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Original Article

Antimicrobial study of the optimized floating microspheres of cefixime

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ABSTRACT

Floating microspheres of cefixime were prepared by ionotropic gelation method. The method was optimized by 3^2 full factorial experimental designs. Antimicrobial activity of the formulation was evaluated by cup plate method in Muller Hinton Agar (MHA) medium. The microorganisms were inoculated aseptically by pour plate method. The zone of inhibition of the formulation was measured and compared with pure drug and the results were statistically evaluated. The results showed that the drug released from the formulation was effectively inhibiting the growth of microorganisms at concentrations of 125µg/ml and 250µg/ml. The floating microspheres of cefixime trihydrate show promising results in antimicrobial studies.

Key words: Floating microspheres, cefixime, MHA medium, microorganisms, zone of inhibition

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INTRODUCTION

Floating microspheres are multiple unit GRDDS, enabling delivery of drug at absorption site for a prolonged period of time [1,2]. The principle of buoyant preparation offers a simple and practical approach to achieve increased gastric residence time for the dosage form and sustained drug release [3]. In this study floating microspheres of cefixime trihydrate was prepared by ionotropic gelation method using polymeric carriers such as alginate and chitosan. The method was optimized by 3^2 full factorial designs and the optimized formulation was developed by setting the goals as maximizing the buoyancy and cumulative drug release%. Cefixime is an orally active 3rd generation cephalosporin antibiotic active against enterobacteriaceae, *Haemofilus influenzae*, *Streptococcus pyogenes*, *Streptococcus pneumoniae, Moraxella, E.coli, Protease, Neisseria gonorrheae* and is resistant to many β lactamases [4]. It is incompletely absorbed after oral administration which results in poor bioavailability of 40- 50% [5, 6]. So formulating into floating microspheres will prolong gastric residence time of cefixime containing formulation and allow it to float in the stomach for a long period of time which helps in increasing the oral bioavailability.

The aim of the present study is to conduct the antimicrobial activity of the optimized formulation. Antimicrobial studies were conducted to find out the drug release from the formulation and its efficacy to inhibit the growth of microorganisms. As various release retardants are used in the formulations, the effect of these on the antibacterial potency of the drug has to be tested to determine the efficacy of the formulation. Thus the optimized formulation along with pure drug was tested for the antibacterial activity. Beg S et al [7] evaluated the antimicrobial activity of bilayer tablets of amoxicillin trihydrate by agar plate diffusion method.

MATERIALS AND METHODS

Materials: Cefixime was gifted from Sance Pharmaceuticals Pala, Kerala, India. Sodium alginate was purchased from Loba Chemie Pvt.Ltd. Mumbai. Chitosan was gifted by India Sea Foods, Kochi, Kerala. Calcium carbonate was purchased from S.D fine laboratories. Muller Hinton Agar Medium was purchased from Himedia Laboratories Pvt. Ltd. Mumbai. All other chemicals used were analytical grade.

Methods:

Method of preparation and characterisation of floating microspheres: Floating microspheres were prepared by ionic gelation method. 3^2 full factorial experimental design was used to optimize the formulation of the floating microspheres. From the optimization studies 2.92 % alginate and 1.48% chitosan was selected as the optimum concentration of the poymers for the formulation to obtain maximum buoyancy and cumulative drug release percentage.

Alginate was dissolved in 100 ml distilled water. 100 mg of the drug cefixime and $CaCO_3$ (0.75:1), i.e. $CaCO_3$: alginate, w/w) was added and stirred thoroughly. The gelation medium was prepared by dissolving calcium chloride ($CaCl_2$) 0.5% w/v in 2 % glacial acetic acid. To 100 ml of the gelation medium, chitosan was added. The homogenous alginate solution was extruded using a 21G syringe needle into the gelation medium. The dropping rate was 30 drops/minute and the falling distance was 5cm. The solution containing the suspended microspheres was stirred with a magnetic stir bar for 10 min to improve the mechanical strength of the microspheres and was allowed to complete the reaction to produce gas. The microspheres were collected, washed twice with distilled water and subsequently air dried. The prepared microspheres were characterized for micromeritic properties, entrapment efficiency, buoyancy and cumulative drug release study [8].

