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UREA AND RETINOIC ACID IN ICHTHYOSIS AND THEIR EFFECT ON TRANSEPIDERMAL WATER LOSS AND WATER HOLDING CAPACITY OF STRATUM CORNEUM

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Abstract. Fourteen patients with ichthyosis (8 sex-linked, 6 autosomal dominant) were treated in a double-blind trial with urea cream or its base and with retinoic acid. Urea cream caused clinical improvement, increased water-holding capacity of the stratum corneum by 100%, and had little effect on the epidermal water barrier. Retinoic acid was useful for treating keratosis pilaris, but caused irritation. The epidermal barrier and water-holding capacity were little affected.

Several reports have suggested that both 10% urea cream (Calmurid®—Pharmacia Ltd., London, G.B.) (2, 10, 11, 12) and retinoic acid (Vitamin A acid) (3, 6, 8, 9, 13) are of value in the management of ichthyosis. We therefore investigated their possible mode of action in vivo by studying their effects on two parameters of epidermal function. The parameters chosen were:

1. Transepidermal water loss (TEWL) as an indicator of the integrity of the epidermal water barrier.

2. Hydration of epidermal scales in vitro at various humidities as an indication of the water-holding capacity (WHC) of corneal cells. Urea and retinoic acid were applied *in vivo* and the water-holding capacity of the scales was subsequently measured in vitro.

METHOD

Fourteen patients with ichthyosis were studied. Eight had the sex-linked variety and 6 the autosomal dominant type (14). The ages and sexes of the patients are shown in Table I. Keratosis pilaris was present in all the autosomal dominant cases but in none of the sex-linked cases. There was a past history of hand eczema in case 6 and urticaria in case 8.

PROCEDURE

For 3 weeks before the start of the trials no topical applications were used and no drugs taken.

In a double-blind trial urea cream was applied to one upper limb and the base was applied to the opposite limb (14 subjects, in cases 7 and 8 the lower legs were used). The creams were applied twice daily using the method suggested by the manufacturers, i.e. applied as a thick film which was left undisturbed for 3–5 minutes before being rubbed into the skin.

Retinoic acid was applied to one lower leg and left overnight under polythene occlusion. The opposite limb was left untreated and acted as the control for retinoic acid (13 subjects, in case 7 the upper limbs, and in case 8 the thighs were tested).

The three skin preparations used were:

1. 10% urea cream (Calmurid)
2. The base of Calmurid (a hydrous cream in which the urea, betaine and lactic acid had been removed.
3. retinoic acid 0.1% in soft yellow paraffin.

At the end of the 3 week period all treatment was stopped for 1½ days and the skin parameters remeasured.

1. *Epidermal water barrier*

The water loss by diffusion through skin, when sweat glands are inactive, is known as transepidermal water loss (TEWL). Measurement of TEWL can be used as a method of assaying the effect of topical skin applications on the skin water barrier function. Damage to the barrier increases TEWL.

TEWL was measured by a standard technique using an electrolytic hygrometer (Meeco), after abolition of eccrine sweating by the application of poldine methosulphate. The galvanic skin resistance (GSR) was measured to ensure that sweating had been completely abolished. Skin temperature and ambient humidity were measured and TEWL corrected for skin temperature (4).

2. *Water holding capacity of stratum corneum scales*

The capacity of stratum corneum scales to take up water was assessed by allowing them to equilibrate in an

Table I. *Effect of urea cream and retinoic acid on transepidermal water loss and on clinical state*

