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ANALYSIS OF ANTIBIOTIC SENSITIVITY PATTERNS IN DIFFERENT HOSPITALS OF KATHMANDU AND DHULIKHEL HOSPITAL

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ABSTRACT

Introduction: Hospital-acquired infections and community-acquired infections are both linked to Staphylococcus aureus. It has been discovered that staphylococcal isolates from tertiary care hospitals are resistant to widely used antimicrobial drugs. Hospital patient morbidity and death rates have been linked to methicillin-resistant S. aureus (MRSA) with intrinsically established antibiotic resistance. The purpose of this research was to determine the pattern of antibiotic sensitivity of staphylococcal isolates, with a focus on methicillin-resistant S. aureus.

Methods: All S. aureus isolates were included in the research after clinical specimens obtained by Kathmandu Medical College-Teaching Hospital between July 2009 and July 2010 were analyzed. The isolations were identified using conventional laboratory techniques.All staphylococcal strains' antibiotic susceptibility patterns were established using the modified Kirby Bauer antibiotic sensitivity technique.

Out of 111 isolates of S. aureus, 29 (or 26.12%) were found to be MRSA. For MRSA, the rate of multidrug resistance was 75.86%, while for MSSA, it was 6.09%. Every staphylococcal isolate tested negative for penicillin. All strains, meanwhile, responded well to vancomycin.

Conclusions: The Kathmandu Valley's tertiary care hospital has a high frequency of MRSA, according to this research. To lower the frequency of MRSA in hospitals and stop it from spreading to the community, regular surveillance of hospital-associated infections and trend monitoring of antibiotic sensitivity are required. The current research unequivocally demonstrates that the first line of therapy for MRSA infection is still vancomycin. The use of vancomycin should be restricted to situations when it is obviously necessary in order to retain its usefulness.

Key words: Vancomycin, Staphylococcus aureus, MRSA, MSSA, and Antibiotic Sensitivity.

I. INTRODUCTION

S .aureus remains a potent human pathogen, since it is one of the most common cause of nosocomial as well as community acquired infection. 1 It is also the most signiÞcant pathogen known for causing sporadic infections and epidemics. 2 Most of the S. aureus infections are caused by Methicillin- sensitive S. aureus strains (MSSA)that are usually susceptible to major classes of anti-staphylococcal antibiotics.But Resistance to multiple antibiotics among the staphylococci isolates in hospitals has been recognized as one of the major challenges in hospital infection control. 3

In the recent years, the widespread use of antibiotics has undoubtedly accelerated the evolution of MRSAand led to the emergence of strains that have systematically acquired multiple resistance genes. 4 With the current emergence of multi-drug resistant MRSA in hospitals on the one hand 5 and the dramatically increased incidence of hyper-virulent community-associated MRSA on the other hand,

MRSA has been able to evolve rapidly and create new clinical problems. 6 These strains are frequently implicated in serious infections and nosocomial outbreaks, which appear to be disseminated globally in adult, pediatric, and neonatal intensive care units (ICUs).7

The prevalence of MRSA infection varies depending on the characteristics and size of the hospital. In Australia, 31.9% of S. aureus samples taken from 32 laboratories from all states and territories of the country were resistant to methicillin. 8 The number of infections with MRSA in United States hospitals alone rose to nearly 369000 in 2005.9 W hile study done in India also shows prevalence rate as high as 31.1% in clinical samples.10In an earliest study from Nepal, Rai et al.6found that 29% of S. aureus isolates were resistant to methicillin. 11

MRSAare usually found to be resistant to most commonly used antibiotics against staphylococcal isolate. In fact, many stains of MRSA exhibit resistance to both -lactams and aminoglycosides. 12 These strains are seen possessing elevated resistance to a wide range of antibiotics, limiting the treatment optionstovery few agentssuch asvancomycin and teicoplanin. 13 Hence, knowledge of prevalence of MRSA and MSSA and their antimicrobial proÞle becomes necessary in the selection of appropriate empirical treatment of these infections and controlling nosocomial infection. This study was done to know the antibiotic sensitivity pattern of staphylococcal isolates with special reference to Methicillin resistant S. aureus at KMC Teaching Hospital.

II. METHODS

Clinical specimens from July 2009 to July 2010received in clinical microbiology lab of KMC Teaching Hospital were processed and all S. aureus isolates were included in the study. One hundred and eleven (111)isolates of S. aureus were collected from culture samplesreceived from different departments of the hospital. The isolates were consecutive and non repetitive (One per patient). One sample from one patient was inclusion criteria of study data, second sample from other site of same patient was not considered for study.

