ISSN:0975-1459
Journal of Pharmaceutical Sciences and Research
www.jpsr.pharmainfo.in
Jayaraj Kumar, K. et al, /J. Pharm. Sci. & Res. Vol.2(2), 2010, 116-122

Formulation of Thermoresponsive and Buccal Adhesive In Situ Gel for Treatment of Oral Thrush containing Itraconazole

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Abstract:
The aim of the present work is to formulate and evaluate in situ oral topical gels of itraconazole based on temperature induced systems for the treatment of oral condidiasis. . The system using polaxamer (10% to15%w/w) along with carbopol 934 (0.01 to 0.4 % w/w) and Itraconazole (1%w/w). The formulations were evaluated for physiochemical parameter, gelation Temperature, viscosity, gel strength, content uniformity mucoadhesive force, FTIR and DSC.

Keyword: Temperature induced, topical in situ gel, oral candidiasis, Itraconazole.

Introduction:

Candidiasis is an opportunistic infections condition caused by ubiquitous, saprophytic fungi of the genus Candida which includes eight species of fungi, the most common of which is Candida the most common of which is Candida albicans. Oral candidiasis is one of the most common pathological conditions affecting the oral mucosa[1, 2.] Local delivery of drugs to the tissue of the oral cavity has a number of applications including the treatment of toothache, periodontal diseases, dental caries, bacterial and fungal infections. The conventional formulations for the local delivery of drugs to the oral cavity are the mouth paints, rinses, troches, creams and suspensions. One way to improve the efficacy in eradicating the infection is to deliver the antifungal locally in the oral cavity are the oral cavity. Better stability and longer residence time will allow more of the antifungal to penetrate through the oral mucous layer to act on Candida species. The reason for incomplete eradication of candidacies in most cases may be due to the short residence time of antifungal agents in the oral cavity. The other reason may be degradation of antifungal agents in salivary fluid. Therefore, researchers have prepared and reported new formulation such as gels, mucoadhesive tablets, PH sensitive, temperature induced in situ gel excipients composition mucoadhesive microspheres, etc.,Pyrrolidone and its derivatives are reported to interact with both keratin[12] and with lipids[13] in the skin. Azone is known to show significant accelerant effects at low concentration for both hydrophilic and hydrophobic[14] drugs and is one of the few enhancers that have been developed commercially. Differential scanning calorimetric studies have shown that azone affects lipid structure of stratum corneum[15]. In addition, azone is reported to decrease transition temperature with in lipid bilayer to induce formation of a liquid phase with a resultant increase in lipid fluidity[16].

Surface active triblock copolymers of polyoxyetcylne– polyoxypropylene – polyoxyethylene (PEOn – ppo_n – PEO_n) (pluronics) are used in medical, pharmaceutical and cosmetic systems. The unique characteristics of this copolymer are reverse thermal gelation: concentrated solution (20-30% w/w) of the polymer are fluid at refrigerator temperature (4-5˙)˙but are soft get at body temperature. The core of the micelles is hydrophobic consisting of polypropylene oxide, while a hydrophilic corona consists of ethylene oxide self assembly is temperature depended and at a given polymer concentration a critical mica temperature (CMT) exists[3]. Pluronics are available in wide range of molecular hydrophilic regions. The high solubilizing capacity and non-toxic properties of pluronics make if suitable for drug delivery.

Pluronic is more soluble in cold water than not water. The hot solution process has been attributed to excessive hydrogen bonding between water molecules and ethereal oxygen of the polymer. The pluronics foliations are transformed from low viscosity transparent solutions at 50^{o}C to solid gels on heating to body temperatures. The temperature dependent gelling process is micellar in nature, being constructed from cubic orientation of micellar subunits[5]. The micellar mode of association has been useful as drug delivery systems[6]. The aqueous solution of pluronic is known to exhibit the phenomenon of reverse thermal gelation, remaining as solution at low temperature.[7, 8] the reversal thermal gelation exhibited by pluronic aqueous solutions has been used as drug delivery system for Opthalmic, [9] Partentral[10], rectal[11] and percutaneous use[12].

Material:

Poloxamer 188 were kindly gifted by pharmaceutical Pvt, (Bombay) Itraconazole was used as a standard dry carbopol 934 and 2- ethyl 2 -methyl pyrolidone was supplied by Merck, Mumbai, methyl paraben were gifted by fourt's India Chennai ,all other chemical used were of analytical grade.

