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EVIDENCE SEARCH REPORT

RESEARCH QUESTION: What is the time course of antibody development in the clinical course of COVID-19?	UNIQUE IDENTIFIER: LAB041402-01
REQUESTED RESOURCES: • medRxiv • CDC website/database • Google • Google Scholar • OVID Medline • PubMed • WHO Global Research on COVID-19 • PHAC COVID-19 • Up to Date	
LIMITS/EXCLUSIONS/INCLUSIONS: English	
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CITE AS: Duncan, V. At what time in the disease timeline of COVID-19 do antibodies develop? 2020 Apr 14; Document no.: LAB041402-01 ESR. In: COVID-19 Rapid Evidence Reviews [Internet]. SK: SK COVID Evidence Support Team, c2020. 42 p. (CEST evidence search report)	

LIBRARIAN NOTES/COMMENTS

The searcher employed a broad search strategy, and asked the researcher to identify relevant articles. Articles citing a very relevant article were identified, and reviewed.

SEARCH RESULTS

To obtain full-text articles email library@saskhealthauthority.ca.

SUMMARIES, GUIDELINES & OTHER RESOURCES

Clinical Evidence

Include:

DynaMed, CADTH reports, Nursing Reference Center/Rehabilitation Reference Center, websites, policies, links from Google Scholar, Google, etc.

Recommendations

Guidance from societies, etc.

Grey Literature

Sub-headings may be created for reading clarity. For example: Websites, Policies, etc.

ARTICLES FROM LIBRARY DATABASES

Note: References are in alphabetical order by author.

1. **Amanat F, Nguyen T, Chromikova V, et al. A serological assay to detect SARS-CoV-2 seroconversion in humans. *medRxiv*. 2020:2020.03.17.20037713. DOI:** 10.1101/2020.03.17.20037713

Introduction: SARS-Cov-2 (severe acute respiratory disease coronavirus 2), which causes Coronavirus Disease 2019 (COVID19) was first detected in China in late 2019 and has since then caused a global pandemic. While molecular assays to directly detect the viral genetic material are available for the diagnosis of acute infection, we currently lack serological assays suitable to specifically detect SARS-CoV-2 antibodies. Methods: Here we describe serological enzyme-linked immunosorbent assays (ELISA) that we developed using recombinant antigens derived from the spike protein of SARS-CoV-2. These assays were developed with negative control samples representing pre-COVID 19 background immunity in the general population and samples from COVID19 patients. Results: The assays are sensitive and specific, allowing for screening and identification of COVID19 seroconverters using human plasma/serum as early as 3 days post symptom onset. Importantly, these assays do not require handling of infectious virus, can be adjusted to detect different antibody types and are amendable to scaling. Conclusion: Serological assays are of critical importance to determine seroprevalence in a given population, define previous exposure and identify highly reactive human donors for the generation of convalescent serum as therapeutic. Sensitive and specific identification of coronavirus SARS-Cov-2 antibody titers will also support screening of health care workers to identify those who are already immune and can be deployed to care for infected patients minimizing the risk of viral spread to colleagues and other patients.Competing Interest StatementThe authors have declared no competing interest.Clinical TrialThis is not a clinical trial, just development of a serological assay.Funding Statement Mount Sinai Health System Translational Science Hub (NIH grant U54TR001433) for supporting sample collection. The work of the Personalized Virology Initiative is supported by institutional funds and philanthropic donations. This work was partially supported by the NIAID Centers of Excellence for Influenza Research and Surveillance (CEIRS) contract HHSN272201400008C, the Australian National Health and Medical Research Council (NHMRC) NHMRC Program Grant (1071916) and NHMRC Research Fellowship Level B (#1102792), the Academy of Finland and Helsinki University Hospital Funds (TYH2018322). Finally, we want to thank the three COVID19 patients for their contribution to research and wish them a speedy recovery.Author DeclarationsAll relevant ethical guidelines have been followed; any necessary IRB and/or ethics committee approvals have been obtained and details of the IRB/oversight body are included in the manuscript.YesAll necessary patient/participant consent has been obtained and the appropriate institutional forms have been archived.YesI understand that all clinical trials and any other prospective interventional studies must be registered with an ICMJE-approved registry, such as ClinicalTrials.gov. I confirm that any such study reported in the manuscript has been registered and the trial registration ID is provided (note: if posting a prospective study registered retrospectively, please provide a statement in the trial ID field explaining why the study was not registered in advance).Yes I have followed all appropriate research reporting guidelines and uploaded the relevant EQUATOR Network research reporting checklist(s) and other pertinent material as supplementary files, if applicable.YesData are available from the corresponding author.

URL: http://medrxiv.org/content/early/2020/03/18/2020.03.17.20037713.abstract

DOI: 10.1101/2020.03.17.20037713

2. **AminJafari A, Ghasemi S. The Possible of Immunotherapy for COVID-19: a Systematic Review. *Int Immunopharmacol*. 2020:106455. DOI:** 10.1016/j.intimp.2020.106455

The novel coronavirus (2019-nCoV) is an emerging pathogen that was first described in late December 2019 and causes a severe respiratory infection in humans. Since the outbreak of COVID-19, international attention has raised to develop treatment and control options such as types of immunotherapies. The immunotherapy is an effective method for fighting against similar viral infections such as SARS-CoV, and MERS-CoV. These methods include several types of vaccines, monoclonal antibody candidates, and etc. This systematic review article was designed to evaluate the existing evidence and experience related to immunotherapy for 2019-nCoV. Web of Science (ISI), PubMed, and Scopus databases were used to search for suitable keywords such as 2019-nCoV, novel coronavirus, Immunotherapy, interleukin, vaccine and the related words for relevant publications up to 24.3.2020. The present systematic review was performed based on PRISMA protocol. Data extraction and quality valuation of articles were performed by two reviewers. 51 articles were the results of the search and based on the inclusions and exclusions criteria, 7 articles were included in the final review. As a conclusion of these studies demonstrated that although no serious research has been done on this subject at the time of writing this article, similar studies on the related viruses showed notable results. So immunotherapy for this virus can also be a suitable option.

URL: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7128194/
DOI: 10.1016/j.intimp.2020.106455

3. **Bai S, Wang J, Zhou Y, et al. Analysis of the first cluster of cases in a family of novel coronavirus pneumonia in Gansu Province. *Chinese Journal of Preventive Medicine*. 2020;54(0):E005-E.**

The epidemiological history and clinical characteristics of 7 cases of COVID-19 and 1 case of close contact in the first family aggregation epidemic of COVID-19 in Gansu Province were analyzed. The first patient A developed on January 22, 2020, with a history of residence in Wuhan, and confirmed severe cases of NCP on January 24, 2020; patient B, on January 23, 2020, diagnosed on January 31, severe cases; patient C, asymptomatic, diagnosed on January 27; patient D, asymptomatic, diagnosed on January 27; patient E, on January 24, diagnosed on January 28; patient F, asymptomatic, diagnosed on January 31; Patient G was asymptomatic and was diagnosed on January 31. In close contact, H was asymptomatic, PCR test was negative and asymptomatic, and he was discharged early. Among the 7 patients, 1 case died of (B) aggravation, and the other patients' condition was effectively controlled after active treatment. Except for the discharged cases, 5 cases were positive for COVID-19 specific IgM antibody and 1 case was negative. In this clustering outbreak, 4 patients remained asymptomatic, but PCR and IgM antibodies were positive, indicating that asymptomatic patients may be the key point to control the epidemic. Specific IgM antibody screening for patients whose pharyngeal swab nucleic acid test is negative but with ground glass-like lung lesions is very important for early detection and early isolation.;

URL: DOI:

4. **Bai SL, Wang JY, Zhou YQ, et al. Analysis of the first cluster of cases in a family of novel coronavirus pneumonia in Gansu Province. *Zhonghua yu fang yi xue za zhi [Chinese journal of preventive medicine]*. 2020;54(0):E005-E. DOI:** 10.3760/cma.j.issn.0253-9624.2020.0005

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URL: https://pubmed.ncbi.nlm.nih.gov/32064855
DOI: 10.3760/cma.j.issn.0253-9624.2020.0005

5. **Bloch EM, Shoham S, Casadevall A, et al. Deployment of convalescent plasma for the prevention and treatment of COVID-19. *J Clin Invest*. 2020;2020/04/08. DOI:** 10.1172/JCI138745.; ID: 9765
10.1172/jci138745
Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2), the cause of coronavirus disease (COVID-19), has spurred a global health crisis. To date, there are no proven options for prophylaxis for those who have been exposed to SARS-CoV-2, nor therapy for those who develop COVID-19. Immune (i.e. "convalescent") plasma refers to plasma that is collected from individuals, following resolution of infection and development of antibodies. Passive antibody administration through transfusion of convalescent plasma may offer the only short-term strategy to confer immediate immunity to susceptible individuals. There are numerous examples, where convalescent plasma has been used successfully as post-exposure prophylaxis and/or treatment of infectious diseases, including other outbreaks of coronaviruses (e.g., SARS-1, Middle East Respiratory Syndrome MERS]). Convalescent plasma has also been used in the COVID-19 pandemic; limited data from China suggest clinical benefit, including radiological resolution, reduction in viral loads and improved survival. Globally, blood centers have robust infrastructure to undertake collections and construct inventories of convalescent plasma to meet the growing demand. Nonetheless, there are nuanced challenges, both regulatory and logistical, spanning donor eligibility, donor recruitment, collections and transfusion itself. Data from rigorously controlled clinical trials of convalescent plasma are also few, underscoring the need to evaluate its use objectively for a range of indications (e.g., prevention vs treatment) and patient populations (e.g., age, comorbid disease). We provide an overview of convalescent plasma, from evidence of benefit, regulatory considerations, logistical work flow and proposed clinical trials, as scale up is brought underway to mobilize this critical resource. â€ƒ.

URL: DOI: 10.1172/JCI138745.; ID: 9765
10.1172/jci138745

6. **Chan JF, Zhang AJ, Yuan S, et al. Simulation of the clinical and pathological manifestations of Coronavirus Disease 2019 (COVID-19) in golden Syrian hamster model: implications for disease pathogenesis and transmissibility. *Clin Infect Dis*. 2020. DOI:** 10.1093/cid/ciaa325.; ID: 4657
10.1093/cid/ciaa325
BACKGROUND: A physiological small animal model that resembles COVID-19 with low mortality is lacking. METHODS: Molecular docking on the binding between angiotensin-converting enzyme 2 (ACE2) of common laboratory mammals and the receptor-binding domain of the surface spike protein of SARS-CoV-2 suggested that the golden Syrian hamster is an option. Virus challenge, contact transmission, and passive immunoprophylaxis were performed. Serial organ tissues and blood were harvested for histopathology, viral load and titre, chemokine/cytokine assay, and neutralising antibody titre. RESULTS: The Syrian hamster could be consistently infected by SARS-CoV-2. Maximal clinical signs of rapid breathing, weight loss, histopathological changes from the initial exudative phase of diffuse alveolar damage with extensive apoptosis to the later proliferative phase of tissue repair, airway and intestinal involvement with virus nucleocapsid protein expression, high lung viral load, and spleen and lymphoid atrophy associated with marked cytokine activation were observed within the first week of virus challenge. The lung virus titre was between 10^{5}-10^{7} TCID50/g. Challenged index hamsters consistently infected naïve contact hamsters housed within the same cage, resulting in similar pathology but not weight loss. All infected hamsters recovered and developed mean serum neutralising antibody titre ≥1:427 fourteen days post-challenge. Immunoprophylaxis with early convalescent serum achieved significant decrease in lung viral load but not in lung pathology. No consistent non-synonymous adaptive mutation of the spike was found in viruses isolated from infected hamsters. CONCLUSIONS: Besides satisfying the Koch's postulates, this readily available hamster model is an important tool for studying transmission, pathogenesis, treatment, and vaccination against SARS-CoV-2.

URL: DOI: 10.1093/cid/ciaa325.; ID: 4657
10.1093/cid/ciaa325

7. **Chan JF-W, Zhang AJ, Yuan S, et al. Simulation of the clinical and pathological manifestations of Coronavirus Disease 2019 (COVID-19) in golden Syrian hamster model: implications for disease pathogenesis and transmissibility. *Clinical Infectious Diseases*. 2020. DOI:** 10.1093/cid/ciaa325

Background A physiological small animal model that resembles COVID-19 with low mortality is lacking. Methods Molecular docking on the binding between angiotensin-converting enzyme 2 (ACE2) of common laboratory mammals and the receptor-binding domain of the surface spike protein of SARS-CoV-2 suggested that the golden Syrian hamster is an option. Virus challenge, contact transmission, and passive immunoprophylaxis were performed. Serial organ tissues and blood were harvested for histopathology, viral load and titre, chemokine/cytokine assay, and neutralising antibody titre. Results The Syrian hamster could be consistently infected by SARS-CoV-2. Maximal clinical signs of rapid breathing, weight loss, histopathological changes from the initial exudative phase of diffuse alveolar damage with extensive apoptosis to the later proliferative phase of tissue repair, airway and intestinal involvement with virus nucleocapsid protein expression, high lung viral load, and spleen and lymphoid atrophy associated with marked cytokine activation were observed within the first week of virus challenge. The lung virus titre was between 105-107 TCID50/g. Challenged index hamsters consistently infected naïve contact hamsters housed within the same cage, resulting in similar pathology but not weight loss. All infected hamsters recovered and developed mean serum neutralising antibody titre ≥1:427 fourteen days post-challenge. Immunoprophylaxis with early convalescent serum achieved significant decrease in lung viral load but not in lung pathology. No consistent non-synonymous adaptive mutation of the spike was found in viruses isolated from infected hamsters. Conclusions Besides satisfying the Koch's postulates, this readily available hamster model is an important tool for studying transmission, pathogenesis, treatment, and vaccination against SARS-CoV-2.

URL: https://academic.oup.com/cid/advance-article/doi/10.1093/cid/ciaa325/5811871
DOI: 10.1093/cid/ciaa325

8. **Chen C, Zhang XR, Ju ZY, et al. Advances in the research of cytokine storm mechanism induced by Corona Virus Disease 2019 and the corresponding immunotherapies. *Zhonghua shao shang za zhi = Zhonghua shaoshang zazhi = Chinese journal of burns*. 2020;36(0):E005-E. DOI:** 10.3760/cma.j.cn501120-20200224-00088

Corona Virus Disease 2019 (COVID-19) has seriously affected the treatment of patients and social stability. In the later stage of disease, some COVID-19 patients may develop into acute respiratory distress syndrome or even multiple organ failure. However, one of the most important mechanism underlying the deterioration of disease is cytokine storm. At present, some therapies such as interleukin-6 antibody blocker, stem cell therapy, and transfusion of convalescent plasma have been applied to counteract the cytokine storm and have made some progress. This article reviews the influences of cytokine storm syndrome on the COVID-19 and the corresponding immunotherapies to resist cytokine storm.

URL: https://pubmed.ncbi.nlm.nih.gov/32114747
DOI: 10.3760/cma.j.cn501120-20200224-00088

9. **Chen X, Ran D, Zeng L, et al. Immunoassay of cooked wild rat meat by ELISA with a highly specific antibody targeting rat heat-resistant proteins. *Food Agric Immunol*. 2020;31(1):533-44. DOI:** 10.1080/09540105.2020.1740180

The 2019 new coronavirus epidemic potentially induced by wild animals has drawn tremendous attention. Wild animal meat contamination and adulteration have become increasingly serious, particularly for highly cooked wild animal meats that are difficult to be detected. In this study, a highly specific polyclonal antibody targeting the cooked rat proteins was developed. The corresponding sandwich ELISA (swELISA) was developed and found highly sensitive and specific for cooked rat meat, while there are no cross-reactions to the cooked chicken, pork and beef meats. The limit of detection (LOD) is determined to be as low as 0.01 ug/L based OD values. The coefficient variation (CV) is 5% and 8% for intra and inter assays, respectively. The recovery efficiencies are between 90% and 110%. The sandwich ELISA can detect both raw and cooked rat meat and is also suitable for Swab test of rat contamination. The results indicated a highly reliable and robust ELISA-based assay for cooked rat meat identification and contamination. © 2020, © 2020 The Author(s). Published with license by Taylor and Francis Group, LLC.

URL: https://www.scopus.com/inward/record.uri?eid=2-s2.0-85082764714&doi=10.1080%2f09540105.2020.1740180&partnerID=40&md5=9c711b7a85954c794003d570d69a760f
DOI: 10.1080/09540105.2020.1740180

10. **Chenoweth AM, Wines BD, Anania JC, et al. Harnessing the immune system via FcgammaR function in immune therapy: A pathway to next-gen mAbs. *Immunol Cell Biol*. 2020;11:11. DOI:** https://dx.doi.org/10.1111/imcb.12326

The human FcgammaRs interact with antigen-complexed IgG ligands to both activate and modulate a powerful network of inflammatory host-protective effector functions that are key to the normal physiology of immune resistance to pathogens. More than 100 therapeutic monoclonal antibodies (mAbs) are approved or in late stage clinical trials, many of which harness the potent FcgammaR-mediated effector systems to varying degrees. This is most evident for antibodies targeting cancer cells inducing antibody-dependent killing or phagocytosis but is also true to some degree for the mAbs that neutralise or remove small macromolecules such as cytokines or other immunoglobulins. The use of mAb therapeutics has also revealed a "scaffolding" role for FcgammaR which, in different contexts, may either underpin the therapeutic mAb action such as immune agonism or may trigger catastrophic adverse effects. The still unmet therapeutic need in many cancers, inflammatory diseases or emerging infections such as SARS-CoV-2, requires increased effort on the development of improved and novel mAbs. A more mature appreciation of the immunobiology of individual FcgammaR function and the complexity of the relationships between FcgammaRs and antibodies is fuelling efforts to develop more potent "next-gen" therapeutic antibodies. Such development strategies now include focused glycan or protein engineering of the Fc to increase affinity and/or tailor specificity for selective engagement of individual activating FcgammaRs or the inhibitory FcgammaRIIb or alternatively, for the ablation of FcgammaR interaction altogether. This review touches on recent aspects FcgammaR and IgG immunobiology and its relationship to the present and future actions of therapeutic mAbs.

URL:

http://ovidsp.ovid.com/ovidweb.cgi?T=JS&CSC=Y&NEWS=N&PAGE=fulltext&D=medp&AN=32157732http://sfx.library.cdc.gov/cdc?sid=OVID:medline&id=pmid:32157732&id=10.1111%2Fimcb.12326&issn=0818-9641&isbn=&volume=&issue=&spage=&pages=&date=2020&title=Immunology+%26+Cell+Biology&atitle=Harnessing+the+immune+system+via+FcgammaR+function+in+immune+therapy%3A+A+pathway+to+next-gen+mAbs.&aulast=Chenoweth&pid=%3Cauthor%3EChenoweth+AM%2CWines+BD%2CAnania+JC%2CMark+Hogarth+P%3C%2Fauthor%3E&%3CAN%3E32157732%3C%2FAN%3E&%3CDT%3EJournal+Article%3C%2FDT%3E

DOI: https://dx.doi.org/10.1111/imcb.12326

11. **Chenoweth AM, Wines BD, Anania JC, et al. Harnessing the immune system via FcÎ³R function in immune therapy: A pathway to next-gen mAbs. *Immunology and cell biology*. 2020:10.1111/imcb.12326. DOI:** 10.1111/imcb.12326

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URL: https://pubmed.ncbi.nlm.nih.gov/32157732
DOI: 10.1111/imcb.12326

12. **Correa Giron C, Laaksonen A, Barroso da Silva FL. On the interactions of the receptor-binding domain of SARS-CoV-1 and SARS-CoV-2 spike proteins with monoclonal antibodies and the receptor ACE2. *bioRxiv*. 2020:2020.04.05.026377. DOI:** 10.1101/2020.04.05.026377

A new betacoronavirus named SARS-CoV-2 has emerged as a new threat to global health and economy. A promising target for both diagnosis and therapeutics treatments of the new disease named COVID-19 is the coronavirus (CoV) spike (S) glycoprotein. By constant-pH Monte Carlo simulations and the PROCEEDpKa method, we have mapped the electrostatic epitopes for four monoclonal antibodies and the angiotensin-converting enzyme 2 (ACE2) on both SARS-CoV-1 and the new SARS-CoV-2 S receptor binding domain (RBD) proteins. We also calculated free energy of interactions and shown that the S RBD proteins from both SARS viruses binds to ACE2 with similar affinities. However, the affinity between the S RBD protein from the new SARS-CoV-2 and ACE2 is higher than for any studied antibody previously found complexed with SARS-CoV-1. Based on physical chemical analysis and free energies estimates, we can shed some light on the involved molecular recognition processes, their clinical aspects, the implications for drug developments, and suggest structural modifications on the CR3022 antibody that would improve its binding affinities for SARS-CoV-2 and contribute to address the ongoing international health crisis.Competing Interest Statement

URL: http://biorxiv.org/content/early/2020/04/10/2020.04.05.026377.abstract
DOI: 10.1101/2020.04.05.026377

13. **D'Annessa I, Marchetti F, Colombo G. Differential Antibody Recognition by Novel SARS-CoV-2 and SARS-CoV Spike Protein Receptor Binding Domains: Mechanistic Insights and Implications for the Design of Diagnostics and Therapeutics. *bioRxiv*. 2020:2020.03.13.990267. DOI:** 10.1101/2020.03.13.990267

The appearance of the novel betacoronavirus SARS-CoV-2 represents a major threat to human health, and its diffusion around the world is causing dramatic consequences. The knowledge of the 3D structures of SARS-CoV-2 proteins can facilitate the development of therapeutic and diagnostic molecules. Specifically, comparative analyses of the structures of SARS-CoV-2 proteins and homologous proteins from previously characterized viruses, such as SARS-CoV, can reveal the common and/or distinctive traits that underlie the mechanisms of recognition of cell receptors and of molecules of the immune system.Herein, we apply our recently developed energy-based methods for the prediction of antibody-binding epitopes and protein-protein interaction regions to the Receptor Binding Domain (RBD) of the Spike proteins from SARS-CoV-2 and SARS-CoV. Our analysis focusses only on the study of the structure of RBDs in isolation, without making use of any previous knowledge of binding properties. Importantly, our results highlight structural and sequence differences among the regions that are predicted to be immunoreactive and bind/elicit antibodies. These results provide a rational basis to the observation that several SARS-CoV RDB-specific monoclonal antibodies fail to appreciably bind the SARS-CoV-2 counterpart. Furthermore, we correctly identify the region of SARS-CoV-2 RBD that is engaged by the cell receptor ACE2 during viral entry into host cells.The data, sequences and structures we present here can be useful for the development of novel therapeutic and diagnostic interventions.

