Investigation the effect of different concentrations of carbomer & Co solvent propylene glycol on the releasing process of tinidazole from vaginal aqueous gel

Salmo M. Hiba*

Received 26/10/2005 ; accepted 12/3/2006

الخلاصة

إن قابلية الالتصاق المخاطي الموضعي للهلام المائي المهبلي لإخراج المضاد البكتيري والمضاد الطفيلي لمادة التينيدازولمن الوسط الناقل له يعتمد على نفاذه من مذيبة إلى الطبقة المخاطية المهبلية لإعطاء تأثيره الموضعي وهذا يعتمد على الصفات الفيزياوية لكل من الدواء والمذيب ومكونات الوسط المحرر المستعمل.

لقد تمت دراسة تأثير التراكيز المختلفة للكاربوميز (٠,٥، ١، ٢٪) على تحرير التينيدازول (٢٪) إلى الوسط المشابه للسائل المهبلي من قاعدة الهلام المائي بوجود وعدم وجود تراكيز مختلفة من البروبلين كلايكول وقد تبين إن أعلى نفاذ تم الحصول عليه من التركيز القليل للكاربوميز (٠,٥) وكذلك في التركيز القليل للبروبلين كلايكول في قاعدة الهلام المائي. وأيضا قد تمت دراسة ذوبان التينيدازول في تراكيز مختلفة من خليط محلول الماء والبروبلين كلايكول. إن الدراسة تمت باستخدام نوعين من الوسط المحرر وهو الوسط المشابه للسائل المهبلي بالآس الهيدروجيني ٤,٢ ومحلول الستريت الدارئ و بالآس الهيدروجيني٤,٢. ولقد تبين إن استعمال الوسط المشابه للسائل المهبلي يعتبر مفيد في تقييم المستحضرات المهبلية الموضعية.

إضافة إلى ذلك قد تم حساب نسبة الانتفاخ في ١غم من الكاربوميز الجاف بعد أربعة ساعات حضانة في الوسط المشابه للسائل المهبلي, وكذلك دراسة سرعة الانتفاخ الأولية والانتفاخ المساوي للكاربوميز الجاف (٠,٥، ١، ٢٪).

في نفس الوسط, ولقد تبين من هذه الدراسة إن تركيز (٠,٥٪) للكاربوميز في قاعدة الهلام أعطى أعلى سرعة للانتفاخ الأولي والانتفاخ إلى الحجم المساوي, إما تركيز (١، ٢٪) فقد أعطى اقل سرع للانتفاخ الأولي واقل حجم مساوي من الهلام المنتفخ.

ABSTRACT

The ability of local mucoadhesive vaginal aqueous gel to deliver the antibacterial, antiprotozoal tinidazole (TND) to vaginal endolayers is dependent on its releasing out of the drug from vehicle to the mucosal layer of the vagina and then exert its local effect. This depends upon the physicochemical properties of the drug, vehicle & the composition of the releasing medium used.

The effect of different concentration of carbomer 941(0.5, 1&2% w/w) on the release of TND (2%w/w) from the aqueous gel base in absence & presence of various concentration of propylene glycol (PG) to the stimulant vaginal fluid (S.V.F) medium was investigated. The release of TND from low concentration of carbomer (0.5% w/w) aqueous gel base was higher & its followed fickian mechanism of the release while an anomalous mechanism holds the releasing of drug from a high concentration of carbomer (1&2% w/w) aqueous gel base, the release of TND from aqueous gel containing low concentration of PG was markedly higher than that of aqueous gel having a higher concentration of PG. The solubility of TND in different concentration of PG : water solution were also investigated. The releasing method were carried out using both S.V.F (pH4.2) & citrate buffer (pH4.2). It was found that the using of S.V.F considered to be useful for the evaluation of topically prepared vaginal dosage form as a releasing medium.

Furthermore the swelling index or (swelling percent) of 1gm carbomer polymer in S.V.F was measured after 4 hours incubation of the dry polymer in the fluid. The initial rate of swelling & the swelling equilibrium of (0.5, 1&2% w/w) dry carbomer in the S.V.F were also investigated, 0.5 % w/w cabomer aqueous gel has a high rate of swelling & swelling to equilibrium size rapidly, while 1&2% w/w carbomer aqueous gel having a low rate of swelling & swelling to equilibrium size slowly.

* Pharmaceutics Department, College of Pharmacy, Baghdad University, Baghdad–Iraq.

INTRODUCTION :

The presence of dense network of blood vessels has made the vagina an excellent rout of drug delivery for both systemic & local effect. The main advantages of vagina drug delivery over conventional drug delivery are the ability to by–pass first pass metabolism, ease of administration & high permeability for low molecular weight drugs[1].

