

In Situ Gel Formulation of Ornidazole for the Treatment of Periodontal Disease

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Abstract

A biocompatible and biodegradable syringeable *in-situ* gel formulation of Ornidazole having controlled release characteristics for direct placement into the periodontal pocket was developed using Poloxamer 407 (Pluronic F-127) to inject without incision. Ornidazole specifically acts on gram negative anaerobic, facultative bacteria which are responsible for periodontal disease. Ornidazole requires a very low minimum inhibitory concentration to inhibit the growth of periodontal pathogens as compared to that of Metronidazole. The drug and polymer were characterized for molecular weight, solubility, refractive index, specific gravity, thermal analysis, UV Spectra analysis, hydrolysis and swellability. Pluronics, are ABA type of copolymers and it showed characteristic property of thermo reversible gelation. The drug delivery systems were prepared by two methods the hot process and cold process, cold method is preferred as lump formation takes place in case of hot process. The developed formulations were evaluated for various parameters like gelation temperature, drug content, bioadhesive strength, syringeability, viscosity, *in vitro* drug release and antibiotic activities. *In-vitro* drug release showed that SO₁ formulation released the drug completely within 8 hour. For *in-vitro* antibacterial activity, isolation, characterization, and identification of bacterial strain were carried out from dental plaque sample collected from Periodontal diseased patients. The antibiotic assay of Ornidazole gel was performed against *E.coli*, *S.aureus* and isolated coagulase negative *Staphylococcus spp.* *In-vitro* antibacterial study showed higher zone of inhibition as compared to marketed formulation. The results of study indicate that, Pluronic F-127 is promising polymer to develop *in-situ* gel formulation for periodontal disease. The formulation stored at 4°C before application, which is syringeable through 21 gauge needle. This formulation was made to inject directly in to periodontal pocket where it immediately converts in to gel form at body temperature. Usually 0.2 ml of gel formulation can be injected in to periodontal pocket.

Keywords

Periodontitis, Poloxamer 407, Gelation temperature, Bioadhesivity, Mucoadhesive, Syringeability, subgingival dental plaque

Introduction

Dental diseases are recognized as the major public problem throughout the world. Dental diseases are amongst the most widespread chronic disorders affecting the mankind. Periodontal, an inflammation of the gingival and the deeper periodontal tissues. Periodontitis is preceded and accompanied by gingivitis. However, gingivitis may persist without progressing to periodontitis leading to the loss of supporting structure of tooth and the deeper periodontal tissues which affect the supportive structures of the teeth (Lisgarten, 1987).

The periodontal pocket provides diverse environment for the colonization of gram negative facultative or obligate anaerobes like *Porphyromonas gingivalis*, *Bacteroides spp.*, *Capnocytophaga spp.* and *Actinobacillus actionomycetemcomitns*. The bacteria accumulate in the periodontal pocket that develops between the roots of affected teeth and soft tissues (Slots, et al., 1984). If the disease is allowed to progress, increased tooth mobility and possibly tooth loss may result. As a result of pathologic changes, the gingival attachment to the tooth may become displaced and a space (Fig.1) forms between this detached gingiva and the tooth is called a pocket (Medlicott et al., 1994). The ultimate result of progressive pocket

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formation leads to bone loss and tooth mobility. Systemic administration has been useful in treating periodontal pockets, but repeated and long term use of systemic drugs is fraught with potential danger including resistant strains and superimposed infections. To overcome these disadvantages of conventional preparations, an attempt has been made in the present work to develop and evaluate suitable drug delivery system with the aim of improving patient compliance, therapeutic efficacy, reduced dosage regimen and targeted drug action with mucoadhesive and biodegradable polymeric systems. Local administration provides a useful answer to these problems. Ornidazole has shown a marked antibacterial and antiprotozoal activity under *in vitro* conditions and also in laboratory animals. The antibacterial spectrum of ornidazole includes bacteria such as the Peptostreptococcus species, Clostridium difficile, the clostridium species, Bacteroids species, Bacteroids fragilis, Prevotella species, Porphyromonas species, Fusobacterium species, Actinomycetes, Propionibacterium species and the Eubacterium species. The drug is highly effective against various protozoa, namely trichomonas vaginalis, Entamoeba histolytica, and Giardiasis intestinalis. Bioadhesion may be defined as the state in which two materials, at least one of which is of a biological nature, are held together for extended periods of time by interfacial forces. For drug delivery purposes, the term bioadhesion implies attachment of a drug carrier system to a specific biological location. The biological surface can be epithelial tissue, or the mucus coat on the surface of a tissue. If adhesion attachment is to a mucus coat, the phenomenon is referred to as mucoadhesion (Duchene et al., 1988). Recently, many researchers have focused their attention on placing a drug or drug delivery system in a particular region of the body by using bioadhesive hydrophilic polymers to control the delivery of biologically active agents systemically or locally (Chen et al., 1992). Mucoadhesive drug delivery system is one of such drug delivery system. Biodegradable polymers have been used in drug delivery devices. Because of their biodegradability, there is no need to remove the drug depleted delivery devices. The use of bioabsorbable delivery devices represents a major step forward in the treatment of periodontal diseases. There remains a need in the art for the device, which can be prepared more quickly and easily placed in a

periodontal pocket for the release of beneficial agent for longer duration in order to treat the disease condition

Materials and Methods

Materials

Ornidazole and Pluronic F-127 were obtained from Wockhardt Ltd (Aurangabad, India.). Carbopol 934, 934 P, 940, HPMCK-100, PVPK-30, Silicon dioxide was of pharmacopoeial grade purchased from Research Lab (Mumbai, India). Dialysis Membrane MWCO 12000 Da .Purchased from Hi Media (Mumbai, India) Deionised water was used as the aqueous component.

