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Research Article

**ANTIMICROBIAL & SYNERGISTIC ACTIVITY OF  
ANTIBIOTICS AGAINST COMMON CLINICAL ISOLATES.**Zehra Ashraf<sup>1</sup>, Mirza Tasawer Baig<sup>2</sup>, Waseem Abbas<sup>3\*</sup>, Ifrah Athar<sup>1</sup>, Nimra Waheed<sup>1</sup><sup>1</sup>Faculty of Pharmacy, Jinnah University for women, Karachi, Pakistan<sup>2</sup>Faculty of Pharmacy Ziauddin University Karachi<sup>3</sup>Department of Pharmacy Shaheed Mohtarma Benazir Bhutto Medical University Larkana

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**Abstract:**

**Object:** This experiment was carried out to check the anti-bacterial & synergistic effect of different antibiotics on gram negative (*E-coli*) & gram positive (*S-aureus*) bacteria.

**Method:** Basically, the whole experiment was carried out in two parts, 1<sup>st</sup> by looking for the individual effect of anti-bacterial drugs and 2<sup>nd</sup> by evaluating the synergistic effect taking Amoxicillin (50µg) as standard. Hundred bacterial samples taken from the laboratory were collected from different sites of infected person. The experiment was carried out by disc diffusion method & by considering CLSI standards to check the resistance result. **Result:** We have observed that the male patients are more prone to infections caused by *E-coli*. The greater occurrence of *E. coli* & *S.aureus* was obtained from Respiratory Tract. *E. coli* & *S.aureus* showed 100 % resistance against Cefotaxime. In *S.aureus* isolates the most effective therapy was Amoxicillin-Ciprofloxacin & in *E.coli* isolates best effective therapy was Amoxicillin-Gentamycin.

**Key words:** Anti-bacterial activity, Disc Diffusion Method, Synergy, Zones.

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**INTRODUCTION:**

Nowadays lethal infections are causing by the human original flora. (1). The bacteria *E-coli* normally living in human intestine but its certain types are causing intestinal infection which are characterized by bloody diarrhea, abdominal pain, fever & dehydration, even some time kidney failure. Pregnant women, People with weak immunity, weak children are more prone to such infections. (2). *S.aureus* is also a part of human flora present in Respiratory tract, skin & nose, but can cause infections like sinusitis, respiratory infections, and abscesses. Methicillin-Resistant *S.aureus* (MRSA) is still the specie which is the worldwide problem for clinical medicine and even after so many researches there is no authentic and approved vaccine for *S. aureus*.(3). There are many facts which are making bacteria resistant to antibiotics, one of the factor is improper dose taking which leads to activation of inhibitory enzyme activation against the drugs other factors include desensitization and increased tolerance level of bacteria by their genetic mutations or changes.(4,5)There are so many antibiotics regimen which give us the helpful result for providing better treatment and also helps us by decreasing the treatment failure rate (6).So many antibiotics in double, triple or even in multiple combinations were being tested through disc diffusion method and the results were quiet amazing (7, 8).

**MATERIALS AND METHODS:**

Since April 12th 2017 till the March 12th 2018 around 100 samples of infectious patients were being collected and given to the laboratory, half of which (50) were of *E-coli* and other half (50) were of *S.aureus*. The identification and confirmation of the isolates were done by differential techniques & convectional biochemical methods. (9) The susceptibility of *E-coli* & *S.aureus* isolates pattern were obtained by Disc diffusion method in which the streaking of the culture through wire loops were done on the petri plate containing Mueller Hinton Agar. For the checking of synergistic activity the discs of antibiotics i.e. Amoxicillin (Amx) 10µg, Ciprofloxacin (Cip) 5µg, Cefoxitin (Fox) 30µg& Gentamicin (Gen) 120µg were placed in the petri plate. Considering Amoxicillin as the standard, each of 3 antibiotics disc were then tested by placing them in the incubator for 4-6hrs at 37 0C. For the antimicrobial activity the discs were placed individually after the streaking of sample then placed in the incubator for 24 hrs at 37 0C. The CLSI was kept as the standard for the evaluation of result and also for the checking of zone of inhibition and synergistic zone visualization. The procedure was done for the 50 samples of *E-coli* & 50 samples of *S. aureus*. (10).

