Impact of rapid in situ hybridization testing on coagulase-negative staphylococci positive blood cultures

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Objectives: To evaluate the impact of the rapid differentiation of Staphylococcus aureus from coagulase-negative staphylococci (CoNS) in blood cultures using peptide nucleic acid fluorescence in situ hybridization (PNA FISH) on vancomycin usage, length of patient hospital stay and hospital costs.

Design: This was a retrospective, cost-effective analysis of PNA FISH in its initial 3 month implementation period in 2004 in a 650 bed academic medical centre. Blood cultures with Gram-positive cocci in clusters (GPCC) that were negative for S. aureus using the PNA FISH assay were compared with an untested control group in the same period that had similar illness severity and location. We evaluated the effectiveness of the early identification of CoNS by ruling out S. aureus in conjunction with an antimicrobial team (AMT) on antimicrobial therapy, patient length of stay and hospital costs.

Results: A total of 139 blood cultures positive with GPCC had PNA FISH results while 84 in the control group did not. Evaluable criteria were met in 53 patients in the PNA FISH group and 34 in the control group. When comparing the results obtained from using the PNA FISH assay with those for the control group, there was a significant reduction in median length of hospital stay from 6 to 4 days (P < 0.05, CI 0.95–1.87) and a trend towards less vancomycin usage with a decrease in associated hospital costs of $4000 per patient.

Conclusions: The PNA FISH assay is rapid, accurate and reliable and in association with an AMT could decrease hospital length of stay in patients with CoNS bacteraemia in non-intensive care unit settings and prevent excessive vancomycin usage.

Keywords: antimicrobial management, cost-savings, vancomycin, length of stay

Introduction

Staphylococci remain the most common organism isolated from blood cultures with the majority being coagulase-negative staphylococci (CoNS).1 CoNS constitute a frequent contaminant of blood cultures and can result in excessive use of vancomycin and consequently an increased length of hospital stay.2–4 This can result in excessive hospital costs due to decreased bed utilization and increased pharmacy costs. In addition, the patient may have a greater risk of acquiring an antibiotic-resistant organism or other nosocomial infection.5–11

Previously, several laboratory-based algorithms and tests have been developed to differentiate contamination from true bacteraemia. However, all such algorithms and tests have been shown to be of limited value due to poor sensitivity and specificity, and they rarely result in a decrease in antibiotic utilization.2,6,7,12,13 Recently, a peptide nucleic acid fluorescence in situ hybridization (PNA FISH) (AdvanDx, Woburn, MA, USA) assay to rapidly detect Staphylococcus aureus became available for use. This assay targets the 16S rRNA of S. aureus directly from blood cultures, with results available in ~3 h after the observation of Gram-positive cocci in clusters (GPCC).14,15

Early in 2004, the University of Maryland Medical Center (UMMC) initiated the use of the PNA FISH test on blood cultures positive with GPCC. A member of the antimicrobial team (AMT), which includes an infectious disease physician,
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a dedicated clinical pharmacist and the infectious disease fellows, was notified daily of the PNA FISH results. This article reviews the clinical impact of PNA FISH assay on vancomycin usage, on length of hospital stay and on hospital costs.

Methods

A retrospective chart review to evaluate the effect on patient care of an FDA licensed FISH assay (PNA FISH) for the detection of *S. aureus* bacteraemia was performed following approval from the Office of Human Protections.

Setting

The UMMC is a 650 bed tertiary care medical centre and a 90 bed shock trauma centre. The patient population treated includes a large inner city population. There is also an inpatient stem cell and leukaemia unit, a large solid organ transplant programme and an inpatient HIV service. Since 2000, the rates of methicillin-resistant *S. aureus* (MRSA) from all cultures has risen to over 50%, with higher rates in patients admitted with HIV, with substance abuse, or with a history of hospitalization or incarceration.16–18

Patient selection

A total of 223 episodes of bacteraemia due to GPCC in 223 patients were reviewed in the 3 month trial period. We compared all PNA FISH results negative for *S. aureus* with a control group of patients with positive blood cultures with GPCC that were not tested in this time period. The control group was similarly matched by age, sex, location and diagnosis as the PNA FISH group. The admitting diagnosis [as defined by the International Classification of Diseases (ICD-9)] was used to establish a similar level of illness severity and to standardize hospital costs.

