Short Communication

Frequency and anti-biogram pattern of coagulase negative Staphylococcus in clinical specimens of Shahid Mohammadi Hospital in patients, Bandar-Abbas, Iran

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Acceptance 7 June, 2017

Coagulase negative staphylococci (CoNS), commonly known as a part of normal-biota of human body, have become predominant pathogens over the last decades. Due to the vast use of antibiotics for therapy or prophylaxis, those CoNS strains acquired in the hospitals have become resistant to various antimicrobial agents. This descriptive cross-sectional study was carried out from December 2007 to June 2008 on 5063 clinical samples. Identification of CoNS was performed by routine microbiological methods using gram staining, catalase and coagulase tests. Susceptibility of microorganisms to 14 antibiotics was determined by Disc Diffusion Method. Among 1573 positive cultures, 17.5% of isolates were CoNS. About 74% of isolates belonged to out patients and 26% of them were obtained from hospitalized patients. Most of CONs were isolated from internal (38.9%) and internal emergency wards (34.7%). Among different clinical samples, CoNS were mainly isolated from urine cultures (78.6%) in women. Vancomycin (100%) and Imipenem (94%) were found to be the most effective antibiotics. Ciprofloxacin, norfloxacin and aminoglycosides were effective antibiotics to combat CoNS in out patient department (OPD patients).

Key words: Coagulase negative staphylococci, inpatients, antibiogram pattern.

INTRODUCTION

Coagulase negative staphylococci (CoNS), once considered as a common part of the normal-biota of human body, are now a major cause of nosocomial infections, mainly because of increasing use of invasive procedures in hospitals (Devedima et al., 2000; Couto et al., 2001). CoNS are a hetero-genus group of bacteria, consisting of approximately 32 species, among which fourteen species are known to cause infection in human (Lee and Park, 2001). The two most isolates, *Staphlococcus epidermidis* and *Staphlococcus saprophyticus* are well recognized as pathogens isolated from indwelling foreign devices and urinary tract infection, respectively (Kloos and Bannerman,

1995). Seriously ill and immuno-compromised hospitalized patients are the most vulnerable to CoNS infection. The organ sites of infections are different, but virtually all infections involve contamination of prosthetic medical devices (Huebner and Goldman, 1999). CoNS are currently responsible for most late onset infections in neonates (Stoll et al., 2002). In a duration of ten years study, from 1991 to 2000 in 18 Australasian neonatal nurseries, 1281 cases of CoNS sepsis were observed which comprised 57% of all late onset infections (Isaacs, 2003). The widespread use of antibiotics for prophylaxis or therapy has provided a reservoir of antibiotic-resistant strains in hospitals, which show wide variability in their pattern of resistance to antimicrobial agents (Mulder et al., 1997; Cimolia and Carter, 2002). Considering CoNS increasing infections, the aim of this study was to determine frequency and antibiogram pattern of CONS in

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 Table 1. Clinical Sources of coagulase-negative staphylococci isolates.

Sample	Frequency	Percent
Urine	217	78.6
Blood	30	10.9
Wound	13	4.6
Acuities fluid	3	1.1
Eye discharge	3	1.1
Trachea tube	3	1.1
Sinuses derange	2	0.7
Sputum	2	0.7
Peritoneum fluid	1	0.4
Ear discharge	1	0.4
Throat	1	0.4
Total	276	100

clinical specimens submitted to Shahid Mohammadi Hospital Laboratory.

MATERIALS AND METHODS

The present descriptive cross-sectional study was carried out from December 2007 to June 2008 on 5063 different clinical specimens, submitted to Shahid Mohammadi Hospital laboratory. Isolation of the bacteria was carried out by culturing the specimens on appropriate bacteriological media, including Blood agar, Chocolate agar, thioglycollate, EMB or MacConkey agar media. Cultures were incubated at 37°C for 24 - 48 h. Blood samples were inoculated in Trypticase soy broth bottles and incubated for at least 7 days at 37°C. Identification of CoNS was performed by routine microbiological methods using Gram staining, catalase and coagulase tests. In vitro susceptibility of the isolates to 14 antibiotics was determined by standard disc diffusion method, on Mueller-Hinton agar. According to the guidelines of the CLSI, cell suspension inoculates were prepared from 18 - 24 h old pure cultures, in 0.85% sterile saline and adjusted to match a 0.5 Mc Farland standard tubes. Discs contained the following antibiotics, at the specific concentrations indicated in parentheses: Imipenem (10 µg), methicillin (5 µg) vancomycin(30 µg), clindamycin (2 µg), ciprofloxacine (5 µg), ofloxacine (5 µg), norfloxacine (10 µg), lomefloxacine (10 µg), amikacin (30 µg), kanamycin (30 µg), tobramycin (10 µg), gentamicin (10 µg), nethilmicin (30 µg) and rifampin (5 µg) which were purchased from Himedia, India.

