International Journal of Advanced Biotechnology and Research (IJBR)
ISSN 0976-2612, Online ISSN 2278–599X,
Vol-8, Issue-4, 2017, pp744-750
http://www.bipublication.com

Research Article

Investigating the Prevalence of Staphylococcus types in the Samples of Children's Blood Culture in Hospitalized Patients at Pediatric Wards

Zahra Gorjian[1] and Masoumeh Ghasemnezhad[2]*

[1]Faculty Member of Nursing Department. Abadan School of Medical Sciences, Abadan, Iran.
[2]MSc in Medical Microbiology.Student Research Committee,
Abadan School of Medical Sciences, Abadan, Iran.
Corresponding author: Masoumeh Ghasemnezhad, Student Research Committee,
Abadan School of Medical Sciences, Abadan, Iran

ABSTRACT

Introduction: Septicemia is a common cause of death toll among hospitalized patients at the pediatric wards. In this survey, the *staphylococc i*infections were investigated regarding their sensitivity and antibiotics resistance.
Materials and methods:In this cross sectional study, the research was conducted through using descriptive research which dealt with the children's blood cultures inoculated three times on blood agar and methyl red eosin medium they were also identified through using Gram colors, standard biochemical tests, catalase and coagulase. Finally, the antibiotic resistance of the *staphylococci* infections was evaluated with the disk diffusion method.
Findings: In this study, the research was lasted 6 months in Abadan Taleghani Hospital. There were121 blood cultures were selected among neonatal and children's samples(i.e., 57 samples related to girls (47.1%), 64 samples are related to boys (52.9%).13 cases of blood culture was positive (10.7%) and 108 cases (89.3%) were negative in terms of *staphylococci*growth.21 children were below1 month (18.4%) and 22 cases were between 1 and 6 months (19.3%).16 cases were patients between 6 months to 1 year (14%),33 cases were between 1 to 2 years old (28.9%), and 22 cases were above 2 years (19.3%).
Results: The most bacterial resistance in Gram-positive starins was seen against Ampicillin and in Gram-negative strains was seen against Ceftoxim. The Gram-positive strains were also mostly sensitive to Vancomycin while the Gram-negative strains were sensitive to Ciprofloxacin.
Conclusion: The results of this study indicated the importance of paying more attention to *staphylococcal* infection control, especially negative Coagulase *staphylococci* which were not common in blood cultures of infants that revealed effective antibiotic usage with high efficacy.

Key words*: Staphylococci*, Blood culture, Infants,kids, Taleghani Hospital of Abadan

INTRODUCTION

Staphylococcus is Gram positive bacteria, which are the most common organisms in medicine. They are also considered as a cause for more than 80% of septic infections and inflammations. *Staphylococcus* is the cause of most chronic infections which one created through using of medical devices and medical equipments. Increasing the use of medical devices has a significant impact on *Staphylococcus* role in nosocomial infections. The frequency of *staphylococcal* types on human skin and repetitive inoculation of devices are the important causes which increase the prevalence of hospitalized patients 'infections (1). In general, Staphylococcus positive catalase can ferment the glucose (2). *Staphylococcus* includes 40 types and

many of them do not cause diseases in humans. However, they are seen on the surface of skin, mucus and other domesticated animals such as dogs, goat, sheep,etc. *Staphylococcus* types are anaerobic and all species of this group also have the potential growth in presence of bile salts. *Staphylococcus* is seen on skin, mucous and membranes, among animals and humans (3) while it is separated from the blood circulation(4-6).Mannitol fermentation and coagulase production help us to distinguish *Staphylococcus aureus(S.*aureus) from other bacteria(2). Some of them are not able to ferment like sugar while these types are considered as aureus. In fact, *Staphylococcus* aureus is one of the most important human and animal pathogens and it is one of the most common causes of nosocomial infections. These infections are spread everywhere and are able to produce a wide range of infections and chronic diseases such as Endocarditis, Osteomyelitis, soft tissue infections and other infections (7-14).With increasing the resistance of bacteria against antibiotics, it is estimated that hospital blood infections as 14% for some types such as negative *staphylococcus*coagulase (15). *Staphylococcus aureus*can be found in different samples of infections related to dialy sisperitoneal tubes, intravenous catheters, blood cultures, tracheotomy tubes, and the infections which are caused by eye contact lenses and other medical devices (16).

