better outcomes when LPV/r was started early (within 12 days of symptom onset) was considered worthy of further study and there are many trials investigating LPV/r recruiting now or imminently. Of note, antivirals including LPV/r may offer more benefit when used in early [11] or less severe COVID-19 and immunomodulatory therapies, which we will not cover here, may be a better option for critically ill, hospitalized patients. Indeed, the lesson from the early reports and trials that have (understandably) focused on critically ill individuals may not necessarily be transferable to mild or moderate disease, or to prevention strategies, and we will learn more about the best place for therapies over time. Of note, the LPV/r arm of the large RECOVERY trial was closed at the end of June 2020 due to lack of benefit [41].

Ritonavir-boosted lopinavir at least demonstrates *in vitro* activity against SARS-COV-2 [12], unlike some of the other ARVs under investigation. The ARV effect of LPV/r is through its inhibition of HIV's aspartic protease enzyme and activity against SARS-CoV-2 is mediated through inhibition of its cysteine protease. However, the concentration of LPV/r required to inhibit SARS-CoV-2 is 4000- to 8000-fold higher that that required to inhibit HIV [13] so it perhaps entirely unsurprising that the dose that is effective in treating HIV may be ineffective for COVID-19. The penetration of drug into target sites is also crucial; the protein-adjusted the concentration of drug required for 90% inhibition (IC90) values of lopinavir required to inhibit SARS-CoV-2 replication in plasma, pulmonary epithelial lining fluid and cerebrospinal fluid are, respectively, 200-fold, 20-fold and 2000-fold higher than the concentrations measured *in vivo* in COVID-19 patients [13] – arguably the results of the LPV/r COVID-19 trials so far were predictable. LPV/r dose will be limited by toxicity and the higher concentrations of LPV described in COVID-19, compared with those observed in people on LPV/r for antiretroviral therapy (ART), are already significantly higher, probably due to the impact of coronavirus on liver function [14].

As the protease binding of LPV/r is probably less selective than more novel HIV protease inhibitors, one cannot assume all will have activity against non-HIV viruses. Following anecdotal reports of the efficacy of darunavir, another HIV protease inhibitor, its manufacturer Janssen released a statement confirming no *in vitro* activity against SARS-CoV-2 and no evidence of benefit in a small, single-arm study [15]. Despite this, on the ClinicalTrials.gov website alone, there are two trials investigating darunavir as a therapeutic option for COVID-19 (one recruiting and one pending) [5,6]. The HIV protease inhibitor atazanavir, listed as an experimental COVID-19 therapy on the Liverpool drug interaction website [16], does demonstrate *in vitro* activity [17] yet there are no trials listed on ClinicalTrials.gov investigating its potential for treatment or prevention of COVID-19.

Antiretrovirals for COVID-19 prevention

Effective prophylaxis against SARS-CoV-2, particularly for healthcare workers who are at higher risk of exposure, is desirable. Anecdotal reports of fewer cases of severe COVID-19 in Spain in HIV-positive people on ART has prompted a large, randomized trial in Spain investigating the use of tenofovir-disoproxil fumarate/emtricitabine (TDF/FTC) and low-dose hydroxychloroquine in Spanish healthcare workers [4]. A New York study suggesting lower than expected numbers of people with HIV amongst COVID-19 hospitalization has been interpreted by some as indicative of some degree of protection, but the difference was small and, without adjustment for confounders such as age, socio-economic status and social distancing practices, firm conclusions cannot be drawn [18].

To date, TDF has shown no efficacy, *in vitro* [12] or *in vivo*, against SARS-CoV-2. Molecular docking work suggests that TDF might be active due to its tight union to SARS-CoV-2 RdRp [19]. However, docking studies, though useful in pre-screening large numbers of compounds, are not a replacement for *in vitro* activity assessment [20]. Indeed, *in silico* evidence, of which docking is an example, sits lowest in the hierarchy of evidence topped by clinical evidence in COVID-19.

