METHICILLIN RESISTANT STAPHYLOCOCCUS AUREUS IN A TEACHING HOSPITAL OF KARACHI - A LABORATORY STUDY

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Abstract
A search was made for Methicillin Resistant Strains of Staphylococcus Aureus (MRSA) among staph aureus cultures isolated at a teaching hospital in Karachi. Of 100 staphylococcus aureus isolated in 1987-88, 5 were MRSA, four from admitted patients and one from outpatient. These MRSA were resistant to Gentamicin as well as to other anti biotics. The presence of MRSA in the in-patients is a serious problem as it can act as reservoir to cause outbreak of colonisation and infection. No MRSA was isolated from 50 samples studied from Quetta (JPMA 39: 6 , 1989).

INTRODUCTION
Strains of Methicillin Resistant Staphylococcus Aureus (MRSA) have caused outbreaks of colonization and infection in intensive care and other special units in hospitals throughout the world. Difficulty in controlling cross infection and the alarm in having only one reserve antibiotic Vancomycin, despite its potential toxicity available for treatment have led to the publication of guidelines for control by a combined working party of the Hospital Infection Society and the British Society for Antimicrobial Chemotherapy. Because of the ease with which these strains spread, the guideline call them the epidemic resistant staphylococcus aureus. Methicillin resistance was reported in staph aureus in 1959 and increased in early 1960. Outbreaks of hospital infection occurred in 1960s and then declined. In 1976 the first outbreak of hospital infection with a strain resistant to both Methicillin and Gentamicin was described in London and a few years later, in Australia there was a nation-wide outbreak of hospital infection with staphylococcus aureus resistant to Penicillin, Methicillin, Gentamicin, Cephalosporins, Erythromycin and Tetracycline. Since then outbreaks have been described in Europe and USA, South Africa and the Middle East. The initiation of many hospital outbreaks has been the introduction of MRSA strain by Infected or colonised patients, or hospital staff. Once within a hospital, resistant staphylococcus aureus spreads like any other staphylococcus aureus predominantly by contact transfer on the hands. In order to identify and control MRSA infection outbreaks, in Karachi and Quetta, a continuous laboratory based surveillance programme has been set up. The present report describes the frequency of MRSA infection in both the cities.

MATERIAL AND METHODS
Material
One hundred clinical isolates of staphylococcus aureus were tested from March 1987 to February 1988 in Jinnah Postgraduate Medical Centre, Karachi and during March 1988 in Quetta for sensitivity to Methicillin.
Methods
Staphylococcus Aureus Strains: Identification of staphylococcus aureus was based on colonial
morphology on blood agar plates incubated at 37° overnight. Gram staining and catalase test (after subculture on nutrient agar media) were done. Gram positive cocci, catalase positive (staphylococci) were confirmed as staph aureus by slide and the coagulase (plasma broth) tests. (Coagulase tests is considered sufficient for research purposes for this kind of work). Two separate colonies of each culture were selected for identification and antimicrobial sensitivity. All tests were performed by Standard methods. Primary plates were reincubated for 48 hours to isolate any further growth of staph aureus.

