Biomarkers for ARDS – not so simple

John Laffey
Critical Illness and Injury Research Centre
St Michael’s Hospital,
University of Toronto,
CANADA
<table>
<thead>
<tr>
<th></th>
<th>Mild</th>
<th>Moderate</th>
<th>Severe</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Acute Onset</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Low Oxygen (Hypoxia)</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Lung Water</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Abnormal Chest X-Ray</strong></td>
<td></td>
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</tr>
</tbody>
</table>

**No definitive test for ARDS**
‘Hallmark’ of ARDS

Diffuse Alveolar Damage
Comparison of the Berlin Definition for Acute Respiratory Distress Syndrome with Autopsy

Arnaud W. Thille\textsuperscript{1,3}, Andrés Esteban\textsuperscript{1}, Pilar Fernández-Segoviano\textsuperscript{2}, José-Maria Rodriguez\textsuperscript{2}, José-Antonio Aramburu\textsuperscript{2}, Oscar Peñuelas\textsuperscript{1}, Irene Cortés-Puch\textsuperscript{1}, Pablo Cardinal-Fernández\textsuperscript{1}, José A. Lorente\textsuperscript{1}, and Fernando Frutos-Vivar\textsuperscript{1} Am J Respir Crit Care Med Vol 187, Iss. 7, pp 761–767, Apr 1, 2013

712 autopsies over a 20 year-period

Exclusion: \( N = 264 \)
- Dead at admission: \( N = 50 \)
- Dead without mechanical ventilation: \( N = 38 \)
- Absence of risk factor for ARDS: \( N = 51 \)
- Missing data: \( N = 30 \)
- Acute Pulmonary Edema: \( N = 88 \)
- Pulmonary fibrosis: \( N = 7 \)

Patients with risk factors of ARDS: \( N = 448 \)

Patients with clinical criteria of ARDS: \( N = 356 \)

Mild ARDS: \( N = 49 \) (14%)
Moderate ARDS: \( N = 141 \) (40%)
Severe ARDS: \( N = 166 \) (46%)
Comparison of the Berlin Definition for Acute Respiratory Distress Syndrome with Autopsy

Arnaud W. Thille¹,³, Andrés Esteban¹, Pilar Fernández-Segoviano², José-Maria Rodriguez², José-Antonio Aramburu², Oscar Peñuelas¹, Irene Cortés-Puch¹, Pablo Cardinal-Fernández¹, José A. Lorente¹, and Fernando Frutos-Vivar¹

Am J Respir Crit Care Med  Vol 187, Iss. 7, pp 761–767, Apr 1, 2013
Current diagnostic Precision of ARDS Definition

- ARF Patients
- Patients fulfilling ARDS Diagnostic Criteria
- ARF Patients with ARDS

- ARF Patients that don’t have ARDS
Biomarkers – what are they and how might they help?

2001 National Institutes of Health definition, a biomarker is “a characteristic that is objectively measured and evaluated as an indicator of normal biological processes, pathogenic processes, or pharmacologic responses to a therapeutic intervention”
Biomarkers—focusing on biology of ARDS
Potential of Biomarkers

- To enhance diagnostic precision of current clinical definition of ARDS
- To enhance recognition of ARDS
- To predict ARDS in at risk populations
- To yield insights into pathogenesis
- To enhance risk stratification
- To facilitate ‘splitting’ of ARDS into meaningful subtypes [endotypes] to reduce heterogeneity
- To predict response to therapy / Drug responsiveness
Enhancing Precision of ARDS Definition

ARF Patients that don’t have ARDS

ARF Patients Fulfilling ARDS Criteria
Biomarkers to enhance ARDS Recognition

• Potential to implement evidence based strategies to prevent further injury to the Lung
  – Protective Ventilation
  – Fluid Restriction
  – Neuromuscular Blockade
  – Prone Positioning

• Evidence that these are under-utilized especially in patients in whom ARDS is not recognized.
ARDS definition:
- P/F ratio ≤300
- Bilateral opacities on CXR
- PEEP or CPAP≥ 5 cmH₂O
- Rule out cardiac origin
- Onset less than 1 week

<table>
<thead>
<tr>
<th>Extent of ARDS Recognition</th>
<th>Mild</th>
<th>Moderate</th>
<th>Severe</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Recognition at Day 1 (%)</td>
<td>177 (25%)</td>
<td>364 (33%)</td>
<td>234 (42%)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Recognition at any time (%)</td>
<td>366 (51%)</td>
<td>710 (65%)</td>
<td>434 (78%)</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

Unpublished Data
Biomarkers could enhance prediction of ARDS development, allowing early targeted interventions

- Potential to prevent ARDS in ‘at risk’ patients.