Tenofovir-disoproxil fumarate appears to have immunomodulatory properties; one *in vitro* study found that tenofovir altered inflammatory cytokine production, with reductions in interleukin-8 (IL-8), IL-10 and monocyte chemoattractant protein 1 (MCP-1) and increases in IL-12. While this may result in enhance pathogen-directed immune responses it may also be detrimental to patients with heightened COVID-19-induced inflammatory profiles [21]. The observation that there have been numerous

COVID-19 infections in HIV patients on TDF or tenofovir alafenamide (TAF) [22,23] at least speaks against a complete protection conferred by these agents. A cohort analysis of 60 Spanish hospitals deomstrated a lower risk of hospitalisation amongst people with HIV who were on TDF-based therapy compared to TAF-based and other regimens but residual confounding by comorbid conditions could not be completely excluded [24]. Clearly, the pre-exposure prophylaxis trial results must be awaited to shed light on the usefulness of this strategy.

Hydroxychloroquine for COVID-19 treatment

Although chloroquine (CQ) and hydroxychloroquine (HCQ) are not ARVs, they too were mooted as potential treatments for HIV, thanks to their immunomodulatory actions, with 'promising' pilot results [25] but no benefit (indeed a degree of harm) in the only randomized trial of sufficient duration [26]. The mechanism of any specific antiviral action of HCQ is unclear, but it is known to decrease endosome acidity, which might prevent release of virus into the cytoplasm [27]. HCQ has also been trialled for other viral diseases after favourable *in vitro* results, but never shown to be efficacious, and even deleterious in the case of Chikungunya [28]. Unfortunately, a small, poorly designed study with non-clinical endpoints [29] and media hysteria have led to HCQ and azithromycin (the drug it was partnered with) being the top two treatments used for COVID-19 in the 'COVID019 real-time barometer' study of over 20 000 physicians in 30 countries, with 55% reporting HCQ use at the time of writing [30]. That HCQ and azithromycin were rapidly incorporated into some COVID-19 treatment guidelines [31] would be brave enough if this drug combination were harmless, but concerns of cardiovascular toxicity, including an increased risk of cardiovascular mortality in a pre-print study [32], prompted the early termination of a CQ + azithromycin trial in Brazil [33], and led the U.S. Food and Drug Administration to warn about conduction abnormalities associated with HCQ or CQ and advise that clinicians should prioritize clinical trials if considering the use of these agents to treat COVID-19 [34]. Since then, a retrospective analysis of cardiovascular events associated with HCQ and azithromycin in the World Health Organization Pharmacovigilance Database, undertaken prior to their use for COVID-19, described a significant association with prolonged QT interval and ventricular tachyarrhythmia for each drug, which was even greater when both were given in combination [35].

The potential for harm, both direct toxicity and indirect in terms of drug shortages for people with established indications for these drugs [36], combined with lack of evidence of benefit in large observational studies with controls from the same population [37,38] surely means that CQ and HCQ should be used only within clinical trials? Of note, the latest findings, in over 1000 people with predominantly mild COVID-19, from the same Marseilles group who first espoused the apparent benefits of HCQ + azithromycin [39] is single-arm with no comparator at all, even to historic outcomes.

At the time of writing a large retrospective multinational analysis concluding lack of benefit of CQ or HCQ, alone or with a macrolide, and indeed decreased in-hospital survival and more ventricular arrhythmias [40] has been retracted due to serious methodological concerns [41].

HIV and COVID-19 may be very different conditions but the 'hypothesis > promising pilot > ineffective larger trial' cascade could have been anticipated and, arguably, had better designed trials been implemented earlier and the results of a flawed study not been promoted so widely by news outlets and political leaders, a potentially harmful treatment may not have gained such a firm foothold in some guidelines [31]. HCQ was one of the interventions under study in the large Recovery trial but, following an interim data review, the independent Data Monitoring Committee recommended that trial investigators review the unblinded data of the HCQ arm. Based on findings of no beneficial effect of HCQ in patients hospitalized with COVID-19, the HCQ arm was terminated immediately [42]. At the time of writing the Solidarity [43] and Discovery trials [44] trials, also investigating HCQ for COVID-19, were ongoing, the results of which are awaited eagerly.

