THE EPIDEMIOLOGY ADVENTURE: BEING A DISEASE DETECTIVE
20.04.2020

Episode 6: Population screening "What to test, how to test, who to test?"

For many people, the main current question is when, and how, will it end? While no vaccine is yet available for COVID-19, public health and social measures will continue to play a key role in reducing infections. According to WHO Director General Tedros Adhanom Ghebreyesus, **no single step will suffice:** "Not testing alone, not contact tracing alone, not quarantining alone, not social distancing alone. Do it all". "Countries cannot fight this pandemic blindfold". This episode covers the use of screening tests to control population exposure, to monitor the spread of the virus and to measure herd immunity.

Testing approaches and strategies

According to WHO, the most effective way to prevent infections is breaking the chains of transmission, is to **test every suspected case** (rapid diagnosis) and **immediately isolate** confirmed cases, and close contacts should be rigorously **traced for self-isolation**.

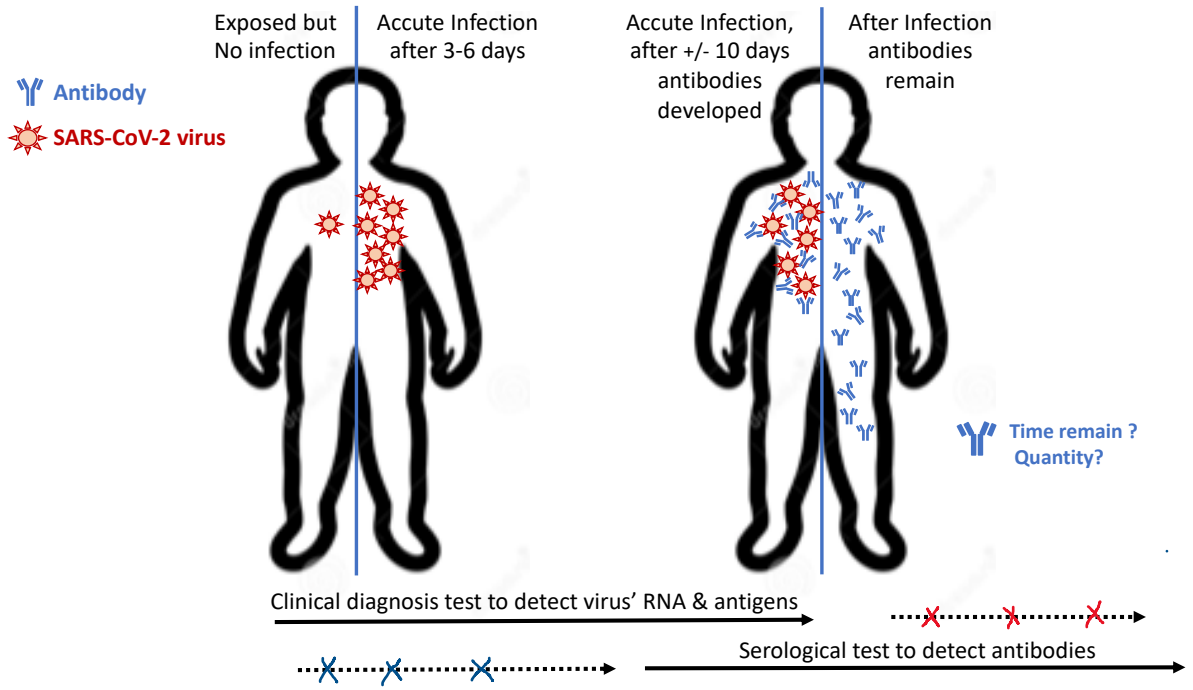
For better community testing, alternative approaches to traditional health-care settings have been introduced to reduce patients and health-care professionals' contamination risks from staying and waiting in hospitals for COVID-19 tests and results. These include **drive-in testing** and home-based self-testing allowing to decrease the risk of contamination in waiting rooms. Drive-through centers were extensively used in South Korea, but now spring up all over the world in order to scale up levels of testing.

