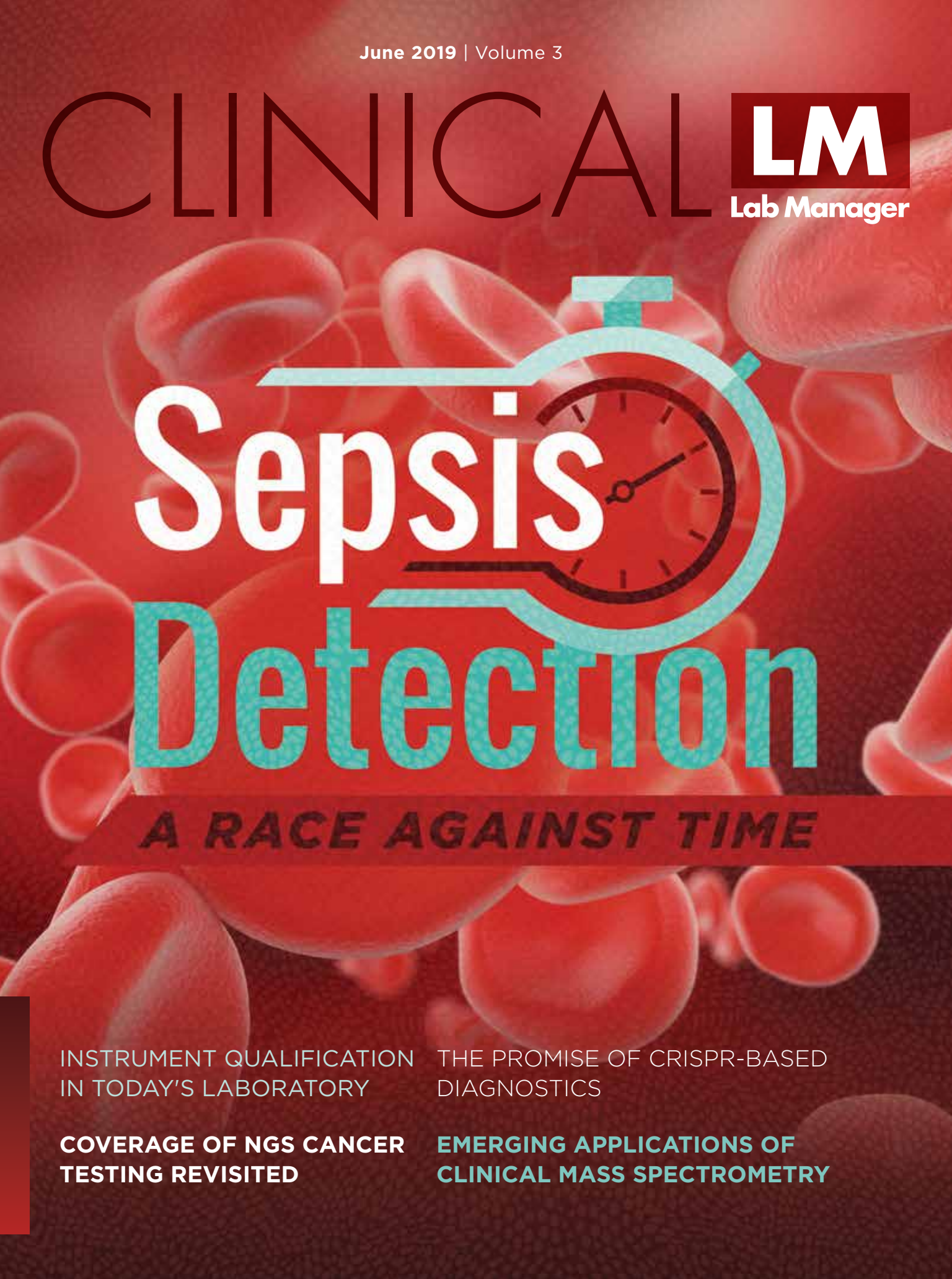
June 2019 | Volume 3

CLINICAL LM Lab Manager



Sepsis Detection

A RACE AGAINST TIME

INSTRUMENT QUALIFICATION IN TODAY'S LABORATORY

THE PROMISE OF CRISPR-BASED DIAGNOSTICS

COVERAGE OF NGS CANCER TESTING REVISITED

EMERGING APPLICATIONS OF CLINICAL MASS SPECTROMETRY

CLINICAL LM Lab Manager

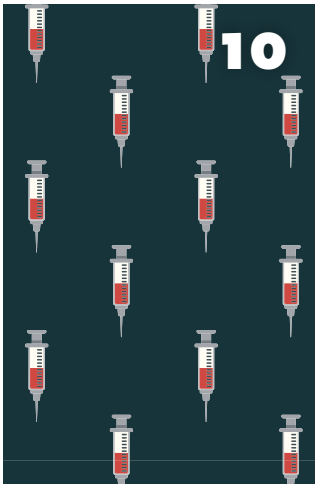
June 2019

CONTENTS

ClinicalLabManager.com

FEATURES

IN EVERY ISSUE



MilliporeSigma

Milli-Q® CLX 7000

Connected water purification systems, for maximum lab productivity

"Great instrument and great customer service! Much easier to use than our prior water system. Very easy to maintain and the water purity seems to be much better."

– Kimberly Bartlett, Chemistry Supervisor, Athens-Limestone Hospital, Alabama, USA

Now with Milli-Q® Connect remote service and monitoring capability

- Constant CLRW-quality water for analyzers, up to 3000 L daily
- Low and predictable running costs with minimal maintenance thanks to patented Elix® electrodeionization and E.R.A.™ technologies
- Reliable high-performance system, with 24/7 remote access and control, for maximal uptime
- Save time with rapid, online diagnostics—and even remote repair—by our service team
- Powerful data management facilitates data traceability, audits and lab accreditation

EMDMillipore.com/biomedical

2019 - 22892 05/2019

The life science business of Merck KGaA, Darmstadt, Germany operates as MilliporeSigma in the U.S. and Canada.

Milli-Q®

Lab Water Solutions

Transformational Technologies

Reading this month's cover story, I learned some startling statistics: Each year in the US, approximately 1.7 million people develop sepsis and nearly 270,000 people die as a result, and sepsis is responsible for one in three hospital deaths. For every hour that sepsis patients don't receive antibiotics, their risk of mortality increases dramatically, meaning sepsis detection is truly a race against time. "Speeding Up Sepsis Detection" discusses how point-of-care (POC) testing performed at or near the site of care has the potential to hasten sepsis diagnosis and, ultimately, improve patient outcomes. We delve into the latest POC sepsis testing devices and take a peek at what's on the horizon.

This issue focuses on technologies that are changing the face of the clinical laboratory. We sat down with co-founder and chief research officer of Mammoth Biosciences Janice Chen to learn how CRISPR, a revolutionary gene-editing tool, is also poised to revolutionize diagnostic testing. Our Technology feature, "Emerging Applications of Clinical Mass Spectrometry," explores how clinical mass spectrometry is making inroads into infectious disease and cancer diagnostics. Automation solutions for the clinical lab receive special attention in our latest Product Roundup. To round out the issue, this month's thought leaders explain why hospitals are increasingly insourcing their next-generation sequencing and outline the benefits of droplet digital PCR-based liquid biopsy.

There's plenty more inside—a lesson on installation qualification/operational qualification/performance qualification, tips for reducing repetitive laboratory testing, an update on coverage for certain types of cancer testing, and our top picks from the peer-reviewed literature. There's even more going on outside the issue, so be sure to check out ClinicalLabManager.com to read our original digital content and curated news, download our e-books and infographics, and subscribe to our weekly Clinical Tools & Techniques newsletter.

Erica Tennenhouse

Erica Tennenhouse, PhD, Editor

[Direct from whole blood]

[3 to 5 hours species ID]

[>90% sensitivity]

Stewardship Impact:

Only T2 enables changes in therapy in 3-5 hours

T2Direct Diagnostics: results you can rely on in just a few hours—direct from whole blood

Determine earlier effective antibiotic therapy through culture-independent diagnostics.

Getting results from traditional blood culture methods can take anywhere from 1 to 5 days, making it difficult to quickly treat patients with the targeted antimicrobial therapy they need. This can be compounded by a lack of sensitivity and accurate results with these methods.

T2Direct Diagnostics is the first and only FDA-cleared system to identify deadly sepsis-causing pathogens directly in whole blood **without the need to wait for blood culture results.**

Attending ASM?

Join us for these events:

Industry & Science Showcase

Theater North, Exhibit Hall
Friday, June 21
12:00–12:45 PM

In-booth Presentations

Daily with clinical experts

Instrument Demos

Daily with the T2Dx® Instrument

T2Biosystems®

One T2 result = **3.8** blood culture sets.

Visit us at Booth 1443

Our Top Picks from the Literature

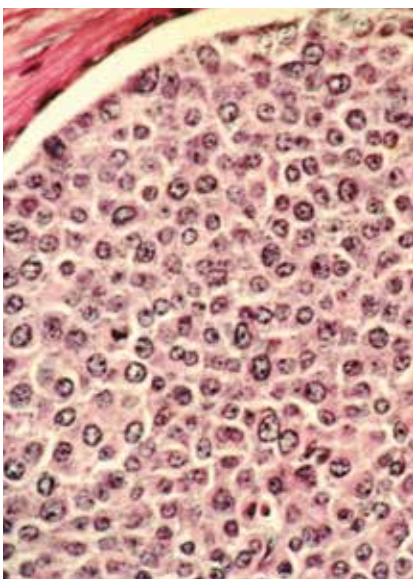
CTE Patients Share Distinct Tau Filament Structure

A novel tau protein filament structure has been found in the brains of individuals with chronic traumatic encephalopathy (CTE). There is no current therapeutic treatment for CTE, which occurs in those who have endured repeated head injuries or exposure to blast waves. As reported in *Nature* in March 2019, researchers scanned brain tissue from three individuals with CTE and examined their tau filament structures using cryo-electron microscopy. They found tau filaments that were identical to one another but structurally different from the tau filaments of patients with Alzheimer's and Pick's diseases. Despite structural similarities in the tau filaments associated with all these conditions, CTE patients have a variant orientation of the tau β-helix region with a hydrophobic cavity. This cavity encloses an additional density not linked to a tau, suggesting that cofactors may impact tau buildup in CTE. In contrast, tau filaments in the brains of Alzheimer's patients are not linked to hydrophobic cavities. These findings demonstrate that different tau protein assemblages are present in different neurodegenerative pathologies.

Falcon, Benjamin, *et al.* "Novel tau filament fold in chronic traumatic encephalopathy encloses hydrophobic molecules." *Nature* 568 (2019): 420-423.

How Cancer Drugs Impact the Immune Microenvironment

Researchers have identified potential cancer drug targets that could improve breast cancer patients' sensitivity to chemotherapy and immunotherapy. Changes to the immune system microenvironment that follow neoadjuvant treatment for primary breast cancer have been poorly understood. To elucidate how immune function is affected,



researchers determined tumor infiltrating lymphocyte (TIL) counts and PD-L1 immunohistochemistry in 60 paired pre- and post-treatment samples. They identified a large number of immunobiological processes and genes associated with higher immune cell infiltration and better response to chemotherapy, as well as genes and processes associated with worse response. The presence of activated T cells in the tumor microenvironment was associated with pathologic complete response, whereas stromal functions were associated with residual disease. Most immune functions decreased during neoadjuvant chemotherapy, the researchers reported in the *Journal for ImmunoTherapy of Cancer* in April 2019. However, immunotherapy targets PD-L1, IL6, and IL8 remained expressed in residual disease, suggesting they potentially could be exploited as therapeutic targets in residual cancer.

Li, Xiaotong, *et al.* "Immune profiling of pre- and post-treatment breast cancer tissues from the SWOG S0800 neoadjuvant trial." *Journal for ImmunoTherapy of Cancer* 7 (2019): 88.

Machine Learning Predicts Hospital Readmission

Machine learning is a more accurate predictor of hospital readmissions than standard methods, according to a study published in March 2019 in *JAMA Network Open.* Researchers assessed 16,649 adult discharges from three hospitals in Maryland during the fall of 2016 to generate a Baltimore score (B score) machine learning rank. A 30-day unexpected readmission rate was predicted using the B score and compared with results generated by

traditional scoring methods, such as the modified LACE score, the HOSPITAL score, and the Maxim/RightCare score. Unlike previously developed machine learning methods, the B score produces scores individualized for hospitals rather than for individual patients. The researchers found that the B score predicted individual hospital readmission rates with 25.5–54.9 percent greater accuracy than the other methods. The findings suggest that machine learning has great potential for predicting patient readmission, meaning it could be used to help reduce patient readmission in the future.

Morgan, Daniel J., *et al.* "Assessment of machine learning vs standard prediction rules for predicting hospital readmissions." *JAMA Network Open* 2 (2019): e190348.

