COVID-19 Decentralized Testing Proposal:

Implementation and Regulatory Strategies

Rule-Out Covid19 Working Group / ruleout.org

Draft v200407v1

Introduction: "Rule-Out" COVID-19 Testing

The COVID-19 pandemic has stretched diagnostic testing capacity; the number of tests available are inadequate to meet population needs. Current tests are designed for "rule in" use: the result is aimed toward confirming that suspected patients HAVE the disease. With wider deployment, current tests may eventually also be used to try to find asymptomatic people WITH the disease--but that has been a slow process To deal more quickly with the vast number of people needing testing, we propose an additional approach: a Rule-Out Test.

The proposed testing approach utilizes existing equipment, pulls from areas of the supply chain not currently under greatest strain, and recruits non-medical but highly trained laboratory personnel to assist in testing, without increasing their risk of infection.

Ruling out COVID-19 viral infection in large numbers of people is important in that it:

- Provides helpful testing in a population with unmet needs (perceived or real)[1]
- Serves as a gateway to isolate asymptomatic but likely positive patients,
- Could serve as a screening tool for returning to workplaces and schools, and most importantly
- Allows focused allocation of the limited conventional "rule-in" tests to the patients who most need them.

The proposed approach is flexible and allows for a mobile rapid deployment to hotspots as they emerge. For example, there are 326 accredited hospitals across California, who could add up-to 500 tests/day in capacity on a 10 hours shift using this test. This would add over 163,000 tests/day capacity to the system in California alone. Moreover, if unnecessary regulatory barriers were suspended during this emergency, additional capacity could be put in place at associated testing locations.

We are a group of volunteers sponsored by a 501(c)(3) non-profit. We will make the tools we have developed available to others on an open source basis.

Problem Statement

Progress has been made recently on additional COVID-19 testing capacity through the mobilization of additional private and quasi-public actors: FDA emergency policies allows highly capable clinical and research labs to self-validate a "Lab Developed Test" (LDT) for the SARS-CoV-2 virus to notify the FDA, and then to deploy that test at their facility. On a similar FDA path, highly capable commercial labs can develop and validate test kits that allow test samples to be collected remotely from the commercial lab and returned to those labs to be read. On both paths, the FDA expects labs to show that their tests can identify both positive COVID-19 cases and uninfected cases with a low error rate.[2] However, there are now some reports that COVID-19 infections are not being detected in as many as 30% of tested asymptomatic cases, i.e., that many no-infection conclusions are incorrect.[3]

These FDA policies have allowed new LDTs and test kits to be deployed while applications for EUAs are pending. But scaling up has been difficult, total available testing capacity is still grossly inadequate, and turn-around times can be too long.

In addition, relying on a specimen reading step that is often remote in space and time from sample collection requires too many hand-offs, too much transport, too much tracking, and too much delay—which means critical decisions about admissions, treatment, isolation, whether health care workers have been exposed, contact tracing, and returning to

[1] First responders, hospital employees, maintenance and cleaning staff, grocery workers, delivery workers, employees with large public contact, persons with close family/work contacts positive for coronavirus, etc.
[2] See: https://www.aacc.org/global-health-outreach/how-labs-can-prepare-for-coronavirus-and-other-outbreaks
[3] This paper finds a 72% sensitivity for a single NP swab. https://pubs.rsna.org/doi/10.1148/radiol.2020200432

work are uninformed or delayed. These problems are most acute where health care decisions must be made at smaller and/or rural hospitals and health/employment areas that realistically cannot bring COVID-19 testing in house without substantial help.

Finally, in practice, testing that is still limited to attempts to confirm the status of already suspected infections cannot support reliable inferences about infection rates in a community at large. That leaves public health officers, policy makers, and the public to surmise the state of the world, and undermines public health messaging and decision making.

A widely deployed and robust *screening capability* that is highly reliable in identifying negative cases is needed now. Higher volume testing would help to identify still asymptomatic individuals who might already be infected, for follow-up testing and earlier isolation. Broadly available testing would also provide useful aggregate population information, and could help to ensure that conventional testing resources can be well-targeted.

Our Solution

Phase One: 'On-Campus' Test Sites

The Rule-Out group will build testing capacity in two phases. In our first phase, we will work with partners to deploy our protocol at as many interested and appropriately certified laboratories as possible. We will also allow others who can do so to use our open-source tools to stand up additional laboratory-tied sites on their own. This is Phase One because these deployments will not depend on any further changes in currently applicable federal or state laboratory oversight rules and policies. FDA emergency policies issued in February and March already allow any laboratory certified pursuant to the Clinical Laboratory Improvement Act (CLIA) for high complexity testing to validate and deploy a Lab Developed Test (LDT) for SARS-CoV-19, while an application for emergency use authorization (EUA) is pending. Those policies also allow LDT validations and approved EUAs to be extended to additional laboratories more easily.

