

THE EFFECT OF UREA AND LACTIC ACID ON THE PERCUTANEOUS ABSORPTION OF HYDROCORTISONE

J. E. Wahlberg and G. Swanbeck

From the Departments of Dermatology and Occupational Dermatology, Karolinska sjukhuset, Stockholm, Sweden

Abstract. To study the influence of urea and lactic acid on the percutaneous absorption of hydrocortisone, investigations were performed on excised human and guinea pig skin. Isotope-labelled hydrocortisone in solution or cream base, without or with urea or lactic acid, were applied to the epidermal side. The bathing solution on the dermal side was then collected and analysed for hydrocortisone content. Addition of lactic acid increased the absorption of hydrocortisone, probably due to a decrease in pH. Addition of urea caused an insignificant increase in hydrocortisone absorption through human skin and a considerable decrease through guinea pig skin. The increased hydrocortisone absorption observed in the presence of urea and lactic acid must be attributed to the latter substance. The absorption of hydrocortisone from a cream base was minimal and addition of urea caused no increase. No systemic effects after topical application of urea- and hydrocortisone-containing creams need be feared. The absorption of urea was small, being only about 1% of the amount applied during 2nd 24 hour period.

A favourable clinical effect of urea cream was noted in various skin diseases and its mechanism of action has been described by Swanbeck (6-8). As urea is an endogenous and nontoxic substance no side effects need be expected when urea is used in a 10% concentration for topical treatment. From a therapeutic point of view it is sometimes desirable to apply hydrocortisone concomitantly. It might then be possible that the urea and lactic acid present in the preparation could so increase the percutaneous absorption of hydrocortisone that systemic effects appear. The purpose of the following report was to elucidate this question. Some pilot experiments on the absorption of urea were also performed.

MATERIAL AND METHODS

A detailed description of the *in vitro* method has been reported previously (11-13). In short, freely prepared

pieces of whole skin are fixed in diffusion chambers made of plexi-glass. The chamber is placed on a hot-plate with regulated temperature in order that the absorption experiments can be performed under constant temperature conditions. The substance studied is applied to the epidermal side (area 3.1 cm²) and the dermal side is bathed by a continuous flow of saline. The bathing solution is then collected in an automatic fraction collector at previously determined intervals. Analytical methods are described below.

Skin. Human skin was obtained from the Plastic Surgery Clinic at the Karolinska Hospital, from patients who had undergone mammary plastic surgery. Skin from the backs of white guinea pigs was excised. After free preparation the resistance was measured in order to determine any possible injury to the skin (11). Only values less than 5 μ A/1 V was accepted.

Test substances. ¹⁴C-labelled hydrocortisone and urea were obtained from the Radiochemical Centre, Amersham, England.

Vehicles for hydrocortisone.

1. Cream base containing 10% urea (pH = 3)
2. Cream base without urea (pH = 3)
3. 2.5% Tween 20 + dist. water
4. 2.5% Tween 20 + 10% urea + dist. water
5. 2.5% Tween 20 + 10% urea + 5% acid lact. + dist. water
6. 2.5% Tween 20 + 5% acid lact. + dist. water

Vehicles for urea.

1. Distilled water
2. 2.5% Tween 20 + dist. water
3. 2.5% Tween 20 + 5% acid lact. + dist. water

Radioactivity was measured in a liquid scintillation spectrometer (Packard Tri-carb, model 3314). The solvent was 1 part of xylene + 3 parts of dioxane + 1 part of cellosolve. In this mixture were dissolved 1.4% PPO, 0.07% dimethyl-POPOP and 11.2% naphthalene (w/v). 10 ml of this solution was mixed with 4 ml of cellosolve and 1 ml of the bathing solution.

RESULTS

The results are presented in Tables I-III and Figs. 1-3. The absorption is given as a percentage

Table I. Percutaneous absorption of hydrocortisone (0.03%) in 2.5% Tween 20 and distilled water without and in the presence of urea and lactic acid. Excised human and guinea pig skin