In Microbiology Lab, samples were cultured on Blood, Mac-Conkey, and Chocolate agars for 24-48 hours. Samples for blood culture were inoculated in Brain Heart Infusion broth and subcultured on 24 and 72 hours on Blood, Mac-Conkey agar. IdentiÞcation of organisms was carried out by standard laboratory operating procedures . 14 (Gram staining, Catalase test, Mannitol fermentation, Slide coagulase and Tube coagulase test).

The antibiotic susceptibility pattern of all the strains was determined by modiÞed Kirby Bauer disc diffusion method against the following antibiotics:penicillin (10 units), gentamicin (10mcg), erythromycin (15mcg), tetracycline (30mcg), co-trimoxazole (25mg), amikacin (30mcg), cephalexin (30mcg), ciproßoxacin (5mg)and vancomycin (30mg). For identiÞcation of MRSA, oxacillin discs (1unit)obtained from Hi-Media Laboratories Pvt. Ltd. was used. A zone of inhibition less than 10mm. or any discernible growth within zone of inhibition was indicative of methicillin resistance. S. aureus ATCC 25923 was used as a standard control strain. Methicillin resistance was conÞrmed for all the MRSA isolates by the agar screening method using Mueller Hinton agar supplemented with 4% NaCl and 6 !g/Lof oxacillin. 15

Multidrug resistance (MDR)is deÞned for this report as resistance to three or more antimicrobial classes. Statistical analysistool (SPSS17)was used to calculate P-value (<0.05 signiÞcant)was calculated using Pearson Chi Square test.

III. RESULTS

Out of 111 S. aureusisolated fromvarious clinicalspecimen. Highest number of isolates was from pus and wound swab and least number from urine. (Table 1)29 (26.12%)of isolates were MRSA (Table 1). Out of 29 MRSA isolated 22(75.86%)were MDR strain. Only 5(6.09%)among 82(73.87%)MSSAwere MDRstrain (Fig. 1).

Table 1: Distribution of S aureus and MRSA in various clinical specimens

Clinical specimen	No of S aureus isolate (%)	MRSA (%)
Pus and wound swab	87 (78.37)	25 (28.73)
Blood	19 (17.11)	3 (15.78)
Urine	05 (04.50)	1 (20.00)
Total	111 (100)	29 (26.12)

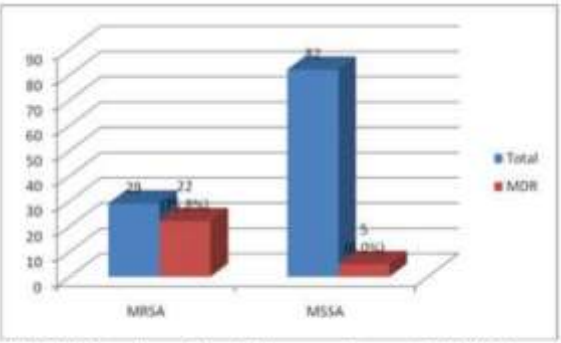

Fig 1: Detection of multidrug resistance in MSSA and MRSA

The antimicrobial susceptibility pattern of MRSA and MSSAisolates against antimicrobial agents are summarized in Table 2. More than 25% of MRSAisolates were resistant to penicillin, oxacillin, cephalexin, co-trimoxazole, erythromycin. Least amount of resistance was observed in vancomycin (0%)ciproßoxacin(17.03%)tetracycline and gentamicin (20.68%). Last but not the least amikacin (24.13%). -lactam antibiotics like penicillin (100% resistance)and cephalexin (30.48% resistance)were found to be ineffective against MSSA too. Rest of the antibiotic showed less than 20% resistance towards the isolated MSSA.

Table 2: Antibiotic resistance pattern of MRSAand MSSA

Antibiotic	MRSA		MSSA		P-value
	Number(n=29)	%	Number(n=82)	%	
Penicillin	29	100	82	100	N/A*
Oxacillin	29	100	00	00	0.0001
Vancomycin	00	00	00	00	N/A*
Gentamicin	06	20.68	05	0.6	0.024
Amikacin	07	24.13	02	2.43	0.001
Co-trimoxazole	13	44.82	17	20.07	0.012
Tetracycline	06	20.68	09	10.97	0.188
Cephalexin	22	75.86	25	30.48	0.0001
Erythromicin	13	44.82	07	8.53	0.0001
Ciprofloxacin	05	17.03	14	17	0.984