Bacteria in Cervix May Signal Premature Births

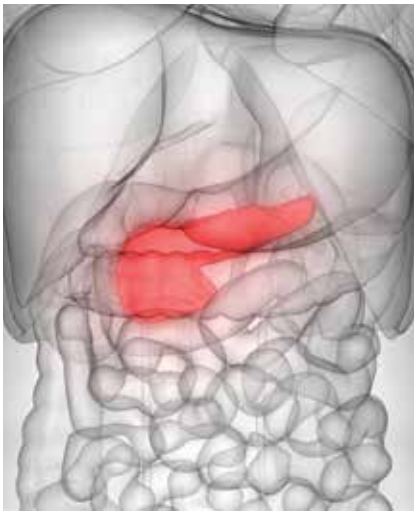
Several types of cervicovaginal bacteria are associated with increased risk of spontaneous preterm birth (sPTB), according to a study published in *Nature Communications* in March 2019. Spontaneous preterm birth, defined as birth occurring before 37 weeks of gestation, is a leading cause of neonatal death and harm worldwide. Until now, an inability to understand the causes of sPTB has limited the potential for prevention and treatment. In the study, researchers swabbed the cervicovaginal areas of 2,000 pregnant women during three different stages of their pregnancies to determine which bacteria were present. They found that seven bacterial taxa were associated with sPTB, with African American women showing the strongest effects. The bacteria most strongly linked to increased risk

of sPTB were *Mobiluncus curtisii/mulieris* and *Sneathia sanguinegens*. The researchers also found that having higher vaginal levels of β-defensin-2, a human immune system agent with strong antimicrobial activity, lowered the risk of sPTB, while lower levels increased the risk. These findings suggest treatment using immune modulators and microbiome-based therapeutics early in pregnancy could aid in prevention of sPTB.

Elovitz, Michal A., *et al.* "Cervicovaginal microbiota and local immune response modulate the risk of spontaneous preterm delivery." *Nature Communications* 10 (2019): 1305.

Earlier Detection of Pancreatic Cancer

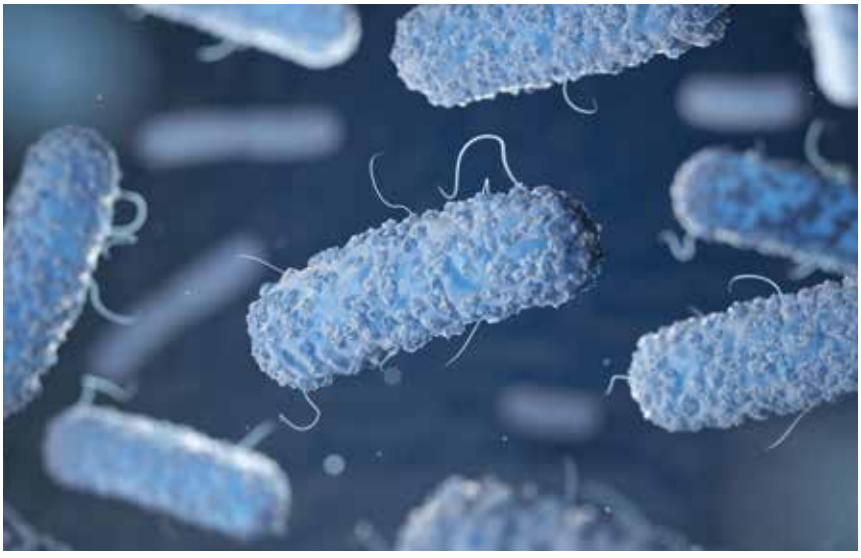
Researchers have performed the largest-ever real-time clinical genomic characterization of known targetable alterations and predictive biomarkers in pancreatic ductal adenocarcinomas (PDACs). Historically, pancreatic cancers have low five-year survival rates. PDACs are difficult to detect and treat because of differences in genome alterations across patients. To assess the full spectrum of genomic alterations in PDACs, the researchers analyzed 3,594 PDAC samples from an international cohort and found mutations in 317 genes. Of these gene changes, *KRAS* mutations were present in 88 percent of samples, while other commonly altered genes included *TP53*, *CDKN2A*, and *SMAD4*, the researchers reported in *Gastroenterology* in March 2019. They found that 17 percent of the tumor cells contained genetic alterations that could make them vulnerable to currently used chemotherapy treatments. These therapeutically relevant changes occurred primarily in genes in the BRCA-FANC family. Furthermore, the researchers found mutations in PDACs that could be used as warning signs that malignancy may develop, allowing for early detection of some pancreatic cancers.



Singhi, Aatur D., *et al.* "Real-time targeted genome profile analysis of pancreatic ductal adenocarcinomas identifies genetic alterations that might be targeted with existing drugs or used as biomarkers." *Gastroenterology* (2019).

Toward Precision Dosing of Vancomycin

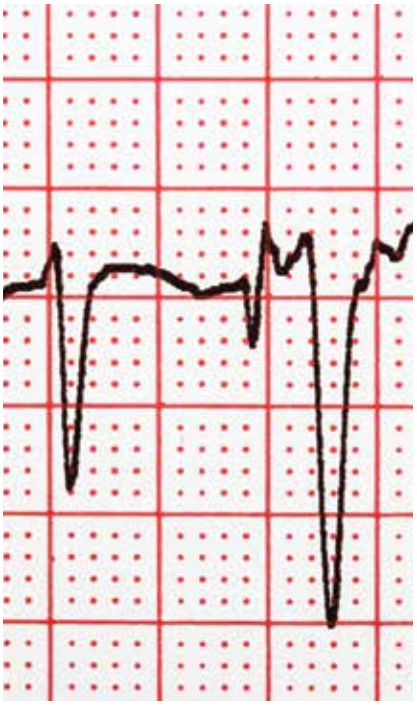
A new study determines which pharmacokinetic model for vancomycin has the best predictive performance. A crucial treatment option for patients suffering from serious infections, vancomycin works by stopping the growth of bacteria. However, dosage needs to be closely monitored to prevent patient harm. Researchers used 31 different published pharmacokinetic models and Bayesian forecasting to assess how accurately vancomycin concentration could be predicted. They used data from 292 patients treated with vancomycin to forecast the antibiotic's trough concentrations based on patient characteristics and known vancomycin concentrations following previous doses. Models that included patient body weight and creatinine clearance were most accurate, the researchers reported in March 2019 in *Clinical Microbiology and Infection*. The model by Goti and colleagues best predicted vancomycin trough concentration. According to the researchers, use of the Goti



model could improve precision dosing in hospitalized patients.

Broeker, A., *et al.* "Towards precision dosing of vancomycin: A systematic evaluation of pharmacometric models for Bayesian forecasting." *Clinical Microbiology and Infection* (2019).

Biomarkers Predict the Risk of Atrial Fibrillation



Two biomarkers have been identified that are associated with the risk of atrial fibrillation (AF), according to research published in the *Journal of the American College of Cardiology* in April 2019. AF—an abnormal heart rhythm with irregular pulsing of the atria—affects millions of people worldwide and is associated with substantial morbidity and mortality. The main substrate for AF perpetuation is left atrial myocardial interstitial fibrosis (MIF). Researchers discovered that the CCL+ and CD+ collagen biomarkers released due to MIF were significantly associated with atrial fibrillation. Of 392 patients studied, those with CCL+ and CD+ biomarkers had significantly lower left atrial voltage than patients with CCL-, CD-, or other biomarkers. The CCL+ and CD+ circulating biomarkers showed excessive myocardial type-1 deposition and cross linking and reveal that AF is more likely to be present after ablation treatment.

Ravassa, Susana, *et al.* "Combination of circulating type I collagen-related biomarkers is associated with atrial fibrillation." *Journal of the American College of Cardiology* 73 (2019): 1398-1410.

Pembrolizumab as a First-Line Therapy for Advanced Non-Small Cell Lung Cancer

Pembrolizumab therapy is an appropriate first-line therapy for cancer patients with advanced or metastatic non-small cell lung cancer (NSCLC), according to research published in *The Lancet* in April 2019. NSCLC, which represents the majority of lung cancers, often results in patient mortality. To evaluate overall survival following pembrolizumab monotherapy treatment, researchers assessed 1,274 patients with a PD-L1 tumor proportion score (TPS) of more than 1 percent from 213 medical centers across 32 countries. Half of the patients received pembrolizumab treatment, while the other half received a platinum-based chemotherapy treatment chosen by local clinicians. Both overall survival and progression-free survival were enhanced in patients receiving pembrolizumab therapy whose metastatic NSCLC had been previously untreated. The researchers concluded that the benefits of pembrolizumab monotherapy outweighed the risks in combating cancers with low PD-L1 TPS and without sensitizing *EGFR* or *ALK* alterations.

Mok, Tony S.K., *et al.* "Pembrolizumab versus chemotherapy for previously untreated, PD-L1-expressing, locally advanced or metastatic non-small-cell lung cancer (KEYNOTE-042): a randomised, open-label, controlled, phase 3 trial." *The Lancet* 393 (2019): 1819-1830.

The ABCs of Reducing Repetitive Laboratory Testing

TRIED AND TESTED APPROACHES TO CURB OVERUTILIZATION OF DAILY LABS

by Joesph R. Wiencek, PhD, and Andrew S. Parsons, MD, MPH

Low-value services are a major contributor to healthcare waste in the US. Commonly defined as patient care that could be eliminated without reducing health quality, these services carry an estimated cost of around $800 billion per year.[1]

When ordered in a clinically stable hospitalized patient, repetitive laboratory testing (aka daily labs) qualifies as one of these low-value services. Repetitive testing can put patients at risk: it can lead to iatrogenic anemia in an estimated 20 percent of hospitalized patients,[2] and ordering providers may act on clinically irrelevant abnormals or trigger the testing cascade, subjecting patients to further unnecessary and potentially harmful diagnostic and therapeutic interventions.[3]

Repetitive laboratory testing is therefore an ideal target for quality improvement interventions. Such interventions have been highlighted by multiple medical societies as part of the Choosing Wisely campaign, an initiative led by the American Board of Internal Medicine Foundation to educate providers and patients on potential areas of overuse. Effective initiatives to reduce unnecessary laboratory utilization bend the cost curve and fulfill the physician's first oath to do no harm.[4] In this article, we aim to highlight effective approaches to reducing overutilization of daily labs.

Audit. Individualized and group feedback on laboratory ordering practices is an effective way to reduce unnecessary utilization.[4] Studies have found that emails detailing provider-specific test ordering information sent at routine intervals or easily accessible clinician dashboards reduce the number of common laboratory tests ordered and laboratory costs per patient day.[5,6] Both anonymous comparison data and individualized reports appear to be effective. These reports can be sent to all ordering providers or they can be targeted to high utilizers. The limitation with this approach, if used alone, is sustainability, because laboratory ordering rates tend to return toward baseline rates after feedback reports are discontinued.[4]

Buy-in. Institutional buy-in is vital to transitioning from a culture of waste to one of high-value care.[7] To promote long-lasting change, the buy-in must have a multidisciplinary approach. First and foremost, buy-in must come from the institutional leadership and key chief executives, commonly referred to as the C-suite. Discussions with these key individuals must be strategically aligned with the institution's immediate and long-term goals. Support from the C-suite can often provide the leverage needed to facilitate pilot studies as well as resources such as project managers who could help tackle larger initiatives. It is important that these key members are updated on a regular basis about current and future projects, projected cost savings, and overall impact on patient care. Second, outside the C-suite, it is important to engage colleagues about various stewardship efforts and assure them that the primary focus is on appropriate care of the patient rather than on reducing costs. Sharing data and having open discussions in the form of departmental grand rounds, other small presentations, or informal one-on-one conversations can help spread the message of high-value care.

"Effective initiatives to reduce unnecessary laboratory utilization bend the cost curve and fulfill the physician's first oath to do no harm."

Competition. Even when done anonymously, comparing ordering practices among clinicians provides an incentive for change. At the University of Virginia Medical Center, we have an ongoing chief resident–led initiative in which internal medicine residents receive a weekly email containing individualized and team daily lab-ordering practices in comparison with those of their peers. This intervention, based on a similar initiative at Vanderbilt University Medical Center and paired with an educational online module, has shown an immediate and sustainable reduction in repetitive laboratory utilization.[8]

Decision support. The electronic health record (EHR) may be an underexplored resource to reduce unnecessary lab tests.[9] Through EHRs, clinical care teams can gather information from a patient's visit in real time and track lab tests with a click of a button. EHRs can also provide decision support at the point of entry and restrict tests to specific locations, patient populations, or ordering clinicians. The support tools can include rules on frequency (e.g., daily labs), sex-specific tests (e.g., prostate-specific antigen), costly send-out tests (e.g., genetics), and many more. They are typically vetted by

decision support committees that determine the best approach to implementation. There are also additional third-party decision support systems that can be incorporated into EHRs. However, these systems are in the early stages of development; it is therefore important that organizations evaluate and validate support tools before full adoption. Regardless of internal or external options, a well-designed decision support system may help clinicians make better use of laboratory tests, but more data is needed.

Education. Interventions focused solely on education have found mixed results.[10] Educating clinicians commonly leads to an initial decrease in laboratory ordering practices, but the approach lacks sustainability. There is evidence that resident trainees are particularly prone to overutilization of common laboratory tests, which is not surprising given the known association between diagnostic uncertainty and increased laboratory ordering.[11] Additionally, a recent study found that nurses and advanced-practice clinicians may be more prone to overutilization of common laboratory tests compared with residents and attending physicians.[12] We recommend educational initiatives at all levels. Medical students should be taught principles of high-value care, and attending physicians should role-model high-value practices for residents. At the University of Virginia School of Medicine, we have developed a case-based introduction to laboratory medicine curriculum for first- and second-year medical students and are using the SOAP-V (Subjective, Objective, Assessment, Plan, Value) framework as an assessment tool for Entrustable Professional Activity 3 (EPA3) on clerkship.[13] The High Value Practice Academic Alliance (http://hvpaa.org/) provides a number of resources to guide teaching in addition to the Choosing Wisely guidelines.