Method:

Different formulations were prepared with various ratios of polaxmer 188 and carbopol 934 (10% to 15% w/w**):** (0.01% to 0.4% w/w)

Step I: The method involved slow addition polaxmer 188 and methyl paraben were solubilized in required quantity of cold distilled water.

Step II : Required quantities of carbopol 934 were kept overnight for swelling.

The polymer solution taken in a beaker with continuous stirring (magnetic stir) until uniform solution obtained.

Step III: After the mixture had been kept at ambient Temperature for 24 hrs. a small amount of trienthanolamine was added to adjust the PH 7.

Step IV: An appropriate amount of Itraconazole Solubilized in physiologically compatible solvent such as 2 – methyl – 2 pyrrolidone with continues stirring until uniform Drug solution obtained. Thermo reversible gels were prepared using cold technique[17.]

Step V: Drug solution was added to this preformed gel before invitro studies.

Table 1: Composition of optimized formulation

S.No	Ingredient	Optimized formulation		
		k1 w/w	k6 w/w	k7 w/w
1	Itraconazole	1%	1%	1%
2	Polaxamer 188	15%	15%	15%
3	Carbopol 934	---	0.02	0.03
4	2-ethyl-methyl-2 pyrrolidone	q.s	q.s	q.s
5	Methyl paraben	0.001	0.001	0.001
6	triethanolamine	q.s	q.s	q.s
7.	Cold distilled water	q.s	q.s	q.s

Evaluation of buccal gel

Preliminary evaluation of gel (gelation temperature): The different formulations of poloxamer 188 and poloxamer 188**:** Carbopol 934 combinations were evaluated for geletion temperature. The gelation temperature was determined by heating the solution (1-2^{o}c) min in a test tube with gentle stirring until gel was formed. The gel was said to have formed when there was no flow after container was overturned[18].

Determination of PH: The PH of the gel was determined using a calibrated pH meter. The readings were taken for average of 3 samples.

Measurement of Gel Strength: A sample of 50 Gms of gel was placed in a 100ml graduated cylinder and gelled in a thermostat at 37c.

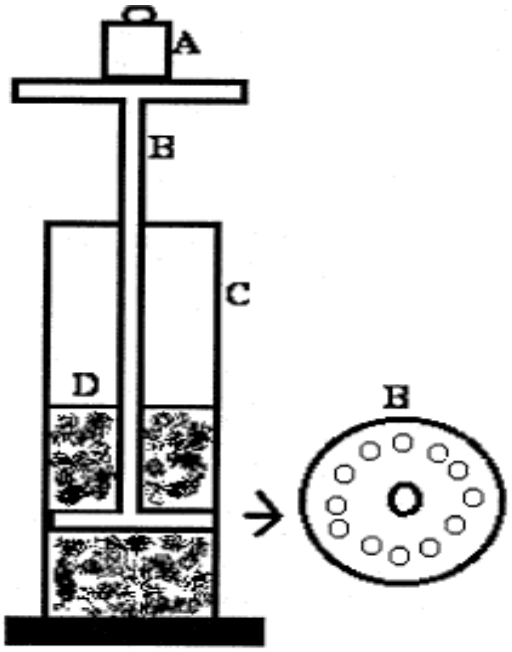


Fig. 1. Gel strength-measuring device, (A) weights; (B) device; (C) mess cylinder; (D) poloxamer gel.

The apparatus for measuring gel strength (weigh or apparatus as shown in figure 1, weighing 27 gm) was allowed to penetrate in polaxamer 188 gel. The gels strength, which means the viscosity of the gels at physiological temperature, was determined by the time (seconds), the apparatus took to sink 5cm down through the prepared gel.

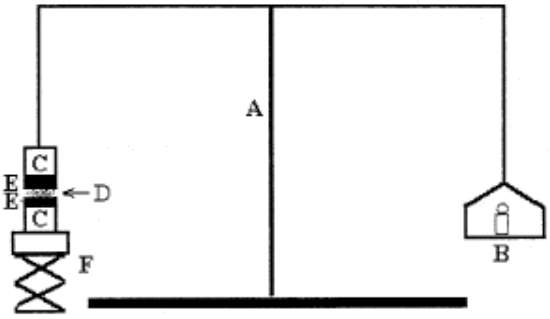


Fig. 2. Bioadhesive force-measuring device, (A) modified balance; (B) weights; (C) glass vial; (D) poloxamer gel; (E) rectal tissue; (F) height-adjustable pan.

