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Title : The Antimicrobial Resistance Pattern of pathogens isolated from patients with Health Care Associated Infections in a Tertiary Care Hospital of Chhattisgarh, India.

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Background: Hospital Acquired infections are now frequently recognised as a threat to treatment failure in intensive care units. More than 70% of critically sick patients receive an antibiotic during their stay in the ICU, either for prophylaxis or for treatment. Injudicious use of broad-spectrum antimicrobial agents has led to the emergence of multi drug resistant organisms (MDROs), which can cause addition of more antibiotic resistance organism to Hospital Flora. **Objectives:** The objective of this study was to know the antimicrobial resistance pattern of the pathogens isolated from patients with HCAIs, including identification of Methicillin resistant Staphylococcus aureus (MRSA) and Extended Spectrum Beta Lactamases (ESBL). **Method:** This is a laboratory-based targeted surveillance conducted from 1st June 2019 to 30th September 2019. The study included all the patients admitted in the ICUs, surgical wards and burn ward within the study period and had positive culture result of their clinical specimen. **Results:** Most of the 3rd and 4th generation Cephalosporins were found resistant. Fluroquinolones were resistant in more than half of cases. Most of the organism were resistant to aminoglycosides too. Piperacillin-Tazobactam and Carbapenems were also found resistant in about 70 % of A. baumannii, about 80 % in Pseudomonas and about 90 % in Klebsiella. For many Klebsiella and Pseudomonas isolates, Colistin was the only available option to treat the infection. **Conclusions:** This study showed

the alarming trends of resistance especially in gram negative bacilli isolated from clinical specimens of patients with HCAIs to the various classes of antimicrobials.

Keywords: Health Care Associated Infection. Antimicrobial resistance. Methicillin resistant Staphylococcus aureus, Extended Spectrum Beta Lactamases. Antimicrobial Stewardship.

Introduction

Due to weakened host defences, frequent invasive medical device use, administration of multiple drugs, cross-transmission of pathogens between patients and staff, and insufficient infection control procedures, healthcare associated infections (HCAIs) in particular pose a significant risk to critically ill patients in the ICU .(1)(2) As a result, hospital infections are now frequently recognised as occurring in intensive care units (ICUs).(3) A significant surveillance research found that more than 70% of critically sick patients receive an antibiotic during their stay in the intensive care unit (ICU), either for prophylaxis or for treatment.(4)

In addition, use of broad-spectrum antimicrobial agents has led to the emergence of multi drug resistant organisms (MDROs) which can cause addition of more antibiotic resistance organism to Hospital Flora. This antibiotic resistance can be transferred to strains not having resistance if a patient infected by two different strains. This type of resistance is seen for Penicillin, Cephalosporins, Aminoglycosides, Chloramphenicol, Tetracyclines, Sulphonamides and Vancomycin.(5) On the other hand, due to heavy workload and low staffing levels, healthcare personnel working in ICUs have low compliance with hand hygiene and other basic infection prevention and control (IPC) measures, resulting in cross-infection of microorganisms from patient to patient.(6)

However, in recent years, therapeutic medications have become progressively less successful at treating bacterial infections, endangering the efficacy of standard medical care.(7) Increased patient morbidity, death, medical expense, and treatment failure are the main effects of this issue.(8)(9)

Objectives: Our study was aimed to know the antimicrobial resistance pattern of the pathogens isolated from patients with HCAIs, including identification of multi-drug resistant organisms like Methicillin resistant Staphylococcus aureus (MRSA) and Extended Spectrum Beta Lactamases (ESBL).

MATERIALS AND METHODS

1) **Design of study**: Laboratory based targeted HCAI surveillance. The microbiologists retrospectively visited the ward with the positive culture report to correlate with the clinical data and history.

2) **Place of study**: The study was conducted in the Department of Microbiology, Late Shri Lakhiram Agrawal Memorial Government Medical & Hospital, Raigarh, Chhattisgarh. The clinical specimens were received from various Intensive Care Units, surgical ward and burn ward. The ICUs and wards included in the study were Surgery ward, Orthopaedics ward, Medicine ICU, Neonatal ICU, and burn ward.