Exclusion criteria

Duplicate blood culture specimens from the same patient that were drawn from separate sites were excluded from the analysis as these were considered significant. Cancer centre and trauma patients were also excluded since these services are not covered by the AMT because there is coverage by full-time infectious disease physicians. Major exclusion criteria included patients with non-removable intravascular prostheses (prosthetic heart valves, pacemakers, arteriovenous grafts, ventricular assist devices, etc.) where CoNS is unlikely to be a contaminant and require therapy.

Blood culture policy

A physician order is required for the initiation of drawing blood cultures. Any persons drawing blood cultures are to adhere to the following guidelines.

The caps are removed from the blood culture bottles and the rubber stopper of each bottle is cleansed with 70% alcohol before inoculating with blood. After selecting the venipuncture site, non-sterile gloves are put on and the skin is prepared using 70% alcohol for a minimum of 30 s. Once the alcohol has air-dried, chlorhexidine is applied in concentric circles away from the puncture site covering a circular area 1–2 inches in diameter. The skin antiseptic is allowed to air dry before venipuncture. From 8 to 10 mL of blood is collected into the blood culture bottle (Bactec 9000, Becton Dickinson, MD, USA) from two separate sites. The bottle is gently inverted to allow the medium to mix with the blood. After delivery to the laboratory, the bottles are placed in a continuous automated detection incubator (BacT/Alert). Blood draws from central lines are not to be performed unless directed by a physician.

Laboratory methods

The PNA FISH assay was batched daily in the evaluation period to minimize labour and laboratory costs. The start-up costs were minimal, requiring the purchase of a water bath and a lens filter for the UV light source microscope. The test takes ~3 h to run, which includes 45 min of technician’s time per batch (~$12 in labour).1 Costs for materials and controls are dependent on the number of tests and controls per batch. To test a single patient using the list reagent kit’s list price would cost $68. Presently, reimburse- ment is ~$30; however, by batching blood cultures with GPCC and testing daily (an average of five specimens per batch) we minimized the cost per test to have the reimbursement cover our costs. Prior to the implementation of reporting the PNA FISH results, the laboratory’s evaluation of the test showed a 100% accuracy for 549 blood cultures positive with GPCC that included 115 *S. aureus* and 434 CoNS (data not included).

Organisms

When GPCC were identified by Gram staining of signal-positive blood cultures, they were plated onto standard isolation plates as per standard protocol. When there was growth on the plates, the species of CoNS (catalase positive, coagulase negative) was identified using catalase (3% H2O2) and a latex coagulase test (Staphaurex, Murex Biotech Ltd, Dartford, UK).19

Result management

The Gram stain results of blood cultures with GPCC were reported to the primary clinical service as per normal protocol, with a second call later reporting the PNA FISH results. The AMT was informed of all PNA FISH results for GPCC since their approval was required to release vancomycin to the requesting physician. In the PNA FISH group, the AMT could determine the need for vancomycin usage based on the exclusion criteria, the negative result for *S. aureus*, and any patient haemodynamic instability, with the ability to limit doses of vancomycin released. In the control group, vancomycin would be released as normal pending culture results to the requesting physician and would require subsequent AMT intervention when the final culture results were returned. Because the AMT can only recommend discontinuation of vancomycin usage, once it is released for treatment, it requires the treating physician to accept the recommendation of the AMT to write the stop order.

Data analysis

The PNA FISH results were compared with final culture identification for accuracy. All GPCC were identified according to the standard laboratory protocols. The defined daily doses (DDD) of vancomycin were obtained from the clinical pharmacy and calculated using standard WHO guidelines.20 The criteria for determining contaminated blood cultures during the chart review were based on the following: admission diagnosis, date and site of blood draw, signs of systemic infection when result reported, presence of a non-removable intravascular source, AMT intervention notes and the treating physician orders, which was the key determinant in vancomycin discontinuation. Patient stay and costs (including pharmacy and laboratory) were obtained from the UMMC’s central data repository. All monetary values are in US dollars. Statistical analysis was performed using the Cox hazard analysis, Wilcoxon Rank Sum (Mann–Whitney) test, Fisher’s exact test and χ² test where appropriate with SPSS software (version 13 for windows).
Results

In the review period, 139 blood cultures with GPCC were subjected to PNA FISH testing and 84 blood cultures to conventional laboratory reporting. Of the 139 cultures that underwent FISH testing, 28 patients had documented S. aureus, while 58 met exclusion criteria (20 patients in cancer centre, 18 in shock trauma, 13 with non-removable intravascular material and 7 duplicates), leaving 53 out of 111 evaluable patients with CoNS for review. Of the 84 patients in the control arm, 18 patients had S. aureus, while 32 had exclusion criteria (15 in cancer centre, 5 in shock trauma, 7 with non-removable intravascular material and 5 duplicates), leaving 34 out of 66 patients for evaluation. Of those patients with CoNS bacteremia who were excluded because of an intravascular foreign body, there were 8/111 (7.2%) patients in the PNA FISH group and 6/66 (9.1%) in the control group (P = 0.77, not significant). All these patients received an appropriate course of vancomycin therapy.