Series no.	Urea conc. %	Lactic acid conc. %	pH	Type of skin	No of exp.	% absorption of amount applied					
						0-24 h		25-48 h		49-72 h	
						Range	Mean ± S.E.	Range	Mean ± S.E.	Range	Mean ± S.E.
1-5	0	0	6.20	Guinea pig	5	0.06-0.31	0.12 ± 0.05	0-0.55	0.25 ± 0.14	0.07-1.03	0.70 ± 0.23
6-7	0	0	6.20	Human	2	0-0.07	(0.04)	0-0.07	(0.04)	0-0.08	(0.04)
8-12	10	0	6.80	Guinea pig	5	0-0.04	0.04 ± 0.02	0.03-0.19	0.12 ± 0.03	0.10-0.34	0.19 ± 0.08
13-14	10	0	6.80	Human	2	0.03-0.17	(0.10)	0-0.17	(0.09)	0.02-0.22	(0.12)
15-19	10	5	2.55	Guinea pig	5	0.06-0.49	0.29 ± 0.09	0.16-1.54	0.69 ± 0.30	0.44-1.63	0.94 ± 0.25
20-21	10	5	2.55	Human	2	0.15-0.30	(0.22)	1.31-2.30	(1.80)	2.80-2.90	(2.85)
22-26	0	5	2.15	Guinea pig	5	0-0.36	0.15 ± 0.07	0.04-0.73	0.50 ± 0.12	0.08-1.42	0.97 ± 0.30
27-29	0	5	2.15	Human	3	0-1.13	(0.48)	0.12-3.70	(1.80)	1.27-4.12	(2.33)

of the amount applied, both per 24 hour period, and also cumulatively.

Effect of urea and lactic acid on the absorption of hydrocortisone
(Table I, Figs. 1, 2)

Addition of 10% urea increased the absorption of hydrocortisone through human skin to an insignificant extent (series 13-14 compared with series 6-7) while the absorption through guinea pig skin decreased (series 8-12 compared with 1-5).

Addition of 5% lactic acid increased the absorption both through human skin (series 27-29 compared with series 6-7) and through guinea pig skin (series 22-26 compared with 1-5).

Addition of both urea and lactic acid (series 15-19) gave, throughout, the highest absorption for guinea pig skin. For human skin (series 20-21) a higher absorption was obtained than by using the solution without addition (series 6-7) or with urea alone (series 13-14). Lactic acid alone (series 27-29) failed to modify the absorption, though it did increase gradually with time for both types

of skin. There was less absorption through guinea pig skin than through human skin when the test solutions contained lactic acid.

Effect of urea on the absorption of hydrocortisone in cream base (Table II)

Absorption was minimal and could be detected in series 31 during the 2nd day, in series 32-33 during the 3rd day. In series 30 and 34 no absorption at all could be observed. In those 3 experiments where the investigation could be completed during seven 24 hour periods the absorption was, in series 30, less than 0.04 and in series 31, 0.93% (both without urea) compared with 0.20% (series 33) with urea.

Absorption of urea (Table III, Fig. 3)

The absorption usually increased gradually with time and no steady state was observed. For the 20% urea solutions 1% was absorbed during the 2nd 24 hour period, after seven 24 hour periods 5-9% was absorbed. Addition of lactic acid increased absorption slightly.

Table II. Percutaneous absorption of hydrocortisone (1.0%) in a cream base, without and in the presence of urea. Excised guinea pig skin

Series no.	Urea conc. %	Amount cream applied (g)	% absorption of applied hydrocortisone					Cumulative absorption (%)					
			Day 1	Day 2	Day 3	Day 4	Day 5	Day 3	Day 4	Day 5	Day 6	Day 7	Day 10
			30	0	1.074	0	0	0	0	0	0	0	0
31	0	0.387	0	0.04	0.08	0.05	0.30	0.12	0.17	0.47	0.83	0.93	—
32	0	0.695	0	0	0.11	0.30	—	0.11	0.41	—	—	—	—
33	10	0.780	0	0	0.03	0.07	0.06	0.03	0.10	0.16	0.20	0.20	0.54
34	10	0.969	0	0	0	—	—	0	—	—	—	—	—

Table III. Percutaneous absorption of urea from solutions. Excised human and guinea pig skin