However, the shortage in laboratory supplies and reagents has become a bottleneck for testing in some parts of the EU, UK and US. This shortage affects diagnosis capacity and hampers the epidemic response at national and local levels [3]. Optimized testing strategies have to be considered.

Current test method for COVID-19

It is important to distinguish between two test methods which respectively detect different aspects of the SARS-CoV-2 virus (Fig. 1), i.e. the presence in the body of the virus (exposed cases), or the presence of antibodies (immunity level). They have different purposes, benefits and limitations, and are not performed at the same stage of the COVID-19 disease.

Figure 1: Testing at different stages of COVID-19 disease



A diagnosis test is used (1) to detect current infection or the presence of the virus to identify current cases. A serological test can be used (2) to verify the presence of antibodies to verify whether the person has been infected in the past.

1. **Testing the current presence of the virus, a diagnosis test for infected cases**. Tests can be performed for detecting either the SARS-CoV-2 genetic material (the virus' RNA) or to detect viral proteins (the virus' antigens):
   1.1. **Laboratory RNA test for the presence of the virus:** The RT-PCR[1] test is currently recommended for COVID-19 disease diagnosis by the European Center for Disease Prevention and Control and WHO. This test detects the virus' genetic material called RNA (appendix 1: more on RNA viruses). It is used to confirm whether an individual is **currently infected**. But once you have recovered and the virus is eliminated, this test can no longer tell you if you have been infected. The WHO testing protocol includes: (1) specimen collection on respiratory samples or sputum samples obtained for example from nasal secretions from the back of the nose; (2) storage and shipment of the specimens; (3) communication with the laboratory; (4) laboratory testing; (5) results reporting. This test can be seen as a "gold standard" for diagnosing an infectious agent, an accurate and sensitive test which can detect as little as one virus particle (appendix 2: more on accuracy of tests). The test allows for consistent data but requires sophisticated laboratory equipment, technical expertise, takes time and has stretched capacity in light of the large COVID-19 disease outbreak and the speed of the epidemic. PCR tests are easy to design but difficult to miniaturize due to the need for amplification to detect RNA. However, with the latest technologies, portable devices may become more available, hence, time to results could be reduced to 2- 4 hours, while it takes up to 1-2 days if samples must be transported to a testing laboratory.

   1.2. **Testing outside laboratory settings: Point-of-Care Testing (POCT[2]).** To overcome supply shortages of laboratory-based testing, the availability of reliable easy-to-use test kits are needed to deploy widespread testing, without shipment of specimen or laboratory involvement. Such medical testing devices can be used, at or near the place of patient-care, for the **rapid detection of infection, to accelerate clinical decision-making**, and influence the way patients are treated. **Timely and accurate tests are an essential** part of the management of COVID-19 infections.

   An example of POCT is the Rapid Diagnosis Tests (RDT) for antigen detection. Like RNA, antigens are also present in the respiratory tract of infected individuals (sputum, throat swab), and can be used to diagnose acute phase infection. Antigens are proteins on the surface of viruses that are recognized by our immune system. Antigens are "targeted" by antigen-specific antibodies. If the target antigen is present in sufficient concentrations in the sample, it will generate a visually detectable signal, typically within 30 minutes. The antigen will only be detected when the virus is actively replicating, hence these tests are best to **identify acute or early infection**. The **quality for these tests varies** and sensitivity could be ranging from 34% to 80%[3]. Even with 80% accuracy, one in five test results could be wrong. Also, false-positives can result if the test strip recognize antigens of other coronaviruses like the one that causes a common cold. As accuracy of such tests becomes adequate, these antigen tests could be used as **triage tests to rapidly identify patients** very likely to have COVID-19, and it would reduce the need for expensive laboratory RT-PCR testing.

