1 “Friend or Foe” — Oxygen Therapy for COVID-19 patients, a New Perspective

2
3 Shoaib Ashraf^a (BS (MLT), DVM, PhD), Muhammad Faisal Nadeem^b (BS (MLT),
4 PharmD, PhD), Muhammad Ahmad Imran^c (MBBS), Shafaat Yaar Khan^d, (PhD), Anwer
5 Hasil Kottarampatel^e, (PharmD, PhD), Saffa Khalid^f (MS2), Sagheer Ahmad^g (MBBS,
6 FCPS), Muhammad Bilal^h (DVM), Suzanne Samaraniⁱ (PhD), Rizwan Saifullah^j (BS
7 (MLT)), Sidra Ashraf^k (BS (MLT)), Moneeb Ashraf^l (MBBS), Sundas Rafique^m (MBBS),
8 Nazish Faisalⁿ (PharmD), Xin Zhao^h (PhD), Sohaib Ashraf^{o*} (MBBS), Muhammad
9 Ashraf^{p*} (DVM, PhD), and Ali Ahmad^{i,p*} (DVM, PhD).

10
11 ^aWellman Center for Photomedicine, Massachusetts General Hospital and Harvard
12 Medical School, Boston, Massachusetts, USA;

13 ^bInstitute of Pharmaceutical Sciences, University of Veterinary and Animal Sciences,
14 Lahore, Pakistan

15 ^cDepartment of Cardiology, Shaikh Zayed Hospital, Lahore, Pakistan;

16 ^dDepartment of Zoology, University of Sargodha, Sargodha, Pakistan;

17 ^eDepartment of Chemistry and Biological Sciences, University of Ottawa, Ottawa, Canada;

18 ^fSt George’s University, St. Georges, Grenada;

19 ^gPakistan Air Force Hospital, Jacobabad, Pakistan^f;

20 ^hDepartment of Animal Sciences, McGill University, Sainte-Anne-de-Bellevue, Quebec,
21 Canada;

22 ⁱCHU Sainte-Justine, University of Montreal, Canada;

23 ^jDepartment of Forensic Medicine and Toxicology Development, King Edward Medical
24 College University, Lahore, Pakistan;

25 ^kDepartment of Biochemistry, University of Veterinary and Animal Sciences, Lahore,
26 Pakistan

27 ^lDepartment of Pharmacology, Post Graduate Medical Institute, Lahore, Pakistan;

28 ^mDepartment of Medicine, King Edward Medical College University, Lahore, Pakistan;

29 ⁿDepartment of Pharmacy, Quaid-e-Azam University, Islamabad, Pakistan

30 ^oDepartment of Pharmacology & Toxicology, University of Veterinary and Animal
31 Sciences, Lahore, Pakistan

32 ^pDepartment of Microbiology, Infectious Diseases & Immunology, University of Montreal,
33 Montreal, Quebec, Canada.

***Correspondence:**

Sohaib Ashraf: sohaib-ashraf@outlook.com;

Muhammad Ashraf: drashraf2001@uvas.edu.pk;

Ali Ahmad: ali.ahmad@recherche-ste-justine.qc.ca;

Abstract

In late December 2019, a disease known as Coronavirus Disease (COVID-19), later known to be caused by a novel coronavirus, SARS-CoV-2, was first reported from Wuhan, China. In an unprecedented event, this highly contagious disease would become a global pandemic. The hospitalized patients are being treated mainly for a typical viral pneumonia with supplemental oxygen therapy and mechanical ventilation. This treatment ought to be helpful for COVID-19 patients, but the clinical outcomes are, thus far, not very promising. Therefore, there is an urgent need of new insights towards the pathophysiology of COVID-19 leading to the basis for better and more effective treatments. Recent research hints that SARS-CoV-2 may impair hemoglobin's ability to perform gaseous exchange, which leads to oxidative stress. In this complex situation, oxygen therapy could only be of limited utility and would rather aggravate the oxidative stress and its' downstream pathologies. We propose here that ideally these patients should be treated with exchange blood transfusions for immediate relief and resuscitation.

