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Effect of primary alcohols on percutaneous absorption

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Summary

The penetration enhancement of indomethacin by a series of primary alcohols with alkyl chains (C4–C16) was evaluated by the steady-state flux of indomethacin through shed skin of black rat snake. A parabolic relationship between the enhancement and the carbon atom number of the alcohols was established with tridecanol showing the maximum effect.

Introduction

Skin provides a superior route for drug administration in that the transdermal device can be removed immediately to terminate further supply of medication when complications arise. However, skin is resistant to most external chemicals and drugs. In the presence of transdermal penetration enhancers, it is possible that a drug could be administered transdermally and maintained at an effective blood concentration. Enhancers such as surfactants, aprotic solvents and Azone have been investigated extensively for some years to evaluate their enhancing effect. The mechanism by which enhancers affect the skin is not clear, but it is known that the lipophilicity of enhancers is an important factor in determining their efficiency. In this regard, long-chain primary alcohols can be useful, cheap and non-toxic enhancers. The purpose of this work is to evaluate the enhancing effects of a series of primary alcohols (butanol, octanol, decanol, undecanol, dodecanol, tridecanol, tetradecanol, hexadecanol) on the penetration enhancement of indomethacin through shed skin of black rat snake. The usefulness of snake skin as a skin model has already been reported previously by Higuchi and Konishi (1987).

Materials and Methods

Materials

Azone was supplied by Nelson Research and Development (Irvine, CA, U.S.A.) and indomethacin was purchased from Sigma Chemical Co. (St. Louis, MO, U.S.A.) and both were used as supplied. Alcohols obtained commercially were of reagent grade and white petrolatum was USP grade.

Preparation of ointment

The ointments were prepared by mixing 1% indomethacin, 5% enhancer (Azone or alcohols)

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and 94% petrolatum by weight. The indomethacin was first mixed with the enhancer and then combined with the petrolatum by stirring the mixture at 55–60 °C to obtain a homogeneous ointment.

Hydration of snake skin

The shed snake skin was cut into pieces of desired size and they were soaked for several minutes in water to remove the dirt from their surface. Each piece was spread on a plastic weighing pan which was placed in a water bath at 32 °C overnight to hydrate the skin. The water bath was covered with an aluminum foil during the hydration process.

Diffusion studies

Vertical diffusion cells (Fig. 1) were used in the in vitro diffusion studies. Approximately 30 mg of the ointment was applied evenly over 1.8 cm^2 of the snake skin and the skin was mounted on the receptor cell provided with a small magnetic bar and filled with about 9 ml of isotonic buffer (0.005 M NaH_2PO_4–Na_2HPO_4, pH = 7.2) of ionic strength 0.15 M. A rubber ring was placed on top of the receptor cell followed by the skin. After examining for bubbles which might be trapped in the receptor cells, the diffusion cell assembly was clamped together and transferred to a water bath (32 °C) under which were placed several magnetic stirrers. The content of the receptor cells was stirred continuously except during the period of sampling. Sampling was carried out periodically using gravity to collect the sample by putting in fresh isotonic buffer. The volume of sample withdrawn was 0.2 ml.

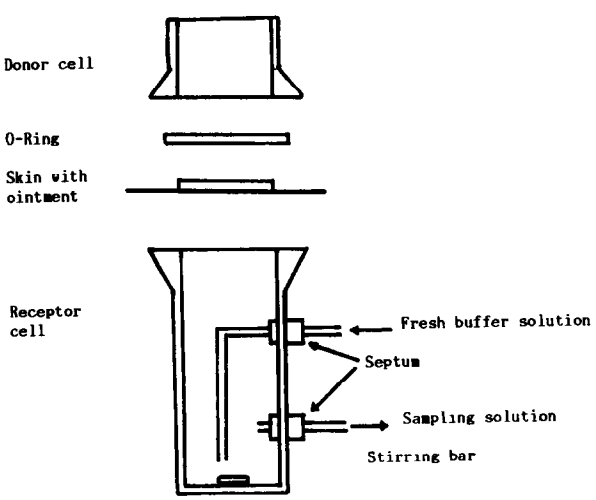

Fig. 1. Vertical diffusion cell assembly.

Chromatographic analysis for indomethacin

The amount of indomethacin in each sample was determined by HPLC using a Perkin-Elmer HPLC apparatus consisting of a UV detector (LC 90), a pump control unit (410 LC), an auto sampler (ISS-100) and an integrator (LCI-100). The chromatographic conditions were as follows: column, 100 mm × 4.6 mm i.d., RP-8 SPHERI-5, Brownlee Labs (Santa Clara, CA), mobile phase, acetonitrile–phosphate buffer (0.01 M NaH_2PO_4 $-H_3PO_4$, pH = 3.0) (1 : 1); flow rate, 1.0 ml/min.; UV detector, 260 nm; attenuation, 1.28 a.u.f.s. A sample volume of 15 μl was injected into the HPLC apparatus.

