Department of Pharmaceutics, College of Pharmaceutical Sciences, Zhejiang University, Hangzhou, P.R. China

A comparative study of electroporation and iontophoresis for percutaneous penetration of naproxen

Q. H. Hu, W. Q. Liang

Received August 20, 2002, accepted September 12, 2002

Dr. Qiao-Hong Hu, College of Pharmaceutical Sciences, Zhejiang University, 353 Yan'an Road, Hangzhou 310006, P.R. China
Hu_qh@mail.hz.zj.cn

Pharmazie 58: 192–194 (2003)

Using side-by-side compartment diffusion cell and rat skin, the *in vitro* percutaneous penetration of naproxen by electroporation and iontophoresis with different energy was investigated and compared. Both electroporation and iontophoresis could obviously increase the penetration of naproxen through rat skin. The enhancing effect of electroporation was not always better than that of iontophoresis. The enhancing effect was dependent on the energy of the electrical field.

1. Introduction

Many methods have been explored to enhance percutaneous penetration of drugs, including chemical enhancers, ultrasound, iontophoresis and electroporation etc. Electroporation is a method of reversibly permeabilizing lipid bilayers, involving the creation of transient aqueous pathways by a short, high-voltage pulse. It was firstly used in cell biology such as DNA introduction, cell killing under nontoxic conditions and insertion of membrane macromolecules into the cell membrane [1]. Recently it was used in electro-chemotherapy to enhance the introduction of drugs into tumor [2], and in gene delivery [3]. From 1990s on, there have been many reports about enhanced percutaneous penetration of drugs by electroporation [4–10]. Wang et al. [4] investigated the influence of electroporation and iontophoresis on the percutaneous penetration of cyclosporin A. Electroporation could significantly increase the permeation of cyclosporin A. But there was no obvious difference between iontophoresis and passive diffusion concerning drug in the skin and receiver solution. Is electroporation better than iontophoresis to enhance percutaneous penetration of drugs in any conditions? Here we use a cationic drug, naproxen ($M_w = 230.26$, -1 valence) as a model drug to compare the influence of electroporation and iontophoresis with different energy on its *in vitro* percutaneous penetration.

2. Investigations and results

The cumulative quantities and fluxes of naproxen by different electroporation, iontophoresis and passive diffusion are shown in Fig. 1 and Fig. 2. The cumulative quantity of naproxen by electroporation I (voltage = 400 V, capacity = 2.2 μF, pulse frequency = 20 pulses/min, pulse time = 6.0 ms, pulse number = 100, total energy = 17.6 J) was obviously smaller than iontophoresis I (1 mA/cm^2, last for 6 h, total energy = 129.6 J) and iontophoresis II (0.5 mA/cm^2, last for 4 h, total energy = 43.2 J). The flux of naproxen at 0.5 h by electroporation I (43.5 ± 6.3 μg/cm^2 · h) was obviously smaller than that by iontophoresis I

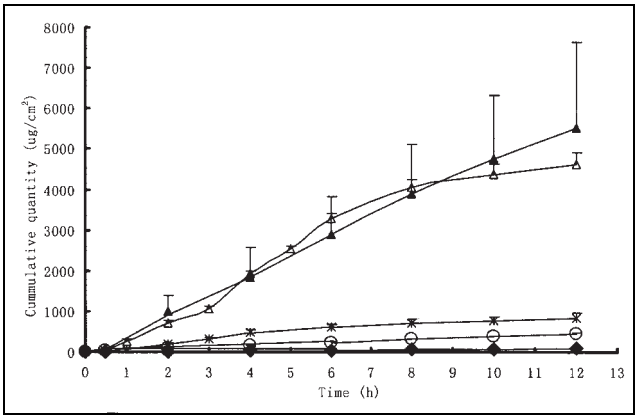

Fig. 1: Cumulative quantities of naproxen under different conditions
—◆— Passive diffusion; —○— Electroporation I; —▲— Electroporation II; —△— Iontophoresis I; —✳— Iontophoress II

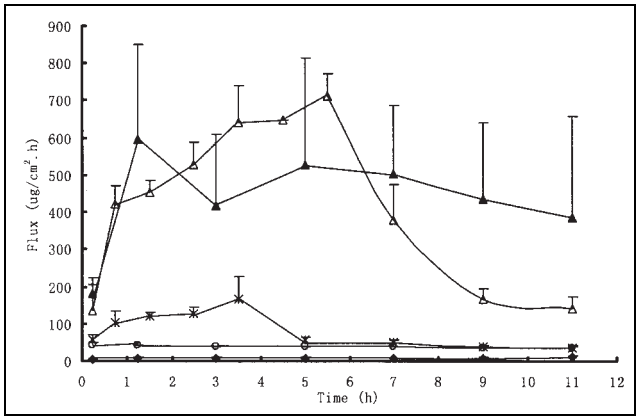

Fig. 2: Fluxes of naproxen under different conditons
—◆— Passive diffusion; —○— Electroporation I; —▲— Electroporation II; —△— Iontophoresis I; —✳— Iontophoress II

($134.9 \pm 69.5\ \mu g/cm^2 \cdot h$), but was not significantly different from that by iontophoresis II ($56.4 \pm 14.1\ \mu g/cm^2 \cdot h$).

