MAY 2020 # 65

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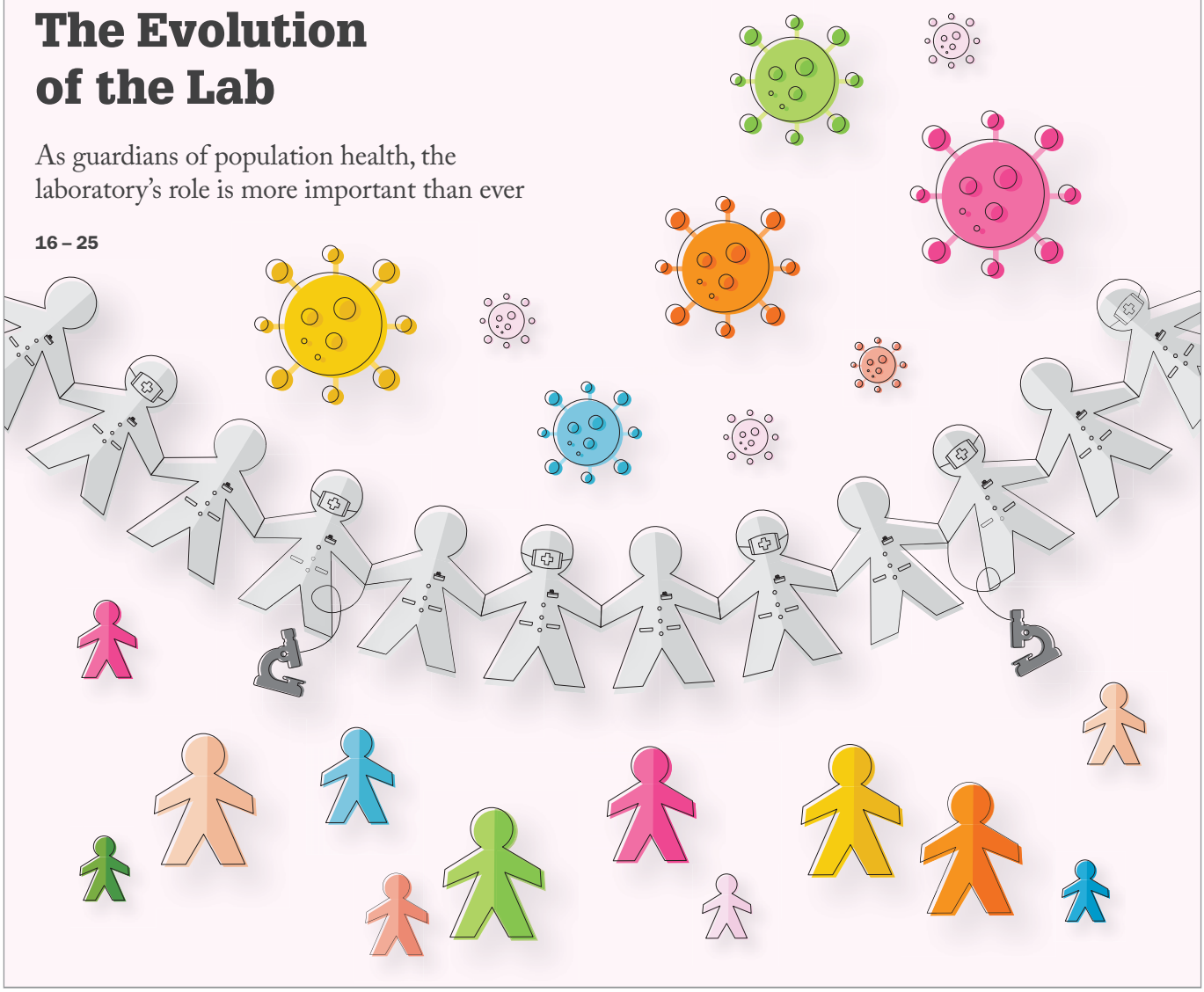

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Trying Times

In the midst of a pandemic, it's not just physical health that matters

Around the world, new traditions have formed. In New York City, for instance, people open their doors and windows at seven o'clock every night, look out of their homes, and applaud the healthcare workers who are keeping the country running. The UK has a similar tradition on Thursday evenings. The "balcony concerts" given by Italians in lockdown have struck international fame via the Internet. Although media coverage focuses on doctors and nurses on the front lines (and, in a rare move, occasionally mentions those involved in laboratory testing), the conversations on social media and in local hubs include other essential workers; for instance, the people who ensure that hospitals and laboratories are kept clean, or those who answer the COVID-19 telephone hotlines.

Cheery news abounds: the 100-year-old man who has raised £32 million for the National Health Service; the children setting up "take what you will" stands outside their homes; the people sewing face masks for bus drivers, retail workers, and other vital (and often overlooked) professions. But on social media, I see a different story. Every day, another pathologist or laboratory medicine professional on Twitter posts that they're taking a break for their mental health. Every day, another voice on Facebook or YouTube or Instagram goes silent to focus inward, rather than deal with the seemingly endless flow of (not always good, often politically motivated) news.

In the midst of one health crisis, are we missing another?

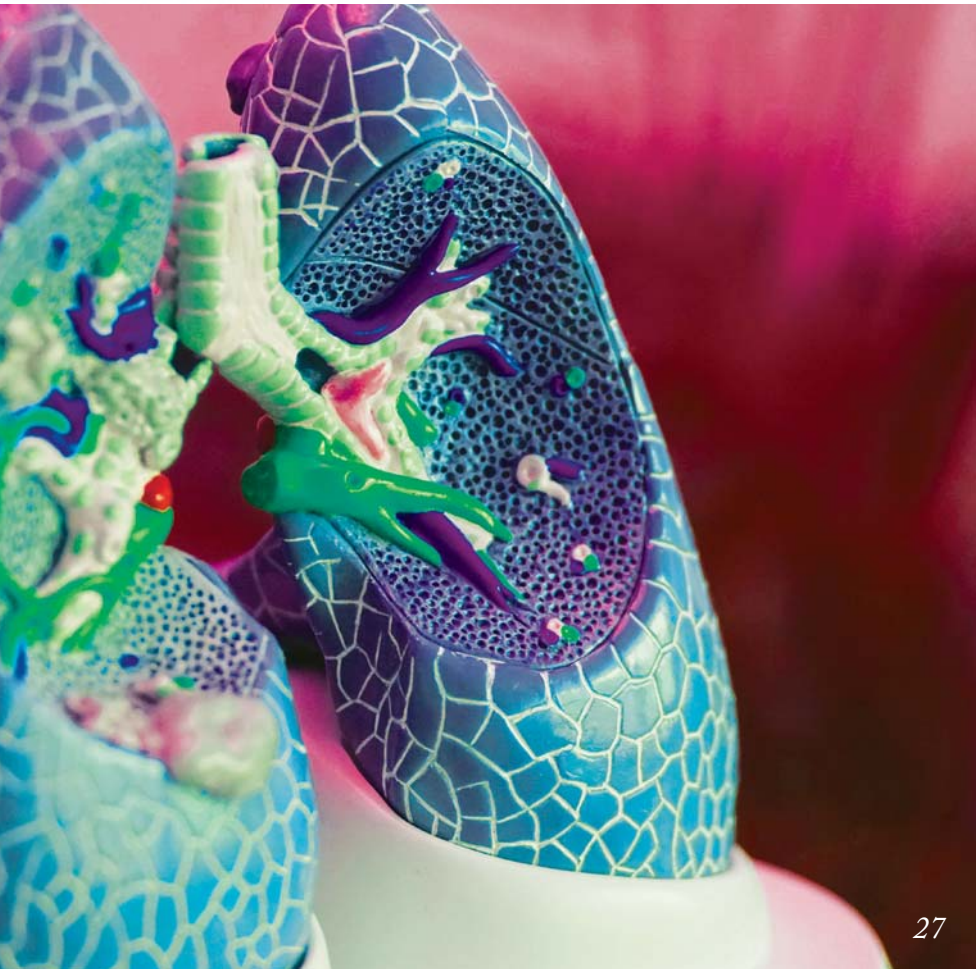
COVID-19 may be grabbing the spotlight at the moment, but it is perhaps more important than ever to be aware of the emotional and psychological effects this pandemic is having – not just on the doctors and nurses patrolling the intensive care wards, but also everyone else in the chain, from the driver who transports the swabs to the pathologist who writes the reports. In the latest installment of our online-only "Pandemic Perspectives" (1), Marisa Saint Martin shares her own approach to maintaining balance and wellness in the midst of a storm. Whether you prefer mindfulness, a workout, a social media break, or something else entirely, remember to pause and take a moment for your own health. Fit your oxygen mask first – before helping anyone else with theirs.

Michael Schubert
Editor

References

1. *M Schubert, "Keeping Pace With the Pandemic", The Pathologist (2019). Available at: https://bit.ly/3buP867.*

Contents



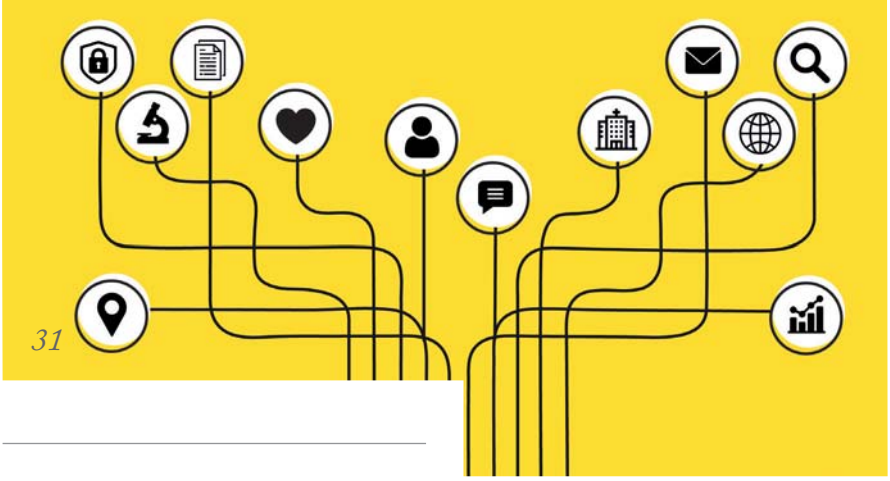
On The Cover

Artist's representation of the laboratory defending the population against diseases such as COVID-19.

Upfront

In My View

From The ASCP



Feature

In Practice

NextGen

Reports

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Sitting Down With

the Pathologist

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Upfront

Research
Innovation
Trends

No More Unknowns?

Scientists have catalogued almost every variant of the *NUDT15* gene to understand its pharmacogenetic effects

Thiopurine drugs serve a wide variety of purposes – from treating childhood leukemias to managing autoimmune disorders. However, not all patients tolerate thiopurines equally well. Until recently, doctors couldn't predict how individual patients might react to treatment – but a research group at St. Jude Children's Research Hospital in Memphis, Tennessee, recently catalogued almost every variant in the NUDT15 enzyme to better understand the potential for side effects (1). Senior author Jun Yang tells us more…

What prompted you to investigate NUDT15?

We first discovered that NUDT15 regulates drug toxicity in 2015 – and the evidence that the *NUDT15* gene can predict side effects of thiopurines continues to grow. There are many variants in this gene, but the vast majority have been not studied carefully, so we do not know if they cause drug toxicity. A couple of years ago, we launched a major effort to map every possible pharmacogenetic variant in the *NUDT15* gene – a lofty goal, but one whose results are very exciting.

What does NUDT15 do?

NUDT15 breaks down thiopurine drug metabolites; its activity is important to keep toxicity in check. Some genetic variants disrupt the protein's function. Patients with these loss-of-function genetic variants cannot break down thiopurine drugs and have excessive toxicities.

Tell us about your new assay…

There are lots of *NUDT15* variants in humans. Traditional characterization requires the creation of each variant protein one at a time – extremely tedious and obviously not scalable. Our new high-throughput method studies the function of thousands of variants simultaneously by introducing each one into a single cell and then characterizing tens of thousands of cells.

Our results offer a comprehensive reference of potential pharmacogenetic variants in *NUDT15*. Most diagnostic laboratories currently test only a couple of variants in the context of thiopurine pharmacogenetics – but there are many more variants equally likely to cause thiopurine toxicities. The data is particularly relevant when *NUDT15* is sequenced and novel variants identified. In the past, these variants would have been considered of "unknown significance." Now, labs can look up their variants in our data for an improved understanding of their results.

Reference

1. *CC Suiter et al., Proc Natl Acad Sci USA, 117, 5394 (2020). PMID: 32094176.*

TIMELINE

The Pandemic To Date

A timeline of the most significant moments in the COVID-19 pandemic so far

December 31, 2019: Pneumonia of unknown cause is detected in the city of Wuhan and reported to the WHO.

January 23, 2020: WHO's Emergency Committee meets to consider the outbreak, with multiple countries now reporting cases.

February 11, 2020: The disease caused by the novel coronavirus is given a name: COVID-19.

March 2, 2020: WHO says the virus is capable of community transmission, but can still be contained.

SPECIAL SERIES
Infectious Disease

BUSINESS IN BRIEF

How is the business world reacting to the pandemic?

Information Exchange
The UK's National Health Service has issued an adoption directive instructing all laboratories to use the National Pathology Exchange (NPEx) for electronic transfer of COVID-19 test requests and results. The aim of 100 percent adoption is to minimize manual processes and the resulting potential for errors and delays (1).

A Digital Blood Count
A new "dry" analyzer needs only two drops of blood to return a complete blood count. OLO first digitizes blood samples by taking over 1,000 images, then automates cell identification and counting. The device is well-suited to quarantine settings in which it can be used to assess the health of COVID-19 patients (2).

Aid From AI
A new artificial intelligence tool that analyzes X-ray images and helps healthcare professionals manage patients with COVID-19 can be accessed for free by hospitals and academic institutes around the globe. Thirona and Delft Imaging launched CAD4COVID to help triage infected patients by indicating affected lung tissue with an abnormality score between 0 and 100 (3).

Trial Tracker
A new COVID-19 global clinical trial tracker has been launched to improve collaboration between researchers, clinicians, philanthropists, policymakers, and other critical stakeholders. The live dashboard details hundreds of ongoing trials, indicating the most promising efforts and helping decision-makers to channel resources appropriately (4).

Visit tp.txp.to/COVID19/BiB to read the full collection of COVID-19 business news!

References

1. *NPEx (2020). Available at: https://bit.ly/2SccEOk.*
2. *ight Diagnostics (2020). Available at: https://bit.ly/2UFqeeK.*
3. *Delft Imaging (2020). Available at: https://bit.ly/3eMIj2v.*
4. *Global Coronavirus COVID-19 Clinical Trial Tracker (2020). Available at: https://bit.ly/3aAWTqq.*

Why Didn't They Teach This in Med School?

A series on new (and not-so-new) medical terms and diagnoses that most of us (probably) missed in training

Curated by Ivan Damjanov

Aichmophobia
(eɪkmə'foʊbiə)
A fear of sharp objects, including needles.

Approximately 10 percent of adult Americans are scared of needles and will refuse injections they feel are unnecessary, such as an annual flu shot.

Dysbiosis
(dɪsbaɪ'oʊsəs)
An imbalance between bacteria forming the normal resident commensal microbiome in certain parts of the human body and new bacteria colonizing that part of the body for pathological reasons.

More than 10,000 fecal microbiotal transplants, colloquially known as "stool transplants," were performed in the US in 2019 to correct large intestinal bacterial dysbiosis in diseases such as ulcerative colitis or recurrent *C. diff*–related pseudomembranous colitis.

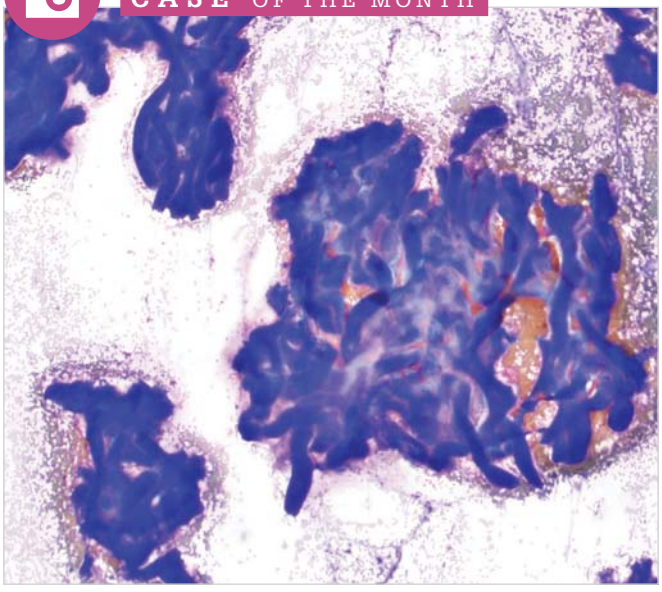
March 11, 2020:
COVID-19 is declared a pandemic by WHO, with the number of global cases now past 100,000.

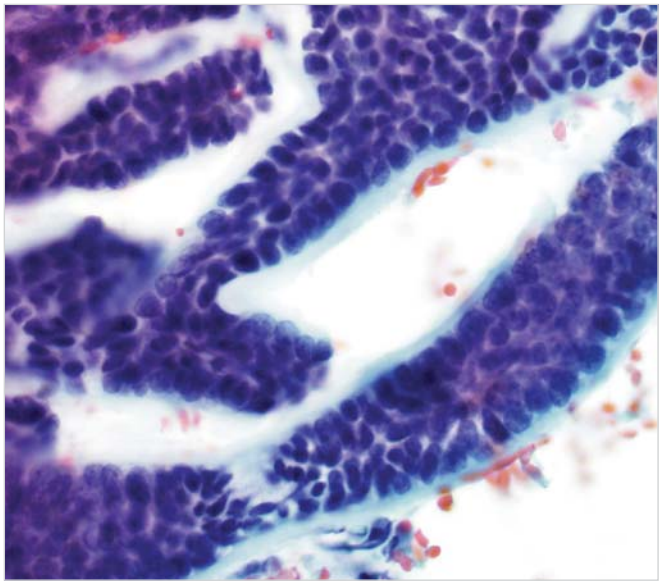
March 13, 2020:
Europe is the new epicenter of the pandemic, with more cases and deaths than the rest of the world combined.

April 13, 2020:
An expert group of scientists, physicians, funders, and manufacturers forms to collaborate on vaccine development.

May 13, 2020:
Over 290,000 people have now died globally from COVID-19.

CASE OF THE MONTH





A 61-year-old woman presents with a 1 cm left postauricular mass that she had initially noticed three months earlier. On imaging, the lesion appears to be well-circumscribed. A fine needle aspiration was performed and yielded cellular smear preparations of which representative findings are displayed in the images below.

What is the most likely diagnosis?

a) Cellular pleomorphic adenoma
b) Basal cell adenoma
c) Adenoid cystic carcinoma
d) Polymorphous adenocarcinoma
e) Epithelial–myoepithelial carcinoma

Answer to last issue's Case of the Month…
d) T-cell prolymphocytic leukemia

T-cell prolymphocytic leukemia (T-PLL) is an aggressive T-cell leukemia that involves peripheral blood, bone marrow, lymph nodes, spleen, liver, and occasionally skin. On peripheral blood smear, the prolymphocytes are small to medium-sized, with round to irregular (and occasionally cleaved) nuclei, visible nucleoli, and cytoplasmic blebs. Immunophenotypically, T-PLL cells are positive for CD3, CD2, CD5, CD7, TCL1, and CD52. Most cases are CD4-positive (rarely CD8-positive);t double CD4/CD8 expression (as observed in this case) is seen in around 25 percent of cases. This "double-positive" phenotype is not usually observed in other peripheral/post-thymic T-cell lymphomas. The most common cytogenetic abnormality, seen in 80 percent of patients, is inv(14)(q11.2q32.1) or, rarely, variant t(14;14)(q11.2q32.1), leading to juxtaposition of the T-cell receptor TRA at 14q11.2 with the *TCL1A* and *TCL1B* genes at 14q32.1.

Differential diagnosis of T-PLL includes T-lymphoblastic leukemia/lymphoma, an immature neoplasm, and other mature T-cell leukemias/lymphomas can be challenging due to overlapping morphologic and immunophenotypic features. Morphologic evaluation, in combination with different ancillary studies and clinical correlation, is usually needed.

Contributed by Laura Baugh and Lina Shao, Department of Pathology University of Michigan, Ann Arbor, Michigan, USA.

