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BAC' OF B OF A	TERIOLOGICAL PROFILE AND ANTIBIOGRAM ACTERIAL ISOLATES FROM BLOOD CULTURES TERTIARY CARE TEACHING HOSPITAL : A ROSPECTIVE TRIAL	KEY WORDS: bloodstream infections, antibiotic sensitivity , bacterial profile.					
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Introduction: Blood stream infections, ranging from self limiting bacteremia to life threatening septicemia, remain one of the most important cause of morbidity and mortality worldwide. Microorganisms present in circulating blood whether continuously, intermittently or transiently are a threat to every organ in the body. Blood culture is a vital tool for the detection of BSI and remains the gold standard for bacteremia detection. Aims and Objectives: To Study the bacterial profile of blood stream infections and the drug sensitivity pattern of the isolate bacteria in patients admitted in various departments of AMCH Shahbad. Material & Methods: A study was carried out from June 2018 to November 2018; blood samples were collected and processed by automated blood culture system BACTEC BD Fx40.Positive samples were further plated for isolation and identification of causative organism and isolate further tested by Kirby bauer disc antibiotic susceptibility test to know the sensitivity pattern of the isolate. **Results:** During the six months study period 150 non repetitive blood samples collected from patients attending AMCH Shahbad. Culture positivity was seen in 40(26.67%) samples and 110(73.3%) samples were sterile. Gram positive cocci was the most common organism isolated with 23(57.5%) .Staphylococcus aureus being the most common organism among gram positive cocci and Klebsiella pneumoniae commonest among Gram negative bacilli 12(30%). There were 2(5%) samples in which Candida albicans was isolated. Antibiotic resistance pattern among Staphylococcus aureus were MRSA 60% and MSSA 40%. Conclusion: Local microbiological analysis suggesting possible causative agents and the best choice of antibiotics are important for the local physicians when treatment has to be initiated before the result of the blood culture is known.

INTRODUCTION

ABSTRACT

Bacteremia signifies the presence of bacteria in the blood stream . Bacteremia may be transient, continuous or intermittent. Microorganisms present in the circulating blood, whether continuously, intermittently, or transiently, are a threat to every organ in the body. They can have serious consequences like shock, multiple organ failure, disseminated intravascular coagulation, etc. Thus, the blood stream infections constitute one of the most serious situations and, as a result, timely detection and identification of blood stream pathogen is important⁽¹⁾.

Bloodstream infections (BSIs) occur due to the failure of our immune system to restrict infection at a focal site; leading to widespread disease. It is a major cause of morbidity and mortality. The occurrence of these infections, their epidemiology, and the invading pathogens have altered in parallel with the evolution in medical care, particularly with emergence of increasingly ill and immunocompromised population of hospitalized patients who are often heavily reliant on medical support and indwelling devices. The responsible bacteria are usually human pathogens. In contrast, many cases of severe sepsis that occur currently are associated with bacterial or fungal microbes that are members of the patient's own microbial flora.^(#).

Early diagnosis plays a crucial role in managing BSI, and hence, prompt detection of such infections is a critical function of clinical microbiology laboratories.Blood culture is a vital tool for the detection of BSI and remains the gold standard for bacteremia detection^{(3).} The successful recovery of microorganism from blood by possible types of bacteremia depends upon specimen collection methods, blood volumes, the number and timing of blood cultures, interpretation of results and the type of patient's population being served by the laboratory .There is a wide variation in the incidence and clinical characteristics of invasive infections caused by different species of bacteria. Identifying the causative agents and characterizing the clinical significance in a particular age group is essential for the prevention and treatment of these infections⁽¹⁾

Empiric antimicrobial therapy is based on knowledge of the microbial profile and their antimicrobial sensitivity patterns, clinical and epidemiological data. Irrational use of drugs has led to an increase of multidrug-resistant bugs and thus worsened the condition. Prevalence and susceptibility patterns of microorganism vary according to geography and even within the same hospital with time[®] It is therefore necessary to periodically document research results obtained from analysis of blood culture, to assist clinicians have the needed idea to commence treatment for desperately sick patients, awaiting blood culture results[®].

