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Antimicrobial Resistance in Gram Negative Bacteria Causing Nosocomial Infection in General Hospital

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ABSTRACT

During the study of nosocomial infection different types of microorganisms are isolated from the various sources. Near about 12500 patients were observed for the nosocomial infection. The samples were collected from the patients of various units of the hospital such as chest surgery unit, ICU's, cardiology unit, cardiac surgery, medical unit, orthopedic unit, Pediatric unit, burn unit, ophthalmology unit, neuro surgery, surgical unit, urology, oncology unit and dermatology unit. All the samples were analyzed by different techniques such as isolation, identification, biochemical and biological characterization. During the period of study various pathogenic microorganism such as *Acinetobacter, Pseudomonas, Staphylococcus, streptococcus, E.coli, Enterococcus, Enterobacter, Proteus, Haemophilus, serratia*etc. are isolated. The materials taken from the various sources are analyzed properly. The various patient samples were collected from blood, urine, sputum, pus and wound swab, ulcer, burns swab, endotracheal secretion, cerebrospinal fluid, plural fluid, ascetic fluid, superficial and deep seated, fecal and drainage, medical devices or equipments present in the body and closed abscesses. For the antibacterial study against nosocomial infecting producing microorganism various antibiotics were used these antibiotics are ampicillin, amikacin, cefepime, cefotaxime, ciprofloxacin, gentamicin, piperacillin, and tetracyclines.

KEYWORDS: Nosocomial infection, Antimicrobial resistance, Endotracheal secretion, Cerebrospinal fluid.

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INTRODUCTION

Antibiotic resistance nosocomial infections are increasing dayby day. Patient with immune suppression, illness, poor functional status and who undergo various medical procedures have been reported to be at a high risk of nosocomial infection[1]. According to previous studies 17 different classes of antibiotics have been produced. Unfortunately from the above classes one or many times of more than one antibiotics resistance developed over the years. Antibiotic resistance can ne natural or acquired and can be transmitted horizontally or vertically. The natural form of antibiotic resistance is caused by spontaneous gene mutation in the lack of selective pressure to the presence of antibiotic[2,3].Table-1 showing the major classes of antibiotics with their mechanism of action-

Table 1- List of various antibiotics and their mechanisms of action.

Class of antibiotics	**Mechanism of action**
Beta-lactams (penicillins, cephalosporins, carbapenems, monobactams); glycopeptides; cyclic lipopeptides (daptomycin)	Inhibition of cellwall synthesis
Tetracyclines; aminoglycosides; Polymyxins (Polymyxin-B, Colistin); oxazolidonones (linezolid); streptogramins (quinupristin-dalfopristin); ketolides; macrolides; lincosamides,	Inhibition of proteinsynthesis
Fluoroquinolones	Inhibition of DNAsynthesis
Rifampin	Inhibition of RNAsynthesis
Sulfonamides; trimethoprim	Competitive inhibition of folic acid synthesisInhibition
Polymyxins (Polymyxin-B, Colistin)	Membrane disorganizing agents

There are various gram negative bacteria showing resistance with antibiotics such as extended-spectrum carbapenem-resistant Pseudomonas aeruginosa and Acinetobacterbaumannii, cephalosporin-resistant Enterobacteriaceae and ciprofloxacin-resistant Enterobacteriaceae and non-fermentative Gram-negative bacilli are of great concern because antimicrobial therapy for infections due to these resistant microbial pathogens remains a clinical dilemma in hospitalised patients[4,5].

MATERIALS AND METHODS

Clinical strains:

A total of 12500 samples were collected from the general hospital. These samples were collected from the various wards of the hospital such as chest surgery, ICU's, cardiology dpt. cardio surgery, medical unit, orthopedic dpt. pediatric unit, burn unit, ophthalmic unit, neuro surgery, surgical unit, urology, oncology, dermatology etc. All samples were collected from a single hospital and all the samples were examined in detail.

