Available online at www.sciencedirect.com
SCIENCE DIRECT
European Journal of Pharmaceutics and Biopharmaceutics 60 (2005) 67–71
European Journal of Pharmaceutics and Biopharmaceutics
www.elsevier.com/locate/ejpb

Research paper

Development of tretinoin gels for enhanced transdermal delivery

Sang-Chul Shin[a], Hee-Jung Kim[a], In-Joon Oh[a], Cheong-Weon Cho[b], Kyu-Ho Yang[c,*]

[a]College of Pharmacy, Chonnam National University, Gwangju, South Korea
[b]CJ Crop., R&D Center of Pharmaceuticals Ichonsi, Gyonggido, South Korea
[c]College of Dentistry, Chonnam National University, Gwangju, South Korea

Received 6 October 2004; accepted in revised form 31 January 2005
Available online 26 March 2005

Abstract

To develop the new gel formulations that show sustained release for a period of time, the bioadhesive carbopol gels containing tretinoin were prepared. The release characteristics of drug from the carbopol gel were studied according to temperature, receptor medium and drug concentration. For the enhancement of its percutaneous absorption, some kinds of penetration enhancer were used. As the concentration of drug increased, the release of drug from the gel increased, showing concentration dependency. The increase of temperature showed the increased drug release, depending on the activation energy of permeation. Among the enhancers used such as the glycols and the non-ionic surfactants, polyoxyethylene 2-oleyl ether showed the best enhancing effect. The carbopol gels of tretinoin containing an enhancer could be developed for the enhanced transdermal delivery of drug.

Keywords: Tretinoin; Penetration enhancer; Diffusion; Carbopol gels; Transdermal delivery

1. Introduction

Tretinoin has been used in the management of skin disorder to improve the appearance and texture of skin. Topically, tretinoin has been used for treatment of ichtyosis, psoriasis, acne vulgaris, neoplasia and other skin disease [1–7] at various dosage forms, such as solutions, lotions, ointments and creams.

Generally, in case of application of such formulations onto face tissue, it is difficult to expect their effects for a significant period of time, because they are easily removed by wetting, movement, and contacting. Therefore, the bioadhesive gels that have good accessibility could be developed for localized, percutaneous administration. Among the various hydrogel bases, carbopol is used because of its high stability, compatibility and low toxicity. Carbopol 934 is a hydrophilic polyacrylic acid polymer and its carboxyl groups become highly ionized after neutralization, forming a gel due to electrostatic repulsion among charged polymer chain [8–10].

To improve the permeability of drugs, the use of penetration enhancers is a logical approach to increase the drug flux across the epithelium. It has been shown that dermal penetration can be improved by using compounds, which have been proven to be effective enhancers on skin. The effect of various classes of transdermal penetration enhancers such as the bile salts, the surfactants, the glycols and derivatives, and chelators have been studied [11–15] to determine the diffusion properties of drugs in the semisolid vehicles especially when the release of drugs at the application site is likely to be rate-limited by the diffusion of drug. Although most topical formulations consist of rather simple components, the ability of a vehicle to release the drug at the local site is limited by numerous factors such as drug-vehicle, drug-skin and vehicle-skin interaction [16]. In this paper, the influence of receptor medium, temperature, pH, and drug concentration and the effects of penetration enhancer on drug diffusion from the carbopol gels were investigated in order to develop the effective semisolid topical formulation of tretinoin for treatment of skin disorder.

* Corresponding author. College of Dentistry, Chonnam National University, Gwangju 501-190, South Korea. Tel.: +82 62 220 5470; fax: +82 62 234 1271.
E-mail addresses: shinsc@chonnam.ac.kr (S.-C. Shin), ppappalee@hanmail.net (H.-J. Kim), ijoh@chonnam.ac.kr (I.-J. Oh), morning-garden@hanmail.net (C.-W. Cho), khyang@chonnam.ac.kr (K.-H. Yang).

0939-6411/$ - see front matter © 2005 Elsevier B.V. All rights reserved.
doi:10.1016/j.ejpb.2005.01.008

2. Materials and methods

2.1. Materials

Tretinoin was kindly supplied from Taegeug Pharm. Co., Ltd (Korea). Carbopol 934 was obtained from BF Goodrich (USA). Polyoxyethylene 2-stearyl ether, polyoxyethylene 2-oleyl ether, polyoxyethylene 23-lauryl ether, tetraethylene glycol (TEG), and diethylene glycol (DEG) were purchased from Sigma Chemical Co. (USA). All reagents of analytical grade were used without further purification. Acetonitrile and anhydrous ethyl alcohol were HPLC grade from J.T. Baker Inc. (USA).