Results and Discussion

Fig. 2 shows the plot of the amount of indomethacin transported through the snake skin from the ointment in the presence of Azone or octanol vs time (h). Each data point represents the mean of 4 experimental trials. The bars indicate S.D. and the line of best fit was determined by linear regression. Indomethacin transported through the snake skin into the receptor cells from the ointment at a constant rate and no plateau was observed after 3 days. The transport of indomethacin from the control ointment without enhancer was insignificant.

The steady-state flux, J, can be calculated from the straight line plotted in Fig. 2 by Fick's first law.

$$J = \Delta M / S \cdot \Delta t$$

where ΔM (μg) is the amount of indomethacin transported; Δt, unit time (h) and S, unit area (cm^2). Table 1 shows the average steady-state flux for each experiment. Because the transport of indomethacin through snake skin varied from one

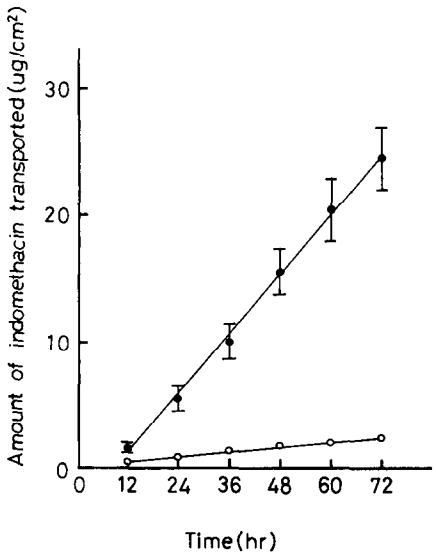

Fig. 2. Transport of indomethacin from 1% w/w indomethacin ointment through snake skin at 32°C. (●———●), Azone; (○———○), octanol. Each point represents the mean ± S.E.M. of 4 trials.

skin to another, Azone is used as the internal standard of comparison. In this way, the transdermal data are reproducible and they become 'relative penetration enhancement', Table 1.

It is well known that the partition coefficient of a series of straight alkyl chain compounds depends linearly on the number of carbon atoms

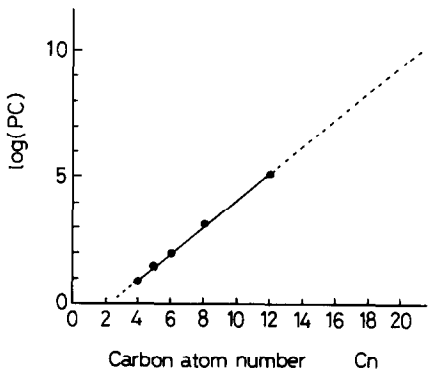

Fig. 3. The regression line between the logarithm of partition coefficients of primary alcohols and their carbon atom numbers.

(Leo et al., 1981). When the partition coefficients of several alcohols from literature (Valvani et al., 1981) was plotted against their number of carbon atoms, a straight line was obtained with a slope of 0.53, Fig. 3. From the plot, the partition coefficients for the other alcohols can be calculated (Table 1).

In Fig. 4, the relative steady flux of each alcohol to that of Azone was plotted against the carbon atom number of the alcohol. The enhancing effect of the alcohols increases as their carbon number increases reaching a maximum beyond which it decreases as the carbon number increases further. A parabolic relationship was obtained,

TABLE 1

Transdermal data of indomethacin through snake skin from petrolatum ointment at 32°C

	Alcohols			Azone	Relative permeability (%) [c]
	C_n [a]	log(PC) [b]	Steady-state flux ($\mu g/cm^2 \cdot h$)	Steady-state flux ($\mu g/cm^2 \cdot h$)	
Butanol	4	0.89 *	0.052 ± 0.009 [d]	0.644 ± 0.092 [d]	8.1 ± 1.4 [d]
Octanol	8	3.15 *	0.033 ± 0.003	0.392 ± 0.073	8.4 ± 0.8
Decanol	10	4.12	0.150 ± 0.071	0.542 ± 0.066	27.7 ± 13.1
Undecanol	11	4.65	0.297 ± 0.043	0.762 ± 0.028	39.0 ± 5.6
Dodecanol	12	5.13 *	0.392 ± 0.036	0.644 ± 0.092	60.9 ± 5.6
Tridecanol	13	5.71	0.471 ± 0.107	0.762 ± 0.028	61.8 ± 14.0
Tetradecanol	14	6.24	0.219 ± 0.039	0.542 ± 0.066	40.4 ± 7.2
Hexadecanol	16	7.29	0.103 ± 0.013	0.644 ± 0.092	16.0 ± 2.0

[a] Carbon atom number.

[b] Partition coefficient (* Experimental; in octanol-water system) taken from Leo et al., 1971.

[c] This ratio was obtained by dividing the steady-state flux of alcohol by that of Azone and multiplied by 100%.