Main

SARS-CoV-2 — Structure

SARS-CoV-2 is an enveloped virus with a single positive sense 30 kb RNA. The viral genome contains 14 ORFs, which encode four structural proteins namely Spike (S), Envelope (E), Membrane (M) and Nucleocapsid (N); and twenty-seven accessory (non-structural) proteins that are required for viral replication and immune evasion [1]. The S protein binds the Angiotensin Converting Enzyme (ACE2), a carboxypeptidase expressed

on the surface of target cells (epithelial cells of the respiratory tract, intestine and distal glomerular ducts as well as on vascular endothelial cells). The binding initiates the infection process. The process also requires activation of the S protein by the type-II transmembrane serine protease (TMPRSS2) [2, 3]. Additionally, the virus was recently shown to bind CD147, also known as Basigin, a highly glycosylated multifunctional transmembrane protein belonging to the Immunoglobulin superfamily [4]. The virus was not able to infect certain target cells lacking CD147. CD147 activation is known to induce expression and activation of several matrix metalloproteases, interact with integrins on the cell surface, and promote homo- and heterotypic cell adhesions [5].

COVID-19 — Clinical Conundrum

COVID-19 seems to be a complex disease with varying pathology as the patients often present themselves with one or more of the following clinical symptoms: fever, dry cough, headache, myalgia, fatigue, dyspnoea, chills, diarrhoea, nausea, mental confusion and loss of smell and/or taste [6, 7]. There is no specific therapy for the disease, although a variety of antiviral, anti-parasitic, and other kinds of drugs are being tested. Hospitalized patients are mainly treated with oxygen therapy depending on their oxygen saturation levels. Oxygen therapy-refractory patients require mechanical ventilation and, in extreme cases, extra-corporeal membrane oxygenation. Despite these treatments, up to 15% of the critically ill patients die. With a staggering figure of ~4.0 million infections worldwide and an overwhelming of intensive care units (ICU) with COVID-19 patients, there is an urgent need to find novel and more effective treatment modalities. The hospitalized patients are treated as per guidelines set for viral pneumonia and acute respiratory distress syndrome (ARDS). The patients often show bilateral ground glass opacities in lungs in chest computed tomography (CT) scans. In the case of COVID-19, most of the patients are asymptomatic, which is clinically odd for a classical viral pneumonia. Furthermore, the patients with a typical viral pneumonia usually respond to oxygen therapy quite predictively. However, COVID-19 patients' response to supplemental oxygen has been quite discouraging. Consequently, up to 97% of the patients on invasive ventilation succumb to death [8]. Moreover, we do not see significant clinical findings apart from tachypnea presenting with hypocapnic hypoxia and end tidal CO₂ levels in the 1.4-2.0 kPa

range. These findings have challenged physicians across the globe as the clinical outcome of this disease remains grim.

Figure 1. A schematic presentation of a novel aspect of COVID-19 pathogenesis and its implications for treatment. Right: The patients are given oxygen therapy based upon

unreliable oximeter profiles that could cause oxidative stress, inflammation and tissue damage. Left: Patients' hemoglobin is compromised its affinity for oxygen as SARS CoV-2 and/or its proteins attack and remove iron from heme. This leads to iron overload and porphyria. Potential treatments are indicated in bold text in the Figure. For details, see text.

