

## STUDIES ON BACTERIAL POPULATION IN INTENSIVE CARE UNIT OF THANJAVUR MEDICAL COLLEGE

TAMIZHAZHAGAN V.<sup>1</sup>, ASHOK K.\* , RAJESH S.<sup>1</sup>

<sup>1</sup>Department of Zoology, Rajah serfoji Government College, Thanjavur, Tamil Nadu, India, <sup>2</sup>PG and Research Department of Zoology, Presidency College, Chennai 600005, Tamil Nadu, India.  
Email: ashok1984biotech@gmail.com

Received: 22 August 2014, Revised and Accepted: 10 September 2014

### ABSTRACT

In the present study the following bacterial species were isolated and identified from Intensive Care Unit / Intensive Critical Care Unit of Thanjavur Medical College Hospital (TMCH), Thanjavur. Totally five species of bacteria were isolated and identified based on colony morphology, Gram staining and biochemical studies. The identified bacteria namely *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Staphylococcus aureus* and *Serratiamarcensis* spp. All the organisms were tested against antibiotic sensitivity test, the three commercially available common antibiotic discs like Ciproflaxacin, Penicillin G and Streptomycin were used all the organisms were sensitive. Thus, it promotes the treatment against the original pathogen and reduces the consumption of wide spectrum of antibiotic which in turn reduces the consumption of wide spectrum of antibiotic which in turn reduces a wide spectrum of side effects and thus save the humans from the lower extremity amputations in time.

**Keywords:** Biochemical studies, Antibiotics and antibiotic sensitivity test.

### INTRODUCTION

The widespread use of antibiotics both inside and outside of medicine is playing a significant role in the emergence of resistant bacteria [6]. Although there were low levels of preexisting antibiotic-resistant bacteria before the widespread use of antibiotics Caldwell and Lindberg (2011) [2]. Evolutionary pressure from their use has played a role in the development of multidrug resistance varieties and the spread of resistance between bacterial species Hawkey and Jones (2009) [7]. In some countries, antibiotics are sold over the counter without a prescription, which also leads to the creation of resistant strains. In human medicine, the major problem of the emergence of resistant bacteria is due to misuse and overuse of antibiotics by doctors as well as patients WHO (2002) [22]. Other practices contributing towards resistance include the addition of antibiotics to livestock feed [5]. Household use of antibacterial in soaps and other products, although not clearly contributing to resistance, is also discouraged (as not being effective at infection control) [3]. Also unsound practices in the pharmaceutical manufacturing industry can contribute towards the likelihood of creating antibiotic-resistant strains [4].

Certain antibiotic classes are highly associated with colonization with "superbugs" (highly antibiotic resistant bacteria) compared to other antibiotic classes. The risk for colonization increases if there is a lack of sensitivity (resistance) of the superbugs to the antibiotic used and high tissue penetration, as well as broad-spectrum activity against "good bacteria". In the case of MRSA, increased rates of MRSA infections are seen with glycopeptides, cephalosporin and especially quinolones [15]. In the case of colonization with *Clostridium difficile* the high risk antibiotics include cephalosporins and in particular quinolones and clindamycin [17].

The volume of antibiotic prescribed is the major factor in increasing rates of bacterial resistance rather than compliance with antibiotics [13]. A single dose of antibiotics leads to a greater risk of resistant organisms to that antibiotic in the person for up to a year [4].

Inappropriate prescribing of antibiotics has been attributed to a number of causes, including: people who insist on antibiotics, physicians simply prescribe them as they feel they do not have time to explain why they are not necessary, physicians who do not know when to prescribe antibiotics or else are overly cautious for medical legal reasons. For example, a third of people believe that antibiotics are effective for the common cold [11, 12].

Antibiotic sensitivity study is prime important in clinical management of ailing cases caused by various pathogenic

organisms. Several attempts were directed to find out the causal agent of wound and their eradication by antibiotic therapy in man [13, 14]. Specific causal agents of other infectious diseases of humans were diagnosed. Penicillin was the first antibiotic discovered for the treatment of bacterial infection and its indiscriminate use causes emergence of resistant organisms. Currently some important organisms are developing resistance rapidly, including those that cause skin and bloodstream infections, *S. aureus* [15].

