약제학회지(2001), 제 31 권 제 3 호
J. Kor. Pharm. Sci., Vol. 31, No. 3, 181-184 (2001)

Development of Transdermal Drug Delivery System for the Combination of Physostigmine and Procyclidine

Soon-Cheol Park and Hoo-Kyun Choi†

College of Pharmacy, Chosun University, Kwangju 501-759, Korea

(Received August 2, 2001)

ABSTRACT–The purpose of this study was to develop transdermal drug delivery system (TDDS) for the combination of physostigmine and procyclidine. The effects of various pressure sensitive adhesives (PSA) on the percutaneous absorption of procyclidine across hairless mouse skin were evaluated to select an appropriate PSA. In addition, the influences of various vehicles on the percutaneous absorption of procyclidine from PSA matrix across hairless mouse skin were evaluated using flow-through diffusion cell system at 37°C. Physostigmine did not have any influence on the permeation rate of procyclidine. The flux of procyclidine was the highest in silicone and PIB and was relatively lower in SIS, Acryl, and SBS adhesive matrices, however, their use was limited by the crystallization of the drug in the matrix. Among acrylic adhesives, the permeability of procyclidine was the highest from poly (ethylene oxide) grafted acrylic adhesive. Some enhancers show different enhancing effect depending on the drug, however, many of the tested enhancers showed enhancing effect for the permeation of both procyclidine and physostigmine to some extent. Crovol® EP 40 showed the highest enhancing effect on the permeation of both compounds. The size of TDDS to provide required permeation rate was estimated to be 35 cm^2 based on available information.

Keywords–Transdermal drug delivery, Physostigmine, Procyclidine, Pressure sensitive adhesive, Enhancer

It has been reported that pretreatment with carbamate prophylactics followed by treatment with cholinolytic atropine is very effective in reducing the lethal effects of diverse organophosphate compounds.[1,2] In spite of their potential for reducing the lethality of organophosphates, the combination could not prevent organophosphate-induced brain damages. However, procyclidine (1-cyclohexyl-1-phenyl-3-(pyrrolidin-1-yl)-propan-1-ol) possessing both cholinolytic and NMDA-antagonistic actions could be a promising antidote, substituting for atropine, for the prevention of lethality and brain damages induced by organophosphate poisoning.[3,4]

Physostigmine and pyridostigmine, both carbamate prophylactics, have been shown to be effective for the treatment of cholinergic disorders and as a pretreatment against organophosphate poisoning. Physostigmine, being a tertiary amine that readily enter the central nervous system, was shown to be more effective in the protection against the organophosphate poisoning than the quaternary, peripherally acting pyridostigmine.[5-7] However, the short half-life and narrow therapeutic window of physostigmine limited its clinical use.[8,9]

In order to overcome this difficulty, transdermal delivery of the drug is promising route of administration.[10,11] The medication of physostigmine by transdermal drug delivery system (TDDS) has some advantages. The drug delivered by TDDS can avoid first-pass metabolism. The risks and inconveniences of parental therapy and the variable absorption and metabolism associated with oral therapy can be avoided. Also, TDDS can provide the capacity for multiday therapy with a single application, thereby improving patient compliance. In spite of these advantages of TDDS, only a limited number of drugs have been used to develop the system due to excellent barrier function of the skin. Currently, the most widely utilized approach to overcome the barrier function of skin involves the use of chemical permeation enhancers.

Until now, the study for transdermal delivery of procyclidine has not been reported. A limited number of studies have been done to develop transdermal delivery system for physostigmine, however, they didnt consider the effects of PSA on the permeation of physostigmine and the effect of various enhancers in PSA.[5,10-13] Considering the fact that the effects of enhancers in solution formulations can be different from those in the adhesive matrix of TDDS, their studies have limitations in developing the matrix type TDDS for physostigmine and procyclidine.[14] Therefore, to develop the transdermal drug delivery system for the combination of physostigmine and procyclidine we investigated the effect of PSA on permeation of

†본 논문에 관한 문의는 이 저자에게로
Tel : (062)230-6367 E-mail : hgchoi@chosun.ac.kr

physostigmine and procyclidine. The effects of enhancers within PSA were also studied.