Preformulation studies

Tests were carried out on the sample of the drug to establish its identity and purity as per specification reported in European Pharmacopoeia. The parameters studied include appearance, solubility in different solvents, melting point, infrared spectra, determination of λ_{max} in saline phosphate buffer pH 7.4 and ethanol. UV spectra were measured on Jasco V- 630 UV-VIS (Jasco Corporation, Tokyo, Japan) double beam spectrophotometer to construct the standard curve for Ornidazole in phosphate buffer pH 7.4. (Chandur et al, 2007, Kastureet al, 2004) and in ethanol at 320nm (Table 9 and Figure 20 Table 10 and Figure 21 respectively).The melting point of Ornidazole and poloxamer was DSC studies (Figure 18 and 19). Infra red spectra were analyzed by FTIR (JASCO-FTIR 5300, Tokyo, Japan).

Preparation of gel formulation

There are two preparation methods of gels, cold method and hot method. Both methods of preparations generally yield gels with comparable properties. Cold method is preferred. In case of hot process, lumps formation of polymer occurs.

Cold process

Gels were prepared on a weight basis using a cold process. Carefully weighed an amount of Pluronic F-127 sufficient to yield 20% was slowly added to cold water (5°C); constant stirring was maintained. Each dispersion was then refrigerated until a clear solution is formed (5hr's). Active substances that are insoluble in water are dissolved prior to addition in Ethanol, Isopropyl alcohol or Propylene glycol at 5°C to form a homogeneous mass.

Hot process

Pluronic F-127 dissolved in water approximately at 70°C. Active substances that are insoluble in water are dissolved in Ethanol, Isopropyl alcohol or

Propylene glycol at 70°C and mixed with warm aqueous phase to form a homogeneous mass before addition. The gel forms when the solution cools to room temperature.

Characterization of formulations

Various formulations prepared were characterized for the following properties; Gelation temperature by Miller and Donovan technique (Miller et al., 1982). Bioadhesivity were studied (Perioli et al, 2004, Jadhav et al, 2004) in a locally assembled apparatus and was a modification of the apparatus previously applied by Paroli et al, 2004. (Photograph No.2), Mucoadhesive force strength were also determined and reported, surface pH of the gel determined by the method similar to Bottenburg et al, 1991. The viscosity of gels were measured using Brookfield Viscometer CAP 2000⁺ 1.10 programmable viscometer (Brookfield Engineering Laboratories Inc, Middleboro, MA) and Syringeability Study (Mahishwari et al, 2006) was performed as the product must be able to delivered from a syringe through 21 gauge needle, in order to fulfill the requirement of ease of application.

Estimation of drug content in gel formulations

Formulations containing 1 mg drug was taken in 10 ml volumetric flask, dissolved in Ethanol, made up the volume to 10 ml with ethanol and then filtered. Absorbance values were measured with suitable dilutions at (λ_{max}) 320 nm. Concentrations of drug were calculated from the standard calibration curve prepared in Ethanol (Ahuja et al, 2006).

In vitro drug release

The release studies were performed (Mahishwari et al, 2006) using the dialysis method. Typically, 1g of pluronic gel was placed in a dialysis tube (MW 12000 cutoff). The dialysis tube (2.3cm in diameter) was then placed in a vessel containing 100ml of phosphate buffer pH 7.4, (simulated pH of GCF) stirred at 100 rpm. Samples were collected periodically and replaced with fresh medium of phosphate buffer pH 7.4. After filtration through the Whatman filter paper 41, the concentration of Ornidazole was determined spectrophotometrically at 320 nm. The kinetic analysis of release data was done using PCP –Disso 2.08 software.

In-vitro antibacterial activity

Out of various micro flora we have selected E.coli, S.aureus. (Ahuja et al, 2006) The standard culture of E.coli and S.aureus obtained from Department of Microbiology, Maulana Azad College of

Postgraduate and Research Centre, Aurangabad. Media used were Blood agar (Mackie et al, 1979) and Nutrient agar. Flora obtained were gram stained and Gram positive coccus in cluster was observed. No zone of hemolysis was observed around the colonies and Non diffusible yellow pigmented colonies were observed, cocci in cluster were observed. No coagulation was observed in test when compared to positive and negative control.

