International Journal of Pharmaceutics, 66 (1990) 243-252
Elsevier

IJP 02281

Percutaneous absorption of ibuprofen and naproxen: Effect of amide enhancers on transport through rat skin

W.J. Irwin, F.D. Sanderson* and A. Li Wan Po+

Drug Development Research Group, Pharmaceutical Sciences Institute, Aston University, Aston Triangle, Birmingham B4 7ET (U.K.)

(Received 6 June 1990)
(Modified version received 2 August 1990)
(Accepted 10 August 1990)

Key words: Azone; Dimethylamide; Ibuprofen; Naproxen; Penetration enhancer; Percutaneous absorption; Mode of action

Summary

n-Alkanoic *N,N*-dimethylamides were found to act as penetration enhancers for the transport of ibuprofen and naproxen from suspensions in 50% aqueous propylene glycol vehicles across rat skin. Stripped skin and separated dermis were used to establish the maximum potential for enhancement and comparisons with established enhancers such as azone and *N*-methylpyrollidone were undertaken. Greatest enhancement was observed with naproxen but both drugs demonstrated a bell-shaped dependence on the alkyl chain length of the enhancer. Maximum effect was observed with *N,N*-dimethyloctanamide and *N,N*-dimethyldecanamide. Measurement of the skin-vehicle partition coefficients indicated that the partition of the drug into the skin was also maximal when these enhancers were incorporated into the vehicle. Permeation studies monitoring the flux of enhancer indicated that these compounds also penetrated the skin most effectively. In contrast, the enhancers had little effect on delivery from liquid paraffin vehicles.

Introduction

Much current work is progressing on the use and mode of action of enhancers of percutaneous absorption (Woodford and Barry, 1986; Barry, 1987; Green et. al., 1988; Walters, 1989). Many of the compounds used are of diverse structures and properties and few studies have appeared using homologous families to reveal structure-activity relationships. To enable this aspect of penetration enhancement to be studied a series of *n*-alkanoic *N,N*-dimethylamides [$(CH_3)_2N$-CO-$(CH_2)_n$-CH_3] have been used to promote the percutaneous absorption of ibuprofen and naproxen. Compounds of this type which have previously been used as absorption enhancers include small amides such as *N,N*-dimethylformamide and *N,N*-dimethylacetamide which have shown penetration enhancement at high concentrations. Longer chain amides appear to be active at much smaller concentrations and include Azone. The potential of this series is further indicated as higher derivatives of dimethylsulphoxide, such as decylmethylsulphoxide, have been used effectively as enhancers at low concentrations (Sekura and Scala, 1972). Moreover, evidence indicates that these amides enhance the percutaneous absorption of an orga-

Correspondence: W.J. Irwin, Drug Development Research Group, Pharmaceutical Sciences Institute, Aston University, Aston Triangle, Birmingham B4 7ET, U.K.
*Present address: SmithKline-Beecham, Research Division, Clarendon Road, Worthing, West Sussex BN14 8QH, U.K.
+Present address: Department of Pharmacy, Queen's University of Belfast, 97, Lisburn Road, Belfast BT9 7BL, U.K.

nophosphorus anticholinesterase (Wiles and Narcisse, 1971) during a study of the 50% death time in mice and rabbits using topical formulations in amides. They found that compounds from *N,N*-dimethylhexanamide to *N,N*-dimethyldodecanamide were most effective in mice and the *N,N*-dimethylhexadecanamide in rabbits for reducing the time of death and therefore enhancing the penetration. Additionally, *N,N*-dimethylamides have been patented as insect repellents (Jacobi and Lust, 1961), they have proved useful as insecticides (Hwang and Mulla, 1980) and have shown antimicrobial activity against *Staphylococcus aureus* (Bistline et al., 1980). Technical sources of these compounds are available, generally as mixtures, (Hallcomids) and they have suggested uses as solvents, solubilisers, surfactants, chemical reaction media and grinding agents. In this study technical and synthesised *N,N*-dimethylamides, particularly the higher derivatives from *N,N*-dimethylhexanamide to *N,N*-dimethyloctadecanamide were studied to expose any structure-activity relationships in the enhancement profiles.

Experimental

Materials

N,N-Dimethylamides (C_3–C_{10}) were synthesised from the appropriate acid chloride [propionic (C_3), butyric (C_4), valeric (C_5) caproic (C_6), caprylic (C_8), capric (C_{10})] and dimethylamine. Higher derivatives, *N,N*-dimethyldodecanamide (C_{12}), *N,N*-dimethyltetradecanamide (C_{14}), *N,N*-dimethylhexadecanamide (C_{16}) and *N,N*-dimethyloctadecanamide (C_{18}), were prepared from the alkanoic acid [lauric (C_{12}), myristic (C_{14}), palmitic (C_{16}), stearic (C_{18})] and thionyl chloride followed by treatment with dimethylamine. Hallcomids were supplied by C P Hall, Chicago. M-18-OL is a mixture consisting of 5% C_{14}, 5% C_{16}, 5% C_{18}, 80% *N,N*-dimethyloleamide, and 5% *N,N*-dimethyllinoleamide. M-8,10 is a mixture containing 5% C_6, 50% C_8, 40% C_{10}, and 5% C_{12} amides.

Rat skin was prepared as described earlier (Irwin et al., 1990).

