Passive Drug Diffusion via Standardized Skin Mini-erosion; Methodological Aspects and Clinical Findings with New Device

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Received April 9, 1996; accepted June 25, 1996

Purpose. To develop a clinical alternative to drug administration by injection or infusion.

Methods. A simple, mechanical device (Cellpatch) enables both the formation of a standardized small epidermal bleb and exposure of the circular base of the bleb to drug. The epidermis is split off by suctioning without bleeding or discomfort in a layer superficial to dermal capillaries and nociceptor nerves. Transdermal invasivity is thus avoided. Absorption of dextran test drug in aqueous solution vs molecular weight (3 kDa–70 kDa) and erosion area (3 kDa, diameter: 3–10 mm) were studied in healthy volunteers. The feasibility of using Morphine cellpatch (cell filled with 20 mg/ml morphine hydrochloride, aqueous solution, erosion diameter 6 mm) for post-operative pain relief was studied in two different patient groups; the Cellpatch was removed after 48 hours. Plasma morphine concentrations were determined at intervals.

Results. Dextrans of all sizes were efficiently absorbed transdermally, although absorption decreased with increasing molecular weight. The degree of absorption was directly related to the area of the mini-erosion. There were no sign of dose-dumping even with the largest erosions. The Cellpatch performed well in the demanding conditions of the postoperative unit, and was considered easy to use. Pharmacokinetically, the postoperative morphine delivery was related to that of a continuous infusion, with variability and dose in the same range as a continuous morphine infusion used clinically for providing basal pain relief. There were no bacterial growth in the morphine cells at 48 h. Reepithelialization of the erosion was rapid.

Conclusions. The feasibility of administering drugs in a wide size range by passive diffusion through a standardized skin mini-erosion was demonstrated; the rate of absorption decreased with increasing molecular weight. The small area of the erosion restricts and controls the concentration driven diffusion of drug into the circulation. As a consequence of the favorable findings, three placebo-controlled clinical studies using Morphine cellpatch for postoperative pain relief are currently underway.

KEY WORDS: transdermal administration; microcirculation; morphine; dextrans.

INTRODUCTION

In order to optimally produce its effects, a drug must be present at its site of action at the right time and at an appropriate concentration. In many situations, it would appear to be advantageous if a single drug delivery procedure could allow the clinician to pre-select an appropriate concentration level which could be maintained for a suitable number of days without further action from the patient or medical personnel. With the current state of the art, parenteral delivery remains the gold standard in this respect. It allows titration of dosage but requires surveillance and cannot be used safely in out-patients (Table I). Controlled release deliveries (1,2) have important clinical applications but titration of dosage may be difficult. An ingested or implanted delivery system cannot easily be retrieved if the need should arise. Transdermal drug patches may entail disturbing lag phases before an adequate dose level can be obtained and before all drug has left the body. Their major drawback is, however, the restriction to a small number of drugs that combine quite low molecular weight with lipophilicity. Moreover, transdermal drug absorption may vary unpredictably due to differences in thickness and composition of the skin barrier and variation in skin blood supply.

In a mechanically simple but biologically more sophisticated alternative evolved by us, drug is absorbed transdermally through a small skin erosion usually 5–6 mm in diameter after complete removal of the epidermal barrier by suction (3) (Fig. 1). Despite the small size of the erosion, potent drugs which are not normally absorbed transdermally (morphine and the antidiuretic peptide dDAVP, molecular weights 0.285 kDa and 1 kDa, respectively) were delivered at therapeutically effective plasma concentrations (3–5). A noteworthy finding was that administration could proceed at therapeutic rates through the same erosion for at least six days (dDAVP) (5). The pharmacokinetics were also favourable: our study of transdermal morphine in healthy volunteers showed that relatively constant plasma morphine concentrations in a narrow range were produced within about 10 min. The bioavailability at 24 h was as high as 75 per cent (mean) (4). The epidermal bleb was originally formed using a simple vacuum device, and its roof was then removed using a pincet (3). Subsequently, a device integrating a vacuum source, an epidermotome for removing the epidermal barrier and a drug cell has been developed (Cellpatch, see Fig. 3).