Antibiotic assay

Most of microorganisms exhibit great variation in susceptibility to antibiotics and chemotherapeutic agent. This is particularly in the case of S. aureus and many gram negative bacilli. Hence it is essential to determine the susceptibility of isolates to antibiotic that are likely to be used in treatment.

Antibiotic sensitivity tests are of two types

- Diffusion tests (Lipipun et al, 2002): Here the test bacterium is seeded in the medium and wells are digged into the agar plate with sterile borer. Then add antibiotic into it. Allow to diffuse the drug and observe the effect of drug on seeded bacterium in the form of zone of inhibition
- Dilution tests (Rautenbach, 2000): is used to find out the minimum inhibitory concentration of drug on test organism. Here, serial dilutions of drug are prepared and inoculate with test bacterium. After incubation, read MIC by noting the lowest concentration of the drug that inhibits growth. Then take tube which do not shows growth and inoculate it in fresh medium not containing drug. Then keep tube for incubation. If turbidity observed, the effect of drug supposed to be bacteriostatic and if no growth observed it means that drug shows bactericidal effect. This process is called as a minimum Bactericidal Concentration i.e. MBC. Observations were computed in table 19 and photograph 9 to 17 Out of all formulations, formulation SO11 shows maximum antibacterial activity on both microorganisms.

Results and Discussion

In the present investigation, an attempt was made to develop and evaluate mucoadhesive, syringeable in-situ gel formulation of Ornidazole having controlled release characteristics for direct placement into the periodontal pocket. Ornidazole specifically acts on gram negative anaerobic, facultative bacteria which

are responsible for periodontal disease. Ornidazole requires a very low minimum inhibitory concentration to inhibit the growth of periodontal pathogens (Ellie et al, 1978) as compared to that of Metronidazole (Metrogel 1% Marketed formulation). Preformulation study of drug was carried out as per literature survey and official text. In this study, the thermo reversible polymer, Pluronic has been used for formulation of *in-situ* gel of Ornidazole. The developed formulations were evaluated for various parameters like gelation temperature, drug content, bioadhesive strength, syringeability, viscosity, *in vitro* drug release and antibacterial activity. For *in-vitro* antibacterial activity, isolation, characterization, and identification of bacterial strain were carried out from dental plaque sample collected from Govt. Dental College Aurangabad. The antibiotic assay of Ornidazole gel was performed against *E.coli*, *S.aureus* and isolated coagulase negative *Staphylococcus spp.* All the developed formulations shows significant zone of inhibition against *E.coli* and isolate coagulase negative *Staphylococcus spp.* *In-vitro* antibacterial study shows that, all formulations of Ornidazole gel (0.5%) shows significantly higher zone of inhibition as compared to marketed formulation and out of all developed formulations, SO₁₁, shows highest zone of inhibition. The results of study indicate that, Pluronic F-127 is promising polymer to develop *in-situ* gel formulation for periodontal disease. The formulation stored at 4°C before application, which is syringeable through 21 gauge needle. This formulation is directly injected in to periodontal pocket where it will immediately convert in to gel form at body temperature. Usually 0.2 ml of gel formulation can be injected in to periodontal pocket.

Preformulation studies

To confirm the identity, purity and suitability of drug for formulation and to establish a drug profile, preformulation studies were undertaken which includes: Description, Solubility in water and phosphate buffer pH 7.4 and were found to be 17.35 and 17.45mg/ml in deionized water and Phosphate buffer pH 7.4 respectively. The identity and purity of the drug and polymer were confirmed by FTIR (fig1 and 2) and DSC (fig3 and 4) thermogram. Melting point was determined and was found to be 90°C. In preformulation study the I.R. spectrum obtained for drug alone and with a formulation excipient revealed that there is no interaction

between drug and formulation excipient .
Preparation of standard calibration curve of Ornidazole in phosphate buffer pH 7.4 and in ethanol

Standard Calibration curve of Ornidazole was prepared in phosphate buffer pH 7.4. PBS pH 7.4 simulates GCF thus facilitates the extrapolation of results to *in vivo* conditions. It was also prepared in ethanol to ascertain the drug content in the *in situ* gel formulations. The graded concentrations were chosen within the range in of beer-lambert's law (fig 5 and 6)

Preparation of gel formulation

Gel was prepared on a weight basis using the cold method. An amount of Pluronic F-127 sufficient to yield 20% gel was slowly added to cold water (5°C); constant stirring was maintained. Prepared dispersion was refrigerated until a clear solution was formed (5hours). To control the release of drug from the gel, the different concentration of mucoadhesive polymers like Carbopol 934P, Carbopol 940, HPMCK-100, PVPK-30 Silicon dioxide were added to pluronic gel .The concentration of drug (5mg/gm) Ornidazole was kept constant in all the batches. Table 1 represents Ornidazole gel formulation

Characterization of formulations

The gelation temperature: The gelation temperature of gel decreases with increasing the concentration of mucoadhesive polymers

Determination of drug content uniformity of gel formulations: Known amount of gel containing 1mg of drug was dissolved in ethanol. The solutions were analyzed for drug content spectrophotometrically at λ_{max} 320 nm. The drug content was estimated by measuring the amount of drug present in known quantity of gel. All the formulations exhibited fairly uniform drug content. This ensures intended delivery of drug to the site after administration of the gel formulation. Results revealed that drug content of all developed formulations were in the range of 97 to 99%.

