POR Testing for Platelet Bacterial Contamination: An ongoing risk under continuous improvement

8th July 2012.
Wm. Andrew Heaton¹ MD
NSLIJ Health System & Hofstra NSLIJ School of Medicine
The views are those of the presenter and not of the NSLIJ Health System or Hofstra

¹Andrew Heaton has received research support and/or honoraria from Verax, Hemonetics, Fenwal, & Immunetics, and has consulted for Beckman-Coulter, Verax, & Novartis Diagnostics
Objectives:

1. Describe platelet bacterial risks and recent intervention effects.
2. Discuss residual risk due to false negative bacterial cultures.
3. Relate residual bacterial contamination to clinically effects.
4. Review the Point of Release testing:
   - Outcome & Feasibility
5. Effect on Transfusion Related Death & US Regulatory process.
6. Summarize the policy related questions.

Current Situation:

- Reported US platelet bacterial contamination fatality rate is \(~1.5\) deaths per million PC doses transfused (\(~3\) deaths/year)
- The Bacterial Testing Issue:
  - Culture as a release test has a \(~26\%)\ sensitivity
  - 231 out of 893/million contaminated units detected
  - Only \(~10\%)\ transfusion sepsis is reported
Oct 95 – Sep 04, 60 FDA contaminated PC reports of Post-Tx fatality
- 38 of the 60 (63.3%) cases were gram-negative organisms
- ~ 2/3 of post-transfusion sepsis organisms were gram-positive

Effect of Skin Preparation, Inlet-line Diversion & Culture Upgrade
- ARC septic reactions decreased from 1:40,000 to 1:86,000, ~ 50% reduction
- JHU decrease from 7.45/100K to 2/100K transfusions, 70% reduction

FDA reported death decreased 60% (7/yr in 2001-3) to 2.8/yr 2006-10

aaBB Bacterial Assay Task Force 2012
Confirmed positive cases (3/1/2004 – 1/31/2007):

**Gram positive (n=196):**
- Staphylococcus, coagulase negative 87
- Staphylococcus epidermidis 22
- Staphylococcus aureus 13
- Staphylococcus (others) 13
- Streptococcus sp. 43
- Bacillus sp. 8
- Enterococcus sp. 3
- Listeria monocytogenes 4
- Lactobacillus/Micrococcus/Unspecified 1 each

**Gram negative (n=29):**
- Escherichia coli 12
- Serratia marcescens 6
- Klebsiella sp. 8
- Citrobacter/Enterobacter/Unspecified 1 each

Eder AF et al, Transfusion 2007
Bacterial Safety Interventions and Effects

**Inline Diversion – Prestorage PC**

- BacT/ALERT Positive per 10^6 Cultures
  - No Diversion: 2000
  - Diversion: 500
  - O.R. = 0.46 (95% C.I. 0.21-0.95)


**Arm Prep – Chloraprep Vs Povidone I₂**

- O.R. = 0.47 (95% C.I. 0.21-1.03)

**Platelets, USA 1995 to 2010**

- Average number of cases per year
  - Gram negative: 0.8 (1995-2004)


2005-2010:

- Pooled Platelets microorganisms: S. aureus (1), E. coli (2), S. dysgalactiae (1), S. pneumoniae (1)
- Platelets Pheresis microorganisms: S. aureus (6), S. marcescens (1), S. lugdunensis (1), S. epidermidis (2), E. limosum (1), E. coli (1), M. morganii (1), K. oxytoca (1), S. viridans (1), S. warneri (1)

North Shore LIJ
North Shore-Long Island Jewish Health System

## Bacterial Residual Risk post BacT/Alert Screen

<table>
<thead>
<tr>
<th></th>
<th># Tested</th>
<th>Confirmed +’ve</th>
<th>Rate /10⁶</th>
<th>Ref.</th>
</tr>
</thead>
<tbody>
<tr>
<td>PASSPORT</td>
<td>6,039</td>
<td>4</td>
<td>662 (1:1,509)</td>
<td>Dumont 2010</td>
</tr>
<tr>
<td>Irish BS Day 8</td>
<td>8,282</td>
<td>7</td>
<td>1,183 (1:850)</td>
<td>Murphy 2008</td>
</tr>
<tr>
<td>Irish BS Day 4</td>
<td>3,310</td>
<td>1</td>
<td>3,310 (1:302)</td>
<td>Murphy 2008</td>
</tr>
<tr>
<td>Welsh BS</td>
<td>6,438</td>
<td>6</td>
<td>931 (1:1,074)</td>
<td>Pearce 2011</td>
</tr>
<tr>
<td>Combined</td>
<td>24,369</td>
<td>18</td>
<td>1,353 (1:740)</td>
<td></td>
</tr>
</tbody>
</table>

### Sensitivity of culture

- **U.S. standard practice**: 25.9%
- **best practice**: 33%

**Passive Surveillance results in 10.6 times less likely to detect a septic reaction**

---

Murphy et al. Vox Sanguinis 2008
Dumont et al. Transfusion: 50; 589; 2010.
Limitations of Early Culture Testing

Modeling the effect of concentration on bacterial detection when a 300 mL unit is contaminated with 0-300 CFUs (0-1 CFU/mL). The figure shows the probability curves for an 8-mL sample divided into two culture bottles.

