

The complete picture



Jonathan Hadgraft

The School of Pharmacy, University of London



HALCYON MEDICA LTD



21st annual CRS meeting, Nice, France, 1994
Jean-Paul Marty, co-Scientific Chairman

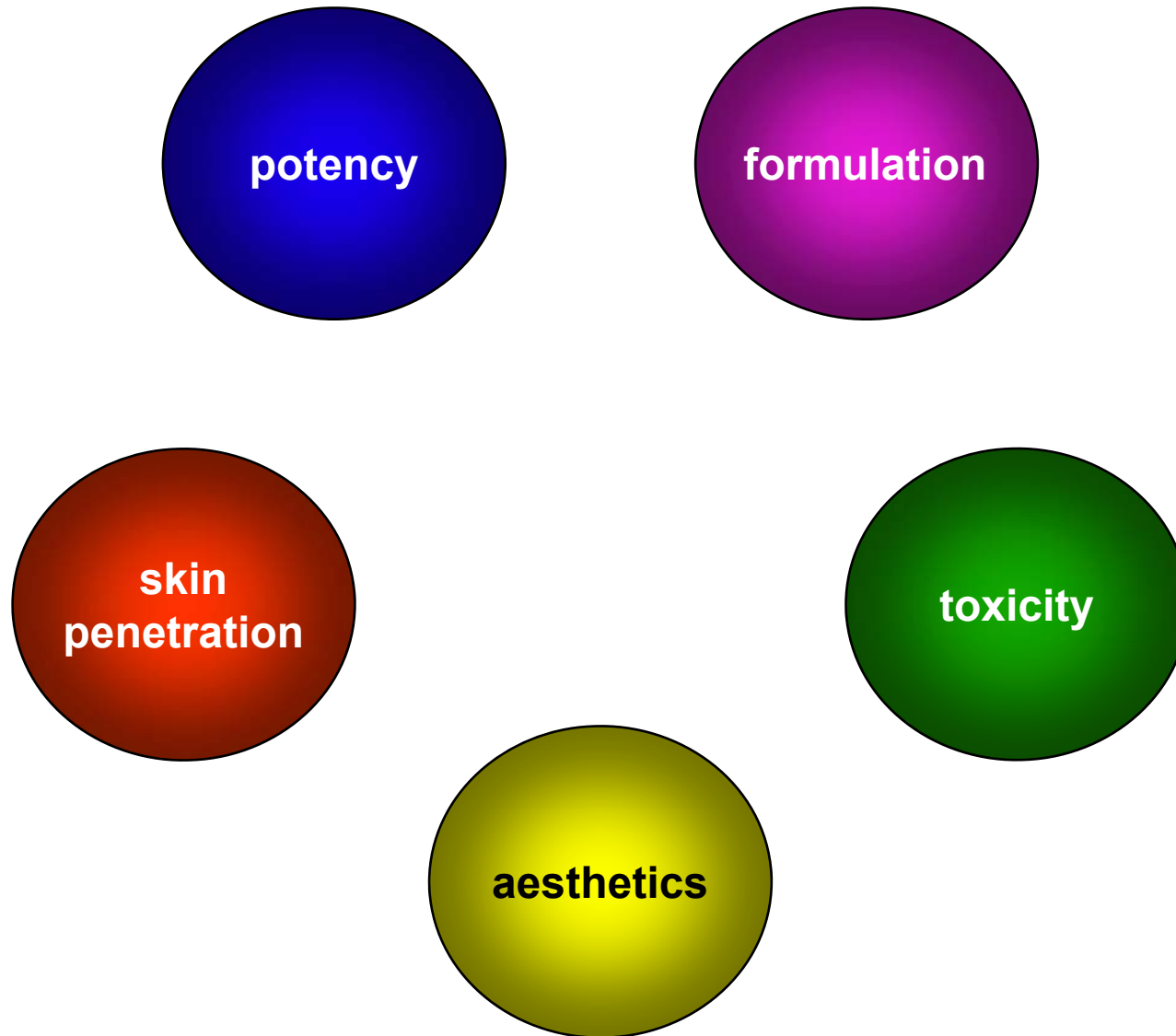


When I think of Jean-Paul I
see his ever smiling face and
remember his love of life



