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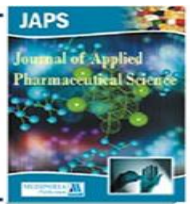

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Comparative *in vitro* antibacterial activities study of some mucoadhesive polymeric suspensions containing fluoroquinolones

Chandra Kanti Chakraborti, Subhashree Sahoo, Pradipta Kumar Behera and Anjulata Manjari Sahoo

Chandra Kanti Chakraborti, Subhashree Sahoo
Department of Pharmaceutics, Kanak Manjari Institute of Pharmaceutical Sciences, Rourkela 769015, Orissa, India.

Pradipta Kumar Behera
School of Chemistry, Sambalpur University, Jyoti Bihar 768019, Orissa, India.

Anjulata Manjari Sahoo
Department of Pharmaceutical Biotechnology, Kanak Manjari Institute of Pharmaceutical Sciences, Rourkela 769015, Orissa, India.

For Correspondence
Dr. Chandra Kanti Chakraborti
Department of Pharmaceutics
Kanak Manjari Institute of Pharmaceutical Sciences
Rourkela 769015, Orissa, India
Ph +91-9776145092
Fax +91-661-2480752

ABSTRACT

To study *in vitro* antibacterial activities of mucoadhesive suspensions containing Norfloxacin, Ciprofloxacin and Ofloxacin, three different formulations of each drug were prepared by using three polymers, such as Hydroxypropyl methylcellulose (HPMC), Carbapol934 (C934) and Carbapol940 (C940), along with some common ingredients (bases). For the antibacterial activities study of the samples, agar well diffusion method was performed taking *Staphylococcus aureus* (ATCC 25923), *Bacillus subtilis* and *Escherichia coli* (ATCC 25922). Considering the overall antibacterial activities, it could be mentioned that HPMC containing formulations were superior to others in most of the cases. Amongst C934 and C940 containing suspensions, the former was more potent than the later. Antibacterial activities of most of the formulations were either more effective or similar to those of corresponding drugs in water against the strains used in the study. Only few formulations were inferior to the corresponding drugs in water. Ciprofloxacin in citrate buffer was not better than Ciprofloxacin containing suspensions. Samples like both marketed suspensions and discs of different drugs were inferior to all the formulations and corresponding drugs in water/Ciprofloxacin in citrate buffer. The negative controls of the study, i.e., the different bases, distilled water and citrate buffer did not show any antibacterial activity.

Keywords: Norfloxacin; Ciprofloxacin; Ofloxacin; HPMC; C934; C940

INTRODUCTION

Now-a-days mucoadhesive suspensions are being prepared for several purposes (Jain *et al*., 2011, Sahoo *et al*., 2011). In some of these suspensions, antibacterial substances are also incorporated. To study their antibacterial activities very few *in vitro* methods are available (Sahoo *et al*., 2011). Such investigations are essential to know the availability/release of the drug from a base (containing a polymer with other substances). Sometimes drug release from the base is reduced, as a result of which that formulation may not be considered as a suitable preparation to control bacterial infections effectively. Considering the importance of the availability of the drug from the suspensions, different mucoadhesive suspensions of some fluoroquinolone antibacterial agents such as Norfloxacin (Norflox), Ciprofloxacin (Cipro) and Ofloxacin (Oflox) were prepared in the present study.