Identification of microorganisms:

Several microorganisms were isolated from a hospital collected samples. All the isolated were identified by various staining methods, biochemical tests, as well as by commercial identification kits etc(4).From 2943isolates;449 isolates of *E. coli,* 213 isolates of *Enterococcus,* 298 isolates of *Enterobactor,*72 isolates of Haemophylus, *109* isolates of klebsiella,115 isolated of proteus, 412 isolates of *Pseudomonas,*758 isolates of *Staphylococcus,*443 isolates of *streptococcus and* 74isolates of *Serratia*were obtained[6,7].

Antibiotic sensitivity determination:

The antibiotic sensitivity assay was determined by disc diffusion or cup plate method and serial dilution method. For this assay method nutrient agar media or LB agar media were used. The antibiotic used against the bacterial isolates were ampicillin (10µgm/ml), amikacin (3µgm/ml), cefepime (1µgm/ml), cefotaxime (1µgm/ml), ciprofloxacin (1µgm/ml), gentamicin (12µgm/ml), piperacillin (50µgm/ml) and tetracycline (10µgm/ml)[8].

Antibiotic resistance:

Any patient in whom *E. Coli, Entrococcus, Entrobactor, Pseudomonas, Staphylococcus, Streptococcus* and *Haemophilus* spp. were isolated in at least one set of blood sample was considered to have microbiologically confirmed nosocomial infection[9,10].

Incidence of nosocomial infection:

Nosocomial infection is mainly caused by gram negative bacteria. In hospitals the major area of nosocomial infection are medical unit, surgical unit, ICU's, urology, chest surgery unit, cardiology unit,

burn unit etc. In human body the main body parts from where nosocomial infection spread are respiratory tract, urinary tract, gastrointestinal tract, surgical site, blood stream etc. Main gram negative microorganisms which cause hospital acquired infection are *E. Coli, Entrococcus, Entrobactor, Pseudomonas, Haemophilus, Proteus, Klebsiella Staphylococcus, Streptococcus* and *Serratiaetc*[4,11].

RESULT

A total number of 12500 indoor patients during the study period from the various areas of general hospital were observed. All of them were considered to be at risk of contracting the nosocomial infections. The patients who developed various kinds of infections other than their original disease were considered to be nosocomial infections or hospital-acquired infection. Clinical samples were collected from the various parts of the patient. The total samples collected were 186 blood sample, 567 urine sample, 489 sputum sample, 743 pus and wound swabs sample, 40 ulcers sample, 84 burns swabs sample, 220 endotracheal secretions sample, 25 cerebrospinal aspirates sample, 26 pleural fluids sample, 96 fecal and drainage sample and 194 medical devices samples present in the body. Out of 12500; 2670 cases of nosocomial infection were selected and processed for bacteriological analysis. The details of various types of samples collected from patients in various hospital wards are shown in Table-2.

Samples collected from the various sources were analyzed by different techniques such as isolation, identification, biochemical and biological characterization. During the period of study various pathogenic microorganism such as*E.coli, Enterococcus, Enterobacter, Haemophilus, Klebsiella, Proteus, Pseudomonas, Staphylococcus, streptococcus, serratia*are obtained. The materials taken from the various sources are analyzed properly.

Bacteriological studies:

Samples which are obtained from the hospital acquired infected persons were analyzed for the detection of various type microorganisms which are listed in table 3. These microorganisms produce various types of infections which may cause the death of patient. The incidence is increasing; particularly for certain organisms such as multi-resistant coagulase-negative *Staphylococcus* and *Streptococcus* spp. Infection may occur at the skin entry site of the intravascular device, or in the subcutaneous path of the catheter (tunnel infection). Organisms colonizing the catheter within the vessel may produce bacteraemia without visible external infection.

Table 2-List of various hospital departments from samples were collected.