Methods

Apparatus. HPLC analyses were undertaken using a system constructed from an Altex 100A dual-piston reciprocating solvent-metering pump and a reversed-phase stainless-steel Shandon-type column (10 cm × 4.6 mm i.d.) packed with Hypersil-ODS (5 μm). Samples were introduced by means of a Rheodyne 7125 injection valve, fitted with a 20 μl loop, and detection was accomplished with a Pye LC3 variable wave-length UV detector, fitted with an 8 μl flow cell, and operated at a wavelength of 225 nm (Ibuprofen) or 245 nm (Naproxen) with a sensitivity of 0.01–0.02 AUFS. The mobile phase for ibuprofen consisted of aqueous acetonitrile (55%), adjusted to pH 2.0 with phosphoric acid (0.45%) while that for Naproxen was 40% acetonitrile with 0.32% phosphoric acid, also at pH 2.0. Each was delivered at 1 ml min^{-1}. Maximum concentrations of 45 μmol l^{-1} (9.3 μg l^{-1}, ibuprofen) and 25 μmol l^{-1} (5.8 μg l^{-1}, naproxen) were used and calcium fenoprofen 1.1 μmol l^{-1} (0.6 μg ml^{-1}) in 50% aqueous methanol was used as the internal standard for ibuprofen and ethyl paraben 3 μmol l^{-1} (0.5 μg ml^{-1}) in aqueous acetonitrile for naproxen. In general, 1 ml of the test solution was treated with 4 ml of the internal standard solution and 10–20 μl were injected into the HPLC. Conditions for the analysis of the *N,N*-dimethylamides are held in Table 1.

Diffusion experiments were undertaken in a glass diffusion cell as described earlier (Irwin et al., 1990).

Formulation of N,N-dimethylamides. Saturated solutions of the higher derivatives (*N,N*-dimethylhexadecanamide (C_{16}), *N,N*-dimethyltetradecanamide (C_{14}), *N,N*-dimethyldodecanamide (C_{12}), Azone (Nelson Research) and Hallcomid M-18-OL) were prepared by adding excess to 50 ml aliquots of the 50% propylene glycol and shaking overnight at 25°C in a water bath. The solutions were centrifuged or filtered depending on whether the amide was liquid or solid. The lower homologues (1% *N,N*-dimethylhexanamide (C_6), 1% *N,N*-dimethyloctanamide (C_8), *N,N*-dimethyldecanamide (C_{10}), 10% M-8,10, 10% *N*-methylpyrrolidine and 10% *N*-methylpyrrolidone) were dissolved separately in the 50% propylene glycol vehicle. Suspensions of ibuprofen were pre-

TABLE 1

HPLC conditions for the analysis of N,N-dimethylamides

Enhancer	Maximum concentration mmol l^{-1} (mg ml^{-1})		Mobile phase acetonitrile (%)	AUFS
N,N-Dimethylhexanamide	18	(2.6)	40	0.64
N,N-Dimethyloctanamide	2	(3.4)	50	0.08
N,N-Dimethyldecanamide	1.5	(0.3)	70	0.16
N,N-Dimethyldodecanamide	0.45	(0.102)	90	0.02
N,N-Dimethyltetradecanamide	0.45	(0.115)	90	0.02
N,N-Dimethylhexadecanamide	8	(2.2)	95	0.16
Azone	0.25	(0.0703)	90	0.02

Detection for all amides was at 225 nm. Calcium fenoprofen 4.6 μmol l^{-1} (2.6 μg ml^{-1}) in 50% methanol was used as the internal standard for *N,N*-dimethyloctanamide; C_{12} (506 μmol l^{-1}, 115 μg ml^{-1}) for *N,N*-dimethyldecanamide and C_{14} (196 μmol l^{-1}, 50 μg ml^{-1}) for *N,N*-dimethylhexadecanamide. Typically, sample (1 ml) was treated with the internal standard solution (4 ml) and 100 μl were injected.

pared in these vehicles. Formulations in liquid paraffin consisted of a control with no additives and four test solutions containing 1% C_{18}, 1% C_{14}, 1% C_8, and 1% *N,N*-dimethylbutyramide (C_4). To the solutions were added excess naproxen to provide suspensions. Additionally, a suspension of naproxen in triacetin was also used. Drug permeation was monitored by HPLC using 50% propylene glycol as the receptor phase. The solubility of naproxen in the vehicles was determined by extracting 1 ml of the filtered liquid paraffin or triacetin suspension with 5 ml of 0.01 M sodium hydroxide. 1 ml of the sodium hydroxide was neutralised with 1 ml of 0.01 M hydrochloric acid and diluted. Concentrations were determined by HPLC.

Determination of partition coefficients. The 50% propylene glycol (pH 5.5) vehicle was presaturated overnight with the lipid phase in a water bath at 25°C and solutions of ibuprofen (70 mg in 250 ml) or naproxen (45 mg in 250 ml) in the vehicle were prepared. Aliquots (40 ml) of this solution were shaken with 5 ml aliquots of the presaturated lipid phase and the drug content was determined in both phases by HPLC. For skin partitions stock solutions of ibuprofen (120 μg ml^{-1}) or naproxen (140 μg ml^{-1}) were prepared in the vehicles and these were diluted 1 in 20 with the appropriate drug-free vehicle. Aliquots (10 ml) were added to vials containing 0.35 g of whole rat skin and to two empty vials to act as controls. The solutions were allowed to equilibrate for four weeks and the solutions analysed by HPLC. The density of the vehicle was determined using a 25 ml density bottle, so that weight ratios instead of volume ratios could be used in partition coefficient determination. The solutions were analysed by HPLC. For vehicles containing C_8 and M-8,10, samples (5 ml) of these vehicles after equilibrium were added to 0.01 M HCl (1 ml) and the solution was extracted with chloroform (2 × 3 ml). The combined chloroform solutions were extracted with 0.1 M NaOH (3 ml) and an aliquot (1 ml) was added to 0.1 M HCl (1 ml). The standard solutions were treated similarly and were analysed by HPLC.