Their *in vitro* antibacterial activities were compared with those of the corresponding drugs in water; Cipro in citrate buffer; marketed suspensions and discs containing Norflox, Cipro and Oflox; different bases; distilled water and only in case of Cipro, citrate buffer against *Staphylococcus (S.) aureus* (ATCC 25923), *Bacillus (B.) subtilis* and *Escherichia (E.) coli* (ATCC 25922). For the above-mentioned study, mucoadhesive suspensions of each drug were prepared using bases containing three different polymers. Hydroxypropyl methylcellulose (HPMC) and two grades of carbopol polymer, having different crosslinking agents such as C934 and C940, were selected for our investigation. HPMC is propylene glycol ether of methyl-cellulose having high swellability upon contact with water (Fatimi *et al*., 2008). It is one of the most commonly used hydrophilic biodegradable polymers for developing mucoadhesive formulations, because it works as a pH-independent gelling agent (Gao et al., 1996, Siepmann and Peppas, 2001, Phaechamud, 2008). On the other hand, carbopol polymers form hydrogel that change their swelling behaviour upon exposure to an external stimulus, such as change in pH (Qiu and Park, 2001), temperature (Bromberg and Ron, 1998), light, or electric field, and are known as "environmentally responsive polymers" or "smart gels" (Galaev and Mattiasso, 1999). Carbopol polymers have recently attracted considerable interest in the field of drug delivery as the means of providing an on-off release by shrinking and swelling in response to the change in pH (Jeong and Gutowska, 2001, Gupta *et al*., 2002).

MATERIAL AND METHODS

Material

The following materials were used for the study: Norfloxacin, Ciprofloxacin and Ofloxacin were obtained from Dr. Reddy's Lab, Hyderabad, India, as gift samples. Hydroxypropyl methylcellulose (HPMC E15 LV Premium) was supplied by Loba Chemie Pvt. Ltd., India. It was having methoxy group (23.8%) and hydroxypropoxy group (8.3%). Pluronic F 68 and Soya lecithin were purchased from Himedia Laboratories Pvt. Ltd., India. C934, C940, Glycerol, Citric acid, Sodium citrate, Methyl praraben sodium, Propyl paraben sodium, Sorbitol solution I.P. and Sucrose were supplied by Cosmo Chem. Laboratory, Pune, India. Tri-sodium citrate dehydrate purified was obtained from Merck Specialities Private Limited, Mumbai, India. Ultra pure water was obtained from a Millipore Milli-Q UV water filtration system. For the antibacterial activity study, different media, and Norfloxacin, Ciprofloxacin and Ofloxacin Susceptibility test discs were obtained from Himedia Laboratories Pvt. Ltd., India.

Samples Used

Formula for the Preparation of Mucoadhesive Suspensions

(Percentage with respect to Norfloxacin/Ciprofloxacin/Ofloxacin)

Polymer ($S_1/S_2/S_3$)*	5%
Pluronic F 68	5%
Soya lecithin	1%
Sorbitol Solution (80%)	7.2%
Glycerin	0.8%
Simple Syrup IP	40%
Distilled water q.s. up to	100ml

Concentration of Norfloxacin used in the formulation – 500mg/25ml of distilled water

Concentration of Ciprofloxacin used in the formulation - 1.25g/25ml of distilled water

Concentration of Ofloxacin used in the formulation – 250mg/25ml of distilled water

*S_1- HPMC; S_2 – C934; S_3 – C940.

Other Samples

S_{1b}-S_1 without drug;

S_{2b}- S_2 without drug;

S_{3b}- S_3 without drug;

S_4 - Drug in 25ml of distilled water;

S_{4c} - Cipro in 25ml of Citrate buffer of pH 5.5;

S_5 - Marketed products –

BACIGYL Suspension (ARISTO Pharmaceutical Pvt. Ltd. Mumbai): Norfloxacin

suspension each 5ml contain 100mg Norfloxacin;

CIPROLAR Suspension (Lark Laboratotries (India) Ltd, New Delhi): Ciprofloxacin suspension each 10ml contain 250mg Ciprofloxacin;

ONOFF (Ranbaxy Laboratories Ltd, New Delhi): Ofloxacin suspension each 5ml contain 50mg Ofloxacin;

S_6 - Disc concentration - 10 µg Norfloxacin/disc; 10 µg Ciprofloxacin /disc; 5µg Ofloxacin /disc

S_7 – Distilled water

S_8 – Citrate buffer of pH 5.5

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Methods

Preparation of Formulations

Praparation of Bulk A

In a beaker, 6 ml of distilled water was heated up to 80° C. Sucrose (10 g) was added under continuous stirring. The temperature was monitored in such a way so that it should not fall below 70° C, till the sucrose was completely dissolved. The prepared syrup was cooled properly at room temperature and kept overnight. Syrup was filtered using 120 mesh nylon cloth.