Staphylococcus aureus is the major cause of hospital infections among human beings (2). Since 1991, *Staphylococcus* has been identified as one of the most important types which resists to Methicillin, and it is also identified as the cause of acute and chronic infections. Negative Coagulase staphylococcus, especially *Staphylococcus aureus* is one of the most common causes of infections related to Dialysis patients. These bacteria often indicate multi-drug resistance (16).

High prevalence of antibiotic resistance, particularly in strains of *S.*aureus, and sometimes other Staphylococcusare the result of horizontal transfer of antibiotic resistance genes that may make the investigation of antibiotic resistance necessary. This study is aimed to examine the frequency of *Staphylococcal* infections in separated blood culture samples of children and infant who were hospitalized inthe pediatric sections. It also investigated theresistance and antibiotic susceptibility and the efficacy of antibiotics. The study was conducted by Moosavian et al (2009), Iran88% of bacteremia bacteremia which were caused by *Staphylococcus* and (41%) caused by *Staphylococcus* aureus in four hospitals of Ahvaz. Cases of bacteremia caused which were by different typesof coagulase-negative included: *Staphylococcus epidermidis*, hemolyticus.*Cohen* were equal to 32 (36%), 10 (11%), 6 (7%) 2 (2.5%) and 2 (2.5%). The most value of separating *Staphylococcal* was related to the patients,who were hospitalized in Burn sections of Taleghani Hospital (19.4%) and Chemotherapy of Shafa hospital (17 %). In addition, the highest number of *Staphylococcus* isolation (60.2%) belonged to the patients with bacteremia and most of them (about 85 percent) were Coagulase-negative *staphylococci* (17). Bakhshi et al (2011) conducted a study and did 402 blood cultures samples of infants who were hospitalized in Urumiah Arefian Hospital in 1.5 year. In this study, 8.9% of blood cultures were positive and prevalence of infection was 61.1% among boys. The highest bacterial resistance in Gram positive was against Ampicillin and the highest sensitivity belonged to Gram-positive strains against Vancomycin and Gram-negative bacteria was against Ciprofloxacin (18).

METHOD

This study, was conducted in Taleghani Hospital of Abadan throughout 6 months. The research sample was selected both morphologically and biochemically. In this study, at first venous blood samples were collected as1-3CC in bottles containing 30 cc of compounds gelatin environment with Peptone 2 g of sodium poly-l and 0.25 grams per liter of ethanol sulfate (i.e., blood culture test technician was MirMedia

Company). The samples were cultured for 24 hours at 37 ° C and then they were put on blood agar medium supplemented with 5% sheep blood and TioGilycolate Broth. Then they were placed for 24 hours at 37° C. Results showed the negative blood culture for 1 week at 37 degrees and they samplesbe re-cultivated three separate times on blood agar medium inoculated to achieve the final results. Dealt with the identification of the bacteria was focused in the second week of the study. Then , in order to separate the desired bacteria Gram stains and prove that the bacteria wereGram-positive, the single colonies were obtained with a morphology similar to colony *Staphylococcus* biochemical testing of coagulase and catalase of the Staphylococcal types. The types of gram positive were obtained from Peru referee Clos (19) for differentiating the *Staphylococcus* and Warner Antibiotic susceptibility testing which were used for other than *S.* saprophiiticcus. This is oxidative ability which grew family S. and Bacitracin 0.4 resistant but Furazolidone 100 sensitive Banners hand, *Staphylococcus epidermidis* to sensitive to Acid-resistant. However, the Saprophiiticcus Novobiocin resisted and the Urease positive and Lipase positive intended to be phosphatase test Negatives.

