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COMPARISON OF EFFICACY OF FIVE VARIOUS COMMERCIAL PRODUCTS OF CIPROFLOXACIN AGAINST *STAPHYLOCOCCUS AUREUS* AND *PSEUDOMONAS AERUGINOSA*

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ABSTRACT

A study was undertaken to evaluate the efficacy of five various commercial products of ciprofloxacin (a broad spectrum antibiotic), manufactured by different pharmaceutical companies. The drug was evaluated for their performance based on equivalent MIC breakpoint, which is a measure of efficacy of drugs against *Staphylococcus aureus* ATCC6538 and *Pseuomanas aeruginosa* ATCC25668 when tested *in vitro*. The data showed variation for all manufacturers with significant difference ($p < 0.0001$) in efficacy of these drugs at the final dilution level (0.02-0.1µg/µl) against *P.aeruginosa*. Thus the ciprofloxacin, prepared by various manufacturers are not equally efficacious in inhibiting the growth of *S.aureus* and *P.aeruginosa.* Therefore, manufacturers should pay special attention for stringent quality control of drug, and clinicians are suggested to use these antibiotics judiciously in the treatment of patients for these infections.

INTRODUCTION: Pharmaceutical products play an important role in improving the health and encouraging the well-being of every individual. These medicines bring succor in the prevention and treatment of diseases, disorders, or conditions. The three criteria considered the cornerstone for these products-quality, effectiveness, and safety- should be demonstrated and verified prior to their rational use [1].

However, the overuse and misuse of these products are leading to the emergence of resistance to these life-saving drugs. Consequently, it is now imperative for clinical microbiologists to provide the clinicians with accurate information required for selecting antibiotics for patient's therapy and care [2]. The method of choice for the clinical microbiologist for the *in vitro* antimicrobial susceptibility testing is still the disc diffusion method.

The Kirby-Bauer technique [3] for disk susceptibility testing has been recommended by CLSI, which is approved by US FDA, and it is also recommended by WHO. Standardization of the techniques controls variation in results, and interpretation is based on a comparison the diameter of the zone of inhibition with published criteria [4].

Nowadays, lots of pharmaceutical companies are involved in formulation and manufacturing of drugs with different trade names. It is reasonable to expect that all the drugs contain the same salt and quantity. Thus, a clinician prescribes the medicine with its trade name only [5].

Therefore, this study reviews to find out that, is there any significant difference in the efficacy of these drugs manufactured by different companies with a focus on antimicrobial susceptibility testing against

Staphylococcus aureus ATCC6538 and *Pseuomanas aeruginosa* ATCC25668. For this purpose, we examine five various commercial products of Ciprofloxacin which is an example of 4-Quinolones that inhibiting DNA gyrase, an enzyme responsible for the tight packaging necessary to fit bacterial DNA into the cell [6, 7, 8].

MATERIALS AND METHODS:

Bacterial Culture: Lyophilized cultures of *S.aureus* ATCC6538 and *P.aeruginosa* ATCC25668 purchased from Hi-Media were revived, and maintain the culture according to instruction given by Hi-Media.

Antibiotics: Various products of ciprofloxacin viz. Cifran 250mg, Ciprobid 250mg, Ciplox 250mg, Zoxen 250mg, and Deplox 250mg manufactured by Ranbaxy, Zydus Alidac, Cipla LTD, FDC Limited, Daffodillus Pharmaceutical LTD respectively, were purchased from market in tablet form, and coded as the sample A, B, C, D and E. Stock solution of these antibiotics were prepared to concentration 5µg/µl for each sample.

Antimicrobial Susceptibility Testing: Antimicrobial susceptibility testing was performed on Mueller-Hinton Agar (Hi-media) plates with sterilized dry filter paper disc [5] loaded with different quantum of the antibiotics ranging from 5 to 0.02 µg by the Kirby-Bauer disc diffusion method.

Briefly, three to five well-isolated colonies from both the organisms of the same morphological type were selected from agar plates and transferred separately into a tube containing 4 to 5 ml of peptone water. The broth cultures were incubated at 35°C until it achieves the turbidity of the 0.5 McFarland standards.

Three sets of Muller Hinton agar plates were prepared with a uniform depth of 4mm for each organism. One set contained five plats for each sample.

The sterile cotton swab dipped into the inoculums and then streaked on all dried MHA plates to ensure an even distribution than plates are incubated for activation of culture for 30 minutes [6-8].

In first set, five various products of ciprofloxacin loaded disc, rang from 1 to 5µg were placed onto the dried surface of the inoculated agar plate with distilled water loaded disc, which was a negative control. Therefore, six filter paper disc loaded with different content of drug were placed on 90mm diameter Petri plate.

In second set, drug range from 0.2 to 1µg, were placed and in third set drug content of a disc were 0.02 to 0.1µg for each product. All the plates were incubated at 37^{o}C for 18 to 24 hours. After incubation period, plates were observed and results were recorded and interpreted as per CLSI guidelines. The procedure was repeated in triplicate.

Statistical Analysis: One-way ANOVA (at significant label 0.05) has been performed with the mean diameter of the zone of inhibition for all samples by Graph pad prism 5 software.

RESULTS: MHA plates had shown the zone of inhibition of various diameters with respect to the different contents of the antibiotic discs. The third set showed the equivalent MIC breakpoint with every sample. Sample A when tested against *S. aureus* ATCC6538 the equivalent MIC breakpoint was 0.02µg/µl, and for sample B, C, D and E were 0.08 µg/µl.

When the sample A was tested against *P. aeruginosa* ATCC25668, the breakpoint was 0.02 µg/µl, however, sample C and D showed 0.2 µg/µl, and sample B and E 0.4 µg/µl as shown in **table 1**.