Formulary. Many institutions have developed and implemented a laboratory formulary committee to eliminate unnecessary laboratory testing.[14] Laboratory formularies are typically modeled after the Pharmacy and Therapeutics committee (P&T committee), which is often tasked with determining whether a specific medication or treatment should be added to the pharmacy formulary. Similar to the P&T committee, laboratory formularies carefully review diagnostic laboratory tests offered and supportive literature to determine the appropriateness or inappropriateness in their respective patient populations.

The University of Rochester has demonstrated great success with their formulary, which relies on a tiered testing approach (see table below).

In the first year of implementation of this approach, the University of Rochester was able to reduce send-out test volumes with initial savings of approximately half a million dollars.[14] Other institutions have adopted a

Tiered Test Features of the University of Rochester's Laboratory Formulary

	Tier 1	Tier 2	Tier 3
Description	Common tests with clear and well-proven clinical utility	More specialized tests with narrow clinical indication	Unclear, controversial tests, poorly proven or with very limited clinical indications
Ordering Requirements	No restrictions	Restricted to subspecialties	Restricted to subspecialties; Laboratory Diagnostic Committee approval required
Examples	Cystic fibrosis 32 mutations; thyroid-stimulating hormone receptor antibody	1,25-Dihydroxy Vitamin D; fecal pancreatic elastase	Hepatitis C recombinant immunoblot assay; β-Thalassemia gene

Source: Zhang, Y. Victoria, Bruce R. Smoller, and Paul C. Levy. "Laboratory formulary: a model for high-value evidence-based medicine." Clinical Chemistry *63.7 (2017): 1299-1300.*

similar tiered approach, including Vanderbilt University Medical Center, which has reported institutional savings of more than $1 million.[15] The success of these two programs relied heavily on institutional buy-in as well as a widespread membership that includes representatives from inside and outside the laboratory.

"Institutional buy-in is vital to transitioning from a culture of waste to one of high-value care."

Summary

The most effective interventions for reducing repetitive laboratory testing are multimodal, combining a number of the approaches described above in an effort to produce meaningful and sustainable advances.[4] Targeting academic institutions for improvement interventions would seem to be the most effective and efficient means of effecting change, but all institutions should seek high-level buy-in to begin or enhance improvement initiatives specific to their own needs. We hope clinicians and laboratorians will find the above approaches useful in these endeavors.

References

1. The Healthcare Imperative: Lowering Costs and Improving Outcomes—workshop series summary. The National Academies of Science Engineering Medicine. http://iom.nationalacademies.org/Reports/2011/The-Healthcare-Imperative-Lowering-Costs-and-Improving-Outcomes.aspx?_ga=1.219462233.1572788654.1438188089.
2. Salisbury, Adam C., *et al.* "Diagnostic blood loss from phlebotomy and hospital-acquired anemia during acute myocardial infarction." *Archives of Internal Medicine* 171.18 (2011): 1646-1653.
3. Fetter, John E., and JS Blumenthal-Barby. "Cascade effects in critical care medicine: a call for practice changes." *American Journal of Respiratory and Critical Care Medicine* 188.12 (2013): 1384-1385.
4. Eaton, Kevin P., *et al.* "Evidence-based guidelines to eliminate repetitive laboratory testing." *JAMA Internal Medicine* 177.12 (2017): 1833-1839.
5. Corson, Adam H., *et al.* "A multifaceted hospitalist quality improvement intervention: decreased frequency of common labs." *Journal of Hospital Medicine* 10.6 (2015): 390-395.
6. Lee, Vivian S., *et al.* "Implementation of a value-driven outcomes program to identify high variability in clinical costs and outcomes and association with reduced cost and improved quality." *JAMA* 316.10 (2016): 1061-1072.
7. Dickerson, Jane A., *et al.* "Transforming laboratory utilization review into laboratory stewardship: guidelines by the PLUGS® National Committee for Laboratory Stewardship." *The Journal of Applied Laboratory Medicine* (2017): JALM-2017.
8. Iams, Wade, *et al.* "A multidisciplinary housestaff-led initiative to safely reduce daily laboratory testing." *Academic Medicine* 91.6 (2016): 813-820.
9. Delvaux, Nicolas, *et al.* "The effects of computerized clinical decision support systems on laboratory test ordering: a systematic review." *Archives of Pathology & Laboratory Medicine* 141.4 (2017): 585-595.
10. Miyakis, Spiros, *et al.* "Factors contributing to inappropriate ordering of tests in an academic medical department and the effect of an educational feedback strategy." *Postgraduate Medical Journal* 82.974 (2006): 823-829.
11. Allison, Jeroan J., *et al.* "The association of physician attitudes about uncertainty and risk taking with resource use in a Medicare HMO." *Medical Decision Making* 18.3 (1998): 320-329.
12. Gonzales, Ralph, Christy Boscardin, and Andrew Auerbach. "Communicating context in quality improvement reports." *JAMA Internal Medicine* 177.6 (2017): 817-818.
13. Moser, Eileen M., et al. "SOAP-V: Introducing a method to empower medical students to be change agents in bending the cost curve." *Journal of Hospital Medicine* 11.3 (2016): 217-220.
14. Zhang, Y. Victoria, Bruce R. Smoller, and Paul C. Levy. "Laboratory formulary: a model for high-value evidence-based medicine." *Clinical Chemistry* 63.7 (2017): 1299-1300.
15. Zutter, Mary, Julie Field, and Gordon Bernard. "Improving care and cutting costs: Implementation of a laboratory formulary to facilitate better laboratory ordering practices." *NEJM Catalyst.* 2017.

***Andrew S. Parsons**, MD, MPH, is an assistant professor of medicine and hospitalist at the University of Virginia (UVA) Health System in Charlottesville, Virginia. He directs the clinical skills course for the UVA School of Medicine and is assistant program director for the Internal Medicine Residency Program. He currently leads a task force committed to implementation of high-value care concepts using EPA3 across the four-year medical school curriculum. He is Director of Continuing Education for HVPAA.*

***Joesph R. Wiencek**, PhD, is associate director of clinical chemistry at the University of Virginia (UVA) Health System in Charlottesville, Virginia, and an assistant professor of pathology at UVA School of Medicine. He currently serves as the co-chair of the Laboratory Stewardship Committee at UVA.*

Coverage of NGS Cancer Testing Revisited

CMS HAS REOPENED ITS DECISION ON NEXT-GENERATION SEQUENCING IN PATIENTS WITH ADVANCED CANCERS

by Kimberly Scott

The Centers for Medicare and Medicaid Services (CMS) has reopened its controversial national coverage decision (NCD) on the use of next-generation sequencing (NGS) in patients with advanced cancers, following pushback from healthcare groups concerned that the NCD would have a negative impact on certain patients.

Tamara Syrek Jensen, CMS director of the Coverage and Analysis Group, announced the decision to reopen the NCD at the annual meeting of the American Clinical Laboratory Association (ACLA) in March, saying that CMS is sensitive to the concerns of its stakeholders regarding the interpretation of this policy. CMS officially reopened the NCD on April 29, saying it would accept comments until May 29.

CMS published its decision memo on NGS in advanced cancers in March 2018. Under the NCD, any diagnostic test using NGS that is approved or cleared by the Food and Drug Administration (FDA) as a companion diagnostic for patients who meet the criteria for recurrent, relapsed, refractory, metastatic, and advanced stage III or IV cancer would be covered nationally.

While the NCD was originally requested for a somatic-based test, CMS instructed Medicare Administrative Contractors (MACs) to apply the terms of the NCD to both somatic and germline NGS-based testing for patients with cancer. In a Feb. 1 letter sent to CMS administrator Seema Verma, a number of healthcare groups argued that this interpretation will restrict patients' access to medically necessary testing of germline mutations. The letter was signed by ACLA, the College of American Pathologists, the Association for Molecular Pathology, and 60 more organizations.

NGS testing is most often performed to determine germline mutations in BRCA1/2 and gauge the risk of breast and ovarian cancer. Medicare contractors historically have covered such tests for patients who already had cancer, but didn't limit testing depending on the cancer stage. MACs previously have implemented local coverage determinations (LCDs) that provide coverage for germline testing of cancer when supported by clinical guidelines, including NGS-based tests for germline mutations for breast and colon cancers, the groups note in the letter.

This NCD, they say, will supersede existing LCDs that provide coverage for NGS-based testing for hereditary breast and ovarian cancer syndromes and Lynch syndrome in patients who do not have advanced cancer.

> "The implication of this interpretation is that both germline and somatic NGS-based testing will become non-covered for Medicare beneficiaries with early-stage cancer."

"The implication of this interpretation is that both germline and somatic NGS-based testing will become non-covered for Medicare beneficiaries with early-stage cancer," the groups write. "Our organizations believe that the inclusion of NGS-based testing for germline mutations represents significant policy overreach by CMS that will have unintended consequences on the care delivered to Medicare beneficiaries, particularly those who may have a genetic predisposition to cancer based on a family history or other relevant criteria."

Evolution of the NCD

Since receiving the letter, CMS has issued a new transmittal (No. 214, Change Request 10878) on the NCD that provides a new implementation date of April 8, 2019, and acknowledged stakeholder concerns regarding the interpretation of the policy. CMS intends to continue with implementation of the policy even as the decision is reopened.

AMP called for CMS to revise its current interpretation of the NCD by limiting it to somatic tumor testing, and requested that this be achieved via communication to the MACs and not through a formal reconsideration process, says Samuel Caughron, MD, FCAP, chair of AMP's Economic Affairs Committee and lab medical director and chair of pathology at AdventHealth Shawnee Mission (Kansas City).

"We remain committed to working with CMS, local MACs, and other stakeholders to preserve broad patient access to all of the thousands of clinically- and analytically-validated NGS-based testing for cancer and other conditions that benefit patients each and every day," he says.

In reopening the NCD, it appears that CMS would like to narrow the policy to focus just on somatic and germline testing when done in advanced cancer patients, noting that other assessments are not within the scope of the coverage terms. "Specifically, we are only reconsidering the evidence available for tests of germline mutations to identify those with hereditary cancer who may benefit from targeted treatments based on results of the test; all other tests are beyond the scope of this investigation," the agency said.

While AMP is encouraged by CMS's intent to thoroughly engage with the laboratory community and other stakeholders during the open comment period, Caughron says the association is concerned that during the reconsideration process, testing utilizing older methods will be the only covered option for early-stage cancer patients who need germline NGS-based testing.

"AMP would like to see this particular issue addressed more urgently because this arrangement is suboptimal for patient care and unnecessarily more difficult and costly for laboratories," says Caughron.

A request for parallel review

The NCD is the result of a request by Foundation Medicine for a parallel review from the FDA and CMS for FoundationOne CDx, a test that gauges somatic variants in 324 genes for tumor tissue samples and guides treatment decisions based on the results. In the NCD, CMS not only approved coverage from that test but also for other similar FDA-approved NGS companion diagnostics.

Since 2010, the FDA-CMS Parallel Review Program has been a collaborative effort intended to reduce the time between FDA marketing approval or clearance and a CMS national coverage determination. Typically, CMS does not engage with manufacturers until after FDA approval. If the manufacturers engage FDA and CMS together while under FDA review, a stronger evidentiary base could be developed in a more efficient manner, accelerating patient access to innovative medical devices, according to CMS.

Since approval of FoundationOne's CDx test, there have been three other NGS genomic oncology panel tests for advanced cancers approved by the FDA: FoundationFocus CDxBRCA (Foundation Medicine), Oncomine Dx Target Test (Thermo Fisher Scientific), and Praxis Extended RAS Panel (Illumina). In addition, the FDA has granted marketing authorization for MSK-IMPACT (Memorial Sloan Kettering Cancer Center).

Kimberly Scott *is a freelance writer specializing in healthcare and medical diagnostics.*

Emerging Applications of Clinical Mass Spectrometry

MS MAKES THE JOURNEY FROM LABORATORY TESTS THROUGH HISTOLOGICAL SAMPLES TO THE SURGICAL SUITE

by Masha G. Savelieff, PhD

Only a decade ago, a mass spectrometer may have looked out of place in a clinical laboratory. The technology, once mostly confined to chemical research facilities, debuted on the clinical scene around ten years ago with the release of commercial FDA-cleared microbial identification mass spectrometry (MS) platforms. But even before that, MS was recognized as a more specific alternative to immunoassays, with fewer false positives, in illicit drug testing. Since then, the breadth of analytes reliably evaluated by MS in laboratory tests has increased tremendously, ranging from small molecules and metal analytes to biomolecules. Applications abound in identification of infectious diseases, inborn errors of metabolism, and cancer diagnostics.

For clinical applications, the most frequently performed MS techniques are gas chromatography MS (GC-MS), liquid chromatography MS (LC-MS), tandem MS (MS/MS, GC-MS/MS, LC-MS/MS), and matrix-assisted laser desorption/ionization time of flight MS (MALDI-TOF MS).

As a clinical tool, MS boasts high analytical specificity and sensitivity, multiplexing capability, and low sample cost.[1] Runs can be staggered into the machine to maximize instrument time. MS is also versatile since protocols can be developed for a new biomarker without waiting for an FDA-approved kit or assay.

Despite its benefits, there are some hurdles to widespread adoption of MS in clinical labs. The instruments come with high price tags. Skilled operators are required to develop and validate protocols for applications that lack an FDA-cleared, end user–friendly MS platform. MS is also labor intensive, since the process is not fully automated. The regulatory landscape for the majority of clinical applications is still uncertain; laboratory-developed tests, (i.e., protocols developed in-house), will eventually need to conform to FDA guidelines, but when the final guidelines will be released is at present unclear.

