

# Study on Study on Pathogenic *Escherichia coli* in a slum area of Khulna, Bangladesh

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Key Words: *Pathogenic E.coli*, *Khulna*, *Bangladesh*, *Slum*, *Sanitation*, *Next-Generation Sequencer*

## 1. Background and Objectives

*E.coli* has been widely used as a fecal pollution indicator as they do not usually exist outside fecal matter and are also non-pathogenic. It is important to understand the mechanism of pathogenic *E.coli* contamination as it poses health risks, like cholera, acute gastroenteritis. The survey area, Khulna, Bangladesh, has huge number of slums with no access to clean water and basic sanitation. The objectives of this study are to (1) understand and quantitatively analyze fecal bacteria contamination in the area; (2) elucidate the risk pathways of all species of bacteria present in the area; and (3) to understand and quantitatively analyze the contamination of several pathogenic bacteria in the area.

## 2. Materials and Methods

The surveys and experiments were conducted in a slum area of Khulna, Bangladesh, from Aug. to Oct. in 2014. *E.coli* and *E.coli* O157 in the area were measured: the samples of stored water in house, tube well, soil, pond, hand, dishes and glasses, and vegetables were analyzed by membrane filter method; the samples of excreta were analyzed by plate smear method. To identify the bacteria present in the samples, community analyses were conducted by sequence determination of 16S rRNA genes. For the stored water and fecal samples, colonies were isolated using XM-G and CHROMagar media. About eight kinds of Pathogenic *E.coli* were identified using PCR and Next-generation sequencer. Moreover, origin estimation was conducted to confirm the origin of isolated *E.coli*.

## 3. Results and Discussion

*E. coli* was detected in the sample of stored water in 17 out of 18 houses in the study area. (Maximum of 100 CFU/100 mL) *E. coli* was not detected in the sample of drinking tube well water (0/4 points) but *E. coli* was detected in the sample of non-drinking tube well water (2/4 points). Results of community analyses of 16S rRNA showed that the exposure pathways of the bacteria are different and even if they have the same pathway, they would have different genes. Nine out of 10 samples (9/10) of stored water in house, 6/6 of tube well water, 3/4 of soil, 2/2 of pond, 8/8 of hand had pathogenic *E.coli* genes included in the top 10 genes present in them. The results of identification of eight kinds of Pathogenic *E.coli* are shown in **Fig. 1**. Results showed the risks for humans were different because each sample had different genes of Pathogenic *E.coli*. The results of origin estimation implied that stored water in house was contaminated by human excreta because the 7.8% of genes of in the samples came from humans.

## 4. Conclusion

*E. coli* was detected in the sample of surrounding environment in the slum area and stored water in houses. The water in the storage drums might have been contaminated from the way they were stored. The 16S rRNA genes and community structure of bacteria in the environmental samples were shown in this study. It was found out that *Acinetobacter* genes (including the pathogenic ones) are superior. Pathogenic *E.coli* species were also surveyed and analyzed.

Fig.1 The percentage of Pathogenic *E.coli*

Sample	the number of colonies	Pathogenic <i>E.coli</i>	detected number	percentage (%)	Concentration of <i>E.coli</i> (cfu/100mL)	Concentration of Pathogenic <i>E. coli</i> (cfu/100mL)
Tube Well Water (XM-G agar media)	357	EAEC	1	0.28	17.10	$4.8 \times 10^{-2}$
		EIEC	1	0.28		$4.8 \times 10^{-2}$
		ETEC	3	0.84		$1.4 \times 10^{-1}$
		ExPEC	3	0.84		$1.4 \times 10^{-1}$
		EPEC	4	1.12		$1.9 \times 10^{-1}$
excreta (XM-G agar media)	264	EAEC	2	0.76	$3.54 \times 10^8$	$2.7 \times 10^6$
		ExPEC	3	1.14		$4.0 \times 10^6$
		ETEC	55	20.83		$7.4 \times 10^7$
excreta (CHROMagar O-157)	96	EIEC	1	1.04	$8.00 \times 10^7$	$8.3 \times 10^5$
		ExPEC	1	1.04		$8.3 \times 10^5$
		ETEC	36	37.50		$3.0 \times 10^7$