Hydroxychloroquine for COVID-19 prevention

One published trial of HCQ post-exposure prophylaxis (PEP) showed no impact on subsequent SARS-CoV-2 acquisition in people reporting exposure, but more adverse events, compared with placebo [45] The excellent accompanying editorial flagged some important issues, including the optimal timing of PEP (probably earlier than the average of 3 days post-exposure in this trial) and the impact of these findings on the more than 200 trials investigating HCQ prophylaxis as of 1 June 2020 [46].

Conclusions

When dealing with a novel disease, we must not forget the hierarchy of evidence that should guide the interpretation of trials. In particular, drug modelling, as we have

outlined, may be a poor predictor of clinical effectiveness.

Perhaps we should follow the advice from those websites that share pre-publication research: 'Preprints are preliminary reports of work that have not been certified by peer review. They should not be relied on to guide clinical practice or health-related behaviour and should not be reported in news media as established information' [7,8]. While the peer review process may be flawed [47], it offers a buffer that filters out some of the more ludicrous COVID-19 assertions, and, in our view, continues to play an important role.

While a major pandemic understandably creates an urgent need for effective interventions, that does not mean the basic principles of clinical trials do not apply. Rapid assessment of *in vitro* drug activity, use of consistent consensus endpoints in case series and pilots, urgent modified peer review and prompt design of appropriately controlled trials will ultimately do a better service.

References

1 https://www.ecdc.europa.eu/en/geographical-distribution-2019-ncov-cases accessed 5th June 2020.

2 Rojek A, Horby P, Dunning J. Insights from clinical research completed during the west Africa Ebola virus disease epidemic. *Lancet Infect Dis* 2017; **17**: e280–e292.

3 Farha MA, Brown ED. Drug repurposing for antimicrobial discovery. *Nat Microbiol* 2019; **4**: 565–577.

4 ClinicalTrials.gov [Internet]. Bethesda (MD): National Library of Medicine (US). 2000 Feb 29. Identifier NCT04334928, Randomized Clinical Trial for the Prevention of SARS-CoV-2 Infection (COVID-19) in Healthcare Personnel (EPICOS); 2020 Apr 6 [cited 2020 Jun 12]; Available from: https://clinicaltrials.gov/ct2/show/NCT04334928.

5 ClinicalTrials.gov [Internet]. Bethesda (MD): National Library of Medicine (US). 2000 Feb 29. Identifier NCT04303299, Various Combination of Protease Inhibitors, Oseltamivir, Favipiravir, and Hydroxychloroquine for Treatment of COVID-19: A Randomized Control Trial (THDMS-COVID-19); 2020 Mar 11 [cited 2020 Jun12]; Available from: https://clinicaltrials.gov/ct2/show/NCT04303299.

6 ClinicalTrials.gov [Internet]. Bethesda (MD): National Library of Medicine (US). 2000 Feb 29. Identifier NCT04252274, Efficacy and Safety of Darunavir and Cobicistat for Treatment of COVID-19 (DC-COVID-19); 2020 Feb 5 [cited 2020 Jun 12]; Available from: https://clinicaltrials.gov/ct2/show/NCT04252274.

7 https://www.biorxiv.org/ accessed 10th May 2020.

8 https://www.medrxiv.org/ accessed 10th May 2020.

9 Ford N, Vitoria M, Rangaraj A, Norris SL, Calmy A, Doherty M. Systematic review of the efficacy and safety of antiretroviral drugs against SARS, MERS or COVID-19: initial assessment. *J Int AIDS Soc* 2020; **23**: e25489. https://doi.org/10.1002/jia2.25489.

10 Li Y, Xie Z, Lin W, Cai W, Wen C, Guan Y*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 preprint.

11 Cao B, Wang Y, Wen D *et al*. A trial of lopinavir-ritonavir in adults hospitalized with severe Covid-19. *N Engl J Med* 2020; **382**: 1787–1799. https://doi.org/10.1056/NEJMoa2001282.

