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Rapid Communication

The Control of Skin-Permeating Rate of Bisoprolol by Ion-Pair Strategy for Long-Acting Transdermal Patches

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Abstract. A moderate drug permeating rate (flux) is desirable for long-acting transdermal patches. In this work, a novel simple method of controlling bisoprolol (BSP) flux by ion-pair strategy was initiated. Different ion-pair complexes including bisoprolol maleate (BSP-M), bisoprolol tartarate, bisoprolol besilate, and bisoprolol fumarate were prepared and their fluxes through rabbit abdominal skin were determined separately *in vitro*. Furthermore, permeation behavior from isopropyl myristate, solubility index in pressure-sensitive adhesives, determined by DSC, and *n*-octanol/water partition coefficient (log *P*) were investigated to illustrate the mechanism of drug permeation rate controlling. The results showed that compared to free BSP (J=25.98±2.34 μg/cm^2/h), all BSP ion-pair complexes displayed lower and controllable flux in the range of 0.11 to 4.19 μg/cm^2/h. After forming ion-pair complexes, the capability of BSP to penetrate through skin was weakened due to the lowered log *P* and increased molecule weight. Accordingly, this study has demonstrated that the flux of BSP could be controlled by ion-pair strategy, and among all complexes investigated, BSP-M was the most promising candidate for long-acting transdermal patches.

KEY WORDS: bisoprolol; flux; ion-pair; transdermal.

INTRODUCTION

Bisoprolol, a selective type β_1 adrenergic receptor blocker, has been widely used for hypertension treatment (1). US FDA has approved the marketing of bisoprolol tablet, which is administered once daily. However, patients are required to keep taking medicine everyday for a very long time since hypertension is a chronic disease. Therefore, some accidents like missing or repeat of taking medication frequently occur resulting in bad even life-threatening effects. This is particularly dangerous to the elder. An effective transdermal patch which can provide long-acting effects and improve patients' compliance is desirable. The key to achieve long-acting effects for transdermal patches is to control the drug skin-permeating rate (flux) in a moderate range, that is, neither too high and leading to a relatively high blood concentration nor too low to obtain the therapeutic effect. As shown in Table I, our preliminary work showed the flux of bisoprolol (BSP) in 24 h (J= 25.98±2.34 μg/cm^2/h) is too high with reference to the required flux of transdermal administration (2). Reducing drug content in patches can lead to the decrease of flux directly, but it also could bring about the problem of dose depletion for long-acting transdermal patches. Therefore, it was more practically meaningful to control the flux by using different techniques. To date, the most common strategies of controlling flux include the use of release controlling membranes (3,4) and suitable adhesive matrixes (5). In this work, we initiated a simple chemical way of controlling flux, *i.e.*, ion-pair strategy, which might simplify bisoprolol long-acting transdermal patches manufacturing procedures. Since Hadgraft (6) succeeded in enhancing permeation of sodium salicylate using an ethoxylated amine, ion-pair complexes has been extensively employed to enhance skin permeation (7,8). However, to our knowledge, ion-pair strategy has never been reported to control flux before.

The goal of this work was to control BSP flux to a moderate level by applying ion-pair strategy. bisoprolol maleate (BSP-M), bisoprolol fumarate (BSP-F), bisoprolol tartarate (BSP-T), and bisoprolol besilate (BSP-B) were synthesized and the fluxes of them from patches were examined. The permeation experiment from isopropyl myristate (IPM), determination of *n*-octanol/water partition coefficient (log *P*) and solubility index in pressure-sensitive adhesives (PSA) were conducted to explain the permeation controlling mechanism.

EXPERIMENTAL

Materials

Bisoprolol fumarate (Zhuhai, China); fumaric acid (F), maleic acid (M), and tartaric acid (T) (Tianjin, China); benzenesulfonic acid(B) (Shanghai, China); PSA DURO-TAK® 87-4098 (Bridgewater, USA), release liner ScotchPak® 9744; and backing film CoTran® 9700 (St. Paul, USA) were used in this study.