COVID-19 — Foiled

Classically when a patient comes to triage with respiratory distress, the nurse measures blood gas related indices including SpO₂ using pulse oximetry, which measures percentage of oxygen saturation in the blood. However, in the case of COVID-19, SpO₂ levels might not evidently indicate the patients' degree of deterioration, as pulse oximetry fails to take tissue oxygenation into account. Hence, body organs might still be in a hypoxic state despite administration of supplemental oxygen, even though the SpO₂ levels rise. Similar to pulse oximetry, parameters derived through arterial blood gas analysis could also be misleading. For instance, total O₂ content and SaO₂ can appear normal in plasma in the case of dyshemoglobinemias [9]. Similarly, anemic hypoxia due to possible hemoglobin breakdown will affect total O₂ content only, while PaO₂ and SaO₂ remain within normal ranges. In such circumstances, we propose that CO-oximetry (which is not being routinely used) could provide much better information regarding status of different hemoglobin types including oxyhemoglobin. However, as proposed in this special report, since SARS-CoV-2 may likely create a new hemoglobin variant (see below), CO-oximetry will also be of limited utility. Furthermore, raising FiO₂ settings in such cases to 100% might still not improve the respiratory difficulty, as oxygen alone will fail to make up for decreased hemoglobin levels and O₂ carrying capacity of the defective hemoglobin variant, for which we coin the term "Sarshemoglobin". We are currently performing experiments to investigate our proposed "Sarshemoglobinemia" model.

Oxygen Therapy — Pros and Cons

Real-time data suggest that oxygen therapy has biased after-effects and the outcomes are contradictory. Several anecdotal and published reports have raised concerns about the

treatment of COVID-19 patients as a “typical viral pneumonia”; where patients often require supplemental oxygen. Thus, we reason and emphasize that SARS-CoV-2 associated pneumonia or ARDS is quite atypical and is merely a piece of the COVID-19’s confusing puzzle. In Pakistan, we are experiencing that often patients having low oxygen saturation levels (~%80) show no respiratory distress. Furthermore, these COVID-19 patients present with a paradoxical feeling of calm and do not gasp for air. These levels, in routine viral pneumonia warrant oxygen therapy, which is not true for these patients.

For this reason, we along with many other emergency room physicians across the world describe COVID-19 more like a “high altitude sickness”. In such cases lungs are functioning normally and are compliant but their hypoxemia results from reduced oxygen supply (e.g., visit: <https://twitter.com/cameronks/status/1250798158948507648?lang=en>; <https://twitter.com/sarchotic93/status/1257346489841602567?s=19>). Conversely, in COVID-19 patients, reduced oxygen supply may be more due to the decreased oxygen-carrying capacity of hemoglobin rather than its availability in pulmonary alveoli. In our opinion, clinicians are rushing the patients too early to oxygen therapy and mechanical ventilation. We suggest that oxygen therapy should be given with great caution as it may act as a double-edged sword. While supplemental oxygen may give COVID-19 patients temporary relief, it can result in hyperoxemia and cause oxidative stress leading to a cascade of systemic events. Consequently, exposing the alveoli to excessive oxygen may induce production of dyshemoglobins via generation of reactive oxygen species and cause damage to airways and lungs. This could lead to the production of cytokines, chemokines and pro-coagulation mediators (so-called a cytokine storm), which could deteriorate the patient’s health further by promoting inflammation and widespread coagulopathies (Figure 1). Our hypothesis is derived from historical data, where cytokine storms and coagulopathies are leading pathologies that culminate into death of COVID-19 patients.

To this end, a pro-coagulation role of cytokines has also been well described [10] and recent studies report that pulmonary autopsies of terminally ill COVID-19 patients show presence of platelet-fibrin thrombi indicative of profound bleeding disturbances [11]. Thus, supplemental oxygen may aggravate respiratory distress and prompt mechanical

ventilation. High pressure intubations required by invasive ventilation would cause more damage than good in the patients and result in tracheal scarring and tissue damage. These hyperoxemia associated events may lead to systemic pathologies with poor prognosis.