Permeation studies. The donor solution was a suspension of ibuprofen or naproxen (5 ml) in 50% propylene glycol (pH 5.5). Samples (1 ml) were taken from the receiver compartment at 1 h intervals for a period of 12 h and these were analysed by HPLC.

Penetration of amides through rat skin. Vehicles were prepared as above containing C_6, C_8, C_{10}, C_{12}, C_{14} and Azone. The penetration profiles were determined for C_8 and C_{10} by sampling at 1 h intervals. In addition, the amount penetrated after 24 h was determined for all amide systems using HPLC.

Results and Discussion

The percutaneous absorption of ibuprofen and naproxen through rat skin was studied from suspensions. Where possible a 1% solution of the di-

methylamide was used, however, if this was not possible the standard vehicle, 50% propylene glycol in aqueous buffer pH 5.5, was presaturated with the amide and then a suspension of the drug was prepared. The solubility of the higher amides in this vehicle are listed in Table 2. Azone was used as a standard in this study together with *N*-methylpyrrolidone as examples of two well characterised enhancers.

Typical penetration profiles for ibuprofen from various vehicles are shown in Fig. 1. To preserve clarity, no error bars are plotted but these are numerically listed in Table 3 which also includes additional systems.

Several initial experiments were carried out on stripped and separated skin to see the effect of the removal of stratum corneum on the penetration characteristics of ibuprofen. These included successive stripping of the rat skin with adhesive tape (× 30) and also chemical separation with sodium bromide (Walker and Scott, 1984). Removal of the epidermis was achieved by gently rubbing the

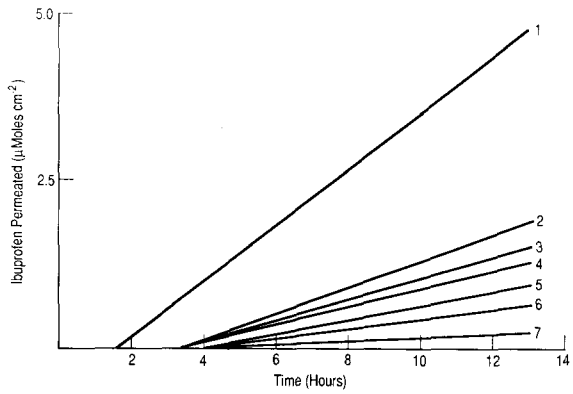

Fig. 1. Comparison of permeation profiles of ibuprofen from various suspension formulations containing amide enhancers in 50% propylene glycol (pH 5.5). Systems are: (1), 50% propylene glycol through separated dermis; (2) 10% *N*-methylpyrollidone; (3) 1% *N,N*-dimethyloctanamide; (4) 1% *N,N*-dimethyldecanamide; (5) 0.145% *N,N*-dimethyldodecanamide; (6) 50% propylene glycol; (7) aqueous buffer, pH 5.5, (2–7) whole skin.

TABLE 2

Solubility of higher N,N-dimethylamides and Azone in 50% aqueous propylene glycol, pH 5.5. (Numbers in parentheses are standard deviations)

Amide	Solubility (mg ml^{-1})
N,N-Dimethylhexadecanamide	0.01985 (0.00195)
N,N-Dimethyltetradecanamide	0.3425 (0.00820)
N,N-Dimethyldodecanamide	1.4520 (0.03535)
Azone	0.2475 (0.01639)

TABLE 3

Flux, lag time and permeability coefficient for the transport of ibuprofen through rat skin from suspensions in 50% propylene glycol (pH 5.5) containing penetration enhancers

Vehicle	Flux(×10^3) (μmol cm^{-2} h^{-1})	Lag time (h)	K_p(×10^3)(cm h^{-1})	*n*
Aqueous buffer (WS)	25.2 (5.9)	3.24 (0.12)	11.27 (2.64)	5
50% PG (SS)	424.1 (38.1)	2.30 (0.23)	28.36 (2.55)	6
50% PG (SD)	418.3 (25.7)	1.55 (0.25)	27.97 (1.72)	6
50% PG (SDF)	615.9 (28.8)	1.30 (0.16)	41.12 (1.93)	6
50% PG (WS)[a]	70.1 (4.4)	3.13 (0.15)	4.69 (0.29)	15
1% C_8, 50% PG (WS)	159.2 (7.3)	3.36 (0.04)	9.03 (0.43)	6
1% C_{10}, 50% PG (WS)	135.6 (8.6)	3.26 (0.10)	8.32 (0.53)	6
0.145% C_{12}, 50% PG (WS)	107.5 (18.0)	3.79 (0.16)	7.32 (1.23)	5
0.034% C_{14}, 50% PG (WS)	114.6 (5.8)	4.23 (0.11)	7.87 (0.40)	5
M-18-OL, 50% PG (WS)	106.0 (21.5)	4.29 (0.26)	7.80 (1.58)	5
10% NMPi, 50% PG (WS)	59.9 (4.2)	4.18 (0.09)	2.16 (0.15)	6
10% NMPo, 50% PG (WS)	203.0 (23.0)	3.40 (0.35)	3.57 (0.40)	6
0.025% Azone, 50% PG (WS)	65.9 (8.5)	4.33 (0.18)	4.40 (0.57)	6

[a]Control; WS, whole skin; SS, skin stripped with tape; SD, separated dermis; SDF, separated dermis stored frozen. C_8, dimethyloctanamide; C_{10}, dimethyldecanamide; C_{12}, dimethyldodecanamide; C_{14}, dimethyltetradecanamide; M-18-OL, Hallcomid; NMPi, *N*-methylpyrollidine; NMPo, *N*-methylpyrrolidone. Figures in parentheses are standard errors.