Bacterial Contamination of Platelets

University Hospitals Case Medical Center, Cleveland, OH
1991-2010  N=68

No. of cases

Coag-neg. staphylococcus
Pseudomonas aeruginosa
Bacillus cereus
Staphylococcus aureus
Serratia marcescens
Streptococcus bovis
Viridans group streptococcus
VGS+CoNS
Staphylococcus lugdunensis
Acinetobacter baumannii

No active surveillance performed

Bacterial load, species virulence, & Tx reaction

**Bacterial load ≥10^5 vs. <10^5 cfu/ml:**
- Any reaction: 16/23 vs 4/23 OR 4.0 (1.5-5.8)
- Severe reaction: 8/23 vs 0/23 OR >34

**Reaction grade:**
- None
- Mild
- Moderate
- Severe
- Life-threatening
- Fatal

Bacterial Contamination Sampling Time Issues

Issues:
- Poisson sampling
- Late growers

Positive units (Gram- ‘ve+ ) are Interdicted
- Day 2+ Negative Units released:
- Any late positives recalled

Days 3-5 Late Positives:
Usually Gram +’ce cocci

Arm Prep
↓ Risk ~ 0.47

Inlet Diversion
↓Risk ~ 0.46

Benjamin, Kline et al. Transfusion 2008
Benjamin et al Transfusion 2011
Multi-site Study of 27,682 PC with PGD® Assay

- Study performed at 18 study sites by over 160 technologists on apheresis units previously tested by culture negative (BacT/ALERT or eBDS) PC
- Doses tested by Platelet PGD test on day of issue (16 sites) or shortly after issue (2 sites) according to the manufacturer’s directions
- Positive PGD results repeated in duplicate and plate cultures performed
- Concurrent aerobic plate cultures were also performed on 10,430 units at three of the study sites, with quantitation of positives at one study site
- Single-use, qualitative test
- Detects the presence of conserved bacterial surface cell wall antigens, lipoteichoic acid and lipopolysaccharide, using specific antibodies

Analytic sensitivity in LRAP

- Staphylococcus aureus
- Pseudomonas aeruginosa
- Staphylococcus epidermidis
- Enterobacter aerogenes
- Bacillus cereus
- Klebsiella pneumoniae
- Escherichia coli
- Streptococcus agalactiae
- Clostridium perfringens
- Serratia marcescens
Multi-site Study of 27,682 PC with PGD® Assay

Valid PGD test results
N=27,620

Reactive
N=151

Culture Positive
N=9

Culture Negative
N=142

Non-reactive
N=27,469

Culture at 3 sites
N=10,344

Culture Positive
N=2

Culture Negative
N=10,342

Jacobs: Transfusion 51: 2573: 2011
# Platelet Bacterial Contamination – TP results

<table>
<thead>
<tr>
<th>Bacterial species isolated by culture at issue</th>
<th>Age of unit (days)</th>
<th>Confirmation method*</th>
<th>Bacterial load (CFU/ml)**</th>
<th>Transfusion status</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bacillus sp; P. acnes</td>
<td>3</td>
<td>BC</td>
<td>NT</td>
<td>Not Tx</td>
</tr>
</tbody>
</table>
| CoNS  
\[
\text{split collection}
\] | 3                  | PC, GS               | NT                        | Not Tx            |
| CoNS                                           | 3                  | PC, GS               | NT                        | Not Tx            |
| Enterococcus faecalis                         | 3                  | PC, GS               | NT                        | Not Tx            |
| CoNS; Peptostrep                              | 4                  | PC, BC, GS           | NT                        | Not Tx            |
| CoNS                                           | 4                  | PC                   | NT                        | Not Tx            |
| CoNS                                           | 5                  | PC, GS               | 1.3 x 10^6                | Tx – no rxn       |
| Bacillus sp.                                   | 5                  | PC, GS               | 1 x 10^7                  | Not Tx            |
| CoNS                                           | 5                  | PC, GS               | 1.2 x 10^7                | Tx – septic shock*** |

