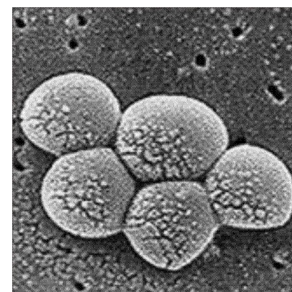


ORIGINAL

PROF-900

PREVALENCE AND SENSITIVITY OF DIFFERENT STRAINS OF BACTERIA; SPECIAL REFERENCE TO MRSA IN A SURGICAL UNIT OF MAYO HOSPITAL LAHORE



DR. I. AHMAD,
MBBS, M.Phil (Microbiology)
Assistant Professor of Pathology
King Edward Medical College Lahore

DR. NAVEED I ANSARI,
MBBS, M.Phil
Associate Professor Pharmacology
K. E. Medical College Lahore

DR. D. LONE,
MBBS, M.Phil (Microbiology)
Senior Demonstrator Bacteriology
K. E. Medical College Lahore

Dr. H. Jamal, MBBS
Sr Medical Officer
Orthopedic Unit II Mayo Hospital Lahore

Dr. M. Awais,
MBBS, MS Orthopedics
Professor, Orthopedic Unit II
Mayo Hospital Lahore

Dr. I.A. Naveed
MBBS, M.Phil (Histopathology)
Professor & Head Pathology Department
K. E. Medical College Lahore
Copyright: 27th March, 2005.

ABSTRACT... Objectives: The correct registration, reporting and analysis of the resistance situation within a hospital is the first step in halting the emergence of antibiotic resistance., we therefore decided o analyze prevalence of bacteria and current trends of antibiotic resistance within our hospital. **Setting:** Orthopaedic ward of Mayo Hospital Lahore. **Period:** Nov 2003 to Apr 2004. **Method & Method:** Isolates were taken from 157 patients admitted in Orthopaedic ward of Mayo Hospital Lahore during 6 months period randomly. The samples were collected from patient's wounds, the patients were not given any antibiotics 48 hours prior to collection of the specimen. Isolates were also taken from the environment (Rooms, AC ducts, corridors of the wards, operation theatre) and OT instruments. Isolates were inoculated on standard media in optimum environment and standard internationally accepted methods were applied for identification of bacteria. **Results:** We found out that the most prevalent bacterium to be staphylococcus aureus 33 (18.75%)and seven were MRSA susceptible to Amikacin only. Where as the other bacteria pseudomonas auregenosa 38(21.59%), E.Coli 19(10.79%). Proteus14(7.95%). Streptococcus pyogenes10(5.68%), enterobacter 8(4.55%), Klebsiella 5(2.84%) more or less showed sensitivity to Amikacin. No growth was seen in 36 cases From among the 50 environmental isolates, Bacillus 18(36%) Staphylococcus in combination with bacillus 23(46%) and No growth in 9(18%).

INTRODUCTION

Health and diseases have always been associated as

part of human life since its origin. Taking these aspects of life into consideration one can not neglect the part played by micro-organisms. History is witness to the fact

that man has always been in search of agents to cure disease. This led to the search for materials, which could undo the detrimental effects of fungi, bacteria and viruses on human and animal life.

Staphylococci are among the most important causes of both hospital- and community- acquired infections world wide¹. *Staphylococcus aureus* causes superficial and deep skin and soft tissue infections, bacteremia with metastatic abscess formation, and wide variety of toxin-mediated infections, including gastroenteritis, staphylococcal scalded skin syndrome and toxic shock syndrome⁸.

MRSA is known to be one of the most prevalent nosocomial pathogens throughout the world and to be capable of causing a wide range of hospital-linked infections⁶.

The availability of a wide selection of antibacterial drugs constitute a therapeutic cornerstone in modern medicine, providing effective therapy for most bacterial infections, prophylaxis in surgical procedures with significant infection risks, and the control of infections in immunocompromised hosts. Until some 10-15 years ago, the number of infectious diseases that could be cured or controlled with antimicrobial drugs increased continuously, but during the last decade, the combination of increasing antibiotic resistance rates and the introduction of few truly new drugs has reversed this favorable situation^{2,5}.

Emergence of resistance is seen in community pathogens as well as in significant hospital pathogens. Severe problems have been particularly frequent within hospitals, notably in units with high consumption of antibiotics and a high risk of spread of resistance bacteria. The emergence of resistance can often be correlated with the over-use of single or multiple antibiotic drugs^{3,4}.

MATERIALS AND METHODS

Isolates were taken from 157 patients admitted in Orthopaedic ward of Mayo hospital Lahore during 06 months period randomly. The samples were collected

from patient's wounds, the patients were not given any antibiotics 48 hours prior to collection of the specimen. The swabs were sent to the department of Pathology KEMC Lahore with in 30 minutes of collection.

Isolates were also taken from the environment (Rooms, AC ducts, corridors of the wards, operation theatre) and OT instruments.

Identification of Bacteria & Antibiotic susceptibility

Identification of isolated organism was performed using standard method⁷.

The smears were also prepared for Gram & ZN Staining.