TND is an antimicrobial drug with high activity against anaerobic bacteria & protozoa; ncluding trichomonal vaginitis, bacterial vaginosis also it is used for surgical & gynecological sepsis[2,3]. TND is usually administered as a single daily dose by mouth, I.V infusion & as vaginal pessaries[4], TND is similar to metronidazole but has a longer duration of action & has many side effect such as nausea, vomiting, unpleasant taste[5].

In treatments of bacterial vaginosis metronidazole vaginal gel or cream are found to be nearly as effective as orally administered drug[6].

It has been recognized that the release of drug from topically or locally dosage form is affected by the composition of the vehicle & the thermodynamic activity of the drug in the vehicle[7].

Therefore, in this study in order to examine the influence of the vehicle containing different concentrations of mucoadhesive aqueous carbomer polymer & co solvent PG, the release of the TND was determined, S.V.F (pH4.2) was employed as the releasing medium & its effect on the in vitro release was compared with the result obtained from citrate buffer (pH4.2).

MATERIALS AND METHODS:

Materials:

TND was obtained from Sigma chemical co.Carbomer 941 from Goodrich U.S.A, sodium chloride, acetic acid, glycerol & glucose from Riedel-dehaen, Germany, potassium hydroxide from Fisons scientific LTD, England, calcium hydroxide, sodium hydroxide & propylene glycole from BDH chemicals LTD, England, lactic acid from Fluka AG, Switzland, urea from Merck, citric acid from Seelze, Germany, hydrochloric acid from AJAX chemical, Australia, bovine serum albumin from CSL limited biopharmaceutical for life®, Australia .

Methods:

Preparation of simulant vaginal fluid (S.V.F) (pH 4.2)

S.V.F was prepared by dissolving 3.51gm sodium chloride, 1.4gm potassium hydroxide, 0.22gm calcium hydroxide, 0.018gm bovine serum albumin, 2gm lactic acid, 1gm acetic acid, 0.16gm glycerol, 0.4gm urea & 5gm glucose in 1 liter distilled water[8].

Preparation of citrate buffer (pH4.2)

Citrate buffer was prepared by mixing a 600ml of solution A (0.1M disodium citrate) & 400ml of solution B (0.1 N HCL).

Preparation of carbomer aqueous gel base

The formulas of aqueous gel tested are shown in (table -1-) carbomer powder was first dispersed in cold freshly distilled water with the aid of a high speed stirrer, until a solution was complete : the stabilizer (0.1% disodium EDTA), preservatives 0.15% methylparaben (m.p) & 0.05% propylparaben (p.p) were then dissolved. The solution was then neutralized with NaOH (400mg/1gm carbomer)[9,10], the mixture of the 2% drug & different concentrations of PG were then added to the gel base with gentle mixing. The final pH value for the prepared aqueous gel was about 5-5.32.

Table-1- the composition of model aqueous gel formulas

No. of formulas	TND gm	Carbomer a(0.5%w/w), b(1%w/w), c(2%w/w)	NaOH gm	PG gm	m.p gm	p.p gm	Di Na EDTA gm	D.W up to 100ml	Solubility of TND mg/ml
1	2	a,b,c	q.s	10	0.15	0.05	0.1		50
2	2	a,b,c	q.s	20	0.15	0.05	0.1		52
3	2	a,b,c	q.s	30	0.15	0.05	0.1		54
4	2	a,b,c	q.s	40	0.15	0.05	0.1		63.3
5	2	a,b,c	q.s	50	0.15	0.05	0.1		72
6	2	a,b,c	q.s	60	0.15	0.05	0.1		75
7	2	a,b,c	q.s	0	0.15	0.05	0.1		6.1

In vitro TND release test from aqueous gel

A basket with 2.5cm in a diameter (dissolution apparatus I) was enclosed with a multifold filter paper in order to be filled with 1gm of each base containing 2% w/w of TND. After connecting to a stirrer motor, the dialysis cell was immersed to about 1cm of its surface in 500ml of S.V.F pH4.2 (collecting medium) containing in a flask of the dissolution apparatus. The system maintained at 37c◦, the collecting medium was stirred at 100r.p.m for five hours of incubation. Samples were withdrawn from the collecting medium after 0.5,1,2,3,4&5 hours & replaced with an equal volume of the fluid solution at 37c◦. The samples were then analyzed for their drug content at its λmax. 320nm[11,12].

Evaluation the effect of carbomer concentration on the mechanism of TND release from the aqueous gel base

The effect of different concentration of carbomer polymer (0.5, 1&2% w/w) on the mechanism of the drug release from aqueous gel were studied.

