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MULTIPLE ANTIBIOTIC RESISTANCE INDEXING OF *PSEUDOMONAS AERUGINOSA* FROM DRINKING WATER

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ABSTRACT

Pseudomonas aeruginosa is a common organism found in water and its growth can cause problems with color, taste, odour and turbidity of the drinking water. In the present study, a total of 210 water samples were analysed and 40 strains of *Pseudomonas aeruginosa* were isolated and identified from various drinking water sources from different localities in Amravati city. Antimicrobial susceptibility tests were carried with 24 antibiotics, which showed 47% resistance, and 53% sensitive. The studies reported high incidence (30%) of *Pseudomonas aeruginosa* in drinking water showing danger to human health due to its biofilm producing ability and indicating emergence of antibiotic resistance due to the indiscriminate use of antibiotics.

INTRODUCTION

Pseudomonas aeruginosa is a common organism found in water and its growth can cause problems with color, taste, odour and turbidity of the drinking water. *Pseudomonas aeruginosa* is a typical biofilm-producing organism that grows on tubing, fittings and showerheads as well as in sinks and sink drains (Schoenen et al. 1985, Keevil et al. 1989). Contamination of *Pseudomonas aeruginosa* in drinking water resulted from the main water supply or through poor cleaning practices or due to sporadic or low flow rates in the units. The bacteria can attach to the internal pipe work surfaces and form a biofilm to protect them (Wende & Characklis 1990).

The occurrence of *Pseudomonas aeruginosa* in drinking water distribution systems indicates deterioration of the bacteriological quality of the drinking water (Armstrong et al. 1981, Michel et al. 1995, Mckeon et al. 1995). Kavitha & Priya (2005) isolated *Pseudomonas aeruginosa* from bottled and packed mineral water. Antibiotic resistance is an ever-increasing problem in *Pseudomonas aeruginosa* (Basustaoglu et al. 1995), which is normally associated with increased morbidity, mortality and costs. The appearance of multiresistant *Ps. aeruginosa* adds a new dimension to hospital outbreaks (Bert et al. 1998). Antibiotic-resistant bacteria and antibiotics discharged in various amounts in the environment, and indiscriminate use of antibiotics in medical, veterinary and agricultural practices leads to multiple antibiotic resistance in bacterial pathogens (Diab et al. 2002). The antibiotics and antibiotic resistant bacterial pathogens mix with ground water through the phenomenon of percolation and contaminate the ground water. The success of antimicrobial therapy in various waterborne diseases depends upon proper selection of the drugs. Penna et al. (2002), showed prevalence of *Ps. aeruginosa* (32.05%) in drinking and purified water during monitoring the water purification system. Sumitha et al. (2003) demonstrated the presence of *Ps. aeruginosa* in Bangalore water supply scheme.

Therefore, the study was aimed to evaluate the presence of *Pseudomonas aeruginosa* in drinking water available from various sources such as tube wells, open wells, and hotels and restaurants in Amravati city and determined their antibiotic resistance pattern/index for proper use of antibiotics.

MATERIALS AND METHODS

To study the antibiotic resistance of *Pseudomonas aeruginosa* in drinking water, the total 210 drinking water samples were collected from open wells (85), tube wells (85), and hotels and restaurants (40) from different localities of Amravati city by using sterilized plastic water sample collection bottles. All the water samples were examined by MPN test for its potability and *Ps. aeruginosa* were isolated from MPN positive test by using cetrimide agar and identified on the basis of standard test.

Antibacterial susceptibilities were determined according to the CLSI (formerly NCCLS). All bacteriological media and antibiotic discs and media were obtained from Hi-media Pvt. Ltd., Mumbai (Table 1). The MAR indices for an isolate and antibiotics were calculated by following formulae.

$$\text{The MAR index of isolate} = \frac{\text{Number of antibiotic resistant isolates}}{\text{Number of isolates}}$$

$$\text{The MAR index of antibiotics} = \frac{\text{Number of resistant antibiotics}}{\text{Number of antibiotics tested}}$$

RESULTS AND DISCUSSION

The primary aim of the study was to determine the occurrence of *Pseudomonas aeruginosa* in drinking water and its susceptibility to commonly used antibiotics. Forty species of *Pseudomonas aeruginosa* were isolated and identified (19 from open wells, 18 from tube wells and 3 from hotel and restaurants) from the water samples. Antimicrobial susceptibility tests were carried out with 24 antibiotics against 40 isolated species of *Ps. aeruginosa*, which showed 47% resistant and 53% sensitive to tested antibiotics (Table 2). The present studies reported a high incidence (30%) of *Pseudomonas aeruginosa* in drinking water indicating danger to human health due to its biofilm producing ability, which supported the finding of previous researchers (Mosso et al. 1994, Vachee et al. 1997, Sumitha et al. 2003).