Praparation of Bulk B

Five millilitre of distilled water was taken in a beaker to which 1.8 ml of sorbitol solution and 0.2 ml glycerin were added. The mixture was stirred properly. To this solution, pluronic F 68 (5%), soya lecithin (1%) and 5% of each polymer in w/w of drug were added with continuous stirring.

Preparation of Mucoadhesive Suspension and Ultrasonication

Five millilitre of distilled water was taken in another beaker to which a drug (500mg of Norflox / 1.25g of Cipro / 250mg of Oflox) was added. To the drug suspension, the bulk B and bulk A were added with continuous stirring. The volume was made up to 25 ml by Ultra pure water. The pH of formulations containing Norflox and Oflox was 5.5, while in case of Cipro it

was adjusted to 5.5 by citrate buffer. Homogenization was carried out for at least 20 min by ULTRASONIC HOMOZENIZER LABSONIC^R M (SARTORIUS), having operating frequency 30 KHZ and line voltage 230 V/50 HZ, using the probe made up of Titanium of diameter 7 mm and length 80 mm. The setting knob "cycle" was adjusted to 0.8, indicating sound was emitted for 0.8 s and paused for 0.2 s. In this manner, we could expose our sample with 100% amplitude, while reducing the heating effect to 80%. This LABSONIC^{R}M generates longitudinal mechanical vibrations with a frequency of 30,000 oscillations / s (30 KHZ). The probes bolted to the sound transducer were made of high-strength Titanium alloys, built as λ /2 oscillators. It amplified the vertical oscillation, and transferred the ultrasonic energy via its front surface with extremely high power density into the sample that was to be subjected to ultrasonic waves. In our study, stress applied was sound wave and in addition, mild rise in temperature of the sample occurred during ultrasonication which helped in the homogenization of the suspension.

Method of Antibacterial Activity Study

The nutrient agar well diffusion method as described by Schillenger and Luke (1989) was performed for our study. Sterile nutrient agar medium was inoculated with 0.1ml of fresh overnight nutrient broth culture of each bacterium (approx.10^7CFU/ml) and poured into sterile petriplates (Bayoub *et al*., 2010). In each plate, wells of 6mm in diameter were punched using a sterile borer and the plates were allowed to dry for 5min (Ganjewala *et al*., 2009, Bayoub *et al*., 2010). For the present study, mucoadhesive suspensions of Norflox / Cipro / Oflox with HPMC containing base (S_1), Norflox / Cipro / Oflox with C934 containing base (S_2) and Norflox / Cipro / Oflox with C940 containing base (S_3), Norflox / Cipro / Oflox in distilled water (S_4), pure Ciprofloxacin in citrate buffer (S_{4c}), marketed suspension of Norflox / Cipro / Oflox (S_5), disc containing Norflox / Cipro / Oflox (S_6), bases (S_{1b}, S_{2b} and S_{3b}) (at different concentrations as mentioned earlier), distilled water (S_7) and citrate buffer (S_8) were used against *S. aureus* (ATCC 25923), *B. subtilis* and *E. coli* (ATCC 25922).

Fifty microliter of each sample was dispensed into different wells using sterile micropipettes. For our study S_{1b}, S_{2b}, S_{3b}, S_7 and S_8 were used as negative controls. After holding the plates at room temperature for 2 h to allow diffusion of the samples and controls into the nutrient agar medium, the plates were incubated at 37 ^{0}C for 24h. The plates were examined for inhibition of the bacterial growth around the wells after the incubation period. The diameters of the zones of inhibition in each case were measured (Bayoub *et al*., 2010).