Experimental

Materials

Procyclidine base was obtained from its HCl salt purchased from Sigma Chemical Co. (St. Louis, MO). Physostigmine base was purchased from Sigma Chemical Co. PEG-6 glyceryl menooleate (Labrafil® 1944), PEG-6 glyceryl linoleate (Labrafil® 2609), caprylic/capric triglyceride (Labrafac® CC), PG caprylate/caprate (Labrafac® PG), PEG-8 glyceryl catrylate/caprate (Labrasol®) and polyglyceryl-3 oleate (Plurol oleique® C497) were obtained from Gattepose Korea (Seoul, Korea). Propylene glycol dicaprylate/dicaprate (Miglyol® 840) was obtained from Hüls America (Edison, NJ). PEG-20 evening primrose glycerides (Crovol® EP40), PEG-20 almond glyceride (Crovol® A40), PEG-60 almond glyceride (Crovol® A70), corn oil and PEG-30 castor oil (Incrocas® 30) were obtained from Croda Inc. (Parsippany, NJ). Sorbitan monolaurate (Span® 20), sorbitan monooleate (Span® 80), PEG sorbitan monolaurate (Tween® 20), PEG sorbitan monooleate (Tween® 80), oleic acid, oleyl alcohol and propylene glycol were purchased from Junsei Chemical Co. (Japan). Polystyrene-polybutadiene-polystyrene (SBS) and acrylic pressure sensitive adhesive solutions in organic solvents were obtained from National Starch and Chemical Company (Bridgewater, NJ). Polyisobutylene (PIB) (Vistanex LM-MH, Vistanex MML -100) were obtained from Jeil Pharm. Co. (Seoul, Korea). Polystyrene-polyisoprene-polystyrene (SIS) and silicone pressure sensitive adhesive was obtained from Shell Chemicals (Stanlow, UK) and Dow Corning (Midland, MI), respectively. All other chemicals were reagent grade or above and were used without further purification.

Preparation of adhesive matrices

SBS, PIB, SIS, silicone or acrylic adhesive solution in organic solvent mixture was mixed with procyclidine and/or physostigmine solution in ethylacetate with or without enhancer according to the study protocol. Pressure sensitive adhesive matrix was prepared by casting the above solutions on polyester release liner using a casting knife. It was set at room temperature for 20 minutes and was subsequently oven-dried at 80°C for about 15 minutes to remove the residual organic solvents. The dried film was laminated onto a backing film.

Penetration studies

A flow-through diffusion cell system, the preparation of hairless mouse skins, procedure of the penetration studies, and data reduction methods have been described in an earlier study.[14] The penetration samples were collected every 4 hr for 36 hr or longer.

Analytical conditions

The HPLC method was used to analyze procyclidine and physostigmine. A reverse-phase column (Alltima C8, Alltech Ass., IL) was used. The column temperature was maintained at 30°C by a thin foil temperature controller (CH1445, Systec, MN). The flow rate was 1 ml/min. The wavelengths of UV detector for analyzing procyclidine and physostigmine were 210 nm and 235 nm, respectively. The compositions of mobile phase for procyclidine and physostigmine were acetonitrile/water/triethylamine/phosphoric acid (449/549/1/1) and methanol/water/triethylamine/phosphoric acid (299/699/1/1), respectively.

