Rediscovering urinalysis

Biomarkers for lung cancer
New diagnostics for STIs
Microfluidics in the clinical lab

EXECUTIVE SNAPSHOT
Norman Sharples, CEO
COPAN Diagnostics, Inc.
The right combination.

ProFlu™+

ProFAST®+

ProParafiu˚+

Pro hMPV˚+

ProAdeno˚+

Why test for everything every time?

The Prodesse® Plus series of real-time PCR assays allows you to choose which combination of respiratory pathogens you need to test for. Now, you can test for any combination of Influenza A/B, respiratory syncytial virus (RSV), Influenza A subtyping, Parainfluenza 1/2/3, human metapneumovirus (hMPV) and adenovirus, without unnecessary extra tests.

Choose the Prodesse Plus Series – Providing flexibility and personalized patient testing that physicians and laboratories require.
Santa will be relying on his glucose results after eating so many cookies.

Make sure your lab is ready. Introducing: Blood Glucose Linearity & Daily QC.

Control DROP LQ Blood Glucose
Order Number: K078M-8
Levels: Two
Type: Liquid
Open Vial: 7 days when stored at 2-8°C
Analytes: Glucose

Linearity DROP LQ Blood Glucose
Order Number: K736M-5
Levels: Five
Type: Liquid
Open Vial: 7 days when stored at 2-8°C
Analytes: Glucose

For more information | 866.25.AUDIT (28348) customerservice@auditmicro.com | auditmicro.com
FEATURES

CLINICAL ISSUES
18 A blood test for early detection of lung cancer
By Laura J. Peek, PhD, and James R. Jett, MD

THE PRIMER
20 MicroRNA: A tiny molecule yields big insights into disease states
By John Brunstein, PhD

SPECIAL FEATURE
22 MALDI-ToF is poised to speed diagnosis for bacterial and fungal infections
By Janet Cromien and Dale Schwab, PhD

EDUCATION
24 New testing options for Trichomonas vaginalis respond to growing awareness
By Barbara Van Dei Pol, PhD, MPH

28 Managing Mycoplasma genitalium infections during a rapid upsurge in antibiotic resistance
By Tim Read, PhD, MBBS, and Catriona Bradshaw, PhD, MBBS (Hons)

30 Neisseria gonorrhoeae susceptibility to ciprofloxacin
By Lao-Tzu Allan-Blitz and Jeffrey D. Klausner, MD, MPH

LAB MANAGEMENT
32 As ACOs evolve, the clinical lab’s role grows
By Irwin Z. Rothenberg, MBA, MS, MT(ASCP)

FUTURE BUZZ
34 Microfluidics in the clinical lab
By Richard Day, PhD

CONTINUING EDUCATION

12 Rediscovering urine chemistry—and understanding its limitations
By Michelle D. Dumonceaux, BS, MBA, and Mira Gamez, BS, MT(ASCP) CLS

15 Urinalysis: Are we done spinning yet?
By Roy Midyett, CLS

EDUCATION

4 From the editor

DEPARTMENTS

4 From the editor
10 The observatory
35 Washington report

PRODUCTS

36 Product focus: POCT
39 New products

MARKETPLACE

38 Clinical spotlights
39 Advertiser index

EXECUTIVE SNAPSHOT

40 Microbiology enters the Age of Automation
Norman Sharples, CEO
COPAN Diagnostics, Inc.
“Testing simultaneously for CT, GC, and Trichomonas is optimal for detection of the most common treatable STIs in female patients.”

2013 IDSA & ASM Guidelines

“How can you achieve

MAX

IMPACT?

ELEVATE THE STANDARD OF CARE

BD MAX™ CT/GC/TV

Twenty million new sexually transmitted infections (STIs) occur in the US annually. The new BD MAX™ CT/GC/TV assay can help, with comprehensive, expedited testing.

► Syndromically focused to address the 3 most prevalent non-viral STIs in a single assay — Chlamydia trachomatis, Neisseria gonorrhoeae, Trichomonas vaginalis (CT/GC/TV)
► Strong solutions to provide quality patient care and to support clinical decision accuracy and patient management
► Versatility — fully integrated with BD MAX™ System

START MAXIMIZING YOUR IMPACT

VISIT BD.COM/DS
FROM THE EDITOR
By Alan Lenhoff, Editor

A supplement for empathy?

In his 1972 novel Breakfast of Champions, the American writer Kurt Vonnegut, Jr. (1922-2007), wrote, “I tend to think of human beings as huge, rubbery test tubes, with chemical reactions seething inside.” Vonnegut was speaking ironically; he didn’t really think of human beings that way: if there is one over-arching theme in his body of work, it is the question of how we remain human in an increasingly technological world. But he was making a serious point about how chemical influences affect psychological states, and I think he would have been interested in a news release that recently crossed my desk.

The released description research conducted at the University of Cardiff (Wales) and presented at the Society of Endocrinology annual conference. It concerned a study that indicated that people with low levels of the hormone oxytocin exhibit reduced empathy. That is, in the study, people suffering medical conditions causing low levels of oxytocin performed worse on empathy tasks. The research suggests that hormone replacement could improve the psychological well-being of those living with the low levels.

Oxytocin, which has sometimes been called “the love hormone,” has a role in sexual arousal, recognition, trust, anxiety, and mother-infant bonding. It is produced by the hypothalamus and stored in the pituitary gland.

The researchers investigated empathic behavior in people whom they suspected of having reduced oxytocin levels due to one of two medical conditions caused in response to pituitary surgery. The study assessed 20 people with cranial diabetes insipidus (CDI). In CDI, the body has reduced levels of ADH, a chemical also produced in the hypothalamus and structurally very similar to oxytocin. They also assessed 15 people with hypopituitarism (HP), a condition in which the pituitary gland does not release enough hormones. These two patient groups were compared to a group of 20 healthy controls.

The researchers gave all participants two tasks designed to test empathy, both relating to the recognition of emotional expression. Both tasks required participants to identify which emotion or feeling best described a series of images of human faces.

The scientists measured each group’s oxytocin levels and found that the 35 CDI and HP participants had slightly lower oxytocin compared to the healthy controls. They also saw that the CDI and HP groups performed significantly worse on empathy tasks, compared to controls. In particular, CDI participants’ ability to identify expressions was predicted by their oxytocin levels: those with the lowest levels of oxytocin produced the worst performances.

“This study looks at low oxytocin as a result of medical, as opposed to psychological, disorders,” said Katie Daughters, lead researcher. “The results from our patient groups suggest it is also important to consider medical conditions carrying a risk of low oxytocin levels.”

“Patients who have undergone pituitary surgery, and in particular those who have acquired CDI as a consequence, may present with lower oxytocin levels. This could impact their emotional behavior, and in turn affect their psychological well-being. Perhaps we should be considering the introduction of supplements to make them, well, nicer, need to be addressed. And if one ex-

Now, ethical questions can have ethical answers, and science should not fear that could be a slippery slope, at least. Perhaps. But empathy is subjective, and how much of it someone should have is a matter of opinion; the ethical questions that might be raised by the idea of giving supplements to people who seem to lack the optimal amount of empathy for any reason—that could be a slippery slope, at least.

Now, ethical questions can have ethical answers, and science should not fear.
QUALITY MEANS CERTAINTY

The integrity and accuracy of genetic analysis relies on the quality of the reagents and components utilized in your workflow. Every laboratory – every doctor – and every patient relies on the quality of our products to deliver certainty in their results.

Learn More about our complete offering for the Genetic Analysis Workflow:

Sample Preparation
• GenElute™ DNA & RNA Purification Kits
• Extract-N-Amp™ Kits

Amplification
• SeqPlex™ DNA & RNA Amplification Kits
• JumpStart™ Taq Polymerase and PCR Reagents

Sequencing
• ISO13485 Adapters and Probes

Available in package sizes to fit your needs.

WE ARE QUALITY.
sigma-aldrich.com/clinical
Identify EGFR T790M mutation in patients who progress on or after EGFR TKI therapy

• Nearly 2 out of 3 cases of progression with first-generation EGFR TKIs are related to the acquired T790M mutation

• Testing at progression provides the opportunity to identify mechanisms of resistance, including the T790M mutation

TAGRISSO demonstrated efficacy and safety in two clinical trials

• TAGRISSO was researched in two separate, global, Phase II, single-arm, open-label clinical trials in patients with EGFR T790M mutation–positive NSCLC who had progressed on or after EGFR TKI therapy

• A 59% objective response rate (95% CI: 54–64) observed in patients (N=411) who progressed with previous EGFR TKI therapy

• The most common adverse reactions (>20%) observed in TAGRISSO patients were diarrhea (42%), rash (41%), dry skin (31%), and nail toxicity (25%)

• Interstitial lung disease (ILD) was reported in 3.3% of patients and was fatal in 0.5% of TAGRISSO patients

Visit TAGRISSOhcp.com for more information
IDENTIFY PATIENTS APPROPRIATE FOR TAGRISSO

Visit cobasEGFRtest.com today to learn how.

IMPORTANT SAFETY INFORMATION

• There are no contraindications for TAGRISSO

• Interstitial Lung Disease (ILD)/Pneumonitis occurred in 3.3% and was fatal in 0.5% of 813 TAGRISSO patients. Withhold TAGRISSO and promptly investigate for ILD in any patient presenting with worsening of respiratory symptoms indicative of ILD (e.g., dyspnea, cough and fever). Permanently discontinue TAGRISSO if ILD is confirmed

• QTc interval prolongation occurred in TAGRISSO patients. Of the 411 patients in two Phase II studies, 0.2% were found to have a QTc greater than 500 msec, and 2.7% had an increase from baseline QTc greater than 60 msec. Conduct periodic monitoring with ECGs and electrolytes in patients with congenital long QTc syndrome, congestive heart failure, electrolyte abnormalities, or those who are taking medications known to prolong the QTc interval. Permanently discontinue TAGRISSO in patients who develop QTc interval prolongation with signs/symptoms of life threatening arrhythmia

• Cardiomyopathy occurred in 1.4% and was fatal in 0.2% of 813 TAGRISSO patients. Left Ventricular Ejection Fraction (LVEF) decline >10% and a drop to <50% occurred in 2.4% of (9/375) TAGRISSO patients. Assess LVEF before initiation and then at 3 month intervals of TAGRISSO treatment. Withhold TAGRISSO if ejection fraction decreases by 10% from pretreatment values and is less than 50%. For symptomatic congestive heart failure or persistent asymptomatic LV dysfunction that does not resolve within 4 weeks, permanently discontinue TAGRISSO

• Advise pregnant women of the potential risk to a fetus. Advise females of reproductive potential to use effective contraception during TAGRISSO treatment and for 6 weeks after the final dose. Advise males with female partners of reproductive potential to use effective contraception for 4 months after the final dose

• The most common adverse reactions (>20%) observed in TAGRISSO patients were diarrhea (42%), rash (41%), dry skin (31%) and nail toxicity (25%)

INDICATION

TAGRISSO is indicated for the treatment of patients with metastatic epidermal growth factor receptor (EGFR) T790M mutation-positive non-small cell lung cancer (NSCLC), as detected by an FDA-approved test, who have progressed on or after EGFR tyrosine kinase inhibitor therapy.

This indication is approved under accelerated approval based on tumor response rate and duration of response. Continued approval for this indication may be contingent upon verification and description of clinical benefit in confirmatory trials.

Please see Brief Summary of full Prescribing Information on adjacent pages.

TAGRISSOTM (osimertinib) tablets, for oral use

Brief Summary of Prescribing Information.

For complete prescribing information consult official package insert.

INDICATIONS AND USAGE

TAGRISSO is indicated for the treatment of patients with metastatic epidermal growth factor receptor (EGFR) T790M-mutation-positive non-small cell lung cancer (NSCLC), as detected by an FDA-approved test, who have progressed on or after EGFR tyrosine kinase inhibitor (TKI) therapy.

This indication is approved under accelerated approval based on tumor response rate and duration of response [see Clinical Studies (14) in full Prescribing Information]. Continued approval for this indication may be contingent upon verification and description of clinical benefit in confirmatory trials.

DOSEAGE AND ADMINISTRATION

Patient Selection

Confirm the presence of a T790M EGFR mutation in tumor or, in the absence of tumor, plasma specimens prior to initiation of treatment with TAGRISSO [see Indications and Usage (1) and Clinical Studies (14) in full Prescribing Information]. Testing for the presence of the mutation in plasma specimens is recommended only in patients for whom a tumor biopsy cannot be obtained. If this mutation is not detected in a plasma specimen, re-evaluate the feasibility of biopsy for tumor tissue testing. Information on FDA-approved tests for the detection of T790M mutations is available at http://www.fda.gov/companiondiagnostics.

Recommended Dosage Regimen

The recommended dose of TAGRISSO is 80 mg tablet once a day until disease progression or unacceptable toxicity. TAGRISSO can be taken with or without food.

If a dose of TAGRISSO is missed, do not make up the missed dose and take the next dose as scheduled.

Administration to Patients Who Have Difficulty Swallowing Solids

Disperse tablet in 60 mL (2 ounces) of non-carbonated water only. Stir until tablet is dispersed into small pieces (the tablet will not completely dissolve) and swallow immediately. Do not crush, heat, or ultrasonicate during preparation. Rinse the container with 120 mL to 240 mL (4 to 8 ounces) of water and immediately drink.

If administration via nasogastric tube is required, disperse the tablet as above in 15 mL of non-carbonated water, and then use an additional 15 mL of water to transfer any residues to the syringe. The resulting 30 mL liquid should be administered as per the nasogastric tube instructions with appropriate water flushes (approximately 30 mL).

Dosage Modification

Adverse Reactions

Table 1 Recommended Dose Modifications for TAGRISSO

<table>
<thead>
<tr>
<th>Target Organ</th>
<th>Adverse Reactiona</th>
<th>Dose Modification</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pulmonary</td>
<td>Intestinal lung disease (ILD)/Pneumonitis</td>
<td>Permanently discontinue TAGRISSO</td>
</tr>
<tr>
<td>QTc interval greater than 500 msec on at least 2 separate EGSb</td>
<td>Withhold TAGRISSO until QTc interval is less than 481 msec or recovery to baseline if QTc is greater than or equal to 431 msec, then resume at 40 mg dose.</td>
<td></td>
</tr>
<tr>
<td>QTc interval prolongation with signs/symptoms of life-threatening arrhythmia</td>
<td>Permanently discontinue TAGRISSO.</td>
<td></td>
</tr>
<tr>
<td>Cardiac</td>
<td>QTc interval prolongation with signs/symptoms of life-threatening arrhythmia</td>
<td>Permanently discontinue TAGRISSO.</td>
</tr>
<tr>
<td>Symptomatic congestive heart failure</td>
<td>Withhold TAGRISSO for up to 4 weeks. If improved to baseline use TAGRISSO, permanently discontinue.</td>
<td></td>
</tr>
<tr>
<td>Grade 3 or higher adverse reaction</td>
<td>Withhold TAGRISSO for up to 3 weeks.</td>
<td></td>
</tr>
<tr>
<td>If improvement to Grade 0-2 within 3 weeks</td>
<td>Resume at 80 mg or 40 mg daily.</td>
<td></td>
</tr>
<tr>
<td>If no improvement within 3 weeks</td>
<td>Permanently discontinue TAGRISSO.</td>
<td></td>
</tr>
</tbody>
</table>

a Adverse reactions graded by the National Cancer Institute Common Terminology Criteria for Adverse Events version 4.0 (NCI CTCAE v4.0).
b EGS = Electrocardiograms.

Drug Interactions

Strong CYP3A4 Inducers

If concurrent use is unavoidable, increase TAGRISSO dosage to 160 mg daily when coadministering with a strong CYP3A4 inducer. Resume TAGRISSO at 80 mg 3 weeks after discontinuation of the strong CYP3A4 inducer [see Drug Interactions (7), and Clinical Pharmacology (12.3) in full Prescribing Information].

CONTRAINDICATIONS

None.

WARNINGS AND PRECAUTIONS

Interstitial Lung Disease/Pneumonitis

Across clinical trials, interstitial lung disease (ILD)/pneumonitis occurred in 3.3% (n=27) of TAGRISSO treated patients (n=813); 0.5% (n=4) were fatal.

Withhold TAGRISSO and promptly investigate for ILD in any patient who presents with worsening of respiratory symptoms which may be indicative of ILD (e.g., dyspnea, cough and fever). Permanently discontinue TAGRISSO if ILD is confirmed [see Dosage and Administration (2.4) and Adverse Reactions (6) in full Prescribing Information].

QTc Interval Prolongation

If the heart rate-corrected QT (QTc) interval prolongation occurs in patients treated with TAGRISSO. Of the 411 patients in Study 1 and Study 2, one patient (0.2%) was found to have a QTc greater than 500 msec, and 11 patients (2.7%) had an increase from baseline QTc greater than 60 msec [see Clinical Pharmacology (12.2) in full Prescribing Information].