Duration of Study - 1st June 2019 to 30th September 2019

Ethical Clearance : Ethical clearance was obtained from the Institutional ethics committee before starting the data collection (vide S.No./27/IEC/GMC/2019 dated 27/04/2019)

Inclusion criterion:

- All the patients who were admitted in the ICUs & Burn ward of the hospital for a period of 48 hours or more and up to 3 days after being discharged or within 30 days of a surgical procedure are considered as population at risk. Informed Consent was taken from all the patient's/guardians.

Exclusion Criteria:

- Those who were admitted in the hospital with fever or any sign and symptoms of infection or developed fever or sign and symptoms of infection within 48 hours of admission are excluded from the study.

Study subjects:

- The Four Major HCAIs are Catheter-Associated Urinary tract Infection (CAUTI), Surgical Site Infections (SSI), Catheter-Related Bloodstream Infection (CRBSI)) and Ventilator- Associated Pneumonia (VAP) and also Burn Associated Skin Infection.

Sample Collection and Laboratory testing: Blood, pus, Swab, urine, ET aspirate and tip of IV catheter were collected using guideline mentioned in SOP-ICMR-AMP from patients showing clinical signs of nosocomial infections.(10)

1. **Culture:** The collected samples was inoculated in appropriate culture media.
   - Blood agar (BA) plate was incubated at 37^{o}C.
   - MacConkey agar plate was incubated aerobically at 37^{o}C.
   - Robertson cooked meat (RCM) broth was incubated at 37^{o}C.
   - Sabouraud dextrose agar was incubated at 37^{o}C and 25^{o}C.

Inoculated primary plates were incubated for 48 to 72 hours and was discarded as negative after examining once daily for 72 hours.

Identification of causative organism

- **Culture:** The colony morphology and cultural characteristics were used to identify the organism.
- **Gram stained smear**: Gram staining was performed to identify whether the causative organism is gram positive or gram negative.
- **Biochemical reactions** – Biochemical tests were used for identification. Some of commonly used tests were Indole test, Citrate utilization test, Urea hydrolysis test, Triple sugar iron test, Coagulase test and Oxidase test.

Antibiotic susceptibility:

- The antibiotic susceptibility was determined by Kirby Bauer disk diffusion method as per CLSI 2018 recommendations.
- The isolates were also tested for Methicillin resistant Staphylococcus aureus (MRSA), Extended Spectrum Beta Lactamases (ESBL) as per CLSI 2018 recommendations.

Detection of Extended Spectrum Beta Lactamases

- Screening with Cefotaxime (30ug) and Ceftazidime (30ug), for Klebsiella species and E. coli, if the zone diameter was found less or equal (as suggested in CLSI 2018), Combined disk method for Cefotaxime in one side and Cefotaxime + Clavulanic Acid on other side and Ceftazidime in one side and Ceftazidime + Clavulanic Acid on other side.

- A ≥5-mm increase in a zone diameter for either antimicrobial agent tested in combination with Clavulanic Acid vs the zone diameter when tested alone was considered as be ESBL producers.

Detection of Methicillin resistant Staphylococcus aureus (MRSA)

Detection with Cefoxitin Disc Diffusion test as per CLSI 2018 guidelines

Data entry and Statistical analysis: The data was entered in Excel sheet and was analysed in SPSS v 20 provided by Public Health Foundation of India (PHFI) and descriptive statistics were used wherever required. For citation and bibliography, Zotero version 6.0.5 was used..

OBSERVATION & RESULTS

Table 1: Antibiotic Resistance* Pattern of Gram-Negative Bacteria

Antibiotics	*K. pneumoniae* (22)	*P. aeruginosa* (12)	*A. baumannii* (10)	*E.coli* (7)	*C.koseri* (2)	*NFGNB* (2)
Ampicillin	NT	NT[#]	NT	100%	100%	NT
Cefazolin	100 %	NT	NT	100%	100%	NT
Cefuroxime	100%	NT	NT	100%	100%	NT
Amoxicillin-Clavulanate	100%	NT	NT	100%	100%	NT
Cefotaxime	100%	NT	100%	85.7%	100%	100%
Ceftazidime	90.9%	100%	100 %	85.7%	100%	100%
Cefoperazon-Sulbactam	90.9%	83.3%	70%	57.1%	100%	100%
Cefepime	95.4%	91.6%	70%	57.1%	50%	100%
Gentamycin	95.4%	66.6%	60%	42.8%	50%	!00%
Tobramycin	90.9%	75%	80%	42.8%	100%	100%
Amikacin	81.8%	75%	50%	14.2%	50%	50%
Piperacillin-Tazobactam	90.0%	83.3%	70%	57.1%	50%	50%
Imipenem	86.3%	100%	70%	71.4%	0%	50%
Meropenem	86.3%	75%	20%	71.4%	0%	50%
Co-trimoxazole	95.4%	NT	80%	100%	0%	50%
Tetracycline	72.7%	NT	50%	71.4%	100%	100%
Colistin	0%	0%	0%	0%	0%	0%
Tigecycline	18.8%	NT	30%	0%	0%	0%
Levofloxacin	81.8%	58.3%	70%	57.1%	0%	100%
Ciprofloxacin	81.8%	58.3%	70%	57.1%	0%	100%
Chloramphenicol	68.1%	NT	NT	71.4%	NT	NT
Fosfomycin	NT	NT	NT	50%[$]	NT	NT