TABLE 4

Ratio of steady-state flux and permeability coefficient for ibuprofen through rat skin from 50% propylene glycol (pH 5.5) using various amide enhancers

Vehicle	Flux ratio	Permeability coefficient ratio
Aqueous buffer (WS)	0.4	2.4
50% PG (SS)	6.0	6.0
50% PG (SD)	6.0	6.0
50% PG (SDF)	8.8	8.8
50% PG (WS)[a]	1.0	1.0
1% C_8, 50% PG (WS)	2.3	2.0
1% C_{10}, 50% PG (WS)	1.9	1.8
0.145% C_{12}, 50% PG (WS)	1.5	1.6
0.034% C_{14}, 50% PG (WS)	1.6	1.7
M-18-OL, 50% PG (WS)	1.5	1.7
10% NMPi, 50% PG (WS)	0.9	0.5
10% NMPo, 50% PG (WS)	2.9	0.8
0.025% Azone, 50% PG (WS)	0.9	0.9

[a]Control; WS, whole skin; SS, skin stripped with tape; SD, separated dermis; SDF, separated dermis stored frozen. C_8, dimethyloctanamide; C_{10}, dimethyldecanamide; C_{12}, dimethyldodecanamide; C_{14}, dimethyltetradecanamide; M-18-OL, Hallcomid; NMPi, *N*-methylpyrollidine; NMPo, *N*-methylpyrrolidone.

skin surface. The steady state flux for the stripped and chemically separated skin are almost identical at 424.1×10^{-3} and 418.3×10^{-3} μmol cm^{-2} h^{-1}, respectively, and indicates the maximum steady-state flux obtainable when the stratum corneum transport barrier is not present. This is a theoretical limiting value for any enhancement assuming that the enhancers exert their mode of action by an increased penetration through the stratum corneum only. When the chemically separated skin was stored frozen prior to use the steady-state flux rose to 615.0×10^{-3} μmol cm^{-2} h^{-1}. This is probably due to the high water content of the dermis which, on freezing, causes cellular damage leading to a more rapid penetration rate.

The flux and permeability coefficients of ibuprofen are normalised, relative to transport through whole skin from the 50% aqueous propylene glycol (pH 5.5) vehicle without enhancers, in Table 4.

The greatest increase in flux was achieved with *N*-methylpyrrolidone which was some 2.9 times higher than that of the control vehicle. However, due to the high level of this enhancer in the formulation and to the much increased solubility of ibuprofen in the vehicle (11.75 compared to 3.09 mg ml^{-1}) this flux is largely solubility-driven and the permeability constant is actually less than that of the control. In contrast, the octanamide and decanamide derivatives produced an approximate 2-fold increase in steady-state flux and permeability constant, whereas Azone caused no change. This

TABLE 5

Solubility and skin partition coefficients of ibuprofen in 50% propylene glycol (pH 5.5) containing various amide penetration enhancers

Vehicle	Solubility (mg ml^{-1})	Partition coefficient (P_m)	
		Dry	Wet
Aqueous buffer (WS)	0.461 (0.023)	6.08 (0.73)	6.58 (0.59)
50% PG (SS)	3.085 (0.196)	5.89 (0.62)	4.94 (0.55)
50% PG (WS)[a]	3.085 (0.196)	4.33 (0.21)	4.46 (0.18)
1% C_6, 50% PG (WS)		4.70 (0.64)	5.35 (0.79)
1% C_8, 50% PG (WS)	3.547 (0.133)		
1% C_{10}, 50% PG (WS)	3.362 (0.085)	42.79 (2.06)	43.42 (1.89)
0.145% C_{12}, 50% PG (WS)	3.029 (0.052)	9.47 (0.79)	9.04 (0.68)
0.034% C_{14}, 50% PG (WS)	3.033 (0.047)	6.71 (0.33)	6.71 (0.33)
2×10^{-3}% C_{16}, 50% PG (WS)		5.05 (0.29)	5.35 (0.37)
M-18-OL, 50% PG (WS)	2.803 (0.107)	6.96 (0.92)	6.59 (0.80)
10% NMPi, 50% PG (WS)	5.732 (0.092)	4.69 (0.15)	5.15 (0.27)
10% NMPo, 50% PG (WS)	11.746 (1.066)	1.85 (0.24)	2.21 (0.26)
0.025% Azone, 50% PG (WS)	3.091 (0.045)	2.73 (0.53)	2.63 (0.49)

[a]Control; WS, whole skin; SS, skin stripped with tape; SD, C_6, dimethylhexanamide; C_8, dimethyloctanamide; C_{10}, dimethyldecanamide; C_{12}, dimethyldodecanamide; C_{14}, dimethyltetradecanamide; C_{16}, dimethylhexadecanamide; M-18-OL, Hallcomid; NMPi, *N*-methylpyrollidine; NMPo, *N*-methylpyrrolidone.

may be due to the very low solubility of Azone in the vehicle with insufficient being present to exert an effect on penetration rates.