The cumulative quantity of naproxen by electroporation II (voltage = 380 V, capacity = 22 μF, pulse frequency = 4 pulses/min, pulse time = 7.4 ms, pulse number = 100, total energy = 158.8 J) was obviously larger than iontophoresis II, but was not significantly different from iontophoresis I. The flux of naproxen at 0.25 h by electroporation II ($180.6 \pm 41.6\ \mu g/cm^2 \cdot h$) was obviously larger than that by iontophoresis II ($56.4 \pm 14.1\ \mu g/cm^2 \cdot h$), but was not significantly different from with that by iontophoresis I ($134.9 \pm 69.5\ \mu g/cm^2 \cdot h$).

The cumulative quantities and fluxes of naproxen by electroporation and iontophoresis were obviously larger than those by passive diffusion.

Fig. 2 shows that the time to achieve steady state of naproxen by electroporation or iontophoresis was obviously shorter than that by passive diffusion. Times were 3 h, 3.5 h and 5 h for electroporation, iontophoresis and passive diffusion, respectively.

Fig. 2 also shows that the fluxes of naproxen by electroporation maintained at a high level even if the electrical field was stopped. The flux of naproxen at 7 h by electroporation I was $37.8 \pm 0.6\ \mu g/cm^2 \cdot h$, and that by electroporation II was $503.9 \pm 182.6\ \mu g/cm^2 \cdot h$. But in iontophoresis, the fluxes of naproxen decreased quickly after the electrical field was stopped. For iontophoresis II, the flux at 3.5 h was $166.0 \pm 59.3\ \mu g/cm^2 \cdot h$ and that at 5 h was $57.6 \pm 6.4\ \mu g/cm^2 \cdot h$. For iontophoresis I, the flux at 5.5 h was $713.8 \pm 60.4\ \mu g/cm^2 \cdot h$ and that at 7 h was $378.1 \pm 97.4\ \mu g/cm^2 \cdot h$.

3. Discussion

When the electrical field is present, the flux of ionic drug through skin is in accordance with the Nernst-Plank equation (1).

$$J_i = D_i \frac{dC_i}{dx} + \frac{D_i z_i e E C_i}{KT} + V_i C_i \qquad (1)$$

In this equation, $D_i \frac{dC_i}{dx}$ represents the flux of drug caused by passive diffusion; $\frac{D_i z_i e E C_i}{KT}$ represents the flux of drug caused by the migration of ion in the electrical field; $V_i C_i$ represents the flux of drug caused by electro-osmotic coupling of drug and solvent flows in the skin. In the condition of iontophoresis, the presenting time of electrical field was long (6 h for iontophoresis I, 4 h for iontophoresis II), so the contribution of migration and convection flow to enhanced flux of naproxen was large. Under the condition of a pulse electrical field, when the actual time of electrical field was very short (0.6 s for electroporation I, 0.74 s for electroporation II), the flux caused by migration and solvent convection flow in the electrical field could be omitted. The high level flux of naproxen after stopping of the electrical field implied that the permeation property of the skin was changed by electroporation.

The permeation of naproxen through the skin was dependent on the energy of the electrical field. The cumulative quantity of naproxen increased with the increasing energy of the electrical field. The energy of electroporation II was near that of iontophoresis I, so the cumulative quantity of naproxen showed no obvious difference between these two conditions.

In the condition of electroporation or iontophoresis, the skin resistance was changed. But the skin resistance was obviously different between electroporation and iontophoresis. For electroporation, the resistance between the electrodes can be calculated according to $R = \tau/C$ (τ is pulse time, C is the capacity of the capacitor). For iontophoresis, the resistance between the electrodes can be calculated according to $R = I/U$. The resistances were 24000 Ω, 28000 Ω, 2727 Ω and 337 Ω for iontophoresis I, iontophoresis II, electroporation I and electroporation II, respectively. The resistance decreased significantly under the condition of electroporation. It was in accordance with other results [11, 12]. This also implies that skin structure was changed during electroporation.

Although the enhancing effect achieved by electroporation was not always larger than that by iontophoresis, the enhancing effect achieved by electroporation lasted longer than that by iontophoresis after the electrical field was stopped. The actual time of electrical field during electroporation was obviously shorter than that during iontophoresis, so electroporation could possibly avoid skin stimulation.