COVID-19 Pathophysiology — Perplexity

The pathology of COVID-19 suggests that the complex disease is more than a viral pneumonia and is rather a systemic syndrome. A wide variety of additional clinical presentations manifested by COVID-19 patients supports this notion. The presentations include, but are not limited to, a hypercoagulable state causing myocardial infarction with or without obstructive coronary arteries, Guillain Barre Syndrome, syncopal episodes with normal chest X-ray, so-called ‘COVID toes’ and nervous system manifestations, etc. Interestingly, the biochemical laboratory findings of COVID-19 patients (decreased hemoglobin levels, and increased levels of heme, ferritin, bilirubin, CRP and ESR) suggest abnormalities in the blood compartment of the patients [12]. COVID-19 patients reveal abnormal erythrocyte morphology with decreased deformability and rapid clearance of erythrocytes [13]. In support of this, it was also demonstrated that macaques experimentally infected with SARS-CoV-2 have decreased numbers of RBCs in their circulation [14]. Moreover, the RBC distribution width (RDW) increases in COVID-19 patients due to their increased turnover. The RDW variation is a strong prognostic indicator for COVID-19 disease monitoring [15]. Together, these observations indicate defects in the erythroid compartment in COVID-19 patients that could adversely affect hemoglobin’s oxygen carrying capacity. Additionally, supplemental oxygen therapy may also aggravate the pathogenetic process by inducing hyperoxemia.

SARS-CoV-2 — Heme Interplay

Using in silico techniques, Liu and Li have recently shown that the viral proteins coordinately attack heme and hemoglobin and displace iron [16]. Despite its certain limitations, this is a provocative study and deserves due attention. It provides important insights in the pathogenesis of the COVID-19 syndrome. Heme forms the iron-containing prosthetic group of hemoglobin, is essential for the latter’s ability to bind oxygen and

carbon dioxide to maintain a physiological gaseous exchange [13]. By removing iron from hemoglobin's prosthetic group (heme), SARS-CoV-2 may impair hemoglobin's ability to bind O₂ in lungs and hence deliver it to tissues and organs (Figure 1). Although yet to be confirmed by experimental evidence, these observations are in line with clinical observations of COVID-19 and bear implications as to how these patients should be treated. It remains to be seen how Fe-deficient hemoglobin may affect oxygen saturation levels and oximeter readings. As discussed above, it may be showing lower than the actual saturation readings prompting clinicians to put the patients on oxygen therapy. Furthermore, the heme-invading properties of the viral proteins may result in the accumulation of porphyrin in body tissues mimicking acute porphyria. Porphyrins can cause internal organ damage by binding with and causing aggregation of a myriad of proteins even in the absence of light, resulting in an unfolded protein response and endoplasmic reticulum (ER) stress [17]. In fact, it has been suggested that COVID-19 patients suffering from moderate to severe form of viral pneumonia are experiencing a form of acquired acute porphyria [18].

SARS-CoV-2 — Erythrocyte Relationship

To attack and destroy hemoglobin, virus and/or viral proteins may enter RBCs, reticulocytes and/or their erythroid progenitors in the bone marrow. There is no evidence at this moment that virus targets and enters cells of the erythroid compartment. However, the erythroid progenitors do express ACE2 [19, 20] as well as CD147 [21]. Thus, we hypothesize that SARS-CoV-2 might be entering the RBCs, reticulocytes and/or their progenitor cells. It is noteworthy that *Plasmodium falciparum* merozoites use CD147 as a target to invade and enter RBCs. Binding of the parasite's reticulocyte-binding protein homologue (PfRh5) complex with CD147 is an essential step in this invasion process [22]. It would be very interesting to investigate whether SARS-CoV-2 can enter the erythroid cells via CD147. Another potential route for the viral entry into the erythroid compartment may be via Annexin-A2, a phospholipid binding protein expressed on the plasma membrane of many types of cells such as pulmonary epithelial cells [23]. Like other Corona viruses, SARS-CoV-2 is likely to incorporate it into its phospholipid bilayer [24].

The protein may mediate binding of the virus to the host cells including cells of the erythroid compartment. Post entry, the viral structural protein S may attack and remove iron from the prosthetic group of hemoglobin. Furthermore, reticulocytes and erythroid progenitors, unlike mature RBCs, contain cellular organelles and may be able to translate some viral proteins from the positive sense viral RNA. Alternately, viral proteins may be released from the infected epithelial cells into the circulation and may gain access to erythroid compartment and remove iron from their hemoglobin. Interestingly, CD147, the carrier of the Ok histo-blood group, plays an essential role in the normal circulation of RBCs in the body [5]. Binding of the SARS-CoV-2 to it via S protein may also cause abnormalities in the normal circulation pattern of RBCs in the body [25]. SARS-CoV-2 and/or its proteins may invade RBC compartment only to scavenge iron as RBC are inhospitable host to viruses. In fact, attempts were made in the past to lure viruses to RBC using the latter as viral traps [26].