Table 5 records the solubility of ibuprofen in the various vehicles and compares the partition coefficients between skin and vehicle based upon both the initial dry weights of skin and also the final weights of the equilibrated samples.

In contrast to pH effects (Irwin et al., 1990) no large changes in the weight of the skin before and after these partitioning experiments was noted suggesting that no major compromising of the skin was occurring. *N,N*-Dimethyloctanamide, and its presence in the Hallcomid M-8,10, interfered with the ibuprofen peak during HPLC analysis and an extraction procedure was thus required. This involved acidification of the vehicle, extraction into chloroform, and then re-extraction of ibuprofen into the sodium hydroxide layer and of the amide into the chloroform layer. Standards and the controls, were treated similarly and good correlation was obtained. Although solubilities were measured satisfactorily with this technique, partition coefficients in the presence of the *N,N*-dimethyloctanamide were rather high. To overcome this effect and also because the penetration of ibuprofen could only be increased by a factor of two with the above enhancers naproxen, another nonsteroidal anti-inflammatory agent was also examined. Naproxen is less soluble in the 50% propylene glycol (pH 5.5) vehicle (1.92 compared to 3.09 mg ml^{-1}) and partition coefficients (octanol, 15.09; isopropyl myristate, 2.90) are lower than those of ibuprofen (octanol 95.9; isopropyl myristate, 21.5). This trend is continued in the partition coefficients in various organic vehicles and skin as all are lower for naproxen than ibuprofen and manifests itself as a decreased steady state flux through skin. However, when the skin was stripped the flux, in molar terms, was almost identical for both drugs and indicated a theoretical maximum equivalent to a twenty four-fold increase in flux for naproxen through normal skin. This compares with the six-fold increase available for ibuprofen. A number of vehicles were prepared containing the amides and the penetration rates of naproxen in suspensions were determined. Data are recorded in Table 6, solubility and skin partition results are

TABLE 6

Flux, lag time and permeability coefficient for the transport of naproxen through rat skin from suspensions in 50% propylene glycol (pH 5.5) containing penetration enhancers

Vehicle	Flux($\times 10^3$) (μmol cm^{-2} h^{-1})	Lag time (h)	K_p($\times 10^3$)(cm h^{-1})
Aqueous buffer (WS)	23.3 (1.0)	3.66 (0.25)	19.24 (0.83)
50% PG (SS)	376.6 (35.7)	1.49 (0.12)	45.14 (4.28)
50% PG (SD)	421.2 (22.7)	1.12 (0.10)	50.49 (2.72)
50% PG (SDF)	576.7 (66.1)	1.02 (0.15)	69.13 (7.92)
50% PG (WS)[a]	13.1 (2.1)	2.90 (0.10)	1.57 (0.25)
1% C_6, 50% PG (WS)	19.8 (7.0)	4.53 (0.53)	1.68 (0.59)
1% C_8, 50% PG (WS)	93.4 (13.7)	2.85 (0.28)	7.12 (1.04)
1% C_{10} 50% PG (WS)	131.3 (11.1)	3.28 (0.30)	12.10 (1.02)
0.145% C_{12}, 50% PG (WS)	42.4 (2.5)	2.05 (0.75)	5.81 (0.34)
0.034% C_{14}, 50% PG (WS)	68.2 (5.0)	4.05 (0.11)	7.17 (0.53)
2×10^{-3}% C_{16}, 50% PG (WS)	11.0 (2.2)	5.85 (0.36)	1.09 (0.22)
M-18-OL, 50% PG (WS)	78.6 (15.5)	4.41 (0.24)	11.37 (2.24)
10% M-8,10, 50% PG (WS)	105.3 (9.5)	3.98 (0.19)	1.61 (0.15)
10% NMPi, 50% PG (WS)	8.4 (2.5)	3.56 (0.20)	0.40 (0.12)
10% NMPo, 50% PG (WS)	28.3 (6.5)	4.93 (0.74)	0.80 (0.18)
0.025% Azone, 50% PG (WS)	2.89 (0.22)	2.89 (0.22)	4.26 (0.23)

[a]Control; WS, whole skin; SS, skin stripped with tape; SD, separated dermis SDF, separated dermis stored frozen. C_6, dimethylhexanamide, C_8, dimethyloctanamide; C_{10}, dimethyldecanamide; C_{12}, dimethyldodecanamide; C_{14}, dimethyltetradecanamide; C_{16}, dimethylhexadecanamide; M-18-OL, M-8,10, Hallcomids; NMPi, *N*-methylpyrollidine; NMPo, *N*-methylpyrrolidone.

shown in Table 7 and ratios of flux and permeability coefficient are listed in Table 8.

N-methylpyrollidone again shows enhancement (2-fold) which is solubility-driven with a real decrease in the permeability coefficient of 50%. Tables 6 and 8 show that *N,N*-dimethyloctanamide (7-fold) and *N,N*-dimethyldecanamide (10-fold) are the most effective enhancers together with Hallcomid M-8,10 (8-fold) which is essentially a mixture of these two amides. Azone, despite the low concentration used in this study improves the penetration flux of naproxen threefold. These enhancements reflect genuine increases in permeability as the solubility, although modified in these formulations, changes but little. The single exception to this is the case of Hallcomid M-8,10, a technical mixture. A parallel increase in partition from the vehicle into the skin is noted as the steady state flux increases. These results are shown clearly in Figs 2 and 3 where steady state flux, solubility and partition coefficient are plotted against length of alkyl chain. A parabolic relationship is noted in both cases with a peak at the *N,N*-dimethyldecanamide.