Viscosity Studies: The rheological studies were carried out using Brookfield programmable DVII+Model pro II type (USA). The gel under study was placed n a small sample holder with a facility of water circulation. Water was circulated in the jacket with the help of Water immersion pump. Initially ice-cold water was circulated and then hot water to raise the temperature gradually. The temperature – sensing probe was lowered in the gel was recorded. Spindle number SS64 was lowered vertically in it. The spinnle was rotated at varying speed.

Determination of mucoadhesive Force: The mucoadhesive force of all the optimized batches was determined as follows, a section of mucosa was cut from the chicken cheek portion and instantly fixed with mucosal side out onto each glass vial using rubber

band. The vial with chicken cheek mucosa was connected to the balance in inverted position while first vial was placed on a height adjustable pan. Oral gel was added onto the nasal mucosa of first vial. Then the height of second vial was so adjusted that the mucosal surfaces of both vials come in intimate contact.

Table 2: Physicochemical Evaluation of Itraconazole solution

Physicochemical Evaluation of Itraconazole solution								
Tests	K_1	K_2	K_3	K_4	K_5	K_6	K_7	K_8
Visual appearance	+	+	+	+	+	+	+	+
Clarity	+	+	+	+	+	+	+	+
PH	4.9	4.6	5.0	5.0	4.9	4.7	4.9	5.0

Two minutes time of contact was given. Then weight was kept rising in the pan until vials get detached. Mucoadhesive force was the minimum weight required to detach two vials. The cheek mucosa was changed for each measurement.

Content Uniformity: Buccal cavity of Isolation of chicken cheek mucosa from the anterior healthy chicken was obtained from the local slaughter house. It was cleaned and the mucosa was re-moved from the anterior buccal cavity. The mucosa was stored in normal saline with few drops of gentamycin sulphate injection, to avoid bacterial growth. After the removals of blood from the mucosal surface it become ready for use

Diffusion Medium: The diffusion medium used was phosphate buffer PH 6.6 Assembly of diffusion cell-For in – vitro diffusion studies the oral diffusion cell was designed as per the dimension given. The diffusion cells were placed on the magnetic stirrers. The outlet of the reservoir maintained at 37±0.5· C and was connected to water jacket of diffusion cell using rubber latex tubes. The receptor co presentment was filled with fluid. Then the prepared chicken cheek mucosa was mounted on the cell carefully so as to avoid the entrapment of air bubble under the mucosa. Intimate contact of mucosa was ensured with receptor fluid by placing it tightly with clamp. The speed of the sitting was kept content throughout the experiment .With the help of micropipette 1ml of sample was withdrawn at a time intervals of one hour from sampling port of receptor compartment and same volume was the replaced with receptor fluid solution in order to maintain sink condition. The samples were appropropriately diluted and the absorbance was measured at 254 nm using Shimadzu 1700UV-VIC spectrophotometer.

Results and discussion:

Gelation temperature: Viscosity of temperature induced gel formulation were increases with increase in concentration of polaxamer 188 (K1, K2, K3, K4) at35· C, 34· C, 25· C, 23· C. In addition of carbopol 934 with polaxamer188 ratio (15:.01), (15:.02), (15:.03).it was found that the gradual increase in the gelation temperature of formulation (K5,K6,K7).the buccal gel prepared with high concentration of carbopol 934 (0.1 to 0.4) with polaxamer 188 (15%) exhibited gelation above body

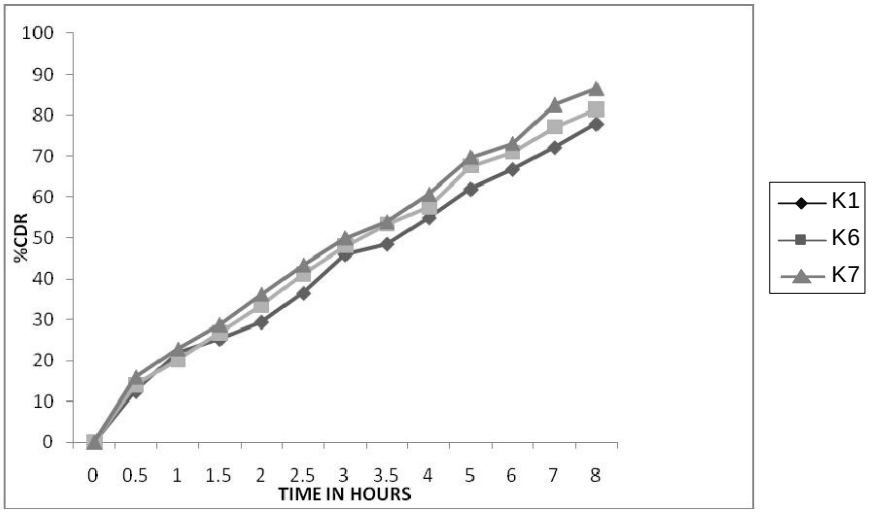
Viscosity study: From viscosity study on formulation K1, K6 and K7 optimized formulation showed increase in viscosity at 37· C whereas viscosity was decreased in