Case no.	Age	Sex	Autosomal dominant (AD) or X-linked (SL)	Topical application (Urea cream: urea cream base: retinoic acid RA) Pre-trial readings = 1 Post-trial readings = 2	Trans-epidermal water loss (mg cm^{-2} h^{-1})	Physician's assessment: ++ Great improvement + Improved 0 No change − Worse	Patient's assessment
1	61	♂	SL	Control 1	0.8	0	0
				Control 2	0.8	0	0
				Urea 2	1.0	++	++
				Base 2	1.1	0	0
2	17	♂	SL	Control 1	1.2	0	0
				Base 2	1.1	0	0
				Urea 2	1.1	++	++
				RA 2	0.7	++	++
				Control 2	1.0	0	0
3	18	♂	SL	Control 1	0.7	0	0
				Urea 2	0.7	++	++
				Base 2	0.6	0	0
				Control 2	0.9	0	0
				RA 2	0.7	++	++
4	18	♀	AD	Control 1	0.9	0	0
				Urea 2	0.6	++	++
				Base 2	0.9	0	0
				RA 2	1.2	++	−
				Control 2	0.7	0	0
5	12	♂	SL	Control 1	0.8	0	0
				Base 2	0.7	++	++
				Urea 2	0.7	+	0
				RA 2	2.8	++	++
				Control 2	0.9	0	0
6	12	♂	AD	Control 1	0.9	0	0
				Base 2	0.9	0	0
				Urea 2	1.0	0	0
				Control 2	1.0	0	0
				RA 2		—	—
7	18	♀	AD	Control 1	0.6	0	0
				Base 2	0.7	0	0
				Urea 2	0.5	++	++
				Control 2	0.9	0	0
				RA 2	1.5	++	—
8	53	♂	AD	Control 1	1.5	0	0
				Urea 2	0.4	+	+
				Base 2	0.4	0	0
				RA 2	0.4	++	−
				Control 2	0.3	0	0
9	55	♀	AD	Control 1	0.6	0	0
				Urea 2	0.5	++	++
				Base 2	0.7	++	++
				Control 2	0.5	0	0
				RA 2	0.5	++	++
10	27	♂	SL	Control 1	0.8	0	0
				Base 2	0.8	0	0
				Urea 2	0.6	+	+
				Control 2	0.7	0	0
				RA 2	0.9	0	0
11	15	♀	AD	Control 1	0.9	0	0
				Urea 2	0.6	+	++
				Base 2	0.8	0	0
				RA 2	0.7	++	0
				Control 2	0.7	0	0

Table I (*continued*)

Case no.	Age	Sex	Autosomal dominant (AD) or X-linked (SL)	Topical application (Urea cream: urea cream base: retinoic acid RA) Pre-trial readings = 1 Post-trial readings = 2	Trans-epidermal water loss (mg cm^{-2} h^{-1})	Physician's assessment: ++ Great improvement + Improved 0 No change − Worse	Patient's assessment
12	56	♂	SL	Control 1	0.7	0	0
				Base 2	0.8	+	+
				Urea 2	0.9	+	++
				Control 2	0.8	0	0
				RA 2	1.9	++	−
13	48	♂	SL	Control 1	0.5	0	0
				Urea 2	0.3	++	++
				Base 2	0.6	0	0
				RA 2	0.9	+	+
				Control 2	0.9	0	0
14	14	♂	SL	Control 1	0.9	0	0
				Urea 2	1.5	++	++
				Base 2	1.6	+	+
				RA 2	2.2	++	0
				Control 2	1.8	0	0

atmosphere of known humidity. The gain in weight from the dry weight (established by initial desiccation) was a measure of the water-holding capacity (or water content) of the corneum at a given humidity.

Surface scales were scraped off the stratum corneum and weighed in a polytetra fluoroethylene (PTFE) cup. The scales were left in a desiccator and re-weighed daily. When equilibrium was reached (usually in 4–8 days) and there was no further weight loss, the scales in the PTFE cup were transferred to the high humidity atmosphere. A saturated solution of sodium chloride in the base of a desiccator jar produced a relative humidity (RH) of 82% (1). The scales were allowed to reach equilibrium at RH 82% and were reweighed. The method is similar to one used by Swanbeck (12) for measuring the water content of ichthyotic scales after he had soaked them in urea.