URL: http://biorxiv.org/content/early/2020/03/15/2020.03.13.990267.abstract
DOI: 10.1101/2020.03.13.990267

14. **D'Annessa I, Marchetti F, Colombo G. Differential Antibody Recognition by Novel SARS-CoV-2 and SARS-CoV Spike Protein Receptor Binding Domains: Mechanistic Insights and Implications for the Design of Diagnostics and Therapeutics. *bioRxiv*. 2020:2020.03.13.990267. DOI:** 10.1101/2020.03.13.990267

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URL: http://biorxiv.org/content/early/2020/03/15/2020.03.13.990267.abstract
DOI: 10.1101/2020.03.13.990267

15. **Desautels T, Zemla A, Lau E, et al. Rapid in silico design of antibodies targeting SARS-CoV-2 using machine learning and supercomputing. *bioRxiv*. 2020:2020.04.03.024885. DOI:** 10.1101/2020.04.03.024885

Rapidly responding to novel pathogens, such as SARS-CoV-2, represents an extremely challenging and complex endeavor. Numerous promising therapeutic and vaccine research efforts to mitigate the catastrophic effects of COVID-19 pandemic are underway, yet an efficacious countermeasure is still not available. To support these global research efforts, we have used a novel computational pipeline combining machine learning, bioinformatics, and supercomputing to predict antibody structures capable of targeting the SARS-CoV-2 receptor binding domain (RBD). In 22 days, using just the SARS-CoV-2 sequence and previously published neutralizing antibody structures for SARS-CoV-1, we generated 20 initial antibody sequences predicted to target the SARS-CoV-2 RBD. As a first step in this process, we predicted (and publicly released) structures of the SARS-CoV-2 spike protein using homology-based structural modeling. The predicted structures proved to be accurate within the targeted RBD region when compared to experimentally derived structures published weeks later. Next we used our in silico design platform to iteratively propose mutations to SARS-CoV-1 neutralizing antibodies (known not to bind SARS-Cov-2) to enable and optimize binding within the RBD of SARS-CoV-2. Starting from a calculated baseline free energy of −48.1 kcal/mol (± 8.3), our 20 selected first round antibody structures are predicted to have improved interaction with the SARS-CoV-2 RBD with free energies as low as −82.0 kcal/mole. The baseline SARS-CoV-1 antibody in complex with the SARS-CoV-1 RBD has a calculated interaction energy of −52.2 kcal/mole and neutralizes the virus by preventing it from binding and entering the human ACE2 receptor. These results suggest that our predicted antibody mutants may bind the SARS-CoV-2 RBD and potentially neutralize the virus. Additionally, our selected antibody mutants score well according to multiple antibody developability metrics. These antibody designs are being expressed and experimentally tested for binding to COVID-19 viral proteins, which will provide invaluable feedback to further improve the machine learning-driven designs. This technical report is a high-level description of that effort; the Supplementary Materials includes the homology-based structural models we developed and 178,856 in silico free energy calculations for 89,263 mutant antibodies derived from known SARS-CoV-1 neutralizing antibodies.Competing Interest Statement

URL: http://biorxiv.org/content/early/2020/04/10/2020.04.03.024885.abstract
DOI: 10.1101/2020.04.03.024885

16. **Diao B, Wen K, Chen J, et al. Diagnosis of Acute Respiratory Syndrome Coronavirus 2 Infection by Detection of Nucleocapsid Protein. *medRxiv*. 2020:2020.03.07.20032524. DOI:** 10.1101/2020.03.07.20032524

BACKGROUND Nucleic acid test and antibody assay have been employed in the diagnosis for SARS-CoV-2 infection, but the use of viral antigen for diagnosis has not been successfully developed. Theoretically, viral antigen is the specific marker of the virus and precedes antibody appearance within the infected population. There is a clear need of detection of viral antigen for rapid and early diagnosis. METHODS We included a cohort of 239 participants with suspected SARS-CoV-2 infection from 7 centers for the study. We measured nucleocapsid protein in nasopharyngeal swab samples in parallel with the nucleic acid test. Nucleic acid test was taken as the reference standard, and statistical evaluation was taken in blind. We detected nucleocapsid protein in 20 urine samples in another center, employing nasopharyngeal swab nucleic acid test as reference standard. RESULTS We developed a fluorescence immunochromatographic assay for detecting nucleocapsid protein of SARS-CoV-2 in nasopharyngeal swab sample and urine within 10 minutes. 100% of nucleocapsid protein positive and negative participants accord with nucleic acid test for same samples. Further, earliest participant after 3 days of fever can be identified by the method. In an additional preliminary study, we detected nucleocapsid protein in urine in 73.6% of diagnosed COVID-19 patients. CONCLUSIONS Those findings indicate that nucleocapsid protein assay is an accurate, rapid, early and simple method for diagnosis of COVID-19. Appearance of nucleocapsid protein in urine coincides our finding of the SARS-CoV-2 invading kidney and might be of diagnostic value.Competing Interest StatementThe authors have declared no competing interest.Funding StatementThis research was supported by grants from National Key R&D Program of China (2016YFA0502204); Chongqing Health Commission COVID-19 Project (2020ZX01).Author DeclarationsAll relevant ethical guidelines have been followed; any necessary IRB and/or ethics committee approvals have been obtained and details of the IRB/oversight body are included in the manuscript.YesAll necessary patient/participant consent has been obtained and the appropriate institutional forms have been archived.YesI understand that all clinical trials and any other prospective interventional studies must be registered with an ICMJE-approved registry, such as

ClinicalTrials.gov.I confirm that any such study reported in the manuscript has been registered and the trial registration ID is provided (note: if posting a prospective study registered retrospectively, please provide a statement in the trial ID field explaining why the study was not registered in advance).Yes I have followed all appropriate research reporting guidelines and uploaded the relevant EQUATOR Network research reporting checklist(s) and other pertinent material as supplementary files, if applicable.YesThe data used to support the findings of this study are included within the article.

URL: http://medrxiv.org/content/early/2020/03/10/2020.03.07.20032524.abstract
DOI: 10.1101/2020.03.07.20032524

17. **Du Z, Zhu F, Guo F, et al. Detection of antibodies against SARS-CoV-2 in patients with COVID-19. *Journal of Medical Virology*. 2020;n/a(n/a). DOI:** 10.1002/jmv.25820

Abstract Detection of IgM and IgG against SARS-CoV-2 is a fast and simple screening method. As an effective supplement to RNA testing, antibody detection is of epidemiological significance and an important means to understand the occurrence, development, prognosis, and outcome of COVID-19. However, more medical research is needed on the expression level of antibodies against SARS-CoV-2 and the prognosis of COVID-19. This article is protected by copyright. All rights reserved.

URL: https://doi.org/10.1002/jmv.25820
DOI: 10.1002/jmv.25820

18. **Duan K, Liu B, Li C, et al. The feasibility of convalescent plasma therapy in severe COVID-19 patients: a pilot study. *medRxiv*. 2020:2020.03.16.20036145. DOI:** 10.1101/2020.03.16.20036145

Currently, there are no approved specific antiviral agents for 2019 novel coronavirus disease (COVID-19). In this study, ten severe patients confirmed by real-time viral RNA test were enrolled prospectively. One dose of 200 mL convalescent plasma (CP) derived from recently recovered donors with the neutralizing antibody titers above 1:640 was transfused to the patients as an addition to maximal supportive care and antiviral agents. The primary endpoint was the safety of CP transfusion. The second endpoints were the improvement of clinical symptoms and laboratory parameters within 3 days after CP transfusion. The median time from onset of illness to CP transfusion was 16.5 days. After CP transfusion, the level of neutralizing antibody increased rapidly up to 1:640 in five cases, while that of the other four cases maintained at a high level (1:640). The clinical symptoms were significantly improved along with increase of oxyhemoglobin saturation within 3 days. Several parameters tended to improve as compared to pre-transfusion, including increased lymphocyte counts (0.65*109/L vs. 0.76*109/L) and decreased C-reactive protein (55.98 mg/L vs. 18.13 mg/L). Radiological examinations showed varying degrees of absorption of lung lesions within 7 days. The viral load was undetectable after transfusion in seven patients who had previous viremia. No severe adverse effects were observed. This study showed CP therapy was well tolerated and could potentially improve the clinical outcomes through neutralizing viremia in severe COVID-19 cases. The optimal dose and time point, as well as the clinical benefit of CP therapy, needs further investigation in larger well-controlled trials.Competing Interest StatementThe authors have declared no competing interest.Clinical TrialChiCTR2000030048Funding StatementThis study was funded by Key projects of the Ministry of Science and Technology China "Preparation of specific plasma and specific globulin from patients with a recovery period of COVID-19 infectionÂ" (project number: 2020YFC0841800).This work was also supported by Shanghai Guangci Translational Medicine Development Foundation.Author DeclarationsAll relevant ethical guidelines have been followed; any necessary IRB and/or ethics committee approvals have been obtained and details of the IRB/oversight body are included in the manuscript.YesAll necessary patient/participant consent has been obtained and the appropriate institutional forms have been archived.YesI understand that all clinical trials and any other prospective interventional studies must be registered with an ICMJE-approved registry, such as ClinicalTrials.gov.I confirm that any such study reported in the manuscript has been registered and the trial registration ID is provided (note: if posting a prospective study registered retrospectively, please provide a statement in the trial ID field explaining why the study was not registered in advance).Yes I have followed all appropriate research reporting guidelines and uploaded the relevant EQUATOR Network research reporting checklist(s) and other pertinent material as supplementary files, if applicable.YesAll data associated with the manuscript were available on request

URL: http://medrxiv.org/content/early/2020/03/23/2020.03.16.20036145.abstract
DOI: 10.1101/2020.03.16.20036145

19. **Fast E, Altman RB, Chen B. Potential T-cell and B-cell Epitopes of 2019-nCoV. *bioRxiv*. 2020:2020.02.19.955484. DOI:** 10.1101/2020.02.19.955484

As of Feb 16th 2020, 2019-nCoV has infected more than 51,857 people across 26 countries and claimed 1666 lives. 2019-nCoV is a novel form of coronavirus that causes COVID-19 and has high similarity with SARS-CoV. No approved vaccine yet exists for 2019-nCoV or any form of coronavirus. Here we use computational tools from structural biology and machine learning to identify 2019-nCoV T-cell and B-cell epitopes based on viral protein antigen presentation and antibody binding properties. These epitopes can be used to develop more effective vaccines and identify neutralizing antibodies. We identified 405 viral peptides with good antigen presentation scores for both human MHC-I and MHC-II alleles, and two potential neutralizing B-cell epitopes near the 2019-nCoV spike protein receptor binding domain (440-460 and 494-506). Analyzing mutation profiles of 68 viral genomes from four continents, we identified 96 coding-change mutations. These mutations are more likely to occur in regions with good MHC-I presentation scores (p=0.02). No mutations are present near the spike protein receptor binding domain. We validated our computational pipeline with SARS-CoV experimental data.

URL: http://biorxiv.org/content/early/2020/02/21/2020.02.19.955484.abstract
DOI: 10.1101/2020.02.19.955484

20. **Fast E, Chen B. Potential T-cell and B-cell Epitopes of 2019-nCoV. *bioRxiv*. 2020:2020.02.19.955484. DOI:** 10.1101/2020.02.19.955484

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URL: http://biorxiv.org/content/early/2020/02/21/2020.02.19.955484.abstract
DOI: 10.1101/2020.02.19.955484

21. **Fu J, Chen R, Hu J, et al. Identification of a Novel Linear B-Cell Epitope on the Nucleocapsid Protein of Porcine Deltacoronavirus. *International journal of molecular sciences*. 2020;21(2):E648. DOI:** 10.3390/ijms21020648

Porcine deltacoronavirus (PDCoV), first identified in 2012, is a swine enteropathogen now found in many countries. The nucleocapsid (N) protein, a core component of PDCoV, is essential for virus replication and is a significant candidate in the development of diagnostics for PDCoV. In this study, monoclonal antibodies (mAbs) were generated and tested for reactivity with three truncations of the full protein (N1, N2, N3) that contained partial overlaps; of the five monoclonals chosen tested, each reacted with only the N3 truncation. The antibody designated 4E88 had highest binding affinity with the N protein and was chosen for in-depth examination. The 4E88 epitope was located to amino acids 308-AKPKQQKKPKK-318 by testing the 4E88 monoclonal for reactivity with a series of N3 truncations, then the minimal epitope, 309-KPKQQKKPK-317 (designated EP-4E88), was pinpointed by testing the 4E88 monoclonal for reactivity with a series of synthetic peptides of this region. Homology analysis showed that the EP-4E88 sequence is highly conserved among PDCoV strains, and also shares high similarity with sparrow coronavirus (HKU17), Asian leopard cat coronavirus (ALCCoV), quail coronavirus (UAE-HKU30), and sparrow deltacoronavirus (SpDCoV). Of note, the PDCoV EP-4E88 sequence shared very low similarity (<22.2%) with other porcine coronaviruses (PEDV, TGEV, PRCV, SADS-CoV, PHEV), demonstrating that it is an epitope that can be used for distinguishing PDCoV and other porcine coronavirus. 3D structural analysis revealed that amino acids of EP-4E88 were in close proximity and may be exposed on the surface of the N protein.

URL: https://www.ncbi.nlm.nih.gov/pubmed/31963776
DOI: 10.3390/ijms21020648

22. **Gilchuk P, Murin CD, Milligan JC, et al. Analysis of a Therapeutic Antibody Cocktail Reveals Determinants for Cooperative and Broad Ebolavirus Neutralization. *Immunity*. 2020;52(2):388-403.e12. DOI:** 10.1016/j.immuni.2020.01.001; 11
10.1016/j.immuni.2020.01.001
Structural principles underlying the composition of protective antiviral monoclonal antibody (mAb) cocktails are poorly defined. Here, we exploited antibody cooperativity to develop a therapeutic mAb cocktail against Ebola virus. We systematically analyzed the antibody repertoire in human survivors and identified a pair of potently neutralizing mAbs that cooperatively bound to the ebolavirus glycoprotein (GP). High-resolution structures revealed that in a two-antibody cocktail, molecular mimicry was a major feature of mAb-GP interactions. Broadly neutralizing mAb rEBOV-520 targeted a conserved epitope on the GP base region. mAb rEBOV-548 bound to a glycan cap epitope, possessed neutralizing and Fc-mediated effector function activities, and potentiated neutralization by rEBOV-520. Remodeling of the glycan cap structures by the cocktail enabled enhanced GP binding and virus neutralization. The cocktail demonstrated resistance to virus escape and protected non-human primates (NHPs) against Ebola virus disease. These data illuminate structural principles of antibody cooperativity with implications for development of antiviral immunotherapeutics.; Structural principles underlying the composition of protective antiviral monoclonal antibody (mAb) cocktails are poorly defined. Here, we exploited antibody cooperativity to develop a therapeutic mAb cocktail against Ebola virus. We systematically analyzed the antibody repertoire in human survivors and identified a pair of potently neutralizing mAbs that cooperatively bound to the ebolavirus glycoprotein (GP). High-resolution structures revealed that in a two-antibody cocktail, molecular mimicry was a major feature of mAb-GP interactions. Broadly neutralizing mAb rEBOV-520 targeted a conserved epitope on the GP base region. mAb rEBOV-548 bound to a glycan cap epitope, possessed neutralizing and Fc-mediated effector function activities, and potentiated neutralization by rEBOV-520. Remodeling of the glycan cap structures by the cocktail enabled enhanced GP binding and virus neutralization. The cocktail demonstrated resistance to virus escape and protected non-human primates (NHPs) against Ebola virus disease. These data illuminate structural principles of antibody cooperativity with implications for development of antiviral immunotherapeutics.

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DOI: 10.1016/j.immuni.2020.01.001; 11
10.1016/j.immuni.2020.01.001

23. **Han Y, Jiang M, Xia D, et al. COVID-19 in a patient with long-term use of glucocorticoids: A study of a familial cluster. *Clinical Immunology*. 2020:108413-. DOI:** https://doi.org/10.1016/j.clim.2020.108413
Clusters of patients with novel coronavirus disease 2019 (COVID-19) have been successively reported globally. Studies show clear person-to-person transmission. The average incubation period is 2–14 days, and mostly 3–7 days. However, in some patients, this period may be longer. Here, we report a familial cluster of COVID-19 where a 47-year-old woman with long-term use of glucocorticoids did not develop any symptoms within the 14-day quarantine period but was confirmed with COVID-19 by tested positive of antibody on day 40 after she left Wuhan. Almost at the same time, her father and sister were diagnosed with COVID-19. The results suggest that the long-term use of glucocorticoids might cause atypical infections, a long incubation period, and extra transmission of COVID-19.

URL: http://www.sciencedirect.com/science/article/pii/S1521661620302059
DOI: https://doi.org/10.1016/j.clim.2020.108413

24. **Holland M, Negrón D, Mitchell S, et al. BioLaboro: A bioinformatics system for detecting molecular assay signature erosion and designing new assays in response to emerging and reemerging pathogens. *bioRxiv*. 2020:2020.04.08.031963. DOI:** 10.1101/2020.04.08.031963
Background Emerging and reemerging infectious diseases such as the novel Coronavirus disease, COVID-19 and Ebola pose a significant threat to global society and test the public health community's preparedness to rapidly respond to an outbreak with effective diagnostics and therapeutics. Recent advances in next generation sequencing technologies enable rapid generation of pathogen genome sequence data, within 24 hours of obtaining a sample in some instances. With these data, one can quickly evaluate the effectiveness of existing diagnostics and therapeutics using in silico approaches. The propensity of some viruses to rapidly accumulate mutations can lead to the failure of molecular detection assays creating the need for redesigned or newly designed assays.Results Here we describe a bioinformatics system named BioLaboro to identify signature regions in a given pathogen genome, design PCR assays targeting

those regions, and then test the PCR assays in silico to determine their sensitivity and specificity. We demonstrate BioLaboro with two use cases: Bombali Ebolavirus (BOMV) and the novel Coronavirus 2019 (SARS-CoV-2). For the BOMV, we analyzed 30 currently available real-time reverse transcription-PCR assays against the three available complete genome sequences of BOMV. Only two met our in silico criteria for successful detection and neither had perfect matches to the primer/probe sequences. We designed five new primer sets against BOMV signatures and all had true positive hits to the three BOMV genomes and no false positive hits to any other sequence. Four assays are closely clustered in the nucleoprotein gene and one is located in the glycoprotein gene. Similarly, for the SARS-CoV-2, we designed five highly specific primer sets that hit all 145 whole genomes (available as of February 28, 2020) and none of the near neighbors.ConclusionsHere we applied BioLaboro in two real-world use cases to demonstrate its capability; 1) to identify signature regions, 2) to assess the efficacy of existing PCR assays to detect pathogens as they evolve over time, and 3) to design new assays with perfect in silico detection accuracy, all within hours, for further development and deployment. BioLaboro is designed with a user-friendly graphical user interface for biologists with limited bioinformatics experience.Competing Interest Statementbpbase pair (bps, plural base pairs)BDBVBundibugyo ebolavirusBLASTBasic Local Alignment Search ToolBOMVBombali ebolavirusCFRCase Fatality RatioCOVID-19Coronavirus Disease 2019DNAdeoxyribonucleic acidDRCDemocratic Republic of the CongoEBOVZaire ebolavirusEMAEuropean Medicines AgencyEVDEbola Virus DiseaseFDAFood and Drug AdministrationFNFalse NegativeFPFalse PositiveGCguanine cytosineGPglycoproteinGUIGraphic User interfaceIDidentifiermAbmonoclonal antibodyMAFFTMultiple Sequence Alignment using Fast Fourier TransformMCMmedical countermeasureNnucleocapsid phosphoproteinN/ANot availableNCBINational Center for Biotechnology InformationNIAIDNational Institute of Allergy and Infectious DiseasesNPnucleoproteinNPC1Niemann-Pick C1ORFOpen Reading FramePALMPamoja Tulinde Maisha studyPCRpolymerase chain reactionPSETPCR signature erosion toolRAMrandom access memoryRESTVReston ebolavirusRNAribonucleic acidrRT-PCRreal-time reverse-transcription polymerase chain reactionrVSVrecombinant vesicular stomatitis virusSARSsevere acute respiratory syndromeSNPsingle nucleotide proteinSUDVSudan ebolavirusTAFVTaï Forest ebolavirusTNTrue NegativeTPTrue PositiveWGSWhole Genome SequenceWHOWorld Health Organization

URL: http://biorxiv.org/content/early/2020/04/10/2020.04.08.031963.abstract
DOI: 10.1101/2020.04.08.031963

25. **Hu L-J, Meng Y, Zhou Y-L, et al. Establishment and application of indirect ELISA for detection of bovine coronavirus antibody. *Weishengwuxue Tongbao* = *Microbiology*. 2020(1):330.**

[Background] Bovine coronavirus(BCoV) is one of the main causes of neonatal calf death, and effective detection is the prerequisite to prevent and control the disease. [Objective] At present, BCoV ELISA detection method has some defects, such as low sensitivity, instability and so on. This study aims to improve these defections to establish indirect ELISA detection method. [Methods] The method of indirect ELISA was established by using the soluble recombinant N protein of the epidemic BCoV-CD strain as an antigen. The epitope of the N protein was predicted by DNAStar soft, and was prepared by prokaryotic expression in the non-denatured condition. The seroepidemiological investigation of BCoV infection in Heilongjiang province in recent 5 years was carried out by using this method. [Results] The optimum working conditions of the ELISA method were as follows: the coating solution was 50 mmol/L pH 9.6 carbonate, and the antigen coating concentration was 2.5 μg/mL; The sample diluent was PBST, the dilution concentration was 1 μg/mL, and incubated at 37 °C for 1.5 h; The dilution concentration of HRP-labeled secondary antibody was 1:7 500, and incubated at 37 °C for 1.0 h; The blocked condition was 1% gelatin at 37 °C for 30 minutes. The negative-positive cut off value was 0.225. The method had no cross-reaction with positive serum of bovine rotavirus, bovine viral diarrhea virus, bovine respiratory syncytial body, bovine infectious rhinotracheitis, bovine parainfluenza virus type 3 and Escherichia coli. The intra-and inter-assay coefficient of variation was less than 10%, and the coincident rate with virus neutralization test was 93.5%. The results showed that the positive rate of BCoV antibody was 98.84% in 603 serum samples of cows in some areas of Heilongjiang Province. [Conclusion] The ELISA method established in this study has strong specificity, high sensitivity and good stability, which provides a technical basis for the further development of ELISA kit.