RESULT AND DISCUSSION

Against *S. aureus*, both Norflox with HPMC containing base (NS_1) and Norflox with C934 containing base (NS_2) were more effective than Norflox in distilled water (NS_4), while Norflox with C940 containing base (NS_3) was more or less similar to NS_4. Between NS_1 and NS_2, it was observed that NS_1 was more potent than NS_2 against that strain. In case of NS_1, the zone of inhibition was more than NS_4 against *B. subtilis*, on the other hand, it was more or less similar in NS_2, NS_3 and NS_4. All the suspensions (NS_1, NS_2 and NS_3) were more potent than NS_4 against *E. coli*. The zones of inhibition produced by Norflox containing marketed product (NS_5) and Norflox Disc (NS_6) were smaller than those of NS_4 against all the strains. The negative controls of the study, i.e., the bases without Norflox (NS_{1b}, NS_{2b} and NS_{3b}) and distilled water (S_7) did not show any antibacterial activity (Table 1 and Fig. 1).

Table. 1: Antibacterial activities of different mucoadhesive suspensions, pure Norflox in distilled water, marketed suspension and discs containing Norflox, and negative controls.

Micro-organisms	Average Zone of Inhibition (mm)									
	NS_1	NS_{1b}	NS_2	NS_{2b}	NS_3	NS_{3b}	NS_4	NS_5	NS_6	S_7
S.aureus	40	0	34.3	0	27.3	0	24.7	24	20	0
B.subtilis	30.3	0	28	0	29.3	0	28	23	18	0
E. coli	24	0	21.3	0	23.3	0	18.3	23	12	0

NS_1– Norflox with HPMC containing base; NS_{1b} - HPMC containing base; NS_2 - Norflox with C934 containing base; NS_{2b}- C934 containing base; NS_3 - Norflox with C940 containing base; NS_{3b} - C940 containing base; NS_4– Norflox in distilled water; NS_5– Marketed Norflox suspension; NS_6– Norflox containing disc; S_7 – Distilled water

Table. 2: Antibacterial activities of different mucoadhesive suspensions, pure Cipro in distilled water and citrate buffer, marketed suspension and discs containing Cipro, and negative controls.

Micro-organisms	Average Zone of Inhibition (mm)											
	CS_1	CS_{1b}	CS_2	CS_{2b}	CS_3	CS_{3b}	CS_4	CS_{4c}	CS_5	CS_6	S_7	S_8
S. aureus	54.8	0	57.7	0	50.8	0	50.5	47.8	38.8	28.3	0	0
B. subtilis	45.4	0	40.8	0	35.5	0	47.3	38.3	38.7	27.0	0	0
E. coli	32.6	0	32.5	0	32.3	0	32.4	32.3	11.7	12	0	0

CS_1 – Cipro with HPMC containing base; CS_{1b}- HPMC containing base; CS_2 - Cipro with C934 containing base; CS_{2b} - C934 containing base; CS_3- Cipro with C940 containing base; CS_{3b} - C940 containing base; CS_4– Cipro in distilled water; CS_{4c}– Cipro in citrate buffer; CS_5 –Marketed Cipro suspension; CS_6– Cipro containing disc; S_7 – Distilled water; S_8– Citrate buffer.

Table. 3: Antibacterial activities of different mucoadhesive suspensions, pure Oflox in distilled water, marketed suspension and discs containing Oflox, and negative controls.