Hinton agar disk diffusion was method as a disk diffusion of culture and antibiotic was placed on Penicillin, Doxycycline, Tetracycline, Amoxicillin, Cefazolin, Ampicillin, Gentamycin, Oxacillin, Azithromycin, Cefexime, Ciprofloxacin, Vancomycin, Cefterxion, Imipenem, Ceftazidime, Clindamycin, Cephalotin, Amikasin, Co-amoxiclave, Co-trimazol, Cloxacilin, Cefizoxim, piperacillin and Cefotaxime.

After 14 or 16 hours of incubation at 37 ° C , the results showed the sensitivity of antibiotic resistance.

Findings

In this study, 121 blood cultures of children were used among children who were hospitalized in Taleghani Hospital in Abadan in 2015 during six months. 57 samples of girls (47.1%) and 64 cases (52.9%) of boys were selected(Table1).

Boys were more than girls among 13 patients with positive blood culture results (10.7%) and had a growth of Staphylococcal families and 108 (89.3%) were negative Staphylococcal growth (Table 2).

In this study, 21 children were participated with ages less than one month (18.4%). There were also 22 individuals who were between 1 and 6 months (19.3%), 16 cases of children were between 6 months andone year (14%), 33 individuals aged between 1 and 2 years old (28.9%), and 22 children were above 2 yearsold (19.3%).(Table3)and(figure1).

The studies which investigated the resistance and sensitivity of antibiotics showed that the highest degree of sensitivity was seen among the antibiotics including Sephtoximine, Amoxicillin, and Sephtoxim on one hand. The most medical resistance was seen among the antibiotics concerning Erythromycin, Gentamycin, Doxycycline, and Exacyline on the other hand. In the present study, the antibiotic resistance was equally seen in Sephtoxim and Doxycycline(Table 4, Figure2).

Table 1. Examination of gender-based Staphylococcal infections

Percentage	No.	Gender
47.1	57	Girl
52.9	64	Boy

Table2: Positive and negative results in staphylococcal infection

Percentage	No.	Result
10.7	13	Positive
89.3	108	Negative

Table 3. Classification of the participants' age

Percentage	No.	Age
18.4	21	Less than one month
19.3	22	1 to 6 months
14	16	6 months to 1 year
28.9	33	1 to 2 years
19.3	22	More than 2 years

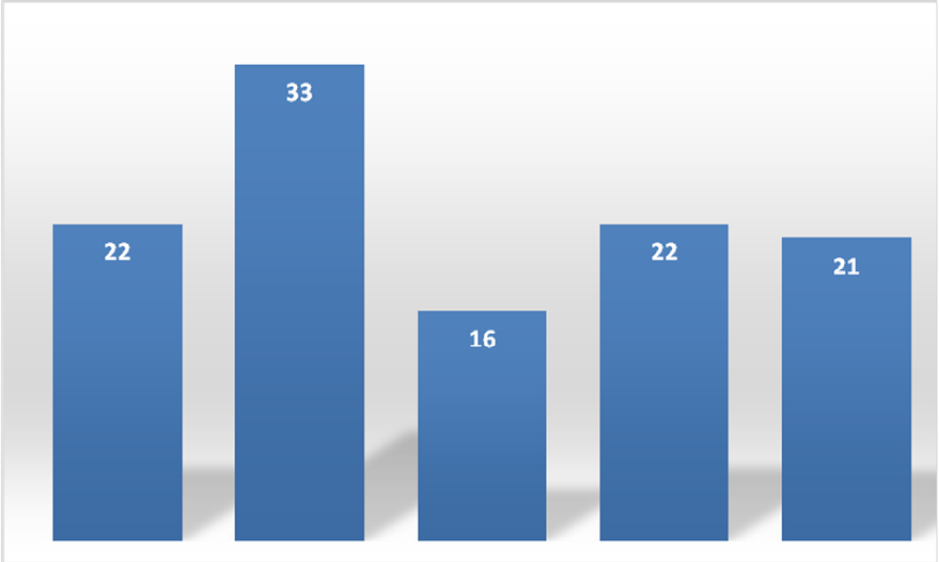

Figure1: Classification based on age range. Above 2 years (22) - 1 to 2 years (33) - Six months to 1 year (16) - Less than 1 to 6 months (21) .