In this methodologically oriented report, we determined the absorption rates for dextran test drugs as function of molecular size and area of the skin mini-erosion in healthy volunteers. The feasibility of using Morphine cellpatch for postoperative pain relief was studied in two different patient groups.

MATERIALS AND METHODS

Subjects

The experiments were undertaken in consenting volunteers after approval by the Ethics Committee of Lund and Uppsala Universities and by the Swedish Medical Products Agency. The research followed the tenets of the declaration of Helsinki promulgated in 1964. The groups of volunteers are presented separately below.

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0724-8741/96/0900-1354$09.50/0

Table 1. Parental Techniques vs Passive Transdermal Diffusion

Parenteral administration (active propulsion)	Passive transdermal absorption
Active propulsion failures: 'no dose' 'dose dumping'	Passive diffusion: standardized 'hour glass'-restricted controlled release
Cannula may channel bacteria, infection	No dermal invasivity
Pain inflicted & provoked by movement	No discomfort
Cannula may produce significant tissue damage (bleeding, inflammation, thrombosis)	No such damage
Incomplete tissue restitution may result in protracted painful condition, disability	Transient pigmentation

Skin Mini-erosion

The skin mini-erosion is formed without discomfort to the patient and in a standardized way by vacuum suctioning and removal of the roof of the epidermal vesicle so created (Fig. 1). No needle is used for accessing the tissue. The epidermis is always split external to the superficial capillaries and nociceptor nerves through the loosely fibrous superficial layer of the epidermal basement membrane (6). A light tingle may be experienced transiently during the split. The dermis, including the superficial dermal capillaries, is left essentially intact, and the few layers of viable epidermal cells that are removed are regenerated (6,7). Local skin perfusion is markedly increased for days (8–11) (Fig. 2). The variability of dermal blood flow is considerably less in the erosion than in intact skin (3,8), where dermal blood flow varies within wide limits between different skin sites, over time and inter-individually. During the suctioning, the plasma filtrate that eventually fills the epidermal bleb leaks out through the outermost capillaries of the dermis. Low perfusion in the intact skin, which can be seen especially in young adults, may delay formation of the vesicle beyond the usual 30–90 min. By simply applying a (re-usable) finger warming pad during suctioning the procedure takes maximally about 80 min even in the postoperative state (below) when blood is shunted away from the skin as part of a stress response.

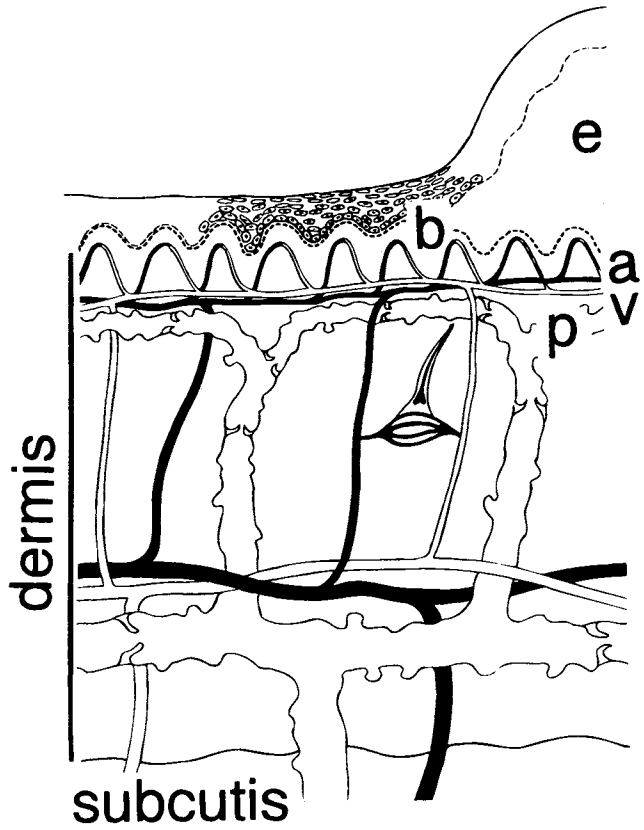

Fig. 1. Skin erosion (bleb) and dermal microcirculation. e = erosion, b = lamina densa of basement membrane, a = arteriole, v = venule, p = primary lymphatics. The skin split always occurs through the stratum lucidum of the epidermal basement membrane, leaving the collagenous and elastic network of the lamina densa adherent to the dermis. Drug is absorbed through blood capillaries and venules, and also through the lymphatic system.