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Table I. Fluxes from Patches, Fluxes from IPM, Log *P*, MW of BSP, and Its Ion-Pair Complexes

	MW	Log *P*	Flux from patches(μg/cm^2/h)	Flux from IPM (μg/cm^2/h)
BSP	325	0.93	25.98±2.34	949.18±87.20
BSP-M	383	−0.62	4.19±0.35	275.99±25.36
BSP-F	383	−1.10	1.30±0.11	276.76±26.20
BSP-T	400	−1.69	0.65±0.07	54.87±6.88
BSP-B	501	−0.61	0.11±0.02	7.85±0.87

Free BSP was extracted by ethyl acetate from BSP-F solution adjusted to pH 12 using a solution of diluted sodium carbonate as described in the method before (10). All complexes were obtained by ion-pair interaction of BSP and F, M, T, and B acid respectively at a molar ratio of 2:1, 2:1, 2:1, and 1:1 in acetone after mechanical agitation for 1 h, and the complexes were characterized by DSC and FTIR.

Preparation of Patches

Adhesive patches containing 16% BSP or ion-pair complexes corresponding to the same amount of BSP were prepared by dissolving drug and PSA in ethanol and mixed thoroughly. The resulting formulation was coated onto release liner (9). The coated release liner was oven-dried at 50°C for 15 min, and then it was laminated with backing film.

Permeation Experiments

A two-chamber side-by-side glass diffusion cell (effective diffusion area=0.95 cm^2) with a water jacket connected to a water bath at 32°C was used to investigate the flux of BSP and its ion-pair complexes (10). The excised rabbit abdominal skin was used as a model and prepared prior to experiments as described in previous study (11). The dermal side of the skin faced the receiver solution, and the receptor cell was filled with 3 mL phosphate-buffered solution (pH 7.4).

As for permeation experiment from patches, the patch was pressed on the skin with the adhesive side facing stratum corneum (SC). For permeation experiment from IPM, the donor cell was filled with a 3 ml suspension of BSP or its ion-pair complexes in IPM. Then 2.0 mL of receptor medium was withdrawn at predetermined time intervals for analysis and replaced with the same volume of fresh receptor medium to maintain sink conditions.

Determination of Log *P*

The classic shake-flask method was applied to determine log *P* (12). Equal volumes (4 ml) of distilled water and *n*-octanol and 8 mg drug were added into a glass-stoppered tube and agitated for 48 h in a thermostatic bath at 32°C. After centrifuging and proper dilution, the drug concentration in the water layer and in the *n*-octanol layer was analyzed by HPLC.

Solubility Index Measured by DSC

The solubility indexes in PSA were measured by DSC (DSC-1 Mettler Toledo, Switzerland) (13). Pieces of adhesive layer of the same size containing 2%, 8%, 16%, 24%, 32% (*w*/*w*) BSP-M or BSP-F were packed into an aluminum pan. The oven temperature was set from 60 to 140°C at an increasing rate of 10°C/min. The heat of fusion for solid drug in PSA was detected as an endothermic peak area and was proportionally increased with the increase of drug concentration. The concentration at the bending point in the profile of heat of fusion *versus* drug concentration in PSA was defined as solubility index.

Quantitative Analysis

The HPLC system was equipped with a Hitachi instrument (UV Detector L-2420 and Pump L-2130) and Diamonsil C-18 reversed-phase column (200×4.6 mm, 5 μm; Dikma Technologies, Beijing China). The mobile phase was a mixture of methanol and 0.05 mol/L $(NH4)_2HPO_4$ water solution (50:50) and the pH was adjusted to 4.0 with phosphoric acid at a flow rate of 1 mL/min. The wavelength was set at 225 nm.

Statistical Analysis

All *in vitro* experiments were replicated at least four times, and the data were calculated and presented as mean ±S.E. For comparison between two groups of data, significance was determined by Student's *t* test. Data were considered significant at $p<0.05$.