Reference

1. *SH Swerdlow et al., WHO Classification of Tumours of Haematopoietic and Lymphoid Tissues, 4th edition. IARC: 2017.*

To register your guess, please go to http://tp.txp.to/0520/case-of-the-month
We will reveal the answer in next month's issue!

Case of the Month is curated by Anamarija M. Perry, University of Michigan, USA.

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In My View

Experts from across the world share a single strongly held opinion or key idea.

Preparing for a Pandemic

The best way to be ready for COVID-19 is to be ready for anything

By Andrew Jaeger, Senior Medical Planner and Principal, HKS Architects, Detroit, Michigan, USA

Is your laboratory prepared to handle the COVID-19 pandemic? The vast majority of laboratories have been seriously thinking about and making plans for a "pandemic situation" for some time, after a surge of renewed interest during the recent Ebola outbreak. However, labs typically need space, staff, and funds to implement their plans – all of which are in short supply. There always seem to be more pressing demands on hospitals' and health systems' limited funding: patient rooms, operating rooms, imaging equipment, emergency department treatment bays, nurses, and more. Another barrier to laboratory expansion is that the equipment and analyzers specifically purchased to provide rapid response testing in a pandemic situation sit idle when they are not needed – and must therefore be revalidated and certified before they can be brought into service. Often, they require unique reagents whose limited shelf life means that they expire before they are needed and must then be restocked.

There has been a fair amount of criticism regarding the slow response of the laboratory community to the need for COVID-19 testing. It's important to keep in mind that, for this (as with all new pathogens), there were no "off the shelf" tests available – and the vast majority of hospital labs don't have the infrastructure or trained staff required to self-develop new, non-FDA-approved molecular testing. Not every lab has facilities for DNA and RNA extraction, "Master Mix" creation, or the development of other specialty reagents not used in routine clinical testing.

But that's not to say that labs can't optimize their available space. Open planning concepts – such as "floors-free" services (power, data, water, and gas from overhead) and end-user reconfigurable mobile benches and workstations – offer the ability to quickly reconfigure areas of the laboratory. The traditional fixed casework and "honeycomb" of rooms found in most laboratories, in contrast, are inherently inflexible and require time, funds, and construction to adapt.

Most clinical labs are adept at flexing their workflows – a daily occurrence in pretty much every hospital-based laboratory. Staffing levels expand and contract with every shift and with the seasons, as do testing volumes. But what happens when the resources run out? "Lean" supply chains and "just in time" delivery models save institutions money by reducing the capital tied up in inventory and can free up valuable laboratory space – but recent emphasis on these approaches has contributed to today's chronic supply shortages. When supplies become scarce, these models tend to crack… or fail completely. If even one link in the supply chain is disrupted, labs can't access the materials they need.

Many labs have also been struggling for years with serious staff shortages. Some have resorted to hiring staff from overseas (at a fairly high cost) or opening their own training schools, but these initiatives require time and space. Investing in a higher degree of automation can compensate for a lack of staff, but only goes so far toward solving a problem that runs much deeper than individual laboratories.

"The vast majority of hospital labs don't have the infrastructure or trained staff required to self-develop new, non-FDA-approved molecular testing."

It's fortunate that laboratory staff are both dedicated and versatile – and, in today's climate, equally good that they are knowledgeable about protecting themselves. Ultimately, however, what laboratories need – especially in times of crisis – is the same as what every other medical department needs: enough supplies, enough staff, and enough flexibility to cope with unprecedented demand.

Bottom-Up to 2020

How can digital champions rise up and lead the way with AI for primary diagnosis?

By Liron Pantanowitz, Professor of Pathology and Biomedical Informatics and Vice Chairman for Pathology Informatics at the University of Pittsburgh Medical Center, Pittsburgh, Pennsylvania, USA

"When will we see mainstream adoption of artificial intelligence (AI) in pathology?" It's a question I hear at every digital pathology meeting across the world – but one that has no definite answer. I know of a handful of labs that use AI every day; some use it prospectively before the pathologist sees the slides, whereas others use algorithms retrospectively to confirm diagnoses. Although small in number, these labs demonstrate that there is a real appetite for AI in pathology and that it can be used successfully. But exactly how long do I think widespread adoption will take? In my view, at least five years. Many people speak of a "third revolution" in pathology; however, before labs can capitalize on AI, it's crucial to first have a proper digital pathology platform – a key obstacle for many slow adopters.

Our advantage at the University of Pittsburgh Medical Center (UPMC) is that we already have a digital pathology platform in place. Although the journey to a fully digital workflow is difficult for any lab (not least because the chance of reimbursement is initially minimal), one of AI's benefits is that it gives clear justification for the implementation of a fully digital platform. The combination of AI and digital pathology makes it a much more exciting journey and increases the chance of buy-in from both pathologists and non-pathologists.

The UPMC healthcare system has 41 hospitals and travel between them can take up to five hours. We run a very distributed operation, with generalists on the periphery and academic medical centers in the heart of Pittsburgh, so we often move difficult cases to the central hospitals. AI will support our distributed model by providing expertise to community pathologists, preventing the need to routinely send cases elsewhere. I am particularly excited by the increased accuracy that AI promises for every single case – and the potential cost savings that result.

As an academic medical center, AI's potential in research is an appealing opportunity for our staff. We've already started to probe images with AI tools to make discoveries not possible through human investigation alone. We have also experienced the reputational benefits of AI. We now commonly have prospective residents ask us whether we are implementing AI. Doing so makes ours a highly attractive program and increases enrolment. Residents want to be trained for the future.

Pathologists are often scared to be the first to do something because of the possible risks (bad press, medico-legal risk, and potential patient harm). For these reasons, we tend to follow the flock. And that's where I think pathology colleges – both in the US and around the world – could do more in terms of setting guidelines for the use of AI. Leading the charge in this way would endorse the adoption of AI and prevent it from being perceived as a rogue activity. And, crucially, it would provide some much-needed clearance on regulatory methods.

Because there isn't a wealth of experience to draw upon when using AI, it's important to have guidance on validation – something I struggled with when validating an algorithm for clinical use at UPMC. I was unable to seek advice from other labs, so I contacted various organizations to ask what guidelines I should follow. There were none. By applying good scientific and laboratory practice, we successfully ensured that the algorithm is safe to use at UPMC – but it certainly delayed the validation process. College guidelines would undoubtedly simplify and expedite the adoption of AI and ensure that everything is standardized for safe practice. We faced a similar hurdle when whole-slide imaging first became commercially available; many colleges stepped up and provided guidelines for validation, which improved adoption and made pathologists feel more comfortable using the technology. That should act as a precedent for AI.

"Our ultimate aim is to implement AI for primary diagnosis in 2020."

Our ultimate aim is to implement AI for primary diagnosis in 2020. We know that will be a challenge – not least because some of our pathologists are determined to stick to traditional microscopes – but we have noticed an increase in requests for digital pathology tools. Instead of a top-down approach, where digital pathology would be forced upon our staff, we've instead opted for a bottom-up strategy that allows champions to rise up, request digital pathology, and start using AI for routine diagnosis. Such an approach will allow a much smoother transition with stepwise changes across different subspecialties – and we hope to reap its rewards in the near future!

Precision Medicine: The Next Generation

Preparing your laboratory for a rising tide of diagnostic tests

By David Fenstermacher, Vice President of Precision Medicine and Data Services at DNANexus, Mountain View, California, USA

Next-generation sequencing (NGS)-based diagnostics are overtaking traditional approaches in a wide variety of indications, including infectious diseases, rheumatology, transplants, human hereditary disorders, and non-invasive prenatal testing (1). Increasingly, NGS technologies are also used for the molecular characterization of tumor subtypes, thereby unlocking the use of targeted therapies in early- and late-stage cancers. Add to this the fact that NGS sequencing costs continue to drop, reimbursement is improving, and patients are gaining more education on what's available to them, and it's easy to see why the number of available NGS tests is only expected to grow (2).

NGS has reached a turning point in diagnosing and treating rare and inherited diseases, which are often difficult to identify clinically. In these complex diagnostic cases, performing whole exome or genome sequencing can detect these rare diseases sooner and direct care appropriately. As the cost decreases and performance improves, more payors are seeing the benefit of preventing expensive and unnecessary tests and avoiding treatment delays. Indeed, diagnostic testing for rare diseases is one of the fastest-growing market segments (3).

Perhaps you have already had to respond to the rapid rise in genetic testing. But are you satisfied with your solutions? Are you confident that they will scale to meet future demands for turnaround time, quality, and accuracy?

The first step for any NGS provider, if you haven't already taken it, leverages another major technological advance: the cloud. Moving your NGS analysis to a cloud-based informatics platform provides an environment that can flexibly scale to meet the demand for increased test volume, while saving time and money. Cloud-based systems enable you to optimize analysis pipelines for quality, speed, runtime, and cost, and can help eliminate bottlenecks in processing queues and server capacity.

If you are looking to expand your footprint, either locally or globally, the cloud can help bring your production pipelines into a single, unified environment, with version-controlled updates rolled out simultaneously across your locales. The cloud-based systems also allow you to decentralize your sequencing among multiple sites or global lab partners, and ensures compliance and intellectual property (IP) protection by keeping your proprietary pipelines centralized and secure in your home region.

Of course, IP is not the only thing you have to protect. Make sure your cloud-based informatics platform has version-controlled tools that allow team members to share large datasets and analyses securely and efficiently, with encryption and tracking to ensure auditability and reproducibility. Ideally, the platform will also facilitate easy compliance with the industry's strict privacy regulations, which are constantly changing and often vary by region. However you choose to develop your informatics approach, you will always have to consider security and compliance – so keep them in mind from the start to avoid trouble down the line.

For a quick "informatics health check," ask yourself the following questions:

- How will the changing genetic testing landscape impact my operations and support needs?
- Is my informatics system sufficient?
- Is it scalable?
- Does it give me the flexibility I need?
- How does it handle quality, security, and compliance?
- Can it help me improve my sample turnaround time or pipeline development?

With the coming deluge of genomic tests in the next five years or less, how can you ensure you're making the right investments today? Should you continue building your own infrastructure or upgrade to a purpose-built NGS informatics platform?

Consider the growth you anticipate in the next few years and the impact on your existing systems. Also consider the hidden costs associated with managing your own NGS informatics infrastructure. For instance, slow compute times, backlogged queues, and cumbersome processes for accessing test data create bottlenecks that can increase your turnaround time. And don't forget the opportunity costs you lose by tying up your resources and headcount on software development. Do you want to continue to invest time, money, and people in operating, scaling, maintaining, securing, and providing support on genomic analysis infrastructure? There are clear benefits to keeping the process in-house – but don't underestimate your own need for support, and don't hesitate to call on it when necessary. By keeping pace with technology and industry innovations in the NGS and genomics field, you can ensure that you are not only ahead of the tide, but making your own waves.

References

1. *1. C Di Resta et al., EJIFCC, 29, 4 (2018). PMID: 29765282.*
2. *2. KA Phillips, MP Douglas, (2018). Available at: bit.ly/37k2nWi.*
3. *3. Concert Genetics, (2018). Available at: bit.ly/2KDmKEc.*

At the Eye of the Storm

COVID-19 has thrust the laboratory into the spotlight

By E. Blair Holladay

At the start of the COVID-19 outbreak, before most of the United States was encouraged to shelter in place, I had the opportunity to visit some of the major laboratories handling the initial cases and see firsthand how they were navigating this uncharted territory. These labs were the epicenters of testing – healthcare leaders getting the right tests to the patients who needed them most. My conversations with their laboratory directors and the teams of medical laboratory professionals highlighted their unprecedented levels of dedication to getting answers for patients and health officials alike. They selflessly put the needs of others ahead of their own, knowing that safe, rapid testing was critical to stopping the spread of COVID-19. That attitude will be essential to fighting our way through to the other side of this pandemic.

The question we face now is, "What does that other side look like?" Unfortunately, we can't know what the world will look like six months – or even one month – from now. What we do know is that we, as pathology and laboratory professionals, are the ones who can shape how the COVID-19 pandemic is handled and, ultimately, how it ends. As testing increases, we have the data that informs health systems and government officials about the crisis and directs care for both individual patients and the population as a whole. And we know that the research we discover and share during this time is essential to moving off the precarious path that COVID-19 has set before us and onto safer ground.

It is medical laboratory scientists' skills and expertise that make us critical to the fight against COVID-19. And it is on behalf of the urgent and demanding work being done by the labs across the country that we at the American Society for Clinical Pathology are pushing the laboratory into the spotlight, emphasizing the integral role the lab plays before, during, and after a crisis.

Throughout the pandemic, we have continued to promote and advocate on the behalf of the medical laboratory community. We've sent Action Alerts urging the federal government to expand testing capacity and allow for remote pathology services. We've called for a national testing strategy. Our subject matter experts have given countless interviews for print, radio, and television discussing the essential nature of the laboratory. We have published multiple editorials around COVID-19 discoveries in our journals. When I visited the medical centers in those early days of the outbreak, we were lucky enough to film people hard at work; from that, we created a docuseries that takes a behind-the-scenes look at the work our laboratories are doing to fight this pandemic. I invite you to see for yourself just how much dedication the laboratory is pouring into the fight against the coronavirus pandemic at ascp.org/COVID-19. We continue to update this page as more resources become available, so we can keep the laboratory community informed.

"We have the data that informs health systems and government officials about the crisis and directs care for both individual patients and the population as a whole."

We are working in challenging times with many unknowns. But we do know this: the pandemic will end. The laboratory will not.

Testing for Gene Fusions

A comparison exercise among commonly used NGS assays

Based on an educational webinar with Wei Song and Phillip Jermann

Genetic rearrangements with contiguous, but unrelated, nucleic acid sequences – so-called fusion genes – often drive malignant transformation (1). Their identification is important and growing in precision oncology research, but can present some challenges. First, it can be time-consuming. Second, the trend toward smaller biopsies and limited tumor content makes high demands on assay performance, including starting input and limit of detection (LoD). Finally, the continuous discovery of new oncogenic rearrangements means that specific assays are not available for all fusion genes.

Can recent advances in NGS instrumentation and solutions address these difficulties?

NGS-based tests that use RNA (rather than DNA) as the input analyte are particularly useful for fusion detection. These RNASeq assays enable us to both detect the fusion and also to measure its expression level. Furthermore, unlike DNA-based tests, RNASeq assays can accommodate sequences with large intronic regions (for example, *NTRK* genes). But how do these tests perform in the real world? After all, for a gene fusion assay to be useful, it must meet specific requirements – not least the following:

- Rapid turnaround time to permit simultaneous return of both NGS and immunohistochemistry data (ideally in a fully integrated report)
- Minimal input requirement to consistently accommodate very small biopsies and low tumor cell counts; the ability to detect gene fusions from low transcript levels and retain high sensitivity and specificity without an unacceptable level of false positives
- Reliably identify all known gene fusions, as well as novel fusion isoforms

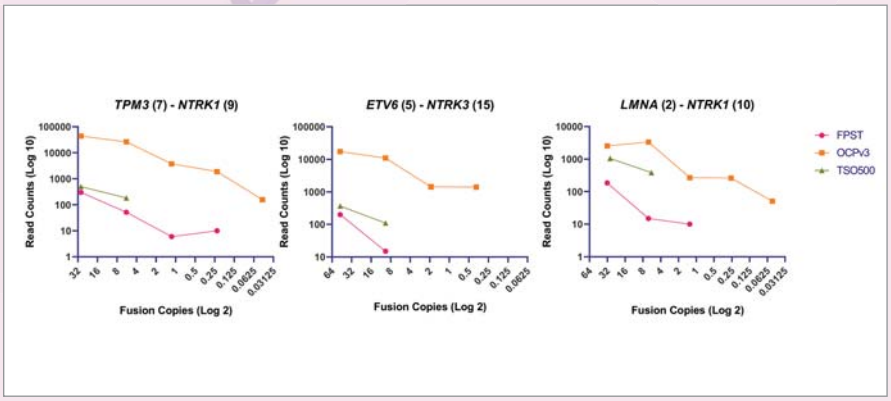


Figure 1. LoD comparison. OCPv3 enables the detection of low abundant fusion events, requiring lower fusion copies to generate higher read counts compared with FPST or TSO500.

How do existing RNASeq technologies perform against the above criteria?

The Song laboratory has compared the ability of three different NGS-based RNASeq tests – the FusionPlex™ Solid Tumor (FPST) from ArcherDx, the TruSight™ Oncology 500 (TSO500), and the Oncomine™ Comprehensive Assay v3 (OCPv3) – to detect *NTRK* fusions. First, Song's team showed that all three assays successfully detected the fusions present in the SeraSeq RNA standards for which they had compatible probes (FPST and TSO500 detected 15/15 SeraSeq FFPE *NTRK* fusion samples, and OCPv3 detected 13/13). Next, they assayed 16 clinical research specimens from glioblastoma patients, all bearing either an *NTRK1* or *NTRK3* fusions, except one with an *NTRK2* fusion. Although the TSO500 detected all fusions, FPST and OCPv3 did not detect the *NTRK2* rearrangement. Finally, again using the SeraSeq RNA standards, Song's lab compared the assays' LoDs using a dilution-based approach. Notably, LoD varied substantially between platforms; for example, to detect the *TPM3-NTRK1* fusion, the TSO500 required ~20 copies, whereas FPST and OCPv3 required only one copy; for *ETV6-NTRK3*, the FPST and TS500 required ~50 copies and the OCPv3 needed only one copy; and lastly, for *LMNA-NTRK1*, the TSO 500 needed ~20 copies, the FPST two copies, and OCPv3 only one copy. Finally, although these examples highlight that LoD is a major differentiator between the three assays, another critical parameter enhancing the differences between them is the assay input requirements. The FPST requires 25–250 ng input RNA, TSO500 requires 45–85 ng, and OCPv3 needs only 1–20 ng.

Jermann's team evaluated the recently launched Oncomine™ Precision Assay, in combination with the latest Ion Torrent™ sequencer, the Genexus™ System. This new panel, unlike the OCPv3 assessed by Song's team, uses novel FusionSync™ technology to identify both known and novel gene fusions (see sidebar: Detecting known and novel fusions with FusionSync). By testing SeraSeq RNA standards, Jermann showed that the Oncomine™ Precision

Detecting known and novel fusions with FusionSync™

FusionSync™ consists of two underlying technologies:

i. Detection of known fusion isoforms (as per previous Oncomine™ assays) by reverse transcription of sample RNA into cDNA, followed by multiplexed PCR amplification of specific fusion genes;

ii. Detection of fusions involving known driver genes (*ALK, RET, FGFR1, 2* and *3* and *NTRK1, 2* and *3*) and unknown partners, by means of the tiling imbalance assay; such fusion events would have been missed by previous versions of Oncomine assays.

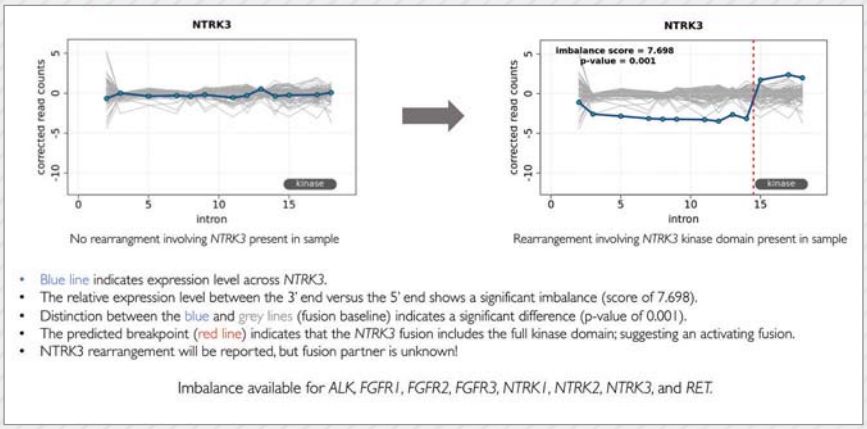


Figure 2. NTRK3 fusion analysis by FusionSync™. Exon expression levels in the test sample (blue line) are compared with baseline expression of non-rearranged DNA (grey lines; several readings are taken to account for sample-to-sample variation). A flat line (left) indicates absence of gene fusion. An expression imbalance (right) indicates rearrangement; here, the expression is below baseline until exon 15, and jumps above baseline thereafter. Red dotted line = predicted breakpoint.