In Study 1 and 2, patients with baseline QTc of 470 msec or greater were excluded. Conduct periodic monitoring with ECGs and electrolytes in patients with congenital long QT syndrome, congestive heart failure, electrolyte abnormalities, or those who are taking medications known to prolong the QTc interval. Permanently discontinue TAGRISSO in patients who develop QTc interval prolongation with signs/symptoms of life-threatening arrhythmia [see Dosage and Administration (2.4) in full Prescribing Information].

Cardiomyopathy

Across clinical trials, cardiomyopathy (defined as cardiac failure, pulmonary edema, ejection fraction decreased or stress cardiomyopathy) occurred in 1.4% (n=11) of TAGRISSO treated patients (n=813); 0.2% (n=2) were fatal.

In Study 1 and Study 2, Left Ventricular Ejection Fraction (LVEF) decline >10% and a drop in LVEF ≥ 50% occurred in 2.4% (8/357) of patients who had baseline and at least one follow-up LVEF assessment.

Assess LVEF by echocardiogram or multigated acquisition (MUGA) scan before initiation of TAGRISSO and then at 3 month intervals while on treatment. Withhold treatment with TAGRISSO if LVEF <50% from pretreatment values and is less than 50%. For symptomatic congestive heart failure or persistent, asymptomatic LV dysfunction that does not resolve within 4 weeks, permanently discontinue TAGRISSO [see Dosage and Administration (2.4) in full Prescribing Information].

Embryo-Fetal Toxicity

Based on data from animal studies and its mechanism of action, TAGRISSO can cause fetal harm when administered to a pregnant woman. In animal reproduction studies, osimertinib caused post-implantation embryonic loss at plasma exposures of approximately 0.5-times those observed in patients at the 80 mg dose level.

Advise pregnant women of the potential risk to a fetus.

Advise females of reproductive potential to use effective contraception during treatment with TAGRISSO and for 6 weeks after the final dose. Advise males with female partners of reproductive potential to use effective contraception for 1 month after the final dose [see Use in Specific Populations (8.1), (8.3) and Clinical Pharmacology (12.3) in full Prescribing Information].

ADVERSE REACTIONS

The following adverse reactions are discussed in greater detail in other sections of the labeling:

Interstitial Lung Disease/Pneumonitis [see Warnings and Precautions (5.1) in full Prescribing Information]

QTc Interval Prolongation [see Warnings and Precautions (5.2) in full Prescribing Information]

Clinical Trials Experience

Because clinical trials are conducted under widely varying conditions, adverse reaction rates observed in the clinical trials of a drug cannot be directly compared to rates in the clinical trials of another drug and may not reflect the rates observed in practice.

The data described below reflect exposure to TAGRISSO (80 mg daily) in 411 patients with EGFR T790M-mutation-positive non-small cell lung cancer who received prior EGFR TKI therapy, in two single-arm studies, Study 1 and Study 2. Patients with a past medical history of ILD or radiation pneumonitis that required steroid treatment, serious arrhythmia or baseline QTc interval greater than 470 ms were excluded from Study 1 and Study 2. Baseline patient and disease characteristics were: median age 63 years, 13% of patients were ≥75 years old, female (58%), White (38%), Asian (9%), metastatic (98%), sites of brain metastases (39%), World Health Organization (WHO) performance status of 0 (37%) or 1 (63%), 1 prior line of therapy [EGFR-TKI treatment only, second line, chemotherapy-naive (31%), 2 or more prior lines of therapy (58%). Of the 411 patients, 333 patients were evaluated for LVEF at least 6 months; 97 patients were exposed for at least 9 months; however, no patient was exposed to TAGRISSO for 12 months.

In Studies 1 and 2, the most common (>20%) adverse reactions (all grades) observed in TAGRISSO-treated patients were diarrhea (42%), rash (41%), dry skin (31%), and nail toxicity (22%). Dose reductions occurred in 4.4% of patients treated with TAGRISSO. The most frequent adverse reactions that led to dose reductions or interruptions were: electrocardiogram QTc prolonged (2.2%) and neutropenia (1.9%). Serious adverse reactions reported in ≥2% or more patients were pulmonary and pulmonary embolus. There were 4 patients (1%) treated with TAGRISSO who developed fatal adverse reactions of ILD/pneumonitis. Other fatal adverse reactions occurring in more than 1 patient included pneumonia (4 patients) and CVA/cerebral hemorrhage (2 patients). Discontinuation of treatment due to adverse reactions occurred in 5.6% of patients treated with TAGRISSO. The most frequent adverse reactions that led to discontinuation were ILD/pneumonitis and cerebrovascular accidents/faintings.

Tables 2 and 3 summarize the common adverse reactions and laboratory abnormalities observed in TAGRISSO-treated patients.
substrate compared to administering the BCRP substrate alone. Coadministering TAGRISSO with a BCRP substrate increased the exposure of the BCRP substrate.

**Effect of Osimertinib on Other Drugs**

If coadministering TAGRISSO with a strong CYP3A4 inducer decreased the exposure of osimertinib compared to administering TAGRISSO alone [see Clinical Pharmacology (12.3) in full Prescribing Information]. Increased BCRP substrate exposure may increase the risk of exposure-related toxicity. Monitor for adverse reactions of the BCRP substrate (e.g., rosuvastatin, sulfasalazine, topotecan), unless otherwise instructed in its approved labeling, when coadministered with TAGRISSO.

**USE IN SPECIFIC POPULATIONS**

### Pregnancy

**Risk Summary**

Based on data from animal studies and its mechanism of action, TAGRISSO can cause fetal harm when administered to a pregnant woman. There are no available data on TAGRISSO use in pregnant women. Administration of osimertinib to pregnant rats was associated with embryolethality and reduced fetal growth at plasma exposures 1.5 times the exposure at the recommended human dose [see Data]. Advise pregnant women of the potential risk to a fetus. The in U.S. general population, the estimated background risk of major birth defects and miscarriage in clinically-recognized pregnancies is 2% to 4% and 15% to 20%, respectively.

**Data**

**Animal Data**

When administered to pregnant rats prior to embryonic implantation through the end of organogenesis (gestation days 2-20) at a dose of 20 mg/kg/day, which produced plasma exposures of approximately 1.5 times the clinical exposure, osimertinib caused post-implantation loss and early embryonic death. When administered to pregnant rats from implantation through the closure of the hard palate (gestation days 6 to 16) at doses of 1 mg/kg/day and above (0.1-times the AUC observed in patients at the recommended dose of 80 mg), an increased incidence in the rate of fetal malformations and variations was observed in treated litters relative to those of concurrent controls. When administered to pregnant dams at doses of 30 mg/kg/day during organogenesis through lactation Day 6, osimertinib caused an increase in total litter loss and postnatal death. At a dose of 20 mg/kg/day, osimertinib administration during the same period resulted in increased postnatal death as well as a slight reduction in mean pup weight at birth that increased in magnitude between lactation days 4 and 6.

**Lactation**

**Risk Summary**

There are no data on the presence of osimertinib in human milk, the effects of osimertinib on the breastfed infant or on milk production. Administration to rat during gestation and early lactation was associated with adverse effects, including reduced growth rates and neonatal death [see Use in Specific Populations (8.1) in full Prescribing Information]. Because of the potential for serious adverse reactions in breastfed infants from osimertinib, advise a lactating woman not to breastfeed during treatment with TAGRISSO and for 2 weeks after the final dose.

**Females and Males of Reproductive Potential**

**Contraception**

**Females**

Advise females of reproductive potential to use effective contraception during treatment with TAGRISSO and for 6 weeks after the final dose [see Use in Specific Populations (8.1) in full Prescribing Information].

**Males**

Advise male patients with female partners of reproductive potential to use effective contraception during treatment with TAGRISSO.

**Females and Males of Reproductive Potential**

**Infertility**

Based on animal studies, TAGRISSO may impair fertility in females and males of reproductive potential. The effects on female fertility showed a trend toward reversibility. It is not known whether the effects on male fertility are reversible [see Nonclinical Toxicology (13.1) in full Prescribing Information].

**Pediatric Use**

The safety and effectiveness of TAGRISSO in pediatric patients have not been established.

**Geriatric Use**

One hundred eighty-seven (45%) of the 411 patients in clinical trials of TAGRISSO were 65 years of age and older, and 54 patients (13%) were 75 years of age and older. No overall differences in effectiveness were observed based on age. Exploratory analysis suggests a higher incidence of adverse effects [see Nonclinical Toxicology (13.1) in full Prescribing Information].

**Renal Impairment**

No dose adjustment is recommended in patients with mild [creatinine clearance (Clcr) 60-89 mL/min, as estimated by the Cockcroft Gault method (CG)] or moderate (Clcr 30-59 mL/min, as estimated by CG) renal impairment. There is no recommended dose of TAGRISSO for patients with severe renal impairment (Clcr <30 mL/min) or end-stage renal disease [see Clinical Pharmacology (12.3) in full Prescribing Information].

**Hepatic Impairment**

No dose adjustment is recommended in patients with mild hepatic impairment [total bilirubin less than or equal to upper limit of normal (ULN) and AST greater than ULN or total bilirubin between 1.0 to 1.5 times ULN and any AST]. There is no recommended dose for TAGRISSO for patients with moderate or severe hepatic impairment [see Clinical Pharmacology (12.3) in full Prescribing Information].

**Distributed by:**

AstraZeneca Pharmaceuticals LP, Wilmington, DE 19805

TAGRISSO is a trademark of the AstraZeneca group of companies ©AstraZeneca 2015

**Iss. 09/16 3296602 10/16**

---

**Table 2: Adverse Reactions (>10% for all NCI CTCAE Gr 3-4) in Study 1 and Study 2**

<table>
<thead>
<tr>
<th>Adverse Reaction</th>
<th>TAGRISSO N=411</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>All Grades</td>
</tr>
<tr>
<td>Gastrointestinal disorders</td>
<td>%</td>
</tr>
<tr>
<td>Diarrhea</td>
<td>42</td>
</tr>
<tr>
<td>Nausea</td>
<td>17</td>
</tr>
<tr>
<td>Decreased appetite</td>
<td>16</td>
</tr>
<tr>
<td>Constipation</td>
<td>15</td>
</tr>
<tr>
<td>Stomatitis</td>
<td>12</td>
</tr>
<tr>
<td>Skin disorders</td>
<td></td>
</tr>
<tr>
<td>Rash&lt;sup&gt;a&lt;/sup&gt;</td>
<td>41</td>
</tr>
<tr>
<td>Dry skin&lt;sup&gt;b&lt;/sup&gt;</td>
<td>31</td>
</tr>
<tr>
<td>Nail toxicity&lt;sup&gt;c&lt;/sup&gt;</td>
<td>25</td>
</tr>
<tr>
<td>Pruritus</td>
<td>14</td>
</tr>
<tr>
<td>Eye Disorders</td>
<td></td>
</tr>
<tr>
<td>Respiratory</td>
<td></td>
</tr>
<tr>
<td>Cough</td>
<td>14</td>
</tr>
<tr>
<td>General</td>
<td></td>
</tr>
<tr>
<td>Fatigue</td>
<td>14</td>
</tr>
<tr>
<td>Musculoskeletal</td>
<td></td>
</tr>
<tr>
<td>Back pain</td>
<td>13</td>
</tr>
<tr>
<td>Central Nervous System</td>
<td></td>
</tr>
<tr>
<td>Headache</td>
<td>10</td>
</tr>
<tr>
<td>Infections</td>
<td></td>
</tr>
<tr>
<td>Pneumonia</td>
<td>4</td>
</tr>
<tr>
<td>Vascular events</td>
<td></td>
</tr>
<tr>
<td>Venous thromboembolism&lt;sup&gt;a&lt;/sup&gt;</td>
<td>7</td>
</tr>
</tbody>
</table>

<sup>a</sup> NCI CTCAE v4.0

<sup>b</sup> Includes cases reported within the clustered terms for rash adverse events: Rash, rash generalized, rash erythematous, rash macular, rash maculo-papular, rash papular, rash pustular, erythema, folliculitis, acne, dermatitis and acneform dermatitis.

<sup>c</sup> Includes nail disorders, nail bed disorders, nail bed inflammation, nail bed tenderness, nail discoloration, nail disorder, nail dysplasia, nail infection, nail ridging, onychodystrophy, onycholysis, onychoschizia, paronychia.

**Table 3: Laboratory Abnormalities (>20% for all NCI CTCAE Grades) in Study 1 and Study 2**

<table>
<thead>
<tr>
<th>Laboratory Abnormality</th>
<th>Change from Baseline to Grade 3 (All Grades (%)</th>
<th>Change from Baseline to Grade 4 (All Grades) (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Clinical Chemistry</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hypokalemia</td>
<td>26</td>
<td>3.4</td>
</tr>
<tr>
<td>Hypomagnesemia</td>
<td>20</td>
<td>0.7</td>
</tr>
<tr>
<td>Hematologic</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lymphopenia</td>
<td>63</td>
<td>3.3</td>
</tr>
<tr>
<td>Thrombocytopenia</td>
<td>54</td>
<td>1.2&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Anemia</td>
<td>44</td>
<td>0.2</td>
</tr>
<tr>
<td>Neutropenia</td>
<td>33</td>
<td>3.4</td>
</tr>
</tbody>
</table>

<sup>a</sup> The only grade 4 laboratory abnormality was 1 patient with grade 4 thrombocytopenia.
The link between HPV and cancer

40 is the number of HPV strains.

12 is the number of HPV strains that can cause cancer.

17,600 women are affected by HPV-causing cancers each year in the U.S.

9,300 men are affected by HPV-causing cancers each year in the U.S.

11,000 women in the U.S. get cervical cancer each year.

HPV strains 16 and 18 cause 70% of all cervical cancers.

HPV strains 16 and 18 can be prevented with HPV vaccines.

Two doses


Immunization

CDC recommends only two HPV shots for younger adolescents. The Centers for Disease Control and Prevention (CDC) has recommended that 11-12 year olds receive two doses of human papillomavirus (HPV) vaccine at least six months apart, rather than the previously recommended three doses, to protect against cancers caused by HPV infections. Teens and young adults who start the series later, at ages 15 through 26 years, will continue to need three doses of HPV vaccine to protect against cancer-causing HPV infection.

The Advisory Committee on Immunization Practices (ACIP), a panel of experts that advises the CDC on vaccine recommendations in the U.S., voted to recommend a two-dose HPV vaccine schedule for young adolescents. CDC Director Thomas Frieden, MD, MPH, approved the committee’s recommendations shortly after the vote.

The CDC and ACIP reviewed data from clinical trials showing that two doses of HPV vaccine in younger adolescents (aged 9 to 14 years) produced an immune response similar to or higher than the response in young adults (aged 16 to 26 years) who received three doses.

Generally, preteens receive HPV vaccine at the same time as whooping cough and meningitis vaccines. Two doses of HPV vaccine given at least six months apart at ages 11 and 12 years will provide safe, effective, and long-lasting protection against HPV-associated cancers. Adolescents ages 13 and 14 are also able to receive HPV vaccination on the new two-dose schedule.

The CDC will provide guidance to parents, healthcare professionals, and insurers on the change in recommendation. On October 7, 2016, the U.S. Food and Drug Administration (FDA) approved adding a two-dose schedule for 9-valent HPV vaccine (Gardasil 9) for adolescents ages 9 through 14 years. The CDC is encouraging clinicians to begin implementing the two-dose schedule in their practice.

Diagnostics

Should men get a PSA test for prostate cancer? When the U.S. Preventative Services Task Force (USPSTF) recommended against prostate-specific antigen screening for prostate cancer in 2012, researchers began studying what effect this would have on diagnosing and treating prostate cancer in medical practices nationwide.

Last month, JAMA Surgery online reported a significant decrease in prostate biopsies and prostate cancer surgeries since the USPSTF’s recommendation. As recently as the 1990s, population-based screening programs were widely implemented in the U.S.

Jason Hafron, MD, a urologist and surgeon at Michigan-based Beaumont Health, insists that PSAs remain a valuable tool in diagnosing prostate cancer in certain men, and patients and their physicians should continue to discuss the risks and benefits of PSA screening. He adds that new technology, such as the use of biomarkers, advanced MRI imaging, and genetic testing, are also beneficial in preventing and treating prostate cancer.

Adherents of the PSA blood test say that men who might benefit include those who are ages 55 to 69, have a family history of prostate cancer, are African-American, or have had an abnormal result from a prostate exam.

“Urologists are now focusing on refining and identifying prostate cancer that is potentially lethal and should be treated,” says Hafron. “I agree we need to be smarter and selective of who we are screening and when, but we should not stop using PSA altogether as recommended by the USPSTF.”

Genetics/Genomics

Genetic risk factor for binge eating discovered. Researchers have identified a gene (CYFIP2) associated with binge eating. This finding represents one of the first examples of a genome-wide significant genetic factor to be identified for binge eating in model organisms or humans.