URL: https://search.proquest.com/docview/2387271413?accountid=26724http://sfx.library.cdc.gov/cdc/?url_ver=Z39.88-2004&rft_val_fmt=info:ofi/fmt:kev:mtx:journal&genre=article&sid=ProQ:ProQ%3Amicrobiologyb&atitle=Establishment+and+application+of+indirect+ELISA+for+detection+of+bovine+coronavirus+antibody&title=Weishengwuxue+Tongbao+%3D+Microbiology&issn=02532654&date=2020-01-01&volume=&issue=1&spage=330&au=Hu%2C+Lin-Jie%3BMeng%2C+Ye%3BZhou%2C+Yu-Long%3BJia%2C+Wei-Qiang%3BZhai%2C+Hai-

Rui%3BWu%2C+Rui%3BHou%2C+Xi-Ling&isbn=&jtitle=Weishengwuxue+Tongbao+%3D+Microbiology&btitle=&rft_id=info:eric/&rft_id=info:doi/

DOI:

26. **Iacobucci G. Covid-19: government promises 100 000 tests per day in England by end of April. *BMJ*. 2020;369. DOI:** http://dx.doi.org/10.1136/bmj.m1392

In particular, the government's failure to fulfil promises to roll out testing for NHS staff has left medical leaders frustrated,1 with large numbers of doctors having to self-isolate because they are unsure if they have the virus. Scale up swab testing in Public Health England laboratories and NHS hospitals for those with a medical need and key workers to 25 000 a day by mid to late April Deliver increased commercial swab testing for critical key workers in the NHS, later expanding to key workers in other sectors Develop antibody blood tests, currently being tested for validation, but not yet launched Conduct surveillance testing to learn more about the virus's spread and help develop new tests and treatments Build mass testing capacity at "a completely new scale" by working with industry, academia, and the NHS John Newton, Public Health England's director of health improvement, has been appointed to help deliver the new plans. In a separate move the charity Sense About Science has written to the prime minister urging him to "start publishing the government's evolving plans for coronavirus testing."

URL: https://search.proquest.com/docview/2385884657?accountid=26724http://sfx.library.cdc.gov/cdc/?url_ver=Z39.88-2004&rft_val_fmt=info:ofi/fmt:kev:mtx:journal&genre=article&sid=ProQ:ProQ%3Asciencejournals&atitle=Covid-19%3A+government+promises+100+000+tests+per+day+in+England+by+end+of+April&title=BMJ+%3A+British+Medical+Journal+%28Online%29&issn=&date=2020-04-03&volume=369&issue=&spage=&au=Iacobucci%2C+Gareth&isbn=&jtitle=BMJ+%3A+British+Medical+Journal+%28Online%29&btitle=&rft_id=info:eric/&rft_id=info:doi/10.1136%2Fbmj.m1392

DOI: http://dx.doi.org/10.1136/bmj.m1392

27. **Imai Y. Treatment to prevent the development of severe COVID-19. *Proceedings for Annual Meeting of The Japanese Pharmacological Society*. 2020;93(0):2-ES. DOI:** 10.1254/jpssuppl.93.0_2-ES-4

The respiratory virus infection COVID-19 caused by the new coronavirus SARS-CoV2 has been reported in China since December 2019. It has been reported that COVID-19 tends to be more severe in the elderly and in patients with underlying diseases including diabetes, heart disease, and chronic lung disease. In severe cases, patients require intensive cares including mechanical ventilation in the ICUs. So far, no biomarker that predicts the severity, or no therapeutic strategies to prevent the development of severe diseases has been established. Pathology of severe COVID-19 has two aspects: viral overgrowth and excess pulmonary inflammation. For the former, clinical trials using existing drugs such as remdesivir (nucleic acid drug), lopinavir/ritonavir combination drug (protease inhibitor), favipiravir (polymerase inhibitor), and interferon (antiviral drugs) are being conducted in patients with severe COVID-19 in China. Furthermore the interest has been focused on immune globulin preparations enriched with pathogen-specific antibodies collected from the plasma of recovered patients. For the latter, clinical studies using tocilizumab (IL-6 receptor antibody) and ACE2 protein have been conducted with the purpose of reducing excessive inflammation of the lung. In addition, single cell analysis of immune cells and comprehensive reperto ire analysis of TCR/BCR using patient blood are in progress overseas, which are useful to elucidate the mechanism of the severe disease progression and identify the useful biomarkers for it.

URL: https://www.jstage.jst.go.jp/article/jpssuppl/93/0/93_2-ES-4/_article/-char/ja/

DOI: 10.1254/jpssuppl.93.0_2-ES-4

28. **Jiang H-w, Li Y, Zhang H-n, et al. Global profiling of SARS-CoV-2 specific IgG/ IgM responses of convalescents using a proteome microarray. *medRxiv*. 2020:2020.03.20.20039495. DOI:** 10.1101/2020.03.20.20039495

COVID-19 is caused by SARS-CoV-2, and has become a global pandemic. There is no highly effective medicine or vaccine, most of the patients were recovered by their own immune response, especially the virus specific IgG and IgM responses. However, the IgG/IgM responses is barely known. To enable the global understanding of SARS-CoV-2 specific IgG/IgM responses, a SARS-CoV-2 proteome microarray with 18 out of the 28 predicted proteins was constructed. The microarray was applied to profile the IgG/IgM responses with 29 convalescent sera. The results suggest that at the convalescent phase 100% of patients had IgG/IgM responses to SARS-CoV-2, especially to protein N, S1 but not S2. S1 purified from mammalian cell demonstrated the highest performance to differentiate COVID-19 patients from controls. Besides protein N and S1, significant antibody responses to ORF9b and NSP5 were also identified. In-depth analysis showed that the level of S1 IgG positively correlate to age and the level of LDH (lactate dehydrogenase),

especially for women, while the level of S1 IgG negatively correlate to Ly% (Lymphocyte percentage). This study presents the first whole picture of the SARS-CoV-2 specific IgG/ IgM responses, and provides insights to develop precise immuno-diagnostics, effective treatment and vaccine.Competing Interest StatementThe authors have declared no competing interest.Funding StatementThis work was partially supported by National Key Research and Development Program of China Grant (No. 2016YFA0500600), National Natural Science Foundation of China (No. 31970130, 31600672, 31670831, and 31370813).Author DeclarationsAll relevant ethical guidelines have been followed; any necessary IRB and/or ethics committee approvals have been obtained and details of the IRB/oversight body are included in the manuscript.YesAll necessary patient/participant consent has been obtained and the appropriate institutional forms have been archived.YesI understand that all clinical trials and any other prospective interventional studies must be registered with an ICMJE-approved registry, such as ClinicalTrials.gov. I confirm that any such study reported in the manuscript has been registered and the trial registration ID is provided (note: if posting a prospective study registered retrospectively, please provide a statement in the trial ID field explaining why the study was not registered in advance).Yes I have followed all appropriate research reporting guidelines and uploaded the relevant EQUATOR Network research reporting checklist(s) and other pertinent material as supplementary files, if applicable.YesThe SARS-CoV-2 proteome microarray data are deposited on Protein Microarray Database (http://www.proteinmicroarray.cn) under the accession number PMDE241. Additional data related to this paper may be requested from the authors.http://www.proteinmicroarray.cn/index.php/experiment/detail?experiment_id=241

URL: http://medrxiv.org/content/early/2020/03/27/2020.03.20.20039495.abstract
DOI: 10.1101/2020.03.20.20039495

29. **Joyce MG, Sankhala RS, Chen W-H, et al. A Cryptic Site of Vulnerability on the Receptor Binding Domain of the SARS-CoV-2 Spike Glycoprotein. *bioRxiv*. 2020:2020.03.15.992883. DOI:** 10.1101/2020.03.15.992883

SARS-CoV-2 is a zoonotic virus that has caused a pandemic of severe respiratory diseaseâ€"COVID-19â€"within several months of its initial identification. Comparable to the first SARS-CoV, this coronaviruses surface Spike (S) glycoprotein mediates cell entry via the human ACE-2 receptor, and, thus, is the principal target for the development of vaccines and immunotherapeutics. Molecular information on the SARS-CoV-2 S glycoprotein remains limited. Here we report the crystal structure of the SARS-CoV-2 S receptor-binding-domain (RBD) at a the highest resolution to date, of 1.95 Ã…. We identified a set of SARS-reactive monoclonal antibodies with cross-reactivity to SARS-CoV-2 RBD and other betacoronavirus S glycoproteins. One of these antibodies, CR3022, was previously shown to synergize with antibodies that target the ACE-2 binding site on the SARS-CoV RBD and reduce viral escape capacity. We determined the structure of CR3022, in complex with the SARS-CoV-2 RBD, and defined a broadly reactive epitope that is highly conserved across betacoronaviruses. This epitope is inaccessible in the closed prefusion S structure, but is accessible in open conformations. This first-ever resolution of a human antibody in complex with SARS-CoV-2 and the broad reactivity of this set of antibodies to a conserved betacoronavirus epitope will allow antigenic assessment of vaccine candidates, and provide a framework for accelerated vaccine, immunotherapeutic and diagnostic strategies against SARS-CoV-2 and related betacoronaviruses.

URL: http://biorxiv.org/content/early/2020/03/17/2020.03.15.992883.abstract
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30. **Joyce MG, Sankhala RS, Chen W-H, et al. A Cryptic Site of Vulnerability on the Receptor Binding Domain of the SARS-CoV-2 Spike Glycoprotein. *bioRxiv*. 2020:2020.03.15.992883. DOI:** 10.1101/2020.03.15.992883

SARS-CoV-2 is a zoonotic virus that has caused a pandemic of severe respiratory disease—COVID-19—within several months of its initial identification. Comparable to the first SARS-CoV, this novel coronavirus's surface Spike (S) glycoprotein mediates cell entry via the human ACE-2 receptor, and, thus, is the principal target for the development of vaccines and immunotherapeutics. Molecular information on the SARS-CoV-2 S glycoprotein remains limited. Here we report the crystal structure of the SARS-CoV-2 S receptor-binding-domain (RBD) at a the highest resolution to date, of 1.95 Å. We identified a set of SARS-reactive monoclonal antibodies with cross-reactivity to SARS-CoV-2 RBD and other betacoronavirus S glycoproteins. One of these antibodies, CR3022, was previously shown to synergize with antibodies that target the ACE-2 binding site on the SARS-CoV RBD and reduce viral escape capacity. We determined the structure of CR3022, in complex with the SARS-CoV-2 RBD, and defined a broadly reactive epitope that is highly conserved across betacoronaviruses. This epitope is inaccessible in the "closed" prefusion S structure, but is accessible in "open" conformations. This first-ever resolution of a human antibody in complex with SARS-CoV-2 and the broad reactivity of this set of antibodies to a conserved betacoronavirus epitope will allow antigenic assessment of vaccine

candidates, and provide a framework for accelerated vaccine, immunotherapeutic and diagnostic strategies against SARS-CoV-2 and related betacoronaviruses.HIGHLIGHTS High resolution structure of the SARS-CoV-2 Receptor-Binding-Domain (RBD).Recognition of the SARS-CoV-2 RBD by SARS-CoV antibodies.Structure of the SARS-COV-2 RBD in complex with antibody CR3022.Identification of a cryptic site of vulnerability on the SARS-CoV-2 Spike.

URL: http://biorxiv.org/content/early/2020/03/17/2020.03.15.992883.abstract
DOI: 10.1101/2020.03.15.992883

31. **Khan S, Nakajima R, Jain A, et al. Analysis of Serologic Cross-Reactivity Between Common Human Coronaviruses and SARS-CoV-2 Using Coronavirus Antigen Microarray. *bioRxiv*. 2020:2020.03.24.006544. DOI:** 10.1101/2020.03.24.006544

The current practice for diagnosis of SARS-CoV-2 infection relies on PCR testing of nasopharyngeal or respiratory specimens in a symptomatic patient at high epidemiologic risk. This testing strategy likely underestimates the true prevalence of infection, creating the need for serologic methods to detect infections missed by the limited testing to date. Here, we describe the development of a coronavirus antigen microarray containing immunologically significant antigens from SARS-CoV-2, in addition to SARS-CoV, MERS-CoV, common human coronavirus strains, and other common respiratory viruses. A preliminary study of human sera collected prior to the SARS-CoV-2 pandemic demonstrates overall high IgG reactivity to common human coronaviruses and low IgG reactivity to epidemic coronaviruses including SARS-CoV-2, with some cross-reactivity of conserved antigenic domains including S2 domain of spike protein and nucleocapsid protein. This array can be used to answer outstanding questions regarding SARS-CoV-2 infection, including whether baseline serology for other coronaviruses impacts disease course, how the antibody response to infection develops over time, and what antigens would be optimal for vaccine development.

URL: http://biorxiv.org/content/early/2020/03/25/2020.03.24.006544.abstract
DOI: 10.1101/2020.03.24.006544

32. **Kruse RL. Therapeutic strategies in an outbreak scenario to treat the novel coronavirus originating in Wuhan, China. *F1000Research*. 2020;9:72. DOI:** 10.12688/f1000research.22211.2

A novel coronavirus (2019-nCoV) originating in Wuhan, China presents a potential respiratory viral pandemic to the world population. Current efforts are focused on containment and quarantine of infected individuals. Ultimately, the outbreak could be controlled with a protective vaccine to prevent 2019-nCoV infection. While vaccine research should be pursued intensely, there exists today no therapy to treat 2019-nCoV upon infection, despite an urgent need to find options to help these patients and preclude potential death. Herein, I review the potential options to treat 2019-nCoV in patients, with an emphasis on the necessity for speed and timeliness in developing new and effective therapies in this outbreak. I consider the options of drug repurposing, developing neutralizing monoclonal antibody therapy, and an oligonucleotide strategy targeting the viral RNA genome, emphasizing the promise and pitfalls of these approaches. Finally, I advocate for the fastest strategy to develop a treatment now, which could be resistant to any mutations the virus may have in the future. The proposal is a biologic that blocks 2019-nCoV entry using a soluble version of the viral receptor, angiotensin-converting enzyme 2 (ACE2), fused to an immunoglobulin Fc domain (ACE2-Fc), providing a neutralizing antibody with maximal breath to avoid any viral escape, while also helping to recruit the immune system to build lasting immunity. The ACE2-Fc therapy would also supplement decreased ACE2 levels in the lungs during infection, thereby directly treating acute respiratory distress pathophysiology as a third mechanism of action. The sequence of the ACE2-Fc protein is provided to investigators, allowing its possible use in recombinant protein expression systems to start producing drug today to treat patients under compassionate use, while formal clinical trials are later undertaken. Such a treatment could help infected patients before a protective vaccine is developed and widely available in the coming months to year(s).

URL: https://pubmed.ncbi.nlm.nih.gov/32117569 https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7029759/
DOI: 10.12688/f1000research.22211.2

33. **Kruse RL. Therapeutic strategies in an outbreak scenario to treat the novel coronavirus originating in Wuhan, China [version 2; peer review: 2 approved]. *F1000Research*. 2020;9. DOI:** 10.12688/f1000research.22211.2

A novel coronavirus (2019-nCoV) originating in Wuhan, China presents a potential respiratory viral pandemic to the world population. Current efforts are focused on containment and quarantine of infected individuals. Ultimately, the outbreak could be controlled with a protective vaccine to prevent 2019-nCoV infection. While vaccine research should

be pursued intensely, there exists today no therapy to treat 2019-nCoV upon infection, despite an urgent need to find options to help these patients and preclude potential death. Herein, I review the potential options to treat 2019-nCoV in patients, with an emphasis on the necessity for speed and timeliness in developing new and effective therapies in this outbreak. I consider the options of drug repurposing, developing neutralizing monoclonal antibody therapy, and an oligonucleotide strategy targeting the viral RNA genome, emphasizing the promise and pitfalls of these approaches. Finally, I advocate for the fastest strategy to develop a treatment now, which could be resistant to any mutations the virus may have in the future. The proposal is a biologic that blocks 2019-nCoV entry using a soluble version of the viral receptor, angiotensin-converting enzyme 2 (ACE2), fused to an immunoglobulin Fc domain (ACE2-Fc), providing a neutralizing antibody with maximal breath to avoid any viral escape, while also helping to recruit the immune system to build lasting immunity. The ACE2-Fc therapy would also supplement decreased ACE2 levels in the lungs during infection, thereby directly treating acute respiratory distress pathophysiology as a third mechanism of action. The sequence of the ACE2-Fc protein is provided to investigators, allowing its possible use in recombinant protein expression systems to start producing drug today to treat patients under compassionate use, while formal clinical trials are later undertaken. Such a treatment could help infected patients before a protective vaccine is developed and widely available in the coming months to year(s).

URL: https://doaj.org/article/33b8fde0a6f34e978d3470caa9a491b5
DOI: 10.12688/f1000research.22211.2

34. **Ku J, Kim S, Park J, et al. Reactive Polymer Targeting dsRNA as Universal Virus Detection Platform with Enhanced Sensitivity. *Biomacromolecules*. 2020. DOI:** 10.1021/acs.biomac.0c00379

Reactive poly(pentafluorophenyl acrylate) (PPFPA) grafted surfaces offer a versatile platform to immobilize biomolecules. Here, we utilize PPFPA grafted surface and double-stranded RNA (dsRNA) recognizing J2 antibody to construct a universal virus detection platform with enhanced sensitivity. PPFPA on silicon substrates are prepared, and surface hydrophilicity is modulated by partial substitution of the pentafluorophenyl units with poly(ethylene glycol). Following dsRNA antibody immobilization, the prepared surfaces can distinguish long dsRNAs from single-stranded RNAs of the same length and short dsRNAs. As long dsRNAs are common byproducts of viral transcription/replication, these surfaces can detect the presence of different kinds of viruses without prior knowledge of their genomic sequences. To increase dsRNA detection sensitivity, a two-step method is devised where the captured dsRNAs are visualized with multiple fluorophore-tagged J2 antibodies. We show that the developed platform can differentiate foreign long dsRNAs from cellular dsRNAs and other biomolecules present in the cell lysate. Moreover, when tested against cells infected with hepatitis A or C viruses, both viruses are successfully detected using a single platform. Our study shows that the developed PPFPA platform immobilized with J2 antibody can serve as a primary diagnostic tool to determine the infection status for a wide range of viruses.

URL: https://doi.org/10.1021/acs.biomac.0c00379
DOI: 10.1021/acs.biomac.0c00379

35. **Leist SR, Cockrell AS. Genetically Engineering a Susceptible Mouse Model for MERS-CoV-Induced Acute Respiratory Distress Syndrome. *Methods in molecular biology (Clifton, NJ)*. 2020;2099:137-59. DOI:** 10.1007/978-1-0716-0211-9_12

Since 2012, monthly cases of Middle East respiratory syndrome coronavirus (MERS-CoV) continue to cause severe respiratory disease that is fatal in ~35% of diagnosed individuals. The ongoing threat to global public health and the need for novel therapeutic countermeasures have driven the development of animal models that can reproducibly replicate the pathology associated with MERS-CoV in human infections. The inability of MERS-CoV to replicate in the respiratory tracts of mice, hamsters, and ferrets stymied initial attempts to generate small animal models. Identification of human dipeptidyl peptidase IV (hDPP4) as the receptor for MERS-CoV infection opened the door for genetic engineering of mice. Precise molecular engineering of mouse DPP4 (mDPP4) with clustered regularly interspaced short palindromic repeats (CRISPR)/Cas9 technology maintained inherent expression profiles, and limited MERS-CoV susceptibility to tissues that naturally express mDPP4, notably the lower respiratory tract wherein MERS-CoV elicits severe pulmonary pathology. Here, we describe the generation of the 288-330(+/+) MERS-CoV mouse model in which mice were made susceptible to MERS-CoV by modifying two amino acids on mDPP4 (A288 and T330), and the use of adaptive evolution to generate novel MERS-CoV isolates that cause fatal respiratory disease. The 288-330(+/+) mice are currently being used to evaluate novel drug, antibody, and vaccine therapeutic countermeasures for MERS-CoV. The chapter starts with a historical perspective on the emergence of MERS-CoV and animal models evaluated for MERS-

CoV pathogenesis, and then outlines the development of the 288-330(+/+) mouse model, assays for assessing a MERS-CoV pulmonary infection in a mouse model, and describes some of the challenges associated with using genetically engineered mice.

URL: https://www.ncbi.nlm.nih.gov/pubmed/31883094
DOI: 10.1007/978-1-0716-0211-9_12

36. **Li E, Yan F, Huang P, et al. Characterization of the Immune Response of MERS-CoV Vaccine Candidates Derived from Two Different Vectors in Mice. *Viruses*. 2020;12(1):20. DOI:** https://dx.doi.org/10.3390/v12010125

Middle East respiratory syndrome (MERS) is an acute, high-mortality-rate, severe infectious disease caused by an emerging MERS coronavirus (MERS-CoV) that causes severe respiratory diseases. The continuous spread and great pandemic potential of MERS-CoV make it necessarily important to develop effective vaccines. We previously demonstrated that the application of Gram-positive enhancer matrix (GEM) particles as a bacterial vector displaying the MERS-CoV receptor-binding domain (RBD) is a very promising MERS vaccine candidate that is capable of producing potential neutralization antibodies. We have also used the rabies virus (RV) as a viral vector to design a recombinant vaccine by expressing the MERS-CoV S1 (spike) protein on the surface of the RV. In this study, we compared the immunological efficacy of the vaccine candidates in BALB/c mice in terms of the levels of humoral and cellular immune responses. The results show that the rabies virus vector-based vaccine can induce remarkably earlier antibody response and higher levels of cellular immunity than the GEM particles vector. However, the GEM particles vector-based vaccine candidate can induce remarkably higher antibody response, even at a very low dose of 1 micro g. These results indicate that vaccines constructed using different vaccine vector platforms for the same pathogen have different rates and trends in humoral and cellular immune responses in the same animal model. This discovery not only provides more alternative vaccine development platforms for MERS-CoV vaccine development, but also provides a theoretical basis for our future selection of vaccine vector platforms for other specific pathogens.