Results and Discussion

Effect of pressure sensitive adhesive

The permeation rate of procyclidine across hairless mouse skin from silicone, SIS, SBS, PIB, and acrylic pressure sensitive adhesive matrices (PSA) were tested. The amount of procyclidine in each PSA tested was 10% of the weight of the adhesive polymer. Figure 1 shows the effect of various adhesive matrices on the permeation of procyclidine across hairless mouse skin. The flux of procyclidine was the highest in silicone and PIB and was relatively lower in SIS, Acryl, and SBS adhesive matrices. It was observed that the procyclidine crystallized in silicone and PIB adhesive matrices, in which the drug showed the highest permeability. The crystallization of procyclidine indicates that the drug is saturated in the adhesive

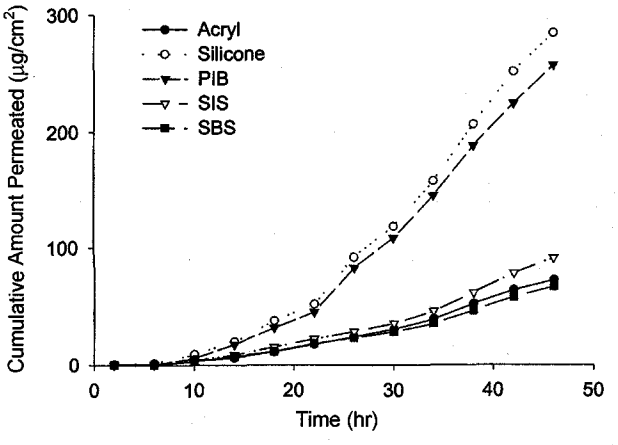

Figure 1–Effect of various adhesive matrices on the permeation of procyclidine across hairless mouse skin.

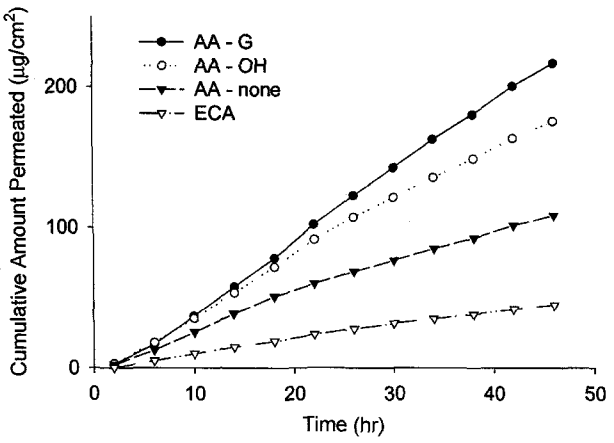

Figure 2–Effect of functional group of acrylic adhesive on the permeation of procyclidine across hairless mouse skin.

matrix due to low solubility. It also indicates that the drug has the maximum thermodynamic activity in silicone and PIB matrices, resulting in higher permeability.[15]

Figure 2 shows the effect of various functional groups of acrylic adhesive on the permeation of procyclidine across hairless mouse skin. The amount of procyclidine in each PSA tested was 10% of the weight of the adhesive polymer. The highest permeability of procyclidine was obtained from poly (ethylene oxide) grafted acrylic adhesives (AA-G) followed by acrylic adhesive with hydroxyl group (AA-OH), without functional group (AA-none), and highly cross-linked enhancer compatible acrylic adhesive (ECA). It was interesting to note that no significant difference in the permeability of procyclidine was observed within a same type of acrylic adhesives having same functional groups (Data not shown). Even though all the acrylic adhesives tested had similar solubility parameter, the permeabilities were widely different depending the functional group. The results indicate that not only lipophilicity of the matrix but the potential interaction of functional group of the adhesive and the drug are important in determining the release rate of the drug from TDDS.

In our previous study, the effects of various adhesive matrices on the permeation of physostigmine were studied (data not shown). The order of permeability of physostigmine from PSA was same with that of procyclidine. The fluxes of procyclidine and physostigmine from silicone and PIB matrix were higher than from other adhesives tested. However, incorporation of drugs into silicone and PIB significantly decreased the tack of PSA. To select appropriate PSA for developing TDDS, the tack as well as the permeability should be considered. Therefore, the effect of enhancer was investigated using AA-G based on its highest permeability among acrylic adhesives.