Implementation of a ‘bundle’ of Strategies to prevent ARDS including:

- Optimal mechanical ventilation
- Aggressive resuscitation
- Reduction in transfusion
- Prevention of common complications

Reduced ARDS incidence in Olmsted County from 81 to 38.3 cases per 100,000 person-years [Li et al, AJRCCM 2012]

- Decrease was driven by reduction in ‘hospital acquired’ ARDS
Prognosticating outcome in ARDS

• Understanding key factors contributing to poor outcome in ARDS

• Focusing high cost and/or high complexity interventions to population with greatest mortality risk

• Target for future studies of interventions for ARDS
What are the promising Biomarkers?
Candidate Biomarkers for ARDS

Lung Epithelium
- Surfactant protein B, D
- sRAGE (type 1 pneumocytes)
- KL-6
- CC-16; CCSP

Inflammatory Response
- TNFα; IL-8; IL-6, PAI-1, IL-18

Lung Endothelium
- vWF; Ang2
- P selectin; ICAM-1

Other
- Physiologic Data
- Gene Polymorphisms
The ‘ideal’ Biomarker

- Easily, cheaply, quickly and reliably measured...ideally near bedside

- Related to Pathogenesis – insights into injury/inflammatory process
  - Related to phase of disease?

- Low or non-invasive test
  - Exhaled condensate vs. plasma vs. BALF vs. Tissue sample

- High Sensitivity and Specificity

- Provides diagnostic and classification information

- Provides prognostic information
Diagnosis of ARDS
# Prediction of outcome of ARDS

## Plasma Biomarkers for Acute Respiratory Distress Syndrome: A Systematic Review and Meta-Analysis

Matty L. Terpstra, BSc; Jurjan Aman, MD; Geerten P. van Nieuw Amerongen, PhD; A. B. Johan Groeneveld, MD, PhD, FCCP, FCCM

Critical Care Medicine  March 2014 • Volume 42 • Number 3

### IL-1β

<table>
<thead>
<tr>
<th>Study name</th>
<th>Sample size</th>
<th>Odds ratio (95% CI)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lee '10</td>
<td>44</td>
<td>1.5 (0.4 - 5.5)</td>
<td>0.52</td>
</tr>
<tr>
<td>Lin '10</td>
<td>63</td>
<td>1.4 (0.6 - 3.4)</td>
<td>0.60</td>
</tr>
<tr>
<td>Bauer '00</td>
<td>46</td>
<td>1.9 (0.6 - 6.3)</td>
<td>0.27</td>
</tr>
<tr>
<td>Meduri '95</td>
<td>21</td>
<td>50.3 (7.0 - 358.5)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Headley '95</td>
<td>34</td>
<td>15.6 (4.0 - 63.3)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>206</strong></td>
<td><strong>4.3 (1.3 - 14.4)</strong></td>
<td><strong>&lt;0.01</strong></td>
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</table>

### TNF-α

<table>
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<th>Sample size</th>
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<th>P-value</th>
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<tbody>
<tr>
<td>Lin '10</td>
<td>63</td>
<td>1.6 (0.6 - 3.7)</td>
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<tr>
<td>Lee '10</td>
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<td>2.6 (0.8 - 10.2)</td>
<td>0.12</td>
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<tr>
<td>Chen '09</td>
<td>88</td>
<td>2.2 (1.0 - 4.8)</td>
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<tr>
<td>Bauer '00</td>
<td>46</td>
<td>1.8 (0.6 - 5.9)</td>
<td>0.32</td>
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<tr>
<td>Headley '95</td>
<td>34</td>
<td>15.6 (4.0 - 63.3)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Meduri '95</td>
<td>21</td>
<td>50.3 (7.0 - 358.5)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>206</strong></td>
<td><strong>3.9 (1.6 - 9.4)</strong></td>
<td><strong>&lt;0.01</strong></td>
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### IL-8

<table>
<thead>
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<th>P-value</th>
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<tbody>
<tr>
<td>Lee '10</td>
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<td>2.4 (0.7 - 8.8)</td>
<td>0.18</td>
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<tr>
<td>Ware '10</td>
<td>628</td>
<td>1.8 (1.3 - 2.5)</td>
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<tr>
<td>Lin '10</td>
<td>63</td>
<td>2.9 (0.8 - 5.5)</td>
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<td>McClintock '08</td>
<td>50</td>
<td>5.5 (1.9 - 16.0)</td>
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<tr>
<td>Parsons '05</td>
<td>780</td>
<td>1.7 (1.3 - 2.2)</td>
<td>&lt;0.01</td>
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<tr>
<td>Headley '95</td>
<td>34</td>
<td>15.8 (4.0 - 63.3)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Meduri '95</td>
<td>21</td>
<td>50.3 (7.0 - 358.5)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Groeneveld '95</td>
<td>13</td>
<td>7.3 (0.8 - 64.1)</td>
<td>0.07</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>1533</strong></td>
<td><strong>3.4 (2.0 - 5.7)</strong></td>
<td><strong>&lt;0.01</strong></td>
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### IL-6