In addition, the researchers discovered a network of downregulated genes involved in myelination (the process of forming a sheath around a nerve fiber to allow nerve impulses to move quickly) that also was associated with binge eating. These findings, which appear online in the journal Biological Psychiatry, could potentially lead to treatments targeted to normalize eating behaviors.

Using gene mapping and gene validation, researchers from Boston University School of Medicine were able to identify cytoplasmic FMR1-interacting protein 2 (CYFIP2) as a major genetic risk factor for binge eating. In addition, they observed that decreased myelination could be a neuropathological consequence of binge eating.

“Because we found changes in the brain as a consequence of binge eating that were predictive of decreased myelination, therapeutically promoting remyelination may represent a novel treatment avenue for promoting recovery from negative feeding behaviors in eating disorders,” explains corresponding author Cameron Bryant, PhD.

Bryant and his colleagues believe these findings may lead to new therapeutic treatments which could ultimately save lives and restore healthy eating behaviors in conditions such as compulsive overeating, bulimia nervosa, anorexia nervosa, and even substance use disorders.

Correction: In the Continuing Education article published in the November 2016 issue of MLO, “Best Practices for influenza testing: Methodologies and implementation,” by Dr. Robert L. Sautter, please note that the last sentence of the paragraph beginning “Molecular assays are increasingly used in flu testing…” (page 14) should have been deleted. It was left in due to an editing error. Here is the sentence that should have been deleted: The system using RTPCR is able to discriminate among flu A subtypes (H1N1, H3N2 and H1N1/2009) and thus can help identify the presence of novel flu A subtypes.”
Clinical laboratory customers know that when a control is manufactured by Streck, they can rely on its performance. This reputation for excellence is the driving force behind Streck’s urinalysis control products.

**UA-Cellular® Complete**
Comprehensive cellular component and chemistry analyte urinalysis control designed specifically for the Siemens CLINITEK Atlas® and Sysmex® UF-1000i™ combined urinalysis system.

**UA-Cellular® for iQ**
The control thoroughly evaluates the ability of the iQ® 200 to both identify and quantify white blood cells, red blood cells, non-squamous epithelial cells and crystals.

**UA-Cellular® for UF**
Cellular urinalysis control product that contains components at two clinically significant levels. The product evaluates the ability of the Sysmex® UF-1000i to identify and quantify white blood cells, red blood cells, epithelial cells, bacteria, casts and crystals.

Call today to request a free sample.
4. Summarize the findings of an analysis that was performed
3. Explain ideal methods for accurate urinalysis testing.
2. Describe interferences with urine chemistry elements that
1. Discuss the advancement of automation in the urinalysis test.

Upon completion of these articles, the reader will be able to:

To earn CEUs, see test on page 16 or online at www.mlo-online.com under the CE Tests tab.

LEARNING OBJECTIVES

1. Discuss the advancement of automation in the urinalysis test.
2. Describe interferences with urine chemistry elements that cause erroneous results.
3. Explain ideal methods for accurate urinalysis testing.
4. Summarize the findings of an analysis that was performed using spun versus unspun urine when reviewing automated flags from a urine analyzer.

Rediscovering urine chemistry—and understanding its limitations

By Michelle D. Dumonceaux, BS, MBA, and Mirta Gamez, BS, MT(ASCP) CLS

Routine urinalysis is a cost-effective, non-invasive test used as an indicator of health or disease for metabolic and renal disorders, infection, drug abuse, pregnancy, and nutrition. Urine chemistry can be completed in a number of different ways, ranging from manual reading of a visual urine test strip to the use of semi-automated analyzers to loading the sample on a fully automated urine chemistry analyzer. There is one thing that all methods have in common: a urine chemistry reagent strip.

While urinalysis remains a routinely ordered laboratory test, today most of the emphasis focuses on automating urine microscopy to reduce manual, subjective microscopic work. Urine chemistry analysis is viewed by many as a screening tool that can help aid in the diagnosis of some common conditions such as urinary tract infections (UTIs), kidney or liver diseases, or diabetes, among others. It is important to remain focused on urine chemistry and better understand common test interferences.

Urine chemistry reagent strips come in many different configurations, depending on their use. The most common tests include bilirubin, urobilinogen, glucose, ketones, protein, blood, nitrite, leukocyte esterase, and pH. In addition, some manufacturers include urine chemistry reagent pads for specific gravity, ascorbic acid, microalbumin, creatinine, and color. While urine chemistry testing is common, it is important to understand the test and its limitations to ensure accuracy of the test and recognize the factors that can cause incorrect results. Manufacturers have improved urine chemistry analysis by including additional tests to easily identify common interferences.

**Bilirubin**

Bilirubin (BIL) is a waste product of red blood cell (RBC) destruction. The primary source of bilirubin is the daily release of hemoglobin from the breakdown of RBCs in the reticuloendothelial system. In addition, RBC breakdown can occur in the bone marrow or other heme-containing proteins. The liver normally breaks down most of the bilirubin.

Healthy individuals exhibit a “negative” reading, very small amounts (0.02 mg/dL or ≤1 Ehrlich unit) may be found in normal urine. However, the presence of bilirubin can indicate liver dysfunction such as jaundice, hemolytic disease, or obstruction of the bile duct or biliary system. A high amount of bilirubin, especially, affects the brain of newborns.

False positives can be caused by drugs that color the urine red, such as phenazopyridine, or large quantities of chlorpromazine metabolites. False negatives can be caused by the presence of ascorbic acid, increased nitrite concentrations, or improper sample storage.

**Urobilinogen**

Urobilinogen (URO) is a breakdown product of bilirubin. When high concentrations form in the body, the liver may not be able to break down all of the bilirubin present. Urobilinogen is produced in the intestines as bacteria metabolizes bilirubin. Small amounts (≤1 mg/dL or =1 Ehrlich unit) may be found in normal urine. However, the presence of urobilinogen is found with liver dysfunction, excessive destruction of RBC (hemolytic anemia, pernicious anemia and malaria), hepatitis, portal cirrhosis, and congestive heart failure.

Interferences for urobilinogen include formalin, high concentrations of nitrites, and drugs or substances that color the urine. If samples don’t equilibrate to room temperature before testing, that can produce an incorrect result.

**Ketones**

Ketones (KET) are normally not found in urine, but can be present when the body breaks down fat for energy. The body normally obtains energy from carbohydrates. If the carbohydrate supply is reduced, not absorbed properly, or not broken down metabolically, the body will use fat for energy. Ketones are associated with uncontrolled diabetes, vomiting, starvation, fasting, frequent strenuous exercise, and when the body uses fat instead of glucose for energy, which often occurs in people on a high-protein diet.

Agents containing free sulfhydryl groups can cause interference with ketone detection. Highly pigmented urine can result in false positive results, and improper sample or test strip storage may provide false negative results.

**Glucose**

Glucose (GLU) supplies the body with energy. In healthy individuals, glucose is reabsorbed by the kidney tubules and not present in the urine. However, if the concentration of blood glucose becomes too high (160-180 mg/dl), then the tubules can no longer reabsorb glucose and it will pass into the urine. This presence of glucose in the urine is called glycosuria. It is often associated with endocrine disorders such as diabetes, kidney impairment, central nervous system
damage, and pancreatic disease. Other conditions associated with glycosuria include burns, infections, and fractures. Glycosuria is also associated with pregnancy.

High concentrations of ketones, decreased urine sample temperature, and increased specific gravity affect the sensitivity of the glucose pad. Increased ascorbic acid can also pose an interference. Bacterial glycolysis can occur with improper storage and can provide a false negative result.

**Protein**

The presence of protein (PRO) in the urine, otherwise known as proteinuria, is often the first indicator of kidney disease. It can also be indicative of other diseases such as nephrotic syndrome, glomerulonephritis, multiple myeloma, and pre-eclampsia. Exposure to cold, strenuous exercise, high fever, and dehydration can also cause the presence of protein in the urine.

The protein pad is most sensitive to albumin as opposed to other proteins. False positive results can be found with extremely alkaline samples. In addition to protein urine chemistry pads, there are also urine chemistry strips that test for microalbumin and creatinine for further assessment.

**Blood**

Blood (BLD) is not normally present in the urine and may not be visually present. The abnormal presence of RBCs in the urine is called hematuria, and the presence of hemoglobin in the urine is called hemoglobinuria. Blood in the urine is associated with kidney or urinary tract diseases, severe burns, infections, trauma, exposure to toxic chemicals or drugs, pyelonephritis, glomerulonephritis, renal or genital disorders, tumors, transfusion reactions, intravascular hemolysis, and hemolytic anemia. Strenuous exercise and menstruation can cause the presence of blood in the urine. A positive result should be followed up with a microscopic correlation to assess for the presence of RBCs and casts.

Urine specimens must be well mixed to ensure that RBCs have not settled out. Ascorbic acid should be considered an interferent when RBCs are present during a microscopic exam but the blood urine chemistry test is negative.

**Nitrites**

Nitrites (NIT) are consumed in the diet as green vegetables and are normally excreted without nitrite formation. The presence of bacteria in the urinary tract (e.g., bladder, kidney, etc.), can lead to the production of nitrites. Nitrite and leukocyte esterase screening help identify the presence of an infection. This screen should not replace further microscopic examination for bacteria or a culture to identify and quantify the bacteria present. It is used to quickly identify nitrate-reducing bacteria at a low cost.

Proper nitrite screening should be performed on a urine sample collected in the morning or after it has been retained in the bladder for at least four hours. High concentrations of ascorbic acid and improper storage can provide false results.

**Leukocytes**

Normal urine may contain a small number of white blood cells (WBCs) or leukocytes (LEUs). An increase in the presence of leukocyte esterase, an enzyme found in leukocytes, indicates inflammation in the urinary system. A WBC increase can be present with or without bacteruria. If leukocytes are present without bacteria, there is usually a kidney or urinary tract infection (UTI) involving trichomonas, yeast, chlamydia, mycoplasmas, virus, or tuberculosis. A positive nitrite and leukocyte esterase is a good indication for the performance of further microscopic examination.

High glucose, protein, and specific gravity can interfere with the leukocyte-esterase reaction, causing inaccurate results. In addition, specific antibiotics, drugs, and food (beets) can affect the chemical reaction.

**pH**

The kidneys play a major role in maintaining proper pH balance. Urine pH can affect the stability of formed particles in the body. Acidic urine (i.e., 4.5-6.9) is associated with, but not limited to, high-protein diets or the ingestion of cranberries, starvation, severe diarrhea, chronic lung disease, and UTIs with acid-producing bacteria (Escherichia coli) as well as certain medications. Alkaline urine (i.e., 7.0-7.9) is associated with, but not limited to, vegetarian or low-carbohydrate diets, vomiting, hyperventilation, UTIs with urease-producing bacteria, and certain medications. pHs that are below 4.5 should be suspected of adulteration, and pHs that are above 8 are often tied to improperly stored urine specimens.

**Specific gravity**

Specific gravity (SG) is a measure of the density of a urine. The more particles (i.e., salts, glucose, protein, etc.) in a urine, the higher the specific gravity. High specific gravity is caused by dehydration, diarrhea, heart failure, and glucose in the urine (i.e., diabetes). Low specific gravity is caused by kidney failure, diabetes insipidus, renal tubular necrosis, and the intake of too much fluids.

Urine test strips used for visual analysis often have a pH reagent pad. A limitation of the reagent pad is that it only measures the ionic solutions and can be susceptible to pH readings. Fully automated urine chemistry analyzers often use an onboard refractometer to obtain a specific gravity reading. A refractometer can be affected by particle size, temperature, and concentration of the solution as well as light wavelength. Some manufacturers have a specific gravity correction factor for high protein and glucose concentrations.

**Ascorbic acid**

Ascorbic acid, otherwise known as vitamin C, can be found in various foods and supplements. It is also a common interferent with urine chemistry reagent pads. When a urine sample has high levels of ascorbic acid, the reagent pads for blood, glucose, nitrite, and bilirubin may not react properly. This especially interferes with blood measurements at low levels. Clinicians should consider asking whether the patient is taking vitamin C when collecting a urine sample. We see more people taking vitamin C or vitamin C-like substances during the winter months or when traveling by plane, in an effort to boost their immune system.

Not all strip manufacturers have an ascorbic acid detection pad, as ascorbic acid is not commonly reported out. When the sample tests positive for ascorbic acid, the laboratory may append a note with the results identifying potential interferences to the physician.

**Color**

Normal urine ranges from yellow/amber in color to clear or transparent and has a characteristic odor. A change in color, clarity, or odor is not necessarily a sign that something is incorrect. Urine changes color based on the body’s chemistry, food, medication intake, and state of hydration. Below is a list of colors, other than shades of yellow, found during urinalysis testing, along with their associated causes:

- **Orange**: dehydration; certain medications; liver or bile duct issues
- **Blue/green**: dyes in food or for kidney and bladder tests; medications such as amitryptilene, indomethacin (Indocin) and
propofol (Diprivan); familial benign hypercalcemia, also known as blue diaper syndrome; UTIs caused by pseudomonas bacteria
  • Red/pink: UTIs; enlarged prostate; tumors; kidney cysts; long-distance running; kidney or bladder stones; the use of medications such as rifampin (Rifadin, Rimactane) or phenazopyridine (Pyridium); the use of some laxatives; the use of chemotherapy drugs. In addition, eating beets, blackberries, or rhubarb may cause the urine to turn red or pink
  • Brown: liver and kidney disorders; UTIs; extreme exercise; ingesting large amounts of certain foods (e.g., fava beans, rhubarb, or aloe); medications such as the antimalarial drugs chloroquine and primaquine, antibiotics metronidazole (Flagyl) and nitrofurantoin
  • Cloud/murky: urinary tract infection (UTI)

Urine color can interfere with some of the aforementioned tests during the color reaction process that takes place on the pad. For this reason, some manufacturers have a “blank” or color compensation pad on the dipstick. This color compensation pad will identify the color of the urine, and the analyzer will “subtract” the color from other readings to provide a more accurate result.

The lab’s perspective

As noted above, specimen storage is a concern for a number of tests. Most manufacturers require testing within one to two hours of collection. If this is not feasible, samples are often refrigerated or stored in a preservative tube for testing at a later date. It’s important to note that few manufacturers have validated the use of preservative tubes for analysis on their urine analyzers, so lab leaders should assess their needs before purchasing a system.

Michelle D. Dumonceaux, BS, MBA, serves as Marketing Manager for Beckman Coulter. Michelle has more than 18 years of experience in marketing, quality systems, and technical support in medical devices and diagnostics.

Mirta Gamez, BS, MT(ASCP) CLS, serves as a Hematology Product Specialist for Beckman Coulter Diagnostics. She has worked in the lab industry for over 16 years in academic research, boutique, and mid- and high-volume hospital labs, and has managed hematology, molecular, and flow cytometry departments.

In summary, urinalysis remains an informative laboratory test. It is important to understand what is being tested and what can interfere with the test, since certain medications and vitamins interfere with urinalysis testing. For example, during the winter months, more and more people are taking vitamin C in an effort to “starve a cold,” and we see ascorbic acid as an interferer in bilirubin, glucose, blood, and nitrite testing. It is also important to understand the patient’s diet and exercise level, since they can impact results as well. Laboratorians should become very familiar with the manufacturer’s instructions for use to know what the limitations of the analyte are in order to ensure accurate reporting.

Introducing e-Learning for PD-L1 IHC 28-8 pharmDx.

The PD-L1 IHC 28-8 pharmDx interpretation training is a comprehensive online program focused on helping pathologists:
  • Understand the role of immune checkpoint pathways
  • Learn about technical considerations for optimal slide staining results
  • Learn how to evaluate non-squamous NSCLC or melanoma specimens stained with PD-L1 IHC 28-8 pharmDx

Intended Use For in vitro diagnostic use
PD-L1 IHC 28-8 pharmDx is a qualitative immunohistochemical assay using Monoclonal Rabbit Anti PD-L1. Clone 28-8 is intended for use in the detection of PD-L1 protein in formalin-fixed paraffin-embedded (FFPE) non-squamous non small cell lung cancer (NSCLC) and melanoma tissue using Envision FLEX visualization system on Automation Link 48. PD-L1 protein expression is defined as the percentage of tumor cells exhibiting positive membrane staining at any intensity.

Discover e-Learning at dakocom/us/PDL1288-learning
Urinalysis: Are we done spinning yet?

By Roy Midyett, CLS

There are two laboratory tests that have remained basically unchanged for 100 years: manual blood smear differential and urine microscopic analysis. The basic method for the urinalysis is the same as it has always been: centrifuge a standard volume of specimen; decant; examine the remaining sediment microscopically; then identify and enumerate what is seen. There have been slight refinements in such things as the sample volume, the centrifuge times, pipettes, slides, etc. But the basic premise has been unchanged, even with the introduction of automated urine particle analysis.