The tiling assay detects expression imbalances between the 5' and 3' ends of the gene; such imbalances indicate that part of the gene has been translocated and is being controlled by a different promoter, as part of a chimeric gene product. The assay generates an imbalance score and a T value for deviation of expression from baseline. The resulting values are fed into an algorithm, which then returns an estimate of the probability of rearrangement.

Assay detected all known driver gene fusions and *NTRK* rearrangements. The subsequent analysis of clinical research samples showed 100 percent detection concordance (see Table 1) with reference methods (FISH or FPST assays) used for specimen characterization, along with demonstrating the added value of both methodological elements constituting the FusionSync™ technology – the targeted part and the tiling imbalance – with respect to their ability to identify both the driver gene and the break point of the rearrangement.

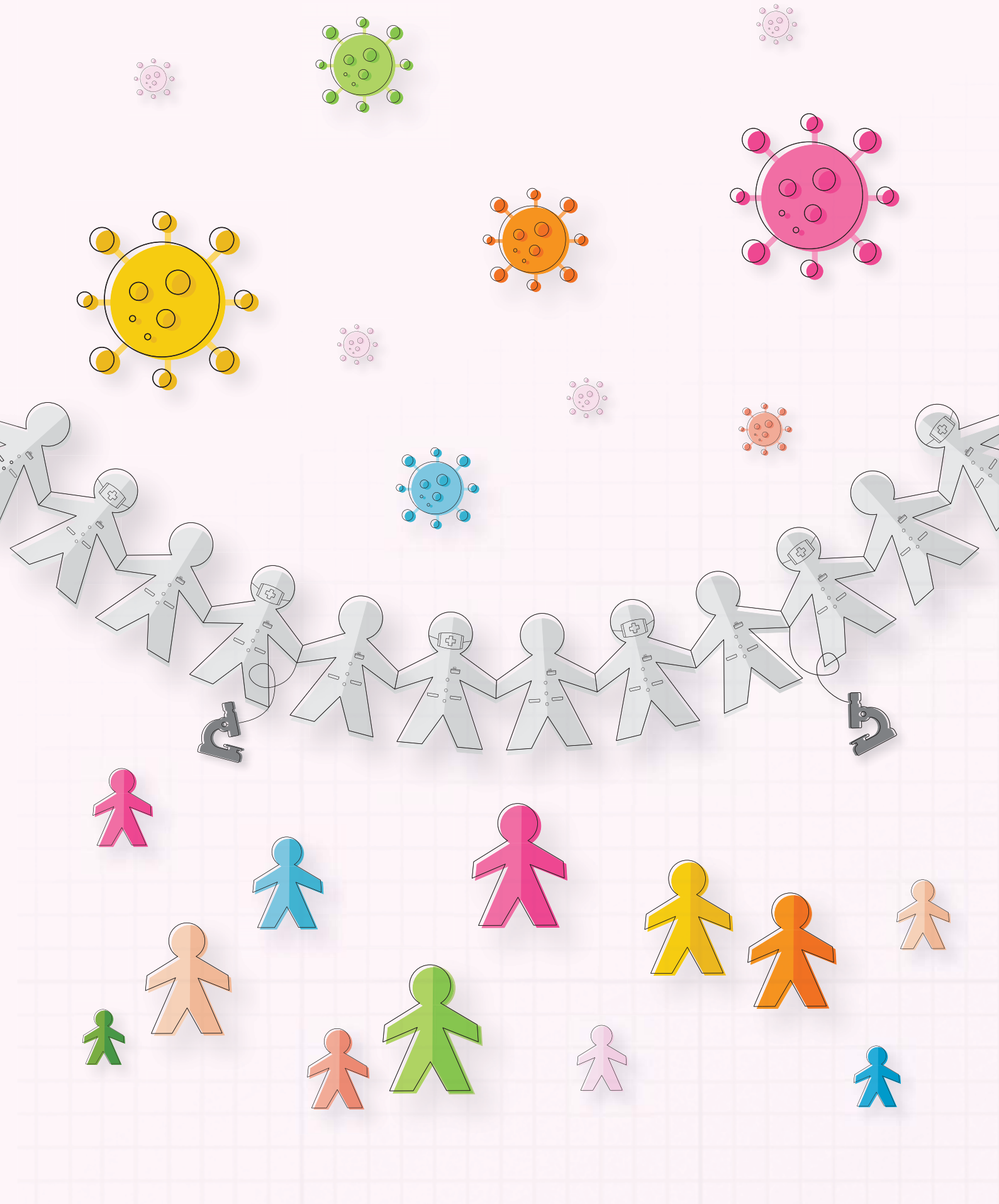
Could this be the future of fusion detection?

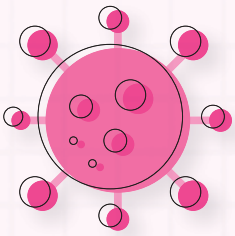
Song's evaluation shows that OCPv3 has the best-performing LoD of the three tested assays, along with lower input requirements. Nonetheless, the OCPv3 could not detect unknown novel fusions, representing a drawback for investigating tumor types in which the landscape of gene fusions has not yet been thoroughly investigated. This limitation does not apply to FPST or TS500. Conversely, Jermann's experiments show that the new Oncomine™ Precision Assay on the Genexus™ System overcomes this limitation – without compromising the typical Oncomine™ characteristics. The Oncomine™ Precision Assay requires only 10 ng of input nucleic acid, which helps laboratories achieve minimal sample rejection rates while working with increasingly small biopsies. The unprecedented level of automation in the Genexus™ System has a direct impact on time to results; the 5–10 days usually required for manual NGS systems can now be reduced to 1–2 days. Such a substantial reduction of the workflow turnaround time enables laboratories to provide both NGS and immunohistochemistry results at the same time as part of a single integrated report. Lastly, but not least, the fully automated NGS technology embodied in the Genexus™ System considerably lowers a major NGS uptake barrier by removing the need to employ highly specialized NGS technicians exclusively for NGS-related operations, extending the audience of laboratories that could embrace this technology.

Wei Song, Director of the Clinical Genomics Laboratory at Weill Cornell Medical College, works on novel methods for analyzing the mutation profiles of cancers, with a particular focus on application of NGS to define the mutation profiles of solid tumors. Phillip Jermann, Head of Molecular Assay Development at the Institute of Medical Genetics and Pathology, University Hospital, Basel, has helped establish NGS-based diagnostic laboratories throughout Europe and works on development and evaluation of novel molecular diagnostic assays.

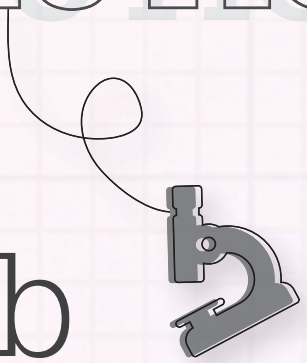
Reference

1. *Q Gao et al., "Driver fusions and their implications in the development and treatment of human cancers", Cell Rep, 23, 227 (2018). PMID: 29617662.*





THE EVOLUTION *of* the Lab



How a grassroots movement is positioning the laboratory at the forefront of healthcare

By Khosrow Shotorbani

With an increasingly global society – not to mention a growing pandemic – the idea of population health is at the forefront of many medical minds. But who is responsible for population health? Is it the epidemiologists, the sociologists, or the politicians? A new movement, termed "Clinical Lab 2.0," suggests that the laboratory is an integral part of population heath – and that laboratory medicine professionals can be leaders in the move from volume- to value-based healthcare. But what is Clinical Lab 2.0, and how does it position the laboratory at the apex of population health?

THE MEANING OF CLINICAL LAB 2.0

An initiative of the Project Santa Fe Foundation, the Clinical Lab 2.0 movement is a grassroots effort to transform the role of the diagnostic laboratory to better support the objectives of population health and value-based healthcare. The effort, launched in 2016, is designed to promote more effective utilization of laboratory data in pursuit of the lab's enormous potential for improving patient and population outcomes, reducing the total cost of care, and strengthening the patient and clinician experience.

The movement was born from a realization among a select group of laboratory leaders that our industry had reached a major inflection point. In other words, the past was no longer reflective of the future. We understood that the diagnostic lab's value proposition needed to evolve dramatically to align with, and support, healthcare's transition from volume to value. At the same time, it was clear that longstanding business models and conventional industry wisdom had not provided much room for innovation. Finally, as the commoditization of clinical testing has accelerated, it has become evident that hospital-based laboratories are at increasing risk of being sold or replaced by outsourced laboratory providers. And that's why developing ways to add value to the lab have become critical.

In the simplest terms, Clinical Lab 2.0's mission is to position the lab as the center of value-based care by promoting new strategies, models, and ideas to empower laboratory leaders – pathologists and management alike – to harness the data we collect in pursuit of population-level initiatives. These efforts can lead to substantial improvements in both outcomes and the cost of care. Underpinning this mission is a recognition that, although in vitro diagnostics account for just two cents of every dollar spent on US healthcare, lab results serve as the basis for over two-thirds of all medical decisions. Given the ubiquity of clinical testing, we believe the laboratory can positively impact virtually all aspects of healthcare and 100 percent of spending.

It has been four years since those laboratory leaders first met in Santa Fe (hence the name of the organization), and our message continues to gain traction both in the US and globally. We've created a nonprofit organization, launched four multi-institutional demonstration projects, hosted three additional closed-door colloquia, and produced three public workshops (all of which have been sold-out events) – and there is more to come. Our meetings continue to be critical to our movement by providing forums for a range of stakeholders to discuss the opportunities presented by the Clinical Lab 2.0 concept.

EXTENDING THE LABORATORY

Clinical Lab 2.0 represents an extension of the laboratory's existing transactional model (Clinical Lab 1.0) to incorporate and reflect quantitative value around the total cost of delivery and cost avoidance. Whereas 1.0 is reactive and focused on "sick care" and de-escalation, 2.0 concentrates on early detection, early escalation, intervention, and prevention (see Tables 1 and 2).

In 2017, we authored an article that we hoped would change the conversation about the potential of the clinical lab (1). We asserted that, in traditional business and care models, the clinical lab has been viewed primarily as an ancillary and increasingly commoditized departmental function. In the 2.0 model, the lab's aggregated data provides vital longitudinal touchpoints to support the full spectrum of integrated health care. Because the lab generates data regardless of where, when, or how the patient receives care, we can serve as a repository of actionable information across the entire care continuum.

Clinical Lab 2.0 can support pre-diagnostic identification and closure of care gaps, as well as deliver post-diagnostic computations of aggregated longitudinal data to enable a range of insights and actions. These include clinical prevention, programmatic clinical interventions, and optimization of diagnostic and therapeutic management. Our goals? Improved patient and population outcomes and management of population risk.

In effect, Clinical Lab 2.0 views lab personnel as "first responders." They're the first to see these critical important data and the best-equipped to understand the implications. As such, they're optimally positioned to manage population health in value-based care.

MY CLINICAL LAB 2.0 STORY

Clinical Lab 2.0 has no borders – it's truly a global movement. I've been excited to see the level of interest and engagement our efforts have elicited in diverse healthcare settings around

"ALTHOUGH IN VITRO DIAGNOSTICS ACCOUNT FOR JUST TWO CENTS OF EVERY DOLLAR SPENT ON US HEALTHCARE, LAB RESULTS SERVE AS THE BASIS FOR OVER TWO-THIRDS OF ALL MEDICAL DECISIONS."

the world. I've heard about the concerns and challenges faced by healthcare systems globally – and what I've learned is that, regardless of the setting, the fundamental principles of Lab 2.0 are universal in their application. Labs can play a critical role by providing population risk stratification relative to the known prevalence of chronic conditions, identifying care gaps and predicting clinical risk, identifying high-risk patients before they are admitted into emergency room or hospital, and facilitating early intervention between care providers and patients. These capabilities and their implications resonate globally.

The Lab 2.0 integrative model cannot exist without a solid Lab 1.0 foundation. The models are iterative and interconnected. In envisioning the lab as the first responder, we're saying that the lab is the first to become aware of a clinical need and therefore in the best position to provide leadership in addressing that need. Reducing the time to diagnosis can help with diagnostic optimization and appropriate laboratory test utilization, which, in turn, leads to care optimization, therapeutic optimization, and appropriate screening and surveillance.

If we don't get the first step – identifying actionable clinical information at the point at which it is generated – right, the entire continuum of care becomes suboptimal, and that can cause significant patient harm. The lab can be the catalyst for improving population health outcomes, reducing the overall cost of care and, importantly, empowering health systems to successfully manage the financial risk of providing value-based care. The central advantage we possess is the ability to produce scientifically measured, structured data at each touchpoint on the care continuum. That means the information we generate is clinically actionable with zero latency.

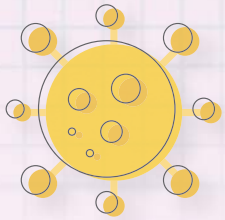
FROM OBSTACLES TO OPPORTUNITIES

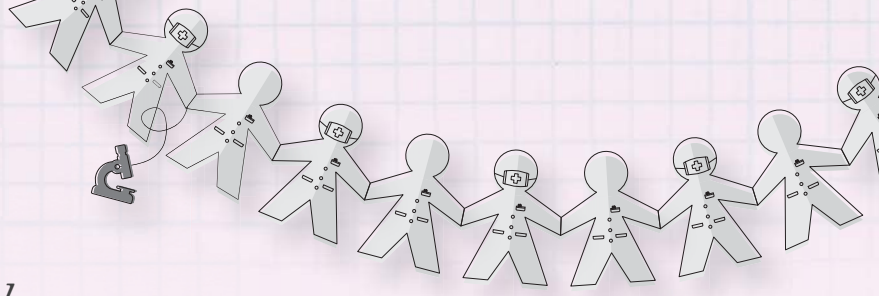
We've identified a number of barriers or obstacles that can impact the transition to Lab 2.0. These include:

- Lack of a common language among providers, data analysts, health systems, and payers with respect to certain clinical conditions and lab results
- Lack of models for comparison and benchmarking
- The inability of existing laboratory information systems to integrate data or provide information for clinical decision support; current systems tend to support only revenue cycle and contract pricing data
- Lack of outcomes-based evidence for laboratory-led innovation
- Difficulty integrating laboratory insights into the existing clinician workflow
- Lack of aligned incentives
- Inadequate leveraging of laboratory data into actionable information, including the absence of detailed data-sharing agreements
- Lack of access to capital for in-system laboratories versus the for-profit sector of laboratory industry
- Lack of access to new and necessary skill sets
- Limited understanding of the laboratory's potential among health system leaders and inadequate engagement of same
- No playbook for providing Lab 2.0 leadership

The Lab 2.0 initiative helps the industry overcome these barriers by emphasizing three fundamental pillars of transformation:

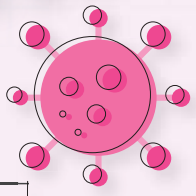
- *Leadership:* Helping clinical lab leaders embrace a new leadership mindset that extends beyond the four walls of the laboratory.
- *Standards:* Measuring what matters – that is, the development of new measurements and benchmarks that support a new clinical value proposition.
- *Evidence:* Developing multi-institutional demonstrations to show how laboratory medicine and pathology affect population health and align with the drivers of value-based care. Our projects focus on providing outcomes-based evidence and producing roadmaps that all labs can follow.

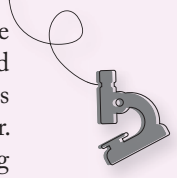




Labs, Population Health and COVID-19

By Khosrow Shotorbani





The question you may be asking is: as guardians of public health, what is the lab's role in the COVID-19 pandemic? Obviously, our ability to serve as leaders goes beyond our duty to provide timely, accurate testing.

The four points we need to highlight – and illustrate by our actions – are:

- The laboratory is the first to know with real-time results.
- Laboratories are the first responders providing recommendations and developing new strategies.
- Laboratories are the "epicenter of informatics." with insights around disease patterns and predicting outbreaks.
- Laboratories should serve as the "command center" managing this pandemic by developing guidance as to who should be tested and when.

In the COVID-19 pandemic response, the lab takes center stage. I have been humbled as I've witnessed my colleagues across the country rise to the challenge. In my opinion, it's impossible to overstate the impact of the laboratory at this time.

I've been asked, "What is the role of Clinical Lab 2.0 in managing this pandemic?"

The Clinical Lab 2.0 model is based on three key actionable pillars:

- Leadership outside the clinical laboratory
- Clinical Lab 2.0 new standards: measuring what matters to provide actionable data that can lead to objective key results
- The science of laboratory medicine: focusing on not just the analytical components of lab medicine, but also the pre- and post-analytical stages

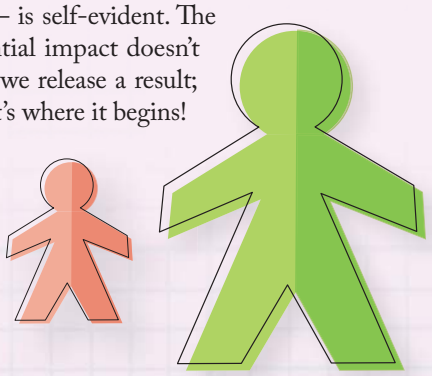
The Clinical Lab 2.0 model argues that laboratory medicine professionals must assume a leadership role outside the lab and engage their health system's stakeholders and public health agencies. Obviously, we have to set up testing to keep up with demand – a key task that, at this point, remains challenging. Clinical Lab 2.0 can then potentially mine longitudinal data (laboratory results, patient demographics, and any pre-existing or past conditions) to proactively determine which patients are potentially at risk of comorbidities. Labs can help their health systems risk-stratify their populations based on historical conditions, such as respiratory syndromes or infections, chronic diseases like diabetes mellitus, or cancer leading to immunosuppression.

It's important to remember that a negative COVID-19 test result doesn't entirely eliminate a patient's risk. Not only are false-negative results possible, but any patient who has not yet been infected remains vulnerable. Labs can identify a high-risk patient pool, then partner with providers and state agencies to develop targeted isolation strategies for prevention and intervention focused on outcome.

COVID-19 has undoubtedly raised the critical, urgent, and quantitatively relevant value of the clinical lab and its clinical assets globally. The lab is the centerpiece of healthcare delivery and provides a method to triage care, as opposed to being an ancillary cost center. The clinical lab is the catalyst managing population health, helping to flatten the curve of not only COVID-19, but also chronic conditions.

We also cannot forget the role of the lab in returning infected patients – and, indeed, the population as a whole – to normal life. Who is infectious? Who is immune? Who can go back to work and who must remain in lockdown? This is an especially vital function as it relates to healthcare workers and first responders on the front lines. Furthermore, the data we gather – and the tests we conduct – are critical to further evaluating the effectiveness of treatments and vaccines, and to detecting (and ideally preventing) future waves if COVID-19 becomes a seasonal affliction. It's our job to provide global surveillance so that not just individual patients, but the entire population, can be protected.

In this pandemic, global healthcare faces the ultimate challenge. Now, more than ever, the tangible value of the clinical laboratory – and the unsung heroes who keep it running every day – is self-evident. The lab's potential impact doesn't end when we release a result; rather, that's where it begins!