The specimen were inoculated on Blood agar, Chocolate agar & Mac Conkey's agar media, plates were incubated at 37°C for 24 hours, observed for growth, in case of no growth kept for another 24 hours.

The organism were identified by the following methods.

1. Observation of colonial morphology & color
2. Gram & ZN Staining
3. Catalase test: catalase positive and negative Staphylococci & Streptococci identified
4. Coagulase test: *Staphylococcus aureus* identified
5. Oxacillin sensitivity: MRSA identified (as methicillin is unstable compound so oxacillin is used instead and gives equal response)
6. Biochemical reactions: Indole, methyl red, Voges proskauer and citrate utilization test Gram negative organisms identified
7. Triple sugar iron agar medium: identification differentiation and to prepare stock cultures of various Gram negative organisms.
8. Oxidase test: *Pseudomonas* species identified.

In case of growths over the plates, various organisms were used to prepare 0.5 Mac Farland standard of turbidity in broth media which were poured over the Muller Hinton agar to perform the antibiotic disc diffusion test. After 24 hours the area of inhibition was measured to issue the report and the complete record was

maintained.

RESULTS

The isolation of various bacterial species was first analyzed for with regard to both absolute number isolated both singly/ mixed infection and percentage of the total number (Table I & II) and antibiotic susceptibility (Table III).

Out of 157 patients 36 patients had no infection and in the remaining 121 patients, total number of bacteria isolated were 140, 102 patients had single bacterial infection and in 19 cases two bacteria were isolated. 140 bacterial isolates and 36 no growths were added (176) for the calculation of percentage. 29 isolates were taken from different instruments and environment, In 9 samples no growth was obtained, whereas in 8 samples only bacillus was isolated and in 12 samples mixed infection (Bacillus, Staph. aureus and coagulase negative staphylococcus) was found.

Staphylococci

Out of 176 bacterial isolates, staphylococcus aureus was isolated from 33 (18.75%) cases and coagulase negative staphylococci from 13 (7.39%) being the most prevalent bacterium 46(26.14%). Alone 36(20.46%) and in mixed

growth with *Pseudomonas auregenosa* 7(3.98%), with *Strept. Pyogenase* 2(1.14%), *Enterobacter* 1(0.57%). It was most susceptible to Amikacin and in mixed infection susceptible to Amikacin along with Fusidic acid and Amoxicillin.

Pseudomonas auregenosa

It was the next prevalent bacterium in our study 38 (21.59%) patients. Alone 24(13.64%) and with *Staph. aureus* 7(3.98%), with *E. Coli* 7(3.98).

It was also susceptible to Amikacin.

E. Coli

It was isolated from 19(10.79%) cases being 11(6.25%) alone, 7(3.98%) with *Pseudomonas* and 1(0.57%) with *proteus*. It was observed that in case of infection with *E.coli* alone and with *Pseudomonas* it was susceptible to Amikacin, whereas in infection with *E. coli* & *proteus* Amikacin and Amoxicillin were effective.

Proteus

Out of 157, *proteus* was present in 14(7.95%) patients, alone 12(6.81%), along with *streptococcus* 1(0.57%) and *E.coli* 1(0.57%).

Table I. Incidence of Bacterial Strains

Organism	Patients		Environment	
	No	% Age	No	% Age
Staphylococcus	46	26.14	23	46
<i>Pseudomonas</i>	38	21.59	-	-
<i>E. Coli</i>	19	10.79	-	-
<i>Proteus</i>	14	07.95	-	-
<i>Streptococcus</i>	10	05.68	-	-
<i>Enterbacter</i>	08	04.55	-	-
<i>Kebsiella</i>	05	02.84	-	-
<i>Bacillus</i>	Nil	Nil	18	36
No Growth	36	20.45	9	18

Table II. Percentage incidence of bacterial strains alone and in mixed infections

Organism	Staph		Pseudo		E. Coli		Proteus		Strepto		Entero		Kleb		Total	
	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%
Staph. aureus	23	13.7	7	3.98	-	-	-	-	2	1.14	1	0.57	-	-	46	2.14
CN Staph	13	7.39	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Pseudeo	7	3.98	24	1364	7	3.98	-	-	-	-	-	-	-	-	38	21.59
E.coli	-	-	7	3.98	-	-	1	.57	-	-	-	-	-	-	19	10.79
Proteus	-	-	-	-	1	.57	12	6.81	1	.57	-	-	-	-	14	7.95
Strepto	2	1.14	-	-	-	-	1	.57	7	3.98	-	-	-	-	10	5.68
Entero	1	0.57	-	-	-	-	-	-	-	-	7	3.98	-	-	8	4.55
Kleb	-	-	-	-	-	-	-	-	-	-	-	-	5	2.84	5	2.85
No growth	-	-	-	-	-	-	-	-	-	-	-	-	-	-	36	20.45