Table 1: Antibiotics used in the study.

Antibiotics	Quantity (mcg/disc)	Antibiotics	Quantity (mcg/disc)	Antibiotics	Quantity (mcg/disc)
Amikacin	30	Ceftriaxone	30	Nalidixic acid	30
Ampicillin	10	Chloramphenicol	30	Nitrofurantoin	300
Anoxyclav	30	Ciprofloxacin	30	Novobiocin	30
Azithromycin	15	Gatifloxacin	5	Ofloxacin	5
Cefaclor	30	Gentamicin	30	Piperacillin	100
Cefepime	30	Imipenem	10	Streptomycin	10
Cefprozil	30	Levofloxacin	5	Tetracycline	30
Ceftazidime	30	Linezolid	30	Ticarcillin	7.5

Table 2: Contamination as per the source of drinking water and antibiotic susceptibility pattern of isolated *Ps. aeruginosa.*

Source	No. of Samples	No. of isolated	Resistance	Sensitivity	Total
Tube Wells	85	19	209	247	456
Open Wells	85	18	205	227	432
Hotels / Restaurants	40	3	33	39	72
Total	210	40	447	513	960

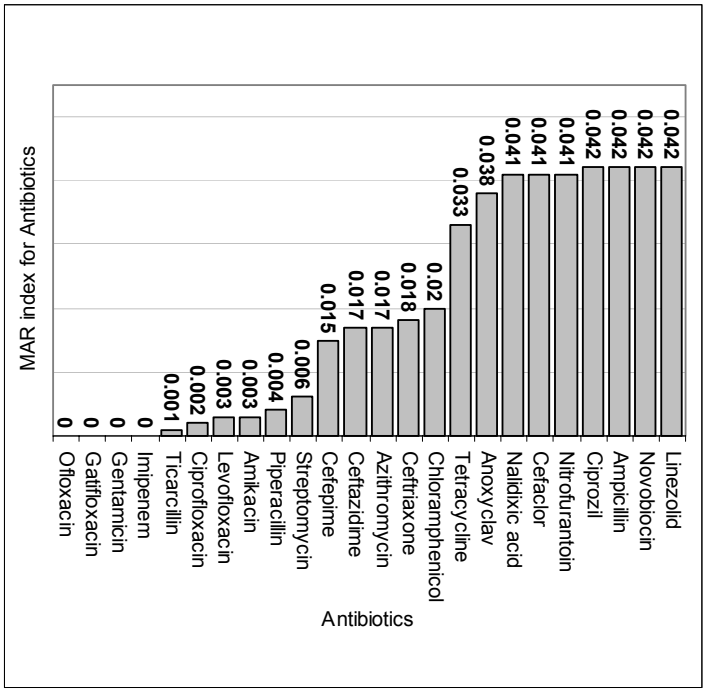

Fig. 1: MAR Index of antibiotics against *Ps. aeruginosa*.

All the isolated strains of *Pseudomonas aeruginosa* were highly sensitive (100%) to gatifloxacin, ofloxacin, gentamicin and Imipenem followed by ticarcillin (97.5%), ciprofloxacin (95%), levofloxacin (92.5%), amikacin (92.5%), piperacillin (90%), streptomycin (85%), cefepime (65%), ceftazidime (60%), azithromycin (60%), ceftriaxone (57.5%), chloramphenicol (52.5%) and highly sensitive to tetracycline (20%), anoxyclav (7.5%), nalidixic acid (2.5%), cefaclor (2.5%), nitrofurantoin (2.5%). These finding supported the observations of several previous studies (Begum et al. 2004, Tambekar et al. 2005). Comparatively high antibacterial sensitivity observed due to rare or occasional use of the drugs and could be attributed to the fact that these drugs were seldom used (Davis & Smith 1978).

While all strains of *Ps. aeruginosa* were resistance (100%) to ciprozil, ampicillin, novobiocin, linezolid followed by piperacillin (10%, MARI 0.004), amikacin (7.5%, MARI 0.003), levofloxacin (7.5%, MARI 0.003), ciprofloxacin (5%, MARI 0.002), ticarcillin (2.5%, MARI 0.001). The antibiotics gatifloxacin, ofloxacin, gentamicin, imipenem were highly sensitive indicating MAR index 0 (Fig. 1).

The present study showed low sensitivity to nalidixic acid which was also reported by Massa et al. (1995), Begum et al. (2004). The higher level resistance to these antibiotics might be attributed to antibiotic and antibiotic resistance bacterial emergence in drinking water sources because of improper and higher use of these common antibiotics. The variation occurred in multiple antibiotics resistance pattern among *Ps. aeruginosa* strains isolated from drinking water, indicated emergence of antibiotic and antibiotic resistance in *Ps. aeruginosa* due to the indiscriminate use of antibiotics.

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