Table 4. Antibiotic sensitivity and resistance

	Staph coagulase(negative)		
	R	I	S
Amikacin	2(40%)	0	3(60%)
Cefexime	4(100%)	0	0
Ampicillin	4(80%)	0	1(20%)
Doxycycline	1(100%)	0	0
Co-amoxiclave	2(50%)	2(50%)	0
Co-trimazole	1(20%)	0	4(80%)
Tetracycline	2(50%)	0	2(50%)
Amoxicillin	1(100%)	0	0
Cloxacilin	4(100%)	0	0
Gentamycin	1(33/33%)	0	2(66/66%)
Imipenem	0	0	1(100%)
Cefizoxim	1(33/33%)	0	2(66/66%)
Ciprofloxacin	1(100%)	0	0
Cephalotin	0	0	2(100%)
piperacillin	0	0	1(100%)
Oxacillin	2(100%)	0	0
Ceftriaxone	1(100%)	0	0
Ceftazidime	2(%66/66)	0	1(33/33%)
Penicillin	5(100%)	0	0
Azithromycin	4(80%)	0	1(20%)
Cefazolin	1(33/33%)	0	2(66/66%)
Clindamycin	2(100%)	0	0
Vancomycin	0	1(%33/33)	2(66/66%)
Cefotaxime	0	0	3(100%)

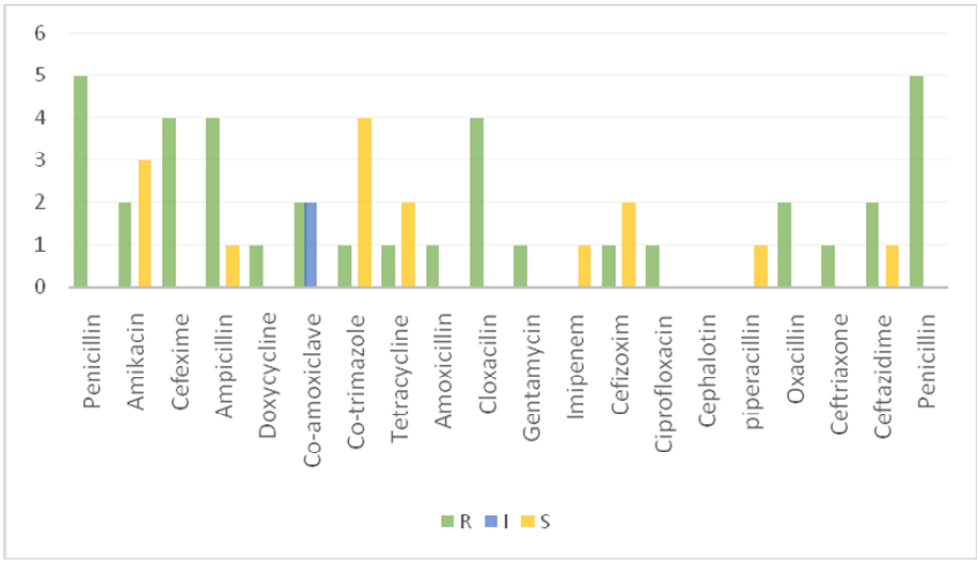

Figure2: Antibiotic sensitivity and resistance

DISCUSSION

The results of study were supported by the study conducted by Asqari et al (2011). This study lasted 6 months and the sample included 121 isolated sample with negative coagulase *staphylococcus*, and most of them were children between 1 and 2 years old. The boys were included the majority of cases. In this study, a wide range of antibiotics were used. Results showed that the most sensitivity to antibiotics was related Tetracycline and Amikacin, and most antibiotics resistance was related to Ampicillin and Cefexime While in the study of Asqari et al (2011), 402 blood culture samples of infants hospitalized in Urumiah Arefian hospital were investigated during 1.5 year. In this study, 8.9% of blood cultures were positive. Prevalence of infections in boys was 61.1%. Most of bacterial resistance in Gram-positive was related to ampicillin and in Gram-negative strains was related to ciprofloxacin.

