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Prevalence of Faecal Carriage of Multidrug Resistance Among Commensal Escherichia Coli and Klebsiellapneumoniaecolonising the Gut of Healthy Adult Population in Tumkur

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ABSTRACT

The emergence of resistance in the normal flora is a major concern in the management of both hospital and community acquired infections. The prevalence of resistant organisms in the community is on the rise. Multidrug resistant (MDR) colonizers act as a reservoir for transmission of antibiotic resistance and are the source of infection. Irrational use of antibiotics is probably a major contributor to rising antibiotic resistance. This study was conducted to find the faecal prevalence of antibiotic resistance among commensal Escherichia coli and Klebsiellapneumoniaein the community. 860 healthy adults were randomly selected from the urban practice area of Sri Siddhartha Medical College, Tumkur. Stool samples were collected from the participants and plated on MacConkey agar plates and further processed according to standard laboratory guidelines for the isolation and identification of Escherichia coli and Klebsiellapneumoniaein. All the isolates were screened for multidrug resistance to commonly used antibiotics by standard methods. Among the total of 860 samples, 64% has shown antibiotic resistance. Maximum resistance was seen for Fluoroquinolones 51% followed by Cephalosporins 48% and Tetracycline 47%.The prevalence of MDR colonizers in the community were found to be 54%.

INTRODUCTION

One of the biggest public health problems at present is Antibiotic resistance^[1]. The prevalence of resistant bacteria in the community is increasing globally. It is known that antibiotic pressure is an established cause of propagation of resistance and inappropriate use of antibiotics has transformed the healthy intestinal gut flora of human into a reservoir of antibiotics resistant microorganisms also known as the gut resistome. This selective pressure has driven adverse effect on the gut microbiome, facilitates these organisms to behave as opportunistic pathogens^[2]. *Escherichia coli* and *Klebsiella pneumoniae* are the most common among the Enterobacteriaceae family and has been shown to be the main carrier of drug resistance genes. *Escherichia coli* and *Klebsiella pneumoniae*, are the important part of fecal flora, are associated with varying human infections and has been reported frequently in the community outbreaks^[3]. Drug resistant genes usually detected in clinical isolates are now seen in the gut flora of healthy individuals at the community level. Most of the data on faecal carriage are from hospitalized patients, who are under antibiotic pressure. There is limited information about the carriage and spread of resistant strains in the community. This study was undertaken to determine the prevalence of MDR strains in the community.

MATERIAL AND METHODS

Study Subjects: A total of 860 healthy individuals of both the sexes aged 18-60 residing in the urban practice area of Sri Siddhartha Medical College, Tumkur were included in the study. This was a prospective cross sectional study conducted from January 2021 to December 2022 in the Department of Microbiology, Sri Siddhartha Medical College, Tumkur. Ethical clearance was obtained from the institutional ethical committee. Individuals with any condition like diabetes mellitus, pregnancy, immunosuppressive disorders, history of recent antibiotic consumption (3months) and history of hospitalization in the past one year were excluded from the study.

Sample Collection and Processing: Participants were provided with a sterile leak proof plastic container to collect the stool sample. Samples were transported on the same day to the laboratory and inoculated into MacConkey agar plates and incubated at 37°C for 24 hours^[11].

Identification of Pathogen: Colony morphology and Gram staining were used to identify the bacteria initially and subjected to catalase, oxidase, indole, urease, citrate, TSI, motility tests for biochemical characterization^[11].

Antibiotic Sensitivity Test: It was done on Muller Hinton agar plates by Kirby-Bauer's disc diffusion method as per CLSI guidelines. Antibiotics namely Ampicillin (10 µg), Amoxicillin/clavulanic acid (20/10 µg), Piperacillin/Tazobactam (100/10 µg), Ciprofloxacin (5 µg), Tetracycline (30 µg), Gentamicin (10 µg), Cotrimoxazole (1.25/23.75 µg), Cefoxitin (30 µg), Ceftazidime (30 µg), Ceftriaxone (30 µg), Gentamicin (30 µg), Imipenem (10 µg) were used with *Escherichia coli* ATCC 25922 and *Klebsiella* ATCC 13883 as a controls. The results were interpreted as sensitive/resistant according to CLSI guidelines^[12].

RESULTS AND DISCUSSIONS

Age and Sex Distribution: Among the 860 participants, majority of the study population falls in the age of 41-60 with a female predominance of 52% (Table 1) (Fig. 1). The male: female ratio among the study sample was 1:91:1. There was female predominance 52% among the isolates obtained from the study population. The maximum number of isolates were from the individuals in the age group of 41-60yrs. 35.9% followed by 21-40 yrs. 30.5%. Identification of organisms: Among the 860 samples, based on colony morphology and biochemical tests, it has been found

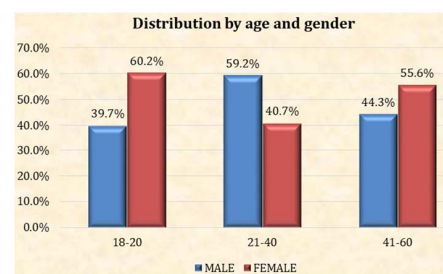


Fig 1: Gender and age wise distribution of study population

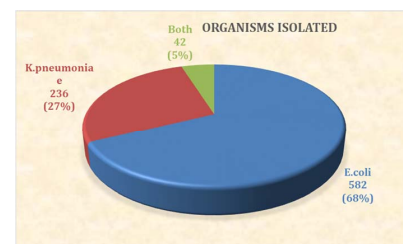


Fig 2: Organisms isolated

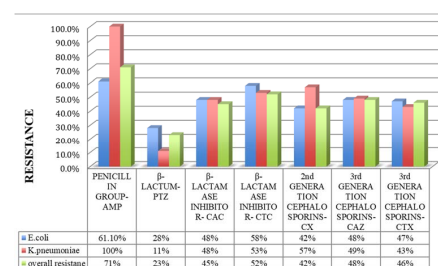


Fig 3: Resistance pattern of *Escherichia coli* and *Klebsiella pneumoniae*

Table 1: Gender and age wise distribution of study population

Age in years	No. of participants		Total	Percentage
	Male (%)	Female (%)		
18-20	109(39.7)	165(60.2)	274	31.8
21-40	164(59.2)	113(40.7)	277	32.2
41-60	137(44.3)	172(55.6)	309	35.9
Total	410(48)	450(52)	860	100