(K8, K9, K10, K11, and K12). When temperature of formulation increased (no sol-gel) this finding may be increased concentration of carbopol 934 in the formulation.

Content Uniformity: All the optimized formulation (K1, K6, K7) were checked for their content uniformity. The content was determined at 254nm, which is of drug using Shimadzu 1700UV-VIS spectrophotometer.

Table 3: Composition of Formulation

formulation	Composition of polymer	Gelation temperature sol-gel.(C)
K1	Polaxamer 188 (10%)	35 ^{o} C
K2	Polaxamer 188 (11%)	34 ^{o} C
K3	Polaxamer 188 (13%)	25 ^{o} C
K4	Polaxamer 188 (15%)	23 ^{o} C
K5	Polaxamer 188: carbopol 934 (15%:0.01%)	28 ^{o} C
K6	Polaxamer 188: carbopol 934 (15%:0.02%)	34 ^{o} C
K7	Polaxamer 188: carbopol 934 (15%:0.03%)	36 ^{o} C
K8	Polaxamer 188: carbopol 934 (15%:0.1%)	42 ^{o} C
K9	Polaxamer 188: carbopol 934 (15%:0.2%)	50 ^{o} C No reverse
K10	Polaxamer 188: carbopol 934 (15%:0.3%)	>60 ^{o} C No reverse
K11	Polaxamer 188: carbopol 934 (15%:0.4%)	>60 ^{o} C No reverse



Change in drug release:K1-78.4%,K6-81.4%,K7-86.6%

Fig 1: Percentage of drug release from the formulation

Diffusion across chicken cheek mucous membrane: The percentage of drug diffused through chicken cheek mucous membrane over the period of 6 hrs for formulation K1, K6, K7 was found to be 75.87%, 78.92% and 85.31% respectely.The diffusion of drug from formulation K1 was less may be due to presence of polaxamer 188 in the gel which retards the drug release rate owing to reduction in dimensition of water channel.

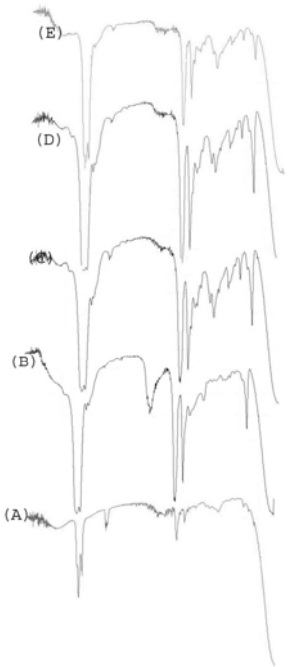
While diffusion of drug through formulation K6, K7 was found to be more may be due to presence of carbopol 934, which undergoes rapid swelling and helps in faster diffusion. (Ref .Grap- 1) Gel Strengh: At 37^{o} C, the gel strength of formulation K6 was found to be more as compared to formulation K1 and gel strength of formulation K7 was found to be more than as compared to formulation K_6. This may be due to reversible gelation of poloxamer at 37 ^{o} C.

