Validating Laboratory Testing During a Novel Pandemic

Aaron Han, MD, PhD, FCAP
Adj Prof MBRU School of Medicine
College of American Pathologists,
Deputy International Commissioner for UAE

September 5, 2020
Opening Comments

• The role of the lab has never been greater than it is today.
• High-quality, adaptable labs are foundational to a strong health care system.
• Accreditation is foundational to building a high-quality, adaptable lab.
• COVID-19 is an excellent case study.
Today’s Presenter: Dr. Aaron Han, MD, PhD, FCAP

• Adjunct Professor of Pathology at Mohammed bin Rashid University (MBRU) School of Medicine
• CAP Deputy International Commissioner for the United Arab Emirates
• Member of the CAP Point of Care Committee
The problem with novel diseases

Graphic Source: https://www.youtube.com/watch?v=Y7XW-mewUm8
What will be addressed during our session

• Development of COVID testing
  o Types of tests
  o Differences between tests

• Thoughts on the near future
  o Serology and convalescent serum
  o Overlap with influenza season

• Pandemic preparedness via accreditation rigor

• Q&A
Usual lab testing framework

- Performance characteristics
- Phases of testing and impact on results
- External quality assurance (EQA), proficiency testing (PT)
- Quality control (QC)
- Validation and verification

FDA marking during the pandemic

- Emergency Use Authorization (EUA)

Graphic Source: https://emmainternational.com/fdas-emergency-use-authorization/
SARS-CoV-2 Proficiency Testing Programs

As laboratories expand patient testing capabilities for detection of or exposure to the novel coronavirus (SARS-CoV-2), we have introduced new proficiency testing (PT) programs to support your efforts.

Enroll Today

Place your order today to ensure material availability.

Order SARS-CoV-2 Serology (COVS) →

Order SARS-CoV-2, Molecular (COV2) →
Types of tests

- Molecular
- Serology

Gold standard is PCR

• Limitations are
  o Pre-analytical
  o Contamination
  o Relapse and detection of dead virus
  o Number of probes in the primer set

What did the UAE do?

LIFE
THE UAE BUILT THE SECOND LARGEST OPERATIONAL CORONAVIRUS TESTING FACILITY IN THE WORLD

The laboratory will be capable of processing tens of thousands of tests a day.

BY SCENE ARABIA - © 02 Apr 2020

Source: https://scenearabia.com/Life/The-UAE-Built-the-Largest-Operational-Coronavirus-Testing-Facility-Outside-of-China

COVID-19 UAE: Dubai conducts more than 600,000 tests

Emirate to continue increasing testing as part of relentless efforts to combat pandemic

Published: June 22, 2020 20:31
Gulf News Report

Source: https://gulfnews.com/uae/government/covid-19-uae-dubai-conducts-more-than-600000-tests-1.1592843566493

Graphic Source: Shutterstock Image 607763987

Source: https://gulfnews.com/uae/government/covid-19-uae-dubai-conducts-more-than-600000-tests-1.1592843566493
PCR in a suitcase – 4 hours to result

Source: Dr. A. Han
Other rapid molecular tests are coming

- Results in 45 minutes or less
- COVID only versus syndromic panel (multiple flu viruses)
- Saliva
- Self/Home sampling
What about the usefulness of a syndromic panel?

<table>
<thead>
<tr>
<th></th>
<th>COVID-</th>
<th>COVID+</th>
</tr>
</thead>
<tbody>
<tr>
<td>RVP-</td>
<td>23</td>
<td>14</td>
</tr>
<tr>
<td>RVP+</td>
<td>60</td>
<td>3</td>
</tr>
</tbody>
</table>

**NPV 95%**
**PPV 38%**

Source: Dr. A. Han, manuscript in review
Serology

• Rapid (fingerstick)
• Non-rapid (ELISA)

• Gold standard is
  o Competent/protective serum
  o Viral assay
  o Plasma therapy

Mayo Clinic launches test to detect COVID-19 immunity

JUNE 12, 2020 BY SEAN WHOOLEY — LEAVE A COMMENT

Mayo Clinic announced that it launched a new SARS-CoV-2 neutralizing antibody test in support of efforts to establish therapies and vaccines for COVID-19.

As part of the Expanded Access Program for Convalescent Plasma and other critical research efforts, Mayo Clinic intends to make the test available to select labs, blood banks and biopharma clients later this month.