12 Choy KT, Wong AY, Kaewpreedee P *et al*. Remdesivir, lopinavir, emetine, and homoharringtonine inhibit SARS-CoV-2 replication in vitro. *Antiviral Res* 2020; **3**: 104786.

13 Cattaneo D. Does lopinavir really inhibit SARS-CoV-2? *Pharmacol Res* 2020; **158**: 104898.

14 Baldelli S, Corbellino M, Clementi E, Cattaneo D, Gervasoni C. Lopinavir/ritonavir in COVID-19 patients: maybe yes, but at what dose? *J AntimicrobChemother* 2020 May 14 : dkaa190. https://doi.org/10.1093/jac/dkaa190.

15 https://www.jnj.com/lack-of-evidence-to-support-darunavir-based-hiv-treatments-for-coronavirus accessed 26th April 2020.

16 https://www.covid19-druginteractions.org/ accessed 12thJune 2020.

17 https://www.biorxiv.org/content/ 10.1101/2020.04.04.020925v1.full.pdf.

18 Richardson S, Hirsch JS, Narasimhan M *et al*. Presenting Characteristics, Comorbidities, and Outcomes Among 5700 Patients Hospitalized With COVID-19 in the New York City Area. *JAMA* 2020: e206775.**323**(20):2052–2059.

19 Elfiky Abdo A. Ribavirin, remdesivir, sofosbuvir, galidesivir, and Tenofovir against SARS-CoV-2 RNA dependent RNA polymerase (RdRp): A molecular docking study. *Life Sci* 2020; **253**: 117592.

20 Ramírez D, Caballero J. Is it reliable to use common molecular docking methods for comparing the binding affinities of enantiomer pairs for their protein target? *Int J Mol Sci* 2016; **17**: 525.

21 Melchjorsen J, Risør MW, Søgaard OS *et al*. Tenofovir selectively regulates production of inflammatory cytokines and shifts the IL-12/IL-10 balance in human primary cells. *J Acquir Immune DeficSyndr* 2011; **57**: 265–275.

22 Härter G, Spinner CD, Roider J *et al*. COVID-19 in people living with human immunodeficiency virus: a case series of 33 patients. *Infection* 2020;**11**: 1–6. https://doi.org/10.1007/s15010-020-01438-z

23 Gervasoni C, Meraviglia P, Riva A *et al*. Clinical features and outcomes of HIV patients with coronavirus disease 2019. *Clin Infect Dis*. 2020 May 14. pii: ciaa579. https://doi.org/10.1093/cid/ciaa579

24 Del Amo J, Polo R, Moerno S et al. Incidence and severity of COVID-19 in HIV-positive persons resceiving antiretroviral therapy: a cohort study (published online ahead, of print, 2020 Jun 26. Ann Intern Med. 2020;10.7326/M20-3689. https://doi.org/10.7326/M20-3689

25 Savarino A, Shytaj IL. Chloroquine and beyond: exploring anti-rheumatic drugs to reduce immune hyperactivation in HIV/AIDS. *Retrovirology* 2015; **12**: 51.

26 Paton NI, Goodall RL, Dunn DT et al. Hydroxychloroquine Trial Team: effects of hydroxychloroquine on immune activation and disease progression among HIV-infected patients not receiving antiretroviral therapy: a randomized controlled trial. *JAMA* 2012; **308**: 353–361.

27 Owens B. Excitement around hydroxychloroquine for treating COVID-19 causes challenges for rheumatology. *Lancet Rheumatol* 2020; **2**: e257.

28 De Lamballerie X, Boisson V, Reynier JC et al. On chikungunya acute infection and chloroquine treatment. *Vector Borne Zoonotic Dis* 2008; **8**: 837–9.

29 Gautret P, Lagier JC, Parola P et al. Hydroxychloroquine and azithromycin as a treatment of COVID-19: results of an open-label non-randomized clinical trial. *Int J Antimicrob Agents* 2020; 105949.