**Table 1. Distribution of *E-coli* in different sites of infections (n=50)**

Site of infection	For <i>E-coli</i> Male n=33(66%)	Female n=17 (34%)
Respiratory Tract	20	12
Blood	04	02
Urine	08	03
Pus	01	00
Total	33 (66 %)	17 (34%)

**Table 2. Distribution of *S.aureus* in different sites of infections (n=50)**

Site of infection	For <i>S.aureus</i> Male n=30(66%)	For <i>S.aureus</i> Female n=20 (40%)
Respiratory Tract	10	09
Blood	04	02
Urine	07	04
Pus	09	05
Total	30 (60 %)	20 (40%)

**Table 2.Age & gender specific distribution among patients**

Age Group (years)	For <i>E-coli</i> Male n=33(66%)	Female n=17 (34%)
00-25	16	10
26-50	08	03
51-75	03	02
76-100	06	07

**Table 3. Age & gender specific distribution among patients**

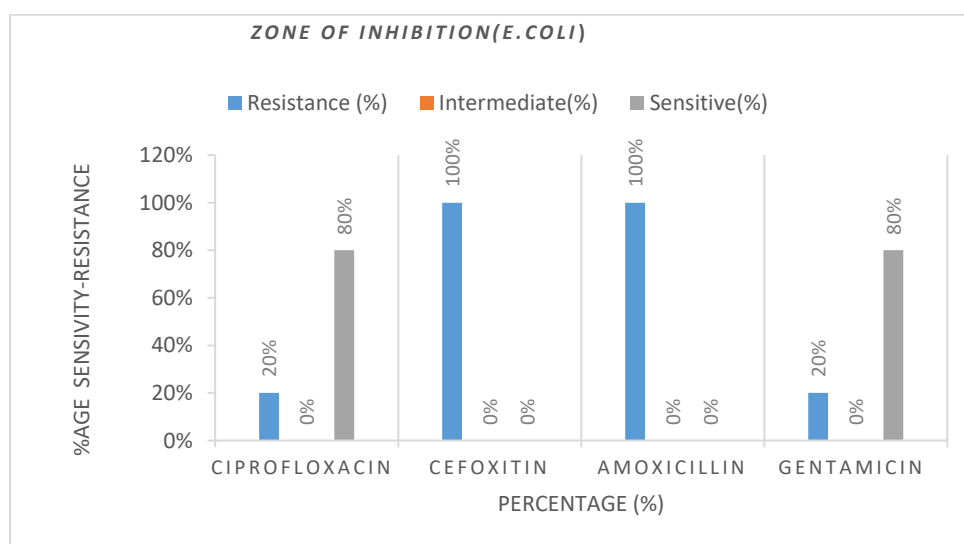
Age Group (years)	For <i>S.aureus</i> Male n=30(66%)	For <i>S.aureus</i> Female n=20 (40%)
00-25	08	04
26-50	08	05
51-75	06	06
76-100	08	05

**Table 4. . Zone diameter interpretive standards for *E.coli* CLSI standards table of antibiotics for *E. coli*.**

Antibiotic	Disc Content	Zone of inhibition (For <i>E.coli</i> )		
		Resistance	Intermediate	Sensitive
Ciprofloxacin	5 µg	≤15	16-20	≥ 21
Cefoxitin	30 µg	≤21	15-17	≥22
Amoxicillin	10 µg	≤ 19	14-17	≥ 20
Gentamycin	10 µg	< 12	13-14	≥15

**Table 5. Zone diameter interpretive standards for *S.aureus* CLSI standards table of antibiotics for *S.aureus***

Antibiotic	Disc Content	Zone of inhibition (For <i>E.coli</i> )		
		Resistance	Intermediate	Sensitive
Ciprofloxacin	5 µg	≤15	16-20	≥ 21
Cefoxitin	30 µg	≤21	15-17	≥22
Amoxicillin	10 µg	≤ 19	14-17	≥ 20
Gentamycin	10 µg	< 12	13-14	≥15

