

**Research**

## **Resistantce Pattern of Clinical Isolates from Cases of Urinary tract Infection against Gatifloxacin**

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### **Abstract**

A urinary tract contamination (UTI) is a disease that influences some part of the urinary tract. When it influences the lower urinary tract it is known as a bladder contamination (cystitis) and when it influences the upper urinary tract it is known as kidney contamination (pyelonephritis). Gatifloxacin is an antibiotic of the fourth generation fluoroquinolone family that like different individuals from that family hinders the bacterial compounds DNA gyrase and topoisomerase IV. The aim of objective of this study to determine the resistant paten of gatifloxacin against UTI pathogens

One hundred thirty five clinical isolates of *Staphylococcus aureus*, *Escherichia coli*, and *Pseudomonas aeruginosa* were collected from different hospitals and labs. These isolates were evaluated against the Gatifloxacin to investigate minimum inhibitory concentration and the resistant pattern. The antibacterial activity of Gatifloxacin was carried out by disc diffusion method.

Gatifloxacin was found to be 49.44%, 21.66% and 37.77% sensitive to *Staphylococcus aureus*, *Escherichia coli* and *Pseudomonas aeruginosa* respectively and shows 50.55%, 78.33% and 62.22% resistance to clinical isolates.

Minimum inhibitory concentration of Gatifloxacin against *Staphylococcus aureus*, *Escherichia coli* and *Pseudomonas aeruginosa* was found to be 0.5 mg/lit.

Hence it has been evaluated that all these clinical isolates have developed resistant to Gatifloxacin, though it is a new fluoroquinolone in our clinical settings but the clinical isolates had developed resistant to different antimicrobial which is very alarming. So fluoroquinolone should not be prescribed unless alternates are available.

### **Introduction**

Antibiotic resistance obstruction is the capacity of a microorganism to withstand the impacts of anti-infection agents. It is a particular kind of medication opposition. Antibiotic resistance obstruction develops normally by means of regular determination following up on irregular transformation, yet it can likewise be designed by applying a developmental weight on a populace. When such a quality is created, microscopic organisms would then be able to exchange the hereditary data in a flat design (between people) by plasmid trade. On the off chance that a bacterium conveys a few opposition qualities, it is called **multiresistant** or, casually, a superbug. The term antimicrobial obstruction is some of the time used to unequivocally include life forms other than microscopic organisms.

### **Mechanism of Antibiotic Resistance**

The four primary mechanisms by which microorganisms show protection from antimicrobials are:

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- 1 Drug inactivation or alteration: e.g. enzymatic deactivation of Penicillin G in some penicillin-safe microscopic organisms through the creation of  $\beta$ -lactamases.
2. Alteration of target site: e.g. change of PBP—the coupling target site of penicillins—in MRSA and other penicillin-safe microscopic organisms.
3. Alteration of metabolic pathway: e.g. some sulfonamide-safe microscopic organisms don't require para-aminobenzoic corrosive (PABA), a critical forerunner for the union of folic corrosive and nucleic acids in microorganisms hindered by sulfonamides. Rather, similar to mammalian cells, they swing to using preformed folic corrosive.
4. Reduced medication amassing: by diminishing medication porousness or potentially expanding dynamic efflux (drawing out) of the medications over the cell surface.

A urinary tract disease (UTI) is a bacterial contamination that influences any piece of the urinary tract. Despite the fact that pee contains an assortment of liquids, salts, and waste items, it more often than not does not have microscopic organisms in it when microorganisms get into the bladder or kidney and increase in the pee, they cause an UTI. The most widely recognized kind of UTI is a bladder disease which is additionally regularly called cystitis. Another sort of UTI is a kidney disease, known as pyelonephritis, and is considerably more genuine. In spite of the fact that they cause distress, urinary tract diseases can more often than not be rapidly and effectively treated with a short course of antibiotics. Studies have demonstrated that breastfeeding can diminish the danger of UTI's in newborn children.

Gatifloxacin is an anti-microbial of the fourth generation fluoroquinolone family that like different individuals from that family hinders the bacterial proteins DNA gyrase and topoisomerase IV. Bristol-Myers presented Gatifloxacin in 1999 under the exclusive name Tequin for the treatment of respiratory tract contaminations, having authorized the drug from Kyorin Pharmaceutical Company of Japan. Allergan produces an eye-drop detailing called Zymar. Gatifloxacin is accessible as tablets and in different watery answers for intravenous treatment.