All labs pursuing an LDT for SARS-CoV-19 benefit from these policies, but our test can be scaled more easily than others in this environment because it mitigates other important constraints.

First, test complexity is itself an obstacle to scaling. For technical reasons, a 'conventional' test that is expected to provide high confidence for both positive and negative results must be more complex in design and execution than a rule-out test, and therefore will inherently be slower to return results and more difficult to scale. Our rule-out test is more streamlined and faster. For any given instrument we can complete many more tests per unit of time.

Second, because 'conventional' tests must be complex, those test protocols tend to be rigid, and that makes these tests logistically fragile—if any part of the kit or protocol is not available (for example, a specific reagent is in short supply), testing is impossible until a fix can be found. Our rule-out test is less fragile.

Third, we expect that our open-source /volunteer / non-profit approach to meeting testing needs will enable us to form effective partnerships to help speed implementation of this concept. Our concept includes partnerships with data system providers to address patient interface, tracking, and reporting. We hope that universities, with encouragement by states, will make instruments available for use at test sites. We will be able to put this protocol in place at existing CLIA-certified laboratories without those laboratories developing or validating an LDT on their own--but we can still rely on in-place Laboratory Directors and Medical Directors to satisfy underlying CLIA requirements, and to oversee this new testing effort once in place. We can work with other non-profit organizations and governments to put any available emergency response or donor funding to work. And if a partner or open-source operator chooses to charge for some or all tests, the efficiency of our protocol will allow those charges to be within CMS guidelines.

Phase Two: Distributed Test Sites

In our phase two, which we are already working to enable, we hope to deploy test sites on a distributed basis, not just on the campuses of CLIA-certified laboratories. We also want to ensure that these additional test sites can be staffed and supervised. To achieve those goals we must persuade federal and state actors to waive or to modify certain regulatory requirements for the duration of this emergency. Doing so would be appropriate: CLIA requirements are about maintaining and improving quality in clinical laboratories, not about dealing with an emergency requirement for much more testing capacity of a particular kind.

This regulatory landscape is still fluid, so we do not set out in this paper the specific regulatory changes we will be seeking. However, we are focused on three main issues. First, finding an acceptable way to allow test sites to be set up where they are needed. That could be done in several different ways. Second, leveraging supervision by qualified laboratory directors and medical directors to these additional testing sites. And third, enabling capable research laboratory staff--graduate students, post-doctoral researchers, and technicians--to staff these additional sites.

The Rule Out team is engaging with federal and state officials on these issues in more specific terms, and will update its suggestions and requests as agencies continue to streamline their requirements.

Implementation Concept

Facilities, Staffing and Oversight

1. Temporary structures; labs deployed where needed
    a. Tents from event planning companies
    b. Mobile donation buses
    c. Temporary labs are allowed “on campus” at CLIA-certified high complexity facilities during this emergency
    d. Other: We will seek flexibility for labs at more remote sites, and for mobile labs

2. Deployed Labs: All test steps will be completed on site
    a. Sample collection
        i. Trained laboratory personnel collect
        ii. Possibly observed self-administration
    b. Tracking: using an existing patient interface platform
    c. Transport: None needed
    d. Sample preparation: simplified and completed on-site
    e. High-throughput Assay
        i. Same day results, or faster
        ii. Automated results reporting
    f. Throughput: Potential throughput of 6,000 tests per site per day

3. Instrumentation: Securing and mobilizing required instruments
    a. Target instruments are in public research facilities
        i. State support will be sought to repurpose these
    b. Instrument installation:
        i. Instruments are dedicated and installed at each location
        ii. Mobile instruments used as the end point for assays at some sites
            1. Rapidly deployable to hot spots if excess capacity is required

2. Rapidly deployable instruments would be used to their capacity

4. Staffing
    a. Technicians
        i. Number: Two laboratory technicians required for each remote or mobile site
        ii. Licensing:
            1. Staffing will conform to current CLIA requirements for licensure and supervision unless requirements are waived or modified
            2. Additional unlicensed technical staff to perform possible unlicensed activity (specimen registration, fixed pipetting, initial processing) can be recruited from the pool of furloughed research associates
        iii. Multiple staffing shifts can be implemented based on need.
    b. Oversight:
        i. Medical Directorship Lab Director licensing in compliance with CLIA regulations (unless waived or modified)
        ii. These Directors and Clinical Lab Specialists will be leveraged for oversight of additional test sites as allowed
    c. Drivers
        i. In a rapid mobile setting (eg---modified blood mobile or van), a driver will also be required