In recent years most of the organisms get resistant power against most of the antibiotics. World health organization recently reported the nosocomial infection is mainly causing most of the disease. Recently Mycobacterium *tuberculosis* bacterial strains get resistant power against antibiotics. So in the present investigation justifiably planned with following objectives:

Clinical sample were collected from ICU / ICCU Thanjavur Medical College Hospital (TMCH), Thanjavur, Isolation and identification of bacteria from the open plate method and to study the antibiotic sensitivity test against isolated organism by using Kirby Bauer method.

### MATERIALS AND METHODS

#### Sample collection

In the present study isolate and identify the bacterial population in Intensive Care Unit / Intensive Critical Care Unit. To study the bacterial population the samples were collected from Thanjavur Medical College Hospital (TMCH). For the sample collection nutrient agar plates were used by open plate method at different time intervals in Intensive Care Unit / Intensive Critical Care Unit. The transport media also used to collect the sample by swab the internal things like equipments and patient beds, bed seat etc. After sample collection the samples were brought to the laboratory immediately and kept in 37° C for further analysis.

#### Isolation of bacteria

For bacteriological analysis, samples were pipette out in the 1<sup>st</sup> test tube containing 9 ml sterile distilled water from transport media. The 1 ml of diluted sample was serially diluted to the following dilution factors such as 10<sup>-6</sup>, and 10<sup>-7</sup>. The 0.1 ml of diluted sample was taken from each dilution factor the 1 ml of diluted sample was taken from each dilution factor (10<sup>-6</sup> and 10<sup>-7</sup>). The aerobic heterotrophic bacteria were enumerated in nutrient agar by serial dilution of the sample, followed by the conventional spread plate method of Saha *et al.*, 1995 [14]. *Aeromonas* sp. and *Pseudomonas* sp.

were similarly enumerated on Aeromonas Isolation Medium Base and *Pseudomonas* Isolation Agar, respectively. All the bacteriological media were used Himedia laboratories Ltd product. After inoculation, the Petri dishes containing the culture media were incubated at 37°C for 48 hrs. The populations of bacteria were expressed in terms of cfus/ml (colony forming units) in water, Arithmetical means from three Petri dishes for each dilution were used in the study.

#### Transport media

Weighed Peptone 3 g, NaCl<sub>2</sub> 5g, and then dissolved in 1000 ml of distilled water. Adjust the pH of the medium 7.0 (using 1 N NaOH or 1% HCl sterilized medium) by autoclave at 121°C, 15 lbs pressure for 15 minutes [18, 21].

#### Identification of bacteria

The isolated organisms were subjected into various physiological and biochemical test. The isolated organisms were characterized by Gram staining technique and the organisms were confirmed by Joseph et al., 1996 [8] and the biochemical tests were carried out according to the method of Koneman et al., 2005 [17].

#### Antibiotic sensitivity test by disc diffusion method

The above identified bacterial colonies were study the antibiotic sensitivity test. The strains are following commercially available antibiotic discs used for the Antimicrobial sensitivity studies. The test was carried out by disc diffusion method on Muller Hinton Agar medium (MHA) following the method of NCCLS, 1999[16].

### RESULT AND DISCUSSION

#### Isolation and identification of bacteria

In the present study the following bacterial species were isolated and identified from Intensive Care Unit / Intensive Critical Care Unit of Thanjavur Medical College Hospital (TMCH), Thanjavur. Totally five species of bacteria were isolated and identified based on colony morphology, gram staining and Biochemical studies. The identified bacteria namely *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Staphylococcus aureus* and *Serratiamarcensis* spp., (Table-1).

Aerobic Gram negative bacilli are common in mixed infections with *Proteus* sp., *Escherichia coli*, *Klebsiella* and *Enterobacter* sp., being isolated most often. It was found to be true in this study also and the mixed growth was noticed in 5 cultures (12.1%). *Klebsiella* and *Escherichia coli* and *Pseudomonas* and *Klebsiella* showed mixed growth. However, the common inhabitant of the pus, *proteus* sp., was absent among the isolates. Mixed growth was obtained rarely, only in 5 cultures among the eight positive samples, this was found to be a deviation from the earlier works, in which mixed growth was frequently present and more often than single incidence[10]. Similar studies were observed by [7,9,1].