The identification accuracy in this period of the PNA FISH test for S. aureus was 100% (28 of 28). Of the 111 negative results, 106 were S. epidermidis, 3 were Micrococcus species and 2 S. hominis.

There were no statistically significant differences between the two groups in numbers, age, sex, location and case mix for evaluation after exclusion criteria were met (Table 1). The majority of evaluable patients with blood cultures positive for CoNS were on non-intensive care unit (non-ICU) services.

In the non-ICU setting there was a trend towards a decrease in the amount of vancomycin used in those tested with PNA FISH for evaluation. Of those patients with CoNS bacteraemia who had S. aureus, while 32 had exclusion criteria (15 in cancer centre, 5 in shock trauma, 7 with non-removable intravascular material and 5 duplicates), leaving 34 out of 66 patients for evaluation. Of those patients with CoNS bacteremia who were excluded because of an intravascular foreign body, there were 8/111 (7.2%) patients in the PNA FISH group and 6/66 (9.1%) in the control group (P = 0.77, not significant). All these patients received an appropriate course of vancomycin therapy.

The major impact of the PNA FISH assay was on hospital costs in the non-ICU setting where there was an overall saving of ~$4000 per patient for bed, laboratory (this includes microbiology, chemistry, haematology and radiology) and pharmacy costs (Table 3). This reflects the shorter length of stay found in the PNA FISH arm and demonstrates the benefit of the test.

Discussion

The main goal of introducing the PNA FISH assay to the laboratory was to accelerate the accurate detection of S. aureus in blood cultures. This analysis was performed to determine the impact on patient care as to whether UMMC would continue to utilize the PNA FISH test.

The clinical impact of the PNA FISH test was the ability of the hospital AMT to get early results. This allowed them to evaluate the patients and to ensure accurate antimicrobial therapy and (4.9 DDD) compared with controls (6.78 DDD) that was not statistically significant (Table 2). However, there was a greater ability for the AMT responsible for patients tested with PNA FISH to release only one dose of vancomycin to the services as compared with the control arm. There was a significant reduction in median length of stay from 6 days in the control group to 4 days in the PNA FISH group (P < 0.05, CI 0.95–1.87, RR 1.33). There was also a trend towards earlier discharge of patients after reporting GPCC negative for S. aureus in the PNA FISH arm as compared with the controls. The control group, even with a similar case mix, had a longer length of stay and more vancomycin usage.

In the ICU population, there was only a 5% reduction in vancomycin usage, from 10.4 to 9.9 DDD (not statistically significant), and there was no effect on median ICU length of stay (8 days for control versus 10 days for FISH) or on total admission costs ($21 800 for control versus $23 650 for FISH, not significant). There were two deaths in each group.

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Table 1. Demographics of the PNA FISH assay and control groups with coagulase-negative staphylococci bacteremia