[d] Mean ± S.D. S.D. = $\sqrt{(X_i - X)/N - 1}$, where X_i, individual measurement; X, mean value; N, the number of degree of freedom.

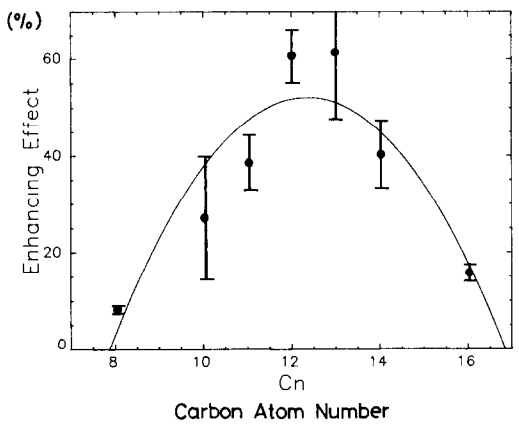

Fig. 4. Parabolic relationship between the enhancing effect and carbon atom number of primary alcohols.

Fig. 4, with tridecanol showing the maximum effect of approximately 62% of Azone. The optimum enhancing effect of a congeneric series of enhancers often occurs when compounds have 12 carbons (Cooper and Berner, 1985). The dependency of permeability of penetrants through skins on lipophilicity has been reported in literature (Higuchi and Konishi, 1987; Durrheim et al., 1980; Hawkins and Reifenrath, 1986 and Walters et al., 1983). The alcohols may play two possible roles in the penetration enhancement of indomethacin observed in this work. Firstly, the alcohols may increase the solubility of indomethacin in the resultant ointment so that the release of indomethacin may be increased and this may in turn increase the penetration rate of indomethacin. The parabolic relationship of penetration, Fig. 4, may be related to the release rate of indomethacin from the resultant ointment with the series of alcohols used in the present study. Secondly, if the alcohol did release from the ointment it may interact with the lipid components of the skin by swelling the skin lipids or causing them to become a more fluid-like material. This may change the permeability characteristics of the skin, thus reducing its resistance to indomethacin.

A quadratic function, $y = ax^2 + bx + c$, was used to simulate the parabolic relationship shown in Fig. 4, where y is enhancing effect and x represents the lipophilicity (carbon atom number) of the alcohols. The coefficients, a, b and c, were

TABLE 2

Polynomial regression coefficients by computer simulation

Coefficient		Estimated S.D.	t-Value
a	$= -2.5905$	0.6004	-4.3144
b	$= 64.043$	14.498	4.4175
c	$= -343.63$	85.113	-4.0373

$y = ax^2 + bx + c$. Correlation coefficient = 0.91288; observed F-value = 10.001. The regression analysis was done on a Macintosh computer.

obtained by computer simulation with the statistical parameters summarized in Table 2. The F-value and t-value are significant at 95% significance level. The correlation coefficient is 0.91 indicating a good correlation between the enhancing effect and carbon atom number. The real mechanism of penetration enhancement may be too complex to be represented by the above simple expression. However, it could provide a predictable expression for some other enhancers with structures similar to primary alcohols. Ibuki (1985) has proposed that the maximum biological activity among homologous compounds may be determined by a balance of their lipophilicity and their thermodynamic activities. Our study seems to support this hypothesis.

Acknowledgements

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References

Cooper, E.R. and Berner, B., In Rieger, M.H. (Ed.), *Surfactants in Cosmetics*, Dekker, New York, 1985, p. 1950.

Durrheim, H., Flynn, G.L., Higuchi, W.I. and Behl, C.R., Permeation of hairless mouse skin I. Experimental methods and comparison with human epidermal permeation by alkanols. *J. Pharm. Sci.*, 69 (1980) 780–786.

Hawkins, G.S. and Reifenrath, W.G., Influence of skin source, penetration cell fluid, and partition coefficient on in vitro skin penetration. *J. Pharm. Sci.*, 75 (1986) 378–381.

Higuchi, T. and Konishi, R., In vitro testing and transdermal delivery. *Ther. Res.*, 6 (1987) 280–288.

Ibuki, R., *Use of Snake Skin as a Model Membrane for Percutaneous Absorption Studies*. Ph.D. Thesis, The University of Kansas (1985).

Leo, A., Hansch, C. and Elkins, D., Partition coefficients and their uses. *Chem Rev.*, 71 (1971) 525.

Valvani, S.C., Valkowsky, S.H. and Roseman, T.J., Solubility and partitioning, IV. Aqueous solubility and octanol-water partition coefficients of liquid non-electrolytes. *J. Pharm. Sci.*, 70 (1981) 502–507.

Walters, K.A., Flynn, G.L. and Marvel, J.R., Physicochemical characterization of the human nail: permeation pattern for water and the homologous alcohols and differences with respect to the stratum corneum. *J. Pharm. Sci.*, 35 (1983) 28–33.