MATERIAL & METHODS

A study was carried out from June 2018 to November 2018; blood samples were collected from patients suspected of having bloodstream infection attending and admitted in

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Adesh Medical College & Hospital Shahbad. Details like hospital identity number, age, gender of the patients, and type and place of collection of specimen were recorded on a formatted proforma. A total of 150 suspected patients attending the various intensive care units (ICUs), out patient departments (OPDs), and indoor wards of Internal Medicine, Pediatrics, Obstetrics and Gynecology, Orthopedics, and General Surgery were included in the study.

Patients of all age groups with fever (both high and low grade) due to infective causes were included. Written informed consent was taken from all who fulfilled the criteria. Patients having both leukocytosis and leucopenia were included in the study.

Blood was collected with all aseptic precautions from the bedside of the patients suspected of having bloodstream infection using a sterile syringe. Approximately 5–10 mL of blood was collected from adult patients, while 1–5 mL blood was collected from pediatric patients, and 1–2 mL from neonates for blood culture. The sample taken was immediately processed for blood culture by BACTEC BD Fx 40 blood culture system . In the BACTEC BD Fx40 Blood Culture System, when growth of bacteria occurred in sufficient amount, the system automatically generated a signal on the front panel. If no growth of bacteria occurred within 5 days of blood culture, then the blood culture was reported as sterile on culture for pyogenic aerobic organisms⁽⁴⁾.

Identification of the bacterial isolates

Subculture was made on MacConkey Agar and Blood Agar from the BACTEC blood culture bottles, which generated a beep in the Automated BACTEC BD Fx40blood culture system indicating growth of the organism. The organisms were identified as per standard protocol.

Gram-negative bacilli. The colony character on culture media was observed, and Gram staining, motility, and biochemical tests – indole, methyl red, Voges–Proskauer, citrate utilization, urease test, phenyl pyruvic acid test, triple sugar iron agar, oxidase, amino acids decarboxylase test, and sugar fermentation reaction – were conducted⁽⁴⁾. Gram-positive cocci. On the basis of colony character, Gram stain, catalase test, and coagulase test.

Antimicrobial susceptibility testing

This was performed by Kirby Bauer disk diffusion method as per the Clinical and Laboratory Standards Institute guidelines(2017). The antibiotics disks (Hi-media, Mumbai) were used.

The following are quality control strains for antimicrobial sensitivity testing:

- (1) Pseudomonas aeruginosa (ATCC27853),
- (2) Escherichia coli (ATCC25922),
- (3) Staphylococcus aureus (ATCC25923).

RESULTS

During the six months study period between June 2018 to November 2018,150 non repetitive blood samples collected from patients attending AMCH Shahbad were studied. Details like Hospital identity No., Lab No., Age, Sex, were recorded in the proforma.

Culture positivity was seen in 40(26.67%) samples and 110(73.3%) samples were sterile as detected with BACTEC BD Fx40 blood culture system. The maximum number of positive blood culture was from age group of 18-60 years and minimum <1 year.

Of the 40 patients 25 were males and 15 females . Gram positive cocci was the most common organism isolated 23(57.5%) with Staphylococcus aureus being the most

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common organism among gram positive cocci and Klebsiella pneumoniae commonest among Gram negative bacilli 12(30%). There were 2(5%) samples in which Candida albicans was isolated while 3(7.5%) samples showed growth of contaminants.

1. Agewise Distribution Of Culture Positive Patients.

	0-1	l month	l year –	5-18	18-	>60	Total
	month	– 1 year	5 year	years	60years	year	
Male	2	-	2	2	13	6	25
Female	3	-	2	1	6	3	15

2.Distribution Of Micro Organism Isolated From Culture.

	Organism Isolated	Number (%) Total 40				
1.	Staphylococcus aureus	20(50%)				
2.	CONS	2(5%)				
3.	Enterococcus fecalis	1(2.5%)				
4.	Klebsiella pneumoniae	4(10%)				
5.	Escherichia coli	2(5%)				
6.	Proteus vulgaris	2(5%)				
7.	Pseudomonas aeruginosa	1(2.5%)				
8.	Acinetobacter baumanii	1(2.5%)				
9.	Salmonella Typhi	2(5%)				
10.	Candida albicans	2(5%)				

3.Antibiotic Sensitivity Pattern of Gram positive Isolates

S.No.		AMC	CD	CIP	CX	TE		TEI	VA	LΖ
1.	Staphyloc	0%	40	20	40	40	70	95	100	100
	occus		%	%	%	%	%	%	%	%
	aureus(n-									
	20)									
2.	Enterococ	NT	NT	100	NT	100	0%	100	100	100
	cus			%		%		%	%	%
	faecalis									
	(n-1)									