CONCLUSION

Due to the fact that more than 50% of Staphylococcal septicemia and bacterial infections occur in hospitals among the patients in the hospitals and non-hospitalized mainly among drug users and persons who link with the weakened immune system. The *Staphylococci* infections have symptoms such as chills, shocking, fever, weakness, lethargy and toxicity. The present study survey was conducted on 121 blood cultures of were done among children over 6 months of the study in Abadan Taleghani hospital. Results showed that all samples were isolated from Coagulase negative *Staphylococci* and highest number of children in the age ranged between 1 and 2 years. A wide range of antibiotics was used and the findings showed that the highest sensitivity in this study was related to Tetracycline and Amikacin and the highest antibiotic resistance was related to Ampicillin and Cefexime. Due to the increasing use of self-medication, the development of antibiotic resistance and bacterial infections, examining the study of bacterial infections and measurement of antibiotic sensitivity is necessary in the future. This study suggests further research is needed to investigate the effect of *Staphylococcus* infections on the children's health.

ACKNOWLEDGMENTS

This article was extracted from the project confirmed by Student Research Committee of Abadan School of Medical Sciences with the code

of 94st-0045 and ethical code of IR.ABADANUMS.REC.1394.65 .The authors thank all the officials who did financial support and the patients who took part in the present study.

REFERENCE:

1. Virtanen S. Gram-positive, motile, cluster-forming cocci as a cause of urinary infection. Journal of clinical pathology. 1974 May 1;27(5):408-12.
2. Diemond-Hernández B, Solórzano-Santos F, Leaños-Miranda B, Peregrino-Bejarano L, Miranda-Novales G. Production of icaADBC-encoded polysaccharide intercellular adhesin and therapeutic failure in pediatric patients with staphylococcal device-related infections. BMC infectious diseases. 2010 Mar 15;10(1):68.
3. El-Mahallawy HA, Loutfy SA, El-Wakil M, El-Al AK, Morcos H. Clinical implications of icaA and icaD genes in coagulase negative staphylococci and Staphylococcus aureus bacteremia in febrile neutropenic pediatric cancer patients. Pediatric blood & cancer. 2009 Jul 1;52(7):824-8.
4. García P, Benítez R, Lam M, Salinas AM, Wirth H, Espinoza C, Garay T, Depix MS, Labarca J, Guzmán AM. Coagulase-negative staphylococci: clinical, microbiological and molecular features to predict true bacteraemia. Journal of medical microbiology. 2004 Jan 1;53(1):67-72.
5. Mack D, Rohde H, Dobinsky S, Riedewald J, Nedelmann M, Knobloch JK, Elsner HA, Feucht HH. Identification of three essential regulatory gene loci governing expression of Staphylococcus epidermidis polysaccharide intercellular adhesin and biofilm formation. Infection and immunity. 2000 Jul 1;68(7):3799-807.
6. Eftekhar F, Dadaei T. Biofilm formation and detection of icaAB genes in clinical isolates of methicillin resistant Staphylococcus aureus. Iranian Journal of basic medical sciences. 2011 Mar 1;14(2):132-6.
7. Mirzaee M, Najar-Peerayeh S, Behmanesh M, Forouzandeh-Moghadam M, Ghasemian AM. Detection of Intracellular Adhesion (ica) Gene and Biofilm Formation Staphylococcus aureus Isolates from Clinical Blood Cultures. Journal of Medical Bacteriology. 2015 Oct 13;3(1-2):1-7.
8. Cue D, Lei MG, Luong TT, Kuechenmeister L, Dunman PM, O'Donnell S, Rowe S, O'Gara JP, Lee CY. Rbf promotes biofilm formation by Staphylococcus aureus via repression of icaR, a negative regulator of icaADBC. Journal of bacteriology. 2009 Oct 15;191(20):6363-73.
9. Ghasemian A, Najar-Peerayeh S, Bakhshi B, Mirzaee M. High Prevalence of icaABCD Genes Responsible for Biofilm Formation in Clinical Isolates of Staphylococcus aureus From Hospitalized Children. Archives of Pediatric Infectious Diseases. 2015;3(3).
10. Smith K, Robertson DP, Lappin DF, Ramage G. Commercial mouthwashes are ineffective against oral MRSA biofilms. Oral surgery, oral medicine, oral pathology and oral radiology. 2013 May 31;115(5):624-9.
11. Beenken KE, Dunman PM, McAleese F, Macapagal D, Murphy E, Projan SJ, Blevins JS, Smeltzer MS. Global gene expression in Staphylococcus aureus biofilms. Journal of bacteriology. 2004 Jul 15;186(14):4665-84.
12. Brady RA, Leid JG, Calhoun JH, Costerton JW, Shirtliff ME. Osteomyelitis and the role of biofilms in chronic infection. Pathogens and Disease. 2008 Jan 1;52(1):13-22.
13. Fitzpatrick F, Humphreys H, O'gara JP. The genetics of staphylococcal biofilm formation—will a greater understanding of pathogenesis lead to better management of device-related infection?. Clinical microbiology and infection. 2005 Dec 1;11(12):967-73.
14. Luong TT, Lei MG, Lee CY. Staphylococcus aureus Rbf activates biofilm formation in