Surface pH measurement: The values were found to be well within the range of neutral pH. This indicates that formulation can be used, and may not cause any irritation in the oral cavity. . The pH of formulation affects the bioadhesive strength, as pH of the formulation decreases, the bioadhesive strength increases or vice versa.

Bioadhesive strength: Mucoadhesive formulations must maintain intimate contact with mucus layer

overlying the epithelial tissue. This parameter is very critical for successful utilization of these dosage forms. So *in-vitro* evaluation of the *in situ* gel was carried out using bovine buccal mucosa. This gives the indirect measurement of bioadhesive strength in grams. Mucoadhesive strength of formulation containing Carbopol 940 showed higher mucoadhesion. Higher mucoadhesive strength of Carbopol 940 may be due to higher percentage of carboxylic acid groups which gradually undergo hydrogen bonding.

Viscosity studies: Viscosity varies with the concentration of polymer. As the concentration of polymer increases, viscosity increases proportionally. As 20% gel offers a good syringeability and optimum viscosity characteristics (Mahishwari et al, 2006), therefore 20% gel has been selected for the present study. Viscosity primarily affects the release of drug from the gel, as viscosity increases the drug release decreases from the gel. It is seen that as the concentration of mucoadhesive polymer increases, viscosity increases proportionally. Pluronic gel shows thermo reversible property. The Pluronic gel at cold temperature converts in to liquid form. As the temperature of the system increases the liquid form converts in to gel at room temperature. Thus pluronic gel shows the temperature dependent thixotropic behavior. This sol-gel transition characteristic plays an important role in development of *in-situ* gel formulations. The viscosities of all the developed gel formulations at 25°C and 37°C were in the range 1200 to 1400 CP and 8000 to 9000 CP respectively.

Syringeability Study (Mahishwari et al, 2006): All the developed formulations were easily syringeable through 21-gauge needle. Results revealed that all the formulations from SO₁ to SO₁₃ were syringeable at cold temperature

***In vitro* drug release**

The prepared *in situ* gel implants are intended for placement in the periodontal pocket. Gingival crevicular fluid (GCF), inflammatory exudate flows continuously in the pocket. The pH of GCF⁹ is 7.2 to 7.6 and a mouth saliva pH 6.4 to 7.4. Hence in the present study, phosphate buffer saline pH 7.4 was used for the *in-vitro* drug release studies of the gel formulations. The release studies were performed using dialysis membrane MWCO 12000 Da. On addition of various mucoadhesive polymers like Carbopol 934, 934P, 940, HPMC K-100, PVP K-30

and Silicon dioxide, the release decreases to an extent level. The result of cumulative percent release is depicted in Table 2 and Figure 7 to Figure 10. *In-vitro* drug release shows that SO₁ formulation released the drug completely within 8 hour. Addition of mucoadhesive polymers extended the drug release up to 14 hr's from the gel formulations. Amongst all developed formulations SO₉, SO₁₁, SO₁₂ and SO₁₃ were selected on the basis of their control release characteristics.

The *In-vitro* antibacterial activity of Ornidazole gel (0.5%)

Kinetic analysis of the *in vitro* release data of Ornidazole from gel formulations are presented in Table No.19. The *in vitro* release data are in favor of Peppas release kinetics for formulations SO₃, SO₄, SO₅, SO₆, SO₇ and SO₁₃. Hixon crowell release for formulation SO₁. First order release kinetics for SO₂, SO₈, SO₉ and SO₁₁ formulation. Formulation SO₁₀, SO₁₂ seems to be Matrix model. Peppas used n value in order to characterize different drug release mechanisms, concluding that n= 0.5 in case for Fickian diffusion, n values falls between 0.5 and 1 for Anomalous transport, n =1 for Case- II transport and for n values higher than 1 for Super –case II transport. In present study, an attempt has been tried to isolate the pathogen that is responsible for the periodontal disease. A subgingival dental plaque sample of a patient suffering from periodontal disease was taken. Gram staining was carried out on that plaque sample, gram positive cocci in clusters were observed. The gram positive cocci are generally *Staphylococcus spp.* They can grow up in aerobic as well as anaerobic condition. The dental plaque samples were streaked on blood agar, which is a selective media to isolate periodontal pathogen. Here inoculated plates were placed inside a large airtight container and a lightened candle kept in it before the lid is sealed. The burning candle is expected to use up all the oxygen inside before it is extinguished and an anaerobic condition was maintained in a jar. Next day colony characteristics were recorded and gram staining was performed. No zone of hemolysis was observed around the colonies. It means that *Staphylococcus* species not active against human erythrocyte. To study the pigment characteristics of an isolate, a culture were streaked on nutrient agar media, after 24 hours non diffusible yellow pigmented colonies were observed and cocci in cluster were observed on gram staining.