URL:

http://ovidsp.ovid.com/ovidweb.cgi?T=JS&CSC=Y&NEWS=N&PAGE=fulltext&D=prem&AN=31968702http://sfx.library.cdc.gov/cdc?sid=OVID:medline&id=pmid:31968702&id=10.3390%2Fv12010125&issn=1999-4915&isbn=&volume=12&issue=1&spage=&pages=&date=2020&title=Viruses&atitle=Characterization+of+the+Immune+Response+of+MERS-CoV+Vaccine+Candidates+Derived+from+Two+Different+Vectors+in+Mice.&aulast=Li&pid=%3Cauthor%3ELi+E%2CYan+F%2CHuang+P%2CChi+H%2CXu+S%2CLi+G%2CLiu+C%2CFeng+N%2CWang+H%2CZhao+Y%2CYang+S%2CXia+X%3C%2Fauthor%3E&%3CAN%3E31968702%3C%2FAN%3E&%3CDT%3EJournal+Article%3C%2FDT%3E

DOI: https://dx.doi.org/10.3390/v12010125

37. **Li K, Li Z, Wohlford-Lenane C, et al. Single-Dose, Intranasal Immunization with Recombinant Parainfluenza Virus 5 Expressing Middle East Respiratory Syndrome Coronavirus (MERS-CoV) Spike Protein Protects Mice from Fatal MERS-CoV Infection. *mBio*. 2020;11. DOI:** 10.1128/mBio.00554-20.; ID: 10296
10.1128/mBio.00554-20

Middle East respiratory syndrome coronavirus (MERS-CoV) can cause severe and fatal acute respiratory disease in humans and remains endemic in the Middle East since first being identified in 2012. There are currently no approved vaccines or therapies available for MERS-CoV. In this study, we evaluated parainfluenza virus 5 (PIV5)-based vaccine expressing the MERS-CoV envelope spike protein (PIV5/MERS-S) in a human DPP4 knockin C57BL/6 congenic mouse model (hDPP4 KI). Following a single-dose intranasal immunization, PIV5-MERS-S induced neutralizing antibody and robust T cell responses in hDPP4 KI mice. A single intranasal administration of 10(4) PFU PIV5-MERS-S provided complete protection against a lethal challenge with mouse-adapted MERS-CoV (MERS(MA)6.1.2) and improved virus clearance in the lung. In comparison, single-dose intramuscular immunization with 10(6) PFU UV-inactivated MERS(MA)6.1.2 mixed with Imject alum provided protection to only 25% of immunized mice. Intriguingly, an influx of eosinophils was observed only in the lungs of mice immunized with inactivated MERS-CoV, suggestive of a hypersensitivity-type response. Overall, our study indicated that PIV5-MERS-S is a promising effective vaccine candidate against MERS-CoV infection.IMPORTANCE MERS-CoV causes lethal infection in humans, and there is no vaccine. Our work demonstrates that PIV5 is a promising vector for developing a MERS vaccine. Furthermore, success of PIV5-based MERS vaccine can be employed to develop a vaccine for emerging CoVs such as SARS-CoV-2, which causes COVID-19.

URL: DOI: 10.1128/mBio.00554-20.; ID: 10296

10.1128/mBio.00554-20

38. **Li Y, Zhang J, Wang N, et al. Therapeutic Drugs Targeting 2019-nCoV Main Protease by High-Throughput Screening. *bioRxiv*. 2020:2020.01.28.922922. DOI:** 10.1101/2020.01.28.922922

2019 Novel Coronavirus (2019-nCoV) is a virus identified as the cause of the outbreak of pneumonia first detected in Wuhan, China. Investigations on the transmissibility, severity, and other features associated with this virus are ongoing. Currently, there is no vaccine or therapeutic antibody to prevent the infection, and more time is required to develop an effective immune strategy against the pathogen. In contrast, specific inhibitors targeting th e key protease involved in replication and proliferation of the virus are the most effective means to alleviate the epidemic. The main protease of SARS-CoV is essential for the life cycle of the virus, which showed 96.1% of similarity with the main proteas eof 2019-nCoV, is considered to be an attractive target for drug development. In this study, we have identified 4 small molecular drugs with high binding capacity with SARS-CoV main protease by high-throughput screening based on the 8,000 clinical drug libraries, all these drugs have been widely used in clinical applications with guaranteed safety, which may serve as promising candidates to treat the infection of 2019-nCoV.

URL: http://biorxiv.org/content/early/2020/01/30/2020.01.28.922922.abstract
DOI: 10.1101/2020.01.28.922922

39. **Li Z, Yi Y, Luo X, et al. Development and Clinical Application of A Rapid IgM-IgG Combined Antibody Test for SARS-CoV-2 Infection Diagnosis. *Journal of medical virology*. 2020:10.1002/jmv.25727. DOI:** 10.1002/jmv.25727

The outbreak of the novel coronavirus disease (COVID-19) quickly spread all over China and to more than 20 other countries. Although the virus (SARS-Cov-2) nucleic acid RT-PCR test has become the standard method for diagnosis of SARS-CoV-2 infection, these real-time PCR test kits have many limitations. In addition, high false negative rates were reported. There is an urgent need for an accurate and rapid test method to quickly identify large number of infected patients and asymptomatic carriers to prevent virus tran smission and assure timely treatment of patients. We have developed a rapid and simple point-of-care lateral flow immunoassay which can detect IgM and IgG antibodies simultaneously against SARS-CoV-2 virus in human blood within 15 minutes which can detect patients at different infection stages. With this test kit, we carried out clinical studies to validate its clinical efficacy uses. The clinical detection sensitivity and specificity of this test were measured using blood samples collected from 397 PCR con firmed COVID-19 patients and 128 negative patients at 8 different clinical sites. The overall testing sensitivity was 88.66% and specificity was 90.63%. In addition, we evaluated clinical diagnosis results obtained from different types of venous and fingerstick blood samples. The results indicated great detection consistency among samples from fingerstick blood, serum and plasma of venous blood. The IgM-IgG combined assay has better utility and sensitivity compared with a single IgM or IgG test. It can be used for the rapid screening of SARS-CoV-2 carriers, symptomatic or asymptomatic, in hospitals, clinics, and test laboratories. This article is protected by copyright. All rights reserved.

URL: https://pubmed.ncbi.nlm.nih.gov/32104917
DOI: 10.1002/jmv.25727

40. **Lin D, Liu L, Zhang M, et al. Evaluations of serological test in the diagnosis of 2019 novel coronavirus (SARS-CoV-2) infections during the COVID-19 outbreak. *medRxiv*. 2020:2020.03.27.20045153. DOI:** 10.1101/2020.03.27.20045153

The ongoing SARS-CoV-2 outbreak has killed over twenty-one thousand and sickened over four hundred thousand people worldwide, posing a great challenge to global public health. A sensitive and accurate diagnosis method will substantially help to control disease expansion. Here, we developed a chemiluminescence-immunoassay method based on the recombinant nucleocapsid antigen and the magnetic beads for diagnosis of SARS-CoV-2 infections and surveillance of antibody changing pattern. Serums from 29 healthy individuals, 51 tuberculosis patients, and 79 SARS-CoV-2 confirmed patients were employed to evaluate the performance of this approach. Compared to the IgM testing, the IgG testing was more reliable in which it identified 65 SARS-CoV-2 infections from the 79 confirmed patients and only two false-positive cases from the 80 control group with a sensitivity and specificity reaching 82.28% and 97.5%, respectively. However, only a slight difference (not statistically significant) in the detected cases of SARS-CoV-2 infections was observed between the IgM and IgG testing manner in patients at a different time of onset of disease. A performance comparison between an ELISA kit using the same nucleocapsid antigen and our chemiluminescence method was undertaken. The same false-positive cases were seen in both methods from the

paired control group, while ELISA kit can only detect half of the SARS-CoV-2 infections from paired SARS-CoV-2 confirmed patients group than that of the chemiluminescence method, indicating a higher performance for the chemiluminescence-immunoassay approach. Together, our studies provide a useful and valuable serological testing tool for the diagnosis of SARS-CoV-2 infections in the community.Competing Interest StatementThe authors have declared no competing interest.Funding StatementThis work is supported by Guangdong Provincial Science and Technology Program (No. 2019b030301009), the National Natural Science Funds of China (81802060), the start-up funding of Shenzhen University and the National Science and Technology Major Project (2017ZX10201301).Author DeclarationsAll relevant ethical guidelines have been followed; any necessary IRB and/or ethics committee approvals have been obtained and details of the IRB/oversight body are included in the manuscript.YesAll necessary patient/participant consent has been obtained and the appropriate institutional forms have been archived.YesI understand that all clinical trials and any other prospective interventional studies must be registered with an ICMJE-approved registry, such as ClinicalTrials.gov. I confirm that any such study reported in the manuscript has been registered and the trial registration ID is provided (note: if posting a prospective study registered retrospectively, please provide a statement in the trial ID field explaining why the study was not registered in advance).Yes I have followed all appropriate research reporting guidelines and uploaded the relevant EQUATOR Network research reporting checklist(s) and other pertinent material as supplementary files, if applicable.YesAll the data referred to in the manuscript has been included in the main text and supplementary files.

URL: http://medrxiv.org/content/early/2020/03/30/2020.03.27.20045153.abstract
DOI: 10.1101/2020.03.27.20045153

41. **Liu L, Liu W, Wang S, et al. A preliminary study on serological assay for severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) in 238 admitted hospital patients. *medRxiv*. 2020:2020.03.06.20031856. DOI:** 10.1101/2020.03.06.20031856

Background The outbreak of the recently emerged novel corona virus disease 2019 (COVID-19) poses a challenge for public health laboratories. We aimed to evaluate the diagnostic value of serological assay for SARS-CoV-2. Methods A newly-developed ELISA assay for IgM and IgG antibodies against N protein of SARS-CoV-2 were used to screen the serums of 238 admitted hospital patients with confirmed or suspected SARS-CoV-2 infection from February 6 to February 14, 2020. SARS-CoV-2 RNA was detected by real time RT-PCR on pharyngeal swab specimens. Findings Of the 238 patients, 194 (81.5%) were detected to be antibody (IgM and/or IgG) positive, which was significantly higher than the positive rate of viral RNA (64.3%). There was no difference in the positive rate of antibody between the confirmed patients (83.0%, 127/153) and the suspected patients (78.8%, 67/85) whose nucleic acid tests were negative. After the patients were defined to the different stages of disease based on the day when the test samples were collected, the analysis results showed that the antibody positive rates were very low in the first five days after initial onset of symptoms, and then rapidly increased as the disease progressed. After 10 days, the antibody positive rates jumped to above 80% from less than 50%. On the contrary, the positive rates of viral RNA kept above 60% in the first 11 days after initial onset of symptoms, and then rapidly decreased. In addition, half of the suspected patients with symptoms for 6-10 days were detected to be antibody positive. Interpretation The suspected patients were most likely infected by SARS-CoV-2. Before the 11th day after initial onset of symptoms, nucleic acid test is important for confirmation of viral infection. The combination of serological assay can greatly improve the diagnostic efficacy. After that, the diagnosis for viral infection should be majorly dependent on serological assay. Keywords. SARS-CoV-2; diagnosis; serological assay; nucleic acid testCompeting Interest StatementThe authors have declared no competing interest.Funding StatementThis work was supported by the National Natural Science Foundation of China (81801984, 81830003); the National Key Research and Development Program of China (2019YFC130030); and the China Postdoctoral Science Foundation (2019M664008).Author DeclarationsAll relevant ethical guidelines have been followed; any necessary IRB and/or ethics committee approvals have been obtained and details of the IRB/oversight body are included in the manuscript.YesAll necessary patient/participant consent has been obtained and the appropriate institutional forms have been archived.YesI understand that all clinical trials and any other prospective interventional studies must be registered with an ICMJE-approved registry, such as ClinicalTrials.gov. I confirm that any such study reported in the manuscript has been registered and the trial registration ID is provided (note: if posting a prospective study registered retrospectively, please provide a statement in the trial ID field explaining why the study was not registered in advance).Yes I have followed all appropriate research reporting guidelines and uploaded the relevant EQUATOR Network research reporting checklist(s) and other pertinent material as supplementary files, if applicable.YesThe data used to support the findings of this study are included within the article.

URL: http://medrxiv.org/content/early/2020/03/08/2020.03.06.20031856.abstract
DOI: 10.1101/2020.03.06.20031856

42. **Liu R, Fu A, Deng Z, et al. Promising methods for detection of novel coronavirus SARS-CoV-2. *View*. 2020;1(1):e4-e. DOI:** 10.1002/viw2.4

Abstract A very recent outbreak of the novel coronavirus, COVID-19, in the city of Wuhan, China, in December 2019 and its subsequent spread within and across China have resulted in several deaths and infections. Presently, nucleic acid amplification test is essential for the confirmation of COVID infection. In this report, we summarized the six promising methods, including whole-genome sequencing, real-time reverse transcription polymerase chain reaction, nanopore target sequencing, antibody-based immunoassay techniques, use of paper-based biomolecular sensors, and the clustered regularly interspaced short palindromic repeats-Cas system-based technology, which can also be deployed for the detection of SARS-CoV-2. We further introduced the principles of these methods, discussed the scope and practicability of application of the available products and methods, and highlighted the potential approaches to develop additional products and techniques for early diagnosis of COVID-19.

URL: https://doi.org/10.1002/viw2.4
DOI: 10.1002/viw2.4

43. **Lu M, Liu Q, Wang X, et al. Development of an indirect ELISA for detecting porcine deltacoronavirus IgA antibodies. *Arch Virol*. 2020:10.1007/s00705-020. DOI:** 10.1007/s00705-020-04541-6

Porcine deltacoronavirus (PDCoV) is a novel coronavirus that can cause vomiting and watery diarrhea in pigs and death in piglets. Since PDCoV was first detected in 2009 in Hong Kong, the prevalence of PDCoV has increased in recent years, resulting in serious economic losses to the swine industry. The coronavirus spike (S) protein is an antigen that has been demonstrated to contain epitopes that induce neutralizing antibodies. The presence of serum and milk IgA antibodies against pathogens that replicate primarily on mucosal surfaces is important for mucosal immunity. Here, an indirect anti-PDCoV IgA antibody enzyme-linked immunosorbent assay (PDCoV S1 IgA ELISA) using the purified S1 portion of S protein as the coating antigen was developed to detect PDCoV IgA antibodies in serum and sow's milk. A receiver operating characteristic (ROC) curve analysis showed high specificity and sensitivity of the PDCoV-S1-IgA-ELISA based on samples confirmed by IFA. Anti-PDCoV IgA antibodies in 152 serum samples and 65 milk samples collected from six farms that had experienced diarrhea outbreaks within previous last two years were detected by this assay, and 62.5% of the serum samples and 100% of the milk samples were positive for PDCoV. The indirect ELISA method established in this study will provide a convenient tool for measurement of serum and milk IgA levels against PDCoV in pig herds, rapid detection of PDCoV infection in pigs, and evaluation of the immunogenicity of vaccines.

URL: https://pubmed.ncbi.nlm.nih.gov/32052195
DOI: 10.1007/s00705-020-04541-6

44. **Luo A. Positive SARS-Cov-2 test in a woman with COVID-19 at 22 days after hospital discharge: A case report. *Journal of Traditional Chinese Medical Sciences*. 2020. DOI:** https://doi.org/10.1016/j.jtcms.2020.04.001

Background In a few discharged patients with coronavirus disease 2019 (COVID-19), the nucleic acid test shows positive results again. Whether this is due to relapse of the disease, reinfection by the virus, or a false-positive result at hospital discharge is worth exploring. Case presentation A woman with COVID-19 was discharged from the hospital after integrative treatment with traditional Chinese and Western medicine because she met the discharge standards. However, she obtained positive results on a nucleic acid test 22 days later. Conclusion Based on this positive test result in a discharged patient with COVID-19, anal tests and coronavirus antibody tests should be combined with throat swab tests to further develop the diagnosis and discharge standards for patients with COVID-19.

URL: http://www.sciencedirect.com/science/article/pii/S2095754820300247
DOI: https://doi.org/10.1016/j.jtcms.2020.04.001

45. **Luo H, Tang QL, Shang YX, et al. Can Chinese Medicine Be Used for Prevention of Corona Virus Disease 2019 (COVID-19)? A Review of Historical Classics, Research Evidence and Current Prevention Programs Potent binding of 2019 novel coronavirus spike protein by a SARS coronavirus-specific human monoclonal antibody. *Chinese journal of integrative medicine Emerging microbes & infections*. 2020;9(1):382-5. DOI:** 10.1007/s11655-020-3192-6 10.1080/22221751.2020.1729069

OBJECTIVE: Since December 2019, an outbreak of corona virus disease 2019 (COVID-19) occurred in Wuhan, and rapidly spread to almost all parts of China. This was followed by prevention programs recommending Chinese medicine (CM) for the prevention. In order to provide evidence for CM recommendations, we reviewed ancient classics and human studies. METHODS: Historical records on prevention and treatment of infections in CM classics, clinical evidence of CM on the prevention of severe acute respiratory syndrome (SARS) and H1N1 influenza, and CM prevention programs issued by health authorities in China since the COVID-19 outbreak were retrieved from different databases and websites till 12 February, 2020. Research evidence included data from clinical trials, cohort or other population studies using CM for preventing contagious respiratory virus diseases. RESULTS: The use of CM to prevent epidemics of infectious diseases was traced back to ancient Chinese practice cited in Huangdi's Internal Classic (Huang Di Nei Jing) where preventive effects were recorded. There were 3 studies using CM for prevention of SARS and 4 studies for H1N1 influenza. None of the participants who took CM contracted SARS in the 3 studies. The infection rate of H1N1 influenza in the CM group was significantly lower than the non-CM group (relative risk 0.36, 95% confidence interval 0.24-0.52; n=4). For prevention of COVID-19, 23 provinces in China issued CM programs. The main principles of CM use were to tonify qi to protect from external pathogens, disperse wind and discharge heat, and resolve dampness. The most frequently used herbs included Radix astragali (Huangqi), Radix glycyrrhizae (Gancao), Radix saposhnikoviae (Fangfeng), Rhizoma Atractylodis Macrocephalae (Baizhu), Lonicerae Japonicae Flos (Jinyinhua), and Fructus forsythia (Lianqiao). CONCLUSIONS: Based on historical records and human evidence of SARS and H1N1 influenza prevention, Chinese herbal formula could be an alternative approach for prevention of COVID-19 in high-risk population. Prospective, rigorous population studies are warranted to confirm the potential preventive effect of CM. The newly identified 2019 novel coronavirus (2019-nCoV) has caused more than 11,900 laboratory-confirmed human infections, including 259 deaths, posing a serious threat to human health. Currently, however, there is no specific antiviral treatment or vaccine. Considering the relatively high identity of receptor-binding domain (RBD) in 2019-nCoV and SARS-CoV, it is urgent to assess the cross-reactivity of anti-SARS CoV antibodies with 2019-nCoV spike protein, which could have important implications for rapid development of vaccines and therapeutic antibodies against 2019-nCoV. Here, we report for the first time that a SARS-CoV-specific human monoclonal antibody, CR3022, could bind potently with 2019-nCoV RBD (KD of 6.3 nM). The epitope of CR3022 does not overlap with the ACE2 binding site within 2019-nCoV RBD. These results suggest that CR3022 may have the potential to be developed as candidate therapeutics, alone or in combination with other neutralizing antibodies, for the prevention and treatment of 2019-nCoV infections. Interestingly, some of the most potent SARS-CoV-specific neutralizing antibodies (e.g. m396, CR3014) that target the ACE2 binding site of SARS-CoV failed to bind 2019-nCoV spike protein, implying that the difference in the RBD of SARS-CoV and 2019-nCoV has a critical impact for the cross-reactivity of neutralizing antibodies, and that it is still necessary to develop novel monoclonal antibodies that could bind specifically to 2019-nCoV RBD.

URL: DOI: 10.1007/s11655-020-3192-6 10.1080/22221751.2020.1729069

46. **Lv H, Wu NC, Tsang OT-Y, et al. Cross-reactive antibody response between SARS-CoV-2 and SARS-CoV infections. *bioRxiv*. 2020:2020.03.15.993097. DOI:** 10.1101/2020.03.15.993097

The World Health Organization has recently declared the ongoing outbreak of COVID-19, which is caused by a novel coronavirus SARS-CoV-2, as pandemic. There is currently a lack of knowledge in the antibody response elicited from SARS-CoV-2 infection. One major immunological question is concerning the antigenic differences between SARS-CoV-2 and SARS-CoV. We address this question by using plasma from patients infected by SARS-CoV-2 or SARS-CoV, and plasma obtained from infected or immunized mice. Our results show that while cross-reactivity in antibody binding to the spike protein is common, cross-neutralization of the live viruses is rare, indicating the presence of non-neutralizing antibody response to conserved epitopes in the spike. Whether these non-neutralizing antibody responses will lead to antibody-dependent disease enhancement needs to be addressed in the future. Overall, this study not only addresses a fundamental question regarding the antigenicity differences between SARS-CoV-2 and SARS-CoV, but also has important implications in vaccine development.

URL: http://biorxiv.org/content/early/2020/03/17/2020.03.15.993097.abstract
DOI: 10.1101/2020.03.15.993097

47. **Lv H, Wu NC, Tsang OT-Y, et al. Cross-reactive antibody response between SARS-CoV-2 and SARS-CoV infections. *bioRxiv*. 2020:2020.03.15.993097. DOI:** 10.1101/2020.03.15.993097

The World Health Organization has recently declared the ongoing outbreak of COVID-19, which is caused by a novel coronavirus SARS-CoV-2, as pandemic. There is currently a lack of knowledge in the antibody response elicited from

SARS-CoV-2 infection. One major immunological question is concerning the antigenic differences between SARS-CoV-2 and SARS-CoV. We address this question by using plasma from patients infected by SARS-CoV-2 or SARS-CoV, and plasma obtained from infected or immunized mice. Our results show that while cross-reactivity in antibody binding to the spike protein is common, cross-neutralization of the live viruses is rare, indicating the presence of non-neutralizing antibody response to conserved epitopes in the spike. Whether these non-neutralizing antibody responses will lead to antibody-dependent disease enhancement needs to be addressed in the future. Overall, this study not only addresses a fundamental question regarding the antigenicity differences between SARS-CoV-2 and SARS-CoV, but also has important implications in vaccine development.

URL: http://biorxiv.org/content/early/2020/03/17/2020.03.15.993097.abstract
DOI: 10.1101/2020.03.15.993097

48. **Normile D. Singapore claims first use of antibody test to track coronavirus infections. *Science*. 2020.**
In what appears to be a first, disease trackers in Singapore have used an experimental antibody test for COVID-19 to confirm that a suspected patient was infected with the coronavirus. The patient was one of two people who together formed a missing link between two clusters of cases that each occurred in a Singaporean church. Researchers around the world are racing to develop antibody tests, also called serological tests, that can confirm whether someone was infected even after their immune system has cleared the virus that causes COVID-19. The group that developed the test, at Duke-NUS Medical School in Singapore, is among the front-runners, although its assay has to be validated before it is taken into production and deployed widely.