Table I–*Effect of Various Enhancers on the Permeation of Procyclidine and Physostigmine across Hairless Mouse Skin from Acrylic PSA Matrix*

Enhancer	Flux (Average ± STDEV, μg/cm² hr)	
	Procyclidine	Physostigmine
Control (without enhancer)	11.1 ± 0.8	7.5 ± 1.0
Labrafil 1944	12.1 ± 2.0	9.3 ± 2.2
Labrafil 2609	13.9 ± 1.3	11.6 ± 0.8
Labrasol	13.0 ± 2.5	9.0 ± 1.4
Crovol EP 40	16.9 ± 2.8	13.1 ± 0.8
Crovol A 40	13.7 ± 2.7	10.9 ± 0.9
Crovol A 70	12.7 ± 0.7	9.6 ± 0.7
Incrocas 30	13.2 ± 1.0	12.2 ± 1.9
Tween 20	14.6 ± 2.4	8.6 ± 1.7
Tween 80	14.5 ± 2.2	10.6 ± 1.0
Labrafac CC	15.5 ± 1.2	10.7 ± 0.8
Plurol Oleique CC 497	14.7 ± 1.8	10.7± 0.6
Corn oil	16.0 ± 1.4	9.7 ± 1.5
Miglyol 840	11.5 ± 1.6	8.1 ± 1.6
Labrafac PG	16.1 ± 3.2	10.1 ± 1.9
Span 20	15.2 ± 1.2	10.5 ± 0.5
Span 80	14.4 ± 1.6	11.4 ± 1.5
Propylene glycol	16.4 ± 3.9	10.0 ± 1.9
Oleyl alcohol	12.0 ± 3.6	11.0 ± 1.2
Oleic acid	11.5 ± 0.4	11.6 ± 0.3

Effect of enhancer

To develop TDDS that is small enough to be used commercially, the fluxes of procyclidine and physostigmine from acrylic adhesive matrix across hairless mouse skin should be improved. The fluxes of procyclidine and physostigmine within the same patch were evaluated. Procyclidine or physostigmine did not affect significantly the permeability of each other (data not shown). Table I shows the effect of various enhancers on the permeation of procyclidine and physostigmine across hairless mouse skin from acrylic adhesive matrix. The contents of procyclidine, physostigmine and each enhancer used were 10%, 3% and 5% of the weight of adhesive polymer, respectively. Some enhancers show different enhancing effect depending on the drug. Oleic acid enhanced the permeation of physostigmine, however, it did not enhance the permeation rate of procyclidine. On the contrary, Tween® 20 enhanced the permeation rate of procyclidine, but it showed only a marginal effect on the permeation rate of physostigmine. The results suggest that the mechanisms of these enhancers are related to the physicochemical property of drug as well as the change of skin barrier property. Oleic acid has been used for the enhancement of transdermal delivery of physostigmine within pad formulation.[5,11] However, the results in

this study showed that oleic acid is not adequate for transdermal delivery of the combination of procyclidine and physostigmine within adhesive polymer. Many of the tested enhancers showed enhancing effect for the permeation of both procyclidine and physostigmine to some extent. Crovol® EP 40 showed the highest enhancing effect on the permeation of both compounds. Incorporation of Crovol® EP 40 into AA-G increased the fluxes of procyclidine and physostigmine by 1.5 and 1.8 times, respectively.

To be clinically useful, the size of a patch should be considered. The fluxes of procyclidine and physostigmine from the acrylic matrix formulation containing Crovol® EP40 were 17 and 13 $\mu g/cm^2$ hr, respectively. Clinical studies indicated that the physostigmine infused intravenously at 400 μg/hr produced blood cholinesterase inhibitions of approximately 30%, which does cause overt signs of cholinesterase poisoning nor impair performance.[5] Based on daily dose and bioavailability when administered orally, 11 mg/day of procyclidine must be absorbed transdermally to achieve pharmacological effect.[16] The size of TDDS to provide required permeation rate was estimated to be 35 cm^2, indicating that the development of transdermal delivery system of the combination of procyclidine and physostigmine is feasible.

Acknowledgements

This research was supported by the fund provided by the Agency for Defense Development (ADD).

