

Human Journals **Research Article** September 2017 Vol.:7, Issue:3 © All rights are reserved by T. Jaya Chandra et al.

A Study on Identification of Bloodstream Infections among the Patients Admitted In the Hospital



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Submission: 27 August 2017

Accepted:

5 September 2017

Published: 30 September 2017





www.ijsrm.humanjournals.com

Keywords: Bloodstream infections, Gram-positive cocci, gram-negative bacilli,

ABSTRACT

Introduction: Bloodstream infections (BSIs) is a common nosocomial infection caused by gram-positive cocci (GPC), gram-negative bacilli (GNB) and fungi. With this, a study was conducted to find the organisms causing BSIs among the patients admitted to the hospital. Materials and methods: Study was conducted in the Department of Microbiology, GSL Medical College, Rajahmundry from May 2017 to July 2017. After thorough skin disinfection, 10 ml blood was collected by phlebotomy. Samples were inoculated in brain heart infusion biphasic media, incubated at 37°C and checked for turbidity after 1, 2, 3 and 7 days. If any growth observed, that is identified by grams staining and biochemical reactions. Results: The culture positivity was 24% and GPC were isolated more compared to GNB, 59%, 41% respectively. Staphylococcus aureus was the leading (41%) causative agent, followed by Escherichia coli (20%), Coagulase negative staphylococcus (13%), Klebsiella species (13%). Conclusion: Drug resistance was observed to the fine line antimicrobials such Ampicillin, Amoxycillin, as Ciprofloxacin, Hence Cotrimoxazole. Gentamicin. antimicrobials should be used by following proper antibiotic susceptibility test report.

INTRODUCTION:

Bacteremia, a condition where bacteria are present in the blood, cause bloodstream infections (BSIs). BSI is a common nosocomial infection which may usually occur due to the procedures in the hospitalized patient and this may lead to high mortality and morbidity^{1, 2, 3}. In addition, the hospital-acquired BSI may also increase financial burden to the patient. Factors such as the type of antibiotics used, aseptic techniques followed for the invasive procedures, a number of days stayed in the health care setup and so on also influence the BSI as well as causative agents. But immunity of the patient is an important criterion. Because, if the patient is immune competent, sepsis cannot be developed by the floral members such as *Escherichia coli*, Coagulase-negative staphylococcus (CoNS).

Various microorganisms such as both gram-positive cocci (GPC) as well as gram-negative bacilli (GNB) cause BSI. Role of fungi as a causative agent of BSIs is also reported. In this setup increased isolation of CoNS in BSI is notified. In continuation, bacterial resistance to antimicrobial agents is a serious problem in the treatment of BSIs. With these observations, a study was conducted to find the bacterial pathogens associated with BSIs and their susceptibility pattern to various antimicrobials among the patients admitted to the hospital.

MATERIALS AND METHODS:

The study was conducted in the Department of Microbiology, GSL Medical College, Rajamundry for three months, May to July 2017. The study protocol was approved by the institutional ethics committee. An informed verbal consent was taken from all the volunteers. Three blood samples were collected from the participants, at three different time intervals in three different areas. Before phlebotomy, thorough skin disinfection was done with 70% alcohol. A minimum of 10 ml blood was collected. To neutralize the bactericidal effect, blood to BHI broth was used at 1 in 5 ratios.

Immediately after collection, blood samples were inoculated in brain heart infusion (BHI) biphasic media, without opening the cap as well as changing the needle. After inoculation, blood culture bottles were incubated at 37 °C and checked for turbidity after 1, 2, 3 and 7 days. For days 1, 2 and 3 only the bottles showed signs of positive growth were cultured on agar plates. On the day 7, all bottles were sub-cultured on Blood, MacConkey and Nutrient agar plates before being discarded as negative.

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Initially, growth was classified by grams staining (GS). The battery of biochemical tests was used to identify the pathogens^{4, 5, 6}. Coagulase test, Catalase test, **Microdase** test, heat test, Bile esculin agar were used for identification of GPC. To confirm GNB, tests such as Indole, Methyl Red, Voges Proskauer, Citrate utilization, Urease production and growth on Triple Sugar Iron agar were used. Antibiotic sensitivity test (AST) was done on Muller-Hinton agar (MHA) by Kirby bauer disk diffusion method^{7, 8, 9}, *Escherichia coli* ATCC 25922 was used as the control. If fungi were suspected, growth was identified by GS, lactophenol cotton blue mount and slide culture techniques. Chi-square test was used to find the statistical significance, P > 0.05 was considered statistically significant.