<table>
<thead>
<tr>
<th></th>
<th>Control</th>
<th>PNA FISH</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total numbers of GPCC</td>
<td>84</td>
<td>119</td>
<td></td>
</tr>
<tr>
<td>S. aureus</td>
<td>18</td>
<td>28</td>
<td>0.50</td>
</tr>
<tr>
<td>Total number of evaluable patients</td>
<td>34</td>
<td>53</td>
<td></td>
</tr>
<tr>
<td>Age</td>
<td>55 (±15)</td>
<td>56 (±14)</td>
<td>0.80</td>
</tr>
<tr>
<td>Sex (male)</td>
<td>42%</td>
<td>46%</td>
<td>0.67</td>
</tr>
<tr>
<td>Location</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>medical floor</td>
<td>20 (59%)</td>
<td>24 (45%)</td>
<td>0.28</td>
</tr>
<tr>
<td>surgical floor</td>
<td>7 (20.6%)</td>
<td>17 (35%)</td>
<td>0.32</td>
</tr>
<tr>
<td>intensive care unit</td>
<td>7 (20.6%)</td>
<td>12 (20%)</td>
<td>0.57</td>
</tr>
<tr>
<td>Case mix</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>pneumonia</td>
<td>9 (26%)</td>
<td>14 (26%)</td>
<td>1</td>
</tr>
<tr>
<td>skin/soft tissue infection</td>
<td>8 (24%)</td>
<td>15 (28%)</td>
<td>0.80</td>
</tr>
<tr>
<td>meningitis</td>
<td>3 (9%)</td>
<td>3 (6%)</td>
<td>0.67</td>
</tr>
<tr>
<td>renal insufficiency</td>
<td>2 (6%)</td>
<td>7 (13%)</td>
<td>0.47</td>
</tr>
<tr>
<td>(CrCl &lt; 30 mL/min)†</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>otherb</td>
<td>12 (35%)</td>
<td>14 (27%)</td>
<td></td>
</tr>
<tr>
<td>haemodialysis catheter</td>
<td>2</td>
<td>3</td>
<td></td>
</tr>
<tr>
<td>mortality</td>
<td>2 (6%)</td>
<td>2 (4%)</td>
<td>0.64</td>
</tr>
</tbody>
</table>

CrCl, creatinine clearance; GPCC, Gram-positive cocci in clusters. †Creatinine clearance was calculated by pharmacy on admission and documented in chart.

Other diagnoses included cerebrovascular events, liver failure, cardiovascular disease and urinary tract infections.

Case mix: used International Classification of Diseases code (ICD-9) on admission.

Table 2. PNA FISH assay effect on length of stay and defined daily doses of vancomycin usage in patients not in intensive care units

<table>
<thead>
<tr>
<th></th>
<th>Control group</th>
<th>PNA FISH</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total DDD of vancomycin/patient</td>
<td>6.78</td>
<td>4.9</td>
<td>NS</td>
</tr>
<tr>
<td>DDD of vancomycin/patient after GPCC result</td>
<td>4.8</td>
<td>2.55</td>
<td>0.06</td>
</tr>
<tr>
<td>Patients receiving no doses of vancomycin</td>
<td>3/34 (9%)</td>
<td>9/53 (17%)</td>
<td>0.06, NS</td>
</tr>
<tr>
<td>Patients receiving 1 or less doses of vancomycin</td>
<td>5/34 (15%)</td>
<td>23/53 (43%)</td>
<td>&lt;0.005</td>
</tr>
<tr>
<td>Number of patients with LOS &lt; 3 days after GPCC result</td>
<td>6/34 (18%)</td>
<td>20/53 (38%)</td>
<td>0.06, NS</td>
</tr>
<tr>
<td>Median LOS (days)</td>
<td>6</td>
<td>4</td>
<td>&lt;0.05, CI 0.95–1.87</td>
</tr>
</tbody>
</table>

DDD, defined daily doses; NS, not significant; LOS, length of stay; GPCC, Gram-positive cocci in clusters.
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Table 3. Overall cost per patient of PNA FISH assay versus the control group in patients not in an intensive care unit

<table>
<thead>
<tr>
<th></th>
<th>PNA</th>
<th>FISH</th>
<th>Savings</th>
</tr>
</thead>
<tbody>
<tr>
<td>Average bed cost/patient</td>
<td>9002</td>
<td>6298</td>
<td>2704</td>
</tr>
<tr>
<td>Average pharmacy costs/patient</td>
<td>3371</td>
<td>2386</td>
<td>985</td>
</tr>
<tr>
<td>Average laboratory costs/patient</td>
<td>1248</td>
<td>932</td>
<td>316</td>
</tr>
<tr>
<td>Total costs/patient</td>
<td>13 621</td>
<td>9616</td>
<td>4005</td>
</tr>
</tbody>
</table>

*Laboratory costs include radiology, chemistry and hematology costs.

reduce unnecessary usage of vancomycin by preventing or limiting the treatment of CoNS contaminants in blood cultures by ruling out S. aureus.