Antimicrobial study of the optimized formulation: To conduct the antimicrobial study of cefixime in optimized formulation, *Escherichia coli. Salmonella aboney*, *Proteus vulgaris*, *Pseudomonas aeruginosa* and *Staphylococcus aureus* were used. The antimicrobial efficacy test was performed by cup plate method in Muller Hinton Agar (MHA) medium. The medium was prepared by dissolving 3.8 gm MHA and 1gm of agar to 1000 ml of distilled water and sterilized the media by autoclaving. After sterilization, MHA media (20ml) was poured into sterile petriplates and was allowed to solidify for 30 minutes. The microorganisms were inoculated aseptically by pour plate method. Using phosphate buffer, pure drug cefixime (standard) and cefixime microparticles were serially diluted to different concentrations of 125µg/ml and 250µg/ml and were added to the wells made using a sterile borer having diameter 6 mm. Phosphate buffer was used as the blank. The samples were allowed to diffuse for 30 minutes at room temperature and the plates were incubated for 24 hours at 37^0C. The diameter of the zone of inhibition surrounding each well was measured [9-12].

RESULT AND DISCUSSION

The optimized formulation along with pure drug was tested for the antibacterial activity and zone of inhibition was measured and the results are shown in table 1.

Cefixime is found to be active against, *Escherichia coli, Salmonella aboney* and *Proteus vulgaris* as shown in figures 1, 2 & 3. In each case a clear zone of inhibition of more than 10 mm diameter was observed. The study was conducted using two different concentrations of the standard and test as 125 µg /ml and 250 µg/ ml. The results were statistically evaluated by unpaired t- test. When study was performed using *Escherichia coli,* comparison of the zone of inhibition of test with standard at concentration of 125 µg/ml, the zone of inhibition was 28.2±0.65 mm and 26.6±0.72 mm and at 250 µg/ml 35.2±1.04 mm and 33.5±1.02 mm respectively. No significant difference in the zone of inhibition was observed at p values of 0.2431and 0.2360 at different concentrations. In the case of *Salmonella aboney* no significant change in the zone of inhibition was observed in both the concentrations (p values of 0.1569 and 0.299). The zone of inhibition observed at 125µg/ml was 15.6±0.87mm for standard and 12.9±0.93 mm for test. At 250 µg/ml the zone of inhibition was 21.6± 0.76 mm for standard and 18.6 ±0.45 mm for test sample. Using organism Proteus *vulgaris* also no significant difference in the zone of inhibition was observed in the concentration of 125 µg /ml (30.4±1.02 mm for standard and 28.3± 0.71mm for test) as well as 250 µg/ ml(36.4±0.078 mm for standard and 28.6±0.86mm for test). The change was not significant and p value is 0.3291 and 0.1450. No zone of inhibition was observed with *Pseudomonas aeruginosa* and *Staphylococcus aureus.* The drug released from the formulation was effectively inhibiting the growth of microorganisms at concentrations of 125 µg /ml and 250 µg/ ml which proved the potential of the formulation for treatment of infections caused by such microorganisms. Antimicrobial study proved the potential of the formulations in inhibiting the growth of microorganisms.

CONCLUSION

The purpose of antibacterial study was to find out the drug release from the formulation and its efficacy to inhibit the growth of microorganisms. As various release retardants are used in the formulations, the effect of these on the antibacterial potency of the drug has to be tested to determine the efficacy of the formulation. Thus the optimized formulation along with pure drug was tested for the antibacterial activity. The zone of inhibition was measured and statistically evaluated. Cefixime formulation was found to be active against *Escherichia coli. Salmonella aboney and Proteus vulgaris.* As cefixime was resistant to *Pseudomonas aeruginosa* and *Staphylococcus aureus* no zone of inhibition was observed with these microorganisms. It was concluded that the released drug from the formulations were effectively inhibiting the growth of microorganisms at concentrations of 125 µg/ml and 250 µg/ml which proved the potential of the formulations for treatment of infections caused by such microorganisms.

Table 1: Zone of inhibition of the optimized formulation (s_1) using different micro organisms compared with the standard.

Organism used	Concentrations used in mcg/ml				p value&level of significance (95% CI)
	125µg/ml		250 µg/ml		
	Zone of inhibition (mm)		Zone of inhibition (mm)		
	Standard (cefixime)	Test (formulation)	Standard (cefixime)	Test (formulation)	
E.coli	28.2±0.65 (A_1)	26.6±0.72$^{\$}$ (B_1)	35.2±1.04 (A_2)	33.5±1.02 (B_2)	p =0.2431(A_1&B_1)$^{\$}$ not significant p = 0.2360 (A_2& B_2)$^{\$}$ not significant
Salmonella aboney	15.6±0.87 (A_3)	12.9±0.93$^{\$}$ (B_3)	21.6±0.76 (A_4)	18.6±0.45$^{\$}$ (B_4)	p = 0.1569 (A_3&B_3)$^{\$}$ not significant p = 0.229 (A_4& B_4)$^{\$}$ not significant
Proteus vulgaris	30.4±1.02 (A_5)	28.3±0.71$^{\$}$ (B_5)	36.4±0.78 (A_6)	28.6±0.86$^{\$}$ (B_6)	p = 0.3291(A_5&B_5)$^{\$}$ not significant p = 0.1450(A_6&B_6)$^{\$}$ not significant