2.2. Preparation of carbopol gels containing tretinoin

Carbopol gels were prepared by the previous methods [14]. Briefly, an appropriate amount (2 g) of carbopol powder was slowly added into water under constant stirring. A few drops of Tris were added to adjust the pH of carbopol gels. Tretinoin of 0.025 g that was dissolved in 30 ml of DGME solution was added to the above carbopol gels with stirring and water was added to make 100 ml. To minimize the degradation of tretinoin during handling, all procedures were performed in the dark condition.

2.3. HPLC determination of tretinoin

Tretinoin was assayed by modified HPLC methods [17,18]. The column was μ-Bondapak C_{18} (3.9× 300 mm), the mobile phases was a combination of acetonitrile:ethyl alcohol:acetic acid:water (85:85:1:15), and the UV detector was operated at the wavelength of 348 nm. The injection volume of 10 μl and a flow rate of 1.0 ml/min yielded an operation pressure of ≈1200 psi. Under these conditions, tretinoin peak appeared at the retention time of 6 min and DGME approximately at 2.7 min.

2.4. Diffusion study of tretinoin from the carbopol gels

The flux of tretinoin from the carbopol gels was determined using the various concentration of diethylene glycol methyl ether (DGME) as a receptor, which ranged from 0 to 50 (v/v)%. The synthetic cellulose membrane was mounted on the receptor compartment of the diffusion cell. Two grams of prepared carbopol gels containing tretinoin was placed in intimate contact with the cellulose membrane and the donor cap was covered with a parafilm and clamped. The sampling port was sealed with a parafilm to prevent the evaporation of the receptor medium. The receptor medium, 40% DGME solution, was maintained at 37 °C by a circulating water bath and stirred by a magnetic stirring bar. The donor compartment was maintained at ambient temperature of 25 ± 1 °C. The effect of drug concentration on its release from the gels was studied according to drug concentration of 0.005, 0.01, 0.025 and 0.05% (w/w), and the effects of temperature on drug release was performed at 28, 32, 37 and 42 °C by water bath. The total samples from the receptor compartment were withdrawn at predetermined intervals and immediately replaced by the same amount of fresh 40% DGME solution to maintain a sink condition. The sample withdrawn from the receptor compartment was then analyzed by HPLC method. Each data point represents the average of three examinations.

2.5. In vitro skin permeation study

2.5.1. Skin preparation

A male rat (Sprague Dawley) was sacrificed by snapping the spinal cord at the neck. The hair of abdominal area was carefully removed with an electric clipper and a square section of the abdominal skin was excised. After incision, the adhering fats and other visceral debris in the skin were carefully removed from the undersurface with tweezers [19]. The excised skin was used immediately.

2.5.2. Effect of an enhancer on the permeation of tretinoin from the 2% carbopol gels through rat skin

The excised abdominal skin was mounted in a diffusion cell. And other conditions were same as in Section 2.4. The 0.025% tretinoin gels containing 5% (w/v) enhancer were prepared by the previous methods [14]. The enhancers used were the glycols such as diethylene glycol, tetraethylene glycol, and the non-ionic surfactants such as polyoxyethylene-2-oleyl ether, polyoxyethylene-2-stearyl ether, and polyoxyethylene-23-lauryl ether. The amounts of drug permeated from the carbopol gels through rat skin were determined by HPLC. Each data point represents the average of three determinations.

2.5.3. Analysis of permeation data

The cumulative amount of the permeated drug was plotted versus time, and the flux was calculated from the steady-state part of the curve. The effectiveness of penetration enhancer was determined by comparing the flux of tretinoin in the presence and absence of enhancer. It was defined as the enhancement factor (EF) that was calculated using the following equation:

EF = (drug flux of samples containing an enhancer)/(drug flux of control sample without an enhancer)

3. Results and discussion

3.1. Influence of DGME in receptor medium on drug diffusion

In order to obtain sink conditions, it is desirable for the receptor medium to solubilize the drug. DGME is a hydroscopic liquid that is freely miscible with both polar

