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Letter to Editor

Microbiology Section

Multiple Antibiotic Resistance Patterns of the *Enterobacteriaceae* in the Untreated Municipal Sewage

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Dear Editor,

The increasing incidence of bacterial infection and gradual rise in resistance in *Enterobacteriaceae* against the available antibiotics, has highlighted the need to find more alternative antibacterial agents from other source [1]. Despite the pervasive incidence in many parts of the world of this family, surprisingly diminutive is recognized concerning the occurrence of such bacteria in raw sewage in India, although the work was done on hospital sewage [2]. The pathogen residing in the sewage wastes can migrate to ground and surface water and can contaminate them as well, in addition the increased and widespread use of antibiotics by human and animal have resulted in the increased antibiotic resistance in bacteria which can create havoc and can have a devastated impact on public health. The study was undertaken to assess the multiple antibiotic resistance pattern of the *Enterobacteriaceae* in the untreated municipal sewage.

An unprocessed sewage sample having 100ml volume was collected from municipal Wastewater Treatment Plant (WTP) located in Pholriwal village, Jalandhar city having capacity, 100 metric litters per day, from September 2014 to December 2014, every month, by composite sampling method described by Central Pollution Control Board (CPCB), India, in a sterile glass bottle [3]. Sewage was serially diluted then 0.1ml volume was spread onto MacConkey agar plate and incubated at 37°C for 24hours. Total 31 isolates belonging to 16 species of the genera Escherichia, Providencia, Proteus, Serratia, Yersinia, Kluyvera, Budivicia, Salmonella, Shigella and Hafnia were identified after biochemical screening using KB001 HiIMViC Biochemical test Kit by Himedia (indole, methyl red, Voges Proskauer's, citrate utilization, glucose, adonitol, arabinose, lactose, sorbitol, mannitol, rhamnose and sucrose). These were tested against 11 different antibiotics, namely amikacin (30µg), ampicillin (10µg), cefixime (5µg), ceftazidime (30µg), chloramphenicol (30µg), ciprofloxacin (5µg), co- trimoxazole (25µg), doxycycline hydrochloride (30µg), gentamycin (10µg), nalidixic acid (30µg) and tetracycline (30µg) by disc diffusion method [4]. The results were interpreted according to Clinical and Laboratory Standards Institute (CLSI) guidelines (2012) and Multiple Antibiotic Resistance (MAR) index of each individual isolate was also taken into account by method described by Krumperman 1983 [5,6]. All the isolates were resistant to at least two of the antibiotics, as shown in [Table/Fig-1], and the following resistant patterns of all the isolates against 11 antibiotics were found in percentage: Amp^{10} (93.54) > Caz^{30} (74.19) =Cfm^{5} (74.19) > Na^{30} (70.96) > Cip^{5} (41.93) > Te^{30} (38.70) = Do^{30} (38.70) > Cot^{25} (32.35) > Ak^{30} (22.58) > C^{30} (16.12) > Gen10 (12.90). In our study least resistance was observed against gentamycin, which is also in accordance with the previous study done by Silva group in Portugal [7]. In addition 10% resistance was found in the urban waste, Usha group in Indian hospital effluent was found to have 27.2% resistance [2,7]. Results showed and it can be assumed that such bacterial genera are more prone to other group of antibiotics instead of aminoglycosides group. With the increasing use of antibiotics we came across the MAR index which showed us that how the irrational use of antibiotics is not only degrading the particular environment but also making the pathogenic microbes resistant to it. Regular monitoring would be useful in managing the increasing antibiotic resistance in localities to protect public health. Concerned authorities should put major measures to make public aware regarding downfalls of irrational consumption of antibiotics, to protect our environment and for the well-being of humanity.

Enterobacteriaceae	Resistance Pattern	MAR Index
Escherichia coli	AmpCfmCot	0.27
Escherichia coli	NaTeGenAkCfm	0.45
Escherichia coli hermanii	AmpNa	0.18
Escherichia coli hermanii	AmpCipNaTeDoCfm	0.54
Escherichia coli*, inactive*	AmpCipNaTeDoCazCfm	0.63
Escherichia coli*, inactive*	AmpCipNaCazCfm	0.45
Escherichia coli*, inactive*	AmpNa	0.18
Escherichia coli*, inactive*	AmpNaTeDoCazCot	0.54
Escherichia coli*, inactive*	AmpCipNaTeDoCazGenCfmC	0.81
Escherichia coli*, inactive*	AmpCiPNaCazAkCfm	0.54
Providencia rustigianii	NaCazCfm	0.27
Providencia rustigianii	AmpNaCazCfmCot	0.45
Providencia rustigianii	AmpNaCazCfmCot	0.45
Proteus mirabilis	AmpCipNaCazAkCfmCot	0.63
Proteus mirabilis	AmpNaDoCazAkCfmCCot	0.72
Proteus mirabilis	AmpCipNaCazGenAkCfmC	0.72
Proteus mirabilis	AmpCazCfmCot	0.36
Serratia plymuthica	AmpNa	0.18
Serratia plymuthica	AmpCipNaCazCfm	0.45
Proteus vulgaris	AmpNaTeDoCazAk	0.54
Proteus vulgaris	AmpCipNaTeDoCazCfm	0.63
Yersinia frederiksenii	AmpCaz	0.18
Yersinia frederiksenii	AmpCipNaTeDoCfm	0.54
Proteus penneri	AmpNaTeDoCazCfmC	0.63
Kluyvera ascorbata	AmpCazCfmCot	0.36
Budivicia aquatica	AmpTeDoCazGenCfmCot	0.63
Escherichia vulveris	AmpCazCfm	0.27
Salmonella cholerasi	AmpCipCaz	0.27
Yersinia pseudotuberculosis	AmpCipCazAkCfmCCot	0.63
Shigella sonnei	AmpTeDoCazCfmCot	0.54
Hafnia alvei	AmpCipNaTeDo	0.45

[Table/Fig-1]: Multiple antibiotic resistance (MAR) index and resistance pattern of *Enterobacteriaceae* isolated from the raw sewage; AMP= Ampicillin; CFM= Cefixime; CAZ= Ceftazidime; C= Chloramphenicol; CIP= Ciprofloxacin; COT= Co- Trimoxazole; DO= Doxycycline Hydrochloride; GEN= Gentamicin; NA= Nalidixic acid; TE= Tetracycline

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