Un paired t- test, two- tailed. Each value is the mean of three readings ± SEM

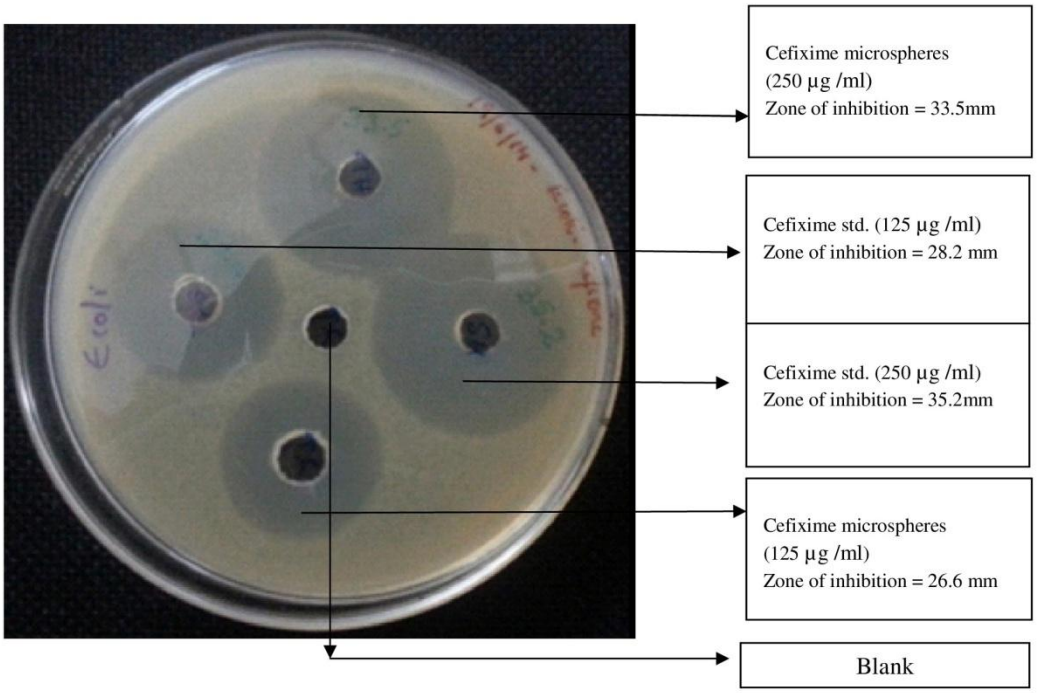

Fig.1:Antimicrobial activity of the formulation and standard drug using *Escherichia coli* as the test organism

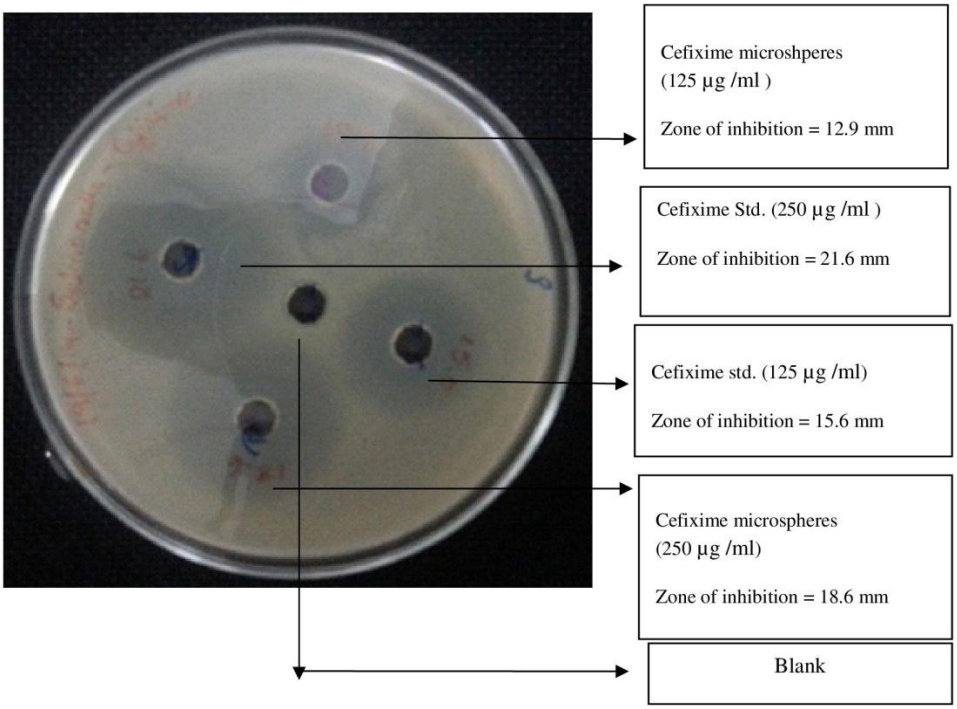

Fig.2: Antimicrobial activity of the formulation and standard drug using *Salmonella aboney* as the test organism