Clinical Lab 1.0 Outcomes-based		Clinical Lab 2.0 Value-based
Sick Care ○ Receive and result tests ○ Passive engagement	→	**Health Optimization** ○ Proactive engagement ○ Precision medicine
Disease Screening ○ Scheduled by treating physician ○ Disease surveillance	→	**Risk Management** ○ Identification & real-time tracking of risk ○ Driving care intervention ○ Reducing negative outcomes
Wellness Program ○ Result-driven ○ Blanket approach to testing	→	**Care Coordination** ○ Diagnostic management teams ○ Closure of care gaps using data ○ Optimizing lab testing in clinical workflow ○ Elimination of care variation
Consensus-based ○ Protocol/guideline-driven	→	**Evidence-based** ○ Predictive analytics ○ Data-driven decisions
Payment Models ○ Cost per test ○ Fee-for-service ○ Laboratory a commodity	→	**Payment Models** ○ Value-based (per member/per month) ○ Impact to total cost of care

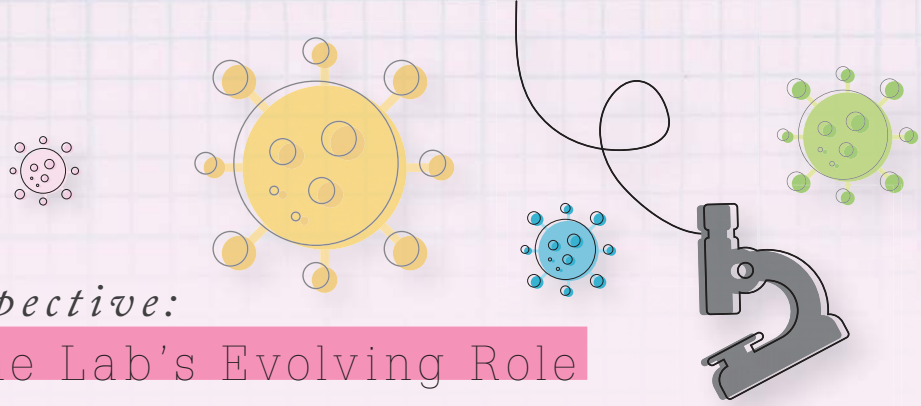
Table 1. Contrasting Clinical Lab 1.0 with Clinical Lab 2.0

Our Project Santa Fe colleagues and participants have encountered many of the opportunities generated by a more engaged and integrated lab. Here are just a few that have been presented in the recent literature:

Diabetic patients (with comorbidity): In most countries, attempts to de-escalate the impact of diabetes don't occur until morbidity is severely advanced, typically when the patient's A1C level is over 9 and the kidney function (EGFR) is below 60. Generally, this means irreversible stage 3 kidney failure. However, if at-risk patients are identified early – when their A1C is 5–7 and EGFR is between 90 and 60 – we can manage their care to improve outcomes and reduce downstream cost. The ability to identify at-risk patients in the pre-diabetic stage can help avoid the progression of the disease which, if uncontrolled, can cost an average of US$10,970 per case (2).

Urinary tract infections: Laboratory-provided insights into urinary tract infections managed in the emergency room (ER) not only diagnose the acute condition, but also offer clues to improving treatment and identifying patients with recurrent infections. These insights "could result in more appropriate drug treatment, improved resource allocation, and decreased ER costs

"OUR PROJECTS FOCUS ON PROVIDING OUTCOMES-BASED EVIDENCE AND PRODUCING ROADMAPS."



A Clinician's Perspective: COVID-19 and the Lab's Evolving Role

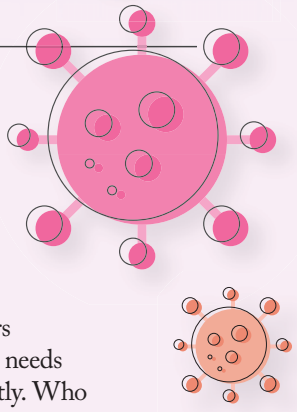
By Jeremy Orr

During the early part of the pandemic, laboratorians mobilized to provide timely, accurate testing for individual patients. In places where testing was limited, lab personnel sometimes enforced prioritization criteria. Fortunately, in many (though not all) parts of the world, tests are now available in greater quantities and rationing is no longer an issue. So it's natural to ask: what lies ahead for the lab as the next stages of the pandemic unfold?

First, let's acknowledge that none of us know for certain how the next few months will go. Different regions are at different points on their case growth curves – and the shapes of those curves are dependent on the circumstances. Will there be a rebound in places where an apparent peak has been reached? Will we see COVID-19 take on seasonal characteristics like influenza? Will containment efforts evolve or corrode? What role will herd immunity play? When will there be a viable vaccine? We just don't know the answer to these questions.

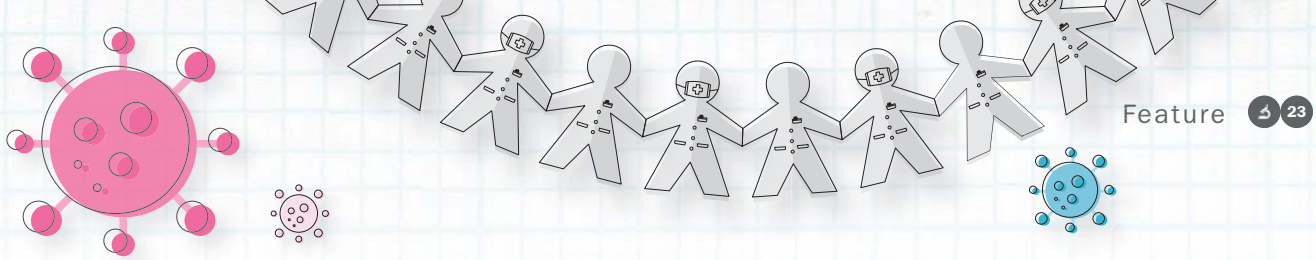
Despite the uncertainty, the laboratory will continue to play a central role – but the nature of that role will evolve. Here are some of the potential future use cases – and how the lab may fit in:

- Patient triage and population health efforts. For patients who have already tested positive, laboratorians are in a strong position to provide risk stratification to guide disposition and follow-up protocols.
- Contact tracing – a best practice in epidemiology, but also a resource-intensive one. Not every COVID-19 positive patient will have complete contact tracing. Because clinicians operate 1:1, they often can't see the connections between events. Labs can see all the data and map the temporal and geospatial relationships between events. There is a long history of labs reporting this data to public health agencies for surveillance purposes, but they can do more. Even tracing within a health system or locality can help prioritize contact tracing and mitigate disease spread.
- Antibody testing. It's fraught with challenges, but has been used for other infectious diseases and will be used for COVID-19 to assay immunity at both the individual and population level. Given the interpretation pitfalls, lab expertise will be needed to guide policy and implementation.
- Vaccine prioritization. An effective COVID-19 vaccine will be critical to long-term containment. But, in the early days of any vaccine, access is often limited and we will have to decide who goes first. Essentially, it's a risk/benefit ratio, and labs – the center of the care data flow – are in an excellent position to help.
- Return to routine. We all know that routine care, including cancer screening and chronic disease management, is being delayed and deprioritized for this phase of the pandemic. How do we get back to par? As routine care efforts rebound, the lab can again play a central role in helping providers understand who needs care most urgently. Who should be at the front of the cue for a colonoscopy or a diabetes check-up? Labs provide critical clues.



Central to all of these use cases is the careful and informed interpretation of data familiar to the lab. Of course, lab personnel cannot alone be responsible for surveying all data available to them for the purposes of powering these use cases – but they don't need to. Newer technologies, including machine learning, are maturing just in time to help. Newer algorithms (full disclosure: including some developed by my company) can systematically analyze structured data and lab results to flag the patients at highest risk for COVID-19 complications, cancers, chronic disease complications, and more. The natural place for running these complications is in the lab – why? Because labs have both the data and the natural expertise to translate insights back to providers.

Jeremy Orr is a practicing, board-certified family physician and Chief Executive Officer and Chief Medical Officer of Medial EarlySign, Aurora, Colorado, USA.



Clinical Lab 1.0 – Transactional	Clinical Lab 2.0 – Integrative
• Did the (new) laboratory testing cause harm? • Was the laboratory testing able to distinguish between patients who had disease versus those who did not? • Was the innovation in laboratory testing able to provide better patient diagnostic information than previous options for such testing? • Did patients benefit from such testing having been done?	• Do innovations, introduced by laboratory leadership, cause harm to patients or populations? • Are populations of individuals subjected to such innovations measurably different from populations not subject to innovation? • Are the differences in a favorable direction? • Did patients (or populations) benefit from such innovation having been introduced

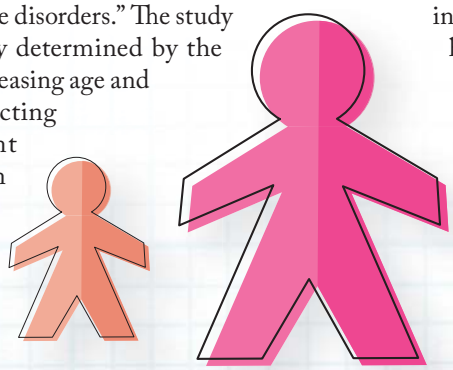
Table 2. Testing the utility of lab services and the value of Project Santa Fe recommendations.

for integrated health systems (3)."

Cost-effective drug therapy regimens for chronic disease: Laboratories are beginning to understand the value of having pharmacists on their staff. These experts can help with antibiotic stewardship and identify appropriate treatments for chronic diseases – especially those that, like rheumatoid arthritis and chronic reoccurring infections, require long-term, high-cost therapy. "To manage these diseases, the cost of drug treatment, monitoring of drug therapy regimens, and treatment adjustments for empiric therapy require post-analytic interpretation of laboratory results, along with drug therapeutics knowledge (3)."

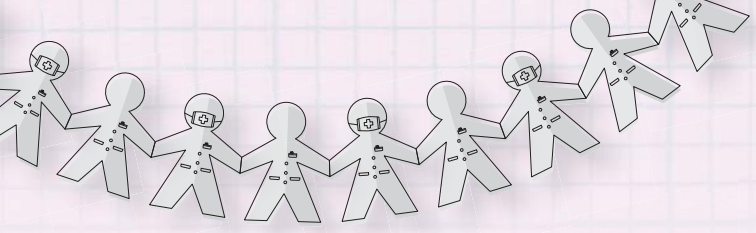
Pregnancy: For women who don't receive routine care – for instance, those on low incomes or without insurance – laboratories can identify pregnancies early, avoid treatment options that could present a pregnancy risk, and monitor prenatal testing patterns and results to identify high-risk pregnancies and women in need of more intensive prenatal care (3).

Opioids and benzodiazepines: We need innovative approaches to tackle the ongoing opioid crisis. "As stewards of health analytic data, laboratories are uniquely poised to approach the opioid crisis differently," says one study (4). The pilot study aimed to "bridge laboratory data with social determinants of health data, which are known to influence morbidity and mortality of patients with substance use disorders." The study found that co-use is largely determined by the patient's providers, with increasing age and geographic area also predicting co-use. "The prominent geographic distribution of co-use suggests that targeted educational initiatives may benefit the communities in which co-use is prevalent. This study exemplifies the Clinical Lab 2.0 approach by leveraging laboratory data to gain insights into the overall health of the patient."



"OUR MOVEMENT CANNOT ACHIEVE ITS OBJECTIVES ALONE – SO KEY PARTNERSHIPS ARE A CRITICAL COMPONENT OF OUR FUTURE ACTIVITIES."

The future of Clinical Lab 2.0 is somewhat academic, but with a sense of agility and urgency. Our vision is to share knowledge through publications and key partnerships, to continue to build the evidence base with expanded multi-institutional demonstration projects, and to continue to host annual scientific colloquia and produce educational workshops. Project Santa Fe Foundation is a member-driven organization. Obviously, our movement cannot achieve its objectives alone – so key partnerships are a critical component of our future activities. These relationships will help us broaden our reach, engage industry partners in the in vitro diagnostics and informatics space, and potentially help influence policies that



Protecting the Population

How "behind the scenes" workers are safeguarding the world's health during the COVID-19 pandemic

Michael Schubert interviews Keren Landsman

How well is the pandemic being handled globally?

That's a very tricky question because each country has a different approach. We don't have a global health department to coordinate efforts worldwide. The closest authority we have is the World Health Organization, but their function is to make recommendations, rather than to take action.

I think the biggest thing we need to learn from this pandemic is how to cooperate internationally on health. We need to understand that viruses do not recognize borders. If we want to stop the next outbreak before it becomes a pandemic, we all need to work together – regardless of a country's size, population, politics…

How are "behind the scenes" healthcare professionals contributing?

There's a lot of work being done that isn't very visible. We all know about the doctors and nurses on the front lines. We see pictures of them in the media every day, wearing masks (if they're lucky) and treating patients. But there are a lot of people you don't see: pathologists, laboratory staff, radiologists, technicians, epidemiologists, cleaners, and more. All of us are working together to mitigate the impact of COVID-19.

How do you think the move from volume- to value-based care will affect COVID-19 testing and management?

In Israel, we are under massive public pressure to increase the volume of testing. The public want us to test as many people as we can, as often as we can, regardless of how sensitive or specific the tests are. As you can imagine, I have a little bit of a problem with that!

I think the general public views testing as very black-and-white. Many common medical tests are presented to patients as either positive or negative; you either have the condition or you don't. Of course, expert diagnosticians know that isn't the case – and COVID-19 is no exception. Someone who tests negative for SARS-CoV-2 today might test positive tomorrow. The result might be a false positive or negative; many currently available tests have low sensitivity or specificity. We don't need more volume; we need more value. We need better tests.

That's not to say we shouldn't be testing more people – it's just that I don't think we should be taking that step right now. First, we need accurate tests – for both disease diagnosis and antibody screening – and then we should roll those tests out to the population. Only then can we begin lifting the precautionary restrictions and returning to our normal lives.

In your opinion, what does the near future look like?

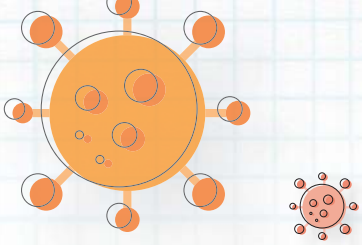
Nobody knows what even the near future holds. We first heard about SARS-CoV-2 in December. The virus didn't even have a name until February. We've only been acquainted with it for four months. There are a lot of theories and guesses at the moment, but there's no such thing as an accurate projection right now. I would caution everyone to be wary of people who claim to know what's going to happen, because no such thing is possible.

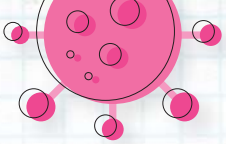
What I can tell you is that, at some point, the immediate threat of the pandemic will die out, the restrictions will be lifted (although, again, each country has a different approach to achieving that), and we will go back to living our lives. But SARS-CoV-2 won't disappear. We will have to learn to live with the virus. There will be a "new normal," although it's impossible to say how that might look.

We will also need to truly grasp the fact that there will be another pandemic – we just don't know when. The only way to mitigate the impact of a future pandemic is to coordinate health efforts around the world as soon as a new disease emerges. If we take one positive from the COVID-19 pandemic, I hope it's a truly global health system.

See an extended version of this interview online at at tp.txp.to/clinlab2.

Keren Landsman is a public health specialist. She is a member of Mida'at, a public health non-governmental organization, and works as an epidemiologist in the Israeli Ministry of Health and as a physician in the Levinsky Clinic, Tel Aviv, Israel.





"WE DON'T HAVE TO BOIL THE OCEAN TO ADD NEW VALUE – JUST GETTING OUT OF THE LAB AND TELLING A DIFFERENT STORY IS MISSION-CRITICAL."

determine the direction of healthcare. Ultimately, our goal is to create a tipping point that elevates the value of the clinical lab, domestically and globally, as healthcare transitions from volume to value and from sick-care to well-care.

HOW TO IMPLEMENT CLINICAL LAB 2.0

Become stewards of data. Create patient-centric longitudinal data sets that make clinical sense. This will support population risk stratification, identification of care gaps, and early identification of high-risk patients. Laboratories enable actionable signals to manage the risks of unfavorable outcomes and inordinate financial resource use. This fundamental step, combined with domain knowledge of pathology, will begin the conversation with key stakeholders outside of the lab.

Get outside the four walls of the lab. Lab leaders must be able to engage the C-suite in discussions about how the lab can impact enterprise-level initiatives and play a pivotal role in population health, value-based care, and mitigating financial risk. We must be able to use C-suite language and address organizational imperatives.

Take a seat at the table. Actively engage in helping design future healthcare delivery models that use the predictive value of the clinical lab data for clinical intervention, prevention, and cost avoidance. The lab must be at the table – perhaps even at the head of the table – to achieve better, more cost-effective care.

Demonstrate value. We need to prove to the C-suite that the lab's value extends beyond simply costs per unit. Look for short-term wins. We must align ourselves with key enterprise objectives by demonstrating the value we can deliver in areas that hospital leaders care about: outcomes, total cost, clinical risk, financial risk, affordability, and increased access. If we fail, we become a target of outsourcing.

We need to think big but act small. We don't have to boil the ocean to add new value – just getting out of the lab and telling a different story is mission-critical. To start, you can mobilize clinical data in a way that makes sense for your local healthcare needs. For example, you could add basic delta checks on some critical assays, start reporting that change to your clinical colleagues, and seek their input.

But we can't do this in a vacuum. Lab 2.0 requires strategic and operational planning that can demonstrate the tangible value of the clinical lab for customers we may never have served before. Don't encumber the Lab 2.0 way of valuation with old ways of doing business. Enterprise health organizations must find ways to improve clinical outcomes, reduce financial risks, and improve patient satisfaction. Otherwise, we'll be at the mercy of inadequate reimbursement models.

A quote from James Crawford, our Project Santa Fe Foundation Chairman of the Board, sums it up best: "There has never been a better time to demonstrate the value of laboratory medicine and pathology in the delivery of healthcare – but it must be quantitatively proven and attributable to the lab's contribution."

Khosrow Shotorbani is President and Executive Director of Project Santa Fe Foundation, CEO and founder of Lab 2.0 Strategic Services, and a Clinical Laboratory 2.0 industry advocate.

We would also like to acknowledge the Project Santa Fe Foundation Board of Directors' institutions: Geisinger Health System, Henry Ford Health System, Intermountain Healthcare Central Laboratory, Kaiser Permanente Northern California, Mayo Clinic Laboratories, Northshore University HealthSystem, Northwell Health, TriCore Reference Laboratories, and The Robert Larner, M.D. College of Medicine, University of Vermont.

References

1. *JM Crawford et al., "Improving American healthcare through 'Clinical Lab 2.0': a Project Santa Fe report", Acad Pathol, 4, 2374289517701067 (2017). PMID: 28725789.*
2. *TM Dall et al., "The economic burden of elevated blood glucose levels in 2012: diagnosed and undiagnosed diabetes, gestational diabetes mellitus, and prediabetes", Diabetes Care, 37, 3172 (2014). PMID: 25414388.*
3. *K Swanson et al., "Improving the delivery of healthcare through clinical diagnostic insights: a valuation of laboratory medicine through 'Clinical Lab 2.0'", J Appl Lab Med, 3, 487 (2018).*
4. *JS Warrington et al., "Integrating social determinants of health and laboratory data: a pilot study to evaluate co-use of opioids and benzodiazepines", Acad Pathol, 6, 2374289519884877 (2019). PMID: 31700992.*

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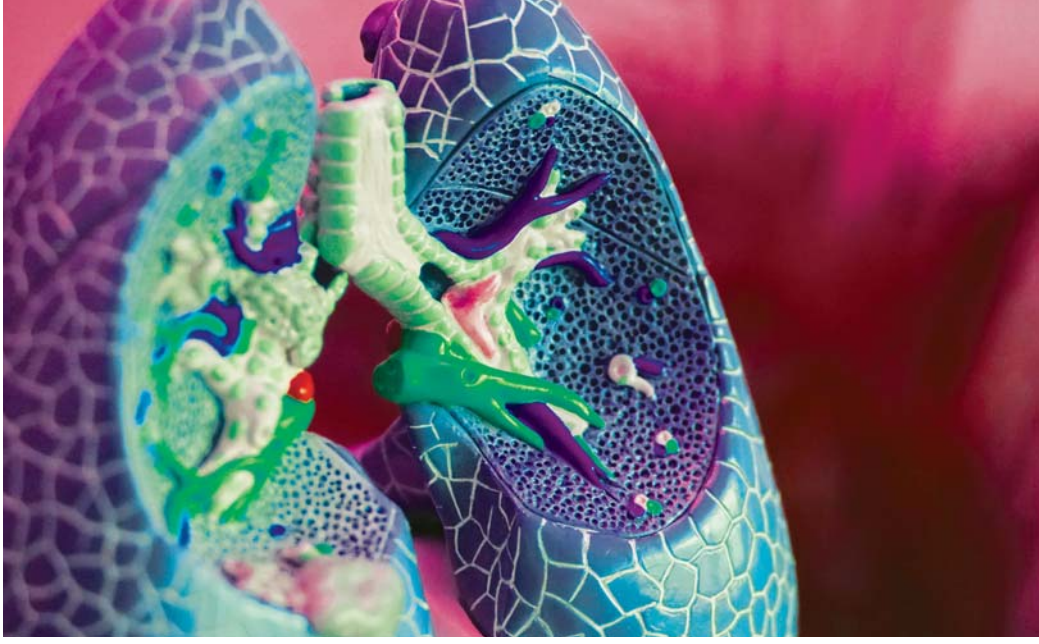
Director of Pathology,
Memorial Sloan
Kettering Cancer Center

ANANT MADABHUSHI
F. Alex Nason Professor II of
Biomedical Engineering, Case
Western Reserve University

In Practice

Technologies and techniques
Quality and compliance
Workflow

Vaping, the use of e-cigarettes, is becoming increasingly popular. Many choose it over traditional smoking because they believe it is safer – but research reveals that vaping can cause acute lung injuries not seen in smoking. More data is needed to determine its long-term effects, but the practice (especially if it involves illicit tetrahydrocannabinol) carries clear risks even in the short term.