The aim for the present investigation was to assess the base inhibitory fixation and safe example of urinary tract contamination against Gatifloxacin

## Method

### Preparation of Broth

Take Tryptic soy broth 7.5gm in a flask pour in the in 250 ml distilled water. Take 2ml broth in small test tube, tube cove it with cotton plug and after that autoclave it at 121°C for 30 minute.

### Preparation of Stock Solution

The minimum inhibitory concentration (MIC) of gatifloxacin is found from the writing overview is 0.25mg/lit.

## Calculation

### Method

1. Take 1000ml volumetric flask and rinse it with distilled water thrice a time.
2. Take butter paper and weight accurately 1 gm of Gatifloxacin by taking the tare of butter paper initially on electrical balance.
3. Take this powder and mix it with small amount of distilled water in volumetric flask with gentle shaking.
4. When homogeneity and clear solution is obtained than make up the volume with distilled water and cover the volumetric flask with its lid and make different dilution with this stock solution.

### Preparation of Dilution

The MIC of Gatifloxacin which is found from literature survey is 0.25mg/lit at 100% potency.

### Calculation of Different Dilutions

1<sup>st</sup> dilution:  $0.00025 \times 1 = 0.00025\text{mg/ml}$

2<sup>nd</sup> dilution:  $0.00025 \times 2 = 0.00050\text{mg/ml}$

3<sup>rd</sup> dilution:  $0.00025 \times 3 = 0.00075\text{mg/ml}$

4<sup>th</sup> dilution:  $0.00025 \times 4 = 0.001\text{mg/ml}$

5<sup>th</sup> dilution:  $0.00025 \times 5 = 0.00125\text{mg/ml}$

### Preparation of Different Concentration

- Take 2.5ml of stock solution in 100ml volumetric flask and make up the volume with distilled water.
- Take 1 ml from dilution 1 into 100ml volumetric flask and make up the volume with distilled water.

$$\text{Stock} \times \frac{2.5}{100} \times \frac{1}{100} = 0.00025 \text{ mg/ml}$$

### 2<sup>nd</sup> Concentration

- Take 5ml of stock solution in 100ml volumetric flask and make up the volume with distilled water.
- Take 1 ml from dilution 1 into 100ml volumetric flask and make up the volume with distilled water

$$\text{Stock} \times \frac{5}{100} \times \frac{1}{100} = 0.00050 \text{ mg/ml}$$

### 3<sup>rd</sup> Concentration

- Take 7.5ml of stock solution in 100ml volumetric flask and make up the volume with distilled water.
- Take 1 ml from dilution 1 into 100ml volumetric flask and make up the volume with distilled water

$$\text{Stock} \times \frac{7.5}{100} \times \frac{1}{100} = 0.00075 \text{ mg/ml}$$

### 4<sup>th</sup> Concentration

- Take 10ml of stock solution in 100ml volumetric flask and make up the volume with distilled water.
- Take 1 ml from dilution 1 into 100ml volumetric flask and make up the volume with distilled water

$$\text{Stock} \times \frac{10}{100} \times \frac{1}{100} = 0.001 \text{ mg/ml}$$

### 5<sup>th</sup> Concentration

- Take 12.5ml of stock solution in 100ml volumetric flask and make up the volume with distilled water.

- Take 1 ml from dilution 1 into 100ml volumetric flask and make up the volume with distilled water

$$\text{Stock} \times \frac{12.5}{100} \times \frac{1}{100} = 0.00125 \text{ mg/ml}$$

### Preparation of Disc

Take specific filter paper (disc paper) or, in which is especially used for antibiotic disc and with the assistance of punch machine, cut the circles. In the wake of cutting of circle, absorbed these plates arrangements arranged from stock. Dispose of the overabundance arrangement from Petri dish and enable it to dry at room temperature and were utilized when totally dried.

### Preparation of Inoculums

The inoculums were prepared by picking of the colonies via sterile wire loop of the isolates and suspending in tube containing 2-3ml of broth. Whole experiments perform via sterile environment. These experiments perform with each isolates (i.e. *Staphylococcus aureus*, *Escherichia. coli* and *Pseudomonas aeruginosa*). After the perform experiments all inoculum incubate in incubator at 37°C for few hours until turbidity reaches McFarland 0.5.