It may be concluded that the person is suffering from *Staphylococcal* infection. Coagulase test were performed on isolated sample, showed negative result. Thus it is concluded that the isolated bacterial strain is coagulase negative *staphylococcus spp.* Thus isolated culture may be *S.epidermis* or *S.saprophyticus* (Ananthnarayan, 1978).

In-vitro antibiotic activity of Ornidazole gel (0.5%)
Antibiotic assay of Ornidazole gel was performed by cup and plate method on *E.coli*, *S.aureus* and

coagulase negative *Staphylococcus spp.* From the results it is seen that the standard culture of *S.aureus* is resistant to Ornidazole gel (0.5%) and Metrogel (1%), but *E.coli* and coagulase negative *Staphylococcus spp.* were shown significant zone of inhibition. Ornidazole requires a very low minimum inhibitory concentration (Ellie et al, 1978) to inhibit the growth of periodontal pathogens as compared to that of Metronidazole (Metrogel 1% Marketed formulation).

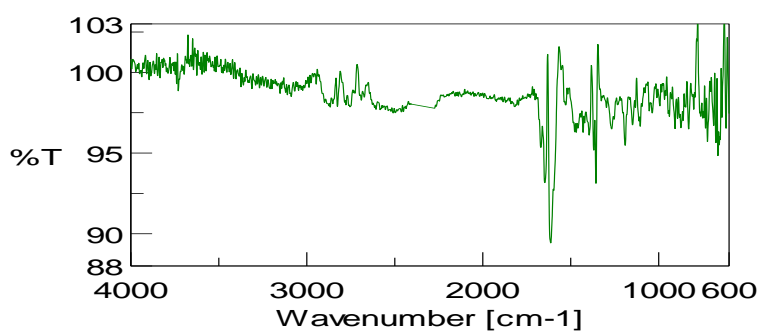


Figure 1: IR spectra of Ornidazole

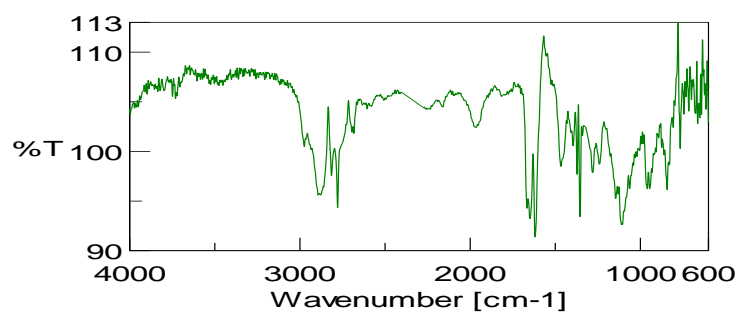


Figure 2: IR spectra of Poloxamer 407

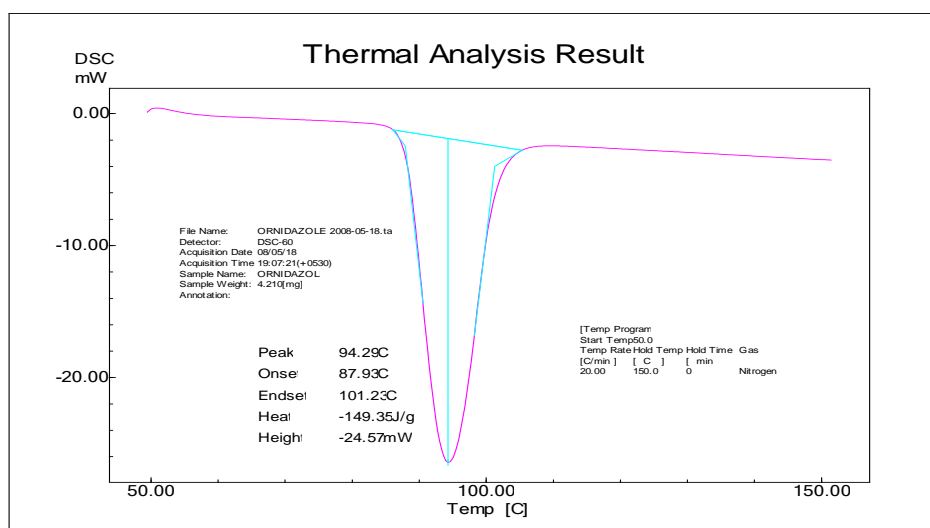


Figure 3: DSC Study of Ornidazole

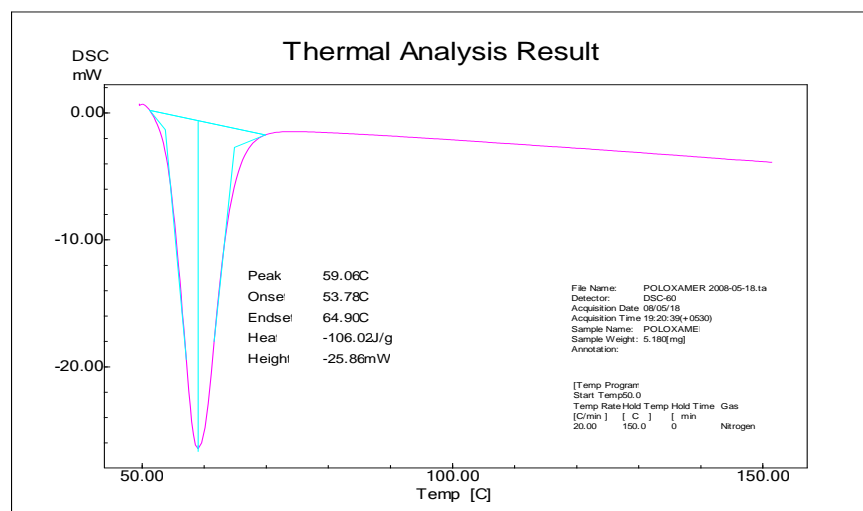


Figure 4: DSC Study of Poloxamer

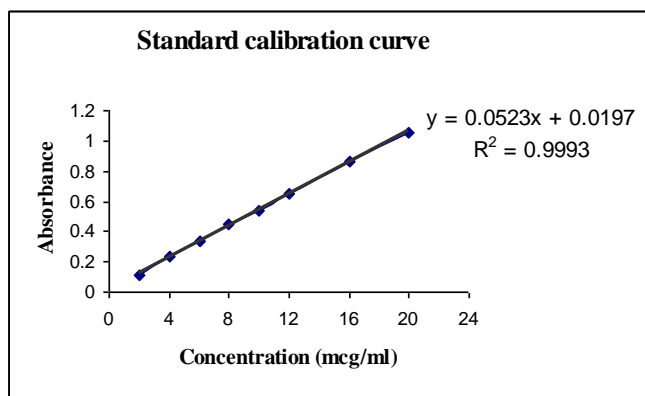


Figure 5: Standard curve of Ornidazole in phosphate buffer pH 7.4

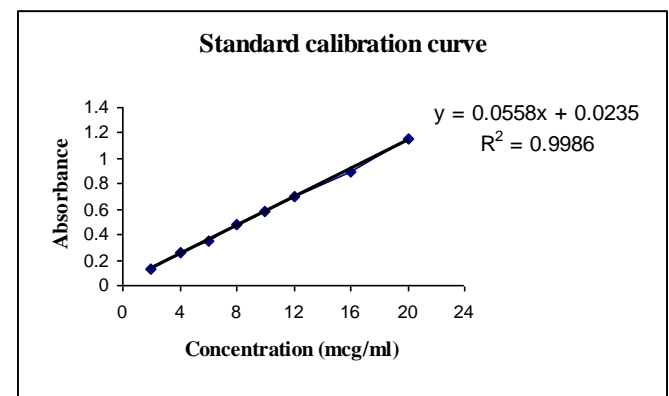


Figure 6: Standard curve of Ornidazole in ethanol at 320nm

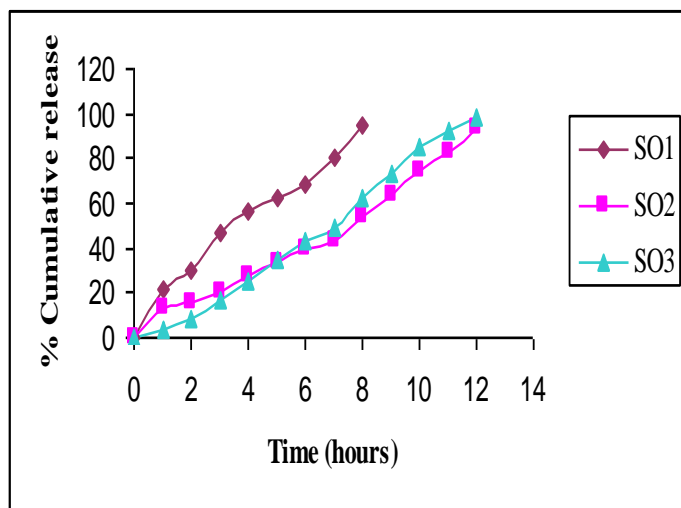


Figure 7: % Cumulative release of SO1-SO3 formulations

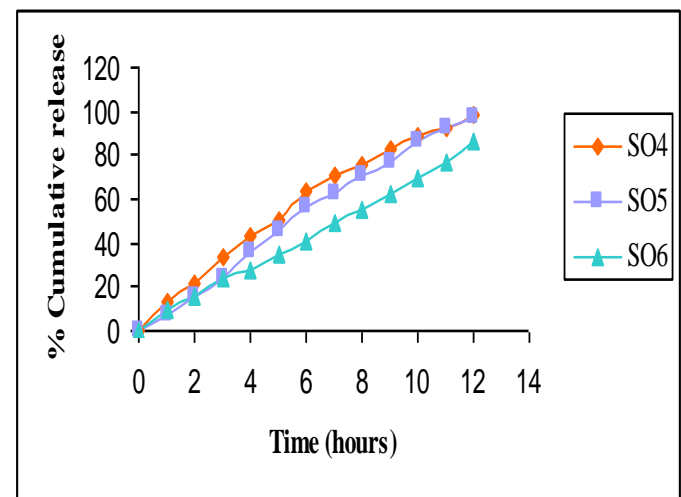


Figure 8: % Cumulative release of SO4-SO6 formulations

