Tissue Engineering

DOI: 10.1089/ten.TEA.2020.0094

The Role of Tissue Engineering in COVID-19 and Future Viral Outbreaks

Alexander M. Tatara, M.D., Ph.D.[1*]

[1] Department of Internal Medicine, Massachusetts General Hospital, Boston, Massachusetts

* Corresponding author

Corresponding author: Alexander M. Tatara

Massachusetts General Hospital

Department of Medicine

55 Fruit Street

Boston, Massachusetts 02114

tel: 617-724-8400

fax: 627-724-0331

e-mail: atatara@partners.org

Keywords: Tissue engineering, biomaterials, pandemic, coronavirus, viral

Abstract

In light of the current novel coronavirus (COVID-19) pandemic as well as other viral outbreaks in the 21st century, there is a dire need for new diagnostic and therapeutic strategies to combat infectious diseases worldwide. As a convergence science, tissue engineering has traditionally focused on application of engineering principles to biological systems, collaboration across disciplines, and rapid translation of technologies from the benchtop to the bedside. Given these strengths, tissue engineers are particularly well-suited to apply their skillset to the current crisis and viral outbreaks in general. This work introduces the basics of virology and epidemiology for tissue engineers and highlights important developments in the field of tissue engineering relevant to the current pandemic, including in vitro model systems, vaccine technology, and small molecule drug delivery. COVID-19 serves as a call to arms for scientists across all disciplines and tissue engineers are well-trained to be leaders and contributors in this time of need.

Impact Statement

Given the steep mortality caused by the recent novel coronavirus (COVID-19) pandemic, there is clear need for advances in diagnostics and therapeutics for viral outbreaks. Tissue engineering has the potential for critical impact on clinical outcomes in viral outbreaks. Tissue engineers, if mobilized, could play key roles as leaders in the outbreak given their ability to apply engineering principles to biological processes, experience in collaborative environments, and penchant for technological translation from benchtop to bedside. In this work, three areas pioneered by tissue engineers that could be applied to the current COVID-19 crisis and future viral outbreaks are highlighted.

Introduction

As of April 2020, the world is facing a pandemic of unfathomable proportions. Per the World Health Organization (WHO) on April 9th, over 1,500,000 patients worldwide have been diagnosed with the novel coronavirus disease 2019 (COVID-2019) caused by a laboratory-confirmed infection of the Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) with more than 84,000 global deaths at this time (1). Our hospital systems are rapidly filling with patients suffering from viral illness and the capacity of resources such as emergency departments, inpatient wards, and intensive care units has become overwhelmed in some regions. There is a dire need for new diagnostic and therapeutic modalities. Early diagnosis is critical in establishing quarantine and limiting the spread of outbreaks; diagnostics later in the course of an epidemic remain important particularly in determining established immunity. For those infected, there is a current paucity in validated anti-viral therapies and no vaccine at this time, although multiple efforts are underway. Even after the predicted resolution of COVID-19, the increasing frequency of viral outbreaks (including the 2003 Severe Acute Respiratory Syndrome Coronavirus (2), 2014 Ebola Virus (3), 2015 Middle Eastern Respiratory Syndrome Coronavirus (4), and 2015 Zika Virus outbreaks (5)) suggests that new mechanisms to combat viral infections are a high priority.

As a convergence science, tissue engineering is uniquely suited to offer solutions to complex clinical questions. Currently, tissue engineering-based technologies are being developed to revolutionize areas in medicine such as high throughput drug discovery (6), personalized cancer therapy (7), immune modulation (8), and organ transplantation (9). Tissue engineers specialize in the application of engineering principles to biological systems which facilitates the generation of fundamental knowledge as well as new technologies which could be key in a pandemic. In addition, tissue engineers are well-versed in collaborative models (10), working closely alongside clinicians, biologists, chemists, physicists, mathematicians, veterinarians, and other specialists which will be critical in a multidisciplinary approach to combating the virus known as SARS-CoV-2. Lastly, tissue engineering as a field has emphasized clinical translation, including creating workflows to optimize bringing the benchtop to the bedside (11) resulting in a 9 billion dollar market with 21 companies selling tissue engineering-based products in the United

States alone as of 2017 (12). In the current setting of limited clinical data and rising patient morbidity and mortality, tissue engineers would be a welcomed and valuable ally in the COVID-19 pandemic.

In this work, the potential impact of tissue engineering in improving clinical outcomes during the COVID-19 pandemic and future viral epidemics is explored (Fig. 1). Relevant background information regarding SARS-CoV-2 is briefly reviewed and pertinent tissue engineering work is highlighted, including development of viral in vitro models, drug delivery systems, and vaccine platforms. Given the current state of the pandemic, it may be challenging to mobilize new efforts within the tissue engineering community in time to change practice in healthcare. New in vitro models, diagnostics, and therapeutics will need to be validated prior to safe implementation at the clinical level. In addition, many major academic centers are currently at limited capacity due to precautions to limit the spread of the pandemic. However, COVID-19 can act as a representative outbreak for which tissue engineers can learn from and begin preparations to lead the way to better prevent and treat the next viral epidemic.

Our current understanding of SARS-CoV-2 is evolving and incomplete; the following information is based on current evidence and will likely change as the virus is more closely studied. Given the urgency of the pandemic, some of the references in this work have yet to receive peer review and should be interpreted with caution.