**Antibiotic Susceptibility Tests**

Antimicrobial susceptibility screening tests were performed on blood agar by the stoke disk diffusion method using Gentamicin 10 ug, Tetracycline 25 ug disks. The ‘Oxford’ strain of S. aureus NCTC 6571 was the control organism in antibiotic susceptibility tests. Methicillin sensitivity with 10 ug disk was done on separate 5 percent salt agar medium plate (Nutrient agar with added 5% NaCl), incubated at 37° for 24 hours and results read. Inoculum of staph aureus which gave confluent growth at 37° after 24 hours was employed (considered best for Methicillin Sensitivity). Oxford Strain of Staph aureus NCTC 6571 was control organism. All apparently resistant cultures to Methicillin were again inoculated from Methicillin sensitivity plate to fresh blood agar plate and organism again identified as Staph Aureus. Disk diffusion sensitivity tests to Methicillin and Gentamicin repeated from single well isolated separate colony. Those isolates that were resistant to Methicillin were then tested for sensitivity to wide range of other antibiotics on diagnostic sensitivity test agar (iso-sensitest agar Oxoid Ltd.) by Stoke Method with the following disks: Penicillin 1.5 units, Tetracycline 30 ug, Erythromycin 15 ug, Gentamicin 10 ug, Fusidic acid 5 ug, Chloromycin 10 ug, Trimethoprim 1.25 ug, Sulphamethoxazole 100 ug, Rifampicin 30 ug, Vancomycin 30 ug, Streptomycin 25 ug, Ofloxacin 5 ug, and Narfioxacin 10 ug. Single disks from oxoid were used. Plates were incubated at 37° overnight and results read. Ten Methicillin sensitive staph aureus strains were saved and disc sensitivity to all above antibiotics was done for comparison purposes.

**RESULTS**

**Isolation of Methicillin Resistant Staph Aureus (MRSA)**

Five Methidillin resistant Staph Aureus strains were isolated from 100 available clinical specimens. All positive samples belonged to males in whom the sample was sent as pus for culture and sensitivity. Four MRSA were isolated in 1987 and one in 1988. All isolates of MRSA were also resistant to gentamicin so called GMRSA (Gentamicin Methicillin Resistant Staph Aureus). Four were isolated from hospital inpatients. The MRSA were isolated from surgical wards, chest ward, outpatients department and an unspecified ward. None of 50 isolates of Staph Aurèus strains from Quetta were Methicillin resistant. Isolation of MRSA (GMRSA) from out-patients One resistant organism was isolated from a patient attending an outpatients department for chronic leg ulcer. Nd information was available regarding past history of any hospital admission. Antimicrobial Sensitivity Tests Results of disk diffusion sensitivity tests with 2 strains of MRSA are shown in Table.
Three specimens were not available, for study by the time all sensitivity disks arrived. MRSA strains were sensitive to Rifampicin, Erythromycin, Chloromycetin, Fusidic acid and Vancomycin and to new quinolones. They were resistant to Penicillin, Tetracycline, Streptomycin, Gentamicin. Sensitivity to Sulphonamide and Trimethoprim was variable. Antimicrobial sensitivity to 10 Methicillin sensitive strains isolated from various specimens were also done.

### DISCUSSION

Methicillin is a valuable drug for the treatment of serious staphylocal infections, so the appearance of MRSA strains is viewed with utmost concern. JPMC receives patients from throughout Karachi City and neighbouring towns; so patients with MRSA once admitted can act as reservoir for cross infection. The detection of first patient with MRSA in a hospital in-patient population is of extreme importance as this can serve as reservoir to cause outbreak of colonization and infection by these organisms. The danger of such strains has been illustrated by major outbreaks of staphylococcus sepsis - in various hospital units. With the identification of the first MRSA positive patient immediate screening programme should be started an other patients and hospital staff. Such an screening programme could not be carried out due to limited available facilities. For the control of MRSA, the working party of Hospital Infection Society and the British Society for Antimicrobial Chemotherapy have set up recommendations which include avoidance of entry of an index case into an unaffected hospital, continuous laboratory based surveillance for the detection of MRSA, prompt and complete definition of the outbreak to include staff nasal carriage and an intensive programme for reducing the reservoir by early discharge of known cases, the isolation of positive patients and the use of antiseptics for colonised skin and superficial sites, i.e. 2% mupiracin nasal ointment four times a day for 5 days, mupiracin skin ointment for skin lesions, 4% chlorohexidine gluconate shampoo daily, triclosan bath concentrates daily. Staff carriers (if only nasal carriers) may be allowed back to work after one day’s treatment with mupiracin (3 weekly negative swabs for wide spread carriage).

### ACKNOWLEDGEMENT

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### TABLE 1. Antimicrobial Sensitivity of MRSA Isolates from Patients.

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R = Resistant
S = Sensitive
REFERENCES