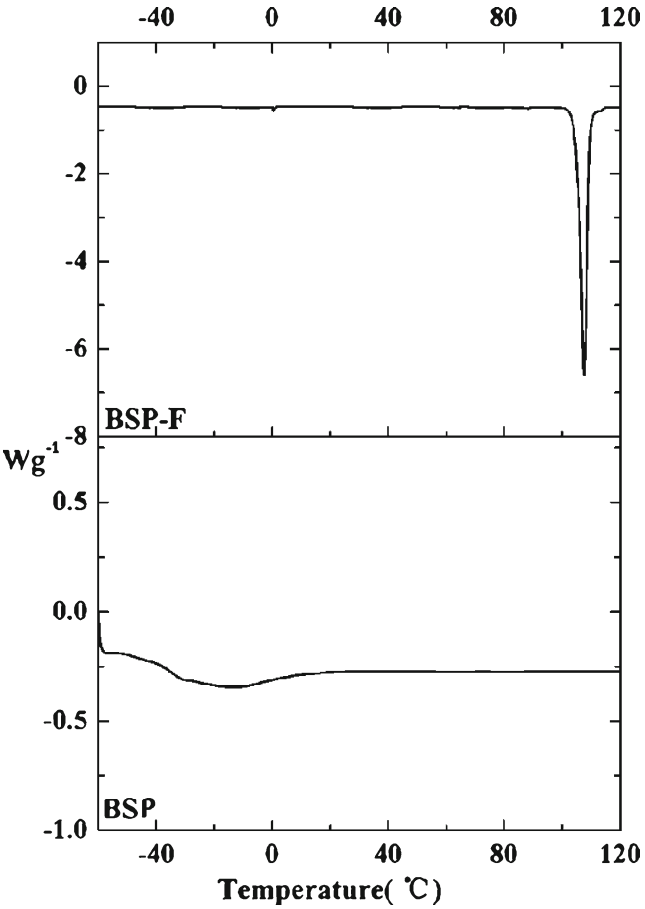

Fig. 1. DSC curves of commercial BSP-F and BSP at a scanning rate of 5°C/min

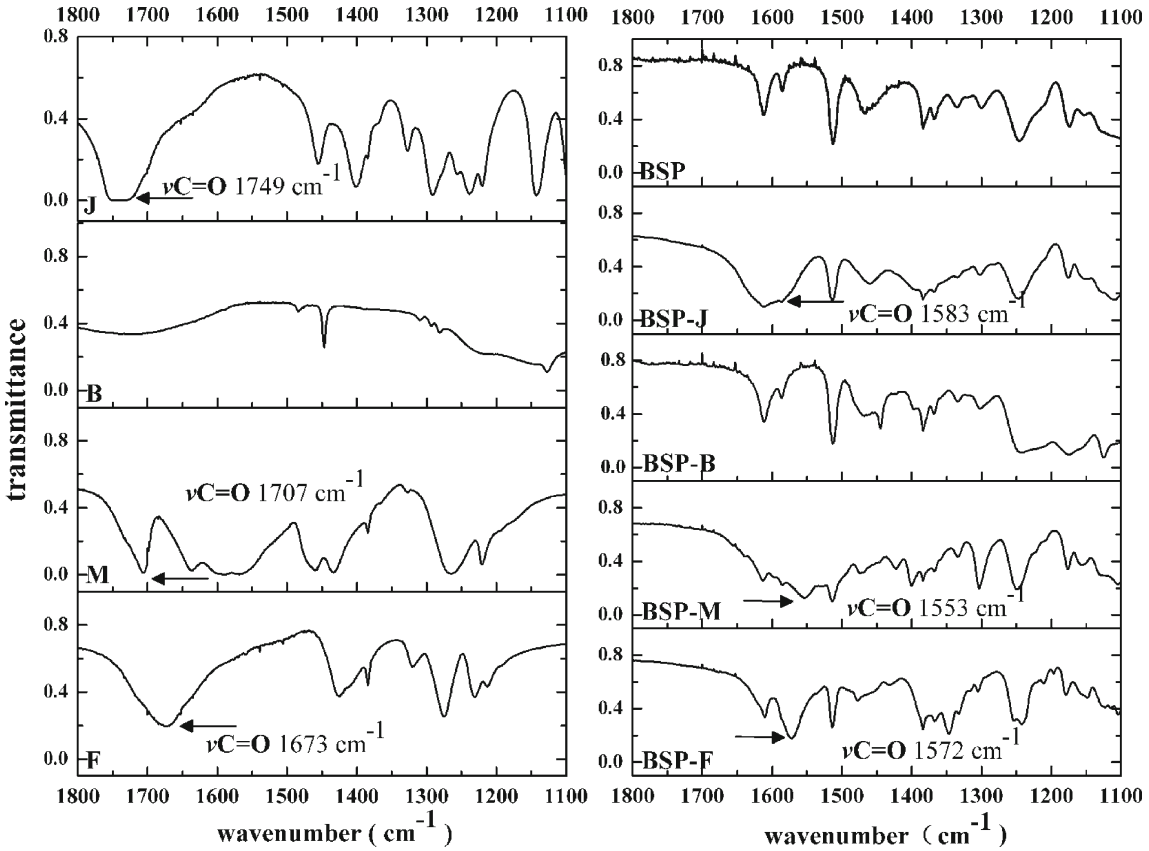

Fig. 2. The IR spectra of BSP and its ion-pair complexes

RESULTS AND DISCUSSION

Characterization of Prepared BSP

The prepared BSP was characterized by DSC. The sharp endothermic peak of BSP-F at 104°C disappeared after synthetic reaction, and a peak of glass transition appeared (Fig. 1), which indicated that BSP might be prepared.

Characterization of BSP Complexes

To prove the formation of different complexes, the IR spectra of them and their corresponding acids were determined (Fig. 2). All of F, M, and J showed a strong signal around 1,700 cm^{-1}, which is the characteristic peak of the carbonyl stretching vibration of acids. However, in the case of BSP-F, BSP-M, and BSP-J, the peak was shifted towards a lower wavenumber, suggesting the existing of ion-pair