Bacteriological profile and antibiotic susceptibility test of blood culture isolates in pediatric patients at a tertiary care teaching hospital

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Abstract
Background: Aim of the study was to know the bacterial profile and antibiotic resistance pattern of blood culture isolates from pediatric patients in a tertiary care teaching hospital.

Methods: Between March 2014–July 2015, blood culture samples from 1346 children (0-18 years) were monitored for the growth in the BACTEC/automated instrument. Samples suspicious of growth as indicated by the instrument were sub cultured and incubated at 37°C and identified by standard procedure. Antibiotic susceptibility testing was performed by Kirby-Bauer disk diffusion method.

Results: Out of 1346 blood culture samples processed, 159 (11.8%) were positive. Among positive isolates Salmonella species were 66.6% and non-Salmonella species were 33.3%. Predominant non-Salmonella species isolated were NFGNB (10.1%), 5% each of CoNS and Enterococcus spp. Among Salmonella spp isolated majority (85%) were S. typhi and (15%) were S. paratyphi A. isolated. The predominant isolates from ICU were NFGNB (30%), CoNS (20%) and Pseudomonas species (13.3%). Both S. typhi and S. paratyphi A revealed 18% and 19% resistance to ciprofloxacin; 8% and 19% resistance to ampicillin respectively. No resistance for chloramphenicol and ceftriaxone were noted in S. paratyphi A. Resistance of 2.2% each to chloramphenicol and cefotaxime, 1.8% to co-trimoxazole was noted, however no resistance was noted to ceftriaxone in S. typhi.

Conclusion: A constant monitoring of blood cultures in pediatric age group is critical to understand bacterial profile and their antibiotic susceptibility pattern in different age groups to provide better patient care.

Keywords: Blood stream infections; Blood culture; Salmonella; Pediatrics.

Introduction
Identification of various organisms in a patient’s blood is of immense diagnostic and prognostic importance. Blood cultures are essential in the diagnosis and treatment of the etiologic agents or sepsis. It has been observed that inadequate therapy is the common reason for antimicrobial resistance. Hence, information on most likely causative organisms and their resistance patterns can increase the likelihood of selecting an effective antimicrobial drug for empirical treatment. Considering the current worldwide changes, information about the occurrence of pathogens and antibiotic resistance pattern are now seen as decisive for optimizing treatment.

Materials and Methods
A prospective study was conducted from March 2014 to July 2015. Total of 1346 blood samples were processed from pediatric age group (0-18 years) at ESIC MC PGIMSR Teaching Hospital, Rajajinagar, Bengaluru.

Inclusion Criteria: All blood culture samples in the age group of 0-18 years irrespective of antibiotic treatment status.

Exclusion Criteria: Blood cultures showing mixed growth (>1 types)

Blood culture submitted to diagnostic microbiology in the age group of 0-18 years was monitored during the study period for the growth in the BACTEC/automated instrument BacT/ALERT3 (BioMérieux, Inc. Durham)
November. *S. typhi* (55.3%), *S. paratyphi* A (10%) and NFGNB (5.3%) were predominant isolates in age group 1-18 years. *Enterococcus* spp (3.8%), NFGNB 5(3.1%), CoNS and *Pseudomonas* spp 2.5% each, were predominant isolates in the age group 0-28 days. (Table 2)

Nineteen percent of isolates were from ICU and 81% were from Non ICU. The predominant organisms isolated from ICU were NFGNB 9(30%), CoNS 6(20%), *Pseudomonas* species 4(13.3%) and *Enterococcus* species 3(10%) (Fig. 1).

Resistance pattern of *Salmonella*—Both *S. typhi* and *S. paratyphi* A revealed 90% and 81% resistance to nalidixic acid; 18% and 19% resistance to ciprofloxacin; 8% and 19% resistance to ampicillin respectively. No resistance for chloramphenicol and ceftriaxone was noted to *S. paratyphi* A. Resistance of 2.2% each to chloramphenicol and cefotaxime, 5.6% to co-trimoxazole was noted, however no resistance was noted to ceftriaxone in *S. typhi* (Fig. 2 & 3).

Predominant Gram Positive Cocci isolated were *Enterococcus* spp and CoNS (5% each). *Enterococcus* showed highest resistance of 75% each to penicillin and ciprofloxacin. CoNS showed highest resistance to penicillin (87.5%) and erythromycin (62.5%). (Fig. 4) Predominant Gram Negative Bacteria other than *Salmonella* was Non fermenters 16 (10%). Highest resistance among these isolates was with ceftazidime and cefotaxime 50% each. Imipenem was found to be most effective with least resistance of 6.25%. (Fig. 5) Since the proportion of Gram negative bacteria other than *Salmonella* were significantly less, detecting their common resistance mechanisms would be improper.