Analyte or biomarker quantification in laboratory tests

The high sensitivity and specificity achievable by MS make it superior to immunoassays for analysis of several drug types, (e.g., steroids, illicit drugs, abused prescription drugs, and therapeutic prescription drugs). For analysis of immunosuppressant prescription drugs, such as tacrolimus, immunoassays fail to yield accurate results due to cross-reactivity with patient autoantibodies.[2] Accurate quantification of tacrolimus is essential as too-low levels run the risk of tissue rejection while high levels run the risk of overdose. In some cases, MS also outstrips immunoassays in the determination of levels of serum thyroglobulin (Tg),[3] a thyroid glycoprotein and biomarker for recurrent thyroid cancer. The immunoassay is unreliable in patients who have serum antithyroglobulin autoantibodies or heterophile antibodies because of cross-reactivity with the Tg immunoassay antibody, which leads to false-positive test results. In such instances, LC-MS/MS is more specific than a Tg immunoassay. Assays for proteins, including monoclonal antibodies, are also emerging, but are still in the early stages of development. Finally, inductively coupled mass spectrometry can be employed to analyze metal content in biosamples, (e.g., blood, plasma, and urine, to evaluate acute or chronic metal exposure).

Infectious disease

The most renowned application for clinical MS is microbial identification by MALDI-TOF MS. This technique has revolutionized microbiology laboratories since the introduction of FDA-cleared instruments for diagnostic identification of patient infections caused by bacteria and yeasts. The process begins by culturing patient-derived biosamples and selecting a microbial colony for evaluation by the MALDI-TOF MS. The spectrum produced by analysis of the microbial colony is matched to the

CyBio® FeliX

Accuracy. Precision. Sample Integrity.

Your Automated Liquid Handling Application Starts Here.

CyBio® FeliX

Providing automated solutions for sample preparation, drug testing, toxicology, NGS, immuonoassays, PCR & more. Optimize your work processes with CyBio FeliX, a liquid handling system adaptable to your requirements.

- Most compact liquid handling platform for any lab bench
- Customer swappable pipetting heads with automatic tip and adapter loading
- 12 deck positions on two levels for microplates, tubes, reservoirs, tips and accessories on a small footprint of 650 x 450 mm
- Precise high parallel 96 or 384 channel pipetting combinable with single channel transfers and pipetting in rows and columns within the unique volume range of 1 – 1000 µl

CyBio FeliX - Accelerating Your Research

For more information:
www.analytik-jena.com
info@us.analytik-jena.com

ASM Microbe | San Francisco, CA | **Booth 636**
ISSCR | Los Angeles, CA | **Booth 722**
AACC | Anaheim, CA | **Booth 1952**

analytikjena
An Endress+Hauser Company

spectrum of a known organism in an approved, FDA-cleared database. At present, analysis of mycobacteria and filamentous fungi is still challenging and not yet approved for clinical diagnostics by the FDA, but research is ongoing. Rare and exotic microbes can be analyzed against a custom-built, research-use-only database, but that process has not received regulatory approval as a diagnostic tool. Additional novel clinical applications on the horizon include antimicrobial susceptibility testing, which would identify the ideal antibiotic treatment, and direct MALDI-TOF MS analysis from blood culture and biofilms, which could substantially shorten the time to identification.

Inborn errors of metabolism

A very early clinical application of MS was in the analysis of acylcarnitine for identifying newborns with inborn errors of fatty acid oxidation.[4] Defects in twenty-five enzymes of this metabolic pathway leads to eighteen conditions associated with human disease, the most frequent of which is medium-chain acyl-CoA dehydrogenase deficiency. Additionally, MS can be used to screen neonates with cystinosis, glutaric aciduria type I, and lysosomal disorders (LSDs). LSDs encompass a broad spectrum of diseases, including Fabry disease; Gaucher disease; Krabbe disease; Mucopolysaccharidoses types I and II; Niemann-Pick disease types A, B, and C; and Pompe disease. In these applications, MS, generally GC- or LC-MS/MS, is applied in a targeted manner to identify key biomarker metabolites associated with the conditions. Alternatively, metabolomics, a systems biology approach that considers the entire metabolome, could be used to identify aberrant metabolic patterns that may be linked to defects in certain biochemical pathways and hence to deactivation of specific genes. Though still in the nascent stages of development, the potential applications of metabolomics for inborn errors of metabolism are vast.

Cancer diagnostics and surgery

MALDI imaging mass spectrometry (MALDI IMS) couples the sensitivity of MS with spatial information. A raster of the MALDI laser beam collects MS data at sampling spots evenly spaced across a matrix-embedded tissue specimen, which are combined to reconstruct a spatial MS map of the histological sample. MALDI IMS has been used predominantly in the analysis of tumor tissue pathology, either in a diagnostic capacity, (i.e., to distinguish tumor from healthy tissue, or in a prognostic capacity, i.e., to identify patients who will benefit from a certain treatment or to predict their survival). Although not yet available in an FDA-cleared platform, the technology is rapidly advancing and could be amenable to automated, user-friendly protocols that would translate well to the clinic.

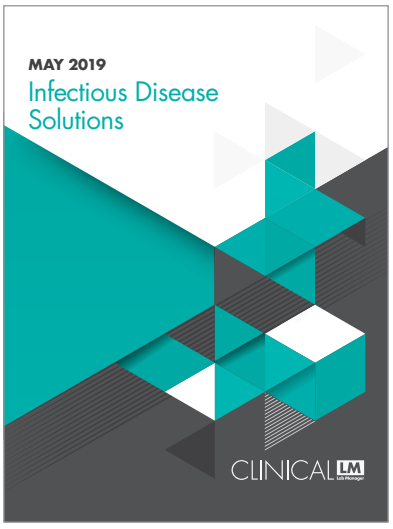
Another MS technique even promises to deliver real-time monitoring of tumor tissue during surgical resection, aiding the surgeon in complete tumor removal. The invention, called the intelligent knife or iKnife,[5] uses rapid evaporative ionization MS to ablate tumor tissue into a mass spectrometer. An algorithm developed using a training set of cancerous and normal tissues allows the iKnife to differentiate tumor from healthy tissue, defining the margin along the tumor mass.

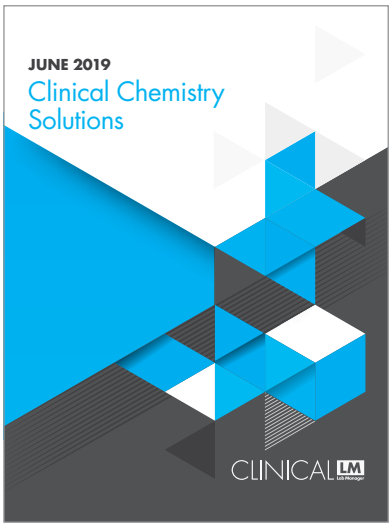
Like many clinical tools, MS is also set to join the 'omics revolution through proteomics, *e.g.*, in patient plasma, metabolomics, and lipidomics analysis and metabolomics flux studies, both for diagnostics and biomarker discovery. Capillary electrophoresis interfaced with MS can offer better analyte separation and hence resolution for biomarker identification. Other technological advances include movement toward miniaturization of MS instruments, which could eventually be applied to diagnostic point-of-care tests.

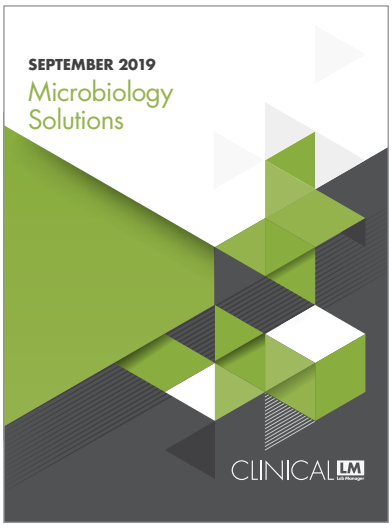
References:

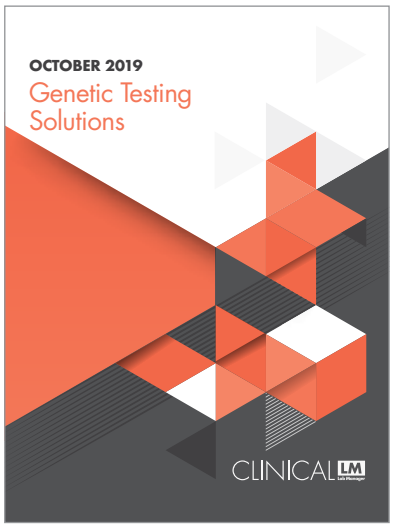
1. Jannetto, Paul J., and Robert L. Fitzgerald. "Effective use of mass spectrometry in the clinical laboratory." *Clinical Chemistry* 62.1 (2016): 92-98.
2. Rostaing, Lionel, *et al.* "Falsely elevated whole–blood tacrolimus concentrations in a kidney–transplant patient: potential hazards." *Transplant International* 23.2 (2010): 227-230.
3. Clarke, Nigel J., Yanni Zhang, and Richard E. Reitz. "A novel mass spectrometry–based assay for the accurate measurement of thyroglobulin from patient samples containing antithyroglobulin autoantibodies." *Journal of Investigative Medicine* 60.8 (2012): 1157-1163.
4. Millington, D.S., *et al.* "Tandem mass spectrometry: a new method for acylcarnitine profiling with potential for neonatal screening for inborn errors of metabolism." *Journal of Inherited Metabolic Disease* 13.3 (1990): 321-324.
5. Balog, Júlia, *et al.* "Intraoperative tissue identification using rapid evaporative ionization mass spectrometry." *Science Translational Medicine* 5.194 (2013): 321-324.

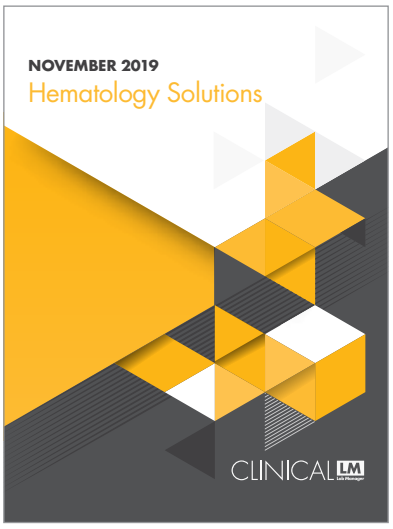
***Masha G. Savelieff**, PhD, is a full-time science writer and co-founder of SciGency Science Communications. She obtained her PhD in chemistry at the University of Illinois at Urbana-Champaign, and performed research in medicinal chemistry, Alzheimer's disease, and cancer biology before becoming a full-time writer.*

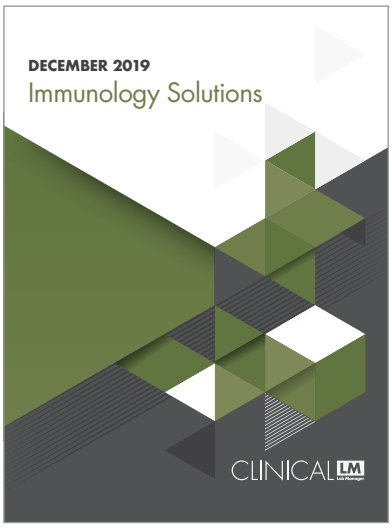












SOLUTIONS eBOOKS

Read about these hot topics in 2019

Visit ClinicalLabManager.com

CLINICAL LM Lab Manager

CLINICAL MASS **SPECTROMETRY** MILESTONES

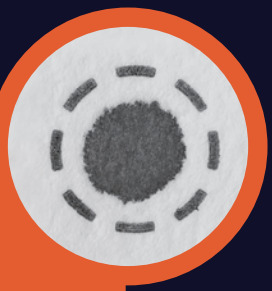
Francis Aston develops the first mass spectrograph that is able to accurately determine the masses of individual atoms. The measurements supported the existence of isotopes in nonradioactive elements and led the pathway toward modern-day mass spectrometry techniques.

The Nimitz accident, a military jet crash, kills 14 service members. Immunoassays revealed the presence of marijuana in members of the crew. At a time when immunoassays had a high rate of false positives, this high-profile case sparked the need for mandatory confirmatory studies in clinical toxicology laboratories.

Gas chromatography-mass spectrometry (GC-MS) becomes the gold standard for confirming drug screens across federal and state agencies, workplaces, and other institutions. The large number of inaccuracies and insensitivity associated with immunoassay and thin-layer chromatography led these tests to be considered preliminary until confirmed by GC-MS.

Millington *et al.*[1] propose tandem MS of dried blood spots for newborn screening. The method enabled screening for multiple organic acid and fatty acid oxidation disorders within a single test. Combining this technology with electrospra ionization (ESI) allowed for the detection of even more disorders with a single test.

1920 1940 **1960** 1980 **1985** 1990 **1995**



**ntroduction
f a multiplex
lectrospray source
hat interfaces with
MS leads to higher
ample throughput
nd cost savings.**
Used in conjunction
with ultra-high
ow rate liquid
hromatography-mass
pectrometry (LC-
MS), the technology
nade rapid and
arallel determination
f pharmaceuticals
resent in plasma
chievable, with
hroughputs reaching
20 samples per hour.

