

STUDIES ON THE ANTIBIOTIC RESISTANCE USING URINARY TRACT ISOLATES OF *E. COLI*.

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ABSTRACT- Urine samples collected from hospitalized male and female patients belonging to different age groups suffering from urinary tract infections were used in the present study. The samples were subjected to isolate the strains of *E.coli*. The isolates were found to be capable of producing biofilm and Extended Spectrum β -Lactamases (ESBLs) especially of CTX – M - type. Both male and female affected individuals exhibited high incidence of *E.coli* in the urine samples. The overall results indicated that the biofilm producing *E.coli* strains with β – lactamase activity had highest antibiotic resistance.

Key words: *E.coli*, Extended Spectrum β –Lactamases (ESBLs), CTX – M – type, biofilm, antibiotics

I. INTRODUCTION

A variety of antibiotics are available to treat the microbial ailments in man including all types of urinary tract infections (UTIs). Unfortunately, there has been a major worldwide increase of antibiotic – resistance in microbes especially in *E.coli*. Multiple antimicrobial resistance among gram negative organisms have been a long term and well recognized problem with urinary tract infections. In tract, the resistance is prevalent in many species of microbes including *Escherichia coli*, *Enterobacter*, *Klebsiella*, *Proteus*, *salmonella* and *pseudomonas*. It is shown that the isolates of *E.coli* from urinary tracts have increased resistance to many antibiotics^[1,2]. Extended Spectrum β – Lactamase (ESBLs) are the major cause for antibiotic resistance in enterobacters^[3].

ESBLs are generally plasmid - mediated bacterial enzymes which are capable of hydrolyzing a wide variety of antibiotics. In other words, the plasmid –mediated sources are the predominant causes for transferable resistance to third generation antibiotics in gram negative bacteria. CTX – M-type is a recently described family of broad spectrum β – Lactamase (CTX – Ms) and more than 70 types have been isolated so far. They have been divided in to 5 clusters based in amino acid sequence namely CTX – M -1, CTX – M -2, CTX – M – 8, CTX – M -9 and CTX – M -25. In last few years, it is established that the *E.coli* bacteria are the ESBL – carrying microbes in urinary tract infections (UTIs). Therefore, the present study has been made to isolate and characterize *E.coli* for CTX – M - type ESBLs in urine samples.

II. Materials and Methods

The hospitalized male and female patients at different age groups (10- 60 years) suffering from urinary tract infections in and around Namakkal town (Tamilnadu) were chosen to collect urine samples. A total of 73 urine samples were subjected to isolation of *E.coli* following the^[4, 5, 6]. The antibiotic stability tests were made on the isolates against 6

antibiotics namely Novobiocin, Bactricin, Vancomycin, Kanamycin, Erythromycin and Co – trimaxazole by standard Kirby – baner disk diffusion method. All the bacterial isolates were tested for the production of β – Lactamases by the method^[7]. The isolates were also evaluated for biofilm formation following the method^[8].

III. Results and Discussion

In the present investigation, *E.coli* are found to prevail in 35 urine samples out of 73 collected samples (47.94 %). Among the patients, females exhibit higher incidence of *E.coli* (50 %) when compared to males (45.16 %) (Table 1). The *E.coli* isolates on administration of antibiotics show highest resistance to Novobiocin and Vancomycin (100 %) followed by co – trimaxazole (94.28 %), Bactricin (88.57 %) and Erythromycin (82.82 %) (Table 2). From Table 3 and 4, it is evident that 80 % of *E.coli* from 35 isolates are biofilm producers in which 42.8 % are strong producers, 25.7% are moderate producers and 11.4 % are weak producers. In the total samples tested, 18 isolates (51.4 %) are found to be β – Lactamase producers and nearly 81.1 % isolates are slime producers (Table 4). Both male and female patients are found to have incidence of *E.coli* (45.16 % and 50.00 % respectively) in the urine samples^[9]. Out of 54 strains, nearly 50 % of gram negative bacteria possess resistance to one or more antibiotics^[10]. The antibiotic resistance in microorganisms is shown to be mainly due to biofilm formation. It is very difficult to treat microbes with biofilm production because the antimicrobial agents are ineffective to penetrate into the microbes via the biofilm. In addition, it is also found that the biofilm formation suppresses the phagocytic activity^[11].

In gram negative bacteria, the prevalence of high level of resistance to antibiotics could be due to β – Lactamase activity especially the plasmid – mediated ESBLs. A total of 34 strains among 93 *E.coli* isolates are found to carry ESBLs^[12]. CTX – M - is a recently described family of ESBLs and this is reported to prevail in many countries including India. Investigators^[13,14] have evidenced that the antibiotic resistance in *E.coli* is mainly due to the ESBL production, mostly CTX – M-type. The overall results of the present study reveals that most of the biofilm producing *E. coli* strains with β – Lactamase activity have antibiotic resistance. This is because these strains express several virulent factors and resistance against phagocytosis and other host defence mechanisms was observed by^[15]. The occurrence of more than one β – Lactamases within the same isolates^[16, 17].

Table 1. Occurrence of *E. coli* in Urine samples.

Nature	Percent occurrence		Total % occurrence
	Males	Females	
<i>E. coli</i> isolates	45.16	50.00	47.55
Biofilm – Producing <i>E. coli</i>	46.20	35.70	40.90

Table 2. Percent response of *E. coli* to different antibiotics.

No.	Antibiotics	Resistance	Intermediate	Sensitive
1	Novobiocin	100	-	-
2	Bacitracin	88.57	2.85	8.57
3	Vancomycin	100	-	-
4	Kanamycin	37.14	34.28	28.57
5	Erythromycin	82.85	17.14	-
6	Co-trimaxazole	94.28	2.85	2.85

Table 3. Nature of biofilm – producing isolates of *E. coli*.

Nature of biofilm production	% isolates
Strong	42.8
Moderate	25.7
Weak	11.4

Table 4. Biofilm production and β - Lactamase activity in isolated *E. coli*.

No.	Strain	Characterization	
		Slime test	β - Lactamase test
1	E - 2	S+	+
2	E - 4	S+	+
3	E - 6	M+	-
4	E - 9	M+	-
5	E - 11	S+	+
6	E - 21	S+	+
7	E - 23	M+	-
8	E - 24	-	+
9	E - 25	W+	-
10	E - 29	S+	+
11	E - 33	S+	+
12	E - 34	M+	-
13	E - 35	S+	+
14	E - 36	-	-
15	E - 37	S+	+
16	E - 39	W+	-
17	E - 40	-	-
18	E - 41	M+	+
19	E - 42	S+	+
20	E - 44	M+	+
21	E - 45	S+	+
22	E - 46	S+	-
23	E - 48	W+	-
24	E - 49	S+	+
25	E - 50	S+	+
26	E - 51	M+	-
27	E - 52	S+	+
28	E - 53	S+	+
29	E - 62	-	-
30	E - 64	M+	-
31	E - 66	W+	-
32	E - 67	-	-
33	E - 68	M+	+
34	E - 71	-	-
35	E - 73	-	-

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