Smoke and Mirrors

A new study reveals the dangers of smoking's "safer" cousin: vaping

Michael Schubert interviews Sanjay Mukhopadhyay

A new craze has been sweeping the smoking world: "vaping," or the use of electronic products designed to act like cigarettes. These e-cigarettes are popularly believed to be safer than traditional tobacco products and can be used to deliver not only nicotine, but also flavored products, marijuana, and other drugs. Because they are relatively new to the market, not much is known about the health risks of using e-cigarettes – but most contain nicotine and all contain other substances that are potentially harmful when aerosolized.

Recent research has raised the concerning issue of e-cigarette or vaping product use-associated lung injury (EVALI) – damage caused to the lungs by vaping. At the moment, little is known about this phenomenon or its development, so we spoke to Sanjay Mukhopadhyay – Director of Pulmonary Pathology at the Cleveland Clinic and lead author of a recent study into the pathology of the disease – to find out more.

What inspired you to investigate the potential dangers of vaping?

In September 2019, as the first large reports on the outbreak of EVALI were coming out in the medical literature, we began to receive lung biopsies from patients who had fallen ill after vaping. We quickly realized that there was no systematic study in the literature on the pathology of this condition. Also, the few descriptions out there were based on bronchoalveolar lavage (BAL) cytology, not lung biopsies, and were leading to an incorrect label ("lipoid pneumonia") for this entity. And that's why we decided to describe the pathology of this disease in lung biopsies.

When we did, we found acute lung injury patterns known as "organizing pneumonia" and "diffuse alveolar damage" (see Figure 1). Lung pathology experts see these patterns frequently in daily practice, because they are common ways in which the lung reacts to injury, regardless of the exact cause.

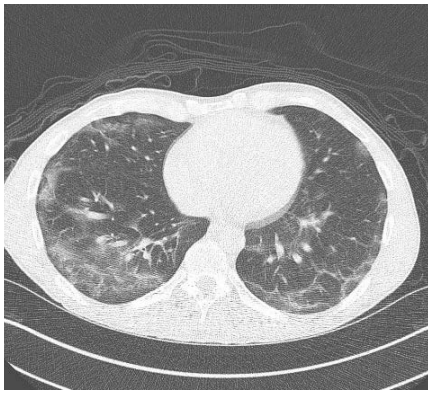


Figure 1. A CT scan showing the lungs of an individual who had vaped THC.

How does vaping damage the lungs?

Most cases of EVALI are caused by vaping illicit tetrahydrocannabinol (THC)-containing oils. The CDC has reported that vitamin E acetate, which is used to "cut" THC-containing oils and mislead customers, is the prime suspect (1). It has been found in counterfeit THC cartridges used by patients with EVALI, as well as in BAL fluid from the lungs of these patients. This chemical probably injures the lung when inhaled (see Figure 2), and it may be especially toxic when heated to high temperatures.

The appearance of these injuries under the microscope is very different to those caused by traditional smoking (see Figure 3). Smoking causes accumulation of a fine, brown pigment within macrophages in the lung, whereas vaping does not. Smoking also causes chronic lung damage in the form of emphysema and fibrosis ("smoking-related interstitial fibrosis") or Langerhans cell histiocytosis, none of which are caused by vaping. In emphysema, the substance of the lung is gradually destroyed over several years and loses elastic recoil, like an inflated balloon that has turned into an empty brown paper bag. In smoking-related interstitial fibrosis, the walls of the lung sacs are thickened by collagen. Pulmonary Langerhans cell histiocytosis results in the formation of numerous tiny collections of abnormal cells within the lungs and can cause scarring and cyst formation in the long term. In contrast, the lung injuries seen in vaping resemble injuries that result from inhaling toxic chemicals like bleach or mustard gas, or from taking drugs like amiodarone or bleomycin.

Overall, the changes caused by vaping develop rapidly (and we consider them acute), whereas those caused by smoking generally develop over several years (and are considered chronic). Unfortunately, we don't yet have any good data on the lung pathology of long-term vaping. This will

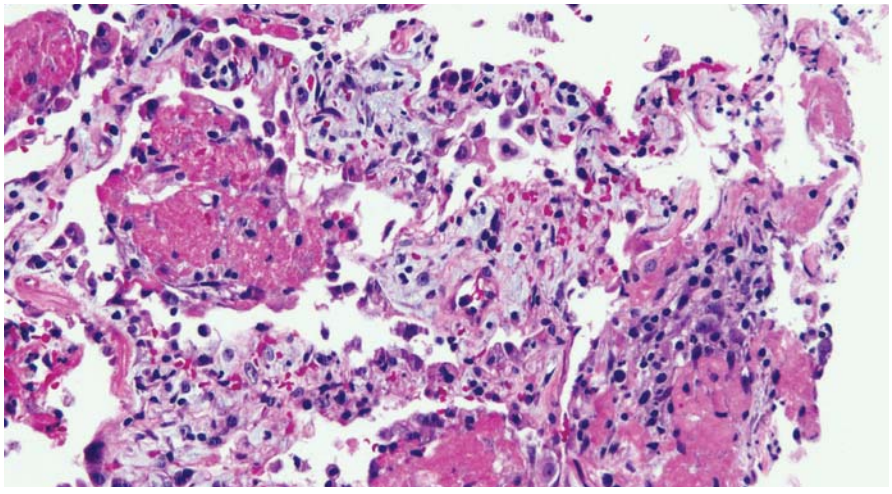


Figure 2. Histology of acute lung injury caused by vaping THC (20X).

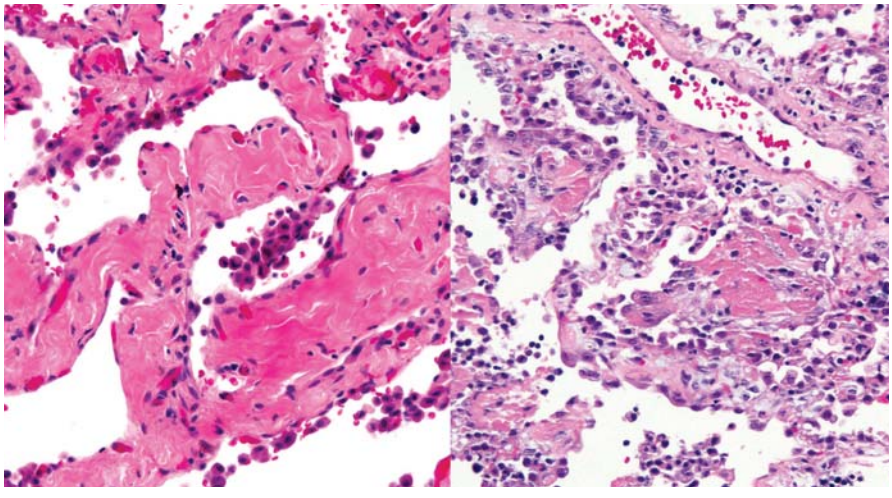


Figure 3. A comparison of smoking-related interstitial fibrosis (left) with acute lung injury in EVALI (right).

require examination of lung biopsies or other lung specimens from patients who have vaped for several years. Nevertheless, I would strongly advise anyone who is a non-smoker to stay away from vaping.

Is vaping actually safer than smoking?

It's hard to say at this point because, other than nicotine addiction, the health effects of vaping store-bought, nicotine-containing e-cigarettes are not well understood. In contrast, we know that smoking is extremely dangerous – probably the worst thing you can do to your lungs. However, vaping illicit THC is very dangerous, can lead to EVALI, and should be strongly discouraged.

We know that children, teenagers, young adults, pregnant women, and individuals who do not currently use tobacco products should never vape. Whether smokers should vape to quit cigarettes is a more difficult question – more research is needed to determine if vaping is "safer" than smoking.

What should other pathologists know about this new type of respiratory injury?

The oil red O stain is not required for the diagnosis of EVALI, and a positive oil red O does not prove "lipoid pneumonia." Lung biopsies have not confirmed a single case of exogenous lipoid pneumonia in EVALI. We are currently conducting a study involving the oil red O stain, with a focus on specificity. From all accounts, pathologists are extremely skeptical of the utility of this stain for the diagnosis of EVALI.

The biggest limitation of pathology is that it does not involve testing for chemicals. Biopsies are helpful only in that they show the type and severity of damage the chemical is causing in the lung. Chemical testing is the way forward; some of this work has already been done by the CDC (2). Specifically, the CDC conducted chemical testing by isotope dilution mass spectometry on BAL fluid samples from the lungs of 29 patients with EVALI in 10 different states. All 29 of the samples tested contained vitamin E acetate; THC was found in 23 of 28 samples tested; and nicotine was found in 16 of 26 samples tested. None of the samples contained plant oil, mineral oil, medium-chain triglyceride oils, or terpenes. No other potential toxins were found (3).

Vitamin E acetate was already a prime suspect in the causation of EVALI based on testing of product samples (vape cartridges) used by EVALI patients. Finding it in biologic samples such as BAL fluid has added another piece of evidence to build the case that this chemical might be causing lung damage in EVALI.

Sanjay Mukhopadhyay is Director of Pulmonary Pathology at the Cleveland Clinic and Associate Editor (Pulmonary) for the American Journal of Clinical Pathology, Cleveland, Ohio, USA.

References

1. *Centers for Disease Control and Prevention, "Outbreak of lung injury associated with the use of e-cigarette, or vaping, products" (2019). Available at: https://bit.ly/2DCaq34.*
2. *Centers for Disease Control and Prevention, "For Healthcare Providers" (2019). Available at: https://bit.ly/2YfM9JB.*
3. *BC Blount et al., "Evaluation of bronchoalveolar lavage fluid from patients in an outbreak of e-cigarette, or vaping, product use-associated lung injury – 10 states, August-October 2019", MMWR Morb Mortal Wkly Rep, 68, 1040 (2019). PMID: 31725707.*

Pathologists: In-House Experts

The in-house laboratory is a valuable resource for both clinicians and patients

An interview with Ruthy Shaco-Levy

Could you describe how pathology fits into precision oncology?

Pathology is developing at light speed compared with other fields of medicine – and one of the fastest-developing areas is precision medicine. I think every pathologist deals with "precision pathology" – it's an integral part of the pathology report. For example, if we find a case of breast cancer, we report on tumor grade, tumor size, and receptor status. Separating molecular pathology is artificial; why would you select one part of the examination process and complete it in a separate place?

Why do you test in-house at your institution?

Israel has a "national health basket" of drugs and diagnostic tests. Hospital laboratories perform all tests included in the basket. Initially, we did a lot of immunohistochemistry and PCR; now, we do much more of our testing with next-generation sequencing (NGS) because it's more efficient and more accurate

It's important for me as a pathologist to correlate my findings with each test result. For example, if I have a breast cancer case that looks like a low-grade lobular or ductal carcinoma and HER2 comes back strongly positive, I have to ask myself some important questions: Do the results make sense? Is my diagnosis correct? Ultimately, I might choose to repeat the tests or to confirm my diagnosis – and that's something that can only really be done if you test in-house.

Also, we must not forget to develop our pathologists. Molecular pathology is an increasingly vital part of our profession and, if pathologists and laboratory medicine professionals aren't given the opportunity to practice, we won't be able to use those tools when we need them. We play a key role in patient care, and we owe it to them to keep our abilities honed.

Have you had experience with centralized testing?

A few years ago, my hospital chain tried to centralize our laboratory testing. Unfortunately, the lab was not equipped to handle our testing needs or connected to a pathology department and, to make a long story short, it failed. Over the few years of our centralization, a wide gap developed between our capabilities and those of hospitals that had not been centralized. Fortunately, since our testing moved back in-house, we've closed that gap.

Last year, there was an initiative to move all non-small cell lung cancer (NSCLC) NGS testing in Israel to a large commercial laboratory from overseas. The Israeli Association of Pathologists and other experts, including oncologists, strongly opposed this for many reasons: long turnaround times, loss of the ability to coordinate the patient care in cross-disciplinary tumor boards on-site, and more. As a result of this opposition, from July 2020 onward, local pathology departments will perform all DNA/RNA NGS analysis for NSCLC patient samples – the best possible outcome for patients, pathologists, and the healthcare system as a whole.

So what are the key benefits of in-house testing?

First, reducing turnaround time to results; some cancer patients have a very rapid clinical course and need test results right away – especially for companion diagnostics. If we send material abroad, it can take weeks to get the results. Patients can't wait that long to start treatment, so they may receive ineffective or even harmful chemotherapy. Turnaround time is critical in pathology in general, and especially in molecular pathology for cancer patients.

Second, preserving precious sample. Many hospitals use smaller panels for their precision testing. In lung cancer, for instance, you can assay a few dozen genes or you can assay hundreds. The more genes you test, the more biopsy tissue you need – and the less remains for future tests. When we test in-house, we carefully select our tests based on the available material. By only asking the most important questions, we make sure there's enough material to get answers.

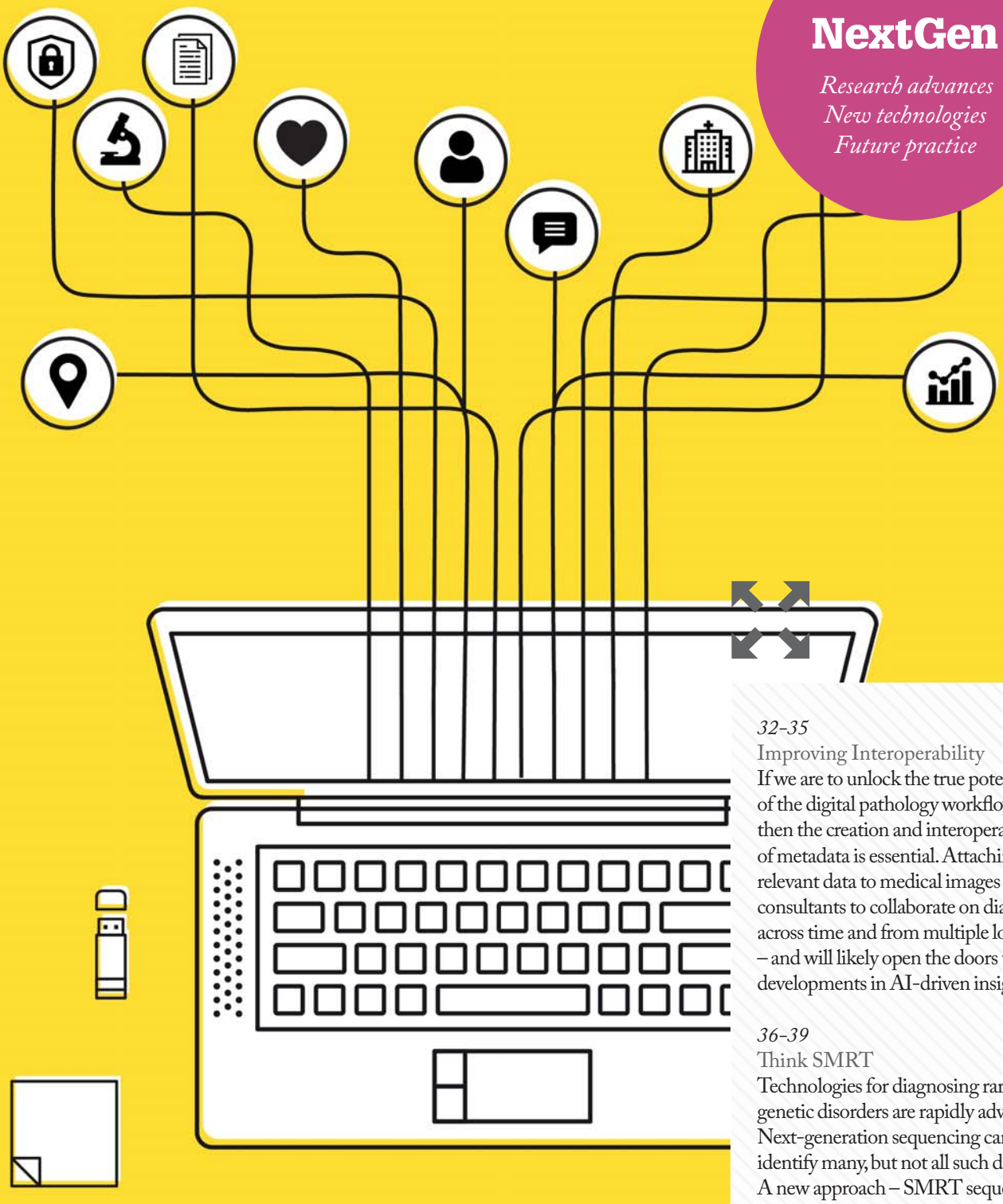
And third, keeping tissue in-house. As noted, biopsy material is precious; sending it out risks loss or damage. Even if the sample reaches its destination safely, we may not receive any material back because other labs may test less conservatively, forcing patients to undergo another biopsy if they need further testing. It's far safer to avoid sending tissue out at all.

It's the pathologist – the expert – who selects the appropriate assay based not only on how much tissue is available, but also its quality. In-house, that decision can be made on a case-by-case basis, but central labs often apply the same large panels to all material – and those panels are "all-or-nothing," so if there isn't enough material, you can't prioritize the most important genes. That means patients with insufficient high-quality tissue must undergo a repeat biopsy or risk having no answers at all – an unacceptable outcome that makes in-house testing vital for true precision oncology.

Ruthy Shaco-Levy is Professor and Head of Pathology at Soroka Medical Center, Clalit Health Services, and Head of the Israeli Pathologists Association, Beer-Sheva, Israel.

NextGen

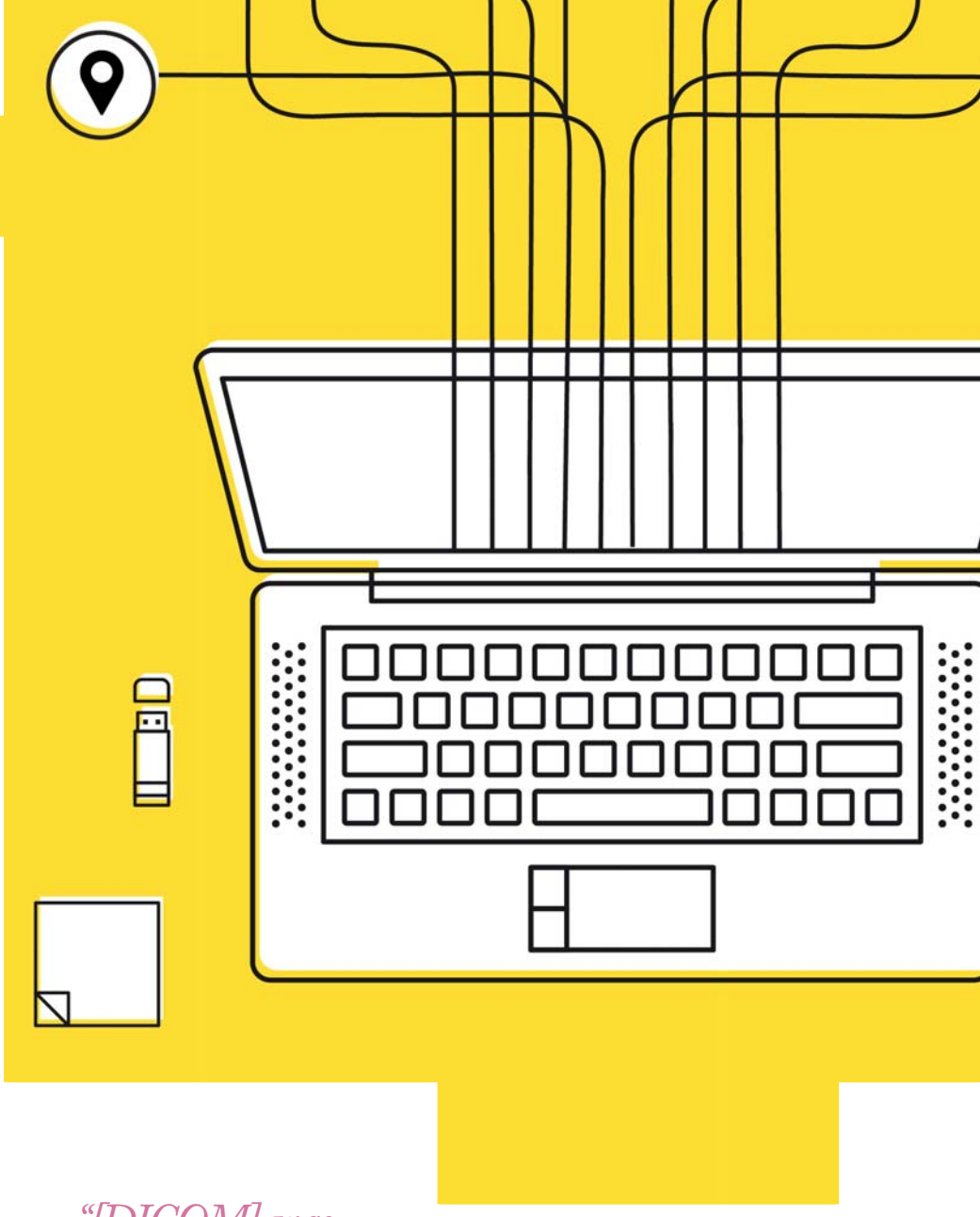
Research advances
New technologies
Future practice



Improving Interoperability

For an efficient AI-powered diagnostic workflow, it is crucial to ensure that pathology images and associated metadata are connected at the source

By David Dimond



After attending a recent digital pathology event, I became excited about the interoperability commitments industry vendors are making. Breakthroughs from standards-based working groups, such as Digital Imaging and Communications in Medicine (DICOM) and the Integrated Healthcare Enterprise (IHE), along with updated guidance from the US Food and Drug Administration, have set the stage for a more sustainable and innovative approach t digital pathology.