SARS-CoV-2 Background

Coronaviruses, or *Orthocoronavirinae*, are enveloped single-stranded RNA viruses. The virus gets its namesake through projections, or “spikes,” emerging from its envelope that appeared crown-like on electron micrography. The major components of SARS-CoV-2 are the envelope protein (E), membrane glycoprotein (M), spike protein (S), nucleocapsid protein (N), and its relatively large RNA genome of ~30 kb (13, 14). Enveloped viruses have a protective lipid bilayer with surface proteins and are generally more vulnerable to harsh environments than non-enveloped viruses. The spike protein is supposed to interact with human angiotensin-converting enzyme 2 (ACE2) membrane protein to induce fusion, endocytosis, and subsequent invasion into the host cell (15). Coronaviruses escape endosomes to the cytoplasm via acid-dependent cleavage of the S protein (14). RNA viruses, with few exceptions, replicate in the cytoplasm. As a positive-stranded virus, SARS-

CoV-2 is not dependent on RNA polymerase for transcription. The virus takes advantage of host ribosomes to replicate by translation and then assembly in the endoplasmic reticulum directed by M and E protein interactions. Viruses are then released by exocytosis to repeat the cycle of infection. Given this pathophysiology, potential targets being explored as therapeutics agents include blocking ACE2 interactions, altering endosome pH to prevent escape, inhibiting viral and/or host enzymes critical to replication, and downregulating host inflammation given that an overexuberant response may lead to acute respiratory distress syndrome (ARDS)(16).

SARS-CoV-2 has highest viral burden in the nares rather than throat and is thought to be spread during coughing and aerosolization of droplets (17). Compared to SARS-CoV-1, spread of the virus appears to be more rapid due to higher asymptomatic carrier rates and longer incubation time prior to symptom onset (18). Initial symptoms commonly include fever, cough, and fatigue and less commonly gastrointestinal manifestations (19). Current biomarkers suggestive of infection include elevated lactate dehydrogenase, ferritin, D-dimer, erythrocyte sedimentation rate, C-reactive protein, and an absolute lymphopenia (20). In the United States, the most widely available diagnostic test is polymerase chain reaction (PCR)-based although antibody-based assays are in development and are available in other countries (21). The virus primarily affects the lungs, causing acute respiratory distress syndrome (ARDS) in up to 5-10% of infected patients (22, 23) although has also been causing myocardial injury suspected to be due to high concentrations of ACE2 in cardiac tissue (15). Mortality is estimated to be 3-4% (18). Given the extreme global morbidity and mortality caused by the pandemic, there is a dire need for a better understanding of molecular mechanisms of host-virus interaction, more rapid methods to screen potential therapeutics, and platforms to facilitate safe clinical translation; these are all areas in which tissue engineers are primed to make significant contributions for COVID-19, the next coronavirus epidemic, or other future viral outbreaks.

In Vitro Models

Development of physiologically representative in vitro models of viral disease can assist in two critical roles during a pandemic: 1) better characterization and understanding of the host-pathogen interface and mechanisms of infection; and 2) as platform for high throughput screening of potential therapeutics. Currently, it is challenging and clinically

necessary to predict the course of patients infected with SARS-CoV-2. Profiling biomarkers may allow for better risk stratification and resource allotment (24); more accurate in vitro modeling to understand host response as well as viral mechanisms for transmission and replication is critical for the identification of biomarkers to pursue as hypothesis-driven diagnostic and therapeutic targets. In addition, a physiologically relevant in vitro model has the potential to fill the critical need for improved drug candidate screening.

The current gold standard for screening antiviral therapeutics is based on static monolayer culture of Vero cells, an interferon-deficient aneuploid line of kidney epithelial cells originally isolated from an African green monkey (25, 26). In a recent study, Vero cells were exposed to a library of 3,000 FDA- and IND-approved drugs and then infected with SARS-CoV-2 to screen for potential therapeutics (27). Because Vero cells lack interferon, they are highly susceptible to viruses and allow for replication so have been an attractive vehicle to screen compounds. However, interferon itself is an important regulator of host binding proteins involved in SARS-CoV-2 (28). In general, the relevance of a drug's ability to inhibit viral infection of a malignant non-human primate kidney cell lacking interferon production is uncertain. For example, while the anti-depressant sertraline was found to have potent in vitro activity against Ebola in the Vero cell model (26), later testing in an in vivo non-human primate model failed to show protection against Ebola (29). Static cultures of xenograft monolayers fail to replicate many of the conditions facing viruses in vivo, including realistic extracellular matrix (ECM), three-dimensional cell-cell interfaces, and shear forces. Coronaviruses and other respiratory viruses, for example, can bind to the ECM components such as sialic acid to assist in infection of the host (30).

In the last decade, tissue engineering has made significant advances regarding *in vitro* human cell culture models. Developments of induced pluripotent stem cells, CRISPR-Cas, microfluidics, 3D printing, and biomaterials have led to technologies such as tissue-on-a-chip and advanced bioreactor models containing co-cultures of cells from ectodermal, mesodermal, and endodermal lineages. These models, utilizing human cells, have been able to mimic complex pathophysiology such as generation of pulmonary edema upon exposure to inflammatory signals such as interleukin-2 (31). For example, in a model to study influenza A virus, 3D tissue-engineered constructs more accurately recapitulated host morphology of cultured human epithelial airway cells compared to 2D culture and

infection with major influenza strains resulted in upregulation of proinflammatory cytokines (32). One coronavirus-specific example of a tissue-engineered platform in which respiratory viruses have been studied is the rotation wall vessel bioreactor. These models simulate low physiologic shear stresses and frequently incorporate multiple pulmonary cell types, including co-culture of human mesenchymal bronchial tracheal cells and human bronchial epithelial cells and challenging them against respiratory syncytial virus and SARS-CoV-1 (33). Unfortunately, SARS-CoV-1 did not replicate in this study (34). In another model, human pulmonary epithelial progenitor cells were grown on a collagen matrix in a serum-free media with a mesenchymal stroma and exposed to virus. It was demonstrated that stem cells were targeted by SARS-CoV-1 which may suggest why normal lung regeneration following viral infection is challenging (35).