Viral Assault — Implications

The observations made by Liu and Li [16] suggest that the infection with SARS-CoV-2 would result in iron overload and build-up of porphyrins. Thus, the virus has the potential to interfere with normal metabolic pathway of heme. Heme per se is pro-inflammatory and pro-oxidant. It is catabolized by the enzyme Heme Oxygenase (HO)-1 into Fe (which is recycled) and anti-inflammatory/antioxidant metabolites namely carbon monoxide (CO) and biliverdin/bilirubin [27]. As discussed above, the heme-invading properties of the viral proteins may result in the enhanced accumulation of porphyrins in body tissues. This may lead to the COVID-19 patients experiencing porphyria. The clinical picture of COVID-19 patients is suggestive of an imbalanced iron equilibrium which might result in iron overload (Figure 1). This may lead to significant abnormalities in their RBC compartment (as discussed above) that are suggestive of their increased turnover rate [13, 15, 18].

In the light of preceding discussion, we posit that COVID-19 patients' blood is compromised for its' ability to mediate normal gaseous exchange, and therefore, the

delivery of supplemental oxygen as a therapy would be of limited utility. Critically, this may rather be detrimental as it may cause oxidative stress, tissue damage and inflammation as mentioned above. Hence, under these circumstances, blood transfusions or exchange blood transfusions (to avoid circulatory overload) from histo-blood group compatible donors should be considered.

SARS-CoV-2 — Iron Acquisition

Clinical and experimental data suggest that SARS-CoV-2 acquires iron from the host, a possible survival mechanism for the virus. It has been well documented that iron is required for many vital biological processes in the body including cell proliferation and activation, mitochondrial electron transport chain and many biologically important oxido-reductive reactions. Not surprisingly iron metabolism is tightly regulated in both eukaryotic and prokaryotic cells [28]. Many viruses use a variety of different strategies to hijack cell's iron regulatory pathways to promote their own proliferation. They need it to enhance metabolism of the virus-infected cells [29]. In this regard, SARS-CoV-2 is not an exception. To this end, it is striking that malaria endemic countries have fewer cases of COVID-19 [30]. It is well known that malaria pathology is usually accompanied by iron deficiency and anemia. Another interesting observation is that males are more susceptible to SARS-COV-2 than females [31]. One of the reasons could be that females have relatively lower hemoglobin levels and hence lower iron concentrations as compared to males. Taken together, we can infer that SARS-CoV-2 replication seems to be iron dependant and has a special affinity for this element, which was recently shown to be the sole element responsible for the origin of life [32].

As stated above the binding of S protein with ACE2 receptors is facilitated by TMPRSS2, a member of the type II trans-membrane serine protease (TTSP). The expression of another member of the TTSP family, namely TMPRSS6, regulates iron homeostasis via cleaving and suppressing the activity of hepcidin, [33-36]. It would be very interesting to investigate possible impact of TMPRSS2 on iron homeostasis. In lieu of the above discussion, we hypothesize that the virus induced dysregulation of iron homeostasis may result in iron overload. This necessitates increased storage of iron in tissues and cells. Increased ferritin

(the molecule used to store iron in the body) concentrations in the circulation of the virus-infected patients support this notion [27]. The iron overload is likely to be a major contributor to the pathogenesis of the disease in SARS Cov-2 infected patients. It would result in inflammation and immunosuppression. In this regard, administration of iron chelators may be host beneficial. A few of them, for example Deferoxamine, are FDA approved. Bioinformatics studies show that Deferoxamine could also bind and inhibit the Papain-like protease of SARS-CoV-2 [37]. To this end, we also suggest that iron supplements should be taken with great caution as it may deteriorate the patients' health.