Micro-organisms	Average Zone of Inhibition (mm)									
	OS_1	OS_{1b}	OS_2	OS_{2b}	OS_3	OS_{3b}	OS_4	OS_5	OS_6	S_7
S. aureus	41.5	0	44	0	45	0	42.3	39	25.5	0
B. subtilis	52.8	0	36.5	0	40	0	41	33	26.5	0
E. coli	34	0	37.3	0	32.7	0	30.2	26	12	0

OS_1 – Oflox with HPMC containing base; OS_{1b}- HPMC containing base; OS_2- Oflox with C934 containing base; OS_{2b}- C934 containing base; OS_3 - Oflox with C940 containing base; OS_{3b}- C940 containing base; OS_4– Oflox in distilled water; OS_5– Marketed Oflox suspension; OS_6– Oflox containing disc; S_7– Distilled water

Table. 4: Antibacterial activities of different Fluoroquinolone mucoadhesive suspensions and their pure suspensions in different vehicles.

Micro-organisms	Average Zone of Inhibition (mm)												
	NS_1	NS_2	NS_3	NS_4	CS_1	CS_2	CS_3	CS_4	CS_{4c}	OS_1	OS_2	OS_3	OS_4
S.aureus	40	34.3	27.3	24.7	54.8	57.7	50.8	50.5	47.8	41.5	44	45	42.3
B.subtilis	30.3	28	29.3	28	45.4	40.8	35.5	47.3	38.3	52.8	36.5	40	41
E. coli	24	21.3	23.3	18.3	32.6	32.5	32.3	32.4	32.3	34	37.3	32.7	30.2

NS_1 – Norflox with HPMC containing base; NS_2- Norflox with C934 containing base; NS_3- Norflox with C940 containing base; NS_4– Norflox in distilled water; CS_1– Cipro with HPMC containing base; CS_2- Cipro with C934 containing base; CS_3- Cipro with C940 containing base; CS_4– Cipro in distilled water; CS_{4c}– Cipro in citrate buffer; OS_1– Oflox with HPMC containing base; OS_2 - Oflox with C934 containing base; OS_3- Oflox with C940 containing base; OS_4– Oflox in distilled water

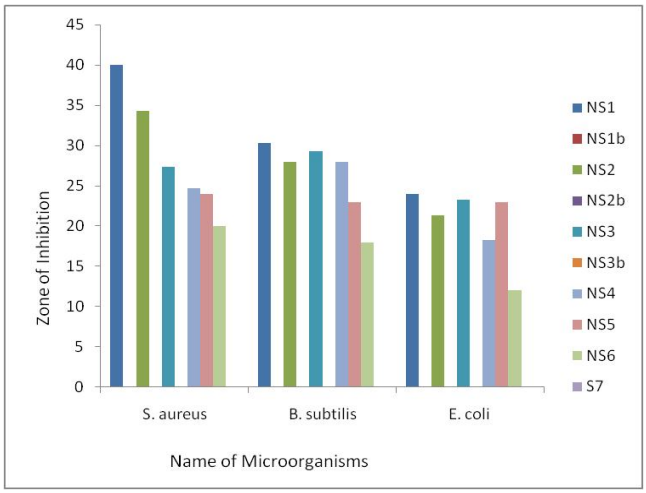

Fig. 1: Comparative zones of inhibition of different samples containing Norfloxacin against some microorganisms.
NS_1– Norflox with HPMC containing base; NS_{1b} - HPMC containing base; NS_2 - Norflox with C934 containing base; NS_{2b}- C934 containing base; NS_3 - Norflox with C940 containing base; NS_{3b} - C940 containing base; NS_4– Norflox in distilled water; NS_5– Marketed Norflox suspension; NS_6– Norflox containing disc; S_7 – Distilled water