Hospital Units	**Types of sample**										
	Blood Samples	Urine Samples	Sputum Samples	Pus and wound swabs Samples	Ulcers Sample Samples	Burns swabs Samples	Endotracheal secretions Samples	Cerebrospinal aspirates Sample	Pleural fluids Samples	Fecal and drainage samples	Medical devices present in the body Samples
Chest Surgery	20	25	200	100	-	-	-	-	10	5	15
Intensive care units (ICU)	09	45	15	55	03	-	12	-	-	05	12
Cardiology Dept.	15	10	18	15	02	-	05	-	03	04	08
Cardiac surgery	15	20	12	10	01	-	07	-	-	05	06
Medical units	07	210	175	17	12	08	90	10	09	10	75
Orthopedic Dept.	20	50	-	150	05	-	-	-	-	20	25
Pediatric Dept.	12	35	-	25	05	-	08	-	-	-	-
Burn unit	26	-	-	-	-	50	-	-	-	-	-
Ophthalmology Dept.	10	-	-	12	-	-	-	05	-	-	-
Neuro surgery	08	-	14	06	02	-	-	04	-	-	-
Surgical Units	08	102	55	302	09	26	35	06	04	43	51
Urology Dept.	16	65	-	35	01	-	-	-	-	04	02
Oncology Dept.	10	05	-	07	-	-	56	-	-	-	-
Dermatology Deptt.	10	-	-	09	-	-	07	-	-	-	-
Total	186	567	489	743	40	84	220	25	26	96	194

The resident or transient cutaneous flora is the source of infection. The main risk factors are the length of catheterization, level of asepsis at insertion, and continuing catheter care. From 2670 samples of various hospital wards 2943 isolates of microorganisms were isolated. Out of 2943 isolated strands 449 (15.25%) strands of E.coli, 213 (7.24%) strands of Enterococcus, 298 (10.13%) strands of Enterobacter, 72 (2.45%) strands of Haemophilus, 109 (3.70%) strands of Klebsiella, 115 (3.91%) strands of Proteus,

412 (14%) strands of Pseudomonas, 758(25.75%) strands of Staphylococcus, 443 (15.05%) strands of streptococcus and 74 (2.51%) strands of SerratiaThe total number of isolates of various microorganisms from various hospitals wards was listed in the table-3.

Table 3- List of isolates from the various hospital wards (Total number of isolates= 2943).

Hospital Units	**Types of sample**														
	Chest surgery	ICU's	Cardiology dpt.	Cardio surgery	Medical Unit	Orthopedic dpt	Pediatric Unit	Burn Unit	Ophthalmic unit	Neuro surgery	Surgical Unit	Urology	Oncology	Dermatology	Total
E. coli	18	42	09	13	220	13	09	12	--	03	90	17	03	--	449
Enterococcus	18	35	06	15	85	04	04	08	01	03	33	--	01	--	213
Enterobacter	16	30	03	20	130	06	09	10	--	08	56	02	08	--	298
Haemophilus	10	13	--	06	34	--	--	--	--	--	09	--	--	--	72
Klebsiella	10	11	--	09	50	01	--	04	--	--	18	05	01	--	109
Proteus	02	17	--	--	30	04	01	21	--	--	29	11	--	--	115
Pseudomonas	36	20	--	10	102	20	15	72	--	--	130	07	--	--	412
Staphylococcus	65	32	24	31	152	45	22	41	--	08	310	15	13	--	758
Streptococcus	62	53	08	25	135	09	05	42	--	02	102	--	--	--	443
Serratia	05	08	--	01	32	--	03	11	--	--	14	--	--	--	74
Total	242	261	50	130	970	102	68	221	01	24	791	57	26	00	

Antibiotic resistance:

Two thousands nine hundred forty three (2943) bacterial isolates collected from nosocomial infected patients were tested with various antibiotics. The antibiotics used are ampicillin, amikacin, cefepime, cefotaxime, ciprofloxacin, gentamicin, piperacillin and tetracycline. The gram negative pathogens are highly resistance to nearly all the antimicrobial agents. The detected history of antibiotic resistance is given in the table 4a and 4b.