URL: https://www.sciencemag.org/news/2020/02/singapore-claims-first-use-antibody-test-track-coronavirus-infections
DOI:

49. **Normile D. Singapore claims first use of antibody test to track coronavirus infections | Science | AAAS. *Science Magazine*. 2020.**
In what appears to be a first, disease trackers in Singapore have used an experimental antibody test for COVID-19 to confirm that a suspected patient was infected with the coronavirus. The patient was one of two people who together formed a missing link between two clusters of cases that each occurred in a Singaporean church. Researchers around the world are racing to develop antibody tests, also called serological tests, that can confirm whether someone was infected even after their immune system has cleared the virus that causes COVID-19. The group that developed the test, at Duke-NUS Medical School in Singapore, is among the front-runners, although its assay has to be validated before it is taken into production and deployed widely.

URL: https://www.sciencemag.org/news/2020/02/singapore-claims-first-use-antibody-test-track-coronavirus-infections
DOI:

50. **Okba NMA, MÃ¼ller MA, Li W, et al. Severe Acute Respiratory Syndrome Coronavirus 2-Specific Antibody Responses in Coronavirus Disease 2019 Patients. *Emerg Infect Dis*. 2020;26. DOI:** 10.3201/eid2607.200841.; ID: 10316
10.3201/eid2607.200841
A new coronavirus, severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), has recently emerged to cause a human pandemic. Although molecular diagnostic tests were rapidly developed, serologic assays are still lacking, yet urgently needed. Validated serologic assays are needed for contact tracing, identifying the viral reservoir, and epidemiologic studies. We developed serologic assays for detection of SARS-CoV-2 neutralizing, spike protein-specific, and nucleocapsid-specific antibodies. Using serum samples from patients with PCR-confirmed SARS-CoV-2 infections, other coronaviruses, or other respiratory pathogenic infections, we validated and tested various antigens in different in-house and commercial ELISAs. We demonstrated that most PCR-confirmed SARS-CoV-2-infected persons seroconverted by 2 weeks after disease onset. We found that commercial S1 IgG or IgA ELISAs were of lower specificity, and sensitivity varied between the 2 assays; the IgA ELISA showed higher sensitivity. Overall, the validated assays described can be instrumental for detection of SARS-CoV-2-specific antibodies for diagnostic, seroepidemiologic, and vaccine evaluation studies.

URL: DOI: 10.3201/eid2607.200841.; ID: 10316
10.3201/eid2607.200841

51. **Okba NMA, Muller MA, Li W, et al. Severe Acute Respiratory Syndrome Coronavirus 2-Specific Antibody Responses in Coronavirus Disease 2019 Patients.** ***Emerging Infectious Diseases*****. 2020;26(7):08. DOI:** https://dx.doi.org/10.3201/eid2607.200841

A new coronavirus, severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), has recently emerged to cause a human pandemic. Although molecular diagnostic tests were rapidly developed, serologic assays are still lacking, yet urgently needed. Validated serologic assays are needed for contact tracing, identifying the viral reservoir, and epidemiologic studies. We developed serologic assays for detection of SARS-CoV-2 neutralizing, spike protein-specific, and nucleocapsid-specific antibodies. Using serum samples from patients with PCR-confirmed SARS-CoV-2 infections, other coronaviruses, or other respiratory pathogenic infections, we validated and tested various antigens in different in-house and commercial ELISAs. We demonstrated that most PCR-confirmed SARS-CoV-2-infected persons seroconverted by 2 weeks after disease onset. We found that commercial S1 IgG or IgA ELISAs were of lower specificity, and sensitivity varied between the 2 assays; the IgA ELISA showed higher sensitivity. Overall, the validated assays described can be instrumental for detection of SARS-CoV-2-specific antibodies for diagnostic, seroepidemiologic, and vaccine evaluation studies.

URL:

http://ovidsp.ovid.com/ovidweb.cgi?T=JS&CSC=Y&NEWS=N&PAGE=fulltext&D=medp&AN=32267220http://sfx.library.cdc.gov/cdc?sid=OVID:medline&id=pmid:32267220&id=10.3201%2Feid2607.200841&issn=1080-6040&isbn=&volume=26&issue=7&spage=&pages=&date=2020&title=Emerging+Infectious+Diseases&atitle=Severe+Acute+Respiratory+Syndrome+Coronavirus+2-Specific+Antibody+Responses+in+Coronavirus+Disease+2019+Patients.&aulast=Okba&pid=%3Cauthor%3EOkba+NMA%2CMuller+MA%2CLi+W%2CWang+C%2CGeurtsvanKessel+CH%2CCorman+VM%2CLamers+MM%2CSikkema+RS%2Cde+Bruin+E%2CChandler+FD%2CYazdanpanah+Y%2CLe+Hingrat+Q%2CDescamps+D%2CHouhou-Fidouh+N%2CReusken+CBEM%2CBosch+BJ%2CDrosten+C%2CKoopmans+MPG%2CHaagmans+BL%3C%2Fauthor%3E&%3CAN%3E32267220%3C%2FAN%3E&%3CDT%3EJournal+Article%3C%2FDT%3E

DOI: https://dx.doi.org/10.3201/eid2607.200841

52. **Okba NMA, Muller MA, Li W, et al. SARS-CoV-2 specific antibody responses in COVID-19 patients.** ***medRxiv*****. 2020:2020.03.18.20038059. DOI:** 10.1101/2020.03.18.20038059

A new coronavirus, SARS-CoV-2, has recently emerged to cause a human pandemic. Whereas molecular diagnostic tests were rapidly developed, serologic assays are still lacking, yet urgently needed. Validated serologic assays are important for contact tracing, identifying the viral reservoir and epidemiological studies. Here, we developed serological assays for the detection of SARS-CoV-2 neutralizing, spike- and nucleocapsid-specific antibodies. Using serum samples from patients with PCR-confirmed infections of SARS-CoV-2, other coronaviruses, or other respiratory pathogenic infections, we validated and tested various antigens in different in-house and commercial ELISAs. We demonstrate that most PCR-confirmed SARS-CoV-2 infected individuals seroconverted, as revealed by sensitive and specific in-house ELISAs. We found that commercial S1 IgG or IgA ELISAs were of lower specificity while sensitivity varied between the two, with IgA showing higher sensitivity. Overall, the validated assays described here can be instrumental for the detection of SARS-CoV-2-specific antibodies for diagnostic, seroepidemiological and vaccine evaluation studies.Competing Interest StatementThe authors have declared no competing interest.Funding StatementThis work was supported by the Zoonoses Anticipation and Preparedness Initiative (ZAPI project; IMI grant agreement no. 115760), with the assistance and financial support of IMI and the European Commission, in-kind contributions from EFPIA partners.Author DeclarationsAll relevant ethical guidelines have been followed; any necessary IRB and/or ethics committee approvals have been obtained and details of the IRB/oversight body are included in the manuscript.YesAll necessary patient/participant consent has been obtained and the appropriate institutional forms have been archived.YesI understand that all clinical trials and any other prospective interventional studies must be registered with an ICMJE-approved registry, such as ClinicalTrials.gov. I confirm that any such study reported in the manuscript has been registered and the trial registration ID is provided (note: if posting a prospective study registered retrospectively, please provide a statement in the trial ID field explaining why the study was not registered in advance).Yes I have followed all appropriate research reporting guidelines and uploaded the relevant EQUATOR Network research reporting checklist(s) and other pertinent material as supplementary files, if applicable.YesAll data referred to in the manuscript are available form corresponding authors upon reasonable request.

URL: http://medrxiv.org/content/early/2020/03/20/2020.03.18.20038059.abstract
DOI: 10.1101/2020.03.18.20038059

53. **Okba NMA, Widjaja I, Li W, et al. Serologic Detection of Middle East Respiratory Syndrome Coronavirus Functional Antibodies. *Emerging Infectious Diseases*. 2020;26(5):17. DOI:** https://dx.doi.org/10.3201/eid2605.190921

We developed and validated 2 species-independent protein-based assays to detect Middle East respiratory syndrome coronavirus functional antibodies that can block virus receptor-binding or sialic acid-attachment. Antibody levels measured in both assays correlated strongly with virus-neutralizing antibody titers, proving their use for serologic confirmatory diagnosis of Middle East respiratory syndrome.

URL:

http://ovidsp.ovid.com/ovidweb.cgi?T=JS&CSC=Y&NEWS=N&PAGE=fulltext&D=medp&AN=32150528http://sfx.library.cdc.gov/cdc?sid=OVID:medline&id=pmid:32150528&id=10.3201%2Feid2605.190921&issn=1080-6040&isbn=&volume=26&issue=5&spage=&pages=&date=2020&title=Emerging+Infectious+Diseases&atitle=Serologic+Detection+of+Middle+East+Respiratory+Syndrome+Coronavirus+Functional+Antibodies.&aulast=Okba&pid=%3Cauthor%3EOkba+NMA%2CWidjaja+I%2CLi+W%2CGeurtsvanKessel+CH%2CFarag+EABA%2CAl-Hajri+M%2CPark+WB%2COh+MD%2CReusken+CBME%2CKoopmans+MPG%2CBosch+BJ%2CHaagmans+BL%3C%2Fauthor%3E&%3CAN%3E32150528%3C%2FAN%3E&%3CDT%3EJournal+Article%3C%2FDT%3E

DOI: https://dx.doi.org/10.3201/eid2605.190921

54. **Okba NMA, Widjaja I, Li W, et al. Serologic Detection of Middle East Respiratory Syndrome Coronavirus Functional Antibodies. *Emerg Infect Dis*. 2020;26(5):10.3201/eid2605.190921. DOI:** 10.3201/eid2605.190921

We developed and validated 2 species-independent protein-based assays to detect Middle East respiratory syndrome coronavirus functional antibodies that can block virus receptor-binding or sialic acid-attachment. Antibody levels measured in both assays correlated strongly with virus-neutralizing antibody titers, proving their use for serologic confirmatory diagnosis of Middle East respiratory syndrome.

URL: https://pubmed.ncbi.nlm.nih.gov/32150528
DOI: 10.3201/eid2605.190921

55. **Pan Y, Li X, Yang G, et al. Serological immunochromatographic approach in diagnosis with SARS-CoV-2 infected COVID-19 patients. *medRxiv*. 2020:2020.03.13.20035428. DOI:** 10.1101/2020.03.13.20035428

An outbreak of new coronavirus SARS-CoV-2 was occurred in Wuhan, China and rapidly spread to other cities and nations. The standard diagnostic approach that widely adopted in the clinic is nuclear acid detection by real-time RT-PCR. However, the false-negative rate of the technique is unneglectable and serological methods are urgently warranted. Here, we presented the colloidal gold-based immunochromatographic (ICG) strip targeting viral IgM or IgG antibody and compared it with real-time RT-PCR. The sensitivity of ICG assay with IgM and IgG combinatorial detection in nuclear acid confirmed cases were 11.1%, 92.9% and 96.8% at the early stage (1-7 days after onset), intermediate stage (8-14 days after onset), and late-stage (more than 15 days), respectively. The ICG detection capacity in nuclear acid-negative suspected cases was 43.6%. In addition, the consistencies of whole blood samples with plasma were 100% and 97.1% in IgM and IgG strips, respectively. In conclusion, serological ICG strip assay in detecting SARS-CoV-2 infection is both sensitive and consistent, which is considered as an excellent supplementary approach in clinical application.Competing Interest StatementThe authors have declared no competing interest.Funding StatementThis work was supported by the National Key Research and Development Program of China (2018YFE0204500)Author DeclarationsAll relevant ethical guidelines have been followed; any necessary IRB and/or ethics committee approvals have been obtained and details of the IRB/oversight body are included in the manuscript.YesAll necessary patient/participant consent has been obtained and the appropriate institutional forms have been archived.YesI understand that all clinical trials and any other prospective interventional studies must be registered with an ICMJE-approved registry, such as ClinicalTrials.gov. I confirm that any such study reported in the manuscript has been registered and the trial registration ID is provided (note: if posting a prospective study registered retrospectively, please provide a statement in the trial ID field explaining why the study was not registered in advance).Yes I have followed all appropriate research reporting guidelines and uploaded the relevant EQUATOR Network research reporting checklist(s) and other pertinent material as supplementary files, if applicable.YesThe data that support the findings of this study are available from the corresponding author upon reasonable request.

URL: http://medrxiv.org/content/early/2020/03/17/2020.03.13.20035428.abstract

DOI: 10.1101/2020.03.13.20035428

56. **Park T, Lee S-Y, Kim S, et al. Spike protein binding prediction with neutralizing antibodies of SARS-CoV-2. *bioRxiv*. 2020:2020.02.22.951178. DOI:** 10.1101/2020.02.22.951178

Coronavirus disease 2019 (COVID-19) is a new emerging human infectious disease caused by Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2, also previously known as 2019-nCoV), originated in Wuhan seafood and animal market, China. Since December 2019, more than 69,000 cases of COVID-19 have been confirmed in China and quickly spreads to other counties. Currently, researchers put their best efforts to identify effective drugs for COVID-19. The neutralizing antibody, which binds to viral capsid in a manner that inhibits cellular entry of virus and uncoating of the genome, is the specific defense against viral invaders. In this study, we investigate to identify neutralizing antibodies that can bind to SARS-CoV-2 Sipke (S) protein and interfere with the interaction between viral S protein and a host receptor by bioinformatic methods. The sequence analysis of S protein showed two major differences in the RBD region of the SARS-CoV-2 S protein compared to SARS-CoV and SARS-CoV related bat viruses (btSARS-CoV). The insertion regions were close to interacting residues with the human ACE2 receptor. Epitope an alysis of neutralizing antibodies revealed that SARS-CoV neutralizing antibodies used conformational epitopes, whereas MERS-CoV neutralizing antibodies used a common linear epitope region, which contributes to form the Î²-sheet structure in MERS-CoV S protein and deleted in SARS-CoV-2 S protein. To identify effective neutralizing antibodies for SARS-CoV-2, the binding affinities of neutralizing antibodies with SARS-CoV-2 S protein were predicted and compared by antibody-antigen docking simulation. The result showed that CR3022 neutralizing antibody from human may have higher binding affinity with SARS-CoV-2 S protein than SARS-CoV S protein. We also found that F26G19 and D12 mouse antibodies could bind to SARS-CoV S protein with high affinity. Our findings provide crucial clues towards the development of antigen diagnosis, therapeutic antibody, and the vaccine against SARS-CoV-2.

URL: http://biorxiv.org/content/early/2020/02/27/2020.02.22.951178.abstract
DOI: 10.1101/2020.02.22.951178

57. **Petherick A. Developing antibody tests for SARS-CoV-2. *The Lancet*. 2020;395:1101-2. DOI:** 10.1016/S0140-6736(20)30788-1

URL: https://doi.org/10.1016/S0140-6736(20)30788-1
DOI: 10.1016/S0140-6736(20)30788-1

58. **Qiu R, Zhao C, Liang T, et al. Core Outcome Set for Clinical Trials of COVID-19 based on Traditional Chinese and Western Medicine. *medRxiv*. 2020:2020.03.23.20041533. DOI:** 10.1101/2020.03.23.20041533

Background: Development of a core outcome set (COS) for clinical trials for COVID-19 is urgent because of the pandemic wreaking havoc worldwide and the heterogeneity of outcomes in clinical trials. Methods: A preliminary list of outcomes were developed after a systematic review of protocols of clinical trials for COVID-19. Then, two rounds of the Delphi survey were conducted. Stakeholders were traditional Chinese medicine (TCM) experts, Western medicine (WM) experts, nurses and the public. Patients with confirmed COVID-19 were also invited to participate in a questionnaire written in understandable language. Frontline clinicians, as well as nurse, methodologist, evidence based-medicine researcher, and staff from the Chinese Clinical Trials Registry participated by video conference to vote. Results: Ninety-seven eligible study protocols were identified from 160 clinical trials. Seventy-six outcomes were identified from TCM clinical trials and 126 outcomes were identified from WM clinical trials. Finally, 145 outcomes were included in the first round of the Delphi survey. Then, a COS for clinical trials of TCM and WM was developed. The COS include clinical outcomes (recovery/improvement/progression/death), etiology (SARS-CoV-2 nucleic-acid tests, viral load), inflammatory factor (C-reactive protein), vital signs (temperature, respiration), blood and lymphatic-system parameters (lymphocytes, virus antibody), respiratory outcomes (Pulmonary imaging, blood oxygen saturation, PaO2/FiO2 ratio, arterial blood gas analysis, mechanical ventilation, oxygen intake, pneumonia se verity index), clinical efficacy (prevalence of preventing patients with mild-to-moderate disease progressing to severe disease), symptoms (clinical symptom score). Outcomes were recommended according to different types of disease. Outcome measurement instrument/definition were also recommended. Conclusion: A COS for COVID-19 may improve consistency of outcome reporting in clinical trials.Competing Interest StatementThe authors have declared no competing interest.Clinical Trialhttp://www.comet-initiative.org/Studies/Details/1507Clinical Protocolshttp://www.comet-initiative.org/Studies/Details/1507Funding StatementThis work was supported by the National High-level Personnel of Special Support Program (W02020052).Author DeclarationsAll relevant ethical

guidelines have been followed; any necessary IRB and/or ethics committee approvals have been obtained and details of the IRB/oversight body are included in the manuscript.YesAll necessary patient/participant consent has been obtained and the appropriate institutional forms have been archived.YesI understand that all clinical trials and any other prospective interventional studies must be registered with an ICMJE -approved registry, such as ClinicalTrials.gov.I confirm that any such study reported in the manuscript has been registered and the trial registration ID is provided (note: if posting a prospective study registered retrospectively, please provide a statement in the trial ID field explaining why the study was not registered in advance).Yes I have followed all appropriate research reporting guidelines and uploaded the relevant EQUATOR Network research reporting checklist(s) and other pertinent material as supplementary files, if applicable.YesThe data is from public database and does not include identifiable patient data.

URL: http://medrxiv.org/content/early/2020/04/07/2020.03.23.20041533.abstract
DOI: 10.1101/2020.03.23.20041533

59. **Qiu R, Zhao C, Liang T, et al. Core Outcome Set for Traditional Chinese and Western Medicine Clinical Trials of COVID-19. *medRxiv*. 2020:2020.03.23.20041533. DOI:** 10.1101/2020.03.23.20041533

Background: There are a large number of clinical trials for COVID -19. But the heterogeneity of outcomes may result in some clinical trials cannot be compared or merged. It is emergency to develop a core outcome set (COS) for clinical trials. Methods: A preliminary list of outcomes were developed after a systematic review of protocols of clinical trials for COVID-19. Then two rounds of Delphi survey was conducted. The stakeholders included traditional Chinese medicine (TCM) experts, Western medicine experts, nurses and the public. Patients with confirmed COVID -19 were also invited to participate in a questionnaire with simple language. Frontline clinicians (including TCM and Western medicine clinicians), nurse, methodologist, evidence -based medicine researcher and staff from Chinese Clinical Trials Registry participate in video conference to vote. Results: 97 eligible study protocols were identified from 160 clinical trials. 76 outcomes were identified from TCM clinical trials, 126 outcomes were identified from Western medicine clinical trials. In the end, 145 were included in the first round of Delphi survey. In the end, a COS was developed for clinical trials of TCM and Western medicine was developed. The COS includes Clinical outcome (recovery/ improvement/ progression/ death), etiology (SARS-CoV-2 nucleic acid tests, viral load), inflammatory factor (CRP), vital signs (temperature, respiration), blood and lymphatic system outcomes (lymphocyte, virus antibody), respiratory outcomes (chest imaging, blood oxygen saturation, PaO2/FiO2, arterial blood gas analysis, mechanical ventilation, oxygen intake, pneumonia severity index), clinical efficacy (rate of preventing mild to moderate type patients from progressing to severe type), symptoms (clinical symptom score). The outcomes were recommended according to different types of disease. Outcome measurement instrument/definition were also recommended. Conclusion: A COS for COVID -19 may improve consistency of outcome reporting in clinical trials, which may help identify valued interventions after comparing different trials when the researchers report the same outcomes.Competing Interest StatementThe authors have declared no competing interest.Clinical Trialhttp://www.comet-initiative.org/Studies/Details/1507Clinical Protocolshttp://www.comet-initiative.org/Studies/Details/1507Funding StatementThis work was supported by the National High -level Personnel of Special Support Program (W02020052).Author Declarations All relevant ethical guidelines have been followed; any necessary IRB and/or ethics committee approvals have been obtained and details of the IRB/oversight body are included in the manuscript.YesAll necessary patient/participant consent has been obtained and the appropriate institutional forms have been archived.YesI understand that all clinical trials and any other prospective interventional studies must be registered with an ICMJE -approved registry, such as ClinicalTrials.gov.I confirm that any such study reported in the manuscript has been registered and the trial registration ID is provided (note: if posting a prospective study registered retrospectively, please provide a statement in the trial ID field explaining why the study was not registered in advance).Yes I have followed all appropriate research reporting guidelines and uploaded the relevant EQUATOR Network research reporting checklist(s) and other pertinent material as supplementary files, if applicable.YesThe data is from public database and does not include identifiable patient data.

URL: http://medrxiv.org/content/early/2020/03/27/2020.03.23.20041533.abstract
DOI: 10.1101/2020.03.23.20041533

60. **Ricke D, Malone RW. Medical Countermeasures Analysis of 2019-nCoV and Vaccine Risks for Antibody-Dependent Enhancement (ADE). *SSRN- Lancet prepublication*. 2020.**

Background: In 80% of patients, COVID-19 presents as mild disease. 20% of cases develop severe (13%) or critical (6%) illness. More severe forms of COVID-19 present as clinical severe acute respiratory syndrome, T-predominant lymphopenia, high circulating levels of proinflammatory cytokines and chemokines, accumulation of macrophages and neutrophils in lungs, and immune dysregulation including immunosuppression.Methods: All major SARS-CoV-2 proteins were characterized using an amino acid residue variation analysis method. Results predict that most SARS-CoV-2 proteins are evolutionary constrained, with the exception of the spike (S) protein extended outer surface. Results were interpreted based on known SARS-like coronavirus virology and pathophysiology, with a focus on medical countermeasure development implications.Findings: Antibodies to variable S domains may enable an alternative infection pathway via Fc receptor-mediated uptake. This may be a gating event for the immune response dysregulation observed in more severe COVID-19 disease. Prior studies involving vaccine candidates for FCoV SARS-CoV-1 and Middle East Respiratory Syndrome coronavirus (MERS-CoV) demonstrate vaccination-induced antibody-dependent enhancement of disease (ADE), including infection of phagocytic antigen presenting cells (APC). T effector cells are believed to play an important role in controlling coronavirus infection; pan-T depletion is present in severe COVID-19 disease and may be accelerated by APC infection. Sequence and structural conservation of S suggests that SARS and MERS vaccine ADE risks may foreshadow SARS-CoV-2 vaccine risks. Autophagy inhibitors may reduce APC infection and T-cell depletion. Amino acid residue variation analysis identifies multiple constrained domains suitable as T cell vaccine targets. Evolutionary constraints on antiviral drug targets present in SARS-CoV-1 and SARS-CoV-2 may reduce risk of developing antiviral drug escape mutants.Interpretation: Safety testing of COVID-19 S protein-based B cell vaccines in animal models is strongly encouraged prior to clinical trials to reduce risk of ADE upon virus exposure.