Series no.	Urea conc. %	Vehicle	Volume applied (ml)	Type of skin	% absorption of amount applied									
					Day 1		Day 2		Day 3	Day 4	Day 5	Day 6	Day 7	
					0-12 h	13-24 h	25-36 h	37-48 h						
35	10.0	Dist. water	2.0	Human	0.02	0.01	0.01	0.01	0.04	0.03	0.03	0.06	0.13	
36	20.0	2.5 % Tween 20 + dist. water	1.0	Guinea pig	0.01	0.10	0.35	0.39	1.24	1.02	1.69	2.48	—	
37	20.0	2.5 % Tween 20 + dist. water	1.0	Guinea pig	0.07		0.68		0.25	0.49	0.45	1.68	1.99	
38	20.0	2.5 % Tween 20 + 5.0 % lactic acid + dist. water	1.0	Human	0	0.12	0.16	0.20	1.05	1.68	2.02	1.45	1.68	
39	20.0	2.5 % Tween 20 + 5.0 % lactic acid + dist. water	1.0	Guinea pig	0.11	0.06	1.01	0.63	0.80	—	—	—	—	
40	20.0	2.5 % Tween 20 + 5.0 % lactic acid + dist. water	1.0	Guinea pig	0.04	0.12	0.94	1.46	0.73	0.43	0.57	0.50	4.02	

DISCUSSION

We still lack satisfactory methods to determine the percutaneous absorption of hydrocortisone

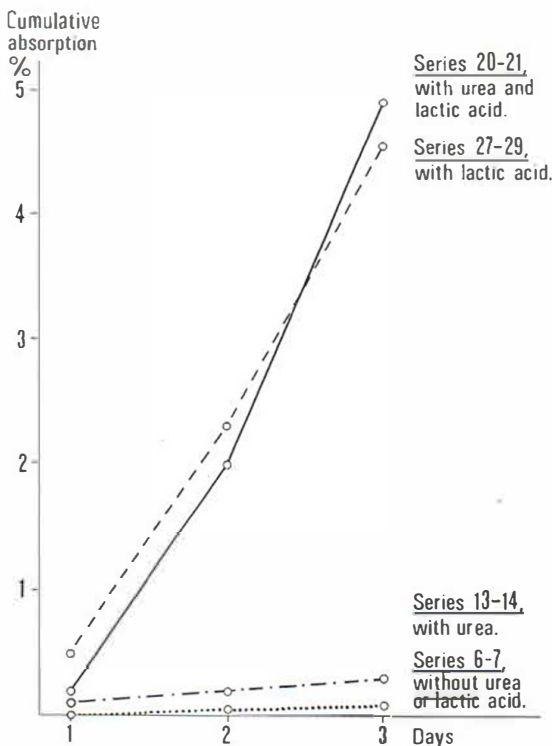


Fig. 1. Mean cumulative absorption of hydrocortisone in Tween 20 and distilled water, without and in the presence of urea and lactic acid. Human skin.

and its derivatives. The techniques tried are: measuring with gas-flow cell (5), urine analysis (1, 2), and vasoconstriction tests (4), but each one of these methods has its disadvantages (14). Therefore, one must for the moment accept that there are no satisfactory in vivo methods for certain substances and in those cases in vitro methods have to be used. Extrapolation to human subjects must be made carefully. Advantages and disadvantages with the in vitro method used in this report were discussed previously (11).

The investigations have shown that lactic acid causes an increase in the absorption of hydrocortisone. The pH of the solutions decreased from 6.2 and 6.8 to 2.55 and 2.15 (Table I) after addition of lactic acid. The gradual in-

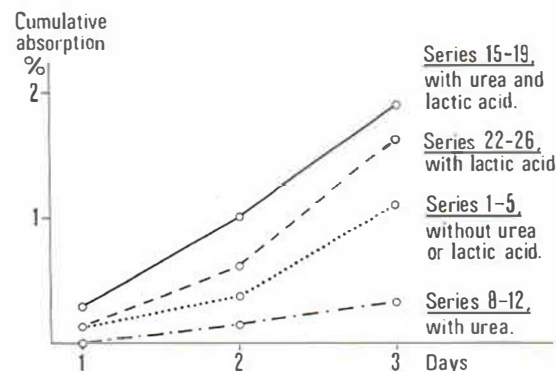


Fig. 2. Mean cumulative absorption of hydrocortisone in Tween 20 and distilled water, without and in the presence of urea and lactic acid. Guinea pig skin.