---

1 Reverse transcription polymerase chain reaction is a laboratory technique primarily used to measure the amount of specific viral RNA in research and clinical settings

2 Point-of-care is defined as medical diagnostic test performed at or near the place of patient care, like for example at the pharmacy or at a private medical practice

3 Based on tests for other respiratory diseases, sensitivity expected to vary between35%-80%. How well test works will depend on viral concentration, quality of specimen, formulation of reagents in the kit. WHO Scientific brief 8 April 2020

TABLE 1: Overview COVID-19 tests for detection of infected cases: RNA or Antigens

Type of test	Time to results	What it tells us	What it cannot tell us
Laboratory RT-PCR test (test tube) Currently the recommended method	Slow (2-4 hours but often up to 1-2 days), and expensive	High accuracy and sensitivity for viral RNA detection. Indicates the current presence of the virus in the body. Allows to identify current infected cases.	PCR relies on amplification of RNA, so is limited to be used when the virus, i.e. RNA, is still present. After recovery, when the virus is eliminated from the body, the test will not detect anything.
RDT Antigen test (nasopharyngeal secretions on paper strip)	Within ±10-30 minutes Low cost	Triage to identify likely infected cases. Antigens detected are expressed only when the virus is actively replicating. Best used for acute or early infection	Quality varies and If low sensitivity, up to half of positives might be missed. After infection the test will not identify past infections.

Source : WHO scientific Briefing 8/4/2020 [11]

RT-PCR tests will remain the backbone of testing performed centralized laboratories, but RDT, especially rapid antigen tests, are needed for suspected cases, to accelerate clinical decision-making and take workload of the laboratories. WHO has identified rapid POCT as a research priority. [11]

2. **Testing for the presence of antibodies of COVID-19**. The aim is to check whether an individual has been infected at some point in the past. Antibodies are Y-shaped proteins produced by the immune system in response to exposure to antigens. They belong to a family of large molecules known as immunoglobulins, playing different roles in the immune defense strategy. Studies suggest that the antibody response to SARS-CoV-2 develops only in the 2nd week after onset of symptoms but lasts much longer in the bloodstream than the virus itself. Antibody detection is an important part of the COVID-19 strategy for three reasons:
   - It provides a sensitive assay to obtain information about how widely the virus has spread throughout the population, even with minor symptoms or asymptomatic. The level of herd immunity can be determined (previous episode 3), and we could investigate the role of specific population groups (children, youth and health professionals) in spreading the virus.
   - Aids in determining who may donate a part of their blood called convalescent plasma, which may serve as a possible treatment.
   - Detection of antibodies is a tool used to check if vaccines work.

   For this type of research, antibody tests are needed (Table 2). Hence antibody tests are mostly useful in providing a **historical picture of the past infections**, but they have a **limited utility for clinical diagnosis** because they cannot quickly diagnose acute infection.

Table 2: Available types of COVID-19 antibody-tests, with its own advantages and limitations.

Type of test	Time to results	What it tells us	What is cannot tell us
Rapid Diagnosis Test (RDT)	10-30 minutes	Check for past infections. The presence or absence (qualitative) of antibodies against the virus present in patient serum. Used best in the recovery face, for people who believed to have been infected.	The quantifiable amounts of antibodies in the serum, or if these antibodies are able to protect against future infection. Limited usefulness for early detection as antibodies only appear in the 2nd week of infection.
Enzyme linked immunosorbent assay (ELISA). Laboratory test	1-5 hours	The presence or absence (quantitative) of antibodies against the virus in patient serum.	If antibodies are able to protect against future infection. Limited usefulness for clinical diagnosis and early detection.
Neutralization assay. Laboratory test	3-5 days	The presence of active antibodies in patient serum that are able to inhibit virus growth ex vivo, in a cell culture system. Indicates if the patient is protected against future infection	The test may miss antibodies to viral proteins that are not involved in replication.

Source: JOHNS HOPKINS CENTER FOR HEALTH SECURITY website 17/4/2020:
https://www.centerforhealthsecurity.org/resources/COVID-19/serology/Serology-based-tests-for-COVID-19.html

Based on these three types of test methods, some are already approved for research, with different degrees of sensibility and specificity, and used in certain countries. Others are under development. However, they are **not yet approved for use as a public health diagnostic tool** or for at-home diagnosis.