The recent increase of activity in the DICOM standards community, for example, is a clear sign of progress. The first meeting of DICOM Working Group 26 (DICOM WG-26) took place in 2005, but the scope of their Connectathon interoperability demonstrations at this year's Pathology Visions conference reached new levels.

For the uninitiated, both the DICOM organization and IHE are standards development organizations, with the goal of improving the interoperability of health information technology (HIT) systems – addressing specific clinical needs in support of optimal patient care. The DICOM organization was established in 1983 to develop a specific standard for the communication of medical imaging information. DICOM WG-26 is a working group within the DICOM organization that focuses on the development of a DICOM imaging standard for whole-slide imaging (WSI) in digital pathology. IHE promotes the coordinated use of established standards to achieve interoperability between HIT systems and effective use of electronic health records (EHRs) – and has a strong relationship with DICOM. Pathology and laboratory medicine (PaLM) is a domain of the IHE that addresses information sharing and workflow related to in vitro diagnostic testing in anatomic pathology, clinical laboratories, and at the point of care. The standards and use cases have been defined and vendors are starting to adopt them – first in academic research organizations

"[DICOM] was established in 1983 to develop a specific standard for the communication of medical imaging information."

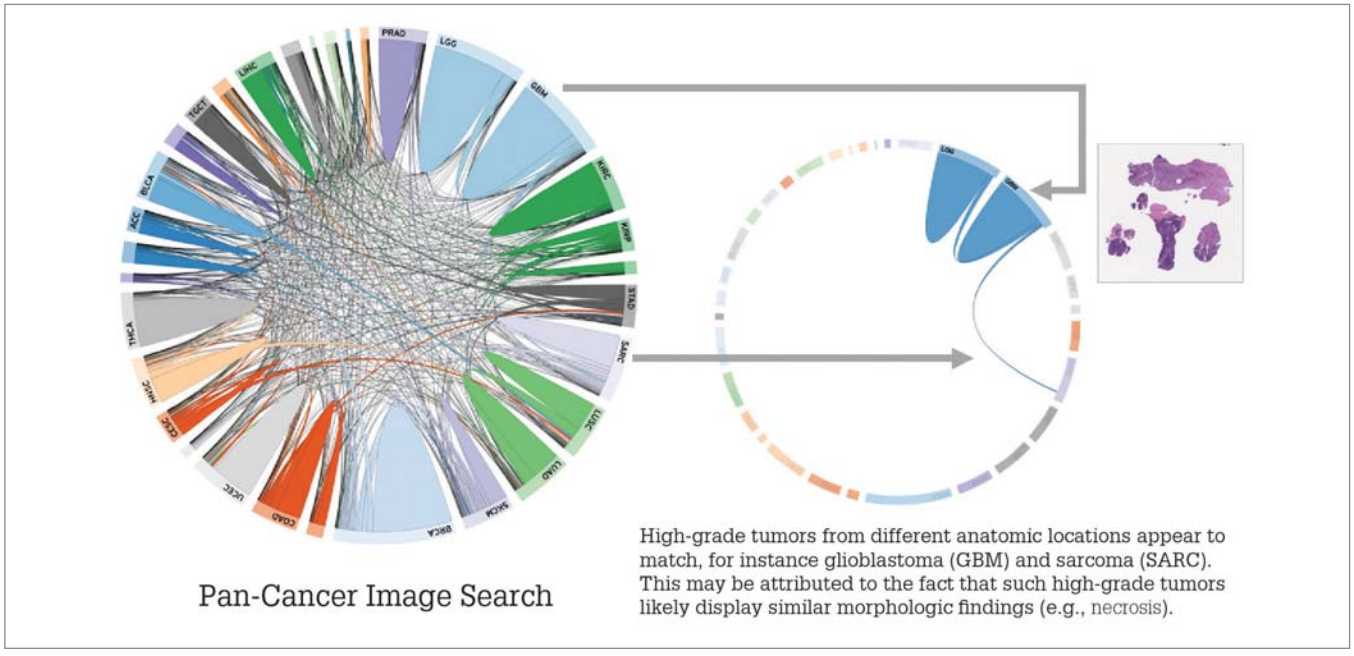


Figure 1: Chord diagram illustrating connections between cancer subtypes using image search and associated slide metadata. Image courtesy of Kimia Lab and Huron Digital Pathology.

and then within traditional healthcare providers.

I've evangelized DICOM and IHE standards for Radiology and Cardiology PACS for years. I've built teams who used the standards in both advisory and consulting work to help select, design, and implement some of the largest PACS systems in the US. These experiences taught me to appreciate the nuances of technology integration in healthcare.

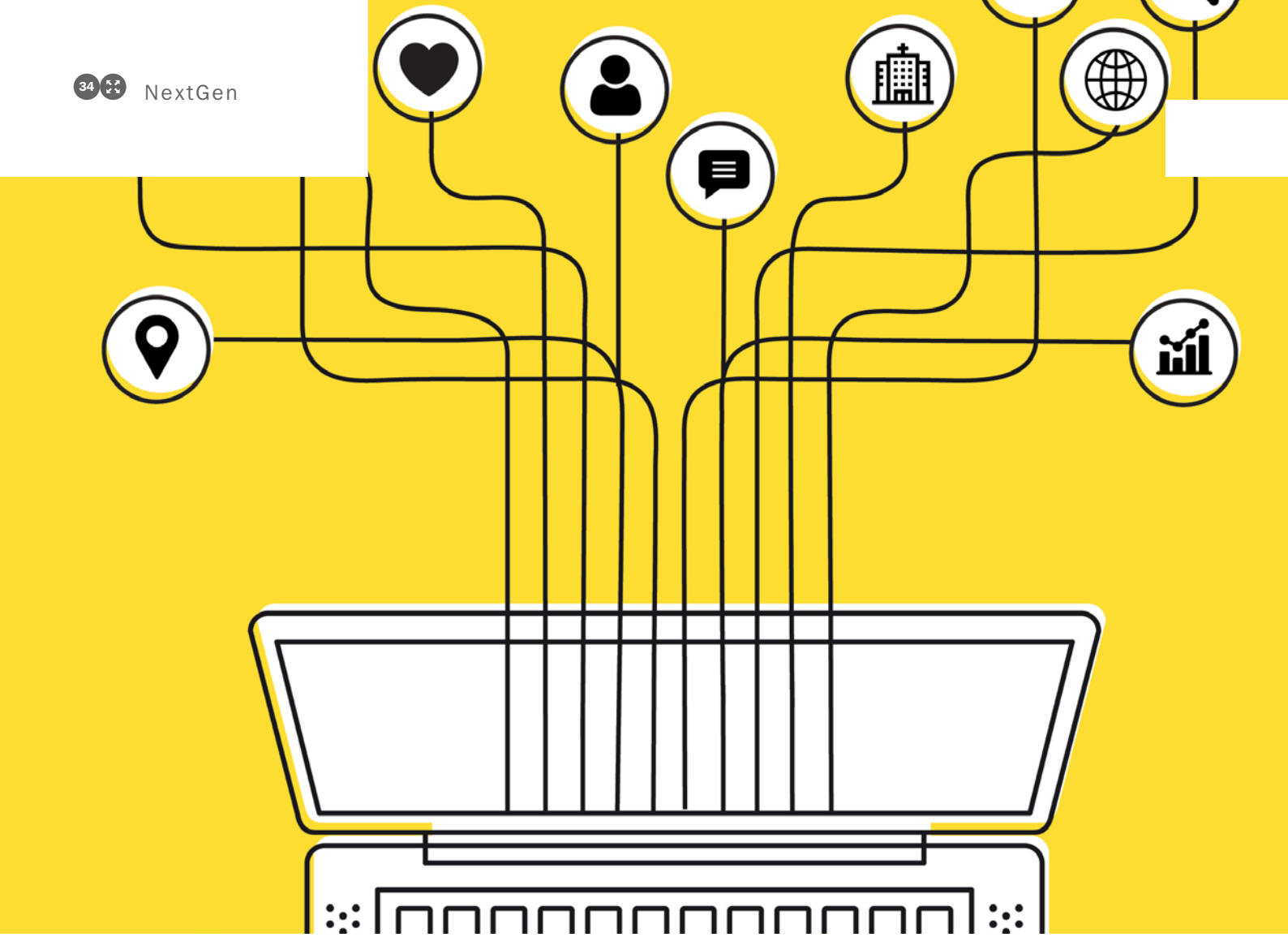
Mining metadata

Although the increase in interest is exciting, there's a key feature at the center of these developments that requires careful consideration. From my observations, what we need going forward are "metadata-enhanced workflows."

Digital pathology has the potential to become the new standard of care, but digitization is only one part of the equation. Digital pathology's biggest strength comes from metadata – the information that can be attached to digital images to become part of an efficient medical workflow. This includes everything relevant to the images: annotations, medical notes, patient identification, clinical order data, modality protocols, and date and time stamps. It is the composite of all information that is associated with a medical image, whether collected at the time of imaging or added after the fact, to create a "packet" needed for workflow enablement, data integrity, provenance, and security.

Without metadata, an archive of WSI files is like storing files on a hard drive using just file and folder names – cumbersome to categorize, search, and use. Conversely, if the pathology images come with embedded data and (more importantly) DICOM-standardized tags, there is a greater opportunity to generate insight during the clinical diagnostic workflow and research phases. Subsequent patient care also becomes interconnected as the metadata builds, ensuring that each WSI file retains an audit trail, and giving diagnosticians and clinicians access to a holistic, longitudinal patient

"Digital pathology's biggest strength comes from metadata – the information that can be attached to digital images to become part of an efficient medical workflow."



view. Researchers also benefit because metadata enhances their ability to harvest high-quality data from biobanks and distributed research archives.

The ability to "line up" the WSI tags with diagnostic images, case histories, and electronic health record (EHR) data also allows pathologists to effectively collaborate with virtual teams, such as tumor boards, for difficult-to-diagnose cases. This has the potential to significantly impact patient survival rates because consultations happen rapidly and collectively. Furthermore, peer reviews and multidisciplinary meetings can become more seamless. Considering the global shortage of pathologists, streamlining their workflow will yield an enormous efficiency benefit, helping them use their valuable time more effectively.

Putting the pieces together

Combining data with images is a complex task that relies heavily on improved data interoperability. When radiologists made the switch to digital, for instance, it took most radiology departments up to 15 years to realize the benefits of enterprise Picture Archiving and Communication Systems (PACS). The prior lack of interoperability inhibited productivity, slowing down adoption rates. The good news is that, in comparison, progress in digital pathology technology is happening at light speed – largely thanks to the groundwork radiology has laid. For example, DICOM Working Group 26 is leveraging DICOM standards by combining WSI images and patient data into one format. It's essential to have a standard like this so that equipment and software from different vendors can interoperate. DICOM Working Group 26 is also collaborating with teams from the IHE initiative in pathology and laboratory medicine to define how data relating to specimens, diagnostic observations, and documentation should be structured.

For pathologists to take full advantage of digital pathology, they must look at it from a workflow-first perspective and determine what can be improved when moving to a digital workflow. The ideal digital workflow includes access to all case-related whole-slide images via an image viewer integrated with the laboratory information system (LIS). Pathologists can then capture regions of interest – preferably

using some automated capabilities such as barcodes and other forms of metadata – and seamlessly export these to the LIS report as needed. They should then be able to share this data with colleagues around the world for secondary consults, using vendor-agnostic communication protocols for improved collaboration. One of the key goals is to invest in an interoperable digital pathology solution, avoiding the pitfalls of potentially obsolete proprietary systems in the future – a problem currently seen with some of the post-PACS radiology Vendor Neutral Archive (VNA) implementations.

I currently work with medical institutions to develop and implement this type of workflow. By partnering with leading digital pathology scanner and PACS vendors, we gather bundled solutions to enable data portability and accessibility. For example, we provide the technology and infrastructure solutions to enable one vendor's unique indexing technologies, which adds metadata in the form of hardware-generated barcodes at the point of scanning to link the relevant data to the patient EHR from the outset. We did this by developing specialized storage solutions to maintain the link between images and metadata to span multiple public and private clouds in various geographic locations. By implementing this approach, healthcare organizations not only gain the data mobility and accessibility needed, but also avoid the "data gravity" problem, in which the sheer size of data impedes its use outside its original repository.

"The ideal digital workflow includes access to all case-related whole-slide images via an image viewer integrated with the laboratory information system."

As the quantity of digital pathology data grows, increasingly sophisticated analytical platforms are needed to glean new insights, opening doors to machine learning and artificial intelligence (AI). There is growing awareness – particularly from IT customers – that there's a wealth of future benefits in store for patient treatment when insights from digital pathology images and data can be combined with other disciplines like genomics and radiology.

By systematically tagging WSI files with metadata, organizations can categorize whole-slide images by patient name, diagnostic site, institution name, and more. They can integrate images with patient reports by cross-referencing demographic data, automatically control access to clinical data based on level of network access, label images for rules-based data retention purposes, and generate end-to-end custodial audit trails from the moment of making the scan. The resultant metadata-enhanced digital pathology files allow healthcare and life sciences organizations to enhance clinical collaboration, streamline reporting, strengthen patient data security, and simplify data management.

Within this highly specialized area of digital pathology, research organizations and industry must work together on solutions that incorporate the standards referenced in this article. By doing so, they will be able to share pathology data seamlessly with other departments, both internally and with other healthcare and life sciences organizations. Once the integration of disparate systems with well-formulated and standardized data tags has been established, digital pathology can take the next step toward an enhanced workflow across the continuum of care. From there, a whole new world of AI-driven, federated data analytics tools will present opportunities that we've only just begun to imagine.

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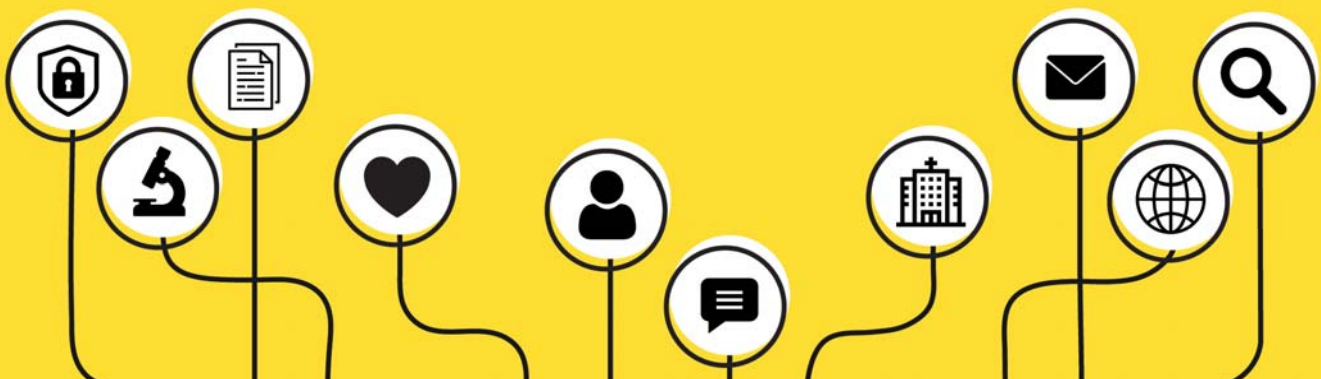


Think SMRT

Solving rare disease with single molecule, real-time sequencing

By Luke Hickey

We are in a golden age of rare disease research. Never before have our laboratory techniques been so successful at identifying rare diseases and elucidating their underlying biological causes. The knowledge we obtain today opens the door to new treatments, giving hope to people who suffer from these rare disorders.

Many of the recent advances in rare disease research stem from technology innovations in DNA sequencing. Falling costs have increased access to whole exome and whole genome sequencing as tools to assess the genetic basis of individual rare disease cases. And in a relatively short time, the genome-scale data these methods produce has transformed the community's understanding of how these diseases arise through rare genetic mutations. There are now more than 7,000 known rare and Mendelian genetic diseases identified – with more added to the databases every year – providing an invaluable information resource for genome-wide screening and exploration.

But even with these next-generation sequencing (NGS) tools, clinical research teams have been unable to explain the genetic basis behind a large percentage of rare disease cases. Solve rates range from 25 to 50 percent, leaving many individuals and their clinical teams without an answer to end the diagnostic odyssey. With an estimated 400 million people worldwide affected by a rare disease, there is still a large underserved population and a pressing need to improve our diagnostic yield.

To that end, scientists have begun deploying a higher-resolution DNA sequencing technology known as single molecule, real-time (SMRT) sequencing. SMRT sequencing differs from previous NGS tools by providing longer reads and even higher accuracy (1). In just the past few years, researchers have used SMRT whole genome sequencing to solve previously intractable rare diseases – and other significant efforts are now underway.

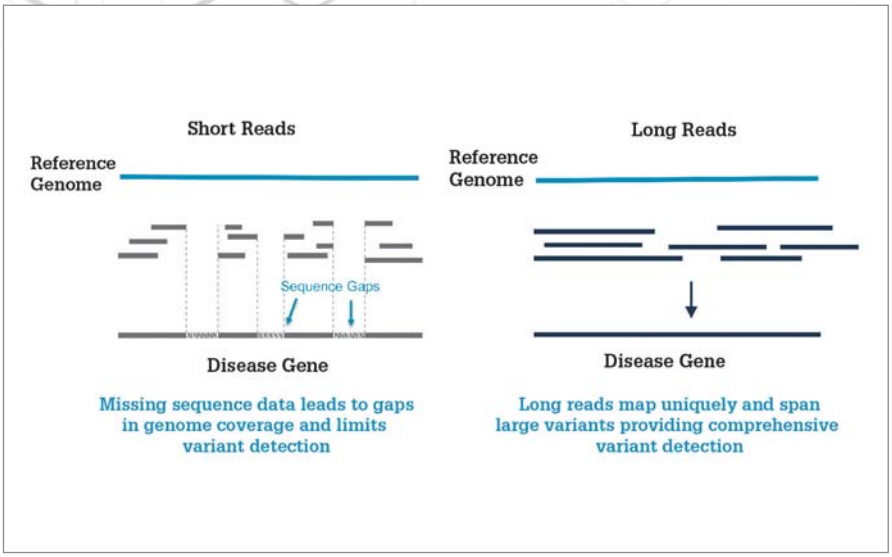


Figure 1. Short-read sequencing produces reads of 50–350 bp, which can lead to sequence gaps and incomplete coverage of disease-causing gene regions. Long-read sequencing produces reads tens of kilobases long, providing high-quality mapping across a genome for comprehensive variant detection.

The long and the short of it

NGS tools use a variety of approaches to generate sequence data. What they have in common, though, is that they all produce short-read data. Massively parallel short-read sequencing platforms have a low cost per run, but generate sequence reads that are typically only 50–350 base pairs long. To identify genetic abnormalities, these short reads are either mapped to a reference genome or bioinformatically "stitched back together" in a complex assembly process.