**Figure 1. Sensitivity pattern of *E.coli* against four broad spectrum antibiotics**

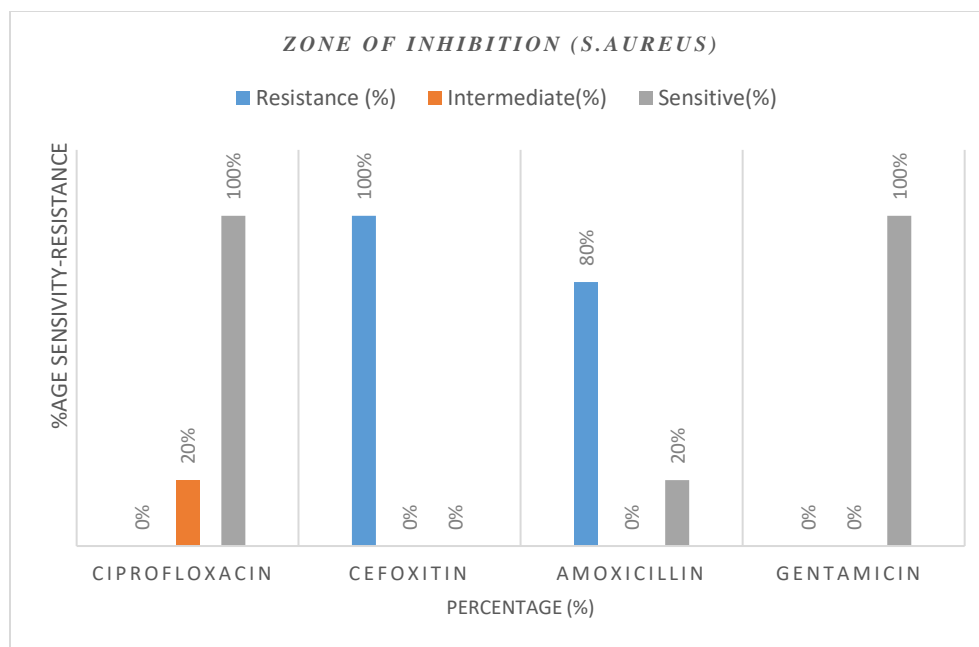


Figure 2.Sensitivity pattern of *S.aureus* against four broad spectrum antibiotics

Table 6.Double disc synergy results for different antimicrobial combinations checked against *E.coli* & *S.aureus* isolated

<i>For E.coli</i>				
S.NO.	Drug combination	Synergy n (%)	Indifferent n (%)	Antagonism n (%)
1	AMOXICILLIN-CEFOXITINE	60%	40%	00
2	AMOXICILLIN-CIPROFLOXACIN	80%	10%	10%
3	AMOXICILLIN-GENTAMYCIN	100%	00	00
<i>For S.aureus</i>				
1	AMOXICILLIN-CEFOXITINE	70%	20%	10%
2	AMOXICILLIN-CIPROFLOXACIN	100%	00	00
3	AMOXICILLIN-GENTAMYCIN	85%	15%	00

### RESULTS:

It was marked that male patients were found to be affected by infection of *E.coli*. The greater occurrence of *E.coli* & *S.aureus* was obtained from Respiratory Tract shown in table 1 & 2. *E.coli* showed 100 % resistance against Cefotaxine and Amoxicillin shown in fig 1. *S.aureus* also showed 100% resistance against Cefotaxine represented in fig 2. In *S.aureus* isolates the most effective therapy was Amoxicillin-Ciprofloxacin which indicated 100% synergistic effect and in *E.coli*

isolates best effective therapy was Amoxicillin-Gentamycin showed 100% synergistic effect as indicated in table 7.

### DISCUSSION:

By the above collected samples we have seen the antimicrobial effects of different antibiotic on the different samples of *E.coli* and *S.aureus*. The most effected drug we have concluded was Cip although in one case Gen was also effective. The synergistic effect

on the *E-coli* sample resulted in the marked effect of Amx-Gen with greater synergistic zones & in some cases Amx-Cip also showed effective zone of inhibition. Through the different samples of *S-aureus* the results of antimicrobial drugs showed the best activity of Cip then Gen. The synergistic effect on the samples of *S-aureus* turns out very effective when Amx-Cip were given together.

#### CONCLUSION:

Through this whole experiment conducted on both gram -ve (*E-coli*) & gram +ve (*S.aureus*) organisms we have concluded that day by day bacteria becoming resistant to the drugs, we have been using these drugs for so long but when the drugs were given in combination we have seen a better result and the efficacy of one or both antibiotics have enhanced. The overall result was increased in inhibiting the growth of microorganisms so antibiotics in combination therapy should be given.

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