interaction between BSP and acids. Furthermore, the forming of BSP-B complex might be evidenced from the change of the characteristic of the sulfonic stretching vibration ranging from 1,100 to 1,200 cm^{-1}.

Ion-Pair Complexes Controlled Flux *Via* Controlling Drug Skin Permeability

The *in vitro* permeation experiments from patches were conducted to test whether flux of BSP through rabbit skin can be properly controlled by forming an ion-pair complex. The results were shown in Fig. 3 and the corresponding steady-state fluxes were presented in Table I. As illustrated in Fig. 3 and Table I, among all tested samples, free BSP had the highest flux (J=25.98±2.34 μg/cm^2/h) from 0 to 24 h, but the flux decreased intensely from 24 to 48 h, which may be attributed to dose depletion. The flux of ion-pair complexes

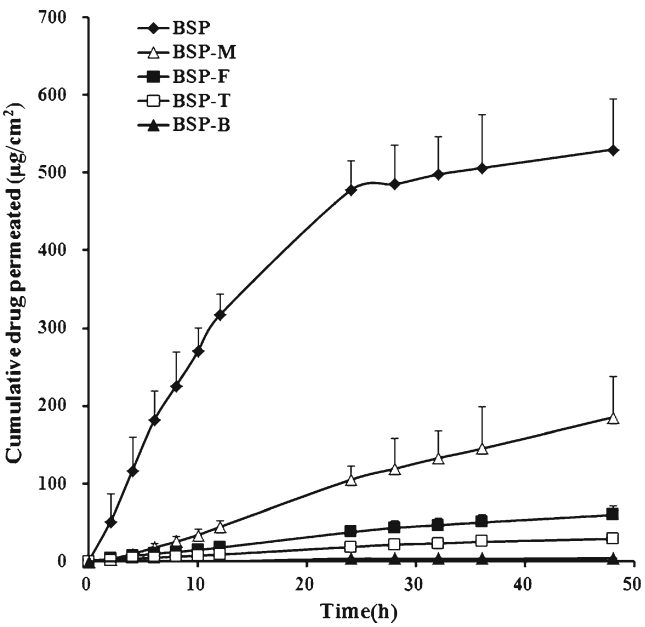

Fig. 3. Effect of ion-pair complexes on the permeation of BSP from patches (n=4)