Measure the swelling index or (swelling percent) of dry carbomer polymer in S.V.F

According to B.P (2001) the swelling index is the volume in ml occupied by 1gm of polymer after it has swollen in aqueous liquid (S.V.F used in the study) for four hours. Place 1gm of dry carbomer polymer in 25ml graduated cylinder, moisten the polymer with 1ml (ethanol 96%), add 25ml of S.V.F & shake vigorously every 10 minute for one hour, then allow to stand for three hours. At one & half an hour after beginning the test release any large volume of liquid retained in the layer of the polymer & any particles of polymer floated at the surface of the liquid, then measure the volume of polymer according to this equation:

$$\% \text{ swelling} = (\text{final volume} - \text{initial volume})/\text{initial volume}$$

Evaluation the effect of PG concentration on the mechanism of TND release from the aqueous gel base

The effect of different concentration of PG (0,10,20,30,40,50&60% w/w) on the mechanism of the drug release from aqueous gel were studied.

Evaluation the thermodynamic activity of TND in the vehicle

The solubility of TND in the vehicle of different PG concentration was measured as follows. After an measured amount of TND powder was added to the mixture of PG–water, the mixture was shaken in an incubator at 35c◦ for 48 hour.

It was then passed through a millipore filter (0.45µm). The solubility of TND in the mixed solvent measured by U.V spectrophotometer after an appropriate dilution with water.
The thermodynamic activity (a), of TND in the gel was defined as : a=Cv/Cs where Cv is the concentration of TND in the gel & Cs is the solubility of TND in gel, Cs was estimated from solubility data of solvent with the same composition[13].

RESULTS & DISCUSSION

In this study, S.V.F (pH4.2) was chosen as a collecting medium for in vitro release of TND from the aqueous vaginal gel[14]. The fraction of TND released from the aqueous gel to the S.V.F (pH4.2) medium compared with the fraction released to citrate buffer (pH4.2) are shown in (figure 1 a,b).

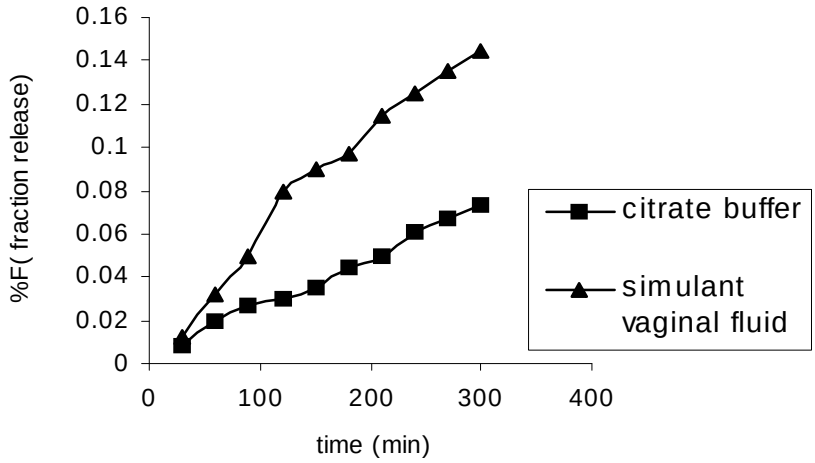

figure 1a- The release of TND from aqueoue gel model 2a to citrate buffer & to simulant vaginal fluid

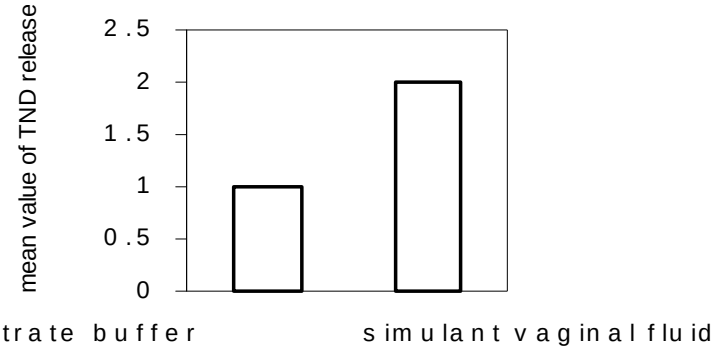

figure 1 b-Mean release of TND from aqueous gel model 2a to citrate buffer & to simulant vaginal fluid

The release of TND from the aqueous gel to the S.V.F medium was higher than that occurring to citrate buffer due to the composition of the fluid medium that promote the increasing in the solubility of TND to be 37mg/ml & 28.3mg/ml in citrate buffer, S.V.F gives a resembling effect & idea to what happened in vivo.

The effect of carbomer concentration (0.5,1&2% w/w) on the mechanism of TND release from the aqueous gel base are shown in (figure 2 a,b).