How we do it now

Many labs now rely on some automation, in the form of flow cytometric microscopic analysis, for the identification and enumeration of urine elements. The majority of automated results are acceptable without further checking or verification, but there will be a percentage of results that, depending on things such as the limits set by the user and the types of specimens, will require manual verification. For flow cytometric analyzers, five elements are identified and counted: WBCs, RBCs, squamous epithelial cells, bacteria, and hyaline casts. Other elements may be detected with user-set flags, including yeast, pathological casts, crystals, and others. Because it is flow cytometry, it is very much like an automated hematology analyzer differential, which reports a five- or six-part differential, and other cells, such as blasts and atypical lymphs, indicated as suspect flags. In both cases, the operator has control over how sensitive these flags are, and even whether to use the flags at all.

In automated flow cytometry of urinalysis, the three elements responsible for most manual microscopic checks are yeast, pathological casts, and hyaline casts. Hyaline casts, one of the five reported elements, benefit from a high-limit check, since the analyzers tend to overcount them. The other four parameters are accurate enough that no limits need be set. If a flag limit is exceeded, the report displays a review flag, and the presence and amount may need to be verified microscopically.

A cursory exam to simply verify the presence or absence of flagged elements is different from doing a complete manual microscopic, where there is no other reference than the strip result. This is analogous to doing a peripheral smear slide check on the automated differential, where, rather than do a 100-cell count, you quickly estimate the accuracy of the flagged result. If a full microscopic isn’t needed on an automated urinalysis result, is a spun sediment really necessary? For verifying automated results, can the spun microscopic be eliminated, and can unspun urine be used to check the results?

A better way? An experiment.

In our lab, we wanted to see if using unspun urine to verify flags for yeast, hyaline casts, and pathological casts would give reliable results, as compared to using the traditional spun urine sediment. We started this study with upper flagging limits based on two years of experience and recommendations from the manufacturer.

• Yeast 4.5/hpf
• Hyaline casts 12/lpf
• Pathological casts 7.4/lpf

These limits have given us a moderate false positive rate, which is preferable to missing too many positives. With these limits, we analyzed 60 samples with any of the three flags, and some samples had more than one flag. Out of 60 specimens, we had 22 true positives, and these are the ones we used to determine if the unspun urine gave different results than the spun sediment. (The false positives were of no use, since neither the spun nor unspun had any of the flagged elements, at least in numbers that we could detect).

As a rule of thumb, the estimate on the unspun urine was multiplied by 10 to extrapolate the results to the spun results. For example, if the unspun result showed two hyaline cast/lpf, the spun results should be around 20/lpf.

We confirmed what we already knew, that sometimes spinning the specimen actually works against identifying elements if the field is too crowded. And if a sample had very high levels of an element, these would be visible easily on unspun urine. It was the moderate and smaller levels that were of more concern.

We always examined the unspun sample first, to avoid bias from the spun sediment, although bias could work both ways: examining a positive spun sediment first might make one look more closely than usual to find something on the unspun no matter what, but looking at a negative unspun sample first might cause one to be less thorough on the spun sample. We decided to check the unspun first for consistency.

In every case where the flag was correct, the element could be seen in the unspun sample before finding it in the spun sediment. Therefore, our data indicates that, using careful criteria, the spun microscopic didn’t give us any better information than using an unspun specimen, and may be unnecessary when simply confirming presence of flagged elements.

Some further takeaways

It is important to note that the lower your flag limits, the more discrepancy you will find between spun and unspun results, because the elements will be harder to find in smaller numbers, and you will be flagging for increasingly insignificant numbers. Obviously setting the yeast flag at 2 per hpf is going to overflag; you will virtually never see yeast in the unspun sample, and you will only rarely see it on the spun. Setting reasonable limits, as in hematology automated differentials, is crucial to this system working.

We used this data as an opportunity to revise our flagging limits, with the goal of reducing the false-positive (FP) rate. This would mean raising the limits if the data so indicated. We took the lowest result that gave a true positive and adjusted the flag limit up to or just below that number. Based on the data, we were able to raise all three levels:

• Yeast 8/hpf
• Hyaline casts 15/lpf
• Pathological casts 10/lpf

This lowered our review rate slightly, from 20 percent to about 17 percent.

The implementation of any new data and limits should be made by an experienced laboratorian, with support from pathologist or medical staff, and with the recommendation from the instrument company specialists. Even after deciding on initial criteria, there will be some period of actually testing the numbers in “real life.”

Clinical laboratory professionals get a feel for whether they are doing too many manual microscopics or too many blood smear slide checks. If you think that is happening, it probably is. Most labs tailor slide check criteria too narrowly, and probably do too many urine microscopics as well. Treating an unspun urine check like a peripheral blood smear check can save time, labor, and money.
TEST QUESTIONS

1. Urine chemistry tests can be completed in a number of ways that include manual testing, semi-automated, and fully automated.
   a. True
   b. False

2. According to the article, what is the main reason behind the interest in automating urine microscopy?
   a. To reduce manual, subjective results
   b. To decrease the turnaround time of the test
   c. To reduce the workload of techs performing the test
   d. None of the above

3. The most common tests in a urine reagent dipstick are bilirubin, urobilinogen, glucose, ketones, protein, blood, nitrite, leukocyte esterase, pH, and ascorbic acid.
   a. True
   b. False

4. False positive bilirubin tests will occur from drugs that color the urine.
   a. green
   b. blue
   c. red
   d. all of the above

5. What must be done to ensure that an incorrect result for urobilinogen will not be reported?
   a. The pH of the specimen should be made neutral before testing.
   b. The specimen should be refrigerated before testing.
   c. The specimen should be allowed to equilibrate to room temperature before testing.
   d. The specimen should be centrifuged before testing.

6. Which process allows for the detection of ketones in urine testing?
   a. When the body uses fat for energy
   b. When the body uses carbohydrates for energy
   c. When the body uses protein for energy
   d. When the body uses minerals for energy

7. What type of urine will cause a false positive result for ketones?
   a. highly acidic urine
   b. highly alkaline urine
   c. highly pigmented urine
   d. urine with a high specific gravity

8. Factors that will affect the sensitivity of the glucose pad include all but:
   a. high concentrations of nitrites.
   b. decreased urine temperature upon testing.
   c. high concentration of ketones.
   d. increased specific gravity.

9. Which positive test on the urine chemistry strip is the first indicator of kidney disease?
   a. leukocytes
   b. nitrites
   c. glucose
   d. protein

10. A positive chemistry strip result for blood should always be followed up with microscopic correlation to assess for the presence of red blood cells and casts.
    a. True
    b. False

11. What is considered an interferent when red blood cells are present during microscopic exam but negative on the urine chemistry strip?
    a. hemoglobin
    b. improper storage of specimen
    c. phenazopyridine
    d. high concentration of nitrites

12. Proper nitrite screening of urine should be on a specimen that has been retained in the bladder for at least
    a. 30 minutes.
    b. 1 hour.
    c. 3 hours.
    d. 4 hours.

13. Which three elements can interfere with the leukocyte esterase reaction on the chemistry strip?
    a. high protein, pH, and ketones
    b. high protein, pH, and specific gravity
    c. high glucose, protein, and pH
    d. high glucose, protein, and specific gravity

14. Urine pH below 4.5 is often tied to improper storage of specimens, and urine pH above 8 is suspected of adulteration.
    a. True
    b. False

15. Specific gravity is a measure of
    a. the pH of the urine specimen.
    b. the density of the urine specimen.
    c. the cloudiness of the urine specimen.
    d. none of the above

16. If a urine specimen has a high level of this, the reagent pads for blood, glucose, nitrite, and bilirubin may not react properly.
    a. phentazopyridine
    b. antibiotics
    c. ascorbic acid
    d. none of the above

17. What have some manufacturers done to their dipstick pads to correct for brilliant urine colors?
    a. an indice measurement for color
    b. a dye to neutralize the color
    c. a blank or color compensation pad
    d. none of the above

18. If prompt urinalysis testing is delayed, a specimen should be refrigerated and stored in a preservative to protect the outcome of the results.
    a. True
    b. False

19. The purpose of the study performed at Presbyterian Hospital was to
    a. eliminate the manual review of all microscopic urine elements.
    b. assess the accuracy of the microscopic elements identified on the urine analyzer.
    c. assess whether spun urine is necessary to manually verify certain microscopic elements.
    d. none of the above

20. The conclusion of the Presbyterian Hospital study was that it is necessary to centrifuge urine specimens in order to manually confirm the presence of certain microscopic elements.
    a. True
    b. False
AIDING IN THE DIAGNOSIS OF URINARY TRACT INFECTIONS

iRICELL combines urine chemistry and microscopy testing in an automated platform. Its urine culture candidate report streamlines the UTI testing workflow. With user-defined thresholds, the report generates work lists of possible patient sample values for review and possible release or reflex to microbiology for urine culture analysis. The unique urine culture indicator checklist combines chemistry nitrite and leukocyte esterase results with urine microscopy bacteria, WBC and small particle counts. iRICELL improves laboratory productivity so you can improve patient care.

Discover more at www.beckmancoulter.com/urinalysis
A blood test for early detection of lung cancer

By Laura J. Peek, PhD, and James R. Jett, MD

An immune response can reveal cancer. As a normal cell undergoes tumorigenesis, it expresses aberrant protein antigens, known as tumor-associated antigens (TAAs). They are recognized as non-self by the immune system, thereby triggering a B-cell response. The production of autoantibodies by B cells is not dependent on the size of a tumor and can occur throughout all stages of the disease. Most important, autoantibodies are produced early in tumorigenesis, when just a few antigens may stimulate a prolific immune response.1 Autoantibodies, therefore, can serve as an early cancer marker.

Biomarkers for lung cancer
A blood test that measures a panel of seven autoantibodies associated with lung cancer is available to clinicians to further assess a patient’s risk of lung cancer being present.2-5 The test provides additional insight into the patient’s lung cancer risk beyond self-reported risk factors, such as age, smoking history, exposure to carcinogens (e.g., radon, asbestos), etc. A tube of blood is drawn from the patient, and the isolated serum is sent to a CLIA-certified laboratory, where autoantibodies are measured utilizing a proprietary platform technology based on indirect enzyme-linked immunosorbent assay (ELISA) principles.

The test results
Results are reported for each autoantibody as well as an overall test result.2-5 Two clinically-derived cut-offs for each autoantibody are utilized to determine whether a patient’s autoantibody levels are low, moderate, or high.2 A low level of autoantibodies does not rule out lung cancer; it simply means that the patient’s lung cancer risk has not changed appreciably from what was estimated by his or her self-reported risk factors.2 Conversely, patients with moderate or high levels of any one or more autoantibodies are at increased risk of having lung cancer, which may warrant additional testing consistent with the patient’s history and overall risk profile.3

Clinical validation
Clinical validation of the autoantibody test utilized serum from three cohorts of newly diagnosed lung cancer patients (n=655) and serum from risk-matched control patients. (Controls were not diagnosed with cancer and were risk-matched to lung cancer cases by gender, age, and smoking history.) The studies demonstrated that the autoantibody test detects all types and all stages of lung cancer equally.5 The panel of autoantibodies was subsequently changed from six to seven autoantibodies, which improved accuracy to 92 percent, PPV (positive predictive value) to one in eight (assumes lung cancer prevalence 2 percent) and specificity to 91 percent at 41 percent sensitivity.2 A second cut-off for each autoantibody was implemented to stratify the positive autoantibody results, with the primary advantage being the enhancement in specificity and PPV with a high level result to 98 percent and one in four (assumes lung cancer prevalence 2.7 percent), respectively.2 The performance characteristics demonstrated by these studies support the test’s utility in helping physicians identify which patients are at highest risk of having a lung cancer. (Note: The test is a rule-in test and cannot be used to rule-out a patient having lung cancer.) A clinical audit was performed on 1,613 U.S. patients whose physicians had ordered the autoantibody test due to their patient’s high risk for lung cancer. Patients provided HIPAA authorization to release their medical records for the clinical audit. Records regarding cancer diagnosis were reviewed six months following testing for all patients with a positive or negative test result. Sixty-one patients (four percent) were identified with lung cancer, 25 of whom had a positive autoantibody test (sensitivity 41 percent). A positive autoantibody test was associated with a five-fold increase in risk of lung cancer versus a negative test. In the lung cancer patients with a positive autoantibody test, where stage was known, 8 of 14 (57 percent) were Stage I or II. The clinical audit also confirmed a highly statistically significant improvement in specificity of the seven-autoantibody panel and demonstrated that the autoantibody test performs the same in routine clinical practice as was shown in the case-control validation studies.3

In another clinical audit study, the autoantibody test was evaluated in 296 high-risk patients with non-calcified pulmonary nodules (lung cancer prevalence 25 percent). The patient’s lung cancer risk was calculated using the Swensen/Mayo Clinic Nodule Calculator.4 A positive autoantibody test resulted in a greater than two-fold increased relative risk of lung cancer.5 Using a “both positive rule” of combining binary tests, adding the autoantibody test to the Swensen risk model improved the diagnostic performance with high specificity (> 92 percent) and positive predictive value (> 70 percent). Accordingly, a positive autoantibody test reflects a significant increased risk of lung cancer in non-calcified nodules 4-20mm in largest diameter.7

Prospective trials
Currently, there are two large prospective trials underway evaluating the autoantibody test. In Scotland, the National Health Service (NHS) is conducting a randomized prospective trial of 12,000 individuals at high-risk for lung cancer. Participants are randomized, with half getting the autoantibody test and half not. Those with a positive autoantibody test are followed-up with a chest x-ray and CT chest scan. (Low-dose CT chest scans are not approved for screening in Scotland.) The goal of the study is to determine if screening continued on page 21
High-Quality, Low-Cost Reagents

Immunoassay Reagents for chemistry analyzers™

Over 30 different assays available

Call today and mention this ad for a free sample!

KAMIYA BIOMEDICAL COMPANY  diagnostics@k-assay.com  |  800-KAMIYA-5

www.k-assay.com/MLO.php
R egular readers of this column will know that there are two main categories of nucleic acids—DNA and RNA. They’ll also know that while for living organisms DNA acts as the genetic data storage center, RNA has a messenger role (mRNAs, transcribed from DNA to direct protein synthesis). Most will also recall that there are other classes of RNA molecules, particularly tRNAs (used to tag and identify amino acids for protein synthesis) and rRNAs (structural components of the ribosome, the cellular “machinery” for protein synthesis). In addition to these, there’s increasing interest in the molecular diagnostics community in a less widely known but no less common RNA form, the microRNA or miRNA. Where do miRNAs come from, how do we analyze them, and what do they have the potential to tell us about the health of patients?

Meet the miRNA

miRNAs are, as the name implies, tiny. They’re generally about 22 nucleotides in length and are most commonly coded for in “non-coding” regions of the DNA (non-coding for protein, that is). These regions can include introns within coding genes, or they can exist buried within other longer noncoding RNA molecules. In either case, the miRNA encoding region is marked by having an adjacent, self-complementary sequence such that after it is transcribed from DNA to a single-stranded RNA precursor, it can fold back on itself to create a double-stranded hairpin configuration. Well-conserved cellular machinery, including the critical cytosolic enzyme Dicer, then act to cut out the miRNA from this precursor and release it in its active, single-stranded form.

In their active form, miRNAs act by binding to fully or partially complementary sequences within mature mRNAs. This in turn acts as a negative regulator on the mRNA being translated to functional protein, by one of several mechanisms: it can interfere with and slow down translation of the mRNA to protein, or it can destabilize the mRNA and trigger its degradation. Expression of an miRNA thus acts as a layer of post-transcriptional negative control on gene expression, and it turns out to be a very common one.

miRNAs are found and well conserved (that is, have genetically highly stable sequences over evolution) in both plants and animals, and even in some viruses. The ubiquity of this regulatory mechanism in humans can be best appreciated by considering that more than 5,500 unique miRNAs have been identified as being expressed, many in a tissue- and developmental stage-specific context. As miRNAs do not require target mRNA complementarity and can exist over their entire 22 bp length to function (a run of as little as six to eight bp can allow for interaction and miRNA regulation, meaning a single miRNA may interact with multiple targets), it becomes apparent that with only approximately 25,000 total human coding genes, many can be subject to miRNA control. In fact, current estimates suggest that nearly 60 percent of all human coding genes are regulated to some extent via miRNA.

As regulatory molecules, then, it stands to reason that miRNAs are expressed in controlled patterns based on cell type, developmental stage, and various external signals. Our interest from the MDx perspective, then, comes about from the idea that if miRNAs show particular reproducible expression patterns in healthy cells, disease states such as cancer or infection may lead to detectable perturbations in pattern of miRNAs expressed. With next-generation sequencing (NGS) technologies ideally suited to the identification of large numbers of short nucleic acid molecules simultaneously, we have technologies in hand which can rapidly produce miRNA expression profiles from input samples, giving the identity (and relative quantity) of the miRNAs found. While we may not know the target(s) of each miRNA, we can observe them as biomarkers whereby variations in the usual profile can indicate disease states, or even in some cases provide detailed insight to the disease state when the profile perturbation is itself statistically characteristic of a particular condition, or indicative of a particular suggested treatment strategy. In some cases, just the levels of a few specific miRNAs may be highly informative as biomarkers.

