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Original Research

Study on chloramphenicol resistance pattern in Central India, Indore

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ABSTRACT

Background: Chloramphenicol (CK) is a drug of choice in many life threatening and serious bacterial infections. Due to the development of resistance and toxicity associated with this drug, it is not in common use now days. This study was done to find out the prevalence of CK resistance pattern. **Materials and Methods:** A total of 100 consecutive pus samples from the surgical site infections from various specialties of index medical college hospital and research center were studied in the microbiology department during the period of 1 year. **Results:** Gram-positive as well as Gram-negative organisms had shown sensitivity to Chloramphenicol. Furthermore, organisms were resistant to commonly used higher antibiotics. **Conclusion:** CK can be used for common serious ICU patients not having contraindication to this drug. Study also reinforces proper and rational use of antibiotics.

Keywords: Chloramphenicol, drug resistance, Gram-positive and Gram-negative organism

INTRODUCTION

Chloramphenicol (CK) is a potent and efficient antibiotic used since years against many pathogens. It is an easily available, cheap antibiotic, having broad spectrum, and widely active in many conditions where other antibiotics fail this purpose. This drug is not in common use now days, due to the emergence of resistance and fear of bone marrow toxicity.[1]

Even being a potent antibiotic with a broad range of spectrum, the use of CAP is limited due to its association with aplastic anemia (AA)[2] and bone marrow suppression.[3] AA is a rare, dose-independent, irreversible, *idiosyncratic*, manifestation of CAP which in most cases is seen years after the treatment[4] and is fatal.[5] Risk of developing AA after CAP administration is 1:30000-1:5000015.[6] Hutchison and Pinkerton[7] have suggested that the incidence of blood dyscrasias due to Chloramphenicol might be 1 in 80,000 patients treated, Only orally administered CAP leads to AA.[8,9] This has made the CAP to be prescribed parenterally by many physicians.

It is not known whether this lowers the incidence of AA or not but yes the risk is obviously lowered. Other than oral and parenterally absorbed CAP, it is also used as ophthalmic preparations where AA is also very rare.[10-12] Generally in common practice, if the patient is admitted in hospitals blood count is done. Hence, this drug can be used in patient not having contraindication to it and with proper blood investigation before and during treatment.

In a survey of the use of antimicrobial drugs in hospitals in and around Philadelphia H. A. Reimann and J. D'Ambola[13] found that 0.84 kg/month was being used in a teaching hospital, and mention five others in which Chloramphenicol accounted for a large proportion, up to one-half, of all the antibiotics prescribed. Meade's 182 doctors issued an average of only 8.5 prescriptions for it in a month. The Drug Safety Committee's statement refers to 24 known cases of marrow aplasia in a period when 1,000,000 prescriptions were issued plus an unknown number in hospitals. This means that our average prescriber would encounter marrow aplasia only once in at least 500 years of practice, a risk which he may be prepared to take. Most large-scale surveys have produced similar results.

This study is done to reemphasize on its use in chronic and life-threatening infections, as due to stoppage of its use by

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practitioners and shifting toward higher antibiotics has made this drug a good candidate for resistant organisms.

MATERIALS AND METHODS

The study was carried out in the department of Microbiology, Index Medical College Hospital and Research center. It is a cross-sectional study which includes 100 consecutive pus samples from surgical site infections from various surgical specialties of Index Medical College Hospital and Research Center, Indore, Madhya Pradesh, India during period of 01 January 2012-31 December 2013.

Specimen Collection

The specimens were collected aseptically on the 1st day when patients presented with clinical evidence of infection (purulent drainage from incision or drain). Using sterile cotton wool, swabs were obtained from the surgical site without contaminating with skin commensals and transported to the laboratory immediately.

- One swab was used for gram staining.
- The second swab was immediately be inoculated onto Blood Agar, MacConkey Agar and nutrient agar. These plates were kept at 37°C for 24-48 h. The isolates were identified by colony morphology and standard biochemical tests.
- Antibiotics susceptibility study will be performed using Kirby-Bauer disc diffusion methods following CLSI guidelines.[14]

RESULT

A total of 100 wound swabs were collected from patients with post-operative wound infections. The age range of patients in this study was- 4-75 years and the mean age was- 35.28 years. Among these, 65% had bacterial growth within 24 h of incubation. The cultures that were positive yielded a total of 70 aerobic bacteria. Gram-positive organisms were more prevalent than Gram-negative bacteria accounting for 47 (67.14%) and 23 (32.85%) of isolates, respectively. The three most commonly isolated bacterial species were *Staphylococcus aureus* 41 (58.6%) *Pseudomonas aeruginosa* 10 (14.3%) and *Escherichia coli* 06 (8.6%) (Figures 1 and 2).