IV. DISCUSSION

MRSA has emerged as a serious threat to public health worldwide. It has added to the burden of patient by prolonging hospital stay and increasing morbidity and mortality rate. Present study showed prevalence rate of MRSA to be 26.12%. Another study done in Kathmanu valley by Shrestha et. al. reported 44.9 % as MRSA from nosocomial S. aureus . 16 Rajbhandari et. al. also reported 54.9% MRSA isolates at Bir Hospital . 1 Study done in Eastern Nepal showed comparable result of 26.14% MRSA. 17W hile recent study done in Bharatpur, Nepal had made known worrisome isolation rate of 39.6%.18 MRSA was isolated at the rate 75.5% from clinical samples in a study conducted by Rijal et. al. in Pokhara Valley. 19 Similar study done in western parts of Nepal by Tiwari et. al. also had shown alarmingly high rate of MRSA isolate (69.1%) which the authors has attributed to indiscriminate use of antibiotics and its accessibility in these 20 Above studies show considerable variations between institutions, often in the same geographical areas, exist, demonstrating that MRSA prevalence, in some settings, signiÞcantly exceeds previous estimate. There could be many explanations for these differences:infection control measures, antibiotic prophylaxis and treatments used in each ward/hospital and, not less important, the clonal and often epidemic nature of these microrganisms. 21, 22 Present study also shows maximum number of S.aureus and MRSA

isolation from pus and wound swab (25/29) ascertaining the role of the organism as cause of pyogenic infection. This is similar to the study done in Nepal, Bharatpur, India and Pakistan. 16, 18, 20, 23, 24 Analysis from previous studies revealed a relationship between methicillin resistance and resistance to other antibiotics 16, 18, 22, 24 . This study showed that all MRSA isolates were signiÞcantly less sensitive to antibiotics as compared with MSSA isolates. SigniÞcant difference (P-value < 0.05) was observed in case of oxacillin, gentamicin, amikacin, co-trimoxazole, cephalaxin, erythromycin. However, the difference observed in case of tetracycline and ciproßoxacin was statistically insigniÞcant (P-value > 0.05). Homogeneous insusceptibility to betalactams like penicillin and cephalexin, characteristic MRSAwas also observed in our study. This may be due to presence of intrinsically developed lactamase in MRSA strain. It also showed the high resistance to cotrimoxazole and erythromycin as these antibiotics are usually used at random to cure generalized and pyogenic infection. Antimicrobials such as amikacin, gentamicin and tetracycline with resistance less than 25% could be used against of MRSA infection. But due to their mode of action, have limited use for empirical therapy of MRSA related infection. Resistance to ciproßoxacillin was observed to be 17.93% in this study. Limiting its indiscriminate use and doing antibiotic susceptibility testing it could be considered as a drug of choice forMRSAinfection and can be recommended for empirical therapy in this setting The multi-drug resistant phenotype is a particular characteristic of the methicillin-resistant S.aureus strains. It has added to the burden of hospital personel to control infection associated with MDR-MRSA. Present study shows alarmingly high rate of MDR strain among MRSA isolates (75.86%). Studies conducted in eastern and western part of Nepal also have reported MDR- MRSAto be as high as 65-78%. 16, 19 Indian literature also shows the isolation of MDR-MRSAas high as 77%23 Though these MDR strains are not found with additional virulence properties, their characteristic multidrug resistance restricts the options available to treat infections caused by this organism. 24 Vancomycin a glycopeptides seems to be the only antimicrobial agent which showed 100% sensitivity through all parts of Nepal and may be used as the drug of choice for treating multidrug resistant MRSA infections. But its toxic side effects like renal impairment and prohibitive cost has limited its use. W hen vancomycin is considered for treatment, choice inevitably requires the need for in vitro susceptibility testing of every isolate of MRSAin the clinical laboratories owing to emergence of Vancomycin resistant Staphylococcus aureus (VRSA)in various parts of world.

V. CONCLUSION

Treatment of MRSA infections with these antibacterial drugs would be unreliable in this province due to the high rates of MRSA isolation and its resistance to cephalexin, trimethoprim, erythromycin, and penicillin.Vancomycin is still the first line of therapy for MRSA infections worldwide, but in order to maintain its usefulness, its use should be restricted to situations in which it is obviously required. Additionally, establishing a definitive antibiotic strategy and conducting routine surveillance of hospital-associated infections, such as tracking MRSA and MSSA antibiotic sensitivity patterns, may help lower the prevalence of MRSA infections. Our research provides a window of opportunity for epidemiologists to comprehend the characteristics of MRSA isolates in this region of Nepal.

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