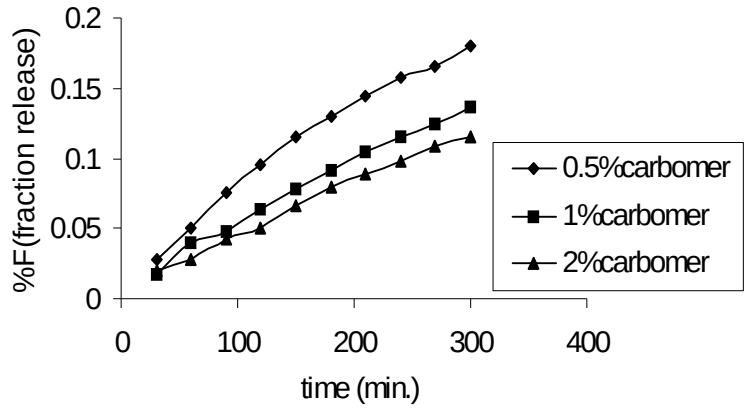

figure 2a- Effect of carbomer concentration on the release of TND from aqueous gel

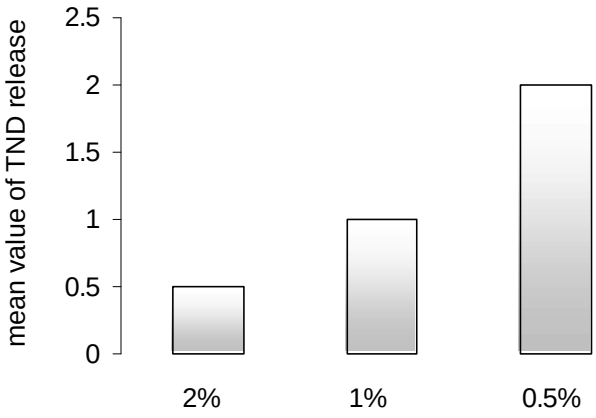

figure 2 b-Mean effect of carbomer concentration on the release of TND from aqueous gel

The releasing mechanism from carbomer cross-link matrices were influenced by carbomer concentration. In general the release mechanism was anomalous (non-Fickian) for 1&2% w/w carbomer concentration, where the release mechanism appears to be followed (Fickian) for 0.5% w/w carbomer concentration[15].

Across-link carbomer with ionizable side chain (high number of carboxylic acid group)[16] swell extensively in aqueous medium, this swelling (taking up of liquid by a gel with an increasing in the volume) depends on the pH of the medium & the presence of electrolytes, the S.V.F pH4.2 has a lot of electrolytes content : sodium chloride, potassium hydroxide, calcium hydroxide. This property (swelling) is important since the releasing of drug from hydrogel depend on the water content of the gel base which is in turn depends on the concentration of the cross-linked polymer, as well as, increasing the concentration of the cross–linking polymer increases the hydrophobicity of a gel & decrease the releasing of drug due to swelling phenomena[17].

The swelling index or (swelling percent) for 1gm dry carbomer in S.V.F is 209.09%.
The kinetics of swelling of carbomer was studied by measuring the increase in the volume of dry polymer in 25ml S.V.F solution placed in 25ml graduated cylinder as a function of time t.A plot of (t/v) t : time in hour, v : volume in ml of aqueous S.V.F absorbed per volume of dry polymer draw against time t in hour gives an idea about the initial rate of swelling (reciprocal of intercept) & swelling equilibrium (reciprocal of slope)[15] as shown in table-2- & figure 3 a,b

Table 2 . initial swelling rate & swelling equilibrium of carbomer polymer in simulant vaginal fluid after fours hour.

Carbomer %w/w	Initial rate of swelling (ml of fluid solution/hr.ml of dry carbomer)	Swelling to equilibrium size (ml of fluid/ml of dry carbomer)	Correlation coefficient (r)
0.5	9.354	2.42	0.9925
1	7.315	0.973	0.9979
2	1.1669	0.34	0.9955