Salmonella species were 106(66.66%) and contamination rate was 3%. Non *Salmonella* species were 53(33.33%). Among the positive isolates *S. typhi* and *S. paratyphi* A were 57% & 10% respectively. Among Non *Salmonella* spp isolated, 16(4%) were NFGNB, 8(4%) each of CoNS and *Enterococcus* spp, 6(3%) were of *Pseudomonas* spp, 4(2%) *Staphylococcus aureus*, 3(1.5%) *Escherichia coli*, 2(1%) each of Enterobacter spp, *Citrobacter* spp, *Streptococcus viridans* were isolated.

Table 1: Distribution of blood culture positive isolates

<table>
<thead>
<tr>
<th>Organisms</th>
<th>Frequency</th>
<th>Percent</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>S. typhi</em></td>
<td>90</td>
<td>56.6</td>
</tr>
<tr>
<td><em>S. Paratyphi</em> A</td>
<td>16</td>
<td>10.1</td>
</tr>
<tr>
<td>NFGNB</td>
<td>16</td>
<td>10.1</td>
</tr>
<tr>
<td>CoNS</td>
<td>8</td>
<td>5</td>
</tr>
<tr>
<td><em>Enterococcus</em> spp</td>
<td>8</td>
<td>5</td>
</tr>
<tr>
<td><em>Pseudomonas</em></td>
<td>6</td>
<td>3.8</td>
</tr>
<tr>
<td><em>Staph.aureus</em></td>
<td>4</td>
<td>2.5</td>
</tr>
<tr>
<td><em>E.coli</em></td>
<td>3</td>
<td>1.9</td>
</tr>
<tr>
<td><em>Citrobacter</em></td>
<td>2</td>
<td>1.3</td>
</tr>
<tr>
<td><em>Enterobacter</em></td>
<td>2</td>
<td>1.3</td>
</tr>
<tr>
<td><em>Strep.viridans</em></td>
<td>2</td>
<td>1.3</td>
</tr>
<tr>
<td><em>Candida alb</em></td>
<td>1</td>
<td>0.6</td>
</tr>
<tr>
<td><em>Gr A strep</em></td>
<td>1</td>
<td>0.6</td>
</tr>
<tr>
<td>Total</td>
<td>159</td>
<td>100</td>
</tr>
</tbody>
</table>

Table 2: Distribution of blood culture positive isolates in different age group

<table>
<thead>
<tr>
<th>Organisms (n)</th>
<th>0-28 Days</th>
<th></th>
<th>1M-1Y</th>
<th></th>
<th>1-18Y</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>No</td>
<td>%</td>
<td>No</td>
<td>%</td>
<td>No</td>
<td>%</td>
<td></td>
</tr>
<tr>
<td><em>S. typhi</em> (90)</td>
<td>1</td>
<td>0.6</td>
<td>1</td>
<td>0.6</td>
<td>88</td>
<td>55.3</td>
</tr>
<tr>
<td><em>S. Paratyphi</em> A</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>16</td>
<td>10.1</td>
</tr>
<tr>
<td>NFGNB (16)</td>
<td>5</td>
<td>3.1</td>
<td>2</td>
<td>1.3</td>
<td>9</td>
<td>5.7</td>
</tr>
<tr>
<td>CoNS (8)</td>
<td>4</td>
<td>2.5</td>
<td>1</td>
<td>0.6</td>
<td>3</td>
<td>1.9</td>
</tr>
<tr>
<td><em>Enterococcus</em> spp (8)</td>
<td>6</td>
<td>3.8</td>
<td>0</td>
<td>0</td>
<td>2</td>
<td>1.3</td>
</tr>
<tr>
<td><em>Pseudomonas</em> spp (6)</td>
<td>4</td>
<td>2.5</td>
<td>0</td>
<td>0</td>
<td>2</td>
<td>1.3</td>
</tr>
<tr>
<td><em>Staphylococcus aureus</em> (4)</td>
<td>1</td>
<td>0.6</td>
<td>0</td>
<td>0</td>
<td>3</td>
<td>1.9</td>
</tr>
<tr>
<td><em>Escherichia coli</em> (3)</td>
<td>1</td>
<td>0.6</td>
<td>1</td>
<td>0.6</td>
<td>1</td>
<td>0.6</td>
</tr>
<tr>
<td><em>Citrobacter</em> spp (2)</td>
<td>0</td>
<td>0</td>
<td>1</td>
<td>0.6</td>
<td>1</td>
<td>0.6</td>
</tr>
<tr>
<td><em>Enterobacter</em> spp (2)</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>2</td>
<td>1.3</td>
</tr>
<tr>
<td><em>Strep. Viridans</em> (2)</td>
<td>1</td>
<td>0.6</td>
<td>0</td>
<td>0</td>
<td>1</td>
<td>0.6</td>
</tr>
<tr>
<td><em>Candida albicans</em> (1)</td>
<td>1</td>
<td>0.6</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>Group A streptococcus (1)</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>1</td>
<td>0.6</td>
</tr>
<tr>
<td>Total</td>
<td>24</td>
<td>15.1</td>
<td>6</td>
<td>3.8</td>
<td>129</td>
<td>81.1</td>
</tr>
</tbody>
</table>