Among the *S. aureus* isolates, all are resistant to penicillin (100%); some are highly resistant to amoxicillin, amoxy clavulanic acid, ciprofloxacin, ofloxacin, and piperacillin; some had low to moderate resistance to co-trimoxazole, cephalexin, erythromycin, and azithromycin; mostly are sensitive to cefazolin, cefuroxime, chloramphenicol and tetracycline. Among *Enterococcus* spp. isolates, both were found to be resistant to penicillin, amoxicillin, amoxy clavulanic acid, ciprofloxacin, ofloxacin erythromycin and cephalexin while one isolate was found sensitive to co-trimoxazole and chloramphenicol. Among CONS isolates, majority resistant to penicillin, amoxicillin, amoxy clavulanic acid, ciprofloxacin, ofloxacin, co-trimoxazole, cefazolin, cefuroxime and piperacillin; while mild to moderate sensitive to cephalexin, erythromycin,

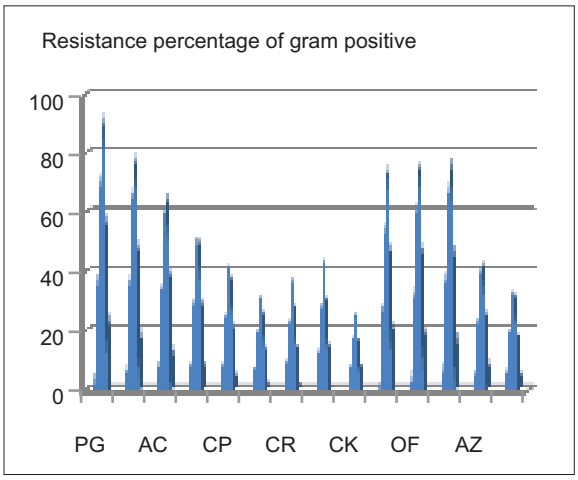

Figure 1: Resistance pattern of Gram positive bacteria

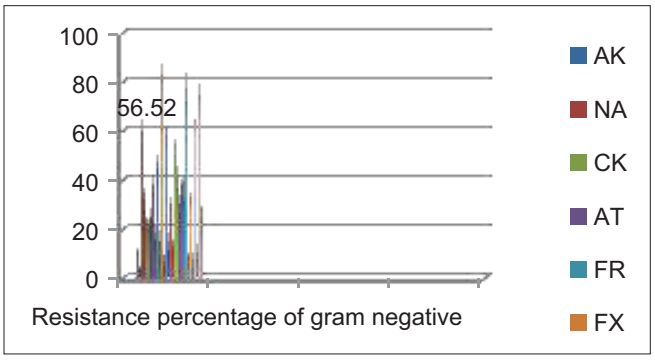

Figure 2: Resistance pattern of Gram negative bacteria

chloramphenicol and tetracycline; and is sensitive to azithromycin. In total, Gram-positive isolates were mostly resistant to penicillin (97.82%), amoxicillin (86.95%), co-amoxiclav (76%), ciprofloxacin (78%), ofloxacin (82.60%) and piperacillin (86.95%); mild to moderate resistant to co-trimoxazole (60.86%), cephalexin (47.82%), erythromycin (45%) and azithromycin (50%) while least resistant to cefazolin (34.78%), cefuroxime (39.13%), chloramphenicol (26.08%) and tetracycline (39.13%).