### Preparation of Media Plates

Muller Hinton Agar was arranged and cleaned as sterilized pure his media into sterile Petri dish around 20-25ml for each plate. Care must be taken to pour on the plates on a level surface with the goal that the profundity of medium is uniform. The plates are then put aside on a level surface and enabled it to set for 15min.

### Inoculation of Cultures

A sterile swab was utilized for this reason. Sterile was plunged into a stock suspension of living being. Evacuate abundance liquid by squeezing and turn the swab against the side of tube over the level of suspension. At that point mark the swab uniformly over the surface of the medium in three ways, turning the plates around 60° to guarantee dispersion. After immunization enable the surface of agar to dry.

### Placement of Antibiotic Disc

By utilizing sterile forceps, the fitting antimicrobial plate of Gatifloxacin was put on the agar surface. Each circle ought to be daintily pushed down to guarantee its contacts with agar. it ought not to be moved once set up. The circle ought to be about 15mm from the edge of the plate and no closer than 25mm from circle to plate.

### Incubation of Plates

Inside 30min. of applying the circle, transform the plates and brood at 37°C for 24hours

### Examination of Plates

After 24hroue of hatching, the plates were analyzed to guarantee the growth. Measure the width of each zone of restraint in mm the end purpose of the hindrance is the place the development begins.

### Results and Discussion

The aim for the present investigation was to assess the base inhibitory fixation and safe example of urinary tract contamination causing living being including *Staphylococcus aureus*, *Escherichia. coli*, and *Pseudomonas aeruginosa* against gatifloxacin. The detaches were gathered from various obsessive research centers. They were recognized and code no was given. The antibacterial action did by plate dissemination strategy and the outcome is displayed in tables and chart no [1, 2, 3, 4].

The resistance is the main consideration constraining the long haul effective utilization of antimicrobial specialists. In the pre-anti-infection period, numerous individuals kicked the bucket of bacterial diseases caused by such pathogens as *Staphylococcus aureus*, *Streptococcus pyrogens*, and *Streptococcus pneumonia*. Anti-microbial has diminished the mortality from irresistible ailment yet not the pervasiveness of these infections .it was not long after the clinical presentation of the principal antimicrobials during the 1950s that the main report of bacterial opposition started to show up. Consume and

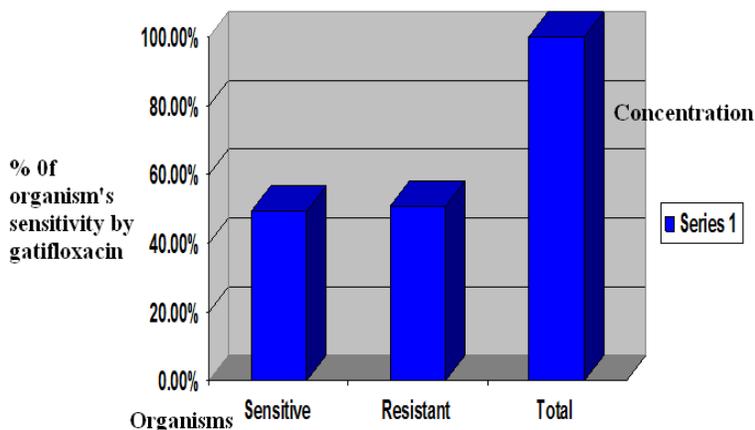
commonly misuse or abuse of antimicrobial specialists has energized the advancement of microscopic organisms toward the opposition, coming about regularly in helpful disappointment. [5]

Gram-positive cocci, for example, *Staphylococcus aureus* and so forth prevail as a reason for nosocomial and network procured diseases. These living beings much of the time uncover a high normal, inherent protection from antimicrobials. These microorganisms can gain protection from every now and again utilized medication quickly through the specific weight of nature and by means of the hereditary development of microscopic organisms. *Staphylococcus aureus* is the most ordinarily separated microscopic organisms causing nosocomial contaminations. Gram-negative microbes, for example, *Escherichia coli* and *Salmonella typhi* and so forth are frequently impervious to antibiotics because of the procurement of safe qualities or quality mutation [6].