Nitrofurantoin	88.8%$	NT	NT	50%$	50%$	NT
Norfloxacin	100%$	NT	NT	57.1%$	50%$	NT
Nalidixic Acid	100%$	NT	NT	57.1%$	50%$	NT
Ampicillin-Sulbactam	100%	NT	50%	71.4%	100%	100%
Minocycline	NT	NT	30%	NT	NT	NT

*Provided value shows percentage of organism found Resistance to the Antibiotic

NT means **NOT TESTED** (not recommended as per CLSI)

$ Only for Urine Isolates

All identified *Klebsiella pneumoniae* are resistant to cefuroxime, cefotaxime/ceftriaxone, Ampicillin-Sulbactam, Amoxicillin-Clavulanate, and Cefazolin. Cefepime, Piperacillin-Tazobactam, Ceftazidime, Cefoperazone-Sulbactam, Gentamicin, Tobramycin, and Cotrimoxazole are all ineffective against more than 90% of isolates. Amikacin, Levofloxacin, Ciprofloxacin, Imipenem, Meropenem, Tetracycline, and Chloramphenicol resistance rates range from 70 to 80 percent. K. pneumoniae is most sensitive to tigecycline and colistin, with tigecycline sensitivity ranging from 80% to 100%.

Ampicillin, Cefazolin, Cefuroxime, Amoxicillin-Clavulanate, and Cotrimoxazole were completely ineffective against *Escherichia coli*. Resistance to Ampicillin-Sulbactam, Cefotaxime/Ceftriaxone, Ceftazidime, Imipenem, Meropenem, Tetracycline, Chloramphenicol, and Cefuroxime is between 70% and 80%. Additionally, more than 40% are resistant to Gentamicin, Tobramycin, Ciprofloxacin, Cefoperazone-Sulbactam, and Levofloxacin. 90% of E. coli samples were sensitive to Amikacin, while 100% of them were sensitive to Tigecycline and Colistin.

All isolated *Citrobacter koseri* are entirely resistant to the antibiotics Amoxicillin-Clavulanate, Ampicillin, Cefazolin, Cefuroxime, Cefotaxime/Ceftriaxone, Ceftazidime, Cefoperazone-Sulbactam, Tobramycin, and Tetracycline. 50% 0f C. Koseri have a resistance to cefepime, gentamycin, amikacin, and piperacillin-tazobactam. Imipenem, Meropenem, Colistin, and Tigecycline have the highest (100%) degree of sensitivity along with Levofloxacin Cotrimoxazole and Ciprofloxacin.

All identified *Acinetobacter baumannii* were totally resistant to ceftazidime. More than 60% to 70% are resistance to Ciprofloxacin, Levofloxacin, Gentamycin, Tobramycin, and Piperacillin-Tazobactam. Amikacin, Tetracyclines, and Ampicillin-Sulbactam resistance accounted for 50% of the cases. The three drugs that were most sensitive to A. baumannii were tigecycline (70%) minocycline (70%) and colistin (100%).

Pseudomonas aeruginosa isolates are all totally resistant to ceftazidime and imipenem. Resistance to Gentamycin, Tobramycin, Piperacillin-Tazobactam, Amikacin, Cefepime, Cefeparazone-Sulbactam, and Meropenem is more than 60%. Ciprofloxacin and Levofloxacin are both resistant in more than 50% of isolated. Only Colistin showed 100 % sensitivity.