All staphylococcal isolates were tested for methicillin susceptibility by disc diffusion using oxacillin 1 µg disc. Of 41 COPS, 20 (48.78%) were methicillin-resistant and 21 (51.21%) were methicillin-sensitive. All the methicillin-resistant staphylococci were resistant to penicillin and amoxicillin (100%). 57 (98.3%) MRSAs were highly resistant (80-95%) to co-amoxiclav, co-trimoxazole, cephalexin, ciprofloxacin, ofloxacin, and piperacillin. MRSA isolates were resistant (60-75%) to cefuroxime, erythromycin, azithromycin and tetracycline while 45-50% resistance shown to cefazolin and chloramphenicol. Out of 21 methicillin-sensitive *S. aureus*, all (100%) strains were resistant to Penicillin. The resistance rate of MSSA to amoxicillin, co-amoxiclav, ciprofloxacin, ofloxacin and piperacillin was 76.19%, 66.66%, 71.42%, 76.19% and 80.95% respectively while low level of resistance fond against co-trimoxazole (42.85%), cephalexin (14.28%), cefazolin (9.52%), cefuroxime (9.52%), erythromycin (19.04%), chloramphenicol (4.76%),

azithromycin (38.09%) and tetracycline (4.76%). Thus in the present study, strains of MRSA were found to be highly resistant to many antibiotic showing only intermediate sensitivity to cefazolin (50%) and chloramphenicol (55%), while MSSA strains showed considerable sensitivity to co-trimoxazole (57.15%), cephalexin (85.72%), cefazolin (90.48%), cefuroxime (90.48%), erythromycin (80.96%), chloramphenicol (95.24%), tetracycline (95.24%) and azithromycin (61.91%).

All Enterobacteriaceae isolates showed high resistance to multiple antimicrobial agents tested but all were highly sensitive to amikacin (86.95%). All Gram-negative organisms tested showed low to moderate resistance (36.84%) to ciprofloxacin. Most common Gram-negative isolates from SSIs were found to be highly resistant to third-generation cephalosporin's frequently used for surgical prophylaxis. Most (90%) of pseudomonas spp. Were sensitive to amikacin, 80% to norfloxacin, (70%) of 10 *P. aeruginosa* isolates were sensitive to both gentamicin, aztreonam and ciprofloxacin; while 60% were sensitive to ceftazidime. mostly pseudomonas spp. Were highly resistant to nalidixic acid, cefixime, nitrofurantoin, cefdinir and cefuroxime. Emerging *Acinetobacter baumannii* isolates (1) was resistant to all antimicrobial agents tested in the study except amikacin. In total Gram-negative isolates were highly resistant to nalidixic acid (82.60%), cefixime (56.52%), cefotaxime (65.21%), nitrofurantoin (82.60%), cefdinir (100%) and cefuroxime (88.88%); moderately resistant to aztreonam (56.52%), ceftriaxone (56.52%), ceftazidime (60.86%) and ofloxacin (64.28%); while least resistant to amikacin (13.04%), norfloxacin (39.13%), gentamicin (39.13%) and ciprofloxacin (36.84%).

DISCUSSION

The discovery and development of antibiotics was undoubtedly one of the greatest advances of modern medicine. Unfortunately, the emergence of antibiotic-resistant bacteria, is threatening the effectiveness of many antimicrobial agents. In the present study samples taken from patients with surgical site infection, we found 65% culture positivity. It is much higher than reports by Taye, 2005;[15] Tesfahunegn *et al*., 2009[16] and Biadglign *et al*., 2009[17] with culture positivity of 14.8%, 44.1% and 53.0% and it is lower than studies conducted by Jonathan *et al*., 2008[18] and Adegoke *et al*., 2010[19] (98.5%-100%). The three most commonly isolated bacterial species were *S. aureus* 41 (58.6%) *P. aeruginosa* 10 (14.3%) and *E. coli* 06 (8.6%). Similar rate of staphylococcal wound infection has been reported by Siddiqi *et al*.[20] [46%], Mohanty *et al*.[21] [38.5%] and by Vidhani *et al*.[22] (41.8%). In this study, Gram-positive isolates were mostly resistant to penicillin (97.82%), amoxicillin (86.95%) and co-amoxiclav (76%), which is consistent with reports in different studies conducted in Ethiopia[17,23,24] and India.[25,26] The remarkably higher prevalence of resistance to the commonly prescribed antibiotics such as amoxicillin, ciprofloxacin, ofloxacin and penicillin noticed in the present study may be due to the easily availability (over the counter drugs) and indiscriminate use of the drugs without prescription.[27] High degree resistance to ciprofloxacin (78%), ofloxacin (82.60%) and piperacillin (86.95%) is found in our study.