References

1) W.K. Berry and D.R. Davies, The use of carbamates and atropine in the protection of animals against poisoning by 1,2,2 trimethylpropyl methylphosphono-fluoridate, *Biochem. Pharmacol.*, **19**, 927-934 (1970).
2) J.J. Gordon, L. Leadbeater and M.P. Maidment, The protection of animals against organophosphate poisoning by pretreatment with a carbamate, *Toxicol. Appl. Pharmacol.*, **43**, 207-216 (1978).
3) Y.B. Kim, K.H. Hur, Y.S. Lee, B.G. Han and S. Shin, A role of nitric oxide in organophosphate-induced convulsions. *Environ. Toxicol. Pharmacol.*, **3**, 53-56 (1997).
4) Y.B. Kim, S. Shin, D.E. Sok and J.K. Kang, Effectiveness of procyclidine in combination with carbamate prophylactics against diisopropylfluorophosphate poisoning, *Environ. Toxicol. Pharmacol.*, **5**, 43-49 (1998).
5) A. Jenner, A. Saleem and D. Swanston, Transdermal delivery of physostigmine. A pretreatment against organophosphate poisoning, *J. Pharm. Pharmacol.*, **47**, 206-212 (1995).
6) R.P. Solana, L.W. Harris, W.H. Carter, J.R. Brian, G. Talbot, R.A. Carchman and C. Gennings, Evaluation of a two-drug combination pretreatment against organophosphorus exposure, *Toxicol. Appl. Pharmacol.*, **102**, 421-429 (1990).
7) L. Leadbeater, R.H. Inns and J.M. Rylands, Treatment of poisoning by soman, *Fund. Appl. Tox.*, **5**, S225-S231 (1985).
8) P. Hartvig, L. Wiklund and B. Lindstrom, Pharmacokinetics of physostigmine after intravenous, intramuscular and subcutaneous administration in surgical patient, *Acta. Anaesthesiol. Scand.*, **30**, 177-182 (1986).
9) S.M. Somani and A. Khalique, Pharmacokinetics and pharmacodynamics of physostigmine in the rat after intravenous administration, *Drug, Metab. Dispos.*, **15**, 627-633 (1987).
10) D. Levy, Y. Meshulam, R. Grunwald, R. Bruckstein and A. Levy, A long-acting Transdermal system for the treatment of organophate poisoning, *Proc. 3rd Int. Symp. Protection Against Warfare Agents Umea Sweden Suppl.*, 151-156 (1989).
11) Y. Meshulam, T. Kadar, A. Wengier, S. Dachir and A. Levy, Transdermal penetration of physostigmine: Effects of oleic acid enhancer, *Drug Dev. Res.*, **28**, 510-515 (1993).
12) A. Pardo, Y. Shiri and S. Cohen, Percutaneous absorption of physostigmine: Optimization of delivery from a binary solvent by thermodynamic control, *J. Pharm. Sci.*, **79**, 573-578 (1990).
13) A. Pardo, Y. Shiri and S. Cohen, Partial molal volumes and solubilities of physostigmine in isopropanol: Isopropyl myristate solvents in relation to skin penetrability, *J. Pharm. Sci.*, **80**, 567-572 (1991).
14) J.-H. Kim, Y.-J. Cho and H.-K. Choi, Effect of vehicles and pressure sensitive adhesives on the permeation of tacrine across hairless mouse skin, *Int. J. Pharm.*, **196**, 105-113 (2000).
15) Y.-J. Cho and H.-K. Choi, Enhancement of percutaneous absorption of ketoprofen: effect of vehicles and adhesive matrix, *Int. J. Pharm.*, **169**, 95-104 (1998).
16) P.D. Whiteman, A.S.E. Fowle, M.J. Hamilton, A.W. Peck, A. Bye, K. Dean and A. Webster, Pharmacokinetics and pharmacodynamics of procyclidine in man, *Eur. J. Clin. Pharmacol.*, **28**, 73-78 (1985).