<table>
<thead>
<tr>
<th>Study name</th>
<th>Sample size</th>
<th>Odds ratio (95% CI)</th>
<th>P-value</th>
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<tbody>
<tr>
<td>Nakamura '11</td>
<td>20</td>
<td>1.4 (0.3 - 6.3)</td>
<td>0.66</td>
</tr>
<tr>
<td>Lee '10</td>
<td>44</td>
<td>4.8 (1.2 - 17.0)</td>
<td>0.16</td>
</tr>
<tr>
<td>Ware '10</td>
<td>621</td>
<td>1.7 (1.2 - 2.4)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Parsons '05</td>
<td>781</td>
<td>1.7 (1.3 - 2.2)</td>
<td>&lt;0.01</td>
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<td>21</td>
<td>50.3 (7.0 - 358.5)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Groeneveld '95</td>
<td>13</td>
<td>64.2 (4.9 - 846.6)</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>1488</strong></td>
<td><strong>3.4 (1.8 - 6.3)</strong></td>
<td><strong>&lt;0.01</strong></td>
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</tbody>
</table>
Prediction of Development of ARDS

Plasma Angiopoietin-2 Predicts the Onset of Acute Lung Injury in Critically Ill Patients

Am J Respir Crit Care Med Vol 187, Apr 1, 2013

Ashish Agrawal¹, Michael A. Matthay²,³,⁴, Kirsten N. Kangelaris³, John Stein⁵, Jeffrey C. Chu⁴, Brandon M. Imp³, Alfredo Cortez⁵, Jason Abbott⁴, Kathleen D. Liu²,³, and Carolyn S. Calfee²,³
Distinct Molecular Phenotypes of Direct vs Indirect ARDS in Single-Center and Multicenter Studies

Carolyn S. Calfee, MD, MAS; David R. Janz, MD; Gordon R. Bernard, MD, FCCP; Addison K. May, MD; Kirsten N. Kangelaris, MD, MAS; Michael A. Matthay, MD, FCCP; Lorraine B. Ware, MD, FCCP; and the NIH NHLBI ARDS Network

CHEST 2015; 147(6):1539-1548

A

\[ p=0.0004 \]

\[ 500 \]

\[ 400 \]

\[ 300 \]

\[ 200 \]

\[ 100 \]

\[ 0 \]

SP-D (ng/ml)

Indirect

Direct

B

\[ p=0.001 \]

\[ 10,000 \]

\[ 8,000 \]

\[ 6,000 \]

\[ 4,000 \]

\[ 2,000 \]

\[ 0 \]

RAGE (pg/ml)

Indirect

Direct

C

\[ p=0.0005 \]

\[ 50,000 \]

\[ 40,000 \]

\[ 30,000 \]

\[ 20,000 \]

\[ 10,000 \]

\[ 0 \]

Ang-2 (pg/ml)

Indirect

Direct

D

\[ p=0.28 \]

\[ 2,000 \]

\[ 1,500 \]

\[ 1,000 \]

\[ 500 \]

\[ 0 \]

IL-6 (pg/ml)

Indirect

Direct
So why are we not using Biomarkers then...?
No ‘ideal’ Biomarker

- Easily, cheaply, quickly and reliably measured...ideally near bedside

- Related to Pathogenesis – insights into injury/inflammatory process
  - Related to phase of disease?

- Low or non-invasive test
  - Exhaled condensate vs. plasma vs. BALF vs. Tissue sample

- High Sensitivity and Specificity

- Provides diagnostic and classification information

- Provides prognostic information
<table>
<thead>
<tr>
<th>Test Result</th>
<th>Actual Result</th>
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</thead>
<tbody>
<tr>
<td>Positive</td>
<td><strong>True Positive</strong></td>
</tr>
<tr>
<td></td>
<td><strong>False Positive</strong> (Statistical analysis)</td>
</tr>
<tr>
<td></td>
<td>$\alpha = 0.05$</td>
</tr>
<tr>
<td>Negative</td>
<td><strong>False Negative</strong> (Power analysis)</td>
</tr>
<tr>
<td></td>
<td>$1 - \beta = 0.20$</td>
</tr>
<tr>
<td></td>
<td><strong>True Negative</strong></td>
</tr>
</tbody>
</table>
Sensitivity and Specificity