From a technological standpoint, miRNAs are generally observed either in bulk (NGS methods) or by targeted qPCR when only a small known handful are likely to be of interest. In either approach, specialized methods have to be applied to adapt these technologies to work with such short targets. Usually this includes the selective removal of longer nucleic acids from the input material, and the ligation of longer nucleic acid “labels” or “handles” onto the miRNAs. These handles then serve as the basis from which to perform sequencing or qPCR, with the miRNA body being selectivity amplified or detected.

With that background on what miRNAs are and what lab methods are used to detect and quantify them, let’s briefly consider a few examples of how they are now coming into clinical utility.

Cardiovascular disease

A common and well known test for acute cardiac injury is the presence of circulating cardiac-specific troponin T. Damage to the cardiac muscle cells leads to membrane permeability and thus leakage of this marker into peripheral blood. A similar approach can be taken based on the observation that some miRNAs are almost exclusively expressed in cardiac muscle. MicroRNAs miR-499-3p, miR-1, miR-133a, miR-208b, and miR-499 appear to be the most promising in this application, with improved predictive value as compared to troponin T testing. Readers interested in more detail on this application are directed to Reference 2, a recent review.

Oncology

Since miRNAs have a regulatory role, it stands to reason that their appearance and levels are likely to be perturbed in states of deregulation of normal cellular development, such as tumors. Indeed, studies have indicated that those miRNAs shown to repress known tumor suppressor genes are often amplified in cancers, while those shown to repress proto-oncogenes are often deleted. Different tumor types have been observed to have distinctive miRNA expression profiles. One study utilizing a panel of just 48 selected miRNAs demonstrated accurate classification of root cancer type even in metastatic sites in nearly 80 percent of cases.

In addition to classifying cancer based on type and tissue of origin, some attempts...
have been made to utilize miRNA profiling to classify subtypes of cancers (such as luminal versus basal breast cancer, or squamous versus non-squamous non-small cell lung cancer). miRNAs have also shown promise as biomarkers in the early detection of some cancers; in the case of ductal adenocarcinoma, one study has indicated that detectable overexpression of just two markers (miR-21, miR-205) is both diagnostic and detectable before overtly visible phenotypic changes detectable by traditional histopathology.

Additionally, work has been done on using miRNA profiling as a means to classify cancers based on likelihood of response to a given therapeutic approach; an example of this is in hepatocellular carcinoma, where suppressed levels of miR-26 has been suggested as a marker for good response to interferon-α treatment. Readers interested in more on this category of miRNA application (and a more detailed look at the history of the discovery of, biogenesis of, and modes of biological action of miRNAs) are directed to Reference 3 as a good starting point.

**Infectious diseases**

Infections represent another situation where abnormal cell function may become apparent through alterations in normal miRNA expression profiles. For instance, chronic hepatitis B as the underlying cause of liver failure has been shown to be detectable at 85 percent sensitivity and 70 percent specificity—less than ideal as a diagnostic, but a promising start—by analysis of just 10 miRNAs in peripheral blood. Analysis of just three miRNAs in peripheral blood (miR-361-5p, miR-889, and miR-576-3p) was demonstrated in another study as approaching 90 percent specific for the detection of pulmonary tuberculosis as distinct from other infections or uninfected controls. These few examples highlight the diversity of applications to which miRNA profiling, following sufficient validation as biomarkers in particular instances, may be usefully applied. Since this class of molecules can be detected and characterized with existing MDx lab equipment—in some of the examples covered here, even with basic real-time qPCR instrumentation—their adoption into routine diagnostic streams has few technical barriers. Validation and regulatory complexities common to all biomarker strategies remain, but as regulatory flows for approving multi-analyte testing become more common, miRNA-based approaches will likely take on many more diverse and useful roles as diagnostic tools for the MDx lab to offer.

**REFERENCES**


**CANCER BIOMARKERS**

with the autoantibody test reduces the number of patients being diagnosed with Stage III or IV lung cancer. This trial completed accrual in June 2016 and follow-up is ongoing.

The second prospective trial, at National Jewish Health in Denver, Colorado, combines screening with low-dose CT chest scan and the autoantibody test. The goal of the study is to determine if screening with both modalities results in a higher rate of detecting early stage lung cancer. To date, more than 1,300 high-risk participants have been enrolled. Encouraging preliminary results of both studies were presented at the World Conference on Lung Cancer in Denver in late 2015.

Currently, only 25 percent to 30 percent of all patients with lung cancer in the United States meet the criteria for lung cancer screening with low-dose CT chest scans. Clearly there is a need for additional tests, such as the autoantibody test described here, to help identify the remaining 70 percent to 75 percent of lung cancers while they are asymptomatic and early stage. Additionally, as the number of patients with non-calciﬁed pulmonary nodules is rapidly growing through CT screening, there is an urgent need for supplemental tests to assist physicians in determining which are malignant and which are benign.

**REFERENCES**


**THE PRIMER**

**CANCER BIOMARKERS**

**CLINICAL ISSUES**

continued from page 18
MALDI-ToF is poised to speed diagnosis for bacterial and fungal infections

By Janet Cromien and Dale Schwab, PhD

Helping clinicians quickly and effectively address the medical concerns of their patients should be the ultimate goal of any clinical laboratory. Delivering on this goal can be especially important when a patient presents with a bacterial or fungal infection. These infections, ranging from methicillin-resistant Staphylococcus aureus (MRSA) cellulitis to Gram-negative sepsis, can manifest rapidly, causing significant disease and even death within hours. Matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-ToF) is being embraced by laboratories primarily in clinical microbiology because it is a fast, accurate, and cost-effective method for identifying bacteria, fungi, and other infectious agents isolated from culture. In another study, Tan et al also found that the MALDI- ToF protocol yielded results more quickly than the standard protocol for most isolates when assessed by organism groups, leading to better patient outcomes. In addition to bettering patient outcomes through earlier diagnosis, MALDI-ToF technology may have future applications in predicting antibiotic resistance. For instance, a third recent study demonstrated that MALDI-ToF can detect the presence of carbapenemases, a type of enzyme which promotes drug resistance to standard antibiotics. This technique, which can identify drug resistance in hours, could help physicians determine which antibiotic to prescribe. This future benefit may be of consideration for some laboratories.

Benefits and limitations

There are many benefits to incorporating MALDI-ToF to aid in identification of microorganisms. Identification with MALDI-ToF is a relatively simple process. Once a slide has been prepared, it is introduced into a high-vacuum environment, where the sample is ionized with a laser burst that releases proteins. These proteins are then accelerated using an electric charge, and the time of flight is recorded. (Lighter proteins travel faster, while heavier proteins travel more slowly.) At the end of the treatment, the proteins are detected with a sensor. The sensor then creates a spectrum representing the protein makeup for each sample. Identification is made when the spectrum from a sample is compared against a database of spectra of characterized bacteria, yeast, and mycobacteria. The process can reduce the time needed for identification and diagnosis from days to hours.

There are, however, some limitations to MALDI-ToF. Some organisms may require repeat analysis and additional processing, and some closely related organisms are not differentiated and may cause challenges with identification. The technique is also generally not useful for direct testing of clinical specimens. Finally, the initial purchase price of the equipment is high, which may limit the types of labs that can adopt it.

The easiest way to see the value of MALDI-ToF for patient care is to think of it in terms of specific life-threatening infections where the time from initial infection to diagnosis, to treatment is highly correlated to patient morbidity. Sepsis is a life-threatening condition that presents itself as a fever, increased heart rate and breathing, and pneumonia-like symptoms. These symptoms can be confused with other conditions, which may delay diagnosis and care. Sepsis is typically treated with intravenous fluids and broad-spectrum antibiotics. Antibiotics are given as soon as possible to avoid septic shock and death. However, delayed diagnosis can delay care, worsening patient outcomes.

Taking the long view

After weighing the benefits and limitations of MALDI-ToF, an increasing number of commercial laboratories will likely be making efforts to incorporate it as their primary means for identification of bacteria, yeast, and mycobacteria. When they do, they will not simply be inserting another piece of equipment into their existing workflow, which could actually slow the process and increase labor; rather, they will be designing a new workflow to optimize the technology and ensure that results get from the lab to clinicians and patients quickly.

While not all laboratories are capable of widely adopting MALDI-ToF, these that can, probably should. MALDI-ToF may promote better patient outcomes through speedier diagnosis and information to influence efficacious treatment for many bacterial, yeast, and mycobacterial infections—greatly improving the quality of clinical management and patient care.

Insights from studies

Researchers have been increasingly addressing the value of MALDI-ToF. Delport et al specifically addressed the use of MALDI-ToF for the diagnosis of sepsis. The investigators found that MALDI-ToF identification significantly decreased the amount of time to identify a specific pathogen, and a short incubation MALDI-ToF identification protocol from positive blood cultures further reduced time to identification by 38.78 hours overall. The study also noted that changes needed to optimize antibiotic therapy in the study population occurred 20.2 hours earlier when MALDI-ToF was used to aid diagnosis, as compared to conventional test methods. Wrote the authors: “The earlier initiation of antibiotics certainly contributed to the overall improvement in outcomes in patients with sepsis and bacteremia.”

Researchers have found that MALDI-ToF can help improve patient outcomes by speeding diagnosis and improving the quality of care.

Methods Mol Biol


Janet Cromien serves as Director, National Laboratory Operations, Microbiology, for Quest Diagnostics.

Dale Schwab, PhD, serves as Senior Scientific Director, Infectious Disease, for Quest Diagnostics Nichols Institute.
Compact integration of ID/AST systems.

**BD Phoenix™ M50 and BD Bruker™ MALDI Biotyper**

State of the art Mass Spectrometry identification coupled with rapid and accurate resistance detection not only supports your goal to maximize laboratory efficiency, but also gives you the confidence you need to accelerate ID/AST result reporting.

The BD BACTEC™ blood culture system, BD Bruker MALDI Biotyper and BD Phoenix M50 ID/AST system, combined with an active antibiotic stewardship program, can positively impact your clinical outcomes.

Adding the BD EpiCenter™ data management system helps streamline integration between instruments, resulting in increased efficiency and traceability. It also provides insightful and customizable reports.

**Build your future lab at [http://BDPhoenixM50.com](http://BDPhoenixM50.com)**

bd.com

© 2016 BD. BD, the BD Logo and all other trademarks are property of Becton, Dickinson and Company.
New testing options for *Trichomonas vaginalis* respond to growing awareness

By Barbara Van Der Pol, PhD, MPH

Despite decades of epidemiologic research demonstrating that *Trichomonas vaginalis* remains the most prevalent non-viral sexually transmitted infection (STI) in the United States and globally, public health efforts to control this infection remain limited. Infection with *T. vaginalis* is not directly monitored by any public health agency, and in the U.S., the recommendations for screening are passive at best. While there are diagnostic recommendations for men with recurring urethritis, there are no general screening recommendations for men. Screening recommendations for women are focused on either women with high-risk behaviors, which may be difficult to define or operationalize, or women living in high-prevalence populations. Unfortunately, this may be circular logic; given that prevalence estimation requires screening data. Many practitioners and laboratorians continue to question the relevance of trichomonas infections despite strong epidemiologic data showing a relationship between trichomonas infections and HIV acquisition and transmission.\(^1\) However, the high-throughput automation mitigates these potential concerns.

Another mid- to high-throughput system, available since 2012, utilizes strand displacement amplification. Sample types include female urine, endocervical samples, and vaginal swabs. This assay can be performed alone or in combination with a chlamydia/gonorrhea assay as well. When used, the patient sample is used for DNA extraction, and the extracted material is used for all of the tests ordered. Adding the trichomonas assay does reduce the total number of samples that can be tested in a single run; however, the system can perform multiple runs during a typical single-shift work day. This system, and those described above, offer solutions for trichomonas detection alone or in combination with chlamydia/gonorrhea testing, that are semi- or fully automated and offer time-saving lab efficiencies for central or reference laboratories that have medium to high demand and test volume.

Of potential interest to smaller, local laboratories is an assay, \(^12\) recently cleared by the U.S. Food and Drug Administration (FDA), which is the first trissexual assay for detection of chlamydia, gonorrhea, and trichomonas to be commercially available in the U.S. This means that a single sample, a single extraction process, and a single set of reagents are used to generate all three results. Sample types include female urine, endocervical specimens, and vaginal swabs. This PCR assay is run on a bench-top platform that can run one to 24 samples and takes approximately 3.5 hours to generate results. The broad menu of the platform also includes various infectious (non-STI) pathogens, making this platform suitable for use in small- to medium-throughput settings that need to minimize the number of platforms in the laboratory. The components are prepackaged in individual strips, stable at ambient temperature and disposable following completion of the test, in an effort to minimize both waste products and the potential for environmental contamination.

Also noteworthy is a more rapid, potentially point-of-care (POC) assay, \(^13\) requiring approximately 90 minutes for test completion, using a compact bench-top instrument. The assay is classified as CLIA moderate complexity. The system is available in single, double, four-slot, and higher instrument sizes in order to meet a variety of demand and volume situations. The platform is cartridge-based and requires minimal hands-on time. The test for *T. vaginalis* is performed in a separate cartridge, and running the trichomonas and the chlamydia/gonorrhea (in a single cartridge) assays requires only one instrument slot per patient. Most interestingly, this system is the first in the U.S. to be FDA-cleared for male urine in addition to female urine and vaginal swabs. This affords an exciting opportunity to test men who are at risk (e.g., as sexual contacts to trichomonas or because of unresolved symptoms of urethritis following routine treatment) and could have a substantial impact on male-to-female transmission. This platform also has a broad infectious-diseases menu that provides rapid results which may be useful in emergency departments and urgent care settings.

Finally, another POC, rapid NAAT \(^15\) is now available for trichomonas, with results available in about 45 minutes. This assay is isothermal and thus requires only basic equipment (heating continued on page 26
WHERE LAB TECHNOLOGY & HEALTHCARE CONNECT

The RX series connects modern technology with the highest quality reagents providing cost savings through consolidation of routine and specialized tests onto a single platform.

The most versatile range of clinical chemistry analyzers in their class, the RX series combines robust hardware and intuitive software with the RX series dedicated test menu putting innovation, ease-of-use, and superior technology at the forefront of your laboratory.

Guaranteeing performance like no other, the RX series provides unrivalled precision, reliability and accuracy for results you can trust.

RX daytona
RX imola

Tel: +1 304 728 2890 | Toll Free 866 472 6369 | therxseries@randox.com | therxseries.com
blocks) and has a lateral flow readout that is performed in a hand-held instrument. While this assay does not have CLIA-waived status and does require (minimal) instrumentation, the test is suitable for use in settings where immediate results can positively impact patient care and management. Laboratory savings resulting from the lack of capital equipment costs may make this a useful tool in many resource-constrained settings. There is no comparable chlamydia gonorrhea testing that could be performed from the same patient sample.

An improving diagnostic landscape

The assays mentioned above offer a wide variety of solutions that make it possible to provide testing for trichomomas in almost any setting. While the POC options do not have CLIA waivers and require more time than patients might be willing to wait in many settings, there are applications for these assays. Single-test options, such as those offered by any of the last three tests described above, may be useful in emergency departments when patients may, first, have an extended wait prior to the conclusion of a visit and, second, may benefit from immediate treatment. Women presenting with discharge and pelvic pain would likely fall into this category. Beyond the POC applications, assays that can be run in small batch sizes, particularly those that can be combined with chlamydia gonorrhea testing on platforms that offer a broad testing menu, may enable local testing at smaller laboratories, obviating the need for referral to centralized or reference laboratories. Finally, the larger platforms all offer combined trichomomas and chlamydia gonorrhea testing and support higher test volumes. Adoption of the solution best suited to each particular laboratory may facilitate clinical adoption of trichomomas testing in many settings. In addition to improved patient care and management, additional testing may improve our understanding of the populations most at risk for this highly prevalent disease. In populations where HIV risk and rates of trichomomas increase, such data could lead to improved public health efforts targeting at reducing risk of both of these diseases.