Fluorescent Dextrans

Fluorescent dextrans (FD) (Pharmacia & Upjohn, Sweden) with narrow size ranges, which are commonly used for defining the porosity of the microcirculation (12) and the competence of the lymphatic system (13), were used as test drugs. The FD's typically exhibit molecular homogeneity, hydrophilic properties and high level of stability. The blood capillary and venule wall allow the passage of test dextrans of all sizes. With increasing diameter the absorption increasingly takes place via the lym-

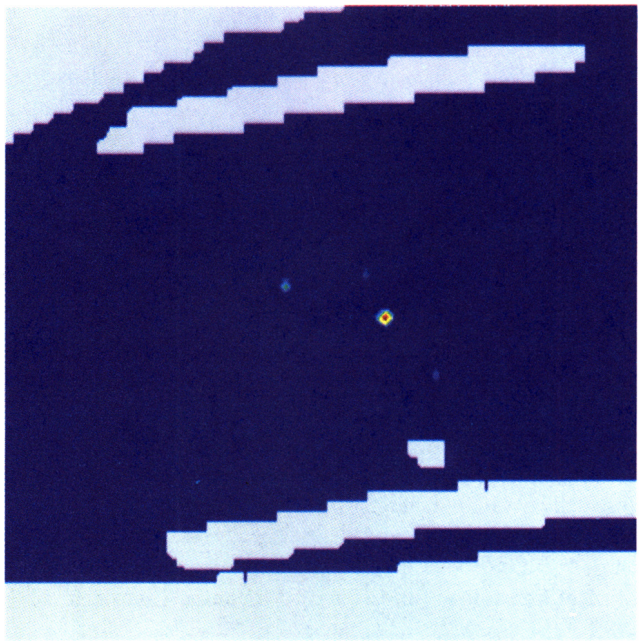

Fig. 2. Typical recording of skin blood perfusion measured with a laser Doppler scanner through the drug cell during postoperative morphine delivery (see Fig 3). The circular red spot with white halo indicates markedly increased blood perfusion (i.e. large number and velocity of red blood cells) in the skin mini-erosion compared with the the adjacent skin.

phatic system (13). Cylindrical cells with removable lid were adhesively adhered by a flange at the cylinder base to the skin circumference directly adjacent to the erosion. These cells contained FD in aqueous solution. Even if acute tests showed that FD remained in solution with no sign of adhesion we cannot exclude local adhesion as a source of systemic error in the experiments. The cells were sampled at intervals. The FD concentration in the cell was measured spectrofluorometrically ('Cytofluor 2300', Millipore, USA) at an excitation wavelength of 480 nm and an emission wavelength of 520 nm. FD absorption from the cell via a mini-erosion (6 mm diameter (D)) into the systemic circulation was verified preliminarily for 20 kDa FD 20 by measuring urinary excretion (n = 3, one woman). 20 kDa FD is the largest of the test molecules that are filtered from the blood into the urine. Each volunteer received a single cell containing 0.5 ml solution (5 mmol). At 24 h, 54.3 per cent (mean) of the total FD dose had been absorbed and the recovery in urine was 12.6 per cent (of the absorbed dose). Some FD degradation (20 kDa) was demonstrated by gel filtrating urine but it was negligible in the context of the experiments. It was not possible to determine the cell volumes of drug solution accurately and the FD test drug absorption was indirectly determined based on concentration measurements in the cell. Intact skin was found to absorb significant volumes of water. This led us to exclude solitary deliveries in which intact skin was exposed to FD test solution due to (1) poor positioning of drug cell (3 mm D erosion) (2) leakage under part of the flange.