Funding and "Business" Expectations

- Rule-Out team members are volunteering their time to launch and oversee this process
- Our tools are open source, but users will be required to accept basic terms addressing liability, reporting, and other legal issues. Agreements for start-up or ongoing support by Rule-Out.org can be arranged.
- Logistical partners and test site operators should expect to absorb or otherwise fund their staffing costs, testing costs, PPE and hazardous waste costs, reagent costs, etc.
- We are arranging for fiscal sponsorship by the Darwin Foundation, which is dedicated to facilitating genomic research and diagnostics. This will allow us to accept charitable donations from individuals and business entities, grants from foundations, and in-kind support from partners. This sponsorship will not extend to government grants, but we are seeking an additional 501(c)(3) fiscal sponsorship with a partner experienced in accepting government grants that include audit requirements.
- We expect instruments and required equipment to be lent (ideally at no cost) for use at sites where there is no per test charge. However, we have no such arrangements in place at this time.
- Estimated direct costs to set up and validate a test site are $15,000 to $20,000, with operating costs of about $50 per test for a site operating at capacity.

Logistical Issues and Planned Solutions

- Validation of test methods per FDA emergency policies
    - Self-administered and reported to FDA
    - Adjustments only require "bridging" data
- Immediate emergency use and eventual EUA per FDA emergency policies
    - Will seek application of EUAs to additional sites based on "bridging study" policy
- A core set of procedures developed by experienced licensed pathologists (laboratory inspectors for the College of American Pathologists) will offer standardized structured guidance via SOPs to validation processes, competency training, testing protocols, quality measurements, and reporting.
- Testing methods are performed under laboratory developed test (LDT) guidelines with local medical director approval

- Set Up Timeline
    - Tent, Buses and Trailers: 1-3 days
    - Assay qualification: 4-10 days
    - Inspection and Qualification: 1 day
- Required Materials
    - Supplies
        - NSP Swabs
            - Possible short-term backorders; companies are ramping up production
        - Viral Extraction Kits
            - Possible short-term backorders; companies are ramping up production
            - Alternative methods to be evaluated
        - Personal Protective Equipment (PPE)
            - Possible short-term backorders; companies are ramping up production
        - Sanitation materials
    - Purchasing: Bulk purchases planned with distribution to partner test sites
- Waste management (Biohazard and Non-biohazard material)
- Funding
    - Some costs absorbed by some partners
    - Foundation grants or donations to fund charitable test sites
    - Cost recovery contracts to fund other test sites
    - We expect to be able to accept government grants in the near future

Sources of Expertise and Equipment

- Laboratory Medicine professionals who are experienced in CLIA and state regulations and have abundant experience as medical directors of CAP-accredited high complexity laboratories
- Company and University labs with required instruments and ancillary equipment
- Staff for set up and running of mobile laboratory units
- Dissemination of unified set of protocols, SOP, methods and resources, and compliance including competency determination