REFERENCES

  mydia trachomatis, Neisseria gonorrhoeae and Tricho-
  monas vaginalis: a cross-sectional analysis of positivity and risk factors in remote Australian Aboriginal com-
3. Centers for Disease Control and Prevention. Sexually
  monas vaginalis infection and human immunodefi-
5. Mavedzenge SN, Van Der Pol B, Cheng H, et al. Epi-
7. Hosts BE, Peterman TA, Terrane EA, Weinstock H, Meites E, Bolan GA. A Trich-y question: Should Tricho-
8. Kissinger P. Epidemiology and treatment of tricho-
9. Nye MB, Schwebke JR, Body BA. Comparison of APTIMA Trichomonas vaginalis transcription-mediated amplification to wet mount microscopy, culture, and poly-
  e181-187.
10. Schwebke JR, Hobbs MM, Taylor SN, et al. Molecu-
11. Ginochio CC, Chapp K, Smith JS, et al. Prevalence of Trichomonas vaginalis and coinfection with Chlamydia trachomatis and Neisseria gonorrhoeae in the United States as determined by the Aptima Trichomonas vag-
12. Van Der Pol B, Williams JA, Taylor SN, et al. Detec-
13. Van Der Pol B, Williams JA, Fuller O, Taylor SN, Hook
  EW, Chlamydia, Gonorchae and Trichomonas Combined testing using the BD MAX CT/GC/QV Assay using Geni-
  tion of a new GeneXpert (Gx) molecular point-of-
  N. Trichomonas vaginalis and human immunodeficiency virus coinfection among women under community su-
17. Lazenby GB, Ural ER, Andrews AL, Simpson K. Cost-

INTENDED USE: The BioPlex® 2200 HIV Ag-Ab assay is a multiplex flow immu

The BioPlex® 2200 HIV Ag-Ab assay is intended for use in testing plasma specimens to screen organ donors when specimens are obtained while the donor’s heart is still beating. The BioPlex® 2200 HIV Ag-Ab assay is not intended for use in screening blood or plasma donors, as the effectiveness of this test for use in the screening of these donors has not been established. However, in urgent situations where traditional licensed blood donor screening tests are unavailable or their use is impractical, this assay can be used as a blood donor screening assay.

WARNING: FDA has approved this test for use with serum and plasma specimens only. Use of this test kit with specimens other than those specifically approved for use with this test kit may result in inaccurate test results. This test is not intended for use in children younger than 2 years of age.

CAUTION: United States federal law restricts this device to sale by or on the order of a physician, or to a clinical laboratory.
The 5th generation BioPlex® 2200 HIV Ag-Ab assay sniffs out acute infection early in the window period

BioPlex® 2200 HIV Ag-Ab assay uses its multiplex technology to simultaneously detect and differentiate:
- HIV Ag-Ab overall result, with
  - HIV-1 p24 Ag
- HIV-1 Ab (Groups M & O)
- HIV-2 Ab

Thanks to its radar-like sensitivity to the HIV-1 p24 antigen and the ability to differentiate between HIV-1 and HIV-2 antibodies, the BioPlex® 2200 HIV Ag-Ab assay detects early acute infections and guides in the choice of supplemental testing through the CDC diagnostic testing algorithm. Early detection enables early treatment, saving and improving patients' lives.

Bio-Rad, your authority in HIV testing
Learn more at bio-rad.com/diagnostics
Managing *Mycoplasma genitalium* infections during a rapid upsurge in antibiotic resistance

By Tim Read, PhD, MBBS, and Catrina Bradshaw, PhD, MBBS (Hons)

*Mycoplasma genitalium* is now an established cause of urethritis in men and pelvic inflammatory disease and cervicitis in women. While *M. genitalium* may not be as well understood as *Neisseria gonorrhoeae* and *Chlamydia trachomatis*, microbiology laboratories will soon be fielding more questions on this troublesome bacterium. This is because, unlike the other bacterial sexually transmitted infections (STIs) that remain sensitive to single-dose antibiotic therapy, *M. genitalium* infections in many countries are developing resistance to the antibiotics commonly deployed against them. The result is a growing number of infected patients with diminishing treatment options. This article reflects on lessons learned during a decade of managing *M. genitalium* at Melbourne Sexual Health Centre, a large public clinic in Australia’s second largest city.

Clinical services around the world have been slow to adopt routine testing outside of research programs, largely because of the lack of commercially available assays for *M. genitalium*. When we began testing for *M. genitalium* in 2004, the test was limited to men with urethritis. Since then, testing has increased, as evidence of pathogenicity has strengthened, to include contacts of *M. genitalium* infection, women with pelvic inflammatory disease and cervicitis, and now proctitis.

### The therapeutic history

The long-acting macrolide, azithromycin, which remains an effective single-dose therapy for *Chlamydia trachomatis* infections, was initially also highly effective against *M. genitalium*. Early in vitro data suggested *M. genitalium* strains had minimum inhibitory concentrations (MICs) for most macrolides and tetracyclines within the susceptible range. But almost as soon as testing for *M. genitalium* began in Melbourne, some azithromycin failures occurred. A recent meta-analysis of studies of single-dose azithromycin treatment showed proportions of patients cured fell from 85 percent before 2009 to 67 percent after 2009. Doxycycline has not proved to be useful, with proportions cured in most studies falling below 40 percent. Resistance to macrolides is conferred by single nucleotide substitutions in domain V in the 23S ribosomal RNA gene. These macrolide resistance mutations (MRMs) have been identified by sequencing or by high-resolution melt analysis but can now be detected by one of the commercially developed diagnostic assays which uses a multiplex polymerase-chain reaction (PCR) for detection and resistance. MRMs are strong predictors of treatment failure (odds ratio 24 in one study). In papers published in 2016, they were present in 40 percent to 60 percent of infections from Canada, the United States, Australia, and Germany. It has also become clear that a proportion of macrolide-susceptible infections treated with single-dose azithromycin 1g will persist and develop detectable MRMs following treatment. While the mechanism is unclear, it is likely that single-dose azithromycin, used alone or in combination for the treatment of urethritis and other STI syndromes, is selecting for macrolide resistance and contributing to the rising rates of MR, and this may also be occurring even with higher dose and duration of azithromycin therapy. The major alternative to prescription of azithromycin is to use seven to 10 days of one of the broad-spectrum fluoroquinolone class of antibiotics, such as moxifloxacin or sitafloxacin, which are expensive and have uncommon but serious side effects.

Resistance to fluoroquinolones is less common than to macrolides, and is rare in Northern Europe and the UK, but it is as high as 15 percent in STI services in Melbourne and Sydney. Until new antibiotic options become available, clinicians face a dilemma: prescribe a cheap, safe antibiotic (azithromycin) to which resistance is increasingly common and which may induce or select resistance, or a fluoroquinolone.

The U.S. Centers for Disease Control and Prevention guidelines, and the Australian guidelines, still recommend single-dose azithromycin as first-line treatment, while the European guidelines recommend 1.5g azithromycin over five days or josamycin over ten days. The U.S. and European guidelines recommend ten days of moxifloxacin for second-line therapy. With azithromycin resistance at or above 50 percent in many countries, some authors are recommending a reduced use of azithromycin as a treatment for STI, and this may be reflected in future guidelines.

### Testing and treatment

How best to approach the treatment dilemma will depend on what, if any, type of *M. genitalium* testing is available.

**No testing available for *M. genitalium***: Culture is not available for *M. genitalium* diagnosis, as it takes months and is only available in a few laboratories globally. Several nucleic acid amplification tests are available, but none are yet approved by the U.S. Food and Drug Administration. In clinics without access to any *M. genitalium* testing, clinicians may still suspect the organism to be present in cases of chlamydia-negative nongonococcal urethritis and pelvic inflammatory disease, with persistent symptoms after treatment. If azithromycin treatment has already failed, clinicians need to consider the risks and benefits of a fluoroquinolone in the absence of laboratory confirmation of infection.

**Testing available for *M. genitalium*** but not for macrolide resistance. Given the propensity for *M. genitalium* to develop MRMs after azithromycin treatment, a test of cure is essential. A positive test of cure always raises the possibility of non-adherence to therapy or of reinfection, but if these risks are considered low, then it can be assumed that MRMs are present whenever azithromycin has failed. There is no role for further azithromycin therapy in these cases, and moxifloxacin is the next option.

**Combined testing for *M. genitalium*** and macrolide resistance is available. These diagnostic tests identify infections that can be treated with azithromycin and those with MRMs that cannot. However, whenever a macrolide is used, the potential for generating further resistance is sufficiently high that clinics should do all they can to ensure that every patient returns for a test of cure. Fluoroquinolones are contraindicated in a number of situations, including pregnancy and, as noted above, resistance is an emerging problem in the Asia-Pacific. In Melbourne we had some early success using oral pristinamycin dosed at 1g, four times daily for ten days, in cases where moxifloxacin had failed or was contraindicated. Subsequently, we have observed a number of treatment failures, and these are the subject of current research.

Tests of cure should be performed three to four weeks after the start of treatment. In one study, all 16S PCR tests that were positive 14 days after treatment were also positive at 28 continued on page 30
TO HER, IT’S MORE THAN JUST A TEST

Receiving her STI result is the moment that could change everything. One wrong result can cause a cascade of emotional and physical distress for patients, and raise the risk of infecting others.

Rely on layers of protection that make a world of difference

- Dual probe design increases sensitivity and specificity
- Internal control verifies that amplification has taken place
- AmpErase virtually eliminates false positive results

And, experience hands-off testing on a single platform by consolidating your HSV and CT/NG testing on the cobas® 4800 system.

cobas® CT/NG v2.0 Test | cobas® HSV 1 and 2 Test

For more information, contact your Roche sales representative or visit www.usdiagnostics.roche.com


COBAS and LIFE NEEDS ANSWERS are trademarks of Roche.

© 2016 Roche Diagnostics. All rights reserved. 472-06901-0416 Roche Diagnostics 9115 Hague Road, Indianapolis, IN 46250
Neisseria gonorrhoeae susceptibility to ciprofloxacin

By Lao-Tzu Allan-Blitz and Jeffrey D. Klausner, MD, MPH

Neisseria gonorrhoeae has developed resistance to all antimicrobials currently available, and there are only a few novel antibiotics in development. New approaches for combating that resistance are urgently needed. One new approach calls for targeted therapy with antibiotics previously thought to be ineffective; it has been made possible by the development of molecular assays that predict in vitro antimicrobial susceptibility. One of those molecular assays for determination of N. gonorrhoeae susceptibility to ciprofloxacin has been studied extensively.

Previous reports have shown that mutation at codon 91 of the gyrA gene of N. gonorrhoeae can reliably predict in vitro susceptibility to ciprofloxacin. While there are other mutations conferring ciprofloxacin resistance, it has been shown that those mutations usually occur in parallel with the gyrA mutation. Additionally, in the United States, it is estimated that approximately 80 percent of N. gonorrhoeae infections are susceptible to ciprofloxacin. The ability to predict susceptibility by mutation at a single locus, and the high prevalence of susceptible infections, make ciprofloxacin an appealing option for targeted therapy. What is needed, however, is the ability to rapidly determine genotype results.

In 2007, we developed a real-time polymerase chain reaction (RT-PCR)-based assay for the determination of mutation at codon 91 of N. gonorrhoeae. Since then, there have been 11 studies (N=4777) with samples from ten countries using RT-PCR techniques to compare gyrA genotype results with traditional susceptibility testing methods. Those studies found between 95.8 percent and 100 percent sensitivity and 93.2 percent and 100 percent specificity of wild-type gyrA N. gonorrhoeae for the prediction of ciprofloxacin susceptibility. Notably, one study reported improved assay sensitivity among urine samples after restricting the assay to amplification of only the Ser91 codon. Furthermore, four studies compared mutation at codon 91 of the gyrA gene in N. gonorrhoeae to other Neisseria species and found 100 percent specificity for N. gonorrhoeae.

Therefore, there is strong evidence that codon 91 gyrA gene determination can reliably predict, with sufficient sensitivity and specificity, N. gonorrhoeae susceptibility to ciprofloxacin; and since RT-PCR returns results within 24 hours, those rapid assays can enable targeted ciprofloxacin therapy of N. gonorrhoeae infections. Other rapid molecular assays for prediction of antimicrobial susceptibility are currently in clinical use, such as the FDA-approved molecular assay for Mycobacterium tuberculosis, which uses RT-PCR for amplification and molecular probes for mutation within the rifampin-resistance determining region; or the RT-PCR amplification of known resistance genes in screening for methicillin-resistant Staphylococcus aureus. Targeted therapy will have many benefits, among which may be the reduction in the emergence of ceftriaxone, as a previous study demonstrated that treatment may be the major driver of resistance. The assay we developed in 2007 was verified in accordance with Clinical Laboratory Improvement Amendments and implemented into routine clinical practice for all N. gonorrhoeae positive specimens at UCLA health in November 2015. Further studies are underway to characterize the impact of that implementation.

Acknowledgments: This research was supported by the United States National Institutes of Health grants R21AI117256, NIH R21AI109005.

Please visit mlo-online.com for references.
Are you still diagnosing Trichomonas with a microscope or sending it to the lab? Trixie here is an amazing swimmer and is a very tricky bug to detect.

The OSOM® Trichomonas Test is the only CLIA waived rapid diagnostic available. It does not rely on the detection of a live organism and can be done in 10 minutes!

What does this mean to you and your patients? Far more accurate detection and the ability to test and treat in one visit which helps improve patient care and reduce costs.

Answers for Healthcare. Awesome...yes, OSOM®. Call your rep today at 800-332-1042.
As ACOs evolve, the clinical lab’s role grows

By Irwin Z. Rothenberg, MBA, MS, MT(ASCP)

There is now general agreement that a value-based model (quality of health outcomes per dollar expended), measured at the patient level, is the only way to achieve true healthcare savings over time and bring about real system transformation. Previous attempts to contain healthcare costs utilizing payment models that relied on controlling unit costs or restricting coverage failed to optimize the use of available funds, restrain costs, or address the healthcare needs of the underserved. These models failed because they did not incorporate improvements in the quality of care delivered, while simultaneously restricting access to that care.1

With the enactment of the Affordable Care Act, Congress emphasized experimentation with new coordinated care and accountable delivery systems under the Medicare Shared Savings program (MSSP). The Accountable Care Organization (ACO) is one model that was developed to achieve this. The Centers for Medicare and Medicaid Services (CMS) defines ACOs as groups of doctors, hospitals, and other healthcare providers who come together voluntarily to give coordinated high-quality care to their Medicare patients. In this healthcare model, incentives are tied to organizations that demonstrate their commitment to the prevention of disease while offering quality and efficient services as patients are placed into a continuum of care. When an ACO succeeds in delivering high-quality care and spending healthcare dollars more wisely, it will share in the savings it achieves for the Medicare program.2

According to Jennifer Hansen, partner at Hooper, Lundy and Bookman, a law firm that represents healthcare providers and suppliers, the benefits of an ACO include “improving patient outcomes, cost efficiency, decreased ER waits, allowing patients better access, and achieving better satisfaction. There are also market advantages that take place which allow providers to keep some individual autonomy while still obtaining the opportunity to work with other providers and have a referral source.”3

The lab as information integrator

The ACO model requires a robust information technology infrastructure across the continuum of “cradle to grave” healthcare, including Health Information Exchanges (HIEs), Electronic Health Records (EHRs), Hospital Information Systems, Laboratory Information Systems (LISs), e-Prescribing, Medical Device, and Diagnostic Imaging systems. It also requires multi-disciplinary, multi-organizational, team-based care models to actively manage newer care plans, modes of care delivery, and compliance with evidence-based care practices across a variety of providers and care settings. In order to accomplish this, organizations need to collect and analyze performance and outcomes measures.4 This need has led to the higher profile and redefinition of the central role of the clinical laboratory as a prime integrator of information and data exchange.

The ACO opportunity

Indeed, the laboratory’s clinical expertise, in combination with its network of physician and patient touch points, make it a central component of an integrated provider organization such as an ACO. By hosting the vast majority of centralized information, laboratories reaffirm the importance of highly functioning physician/laboratory relationships. Through the deployment of a robust connectivity system, laboratories have the ability to streamline physician office workflows, receive test orders and return results to a variety of EMR systems in real time, and play an essential role in building physician relationships. Downstream benefits can include enhanced lab order accuracy, more complete patient and billing information, improved revenue collections, and better patient outcomes—all critical differentiators in an era of reduced costs. The best investment a lab can make in its future is to maximize the efficiency of its internal operations. Labs should focus on the accessibility and convenience of their services and information communications.

Strategic Initiatives

Clinical laboratories must recognize the opportunities ACOs create and respond with strategies that position the lab to reach its full potential within the ACO model. The following strategies are recommended for laboratories to meet the clinical information needs of physicians practicing within ACO organizations, and to demonstrate their value by facilitating decision support and coordinated care:

**Outreach.** Extend laboratory services to all available physician offices, nursing facilities, clinics, and service centers. Create a network of integrated and coordinated services across the continuum of care. Develop the infrastructure and logistics required to serve chronically ill patients, who need to access care periodically in different venues from a variety of providers, in an ambulatory environment. Going forward, a laboratory’s association with physician offices, combined with electronic connectivity, may be the best long-term strategy in the era following healthcare reform.