URL: (2/27/2020). Available at SSRN: https://ssrn.com/abstract=3546070

DOI:

61. **Rios P, Radhakrishnan A, Antony J, et al. Effectiveness and safety of antiviral or antibody treatments for coronavirus. *medRxiv*. 2020:2020.03.19.20039008. DOI:** 10.1101/2020.03.19.20039008

Background: To identify safe and effective medical countermeasures (e.g., antivirals/antibodies) to address the current outbreak of a novel coronavirus (COVID-19) Methods: Comprehensive literature searches were developed by an experienced librarian for MEDLINE, EMBASE, the Cochrane Library, and biorxiv.org/medrxiv.org; additional searches for ongoing trials and unpublished studies were conducted in clinicaltrials.gov and the Global Infectious Diseases and Epidemiology Network (GIDEON). Title/abstract and full-text screening, data abstraction, and risk of bias appraisal were carried out by single reviewers. Results: 54 studies were included in the review: three controlled trials, 10 cohort studies, seven retrospective medical record/database studies, and 34 case reports or series. These studies included patients with severe acute respiratory syndrome (SARs, n=33), middle east respiratory syndrome (MERS, n=16), COVID-19 (n=3), and unspecified coronavirus (n=2). The most common treatment was ribavirin (n=41), followed by oseltamivir (n=10) and the combination of lopinavir/ritonavir (n=7). Additional therapies included broad spectrum antibiotics (n=30), steroids (n=39) or various interferons (n=12). No eligible studies examining monoclonal antibodies for COVID-19 were identified. One trial found that ribavirin prophylactic treatment statistically significantly reduced risk of MERS infection in people who had been exposed to the virus. Of the 21 studies reporting rates of ICU admission in hospitalized SARS or MERS patients, none reported statistically significant results in favour of or against antiviral therapies. Of the 40 studies reporting mortality rates in hospitalized SARS or MERS patients, one cohort study (MERS) and one retrospective study (SARS) found a statistically significant increase in the mortality rate for patients treated with ribavirin. Eighteen studies reported potential drug-related adverse effects including gastrointestinal symptoms, anemia, and altered liver function in patients receiving ribavirin. Conclusion: The current evidence for the effectiveness and safety of antiviral therapies for coronavirus is inconclusive and suffers from a lack of well-designed prospective trials or observational studies, preventing any treatment recommendations from being made. However, it is clear that the existing body of evidence is weighted heavily towards ribavirin (41/54 studies), which has not shown conclusive evidence of effectiveness and may cause harmful adverse events so future investigations may consider focusing on other candidates for antiviral therapy.Competing Interest StatementThe authors have declared no competing interest.Funding StatementTBDAuthor DeclarationsAll relevant ethical guidelines have been followed; any necessary IRB and/or ethics committee approvals have been obtained and details of the IRB/oversight body are included in the manuscript.YesAll necessary patient/participant consent has been obtained and the appropriate institutional forms have been archived.YesI understand that all clinical trials and any other prospective interventional studies must be registered with an ICMJE-approved registry, such as ClinicalTrials.gov. I confirm that any such study reported in the manuscript has been registered and the trial registration ID is provided (note: if posting a prospective study registered retrospectively, please provide a statement in the trial ID field explaining why the study was not

registered in advance).Yes I have followed all appropriate research reporting guidelines and uploaded the relevant EQUATOR Network research reporting checklist(s) and other pertinent material as supplementary files, if applicable.YesAll datasets supporting the conclusions of this article are included within the article.

URL: http://medrxiv.org/content/early/2020/03/23/2020.03.19.20039008.abstract
DOI: 10.1101/2020.03.19.20039008

62. **Rios P, Radhakrishnan A, Antony J, et al. Effectiveness and safety of antiviral or antibody treatments for coronavirus: A rapid review. *medRxiv*. 2020:2020.03.19.20039008. DOI:** 10.1101/2020.03.19.20039008

Background: To identify safe and effective medical countermeasures (e.g., antivirals/antibodies) to address the current outbreak of a novel coronavirus (COVID-19) Methods: Comprehensive literature searches were developed by an experienced librarian for MEDLINE, EMBASE, the Cochrane Library, and biorxiv.org/medrxiv.org; additional searches for ongoing trials and unpublished studies were conducted in clinicaltrials.gov and the Global Infectious Diseases and Epidemiology Network (GIDEON). Title/abstract and full-text screening, data abstraction, and risk of bias appraisal were carried out by single reviewers. Results: 54 studies were included in the review: three controlled trials, 10 cohort studies, seven retrospective medical record/database studies, and 34 case reports or series. These studies included patients with severe acute respiratory syndrome (SARs, n=33), middle east respiratory syndrome (MERS, n=16), COVID-19 (n=3), and unspecified coronavirus (n=2). The most common treatment was ribavirin (n=41), followed by oseltamivir (n=10) and the combination of lopinavir/ritonavir (n=7). Additional therapies included broad spectrum antibiotics (n=30), steroids (n=39) or various interferons (n=12). No eligible studies examining monoclonal antibodies for COVID-19 were identified. One trial found that ribavirin prophylactic treatment statistically significantly reduced risk of MERS infection in people who had been exposed to the virus. Of the 21 studies reporting rates of ICU admission in hospitalized SARS or MERS patients, none reported statistically significant results in favour of or against antiviral therapies. Of the 40 studies reporting mortality rates in hospitalized SARS or MERS patients, one cohort study (MERS) and one retrospective study (SARS) found a statistically significant increase in the mortality rate for patients treated with ribavirin. Eighteen studies reported potential drug-related adverse effects including gastrointestinal symptoms, anemia, and altered liver function in patients receiving ribavirin. Conclusion: The current evidence for the effectiveness and safety of antiviral therapies for coronavirus is inconclusive and suffers from a lack of well-designed prospective trials or observational studies, preventing any treatment recommendations from being made. However, it is clear that the existing body of evidence is weighted heavily towards ribavirin (41/54 studies), which has not shown conclusive evidence of effectiveness and may cause harmful adverse events so future investigations may consider focusing on other candidates for antiviral therapy.Competing Interest StatementThe authors have declared no competing interest.Funding StatementThis work was supported through the Drug Safety and Effectiveness Network funded by the Canadian Institutes of Health Research.Author DeclarationsAll relevant ethical guidelines have been followed; any necessary IRB and/or ethics committee approvals have been obtained and details of the IRB/oversight body are included in the manuscript.YesAll necessary patient/participant consent has been obtained and the appropriate institutional forms have been archived.YesI understand that all clinical trials and any other prospective interventional studies must be registered with an ICMJE-approved registry, such as ClinicalTrials.gov. I confirm that any such study reported in the manuscript has been registered and the trial registration ID is provided (note: if posting a prospective study registered retrospectively, please provide a statement in the trial ID field explaining why the study was not registered in advance).Yes I have followed all appropriate research reporting guidelines and uploaded the relevant EQUATOR Network research reporting checklist(s) and other pertinent material as supplementary files, if applicable.YesAll datasets supporting the conclusions of this article are included within the article.

URL: http://medrxiv.org/content/early/2020/03/25/2020.03.19.20039008.abstract
DOI: 10.1101/2020.03.19.20039008

63. **Shanmugaraj B, Siriwattananon K, Wangkanont K, et al. Perspectives on monoclonal antibody therapy as potential therapeutic intervention for Coronavirus disease-19 (COVID-19). *Asian Pacific journal of allergy and immunology*. 2020:10.12932/AP-200220-0773. DOI:** 10.12932/AP-200220-0773

Last decade witnessed the outbreak of many life-threatening human pathogens including Nipah, Ebola, Chikungunya, Zika, Middle East respiratory syndrome coronavirus (MERS-CoV), Severe Acute respiratory syndrome coronavirus (SARS-CoV) and more recently novel coronavirus (2019-nCoV or SARS-CoV-2). The disease condition associated with novel coronavirus, referred to as Coronavirus disease (COVID-19). The emergence of novel coronavirus in 2019 in Wuhan,

China marked the third highly pathogenic coronavirus infecting humans in the 21st century. The continuing emergence of coronaviruses at regular intervals poses a significant threat to human health and economy. Ironically, even after a decade of research on coronavirus, still there are no licensed vaccines or therapeutic agents to treat coronavirus infection which highlights an urgent need to develop effective vaccines or post -exposure prophylaxis to prevent future epidemics. Several clinical, genetic and epidemiological features of CO VID-19 resemble SARS-CoV infection. Hence, the research advancements on SARS-CoV treatment might help scientific community in quick understanding of this virus pathogenesis and develop effective therapeutic/prophylactic agents to treat and prevent this infection. Monoclonal antibodies represent the major class of biotherapeutics for passive immunotherapy to fight against viral infection. The therapeutic potential of monoclonal antibodies has been well recognized in the treatment of many diseases. Here, we summarize the potential monoclonal antibody based therapeutic intervention for COVID -19 by considering the existing knowledge on the neutralizing monoclonal antibodies against similar coronaviruses SARS-CoV and MERS-CoV. Further research on COVID-19 pathogenesis could identify appropriate therapeutic targets to develop specific anti-virals against this newly emerging pathogen.

URL: https://pubmed.ncbi.nlm.nih.gov/32134278
DOI: 10.12932/AP-200220-0773

64. **Sun C, Chen L, Yang J, et al. SARS-CoV-2 and SARS-CoV Spike-RBD Structure and Receptor Binding Comparison and Potential Implications on Neutralizing Antibody and Vaccine Development. *bioRxiv*. 2020:2020.02.16.951723. DOI:** 10.1101/2020.02.16.951723

SARS-CoV-2 and SARS-CoV share a common human receptor ACE2. Protein-protein interaction structure modeling indicates that spike-RBD of the two viruses also has similar overall binding conformation and binding free energy to ACE2. In vitro assays using recombinant ACE2 proteins and ACE2 expressing cells confirmed th e two coronaviruses' similar binding affinities to ACE2. The above studies provide experimental supporting evidences and possible explanation for the high transmissibility observed in the SARS-CoV-2 outbreak. Potent ACE2-blocking SARS-CoV neutralizing antibodies showed limited cross-binding and neutralizing activities to SARS-CoV-2. ACE2-non-blocking SARS-CoV RBD antibodies, though with weaker neutralizing activities against SARS -CoV, showed positive cross-neutralizing activities to SARS-CoV-2 with an unknown mechanism. These findings suggest a trade-off between the efficacy and spectrum for therapeutic antibodies to different coronaviruses, and hence highlight the possibilities and challenges in developing broadly protecting antibodies and vaccines against SARS-CoV-2 and its future mutants.

URL: http://biorxiv.org/content/early/2020/02/20/2020.02.16.951723.abstract
DOI: 10.1101/2020.02.16.951723

65. **Tárnok A. Machine Learning, COVID-19 (2019-nCoV), and multi-OMICS. *Cytometry Part A*. 2020;97(3):215-6. DOI:** 10.1002/cyto.a.23990

The primary plan of my editorial for this month was to highlight and comment on the special issue of this month: "Machine Learning for Single Cell Data". I wish to emphasize and thank the Guest Editors of this special issue, Yvan Saeys and Greg Finak, for their outstanding success and hard work to assemble excellent manuscripts for this issue. I am referring to their guest editorial giving you more details on aims and scopes and elaborating on specific articles. I started drafting this editorial while attending the annual Photonics West conference in San Francisco, presumably the largest showcase on photonics technologies and instrumentation. Scientifically, the sub-conference BIOS demonstrated the broadness and vividness of photonics technologies in life sciences and particularly in single cell analysis. When searching for relevant literature for this editorial, I was also tracking the actual global developments. This brought me to change my focus and comment on some issues that are rel evant to our field and somewhat related to that of the special issue. First of all, Nature Methods announced their Method of the Year 2019 1. It is: "Single-cell multimodal omics" and acknowledges (among others) the important contribution of highly multipl exed flow cytometry and cell sorting to the increased understanding of single cell biology and cell systems. This is motivating and confirms that cytometry is receiving the focus of attention. Concurrently, in the last weeks the relevance of the recent COVID-19 (2019-nCoV) outbreak and its effects started to become evident as everybody was monitoring the number of cases registered globally 2. As conference chairs, we were facing the fact that many of our speakers and colleagues became unavailable. Not only that, but the eeriness of the infection (it is transmissible already during its asymptomatic latency period of up to two weeks, recent results indicate even longer) gave many attendees an uneasy feeling. This brings me to two points worth discussing. (a) Are large scale conferences with global attendance still state of the art or should they be rethought; and (b) to what extent can cytometry support

global efforts in various fields of epidemic outbreaks of infectious diseases? In the past one to two decades the world faced several outbreaks of different viral infections that were luckily not as disastrous as initially anticipated but still claimed victims. These were coronaviruses such as the Severe Acute Respiratory Syndrome coronavirus (SARS-CoV) in 2002/2003, the Middle East Respiratory Syndrome coronavirus (MERS-CoV) with occasional outbreaks since 2012 or influenza viruses like the H1N1 pandemic in 2009/2010. It is only a matter of time until other pandemics follow. Global travel for work and leisure supports viral spread and can transport the infection to nearly all places of the globe within days. Global conferences contribute to such a rapid spread and one should reconsider this model of scientific exchange from time to time and scrutinize potential alternatives. Models for sustainable international conferences are under testing and combine venues in close proximity to institutions with a substantial expertise on the focus area of the conference with virtual participation and contribution by internet fo r participants on more distant places. Although personal meetings are essential for optimal information exchange, a reduction in travel would not only reduce the risk of dissemination of disease but, as a side effect, could result in budgetary savings, reduce travel-related emissions (in the wake of Greta), and eliminate jetlag. Now, what can Cytometry do for help in pandemics? Cytometry has already supported several achievements as a quick and non-representative literature search shows. Image cytometry methods 3 and bead-based flow cytometry methods 4 are at hand to enable for screening and detecting antibody virus interactions and detect viral antigens. Airway memory T-cells and viral E protein mutations have been identified in CoV infections a potential targets for vaccine strategies 5, 6. Immune responses seem to be indicative of disease severity 6, 7 but more studies are needed to have a practical assay for decision making at hand. In fact, easy to use and rapid assays derived from Cytomics or multi-OMICS approaches are needed to rapidly distinguish severe from mild cases and identify future critically ill individuals before symptom onset. Such a test would clearly take the pressure off of the clinicians because only those with high probability to becomin g critically ill would receive intensive care early. Hopefully we will see more related studies in this journal in the near future.

URL: https://doi.org/10.1002/cyto.a.23990
DOI: 10.1002/cyto.a.23990

66. **Thevarajan I, Nguyen THO, Koutsakos M, et al. Breadth of concomitant immune responses underpinning viral clearance and patient recovery in a non-severe case of COVID-19.** ***medRxiv*****. 2020:2020.02.20.20025841. DOI:** 10.1101/2020.02.20.20025841

We report the kinetics of the immune response in relation to clinical and virological features of a patient with mild-to-moderate coronavirus disease-19 (COVID-19) requiring hospitalisation. Increased antibody-secreting cells, follicular T-helper cells, activated CD4+ and CD8+ T-cells and IgM/IgG SARS-CoV-2-binding antibodies were detected in blood, prior to symptomatic recovery. These immunological changes persisted for at least 7 days following full resolution of symptoms, indicating substantial anti-viral immunity in this non-severe COVID-19.Competing Interest StatementSRLÂ's institution has received funding for investigator initiated research grants from Gilead Sciences, Merck, Viiv Healthcare and Leidos. She has received honoraria for participation in advisory boards and educational activities for Gilead Sciences, Merck, Viiv Healthcare and Abbvie.Clinical TrialN/AFunding StatementThis work was funded by the Australian National Health and Medical Research Council (NHMRC) Investigator Grant to KK (#1173871). CES has received funding from the European UnionÂ's Horizon 2020 research and innovation programme under the Marie SkÅ,odowska-Curie grant agreement No 792532 and University of Melbourne McKenzie Fellowship laboratory support. KK is supported by a NHMRC Senior Research Fellowship Level B (#1102792) and SRL is supported by an NHMRC Practitioner Fellowship and an NHMRC program grant. SYCT is supported by a NHMRC Career Development Fellowship (#1145033). XJ is supported by China Scholarship Council -University of Melbourne joint Scholarship. Author DeclarationsAll relevant ethical guidelines have been followed; any necessary IRB and/or ethics committee approvals have been obtained and details of the IRB/oversight body are included in the manuscript.YesAll necessary patient/participant consent has been obtained and the appro priate institutional forms have been archived.YesI understand that all clinical trials and any other prospective interventional studies must be registered with an ICMJE-approved registry, such as ClinicalTrials.gov. I confirm that any such study reported i n the manuscript has been registered and the trial registration ID is provided (note: if posting a prospective study registered retrospectively, please provide a statement in the trial ID field explaining why the study was not registered in advance).Yes I have followed all appropriate research reporting guidelines and uploaded the relevant EQUATOR Network research reporting checklist(s) and other pertinent material as supplementary files, if applicable.YesData availability The data that support the findings of this study are available from the corresponding author upon request. Raw FACS data are shown in the manuscript.

URL: http://medrxiv.org/content/early/2020/02/23/2020.02.20.20025841.abstract
DOI: 10.1101/2020.02.20.20025841

67. **Tian X, Li C, Huang A, et al. Potent binding of 2019 novel coronavirus spike protein by a SARS coronavirus-specific human monoclonal antibody. *Emerging microbes & infections*. 2020;9(1):382-5. DOI:** 10.1080/22221751.2020.1729069

The newly identified 2019 novel coronavirus (2019-nCoV) has caused more than 11,900 laboratory-confirmed human infections, including 259 deaths, posing a serious threat to human health. Currently, however, there is no specific antiviral treatment or vaccine. Considering the relatively high identity of receptor-binding domain (RBD) in 2019-nCoV and SARS-CoV, it is urgent to assess the cross-reactivity of anti-SARS CoV antibodies with 2019-nCoV spike protein, which could have important implications for rapid development of vaccines and therapeutic antibodies against 2019-nCoV. Here, we report for the first time that a SARS-CoV-specific human monoclonal antibody, CR3022, could bind potently with 2019-nCoV RBD (KD of 6.3 nM). The epitope of CR3022 does not overlap with the ACE2 binding site within 2019-nCoV RBD. These results suggest that CR3022 may have the potential to be developed as candidate therapeutics, alone or in combination with other neutralizing antibodies, for the prevention and treatment of 2019-nCoV infections. Interestingly, some of the most potent SARS-CoV-specific neutralizing antibodies (e.g. m396, CR3014) that target the ACE2 binding site of SARS-CoV failed to bind 2019-nCoV spike protein, implying that the difference in the RBD of SARS-CoV and 2019-nCoV has a critical impact for the cross-reactivity of neutralizing antibodies, and that it is still necessary to develop novel monoclonal antibodies that could bind specifically to 2019-nCoV RBD.

URL: https://pubmed.ncbi.nlm.nih.gov/32065055
DOI: 10.1080/22221751.2020.1729069

68. **Vogel G. New blood tests for antibodies could show true scale of coronavirus pandemic. *Science*. 2020. DOI:** 10.1126/science.abb8028

How many COVID-19 cases have gone undetected? And are those who had mild cases of the disease—perhaps so mild they dismissed it as a cold or allergies—immune to new infections? If so, they could slow the spread of the burgeoning pandemic. Labs and companies around the world have raced to develop antibody tests, and a few have been used in small studies and received commercial approval, including several from China. But so far, large-scale data from such tests—for example showing what fraction of people in the hard-hit city of Wuhan, China, might now be immune—is still lacking or at least not public. Scientists hope that will soon change as more tests become available.

URL: https://www.sciencemag.org/news/2020/03/new-blood-tests-antibodies-could-show-true-scale-coronavirus-pandemic
DOI: 10.1126/science.abb8028

69. **Wan Y, Shang J, Sun S, et al. Molecular Mechanism for Antibody-Dependent Enhancement of Coronavirus Entry. *Journal of virology*. 2020;94(5):e02015-19. DOI:** 10.1128/JVI.02015-19

Antibody-dependent enhancement (ADE) of viral entry has been a major concern for epidemiology, vaccine development, and antibody-based drug therapy. However, the molecular mechanism behind ADE is still elusive. Coronavirus spike protein mediates viral entry into cells by first binding to a receptor on the host cell surface and then fusing viral and host membranes. In this study, we investigated how a neutralizing monoclonal antibody (MAb), which targets the receptor-binding domain (RBD) of Middle East respiratory syndrome (MERS) coronavirus spike, mediates viral entry using pseudovirus entry and biochemical assays. Our results showed that MAb binds to the virus surface spike, allowing it to undergo conformational changes and become prone to proteolytic activation. Meanwhile, MAb binds to cell surface IgG Fc receptor, guiding viral entry through canonical viral-receptor-dependent pathways. Our data suggest that the antibody/Fc-receptor complex functionally mimics viral receptor in mediating viral entry. Moreover, we characterized MAb dosages in viral-receptor-dependent, Fc-receptor-dependent, and both-receptors-dependent viral entry pathways, delineating guidelines on MAb usages in treating viral infections. Our study reveals a novel molecular mechanism for antibody-enhanced viral entry and can guide future vaccination and antiviral strategies.IMPORTANCE Antibody-dependent enhancement (ADE) of viral entry has been observed for many viruses. It was shown that antibodies target one serotype of viruses but only subneutralize another, leading to ADE of the latter viruses. Here we identify a novel mechanism for ADE: a neutralizing antibody binds to the surface spike protein of coronaviruses like a viral receptor, triggers a conformational change of the spike, and mediates viral entry into IgG

Fc receptor-expressing cells through canonical viral-receptor-dependent pathways. We further evaluated how antibody dosages impacted viral entry into cells expressing viral receptor, Fc receptor, or both receptors. This study reveals complex roles of antibodies in viral entry and can guide future vaccine design and antibody-based drug therapy.