Table III. Sensitivity pattern against various antibiotics

Organism	Amox	Ceph	Cef	Cefo	Ceft	Fusi	Gent	Amik	Oflo	Cipro	Clind
Staphylococcus	S	R	R	R	R	S	R	S	R	R	R
Staph (MRSA)	R	R	R	R	R	R	R	S	R	R	R
Pseudomonas	R	R	R	R	R	R	R	S	R	R	R
E. Coli	S	R	R	R	R	R	R	S	R	R	R
Proteus	S	R	R	R	R	R	R	S	R	S	R
Streptococcus	S	S	R	R	R	R	R	S	R	S	S
Enterococci	S	R	R	R	R	R	R	S	R	S	R
Klebsiella	R	R	R	R	R	R	R	S	R	S	R

Amox = Amoxicillin, Ceph = Cephadrine, Cef = Cefaclor, Cefo = Cefotaxime, Ceft = Ceftriaxone, Fusi = Fusidic acid, Amik = Amikacin, Oflo = Ofloxacin, Cipro = Ciprofloxacin, Clind = Clindamycin

Proteus was sensitive to Amikacin in single infection, whereas in mixed infection with E. coli sensitive to Amikacin and Amoxicillin, and in mixed infection with Strep. Pyogenes sensitive to Amoxicillin and Ciprofloxacin.

Streptococcus pyogenes

The incidence of this bacterium was 10(5.68%), alone 7(3.98%), along with staph. 2(1.14%) and proteus

1(0.57%). This bacterium was sensitive to many antibiotics, Cephadrine, Amoxicillin, Ciprofloxacin and clindamycin when causing infection singly, in mixed infection with Staph. aureus susceptible to Amikacin, Fusidic acid and Amoxicillin and in mixed infection with Proteus susceptible to Ciprofloxacin and Amoxicillin.

Enterobacter

The incidence of this bacterium was 08(4.55%), alone

7(3.98%), along with staph. 1(0.57%). This bacterium was sensitive to Amikacin when causing infection singly, in mixed infection with Staph. aureus susceptible to Amikacin, Ciprofloxacin and Amoxicillin.

Klebsiella

The incidence of this bacterium was 05(2.84%) and sensitive to Amikacin and ciprofloxacin.

No growth

There was no growth of any bacterium in 36 cases

Bacterial Incidence in the environment

Out of 29 specimens 9 showed no growth and from the remaining 20 specimens 41 bacteria were isolated, both added up for calculation of percentage (Table I).

Bacillus

Out of 50, 18(36%) specimen were positive for bacillus, 8(16%) occurring singly and 10(20%) in addition to staphylococci.

Staphylococcus

It was present in combination with bacillus 23(46%), 10(20%) being staph aureus and 13(26%) were coagulase negative staph.

No growth

It occurred in 9(18%) 6(12%) in the instruments and 3(6%) in the environment.

CONCLUSION

In our study it was revealed that the most common wound infecting bacterium is Staphylococcus and susceptible to Amikacin, while in mixed infection is also susceptible to Amoxicillin and Fusidic acid. The susceptibility result clearly shows that all the bacteria are susceptible to Amikacin, Amoxicillin has shown sensitivity to most of the bacteria, Cephradine Fusidic acid and Clindamycin are effective in selective bacteria, while most of the organism are resistant to Cephalosporins, Gentamicin and Ofloxacin, This

indicates the injudicious use of these antibiotic to which resistant strains have emerged, while the antibiotics which have been used sparingly are still effective. The incidence of bacterial contamination in the ward, patient rooms is very high (only 3 sites out of 20 were free of bacteria). In the Instruments and OT equipment 3 out of 9 were contaminated.

REFERENCES

1. B. Aygen, A.Yoruk, O.Yyldyz, E.Alp, S.Kocagoz, B.Sumerkan and M.Doganay. **Blood stream infection caused by Staphylococcus aureus in a university hospital in Turkey: Clinical and molecular epidemiology of methicillin resistant Staphylococcus aureus.** Clinical Microbiology and infection, Vol 10 No. 4, April 2004 : 309-314
2. Casadevall A. **Crisis in infectious diseases: time for a new paradigm?** Clin Infect Dis 1996; 23: 790-4
3. deMan P, Verhoeven BA, Verbrugh HA, Vos MC, van den Anker JN. **An antibiotic policy to prevent emergence of resistant bacilli.** Lancet 2000; 355: 973-8
4. Harbarth S, Pittet D, **Multiresistance of gram-negative bacteria in intensive care units: bad news from without.** Crit care Med 1999; 27: 1037-38
5. Hart CA. **Antibiotics resistance: an increasing problem? It always has, been, but there are things we can do.** BMJ 1998; 316: 1255-6
6. Lee J.h. **Methicillin (Oxacillin)-resistant Staphylococcus strains isolated from major food animals and their potential transmission to humans** Applied and Environmental Microbiology, Nov.2003, p. 6489-6494
7. M. Sorberg, A. Farra, U. Ransjo, B. gardlund, M. Rylander, B. settergren, M. Kalin and G. Kronvall. **Different trends in antibiotic resistance rates at a university teaching hospital,** Clinical Microbiology and infection, Vol 9 No. 5, May 2003 : 388-396
8. Waldvogel FA. **Staphylococcus aureus (including staphylococcal toxic shock)** In : Mandell, GL, Bennet, JE, Dolin, R. **Principals and practice of infectious disease.** Philadelphia: Churchill Livingstone: 2000:2069-2092.