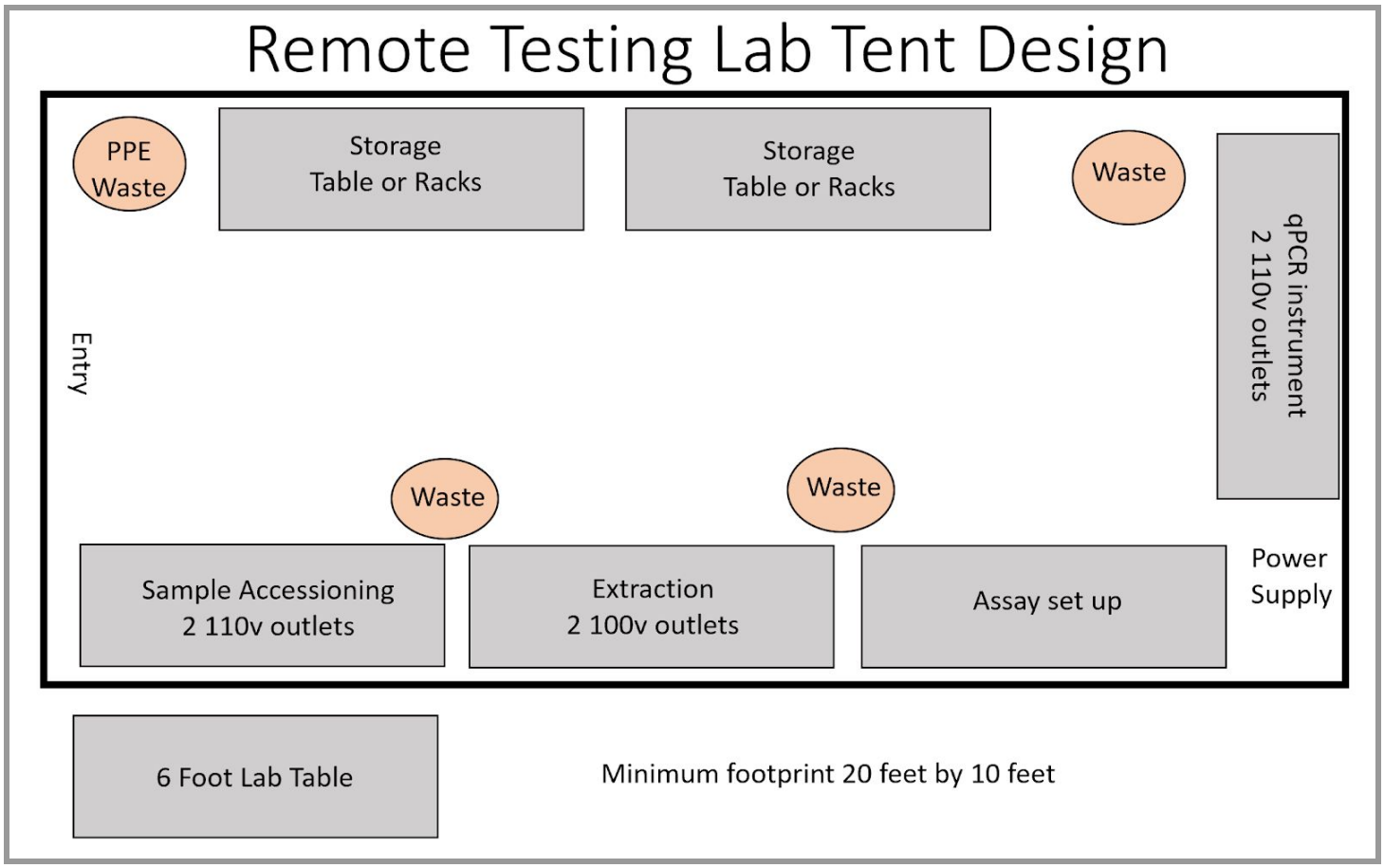
IT Infrastructure

- Website/application development--in house volunteers
- Patient registration interface and results database management--talking to potential partners
- Automated results reporting--talking to potential partners
- Testing proficiency tracking--build on test site partners' capabilities

COVID-19 Testing Technical Considerations

Facilities

To house the necessary equipment an environmentally isolated space of no less than 20' x 10' would be required. This area will need 6 tables, each measuring 6 feet in length. Power requirements are a minimum of 6 standard 110v outlets. This space will need to maintain an internal temperature between 20°C-25°C (68°F to 77°F). Options include tent structures, mobile blood donation buses, or outfitted delivery vans; additional facilities in schools (cafeterias, gyms), universities, churches, neighborhood community centers, etc., could be used if, upon inspection they are deemed adequate.



Pre-Analytical

Coordination with local government will speed the adoption of drive through testing. Traffic patterns in and around the remote testing sites should be set up and controlled by cities. Samples will be taken by trained staff and rapidly handed off to the lab staff. At this point the de-identified sample will be accessioned and entered into a database and placed in the sample preparation queue. This process ideally involves barcoded sample collection tubes.

Isolation

Viral RNA extraction is the first step of laboratory sample preparation. At this stage a solution is added to the biological sample for the purpose of lysing the biological material and separating the RNA from the remaining biological material. This step is currently performed using kits, such as those from Qiagen. There are legitimate concerns that the demand will outpace manufacturing capacity hence alternative forms of RNA isolation need to be evaluated and approved for use. One such method involves the use of Guanidinum (for example, Trizol®). This is an established method used for over 25 years in laboratories across the globe and currently being used to great effect in South Korea.[4] It involves adding the biological sample (NSP Swab) directly to the Trizol reagent. This will inactivate the virus, reducing the risk of infection to the laboratory staff.[3] This protocol requires only 1 tube Trizol, phenol:chloroform and a standard benchtop centrifuge.