**Connectivity.** Build electronic connectivity to providers in a way that integrates data in and out of physician practice EMRs. Laboratories have the opportunity to be instrumental in assisting physicians and demonstrating value to their physician clients by streamlining order and result processes within their offices.

**Lean internal laboratory processes.** One of the most important steps a laboratory can take to position its services for inclusion in an ACO or coordinated-care model is to improve (Lean) every process to eliminate waste, minimize variation, and reduce costs. The best investment a lab can make in its future is to maximize the efficiency of its internal operations. Labs should focus on the accessibility and convenience of their services and information communications.

**Test utilization-management.** Clinical utilization management has the potential to reduce or eliminate unnecessary expenditures. Test-utilization review within a hospital organization can be performed by a multispecialty medical
committee, such as a laboratory formulary committee. This committee can have the scope and authority to recommend the appropriate use or availability of lab tests as well as review the process for referred test orders and protocols for lab workup for specific disease states.

Laboratory professionals are also uniquely qualified to be involved in the development of computerized physician order entry (CPOE) with clinical decision support (CDS), test algorithms, and clinical pathways. Pathologists have the medical training necessary to analyze aggregate clinical data for outcomes and quality. As medical doctors, pathologists are trained to understand the clinical and medical relationships embedded in the data and can use outcomes data to improve diagnostic pathways.

Understanding the big picture. The director of laboratories operating within hospitals or health systems should have a clear understanding of the health system’s clinical and financial objectives, including any plans for an ACO. Successful laboratories will align the laboratory’s strategic objectives with those of the larger organization in areas such as community marketing, physician-alignment, and information technologies. Well-informed laboratories will also be cognizant of efforts from competing laboratories to replace them in an ACO.

It is critical that laboratory directors articulate the value of their laboratory services and the pivotal role of the lab in ensuring that an accurate diagnosis is established early in the continuum of care, and that clinical information is available to coordinate the course and cost of care. Health system executives, managed-care directors, physicians, and administrators of health plans sometimes take for granted the crucial contributions labs make to medical decisions. Successful laboratories, meeting the new challenges of healthcare reform, will leverage outreach relationships, actively participate in the formative stages of ACO development, and prepare for upcoming reimbursement changes. The paradigm shift in healthcare from episodic care to chronic-care management represents a once-in-a-generation opportunity for proactive laboratories to redefine their value in a new, much larger role as integrators of critical clinical information and decision support.¹

REFERENCES
Microfluidics in the clinical lab

By Richard Day, PhD

There has been a great deal of hype for the last 20 years that microfluidics will revolutionize the use of the clinical lab by spawning hand-held, sensitive, and lower-cost diagnostics systems. In fact, the emergence of such systems has been relatively sparse, and the benefit of microfluidics is being seen in the core of larger systems that still intend to be in the clinical lab. Albeit micro in the fluidics, there must be a macro system around it, such as a cartridge of reagents, a manifold for multiple fluid processing steps, and larger still optical detection modules to measure the physics going on at the micro level. Where the real advantage lies is in the key microfluidic principles that govern the behaviour of the small, and it is these principles that are fundamental to the new diagnostic systems that are being used in the clinical lab today.

For example, “liquid biopsy” is a term for sampling blood for oncology diagnostics, with an aim to capture cells to detect or monitor cancers. There is an inherent difficulty in that such cells can be a needle in a haystack, occurring in parts per million or less of blood samples. One currently available system achieves label-free enrichment heterogeneous populations of cancer cells from 7.5ml of patient’s blood. It uses a microfluidic principle called Dean Flow, where liquid flowing in a spiral channel at just the right speed will cause target circulating tumor cells to gather at one side of the channel. By forking the channel, the target cells can be diverted to an output of enriched and purified material that can be detected in laboratory cytology systems.

Without such enrichment, cytology systems would have to trawl through blood samples dominated by normal leukocytes to find the rare cells, taking more time and therefore adding cost. The principle here is that in a spiral channel, liquid flow builds a recirculation flow across the channel, much like a secondary flow makes tea leaves collect in the center of a stirred tea cup. With the knowledge that larger cells are likely to be the circulating tumor cells (CTCs), and that larger cells have a more difficult time recirculating across the spiral channel, they can be separated from smaller, more common cells.

Another principle fundamental to microfluidics is the rapid rate of heat transfer possible when the surface area-to-volume ratio becomes large. Analogous to small droplets of fine spray cooling the skin faster than a single splash, it is possible to change the temperature in a small amount of liquid much more rapidly than a large volume of liquid. This effect is key to PCR reactions inside some clinical lab consumables to amplify DNA. The tiny volumes of sample are heated and cooled for dozens of cycles over 45°C swings so tests can be completed in merely a couple of hours, such as for one commercially available TB test. Inside a microfluidic consumable, the sample is flowed through flat chambers that are placed against the heat source (or sink). The flat shape maintains a high surface area-to-volume ratio, and the thin thickness allows rapid heat transfer, according to the diffusivity law that scales as D²/L²/T, so the time T to diffuse can be driven down with the square of the thickness L.

Flowing a liquid through a porous medium for the purpose of wetting lots of surface area is a principle familiar to anyone making coffee. Small particles have a great deal of surface area to react with the liquid, and chemical processes can be greatly accelerated. The liquid flows among particles in the little voids and follows a sinuous path to the exit. There are many different routes to the exit, so the fluid effectively forms many parallel paths, each with a small flow, and it takes a great deal of pressure to maintain the flow.

A striking example in the clinical lab is the pressure required to squeeze liquid through a long column packed with beads in an HPLC (high pressure liquid chromatography) machine. The liquid molecules in the column react differently to the surfaces of solid particles in the column, due to chemical interactions between the polar molecules in the solvent and the material in the column. Some molecules in the sample are attracted more than others, and spend longer in the column, so by the time the sample has flowed through, the molecules are spread out. This effect is heightened when the beads in the packed column are small, such as a few microns across, so the surface area for the chemical interactions is maximized. The consequence is that the pressure to drive the liquid sample through the column becomes huge.

In HPLC, the sample is driven through the column at hundreds of atmospheres of pressure. The different molecules exit the HPLC column spread out over time, and can be measured separately to generate a chromatography trace in the clinical lab. To increase sensitivity and specificity, one can imagine using smaller and smaller beads, and going to higher and higher pressures. Ultra-high pressure liquid chromatography, or UHPLC, can use particles as small as two microns, requiring pressures of 15,000 psi (more than 1,000 atmospheres) of pressure in a packed bed, which requires using advances in materials and bonding techniques for containing the pressure.

Finally, we are all familiar with an emulsion—for instance, oil and vinegar salad dressing—that clearly shows the refusal of certain liquids to mix. Taking two immiscible liquids down to microfluidic scales, tiny spherical water droplets known as picodroplets remain stable within a channel of oil. These water-in-oil systems can permit the generation of thousands of separate aqueous reaction vessels for rapid drug screening and cellular testing. At small scales, these water droplets are dominated by surface tension and form nearly spherical shapes that can be controlled. In a picodroplet generator, water and oil meet at a microscale crossroads, and the water in the middle pinches off into separate droplets that remain distinct from each other. It is possible to fill these thousands of picodroplets with different drug titrations, different cells, or individual DNA base pairs, which makes each picodroplet like a separate reaction vessel. This way, it is possible to massively increase the throughput of drug discovery, cell analysis, or DNA sequencing.

The hype has not so far been realized, but the fundamental principles of microfluidics will underlie key technologies that will enable the future clinical laboratory.
Medicare's local coverage decision process needs legislative fix

By Richard C. Friedberg, MD, PhD, FCAP

In recent years, the pathologist’s ability to make medical decisions in the best interests of patients has been adversely affected by faulty decisions in Medicare’s local coverage program, a government-approved process lacking in transparency and accountability.

Specifically, the process for drafting Medicare local coverage determinations (LCDs) has interfered with the physician’s medical judgment and what is in the best interests of his or her patient. As all physicians know, these determinations can cover a range of items and services including medical procedures, office visits, imaging, drugs, and laboratory tests. In pathology and laboratory medicine, an erroneous LCD prevents patients from receiving the right test at the right time.

For these reasons, the introduction of new legislation to improve the LCD program is necessary and welcomed. Recently, two new bills were proposed that seek to reform the LCD process. Both H.R. 5721, introduced by Rep. Lynn Jenkins (R-KS) and Rep. Ron Kind (D-WI), and its companion bill S. 3392, introduced by Sen. Johnny Isakson (R-GA) and Sen. Tom Carper (D-DE), deserve the attention of all provider and patient advocacy organizations.

Released in January 2014, a report by the Department of Health and Human Services’ Office of Inspector General (OIG) found more than half of Medicare Administrative Contractors (MACs), which appear to set practice guidelines instead of Medicare, an erroneous LCD prevents patients from receiving the right test at the right time.

The policy then can become of such geographic magnitude that it approaches the very MAC that issued the LCD. Stopping the use of LCDs as a back door to national coverage determinations (NCDs): This will prohibit the CMS from appointing a single MAC, either expressly or in practice, to make determinations that are to be used on a nationwide basis in a given specialty. The CAP has witnessed the carbon-copy adoption of MAC LCDs by other MACs without the benefit of meaningful solicitation or independent assessment of comments and concerns from the public or medical community of the adopting MAC.

For these reasons, the introduction of new legislation to improve the LCD program is necessary and welcomed. Recently, two new bills were proposed that seek to reform the LCD process. Both H.R. 5721, introduced by Rep. Lynn Jenkins (R-KS) and Rep. Ron Kind (D-WI), and its companion bill S. 3392, introduced by Sen. Johnny Isakson (R-GA) and Sen. Tom Carper (D-DE), deserve the attention of all provider and patient advocacy organizations.

Released in January 2014, a report by the Department of Health and Human Services’ Office of Inspector General (OIG) found more than half of Medicare Administrative Contractors (MACs), which appear to set practice guidelines instead of Medicare, an erroneous LCD prevents patients from receiving the right test at the right time.

The policy then can become of such geographic magnitude that it approaches the very MAC that issued the LCD. Stopping the use of LCDs as a back door to national coverage determinations (NCDs): This will prohibit the CMS from appointing a single MAC, either expressly or in practice, to make determinations that are to be used on a nationwide basis in a given specialty. The CAP has witnessed the carbon-copy adoption of MAC LCDs by other MACs without the benefit of meaningful solicitation or independent assessment of comments and concerns from the public or medical community of the adopting MAC.

Reforms contained in the proposed legislation will ensure that Medicare local coverage determinations do not over ride physician medical judgment and deny patients access to medically necessary care. The bills would ensure that coverage decisions are made by qualified independent health experts through a transparent process based on sound medical evidence.

Briefly, the key provisions in the Senate and House bills include:

Open meetings: CAC meetings must be open, public, and on the record. Minutes should be posted to the MAC’s website for public inspection. The gravity of limiting or precluding coverage for both beneficiaries and practitioners heightens the need for transparency where meetings are currently closed.

Upfront disclosure: MACs should include at the outset a description of the evidence considered when drafting an LCD as well as the rationale they rely on to deny coverage. If this information is not provided until the final LCD, it hinders meaningful stakeholder exchange and makes the MAC’s decision to deny coverage almost a foregone conclusion.

Meaningful reconsideration and options for appeal: A meaningful LCD reconsideration process gives Medicare providers and suppliers the opportunity to have a secondary review by a qualified, disinterested party. Under current CMS rules, MAC LCDs are essentially unreviewable by physicians and suppliers without new evidence submitted to the very MAC that issued the LCD.

Richard Friedberg, MD, PhD, FCAP, has served on numerous committees and councils for the College of American Pathologists (CAP), including the Government & Professional Affairs, Accreditation, Quality Practices, Technology Assessment, Transformation, Finance, and Transfusion Medicine. In 2007 and again in 2010, he was elected by the CAP membership to serve on the CAP Board of Governors. In 2013, the CAP membership elected him to serve as CAP President-Elect, and he is serving as CAP President from 2015-2017.
**PRODUCT FOCUS**

**POCT**

**Wireless point-of-care system**

The new wireless BD Veritor Plus System gives healthcare providers and laborato-
rians objective, lab-quality immunoassay test results within minutes. This fast and ac-
urate CLIA-waived solution streamlines the point-of-care diagnostic workflow and
allows providers to quickly review patient results and determine the appropriate treat-
ment in a single consultation, while the patient is still on site. The new system can help
detect infectious diseases such as influenza A and B, respiratory syncytial virus (RSV), and group A strep.

BD Diagnostics, www.rsleads.com/612ml-150

**HbA1c analyzer**

The Quo-Test HbA1c analyzer now comes with a connectivity package, using POCT1-A2 communication protocol as standard. Patient demo-
graphic information and additional test commentary can be added to
each test result, using either the stan-
dard barcode scanner or the new add-on keyboard. This enables patient
test results to be linked and traced throughout the system. Opera-
tor IDs can be added to test results, significantly improving the
traceability and security applied to every HbA1c reading. In addi-
tion to these functions, enhanced quality control is available with multiple user-defined QC lockout options, ensuring that
tests can only be run according to localized QC procedures.

EKF, www.rsleads.com/612ml-151

**Blood gas analyzer**

CueSee Hypoxic is ideal for lowering the re-
portable range of pO2 down to ~15 mmHg
on any blood gas analyzer. The buffered hemo-
globin matrix provides high precision like that of patient samples. Its oxygenation curve al-
ows for its 10-minute open ampule stability.
Highly commutable to provide similar results on point-of-care and bench-top blood gas
analyzers, it is suitable for quality control, method comparisons,
and validating the low end of the Analytical Measurement Range
(AMR), satisfying regulatory requirements for reporting critically low pO2 results.

Eurotrol, www.rsleads.com/612ml-152

**Immunoassay analyzer**

The FastPack IP System is a fully au-
tomated quantitative immunoassay analyzer designed for use in physi-
cian office laboratories. Utilizing sophisticated chemiluminescence
technology, it provides the capability to produce complex immunoas-
say results in 12 minutes or less with a push of a button. It re-
quires minimal space while offering a test menu that will make a clinical impact at the point of care. It also enhances practice efficiency and reduces labor costs; improves patient satisfac-
tion; immediately adjusts therapy; is appropriate for small, medium, and large practices; and boasts an extensive testing menu for vitamin D, testosterone, PSA, TSH, FREE T4 and hCG.

Sekisui Diagnostics, www.rsleads.com/612ml-153

**Multi-parameter hematology control**

Para 4 is a multi-parameter hematology control
for small laboratories and point-of-care applica-
tions that perform a limited number of hemato-
ology tests. Assays include: HemoCue B-Hemogl-
bin Photometer and Hb 201+ analyzer; Separation
Technology, Inc. HemataSTAT II; Spun Hematocrit; and Phase Platelet (Low and Normal values only). Para 4 is available in plastic dropper vials for easy sample allocation with hemoglobin and hematocrit instruments, and is also available in glass vials for auxiliary methods. Para 4 has 110-day closed-vial stability and
14-day open-vial stability.

Streck, www.rsleads.com/612ml-154

**Point-of-care analyzers**

Samsung point-of-care analyzers provide fast, easy, and accurate whole blood testing. Thermo Fisher
Scientific is the exclusive global distribution and development partner for the LABGEO port-
folio: 1) LABGEOPT10 chem-
istry panels provide results in
7 minutes using 70μL sample;
2) LABGEOIB10 cardiac marker panels, PLT (sepsis), TSH, and β-hCG provide results in 20 minutes using 500μL sample. 3) LABGEOHC10 (HC30A for USA) hematology analyzer provides results in 45 seconds using 25μL sample. FDA approval pending.


**Point-of-care blood lead analyzer**

LeadCare II is a CLIA-waived blood lead analyzer that offers a simple, in-office solution to determine a child’s blood lead burden. Using electrochemistry, the portable system features electronic calibration and test kits with everything needed for 48 tests. Now celebrating its 10th anniversary, Lead-
Care II is convenient—CLIA-waived and simple to use; efficient—three minute result while patient is in the office; accurate—quantitative result with 50 μL of blood; and reimbursable—CPT 83655.

Magellan, www.rsleads.com/612ml-156

**Coagulation analyzer**

The Xprecia Stride coagulation analyzer is a Point-of-care (POC) PT/INR device cleared by the FDA based on the new rules published in March 2016. It delivers fast, reliable Prothrombin Time/ International Normalized Ratio (PT/INR) testing for point-of-care monitoring and management of oral anticoagulation ther-
apy with warfarin, a vitamin K antagonist.
No bigger than a smartphone and weighing just 10.5 oz, the Xprecia Stride Coagula-
tion Analyzer can be brought directly to the patient’s finger for efficient and comfortable blood sample application. The Xprecia Stride analyzer uses fresh capillary whole blood, and results are expressed as INR. It utilizes the same Dade Innovin reagent used by Siemens Healthineers central lab analyz-
ers to minimize any potential for variability. Studies have shown the performance to be equivalent to a reference laboratory hemostasis system, with results available within minutes.