• The performance of a laboratory test is commonly reported in terms of sensitivity and specificity defined as:

  • **Sensitivity** = \[ \text{True Positives} \].
    All patients with Disease (TP + FN)

  • **Specificity** = \[ \text{True Negatives} \].
    All patients without disease (TN+ FP)
Plasma Angiopoietin-2 Predicts the Onset of Acute Lung Injury in Critically Ill Patients

Ashish Agrawal¹, Michael A. Matthay²,³,⁴, Kirsten N. Kangelaris³, John Stein⁵, Jeffrey C. Chu⁴, Brandon M. Imp³, Alfredo Cortez⁵, Jason Abbott⁴, Kathleen D. Liu²,³, and Carolyn S. Calfee²,³

\[ p = 0.0008 \]

Option 1

Option 2

\[ \text{N = 148} \]
\[ \text{N = 19} \]
Interpreting a Test Result

- As a clinician examining a positive test, we are most interested in determining whether a patient actually has disease.
PPV and NPV for Serum Ang-2

• Option 1 – ‘Specific’ Option

\[
\text{PPV} = \frac{19}{38} = 0.50
\]

\[
\text{NPV} = \frac{120}{129} = 0.93
\]
What is the future for Biomarkers...?
Plasma Angiopoietin-2 Predicts the Onset of Acute Lung Injury in Critically Ill Patients

Ashish Agrawal¹, Michael A. Matthay²,³,⁴, Kirsten N. Kangelaris³, John Stein⁵, Jeffrey C. Chu⁴, Brandon M. Imp³, Alfredo Cortez⁵, Jason Abbott⁴, Kathleen D. Liu²,³, and Carolyn S. Calfee²,³

Am J Respir Crit Care Med Vol 187, Apr 1, 2013
Clinical Predictors
- APACHE III
- Organ failures
- Age
- Underlying cause
- A-a DO2
- Plateau pressures

Biomarkers
- APACHE III
- vWF antigen,
- Surfactant D
- sTNF receptor-1,
- IL-6
- IL-8
- ICAM -1
- Protein C
- PAI-1

Reduced Model
- APACHE III
- Age
- Surfactant D
- IL-8
Latent Class Modeling [searching for ‘endotypes’]
- Examining large datasets and fitting to data distribution model
- Determine whether data fit in 1 or multiple distributions and find least number of distributions that best explain the dataset

Calfee C et al, Lancet Resp Medicine 2014
- Examined ARMA and ALVEOLI trials and found data fitted a 2 class model
- Class 1 vs Class 2 [hyperinflammatory]
- Class 2 different [more IL-6, sTNFr-1, more vasopressor use, higher mortality]
Subphenotypes in acute respiratory distress syndrome: latent class analysis of data from two randomised controlled trials

Carolyn S Calfee, Kevin Delucchi, Polly E Parsons, B Taylor Thompson, Lorraine B Ware, Michael A Matthay, and the NHLBI ARDS Network

Phenotype 1
(n=404)

Phenotype 2
(n=145)
### Table 4: Association between phenotype assignment and clinical outcomes, adjusted for degree of uncertainty regarding phenotype assignment

<table>
<thead>
<tr>
<th></th>
<th>ARMA cohort</th>
<th>ALVEOLI cohort</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Ph. 1 (n=318)</td>
<td>Ph. 2 (n=155)</td>
</tr>
<tr>
<td>Mortality (at 90 days)</td>
<td>23%</td>
<td>44%</td>
</tr>
<tr>
<td>Ventilator-free days</td>
<td>17.8</td>
<td>7.7</td>
</tr>
<tr>
<td>Organ failure-free days</td>
<td>14.5</td>
<td>8.0</td>
</tr>
</tbody>
</table>

Values are estimated means that take into account the uncertainty of class membership.
Biomarkers...summary

• Biomarkers offer the potential to enhance our capacity to
  – Predict development ARDS
  – Aid in making ARDS diagnosis
  – Prognostication regarding outcome
  – Response to therapy

• However, no single Biomarker likely to meet all needs

• Combining panels of Biomarkers with different sources shows promise
  – Markers of endothelial and epithelial injury, and of inflammation

• Combining with Clinical data enhances predictive validity

• May eventually play a key role in ARDS phenotyping [ARDS ‘endotypes’]

• Likely to guide future clinical trials
  – ‘personalized medicine’