To demonstrate the reality of this correlation it was of interest to determine if any permeation

TABLE 8

Ratio of steady-state flux and permeability coefficient for naproxen through rat skin from 50% propylene glycol (pH 5.5) using various amide enhancers

Vehicle	Flux ratio	Permeability coefficient ratio
Aqueous buffer (WS)	1.8	12.3
50% PG (SS)	28.7	28.7
50% PG (SD)	32.2	32.2
50% PG (SDF)	44.0	44.0
50% PG (WS)[a]	1.0	1.0
1% C_6, 50% PG (WS)	1.5	1.1
1% C_8, 50% PG (WS)	7.1	4.5
1% C_{10}, 50% PG (WS)	10.0	7.7
0.145% C_{12}, 50% PG (WS)	3.2	3.7
0.034% C_{14}, 50% PG (WS)	5.2	4.6
2×10^{-3}% C_{16}, 50% PG (WS)	0.8	0.7
M-18-OL, 50% PG (WS)	6.0	7.2
10% M-8,10, 50% PG (WS)	8.0	1.0
10% NMPi, 50% PG (WS)	0.6	0.3
10% NMPo, 50% PG (WS)	2.2	0.5
0.025% Azone, 50% PG (WS)	3.0	2.7

[a]Control; WS, whole skin; SS, skin stripped with tape; SD, separated dermis; SDF, separated dermis stored frozen. C_6, dimethylhexanamide, C_8, dimethyloctanamide; C_{10}, dimethyldecanamide; C_{12}, dimethyldodecanamide; C_{14}, dimethyltetradecanamide; C_{16}, dimethylhexadecanamide; M-18-OL, M-8,10, Hallcomids; NMPi, *N*-methylpyrollidine; NMPo, *N*-methylpyrrolidone.

TABLE 7

Solubility and skin partition coefficients of naproxen in 50% propylene glycol (pH 5.5) containing various amide penetration enhancers

Vehicle	Solubility (mg ml^{-1})	Partition coefficient (P_m)	
		Dry	Wet
Aqueous buffer (WS)	0.279 (0.017)*	7.33 (0.62)	6.16 (0.34)
50% PG (SD)	1.921 (0.073)	4.84 (0.27)	4.02 (0.22)
50% PG (WS)[a]	1.921 (0.073)	1.80 (0.20)	1.75 (0.18)
1% C_6, 50% PG (WS)	2.722 (0.108)	2.91 (0.89)	3.58 (0.62)
1% C_8, 50% PG (WS)	3.022 (0.145)	6.74 (0.37)	7.67 (0.42)
1% C_{10} 50% PG (WS)	2.499 (0.059)	9.92 (0.82)	10.01 (0.84)
0.145% C_{12}, 50% PG (WS)	1.681 (0.160)	3.68 (0.32)	3.69 (0.34)
0.034% C_{14}, 50% PG (WS)	2.190 (0.052)	3.13 (0.75)	3.33 (0.81)
2×10^{-3}% C_{16}, 50% PG (WS)	2.329 (0.029)	2.93 (0.41)	3.11 (0.40)
M-18-OL, 50% PG (WS)	1.591 (0.069)	0.87 (0.17)	0.90 (0.13)
10% M-8,10, 50% PG (WS)	15.060 (0.190)	7.24 (0.39)	7.48 (0.52)
10% NMPi, 50% PG (WS)	4.785 (0.074)	2.38 (0.59)	2.42 (0.56)
10% NMPo, 50% PG (WS)	8.154 (0.439)	0.98 (0.17)	1.17 (0.18)
0.025% Azone, 50% PG (WS)	2.89 (0.22)	1.09 (0.08)	1.04 (0.06)

[a]Control; WS, whole skin; SD, separated dermis; C_6, dimethylhexanamide, C_8, dimethyloctanamide; C_{10}, dimethyldecanamide; C_{12}, dimethyldodecanamide; C_{14}, dimethyltetradecanamide; C_{16}, dimethylhexadecanamide; M-18-OL, M-8,10, Hallcomids; NMPi, *N*-methylpyrollidine; NMPo, *N*-methylpyrrolidone; Values in parentheses are standard errors; *, standard deviations.

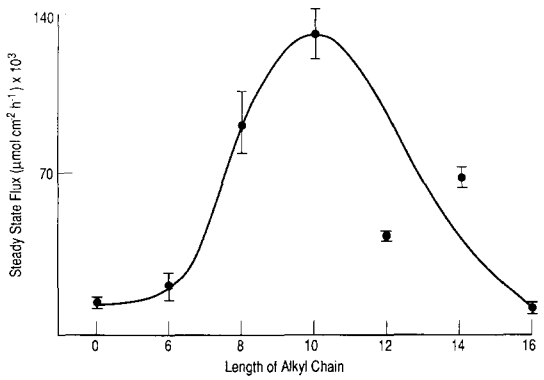

Fig. 2. Steady-state flux of naproxen, from suspensions in 50% aqueous propylene glycol (pH 5.5) containing *N,N*-dimethylamide enhancers, as a function of alkyl chain length.