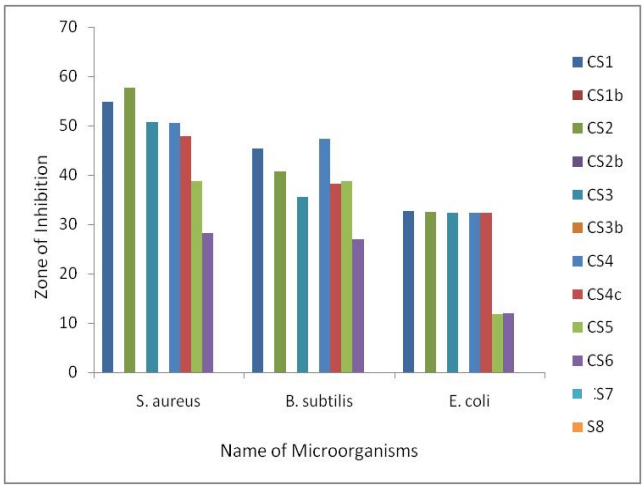

Fig. 2: Comparative zones of inhibition of different samples containing Ciprofloxacin against some microorganisms.
CS_1 – Cipro with HPMC containing base; CS_{1b}- HPMC containing base; CS_2 - Cipro with C934 containing base; CS_{2b} - C934 containing base; CS_3- Cipro with C940 containing base; CS_{3b} - C940 containing base; CS_4– Cipro in distilled water; CS_{4c}– Cipro in citrate buffer; CS_5 –Marketed Cipro suspension; CS_6– Cipro containing disc; S_7 – Distilled water; S_8– Citrate buffer.

Cipro with HPMC containing base (CS_1) and Cipro with C934 containing base (CS_2) were more active than Cipro in distilled water (CS_4), while Cipro with C940 containing base (CS_3) and CS_4 were equally potent against *S. aureus*. Moreover, CS_2 and CS_3 were inferior to CS_4 in case of *B. subtilis*. Against *E. coli*, all the samples (CS_1, CS_2, CS_3 and CS_4) produced more or less similar zones of inhibition. Considering the overall pattern of antibacterial activity, CS_4 was not inferior to Cipro in citrate buffer (CS_{4c}), Marketed Cipro suspension (CS_5) and Cipro containing disc (CS_6). CS_5 and CS_6 were not very effective against the strains used in this study. The negative controls of the study, i.e., the bases without Cipro (CS_{1b}, CS_{2b} and CS_{3b}), distilled water (S_7) and citrate buffer (S_8) did not show any antibacterial activity (Table 2 and Fig. 2). In case of *S. aureus*, antibacterial activity of Oflox in distilled water (OS_4) was more or less retained in all the formulations. While Oflox with C934 containing base (OS_2) was to some extent inferior to OS_4, Oflox with HPMC containing base (OS_1) was more active than OS_4 as far as antibacterial activity against *B. subtilis* was concerned. Both Oflox with C940 containing base (OS_3) and OS_4 were more or less equally effective against *B. subtilis*. In case of *E. coli*, OS_2 produced more antibacterial activity than that of OS_4, while OS_1 and OS_2 were also effective. However, Marketed Ofloxacin suspension (OS_5) and Ofloxacin containing disc (OS_6) were inferior to all the formulations and OS_4 as far as their antibacterial activities were concerned. The negative controls of the study, i.e., the bases without Oflox (OS_{1b}, OS_{2b} and OS_{3b}) and distilled water (S_7) did not show any antibacterial activity (Table 3 and Fig. 3).