RESULTS

Out of 1573 positive cultures, 276 (17.5%) of isolates were CoNS, among which 73.9% belonged to out patient department (OPD) and 26.1% were obtained from inpatients. About 68.5% of CoNS were isolated from women's clinical samples. Table 1 show the frequency of CoNS isolates by body sites. CoNS were most frequently isolated from urine cultures (78.6%). As it is shown in Table 2 the majority of CoNS were obtained from Internal (38.9%) and internal emergency wards (34.7%) patients.

The susceptibility rates of CoNS to tested antibiotics is

presented in Table 3. As it is considered, vancomycin and imipenem with 100 and 94% susceptibility rates were the most active agents, respectively.

DISCUSSION

CoNs are part of the skin and mucus of human normalbiota and a common contaminant of urine, blood and cerebro-spinal fluid cultures. Contamination of specimens complicates the interpretation of culture results and the ability to describe the real history of CoNS infections (Plazzo et al., 2005).

In the present study the overall prevalence of CoNS was found to be 17.5%. Among different clinical samples, CoNS were mainly isolated from urine cultures (78.6%) in young women. The high frequency of CoNS in our urine samples suggests the possibility that some of CoNS positive cultures of urine specimens may be attributed to a contamination rather than a true infection, so more emphasize should be put on mid- stream sampling and sterile condition during collecting urine specimen. In Marsik and Brake study CoNS was mostly isolated from blood cultures (Marsik and Brake, 1982). Prevalence of CoNS strains in blood and wound samples was 10.9 and 4.6%, respectively.

Similar to Chylak and Kholy results, all CoNS strains in this study showed widest susceptibility profile to vancomycin (Chylak and kopka, 2002; Kholy et al., 2003). Tobramycin showed superior activity when compared to other aminoglycosides; and gentamicin with 78% sensitivity rate was the second most active one.

In Chylak study, about 87 - 100% of *Staphylococcus* strains were sensitive to gentamicin, cotrimoxazole and clindamycin. But doxycycline, erythromycin and ampicillin were less effective (Chylak and kopka, 2002).

In Kholy study in Egypt, 23% of CoNs were susceptible to oxacillin but a high resistance rate to erythromycin, cotrimoxazole, clindamycin and doxycycline was observed (Kholy et al., 2003). Ciprofloxacin and gentamicin were the two most potent antimicrobial agents in Pinna's study (Pinna et al., 1999).

The susceptibility rates of our isolates to methicillin and clindamycin were 48 and 42%, respectively. As it is accepted that methicillin resistant strains are resistant to a number of other beta-lactams as well, it is suggested to use penicillins and semisynthetic penicillins just for sensitive strains (Poulsen et al., 2003).

In this study, 31.5% of isolates were resistant to 3 or more antibiotics. Other authors have reported resistance to two or more antibiotics in 29% of staphylococci isolates from patients with chronic belpharitis (Pinna et al., 1999).

Significant increases in multi-drug resistant Gram positive bacteria limits the selection of agents available for therapy, and spread of such strains in hospitals constitutes a threat for immuno-compromised patients. Pattern of antibiotic resistance in coagulase negative

Table 2. Frequency of CoNS isolates in different wards of hospital.

Wards	Frequency	Percent
Internal	28	38.9
Internal emergency	25	34.7
Burn	10	13.9
ICU	3	4.2
Surgical emergency	2	2.8
Orthopedic	2	2.8
ENT	2	2.8
Total	72	100

 Table 3. Antibiogram pattern of coagulase_negative

 Staphylococci strains (expressed as percentage).

Antibiotic	Sensitive	Intermediate	Resistant
Vancomycin	100	0	0
Imipenem	94	4	2
Ciprofloxacin	80	12	8
Tobramycin	80	6	14
Norfloxacin	78	10	12
Gentamicin	78	10	12
Rifampin	78	0	24
Nethilmicin	74	8	8
Amikacin	74	8	8
Kanamycin	66	12	22
Methicillin	51	4	45
Lomefloxacin	42	26	32
Clindamycin	42	28	30
Ofloxacin	28	44	28

Staphylococci are important because they tend to be more resistant than coagulase positive staphylococci. So it is recommended to perform susceptibility tests on all CoNS isolates, considered to be a cause of infection (Marsik and Brake, 1982). According to our results, Ciprofloxacin, norfloxacin and aminoglycosides were effective antibiotics to combat CoNS in OPD patients. But in hospitalized patients who take antibiotics via intravenous, imipenem is recommended, and vancomycin can be administered as the last choice in MDR strains.

In this study a majority of CoNS strains were isolated from patients in internal (38.9%) and internal emergency (34.7%) wards. Considering high rates of infection in these two wards and also because of higher susceptibility of burned-patients to infection, more attention should be paid to disinfection and sterilization procedures in hospitals.

ACKNOWLEDGMENT

The authors are grateful to Shahid Mohammadi laboratory staff for their help with participant enrollment and specimen collection. We also declare no conflicts of interest for this work.

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