URL: https://pubmed.ncbi.nlm.nih.gov/31826992
DOI: 10.1128/JVI.02015-19

70. **Wang H, Hou X, Wu X, et al. SARS-CoV-2 proteome microarray for mapping COVID-19 antibody interactions at amino acid resolution. *bioRxiv*. 2020:2020.03.26.994756. DOI:** 10.1101/2020.03.26.994756

COVID-19 has quickly become a worldwide pandemic, which has significantly impacted the economy, education, and social interactions. Understanding the humoral antibody response to SARS-CoV-2 proteins may help identify biomarkers that can be used to detect and treat COVID-19 infection. However, no immuno-proteomics platform exists that can perform such proteome-wide analysis. To address this need, we created a SARS-CoV-2 proteome microarray to analyze antibody interactions at amino acid resolution by spotting peptides 15 amino acids long with 5-amino acid offsets representing full-length SARS-CoV-2 proteins. Moreover, the array processing time is short (1.5 hours), the dynamic range is ~2 orders of magnitude, and the lowest limit of detection is 94 pg/mL. Here, the SARS-CoV-2 proteome array reveals that antibodies commercially available for SARS-CoV-1 proteins can also target SARS-CoV-2 proteins. These readily available reagents could be used immediately in COVID-19 research. Second, IgM and IgG immunogenic epitopes of SARS-CoV-2 proteins were profiled in the serum of ten COVID-19 patients. Such epitope biomarkers provide insight into the immune response to COVID-19 and are potential targets for COVID-19 diagnosis and vaccine development. Finally, serological antibodies that may neutralize viral entry into host cells via the ACE2 receptor were identified. Further investigation into whether these antibodies can inhibit the propagation of SARS-CoV-2 is warranted. Antibody and epitope profiling in response to COVID-19 is possible with our peptide-based SARS-COV-2 proteome microarray. The data gleaned from the array could provide invaluable information to the scientific community to understand, detect, and treat COVID-19.

URL: http://biorxiv.org/content/early/2020/03/28/2020.03.26.994756.abstract
DOI: 10.1101/2020.03.26.994756

71. **Wang K, Chen W, Zhou Y-S, et al. SARS-CoV-2 invades host cells via a novel route: CD147-spike protein. *bioRxiv*. 2020:2020.03.14.988345. DOI:** 10.1101/2020.03.14.988345

Currently, COVID-19 caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) has been widely spread around the world; nevertheless, so far there exist no specific antiviral drugs for treatment of the disease, which poses great challenge to control and contain the virus. Here, we reported a research finding that SARS-CoV-2 invaded host cells via a novel route of CD147-spike protein (SP). SP bound to CD147, a receptor on the host cells, thereby mediating the viral invasion. Our further research confirmed this finding. First, in vitro antiviral tests indicated Meplazumab, an anti-CD147 humanized antibody, significantly inhibited the viruses from invading host cells, with an EC50 of 24.86 Î¼g/mL and IC50 of 15.16 Î¼g/mL. Second, we validated the interaction between CD147 and SP, with an affinity constant of 1.85E-07M. Co-Immunoprecipitation and ELISA also confirmed the binding of the two proteins. Finally, the localization of CD147 and SP was observed in SARS-CoV-2 infected Vero E6 cells by immuno-electron microscope. Therefore, the discovery of the new route CD147-SP for SARS-CoV-2 invading host cells provides a critical target for development of specific antiviral drugs.

URL: http://biorxiv.org/content/early/2020/03/14/2020.03.14.988345.abstract
DOI: 10.1101/2020.03.14.988345

72. **Watanabe Y, Berndsen ZT, Raghwani J, et al. Vulnerabilities in coronavirus glycan shields despite extensive glycosylation. *bioRxiv*. 2020:2020.02.20.957472. DOI:** 10.1101/2020.02.20.957472

Severe acute respiratory syndrome (SARS) and Middle East respiratory syndrome (MERS) coronaviruses (CoVs) are zoonotic pathogens with high fatality rates and pandemic potential. Vaccine development has focussed on the principal target of the neutralizing humoral immune response, the spike (S) glycoprotein, which mediates receptor recognition and membrane fusion. Coronavirus S proteins are extensively glycosylated viral fusion proteins, encoding around 69-87 N-linked glycosylation sites per trimeric spike. Using a multifaceted structural approach, we reveal a specific area of high glycan density on MERS S that results in the formation of under-processed oligomannose-type glycan clusters, which

was absent on SARS and HKU1 CoVs. We provide a comparison of the global glycan density of coronavirus spikes with other viral proteins including HIV-1 envelope, Lassa virus glycoprotein complex, and influenza hemagglutinin, where glycosylation plays a known role in shielding immunogenic epitopes. Consistent with the ability of the antibody-mediated immune response to effectively target and neutralize coronaviruses, we demonstrate that the glycans of coronavirus spikes are not able to form an efficacious high-density global shield to thwart the humoral immune response. Overall, our data reveal how differential organisation of viral glycosylation across class I viral fusion proteins influence not only individual glycan compositions but also the immunological pressure across the viral protein surface.

URL: http://biorxiv.org/content/early/2020/02/21/2020.02.20.957472.abstract
DOI: 10.1101/2020.02.20.957472

73. **Wippold JA, Wang H, Tingling J, et al. PRESCIENT: platform for the rapid evaluation of antibody success using integrated microfluidics enabled technology. *Lab chip*. 2020;20:20. DOI:** https://dx.doi.org/10.1039/c9lc01165j

Identifying antibodies (Abs) that neutralize infectious agents is the first step for developing therapeutics, vaccines, and diagnostic tools for these infectious agents. However, current approaches for identifying neutralizing Abs (nAbs) typically rely on dilution-based assays that are costly, inefficient, and only survey a small subset of the entire repertoire. There are also intrinsic biases in many steps of conventional nAb identification processes. More importantly, conventional assays rely on simple Ab-antigen binding assays, which may not result in identifying the most potent nAbs, as the strongest binder may not be the most potent nAb. Droplet microfluidic systems have the capability to overcome such limitations by conducting complex multi-step assays with high reliability, resolution, and throughput in a pico-liter volume water-in-oil emulsion droplet format. Here, we describe the development of PRESCIENT (Platform for the Rapid Evaluation of antibody SucCess using Integrated microfluidics ENabled Technology), a droplet microfluidic system that can enable high-throughput single-cell resolution identification of nAb repertoires elicited in response to viral infection. We demonstrate PRESCIENT's ability to identify Abs that neutralize a model viral agent, Murine coronavirus (murine hepatitis virus), which causes high mortality rates in experimentally infected mice. In-droplet infection of host cells by the virus was first demonstrated, followed by demonstration of in-droplet neutralization by nAbs produced from a single Ab-producing hybridoma cell. Finally, fluorescence intensity analyses of two populations of hybridoma cell lines (nAb-producing and non-nAb-producing hybridoma cell lines) successfully discriminated between the two populations. The presented strategy and platform have the potential to identify and investigate neutralizing activities against a broad range of potential infectious agents for which nAbs have yet to be discovered, significantly advancing the nAb identification process as well as reinvigorating the field of Ab discovery, characterization, and development.

URL:

http://ovidsp.ovid.com/ovidweb.cgi?T=JS&CSC=Y&NEWS=N&PAGE=fulltext&D=medp&AN=32196032http://sfx.library.cdc.gov/cdc?sid=OVID:medline&id=pmid:32196032&id=10.1039%2Fc9lc01165j&issn=1473-0189&isbn=&volume=&issue=&spage=&pages=&date=2020&title=Lab+on+a+Chip&atitle=PRESCIENT%3A+platform+for+the+rapid+evaluation+of+antibody+success+using+integrated+microfluidics+enabled+technology.&aulast=Wippold&pid=%3Cauthor%3EWippold+JA%2CWang+H%2CTingling+J%2CLeibowitz+JL%2Cde+Figueiredo+P%2CHan+A%3C%2Fauthor%3E&%3CAN%3E32196032%3C%2FAN%3E&%3CDT%3EJournal+Article%3C%2FDT%3E

DOI: https://dx.doi.org/10.1039/c9lc01165j

74. **Wrapp D, Wang N, Corbett KS, et al. Cryo-EM structure of the 2019-nCoV spike in the prefusion conformation. *Science*. 2020;367:1260-3. DOI:** 10.1126/science.aax0902

The outbreak of a novel coronavirus (2019-nCoV) represents a pandemic threat that has been declared a public health emergency of international concern. The CoV spike (S) glycoprotein is a key target for vaccines, therapeutic antibodies, and diagnostics. To facilitate medical countermeasure development, we determined a 3.5-angstrom-resolution cryo-electron microscopy structure of the 2019-nCoV S trimer in the prefusion conformation. The predominant state of the trimer has one of the three receptor-binding domains (RBDs) rotated up in a receptor-accessible conformation. We also provide biophysical and structural evidence that the 2019-nCoV S protein binds angiotensin-converting enzyme 2 (ACE2) with higher affinity than does severe acute respiratory syndrome (SARS)-CoV S. Additionally, we tested several published SARS-CoV RBD-specific monoclonal antibodies and found that they do not have appreciable binding to 2019-nCoV S, suggesting that antibody cross-reactivity may be limited between the two RBDs. The structure of 2019-nCoV S should enable the rapid development and evaluation of medical

countermeasures to address the ongoing public health crisis. © 2020 American Association for the Advancement of Science. All rights reserved.

URL: https://www.scopus.com/inward/record.uri?eid=2-s2.0-85081889457&doi=10.1126%2fscience.aax0902&partnerID=40&md5=4f6b0cd6bcc5ff5d73415c13ce8efd4b
DOI: 10.1126/science.aax0902

75. **Wrapp D, Wang N, Corbett KS, et al. Cryo-EM structure of the 2019-nCoV spike in the prefusion conformation.** ***Science*****. 2020:eabb2507. DOI:** 10.1126/science.abb2507

The outbreak of a novel betacoronavirus (2019-nCoV) represents a pandemic threat that has been declared a public health emergency of international concern. The CoV spike (S) glycoprotein is a key target for vaccines, therapeutic antibodies, and diagnostics. To facilitate medical countermeasure (MCM) development, we determined a 3.5 Å-resolution cryo-EM structure of the 2019-nCoV S trimer in the prefusion conformation. The predominant state of the trimer has one of the three receptor-binding domains (RBDs) rotated up in a receptor-accessible conformation. We also show biophysical and structural evidence that the 2019-nCoV S binds ACE2 with higher affinity than SARS-CoV S. Additionally, we tested several published SARS-CoV RBD-specific monoclonal antibodies and found that they do not have appreciable binding to 2019-nCoV S, suggesting antibody cross-reactivity may be limited between the two RBDs. The structure of 2019-nCoV S should enable rapid development and evaluation of MCMs to address the ongoing public health crisis.

URL: https://pubmed.ncbi.nlm.nih.gov/32075877
DOI: 10.1126/science.abb2507

76. **Wu F, Wang A, Liu M, et al. Neutralizing antibody responses to SARS-CoV-2 in a COVID-19 recovered patient cohort and their implications.** ***medRxiv*****. 2020:2020.03.30.20047365. DOI:** 10.1101/2020.03.30.20047365

Background The COVID-19 pandemic caused by SARS-CoV-2 coronavirus threatens global public health. Currently, neutralizing antibodies (NAbs) versus this virus are expected to correlate with recovery and protection of this disease. However, the characteristics of these antibodies have not been well studied in association with the clinical manifestations in patients. Methods Plasma collected from 175 COVID-19 recovered patients with mild symptoms were screened using a safe and sensitive pseudotyped-lentiviral-vector-based neutralization assay. Spike-binding antibody in plasma were determined by ELISA using RBD, S1, and S2 proteins of SARS-CoV-2. The levels and the time course of SARS-CoV-2-specific NAbs and the spike-binding antibodies were monitored at the same time. Findings SARS-CoV-2 NAbs were unable to cross-reactive with SARS-CoV virus. SARS-CoV-2-specific NAbs were detected in patients from day 10-15 after the onset of the disease and remained thereafter. The titers of NAb among these patients correlated with the spike-binding antibodies targeting S1, RBD, and S2 regions. The titers of NAbs were variable in different patients. Elderly and middle-age patients had significantly higher plasma NAb titers (P<0.0001) and spike-binding antibodies (P=0.0003) than young patients. Notably, among these patients, there were ten patients whose NAb titers were under the detectable level of our assay (ID50: < 40); while in contrast, two patients, showed very high titers of NAb, with ID50 :15989 and 21567 respectively. The NAb titers were positive correlated with plasma CRP levels but negative correlated with the lymphocyte counts of patients at the time of admission, indicating an association between humoral response and cellular immune response. Interpretation The variations of SARS-CoV-2 specific NAbs in recovered COVID-19 patients may raise the concern about the role of NAbs on disease progression. The correlation of NAb titers with age, lymphocyte counts, and blood CRP levels suggested that the interplay between virus and host immune response in coronavirus infections should be further explored for the development of effective vaccine against SARS-CoV-2 virus. Furthermore, titration of NAb is helpful prior to the use of convalescent plasma for prevention or treatment. Funding Ministry of Science and Technology of China, National Natural Science Foundation of China, Shanghai Municipal Health Commission, and Chinese Academy of Medical SciencesCompeting Interest StatementThe authors have declared no competing interest.Funding StatementThis work was supported by the National Major Science and Technology Projects of China (2017ZX10202102 and 2018ZX10301403), National Natural Science Foundation of China (31771008), Hundred Talent Program of Shanghai Municipal Health Commission (2018BR08), and Chinese Academy of Medical Sciences (2019PT350002). Author DeclarationsAll relevant ethical guidelines have been followed; any necessary IRB and/or ethics committee approvals have been obtained and details of the IRB/oversight body are included in the manuscript.YesAll necessary patient/participant consent has been obtained and the appropriate institutional forms have been archived.YesI understand that all clinical trials and any other prospective interventional studies must be registered with an ICMJE-approved registry, such as

ClinicalTrials.gov.I confirm that any such study reported in the manuscript has been registered and the trial registration ID is provided (note: if posting a prospective study registered retrospectively, please provide a statement in the trial ID field explaining why the study was not registered in advance).Yes I have followed all appropriate research reporting guidelines and uploaded the relevant EQUATOR Network research reporting checklist(s) and other pertinent material as supplementary files, if applicable.YesThe authors confirm that the data supporting the findings of this study are available within the article and its supplementary materials.

URL: http://medrxiv.org/content/early/2020/04/06/2020.03.30.20047365.abstract
DOI: 10.1101/2020.03.30.20047365

77. **Wu Y, Li C, Xia S, et al. Fully human single-domain antibodies against SARS-CoV-2. *bioRxiv*. 2020:2020.03.30.015990. DOI:** 10.1101/2020.03.30.015990

The COVID-19 pandemic is spreading rapidly, highlighting the urgent need for an efficient approach to rapidly develop therapeutics and prophylactics against SARS-CoV-2. We describe here the development of a phage-displayed single-domain antibody library by grafting naive CDRs into framework regions of an identified human germline IGHV allele. This enabled the isolation of high-affinity single-domain antibodies of fully human origin. The panning using SARS-CoV-2 RBD and S1 as antigens resulted in the identification of antibodies targeting five types of neutralizing or non-neutralizing epitopes on SARS-CoV-2 RBD. These fully human single-domain antibodies bound specifically to SARS-CoV-2 RBD with subnanomolar to low nanomolar affinities. Some of them were found to potently neutralize pseudotyped and live virus, and therefore may represent promising candidates for prophylaxis and therapy of COVID-19. This study also reports unique immunogenic profile of SARS-CoV-2 RBD compared to that of SARS-CoV and MERS-CoV, which may have important implications for the development of effective vaccines against SARS-CoV-2.

URL: http://biorxiv.org/content/early/2020/03/31/2020.03.30.015990.abstract
DOI: 10.1101/2020.03.30.015990

78. **Xie L, Sun C, Luo C, et al. SARS-CoV-2 and SARS-CoV Spike-RBD Structure and Receptor Binding Comparison and Potential Implications on Neutralizing Antibody and Vaccine Development. *bioRxiv*. 2020:2020.02.16.951723. DOI:** 10.1101/2020.02.16.951723

SARS-CoV-2 and SARS-CoV share a common human receptor ACE2. Protein-protein interaction structure modeling indicates that spike-RBD of the two viruses also has similar overall binding conformation and binding free energy to ACE2. In vitro assays using recombinant ACE2 proteins and ACE2 expressing cells confirmed the two coronaviruses' similar binding affinities to ACE2. The above studies provide experimental supporting evidences and possible explanation for the high transmissibility observed in the SARS-CoV-2 outbreak. Potent ACE2-blocking SARS-CoV neutralizing antibodies showed limited cross-binding and neutralizing activities to SARS-CoV-2. ACE2-non-blocking SARS-CoV RBD antibodies, though with weaker neutralizing activities against SARS-CoV, showed positive cross-neutralizing activities to SARS-CoV-2 with an unknown mechanism. These findings suggest a trade-off between the efficacy and spectrum for therapeutic antibodies to different coronaviruses, and hence highlight the possibilities and challenges in developing broadly protecting antibodies and vaccines against SARS-CoV-2 and its future mutants.

URL: http://biorxiv.org/content/early/2020/02/20/2020.02.16.951723.abstract
DOI: 10.1101/2020.02.16.951723

79. **Xiong H, Wu Y, Cao J, et al. Robust neutralization assay based on SARS-CoV-2 S-bearing vesicular stomatitis virus (VSV) pseudovirus and ACE2-overexpressed BHK21 cells. *bioRxiv*. 2020:2020.04.08.026948. DOI:** 10.1101/2020.04.08.026948

The global pandemic of Coronavirus disease 2019 (COVID-19) is a disaster for human society. A convenient and reliable in vitro neutralization assay is very important for the development of neutralizing antibodies, vaccines and other inhibitors. In this study, G protein-deficient vesicular stomatitis virus (VSVdG) bearing full-length or truncated spike (S) protein of SARS-CoV-2 and various cell lines for pseudotyped virus package and infection were evaluated. The virus packaging efficiency of VSV-SARS-CoV-2-Sdel18 (S with C-terminal 18 amino acid truncation) is much higher than VSV-SARS-CoV-2-S. A neutralization assay for antibody screening and validation was established based on VSV-SARS-CoV-2-Sdel18 pseudovirus and human angiotensin-converting enzyme 2 (ACE2) overexpressed BHK21 cell (BHK21-hACE2). The detection of pseudovirus through reporter EGFP guarantee the high throughput of pseudovirus based assay. The serum neutralizing titer of COVID-19 convalescent patients measured by VSV-SARS-CoV-2-Sdel18 pseudovirus assay

was correlated well with live SARS-CoV-2 assay. The neutralizing activities of 7 neutralizing mouse monoclonal antibodies (mAbs) targeting receptor binding domain (RBD) of SARS-CoV-2-S were evaluated.Competing Interest Statement

URL: http://biorxiv.org/content/early/2020/04/09/2020.04.08.026948.abstract
DOI: 10.1101/2020.04.08.026948

80. Yang MS, Jang YS, Kim MY, et al., inventors; Industrial Cooperation Foundation Chonbuk National University St George's, University of London, assignee. Polypeptides for antigen delivery, Fc-fusion proteins comprising same and uses patent KR2020008288. 2020.

The title polypeptides for antigen delivery comprises human antibody Fc domain. The polypeptides, which are vaccine delivery platforms applicable to all vaccine development studies, and the Fc fusion proteins obtained by the mol. engineering design based on the polypeptides enhance the affinity and binding force to low -affinity antibody receptors to increase the vaccine efficiency, and can be used for developing vaccines for various diseases.

URL: DOI:

81. **Zegpi RA, He L, Yu Q, et al. Limited Protection Conferred by Recombinant Newcastle Disease Virus Expressing Infectious Bronchitis Spike Protein. *Avian Diseases*. 2020;64(1):53-9. DOI:** https://dx.doi.org/10.1637/0005-2086-64.1.53

Recombinant Newcastle disease virus (NDV) LaSota (LS) expressing secreted trimeric spike (S)-ectodomain (Se) of infectious bronchitis virus (IBV) (rLS/IBV.Se) was developed and evaluated for protection conferred against IBV challenge. The IBV S-ectodomain protein, which is S excluding the transmembrane anchor and short cytoplasmic domain of S2, expressed from recombinant LS corresponds to an Arkansas (Ark)-type IBV. In a first experiment, chickens were primed at 1 day of age or primed at 1 day of age and boosted at 14 days of age with 10 sup 4 /sup 50% embryo infectious doses (EID sub 50 /sub)/bird of rLS/IBV.Se and challenged with a virulent Ark strain. A single vaccination proved completely ineffective at protecting chickens against challenge, whereas priming and boosting reduced clinical signs and tracheal lesions but did not reduce viral load in lachrymal fluids. In experiment 2, the vaccine dose was increased to 10 sup 7 /sup EID sub 50 /sub /bird and a different virulent Ark strain was used for challenge. In addition, chickens were singly immunized on either day 1 or day 10 after hatch. NDV antibody levels detected in vaccinated chickens were moderate, with hemagglutination inhibition titers varying between 4 and 5 log sub 2 /sub . Slightly higher antibody levels to NDV were observed in chickens vaccinated on day 10 versus day 1 but without the difference achieving statistical significance. In contrast, antibody responses measured using recombinant IBV S1 protein-coated ELISA plates were significantly greater in chickens vaccinated on day 10 than on day 1. The use of a higher rLS/IBV.Se dose substantially enhanced the success of a single vaccination compared to experiment 1. Signs and tracheal lesions were reduced more effectively in chickens vaccinated at day 10 after hatch. However, as in experiment 1, vaccination did not reduce the viral loads in tear fluids of challenged chickens. Similar results, in which no reduction in viral load in the trachea was apparent from rLS/IBV.S vaccination, have been obtained by others. Further work is needed to understand the immune responses induced by this recombinant virus that seems to provide some protection against the disease but does not reduce viral loads in the upper respiratory tract.