In all isolated Non-Fermenter-GNBs, Cefoperazon-Sulbactam, Ceftazidime, Gentamycin, Tobramycin, Tetracycline, Levofloxacin, and Ciprofloxacin are entirely (100%) resistant. Amikacin, Piperacillin-Tazobactam, Imipenem, Meropenem, and Co-trimoxazole have 50% NFGNB resistance. Tigecycline and Colistin exhibited the highest levels of sensitivity in NFGNBs (100% and 100%, respectively).

Antibiotic Susceptibility Test for Gram Positive was as follows-

Table 2: Antibiotic Resistance* Pattern of Gram-Positive Bacteria

Antibiotic	*E.faecalis* (6)	*CoNS* (6)	*MRSA* (1)
Penicillin	66.7%	NT[#]	NT
Ampicillin	50%	100%	100%
Co-trimoxazole	NT	33.3%	0%
Erythromycin	100%	66.7%	100%
Clindamycin	NT	50%	100%
Tetracycline	100%	100%	100%
Vancomycin	0%	0%	0%
Linezolid	0%	0%	0%
Levofloxacin	100%	66.7%	100%
Ciprofloxacin	66.6%	66.7%	100%
Nitrofurantoin	16.6%[$]	Not isolated in urine	Not isolated in urine
High Level Gentamicin	66.6%	NT	NT
Fosfomycin	100%[$]	NT[&]	NT
Norfloxacin	100%[$]	NT	NT
Amoxicillin- Clavulanate	NT	100%	100%
Moxifloxacin	NT	NT	100%
Teicoplanin	0%	0%	0%
Gentamycin	NT	83.3%	NT

***Provided value shows percentage of organism found Resistance to the Antibiotic**

 # NT means NOT TESTED (not recommended as per CLSI)

$ Only for Urine Isolates

& Not isolated in urine

All isolated *E. faecalis* were completely resistance (100%) to Erythromycin, Tetracycline and Levofloxacin. 50% to 70% are resistant to Penicillin (66%), Ampicillin (50%), Ciprofloxacin (66%), high level Gentamycin (66%). *E. faecalis* shows maximum sensitivity to Vancomycin (100%) and Linezolid (100%).

All isolated CoNS are completely resistance to Ampicillin, Tetracycline, and Amoxicillin-Clavulanate. More than 50% are resistance to Clindamycin, Levofloxacin, Ciprofloxacin and Erythromycin. CoNS shows maximum sensitivity to co-trimoxazole (67%), Vancomycin (100%), Linezolid (100%) and Teicoplanin (100%).

MRSA were also found resistance to Erythromycin, Clindamycin, Tetracycline, Levofloxacin, Ciprofloxacin, and Moxifloxacin. It was sensitive to Co-trimoxazole, Vancomycin, Linezolid and Teicoplanin.

Table 3: ESBL production rate

Species	No. of isolates	ESBL producing No (%)
E. coli	**21**	**6 (28)**
K. pneumoniae	**63**	**15 (22)**

Out of total isolated E. coli and K. *pneumoniae*., 27% were found to be ESBLs producing.

DISCUSSION

In our study *most of the isolated K. pneumoniae (80-100%)* shown resistance towards 3rd generation Cephalosporins, amoxicillin-clavulanate, Gentamicin, Amikacin, Ciprofloxacin, Levofloxacin, Imipenem and Meropenem. Other studies have found *Klebsiella* to be resistant towards Levofloxacin, Ciprofloxacin, Cefepime, Nitrofurantoin.(11) Some showed sensitivity towards Amikacin, Ceftazidime, Piperacillin and Tazobactam.

About 70 to 100% of isolate*d E. coli* were resistant to Ampicillin, Cefazolin, Cefuroxime, Amoxicillin-Clavulanate, and Cotrimoxazole, Ampicillin-Sulbactam, Cefotaxime/ceftriaxone, Ceftazidime, Piperacillin-Tazobactam, Imipenem, Meropenem, Tetracycline and Chloramphenicol and shown good sensitivity (about 60%) to Gentamicin, Tobramycin, Ciprofloxacin, Cefoperazone-sulbactam and Levofloxacin. E. coli showed maximum sensitivity to Amikacin (90%), Tigecycline (100%) and Colistin (100%). The study by Ghadiri et al showed results where E. coli was resistant to Tetracycline and Ampicillin while showed sensitivity towards Imipenem .(12)

Other studies have found *E. coli* resistance towards Nalidixic acid.(12) A study done by *Albert et al also* shows increasing resistance of *Enterobacteriaceae* family.(13)