Table 2: Organisms isolated

Organisms	Frequency (N = 860)	Percentage
Escherichia coli	582	67.6
Klebsiellapneumoniae	236	27.4
Both	42	4.8

Table 3: Showing the proportion of Escherichia coli and Klebsiella pneumoniae resistant to drugs isolated from healthy individuals

Antimicrobial categories	Drugsug/disc	Breakpoints (mm)			Healthy stool	
		S	I	R	E.coli (%) n = 582	K.pneumoniae(%)n= 278
Penicillin group	AMP (10)	≥17	14	≤13	61.1	100
Piperacillin/Tazobactam	PTZ(100/10)	≥21	16	≤17	28	11
β-lactamase inhibitors	CAC(30/10)	-	18	-	48	48S
CTC(30/10)		-	20		58	48
2ndgeneration Cephalosporins	CX (30)	≥22	-	≤21	42	57S3
3rdgeneration Cephalosporins	CAZ(30)	≥21	18-20	≤17	48	49
CTX(30)	CTX(30)	≥26	23-25	≤22	47	43
Aminoglycosides	GEN (10)	≥15	13-14	≤12	07	04
Fluoroquinolones	CIP (5)	≥21	16-20	≤15	51	49
Carbapenems	IPM (10)	≥23	20-22	≤19	05	13
Tetracycline	TE (30)	≥15	12-14	≤11	47	50
Sulphonamides	COT(1.25/3.78)	≥16	11-15	≤10	48	41

that majority of the samples had shown the presence of *Escherichia coli* (67.6%), *Klebsiellapneumoniae* was isolated from 27.4% of the study population and a combination of both were seen in 8% of the study participants (Table 2) (Fig. 2). *Escherichia coli* (67.6%) is the predominant organism isolated in this study followed by *Klebsiellapneumoniae* (27.4). Antibiotic resistance profile of the isolates from the study participants: Among the 860 isolates, over all resistance was observed in (64%) individuals and faecal carriage rate of MDR colonizers were found to be (54%). Maximum resistance was seen for Fluoroquinolones (51%) followed by Cephalosporins (48%) and Tetracycline (47%) (Table 2) (Fig. 2).

Antibiotic resistance is a looming threat with serious implications on successful treatment of bacterial infections^[1]. In hospitals and also in the community, prevalence of MDR gram negative bacilli especially *Escherichia coli* and *Klebsiella pneumoniae* emerged drastically during the past decade^[2]. The rate of faecal carriage of MDR colonizers among the healthy individuals is on rise. Transmission of these MDR strains in the community leads to community outbreaks^[3]. This study was focused on the faecal carriage of MDR organisms in the community. The results of the present study showed that 54% of isolates were MDR. A study conducted by Antony *et al.* among healthy adults in Pondicherry, South India, 50.1% of MDR isolates were reported^[1]. A study in healthy pregnant women attending urban community centre and rural primary health centers of Vellore (2004) by Luvsansharav *et al.*, showed the prevalence of MDR was 8.4%^[4].

The study done in Ujjain(2011) by Rashid *et al.*, in children aged 3-16 years in the community of Palwa, showed a prevalence rate of 33%^[5]. The study from Vellore had low MDR carriage as the study population were pregnant women who were young and exposure to antibiotics would be presumed to be low. While the study from Pondicherry, South India reported similar carriage rates. In a rural based study from Central India by Shakya *et al.*, children aged 1-3 year. showed the prevalence rate of 70%^[6]. In their study they have also included environmental sampling which included animals, drinking water, common source and waste water and showed resistance rate of 29%, 41%, 30% and 30% respectively^[6]. In our study the source and factors for acquisition of resistance were not studied, since the community studied was congested possible factors for acquisition of resistance could be contamination of household surfaces, food and water due to overcrowding and poor sanitation. In our study maximum resistance was seen for Fluoroquinolones 51% followed by Cephalosporins 48% and Tetracycline 47%. In a study by Gupta *et al.*, from Chandigarh, India in (2020) reported maximum resistance for Cephalosporins 60.4%, followed by Fluoroquinolones 41.5%^[9]. one reason could be the irrational use of these antibiotics in the community, self-prescription, over the counter purchase of drugs, lack of knowledge about rational use of drugs^[7].

CONCLUSION

This study revealed a high faecal carriage rate of MDR organisms in healthy individuals which is

presumed not to be under apparent antibiotic pressure. Detection of commensal strains carrying resistant genes is worrisome. This has a potential of dissemination and also the possibility of getting expressed in the presence of antibiotic pressure. The outcome of spreading of resistant genes will adversely affect the treatment strategy in community acquired infections. Further studies need to be undertaken with environmental sampling and correlation of prescription pattern in the area to determine the reservoirs and vehicles to correlate the cause for dissemination of resistant genes within the community. As a control measure, active surveillance is important to find the true magnitude of gut resistant colonizers in the community, creating awareness on the use and misuse of antibiotics in the community, awareness programed on hygienic practices in the community has to be practiced regularly to stop the dissemination of resistance gene in the community.

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