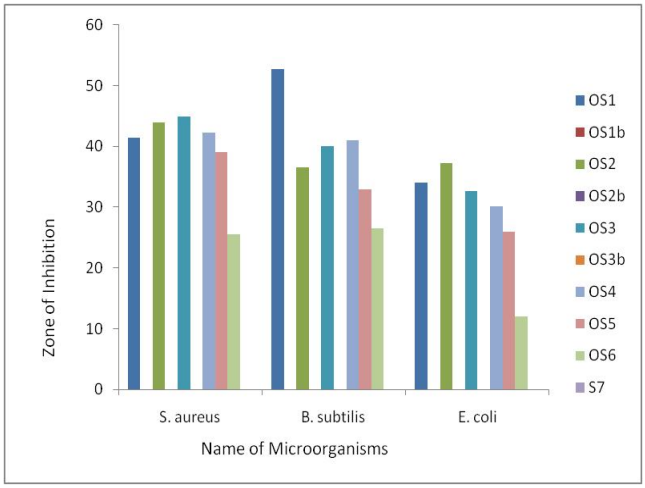

Fig. 3: Comparative zones of inhibition of different samples containing Ofloxacin against some microorganisms.
OS_1 – Oflox with HPMC containing base; OS_{1b}- HPMC containing base; OS_2- Oflox with C934 containing base; OS_{2b}- C934 containing base; OS_3 - Oflox with C940 containing base; OS_{3b}- C940 containing base; OS_4– Oflox in distilled water; OS_5– Marketed Oflox suspension; OS_6– Oflox containing disc; S_7– Distilled water

Antibacterial activities of different Fluoroquinolone mucoadhesive suspensions, their pure suspensions in distilled water and citrate buffer (only in case of Ciprofloxacin) have been mentioned in Table 4. Comparative zones of inhibition of different samples against *S. aureus*, *B. subtilis* and *E. coli* have been demonstrated in Figure 4.

When HPMC was used as a polymer, NS_1 was found to become more potent than the NS_4 against *S. aureus* and *E. coli*, while OS_1 produced more zone of inhibition than OS_4 against *B. subtilis*. On the other hand, both the formulations of Cipro and Oflox with C934, i.e., CS_2 and OS_2, were superior to the corresponding drugs in water (CS_4 and OS_4) against *S. aureus*, and *E. coli*, respectively. With respect to other samples, maximum zone of inhibition was produced by suspension of Oflox containing C940 (OS_3) only against *S. aureus*. Cipro in citrate buffer (CS_{4c}) was not better than other suspensions such as CS_1, CS_2, CS_3 and CS_4. The samples like marketed suspensions and discs of different drugs were inferior to the formulations of those drugs and also the corresponding drugs in water/Cipro in citrate buffer (Table 4 and Fig. 4).

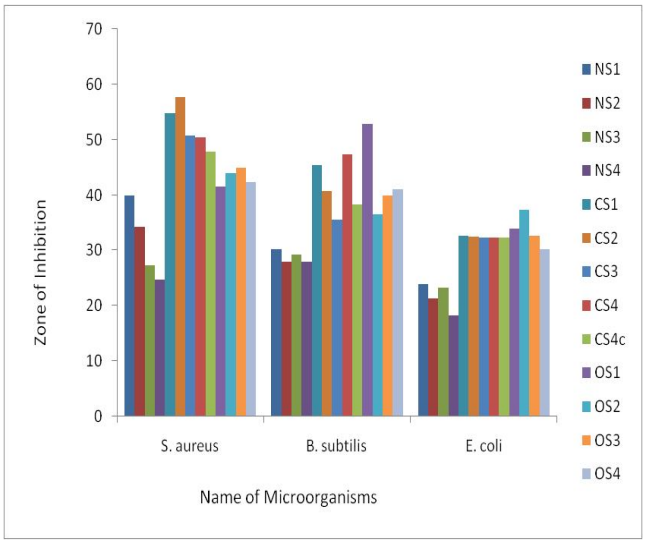

Fig. 4: Comparative zones of inhibition of different samples against some microorganisms.

NS_1 – Norflox with HPMC containing base; NS_2- Norflox with C934 containing base; NS_3- Norflox with C940 containing base; NS_4– Norflox in distilled water; CS_1– Cipro with HPMC containing base; CS_2- Cipro with C934 containing base; CS_3- Cipro with C940 containing base; CS_4– Cipro in distilled water; CS_{4c}– Cipro in citrate buffer; OS_1– Oflox with HPMC containing base; OS_2 - Oflox with C934 containing base; OS_3- Oflox with C940 containing base; OS_4– Oflox in distilled water.