Effect of Molecular Size

The porosity of the capillary membrane and its effect on absorption was assessed by varying FD molecular size. Four FD cells (3, 10, 20 and 70 kDa) (100 μmol, dissolved in 0.5 ml isotonic saline) were applied on 6 mm (D) erosions in 7 healthy volunteers (3 women) weighing 53–102 kg (78 kg) (mean); the delivery lasted 24 h. A fifth cell was adhered on intact skin and served as control. The FD concentration in the cells were determined at 1 h, 2 h, 4 h, 8 h, 16 h and 24 h.

Effect of Erosion Area

Using the experimental set-up described above in 6 volunteers (3 women) weighing 53–102 kg (75 kg), we determined the absorption from skin erosions with different areas (7 mm^2–79 mm^2)(D=3−10 mm). The drug cell contained 3 kDa FD.

Morphine Cellpatch

The Cellpatch device (Epiport Pain Relief AB, Sweden) is shown in Fig. 3. The simple steps required for initiating the drug delivery are indicated. The drug cell contained 53 mmol/l, 5.0 ml morphine hydrochloride in aqueous solution (20 mg/ml).

Clinical Feasibility Studies with Morphine Cellpatch

Study A. Seven women aged 40–61 years (mean: 49 years) and weighing 56–95 kg (71 kg) were studied following elective transabdominal removal of the womb. The morphine delivery was initiated directly after surgery. *Study B.* Eight patients (one woman) aged 42–68 years (62 years) and weighing 54 to 110 kg (80.5 kg) were studied after elective trans-thoracic coronary

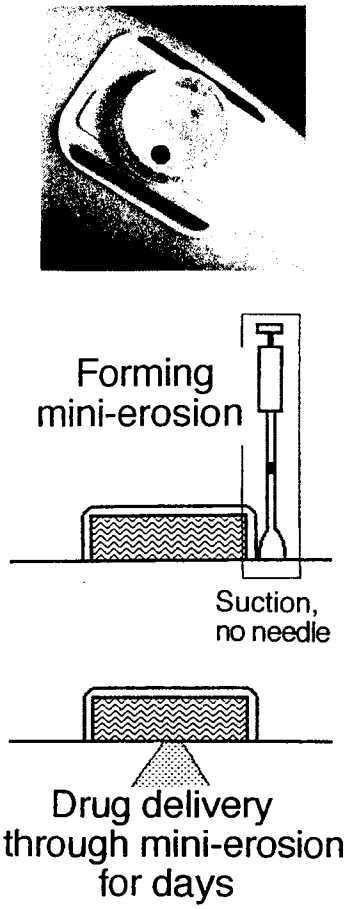

Fig. 3. The 'Cellpatch®' (Epiport Pain Relief AB, Malmö, Sweden) disposable device prefilled with aqueous morphine solution and adhesively applied on the forearm (see photo). Integral with the device, the small epidermal bleb is formed by suctioning and abladed with a built-in epidermotome. The suction cup, tube and volume expander is removed before drug delivery can be initiated.

artery surgery. The 15 patients were otherwise healthy. The morphine delivery was initiated in the morning following operation, once the tracheal tube had been removed. In both studies the morphine delivery lasted for 48 hours. Pethidine hydrochloride (Pharmacia & Upjohn) was used additionally as 'rescue' medication; it was given through an i.v. cannula. The focus of interest in these open studies were threefold: (1) determining the time-courses for morphine in plasma in two distinct patient groups, as a preliminary to a complete pharmacokinetic assessment in a larger study (2) monitoring respiratory function (respiratory depression is the major hazard in opioid overdosing and limits the postoperative use of opioids), and (3) assessing compliance and local factors relating to Cellpatch use.

Respiratory function was monitored during morphine delivery by pulse-oxymetry, a method allowing accurate non-invasive measurement of arterial oxygen tension. Morphine plasma concentrations (studies A and B) and drug cell morphine concentrations (study B) were analysed using an HPLC method with electrochemical and ultraviolet detection (14). The analyses were not interfered with by plasma pethidine. The morphine solution remaining in the drug cell after therapy was taken for bacteriological testing (Study B). Samples were cultured (1)

by aerobic/aerobic incubation on blood agar plates, and on a haematin agar plate; all at 37°C for at least 48 h and (2) with tryptic soy broth likewise incubated for 24 h, subcultured on a haematin agar plate incubated aerobically at 37°C for 48 h.