*S. typhi* (55.3%), *S. paratyphi* A (10%) and NFGNB (5.3%), was the predominant isolates in age group 1-18 years.

*Enterococcus* spp (3.8%), NFGNB 5(3.1%), CoNS and *Pseudomonas* spp 2.5% each, were the predominant isolates in the age group 0-28 days. Highest positivity rate was from age group 1-18 years (81.1%). Least positivity...
rate was from age group 1month – 1year (3.8%). Blood culture positivity was 9.5% (129/1346) in 1-18 years, 1.7% (24/1346) in 0-28 days and 0.5% (6/1346) in 1month – 1year age group.

![Distribution of blood culture isolates (%) from ICU patients](image1)

**Fig. 1:** Distribution of blood culture isolates (%) from ICU patients

Out of 30 isolates from ICU, NFGNB 9(30%) was the predominant organism isolated, followed by CoNS 6(20%), *Pseudomonas* species 4(13.3%) and *Enterococcus* species 3(10%).

![Distribution of antibiotic resistance (%) pattern of *S.typhi*](image2)

**Fig. 2:** Distribution of antibiotic resistance (%) pattern of *S.typhi*

Both *S. typhi* and *S. paratyphi A* revealed 90% and 81% resistance to nalidixic acid; 18% and 19% resistance to ciprofloxacin respectively.8% and 19% resistance to ampicillin. No resistance for chloramphenicol, cefotaxime and
ceftriaxone were noted to \textit{S. paratyphi} A. Resistance of 2.2\% each to cefotaxime, chloramphenicol, 5.6\% to cotrimoxazole noted. However no resistance was noted to ceftriaxone in \textit{S. typhi} (Fig. 2 & 3)

\begin{figure}[h]
\centering
\includegraphics[width=\textwidth]{fig4.png}
\caption{Distribution of antibiotic resistance pattern of CoNS and \textit{Enterococcus}}
\end{figure}

Predominant GPC were \textit{Enterococcus spp} and CoNS, 8(5\%) each. \textit{Enterococcus} showed highest resistance to penicillin and ciprofloxacin, 75\% each. CoNS showed highest resistance to penicillin (87.5\%) and erythromycin(62.5\%).

\begin{figure}[h]
\centering
\includegraphics[width=\textwidth]{fig5.png}
\caption{Distribution of antibiotic resistance (%) pattern of NFGNB}
\end{figure}

Predominant GNB other than Salmonella was Non fermenters 16 (10\%), highest resistance was seen with ceftazidime and cefotaxime 50\% each. Imipenem was found to be most effective with least resistance of 6.25\%.

\section*{Discussion}

The varying microbiological pattern of bacteremia in children warrants the need for an ongoing review of the causative organisms and their antimicrobial susceptibility pattern.\textsuperscript{(7)} In this present study it was observed that there was higher isolation of organisms in males (62\%) than females (38\%). This is comparable to the other studies by Begum S et al\textsuperscript{(9)} and Shrestha NJ et al.\textsuperscript{(9)} The reason for male preponderance is unknown, but this could be due to sex-dependent factors.\textsuperscript{(56)} The synthesis of gamma globulins is probably regulated by X- linked immunoregulatory genes and as males are having one X chromosome, they are more prone for neonatal septicemia than females.\textsuperscript{(10)}

The bacterial isolation rate in this study (11.8\%), was comparable with the other studies like Ethiopia (8.8\%) (Zenebe et al. 2011),\textsuperscript{(31)} India 9.94\% (Manjula et al. 2005)\textsuperscript{(11)} and Nepal 10\% (Usha and Pushpa 2007).\textsuperscript{(12)} It was, however; lower than reports from Gonder, Ethiopia, 24.2\% (Ali and Kebede 2008),\textsuperscript{(13)} Nepal 23.1\% (Amaty et al. 2007),\textsuperscript{(14)} Zimbabwe 37.1\% (Obi and Mazarura 1996),\textsuperscript{(15)} Gambia 34\% (Philip et al. 2007)\textsuperscript{(16)} and India 40\% (Neuma and Chitnis 1996).\textsuperscript{(17)}