Mayo Clinic designed the new test to measure the level of neutralizing antibodies against SARS-CoV-2, the virus causing COVID-19. The test provides semi-quantitative detection of total neutralizing antibodies in human serum. After infection, most will develop an immune response to COVID-19, including the production of neutralizing antibodies against the SARS-CoV-2 spike glycoprotein. However, it’s currently unknown for how long the neutralizing antibodies persist after infection.

The test should only be ordered upon confirmation of prior infection and it should not be used to diagnose SARS-CoV-2 infection, according to a news release.

What was Massachusetts General Hospital (MGH)’s experience?

Source: https://rdcu.be/b6a1q
Update on COVID-19 serologic testing at MGH

What we know – more as of 4/1/2020

In this 9 person cohort:
- Patients start to seroconvert at day 5
  - 50% seropositive by day 7
  - 100% seropositive by day 14
- Infectivity as determined by viral isolation and culture:
  - Naso- and Oropharynx infectious mostly early on in disease
  - Sputum infectious most consistently from beginning through 8 days post-symptom onset
- No live virus isolated from stool (all mild cases)
Update on COVID-19 serologic testing at MGH

Lateral flow assays for Point-of-Care Testing

**Sensitivity of detection**

<table>
<thead>
<tr>
<th>Symptoms</th>
<th>IgM+</th>
<th>IgG+</th>
<th>IgG+ or IgM+</th>
</tr>
</thead>
<tbody>
<tr>
<td>≤7 days</td>
<td>13%</td>
<td>25%</td>
<td>38%</td>
</tr>
<tr>
<td></td>
<td>(1/8)</td>
<td>(2/8)</td>
<td>(3/8)</td>
</tr>
<tr>
<td>&gt;7 days</td>
<td>85%</td>
<td>78%</td>
<td>89%</td>
</tr>
<tr>
<td></td>
<td>(23/27)</td>
<td>(21/27)</td>
<td>(24/27)</td>
</tr>
</tbody>
</table>

Specificity = 100% (0/33)  
(includes 3 non-COVID-19 coronavirus+)

*100% sensitivity at day 12, age <80
Not all assays are similar


Today, the U.S. Food and Drug Administration revoked the emergency use authorization (EUA) of the Chembio Diagnostic System, Inc. (Chembio) DPP COVID-19 IgM/IgG System, a SARS-CoV-2 antibody test, due to performance concerns with the accuracy of the test. Antibody tests, a type of serological test, can help provide information on a person’s and population’s exposure to COVID-19.

“Since the beginning of the COVID-19 public health emergency, the FDA has balanced the urgent need for access to diagnostic and antibody tests with providing a level of oversight that helps to ensure accurate tests are being deployed,” said Jeff Shuren, M.D., director of FDA’s Center for Devices and Radiological Health. “By continuing to monitor authorized tests and emerging scientific evidence, we are able to make changes when appropriate – including taking action when a test’s benefits no longer outweigh its risks.”

Use of serology methods and timing

Before symptom onset | After symptom onset
---|---
Detection unlikely\(^a\) | PCR - Likely positive | PCR - Likely negative\(^b\)

Antibody detection

Source: https://jamanetwork.com/journals/jama/fullarticle/2765837

© College of American Pathologists.

5 September 2020 20
Clinical and immunological assessment of asymptomatic SARS-CoV-2 infections

Quan-Xin Long1,8, Xiao-Jun Tang2,8, Qiu-Lin Shi2,8, Qin Li3,8, Hai-Jun Deng1,8, Jun Yuan1, Jie-Li Hu1, Wei Xu2, Yong Zhang1,2, Fa-Jin Lv4, Kun Su3, Fan Zhang5, Jiang Gong5, Bo Wu6, Xia-Mao Liu7, Jin-Jing Li7, Jing-Fu Qiu6,7, Juan Chen1,8, and Ai-Long Huang1,8

Source: https://www.nature.com/articles/s41591-020-0965-6.pdf
Virology and shedding

Source: https://www.nature.com/articles/s41591-020-0965-6.pdf
Consistent higher IgG response with symptoms

Source: https://www.nature.com/articles/s41591-020-0965-6.pdf
Persistence of IgG in symptomatic patients

Source: https://www.nature.com/articles/s41591-020-0965-6.pdf 5 September 2020
Disease severity and persistence of seropositivity

- **SEVERE DISEASE**: Slower decline may be detectable after 1 year
- **MODERATE DISEASE**: Average of all disease states
- **ASYMPTOMATIC/MILD DISEASE**: Rapid decline in detectable antibody

Serology validation set

• Ideal: Gold standard kit or titers

• Positive samples:
  o PCR+ with immune response (molecular or clinical)
    – PCR clearance
    – Clinical improvement
  o PCR- clinical COVID

• Negative samples: PCR- low risk
  o Other Coronaviruses, HIV

• Avoid comorbid, autoimmune patients
PCR negative results and serology over time

Source: Dr. A. Han
Steps to verifying SARS-CoV-2 antibody assays


**Sample Precision Data**

- **Negative Patient Specimen**
  - Ratio
  - Courtesy of Neil Anderson, MD.