RESULTS

The complete FD cell concentration time-courses showed an early, transitory increase in drug cell concentrations which was most obvious for 20 kDa and 70 kDa FD's. The presence of a transient, osmotically determined shift of water from the drug cell into the tissues was indicated. Assumedly capillary wall pore impedance in the mini-erosion is more increased for the dextrans than for water, in particular it is increased for the larger dextrans at 20 kDa and 70 kDa. At the intact skin control site, 3 kDa FD was not absorbed. On the contrary, the concentration was increased by 20.8 per cent at 24 h. The finding is explained by withholding of water through hydration of the epidermis. The 'stealth' of water from the cell from osmosis and hydration of inadvertently exposed epidermis implies that the values shown underestimates the actual FD absorption. The comparisons are meaningful at 24 h when water shifts and eventual effects of adhesion have tentatively stabilized, see Fig. 4A. In spite of the steath, the dextrans were readily absorbed, though decreasingly with increasing molecular size. The absorption was maximal with 37.9 per cent of the initial dose for 3 kDa FD and it was minimal at 20.1 per cent for 70 kDa FD. The degree of transdermal absorption was directly related to the area of the erosion (Fig. 4B). The absorption of FD 3 kDa was 20.5 per cent through a 3 mm erosion and it was 60.7 per cent through a 10 mm erosion.

In study A (Fig. 5) morphine delivery was associated with a mean Cmax value of 17.3 ± 3.7 nmol/l (CV: 21 per cent), the corresponding value in study B being 20.9 ± 7.7 nmol/l (CV: 33 per cent). After 48 h, 32 ± 6 mg (CV: 19 per cent) of the initial 100 mg morphine dose in the drug cell was found to be absorbed (Study B). The increased plasma concentration variability in study B reflects variable weight (see legend to Fig. 5). No respiratory depression was observed in any of the experiments. The Cellpatch performed well in the demanding conditions of the postoperative unit, and was considered easy to use. The patches adhered completely to the skin for the duration of therapy. While the Cellpatch caused no discomfort, the i.v. cannula produced intermittent light pain and some local stiffness which could be exacerbated when the patients moved the arm or when the projecting part of the cannula was inadvertently moved. According to their ratings (4–5 on a 5-point scale), all patients preferred to receive drugs through the Cellpatch rather than through cannula. When the patch was removed, there was no sign of inflammation in the erosion or the adjacent skin, and the skin under the patch was normal. A week after removal of the Cellpatch the epidermis had regenerated, and the remaining erythema was considered trivial by all patients. At follow-up at 3–6 months (Study A), close examination revealed a very slight and fading pigmentation which had become almost invisible in the first three patients. The bacteriology samples were negative in all cultures.