There are a number o f exciting tissue-engineered human in vitro lung models currently available that could be leveraged for studying viral infection (36, 37) and established in vivo models for respiratory viruses, including non-human primate, already in use (38-40). Elements that may improve the relevance of in vitro models include: 1) human rather than animal cell lines; 2) co-culture of multiple pulmonary cell lines; 3) 3D scaffolds that mimic native pulmonary architecture; and 4) culture methods that permit generation of extracellular matrix prior to viral inoculation. Additional head-to-head studies will need to be performed to determine if these components are necessary to capture the pathophysiology of viral infection. These platforms will be important in conducting hypothesis-driven research to understand the host-pathogen interface. Scaling these models to allow for high throughput drug screening will offer important advantages over the current Vero-based methods to rapidly identify therapeutic candidates for in vivo translation during the major infectious outbreak.

Drug Delivery Systems

During the COVID-19 pandemic, one of the most significant strains on the healthcare system has been the need for inpatient beds both on general wards and in intensive care units (ICUs) (41). As discussed previously regarding in vitro drug screens, there are currently limited therapeutics that have clear clinical evidence of improving outcomes such as days of hospitalization required, need for ICU stay, and need for intubation/ventilation. As new small molecule-based therapies come through the pipeline,

tissue engineers can continue to design drug delivery systems to 1) target medications to specific organ systems to increase bioavailability and 2) extend release of medications such that frequent administration is not necessary.

Classes of molecules that have been suggested as possible therapies include repurposed small molecule drugs, monoclonal antibodies, and oligonucleotide strategies (42). For example, based off of a small clinical study (43), the combination of hydroxychloroquine (a small molecule traditionally used to treat malaria and lupus) and azithromycin (a macrolide antibiotic with anti-inflammatory properties) has been suggested as a means to reduce SARS-CoV-2 viral load although these results are controversial (44). Poly(lactide) and poly(lactide-co-glycolide) (PLGA) microparticles can deliver azithromycin for up to 60 days with zero order release kinetics (45). There are also formulations using fumaryl diketopiperazine microparticles to deliver azithromycin via intratracheal insufflation directly to the lungs, resulting in higher local concentrations in a murine pneumonia model compared to oral and intravenous routes of delivery (46). Given that the half-life of hydroxychloroquine is greater than forty days, extended drug delivery options may not be warranted (47); however, there have been successful delivery systems constructed from PLGA nanoparticles for specific cell targeting (48). While the efficacy of hydroxychloroquine and azithromycin in the prevention and treatment of SARS-CoV-2 remains an area of active study, the above examples serve to show that tissue engineers and biomaterials scientists have been working on drug delivery for decades with a variety of vehicles available for small molecules in general.

In addition to small molecules, monoclonal antibodies are an exciting class of medication given their success in the treatment of Ebola virus (49). Antibodies are a natural part of humoral immunity and can be engineered to block specific ligands or receptors vital for viral function. These therapies generally need to be delivered intravenously to be successful; for the Ebola virus studies, for example, patients required one to three infusion sessions depending on the antibody. A human monoclonal antibody was developed against SARS-CoV-1 and demonstrated to be effective in a ferret model (38). Researchers have screened monoclonal antibodies designed against SARS-CoV-1 and have discovered cross-reactivity of at least one of the antibodies against SARS-CoV-2 (50). Systems designs for controlled extended release of antibodies may be advantageous over

multiple infusion sessions for clinical practice. Nanoporous scaffolds coated with allylamine-based polymer were capable of releasing rituximab, a monoclonal antibody against B cells, for up to thirty days (51). Similarly, an alginate-based drug delivery system was able to deliver a human IgG1 monoclonal antibody in a rat model for at least 28 days with a single dose of the system (52). In addition to the possibility of reducing administration to single dosing by extended release, there has also been development of ingestible injection systems to deliver biomacromolecules through autoinjection during gastric transit- these allowed for insulin delivery in a porcine model and may facilitate oral delivery of medications previously only efficacious in intravenous forms (53).

Lastly, short interfering RNA (siRNA) has also been explored as both prophylaxis and therapy for coronavirus infection (54). For sequences specific to SARS-CoV-1, siRNA was effective in a non-human primate model (39), resulting in diminished viral load and alveolar damage. Four intranasal doses were required over five days in treatment arms. The researchers reported not using additional vehicles to deliver their siRNA such as polyethyleneimine due to possibility of carrier-induced lung inflammation. However, more complex vehicles have since been developed by the field specifically for pulmonary usage, including mesoporous silica nanoparticles (55) and cationic liposomes (56). As siRNA sequences, specific antibodies, and small molecules are identified that specifically mitigate SARS-CoV-2, tissue engineers and biomaterials scientists can continue their work in designing vehicles to specifically and extendedly target areas of high viral load. Even if this work may not come to fruition during the current pandemic, these vehicles may serve vital roles during future viral outbreaks.

Vaccine Platforms

The ability to vaccinate against specific pathogens has played a major role in preventative medicine for the last century. Vaccines exist for both respiratory viruses as such as *Influenzavirus* and bacteria such as *Streptococcus pneumoniae* and *Haemophilus influenzae*. While the success of respiratory viral vaccines varies from season to season, data suggests influenza vaccination generally results in lower probability of complications including ICU stay, mechanical ventilation, and severe outcomes, especially in patients with co-morbidities such as chronic obstructive pulmonary disease (57-59). Work is already

underway to develop vaccines effective against SARS-CoV-2 to prevent disease and mitigate transmission (60).