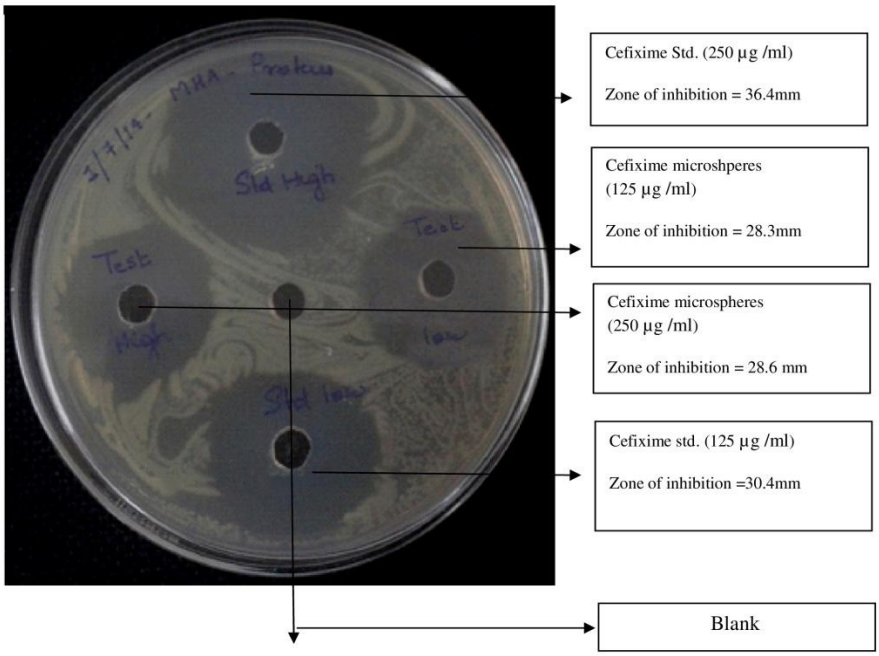

Fig.3: Antimicrobial activity of the formulation and standard drug using *Proteus vulgaris* as the test organism

REFERENCES

1. Chien Y W. Controlled and modulated release drug delivery systems in encyclopedia of pharmaceutical technology. In: Swabrick, Boyalan JC, Eds. New York: Marcel Dekker, 1990: 280-285.
2. Cremer K. Drug delivery: Gastro-remaining dosage forms. The Pharm Journal 1997; 19 (108): 259
3. Gangadharappa HV et al. Development, *In vitro* and *in vivo* evaluation of novel floating hollow microspheres of rosiglitazone maleate. Der Pharma Lettre 2011; 3 (4): 299-316.
4. Product monograph, suprax® (cefixime) 400 mg tablets.Sanofi –aventis Canada inc. 2150 St. Elzear Blvd.West Laval,Quebec H7L A48. Submission Control No.: 155702.s-a Version 5.0 dated July 6, 2012.
5. Sean L, Sweetman. Martindale Complete Drug Release. 33rd ed.; Pharmaceutical Press: UK, 2002.
6. Antony C, David MM, Osselton, Clarks BW. Analysis of Drugs & Poisons. 3rd ed.; Pharmaceutical Press: Great Britain, 2004.
7. Beg S et al. Antimicrobial activity assessment of time-dependent release bilayer tablets of amoxicillin trihydrate. Brazilian Journal of Pharmaceutical Sciences 2012; 2(48): 265-72
8. Sindhumol PG, Sudhakaran Nair CR. Formulation and optimization of floating microspheres of cefixime trihydrate by factorial design. International Journal for Pharmaceutical Research Scholars 2018; 7(1): 55-71.
9. Raosah NS, Narayan BA: Development and evaluation of once a day oral controlled multiparticulate drug delivery Systems of cefixime trihydrate. Indian J Pharm Educ Res 2008; 42 (3):283-93.
10. Kumar N et al. Xanthan gum based once a day matrix tablet of Cefixime Trihydrate: Development and evaluation. Int J Pharm Sci and Res 2013; 4 (9): 3390- 99.
11. Indian Pharmacopoeia1996.Volume II. Delhi: Controller of Publications, Government of India, Ministry of Health & Family welfare; Microbiological assays and tests; A-100- A-108.
12. Abdelmalek et al. Formulation, evaluation and microbiological activity of ampicillin and amoxicillin microspheres. J Mater Environ Sci 2014; 5 (6): 1799-1807.