Conclusion — Succinite Insights

To conclude, we suggest caution in using supplemental oxygen therapy in hospitalized patients. Studies should be conducted on determining total levels of hemoglobin, its variants, and its oxygen carrying capacity as well as iron overload in hospitalized COVID-19 patients. This will provide novel knowledge-based more effective treatment options for these patients (indicated in bold text in Figure 1). Rather than oxygen therapy, exchange blood transfusions should be considered as an immediate support therapy in hospitalized COVID-19 patients.

REFERENCES:

1. Yang, Y., et al., *The deadly coronaviruses: The 2003 SARS pandemic and the 2020 novel coronavirus epidemic in China*. Journal of autoimmunity, 2020: p. 102434.
2. Wrapp, D., et al., *Cryo-EM structure of the 2019-nCoV spike in the prefusion conformation*. Science, 2020. **367**(6483): p. 1260-1263.
3. Hoffmann, M., et al., *SARS-CoV-2 cell entry depends on ACE2 and TMPRSS2 and is blocked by a clinically proven protease inhibitor*. Cell, 2020.
4. Wang, K., et al., *SARS-CoV-2 invades host cells via a novel route: CD147-spike protein*. bioRxiv, 2020.
5. Grass, G.D. and B.P. Toole, *How, with whom and when: an overview of CD147-mediated regulatory networks influencing matrix metalloproteinase activity*. Bioscience reports, 2016. **36**(1).
6. Guan, W.-j., et al., *Clinical characteristics of coronavirus disease 2019 in China*. New England journal of medicine, 2020. **382**(18): p. 1708-1720.
7. Yan, C.H., et al. *Association of chemosensory dysfunction and Covid-19 in patients presenting with influenza-like symptoms*. in *International Forum of Allergy & Rhinology*. 2020. Wiley Online Library.

- 316 8. Weiss, P. and D.R. Murdoch, *Clinical course and mortality risk of severe COVID-19*. The
317 Lancet, 2020. **395**(10229): p. 1014-1015.
- 318 9. Zander, R., *The oxygen status of arterial human blood*. Scandinavian Journal of Clinical
319 and Laboratory Investigation, 1990. **50**(sup203): p. 187-196.
- 320 10. Najem, M.Y., F. Couturaud, and C.A. Lemarié, *Cytokine and chemokine regulation of*
321 *venous thromboembolism*. Journal of Thrombosis and Haemostasis, 2020.
- 322 11. Carsana, L., et al., *Pulmonary post-mortem findings in a large series of COVID-19 cases*
323 *from Northern Italy*. medRxiv, 2020.
- 324 12. Chen, N., et al., *Epidemiological and clinical characteristics of 99 cases of 2019 novel*
325 *coronavirus pneumonia in Wuhan, China: a descriptive study*. The Lancet, 2020.
326 **395**(10223): p. 507-513.
- 327 13. Zini, G., et al., *Morphological anomalies of circulating blood cells in COVID-19*. American
328 Journal of Hematology, 2020.
- 329 14. Munster, V., et al., *Respiratory disease and virus shedding in rhesus macaques inoculated*
330 *with SARS-CoV-2*. BioRxiv, 2020.
- 331 15. Gong, J., et al., *A tool to early predict severe 2019-novel coronavirus pneumonia (COVID-*
332 *19): a multicenter study using the risk nomogram in Wuhan and Guangdong, China*.
333 medRxiv, 2020.
- 334 16. liu, w. and h. Li, *COVID-19: Attacks the 1-Beta Chain of Hemoglobin and Captures the*
335 *Porphyrin to Inhibit Human Heme Metabolism*. 2020.
- 336 17. Maitra, D., et al., *Porphyrin-Induced Protein Oxidation and Aggregation as a Mechanism*
337 *of Porphyria-Associated Cell Injury*. Cellular and molecular gastroenterology and
338 hepatology, 2019. **8**(4): p. 535.
- 339 18. Abrahams, L., *Covid-19: acquired acute porphyria hypothesis (edited)*. 2020.
- 340 19. Li, M., et al., *The SARS-CoV-2 receptor ACE2 expression of maternal-fetal interface and*
341 *fetal organs by single-cell transcriptome study*. PLoS One, 2020. **15**(4): p. e0230295.
- 342 20. Popescu, D.-M., et al., *Decoding human fetal liver haematopoiesis*. Nature, 2019.
343 **574**(7778): p. 365-371.
- 344 21. Odièvre, M.-H., et al., *Modulation of erythroid adhesion receptor expression by*
345 *hydroxyurea in children with sickle cell disease*. Haematologica, 2008. **93**(4): p. 502-510.
- 346 22. Volz, J.C., et al., *Essential role of the PfRh5/PfPrp/CyRPA complex during Plasmodium*
347 *falciparum invasion of erythrocytes*. Cell host & microbe, 2016. **20**(1): p. 60-71.
- 348 23. Taylor, J., J. Skeate, and W. Kast, *Annexin A2 in virus infection*. Front Microbiol 2018; 9:
349 2954. PUBMED.
- 350 24. Zheng, J., et al., *Identification of N-linked glycosylation sites in the spike protein and their*
351 *functional impact on the replication and infectivity of coronavirus infectious bronchitis*
352 *virus in cell culture*. Virology, 2018. **513**: p. 65-74.
- 353 25. Coste, I., et al., *Unavailability of CD147 leads to selective erythrocyte trapping in the*
354 *spleen*. Blood, The Journal of the American Society of Hematology, 2001. **97**(12): p. 3984-
355 3988.
- 356 26. Asher, D.R., A.M. Cerny, and R.W. Finberg, *The erythrocyte viral trap: transgenic*
357 *expression of viral receptor on erythrocytes attenuates coxsackievirus B infection*.
358 Proceedings of the National Academy of Sciences, 2005. **102**(36): p. 12897-12902.
- 359 27. Wu, B., Y. Wu, and W. Tang, *Heme Catabolic Pathway in Inflammation and Immune*
360 *Disorders*. Frontiers in pharmacology, 2019. **10**.
- 361 28. Yiannikourides, A. and G.O. Latunde-Dada, *A short review of iron metabolism and*
362 *pathophysiology of iron disorders*. Medicines, 2019. **6**(3): p. 85.