Successful vaccination against pathogens relies on presenting antigens and stimulating specific elements of the immune system to build recognition and memory in both humoral and cell-mediated branches. With advances in immunology, biomaterials and tissue engineering are being leveraged to elicit specific host immune responses to augment vaccination strategies (8, 61). In many of these systems, the biomaterial acts as both the drug delivery vehicle for the vaccine as well as the adjuvant. Furthermore, the field is characterizing how the size, shape, and other physicochemical properties of biomaterials affects the behavior of immune cells (62-64). The goal of many of these systems is to target antigen-presenting cells such as macrophages and dendritic cells and drive specific responses such as Th1 or Th2 (different helper T classes). For example, conjugation of different receptors to protein-based particles can individually tune Th1 or Th2 response in a murine model (65). Biodegradable polymers, one of the primary workhorses as scaffold material in tissue engineering, have also been explored. PLGA nanoparticles drive Th1 immune response compared to no carriers and other biomaterials in a vaccine against Chagas disease in a murine model (66) and have also been explored for targeted delivery to specific immune populations (67). PLGA microparticles in combination with a chitosan/peptide conjugate coating have also been used as a delivery system to target specific mucosal cells and deliver a swine dysentery vaccine with elevated IgA and IgG production in mice (68). In another murine model, chitosan nanoparticles enhanced T-cell response for a *Mycobacterium tuberculosis* DNA vaccine (69). When chitosan was mannosylated to promote endocytosis, an intranasal vaccine increased IgG levels in a murine model (70). For influenza, chitosan has also been modified to create a thermoresponsive intranasal murine vaccine against H5N1l (71). Silver nanoparticles have also been used to locally deliver inactivated influenza vaccine with some specificity to lung immune cells, resulting in greater IgG titers and less mortality in a murine model (40). While there are currently less studies regarding coronavirus vaccines, there was some success in mice in which nanoparticles were prepared from a SARS-CoV-1 peptide sequence and subsequent sera was successful in preventing infection of Vero cells (72).

In addition to various particle-based platforms, tissue engineering strategies have been harnessed to create scaffold systems for vaccination enhancement. With specific physicochemical properties, such as pore size, and profile of released recruitment signals like granulocyte-colony stimulating factor, scaffolds of PLGA (73, 74) and mesoporous silica rods (64, 75) have been used to recruit and concentrate antigen-presenting cells to vaccine components. This strategy has primarily been applied to tumor vaccines and has demonstrated efficacy in animal models against melanoma and intracranial gliomas (73, 76); it is currently undergoing a phase I clinical trial of 23 patients with melanoma which is estimated to complete in June 2020 (77). This platform of scaffold-based vaccination has also shown efficacy against bacterial pathogens in porcine and murine models (78). Other examples of scaffold-based vaccine systems include those generated from respiratory syncytial virus that were effective in mice as well non-human primates (79, 80). These particle-based and scaffold-based vaccine systems may be promising in translation against SARS-CoV-2; however, the majority have only been studied in mouse models at this point in time and significant translational efforts will need to be undertaken for clinical trials. Modular platforms in which different antigens can be plugged in (78) may be very useful for rapid vaccine development in future pandemics.

Conclusion

The world faces a global healthcare crisis of unheralded magnitude. The rate of infection and mortality from COVID-19 make it unlike any virus seen in this century. Physicians and scientists are banding together to combat the threat of SAR-CoV-2. Tissue engineers have a rare set of tools and can make substantial contributions to our understanding of viral disease and contribute towards critical development of diagnostic and therapeutic platforms. Together, we can overcome this current pandemic and work to prevent and mitigate future viral outbreaks in the future.

Acknowledgements

I would like to thank my co-residents at the Massachusetts General Hospital for their selfless service on the front lines in the treatment of patients in this pandemic.

Author Disclosure Statement:

No competing financial interests exist.

References

1. Coronavirus disease (COVID-19) Pandemic. World Health Organization; 2020.

2. Bell, D.M., World Health Organization Working Group on, I., and Community Transmission of, S. Public health interventions and SARS spread, 2003. Emerg Infect Dis **10**, 1900, 2004.

3. Gomes, M.F., y Piontti, A.P., Rossi, L., Chao, D., Longini, I., Halloran, M.E., and Vespignani, A. Assessing the international spreading risk associated with the 2014 West African Ebola outbreak. PLoS currents **6** 2014.

4. Cho, S.Y., Kang, J.M., Ha, Y.E., Park, G.E., Lee, J.Y., Ko, J.H., Lee, J.Y., Kim, J.M., Kang, C.I., Jo, I.J., Ryu, J.G., Choi, J.R., Kim, S., Huh, H.J., Ki, C.S., Kang, E.S., Peck, K.R., Dhong, H.J., Song, J.H., Chung, D.R., and Kim, Y.J. MERS-CoV outbreak following a single patient exposure in an emergency room in South Korea: an epidemiological outbreak study. Lancet **388**, 994, 2016.

5. Chang, C., Ortiz, K., Ansari, A., and Gershwin, M.E. The Zika outbreak of the 21st century. Journal of autoimmunity **68**, 1, 2016.

6. Mazzocchi, A., Soker, S., and Skardal, A. 3D bioprinting for high-throughput screening: Drug screening, disease modeling, and precision medicine applications. Applied Physics Reviews **6**, 011302, 2019.

7. Fong, E.L., Wan, X., Yang, J., Morgado, M., Mikos, A.G., Harrington, D.A., Navone, N.M., and Farach-Carson, M.C. A 3D in vitro model of patient-derived prostate cancer xenograft for controlled interrogation of in vivo tumor-stromal interactions. Biomaterials **77**, 164, 2016.

8. Vishwakarma, A., Bhise, N.S., Evangelista, M.B., Rouwkema, J., Dokmeci, M.R., Ghaemmaghami, A.M., Vrana, N.E., and Khademhosseini, A. Engineering Immunomodulatory Biomaterials To Tune the Inflammatory Response. Trends Biotechnol **34**, 470, 2016.

9. Khademhosseini, A., and Langer, R. A decade of progress in tissue engineering. Nature protocols **11**, 1775, 2016.

10. Fong, E.L., Watson, B.M., Kasper, F.K., and Mikos, A.G. Building bridges: leveraging interdisciplinary collaborations in the development of biomaterials to meet clinical needs. Advanced Materials **24**, 4995, 2012.