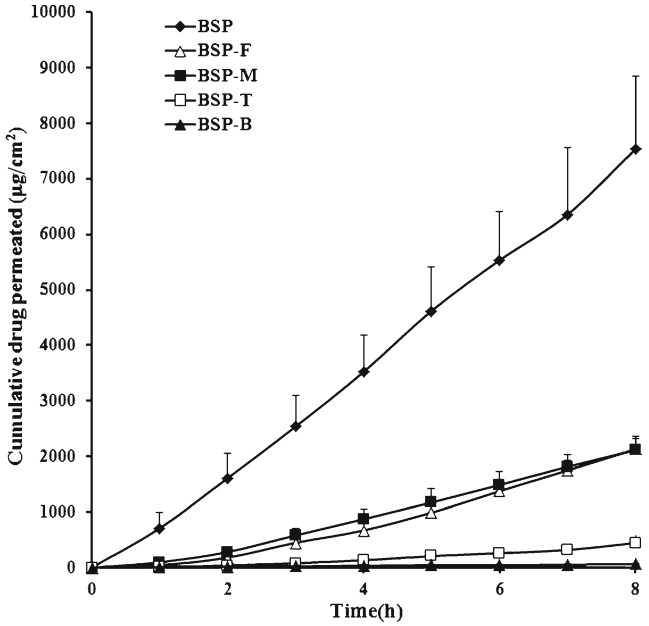

Fig. 4. Effect of ion-pair complexes on the permeation of BSP from IPM (n=4)

was controlled in scope ranging from 0.11 to 4.19 μg/cm^2/h and their fluxes kept constant during the whole experiment period, which suggested that all ion-pair complexes of BSP had the ability to control flux. Nevertheless, it has been defined by Zhao *et al*. (2) that 4 μg/cm^2/h was a critical flux for transdermal administration. Based on this point, BSP-M and BSP-F were more superior than other BSP ion-pair complexes.

IPM is used in cosmetic and topical medicinal preparations where good absorption through the skin is desired. Different from the *in vitro* permeation experiment from patches, the permeation experiment from IPM ignored the complicated influence of patch matrix, thus the flux from IPM can be regarded as an indicator of drug skin permeability. The permeation profiles from IPM and corresponding fluxes were illustrated in Fig. 4 and Table I. In general, the order of fluxes from IPM was consistent with that from patches, which indicated the permeation rate controlling effect by ion-pair complexes from patches was the consequence of controlling drug skin permeability. To further explore the mechanism, the relationships between MW, log *P*, and flux from IPM were investigated.