30 https://app.sermo.com/covid19-barometer?utm_campaign= wwwsermo_covid19 accessed 11th May 2020.

31 Decree No. 2020-314 of March 25, 2020 supplementing Decree No. 2020-293 of March 23, 2020 prescribing the general measures necessary to deal with the Covid-19 epidemic in the context of the state of health emergency. Accessed at https://beta.legifrance.gouv.fr/jorf/id/ JORFTEXT000041755775 12th May 2020.

32 BMJ Best Practice: https://bestpractice.bmj.com/topics/en-gb/ 3000168/emergingtxs#referencePop408 accessed 12th May 2020.

33 Borba MGS, Val FFA, Sampaio VS et al. Effect of high vs low doses of chloroquine diphosphate as adjunctive therapy for patients hospitalized with severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) infection: a randomized clinical Trial. *JAMA Netw Open* 2020; **3**: e208857.

34 https://www.fda.gov/drugs/drug-safety-and-availability/fda-cautions-against-use-hydroxychloroquine-or-chloroquine-covid-19-outside-hospital-setting-or accessed 11th May 2020.

35 Nguyen LS, Dolladille C, Drici MD et al. Cardiovascular Toxicities Associated With Hydroxychloroquine and Azithromycin: An Analysis of the World Health Organization Pharmacovigilance Database. *Circulation* 2020 doi: 10.1161/ CIRCULATIONAHA.120.048238. Online ahead of print.

36 Yazdany J, Kim AHJ. Use of Hydroxychloroquine and Chloroquine During the COVID-19 Pandemic: What Every Clinician Should Know. *Ann Intern Med* 2020; **172**: 754– 755. https://jamanetwork.com/channels/health-forum/fullartic le/2764607 accessed 11th May 2020

37 Geleris J, Sun Y, Platt J et al. Observational Study of Hydroxychloroquine in Hospitalized Patients with Covid-19. *N Engl J Med* 2020. https://doi.org/10.1056/NEJMoa2012410

38 Magagnoli J, Narendran S, Pereira F et al. Outcomes of hydroxychloroquine usage in United States veterans hospitalized with Covid-19. doi: 10.1101/ 2020.04.16.20065920 accessed 11th May 2020.

39 Million M, Lagier JC, Gautret P et al. Early treatment of COVID-19 patients with hydroxychloroquine and azithromycin: A retrospective analysis of 1061 cases in Marseille, France. doi: 10.1016/j.tmaid.2020.101738 accessed 11th May 2020.

40 Mehra MR, Desai SS, Ruschitzka F, Patel AN. Hydroxychloroquine or chloroquine with or without a macrolide for treatment of COVID-19: a multinational registry analysis. *Lancet* 2020; S0140–6736: 31180–6.

41 Mehra MR, Ruschitzka F, Patel AN. Retraction— Hydroxychloroquine or chloroquine with or without a macrolide for treatment of COVID-19: a multinational registry analysis. *Lancet* 2020; **395**: P1820.

42 https://www.recoverytrial.net/; accessed 30th June 2020.

43 https://www.who.int/emergencies/diseases/novel-coronavirus-2019/global-research-on-novel-coronavirus-2019-ncov/solida rity-clinical-trial-for-covid-19-treatments; accessed 9th June 2020.

44 https://clinicaltrials.gov/ct2/show/NCT04315948 accessed 09/ 06/2020.

45 Boulware DR, Pullen MF, Bangdiwala AS et al. A Randomized Trial of Hydroxychloroquine as Postexposure Prophylaxis for Covid-19. *N Engl J Med.* 2020. https://doi. org/10.1056/NEJMoa2016638

46 Cohen M. Hydroxychloroquine for the prevention of Covid-19 — searching for evidence. *N Engl J Med.* 2020. https://d oi.org/10.1056/NEJMe2020388

47 Bruce R, Chauvin A, Trinquart L, Ravaud P, Boutron I. Impact of interventions to improve the quality of peer review of biomedical journals: a systematic review and meta-analysis. *BMC Med.* 2016; **14**(1):85.