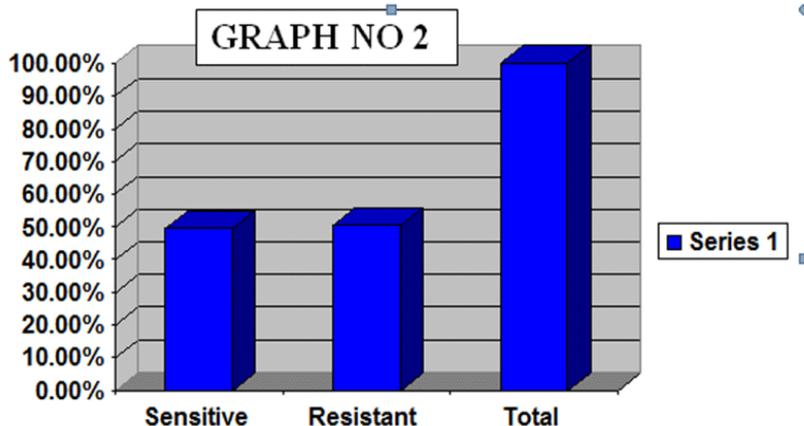
The fluoroquinolones have a wide range of action against microscopic organisms and considered by and large all around endured. Be that as it may, the most recent proof of developing the obstruction in *Streptococcus pneumoniae* segregates has provoked are the advancement of utilization fluoroquinolone, particularly for treatment of diseases for which there are numerous compelling choices accessible [7].

The present examination was led to assess the base inhibitory focus and safe example of aggregate one hundred thirty-five clinical detaches of, *Staphylococcus aureus*, *Escherichia. coli* and *Pseudomonas aeruginosa*, which are the most overwhelming reason for UTI.

The safe example of gatifloxacin was assessed in table no 1 and chart no 1 to diagram no 4

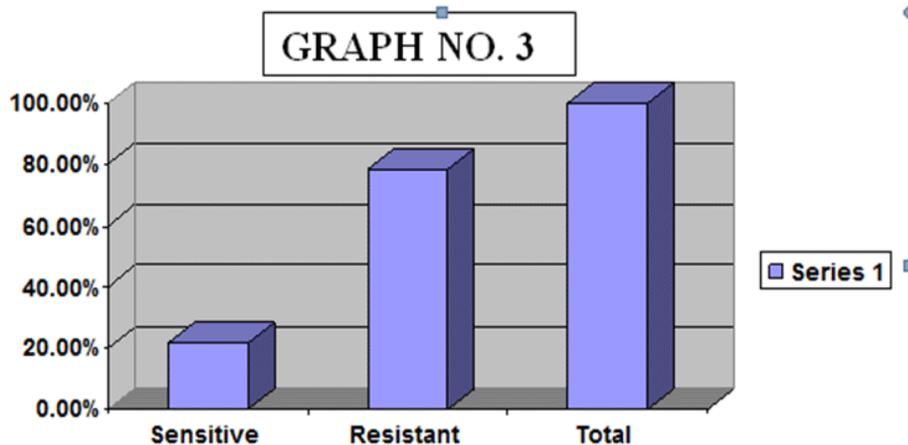


Conc. of antibiotic	S.A ( <i>Staphylococcus aureus</i> )	E.C ( <i>Escherichia coli</i> )	P.A ( <i>Pseudomonas aeruginosa</i> )
0.00025	0%	0%	0%
0.0005	51.11%	44.44%	33.33%
0.00075	46.66%	35.55%	17.77%
0.001	48.88%	35.55%	17.77%
0.00125	51.11%	35.55%	17.77%