- 363 29. Drakesmith, H. and A. Prentice, *Viral infection and iron metabolism*. Nature Reviews
364 Microbiology, 2008. **6**(7): p. 541-552.
- 365 30. Goswami, R.P., D.K. Mittal, and R.P. Goswami, *Interaction between malarial transmission*
366 *and BCG vaccination with COVID-19 incidence in the world map: A changing landscape*
367 *human immune system?* medRxiv, 2020.
- 368 31. Jin, J.-M., et al., *Gender differences in patients with COVID-19: Focus on severity and*
369 *mortality*. Frontiers in Public Health, 2020. **8**: p. 152.
- 370 32. Muchowska, K.B., S.J. Varma, and J. Moran, *Synthesis and breakdown of universal*
371 *metabolic precursors promoted by iron*. Nature, 2019. **569**(7754): p. 104-107.
- 372 33. Antalis, T.M., et al., *The cutting edge: membrane-anchored serine protease activities in*
373 *the pericellular microenvironment*. Biochemical Journal, 2010. **428**(3): p. 325-346.
- 374 34. Casu, C., et al., *Combination of Tmprss6-ASO and the iron chelator deferiprone improves*
375 *erythropoiesis and reduces iron overload in a mouse model of beta-thalassemia*
376 *intermedia*. Haematologica, 2016. **101**(1): p. e8.
- 377 35. Gonçalves, J.L., et al., *Iron intake is positively associated with viral load in antiretroviral*
378 *naïve Brazilian men living with HIV*. Memórias do Instituto Oswaldo Cruz, 2019. **114**.
- 379 36. Meyer, D., et al., *Identification of the first synthetic inhibitors of the type II transmembrane*
380 *serine protease TMPRSS2 suitable for inhibition of influenza virus activation*. Biochemical
381 Journal, 2013. **452**(2): p. 331-343.
- 382 37. Contini, A., *Virtual screening of an FDA approved drugs database on two COVID-19*
383 *coronavirus proteins*. 2020.

384