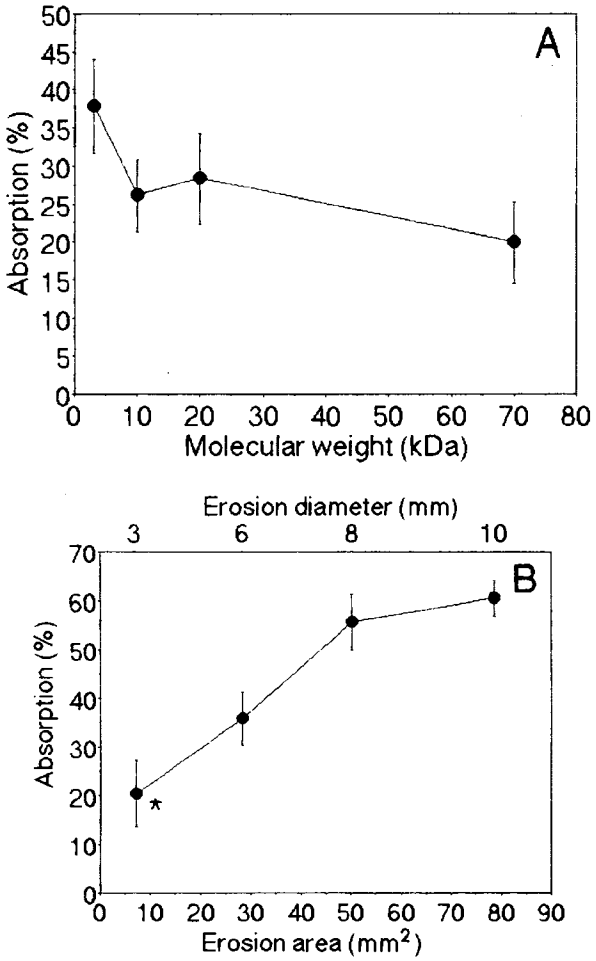

Fig. 4. Per cent absorption of fluorescent dextrans (FD); values shown are means ± SE following 24 h delivery. (A) Effect of FD molecular weight on absorption through 6 mm skin erosion. (B) Absorption (of 3 kDa FD) as a function of the area of skin erosion. *Swelling epidermal edge into tiny cell aperture at 24 h resulted in exclusion, 16 h values are given.

DISCUSSION

We present data indicating that molecules up to 70 kDa are absorbed efficiently and may potentially be used with this new delivery route. The technique may thus become an important key to acieving transdermal, controlled delivery of

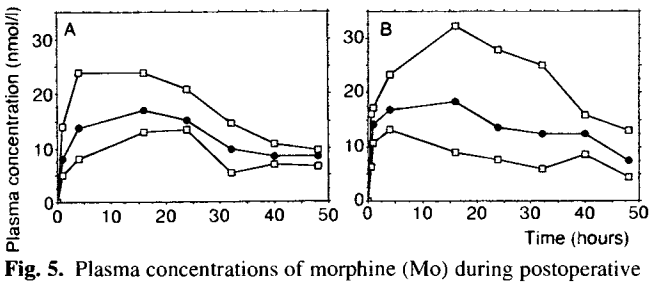

Fig. 5. Plasma concentrations of morphine (Mo) during postoperative passive transdermal administration. Mean values (●) and absolute ranges (□) are given. *Study A.* From elective transabdominal removal of the womb (n:7). *Study B.* Following elective trans-thoracic coronary artery surgery (n:8).

peptide/protein drugs. Passive transdermal absorption is limited by the area of the erosion, as might be expected. Controlled release mode without 'dose-dumping' is observed even with an erosion as large as 10 mm in diameter, and the absorption is significant even at 3 mm (the diameter of a large cannula is 2 mm). Two feasibility studies where morphine was used as the test substance in postoperative patients demonstrate pharmacokinetics related to parenteral infusion without the disadvantages of invasiveness and delivery by active propulsion (Table I).

The feasibility of administering hydrophilic drugs with molecular size of at least 70 kDa by concentration driven, passive diffusion through a 6-mm skin erosion is clearly indicated by the FD results. The blood capillary and venule wall which constitute the main remaining relative barrier to absorption allow the passage of all test dextrans used, as might be expected (12–13). With a molecular size of 70 kDa absorption mainly takes place via the lymphatic plexus. For comparison, Tc-labelled albumin (molecular weight and hydrodynamic radius corresponds to 70 kDa FD), injected into the superficial dermis, is absorbed via the superficial lymphatic plexus (13). The pores of the vascular membrane may be further dilated in response to inflammatory mediators (15). A study of outward permeation through an erosion has shown the ratio between the concentration of proteins in transudate and in plasma to be dependent on the molecular weight, and that the permeation follows the law of diffusion (16). As in parenteral extravascular deposition of drug, the rate of absorption is limited by the area of the absorbing capillary membrane (area of erosion), and the solubility of the substance in the interstitial space.