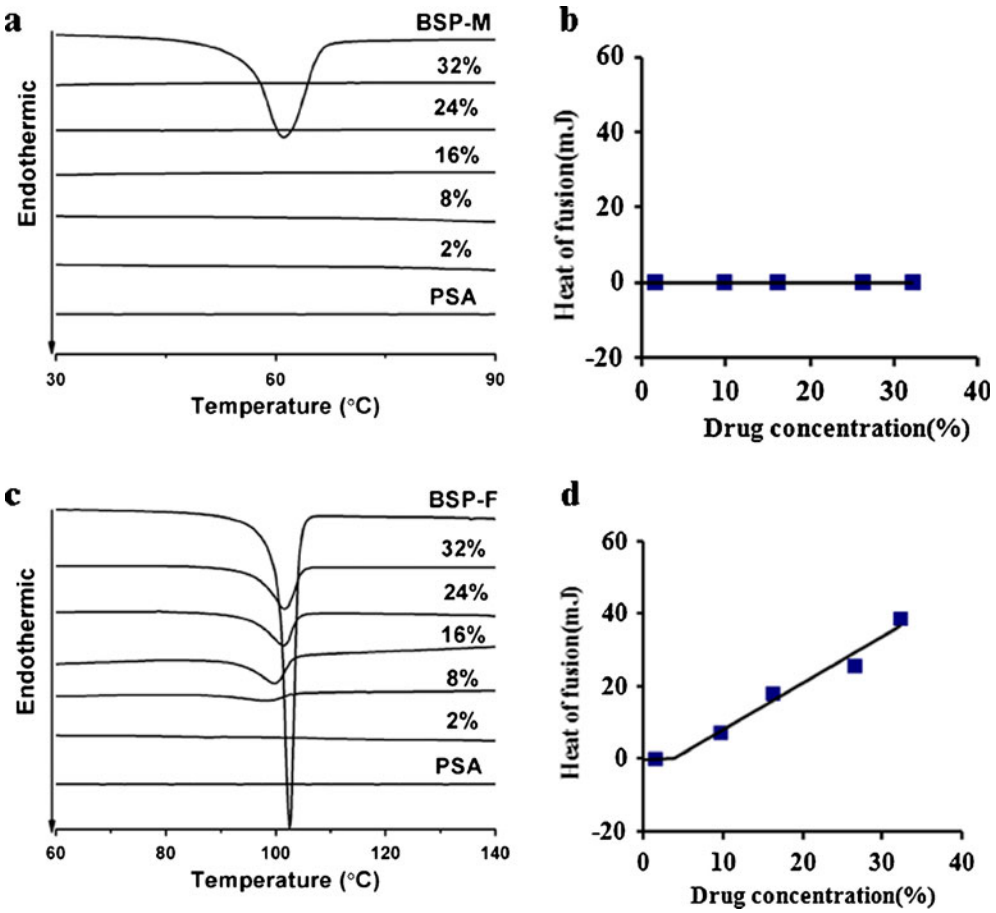

Fig. 5. **a** DSC curves of BSP-M at different concentrations (*PSA* stands for blank matrix, *BSP-M* stands for BSP-M powder). **b** The profile of heat of fusion *versus* BSP-M concentration in PSA. **c** DSC curves of BSP-F at different concentrations (*PSA* stands for blank matrix, *BSP-F* stands for BSP-F powder). **d** The profile of heat of fusion *versus* BSP-F concentration in PSA

Drug Skin Permeability Was Determined by the Combined Effects of Log *P* and MW

Log *P*, MW of BSP, and its ion-pair complexes were also shown in Table I. It was obvious that the flux from IPM was increased with the increase of log *P*, and inversely proportional to MW (r=0.91). Therefore, it can be inferred that the drug skin permeability was affected by both of Log *P* and MW. Skin's structure can be simplified as a two-layer model consisting of lipophilic SC and aqueous viable epidermis (ED). Consequently, drug like BSP with proper log *P* can permeate through both of SC and ED quickly. The ability of BSP ion-pair complexes penetrating through lipophilic SC declined due to their lower Log *P* than that of free BSP, and therefore the flux was well controlled in the moderate range. Ma *et al.* (14) also has proved similar opinion before by showing that the lowering of log *P* to a proper level can lead to the permeation enhancement of flurbiprofen. Furthermore, as highlighted in the research of Magnusson *et al.* (15), the flux was affected by MW greatly. The increase of MW after ion-pair complexes formation, to some extent, hindered the diffusivity of drug molecule and consequently controlled skin permeability.

Comparative Analysis Between BSP-M and BSP-F

Noticeably, it was found that the flux of BSP-M from patches was three times higher than that of BSP-F (P<0.05). But the fluxes of them from IPM were not significantly different (P>0.05), which indicated that these two ion-pair complexes had no significant difference in drug skin permeability. In the process of preparing BSP-F patches, a small amount of drug solid was visible at the surface of the patch. So we assumed that the different flux was associated with different drug dissolution in PSA. To verify the hypothesis, solubility index of two ion-pair complexes in the PSA matrix were determined by DSC. DSC curves of BSP-F or BSP-M at different concentrations and the profiles of heat of fusion *versus* BSP-F or BSP-M concentration in PSA were shown in Fig. 5. The solubility index of BSP-F was 3.67%. However, in patches of BSP-M, no endothermic peak was detectable, which demonstrated that BSP-M was thoroughly dissolved in PSA at the concentration up to 32%. Unfortunately, it was infeasible to prepared BSP-M patch with higher BSP-M content due to the disagreeable character of PSA. So we did not get the solubility index of BSP-M in PSA, but we are sure that it must be higher than 32%. As illustrated in the Fick's first law, the flux in a given direction through skin is directly proportional to the concentration gradient in PSA—the steeper the concentration gradient, the faster the flux. At the concentration of 16%, at which permeation experiments were conducted, BSP-M was completely dissolved in PSA, but BSP-F dissolved partially and left the rest in the form of solid. The different drug amount dissolved in patches resulted in different concentration gradient, thereby led to different fluxes. In consideration of different drug loading capacity resulted from different solubility index, BSP-M was more promising than BSP-F for applying to long-acting transdermal patches.