- **Positive Patient Specimen**
  - Ratio
  - Positive patient near the limit of detection
    \[ CV = \frac{SD}{\text{Mean}} \leq 20\% \]

- **Positive QC**
  - Ratio

**Qualitative Analysis**

- Positive Percent Agreement: 60/60 = 100%
- Negative Percent Agreement: 30/30 = 100%
Importance of timing for optimum serology testing

Source: https://www.captodayonline.com/steps-to-verifying-sars-cov-2-antibody-assays/
What is serology’s usefulness?

- Confirm classic cases which are PCR negative
- Epidemiology
- Immunity, need functional correlate
- Vaccine efficacy
NGS and sequencing in the UAE

- Radial phylogenetic tree generated from open-source tools ‘Augur’ and ‘Auspice’ (nexstrain.org)
  - 182 SARS-CoV-2 genomes used
  - 157 obtained from GISAID and 25 genomes in this study

- Filled circles = viral strains color-coded by country of origin
- Branch length for each strain = date for each sample obtained from GISAID
- Dashed black lines = boundaries of SARS-CoV-2 Types A, B, and C

Accessed through: https://www.biorxiv.org/content/10.1101/2020.05.06.080606v1.full.pdf
Summary

• Ideal to have multiple tests
  o PCR: early detection; identify clusters; contact tracing
  o COVID specific
    – Flu syndromic panels
    – Serology: IgM, IgG

• Other diagnostic tests
  o CBC: neutrophil to lymphocyte ratio (NLR)
  o ESR
  o CXR: ground glass opacities (GGO)
  o Temperature and O₂ saturation
New tests

• Saliva
• T-cell response
• Laser
• Canine

COM.40300 and COM.40350

Test method validation/verification

- Clarified method performance specifications to be verified/validated based on the type of testing performed

<table>
<thead>
<tr>
<th>Requirement</th>
<th>Analytical Accuracy</th>
<th>Analytical Precision</th>
<th>Reportable Range</th>
<th>Analytical Sensitivity</th>
<th>Analytical Specificity</th>
</tr>
</thead>
<tbody>
<tr>
<td>COM.40300</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td></td>
<td></td>
</tr>
<tr>
<td>(FDA-cleared or approved – Verification)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>COM.40350</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>(Modified FDA-cleared or approved and LDTs – Validation)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Role and value of the lab in a novel pandemic

- Quality and timeliness of results
- Know the questions that are and need to be addressed
- Know the limitation of your tests
- Have a multipronged approach
CAP accreditation rigor – benefits in a pandemic

• Provides consistent procedural guidance
  o Confidence that your staff is competent as a result of rigorous competency assessment requirements
  o Clear guidance on verification and validation of tests

• 2020 checklist requirement includes enhanced safety features for infectious pathogens
  o New requirement in the Lab General (GEN) checklist
  o Enhancements to the bloodborne pathogen section

• Running PT/EQA provides benchmarking opportunities
  o Capabilities aligned with the industry
  o Large peer groups
  o Educational opportunities for the laboratorians
Recent Updates on COVID-19

September 3, 2020

- The CAP's Morbidity Committee updates its COVID-19 topic center with guidance on screening and surveillance. [Visit here](#).
- We hear you: CAP Responds to Your COVID-19 Questions. [Read the Q & A](#).
- CAP Infection and Pathology Electronic Reporting (PERT) Committee update answers to questions about COVID-19 reporting requirements. [See here](#).

August 28, 2020

- The CAP's Morbidity Committee updates its COVID-19 topic center with guidance on screening and surveillance. [Learn more](#).
- The CAP opposes HHS' lab pandemic and COVID-19 Reporting Requirements. [Read here](#).
References

• Sources: WHO, CDC, CAP, Mayo; will change over time; vaccine, mutations, etc.
• Sources indicated when not CAP
Thank you for your participation and interest.

For questions, email: international@cap.org.