The frequent empirical prescription of these antibiotics as a treatment and prophylaxis in the study area might contribute for observed high rate of resistance (personal observation). Gram-positive organisms were found to be mild to moderate resistant to cotrimoxazole (60.86%), cephalexin (47.82%), erythromycin (45%) and azithromycin (50%) while least resistant to cefazolin (34.78%), cefuroxime (39.13%), chloramphenicol (26.08%) and tetracycline (39.13%). Earlier studies found high resistance to chloramphenicol,[16,26,28,29] but our study found to be sensitive, this could be due to it might be not commonly used before and/or limited in practice because of its rare but serious side effect of bone marrow suppression. Most common Gram-negative isolates from SSIs were found to be highly resistant to third-generation cephalosporin's frequently used for surgical prophylaxis without any justification. In total Gram-negative isolates were highly resistant to nalidixic acid (82.60%), cefixime (56.52%), cefotaxime (65.21%), nitrofurantoin (82.60%) in line with study done by Mulu *et al*.[23] according to which 100% resistance was found. In our study, Gram-negative isolates were also found highly resistant to cefdinir (100%) and cefuroxime (88.88%); moderately resistant to aztreonam (56.52%), ceftriaxone (56.52%), ceftazidime (60.86%) and ofloxacin (64.28%); while least resistant to, norfloxacin (39.13%), chloramphenicol (39.13%) and ciprofloxacin (36.84%) in line with study done by mulu *et al*. and anguzu *et al*.[23,26] Gram-negative organisms were highly sensitive to amikacin, which is in line with study done in Kanpur.[29]

The resistance of MRSA to a wide range of antibacterials is well documented. The antibiotic sensitivity results showed that all MRSA isolates were more resistant to antibiotics than MSSA isolates. The resistance of MRSA to β lactams like penicillin was 100% in the present study. Similar findings were seen in the studies by Gupta *et al*.,[30] Anupurba *et al*.,[31] Uma choudhary *et al*.,[32] and Anvikar *et al*.[33] Even though 10 isolates of MRSA isolate showed *in vitro* susceptibility to cefazolin in the present study, it should be reported as resistant as recommended by NCCLS, as the drug is ineffective *in vivo*. A high-level ciprofloxacin resistance has emerged very rapidly after its introduction into general use. The resistance rate in the present study was 85%, consistent with the resistance rate of Pulimood *et al*.[34] (90%) and Udaya Shankar *et al*. (95.8%).[35] In the present study, 70% of MRSA isolates were resistant to erythromycin. Higher resistance rate was observed in studies of Anvikar *et al*.[33] (95.9%), and Gupta *et al*.[30] (100%). All the methicillin-resistant staphylococci were resistant to penicillin and amoxicillin (100%). 57 (98.3%) MRSAs were highly resistant (80-95%) to co-amoxiclav, co-trimoxazole, cephalexin, ciprofloxacin, ofloxacin and piperacillin. MRSA isolates were resistant (60-75%) to cefuroxime, erythromycin, azithromycin and tetracycline while 45-50% resistance shown to cefazolin and chloramphenicol. Out of 21 methicillin-sensitive *S. aureus*, all (100%) strains were resistant to Penicillin. The resistance rate of MSSA to amoxicillin, co-amoxiclav, ciprofloxacin, ofloxacin and piperacillin was 76.19%, 66.66%, 71.42%, 76.19% and 80.95%, respectively, while low level of resistance fond against co-trimoxazole

(42.85%), cephalexin (14.28%), cefazolin (9.52%), cefuroxime (9.52%), erythromycin (19.04%), chloramphenicol (4.76%), azithromycin (38.09%) and tetracycline (4.76%). Thus in the present study, strains of MRSA were found to be highly resistant to many antibiotic showing only intermediate sensitivity to cefazolin (50%) and chloramphenicol (55%), while MSSA strains showed considerable sensitivity to co-trimoxazole (57.15%), cephalexin (85.72%), cefazolin (90.48%), cefuroxime (90.48%), erythromycin (80.96%), chloramphenicol (95.24%), tetracycline (95.24%) and azithromycin (61.91%).

CONCLUSION

Both Gram-positive and Gram-negative were sensitive to chloramphenicol that were showing resistance to higher antibiotics. Hence, this study recommends use of parental chloramphenicol where other antibiotics fail for common serious invasive pneumococcal disease patients where these resistant microorganisms are commonly involved. This study also recommends that higher antibiotics should be given a drug holiday for future use.

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PEER REVIEW

Double Blinded externally peer reviewed.

CONFLICTS OF INTEREST

Nil

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