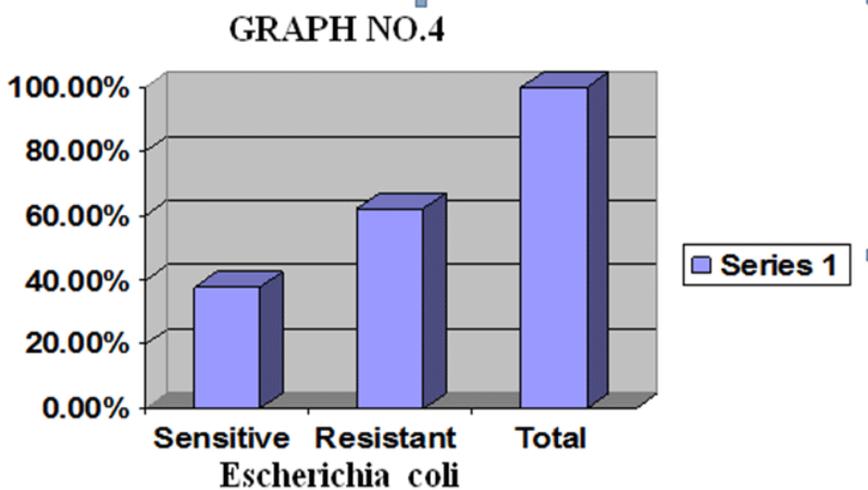
**Staphylococcus aureus**

	Sensitive	Resistant	Total
Series 1	49.44%	50.55%	100%



**Pseudomonas aeruginosa**

	Sensitive	Resistant	Total
Series 1	21.66%	78.33%	100%



	Sensitive	Resistant	Total
Series 1	37.77%	62.22%	100%

Fifty point five percent (50.5%) clinical isolates of *Staphylococcus aureus*, seventy-eight point three percent (78.3%) *Pseudomonas aeruginosa* and sixty two percent (62.2%) *Escherichia coli* are resistant to Gatifloxacin.

The base inhibitory concentration of gatifloxacin for *Staphylococcus aureus*, *Escherichia coli*, and *Pseudomonas aeruginosa* observed to be 0.50mg/lit.

The consequence of *Pseudomonas aeruginosa* are in confirmation with crafted by Stein GE et al(8) has announced 0.5mg/lit, Matsuzaki K et al (1) has detailed 0.06-0.5mg/lit, Naber KG et al (3) has revealed 0.5—0.64mg/lit.

The result of *Escherichia coli* is confirmation with work by Braga PC et al (2) has announced 0.5 to 0.008mg/lit.

The aftereffect of *Staphylococcus aureus* are in confirmation with crafted by Braga PC et al (2) has revealed 0.5 to 0.008mg/lit, Callegan MC et al (4) has announced 0.08-0.57 mg/lit.

Another essential finding of the present investigation was that the living beings are creating protection from anti-infection agents like gatifloxacin and in this manner opposition is expanding which

is exceptionally disturbing sign that fluoroquinolone ought not be endorsed except if no other option is accessible, generally the treatment of some irresistible illnesses wind up unthinkable.

### Conclusion

We concluded in this study that resistant microorganism develop against fourth generation fluoroquinolone Gatifloxacin.

### References

1. Matsuzaki K, Watabe E, Yoshimori K, Shikano M, Sato Y, et al. (2002) Antibacterial activity of gatifloxacin against various fresh clinical isolates in Jpn J antibiot 55(6): 800-807.
2. Braga PC, Dal Sasso M (2002) Effects of sub-minimum inhibitory concentration of gatifloxacin on the inhibition of *Staphylococcus aureus* and *Escherichia coli* adherence. *Arzneimittelforschung* 52(2): 109-112.
3. Naber KG, Hollauer K, Kirchbauer D, Witte W (2000) in vitro activity of gatifloxacin compared with gemifloxacin, moxifloxacin, trovafloxacin, ciprofloxacin and ofloxacin against uropathogens cultured from patients with complicated urinary tract infections. *Int. J Antimicrobial Agents* 16(3): 239-243.

4. Callegan MC, Ramirez R, Kane ST, Cochran DC, Jensen H (2003) antibacterial activity of fourth -generation fluoroquinolones gatifloxacin and moxifloxacin against ocular pathogens. *Adv ther* 20(5): 246-252
5. Straut M, Surdeanu M, Oprisan G, Otelea D, Damian M (1995) antibiotics and bacterial resistance. A few elements of generic basis for this relationship. *Roum Arch Microbial Immunol* 54(4): 241-541.
6. Siu LK (2002) Antibiotics action and resistance in Gram-negative bacteria. *J Microbial Immunol Infect* 35(1): 1-11.
7. Stein GE, Schooley S (2004) urinary concentrations and bactericidal activities of newer fluoroquinolones in healthy volunteers. *Int J Antimicrobial Agents* 24(2): 168-172.