The presence of urea on the skin was checked by using *O*-phthaldialdehyde paint in an area where sweating had been abolished. The paint stained the skin a diffuse black at the site of previous application of urea cream. Juhlin & Shelley (7) showed that *O*-phthaldialdehyde could be used for detecting the presence of sweating by its action on the urea in sweat.

RESULTS

Transepidermal water loss

The results can be seen in Table I. The TEWL was slightly reduced by the application of urea cream (Urea 0.7 ± 0.3 mg cm^{-2} h^{-1}. Base 0.8 ± 0.3

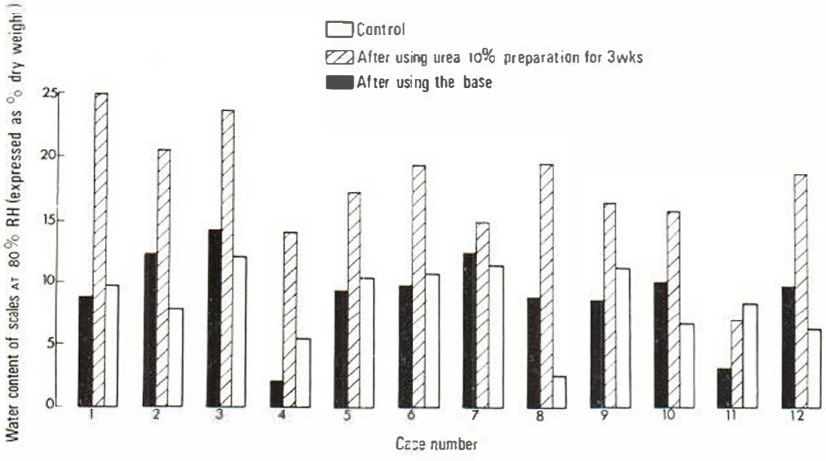

Fig. 1. Effect of urea preparation on water holding capacity of scales in ichthyosis.

Table II. *The effect of 0.1% retinoic acid on the water-holding capacity (WHC) of stratum corneum scales at 82% RH (WHC expressed as % dry weight)*

Case no.	Retinoic acid	Control
1	9.5	10.5
2	18.1	13.2
3	13.5	9.3
7	6.7	4.3
8	9.7	11.6
9	7.4	7.3
10	7.7	6.5
11	8.0	8.7
12	13.4	5.9
13	2.9	3.1
14	9.8	6.9

mg cm^{-2} h^{-1}. In a paired comparison the mean difference was significantly different from zero at the 95% level).

Retinoic acid, however, increased TEWL slightly: (Retinoic acid 1.2 ± 0.7 mg cm^{-2} h^{-1}. Control 0.8 ± 0.3 mg cm^{-2} h^{-1} paired comparison. Significantly different at 95% level).

There was no significant difference between the findings in the two types of ichthyosis, but the number of cases was small.

Water-holding capacity of stratum corneum at 82% RH

As seen in Fig. 1, urea caused a very highly significant increase in the water-holding capacity of the corneocytes (Urea 17.8 ± 4.8%. Base 9.2 ± 3.5%. Significantly positive at the 99.9% level).

Retinoic acid caused a slight increase in the WHC when compared with its control (Retinoic acid 9.7 ± 4.1%. Control 7.9 ± 3.1%. Significantly positive at the 95% level) (Table II).

There was no significant difference in the response of either type of ichthyosis to the two agents: the numbers were very small, however.

In all patients, use of *0*-phthaldialdehyde showed that urea was present in the stratum corneum the day after stopping the urea cream. In 1 patient (case 14) the water-holding capacity was measured up to 24 days after stopping urea treatment (Table III) and it showed that by the third day the WHC had fallen to the pre-treatment level.

Table III. *Repeated measurements of water-holding capacity (WHC) of stratum corneum scales after cessation of urea therapy*

Days after ceasing therapy	1	3	10	17	24
WHC, % dry weight	18.8	11.5	6.7	8.5	7.6

Clinical assessment

Significant clinical improvement occurred at sites treated with urea cream compared with those treated with the base (Table I). None of the ichthyotic patients complained of irritation.