Short reads are useful for detecting certain variant types known to occur in the human genome, such as single nucleotide variants (SNVs) and insertions or deletions (indels) less than 10 base pairs long. But, for larger variants, short reads are of limited utility. The challenge lies in mapping across larger structural variants and indels; for instance, those that occur in repeat expansion disorders, such as fragile X syndrome, amyotrophic lateral sclerosis, and schizophrenia. Mapping issues associated with read length also limit short-read sequencing's ability to call variants across an entire genome. This includes variants in 193 medically relevant genes in the exome. For example, a 200-base sequence read may align to many different regions of the reference genome, leading to sequence gaps and conflating similar regions, such as pseudogenes, repetitive regions, and mobile elements (see Figure 1).

Short reads also tend to defy variant phasing efforts, making it impossible to distinguish maternally from paternally derived haplotypes. Why does this matter? It can be important when determining whether an individual has a functional copy of a gene in cases where multiple mutations are present. Perhaps most importantly, short-read sequence information often misses the structural variants (50 base pairs or longer; see Figure 2) that comprise most of the sequence variation between any two individuals' genomes (2). These larger variants can be called incorrectly or excluded entirely from genomes sequenced with short-read data alone.

In contrast, longer individual reads can fully cover even large structural

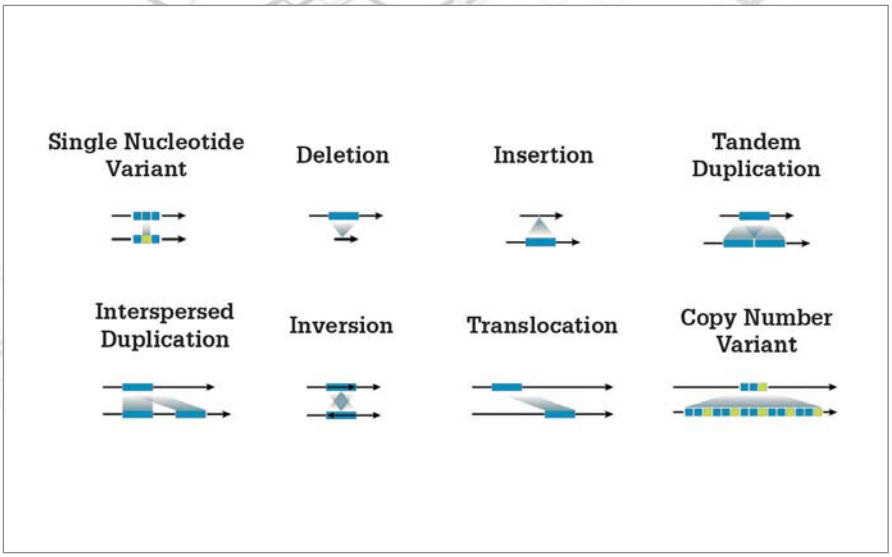


Figure 2. The types of sequence variants found in a human genome. Variants range in size from 1 bp (single nucleotide variant), to >50 bp for larger structural variants such as deletions, insertions, duplications, inversions translocations, and copy number variants (3).

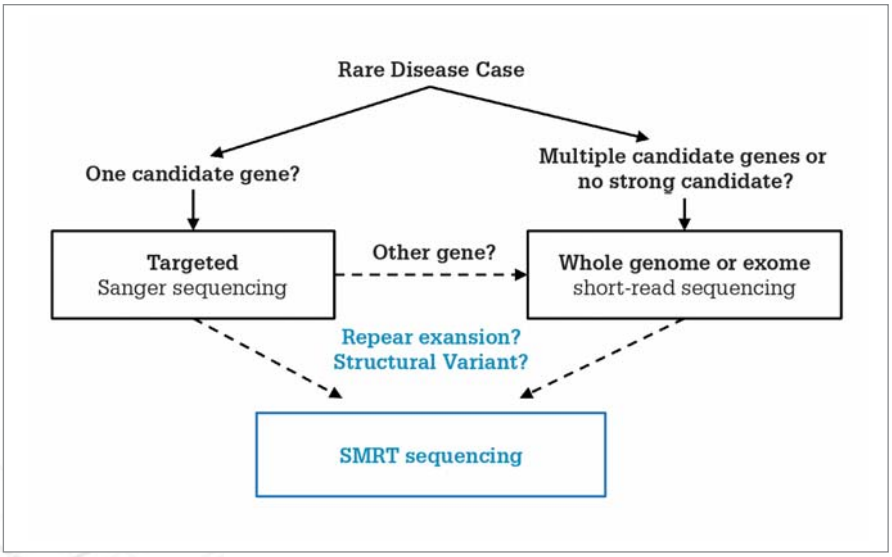


Figure 3. A workflow for identifying pathogenic mutations in rare disease cases. Adapted from (5).

variants, removing assembly ambiguity problems and revealing several times more structural variants, with higher precision and recall, than short reads (3).

Genomic dark matter

As short-read systems became more affordable, scientists were eager to apply these new genetic tools to rare disease cases. Exome sequencing and, eventually, whole genome sequencing with these platforms turned out to be a game-changer. Diseases that had long resisted explanation were suddenly understandable thanks to DNA sequence data. It seemed that there was finally an approach that could discover as-yet unknown pathogenic variants in disease-causing genes, giving affected families long-sought answers.

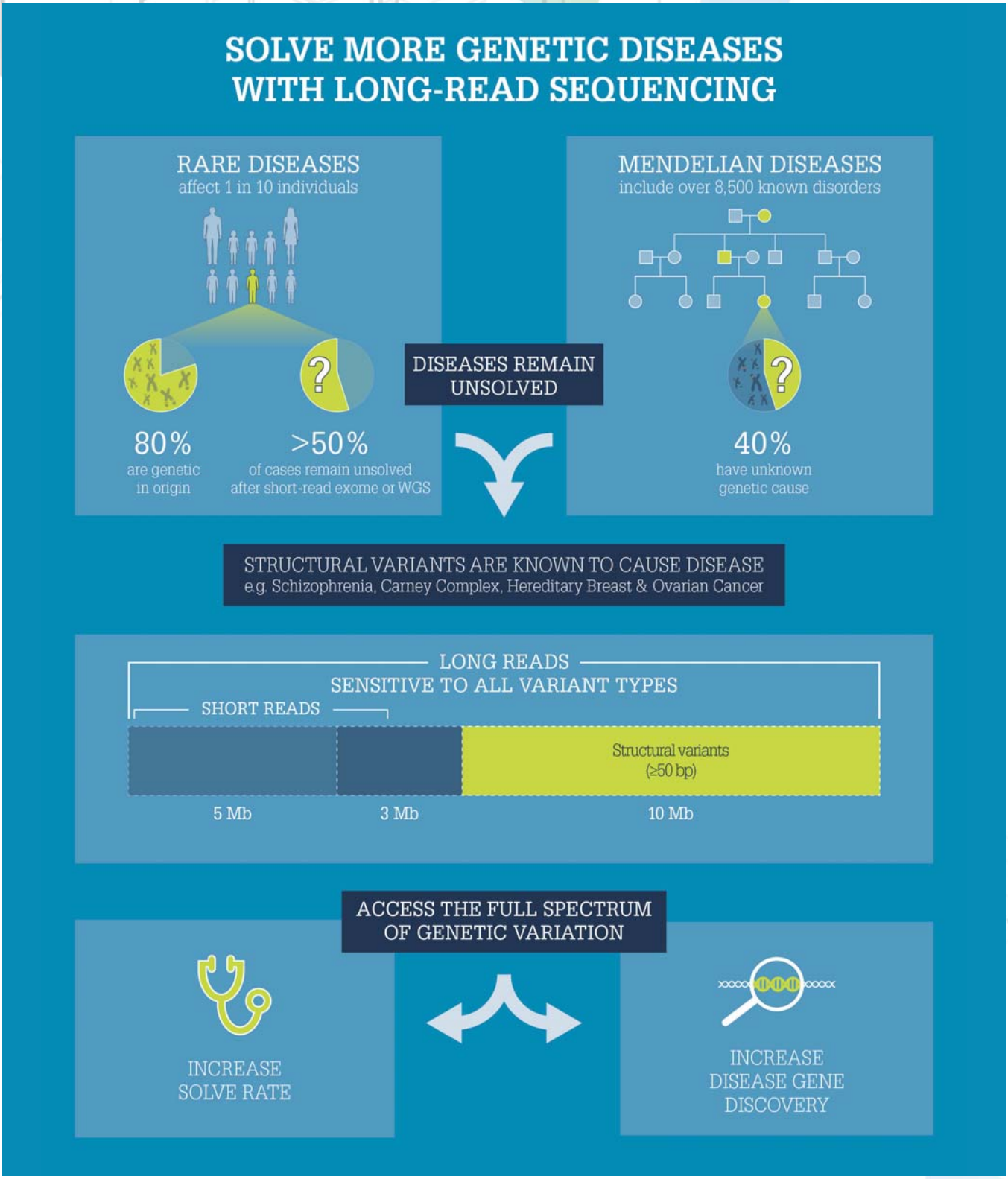
Certainly, short-read NGS tools have significantly increased the diagnostic yield for rare disease cases, but they cannot provide answers for every situation. In fact, scientists estimate that NGS platforms leave more than half of rare disease cases still unsolved.

Could these remaining diseases be caused by something other than genetic mutations? Unlikely, given what we already know about rare diseases; based in part on hereditary patterns and syndromic features, the vast majority appear to be driven by genetic mechanisms (4). What seems more likely is that over 50 percent of genetic mutations accountable for these diseases are invisible to short-read technologies. We do know that several genetic variants can be pathogenic – including repeat expansions, large deletions, complex rearrangements, transposable elements, and more. Now, with growing awareness that short-read NGS tools cannot accurately detect most pathogenic structural variants, scientists are turning to long-read sequencing – and answers have begun to emerge.

Identifying pathogenic structural variants

In one of the first examples of SMRT sequencing on a rare disease, scientists from Stanford University reported the discovery of a disease-causing mutation in an individual who had suffered a series of benign tumors over the course of two decades (5). Although the patient met the clinical criteria for Carney complex, a rare genetic disorder, experts had spent eight years performing various types of genetic analyses without success. Because they could not find the underlying mutation, they were unable to provide a confirmed diagnosis.

Ultimately, Stanford scientists decided to try SMRT sequencing, which led to


SOLVE MORE GENETIC DISEASES WITH LONG-READ SEQUENCING
RARE DISEASES
affect 1 in 10 individuals
80%
are genetic in origin
>50%
of cases remain unsolved after short-read exome or WGS
MENDELIAN DISEASES
include over 8,500 known disorders
40%
have unknown genetic cause
DISEASES REMAIN UNSOLVED
STRUCTURAL VARIANTS ARE KNOWN TO CAUSE DISEASE
e.g. Schizophrenia, Carney Complex, Hereditary Breast & Ovarian Cancer
LONG READS
SENSITIVE TO ALL VARIANT TYPES
SHORT READS
Structural variants (≥50 bp)
5 Mb
3 Mb
10 Mb
ACCESS THE FULL SPECTRUM OF GENETIC VARIATION
INCREASE SOLVE RATE
INCREASE DISEASE GENE DISCOVERY

"Scientists engaged in rare disease research are adopting SMRT sequencing technologies more readily."

the answer – a disease-causing deletion, stretching more than two kilobases, that affects the gene associated with Carney complex. The team sequenced the same region in the individual's parents, finding that neither carried the same mutation, allowing them to classify the mutation as de novo in the affected patient.

In another case, researchers at Yokohama City University and other institutes in Japan deployed SMRT sequencing to investigate the genetic mechanism responsible for the progressive myoclonic epilepsy affecting two siblings in a family (6). But it was not their first attempt to find an answer; previous efforts, including exome sequencing with short-read sequencing tools, had proven unsuccessful. SMRT sequencing allowed the scientists to focus on potentially causative structural variants. A quick filter of the over 17,000 structural variants found across the genome led to a homozygous 12.4 kilobase deletion in a gene known to be associated with a disease that causes similar clinical symptoms to those found in the siblings. Notably, the deletion fell in a region with high GC content, which poses processing challenges to short-read sequencers. Follow-up testing confirmed the deletion and proved that it was pathogenic.

There have been many other rare disease diagnostic victories based on SMRT sequencing (see Figure 3). Among them, advances in repeat expansion disorders stand out. These large runs of repetitive sequence are associated with a wide range of conditions, and the number of repeats is often closely tied to the severity of disease. Long-read sequence data can fully span these large regions and deliver direct, countable results – an approach that has been used for ataxias, fragile X syndrome, myotonic dystrophy, and other disorders (7–11).

Large-scale efforts to solve rare disease

As the number of reported successes ramps up, scientists engaged in rare disease research are adopting SMRT sequencing technologies more readily.

In Europe, the SOLVE-RD consortium consists of nearly two dozen institutions in 10 countries. Funded by a €15 million award from the European Union, SOLVE-RD works to improve the diagnosis and treatment of rare diseases that have evaded explanation. The program will sequence 500 whole genomes with long-read sequencing tools to find disease-causing variants and increase solve rates.

In the USA, the National Institutes of Health-funded Clinical Sequencing Exploratory Research program uses SMRT sequencing as part of a large effort to increase the diagnostic success rate for pediatric cases that have proven challenging with other approaches. Scientists at the HudsonAlpha Institute for Biotechnology are generating whole genome sequences for hundreds of children with intellectual and developmental disabilities for which the genetic cause has not yet been found.

Large-scale programs like these should contribute a significant amount of new knowledge about the genetic mechanisms underlying rare disease, filling in many of the gaps in our understanding today. As SMRT sequencing helps to explain more rare diseases and increase overall diagnostic yield, it should have a profound effect on our ability to diagnose, understand, and ultimately improve treatment for rare disease cases.

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References

1. *AM Wenger et al., "Accurate circular consensus long-read sequencing improves variant detection and assembly of a human genome", Nat Biotechnol, 37, 1155 (2019). PMID: 31406327.*
2. *J Huddleston et al., "Discovery and genotyping of structural variation from long-read haploid genome sequence data", Genome Res, 27, 677 (2017). PMID: 27895111.*
3. *EE Eichler, "Genetic variation, comparative genomics, and the diagnosis of disease", N Engl J Med, 381, 64 (2019). PMID: 31269367.*
4. *National Human Genome Research Institute, "Rare Diseases FAQ" (2020). Available at: https://bit.ly/2V9uz8X.*
5. *J Merker et al., "Long-read genome sequencing identifies causal structural variation in a Mendelian disease", Genet Med, 20, 159 (2018). PMID: 28640241.*
6. *T Mizuguchi et al., "A 12-kb structural variation in progressive myoclonic epilepsy was newly identified by long-read whole-genome sequencing", J Hum Genet, 64, 359 (2019). PMID: 30760880.*
7. *S Mitsuhashi, N Matsumoto, "Long-read sequencing for rare human genetic diseases", J Hum Genet, 65, 11 (2020). PMID: 31558760.*
8. *T Mantere et al., "Long-read sequencing emerging in medical genetics", Front Genet, 10, 426 (2019). PMID: 31134132.*
9. *I Höijer et al., "Detailed analysis of HTT repeat elements in human blood using targeted amplification-free long-read sequencing", Hum Mutat, 39, 1262 (2018). PMID: 29932473.*
10. *B Schüle et al., "Parkinson's disease associated with pure ATXN10 repeat expansion", NPJ Parkinsons Dis, 3, 27 (2017). PMID: 28890930.*
11. *S Ardui et al., "Single molecule real-time (SMRT) sequencing comes of age: applications and utilities for medical diagnostics", Nucleic Acids Res, 46, 2159 (2018). PMID: 29401301.*

Profession

Your career
Your business
Your life

The Pathfinder of Pathology

Christopher D.M. Fletcher discusses the changes that have taken place over a decades-long career in pathology – and the field's promise for the future

Pallavi A. Patil interviews Christopher D.M. Fletcher

Christopher Fletcher is Professor of Pathology at Harvard Medical School, Senior Pathologist and Vice Chair of Anatomic Pathology at Brigham and Women's Hospital, and Chief of Onco-Pathology at the Dana-Farber Cancer Institute. With an extensive history of awards and publications, his long-time focus is on the pathologic diagnosis and classification of soft tissue tumors.

His 35-year-long career has yielded wisdom in many facets of pathology, and he is highly regarded as a mentor by those who have had the privilege of learning from him. Now, Pallavi A. Patil – a fellow in gastrointestinal and liver pathology at Yale University – interviews Fletcher to spread his wisdom to those pathologists and laboratory medicine professionals who have not yet had the opportunity to meet him.

What inspired you to choose pathology?

I discovered I liked pathology in medical school. I did reasonably well at it but, truthfully, I did reasonably well in most subjects – pathology just somehow grabbed my attention. I realized that it's important because it's the underpinning of so many parts of medicine. I understood clearly that pathology was the field for me when I was taking the medical school final examinations. If you knew pathology, you could answer 80 percent of every exam paper. Pathology is incredibly important.

On several occasions, I have been fortunate to identify a new entity in soft tissue pathology. Although it can be a challenge to be the first to report such a discovery to the scientific community, it's also a privilege – and it comes from two things.

First, if you are lucky enough to see a lot of consult cases, you have a better chance of seeing something new – and potentially seeing it more than once. I see cases every single day that I can't identify; some are probably novel disease entities, but I don't see enough of them to recognize them.

Second, a lot of it probably relies on visual memory. I am constantly amazed that we still see and describe previously unrecognized entities even now. We are currently working on a couple for which I started collecting the cases in the early 1990s, nearly 30 years ago. When I think something is going to be a novel disease entity, I try to use certain keywords to make it easier to retrieve the cases from our files. I find it astonishing that, for example, if we pull 50 cases of something that has never been described but is in my head, 45 of them usually look pretty much identical. Fortunately, the keywords make it easy to collate and describe them.

You've edited a key textbook in your field, the Diagnostic Histopathology of Tumors. How did that come about?

Everything in life is an accident – at least, I haven't planned anything in my life. In the early 1990s, Nicholas Wright, Chairman of Pathology at the Royal Postgraduate Medical School in London, suggested to Churchill Livingstone the idea of publishing a new book focused only on diagnostic pathology of tumors. They contacted me and I, in turn, selected the contributors for each individual chapter. (I did, and still always do, edit every chapter to make sure they don't feel dramatically different from one another.) My involvement in the book came about by serendipity, but it's hard work that keeps it up-to-date and in print.

Elsevier eventually bought Churchill Livingstone and now expect a new edition every five years or so. An awful lot of new information is generated in that time; as a result, they considered an online edition that could be edited continuously, but that would be a full-time job for an editor – and no one has the time. Some have suggested delegating the task of keeping topics up-to-date to younger pathologists, but it's important to ensure that each topic has high-quality contributors aware of the latest developments in their fields, because it's works like these that inform how we and our colleagues approach diagnostic medicine. It's not a responsibility to be taken lightly.

Personally, I spend the years in between editions collecting papers that I think are important. I am old-fashioned, so I have them printed out and filed in different folders instead of saved to my computer. When I get a new chapter, I pull out the relevant folder and make sure all of the new information is included. It's a lot of work! Although the current edition is finished and in the proof stage, I still keep copies of important papers – I've been using this system ever since the first edition. I used to receive hard copies of journals, but now I just get content lists and have to view the articles online. It's difficult to find that kind of time, and it makes adding references much harder.

I've just finished work on the fifth edition, but I foresee a change in the book's future. People seem to like smaller books nowadays. It's fashionable to have skinny books with bullet points rather than large amounts of free text. I don't think people will want to use big, older-style books for much longer. Publishers aim to make money; they always want a new product and a new market, but no pathologist gets rich writing books – not even skinny books. Publishers did themselves a disservice by distributing digital editions; after all, if many users (usually in one pathology

department) can share one book, why purchase multiple copies?

What energizes you for your many tasks – managing administration, a large consult service, teaching, and editing a major book?

It's nice to feel that I'm doing something useful. I enjoy helping young pathologists get into their careers. I never thought I would end up managing so many people, but it turns out that I quite like it. It does become somewhat tiring; here in anatomic pathology at the Brigham, we have around 70 faculty and 50 trainees! With so many people, you have to keep an eye on things like workload, logistics, interpersonal issues, trouble at home… In the end, people are important. If you don't care about people, they will not do well.