[4] Kochel et al: https://journals.sagepub.com/doi/full/10.1177/1535676017713739

In an ideal configuration, a single technician can handle batches of 24 samples; batch size is predicated on the fact that centrifuges have 24 positions. Using Trizol, The entire process requires 1 hour of technician time. Scaling up sample numbers is a matter of technicians and benchtop centrifuges. There is an almost inexhaustible supply of centrifuges and tubes in this country. This approach shows no reasonable steps that risk a bottleneck or limiting the supply of reagents or equipment.

The critical step at this stage is the transfer of samples into a 96 well plate. This will require 2 technicians working in concert to assure that the correct barcode is added to the assigned well. There are options to use individual tubes with barcodes affixed to the bottom. This can minimize the chance of incorrectly assigned specimens. This process has well established best practices to ensure correct assignment and greatly lower risk of misassociation or sample swapping.

Each of the above steps can be performed using fixed volume pipettes to minimize the need for licensed personnel performing the task. This would open up the vast numbers of trained laboratory staff from research labs and universities who are currently furloughed and unable to go to work. Eliminating the need for licensed staff to perform each task can assure that staffing does not become the bottleneck.

Testing and Measurement

The process described in Won, et al[5], uses standard qPCR and a 384 well plate. This evaluates 48 samples per run with each sample assay run in duplicate (48 x 2 = 96) and 4 reaction wells used per assay (96 x 4), using the CDC viral specific and human control primers and probes as individual reactions. Many labs have recently forgone the use of one of the viral specific primer/probe sets, increasing throughput by 25%. Our proposal maintains an emphasis on sensitivity and allows incorporation of procedural changes that further increase throughput 33%-50%. This is accomplished using one well for combined viral specific PCR reactions and one well for control PCR reactions. Control reactions confirm that both the extraction and qPCR have worked properly for the patient sample. Again testing in duplicate, this change increases throughput by 50% (96 samples x 2 reaction wells x 2 duplicates).

An additional doubling of throughput is possible using PCR reactions that mimic genotyping approaches when a Taqman qPCR instrument is available. Again emphasizing the need for sensitivity, this method uses two fluorescent probes (i.e. Vic and FAM)—one probe for viral specific reactions and one probe for control reactions. A positive reaction will have results similar to a heterozygous genotyping reaction. This protocol enhancement allows a 384 well plate to perform 192 assays in duplicate.

Finally, we propose a third procedure change that would allow for distributed testing if qPCR instruments are a limiting factor and that is to perform these reactions as end point PCR (there is a large abundance of thermocyclers and small centrifuges) and discard the determination of cycle threshold (Ct value) inferring viral load as viral load is non-contributory in the setting of a screening assay. Confirmatory testing and viral load measurement for putative positives can be accomplished as secondary steps. A significant advantage of the end point PCR approach is that it would decouple the isolation and amplification stages from viral measurement. The small, rapidly deployable, labs can then process specimens in real time and signal measurement can be determined using fixed onsite instruments, portable instruments placed in specially equipped transport systems (such as vans), or transported to a nearby facility with required qPCR instrumentation. The endpoint reading function on qPCR instruments is far more rapid, requiring 10 minutes or less. Throughput can, therefore, be increased by 500% as compared to the standard 40 cycle qRTPCR run performed on the qPCR instrument.

[5] Won J, et al. *Experimental Neurobiology* Pub Online: March 11, 2020 https://doi.org/10.5607/en20009

IT Infrastructure

The IT requirements for this project revolve around physician referral, patient enrollment, sample tracking, result generation and result dissemination. In an effort to speed the drive-through testing, patients would pre-enroll for the test by entering their information on a web or mobile friendly interface. They will then be directed to an appropriate sample collection location along with a QR code to display to the on-site staff. The testing procedure at the drive-through location should involve no more than scanning the patient's QR code pre-generated by the enrollment interface and verification of identity. This will then be associated with the sample collection tube's barcode. Ideally the plate used for the sample storage and assay will also have a barcode. A csv or xls file of the assay plate using de-identified barcodes to identify samples will be required as input for the qPCR instrument used to define the sample well location. Results from the instrument can be exported in a csv format. This file will need to be uploaded to a website that associates the sample barcode with the patient ID followed by a text or email notification of the result to the appropriate state agency, hospital and patient.

Certain aspects of the patient enrollment and reporting standards would differ based on the context of the state legislature. In certain states, a physician order is required before enrollment or test result notification. Similarly, positive or inconclusive test results might have to be reported to the state agency.