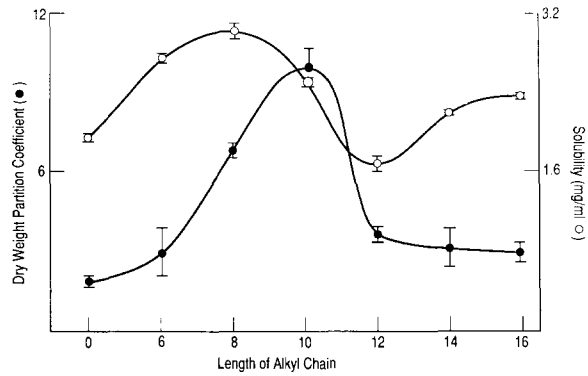

Fig. 3. Solubility and skin partition coefficients of naproxen, using 50% aqueous propylene glycol (pH 5.5) containing *N,N*-dimethylamide enhancers, as a function of alkyl chain length.

through the skin of the penetration enhancers could be detected. Table 9 shows the results of sampling the receptor phase and determining the concentration of the amide by HPLC after 24 h. This procedure was used, rather than the determination of true rates as the concentrations involved were poorly detectable. In the case of *N,N*-dimethyloctanamide and *N,N*-dimethyldecanamide permeation was more rapid and the penetration rate was determined by both the 24 h method and determining a full transport profile. The results were comparable but lag times of 2.73 h (C_8) and 3.85 h (C_{10}) caused some underestimate of the true steady-state flux using 24 h data alone.

These results show that on a molar basis *N,N*-dimethyloctanamide penetrates three-fold and the *N,N*-dimethyldecanamide 4-fold faster than naproxen in the respective vehicle (Table 6). This provides clear evidence that the amides penetrate the skin and are capable of increasing the drug partition into the skin. There is no evidence to suggest that any catastrophic destruction of the barrier is caused by the enhancers as the penetration profile of *N,N*-dimethyloctanamide followed the expected course with a lag time and a steady-state flux with no indication of a continuous increase in penetration rate as might be expected if the barrier were being destroyed. Moreover, the agreement between wet and dry partition coefficients for both ibuprofen and naproxen confirms the integrity of the barrier. Recently, a congeric series of Azone analogues has been tested for toxicity in cell culture (Ponec et al., 1989). Maximum activities in the C_8–C_{14} range were found which confirms the better penetration of medium chain compounds. The mode of action of these enhancers appears to have modification of drug partition into the stratum corneum as a considerable component. It is

TABLE 9

Penetration of some amide enhancers through rat skin (values in parentheses are standard errors

Enhancer	Amount penetrated in 24 h (μmol cm^{-2} 24 h^{-1})	Average flux ($\times 10^3$) (μmol cm^{-2} h^{-1})	K_P ($\times 10^3$) (cm h^{-1})
N,N-Dimethylhexanamide	2.73 (0.06)	113.6 (2.0)	1.63 (0.35)
N,N-Dimethyloctanamide	6.44 (0.31)	268.5 (13.0)	4.60 (0.53)
		318.0 (20.1)[a]	5.44 (0.84)
N,N-Dimethyldecanamide	11.17 (0.60)	465.5 (24.9)	9.26 (0.50)
		504.8 (34.8)[a]	10.05 (0.69)
N,N-Dimethyldodecanamide	0.13 (0.1)	5.4 (0.5)	0.86 (0.07)
N,N-Dimethyltetradecanamide	0.10 (0.01)	4.0 (0.2)	3.01 (0.15)
Azone	0.07 (0.01)	2.7 (0.4)	3.17 (0.46)

[a]*Flux determined by full rate profile*

TABLE 10

Penetration data and solubility of naproxen from lipophilic vehicles (N,N-dimethylamides are dissolved in liquid paraffin)

Vehicle	Flux($\times 10^3$) (μmol cm^{-2} h^{-1})	Lag time (h)	K_p($\times 10^3$)(cm h^{-1})	Solubility (mg ml^{-1})	n
Liquid paraffin	35.1 (3.8)	5.35 (0.27)	476.90 (24.73)	0.0169 (0.0018)	6
1% *N,N*-Dimethylbutanamide	42.7 (2.6)	4.22 (0.19)	35.38 (2.15)	0.278 (0.024)	6
1% *N,N*-Dimethyloctanamide	47.2 (11.8)	4.87 (0.17)	30.97 (7.74)	0.351 (0.004)	5
1% *N,N*-Dimethyltetradecanamide	44.2 (6.3)	5.04 (0.20)	47.12 (6.72)	0.216 (0.027)	5
1% *N,N*-Dimethyloctadecanamide	27.6 (2.1)	4.74 (0.10)	45.10 (3.43)	0.141(0.015)	6
Triacetin	1.6 (0.2)	3.25 (0.24)	0.02 (0.002)	25.013 (0.62)	10

also interesting to observe that a significant enhancement is obtained from the Hallcomid M-18-OL which is largely an oleic acid derivative and compares with the specific enhancement caused by the free acid in other systems, particularly those involving cationic drugs (Green and Hadgraft, 1987; Green et al., 1989). It seems unlikely that these amides act by ion pair formation, as suggested for Azone and salicylic acid (Hadgraft et al., 1985), because they are very weak bases with pK_a values of less than 1 (Higuchi et al., 1962; Adelman, 1964). To produce ions and micelles it is necessary to use strong acids (Menger and Jerkunica, 1979) for example the critical micelle concentration of *N,N*-dimethyldodecanamide is 0.03 ± 0.01 M in 95.2% sulphuric acid. Ion pairing with naproxen or ibuprofen under these conditions would not be expected as these weak acids (pK_a 4.4, ibuprofen) would be exclusively in the unionised form. In the 50% propylene glycol vehicle neither acid nor amide will be ionised sufficiently to promote ion pairing. When *N*-methylpyrollidine, a fairly strong base (pK_a 10.32) was used in the formulations, delivery efficiency was at a minimum. The solubility of both drugs in the vehicle was high, due to ionisation of the acids, but both flux and permeability coefficients were poorest of all despite a small increase in the measured partition coefficient.