This evaluation demonstrated a significant reduction in the median length of hospital stay and there was a trend towards reduced vancomycin usage in patients not in ICU settings. The reasons for the control group to have these prolonged trends are uncertain; however, there appear to be two factors: (i) once standard vancomycin therapy has been initiated it is difficult even for an active AMT to get the treating physician to write a stop order until the results of blood cultures are reported and (ii) the treating physician has to recognize that the final culture is a contaminant to discontinue therapy. Therefore, the advantage of PNA FISH appears to be that, with its rapid and accurate detection of S. aureus, the AMT was able to prevent the initiation of vancomycin therapy for CoNS blood cultures that are probably contaminants, with the subsequent likelihood of earlier discharge. We demonstrated that there were a significantly greater number of patients in the PNA FISH group who received one or less doses of vancomycin than the control group, reflecting the ability of the AMT to intervene before vancomycin is released. Subsequently, with GPCC in the blood, the primary physician using our treatment algorithm could wait for the PNA FISH result before requesting vancomycin unless the patient was clinically unstable or there was a non-removable intravascular device. If there is any uncertainty, a single dose of vancomycin could be released and the AMT could then be utilized to help interpret the results and advise on whether to continue on therapy. Vancomycin would be released for longer treatment if there was clinical instability, endocarditis or a non-removable intravascular foreign body. By appropriately preventing vancomycin release, we were able to impact its usage and achieve these outcomes.

Our study did not show any benefit in the ICU setting in either costs or length of stay. This was due to the low patient numbers being evaluated and their higher severity of illness, where unstable patients are going to be treated for CoNS bacteraemia. There was only a 5% reduction in vancomycin usage. Although no financial benefit from PNA FISH was seen in the ICU, there was strong support for the test for identifying S. aureus bloodstream infections earlier to help direct care. We recognize that this is a single centre review and that we could not reach statistical significance with vancomycin DDD. This was because the treating physicians in the institution had become comfortable using the PNA FISH result in their therapeutic decision-making. We could not proceed with our evaluation past 3 months as the laboratory initiated a second batch for the late afternoon because of physician requests, thereby eliminating our control group.

The use of our combined model of rapid resulting by the clinical laboratory in conjunction with the AMT has enabled the institution to develop and implement simple management algorithms for both CoNS and S. aureus bacteraemias. Previously, the infectious disease physician from the AMT would spend considerable time trying to convince physicians to discontinue vancomycin on presumed contaminated blood cultures based on variables such as time-to-positivity and the number of sets drawn or bottles positive while waiting for the species identification.21 Although these variables are well established, none of them is fully sensitive and specific and also some of these variables have been challenged as a diagnostic tool.21–23 However, physician acceptance of the PNA FISH results was rapid because of its specificity to give a clear result which they can use immediately.14 Subsequently, it has reduced the time the AMT spends on contaminated blood cultures, so they can focus on other antimicrobial issues. As with all programmes, re-education and reinforcement is required and this includes blood culture drawing technique.

We recognize that there are other tests for the rapid diagnosis of S. aureus such as tube coagulase testing (TCT) and API RAPIDDEC Staph (API) (BioMerieux, Durham, NC, USA). TCT although cheap is not as sensitive as the other two tests at 4 h, whereas API is equivalent to PNA FISH.1 We have not evaluated either of these tests in our practice and cannot comment on their likely impact on antimicrobial management. The PNA FISH assay does offer the advantage of viewing the morphology of the organisms while reading fluorescence. This provides an additional safeguard from potential technical errors (i.e. the morphology must match the result).

Lastly, implementing any new test into a microbiology laboratory often encounters resistance from hospitals trying to control budgets and laboratory personnel who are concerned about time consumption on extra procedures. However we have shown that decreasing hospital length of stay may lead to secondary benefits in decreased testing and increased bed usage. Our overall cost-savings for this 3 month period were $110 000; therefore the savings generated for the hospital are significantly greater than the outlay for the laboratory testing.

In conclusion, we have demonstrated how an integrative approach between accurate and rapid testing in the microbiology laboratory in conjunction with an active AMT can lead to reductions in treatment of contaminated blood cultures, earlier patient discharge and considerable cost-savings. The greatest measurable impact in the present investigation was in the non-ICU setting, where we can use PNA FISH to prevent the initiation of vancomycin for treatment of contaminated blood cultures and thus develop a cost-effective treatment algorithm. This is the first report showing the PNA FISH test in conjunction with an AMT reducing vancomycin usage, length of patient stay in hospital and hospital costs.

Transparency declarations

G. N. F. has received financial reimbursement for speaking programmes organized by AdvanDx. R. A. V. has received a speaking honorarium from AdvanDx, while D. P. L. and J. K. J. have received a travel grant from the same company.
References