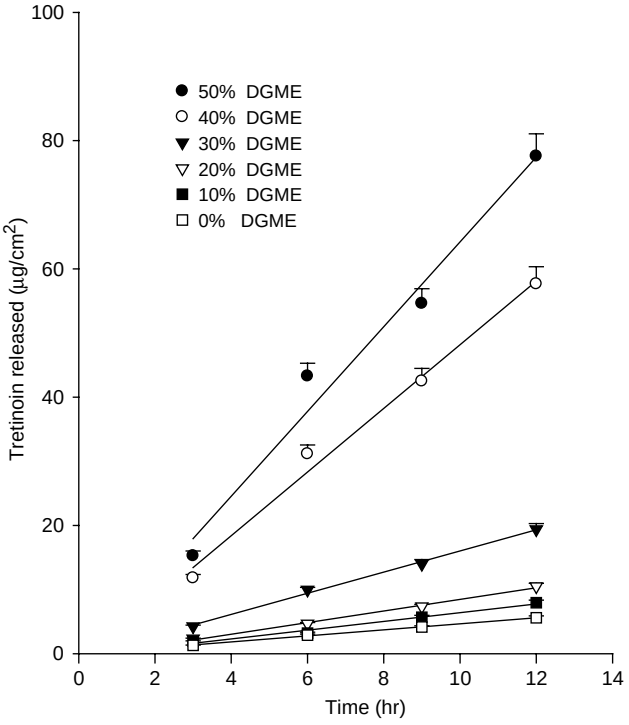

Fig. 1. Effect of DGME in receptor medium on tretinoin diffusion.

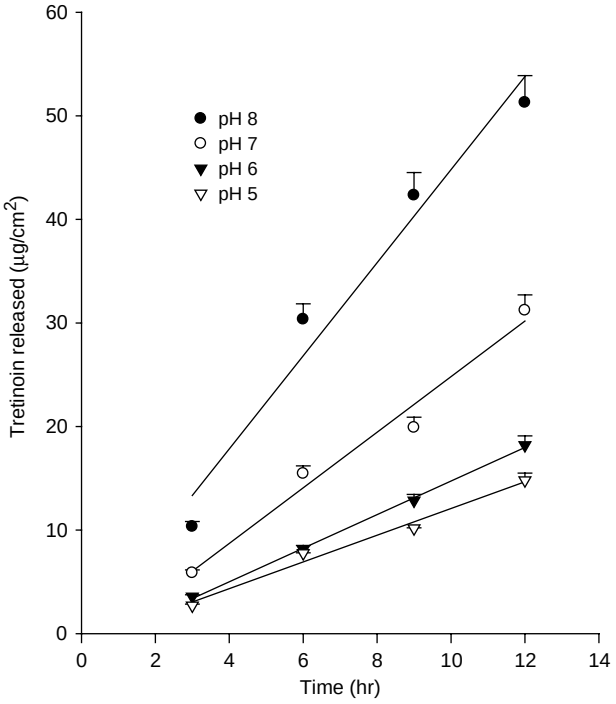

Fig. 2. Effect of pH of 2% carbopol gels on tretinoin diffusion.

and non-polar solvent. The aqueous solubility of tretinoin is extremely low and could be improved by addition of DGME into the aqueous solution as a solubilizer for tretinoin.

The influence of receptor medium on drug diffusion across synthetic cellulose membrane (Spectra/por MW 12-14,000) was studied from the prepared 2% carbopol gels at 37 ± 0.5 °C for 12 h (Fig. 1). The cumulative amount of tretinoin released (Q) versus the time (t) plot showed a good linearity. As the DGME solution in receptor medium increased to about 40%, the release rate of drug increased, thereafter slightly increased, but not significantly. For this, the release of tretinoin was studied using the 40% DGME solution.

3.2. Effect of pH of the carbopol gels on drug diffusion

The pH of vehicle has been shown to be one of the major variables that could influence diffusivity of drugs in semisolid vehicles [10]. In this work, the effect of pH of carbopol gels on diffusion of tretinoin was determined from the hydrogels of pH 5–8, respectively. As the carbopol gels were adjusted from pH 5 to 8, the diffusion of tretinoin from the carbopol gel increased (Fig. 2). The results suggested that diffusion of lipophilic drugs, such as tretinoin is highly dependent upon the pH of the vehicle. In my previous work [9], the increase of pH in the carbopol gels showed increased viscosity and bioadhesive strength, showing the high viscosity when neutralized. For this, the pH of carbopol gel was adjusted to eight for the next experiments.