A nonprofit organization committed to the advancement of MS in the clinical laboratory called Mass Spectrometry: Applications to the Clinical Laboratory is incorporated.
The organization began holding an annual conference to provide education and training on MS in the laboratory setting to leaders and practitioners in the field.

MS is increasingly used in metabolomics—the analysis of cellular metabolites during gene alterations or physiological stimuli.
Its use enabled the capture of either a subset of targeted biological molecules or thousands of molecules via an untargeted approach. MS also started to gain traction in lipidomic, proteomic, and other -omics clinical applications.

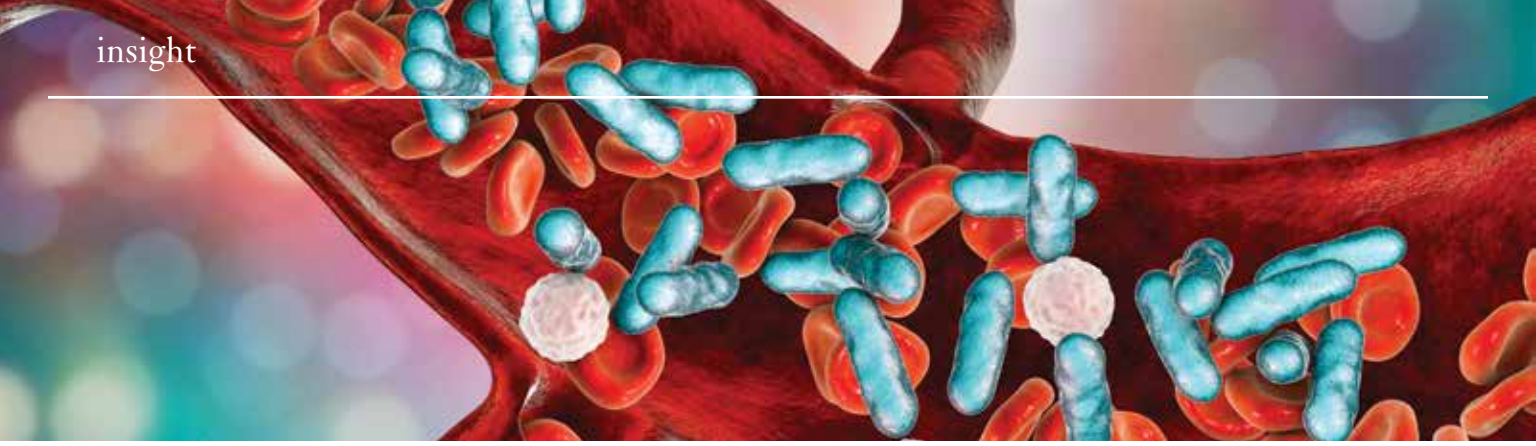
The intelligent knife (iKnife) is used in cancer surgery.
Rapid evaporative ionization mass spectrometry (REIMS) combined with electrosurgery-enabled real-time evaluation of surgical tissue using lipidomic profiles that highly correlate with histopathological analysis.

The Nobel Prize in chemistry goes to John B. Fenn and Koichi Tanaka for the development of ESI and "soft" ionization techniques (shared with Kurt Wüthrich for development of NMR).
The research (conducted in the 1980s) allowed liquid chromatography-tandem mass spectrometry (LC-MS/MS) to simplify MS techniques by eliminating the need for volatile analytes and preliminary sample preparation and shifted clinical applications away from GC-MS.

Matrix-assisted laser desorption ionization-time of flight (MALDI-TOF) MS is successfully cleared by the FDA for microorganism identification.
MALDI-TOF revolutionized the field by providing rapid and robust results. It displaced more time-intensive and expensive methods such as standard culture techniques and susceptibility testing.

2000 **2003** 2006 **2009** 2012 **2015**

1. Millington DS, Kodo N, Norwood DL, Roe CR. "Tandem mass spectrometry: a new method for acylcarnitine profiling with potential for neonatal screening for inborn errors of metabolism." *Journal of Inherited Metabolic Disease* 13(1990): 321-324.



Speeding Up Sepsis Detection

HOW POINT-OF-CARE DEVICES ARE IMPROVING SEPSIS DIAGNOSIS AND PATIENT OUTCOMES

by Raeesa Gupte, PhD

When it comes to sepsis, time is of the essence. Research has shown that for each hour that antibiotic administration to sepsis patients is delayed, there is a linear increase in the risk of mortality.[1]

Approximately 1.7 million individuals in the US develop sepsis each year, yet no gold standard exists for the diagnosis of sepsis in a clinical setting. Identification and treatment are determined based on a combination of tests and the clinician's judgment. Although the field is rife with disagreements on the diagnostic criteria of sepsis, experts agree that early diagnosis is critical.

"Approximately 1.7 million individuals in the US develop sepsis each year, yet no gold standard exists for the diagnosis of sepsis in a clinical setting."

What is sepsis?

The Greek physician Hippocrates used the word "sepidon" circa 400 BC to describe the repugnant process of biological decay. It was not until the 19th century that infectious microorganisms were identified as the cause of that biological decay. Although the term "sepsis" has been used in the medical literature since the late 1800s, physicians still struggle to define it.

The definition of sepsis has been revised several times in recent years. In 1991, Sepsis-1 was defined as an infection leading to the onset of systemic inflammatory response syndrome (SIRS). By this definition, patients were diagnosed with sepsis if they met two of the four SIRS criteria: high heart rate, elevated rate of respiration, hyperthermia, and abnormal white blood cell counts. Sepsis-2, which came into use in 2002, retained the original Sepsis-1 definition but expanded the diagnostic criteria to include altered mental status, elevated plasma levels of C-reactive protein (CRP) and procalcitonin (PCT) as markers of inflammation, hemodynamic parameters including hypotension and oxygen saturation, lactate levels as a marker of tissue perfusion, and organ dysfunction.

The Sepsis-3 definition, coined in 2016, classifies sepsis as "life-threatening organ dysfunction due to a dysregulated host response to infection."[2] In addition, septic shock is defined as a subset of sepsis with circulatory and metabolic dysfunction associated with a greater risk of mortality than sepsis alone. Concerns have been raised about the clinical value of this new definition.[3]

The considerable lack of consensus over the definition of sepsis is one of many reasons its diagnosis is fraught with challenges.

Challenges in sepsis diagnosis

Diagnosing sepsis is challenging primarily because it is a heterogeneous condition with different etiologies ranging from pneumonia and urinary tract infection to wound infections.

In a clinical setting, sepsis is usually diagnosed on the basis of an infection and the host's response to it.

EARLY SEPSIS INDICATOR

KNOW SOONER. ACT FASTER.

First-in-class FDA-cleared hematology biomarker for sepsis

- Augment suspicion of sepsis or risk of sepsis in the first 12 hours when used with current standard of care
- No impact to workflow in the emergency department or the lab
- Automatically available with CBC-diff for adult patients entering the emergency department
- Measure monocyte volume variability in response to bacteria, viruses, or fungi

Learn more at:

www.beckmancoulter.com/earlysepsis

For Beckman Coulter's worldwide office locations and phone numbers, please visit www.beckmancoulter.com/contact

AD-204653

BECKMAN COULTER

SURVIVING SEPSIS

The Surviving Sepsis Campaign guidelines were updated in 2018 to include the hour-1 bundle. The revised guidelines require clinicians to initiate the following measures within an hour of patient presentation to the emergency department with sepsis:

1. Measure lactate level. Remeasure if lactate is >2 mmol/L.
2. Obtain blood cultures prior to administration of antibiotics.
3. Administer broad-spectrum antibiotics.
4. Begin fluid resuscitation or vasopressor therapy to manage hypotension.

Source: Levy, Mitchell M., Laura E. Evans, and Andrew Rhodes. "The Surviving Sepsis Campaign bundle: 2018 update." *Intensive Care Medicine* 44.6 (2018): 925-928.

Symptoms include fever and chills, tachycardia, rapid breathing, rash, and mental confusion or disorientation. Because these symptoms commonly occur in several other conditions, it is difficult to differentiate septic patients from those with a non-sepsis infection. To further complicate matters, elderly sepsis patients often have hypothermia and may not always present with a fever, whereas critically ill patients commonly have hyperthermia, tachycardia, and tachypnea that may not be indicative of sepsis.

Blood and diagnostic imaging tests, such as CT scans, are used to identify the source and location of infection in symptomatic patients. Aberrant white blood cell counts and the presence of inflammatory markers like cytokines, CRP, and PCT in blood are indicative of a host response to infection. However, these indicators may also be altered during non-infectious systemic inflammation in critically ill patients due to acute mesenteric ischemia or adrenal insufficiency. Blood or urine cultures help identify the causative microorganism and distinguish infectious SIRS from non-infectious SIRS. One issue with cultures is that time to results can be slow compared with the speed at which sepsis progresses. In addition, some patients may have negative bacterial cultures due to either prior antibiotic use or unusual viral or fungal infections.

Variable symptoms, inability to distinguish infectious versus non-infectious inflammatory responses, and delays in obtaining pathogen information are the major limitations of commonly used sepsis diagnostics.

How can point-of-care (POC) testing help?

POC testing is performed at or near the site of patient care to provide rapid diagnostic information that can inform clinical decisions and improve outcomes. Given the need for early detection and rapid intervention in sepsis management, POC testing may confer significant benefits.

To be a useful tool for sepsis diagnosis, POC testing must satisfy several requirements. Accurate sepsis diagnosis calls for the ability to distinguish an immune response to infection from an inflammatory or hemodynamic response to non-infectious disease states. The ability to monitor the host response can help stratify patients based on severity, which enables clinicians to predict which patients are more likely to deteriorate and require escalated care. Finally, the ability to obtain information about the causative pathogen allows clinicians to initiate targeted therapies early in the course of infection. Administration of broad-spectrum antibiotics in the face of rising antibiotic resistance is not a sustainable approach.

Accordingly, POC testing for sepsis aims to 1) monitor the host response for accurate diagnosis and patient stratification, and 2) acquire pathogen information for appropriate therapeutic support.

POC devices to evaluate host response

Recent guidelines endorse sustained elevation of lactate as an identifier of sepsis in the emergency department. Several devices have been approved by the US Food and Drug Administration (FDA) for POC lactate measurement. These include near-patient analyzers such as Siemens's RAPIDPoint 500, Roche's Cobas b 221, and IL's GEM Premier 5000 or portable devices such as Nova Biomedical's StatStrip Lactate and Abbott's i-STAT. In one study,[4] in-hospital bedside lactate POC testing significantly reduced test turnaround time and mortality in the emergency department; however, another study found no improvement in pre-hospital diagnostic accuracy when POC analyzers were used by paramedics.[5]

Serum levels of PCT and CRP are significantly elevated in systemic inflammation and are associated with the severity of organ dysfunction.[6] Since PCT

A RACE AGAINST TIME

A retrospective analysis of over 28,000 sepsis patients from 165 ICUs in the US, South America, and Europe found a linear increase in the risk of mortality for each hour that antibiotic administration was delayed.

Time to Antibiotics (Hours)	Probability of Mortality (%)
0-1	24.6
1-2	25.9
2-3	27.0
3-4	27.9
4-5	28.8
5-6	32.3
>6	33.1

Source: Ferrer, Ricard, *et al.* "Empiric antibiotic treatment reduces mortality in severe sepsis and septic shock from the first hour: Results from a guideline-based performance improvement program." *Critical Care Medicine* 42.8 (2014): 1749-1755.

concentrations more closely reflect patient recovery than do CRP concentrations, PCT results are also used to guide antibacterial therapy in critically ill patients. POC devices that measure the levels of these biomarkers offer the ability to rapidly determine the host response to infection, accelerating diagnosis and potentially improving patient outcomes. Several healthcare companies have received FDA approval for their PCT assays to be used on automated testing devices such as BioMérieux's VIDAS 3, Roche's Elecsys BRAHMS, Siemens's Atellica IM, DiaSorin's LIAISON, and Fujirebio's Lumipulse. In 2018, Thermo Fisher Scientific's BRAHMS PCT direct was approved in Europe for use in POC settings. In the US, PCT testing was initially cleared by the FDA to assess the risk of critically ill patients progressing to severe sepsis, but it has now been expanded to determine whether antibiotics can be safely discontinued in hospitalized sepsis patients.

Ultimately, given the diversity of immune responses to infection, one biomarker alone may be inadequate in accurately diagnosing sepsis. Consequently, Nanomix is currently performing clinical trials on a POC device that will concomitantly quantify levels of lactate, PCT, and CRP. SeptiCyte LAB is an FDA-approved diagnostic test that detects and combines the gene expression results of four host mRNA transcripts (CEACAM4, LAMP1, PLA2G7, and PLAC8) to reliably differentiate the host response to infection from inflammation. In addition, the predictive power of a microfluidics-based biochip improved when combined with lactate levels or with data from patients' electronic medical records.[7] Therefore, a POC device that is able to quantify several immune biomarkers from a small quantity of blood within a short time will vastly improve diagnosis and patient stratification.

"Given the need for early detection and rapid intervention in sepsis management, POC testing may confer significant benefits."

POC devices for microbial identification and antibiotic susceptibility

Blood cultures are currently the gold standard for pathogen identification in sepsis. However, cultures require up to five days of incubation, which often results in initiation of inappropriate treatment or prolonged use of broad-spectrum antibiotics while waiting for pathogen information. Nucleic acid amplification technologies (NAATs) that can detect pathogens from positive blood cultures or directly from whole blood can reduce pathogen identification time to 2–12 hours.