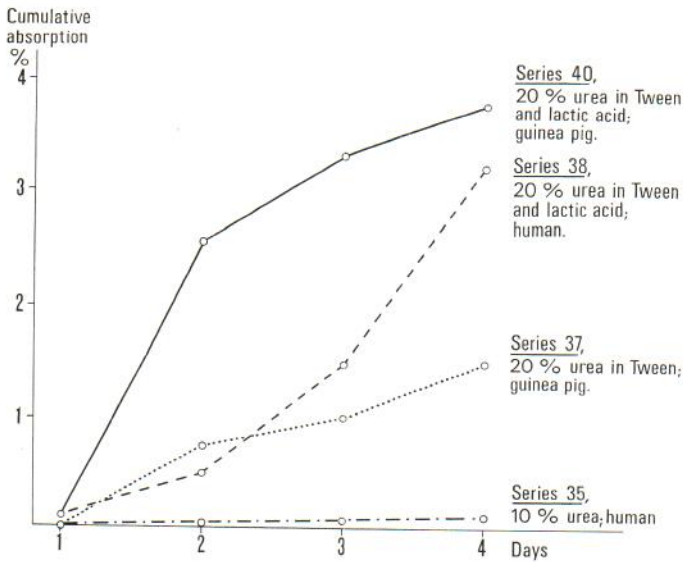


Fig. 3. Cumulative absorption of urea from aqueous solutions, without and in the presence of lactic acid and Tween 20. Human and guinea pig skin.

crease in absorption rate with time indicates an effect on the barrier of the solutions containing lactic acid.

On the other hand, urea had a minimal effect on the absorption of hydrocortisone. Only an insignificant increase was observed through human skin and a decrease through guinea pig skin. Similar changes were observed (14) in skin taken from various species. An increase in hydrocortisone absorption was observed with the addition of urea and lactic acid combined, which must be attributed to lactic acid alone.

The absorption of hydrocortisone from a cream base was minimal (Table II) and the addition of urea caused no increase. However, the method used could not demonstrate increased penetration into the skin. Systemic effects of hydrocortisone should not be expected, therefore.

The absorption of urea was small (Table III), but addition of lactic acid had an accelerating effect in this case too. Its absorption was earlier studied with other methods (3, 9, 10) and the results show a good agreement.

REFERENCES

- Feldmann, R. J. & Maibach, H. L.: Penetration of ^{14}C hydrocortisone through normal skin. *Arch Derm (Chicago)* 91: 661, 1965.
- Percutaneous penetration of ^{14}C hydrocortisone in man. *Arch Derm (Chicago)* 94: 649, 1966.
- Absorption of some organic compounds through the skin in man. *J Invest Derm* 54: 399, 1970.
- McKenzie, A. W. & Stoughton, R. B.: Method for comparing percutaneous absorption of steroids. *Arch Derm (Chicago)* 86: 608, 1962.
- Malkinson, F. D.: Studies on the percutaneous absorption of C^{14} labeled steroids by use of the gas-flow cell. *J Invest Derm* 31: 19, 1958.
- Swanbeck, G.: A new treatment of ichthyosis and other hyperkeratotic conditions. *Acta Dermatovener (Stockholm)* 48: 123, 1968.
- Karbamidkräm mot torr och hyperkeratotisk hud. *Läkartidningen* 65: 2286, 1968.
- Swanbeck, G. & Rajka, G.: Antipruritic effect of urea solutions. *Acta Dermatovener (Stockholm)* 50: 225, 1970.
- Treherne, J. E.: The permeability of skin to some nonelectrolytes. *J Physiol* 133: 171, 1956.
- Vinson, L. J., Singer, E. J., Koehler, W. R., Lehman, M. D. & Masurat, M. T.: The nature of the epidermal barrier and some factors influencing skin permeability. *Toxic Appl Pharmacol* 7: 7, 1965.
- Wahlberg, J. E.: Percutaneous absorption of sodium chromate (^{51}Cr), cobaltous (^{60}Co), and mercuric (^{203}Hg) chlorides through excised human and guinea pig skin. *Acta Dermatovener (Stockholm)* 45: 415, 1965.
- Transepidermal or transfollicular absorption? In vivo and in vitro studies in hairy and non-hairy guinea pig skin with sodium (^{22}Na) and mercuric (^{203}Hg) chlorides. *Acta Dermatovener (Stockholm)* 48: 336, 1968.
- Percutaneous absorption of trivalent and hexavalent chromium (^{51}Cr) through excised human and guinea pig skin. *Dermatologica* 141: 288, 1970.
- Percutaneous absorption. In: *Current Problems in Dermatology*, vol. 5. Karger, Basel, 1972.

Received November 15, 1972

J. E. Wahlberg, M.D.
Department of Occupational Dermatology
Karolinska sjukhuset
S-104 01 Stockholm 60
Sweden