RESULTS:

During the study period, blood samples were collected from 223 participants, the male female ration was 1.3, mean age was 43.1 years. Fifty-four (24%) patient's blood samples were culture positive. Organism wise, *Staphylococcus aureus* was isolated maximum (41%), followed by *Escherichia coli* (20%), CoNS (13%), Klebsiella species (13%) (Table 1).

DISCUSSION:

Blood culture is the sole technique for the diagnosis BSIs causing bacteria. Of the 223 study participants, 126 were male and 97 were female participants. The culture positivity was 31 (25%) and 23 (23%) in male and female participants, respectively; statistically, there was no significant difference in culture positive results between the genders (P>0.05). In the available literature also, the difference in culture positivity among gender was not discussed.

In this study, in spite of sepsis, culture positivity was just 24% (54). This is a tertiary healthcare setup, most of the patients had treated with antibiotics in other centers previously. Ideally, for microbial culture and sensitivity test, the specimen should be collected before initiation of treatment. In addition to this, anaerobes also cause BSIs. But, in this study, anaerobic culture techniques were not included. These may be the reasons for low culture positivity. But, as per the available literature, the range of culture positivity among BSIs was 9.2% to 33.9%⁷.

In this study, GPC was isolated maximum followed by GNB, 59%, 41% respectively. Our findings differ Sahoo et al.⁷ study, where the investigators reported that 30.8%, 69.2% of

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GPC, GNB respectively. But, as per Sumita R et al.⁸ reports, GPC (53.6%) were isolated more compared to GNB (46.4%).

Among GPC, *Staphylococcus aureus* (41%; 22) was isolated maximum, followed by CoNS (13%; 7), Enterococcus (5.5%; 3). In a study by Amit J et al.⁹, *Staphylococcus aureus* was reported as the commonest causative agent of BSIs. The colonization of *Staphylococcus aureus* varied from 8 to 71.2%¹⁰. In a study by Chand W et al., CoNS was reported as the most (20.3%) common blood culture isolate¹¹. CoNS is also reported as the commonest BSI causing bacteria among the ICU patients¹². Chand W et al.¹¹ observed that Enterococcus is an important pathogen (8.4%) of BSIs. This is a floral member of female genital a well as the gastrointestinal tract. In the current report, this may be the reason for getting more number of isolates from female participants.

In GNB, *Escherichia coli* was isolated maximum (20%; 11) followed by Klebsiella species (13%; 7) and *Pseudomonas aeruginosa* (7.4%; 4). Sahoo et al.⁷ also reported that *Escherichia coli* (34.6%) was isolated maximum followed by Klebsiella species (26.9%). Another south Indian study⁸ also reported similar results, 32.6%, 25% isolation of *Escherichia coli*, Klebsiella species; respectively. Unlike the available reports^{7, 8}, in this study fungus was not isolated.

When antibiotic susceptibility pattern was considered, drug resistance was more in GNB compared to GPC. GNB were resistant to common first-line antibiotics such as Ampicillin, Amoxycillin Clavulanic acid, Ciprofloxacin, Cotrimoxazole Gentamicin. But these were sensitive to higher antimicrobials such as Cefaperzone sulbactam, Ceftriaxone, Piperacillin tazobactam. Most of these antibiotics are injectables, costly and also cause side effects on the patients. This may be due to unnecessary usage of antibiotics elsewhere. Hence antimicrobials should be used by following proper AST report.

Acknowledgements: The authors acknowledge the authorities of Andhra University for permitting K. Nayomi to undertake this work as part of their M.Sc. dissertations

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Table 1: Gender wise number of bacteria isolates; n (%)			
Organism	Gender		Total
	Male	Female	Total
Staphylococcus aureus	12 (22)	10 (18.5)	22 (41)
CoNS	2 (3.7)	5 (9.3)	7 (13)
Enterococcus	1 (1.8)	2 (3.7)	3 (5.5)
Esch.coli	8 (14.8)	3 (5.5)	11 (20)
Klebsiella	6 (11)	1 (1.8)	7 (13)
Pseudomonas aeruginosa	0	4 (7.4)	4 (7.4)
Total	29 (53.7)	25 (46.3)	54 (100)