Several NAAT-based tests are able to distinguish between numerous strains of Gram-negative and Gram-positive bacteria as well as some fungi. Most of these—Roche's SeptiFast, Molzym's SepsiTest, and GenMark's ePlex—have received regulatory approval only in Europe. In the US, T2Biosystems' T2Bacteria and T2Candida are the only tests for detection of bacterial and fungal species from whole blood to receive FDA market clearance. All these assays are also capable

of detecting antibiotic resistance genes, which provides added value in informing antimicrobial therapy. However, they all require bulky benchtop equipment for analysis, limiting their use in emergency departments or at the patient's bedside. Miniaturizing detection sensors and simplifying sample processing would improve their use in POC settings.

Future directions

Currently, most sepsis diagnostic devices require access to specialized laboratory equipment, such as thermocyclers for nucleic acid amplification, when used in hospitals and emergency departments. This limits their use in low- to middle-income countries and resource-limited settings where microbial infections may be more prevalent. Several innovative systems have been developed to transform mobile phones into POC diagnostics for human cells and pathogens. Recently, a smartphone-based system reported robust bacterial identification in urine samples from sepsis patients that matched hospital diagnostics but took only a fraction of the time and costs less than $100.[8] These technologies will eventually be harnessed and streamlined for sepsis diagnosis on a wider scale.

The inherent complexity of sepsis warrants adoption of machine learning approaches that can parse several measurements of validated biomarkers, patient data, and pathogen information to accurately diagnose, stratify, and manage sepsis. Recently, an electronic triage system based on machine learning and real-time interactions with patients' electronic health records reliably identified sepsis patients in need of critical care.[9] Similarly, AI Clinician, a computational model using reinforcement learning, is able to suggest individualized treatment strategies that improve outcomes in sepsis patients. Although these technologies require further validation, preliminary findings suggest that integrating POC testing and predictive analytics into clinical workflows may further improve sepsis diagnosis and outcomes.

SEPSIS STATISTICS

In the US…

Approximately **1.7** million individuals develop sepsis each year.

Nearly **270,000** individuals die as a result of sepsis each year.

Sepsis is responsible for **1** out of every **3** hospital deaths.

Source: CDC Sepsis Fact Sheet: https://www.cdc.gov/sepsis/datareports/

References:

1. Ferrer, Ricard, *et al.* "Empiric antibiotic treatment reduces mortality in severe sepsis and septic shock from the first hour: Results from a guideline-based performance improvement program." *Critical Care Medicine* 42.8 (2014): 1749-1755.
2. Singer, Mervyn, *et al.* "The third international consensus definitions for sepsis and septic shock (Sepsis-3)." *JAMA* 315.8 (2016): 801-810.
3. Sartelli, Massimo, *et al.* "Raising concerns about the Sepsis-3 definitions." *World Journal of Emergency Surgery* 13.1 (2018): 6.
4. Singer, Adam J., *et al.* "ED bedside point-of-care lactate in patients with suspected sepsis is associated with reduced time to iv fluids and mortality." *The American Journal of Emergency Medicine* 32.9 (2014): 1120-1124.
5. Boland, Lori L., *et al.* "Prehospital lactate measurement by emergency medical services in patients meeting sepsis criteria." *Western Journal of Emergency Medicine* 17.5 (2016): 648.
6. Castelli, Gian Paolo, *et al.* "Procalcitonin and C-reactive protein during systemic inflammatory response syndrome, sepsis and organ dysfunction." *Critical Care* 8.4 (2004): R234.
7. Hassan, U., *et al.* "A point-of-care microfluidic biochip for quantification of CD64 expression from whole blood for sepsis stratification." *Nature Communications* 8 (2017): 15949.
8. Barnes, Lucien, *et al.* "Smartphone-based pathogen diagnosis in urinary sepsis patients." *EBioMedicine* 36 (2018): 73-82.
9. Levin, Scott, *et al.* "Machine-learning-based electronic triage more accurately differentiates patients with respect to clinical outcomes compared with the emergency severity index." *Annals of Emergency Medicine* 71.5 (2018): 565-574.

***Raeesa Gupte**, PhD, is a freelance science writer and editor specializing in evidence-based medicine, neurological disorders, and diagnostics.*

Lab results giving you a headache?

Have you trained and retrained but are still struggling with hemolysis, QNS, and clotting?

Are redraws "normal" from some collection areas or departments?

Maybe it's time to stop blaming people and practices and find a better product!

If you're ready to get serious about improving sample quality, let SARSTEDT be your partner in the lab.

Visit us at the AACC Clinical Lab Expo in booth #3123 for:

- Our in-booth session on specimen quality, "The numbers don't lie"
- Specimen collection solutions that eliminate sample transfers from untreated syringes, reduce hemolysis and clotting rates, and offer ultra-low volumes for better blood management

Improvement doesn't happen by chance...

It happens by change

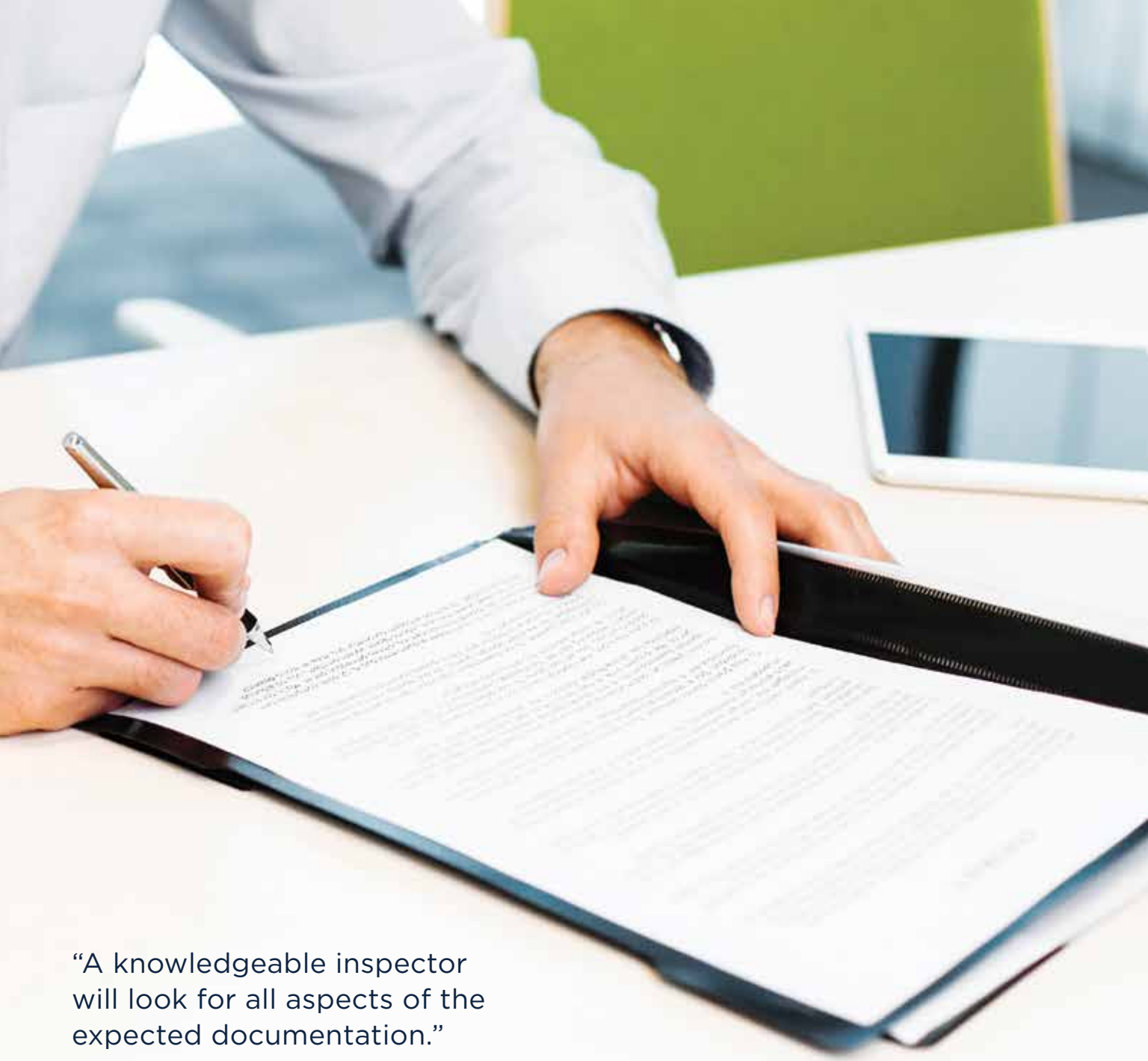
SARSTEDT

www.sarstedt.com • sales.us@sarstedt.com • 800-257-5101

Instrument Qualification in Today's Laboratory

BEST PRACTICES FOR IQ/OQ/PQ IN THE CLINICAL LAB

by Denise Bland, MHA



"A knowledgeable inspector will look for all aspects of the expected documentation."

Instrument reliability is at the heart of accurate laboratory testing. Without the quality assurance of installation qualification/operational qualification/performance qualification (IQ/OQ/PQ), laboratory testing would be riddled with inconsistent results. Accrediting agencies such as the College of American Pathologists (CAP) and the Joint Commission require IQ/OQ/PQ documentation on all new instrumentation.

At the histopathology lab where I work, IQ/OQ/PQ records have been requested and reviewed near the start of my last three inspections. A knowledgeable inspector will look for all aspects of the expected documentation, which will be compared to your department's standard operating procedure for new equipment, test, or method validation. Being prepared is crucial, and it all starts with following a set of best practices.

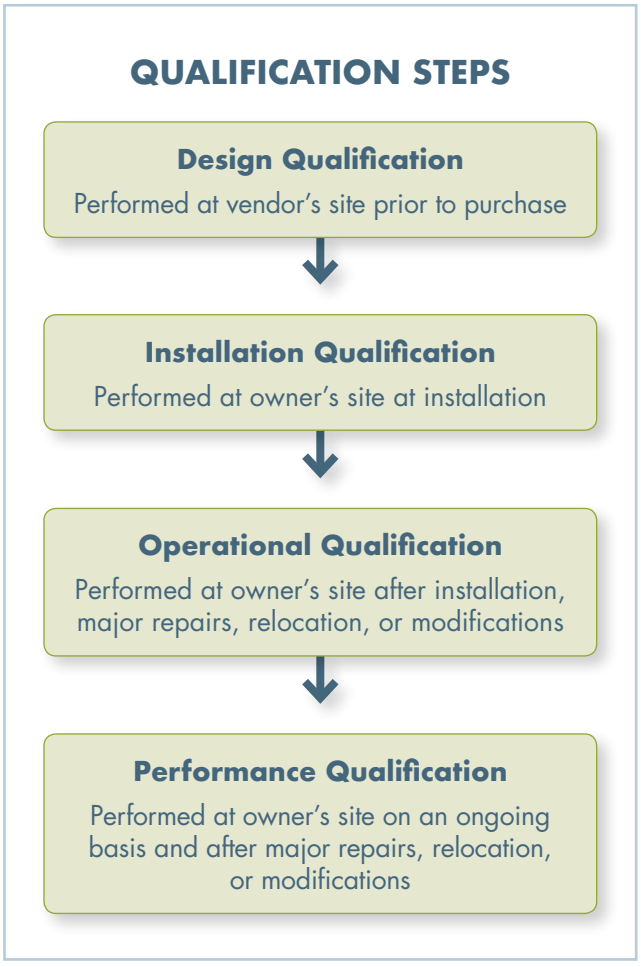
Installation qualification

Installation qualification (IQ) is the documented collection of activities necessary to establish that an instrument is delivered as designed and specified, is properly installed in the selected environment, and that this environment is suitable for the instrument. IQ applies to an instrument that is new or pre-owned and to any instrument that exists

on site but has not been previously qualified. Certain IQ documentation would also apply to a qualified instrument that has been transported to another location or is being reinstalled for other reasons, such as prolonged storage. This documentation should include system components; instrument delivery, assembly and installation; and network and data storage. For installation verification, the vendor or internal qualified personnel performs the initial diagnostics and testing of the instrument after installation.

Operational qualification

After a successful IQ, the instrument is ready for operational qualification (OQ). OQ is the documented collection of activities necessary to demonstrate that an instrument will function according to its operational specifications in the selected environment. This documentation includes secure data storage, backup and archiving, and functional tests. The vendor or user must perform this qualification in the user's environment.



Performance qualification

Performance qualification (PQ) is the documented collection of activities necessary to demonstrate that an instrument consistently performs according to the specifications defined by the user and is appropriate for the intended use. It requires performance checks to be made through a series of tests. Those performing PQ should not choose strictly routine test material, as any minor variabilities will have increased visibility on a rare tissue or fluid type. Natural language searches in a laboratory information system (LIS) can be a useful source to guide appropriate sampling. Users should perform a minimum of 20 tests for positive and negative cases. Validation of the instrument must correlate with the manual or automated method previously used; therefore, the medical director should correlate the results with previous testing on the previous platform when completing required sign-off to document the approval of the performance qualification. When the instrument undergoes major repairs, relocation, or modifications, appropriate OQ and/or PQ tests should be repeated.

> "When the instrument undergoes major repairs, relocation, or modifications, appropriate OQ and/or PQ tests should be repeated. "

Common qualification pitfalls

One major pitfall to qualification can be vendors, as they do not always realize their responsibility for IQ/OQ documentation. If vendors are not aware of or up to date on qualification requirements, their instrument requirements may not meet laboratory needs.