11. Tatara, A.M., Ratcliffe, A., Wong, M.E., Kasper, F.K., and Mikos, A.G. Biomaterials in Regenerative Medicine: Considerations in Early Process Development. Translational Regenerative Medicine: Elsevier; 2015. pp. 141.

12. Kim, Y.S., Smoak, M.M., Melchiorri, A.J., and Mikos, A.G. An overview of the tissue engineering market in the United States from 2011 to 2018. Tissue Engineering Part A **25**, 1, 2019.

13. Chatterjee, S. Understanding the Nature of Variations in Structural Sequences coding for Coronavirus Spike, Envelope, Membrane and Nucleocapsid Proteins of SARS-CoV-2. Envelope, Membrane and Nucleocapsid Proteins of SARS-CoV-2 (March 28, 2020) 2020.

14. Fehr, A.R., and Perlman, S. Coronaviruses: an overview of their replication and pathogenesis. Coronaviruses: Springer; 2015. pp. 1.

15. Zheng, Y.-Y., Ma, Y.-T., Zhang, J.-Y., and Xie, X. COVID-19 and the cardiovascular system. Nature Reviews Cardiology, 1, 2020.

16. Guo, Y.-R., Cao, Q.-D., Hong, Z.-S., Tan, Y.-Y., Chen, S.-D., Jin, H.-J., Tan, K.-S., Wang, D.-Y., and Yan, Y. The origin, transmission and clinical therapies on coronavirus disease 2019 (COVID-19) outbreak–an update on the status. Military Medical Research **7**, 1, 2020.

17. Zou, L., Ruan, F., Huang, M., Liang, L., Huang, H., Hong, Z., Yu, J., Kang, M., Song, Y., and Xia, J. SARS-CoV-2 viral load in upper respiratory specimens of infected patients. New England Journal of Medicine **382**, 1177, 2020.

18. Pedersen, S.F., and Ho, Y.-C. SARS-CoV-2: A Storm is Raging. The Journal of Clinical Investigation 2020.

19. Zhang, J.j., Dong, X., Cao, Y.Y., Yuan, Y.d., Yang, Y.b., Yan, Y.q., Akdis, C.A., and Gao, Y.d. Clinical characteristics of 140 patients infected by SARS-CoV-2 in Wuhan, China. Allergy 2020.

20. Lippi, G., and Plebani, M. Laboratory abnormalities in patients with COVID-2019 infection. Clinical Chemistry and Laboratory Medicine (CCLM), 20200198, 2020.

21. To, K.K.-W., Tsang, O.T.-Y., Leung, W.-S., Tam, A.R., Wu, T.-C., Lung, D.C., Yip, C.C.-Y., Cai, J.-P., Chan, J.M.-C., and Chik, T.S.-H. Temporal profiles of viral load in posterior oropharyngeal saliva samples and serum antibody responses during infection by SARS-CoV-2: an observational cohort study. The Lancet Infectious Diseases 2020.

22. Yang, X., Yu, Y., Xu, J., Shu, H., Liu, H., Wu, Y., Zhang, L., Yu, Z., Fang, M., and Yu, T. Clinical course and outcomes of critically ill patients with SARS-CoV-2 pneumonia in Wuhan, China: a single-centered, retrospective, observational study. The Lancet Respiratory Medicine 2020.

23. Remuzzi, A., and Remuzzi, G. COVID-19 and Italy: what next? The Lancet 2020.

24. Xie, J., Hungerford, D., Chen, H., Abrams, S.T., Li, S., Wang, G., Wang, Y., Kang, H., Bonnett, L., and Zheng, R. Development and external validation of a prognostic multivariable model on admission for hospitalized patients with COVID-19. medRxiv 2020.

25. Adcock, R.S., Chu, Y.K., Golden, J.E., and Chung, D.H. Evaluation of anti-Zika virus activities of broad-spectrum antivirals and NIH clinical collection compounds using a cell-based, high-throughput screen assay. Antiviral Res **138**, 47, 2017.

26. Johansen, L.M., DeWald, L.E., Shoemaker, C.J., Hoffstrom, B.G., Lear-Rooney, C.M., Stossel, A., Nelson, E., Delos, S.E., Simmons, J.A., and Grenier, J.M. A screen of approved drugs and molecular probes identifies therapeutics with anti–Ebola virus activity. Science translational medicine **7**, 290ra89, 2015.

27. Jeon, S., Ko, M., Lee, J., Choi, I., Byun, S.Y., Park, S., Shum, D., and Kim, S. Identification of antiviral drug candidates against SARS-CoV-2 from FDA-approved drugs. bioRxiv 2020.

28. Ziegler, C., Allon, S.J., Nyquist, S.K., Mbano, I., Miao, V.N., Cao, Y., Yousif, A.S., Bals, J., Hauser, B.M., and Feldman, J. SARS-CoV-2 Receptor ACE2 is an Interferon-Stimulated Gene in Human Airway Epithelial Cells and Is Enriched in Specific Cell Subsets Across Tissues. Cell 2020.

29. Honko, A.N., Johnson, J.C., Marchand, J.S., Huzella, L., Adams, R.D., Oberlander, N., Torzewski, L.M., Bennett, R.S., Hensley, L.E., Jahrling, P.B., and Olinger, G.G. High dose sertraline monotherapy fails to protect rhesus macaques from lethal challenge with Ebola virus Makona. Sci Rep **7**, 5886, 2017.

30. Schwegmann-Weßels, C., and Herrler, G. Sialic acids as receptor determinants for coronaviruses. Glycoconjugate journal **23**, 51, 2006.