Retinoic acid was more effective than urea cream in reducing the keratoses, where keratosis pilaris was present, but dermatitis resulted in 2 patients (cases 12 and 13), erythema in 4, and pruritis in 8.

DISCUSSION

In this double-blind trial, urea cream proved subjectively and objectively to be more effective than its base. The stratum corneum after 3 weeks of treatment had increased its capacity to take up water by 100%. This effect was probably due to the presence of the hygroscopic urea in the stratum corneum. Although the tests were performed 24 hours after stopping therapy, *0*-phthaldialdehyde paint showed urea was still present in the stratum corneum. None of the patients complained of irritation and measurement of transepidermal water loss confirmed that there had been no damage to the water barrier. TEWL may be normal or slightly increased in ichthyosis (5).

Retinoic acid caused skin irritation in 8 of the 13 cases. The irritant effect of retinoic acid was reflected in a slight increase in the rate of TEWL which would be compatible with some damage to the epidermal barrier. Keratosis pilaris was greatly improved in the 6 patients who had it. Water-holding capacity of the stratum corneum was little affected by retinoic acid.

The results of these investigations confirm that the effect of urea in ichthyosis is probably due to its hygroscopic effect. The results do not shed any light, however, on the mechanism of the action of retinoic acid. It has been suggested that it affects the lipid sheaths of the keratin fibrils of the stratum corneum (13). From the present findings this is unlikely to be the case, since, were this so, one would have expected retinoic acid to have produced greater destruction of the epidermal barrier and water-holding function of the stratum corneum.

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REFERENCES

1. Acheson, D. T.: Humidity and Moisture Measurements, vol. 3. Reinhold, New York 1965.
2. Ashton, H., Frenk, E. & Stevenson, C. J.: Urea as a topical agent. Brit J Derm *84:* 194, 1970.
3. Frost, P. & Weinstein, G. D.: Topical administration of Vitamin A acid for ichthyosiform dermatoses and psoriasis. JAMA *207:* 1863, 1969.
4. Grice, K., Sattar, H. & Baker, H.: Skin temperature and transepidermal water loss. J Invest Derm *57:* 108, 1971.
5. Grice, K. & Bettley, F. R.: Skin water loss and accidental hypothermia in psoriasis ichthyosis and erythroderma. Brit Med J *4:* 195, 1967.
6. Jarrett, A. & Spearman, R. I. C.: Vitamin A and the skin. Brit J Derm *82:* 197, 1970.
7. Juhlin, L. & Shelley, W. B.: A stain for sweat pores. Nature (Lond.) *213:* 408, 1967.
8. Logan, W. S.: Vitamin A and keratinization. Arch Derm (Chicago) *105:* 748, 1972.
9. Mirrer, E. & McGuire, J.: Lamellar ichthyosis, response to retinoic acid. Arch Derm (Chicago) *102:* 548, 1970.
10. Pope, F. M., Rees, J. K., Wells, R. S. & Lewis, K. G. S.: Out-patient treatment of ichthyosis, a double-blind trial of ointments. Brit J Derm *86:* 291, 1972.
11. Steward, W. D., Danto, J. L. & Maddin, W. S.: Urea cream. Cutis *5:* 1241, 1969.
12. Swanbeck, G.: A new treatment of ichthyosis and other hyperkeratotic conditions. Acta Dermatovener (Stockholm) *48:* 123, 1968.
13. Thomson, J. & Milne, J. A.: The use of retinoic acid in congenital ichthyosiform erythroderma. Brit J Derm *81:* 452, 1969.
14. Wells, R. S. & Kerr, C. B.: Clinical features of autosomal dominant and sex-linked ichthyosis in an English population. Brit Med J *1:* 947, 1966.

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