How did some Asian countries have a set-back on the virus?

Some Asian countries apparently did fight this epidemic without some of the draconian measure of almost complete lockdown. Could it be that some strategies are missing elsewhere:

- **In South Korea,** a central part of the control strategy is based on early widespread testing linked to contact tracing and self-isolation. Mass testing allows them to easily identify possible outbreaks. The government uses intensively artificial intelligence and big data analytics to track down contacts of infected cases. Contact investigation is enhanced by the verification of medical facility records, phone tracking system (GIS), card transactions, and closed-circuit television (video-surveillance), which, while perhaps effective, could raise some concern about data protection. [3]
- **Singapore and Taiwan,** following early recognition of the crisis, were able to quickly mobilize resources and to deploy widespread testing, combined with digital surveillance to trace individual's movements and impose strict quarantines in suspect cases, in addition to building stockpiles of personal protective equipment and masks. Taiwan leveraged its national health insurance database and integrated it with its immigration and customs database to create big data for analysis. It also used new technologies including QR code scanning and online reporting of travel history. As early as 31 December 2019, Taiwanese officials began to board planes on direct flights to assess passengers before they could deplane. Symptomatic passengers were quarantined and those identified as high risk (under home quarantine) were monitored electronically through their mobile phone. Singapore quickly performed aggressive contact tracing and quarantining. All confirmed cases were isolated until two consecutive RT-PCR tests became negative over two days, and those individuals without symptoms were quarantined for 14 days. [7][8]

Overall, certain countries learned from previous coronavirus outbreaks (SARS 2003, MERS 2015) and through major investment in pandemic preparedness, they have strengthened their ability for early recognition and rapid actions to manage infectious outbreaks.

Discussion: Implications for lifting gradually social measures

As social measures will start to be gradually lifted, screening becomes even more important. Testing of suspected cases is an important part of the strategy to timely identify infected cases. Moreover, testing for the presence of the virus is the **start of a sequence of measures to interrupt further transmission**. Until a vaccine becomes available, to go back to a normal life, we need widespread easy reliable **testing for early detection**, rigorous **contact tracing** to limit spreading (facilitated by mobile phone apps), and people's understanding of **quarantining and precautionary self-isolation**. Stockpiles of personal protective equipment should be widely available. Serological screening for antibodies will furthermore provide relevant insights to adjust the public health measures. It is important to consider that in Western democracies, we will additionally all rely on everybody's individual sense of responsibility to **practice measures for self-protection and to protect each other**, in order to respond to resurgent or imported cases and stop the virus spreading. The costs of these control measures may be high in the short term, but efficient control will reduce the economic and social costs of social-distancing and business lock-down measures in the long term. [1][6]

Please excuse any oversights we may be blind to and feel free to contact us and let us know of any "errors and omissions".

Michele Boulade
MSc, MA
Nutritional Epidemiology & Public Health
Michele.boulade@bridging-solutions.ch

Cristina Alvarez
MVD, MSc, PhD
Nutritional Epidemiology & Public Health
cris@suidgeest.com

Appendices

Appendix 1: RNA

Ribonucleic acid (RNA) is a polymeric molecule essential in coding, decoding, regulation and expression of genes. Unlike DNA, RNA is single strand folded onto itself rather than a paired double strand. An RNA virus is a virus that has RNA as its genetic material. Human diseases caused by RNA include the common cold, influenza, SARS-CoV-1, SARS-CoV-2, hepatitis C & E, Ebola, polio and measles. RNA viruses generally have high mutation rates compared to DNA viruses. This is one reason why it is difficult to make effective vaccines-diversity is their strength.

Coronavirus are enveloped RNA viruses This means that they have a lipid membrane (greasy film), which can be dissolved with soap and water or disinfectant, thereby inactivating the virus (OFSP).