As the penetration rate of naproxen could be readily enhanced the effect of other, more lipophilic vehicles was also examined. The effect of delivery from liquid paraffin is recorded in Table 10 where it is seen that the flux was increased some 3-fold over that from the 50% propylene glycol. Moreover, due to the very low solubility of the drug in this vehicle the permeability coefficient is extremely large making this a most efficient delivery system. The addition of enhancers at the 1% level caused no appreciable change in the steady-state flux. Such a result is probably due to the higher affinity of the enhancers for the vehicle, compared to the aqueous propylene glycol system, and confirms the importance of partition and skin permeation of the enhancer. Naproxen was extremely soluble in triacetin and this vehicle also provided an extremely low steady state flux. These observations agree with the expectation that the permeability constant was inversely proportional to solubility in the vehicle (Dugard and Scott, 1986). This rank order relationship is apparent in Table 10 with naproxen being the least soluble in liquid paraffin but the permeability constant the highest. The reverse is true for triacetin.

These data confirm the importance of partition coefficients and their modification when assessing the effectiveness of enhancers of percutaneous absorption.

Acknowledgements

We are grateful to Aston University for the award of a postgraduate research scholarship to F.D.S., to Nelson Research for the supply of Azone and to C.P. Hall, Chicago for samples of Hallcomids.

References

Adelman, R.L., Studies on the base strength of *N,N*-disubstituted amides. *J. Org. Chem.*, 29 (1964) 1837–1898.

Barry, B.W., Penetration enhancers. Mode of action in human skin. *Pharmacol. Skin*, 1 (1987) 121–137.

Bistline, R.G., Maurer, E.W., Smith, F.D. and Linfield, W.M., Fatty acid amides and anilides. Synthesis and antimicrobial properties. *J. Am. Oil Chem. Soc.*, 57 (1980) 98–103.

Dugard, P.H. and Scott, R.C., A method of predicting percutaneous absorption rates from vehicle to vehicle: An experimental assessment. *Int. J. Pharm.*, 28 (1986) 219–227.

Green, P.G., Guy, R.H. and Hadgraft, J., In vitro and in vivo enhancement of skin permeation with oleic and lauric acids. *Int. J. Pharm.*, 48 (1988) 103–111.

Green, P.G. and Hadgraft, J., Facilitated transfer of cationic drugs across a lipoidal membrane by oleic and lauric acid. *Int. J. Pharm.*, 37 (1987) 251–255.

Green, P.G., Hadgraft, J. and Ridout, G., Enhanced in vitro skin permeation of cationic drugs. *Pharm. Res.*, 6 (1989) 628–632.

Hadgraft, J. Walters, K.A. and Wotton, P.K., Facilitated transport of sodium salicylate across an artificial lipid membrane by Azone. *J. Pharm. Pharmacol.*, 37 (1985) 725–727.

Higuchi, T., Barnstein, C.H., Ghassemi, H. and Perez, W.E., Evaluation of amides and other very weak bases in acetic acid, *Anal. Chem.*, 34 (1962) 400–403.

Hwang, Y.-S. and Mulla, M.S., Insecticidal activity of alkanamides against immature mosquitoes. *J. Agric. Food Chem.*, 28 (1980) 1118–1122.

Irwin, W.J., Sanderson, F.D. and Li Wan Po, A., Percutaneous absorption of ibuprofen: Vehicle effects on transport through rat skin. *Int. J. Pharm.*, 66 (1990) 193–200.

Jacobi, E. and Lust, S., Insect repellants. *US Patent*, 3,005,747, 1961.

Menger, F.M. and Jerkunica, J.M., Aggregation in strong acid. A micelle of carbonium ions. *J. Am. Chem. Soc.*, 101 (1979) 1896–1898.

Ponec, M., Haverkort, M., Soei, Y.L., Kempenaar, J., Brussee, J. and Bodde, H., Toxicity screening of *N*-alkylazacycloheptan-2-one derivatives in cultured human cells. *J. Pharm. Sci.*, 78 (1989) 738–741.

Sekura, D.L. and Scala, J., The percutaneous absorption of alkyl methyl sulphoxides. *Adv. Biol. Skin*, 12 (1972) 257–269.

Walker, M. and Scott, R.C., A separation technique for producing intact epidermal membranes from human and rat skin for use in vitro percutaneous studies. *J. Pharm. Pharmacol.*, 36 (1984) 79P.

Walters, K.A., Penetration enhancers and their use in transdermal therapeutic systems. In: Guy, R.H. and Hadgraft, J. (Eds), *Transdermal Drug Delivery: Developmental Issues and Research Initiatives*, Dekker, New York, 1989, pp. 197–246.

Wiles, J.S. and Narcisse, J.K., The acute toxicity of dimethylamides in several animal species. *Am. Ind. Hygiene Assoc. J.*, 32 (1971) 539–545.

Woodford, R. and Barry, B.W., Penetration enhancers and the percutaneous absorption of drugs: an update. *J. Toxicol. Cut. Ocul. Toxicol.*, 5 (1986) 167–177.