CONCLUSION

This work evaluated skin permeation controlling effect of BSP ion-pair complexes and proposed preliminary explanation of the mechanism. It was concluded that BSP ion-pair complexes have the potential to be applied for long-acting transdermal patches and BSP-M was the most promising candidate. Furthermore, the employing of ion-pair complexes in this work will facilitate the manufacturing of long-acting transdermal patches as well as provide new ideas for ion-pair complexes application. We will synthesize various BSP-acid complexes and optimize the formulation of the long-acting patches in further study.

REFERENCES

1. Czuriga I, Riecansky I, Bodnar J, Fulop T, Kruzsicz V, Kristof E, *et al*. Comparison of the new cardioselective beta-blocker nebivolol with bisoprolol in hypertension: the Nebivolol, Bisoprolol Multicenter Study (NEBIS). Cardiovasc Drugs Ther. 2003;17:257–63.
2. Zhao JH, Fu JH, Wang SM, Su CH, Shan Y, Kong SJ, *et al.* A novel transdermal patch incorporating isosorbide dinitrate with bisoprolol: *in vitro* and *in vivo* characterization. Int J Pharm. 2007;337:88–101.
3. Kim J, Shin SC. Controlled release of atenolol from the ethylene-vinyl acetate matrix. Int J Pharm. 2004;273:23–7.
4. Zhan XP, Chen SJ, Tang GC, Mao ZM. Poly (2-hydroxy-3-phenoxypropylacrylate, 4-hydroxybutyl acrylate, dibutyl maleate) membrane controlled clonidine zero-order release. Eur J Pharm Biopharm. 2007;66:429–34.
5. Mukherjeea B, Mahapatraa S, Guptab R, Patraa B, Tiwari A, Arorab P. A comparison between povidone-ethylcellulose and povidone-eudragit transdermal dexamethasone matrix patches based on *in vitro* skin permeation. Eur J Pharm Biopharm. 2005;59:475–83.
6. Hadgraft J, Walters KA, Wotton RK. Facilitated percutaneous absorption: a comparison and evaluation of two *in vitro* models. Int J Pharm. 1986;32:257–63.
7. Cheong HA, Choi HK. Enhanced percutaneous absorption of piroxicam via salt formation with ethanolamines. Pharm Res. 2002;19:1375–80.
8. Nam SH, Xu YJ, Nam H, Jin GW, Jeong Y, An S, *et al.* Ion pairs of risedronate for transdermal delivery and enhanced permeation rate on hairless mouse skin. Int J Pharm. 2011;419:114–20.
9. Ren CS, Fang L, Li T, Wang ML, Zhao LG, He ZG. Effect of permeation enhancers and organic acids on the skin permeation of indapamide. Int J Pharm. 2008;350:43–7.
10. Zhao LG, Li Y, Fang L, He ZG, Liu XT, Wang L, *et al.* Transdermal delivery of tolterodine by O-acylmenthol: *in vitro*/*in vivo* correlation. Int J Pharm. 2009;374:73–81.
11. Li CM, Liu C, Liu J, Fang L. Correlation between rheological properties, *in vitro* release, and percutaneous permeation of tetrahydropalmatine. AAPS PharmSciTech. 2011;12:1002–10.
12. Fang L, Numajiri S, Kobayashi D, Morimoto Y. The use of complexation with alkanolamines to facilitate skin permeation of mefenamic acid. Int J Pharm. 2003;262:13–22.
13. Sato K, Mitsui N, Hasegawa T, Sugibayashi K, Morimoto Y. Potential usefulness of solubility index for prediction of the skin permeation rate of 5-ISMN from pressure-sensitive adhesive tape. J Control Release. 2001;73:269–77.
14. Ma X, Fang L, Guo GP, Zhao NX, He ZG. Effect of counter-ions and penetration enhancers on the skin permeation of flurbiprofen. J Pharm Sci. 2009;99:1826–37.
15. Magnusson BM, Anissimov YG, Cross SE, Roberts MS. Molecular size as the main determinant of solute maximum flux across the skin. J Invest Dermatol. 2004;122:993–9.