3.3. Effect of tretinoin concentration on drug release

The effect of tretinoin concentration on drug release across synthetic cellulose membrane was studied using 40% DGME solution from the prepared 2% carbopol gels at

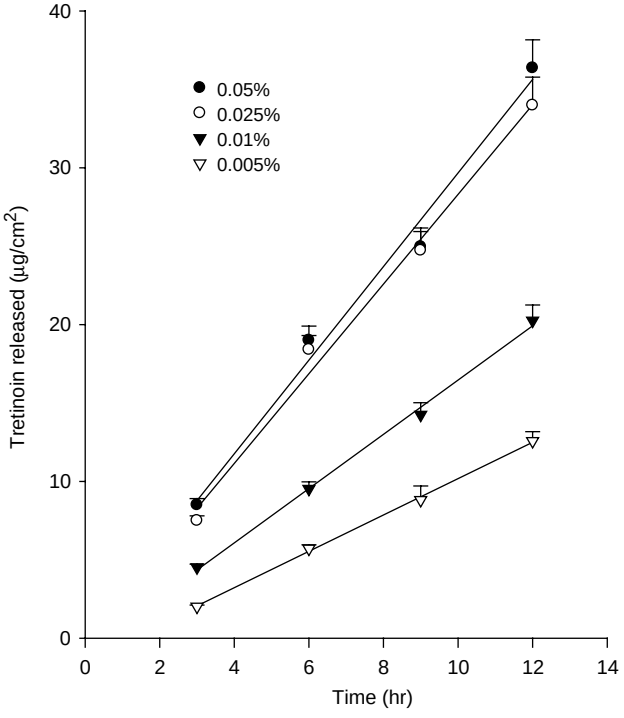

Fig. 3. Effect of tretinoin concentration on drug diffusion from the 2% carbopol gels.

37 ± 0.5 °C. The concentrations tested were 0.005, 0.01, 0.025, and 0.05%, respectively. The flux of tretinoin from the gel formulations through synthetic cellulose membrane (Spectra/por MW 12-14,000) for 12 h was shown in Fig. 3. The release of tretinoin from the gels increased with an increased drug concentration. If the tretinoin concentration is over 0.05%, unpleasant side effects often appear in the form of scaling, erythema, burning and stining [20,21]. Since tretinoin is mostly used at 0.025% concentration in a dermatological preparation, the gels of 0.025% tretinoin concentration was used in permeation test to select an appropriate enhancer.

3.4. Effect of temperature on the drug release

The effect of temperature on of tretinoin release from the gel formulations was evaluated at 28, 32, 37 and 42 °C. All experiments were carried out at least in triplicate. The relationship between the diffusion coefficient and the temperature is as follows:

$$D = D_o\, e^{-E_a/RT} \quad (1)$$

The permeability coefficient is then defined by:

$$P = \text{flux/solubility} \quad (2)$$

$$P = P_o\, e^{-E_a/RT} \quad (3)$$

$$\log P = \log P_o - E_a/R \times 2.303 \times 1000 \times 1000/T \quad (4)$$

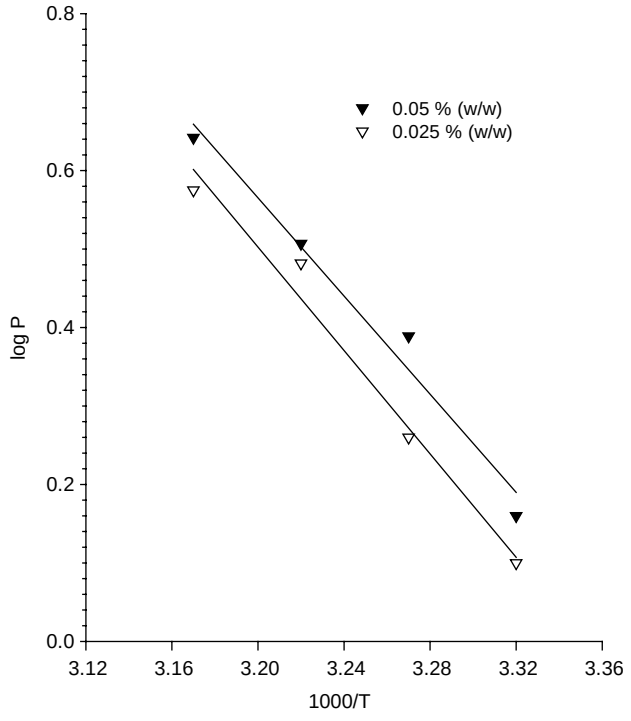

Fig. 4. Effect of temperature on tretinoin diffusion from the 2% carbopol gels containing various drug concentration.