vitro and promotes virulence in a murine foreign body infection model. Infection and immunity. 2009 Jan 1;77(1):335-40.
15. Cramton SE, Gerke C, Schnell NF, Nichols WW, Götz F. The intercellular adhesion (ica) locus is present in Staphylococcus aureus and is required for biofilm formation. Infection and immunity. 1999 Oct 1;67(10):5427-33.
16. Tankersley A, Frank MB, Bebak M, Brennan R. Early effects of Staphylococcus aureus biofilm secreted products on inflammatory responses of human epithelial keratinocytes. Journal of Inflammation. 2014 Jun 3;11(1):17.
17. Martín-López JV, Pérez-Roth E, Claverie-Martín F, Gil OD, Batista N, Morales M, Méndez-Álvarez S. Detection of Staphylococcus aureus clinical isolates harboring the ica gene cluster needed for biofilm establishment. Journal of clinical microbiology. 2002 Apr 1;40(4):1569-70.
18. Gad GF, El-Feky MA, El-Rehewy MS, Hassan MA, Abolella H, El-Baky RM. Detection of icaA, icaD genes and biofilm production by Staphylococcus aureus and Staphylococcus epidermidis isolated from urinary tract catheterized patients. The Journal of Infection in Developing Countries. 2009 Jun 1;3(05):342-51.
19. Cucarella C, Solano C, Valle J, Amorena B, Lasa Í, Penadés JR. Bap, a Staphylococcus aureus surface protein involved in biofilm formation. Journal of bacteriology. 2001 May 1;183(9):2888-96.
20. Kloos WE, Bannerman TL. Update on clinical significance of coagulase-negative staphylococci. Clinical microbiology reviews. 1994 Jan 1;7(1):117-40.
21. Hacek DM, Suriano T, Noskin GA, Kruszynski J, Reisberg B, Peterson LR. Medical and economic benefit of a comprehensive infection control program that includes routine determination of microbial clonality. American journal of clinical pathology. 1999 May 1;111(5):647-54.
22. Stoodley P, Kathju S, Hu FZ, Erdos G, Levenson JE, Mehta N, Dice B, Johnson S, Hall-Stoodley L, Nistico L, Sotereanos N. Molecular and imaging techniques for bacterial biofilms in joint arthroplasty infections. Clinical orthopaedics and related research. 2005 Aug 1;437:31-40.