P. aeruginosa also shown resistance to most of the tested antibiotics tested except Ciprofloxacin, Levofloxacin and Colistin. Other studies have found *Pseudomonas* to be resistant to Cefazolin,(14) Ceftazidime,(15) Ampicillin and Penicillin.(12)

Only 50% of *A. baumannii* were sensitive to Amikacin, Tetracyclines and Ampicillin-sulbactam. *A. baumannii* shown maximum sensitivity to Tigecycline (70%), minocycline (70%) and Colistin (100%). It shown resistance to other tested antibiotics. Similar type of conclusion was drawn by Sohail et al in her study, *Acinetobacte*r isolates were found susceptible to Ampicillin-Sulbactam, Cefepime, Cefotaxime, Ceftazidime, Ceftriaxone, Imipenem, Sulfamethoxazole-Trimethoprim, Amikacin, Gentamicin, Doxycycline, Tigecycline, Ciprofloxacin, Levofloxacin, Tobramycin, Piperacillin-tazobactam.(16)

MRSA showed complete resistance towards Ampicillin, Erythromycin, Clindamycin and Levofloxacin but was sensitive to Vancomycin, Co-trimoxazole, Linezolid and Teicoplanin. Struelens et al states that MRSA is resistant to most of the antibiotics and thus necessitate the use of Vancomycin.(17)

CONS showed maximum sensitivity to co-trimoxazole (67%), Vancomycin (100%), Linezolid (100%) and Teicoplanin (100%). It showed resistance (50-100%) to other tested antibiotics. Some studies have found CoNS to be resistant towards Penicillin and Ampicillin.(12)

All the isolated E. faecalis showed resistance (100%) to Erythromycin, Tetracycline and Levofloxacin. 50% to 70% are resistant to Penicillin (66%), Ampicillin (50%), Ciprofloxacin (66%), high level Gentamycin (66%). E.fecalis shown maximum sensitivity to Vancomycin (100%) and Linezolid(100%). Many studies have been done which shows increasing resistance towards Vancomycin. (18)(19)(20)

About 28% of E. coli were ESBL producing and about 22% of K. pneumoniae were ESBL producing. Study done by Mita D et al shows slightly less value.(21)

The high resistance to antibiotics like Piperacillin- Tazobactam, Meropenem and Imipenem in our study suggests injudicious use of these 2nd line broad spectrum antibiotics. In our setup, Piperacillin- Tazobactam, Meropenem are the most commonly used 1st line antibiotics in ICUs patients. This might be the reason for high degree of resistance toward Piperacillin-Tazobactam and Meropenem. This scenario mandates the need of anti-microbial stewardship programme in hospitals and other health care delivery setups.

CONCLUSION:

Most of the 3rd and 4th generation cephalosporins like Cefotaxime, Ceftriaxone, Ceftazidime and Cefoperazone- Sulbactam and Cefepime were found ineffective. Fluroquinolones like Levofloxacin, Ciprofloxacin and Norfloxacin were resistant in more than half of cases. Most of the organism were resistant to aminoglycosides like Gentamycin, Tobramycin and Amikacin. Piperacillin-Tazobactam and carbapenems (Meropenem and Imipenem) were also found resistant in about 70 % of A.baumannii, about 80 % in Pseudomonas and about 90 % in Klebsiella, which is an alarming situation. For many Klebsiella and Pseudomonas isolates, Colistin (reserve drug for GNB) was the only available option to treat the infection. If this situation worsens, this may be the cause of mortality due to antibiotic failure. So, Anti-microbial stewardship should be started as priority in every hospital.

ESBL and MRSA production rate were high. so, regular surveillance for HAI should be mandatory in healthcare setup.

REFERENCES:

1. Trilla A. Epidemiology of nosocomial infections in adult intensive care units. Intensive Care Med. 1994;20(Suppl 3):S1–4.

2. Majumdar SS, Padiglione AA. Nosocomial infections in the intensive care unit. Anaesth Intens Care Med. 13(5):204–8.

3. Brusselaers N, Vogelaers D, Blot S. The rising problem of antimicrobial resistance in the intensive care unit. Ann Intensive Care. 2011;1:47.

4. Vincent JL, Rello J, Marshall J, Silva E, Anzueto A, Martin CD, et al. International study of the prevalence and outcomes of infection in intensive care units. JAMA. 2009;302(21):2323–9.