Table 1
Effect of various enhancers on tretinoin permeation from the 2% carbopol gels through rat skin

	Enhancer	Flux (ug/cm^2/h)	Enhancement factor
Glycols	Diethylene glycol	1.17 ± 0.11	2.38
	Tetraethylene glycol	1.13 ± 0.11	2.30
Non-ionic surfactants	Polyoxyethylene 2-oleyl ether	1.474 ± 0.13	3.01
	Polyoxyethylene 23-lauryl ether	0.66 ± 0.05	1.34
	Polyoxyethylene 2-stearyl ether	0.62 ± 0.05	1.25
Control	No-enhancer	0.49 ± 0.04	1.00

A linear relationship was observed between the natural logarithm of permeability coefficient (P) and the reciprocal of temperature (T). The apparent diffusion coefficient of tretinoin increased with increased temperature.

As expected from Eq. (4), a plot of log P versus 1000/T yields a straight line (Fig. 4). The E_a (activation energy) was measured from the slope of log P versus 1000/T plots. The activation energy was 14.96 kcal/mol for 0.025% concentration, and 14.55 kcal/mol for 0.05% concentration. The observation indicates clearly that the release of drug from the gels is an energy-linked process [22]. But, for the practical use, the temperature 37 °C was chosen for the permeation experiments to reflect the skin temperature [23].

3.5. Permeation of tretinoin from the gels containing various enhancers across the rat skin

The effects of enhancer on the permeation of tretinoin across the rat skin were investigated. Table 1 represents the permeation data of tretinoin with/without enhancers. In case of gels containing the permeation enhancers such as the glycols, the non-ionic surfactants, showed good enhancement compared to the control. The permeation of tretinoin from the gels containing an enhancer showed better enhancing effect than that without an enhancer. Among the enhancers used, polyoxyethylene 2-oleyl ether showed the best enhancement considering the enhancement factors.

In my previous study [14,24] on the effects of penetration enhancer by thermal analysis, histological examination, the results suggested that incorporation of penetration enhancer decreased the lipid order and shows the fluidizing effect on the lipids of the stratum corneum. The role of penetration enhancer could possibly be explained as an interfacial saturation phenomenon. For the stratum corneum lipids to be solubilized, it is most likely necessary that enhancers such as surfactant, after having penetrated into the tissue, accumulate at the lipid/liquid interface. The enhancer might affect the fluidity of stratum corneum structure and drugs could be permeated better through the rat skin.

4. Conclusions

The release of tretinoin from the carbopol gels was affected by the pH of carbopol gels, showing the best release at pH 8. As the concentration of drug increased, the release of drug from the gel increased, showing concentration dependency. The increase of temperature showed the increased drug release, depending on the activation energy of permeation. Among the enhancers used such as the glycols and the non-ionic surfactants, polyoxyethylene 2-oleyl ether showed the best enhancing effect. The 0.025% tretinoin-carbopol gels containing an enhancer could be developed for the enhanced transdermal delivery of drug.

Acknowledgements

This work was supported by Korea Research Foundation Grant (KRF-2003-002-E00208).