The absence of any sign of rapid drug dispersal through 10 mm erosions underscores even further the reliability of the delivery through still smaller ones. The small, standardized area of the erosion restricts and controls the concentration driven diffusion of drug into the circulation. An hourglass may be a useful analogy. As the area increases, the impedance due to the pores in the capillary membrane becomes increasingly important as an absorption limiting factor. How does this permeation regulation influence absorption of smaller molecules? Will acute dose-dumping be a problem with such substances? Our earlier findings in which 10 mg morphine (at 0.285 kDa) was given through 6 mm erosions may provide a clue. These data indicated that relatively stable plasma morphine concentrations were reached after a lag phase of about 10 minutes. A decrease was evident by 13 h and the bioavailability of 24 h turned out to be in the range 65–85 per cent (4). With a drug of larger molecular size (dDAVP, 1 kDa) a similar plateau was reached in about 2 hours (3,5). This compares favourably with the fentanyl pain relief patch where absorption is negligible at 2 h and remains very low at 4 h (17). Simply altering the concentration of drug in the cell may be a more preferable way of controlling absolute absorption rates than changing the size of the erosion—for instance, there was a 50-fold difference in concentration of 20 kDa FD in this experiment and in the 20 kDa FD control experiment (see methodology section), and relative absorption at 24 h was similar.

A 'conventional' morphine patch on intact skin would need to cover an area of 62.500 cm^2 to provide the equivalent of an i.m. injection of 10 mg morphine (18). Bioavailability achieved by other non-invasive routes ranges from 20 to 40 per cent, and inter-individual variation is wide (18) due to varying individual capacity to metabolise and eliminate the drug. The morphine plasma concentration data from the two patient groups (Fig. 5) are consistent and correspond to those obtained in the previous pharmacokinetic study in healthy volunteers (Cmax of 17.7 ± 4.2 nmol/l; CV: 24 per cent). The morphine dose by passive transdermal diffusion correspond roughly to those of a continuous morphine parenteral infusion used for basal pain relief, and also the CV values are in the range observed for clinical infusions (4). By way of comparison, the CV of Cmax for a fentanyl patch used postoperatively may be 61 per cent (19). Differences in epidermal thickness, degree of hydration and skin perfusion are major sources of the larger variability. Postoperative use of the fentanyl patch is thus contraindicated.

The light hue, consisting of superficially lodged blood pigment, disappears with time. Cosmetically, the skin is normalized (3,5,6,20). The benign course corresponds to that of a natural or traumatically induced skin vesicle. Importantly, there is no foreign body penetration into the tissues which may channel bacteria. Such penetration is the plague of the parenteral techniques, where sepsis is the major risk, especially in more protracted delivery. The absence of bacterial growth in the morphine cells corroborates the accumulated experience gleaned from current clinical studies and from pharmacokinetic studies in volunteers on opiates, including controls containing isotonic saline solution. In summary, these findings demonstrate the feasibility of using Morphine cellpatch for postoperative pain relief, an indication which may at this stage tentatively be limited to providing basal pain relief during 48 h. Three placebo-controlled clinical studies on this indication are currently underway.

The clinical applicability of the skin mini-erosion also derives support from hundreds of studies where suctioning has been used in humans as a means of splitting off the epidermis, mainly for the purposes of dermatological research (6) and for assessment of drug concentrations in the peripheral compartment (21). These studies have been undertaken in patients with different diseases and also in people with different ethnic background. Multiple blisters were mostly used, covering areas of skin much larger than that of the small vesicle that we propose using. No complications were reported.

Traditional transdermal research has focused on weakening the epidermal barrier (2) by toxic chemicals, strong electrical current or ultrasound. Given our simple and reliable means of eliminating the barrier and creating an erosion, our challenge is instead to produce controllable absorption of a particular drug through a limited area of the dermal microvascular wall in the simplest possible way. In this report we have focused on the possibilities inherent in passive transdermal delivery. In the future, mini-erosion absorption may be manipulated by active means, for instance by propulsion of molecules along a pressure gradient, by local movement, by minimal electrical current compared with that presently used in iontophoresis (1) (the considerable epidermal barrier impedance is eliminated), and by pharmacological manipulation of the drug suspension or the tissue.

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