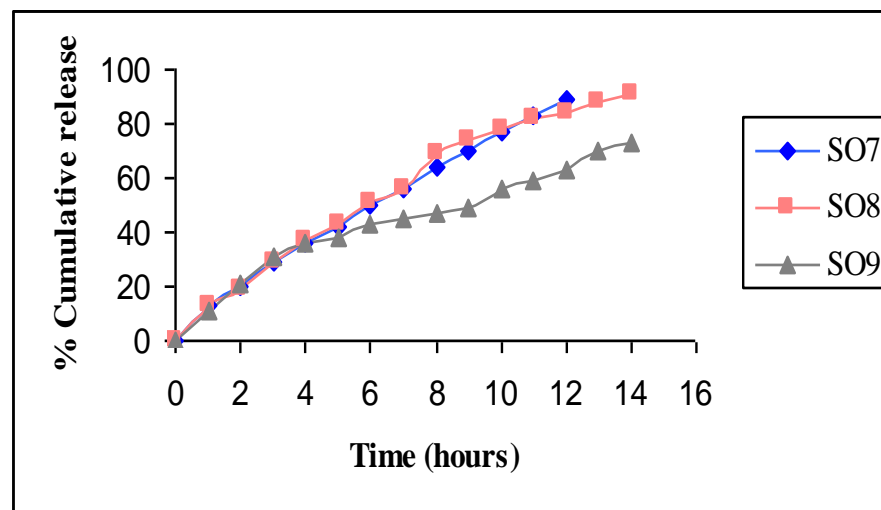


Figure 9: % Cumulative release of SO7-SO9 formulations

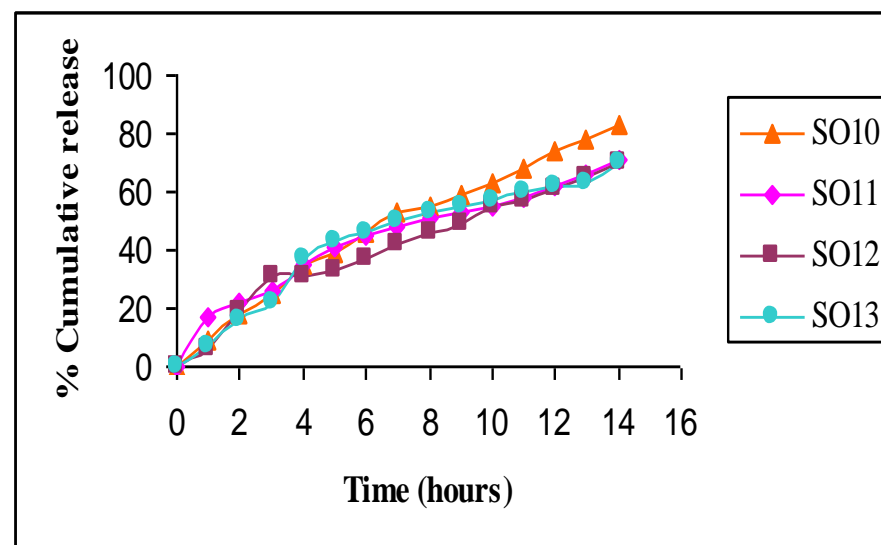


Figure 10: % Cumulative release of SO10-SO13 formulations

Table 1: Composition of In -situ gel formulation of Ornidazole

Formulation code	Drug *	Pluronic gel	PVP K-30	Carbopol 940	Silicon dioxide	Carbopol 934P	Carbopol 934	HPMC K-100
SO1	5 mg	20%	-	-	-	-	-	-
SO2	5 mg	20%	-	0.2%	-	-	-	-
SO3	5 mg	20%	-	-	-	0.2%	-	-
SO4	5 mg	20%	-	-	-	-	0.2%	-
SO5	5 mg	20%	-	-	-	-	-	0.2%
SO6	5 mg	20%	0.2%	-	-	-	-	-
SO7	5 mg	20%	-	-	0.2%	-	-	-
SO8	5 mg	20%	-	0.4%	-	-	-	-
SO9	5 mg	20%	-	-	-	0.4%	-	-
SO10	5 mg	20%	-	-	-	-	0.4%	-
SO11	5 mg	20%	-	-	-	-	-	0.4%
SO12	5 mg	20%	0.4%	-	-	-	-	-
SO13	5 mg	20%	-	-	0.4%	-	-	-

(*0.5 % Ornidazole gel)

Table 2: Kinetic analysis of the release data of Ornidazole from gel formulations

