CONCLUSION

Antibiotic resistance in clinical organisms has emerged as a major health problem in the last few decades. The present study was undertaken to determine the trends of antibiotic resistance among bacteria in Kolkata hospitals. We found high percentage of antibiotic resistance to the twenty three antibiotics tested. Forty-three percent of strains were multi-drug resistant (MDR) bacteria, which were resistant to seven or more antibiotics. These multidrug resistant (MDR) bacteria showed greater resistance to the beta-lactam group of antibiotics. The first and second-generation beta-lactam like cephalaxin showed 96.5% resistance, amoxycillin 95.7% resistance and ampicillin 93.3% resistance. We also observed 11.2% Methicillin-resistant Staphylococcus aureus (MRSA). Initially, a few vancomycin resistant Staphylococcus aureus (VRSA) were observed by DAD but MIC value for these strains did not show resistance. The cause of beta-lactam resistance in 28.5% of the MDR bacteria was due to production of beta-lactamase as found by Microiodometric assay and Nitrocefin spot test. On characterizing of the beta-lactamases, we found that of the 28.5% beta-lactamase producing strains of which 16.2% produced Extended Spectrum Beta-lactamase (ESBL), 6.7% were AmpC beta-lactamase producers, 1.4% showed inducible AmpC beta-lactamase, 1.8% were found to be Metallo-beta-lactamase (MBL) producers and 1.4% to be Inhibitor resistant type beta-lactamase (IRT-BL) producers. IRT-BL was resistant to all commercially available beta-lactamase inhibitors e.g., clavulanic acid, sulbactam and tazobactam. Imipenem resistant Proteus vulgaris found in our study is the first report of metallo-beta-lactamase production in P. vulgaris in Kolkata as well as in India. Though ESBL, AmpC beta-lactamase and MBL producers have been reported from South and North India, there had been no report of these types of beta-lactamase producers from Kolkata or from Eastern India.

Beta-lactamases have different pl on polyacrylamide gel and hence can be classified by using isoelectric focusing. We suspected SHV-18 type of beta-lactamase in Klebsiella spp as it had a pl of 7.8 and TEM-1 type of beta-lactamase in some E. coli as it had a pl of 5.4. Isoelectric focusing of beta-lactamase enzyme showed high intensity of color in Nitrocefin Spot Test. The some of strains producing beta-lactamases were tested for presence of plasmids by using Kado and Liu’s method of
plasmid isolation. The isolates showed the presence of multiple small plasmids of size 2.1 kb to 6 kb and one common plasmid of size greater than 53.4 kb which was seen in most *E. coli* strains, but we have not been able to confirm that these plasmids are responsible for beta-lactamase production.

Livermore suggested that particular resistance patterns are associated with particular beta-lactamase; so the enzyme type can be predicted from the antibiogram. For this the bacteria must first be identified to the species level and then a wide range of beta-lactams should be tested. Susceptibility and resistance of the bacterial strains should be determined quantitatively either as zone of inhibition or as MICs (Livermore 1995).

The present study highlights the fact that the antibiotic resistance problem in the hospital continues to worsen and the bacteria in the hospitals are becoming multi-drug resistant. This study also indicates the various types of beta-lactamases are being produced by the bacterial strains obtained from different city hospitals of Kolkata. These beta-lactamases behave quite differently to different beta-lactam antibiotics as well as to the various beta-lactamase inhibitors i.e., clavulanic acid, sulbactam, tazobactam. For proper beta-lactam antibiotic therapy it is necessary to determine the antibiogram of the pathogen. Before administering the beta-lactam drug to the patient detection of the type of beta-lactamase produced by the causative pathogen is very important as this will help in using suitable antibiotics along with appropriate beta-lactamase inhibitor in patients. This will avoid frequent therapeutic failures as is occurring at present.

To cope with this global problem of multi-drug resistant bacteria, we need operational survey and monitoring of multi-drug resistant Gram-negative bacterial species and Gram-positive cocci by World Health Organization (WHO) and national health authorities as well as by local committees. Guidelines for prudent use of broad-spectrum and extended-spectrum antimicrobial agents in the clinical settings should be set from time to time. This can be achieved by antibiotic stewardship or regulation of antibiotic use keeping in mind the local resistance patterns.