Siemens, www.rsleads.com/612ml-157
Who will educate the next generation of Laboratory Scientists? Spartans Will. At Michigan State University, we believe earning your bachelor’s degree is just the beginning of your journey. Who will be committed to providing you with opportunities for continued growth and education? SPARTANS WILL.

Become a Spartan

Our courses offer you a way to enhance your skills to meet the current demands of advancing technology in the clinical laboratory setting. Whether enrolling for a single course, a certificate program, or pursuing an online Master’s degree, we will help you reach your goals. Who will tailor your studies to fit in with your work and personal life? SPARTANS WILL.

Online Courses | Continuing Education | Online Masters

For more information, contact our online assistant:
Aimee Stewart | (517) 884-3483
bld.natsci.msu.edu/online-education
### Get to the next-level of HIV testing

**Introducing the BioPlex® 2200 HIV Ag-Ab diagnostic test.**
The first and only test that simultaneously detects, differentiates, and reports HIV-1 p24 antigen and antibodies to HIV-1 (Groups M and O) and HIV-2.

**Bio-Rad Laboratories**
www.rsleads.com/612ml-400

### cobas® EGFR Mutation Test v2

**Designed to provide clear, actionable results, the cobas® EGFR Mutation Test v2 is a real-time PCR assay approved as an aid in first-line and subsequent therapy decisions for patients with non-small cell lung cancer (NSCLC).**

**Roche Diagnostics**
www.rsleads.com/612ml-404

### FastPack® IP Immunoassay POC System

**Fully automated quantitative immunoassay analyzer designed for use in any size POL utilizing sophisticated chemiluminescence technology with results in 12 minutes or less. Menu: Vitamin D, Testosterone, PSA, TSH, FREE T4, hCG.**

**Sekisui Diagnostics**
www.rsleads.com/612ml-401

### Quality Means Confidence

**Avoid False-Positive Urine Drug Screens. Use Astec® CHIROBIOTIC® V2 for the rapid, sensitive LC/MS analysis methamphetamine and amphetamine enantiomers in urine without the need for derivatization. See this application note and our full analytical column offering at sigma-aldrich.com/clinical.**

**Millipore Sigma**
www.rsleads.com/612ml-405

### Blood Glucose Linearity and Daily QC

**Introducing Linearity DROP LQ Blood Glucose (Item #K736M-5) and Control DROP LQ Blood Glucose (Item #K078M-8). These ready-to-use liquid products are intended to be used with quantitative assays on clinical laboratory analyzers, simulating human patient samples. For more information please visit: www.auditmicro.com**

**AUDIT MicroControls**
www.rsleads.com/612ml-402

### New EDTA Sed-Rate Instrument

**Streck is the exclusive distributor in the U.S. and Canada of the Diesse MINI-CUBE, an automated instrument for erythrocyte sedimentation rate testing directly from EDTA tubes without consuming the patient sample. Visit streck.com/minicube.**

**Streck**
www.rsleads.com/612ml-406

### Improve analytical quality with Acusera 24.7

**We are pleased to announce that Acusera 24.7 Live Online now features Measurement Uncertainty and Sigma Metrics. Our software will automatically calculate these for you, freeing your time and ensuring results are accurate and reliable.**

**Randox Laboratories**
www.rsleads.com/612ml-403

### cobas® Liat® PCR System

**Whether your flu A/B or strep A result is positive or negative, you can confidently treat patients at the time of visit without confirmation—when they need it most.**

**Roche Diagnostics**
www.rsleads.com/612ml-407
Molecular flu/RSV test

With the flu season upon us, healthcare providers no longer need to choose between a fast test result or an accurate test result for patients who are suspected of influenza infection. Now, they can have both, with Xpert Xpress Flu/RSV from Cepheid, a new test that delivers reliable molecular test results in as little as twenty minutes. Xpert Xpress Flu/RSV is twice as fast as its predecessor, Xpert Flu/RSV XC, but has comparable performance characteristics. The new assay features a novel design, which employs multiple targets for each virus. The built-in redundancy provides a very high test sensitivity, avoiding the impact of seasonal drift that has been a problem historically with molecular tests, while maintaining high specificity.

Cepheid
www.rsleads.com/612ml-158

Blood glucose linearity and control kits

AUDIT MicroControls, Inc., has announced the following new additions to its complete line of calibration verification/linearity and daily quality control products. These ready-to-use liquid products are intended to be used with quantitative assays on clinical laboratory analyzers, simulating human patient samples: (1) The Linearity DROP LG Blood Glucose is a ready-to-use liquid, consisting of five levels that demonstrate a linear relationship to each other when assayed for glucose. This product has an open vial stability of 7 days when stored at 2-8°C. (2) The Control DROP LG Blood Glucose is a ready-to-use liquid, intended to simulate human patient samples for use as assayed quality control materials for glucose. This product has an open vial stability of 7 days when stored at 2-8°C.

Audit MicroControls
www.rsleads.com/612ml-160

IL5 high-speed camera

Fastec Imaging's IL5 High-Speed 5MP Camera is easily mounted on a microscope, enabling users to record high-speed video of microscopic events. Both spatial and temporal magnification work in tandem to clarify understanding in applications such as microfluidics, where particles often move through the field of view very quickly. With four models to choose from, boasting crisp, clean video from 2560 x 2048 @ 230fps to 800 x 600 @ 1650fps, there is an IL5 to fit any application needs. All models record over 3200 fps at VGA resolution and more than 18,000 fps at smaller resolutions. Able to save images to an SSD or SD card while recording high-speed bursts of hundreds or even thousands of images at a time, the IL5 is always ready for the next high-speed snapshot. The IL5 is a cost-effective solution to record high-speed events for slow-motion analysis.

Built for flexibility and ease of use, the Fastec IL5 camera can be controlled over Gigabit Ethernet via Fastec FastMotion software on a PC/Mac or via the built-in web interface with a web browser on a PC, Mac, tablet, or even smartphone. Using the (LR) FasCorder Mode, users can operate the camera as a regular camcorder to record and pause as needed and follow the action, stop recording and review what they have, and then append additional footage at will, even after a power cycle. Unlike traditional high-speed camera systems that only record for a few seconds and require careful triggering, the IL5's Long-Record (LR) option can record at high speed for many minutes at high resolutions, to many hours at reduced resolutions.

Fastec Imaging
www.rsleads.com/612ml-161

INDEX OF ADVERTISERS

<table>
<thead>
<tr>
<th>ADVERTISER</th>
<th>WEB</th>
<th>PAGE</th>
</tr>
</thead>
<tbody>
<tr>
<td>AstraZeneca</td>
<td><a href="http://www.TAGRISSOhcp.com">www.TAGRISSOhcp.com</a></td>
<td>6-7</td>
</tr>
<tr>
<td>AstraZeneca</td>
<td>cobaseEGRFtest.com</td>
<td>8</td>
</tr>
<tr>
<td>AstraZeneca</td>
<td>cobaseEGRFtest.com</td>
<td>9</td>
</tr>
<tr>
<td>AUDIT MicroControls, Inc.</td>
<td><a href="http://www.auditmicro.com">www.auditmicro.com</a></td>
<td>1</td>
</tr>
<tr>
<td>BD Diagnostics</td>
<td><a href="http://BDPhoenixM50.com">http://BDPhoenixM50.com</a></td>
<td>23</td>
</tr>
<tr>
<td>BD Diagnostics</td>
<td>BD.com/DS</td>
<td>3</td>
</tr>
<tr>
<td>Beckman Coulter, Inc.</td>
<td><a href="http://www.beckmancoulter.com/urinalysis">www.beckmancoulter.com/urinalysis</a></td>
<td>17</td>
</tr>
<tr>
<td>Bio-Rad Laboratories</td>
<td>bio-rad.com/diagnostics</td>
<td>26</td>
</tr>
<tr>
<td>Bio-Rad Laboratories</td>
<td>bio-rad.com/diagnostics</td>
<td>27</td>
</tr>
<tr>
<td>DAKO</td>
<td><a href="http://www.dako.com/PDL12c3">www.dako.com/PDL12c3</a></td>
<td>BC</td>
</tr>
<tr>
<td>Dako, An Agilent Technologies</td>
<td>dako.com/us/PDL1288-learning</td>
<td>14</td>
</tr>
<tr>
<td>Hologic - HIV</td>
<td>USApartimaVirology.com</td>
<td>1BC</td>
</tr>
<tr>
<td>Hologic, Inc.</td>
<td><a href="http://www.hologic.com">www.hologic.com</a></td>
<td>IFC</td>
</tr>
<tr>
<td>Kamiya Biomedical Co.</td>
<td><a href="http://www.k-assay.com/MLO.php">www.k-assay.com/MLO.php</a></td>
<td>19</td>
</tr>
<tr>
<td>Michigan State University</td>
<td><a href="http://www.bld.nataci.msu.edu-online-education">www.bld.nataci.msu.edu-online-education</a></td>
<td>37</td>
</tr>
<tr>
<td>Millipore Sigma</td>
<td>sigma-aldrich.com/clinal</td>
<td>5</td>
</tr>
<tr>
<td>Randox Laboratories</td>
<td><a href="http://www.therxseries.com">www.therxseries.com</a></td>
<td>25</td>
</tr>
<tr>
<td>Roche Diagnostics</td>
<td><a href="http://www.usdiagnostics.roche.com">www.usdiagnostics.roche.com</a></td>
<td>29</td>
</tr>
<tr>
<td>Sekisui Diagnostics</td>
<td><a href="http://www.osomtests.com">www.osomtests.com</a></td>
<td>31</td>
</tr>
<tr>
<td>Streck</td>
<td>streck.com</td>
<td>11</td>
</tr>
</tbody>
</table>

This index is provided as a service. The publisher does not assume liability for errors or omissions.
Microbiology enters the Age of Automation

If you were explaining COPAN Diagnostics to someone who is not familiar with the organization, how would you characterize its primary areas of expertise? For someone who is not familiar with COPAN, I would start by describing our unique corporate culture. We strive to create an environment that fosters creativity and rewards innovation. We have a very strong culture and a low employee turnover. Being a family-owned company, we can make decisions based on our convictions and values, keeping in mind that, even though our products are used at the beginning of the diagnostics process, there is a patient at the end.

COPAN has two areas of expertise: sample collection and preservation for analysis, and automation. As for collection and preservation of biological samples, that applies to infectious disease diagnostics, human DNA, environmental testing, forensics investigations, and human genome typing. COPAN entered the automation arena in 2008. Since then, we have become a major player in the revolution of total automation in clinical microbiology, specifically sample processing, image analysis, and robotic work-up to improve efficiency, productivity, and turnaround time—and thus overall patient care.

Your website refers to the company’s “collaborative approach to innovation.” Can you explain that further? COPAN’s development of automated sample processors was done very collaboratively with the clinical microbiology community. We developed the prototype of the Specimen Processor, put it in the back of an RV, and drove it on a 35,000-mile roadshow all across North America to get feedback from microbiologists, lab managers, and bench techs. Before that, the WASP automation existed for specimen processing, but it was restrictive and would only process one sample type, namely urine. With our automation, we wanted a system that would be modular and open and could manage all sample types received in microbiology labs. This approach could only be successful in specimen collection and transport systems and by moving the sample into liquid phase.

The initial feedback that we received about the specimen processor raised excellent questions: Can the system prepare a Gram slide automatically? Can the system apply antibiogram and ID disks? Can the system plant and streak using different manufacturers’ plates? Can the system inoculate enrichment broths, among others? Because COPAN is nimble and creative, we were able to invent different modules to satisfy our customers’ requests.

The evolution of our specimen processor was a perfect example of collaborating with the microbiology community. Microbiologists want to invest in specimen processing automation and digital microbiology because they need to find a solution to replace the vanishing skilled workforce and the lack of new people entering the labor force. COPAN is a completely vertically integrated company, with all the skills and know-how in-house, so this allows us to bring new innovations and product improvements to market quickly.

There is a great deal of emphasis on specimen collection and transport nowadays, both in terms of sample integrity and lab efficiency. How do COPAN solutions address that need? Sometimes small investments can have a big impact on diagnostics. The invention of flocked swabs by COPAN, more than 10 years ago, has revolutionized the quality of sample collection and transport because we were able to move away from traditional fiber-wrapped swabs that trapped more than 90 percent of the sample to flocked technology that releases most of the sample into liquid. The overall impact of this innovation was to increase sensitivity of different assays, amplified and not amplified. Virology laboratories embraced the innovation of flocked swabs because of the dramatic improvement of rapid detection of respiratory pathogens at the point of care and in the laboratory. The rapid migration to flocked swabs was very apparent in 2009, during the H1N1 pandemic, because flocked swabs provided a much easier sample collection alternative than nasopharyngeal aspirates or washes, with equivalent performance.

After the success and rapid adoption of flocked swabs in clinical Virology, the inventive devices caught the attention of Bacteriologists. From a bacteriology perspective, flocked swabs put high volume samples, like swabs, feces, and sputum, in a liquid format. Having the highest volume samples in liquid opened the door for automated specimen processing and digital Microbiology.

I understand that a large study recently affirmed WASP Lab Software’s effectiveness in detecting MRSA on chromogenic agar. Can you expand on that? When labs invest in this technology, they are not doing so simply to automate manual processes. They are also bringing artificial intelligence to the Microbiology lab, because we have to have a medium- to long-term plan to replace the skills of trained Microbiologists that we are losing. We have to be able to digitally read and interpret specimen cultures. Artificial intelligence is achieved by amassing many algorithms that can be applied to a WASP Lab 27 mega pixel image of a culture plate to look for specific organisms like MRSA, Vancomycin-resistant Enterococcus (VRE), or Group B strep, and also algorithms that can distinguish and segregate nongrowth from normal flora from a significant growth of a presumptive pathogen. WASP Lab pixel-rich digital images captured by the system provide a sample size of at least 1,200 pixels per 1mm diameter colony. This is an enormous amount of data available per colony and the bigger the colony, the more data, which ensures a very accurate and reliable growth interpretation. The first in a wave of publications that demonstrate the accuracy and reliability of COPAN WASP Lab algorithms was a recent multi-center study published in the Journal of Clinical Microbiology on COPAN’s MRSA reading algorithm. That publication has already been followed by a similar multicenter study that analyzed more than 100,000 patients on VRE.
In viral load testing

A change in direction is within sight.

COMING SOON

Aptima® HIV-1 Quant Assay

Additional Aptima assay targets in development:
HCV viral load & confirmation  • HBV viral load  • HSV 1 & 2  • M. genitalium  • BV  • Candida

Aptima assays now available:
CT/NG  • Trichomonas vaginalis  • HPV  • HPV GT  • Zika virus*

To get the full picture, visit USAptimaVirology.com

*The Aptima Zika Virus assay:
• This test has not been FDA cleared or approved;
• This test has been authorized by FDA under an EUA for use by authorized laboratories;
• This test has been authorized only for the detection of RNA from Zika virus and diagnosis of Zika virus infection, not for any other viruses or pathogens; and
• This test is only authorized for the duration of the declaration that circumstances exist justifying the authorization of the emergency use of in vitro diagnostic tests for detection of Zika virus and/or diagnosis of Zika virus infection under section 564(b)(1) of the Act, 21 U.S.C.§ 360bbb-3(b)(1), unless the authorization is terminated or revoked sooner.

The Aptima HIV-1 Quant assay is not available for sale in the U.S.
Leading the Way in PD-L1 Testing

PD-L1 IHC 22C3 pharmDx is the only companion diagnostic approved by the FDA as an aid in identifying metastatic NSCLC patients for treatment with KEYTRUDA® (pembrolizumab).

Intended Use

For in vitro diagnostic use.

PD-L1 IHC 22C3 pharmDx is a qualitative immunohistochemical assay using Monoclonal Mouse Anti-PD-L1, Clone 22C3 intended for use in the detection of PD-L1 protein in formalin-fixed, paraffin-embedded (FFPE) non-small cell lung cancer (NSCLC) tissue using EnVision FLEX visualization system on Autostainer Link 48. PD-L1 protein expression is determined by using Tumor Proportion Score (TPS), which is the percentage of viable tumor cells showing partial or complete membrane staining at any intensity. The specimen should be considered to have PD-L1 expression if TPS ≥ 1% and high PD-L1 expression if TPS ≥ 50%.

PD-L1 IHC 22C3 pharmDx is indicated as an aid in identifying NSCLC patients for treatment with KEYTRUDA® (pembrolizumab). See the KEYTRUDA® product label for expression cutoff values guiding therapy in specific clinical circumstances.

Read more at www.dako.com/pdl122c3

KEYTRUDA is a registered trademark of Merck Sharp & Dohme Corp., a subsidiary of Merck & Co., Inc.

1. PD-L1 IHC 22C3 pharmDx - Package Insert.