**Table 1: Isolation and identification of bacteria from sample**

S. No.	Organisms
1	<i>Klebsiella pneumonia</i>
2	<i>Escherichia coli</i> ,
3	<i>Staphylococcus aureus</i>
4	<i>Serratiamarcensis</i>
5	<i>Pseudomonas aeruginosa</i>

**Table 2: Antibiotic sensitivity test by using Ciprofloxacin**

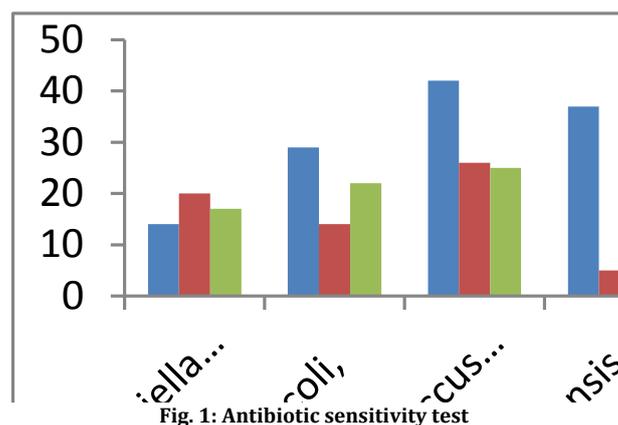
S. No.	Organisms	Zone of inhibition in mm
1	<i>Klebsiella pneumonia</i>	14
2	<i>Escherichia coli</i> ,	29
3	<i>Staphylococcus aureus</i>	42
4	<i>Serratiamarcensis</i>	37
5	<i>Pseudomonas aeruginosa</i>	35

**Table 3: Antibiotic sensitivity test by using Penicillin G**

S. No.	Organisms	Zone of inhibition in mm
1	<i>Klebsiella pneumonia</i>	20
2	<i>Escherichia coli</i> ,	14
3	<i>Staphylococcus aureus</i>	26
4	<i>Serratiamarcensis</i>	5
5	<i>Pseudomonas aeruginosa</i>	14

**Table 4: Antibiotic sensitivity test by using Streptomycin**

S. No.	Organisms	Zone of inhibition in mm
1	<i>Klebsiella pneumonia</i>	17
2	<i>Escherichia coli</i> ,	22
3	<i>Staphylococcus aureus</i>	25
4	<i>Serratiamarcensis</i>	35
5	<i>Pseudomonas aeruginosa</i>	17



**Fig. 1: Antibiotic sensitivity test**

#### Antibiotic sensitivity test

After incubation period, the plates were examined, and the results were observed as zone of inhibition in mm. In antibiotic sensitivity test, three commercially available common antibiotic discs like Ciprofloxacin, Penicillin G and Streptomycin were used. Most of the organisms were sensitive like *Klebsiella pneumonia*, *Escherichia coli*, *Staphylococcus aureus* *Serratiam arcensis* and *Pseudomonas aeruginosa* are sensitive to all three antibiotics and clear zone was formed respectively 14-20 mm, 14-29 mm, 25-42 mm, 5-35 mm and 14-35 mm (Table-2, 3, 4 and Fig. 1). Comparable activities were also observed in Ashok and Jayaprash, 2012 [23]

In these cases, the implementation of standard principles for preventing hospital – acquired infections will result in the prompt eradication of the outbreak. In other hospitals, infections have become endemic, and the clinical and microbiological epidemiology of these infections remain obscure [19, 18, 20, 22]

#### CONCLUSION

These studies will be very much helpful in designing many novel drugs for the beneficial values of human lives.

#### REFERENCE

- Bal A. Dairy animals Foot: magnitude of the problem. J Indian Med Assoc 2002. p. 155-7.
- Roy C, Lindberg, David. Understanding Evolution Mutations arerandom University of California Museum of Paleontology. BMJ 2011;4:3-6.
- CDC. Antibiotic Resistance Questions & Answers [Are antibacterial-containing products (soaps, household cleaners) better for preventing the spread of infection? Does their use add to the problem of resistance?]. Atlanta, Georgia, USA: Centers for Disease Control and Prevention; 2009.