In addition, there are several potential computer-related qualification pitfalls, which users can avoid by educating themselves. It's important to understand the difference between firmware and stand-alone software. Computerized analytical instruments have operating software, called firmware, without which they cannot function properly. Generally, firmware cannot be altered by users and is considered part of the instrument. The firmware version used while testing, and any later changes or upgrades, should be recorded; however, on-site qualification for firmware is not needed. Nonetheless, tests should still be validated under any significant change to the platform by repeating OQ/PQ as appropriate.

GLP Regulations on Equipment

The digital age is enhancing the need to understand multiple aspects of qualification. Good Laboratory Practice (GLP) regulations are appropriate resources to provide guidance on equipment design, calibration, and maintenance. To audit vendor accountability, it is crucial to understand proper equipment design parameters. The following are equipment-related US GLP regulations (see 21 CFR part 58):

- Automatic, mechanical, or electronic equipment used in the generation, measurement, or assessment of data shall be of appropriate design and adequate capacity to function according to the protocol and shall be suitably located for operation, inspection, cleaning, and maintenance.
- Equipment used for generation, measurement, or assessment of data shall be adequately tested, calibrated, and/or standardized.
- Written standard operating procedures shall set forth in sufficient detail the methods, materials, and schedules to be used in routine inspection, cleaning, maintenance, testing, calibration, and/or standardization of equipment and shall specify remedial action to be taken in the event of failure or malfunction of equipment.
- Written records shall be maintained on all inspection operations.

Many computerized instruments are operated via software, by connecting them to a computer. Design qualification (DQ) should be performed on the software along with validation at the vendor's site, and the vendor should provide the user with a validation summary. For on-site validation, holistic validation, which includes the hardware and software, is typically more efficient and may be part of operational qualification. Stand-alone software is separate from software supplied by the vendor; analysts often use stand-alone software to operate analytical instruments or process recorded data. User manuals are typically supplied by the software developer, who also administers the validation process and specifies the appropriate development model for the software.

The vendor should provide the end user with the following computer-related information: system description; scope; assumptions; hardware requirements; software requirements; functional requirements; operational requirements; interface requirements; report requirements; data requirements, including integrity, utility, and environmental requirements; security requirements; and regulatory requirements associated with significant changes.

Document, document, document

When in doubt, document, document, document. Normalization of clinical testing is key for optimal repetition of accurate test results. Qualification of equipment and validation of computer systems are not one-time events. Correlation data across instruments or platforms is essential to accurate test performance. How we monitor software upgrades matters. In the time of computational pathology and as always for best outcomes for patient care, leveraging data interpretation in modern health care systems depends on us to provide the right data.

Further Reading

FDA 21 CFR part 820: Quality system requirements for medical device manufacturers

FDA 21 CFR part 11: Regulations on electronic records and signatures

FDA 42 CFR part 493: Laboratory requirements

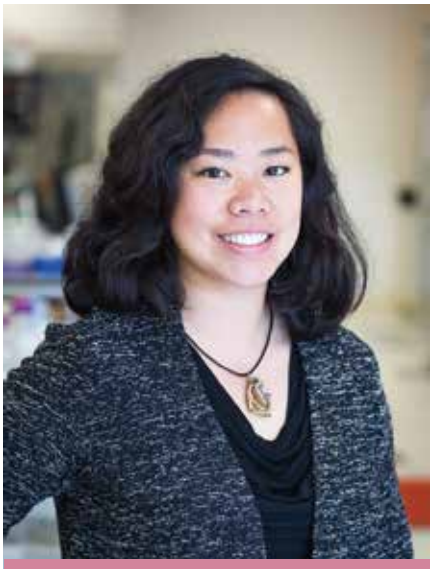
ISO 17025: General requirements for the competence of testing and calibration laboratories

ISO 15189: Requirements for quality and competence in medical laboratories

CAP's Laboratory Accreditation Program: An accreditation program covering the entire spectrum of laboratory test disciplines

Clinical Laboratory Improvement Amendments: Federal regulatory standards that apply to all clinical laboratory testing performed on humans in the US, except clinical trials and basic research

***Denise Bland**, MHA, has been in the field of histopathology for over 28 years. She works at a Boston hospital. Her days are spent striving to bridge the gap between laboratory protocols and best patient outcomes.*



Janice Chen, PhD

ASK THE EXPERT

The Promise of CRISPR-Based Diagnostics

by Laura M. Bolt, PhD

Janice Chen, PhD, is the co-founder and chief research officer of Mammoth Biosciences, a biotechnology company harnessing a revolutionary gene-editing tool called CRISPR for rapid and affordable disease detection. Janice received her PhD from the lab of Jennifer Doudna at University of California, Berkeley. She has authored multiple publications and patents related to CRISPR mechanism and technologies. She also co-invented the programmable CRISPR-based detection technology called DETECTR. Janice was selected as a 2019 Forbes 30 Under 30 in Healthcare, and recently delivered a TEDx talk on the potential for CRISPR to democratize diagnostics.

Q: What is CRISPR and what medical applications does it have?

A: CRISPR is an ancient bacterial immune system that harbors a programmable protein that can cut DNA or RNA. The system can be repurposed as a biological search engine because it uses an RNA molecule that guides the CRISPR protein to search through billions of sequences to find a matching nucleic acid target. The ability to harbor CRISPR proteins for gene editing, gene expression modulation, RNA editing, and DNA/RNA detection has enabled many applications in therapeutics and diagnostics. While CRISPR-based diagnostics are currently in development, the ability to detect and rewrite the underlying cause of genetic disease with CRISPR has huge medical potential.

Q: How can CRISPR be used to diagnose disease?

A: The finding that certain versions of CRISPR proteins can produce a real-time signal when they find a matching DNA or RNA sequence has enabled the use of CRISPR for detecting any disease with nucleic acid biomarkers. For example, CRISPR can be programmed to detect sequences from bacteria, viruses, or genetic mutations within our own cells. The researcher simply designs guide RNAs that allow CRISPR to specifically target a pathogen, such as Zika virus. Once the complex finds the presence of the viral RNA in a given sample, it cleaves a reporter molecule that releases a color change, indicating that the target is present.

"Given that CRISPR diagnostics is a new technology, it hasn't undergone decades of development. We may be scratching only the surface of the possibilities of this technology."

Q: How does the use of CRISPR for diagnostics differ from its gene editing applications?

A: Both applications leverage the programmability of RNA-guided CRISPR proteins to search for a nucleic acid target. Unlike the well-known gene editing tool Cas9, other CRISPR proteins called Cas12, Cas13, and Cas14 used for diagnostics generate indiscriminate cleavage when the protein finds its matching target. This secondary activity was an unexpected mechanistic finding and is the part that enables CRISPR [to serve] as a detection tool, because these proteins have the ability to cleave a reporter molecule that produces a real-time signal only when the target is present.

Q: What research have you conducted in this area?

A: During my time as a graduate student in the Doudna Lab, my colleagues and I discovered a new activity for the Cas12 protein and showed that it could be repurposed to detect HPV in clinical samples. This detection activity has also

been observed in other CRISPR protein families, namely Cas13 and Cas14, and there is active research on the underlying mechanisms of these CRISPR proteins and how they can be further developed for nucleic acid detection.

challenges regarding data sharing and concerns around the need for clinical counseling upon receiving a test result. We're currently having discussions about how to ensure patient privacy and appropriate counseling.

"The simplicity and accuracy of CRISPR-based diagnostics will support increased accessibility of heath information, early disease detection, clinical actionability, and personalized medicine."

Q: What are some of the strengths and weaknesses of CRISPR in disease diagnostics?

A: There are several advantages of CRISPR-based diagnostics compared to traditional PCR-based methods for detection. These include high specificity due to enzymatic recognition of the target, fast turnaround times due to signal amplification, compatibility with low-cost form factors due to isothermal reactions, and rapid test development cycles due to the programmable system.

Given that CRISPR diagnostics is a new technology, it hasn't undergone decades of development. We may be scratching only the surface of the possibilities of this technology. We envision several possibilities for CRISPR-based diagnostic products—reagent kits, integration onto existing bench-top equipment, and fully integrated devices for point-of-care or at-home use. Each class of product will have varying degrees of technical, safety, and regulatory requirements, but the underlying goal will be to improve on existing technologies and enable greater access to diagnostic information. In the case of at-home use, there are potential ethical

Q: What has been the role of Mammoth Biosciences in advancing the use of CRISPR in disease diagnostics?

A: Mammoth is the first company to take the steps toward commercializing CRISPR diagnostics. We are leveraging our expanding toolbox of CRISPR proteins with increased performance features to enable robust DNA and RNA detection. We are also developing the platform that enables rapid design, validation, and prototyping of CRISPR-based diagnostic tests that can be widely used for any nucleic acid biomarker in a simple, low-cost form factor. We think the simplicity and accuracy of CRISPR-based diagnostics will support increased accessibility of heath information, early disease detection, clinical actionability, and personalized medicine.

Q: What do you see happening in CRISPR-based diagnostics in the future?

A: Despite CRISPR diagnostics being a new technology, we are experiencing a significant demand for solutions related to improvements to existing technologies and diagnostic tests that can be performed at point-of-need. We are also observing trends toward personalized medicine and decentralization of healthcare, which will likely increase as hardware, software, and molecular technologies continue to advance. Although CRISPR diagnostics is one link in the larger value chain of decentralized healthcare, it addresses an unmet need for rapid and sensitive diagnostics in the clinic, in the field, and at home.

DETECTR: A Simple Molecular Diagnostics Platform

Last year, Chen and colleagues published a paper in *Science* on a newly discovered behavior of the CRISPR-Cas12a protein. Like Cas9, Cas12a has the ability to generate targeted, double-stranded DNA breaks. The researchers found that binding and cutting of a targeted double-stranded DNA sequence unleashes indiscriminate cutting of single-stranded DNA by Cas12a. They applied this discovery to develop a platform they dubbed DNA endonuclease targeted CRISPR trans reporter, or DETECTR, which combines Cas12a, its guide RNA, a fluorescent reporter molecule, and recombinase polymerase amplification in a single reaction. DETECTR enabled rapid and accurate detection of cervical cancer-associated human papillomavirus types 16 and 18 in patient samples, the authors reported, thus demonstrating its potential application as a simple molecular diagnostics platform.

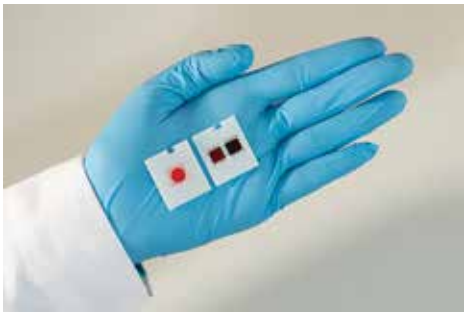
***Laura M. Bolt**, PhD, is a writer, researcher, and university-level educator based in Toronto, Canada. She holds degrees from the University of Cambridge (UK), University of Toronto, and Queen's University (Canada).*

solutions FOR THE CLINICAL LAB

From assays to analyzers, these are some of the latest and greatest products for use in clinical research and diagnostic labs

ORTHO CLINICAL DIAGNOSTICS VITROS® XT MICROSLIDES

Ortho Clinical Diagnostics' VITROS® XT MicroSlides, featuring new, multi-test technology that allows labs to run two tests simultaneously on one MicroSlide, have been cleared for market by the US FDA. By pairing commonly ordered tests so they can be run simultaneously, the MicroSlides, which are available for use on Ortho's VITROS® XT 7600 Integrated System, improve performance and productivity in the clinical lab, enabling a higher test throughput without requiring additional or larger analyzers. This is a significant benefit in a hospital environment, where space is often at a premium. In addition, the miniaturized testing areas on each MicroSlide require less patient blood sample for each of the paired tests. Available test pairs include: triglycerides and cholesterol; urea and creatinine; and glucose and calcium.



OLYMPUS IX73SC MICROSCOPE FOR ASSISTED REPRODUCTIVE TECHNOLOGY

The new Olympus IX73SC inverted microscope system is an assisted reproductive technology solution that facilitates and even eliminates steps in the intracytoplasmic sperm injection (ICSI) workflow as part of the *in vitro* fertilization process. The microscope system improves sperm selection, eases oocyte condition checks through easy metaphase II spindle visualization, increases sample throughput, reduces stress to the oocyte, and standardizes the ICSI process. The semi-motorized version of the IX73SC microscope streamlines the ICSI workflow by combining several steps. Users can switch observation methods and adjust the magnification by pressing a button on the integrated hand switch. Higher-sensitivity manual focusing can be easily achieved in conjunction with the motorized operations.

AKOYA BIOSCIENCES SOLUTION FOR HIGH-PARAMETER TISSUE ANALYSIS

Akoya Biosciences, Inc. has announced the full commercial release of its new solutions for high-parameter tissue imaging. These include the CODEX® System, an ultra-high multiplexing platform for tissue analysis and biomarker discovery, and Phenoptics™ 2.0, the company's next-generation biomarker multiplexing platform. The CODEX System includes a fluidics-based instrument, reagents, and a software analysis suite. It offers a cost-effective means to transform customers' existing fluorescence microscopes into ultra-high-parameter tissue imaging systems. With the release of Phenoptics 2.0, the company is introducing its proprietary MOTiF™ technology, which acquires multispectral images on whole-slide tissue sections of up to seven colors simultaneously, and 20 times faster than previously possible. The ability to process up to 30 whole-slide images per day makes this platform ideal for translational and clinical research studies.