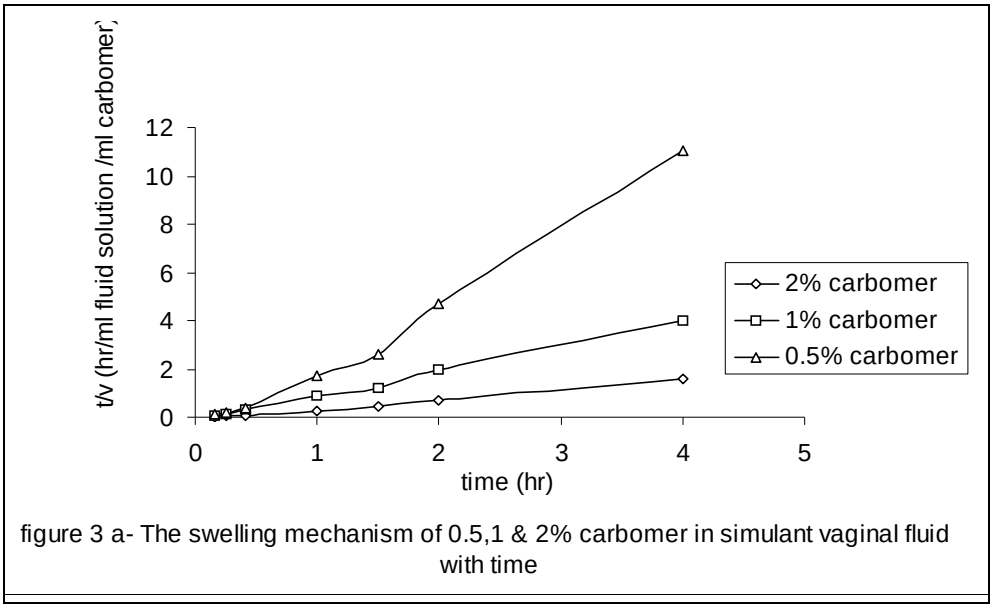

figure 3 a- The swelling mechanism of 0.5,1 & 2% carbomer in simulant vaginal fluid with time

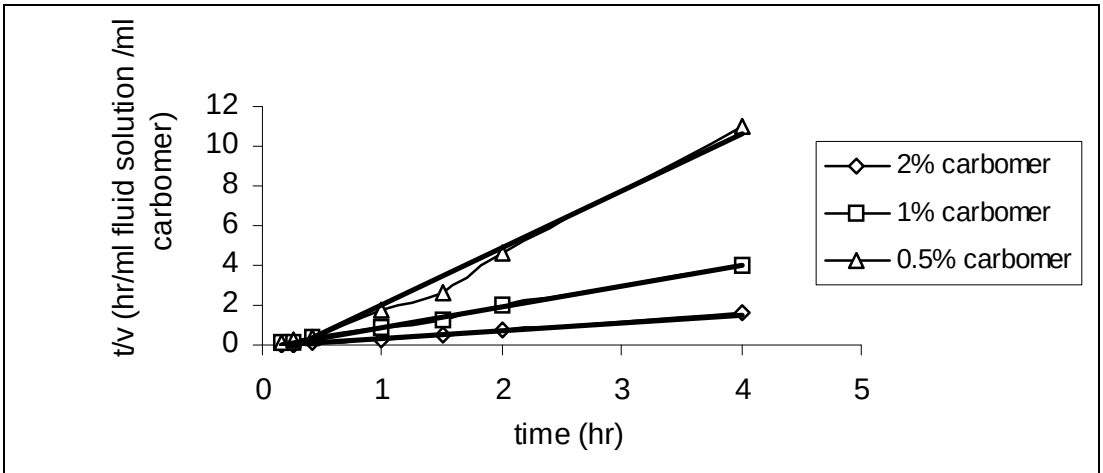

figure 3b- The swelling mechanism of 0.5,1 & 2% carbomer in simulant vaginal fluid with time

This result express the reason behind a high initial rate of swelling for 0.5gm dry polymer to their loosely compacting ordered to be easily penetrated by S.V.F.
Therefore 0.5% w/w aqueous gel base dispersed in the medium, disappeared rapidly & leaves undetectable swollen volume on the surface of multifold filter paper after five hours releasing time, so it holds a Fickian releasing process with an exponent value of 0.5, otherwise the penetration of the fluid into 1 & 2gm dry polymer is slower & less extensive because the powder at these weight is more tightly compact & has a higher density than 0.5gm.
As well as, in 1 & 2% aqueous gel base the swelling takes place at a slower rate to be leaves a significant volume of gel on the surface of multifold filter paper after five hours of the releasing process & to be holds an anomalous releasing mechanism with an exponent value greater than 0.7.

The equation express the fraction (*F*) release of drug from gel

$$F = Mt/Mo = kt^n \quad \text{at time } t$$

Where *Mt* is the amount released at time *t* , *Mo* is the initial amount of drug, *K* is the rate constant & ^{n} is the releasing exponent value indicative the mechanism of drug release, if ^{n} is 0.5 (Fickian) holds the release, if ^{n} is greater than 0.5 this indicate (anomalous) releasing mechanism, ^{n} value could be result from the slope obtained by the drawing of ln. fraction released *(F)* verse ln. time *t* .
In this study, PG was chosen as a main vehicle to solubilize the TND in the aqueous gel since its use has been well defined in the laboratory[18].
Because PG is a good solvent for TND, the solubility of TND in PG increase with PG concentration & a significant effect ($p<0.05$) of PG as a co solvent from 10,20 & 30% w/w to 40,50 &60%w/w concentration was obtained as shown in figure 4 a.
Figures 5 a,b, 6 a,b & 7 a,b showed that the %F fraction release of TND increased with the addition of (10,20 & 30%) PG & decreased as the concentration of PG increased.
The decrease in the release of TND with 40,50 & 60%w/w PG aqueous gel may be due to decrease in the number of free molecules of the drug to be release.
The solubility data were used to estimate the thermodynamic activity of these PG aqueous gels. The thermodynamic activity changes of TND in these aqueous gels are shown in figure 4b.