31. Huh, D., Leslie, D.C., Matthews, B.D., Fraser, J.P., Jurek, S., Hamilton, G.A., Thorneloe, K.S., McAlexander, M.A., and Ingber, D.E. A human disease model of drug toxicity-induced pulmonary edema in a lung-on-a-chip microdevice. Sci Transl Med **4**, 159ra147, 2012.

32. Bhowmick, R., Derakhshan, T., Liang, Y., Ritchey, J., Liu, L., and Gappa-Fahlenkamp, H. A Three-Dimensional Human Tissue-Engineered Lung Model to Study Influenza A Infection. Tissue Eng Part A **24**, 1468, 2018.

33. Gardner, J.K., and Herbst-Kralovetz, M.M. Three-Dimensional Rotating Wall Vessel-Derived Cell Culture Models for Studying Virus-Host Interactions. Viruses **8**2016.

34. Suderman, M., McCarthy, M., Mossell, E., Watts, D., Peters, C., Shope, R., and Goodwin, T. Three-Dimensional Human Bronchial-Tracheal Epithelial Tissue-Like Assemblies (TLAs) as Hosts For Severe Acute Respiratory Syndrome (SARS)-CoV Infection. 2006.

35. Ling, T.Y., Kuo, M.D., Li, C.L., Yu, A.L., Huang, Y.H., Wu, T.J., Lin, Y.C., Chen, S.H., and Yu, J. Identification of pulmonary Oct-4+ stem/progenitor cells and demonstration of their susceptibility to SARS coronavirus (SARS-CoV) infection in vitro. Proc Natl Acad Sci U S A **103**, 9530, 2006.

36. Miller, A.J., and Spence, J.R. In Vitro Models to Study Human Lung Development, Disease and Homeostasis. Physiology (Bethesda) **32**, 246, 2017.

37. Nichols, J.E., Niles, J.A., Vega, S.P., and Cortiella, J. Novel in vitro respiratory models to study lung development, physiology, pathology and toxicology. Stem Cell Res Ther **4 Suppl 1**, S7, 2013.

38. ter Meulen, J., Bakker, A.B., van den Brink, E.N., Weverling, G.J., Martina, B.E., Haagmans, B.L., Kuiken, T., de Kruif, J., Preiser, W., Spaan, W., Gelderblom, H.R., Goudsmit, J., and Osterhaus, A.D. Human monoclonal antibody as prophylaxis for SARS coronavirus infection in ferrets. Lancet **363**, 2139, 2004.

39. Li, B.-j., Tang, Q., Cheng, D., Qin, C., Xie, F.Y., Wei, Q., Xu, J., Liu, Y., Zheng, B.-j., and Woodle, M.C. Using siRNA in prophylactic and therapeutic regimens against SARS coronavirus in Rhesus macaque. Nature medicine **11**, 944, 2005.

40. Sanchez-Guzman, D., Le Guen, P., Villeret, B., Sola, N., Le Borgne, R., Guyard, A., Kemmel, A., Crestani, B., Sallenave, J.-M., and Garcia-Verdugo, I. Silver nanoparticle-adjuvanted vaccine protects against lethal influenza infection through inducing BALT and IgA-mediated mucosal immunity. Biomaterials **217**, 119308, 2019.

41. Emanuel, E.J., Persad, G., Upshur, R., Thome, B., Parker, M., Glickman, A., Zhang, C., Boyle, C., Smith, M., and Phillips, J.P. Fair allocation of scarce medical resources in the time of Covid-19. Mass Medical Soc; 2020.

42. Kruse, R.L. Therapeutic strategies in an outbreak scenario to treat the novel coronavirus originating in Wuhan, China. F1000Research **9**2020.

43. Gautret, P., Lagier, J.-C., Parola, P., Meddeb, L., Mailhe, M., Doudier, B., Courjon, J., Giordanengo, V., Vieira, V.E., and Dupont, H.T. Hydroxychloroquine and azithromycin as a treatment of COVID-19: results of an open-label non-randomized clinical trial. International Journal of Antimicrobial Agents, 105949, 2020.

44. Kim, A.H., Sparks, J.A., Liew, J.W., Putman, M.S., Berenbaum, F., Duarte-García, A., Graef, E.R., Korsten, P., Sattui, S.E., and Sirotich, E. A Rush to Judgment? Rapid Reporting and Dissemination of Results and Its Consequences Regarding the Use of Hydroxychloroquine for COVID-19. Annals of Internal Medicine 2020.

45. Li, X., Chang, S., Du, G., Li, Y., Gong, J., Yang, M., and Wei, Z. Encapsulation of azithromycin into polymeric microspheres by reduced pressure-solvent evaporation method. International journal of pharmaceutics **433**, 79, 2012.

46. Wang, Q., Mi, G., Hickey, D., Li, Y., Tu, J., Webster, T.J., and Shen, Y. Azithromycin-loaded respirable microparticles for targeted pulmonary delivery for the treatment of pneumonia. Biomaterials **160**, 107, 2018.

47. Tett, S., Cutler, D., Day, R., and Brown, K. Bioavailability of hydroxychloroquine tablets in healthy volunteers. British journal of clinical pharmacology **27**, 771, 1989.

48. Mansilla, E., Marin, G.H., Nunez, L., Drago, H., Sturla, F., Mertz, C., Rivera, L., Ichim, T., Riordan, N., and Raimondi, C. The lysosomotropic agent, hydroxychloroquine, delivered in a biodegradable nanoparticle system, overcomes drug resistance of B-chronic lymphocytic leukemia cells in vitro. Cancer Biotherapy and Radiopharmaceuticals **25**, 97, 2010.

49. Mulangu, S., Dodd, L.E., Davey Jr, R.T., Tshiani Mbaya, O., Proschan, M., Mukadi, D., Lusakibanza Manzo, M., Nzolo, D., Tshomba Oloma, A., and Ibanda, A. A randomized, controlled trial of Ebola virus disease therapeutics. New England Journal of Medicine **381**, 2293, 2019.