4. Ceire C, Chris M, Andrew L, Alastair D. "Effect of antibiotic prescribing in primary care on antimicrobial resistance in individual patients: systematic review and meta-analysis". *BMJ* 2010. p. 17-8.
5. Ferber D. "Livestock Feed Ban Preserves Drugs Power". *Sci* 2002;5552:27-8.
6. Goossens H, Ferech M, Vander Stichele R, Elseviers M. "Outpatient antibiotic use in Europe and association with resistance: a cross-national database study". *Lancet* 2005;9459:579-87.
7. Hawkey PM, Jones AM. "The changing epidemiology of resistance". *J Antimicrobial Chemotherapy* 2009;64 Suppl 1:i3-10.
8. Joseph WS, Kosinski MA. Prophylaxis in lower extremity infectious diseases. *Clin Poadiatr Med Sur* 1996;13(4):647-60.
9. Kamal K, Powelt RJ, Sumpio BE. The pathobiology of Dairy animals. Implication for surgeons. *J AM Coll Surg* 1996;183(3):271-89.
10. Kamal MM, Parveen N, Saha S, Amin MM. Bacteriological study on uterine discharge in repeat breeder cows. *Bangladesh Vet J* 2001;35:49-52.
11. McNulty CA, Nichols BP, Clappison P, Davey P. "The public's attitudes to and compliance with antibiotics". *J Antimicrob Chemother* 2007;60:i63-8.
12. Nelson William R, Darwin then. Now: The Most Amazing Story in the History of Science I Universe; 2009. p. 294.
13. Pechère JC. "Patients' interviews and misuse of antibiotics". *Clin Infect Dis* 2001;3:S170-3.
14. Saha SC, Zaman MA, Khan MR, Ali SMK. Common aerobic bacteria in post operative wound infection and their sensitivity pattern. *Bangladesh Med Res Coun Bull* 1995;21:32-7.
15. Tacconelli E, De Angelis G, Cataldo MA, Pozzi E, Cauda R. "Does antibiotic exposure increase the risk of methicillin-resistant *Staphylococcus aureus* (MRSA) isolation: A systematic review and meta-analysis". *J Antimicrob Chemother* 2008;61(1):26-38.
16. NCCLS, Performance Standards for Antimicrobial Susceptibility Testing: Ninth Inf Suppl 1999;19(1):68.
17. Koneman WK, Allen SD, Janda WM, Schreckenberger PC, Propcop GW, Woodsand GL, et al. Philadelphia Color Atlas and Textbook of Diagnostic Microbiology, 6th ed. Lippincott-Raven Publisher; 2005. p. 624-62.
18. Chakraborty SP, Kar Mahapatra S, Somenath Roy BM. Isolation and Identification of Vancomycin Resistant *Staphylococcus aureus* from Post Operative Pus Sample. *Al Ameen J Med Sci* 2011a:4:152-68.
19. Cisneros JM, Rodriguez-Bano J, Nosocomial bacteremia due to *Acinetobacter baumannii*: epidemiology, clinical features and treatment. *Clin Microbiol Infect* 2002;8(11):687-91.
20. Bayuga S, Zeana C, Sahni J, Della-Latta P, el-Sadr W, Larson E. Prevalence and antimicrobial patterns of *Acinetobacter baumannii* on hands and nares of hospital personnel and patients: the iceberg phenomena again. *Heart Lung* 2002;31(5):382-90.
21. Corbella X, Montero A, Pujol M. Emergence and rapid spread of carbapanem resistance during a large and sustained hospital outbreak of multiresistant *Acinetobacter baumannii*. *J Clin Microbiol* 2000;38(11):4086-95.
22. WHO. World Health Report: Reducing risks, Promoting Healthy Life. Geneva: World Health Organization; 2002.
23. Ashok K, Jayaprakash P. Screening of active phytochemicals by GC-MS study and antimicrobial activity in the stem of *Santalum album* 2012;4(3):43-4.
24. Cisneros JM, Rodriguez-Bano J. Nosocomial bacteremia due to *Acinetobacter baumannii*: epidemiology, clinical features and treatment. *Clin Microbiol Infect* 2002;8(11):687-91.
25. NCCLS. Performance Standards for Antimicrobial Susceptibility Testing: Ninth Info Suppl 1999;19(1):68.
26. Bayuga S, Zeana C, Sahni J, Della-Latta P, el-Sadr W, Larson E. Prevalence and antimicrobial patterns of *Acinetobacter baumannii* on hands and nares of hospital personnel and patients: the iceberg phenomena again. *Heart Lung* 2002;31(5):382-90.
27. Corbella X, Montero A, Pujol M. Emergence and rapid spread of carbapanem resistance during a large and sustained hospital outbreak of multiresistant *Acinetobacter baumannii*. *J Clin Microbiol* 2000;38(11):4086-95.