THERMO FISHER SCIENTIFIC SMART QPCR INSTRUMENTS

Thermo Fisher Scientific has launched the Applied Biosystems QuantStudio 6 and 7 Pro Real-Time PCR Systems, the world's first Smart qPCR instruments. Capabilities include facial authentication, voice commands, radio-frequency identification (RFID)-enabled plate scanning, and quick access to service and support with a single touch. QuantStudio Pro instruments use RFID-enabled Applied Biosystems TaqMan Array Plates for seamless upload of plate layout and assay information for setting up the run. This eliminates manual data entry, reduces potential errors, and improves experimental reproducibility. Results from experiments can then be analyzed remotely from any desktop computer or smartphone using the Instrument Connect mobile app. Support on the new system is also now expedited with the new Smart Help feature, which provides instant access to service and support professionals. Through this feature, all relevant diagnostic information is forwarded to an on-call team member and, when possible, corrective action may be completed remotely using augmented reality technology.

CYGNUS TECHNOLOGIES ENDONUCLEASE IMPURITIES IDENTIFICATION KIT

Cygnus Technologies has introduced the EndonucleaseGTP™ ELISA kit for the detection and quantitation of residual endonuclease impurities in recombinant vaccines and viral vectors used for gene therapy. Endonucleases such as Benzonase and Denarase are often used to cleave host cell DNA and RNA during the production of these biologics and must subsequently be removed. The new Cygnus ELISA kit, with a detection limit of ~0.06 ng/ml, is three times more sensitive than the only other commercially available assay for detection and quantitation of these endonuclease impurities. The EndonucleaseGTP ELISA kit contains all the necessary ready-to-use reagents for 96 analyses in microplate format, including a set of calibrated endonuclease standards. The assay is easy to use and can be easily integrated into desired workflow points, from process development to QC to lot release testing.

QIAGEN COMPANION DIAGNOSTIC FOR BLADDER CANCER

QIAGEN has announced the US launch of its novel *therascreen*® FGFR RGQ RT-PCR Kit (*therascreen* FGFR Kit) as a companion diagnostic to help guide the use of the newly approved FGFR kinase inhibitor, BALVERSA™. The test will aid in identifying patients with urothelial cancer whose tumors have certain alterations in the fibroblast growth factor receptor 3 gene. The US FDA co-approved the new test with BALVERSA™. A percentage of urothelial carcinoma tumors have certain FGFR alterations thought to be key drivers of tumor growth. Detection of these alterations utilizing the companion diagnostic will help identify patients eligible for treatment with BALVERSA. The *therascreen* FGFR Kit will run on QIAGEN's Rotor-Gene Q MDx.

SPOTLIGHT ON AUTOMATION solutions

BECKMAN COULTER DXA 5000 TOTAL LABORATORY AUTOMATION SOLUTION

Beckman Coulter has accounted that its DxA 5000 total laboratory automation solution has achieved European CE Mark and China FDA approval. The DxA 5000 delivers rapid and consistent turnaround time, provides a new level of comprehensive pre-analytical sample quality detection, and reduces the number of manual processing steps to significantly improve laboratory efficiency. The DxA 5000 utilizes a universal centrifugation protocol that significantly reduces the pre-analytical processing time by up to 73 percent for connected analyzers across multiple disciplines. Additionally, the DxA 5000 supports laboratories in delivering highly consistent turnaround time to their physicians. Leveraging first-of-its-kind dynamic system software, the DxA 5000 utilizes Intelligent Routing to bring automated patient-centric workflow to the laboratory. By understanding the tests requested, sample volume available, and real-time analyzer capacity and status, the DxA 5000 continuously calculates the most expeditious route for every patient sample—both STAT and routine.

SHIMADZU CLINICAL LABORATORY AUTOMATION MODULE

Shimadzu Scientific Instruments has announced the release of the Clinical Laboratory Automation Module (CLAM-2030), a fully integrated sample pre-treatment module for LCMS biological analysis. This module improves laboratory efficiency, enables fast, precise results, and maintains low operating costs. By automating operations such as dispensing, stirring, filtering, heating, and sample transfer, the system effectively improves data accuracy and achieves the reproducibility needed for clinical research and forensic toxicology. Such steps can be combined to perform tasks such as deproteination, internal standard addition, derivatization, and glucuronidase digestion. Through automation, the CLAM-2030 helps to facilitate a safe working environment for laboratory personnel by minimizing human contact with potentially hazardous biological samples. Following pre-treatment, solid and liquid waste are contained within the body of the CLAM-2030 and collected.

ABBOTT ALINITY™ M DIAGNOSTIC SYSTEM AND ASSAYS

Abbott has announced CE Mark for its Alinity™ m diagnostics system and assays. This new technology will help keep up with the growing demand for infectious disease testing. Alinity m provides flexibility for lab staff and faster testing results for clinicians and patients. Alinity m may also reduce the lab equipment footprint from four to six instruments down to one, decreasing the space requirements and hours spent learning and maintaining different instruments. Now available in countries that recognize CE Mark, Alinity m offers initial assays including virologic testing for human immunodeficiency virus type 1, hepatitis B virus, and hepatitis C virus; sexual health-related testing for *Chlamydia trachomatis*, *Neisseria gonorrhoeae*, *Trichomonas vaginalis*, and *Mycoplasma genitalium*; and high-risk human papillomavirus testing. In the US, Alinity m is in development and is not commercially available for diagnostic use.

CLINICAL LAB MANAGER IS ONLINE

Visit our website at ClinicalLabManager.com

CLINICAL LM Lab Manager

READ THE LATEST NEWS

Malaria Vaccine Pilot Launched

New Genomics Tool Expands Multimodal Single-Cell Analysis

Could Blockchain Ensure Integrity of Clinical Trial Data?

EXPLORE IN-DEPTH FEATURES

Influenza Diagnostic Methods: RT-PCR vs. RIDTs

How Accurate is POC Glucose Testing?

Obstacles to Developing Tailored Normal Test Ranges

FOLLOW OUR THOUGHT LEADERSHIP BLOG

Boosting Lab Efficiency with Quality Management Tools

A Growing Need for Informatics Tools in Clinical Diagnostics

How Point-of-Care Testing Can Add Value to Healthcare Providers

Big Ideas About the Clinical Industry

Why Community-Based Hospitals are Bringing NGS In-House

Insourcing next-generation sequencing saves time and resources and helps improve patient care by Luca Quagliata, PhD

Next-generation sequencing (NGS) began as a revolutionary research tool a decade ago, but it has emerged as the platform of choice for clinical researchers and clinicians looking to advance the way they understand patients' diseases and, more importantly, how to treat them.

In cancer, NGS can reveal precise data about a tumor's genetic makeup, allowing clinicians to match patients with targeted treatments. As recently as five years ago, however, NGS was just making its way out of research labs and was not quite ready for routine use at community hospitals. To get an NGS report, clinicians had to relinquish control and send patient tumor samples for analysis to a reference lab, a process that takes weeks and has a high failure rate. Today, any certified community hospital can perform NGS, and many are in the process of embracing it to save valuable time and money, as well as to improve outcomes for patients.

Bringing NGS analysis in-house helps hospitals overcome the fundamental problem with outsourced testing—the long waiting period for results, which is stressful for both the ordering clinician and the patient. Faster turnaround times are critical for effective cancer treatment decisions, particularly for late-stage patients. While commercial labs can take up to 20 days or more to deliver results—at which point they may no longer be valid or helpful—in-house NGS can produce results in as few as six days. This expedited path enables clinicians to more quickly select the most appropriate targeted therapy, leading to better health outcomes.

Bringing NGS testing in-house also supports stronger stewardship of patient samples and tighter adherence to sequencing protocols to reduce errors and keep patients' genomic data within the hospital. Retaining this critical information protects patient privacy, helps build the hospital's database for further studies, and increases institutional knowledge that can inform treatment for future patients with similar cancers.

Patients also benefit from the stronger collaboration that naturally occurs between pathologists and clinicians when NGS testing is brought in-house. Hospitals now commonly form molecular tumor boards in which oncologists, molecular pathologists, and staff meet to formulate the best treatment options for each patient. The board provides a forum in which pathologists become part of the patient's care team (rather than just the issuer of a written report) and learn how NGS results inform clinical research and treatment. The dialogue creates a unique environment for professional development that ultimately leads to better patient care and furthers cancer clinical research.

As demand for molecular profiling increases, in-house NGS testing makes economic sense for local hospitals. Recent studies have found that compared with sequential single-gene testing, NGS identifies a broader set of actionable drug targets and offers higher reliability, lower dropout rates, minimal tissue requirements, faster turnaround times, and lower cost.

We are already seeing real-world examples of in-house NGS testing globally, driven primarily by cost savings and ease of use. Access to NGS testing in this initial clinical phase has been limited to large academic medical centers, but more and more, community-based hospitals and government health care agencies are changing that paradigm.

Growing demand for molecular profiling, along with the rapid maturation and accessibility of NGS technology and the availability of more effective targeted therapies, is enabling oncologists and clinician teams to leverage an opportunity never seen before in the medical field. The expectation is that this forward momentum will continue to help improve outcomes for patients who need it most.

***Luca Quagliata**, PhD, is the global head of medical affairs for clinical NGS and oncology at Thermo Fisher Scientific.*

Checking Your Answers: Monitoring Immunotherapy Effectiveness with ddPCR

A ddPCR-based liquid biopsy can reveal whether an immunotherapy is working within weeks

by George Karlin-Neumann, PhD

When oncologists decide to use immunotherapy to treat a patient, they must make crucial determinations: Do the benefits outweigh the risks? And is it the best available option for the patient?

Although immunotherapies are effective in treating multiple types of cancers, they may not be effective in a significant fraction of patients who possess particular tumor types. For instance, checkpoint inhibitors, the most commonly used immunotherapy, can fail in up to 85 percent of patients treated for certain cancer types. Immunotherapies fail so often mainly because of our imperfect ability to accurately predict who will benefit.

When immunotherapy drugs do work, they can result in deep, long-lasting positive responses. But because of their unique ability to stimulate the immune system, they can induce a number of severe immune-related adverse events (irAEs) resulting from systemic inflammation. Immunotherapy's limited success rates, paired with its tendency to induce severe irAEs and the inaccuracy of predictive tests, may cause many patients more harm than good. Consequently, oncologists should monitor their patients' responsiveness to therapy and determine as early as possible whether the treatment is working.

The current standard in assessing a patient's response to therapy is the use of computed tomography (CT) imaging or magnetic resonance imaging. However, imaging-based approaches may not tell the whole story. For instance, some cancer patients who receive immunotherapy experience pseudo-progression. This occurs when a scan shows that a tumor has grown, when in reality, it's just temporarily inflamed from an influx of lymphocytes as a result of an effective immunotherapy treatment that will eventually cause it to shrink. Oncologists might not be able to distinguish between pseudo-progression and true progression for more than three months, making it challenging to decide whether the treatment was successful. Incorrectly mistaking pseudo-progression for true progression might put patients at risk of ceasing the use of an effective drug.

A promising alternative to imaging is liquid biopsy, which detects circulating cell-free tumor DNA (ctDNA) shed from dying tumor cells into the blood. Liquid biopsy can detect changes in ctDNA that reflect treatment effectiveness, or lack thereof, within weeks of starting treatment. It can also distinguish true progression from pseudo-progression. This approach is under active investigation using droplet digital PCR (ddPCR).

Unlike other phenotypic markers used as surrogates for tumor response to therapy, such as proteins or RNAs, ctDNA in blood or other bodily fluids is both a genetic and a phenotypic marker of therapy effectiveness. It measures the direct effects of the therapy on tumor cells, such as tumor cell turnover. ctDNA is thus a very specific biomarker for the tumor cells and can efficiently tell an oncologist whether he or she has made the right therapeutic choice for the patient.

Liquid biopsies do harbor some limitations, but these are minor compared with the limitations of current pretreatment predictive tests. For instance, liquid biopsies may not detect some tumors in locations where they do not slough ctDNA into the blood (e.g., brain metastases).

Nonetheless, this plasma-monitoring paradigm of immunotherapy effectiveness is a significant step forward because it removes many of the assumptions that must be made when predicting immunotherapy effectiveness with currently available pretreatment prediction methods. ddPCR enables a short turnaround time and can be used to monitor treatment response more frequently and earlier than can CT scans because it is less invasive than a traditional biopsy and more cost effective than imaging.

Because of their sensitivity, specificity, and speed, ddPCR-based liquid biopsies could provide an early and reliable measure of treatment effectiveness. This would help oncologists make smarter treatment decisions faster, sparing their patients unnecessary side effects and saving precious time from being wasted on ineffective treatment regimens.

***George Karlin-Neumann**, PhD, is the director of Scientific Affairs at Bio-Rad's Digital Biology Center, formerly QuantaLife.*

AirClean® Systems

The Fume Control Experts.

Manufacturing Green Products Since 1992

Combination PCR Workstation

ISO 5 Clean Air and UV Light Sterilization

Features:

- UVTect™ microprocessor controller with HEPA filtration monitoring
- ISO 5 vertical laminar flow air
- Polycarbonate and polypropylene design to reflect UV energy
- Unibody plastic design — no joints or gaps in construction
- Digital UV light timer (0-59 minutes)
- UV shelf with built-in pipettor holder
- Available in 24″, 32″, and 48″ widths; nominal depth 24″
- Shipped fully assembled

www.aircleansystems.com