50. Tian, X., Li, C., Huang, A., Xia, S., Lu, S., Shi, Z., Lu, L., Jiang, S., Yang, Z., Wu, Y., and Ying, T. Potent binding of 2019 novel coronavirus spike protein by a SARS coronavirus-specific human monoclonal antibody. Emerg Microbes Infect **9**, 382, 2020.

51. Simovic, S., Diener, K., Bachhuka, A., Kant, K., Losic, D., Hayball, J., Brownc, M., and Vasilev, K. Controlled release and bioactivity of the monoclonal antibody rituximab from a porous matrix: A potential in situ therapeutic device. Materials Letters **130**, 210, 2014.

52. Schweizer, D., Vostiar, I., Heier, A., Serno, T., Schoenhammer, K., Jahn, M., Jones, S., Piequet, A., Beerli, C., and Gram, H. Pharmacokinetics, biocompatibility and bioavailability of a controlled release monoclonal antibody formulation. Journal of controlled release **172**, 975, 2013.

53. Abramson, A., Caffarel-Salvador, E., Khang, M., Dellal, D., Silverstein, D., Gao, Y., Frederiksen, M.R., Vegge, A., Hubálek, F., and Water, J.J. An ingestible self-orienting system for oral delivery of macromolecules. Science **363**, 611, 2019.

54. Zheng, B.-j., Guan, Y., Tang, Q., Du, C., Xie, F.Y., He, M.-L., Chan, K.-W., Wong, K.-L., Lader, E., and Woodle, M.C. Prophylactic and therapeutic effects of small interfering RNA targeting SARS coronavirus. Antiviral therapy **9**, 365, 2004.

55. Chen, Y., Gu, H., Zhang, D.S.-Z., Li, F., Liu, T., and Xia, W. Highly effective inhibition of lung cancer growth and metastasis by systemic delivery of siRNA via multimodal mesoporous silica-based nanocarrier. Biomaterials **35**, 10058, 2014.

56. Fehring, V., Schaeper, U., Ahrens, K., Santel, A., Keil, O., Eisermann, M., Giese, K., and Kaufmann, J. Delivery of therapeutic siRNA to the lung endothelium via novel lipoplex formulation DACC. Molecular Therapy **22**, 811, 2014.

57. Demicheli, V., Jefferson, T., Ferroni, E., Rivetti, A., and Di Pietrantonj, C. Vaccines for preventing influenza in healthy adults. Cochrane Database Syst Rev **2**, CD001269, 2018.

58. Kaselitz, T.B., Martin, E.T., Power, L.E., and Cinti, S. Impact of vaccination on morbidity and mortality in adults hospitalized with influenza A, 2014–2015. Infectious Diseases in Clinical Practice **27**, 328, 2019.

59. Poole, P.J., Chacko, E., Wood-Baker, R.W., and Cates, C.J. Influenza vaccine for patients with chronic obstructive pulmonary disease. Cochrane Database Syst Rev, CD002733, 2006.

60. Chen, W.H., Strych, U., Hotez, P.J., and Bottazzi, M.E. The SARS-CoV-2 Vaccine Pipeline: an Overview. Curr Trop Med Rep, 1, 2020.

61. Jones, K.S. Biomaterials as vaccine adjuvants. Biotechnology progress **24**, 807, 2008.

62. Chen, X., Yan, Y., Mullner, M., Ping, Y., Cui, J., Kempe, K., Cortez-Jugo, C., and Caruso, F. Shape-Dependent Activation of Cytokine Secretion by Polymer Capsules in Human Monocyte-Derived Macrophages. Biomacromolecules **17**, 1205, 2016.

63. Veiseh, O., Doloff, J.C., Ma, M., Vegas, A.J., Tam, H.H., Bader, A.R., Li, J., Langan, E., Wyckoff, J., and Loo, W.S. Size-and shape-dependent foreign body immune response to materials implanted in rodents and non-human primates. Nature materials **14**, 643, 2015.

64. Li, W.A., Lu, B.Y., Gu, L., Choi, Y., Kim, J., and Mooney, D.J. The effect of surface modification of mesoporous silica micro-rod scaffold on immune cell activation and infiltration. Biomaterials **83**, 249, 2016.

65. Gause, K.T., Yan, Y., O'Brien-Simpson, N.M., Cui, J., Lenzo, J.C., Reynolds, E.C., and Caruso, F. Codelivery of NOD2 and TLR9 ligands via nanoengineered protein antigen particles for improving and tuning immune responses. Advanced Functional Materials **26**, 7526, 2016.

66. Barry, M.A., Wang, Q., Jones, K.M., Heffernan, M.J., Buhaya, M.H., Beaumier, C.M., Keegan, B.P., Zhan, B., Dumonteil, E., Bottazzi, M.E., and Hotez, P.J. A therapeutic nanoparticle vaccine against Trypanosoma cruzi in a BALB/c mouse model of Chagas disease. Hum Vaccin Immunother **12**, 976, 2016.

67. Primard, C., Rochereau, N., Luciani, E., Genin, C., Delair, T., Paul, S., and Verrier, B. Traffic of poly(lactic acid) nanoparticulate vaccine vehicle from intestinal mucus to sub-epithelial immune competent cells. Biomaterials **31**, 6060, 2010.

68. Jiang, T., Singh, B., Li, H.-S., Kim, Y.-K., Kang, S.-K., Nah, J.-W., Choi, Y.-J., and Cho, C.-S. Targeted oral delivery of BmpB vaccine using porous PLGA microparticles coated with M cell homing peptide-coupled chitosan. Biomaterials **35**, 2365, 2014.