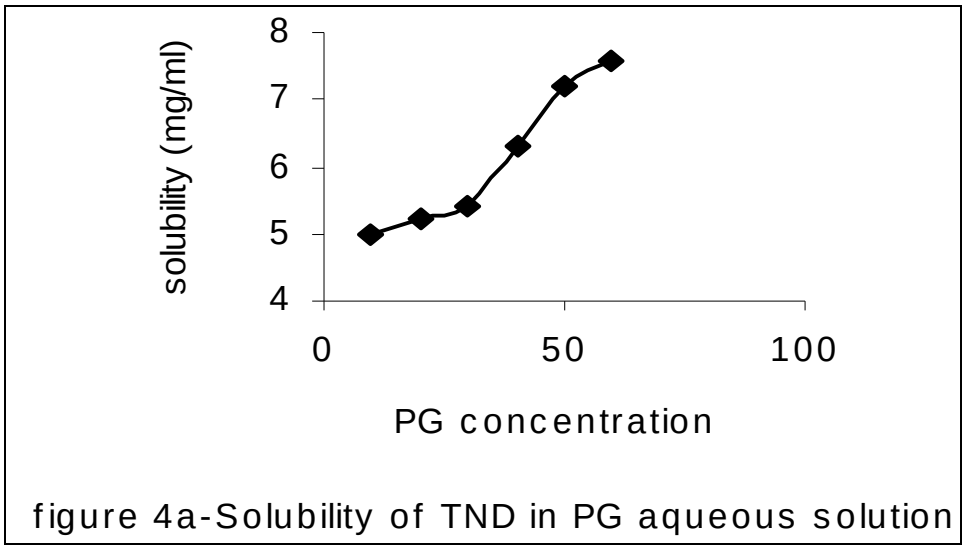

figure 4a-Solubility of TND in PG aqueous solution

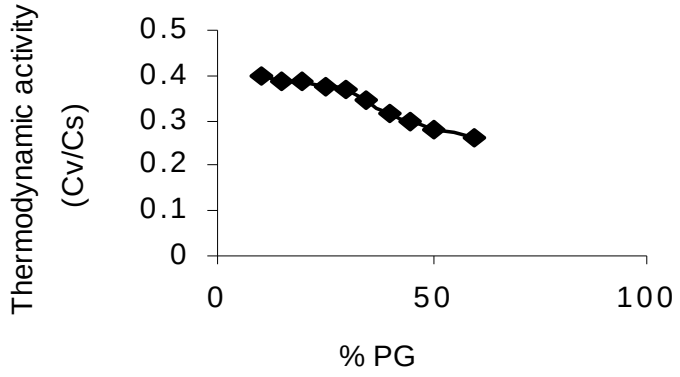

figure 4b-The thermodynamic activity of TND in PG aqueous solution

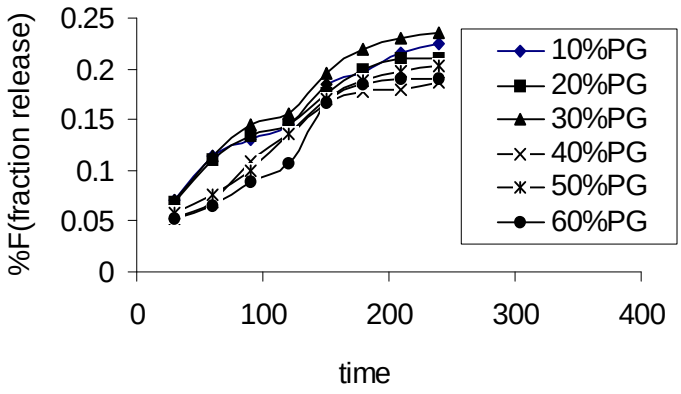

figure 5 a-The effect of PG conc. on the release of TND from 0.5%w/w carbomer aqueous gel

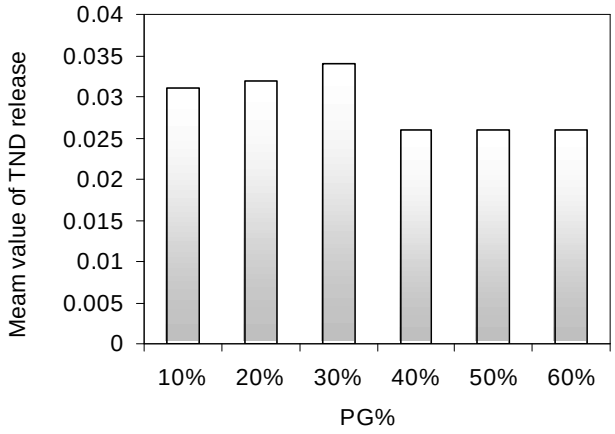

figure 5 b-The mean effect of PG conc. on the release of TND from 0.5%w/w carbomer aqueous gel