\textit{S. typhi}, \textit{S. paratyphi} A, NFGNB, CoNS, \textit{Enterococcus spp}, \textit{Pseudomonas spp}, \textit{S. aureus}, and \textit{Escherichia coli} were the predominant pathogens causing bacteraemia in this study. A similar observations have been seen in cases of bacteraemia in different countries, though, the proportion and prevalence of the bacterial agents varied. (Phetsouvanh
R et al,(16) Vijaya Devi RIMS hospital,(19) Meremo et al. 2012,(20) S. typhi (56.6%) was the predominant isolate in the present study, contrary to the other studies Wasishun et al 2015,(21) Okon KO Negeria 2014,(22) Prakash KP et al Oman 2011(23) where S. aureus was the most common isolate, this prevalence was comparable to the study Phetsouvanh R et al,(18) Vijaya Devi RIMS hospital,(19) Meremo et al Tanzania 2012.(20)

In the present study, 159/1346 paediatric blood cultures yielded bacterial growth, among them prevalence of Salmonella was found to be 106/159(66.6%). Culture positivity varies across the country from 7 – 12.11%.(24) Study showed typhoid prevalence in and around Bangalore to be 78 per 1000 paediatric population (106 Salmonella/1346 blood culture) febrile episodes with blood cultures taken into consideration. In India typhoid prevalence was reported to be 28.1 per 1000 febrile episodes in 2004. In the study Salmonella typhi was the predominant isolate 90(85%) followed by S. paratyphi A 16(15%). Mohanty et al in their study reported Salmonella typhi (75.7%) as the predominant serotype followed by S. paratyphi A (23.8%).(25)

Typhoid fever is among the major widespread diseases affecting both young children and young adults in their productive years. In our study Salmonella isolates were cultured from paediatric age-groups, the median age being 9 years (range 0-18 years). Forty three percent of isolates were obtained from patients in 6-12 years of age. The gender ratio in the study showed male preponderance (male: female = 1.7:1), but this difference was not statistically significant (P=0.521).

Enteric fever cases occurred in all months throughout the year, the maximum number occurred during months (Apr-June) followed by (Nov-Dec). Seasonal distribution of S. paratyphi A isolates was similar to the seasonal distribution of S. typhi. During the summer and monsoon months the water supply and sanitation systems are under a great strain, which could account for the higher incidence in these months.(24,25)

In the present study S. typhi isolates showed significant increase in sensitivity to chloramphenicol (97.7%), ampicillin (95.5%), and cotrimoxazole (94.4%). All S. paratyphi A isolates were 81.3% sensitive to ampicillin, 100% sensitive to chloramphenicol and cotrimoxazole drugs. Similar findings were reported by Nath et al, Tankhiwale et al, Bhatia et al and Sheorey et al from Varanasi, Nagpur, Pune and Mumbai respectively.(26,27,28,29) The results of present study compare favourably with those reported from other parts of India and point towards re-emergence of sensitivity to these classical drugs.(30)

In the present study S. typhi and S. Paratyphi A were 100% sensitive to ceftriaxone, which was similar to the finding of Nath et al from Varanasi and Gautum et al from Haryana.(11,12) The strain was also resistant to ampicillin, ciprofloxacin & nalidixic acid.

The common Gram positive bacteria isolated were CoNS (8, 4%), Enterococcus spp (8, 4%), S. aureus (4, 2%), study in Patan hospital should CoNS as the most common Gram positive bacteria (31%), Adugna negussie et al 23.2% 2015, Al-Rawaqz et al 31% 2012, Prakash KP et al.(2) Wasishun et al 21 2015 10.3%. In all these studies S. aureus was the predominant Gram positive bacteria isolated.

Conclusion

Overall blood culture positivity in paediatric age group was found to be 11.8%. Blood culture positivity was highest in 1-18 years of age with S. typhi and S. paratyphi A as predominant organisms. Non fermenters predominated in the younger age group while Salmonella spp in the older age group. Peak isolation of Salmonella spp were with the onset of monsoon. Chloramphenicol and ceftriaxone were found to be most effective against Salmonella. Isolation of Gram positive cocci and Gram negative bacteria other than Salmonella spp were remarkably less in paediatric age group. A constant monitoring of blood cultures in paediatric age group is critical to understand bacterial profile and their antibiotic susceptibility pattern in different age groups to provide better patient care.

Periodical evaluation of the results will enable precautions including coplying with antisepsis rules during obtaining samples and obtaining sufficient number of samples at the appropriate time by predicting contamination rates. Administration of treatment in accordance with culture antibiogram results instead of empirical treatment will decrease the mortality rates and antibiotic resistance and will enlighten the establishment of antibiotic usage policies.

References