5."A preliminary investigation of antibiotic resistance Enterobacteriaceae isolated from children with diarrhoea from four developing countries" P Shears, CA Hart, G Suliman. *Annals of Tropical Medicine & Parasitology 82 (2), 185-188, 1988*

6.Brusselaers N, Vogelaers D, Blot S (2011) The rising problem of antimicrobial resistance in the intensive care unit. *Ann Inten Care 1: 47*

7. Laxminarayan R, Duse A, Wattal C, Zaidi AKM, Wertheim HFL, Sumpradit N, Vlieghe E, Hara GL, Gould IM, Goossens H, Greko C, So AD, Bigdeli M, Tomson G, Woodhouse W, Ombaka E, Peralta AQ, Qamar FN, Mir F, Kariuki S, Bhutta ZA, Coates A, Bergstrom R,

Wright GD, Brown ED, Cars O. Antibiotic resistance the need for global solutions. The Lancet Infectious Diseases. 2013;13(12):1057–98

8. Vasudevan A, Memon BI, Mukhopadhyay A, Li J, Tambyah PA. The costs of nosocomial resistant gram negative intensive care unit infections among patients with the systemic inflammatory response syndrome- a propensity matched case control study. Antimicrob Resist Infect Control. 2015;4(1):1–8.

9. Horcajada JP, Belvis F, Castells X, Grau S, Riu M, Morales E, et al. Hospital costs of nosocomial multi-drug resistant Pseudomonas aeruginosa acquisition. BMC Health Serv Res. 2012;12

10. Standard_Operating_Procedures_Bacteriology_1stEdition.pdf [Internet]. [cited 2022 Aug 28]. Available from: https://main.icmr.nic.in/sites/default/files/guidelines/Standard_Operating_Procedures_Bacteriology_1stEdition.pdf

11. Jafar Soltani, Bahman Poorabbas, Neda Miri, J. M. & Jafar. Health care associated infections, antibiotic resistance and clinical outcome: A surveillance study from Sanandaj, Iran Jafar. WorldWorld J. Clin. Cases4, 63–70 (2016).

12. Ghadiri, H., Vaez, H., Khosravi, S. & Soleymani, E. *Crit. Care Res. Pract.*2012, (2012).

13. Increasing prevalence of antimicrobial resistance among Enterobacteriaceae uropathogens in Dakar, Senegal: a multicenter studyAlbertDromignya, BabacarNdoyeb, Edgard, AdamMacondoc, Pierre, NabethdTidian,eSibycJean, David, Perrier-Gros-Claudea

14. Rajabi, M., Abdar, M. E., Rafiei, H. & Aflatoonia, M. R. Nosocomial Infections and Epidemiology of Antibiotic Resistance in Teaching Hospitals in South East of Iran. Glob. J. Health Sci.8, 190–197 (2016).

15. Ramana, B. V. & Chaudhury, A. Device associated nosocomial infections and patterns of antimicrobial resistance at a tertiary care hospital. J. Dr. NTR Univ. Heal. Sci.1, 86–89 (2012).

16. Sohail, M. et al. Major Article Antimicrobial susceptibility of Acinetobacter clinical isolates and emerging antibiogram trends for nosocomial infection management. 49,300–304 (2016).

17. Struelens, M. J. The epidemiology of antimicrobial resistance in hospital acquired infections: problems and possible solutions. BMJ317, 652–654 (1998).

18.Amyes SG (May 2007). "Enterococci and streptococci". Int. J. Antimicrob. Agents. 29 Suppl 3: S43–52. doi:10.1016/S0924-8579(07)72177-5. PMID 17659211.

19.Courvalin P (January 2006). "Vancomycin resistance in Gram-positive cocci". Clin. Infect. Dis. 42 Suppl 1: S25–34. doi:10.1086/491711. PMID 16323116.

20.Zhanel GG, Hoban DJ, Karlowsky JA (January 2001). "Nitrofurantoin is active against vancomycin-resistant enterococci". Antimicrob. Agents Chemother. 45 (1): 324–6. doi:10.1128/AAC.45.1.324-326.2001. PMC 90284. PMID 11120989.

21. Phenotypic detection of ESBL and MBL in clinical isolates of Enterobacteriaceae" Mita D.Wadekar, K.Anuradha and D.Venkatesha International journal of current research and academic review ISSN: 2347-3215 Volume 1 Number 3 (2013) pp. 89-95