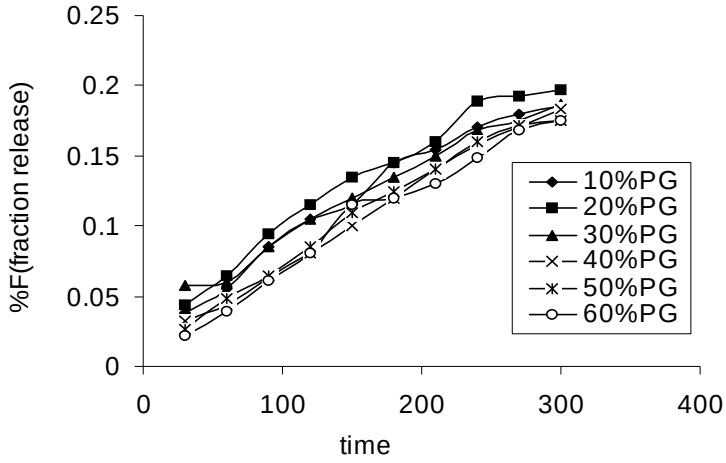

figure 6 a -The release of TND from 1% carbomer aqueous gel base

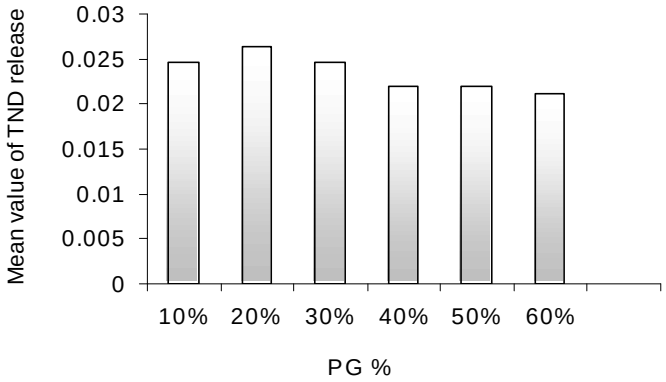

figure 6 b-The mean effect of PG conc. on the release of TND from 1% w/w carbomer aqueous base

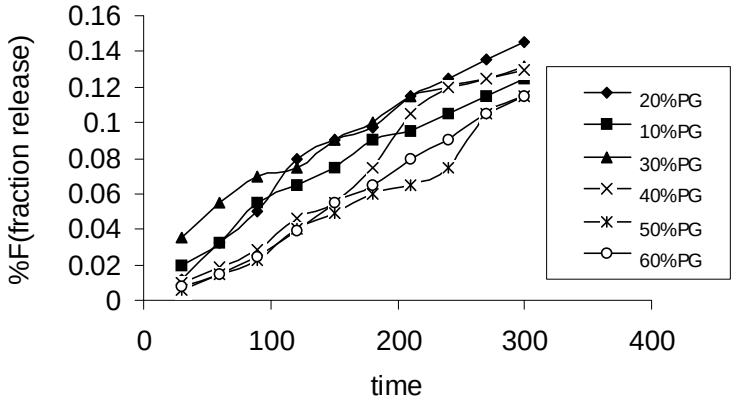

figure 7 a-The effect of PG conc. on the release of TND from 2% carbomer aqueous gel

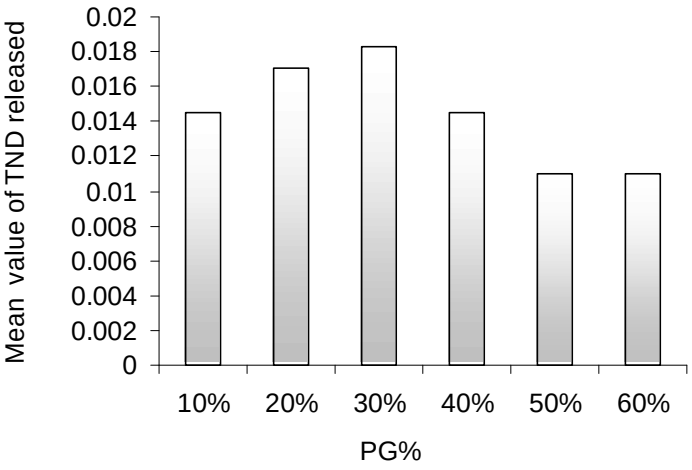

figure 7 b-Mean effect of PG conc. on the release of TND from 2% carbomer aqueous gel

Further more the higher solubility of the drug produces a decreases in the thermodynamic activity of TND in the vehicle[19]. The concept of thermodynamic activity, described by Higuchi[20], represents the escaping tendency of drugs & it was supposed that the increase in this activity leads to an enhanced the releasing of drugs from the vehicle[12]. In addition, the thermodynamic activity is proportional to the solubility of the drug in the vehicle & becomes maximal with minimal amount of co solvent[21].