References

[1] S.A. Mandy, E.G. Thorne, A comparison of the efficacy and safety of tretinoin cream 0.025% and 0.05%, Adv. Ther. 7 (1990) 94–99.
[2] O.H. Mills Jr., A.M. Kligman, Treatment of acne vulgaris with topically applied erythromycin and tretinoin, Acta Derm. Venereol. 58 (1978) 555–557.
[3] J. Nyirady, R.M. Grossman, M. Nighland, R.S. Berger, J.L. Jorizzo, Y.H. Kim, A.G. Martin, A.G. Pandya, K.K. Schulz, J.S. Strauss, A comparative trial of two retinoids commonly used in the treatment of acne vulgaris, J. Derm. Treat. 12 (2001) 149–157.
[4] C.E. Orfanos, R. Ehlert, H. Gollnick, The retinoids, a review of their clinical pharmacology and therapeutic use, Drugs 34 (1987) 459–503.
[5] M.K. Polano, Tretinoin in de dermatologie, Pharm. Weekblad 109 (1974) 908–909.
[6] P. Montassier, D. Duchene, M.C. Poelman, In vitro release study of tretinoin/cyclodextrin derivative complexes, J. Inclusion Phenom. Mol. Recognit. Chem. 31 (1998) 213–218.
[7] H. Gollnick, M. Schramm, Topical drug treatment in acne, Dermatology 196 (1998) 119–125.
[8] B.F. Goodrich, Product information on carbopol, Pharm. Res. 11 (1995) 1396–1400.
[9] S.C. Shin, J.Y. Kim, I.J. Oh, Mucoadhesive and physicochemical characterization of carbopol-poloxamer gels containing triamcinolone acetonide, Drug Dev. Ind. Pharm. 26 (2000) 307–312.
[10] M. Zhou, M.D. Donovan, Intranasal mucociliary clearance of putative bioadhesive polymer gels, Int. J. Pharm. 135 (1996) 115–125.
[11] B.J. Aungst, N.J. Rogers, E. Shegter, Enhancement of naloxone penetration through human skin in vitro using fatty acids, fatty alcohols, surfactants, sulfoxides and amides, Int. J. Pharm. 33 (1986) 225–234.
[12] M. Ishida, Y. Machida, N. Nambu, T. Nakai, New mucosal dosage form of insulin, Chem. Pharm. Bull. 29 (1981) 810–816.
[13] A. Coutel-Egros, Y. Maitani, M. Veillard, Y. Machida, T. Nagai, Combined effects of pH cosolvent and penetration enhancers on the in vitro buccal absorption of propanolol through excised hamster cheek pouch, Int. J. Pharm. 84 (1992) 117–128.
[14] S.C. Shin, J.Y. Kim, Enhanced permeation of triamcinolone acetonide through the buccal mucosa, Eur. J. Pharm. Biopharm. 50 (2000) 217–220.
[15] S.C. Shin, J. Kim, M.K. Yoon, I.J. Oh, J.S. Choi, Transdermal delivery of triprolidine using TPX polymer membrane, Int. J. Pharm. 235 (2002) 141–147.
[16] M. Kartz, B. Polsen, Absorption of drugs through the skin, Handbook Exp. Pharmacol. 28 (1971) 103–174.
[17] R. Wyss, F. Bucheli, Quantitative analysis of retinoids in biological fluids by high-performance liquid chromatography using column switching. I. Determination of isotretinoin and tretinoin and their 4-oxo metabolites in plasma, J. Chromatogr. 424 (1988) 303–314.
[18] A. Zarghi, M. Jenabi, A.J. Ebrahimian, HPLC determination of the stability of tretinoin in tretinoin-minoxidil solution, Pharm. Acta Helv. 73 (1998) 163–165.
[19] H. Durrhein, G.L. Flynn, W.I. Higuchi, C.R. Behl, Permeation of hairless mouse skin I: experimental methods and comparison with human epidermis permeation by alkanols, J. Pharm. Sci. 69 (1980) 781–787.
[20] D.J. Elbaum, Comparison of the stability of topical isotretinoin and topical tretinoin and their efficacy in acne, J. Am. Acad. Derm. 19 (1988) 486–491.
[21] D.A. Schlichting, W.M. Wooding, M. Brown, Optimizing dermatological formulations of retinoic acid, J. Pharm. Sci. 62 (1973) 388–391.
[22] S. Miyazaki, S. Takeuchi, C. Yokouchi, M. Takada, Pluronic F-127 gels as a vehicle for topical administration of anticancer agents, Chem. Pharm. Bull. 32 (1984) 4205–4208.
[23] Y.W. Chien, E.P.K. Lau, Controlled drug release from polymeric delivery devices IV: in vitro–in vivo correlation of subcutaneous release of norgestomet from hydrophilic implants, J. Pharm. Sci. 65 (1976) 488–494.
[24] S.C. Shin, C.W. Cho, I.J. Oh, Effect of non-ionic surfactants as penetration enhancers towards piroxicam from the poloxamer gel through rat skin, Int. J. Pharm. 222 (2001) 193–203.