Detection of previously missed pathogens by a fully-validated metagenomics-based test

Robert Schlaberg, MD, Dr Med, MPH
Assistant Professor (University of Utah)
Medical Director (ARUP Laboratories)
Chief Medical Officer (IDbyDNA Inc.)
From Case Reports to Analytical Performance and Clinical Utility
>90% Agreement with PCR Panel
NP/OP Swabs (Viral Pathogens), RNA-seq & Taxonomer

I. Positives (n=42)
II. Unselected (n=67)

NP Swabs (n=109)

PCR Panel (RVP)

RNA-seq (HiSeq)

5-10 M reads/sample

Graf ... Schlaberg, J Clin Microbiol. 2016 Apr;54(4):1000-7
Putative Pathogens in 30% of Children with Pneumonia of Previously Unknown Etiology

NP/OP Swabs (Viral Pathogens), RNAseq & Taxonomer

Schlaberg et al., J Infect Dis. 2017 May 1;215(9):1407-1415
Putting it Together
A First **Validated Test**
ARUP Laboratories

3500+ Employees
~50,000 Samples per Day

Anatomic Pathology
Genetics
Hemostasis
Immunology

Infectious Disease
Oncology
Pain Management
Pediatrics

Strong Focus on Quality
Automated Sample Handling

Enterprise of the University of Utah

http://www.aruplab.com/
Workflow

Extract → Library Prep QC → Sequence → Analyze Review → Report Interface

PC NC

Spike IC

RNA DNA
Validated: End-to-End Workflow

- External Controls
- Internal Controls

Specimens
- Pos.
- Neg.

Wet Lab
- Sequencing QC
- Sample QC
- Repeat Algorithms

Analysis
- Databases
- Algorithms
- Cutoffs

- Sensitivity
- Specificity
- Accuracy
- Reproducibility
- Stability
Validated: Virtual Specimens

Specimens
Pos. Neg.

Data x10s

Virtual Matrix

Virtual Specimen x1000’s

Pathogens
Near neighbors
Commensals
Contaminants

✓ Sensitivity
✓ Specificity
✓ Accuracy
✓ Reproducibility
✓ Stability
# Diagnostic-Grade Sample-to-Report Metagenomics

Simplifying Workflows for Diagnostic Labs

<table>
<thead>
<tr>
<th>Sample &amp; Library Prep</th>
<th>Sequencing</th>
<th>Analysis &amp; Reporting</th>
</tr>
</thead>
<tbody>
<tr>
<td>Workflow Manager</td>
<td>Unbiased Sequencing</td>
<td>Val. Classification</td>
</tr>
<tr>
<td>Wet Bench Workflow</td>
<td></td>
<td>Reporting</td>
</tr>
<tr>
<td></td>
<td></td>
<td>QC/QA</td>
</tr>
</tbody>
</table>

- Validated
  - Databases
  - Algorithms
  - Cutoffs
Validated Quality Control Parameters

A. Run-Level

B. Sample-Level

C. Run-Level Quality Parameters

D. Sample-Level Quality Parameters

Normalization Improves Reproducibility

- HCoV (n=7)
- HMPV (n=4)
- HRV-A (n=4)
- HRV-B (n=5)
- M. pneumoniae (n=7)
- S. aureus (n=8)
- S. maltophilia (n=9)
- P. jirovecii (n=9)
Diagnostic Yield

A First Application Study
Previously Missed Pathogens in ~40% of Children Immunocompromised, Pneumonia, ICU

A

Explify™ Respiratory Results

- **Viral**
- **Fungal**
- **Bacterial**

Positive

Invalid

B

Number of Pathogens/Case

- **4**
- **2**
- **1**

<table>
<thead>
<tr>
<th>Viruses</th>
<th>Parainfluenza virus</th>
<th>Human Bocavirus</th>
<th>Rhinovirus</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Fungi</strong></td>
<td>Pneumocystis</td>
<td>Mucor</td>
<td>Fusarium</td>
</tr>
<tr>
<td><strong>Bacteria</strong></td>
<td>Staph. aureus</td>
<td>Strep. pneumoniae</td>
<td>S. maltophilia</td>
</tr>
</tbody>
</table>

Graf ... Schlaberg, manuscript in preparation
Adding Additional Layers of Information

- Presence/Absence of pathogen
- Quantification
- Drug resistance
- Epidemiologic typing
- Host-based diagnostics