AMC-Amoxyclav, CD-Clindamicin, CIP- Ciprofloxacin, CX-Cefoxitin, TE- Tetracyclin, GEN- Gentamicin, TEI-Teicoplanin, VA-Vancomycin, LZ-Linezolid

4. Antibiotic Sensitivity Pattern of Gram Negative Bacilli

	Antibiotic	Enterobacteria	Salmonella	Non-fermenters
		cae other than	Typhi (n-2)	(pseudomonas &
		S. Typhi (n-8)		Acinetobacter)
				(n-2)
1.	AMP.	0%	0%	0%
2.	AMC	2.5%	0%	NT
3.	LE	0%	0%	0%
4.	GEN	62.5%	50%	0%
5.	CZ	0%	NT	NT
6.	CXM	0%	100%	50%
7.	CTX	0%	100%	50%
8.	CPM	50%	100%	NT
9.	CAZ	50%	100%	50%
10.	CAC	62.5%	NT	50%
11.	PIT	62.5%	NT	50%
12.	AT	62.5%	100%	50%
13.	IMP	50%	100%	50%

AMP- Ampicillin, AMC-Amoxyclav, LE- Levofloxacin,GEN-Gentamicin, CZ – Cefazolin, CXM – Cefuroxime, CTX-Ceftriaxone, CPM- Cefipime, CAZ-Ceftazidime, CAC- Ceftaclav,PIT-Piperacillin Tazobactum, AT- Aztreonam, IMP-Imipenem,NT-Nottested

DISCUSSION

Bloodstream infections is a challenging problem and

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sometimes, it may be life threatening ;therefore,timely detection , identification and anti microbial susceptibility testing of blood borne pathogens are one of the important functions of diagnostic laboratory.

In this study ,culture positivity of BACTEC BD Fx40 blood culture system was 26.67%. This is consistent with Vasudev et al(2015) whose culture positivity is 31.2% and Kohli and Kochhar et al (2011) whose positivity is 23%.

In our study maximum positive cases were found in age group 18-60 years which is again consistent with Vasudev et al (2015). In this study ,men had higher culture positivity as compared to women which is consistent with study done by Kaur and Singh(2014), similar observation was reported by Vasudev et al (2015).

In the present study maximum blood culture positivity was found in ICUs which could be due to the fact that blood stream infections are more likely in seriously ill patients and patients with medical and surgical interventions leading to nosocomial infections.

The common isolates in blood culture in our study was S.aureus 20(50%) followed by K.pneumoniae 4(10%) which is again consistent with the study by Prabhu et al (2010) which showed prevalence of S. aureus 50.61% and K. pneumoniae 12.3%.

Higher resistance to the methicillin (60%) among S.aureus is noted as compared to other studies which could be due to preponderance of ICUpatients in our study and indiscriminate use of higher antibiotics as emergency empirical therapy. Gram positive coccci isolates showed 95% sensitivity to teicoplanin and 100% sensitivity to vancomycin and linezolid which is consistent with other studies such as Vasudev et al(2015), Fayyaz et al(2013).

Among the Gram negative organisms high resistance was noted against Ampicillin which is consistent with Vasudev et al (2015) and Fayyaz et al (2013). High resistance(100%) was seen against cefazolin and cefuroxime while resistance against cefipime and ceftazidime was 50% which is consistent with study by Prabhu et al (2010). Sensitivity to piperacillin tazobactum and imipenem was noted to be 62.5% which is a pattern of concern.

Salmonella Typhi was isolated from 2(5%) samples which were both resistant to ampicillin while being 100% sensitive to cephalosporins which is comparable to study by Prabhu et al(2010).

CONCLUSION

This study has shown that S. aureus and Klebsiella pneumoniae are the leading causes of septicemia in ICU patients a pattern similar to other low income countries. Also another observation is decline in susceptibility of common pathogens to common antibiotics which calls for rational use of antibiotics. The main driving force behind the increase in antimicrobial resistant bacteria are poor infection control practices and inappropriate use of antibiotics. Specific antibiotic utilization strategies like antibiotic restriction , combination therapy and antibiotic recycling may help to decrease or prevent the emergence of resistance.

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