Table 4a.Antimicrobial resistant percentage on various isolates

Antibiotic	**Percentage of resistant isolates among Gram-negative bacteria**				
	E. coli 449/2943	*Enterococcus* 213/2943	*Enterobacter* 298/2943	*Haemophilus* 72/2943	*Klebsiella* 109/2943
Ampicillin	69%	42.2%	78.4%	41%	89%
Amikacin,	8.8%	15%	10.2%	8.7%	9.6%
Cefepime	7%	5.8%	8%	3.1%	14.4%
Cefotaxime	8.3%	56.3%	7.3%	2.5%	21.7%
Ciprofloxacin	28%%	19.7%	13.2%	16.4%	17.3%
Gentamicin	21%	29.3%	28%	11%	14.3%
Piperacillin	9.1%	11.2%	29.4%	13%	13.6%
Tetracyclines	51%	41%	61.2%	92.4%	88.9%

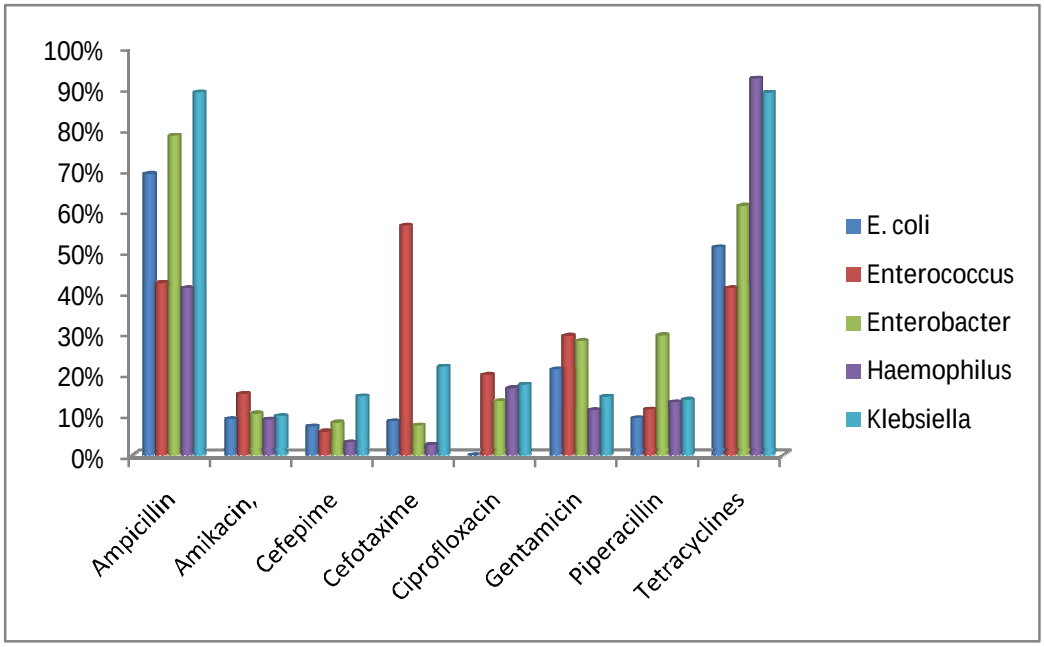

Figure 1. Antimicrobial resistance rate of various isolates against various antibiotics

Table 4b.Antimicrobial resistant percentage on various isolates

Antibiotic	**Percentage of resistant isolates among Gram-negative bacteria**				
	Proteus 115/2943	*Pseudomonas* 412/2943	*Staphylococcus* 758/2943	*Streptococcus* 443/2943	*Serratia* 74/2943
Ampicillin	54%	63%	13.5%	28.4%	91%
Amikacin,	2.2%	16%	6.5%	2.7%	1.5%
Cefepime	1.5%	11.6%	14.3%	11.8%	1.2%
Cefotaxime	3.2%	14.8%	11.7%	10%	3.2%
Ciprofloxacin	5.7%	9.8%	17.9%	31.6%	5.7%
Gentamicin	21.6%	27%	15.3%	8.9%	17.3%
Piperacillin	1%	13.7%	12%	23.6%	1.4%
Tetracyclines	86%	69%	67%	61.4%	89.3%