In conclusion, electroporation and iontophoresis could significantly enhance the permeation of naproxen through the skin. The enhancing effect was dependent on the energy of the electrical field. The enhancing effect achieved by electroporation was not always better but lasted longer than that of iontophoresis.

4. Experimental

4.1. Materials

pH 7.4 phosphate buffer solution: dissolved 1.36 g of monobasic potassium phosphate in 79 ml of 0.1 mol/l sodium hydroxide solution, diluted with water to 200 ml. Donor solution: saturated solution of naproxen (purchased from Xinan No.2 Pharmaceutical Factory, Chongqing, P.R. China) in pH 7.4 phosphate buffer solution. High-performance liquid chromatograph (Waters, 515 HPLC pump, 486 turnable absorbance detector). The electroporative pulse device was a pulse generator which uses capacitor discharge to produce exponentially decaying pulses. The pulse frequency can be selected as 1, 2, 4, 8, 10, 20, 40 pulses per minute. The pulse number was between 1 and 999. It had four capacitors (2.2 μF, 22 μF, 68 μF, 220 μF) to produce pulses with different energy [13]. Skin preparation: Male SD rats with body weight of about 200 g (purchased from Experimemtal Animal Center of Zhejiang University, Hangzhou, P.R. China) were sacrificed. The full-thickness skin on the abdomen was excised after the hair was shaved off. The subcutaneous fat of the skin was removed carefully and then the skin was rinsed in pH 7.4 phosphate buffer solution for about 1 h.

4.2. Permeation of naproxen through the skin

The skin was sandwiched in the two compartments of side-by-side compartment cells (effective diffusion area = 0.5 cm^2, volume = 4.0 ml) with the stratum corneum facing the donor solution. The donor compartments were filled with donor solution and the receiver compartments were filled with pH 7.4 phosphate buffer solution. The solution in donor and receiver compartments were continually stirred by magnetic stirrers and maintained at 32 ± 0.5 °C. Ag/AgCl electrodes were immersed in the receiver compartments and Ag electrodes were immersed in the donor compartments. The electrodes were connected to a electroporative pulse device in case of electroporation and connected to a constant current source in case of iontophoresis. The passive diffusion was carried out. The solutions in the receiver compartments were taken out at intervals and replaced with fresh pH 7.4 phosphate buffer solutions. The concentration of naproxen in the samples taken from the receiver compartments were determined by a HPLC method. Liquid chromatographic analyses were performed on a C18 column (5 μm particle diameter, 15 cm × 4.6 mm i.d.). The mobile phase comprised of methanol: 0.1 mol/l sodium dihydrogen phosphate solution (30 : 70) (V/V). The flow rate was 0.8 ml/min and the detection wavelength was 274 nm. The cumulative quantity and flux of naproxen through the skin were calculated.

References

1 Weaver, J. C.: J. Cell. Biochem., **51**, 426 (1993)
2 Heller, R.; Gilbert, R.; Jaroszeski, M. J.: Adv. Drug Deliv. Rev. **35**, 119 (1999)

3 Jaroszeski, M. J.; Gilbert, R.; Nicolau, C.; Heller, R.: Adv. Drug Deliv. Rev. **35**, 131 (1999)
4 Wang, S.; Kara, M.; Krishnan, T. R.: J. Control. Release **50**, 61 (1998)
5 Vanbever, R.; Lecouturier, N.; Préat, V.: Pharm. Res. **11**, 1657 (1994)
6 Vanbever, R.; Leroy, M. A.; Préat, V.: J. Control. Release **54**, 243 (1998)
7 Jadoul, A.; Lecouturier, N.; Mesens, J.; Caers, W.; Préat, V.: J. Control. Release **54**, 265 (1998)
8 Zewert, T. E.; Pliquett, U. F.; Langer, R. Weaver, J. C.: Biochem. Biophy. Res. Commun. **212**, 286 (1995)
9 Hu, Q. H.; Liang, W. Q.; Bao, J. L.; Ping, Q. N.: Int. J. Pharm. **202**, 121 (2000)
10 Lombry, C.; Dujardin, N.; Préat, V.: Pharm. Res. **17**, 32 (2000)
11 Pliquett, U. F.; Langer, R. Weaver, J. C.: Biochim. Biophys. Acta. **1239**, 111 (1995)
12 Chernomordik, L. V.; Sukharev, S. I.; Abidor, I. G.; Chizmadzhev, Y. A.: Biochim. Biophys. Acta. **736**, 203 (1983)
13 Bao, J. L.; Liang, W. Q.; Hu, Q. H.; Gao, J. Q.: Chinese Journal of Scientific Instrument **21**, 66 (2000)