69. Feng, G., Jiang, Q., Xia, M., Lu, Y., Qiu, W., Zhao, D., Lu, L., Peng, G., and Wang, Y. Enhanced immune response and protective effects of nano-chitosan-based DNA vaccine encoding T cell epitopes of Esat-6 and FL against Mycobacterium tuberculosis infection. PLoS One **8**, e61135, 2013.

70. Yao, W., Peng, Y., Du, M., Luo, J., and Zong, L. Preventative vaccine-loaded mannosylated chitosan nanoparticles intended for nasal mucosal delivery enhance immune responses and potent tumor immunity. Mol Pharm **10**, 2904, 2013.

71. Wu, Y., Wei, W., Zhou, M., Wang, Y., Wu, J., Ma, G., and Su, Z. Thermal-sensitive hydrogel as adjuvant-free vaccine delivery system for H5N1 intranasal immunization. Biomaterials **33**, 2351, 2012.

72. Pimentel, T.A., Yan, Z., Jeffers, S.A., Holmes, K.V., Hodges, R.S., and Burkhard, P. Peptide nanoparticles as novel immunogens: design and analysis of a prototypic severe acute respiratory syndrome vaccine. Chemical biology & drug design **73**, 53, 2009.

73. Ali, O.A., Doherty, E., Mooney, D.J., and Emerich, D. Relationship of vaccine efficacy to the kinetics of DC and T-cell responses induced by PLG-based cancer vaccines. Biomatter **1**, 66, 2011.

74. Ali, O.A., Emerich, D., Dranoff, G., and Mooney, D.J. In situ regulation of DC subsets and T cells mediates tumor regression in mice. Sci Transl Med **1**, 8ra19, 2009.

75. Kim, J., Li, W.A., Choi, Y., Lewin, S.A., Verbeke, C.S., Dranoff, G., and Mooney, D.J. Injectable, spontaneously assembling, inorganic scaffolds modulate immune cells in vivo and increase vaccine efficacy. Nat Biotechnol **33**, 64, 2015.

76. Ali, O.A., Doherty, E., Bell, W.J., Fradet, T., Hudak, J., Laliberte, M.T., Mooney, D.J., and Emerich, D.F. Biomaterial-based vaccine induces regression of established intracranial glioma in rats. Pharm Res **28**, 1074, 2011.

77. Dendritic Cell Activating Scaffold in Melanoma. ClinicalTrials.gov: National Library of Medicine.

78. Super, M., Doherty, E.J., Cartwright, M.J., Seiler, B.T., White, D.A., Langellotto, F., Stafford, A.G., Dimitrakakis, N., Karkada, M., and Graveline, A.R. Modular biomaterials vaccine technology protects against multiple pathogens and septic shock. bioRxiv 2020.

79. Correia, B.E., Bates, J.T., Loomis, R.J., Baneyx, G., Carrico, C., Jardine, J.G., Rupert, P., Correnti, C., Kalyuzhniy, O., Vittal, V., Connell, M.J., Stevens, E., Schroeter, A., Chen, M., Macpherson, S., Serra, A.M., Adachi, Y., Holmes, M.A., Li, Y., Klevit, R.E., Graham, B.S., Wyatt, R.T., Baker, D., Strong, R.K., Crowe, J.E., Jr., Johnson, P.R., and Schief, W.R. Proof of principle for epitope-focused vaccine design. Nature **507**, 201, 2014.

80. Luo, X., Liu, T., Wang, Y., Jia, H., Zhang, Y., Caballero, D., Du, J., Wang, R.E., Wang, D., Schultz, P.G., and Wang, F. An Epitope-Specific Respiratory Syncytial Virus Vaccine Based on an Antibody Scaffold. Angew Chem Int Ed Engl **54**, 14531, 2015.

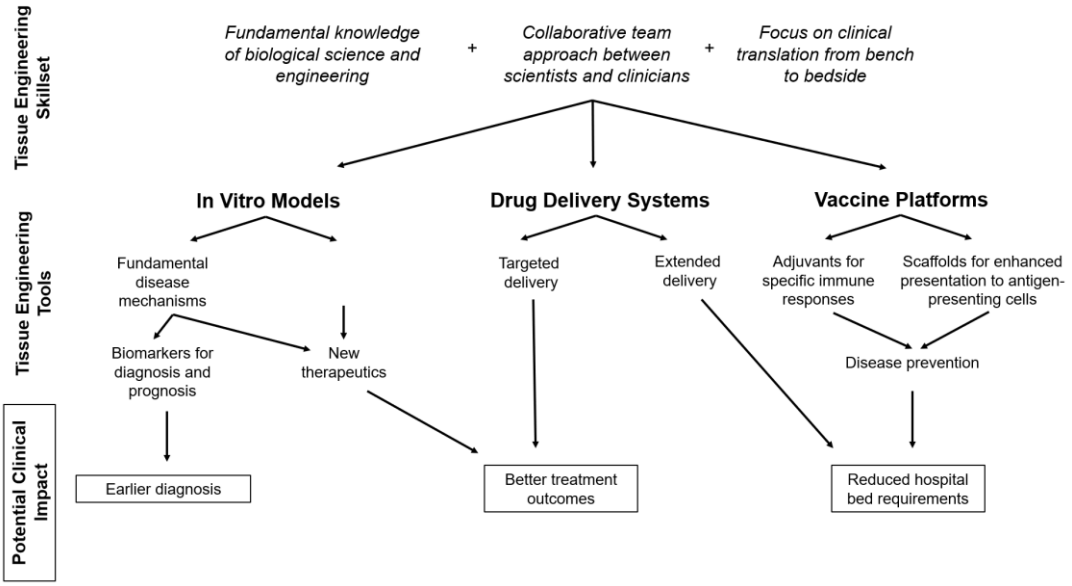

Figure 1. Examples of how tissue engineering skills and tools may be leveraged to have impact on clinical practice in the setting of a viral outbreak