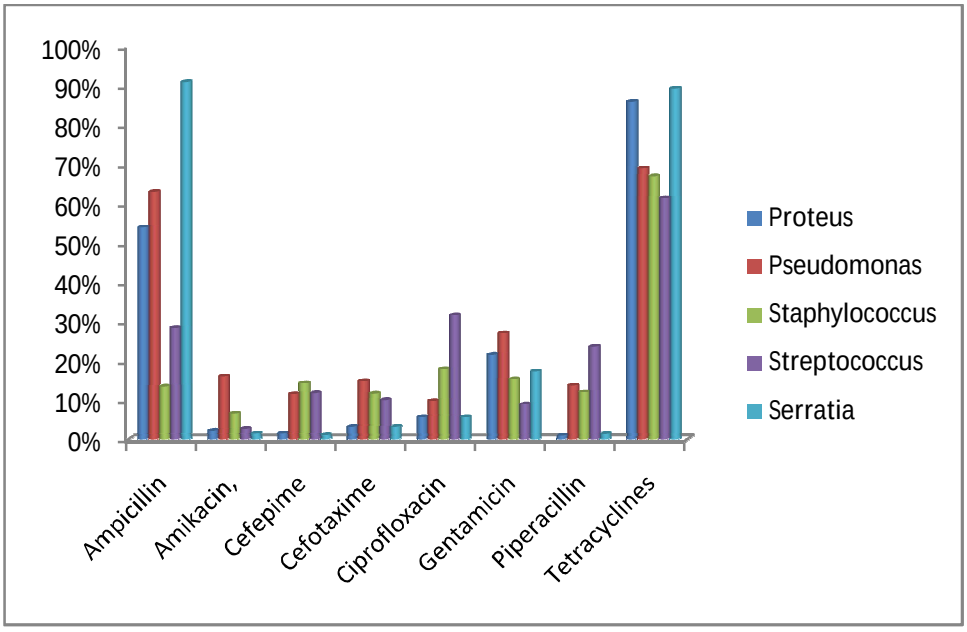

Figure 1. Antimicrobial resistance rate of various isolates against various antibiotics

DISCUSSION

In many countries infection control already plays an important role in hospital management. To assess the importance of active intervention, reliable information on the

consequences of hospital acquired infections has to be obtained. Data from only one, two or a few hospitals mightnot be representative[12].

In the few international studies of hospital acquired infection rates that also deal with the relationship to mortality, a threefold increase has been observed. As is repeatedly described within the literature, hospital acquired infections and their consequences, such as increased mortality and length of hospital stay, are affected by underlying diseases. My study shows that hospital acquired infections have less influence on mortality in hospitals after controlling for age and sex. They do, however, play an important role in overall hospital mortality[13].

All the hospital infections are not preventable even under ideal circumstances. A partial reduction of 15-20% from published rates, which is widely assumed to be a realistic possibility, would produce an important reduction in hospital mortality. This possibility justifies an increased grant allocation for control of nosocomial infection. Taking into account the prolongation of the length of stay in hospital due to hospital acquired infections, this argument is strengthened[14].

CONCLUSION

During the study period 12500 hospital patients were analyzed. Out of which 2670 patients were found to be hospital acquired infected and samples were collected from them. The samples were collected from patient of various wards of the hospital such as chest surgery unit, ICU's, cardiology unit, cardiac surgery, medical unit, orthopedic unit, Pediatric unit, burn unit, ophthalmology unit, neuro surgery, surgical unit, urology, oncology unit and dermatology unit. From 2670 samples, 2943 different type of microorganisms were isolated. The major isolated microorganisms are *E.coli, Enterococcus, Enterobacter, Haemophilus, Klebsiella, Proteus, Pseudomonas, Staphylococcus, streptococcus, serratia.*For the antimicrobial activity various antibiotics such as Ampicillin, Amikacin, Cefepime, Cefotaxime, Gentamicin, Piperacillin and Tetracyclines were used.

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