Considering the overall antibacterial activity, it could be mentioned that HPMC containing formulations were superior to others in most of the cases. Moreover, it is evident that most of the formulations used in the study were found to be more effective than the corresponding drugs in water. In some cases, formulations were similar to the pure drugs in water as far as their antibacterial activity were concerned. However, only few formulations were found to be inferior to the corresponding drugs in water. The result of the study indicates that the antibacterial activities of the drugs was either retained or enhanced when most of the formulations were prepared. Since the bases did not produce any zone of inhibition against the strains, some formulations showing more zones of inhibition than those of the corresponding drugs in water suggested that the bases had got potentiating effect on the antibacterial activities of the different drugs. Moreover, the difference in antibacterial activities between different suspensions may be due to either the effect of bacterial metabolites which may influence the rate of release or the interaction between the drug and the base (Chakraborti *et al*., 2012).

REFERENCES

Bromberg LE, Ron ES. Temperature-responsive gels and thermogelling polymer matrices for protein and peptide delivery. Adv Drug Deliv Rev. 1998, 31: 197-221.

Bayoub K, Baibai T, Mountassif D, Retmane A, Soukri A. Antibacterial activities of the crude ethanol extracts of medicinal plants against *Listeria monocytogenes* and some other pathogenic strains. African J Biotechnol. 2010, 9(27): 4251-4258.

Chakraborti CK, Sahoo S, Behera PK. Antibacterial Activities Study of Ofloxacin Containing Mucoadhesive Suspensions. J Pharm Res. 2012, 5(2): 822-824.

Fatimi A, Tassin JF, Quillard S, Axelos MAV, Weiss P. The rheological properties of silated hydroxypropylmethylcellulose tissue engineering matrices. Biomater. 2008, 29(5): 533-543.

Galaev IY, Mattiasso B. Smart' polymers and what they could do in biotechnology and medicine. Trends Biotechnol. 1999, 17: 335-340.

Ganjewala D, Sam S, Khan KH. Biochemical compositions and antibacterial activities of *Lanatana camara* plants with yellow, lavender, red and white flowers, EurAsia J BioSci. 2009, 3: 69-77

Gao P, Skoug JW, Nixon PR, *et al.* Swelling of Hydroxypropyl Methylcellulose Matrix Tablets. 2. Mechanistic Study of the Influence of Formulation Variables on Matrix Performance and Drug Release. J Pharm Sci. 1996, 85(7): 732-740.

Gupta P, Vermani K, Garg S. Hydrogels: from controlled release to pH-responsive drug delivery. Drug Discovery Today. 2002, 7: 569-579.

Jain V, Kare P, Jain D, Ju RT, Stemm NL, Sung K. Development and Characterization of Mucoadhesive Nanosuspension of Ciprofloxacin. Acta Poloniae Pharmaceutica-Drug Res. 2011, 68(2): 273-278.

Jeong B, Gutowska A. Stimuli-responsive polymers and their biomedical applications. Trend Biotechnol. 2001, 20: 305-311.

Phaechamud T. Variables Influencing Drug Release from Layered Matrix System Comprising Hydroxypropyl Methylcellulose. AAPS Pharm Sci Tech. 2008, 9(2): 668- 674.

Qiu Y, Park K. Environment-sensitive hydrogels for drug delivery. Adv Drug Deliv Rev. 2001, 53: 321-339.

Sahoo S, Chakraborti CK, Mishra SC. Antibacterial activity of a mucoadhesive suspension containing ciprofloxacin. African J Microb Res. 2011, 5(5): 579-581.

Sahoo S, Chakraborti CK, Naik S, Mishra SC, Nanda UN. Qualitative analysis of ciprofloxacin carbopol polymeric composites. Trop J Pharm Res. 2011, 10(3): 273-280. doi: 10.4314/tjpr.v10i3.14.

Siepmann J, Peppas NA. Modeling of drug release from delivery systems based on hydroxypropyl methylcellulose (HPMC). Adv Drug Deliv Rev. 2001, 48: 139–157.