Formulation code	R					Best fit model	Parameters for Kosrmeypappas equation	
	Zero order	First order	Matrix	Peppas	Hix.Crow		K	n
SO1	0.9787	0.9685	0.9887	0.9743	0.9899	Hix.Crow	24.84	0.59
SO2	0.9829	0.9908	0.9766	0.9691	0.9890	First order	10.54	0.7
SO3	0.9904	0.9894	0.9438	0.9940	0.9907	Peppas	4.14	1.26
SO4	0.9585	0.9774	0.9700	0.9813	0.9732	Peppas	13.38	0.85
SO5	0.9934	0.9914	0.9587	0.9967	0.9949	Peppas	7.49	1.09
SO6	0.9829	0.9931	0.9832	0.9955	0.9905	Peppas	9.68	0.77
SO7	0.9649	0.9851	0.9904	0.9918	0.9794	Peppas	13.28	0.68
SO8	0.9213	0.9850	0.9684	0.9512	0.9718	First order	22.02	0.73
SO9	0.9719	0.9885	0.9783	0.9778	0.9848	First order	10.06	0.87
SO10	0.9451	0.9686	0.9879	0.9756	0.9617	Matrix	12.59	0.66
SO11	0.9817	0.9916	0.9723	0.9771	0.9913	First order	14.74	0.76
SO12	0.9466	0.9679	0.9775	0.9438	0.9614	Matrix	8.58	0.85
SO13	0.9566	0.9665	0.9637	0.9795	0.9638	Peppas	8.38	0.92

*R is Coefficient of correlation, K is a constant.

Table 3: Comparitive susceptibility of bacterias for metronidazole and ornidazole⁹⁶

Bacteria	No.of isolates	Drug	MIC (mcg/ml)
<i>B.fragilis</i>	39	Metronidazole	1.8
		Ornidazole	1.6
<i>Clostridium perfringes</i>	9	Metronidazole	1.6
		Ornidazole	0.8
<i>Actinomyces</i>	15	Metronidazole	0.1
		Ornidazole	0.1
<i>Porphyromonas gingivalis</i>	12	Metronidazole	4
		Ornidazole	2
Anaerobic gram positive cocci	24	Metronidazole	0.8
		Ornidazole	0.4

Table 4: *In-vitro* antibacterial activity of Ornidazole gel formulations (0.5%)

MEDIUM		MEDIUM	
Nutrient Agar (Isolated coagulase negative <i>S.Spp.</i>)		Nutrient agar(<i>E.coli</i>)	
Formulation code	Diameter of zone of inhibition (cm)	Formulation code	Diameter of zone of inhibition (cm)
SO ₁	2.4	SO ₁	3
SO ₉	2.8	SO ₉	2.2
SO ₁₁	2.9	SO ₁₁	2.9
SO ₁₂	3.1	SO ₁₂	2.3
SO ₁₃	2.5	SO ₁₃	2.5
Marketed formulation (Metrogel 1%)	2.3	Marketed formulation (Metrogel 1%)	2.1
Placebo	0	Placebo	0

Conclusion

Ornidazole, one of nitroimidazole derivative was selected as drug candidate for the present investigation. Ornidazole specifically acts on gram negative and gram positive anaerobic, facultative bacteria like *Porphyromonas gingivalis*, *Fusobacterium species*, *Bacteroids fragilis*, *S.aureus* and *S.mutans*. Ornidazole has been the most widely used in the management of protozoal anaerobic infection. The antimicrobial activity of Ornidazole has been proposed due to the reduction of nitro group to a more reactive amine that attacks microbial DNA, inhibiting further synthesis and causing degradation of existing DNA. Thus, Ornidazole as local drug delivery may be an advantageous form of treatment since it would probably eliminate side effects, which occur with systemic dosing. So in the present research work, an attempt has made to formulate *in-situ* gel of Ornidazole for the effective management of periodontitis with local delivery into the periodontal pockets. *In situ* gel implants of Ornidazole with mucoadhesive polymers – Pluronic F-127, HPMC K-100, Carbopol 934P, Carbopol 934, Carbopol 940, PVP-K-30 and Silicon dioxide were formulated. The major advantage of Pluronic F-127 is its Thermo reversible nature that used for *in-situ* gel formulation. From the present study it can be concluded that: a) The developed formulation is having enough bioadhesive property, it will remain in to the cavity for sufficient time, which can again protected by periodontal dressing (Coe-pak). b) The local drug delivery system in present study is simple and easy to use. Its syringeability allows easy insertion of gel formulation in to periodontal pocket. c) The developed formulation can release the drug at controlled rate for prolonged duration. d) The results

indicate that these targeted devices for the treatment of periodontal diseases show significant advantages over the conventional therapy. e) Effective and prolonged local levels of an anti-microbial could be achieved without much systemic load with comparatively less frequency of administration. f) This type of drug delivery system can serve as a novel approach for treating periodontal diseases with better patient compliance.

Future prospective: Further optimization and stability studies as per ICH guidelines. Pre-clinical studies in animals like beagle dog and guinea pig. Clinical evaluation of gel formulation and IVIVC studies. An *in vivo* study of the formulation is necessary factor to evaluate the efficacy and safety of the novel drug delivery system.

Reference

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