Appendix 2: Test quality: sensitivity and specificity (Webb and Bain, 2011):

Reliable tests should be accurate. We expect them to be sensitive (find as much as possible cases) and specific (prevent misclassification) (appendix 1). Inadequate tests may miss patients with active infection (low sensitivity) or falsely categorize patients as having the disease (specificity). Moreover, tests should be safe and acceptable, and preferably simple and cheap if we want to screen a large proportion of the population.

We can evaluate a test against a "gold standard" that ideally would give 100% correct results. But this standard might be too costly and time-consuming, or not suitable for routine testing.

There are 4 possible outcomes for a test:

- Infected cases: can test positive (true positive) or negative (false negative)
- Non-infected case: can test positive (false positive) or negative (true negative)

For a test to be accurate it should produce few false-positive and few false-negative results.

The sensitivity of a test measures how well it classifies the infected cases, true positives

$$Sensitivity\ (\%) = True\ positives - all\ infected\ cases$$

The specificity measures how well the test identifies those not infected, true negatives

$$Specificity\ (\%) = True\ negatives - all\ non\ infected\ people$$

A combination of high sensitivity and high specificity is essential.

For large population screening we could imagine a simple screening test and whoever fails (tests positive, the true positives and false positives) will be followed up with formal diagnosis testing to determine the true positives.

So, if a trade cut-off has to be made between sensitivity and specificity, the optimum point has to be selected depending on the consequences of missing a few positives or falsely classify more negatives as positives. That decision will depend on the impact in the population. If early detection greatly reduces mortality or morbidity and if all true positives could be identified quickly so we can isolate and treat them and break transmission mechanisms, then we would tend to favor tests with a high sensitivity.

Sources and references

1. *Anderson, R. M., Heesterbeek, H., Klinkenberg, D., & Hollingsworth, T. D. (2020). How will country-based mitigation measures influence the course of the COVID-19 epidemic?. The Lancet.*
2. *Bedford, J., Enria, D., Giesecke, J., Heymann, D. L., Ihekweazu, C., Kobinger, G., ... & Ungchusak, K. (2020). COVID-19: towards controlling of a pandemic. The Lancet.*
3. *ECDC .europa.EU. Technical report 1 April 2020. An overview of the rapid test situation for COVID-19 diagnosis in EU/EEA https://www.ecdc.europa.eu/en/publications-data/overview-rapid-test-situation-covid-19-diagnosis-eueea*
4. *ECDC.euopa.EU. Technical report 8 April2020. Contact tracing-second update. https://www.ecdc.europa.eu/en/covid-19-contact-tracing-public-health-management*
5. *Nguyen, T., Duong Bang, D., & Wolff, A. (2020). 2019 Novel Coronavirus Disease (COVID-19): Paving the Road for Rapid Detection and Point-of-Care Diagnostics. Micromachines, 11(3), 306.*
6. *Salathé, M., Althaus, C. L., Neher, R., Stringhini, S., Hodcroft, E., Fellay, J., ... & Eckerle, I. (2020). COVID-19 epidemic in Switzerland: on the importance of testing, contact tracing and isolation. Swiss medical weekly, 150(1112).*
7. *Wang, C. J., Ng, C. Y., & Brook, R. H. (2020). Response to COVID-19 in Taiwan: big data analytics, new technology, and proactive testing. Jama.*
8. *Wong, J. E., Leo, Y. S., & Tan, C. C. (2020). COVID-19 in Singapore—current experience: critical global issues that require attention and action. Jama.*
9. *Webb, P., Bain, C., & Page, A. (2016). Essential epidemiology: an introduction for students and health professionals. Cambridge University Press.*
10. *World Health Organization. (2020). Coronavirus disease 2019 (COVID-19): situation report, 72.*
11. *World Health Organization. (2020). Advice on the use of point-of-care immunodiagnostic tests for COVID-19: scientific brief, 8 April 2020 (No. WHO/2019-nCoV/Sci_Brief/POC_immunodiagnostics/2020.1). World Health Organization.*