Table 4: Characteristics of optimized itraconazole gel

formulation	PH	Viscosity at 37· C	content uniformity (% w/w)	mucoadhesive force (dyne/cm2)	gel strength in (sec)
K1	7.0	30943	98.4	3573.12	102
K6	7.1	46860	98.6	4401.14	110
K7	7.0	48740	98.6	4992.06	115

Table 5: FTIR –Spectra

Compound	CH Streching NH Streching	Unsaturated Nitrogen compound	NH Bending	Ar. Tretiary	C-cl	Polaxmer - 1120		Carbopal	
						C-O Polymeric associated	CH_2 CH_3 C O	OH	COOH
Intraconazole	2960	2360	1560	1330	600-800	3400-3200	1260-1350	3650	1650 – 1 1725 – 1700
Polaxmer-188						1380	1320		
Carbopol – 934									
Polaxmer – Carbopol	2960					1380	1320		1440 – 1720 Cluster
Polaxmer – Carbopol Intraconazole	2960	2360	1560	1320		1380	1320	-	1600 - 1720



FTIR SPECTRA :(A) ITRACONAZOLE, (B) CARBOPOL 934, (C) POLAXAMER 188, (D) K-POLYMER COMPINATION, (E) K-POLYMER COMBINATION+ITRCONAZOLE.

Itraconazole compound formed the polymer active with no disturbance the functional group. Itraconazole so polymerized active constituent has no change of effect after polymerizations.

DSC spectra: Itraconazole endothermic peak at 112.61 ^{o}C and 198.87· C, Polaxamer 188 endothermic peak at 62.96 ^{o}C, Combination of polymer endothermic peak at 57.63^{o}C, Combination of polymer and itraconazole peak at 57.52^{o}C, 193.46^{o}C and 242.05^{o} C

DSC- SPECTRA :(A) ITRACONAZOLE,(B) CARBOPOL 934, (C) POLAXAMER 188,(D) K-POLYMER COMPINATION,(E) K-POLYMER COMBINATION+ITRCONAZOLE.

Conclusion: It was seen that as the concentration of poloxamer 188 was increased the gelation temperature decreased. Carbopol 934 tends to increase the gelation Temperature of poloxamer 188. A modulation of the gelation temperature to reach the desired range (35· -C.37· C) Could be achieved through the use of a combination of the polaxmer 188 /carbopol 934. Carbopol 934 has shown a permeation enhancing effect. Thus from the above results it can be concluded that a temperature mediated in situ gelling mucoadhesive buccal gel of itraconazole can be formulated using optimum quantity of polaxmer 188 and carbopol 934 combination of both to have a increase in Buccal residence time and patient comfort.

Reference:

[1] T.W Max Farlane,,in L.P.Samarayake and T.W. Maxfarlane,(Eds.), Buttertworth and Co.,London,1st Edn.,Britan,(1990),pp.20-21

[2] P.G.Fotos,S.D.Vincent and J.W Hallstein,Oral surg.med.oral patho.,74,71

[3] Singhare DS. Khan S. Yeole PG. Poloxamers: Promosing bock co-polymers in drug delivery. Ind.j.pharm. Sci. 2005, sept – oct.523-531.

[4] BASF Wyandotte. Pluronic surfactants, product Brochure BASF Corporation, Mount olive New jersey, 1997.

[5] Collectt JH,Davies MC.Effect of batch variability on molecular properties and in-vivo release characteristics of pluronic PF-127 gels, J. pharm. Pharmacology,1984,36,52-57.

[6] Tomida H. Shinohara M, kuwada n, kiryn s. Invitro release characteristics of diclofenac and hydrocortisone from pluronic F-127 gels, Acta pharm suec., 1987,24,263-272.

[7] Ryu j, chung s, lee M, kim C, Increased bioavailability of propranolol in rats by retaining thermally gelling liquid suppositories in the rectum. Int. J. pharm. 1999, 59,163-172

[8] Sesai SD, Blanchard J. In-vitro evaluation of pluronic F-127 based controlled ocular delivery systems for pilocarpine. J. pharm. Sci., 1998, 87,226-230

[9] pec EA, Wout ZG, Johnston TP. Biological activity of urease formulated in poloxamer 407 after intraperitoneal injection in the rat. J.Pharm. sci. 1992, 81,626-630.

[10] Morikkawa k, Okada F, Hosokawa M,Kobayashi H. Enhancement of the of therapeutic effects of recombinant interleukin-2 on a transplantable rat fibrosarcoma by the use of a sustained release vehical pluronic gel. Cancer Res., 1987, 47, 37-41.

[11] Suh H, jun HW,Dzimianski MT. Pharmacokinetic and local tissue deposition studies of naproxen.

[12] Barry BW. In ry delivery systems;fundamentals and techniques.Johnson P, sloyd- jones for,eds. C hicnester: Ellis Hor wood 1992, P 200-223.