*BC = broth culture; PC = plate culture; GS = Gram stain; **NT = Not Tested for quantity
***documented bacteremia with same organism

Jacobs: Transfusion 51: 2573: 2011
PGD Detection Rates On Day of Release

Day 2 culture negative Apheresis Inventory – sampled on day 1

9/27,620 apheresis units PGD positive

- Rate of detection was 1/3,069 units (95% CI 1/6,711 - 1/1,617)

- Estimated 326 contaminated units per million units (95% CI 149-618)

- Based on 1.7 million LRAP units per year in the U.S., the estimated number of breakthroughs would be expected to be 554 per year (95% CI 253-1051)

Start with 893/MM contaminated:
- Culture detects 150-200/MM
- POR detects 326/MM
- Undetected ~ 192/MM

- Jacobs: Transfusion 51: 2573: 2011
Bacterial contamination @ 1:3,069 doses (326/million; 95% CI 149-618/million)

7 of 9 PGD+ units showed Gram Stain + contamination (~10^7 cfu)

2 false negatives detected in 10,424 doses (192/million) on DOR culture

There were 142 PGD false positives (0.51%)

Based on reaction rate in recipients transfused with >10^5 CFU/mL:
- This could prevent ~300 major Tx reactions & several fatalities/year
NSUH participated in an 18 center evaluation study of a rapid bacterial point-of-care screening assay (PGD®, Verax Biomedical)

- One of 3 sites that performed concurrent culture at issue.
- The PGD Test performed on day of receipt & daily thereafter.

Of the 59% tested, 14% retested @ 48 hours, & 1% @ 72 hours.

Feasibility was confirmed with next steps identified as:

- Definition of ‘acceptable’ hold periods following testing
- Implementation of IT to track inventory testing status
- Identification of the ‘real’ as opposed to ‘reported’ risk
Platelet shipment receipt entered in computer. Units placed on hold and barcode label printed.

Remove Pigtail or: Attach sampling pouch by Sterile docking to bag

• Detach sample, label, and:
  • add to test work sheet.
  • Transfer specimen into Testing tube

Run Manufacturer's Positive and Negative controls
Run two external positive controls (Gram Pos and Gram Neg)

POR testing performed and results Scan into computer system

Supervisory review and specimen released from hold

In-house split units quarantined and pH & gram stain performed

Send samples for:
• pH testing i& gram stain if IR
• Culture if RR

Notify BC if any positive

Discard Unit

Release Unit

2nd Round Testing with Diff Lot: Positive if Result = ½ 0r 2/2 +

First time Positive results

Negative on retest 2 / 2

Results entered and units placed in available inventory

Relabeled with test “Negative” sticker with date and time
### BacTx® Test Train & Sensitivity (510k Approved)

<table>
<thead>
<tr>
<th>Species</th>
<th># 1 Sensitivity</th>
<th># 2 Sensitivity</th>
<th>Overall</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Escherichia coli</em></td>
<td>$5.1 \times 10^3$</td>
<td>$8.7 \times 10^3$</td>
<td>$8.7 \times 10^3$</td>
</tr>
<tr>
<td><em>Pseudomonas aeruginosa</em></td>
<td>$9.6 \times 10^3$</td>
<td>$5.0 \times 10^4$</td>
<td>$5.0 \times 10^4$</td>
</tr>
<tr>
<td><em>Klebsiella oxytoca</em></td>
<td>$6.8 \times 10^3$</td>
<td>$9.9 \times 10^3$</td>
<td>$9.9 \times 10^3$</td>
</tr>
<tr>
<td><em>Serratia marcescens</em></td>
<td>$5.8 \times 10^4$</td>
<td>$6.7 \times 10^3$</td>
<td>$5.8 \times 10^4$</td>
</tr>
<tr>
<td><em>Propionibacterium acnes</em></td>
<td>$7.2 \times 10^3$</td>
<td>$1.1 \times 10^3$</td>
<td>$7.2 \times 10^3$</td>
</tr>
<tr>
<td><em>Staphylococcus aureus</em></td>
<td>$2.1 \times 10^3$</td>
<td>$4.0 \times 10^3$</td>
<td>$4.0 \times 10^3$</td>
</tr>
<tr>
<td><em>Staphylococcus epidermidis</em></td>
<td>$2.0 \times 10^3$</td>
<td>$2.4 \times 10^3$</td>
<td>$2.4 \times 10^3$</td>
</tr>
<tr>
<td><em>Streptococcus agalactiae</em></td>
<td>$3.6 \times 10^3$</td>
<td>$2.7 \times 10^4$</td>
<td>$2.7 \times 10^4$</td>
</tr>
<tr>
<td><em>Clostridium perfringens</em></td>
<td>$2.8 \times 10^3$</td>
<td>$4.5 \times 10^3$</td>
<td>$4.5 \times 10^3$</td>
</tr>
<tr>
<td><em>Bacillus cereus</em></td>
<td>$1.3 \times 10^3$</td>
<td>$1.7 \times 10^3$</td>
<td>$1.7 \times 10^3$</td>
</tr>
</tbody>
</table>
Bacterial Contamination Testing Standards