There is one thing I often used to say about administration and leadership that I don't say much anymore: that if you don't do it yourself, somebody else will, and they may mess it up. There are all sorts of people I would not want to be my boss!

Multitasking has become part and parcel of professional life. What would you advise faculty and trainees regarding burnout?

If you think about it, that whole concept has emerged in the last four or five years. There isn't really a good work-related reason why people should burn out more now than they did 10 or 20 years ago – except that society has changed. Now everybody wants everything fast. Sensory input is more immediate. People (not me) have to deal with social media all the time. I completely avoid social media because I think I would shoot myself. I find it hard enough to keep up with email.

I get 200 to 300 emails a day that are not spam and I do my best to take care of them. Nowadays, you can send a question to someone by email instantly and you often know exactly when they received it, too. In the old days, people would send a letter that took a week to arrive and then you would write a letter back. The pace of interaction was slower. Now, because access is so unlimited, everyone can find everyone else via email. I get emails from doctors and patients around the world every single day. I used to find it very stressful trying to fit them into my day. Now I usually just do many of them in the evening. But despite the stress, it is humbling to have all those people communicating with you – and you learn a lot about disease.

My advice regarding burnout would be: don't overcommit yourself. I will be honest; I think a lot of it is simply a reflection of how modern society works, so it's not something you can control. You will get bombarded with emails, texts, and Twitter messages – and, if that's how you grew up, you think it's a normal part of life. I am lucky not to have grown up dealing with that. I am old enough now that I can ignore them and get away with it. If you are 25 years old, you can't do that or people may think you are a "weirdo." Maybe what we call burnout is just becoming a part of life – there seems to be a constant level of overstimulation.

The other thing is that people nowadays seem to have unrealistic expectations, perhaps because so many things are instantaneous. When we get consults, people (usually clinicians, not pathologists) from referring hospitals start calling within hours of the cases' arrival to find out if there is a report or diagnosis. This happens even when they themselves have worked on the case for a couple of weeks before sending it to Boston. When a case is that difficult,

you have to be patient. The report will be sent when it is ready. Everybody wants everything right now; instant gratification is a very American thing, and the rest of the world always seems to end up copying Americans – have you noticed?

Physicians who directly interact with patients often undervalue pathology or lack a full understanding of how a report is generated. How can we change this?

Medical students see less and less pathology in medical school. They often have no significant pathology course in medical school and, because very few medical students do pathology electives, most have no idea what we do. They don't know whether a test takes 10 minutes (like some blood tests) or multiple days (for a tumor that needs mutational analysis)! We have turned into the Wizard of Oz. We are behind the curtain coming up with a diagnosis and they don't know where we got it from. That's a big problem. For example, when you diagnose a synovial sarcoma, the oncologist might say, "Will you check to see if there is SS18 gene rearrangement?" In straightforward cases, the pathologist should say no – you only need molecular testing when you have a difficult differential diagnosis. But clinicians tend to believe in molecular testing because it appears to be objective, unlike relying on the pathologist's brain.

My consults can be a nightmare in some respects – but, in others, very interesting and humbling. I get 120 to 130 new cases a week, and I have no idea what 20 percent of them are. I have to make my best guess as to what they might be or how I think they might behave, and, of course, that's where some of the new entities come from. It is quite stressful. I find myself thinking about cases all the time but, in most instances, the extra thought brings me no closer to an answer.

If it's a lesion in another specialty, such as pulmonary or gynecologic pathology, I happily show it to experts in that field in our department– but for soft tissue entities that are just plain weird to explain, there often isn't anybody who can really help! Many patients come to the sarcoma clinic here now, and a significant subset of them come because they got a pathology consult from me first. Our sarcoma oncologists have become very good at understanding that, for many cases, there is no black and white answer. Some patients who get my consults go to other disease centers, which may not be familiar with my reports and call me to ask, "You just say it is malignant; you don't say anything else. How are we supposed to treat it?" I have to explain to them that I don't know what specific entity it is, and – although I discuss options with them – it's their job to figure out how to treat it. They seem astonished that pathology cannot definitively identify the disease entity, so I try to educate them that it happens almost every single day. It's a big responsibility when "the buck stops here," and it falls to me to explain that not every slide results in a definitive diagnosis.

How can we address this? We need to educate people. Clinicopathological conferences are an opportunity to teach clinicians how we do what we do, and how long the testing and subsequent reports may take. Pathologists are not handmaidens. In many places, pathologists just do whatever the clinicians ask for. But we are not servants; we are equal players in the care of our patients. Sometimes we must remind them that they don't know what they are talking about. If they have never heard of a particular disease entity, make them realize how dependent they are on us.

You once ran a fee-for-service consultation service in the UK. What happened?

In the UK, where I am from, nobody pays for healthcare except the small minority

who use private practice. It's a socialized system – but there was a time in the early 1990s when the government wanted to make healthcare market-driven. They wanted the referring hospital to pay the receiving hospital for every patient or case that went from one district to another, even though it was all one central budget and the hospitals were all a part of the National Health Service. I was unlucky because they picked me as a pilot project for consultation services. They instructed me to send bills for the consults, starting with those from the UK. Nobody in the UK or Europe had ever billed for a pathology consult before. As you can imagine, people were very angry and called to say, "What is this bill? Nobody else sends bills." I spent hours on the phone explaining that it was the government who wanted to send the bill, not me!

I had a soft tissue tumor unit with two histotechnologists and a secretary. The hospital administrators were instructed to close the unit down if I didn't bring in enough money to cover salaries for the technicians and secretary, as well as lab costs (which, of course, I never did). They put the staff on monthly renewable contracts instead of annual ones. It was quite stressful. European patients generally didn't have to pay for healthcare in other European countries – but then the UK Department of Health decided that I should start billing for European consults as well. That was funny, because cases from Europe were sent anyway without ever sending payments. In the UK, people were wondering whether they could keep sending cases or not, what was going to happen with the bills, and whether they were going to get into trouble. It was in the newspapers and on television. It was horrible – but the government scheme failed and was eventually dropped.

There is an increasing trend in the USA towards mergers and corporatization, particularly in pathology. How do mergers affect pathology?

Medicine in the USA has become more and more corporate, and less and less a profession where you try to heal the sick. Physicians are often employees of a company that is trying to make money. Even not-for-profit organizations need funding to put up new buildings or to buy the latest technology. It has various negative consequences. For example, many places want doctors to do more and more work with less and less support. Even hospitals here in Boston cut back on support staff, such as histotechnologists, secretaries, and so on. They make changes in the name of efficiency – things like asking us to use voice recognition rather than typing assistance. It just puts pressure on doctors to do more with less help.

To maximize billing revenue, administrators want the most complex cases to be done in the big hospitals. Big teaching hospitals have hundreds of trainees, complicated equipment, and lots of expenditures. In hospital networks, they try to push smaller procedures to the smaller hospitals – for example, lumpectomies, herniorrhaphies, or varicose vein surgeries – because their overhead isn't so high. It changes the pathology as well. I have been here at Harvard for almost 25 years and the case mix is constantly changing. I see less and less simple material. Our trainees rarely see a hernia sac, an appendix, or a gallbladder. A lot of the simple things have just disappeared because they're now done in the community hospitals.

How much should pathologists interact directly with patients?

Decades ago, I established a patient consultation practice in the UK. I had no plans to do so, but I had started getting quite a few consults and people wanted treatment advice. We didn't have a sarcoma clinic in the hospital I worked at in the late 1980s. I persuaded an orthopedic surgeon to be interested in limb tumors, a general surgeon to take care of trunk and retroperitoneal tumors, and an oncologist (who did both radiation oncology and chemotherapy back then because there wasn't much chemotherapy) to handle that side of things. This group of practitioners began seeing patients and it grew into an actual scheduled sarcoma clinic. I would see the patients with them, and on my own if they were out of town. If the patients got lung metastases, they often sent me into the room to explain about metastases!

If patients feel they don't understand their disease and need to talk to somebody, clinicians should always consider directing them to pathologists – and pathologists should make themselves available. These interactions happen more by email in the US, which is difficult because you don't necessarily have precise or complete patient details (especially if you're talking to somebody thousands of miles away). You have to be very careful about how you phrase things if you don't know the specific circumstances. That said, patients here sometimes want to come and look at the slides and are generally appreciative. If they ask, I don't think we should ever say no or be shy of talking to patients. I know there is a community hospital in Lowell, Massachusetts, where they recently had clinicians offer patients, after a diagnosis of cancer, the option of talking to the pathologist. Most of the patients who met with the pathologist loved it and felt they knew more about their disease. The conversation helped them visualize the disease and feel better about it. Clinicians like to appear to be the ones to know whether something is benign or malignant, when in fact the pathologist makes the diagnosis. The patients end up appreciating the clinician for work the pathologist has done. Many clinicians don't talk about pathologists too much, so I appreciate the ones who offer patients the opportunity

to speak to us – even if not every patient takes that offer.

One problem is that a lot of people think of pathology as something vaguely creepy that has to do with death. I find even now, when I go to social events, people don't talk very much if I say, "I am a pathologist." If I say instead, "When you have a biopsy, people like me decide whether it is benign or malignant," then they become interested. When their perception is that pathology is autopsies or "weird stuff," they don't want to hear about it. We have an image problem, and I don't know how to change that except to say that we shouldn't hide.

My father, a surgeon, wasn't very pleased when I went into pathology. Remember, this was in 1982; for him, pathologists and anesthesiologists were like servants. Fortunately, he changed his mind after about 15 years! I find that, when I meet residency applicants or new residents, their families may not have much idea about their profession, and some don't even understand that pathologists are "proper" physicians!

People have a lot of ideas for improving the image of pathology. There was a time, about 15 years ago, when pathologists thought it might be a good idea to make FAQ lists for diseases that we could give clinicians to share with patients. That never really caught on, because clinicians didn't necessarily understand the answers or how to explain them. They didn't use them because they felt uncomfortable. If we make the effort to answer their questions in person, we can avoid that discomfort and spread awareness of our profession and its value.

How can we encourage medical students to apply for pathology?

The drop in applications is very troubling. I think that, to solve the problem for the long term, we have to restore more pathology to the medical school curriculum. That will be hard because, in modern education, they shorten the curricula and pack far too much into them. In the way that pathology used to be taught (40 years ago!), systematically, I had a full three-month pathology course in my fourth year of medical school. We learned about etiology, pathogenesis, and microscopic appearances. There was no immunohistochemistry or molecular biology then, but we learned about outcomes and complications of disease. So much of that pathology was invaluable in taking exams across most other medical specialties!

Over the last 20 years, everybody has been focused on social medicine and training good family practitioners. In almost every medical school, pathology (and histology and anatomy) teaching is much more limited than it used to be. The Medical University of South Carolina in Charleston does still have a three-month pathology course just like I had at medical school, and 10 to 15 students go into pathology there each year because they've had that exposure. But here at Harvard, only two or three students each year go into pathology – so the impact is obvious. Those students who do come into pathology residency, no matter how smart they are, often have very little idea what they are doing. It's not their fault; they have probably never seen normal pancreas under a microscope! I think it's very hard to learn pathology from that kind of starting point, and I find it worrying that this is the new normal.

How could pathology training be improved?

Residency programs are successful when faculty like to teach. It's no good when trainees are taken for granted; they shouldn't just be grossing specimens with little guidance or passively sitting on the other side of the two-headed scope. Modern medicine sometimes takes away teaching time; faculty are frustrated by the number of commitments and demands placed on their time and teaching falls by the wayside. Insurance companies often demand shortened turnaround times as a "quality measure," so trainees don't always get to preview things – but that's how I think residents gain the most knowledge. When I met with some insurance company representatives 20 years ago in Massachusetts, they wanted our turnaround times to be shorter. They were trying to find a way to pay less if we didn't meet the turnaround times they wanted to set. Somebody questioned the need for resident preview time to be factored in. I said, "Those are the people who will be looking at your prostate biopsies in about 15 years. Do you want us to teach them or not?" They hadn't thought about it that way. As soon as they realized it could be their specimens under the microscope, they changed their minds!

I think the most important thing is to get good training. One of the reasons residency training in the US was shortened to four years was financial – to save paying for that fifth year. But being well-trained is critical to what we do. It doesn't matter how smart you are, you can't learn without seeing enough cases. There is a tendency for places to use trainee logbooks to tick off, for example, that they have seen 20 colorectal carcinomas – but then the trainee may wrongly imagine that they don't need to see any more. That's not true, because the more you see, the more you realize how heterogeneous human disease is. There is no substitute for experience. The more you see, the better you get, and the more you realize how much you don't know.

People go through phases; quite a few have little or no self-doubt in their 30s or 40s and think that they know almost everything. Most of them, if they are smart, will eventually discover that they don't really know as much as they think. By the time they are in their 60s, they will hedge a little more often or admit to being unsure. As you grow older, it becomes okay to say, "I don't know." People don't like ambiguity when they are younger;

they want everything in black and white. But many things about pathology (and life!) are ambiguous. There are always shades of grey.

What do you foresee in pathology's future?

Genetics and genomics will play a significant role – but they won't be the right solution for everyone. I think every new technology becomes very important for a subset of patients. For example, finding a new therapeutic target via next-generation sequencing is great for 7 to 10 percent of cancer patients, but the other 90 percent don't benefit at all. In most tumors, it is currently of limited help. It is fashionable to try to find something when there are no options. Patients read newspapers or leaflets in oncologists' offices and ask to have tests done. Physicians order the tests because the patients (customers!) want them – and so, medicine is changing. We have become more responsive to what patients want, rather than telling them what should be done. I sometimes deal with patients who demand tests. If the test they want is completely irrelevant to their disease, I try to explain that it is not useful and that all they are doing is throwing money away. Clinicians don't say no very much these days – and sometimes that can lead to crazy tests.

When Gleevec came out, people around the country wanted to test for KIT (CD117) expression in tumor types without any known KIT mutations. American medicine is not very good at saying, "I am sorry. There is nothing more we can do, so let's concentrate on making you comfortable, reducing your pain, and finishing your bucket list." Instead, patients get endless chemotherapy, complications, and ICU stays. I don't know if it is the patients who refuse to give up, or society that has created false hope. It is not good. I wouldn't do it. Most doctors, when extremely sick with cancer, avoid crazy testing, and instead focus on pain relief and support, and know when there is nothing more to be done. It is very telling that most doctors don't want these aggressive or protracted treatment efforts – so why do we do it for so many of our patients?

Telepathology may also become a routine part of our work, but that depends on the specimen type. For a small biopsy with a single slide, it could be very useful – but if you have 30 slides of a pancreatic resection, for example, current technology is still slow and challenging to use. The technology keeps getting better, so I think it will probably become useful, but right now I don't like it much. When we look at a glass slide under the microscope, we all scan the slide at low power – perhaps subconsciously – before we decide what's worth focusing on. It's usually much harder when you have a digital slide to realize what is important at low power, because software is often slow to focus when scanning. When people send me weird cases as digital slides, I find I usually can't make a definitive diagnosis. It takes me much longer than when using a microscope and glass slides.

It would be great to get an opinion on a frozen section consult from someone who is off-campus if you have good software. We set up a system here more than 10 years ago but, in the end, nobody used it very much. Why? Because the person on the receiving end wasn't comfortable enough making a diagnosis that way and would drive to the hospital to make it in person. We older folks grew up in pathology the old-fashioned way and it's hard to change brains that are wired that way. Millennials, on the other hand, have grown up using technology since early childhood, so they may more easily become proficient in using digital images! By the time they are my age, digital pathology may be entirely routine, and pathologists may sign out cases from home without having to come to the hospital. Society changes!

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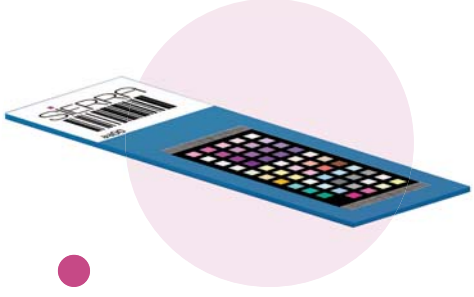

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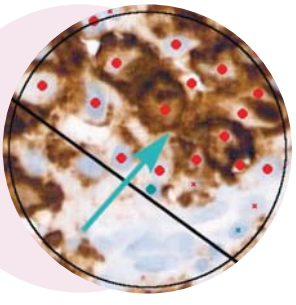

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The Pathologists' Assistant

Sitting Down With… Sarah Garner, Pathologists' Assistant and Director of the Pathologists' Assistant Program at Tulane University, New Orleans, Louisiana, USA

What drew you to pathology and a career as a pathologists' assistant?

Like many who pursue pathology, I fell into it and realized it was perfect for me. During my undergraduate studies, I worked in internal medicine and saw patients daily. One day, a man came in with a large testicular mass and – although I loved speaking with him and wanted to help him – what I really wanted to know was how and why the tumor had formed, what it looked like, and what treatment he would need. Now I know I wanted to understand the epidemiology, pathogenesis, and morphologic features. That's the day I realized I was meant to do something other than see patients – but, at the time, I had no idea what pathology really was (and, unfortunately, none of the physicians I worked with did either).

While working in internal medicine, I took a gross anatomy class with cadaveric dissection. I loved the class so much that I stuck around as a teaching assistant and quickly learned that I wanted my career to involve teaching and learning about the human body. When I stumbled across the pathologists' assistant (PA) profession after Internet searches for careers in anatomy and abnormalities, I knew I had found something that would set my world alight! It seemed like the perfect way to combine all my passions: gross anatomy, dissection, pathology, art, and teaching.

When I speak to other PAs about their route into the career, many of them have similar stories about stumbling upon the profession and realizing it was a perfect fit. It always makes me wonder how many people who would really love this job never find it. We shouldn't have to stumble upon this wonderful profession – or any pathology specialties – and that is why, as a practicing PA and educator, I strive to promote pathology and the PA profession wherever I can.

Can you describe "a day in the life" of a PA?

That's impossible to answer. And that's exactly why I love my job so much! Even if I'm technically doing the same things, every specimen, patient, and day is different. Sometimes I spend the whole day in the gross room, grossing specimens or teaching residents, medical students, and PA students. Equally, I may be working outside the gross room – and the majority of my days are spent doing a little of both. If I had to generalize a typical day, it would go something like this: teach gross anatomy to first-year medical students, spend a few hours in the gross room either grossing or training others, teach a didactic class for PA students, and then one for my graduate and undergraduate students. Other days can include autopsy, presenting at resident conferences, and sometimes teaching for an entire day. I love that I am always doing something different, because nothing ever gets boring and I get to work with a lot of different people.

Are there differences in how you approach teaching pathology residents and PAs?

I designed the curriculum for our PA students and play some role in all of it, so I always know their exact progress and can tailor my teaching techniques to fit their current knowledge. It's highly personalized, and the integrated curriculum teaches them what they need and want to learn. In contrast, residents typically come from a variety of medical schools and backgrounds, so it's more difficult to gauge individual needs. Some might have worked extensively in pathology, whereas others might have no experience at all. Luckily, our residency program is relatively small, so I can get to know each of them and adapt accordingly.

Most of the time, I find it more challenging to teach a first-year pathology resident than a PA student in the gross room, because the PA students have didactic coursework specifically designed to teach them the concepts of grossing and why each section is important. Residents often don't arrive with that knowledge, because it's something they're expected to learn during residency. The residents also get limited time, or their rotations are broken up into months spread throughout the year, so the lack of consistency can make it more challenging for them to learn and familiarize themselves with grossing concepts. I do my best to teach the "why" behind all of the sections and measurements and not just the "how," because I think that leads to better patient care, whether you're a PA or a resident. Our PA curriculum and residency education overlap significantly – and all classes and conferences are available for everyone to attend.

"I do my best to teach the 'why' behind all of the sections and measurements and not just the 'how,' because I think that leads to better patient care."

If you could go back and give yourself one piece of advice at the start of your career, what would it be?

I would tell myself to be patient, which I sometimes find difficult because I like to take on big tasks and I get really excited about what I'm doing. I'm still learning how to be patient, especially now that I am more involved with research. I'm also bad at accepting help from others and even worse at asking for it. I am getting better – but it's something that would have greatly benefited me years ago. You don't have to do everything yourself!

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- *Listeria monocytogenes*
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  - *Staphylococcus aureus*
  - *Staphylococcus epidermidis*
  - *Staphylococcus lugdunensis*
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  - *Streptococcus agalactiae*
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- *Candida auris*
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