CONCLUTION

In this study, the release of a drug from the aqueous gel base was examined using a stimulant vaginal fluid S.V.F as a releasing medium, demonstrating that it is possible to modifying the drug releasing mechanism, by changing the polymer concentration in the aqueous gel base. The drug release was found to be affected by the concentration of co solvent PG in aqueous gel base & to be improved by choosing the appropriate concentration for increasing the thermodynamic activity of the drug in the veh

Acknowledgement

The author is very grateful to Miss. Masar Basim (M.Sc pharmaceutics) for her encouragement, useful suggestions & discussions & to Mr. Haider Kadim (M.Sc pharmaceutics) for his great help in supplying tinidazole.

REFERENCES:

1. Hussain, A., Ahsan, F., The vagina as a route for systemic drug delivery. J. of controlled release. (2005)103, p.301-313.
2. BNF, British national formulary, British medical association and Royal Pharmaceutical society of Great Britain, 47 ed, March 2004, p.287, U.K.
3. Guo, Q.L, Ni, M.X, Wangl, L, Inhibition of tinidazole gel on trichomonas vaginals of human origin in vivo & in vitro. J. china. pharm. Univ., (1996), 27(7), p.432-434.
4. Martindale, The complete drug reference, 32 ed. 3, pharmaceutical press, 1999, p.594.
5. Milani, M., Barcellona, E., Agnello, A., Efficacy of the combination of 2g oral tinidazole and acidic buffering vaginal gel in comparison with vaginal clindamycin alone in bacterial vaginosis : a randomized, investigator-blinded, controlled trial. Euro. J. of Obst. & Gyne., (2003), 109, p.67-71.

6. Dubonchet, L., McGregor, J.A., Ismail, M., McCormack, W.M., A pilot study of metronidazole vaginal versus oral metronidazole for tretmeant of trichomonas vaginalis vaginitis, Sex. Transm. dis., (1998), 25, p.176-179.
7. Poulsen, B.J., Young, E., Coquilla, V., Katz, M., Effect of topical vehicles composition on the in vitro release of fluocinolone acetonide & its acetate ester. J. pharma. sci, (1968), 57, p. 928-933.
8. Owen, D.H., Peters, J.J., Katz, D.F., Rheological properties of contraceptives gels. Contraception, (2000), 62, p. 321-326.
9. Carter, S.J., " Dispensing for Pharmaceutical Students", 12th ed. p.109, Churchill Livingstone, 1987.
10. Kassem, K.A., Attia, M.A., Habib, F.S., Mohamed, A.A., Activity of ophthalmic gels of betamethasone & phenylephrine hydrochloride in the rabbit's eye. Int. J. pharmaceutics, (1986), 32, p. 47-54.
11. Mariee, N.K., Factors affecting the in vitro diffusion of diclofenac sodium from ointment. Iraqi. J. of pharmacy, (2001), 1, p. 62-71.
12. Hui, H.W., Robinson, J.R., Effect of particle dissolution rate on ocular drug bioavailability. J. pharma. sci. (1986), 75, p.280-287.
13. Tanaka, S., Takastima, Y., Murayama, H., Tsuchiya, S., Studies on drug release from ointments. vaginal release of hydrocortisone butyrate propionate from topical dosage forms to silicone rubber. Int. J. pharma.(1985), 27, p.29-38.
14. Chang, J.Y., Kyoung, Y., Choi, H., Kim, Y.B., Kim. C.K., Reological evaluation of thermosensetive & mucoadhesive vaginal gels in physiological conditions Int. J. pharma. (2002), 241, p.155-163.
15. Russo, L.I., Ghaly, E.S., Drug release from carbomer 934 matrices. Health. Sci. J. (2000), 19, p.131-137.
16. Hand Book of Pharmaceutical excipient, 1986, p.41, American pharmaceutical association production, pharmaceutical society of Great Britain production.
17. Martin, A.N., "Physical Pharmacy", 4th ed., 1993, p.497, Waverly, New Delhi.
18. Paul, K.W., Birgitts, M., Jonathan, H., Annie, H., Vehicle effect topical drug delivery III. Effect of Azone on the cutaneous permeation of metronidazole & propylene glycol. Int. J. pharmac. (1985), 24, p.19-26.
19. Pramod, P.S., Joel, L.Z. Evaluation of penetration enhancement of lidocaine by nonionic surfactants through hairless mouse skin in vitro. J. pharma. sci., (1986), 75, p. 176-181.
20. Higuchi, T., physical chemical analysis of percutaneous absorption from cream & ointment. J. soc. cosmet. Chem., (1960). 11, p. 85-97.
21. Doliwa, A. Santoyo, S., Ygartua, P. Influence of piroxicam : Hydroxypropy l-Beta-Cyclodextrine complexation on the in vitro permeation & skin retention of piroxicam. J. pharmacological & biophysical research, (2001), 14, p.97-107.