We also observed that as the dilutions of all samples were increased, the resolution of the zone of inhibition increases (**Figure 1 to 6**).

The One-way ANOVA (at significant label 0.05) results showed that there was significant difference ($p < 0.0001$) in efficacy of these drugs at a final dilution level (drug content 0.02- 0.1µg) against *P. bbaeruginosa*.

TABLE 1: SHOWS THE COMPARISON IN MEAN ZONE DIAMETER OF INHIBITION OF VARIOUS PRODUCTS OF CIPROFLOXACIN

Drug content In µg	Sample A		Sample B		Sample C		Sample D		Sample E	
	S. aureus	*P. aeruginosa*	*S. aureus*	*P. aeruginosa*	*S. aureus*	*P. aeruginosa*	*S. aureus*	*P. aeruginosa*	*S. aureus*	*P. aeruginosa*
	Diameter of zone of inhibition in mm	Diameter of zone of inhibition in mm	Diameter of zone of inhibition in mm	Diameter of zone of inhibition in mm	Diameter of zone of inhibition in mm	Diameter of zone of inhibition in mm	Diameter of zone of inhibition in mm	Diameter of zone of inhibition in mm	Diameter of zone of inhibition in mm	Diameter of zone of inhibition in mm
5	21	21.2	19.3	22.5	19.6	20	20	21.5	19.8	22
4	20.33	19.7	19	21	18.6	20	19.16	21	18.6	20.5
3	19.66	19.5	18.3	20.5	17.6	19.5	15.66	20.7	17	19
2	16.66	19	16	18.5	16.5	17.5	14.3	19.2	16.8	17.5
1	16	16.5	14	13.6	14	13.2	13.6	13.8	16.5	13.2
0.8	15.5	14.5	12.6	12.5	13.3	11.5	13.1	10.5	15	11.5
0.6	15	13.5	12.6	11	13	10	12	9.2	14	9
0.4	13.6	12.5	12.3	9.5*	11.6	10	11.1	8.2	13.3	8*
0.2	12	11.5	10.3	0	9.6	7*	9	8*	11.1	0
0.1	8.3	11.5	7.5	0	7.25	0	7.5	0	7.5	0
0.08	8	10.5	7*	0	7*	0	7*	0	6.7*	0
0.06	7.5	8.5	0	0	0	0	0	0	0	0
0.04	7	8	0	0	0	0	0	0	0	0
0.02	7*	7.5*	0	0	0	0	0	0	0	0
0	No Zone of inhibition	No Zone of inhibition	No Zone of inhibition	No Zone of inhibition	No Zone of inhibition	No Zone of inhibition	No Zone of inhibition	No Zone of inhibition	No Zone of inhibition	No Zone of inhibition

*Shows the mean zone diameter of inhibition at which the inhibition of bacterial growth start.

DISCUSSIONS: Efficacy of the drugs was evaluated on the basis of their performance of antimicrobial susceptibility testing. Results showed that sample A has lowest breakpoint against both the test organisms thus showed better results than the other samples containing the same amount of the salt.

Comparison of drug from different sources was somewhat limited by the fact that all manufacturers do not produce the drug with same efficacy and cost. The differences may be due to differences in the testing methods adopted by manufacturers in various countries [9].

However, laboratory testing of antibiotic susceptibility contributes directly to patients care, and data generated from the test serve as a guideline for deciding therapy [10].

CONCLUSION: Therefore, it is concluded that the ciprofloxacin, prepared by various manufacturers are not equally efficacious in inhibiting the growth of *S. aureus* and *P. aeruginosa* when tested *in vitro*. Then manufacturers should pay special attention for stringent quality control of drug, and clinicians are suggested to use these antibiotics judiciously in the treatment of patients for these infections.

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REFERENCES:

1. Videau J. Making medicines safe (editorial). Bull WHO 2001, 79(2): 87.
2. A Joshi, V Iyer, U Balasubramaniam, A Kagal, R Bharadwaj. Comparison of Efficacy of Three Commercially Available Antibiotic Discs. *Indian journal of Medical Microbiology* (2008) 26(2): 160-2.
3. Bauer, A. W., Kirby, W. M. M., Sherris, J. C., and Turck, M. (1966). Antibiotic susceptibility testing by a standardized single disk method. *Amer. J. clin. Path*., 1966, 45, 493
4. National Committee for Clinical Laboratory Standards. NCCLS document M 100- S15. Performance Standards for Antimicrobial Susceptibility Testing. 8th ed, NCCLS: Waynae; 2004.
5. Kramer J. and Kirshbaum A. Effect of paper on the performance assay in the control of antibiotic sensitivity discs. *Appl Microbiol* 1961: 9; 334-336
6. S.J.Cavalieri, R.J. Harbeck(2005), Mannual of Antimicrobial Suceptivility Testing, ISBN 1-55581-349-6
7. Ira R. Bacteriology, Standard Operative procedure manual for microbiology laboratories, *National Institute of Biologicals.* 1995, 73-97
8. John D.T and James H.J Antimicrobial Susceptibility testing: General Considerations. Manual of Clinical Microbiology 7th edition, Murray P.R, Baron E.J, Pfaller M.A, Turnover F.C, Yolken R, American Society for Microbiology, Washington DC, 1999, 1469-1473
9. D. F. J. Brown and Kothari. Comparison of antibiotic discs from different sources. *J. clin. Path.,* 1975, 28. 779-786
10. Miles RS, Amyes SG. *In*: Mackie and McCartney. Practical Medical Microbiology, 14th ed. Collee, Marmion, Fraser, Simmons, editors. Pub Churchill Livingstone: Laboratory control of Antimicrobial Therapy; 1996. 151.

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