AABB standard 5.1.5.1 (effective March 2004)¹
The blood bank or transfusion service shall have methods to limit and detect bacterial contamination in all platelet components

Apheresis - Collection facilities adopted culture
- FDA cleared culture-based QC (BacT Alert & eBDS)
- Culture at 24hrs, release 12-24hrs later

WBD - Culture not practical for WBD units
- Hospitals validated non FDA cleared tests

AABB standard 5.1.5.1.1 (effective Jan 2011)² for WBDP
Detection methods shall either be approved by the FDA or validated to provide sensitivity equivalent to FDA-approved methods.

First High Profile Litigation affecting Hospital/Blood Center
- Testing and Recall Standards of Practice
- Policies & Procedures pertinent to Transfusion Reaction

1. AABB Standards for Blood Banks, March 2003
2. AABB Interim Proposed Standard, Posted Feb 2010
3. [Link to news article](http://www.tampabay.com/incoming/hillsborough-girl-had-cancer-but-suit-pins-death-on-tainted-blood/1179600)
<table>
<thead>
<tr>
<th>Issue</th>
<th>For action</th>
<th>Opposed to action</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Clinical Issue ?</strong></td>
<td>Well described sepsis/death risk:</td>
<td>None reported locally:</td>
</tr>
<tr>
<td></td>
<td>• Reports credible &amp; conservative</td>
<td>• Small reported fraction = ↓ perceived risk</td>
</tr>
<tr>
<td></td>
<td>Actual sepsis ?? 10 X under-reported:</td>
<td>• No standard-of-care &amp; minimal litigation</td>
</tr>
<tr>
<td></td>
<td>• Clinical significance hard to evaluate</td>
<td></td>
</tr>
<tr>
<td></td>
<td>• Increasingly G+ cocci - skin contaminant</td>
<td></td>
</tr>
<tr>
<td><strong>Economic Question</strong></td>
<td>Reimbursement focused on outcomes</td>
<td>BC’s reluctant to reduce product cost::</td>
</tr>
<tr>
<td></td>
<td>• Quality = purchaser selection criterion</td>
<td>• Low direct cost… not avoidable expense</td>
</tr>
<tr>
<td></td>
<td>• DRG rates affected by readmissions</td>
<td>• Hospital cost of sepsis is not reported</td>
</tr>
<tr>
<td></td>
<td>Culture already factored into unit cost:</td>
<td>Testing not the standard of practice:</td>
</tr>
<tr>
<td></td>
<td>• Maybe avoid BacT/Alert cost</td>
<td>• FDA &amp; aaBB do not require it</td>
</tr>
<tr>
<td></td>
<td>• No studies available</td>
<td>• Low assessment of liability</td>
</tr>
<tr>
<td><strong>Feasibility</strong></td>
<td>NSUH participated in trial (no yield):</td>
<td>Manufacturing not Distribution:</td>
</tr>
<tr>
<td></td>
<td>• Only tested routine units</td>
<td>• BC should ↑ culture sensitivity</td>
</tr>
<tr>
<td></td>
<td>• Showed feasibility in a study</td>
<td>• Hospitals ‘cannot’ test completely</td>
</tr>
<tr>
<td><strong>Options</strong></td>
<td>Test all PC pre-release:</td>
<td>Take no action:</td>
</tr>
<tr>
<td></td>
<td>• Test ‘at-risk’ patient’s PC</td>
<td>• Await regulatory leadership</td>
</tr>
<tr>
<td></td>
<td>• TPGD test reactions (define problem)</td>
<td>• Request BC’s to improve capture rate</td>
</tr>
</tbody>
</table>
Policy Related Questions

- **Clinical Questions:**
  - There is clinical evidence that Point-of-Release Testing is needed.

- **Feasibility Questions:**
  - These tests are do-able in a Blood Bank Environment.

- **Inventory Questions raised:**
  - Tested inventory can be maintained & could dating be extended?

- **Evaluations/studies that are needed:**
  - Larger culture samples & later sampling offer some improvement.
    - Current data suggests that it would be less than equivalent.
  - Affirmative studies are needed to define the test frequency interval.

- **Where we are today:**
  - Simple and effective Point-of-Release Testing is becoming available.
    - aaBB/FDA workshop on 17th July to review evidence Vs. standards.