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DOI: https://dx.doi.org/10.1637/0005-2086-64.1.53

82. **Zhang J, Liu J, Li N, et al. Serological detection of 2019-nCoV respond to the epidemic: A useful complement to nucleic acid testing. *medRxiv*. 2020:2020.03.04.20030916. DOI:** 10.1101/2020.03.04.20030916

Background Pneumonia caused by 2019 novel coronavirus (2019-nCoV) was first reported in Wuhan, Hubei Province, China in December 2019. Then it has been reported in more than 20 countries and regions overseas rapidly. More than eighty thousand cases have been infected, resulting in more than three thousand deaths. Due to the limitation of nucleic

acid detection, many clinical suspected cases cannot be diagnosed in time. Methods We used automated chemiluminescent immunoassay to detect serum IgM and IgG antibodies to 2019-nCoV of 736 subjects including confirmed Corona Virus Disease 2019 (COVID-19) patients, non-COVID-19 fever patients, other disease patients and medical staff as well as healthy people. The dynamic process of antibody production in COVID-19 disease progression were analyzed, and the value of antibody detection in the laboratory diagnosis of COVID-19 were evaluated. Results COVID-19 patients were becoming reactive(positive) for specific anti-2019-nCoV IgM antibodies from 7-12 days after the onset of morbidity, followed closely by the IgG. The levels of specific IgM and IgG antibodies increased with the progression of the disease. The trend of IgM and IgG changes in different cases is not exactly the same. The levels of IgM and IgG and their distributions in different groups were different with that of healthy people. The areas under the ROC curves for IgM and IgG to diagnose COVID-19 were 0.988 and 1.000, respectively. Conclusions Specific IgM or IgG antibody detection had good sensitivity and specificity for the diagnosis of suspected fever cases. D etection of specific antibodies in patients with fever can be a good distinction between COVID-19 and other diseases in low epidemic area.Competing Interest StatementThe authors have declared no competing interest.Funding StatementThis study was funded by â€œthe National Science and Technology Major Project of China (2018ZX10302205)â€□, â€œLiaoning Province Natural Science Foundation Project(20180550523)â€□, â€œLiaoning Province Central Government's special project to guide local scientific and technological development (2019JH6/10400009)â€□, â€œGuangdong Province Major key projects of industrial technology (201902010003)â€□, â€œMajor Special Project of Construction Program of China Medical University in 2018(112/3110118034)â€□ and â€œ345 talent projectâ€□ of Shengjing Hospital of China Medical University. The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.Author DeclarationsAll relevant ethical guidelines have been followed; any necessary IRB and/or ethics committee approvals have been obtained and details of the IRB/oversight body are included in the manuscript.YesAll necessary patient/participant consent has been obtained and the appropriate institutional forms have been archived.YesI understand that all clinical trials and any other prospective interventional studies must be registered with an ICMJE-approved registry, such as ClinicalTrials.gov. I confirm that any such study reported in the manuscript has been registered and the trial registration ID is provided (note: if posting a prospective study registered retrospectively, please provide a statement in the trial ID field explaining why the study was not registered in advance).Yes I have followed all appropriate research reporting g uidelines and uploaded the relevant EQUATOR Network research reporting checklist(s) and other pertinent material as supplementary files, if applicable.YesThe data used to support the findings of this study are available from the corresponding author upon request.

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DOI: 10.1101/2020.03.04.20030916

83. **Zhao R, Li M, Song H, et al. Serological diagnostic kit of SARS-CoV-2 antibodies using CHO-expressed full-length SARS-CoV-2 S1 proteins. *medRxiv*. 2020:2020.03.26.20042184. DOI:** 10.1101/2020.03.26.20042184

WHO has declared COVID-19 a pandemic with more than 300,000 confirmed cases and more than 14,000 deaths. There is urgent need for accurate and rapid diagnostic kits. Here we report the development and validation of a COVID-19/SARS-CoV-2 S1 serology ELISA kit for the detection of total anti-virus antibody (IgG+IgM) titers in sera from either the general population or patients suspected to be infected. For indirect ELISA, CHO-expressed recombinant full length SARS-CoV-2-S1 protein with 6*His tag was used as the coating antigen to capture the SARS-CoV-2-S1 antibodies specifically. The specificity of the ELISA kit was determined to be 97.5%, as examined against total 412 normal human sera including 257 samples collected prior to the outbreak and 155 collected during the outbreak. The sensitivity of the ELISA kit was determined to be 97.5% by testing against 69 samples from hospitalized and/or recovered COVID-19 patients. The overall accuracy rate reached 97.3%. Most importantly, in one case study, the ELISA test kit was able to identify an infected person who had previously been quarantined for 14 days after coming into contact with a confirmed COVID-19 patient, and discharged after testing negative twice by nucleic acid test. With the assays developed here, we can screen millions of medical staffs in the hospitals and people in residential complex, schools, public transportations, and business parks in the epidemic centers of the outbreaks to fish out the "innocent viral spreaders", and help to stop the further spreading of the virus.Competing Interest StatementThe authors have declared no competing interest.Funding StatementThis work is supported by Research Grants from Beijing Science and Technology Commission, and Bill & Melinda Gates Foundation to Le Sun. This work was also supported by the National Natural Science Foundation of China (NSFC) (81702015), and the National Science and Technology Major Project (2018ZX10733403) to H.S.We thank Professor Wenjie Tan from National Institute for Viral Disease Control and Prevention, Chinese Center for Disease Control and Prevention for providing the sequences of SARS-CoV-2 S1

protien.Author DeclarationsAll relevant ethical guidelines have been followed; any necessary IRB and/or ethics committee approvals have been obtained and details of the IRB/oversight body are included in the manuscript.YesAll necessary patient/participant consent has been obtained and the appropriate institutional forms have be en archived.YesI understand that all clinical trials and any other prospective interventional studies must be registered with an ICMJE-approved registry, such as ClinicalTrials.gov. I confirm that any such study reported in the manuscript has been registered and the trial registration ID is provided (note: if posting a prospective study registered retrospectively, please provide a statement in the trial ID field explaining why the study was not registered in advance).Yes I have followed all appropriate research reporting guidelines and uploaded the relevant EQUATOR Network research reporting checklist(s) and other pertinent material as supplementary files, if applicable.YesThe data used to support the findings of this study are available from the corresponding author upon request.

URL: http://medrxiv.org/content/early/2020/03/27/2020.03.26.20042184.abstract
DOI: 10.1101/2020.03.26.20042184

84. **Zhu H, Fohlerová Z, Pekárek J, et al. Recent advances in lab-on-a-chip technologies for viral diagnosis. *Biosensors and Bioelectronics*. 2020;153:112041. DOI:** https://doi.org/10.1016/j.bios.2020.112041

The global risk of viral disease outbreaks emphasizes the need for rapid, accurate, and sensitive detection techniques to spe ed up diagnostics allowing early intervention. An emerging field of microfluidics also known as the lab-on-a-chip (LOC) or micro total analysis system includes a wide range of diagnostic devices. This review briefly covers both conventional and microfluidics-based techniques for rapid viral detection. We first describe conventional detection methods such as cell culturing, immunofluorescence or enzyme-linked immunosorbent assay (ELISA), or reverse transcription polymerase chain reaction (RT-PCR). These methods often have limited speed, sensitivity, or specificity and are performed with typically bulky equipment. Here, we discuss some of the LOC technologies that can overcome these demerits, highlighting the latest advances in LOC devices for viral disease diagnosis. We also discuss the fabrication of LOC systems to produce devices for performing either individual steps or virus detection in samples with the sample to answer method. The complete system consists of sample preparation, and ELISA and RT-PCR for viral-antibody and nucleic acid detection, respectively. Finally, we formulate our opinions on these areas for the future development of LOC systems for viral diagnostics.

URL: http://www.sciencedirect.com/science/article/pii/S0956566320300385
DOI: https://doi.org/10.1016/j.bios.2020.112041

85. **Zhu J, Kim J, Xiao X, et al. The immune vulnerability landscape of the 2019 Novel Coronavirus, SARS-CoV-2. *bioRxiv*. 2020:2020.02.08.939553. DOI:** 10.1101/2020.02.08.939553

The outbreak of the 2019 Novel Coronavirus (SARS-CoV-2) rapidly spread from Wuhan, China to more than 150 countries, areas or territories, causing staggering number of infections and deaths. A systematic profiling of the immune vulnerability landscape of SARS-CoV-2, which can bring critical insights into the immune clearance mecha nism, peptide vaccine development, and antiviral antibody development, is lacking. In this study, we investigated the potential of the SARS-CoV-2 viral proteins to induce class I and II MHC presentation and to form linear antibody epitopes. We created an online database to broadly share the predictions as a resource for the research community. Using this resource, we showed that genetic variations in SARS-CoV-2, though still few for the moment, already follow the pattern of mutations in related coronaviruses, and could alter the immune vulnerability landscape of this virus. Importantly, we discovered evidence that SARS-CoV-2, along with related coronaviruses, used mutations to evade attack from the human immune system. Overall, we present an immunological re source for SARS-CoV-2 that could promote both therapeutic development and mechanistic research.

URL: http://biorxiv.org/content/early/2020/03/23/2020.02.08.939553.abstract
DOI: 10.1101/2020.02.08.939553

86. **Haverkamp A-K, Bosch BJ, Spitzbarth I, et al. Detection of MERS-CoV antigen on formalin-fixed paraffin-embedded nasal tissue of alpacas by immunohistochemistry using human monoclonal antibodies directed against different epitopes of the spike protein. *Veterinary Immunology & Immunopathology*. 2019;218:N.PAG-N.PAG. DOI:** 10.1016/j.vetimm.2019.109939

Middle East respiratory syndrome (MERS) represents an important respiratory disease accompanied by lethal outcome in one third of human patients. In recent years, several investigators developed protective antibodies which could be used as

prophylaxis in prospective human epidemics. In the current study, eight human monoclonal antibodies (mAbs) with neutralizing and non-neutralizing capabilities, directed against different epitopes of the MERS-coronavirus (MERS-CoV) spike (MERS-S) protein, were investigated with regard to their ability to immunohistochemically detect respective epitopes on formalin-fixed paraffin-embedded (FFPE) nasal tissue sections of MERS-CoV experimentally infected alpacas. The most intense immunoreaction was detected using a neutralizing antibody directed against the receptor binding domain S1B of the MERS-S protein, which produced an immunosignal in the cytoplasm of ciliated respiratory epithelium and along the apical membranous region. A similar staining was obtained by two other mAbs which recognize the sialic acid-binding domain and the ectodomain of the membrane fusion subunit S2, respectively. Five mAbs lacked immunoreactivity for MERS-CoV antigen on FFPE tissue, even though they belong, at least in part, to the same epitope group. In summary, three tested human mAbs demonstrated capacity for detection of MERS-CoV antigen on FFPE samples and may be implemented in double or triple immunohistochemical methods. [ABSTRACT FROM AUTHOR] Copyright of Veterinary Immunology & Immunopathology is the property of Elsevier B.V. and its content may not be copied or emailed to multiple sites or posted to a listserv without the copyright holder's express written permission. However, users may print, download, or email articles for individual use. This abstract may be abridged. No warranty is given about the accuracy of the copy. Users should refer to the original published version of the material for the full abstract. (Copyright applies to all Abstracts.)

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DOI: 10.1016/j.vetimm.2019.109939

87. **Wan Y, Shang J, Sun S, et al. Molecular mechanism for antibody-dependent enhancement of coronavirus entry. *Journal of virology*. 2019. DOI:** 10.1128/JVI.02015-19

Antibody-dependent enhancement (ADE) of viral entry has been a major concern for epidemiology, vaccine development and antibody-based drug therapy. However, the molecular mechanism behind ADE is still elusive. Coronavirus spike protein mediates viral entry into cells by first binding to a receptor on host cell surface and then fusing viral and host membranes. Here we investigated how a neutralizing monoclonal antibody (mAb), which targets the receptor-binding domain (RBD) of MERS coronavirus spike, mediates viral entry using pseudovirus entry and biochemical assays. Our results showed that mAb binds to the virus-surface spike, allowing it to undergo conformational changes and become prone to proteolytic activation. Meanwhile, mAb binds to cell-surface IgG Fc receptor, guiding viral entry through canonical viral-receptor-dependent pathways. Our data suggest that the antibody/Fc-receptor complex functionally mimics viral receptor in mediating viral entry. Moreover, we characterized mAb dosages in viral-receptor-dependent, antibody-dependent, and both-receptors-dependent entry pathways, delineating guidelines on mAb usages in treating viral infections. Our study reveals a novel molecular mechanism for antibody-enhanced viral entry and can guide future vaccination and antiviral strategies.Significance Antibody-dependent enhancement (ADE) of viral entry has been observed for many viruses. It was shown that antibodies target one serotype of viruses but only sub-neutralize another, leading to ADE of the latter viruses. Here we identify a novel mechanism for ADE: a neutralizing antibody binds to the virus-surface spike protein of coronaviruses like a viral receptor, triggers a conformational change of the spike, and mediates viral entry into IgG-Fc-receptor-expressing cells through canonical viral-receptor-dependent pathways. We further evaluated how antibody dosages impacted viral entry into cells expressing viral receptor, Fc receptor, or both receptors. This study reveals complex roles of antibodies in viral entry and can guide future vaccine design and antibody-based drug therapy.

URL: http://www.embase.com/search/results?subaction=viewrecord&from=export&id=L630180408 http://dx.doi.org/10.1128/JVI.02015-19 https://jvi.asm.org/content/early/2019/12/05/JVI.02015-19
DOI: 10.1128/JVI.02015-19

88. **Wang L, Xu J, Kong Y, et al. Engineering a Novel Antibody-Peptide Bispecific Fusion Protein Against MERS-CoV. *Antibodies (Basel)*. 2019;8(4):04. DOI:** https://dx.doi.org/10.3390/antib8040053

In recent years, tremendous efforts have been made in the engineering of bispecific or multi-specific antibody-based therapeutics by combining two or more functional antigen-recognizing elements into a single construct. However, to the best of our knowledge there has been no reported cases of effective antiviral antibody-peptide bispecific fusion proteins. We previously developed potent fully human monoclonal antibodies and inhibitory peptides against Middle East Respiratory Syndrome Coronavirus (MERS-CoV), a novel coronavirus that causes severe acute respiratory illness with high mortality. Here, we describe the generation of antibody-peptide bispecific fusion proteins, each of which contains an anti-MERS-CoV single-chain antibody m336 (or normal human IgG1 CH3 domain as a control) linked

with, or without, a MERS-CoV fusion inhibitory peptide HR2P. We found that one of these fusion proteins, designated as m336 diabody-pep, exhibited more potent inhibitory activity than the antibody or the peptide alone against pseudotyped MERS-CoV infection and MERS-CoV S protein-mediated cell-cell fusion, suggesting its potential to be developed as an effective bispecific immunotherapeutic for clinical use.

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DOI: https://dx.doi.org/10.3390/antib8040053

89. **Xu J, Jia W, Wang P, et al. Antibodies and vaccines against Middle East respiratory syndrome coronavirus. *Emerging Microbes & Infections*. 2019;8(1):841-56. DOI:** https://dx.doi.org/10.1080/22221751.2019.1624482

The Middle East respiratory syndrome coronavirus (MERS-CoV) has spread through 27 countries and infected more than 2,200 people since its first outbreak in Saudi Arabia in 2012. The high fatality rate (35.4%) of this novel coronavirus and its persistent wide spread infectiousness in animal reservoirs have generated tremendous global public health concern. However, no licensed therapeutic agents or vaccines against MERS-CoV are currently available and only a limited few have entered clinical trials. Among all the potential targets of MERS-CoV, the spike glycoprotein (S) has been the most well-studied due to its critical role in mediating viral entry and in inducing a protective antibody response in infected individuals. The most notable studies include the recent discoveries of monoclonal antibodies and development of candidate vaccines against the S glycoprotein. Structural characterization of MERS-CoV S protein bound with these monoclonal antibodies has provided insights into the mechanisms of humoral immune responses against MERS-CoV infection. The current review aims to highlight these developments and discuss possible hurdles and strategies to translate these discoveries into ultimate medical interventions against MERS-CoV infection.

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DOI: https://dx.doi.org/10.1080/22221751.2019.1624482

90. Zhang L, Wang X, Jiang L, et al., inventors; Tsinghua University, assignee. Monoclonal antibody MERS-4V2 and its coding gene and application patent CN109666070. 2019.

The invention discloses monoclonal antibody MERS-4V2, which is IgG antibody and consists of heavy chain and light chain, CDR1, CDR2 and CDR3 of the heavy chain variable region have amino acid sequences as 45th-52nd, 70th-77th and 116th-120th from N-terminal end of sequence 1 in the sequence list, and CDR1, CDR2 and CDR3 of the light chain variable region have amino acid sequence as 45th-52nd, 70th-72nd and 109th-119th from N-terminal end of sequence 3 in the sequence list. The invention also discloses the application of the IgG antibody in the preparation of medicament for inhibiting middle east respiratory syndrome coronavirus (MERS-CoV). The invention has wide application prospects for the development of medicaments (vaccines) for treating and/or preventing novel coronavirus MERS-CoV.

991. **Zhao R, Li M, Song H, et al. Serological diagnostic kit of SARS-CoV-2 antibodies using CHO-expressed full-length SARS-CoV-2 S1 proteins. *medRxiv*. 2020:2020.03.26.20042184. DOI:** 10.1101/2020.03.26.20042184

WHO has declared COVID-19 a pandemic with more than 300,000 confirmed cases and more than 14,000 deaths. There is urgent need for accurate and rapid diagnostic kits. Here we report the development and validation of a COVID-19/SARS-CoV-2 S1 serology ELISA kit for the detection of total anti-virus antibody (IgG+IgM) titers in sera from either the general population or patients suspected to be infected. For indirect ELISA, CHO-expressed recombinant full length SARS-CoV-2-S1 protein with 6*His tag was used as the coating antigen to capture the SARS-CoV-2-S1 antibodies specifically. The specificity of the ELISA kit was determined to be 97.5%, as examined against total 412 normal human sera including 257 samples collected prior to the outbreak and 155 collected during the outbreak. The sensitivity of the ELISA kit was determined to be 97.5% by testing against 69 samples from hospitalized and/or recovered COVID-19 patients. The overall accuracy rate reached 97.3%. Most importantly, in one case study, the ELISA test kit was able to identify an infected person who had previously been quarantined for 14 days after coming into contact with a confirmed COVID-19 patient, and discharged after testing negative twice by nucleic acid test. With the assays developed here, we can screen millions of medical staffs in the hospitals and people in residential complex, schools, public transportations, and business parks in the epidemic centers of the outbreaks to fish out the "innocent viral spreaders", and help to stop the further spreading of the virus.Competing Interest StatementThe authors have declared no competing interest.Funding StatementThis work is supported by Research Grants from Beijing Science and Technology Commission, and Bill & Melinda Gates Foundation to Le Sun. This work was also supported by the National Natural Science Foundation of China (NSFC) (81702015), and the National Science and Technology Major Project (2018ZX10733403) to H.S.We thank Professor Wenjie Tan from National Institute for Viral Disease Control and Prevention, Chinese Center for Disease Control and Prevention for providing the sequences of SARS-CoV-2 S1 protien.Author DeclarationsAll relevant ethical guidelines have been followed; any necessary IRB and/or ethics committee approvals have been obtained and details of the IRB/oversight body are included in the manuscript.YesAll necessary patient/participant consent has been obtained and the appropriate institutional forms have been archived.YesI understand that all clinical trials and any other prospective interventional studies must be registered with an ICMJE-approved registry, such as ClinicalTrials.gov. I confirm that any such study reported in the manuscript has been registered and the trial registration ID is provided (note: if posting a prospective study registered retrospectively, please provide a statement in the trial ID field explaining why the study was not registered in advance).Yes I have followed all appropriate research reporting guidelines and uploaded the relevant EQUATOR Network research reporting checklist(s) and other pertinent material as supplementary files, if applicable.YesThe data used to support the findings of this study are available from the corresponding author upon request.

URL: http://medrxiv.org/content/early/2020/03/27/2020.03.26.20042184.abstract
DOI: 10.1101/2020.03.26.20042184

92. **Zhu H, Fohlerová Z, Pekárek J, et al. Recent advances in lab-on-a-chip technologies for viral diagnosis. *Biosensors and Bioelectronics*. 2020;153:112041. DOI:** https://doi.org/10.1016/j.bios.2020.112041

The global risk of viral disease outbreaks emphasizes the need for rapid, accurate, and sensitive detection techniques to speed up diagnostics allowing early intervention. An emerging field of microfluidics also known as the lab-on-a-chip (LOC) or micro total analysis system includes a wide range of diagnostic devices. This review briefly covers both conventional and microfluidics-based techniques for rapid viral detection. We first describe conventional detection methods such as cell culturing, immunofluorescence or enzyme-linked immunosorbent assay (ELISA), or reverse transcription polymerase chain reaction (RT-PCR). These methods often have limited speed, sensitivity, or specificity and are performed with typically bulky equipment. Here, we discuss some of the LOC technologies that can overcome these demerits, highlighting the latest advances in LOC devices for viral disease diagnosis. We also discuss the fabrication of LOC systems to produce devices for performing either individual steps or virus detection in samples with the sample to answer method. The complete system consists of sample preparation, and ELISA and RT-PCR for viral-antibody and nucleic acid detection, respectively. Finally, we formulate our opinions on these areas for the future development of LOC systems for viral diagnostics.

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93. **Zhu J, Kim J, Xiao X, et al. The immune vulnerability landscape of the 2019 Novel Coronavirus, SARS-CoV-2. *bioRxiv*. 2020:2020.02.08.939553. DOI:** 10.1101/2020.02.08.939553

The outbreak of the 2019 Novel Coronavirus (SARS-CoV-2) rapidly spread from Wuhan, China to more than 150 countries, areas or territories, causing staggering number of infections and deaths. A systematic profiling of the immune

vulnerability landscape of SARS-CoV-2, which can bring critical insights into the immune clearance mechanism, peptide vaccine development, and antiviral antibody development, is lacking. In this study, we investigated the potential of the SARS-CoV-2 viral proteins to induce class I and II MHC presentation and to form linear antibody epitopes. We created an online database to broadly share the predictions as a resource for the research community. Using this resource, we showed that genetic variations in SARS-CoV-2, though still few for the moment, already follow the pattern of mutations in related coronaviruses, and could alter the immune vulnerability landscape of this virus. Importantly, we discovered evidence that SARS-CoV-2, along with related coronaviruses, used mutations to evade attack from the human immune system. Overall, we present an immunological resource for SARS-CoV-2 that could promote both therapeutic development and mechanistic research.

URL: http://biorxiv.org/content/early/2020/03/23/2020.02.08.939553.abstract
DOI: 10.1101/2020.02.08.939553

SEARCH STRATEGIES

[PubMed strategy: ((((((coronavirus OR “corona virus” OR coronavirinae OR coronaviridae OR betacoronavirus OR covid19 OR “covid 19” OR nCoV OR “CoV 2” OR CoV2 OR sarscov2 OR 2019nCoV OR “novel CoV” OR “wuhan virus”) OR ((wuhan OR hubei OR huanan) AND (“severe acute respiratory” OR pneumonia) AND (outbreak)) OR “Coronavirus”[Mesh] OR “Coronavirus Infections”[Mesh] OR “COVID-19” [Supplementary Concept] OR “severe acute respiratory syndrome coronavirus 2” [Supplementary Concept] OR “Betacoronavirus”[Mesh]) AND (((detect* OR develop*)) AND (((((((antibody[Title/Abstract] OR antibodies[Title/Abstract])))) AND ((antibody[Text Word] OR antibodies[Text Word])))) OR ("Antibodies"[Mesh] AND ((((“Immune System"[Mesh] OR ("immune system")

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