The complete skin scientist

Pharmaceutical and cosmetic skin delivery



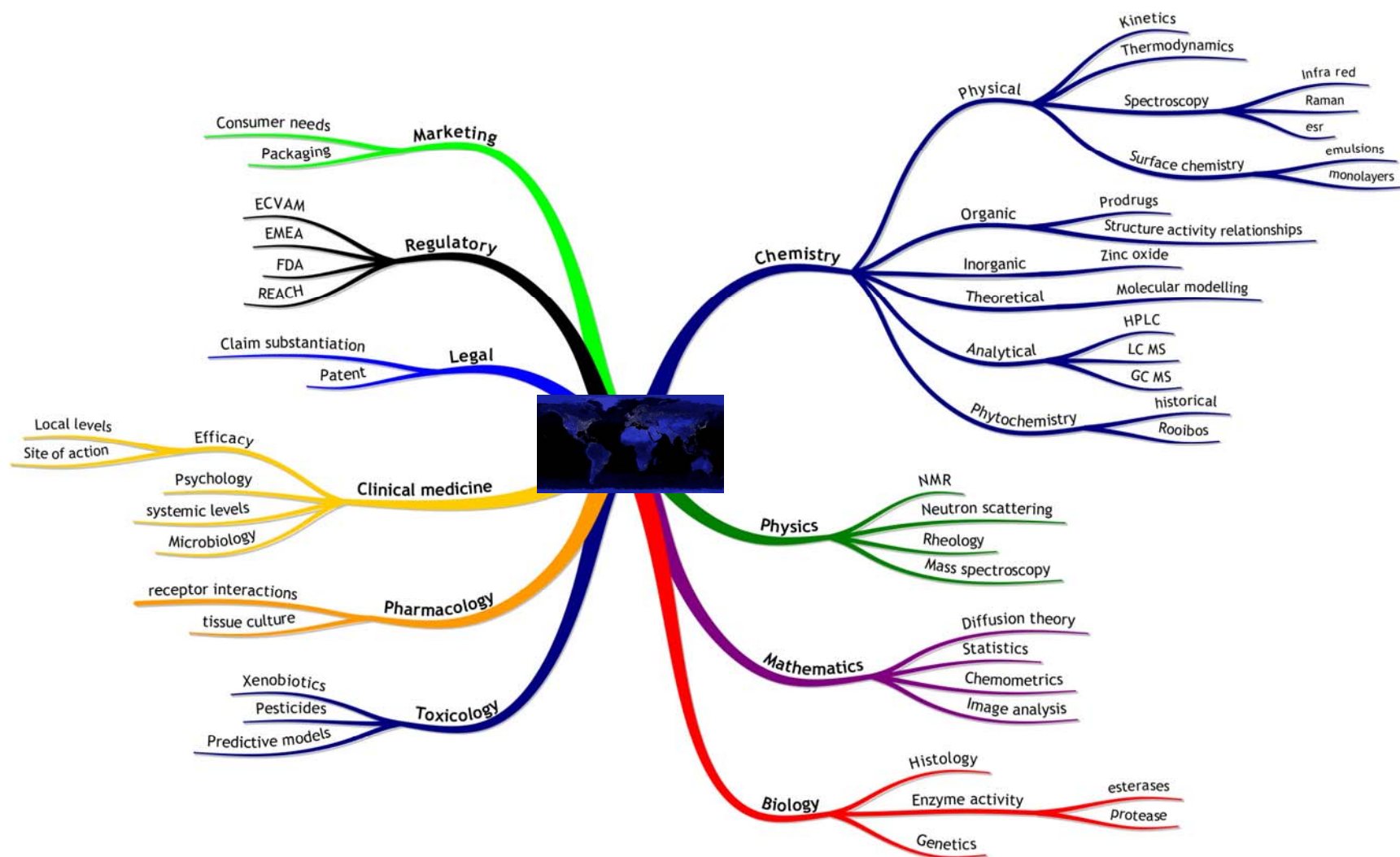
Gold star: effective product



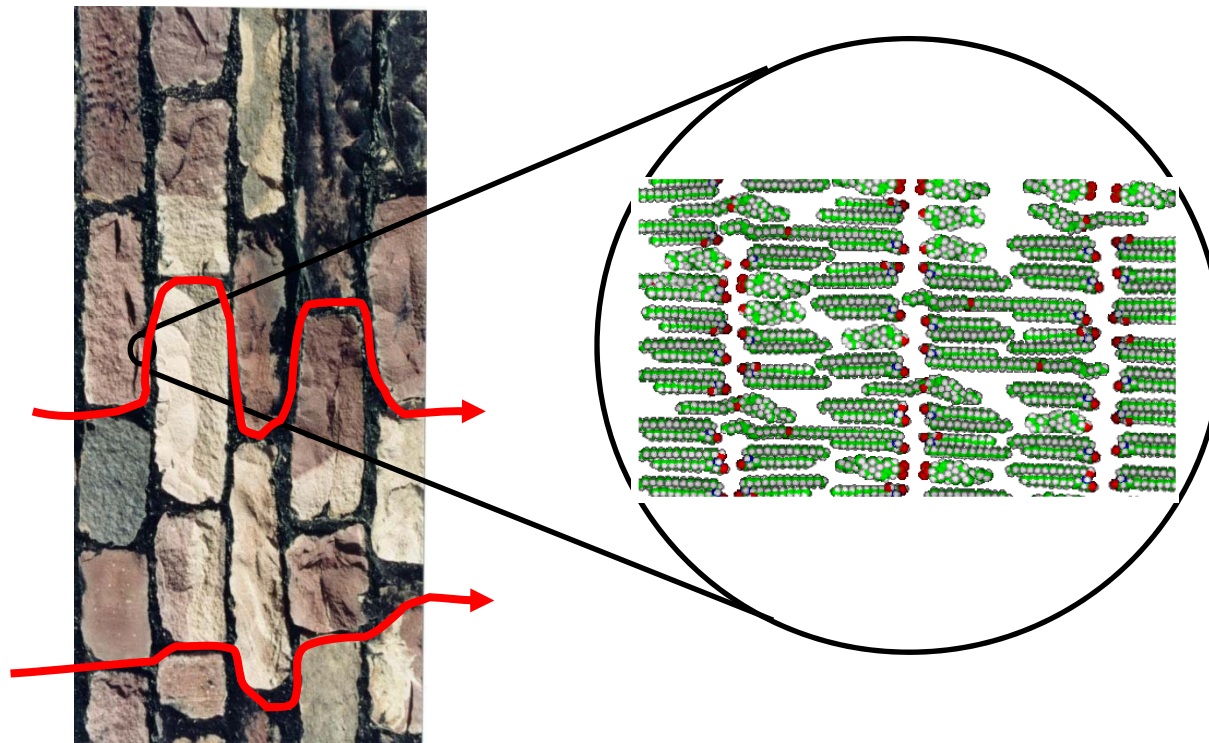
WESTMINSTER DIARY Comment from Tam Dalyell



Spike Gerrell



Composition & structure of intercellular channels



ceramides (50%)
cholesterol (25%)

cholesteryl sulphate (5%)
free fatty acids (15%)

Michaels et al.
1975

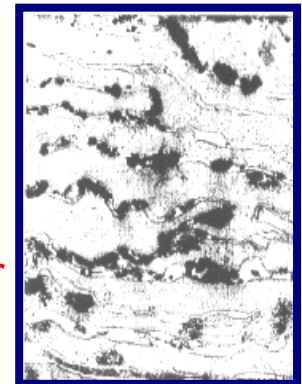
Albery
& Hadgraft

Elias

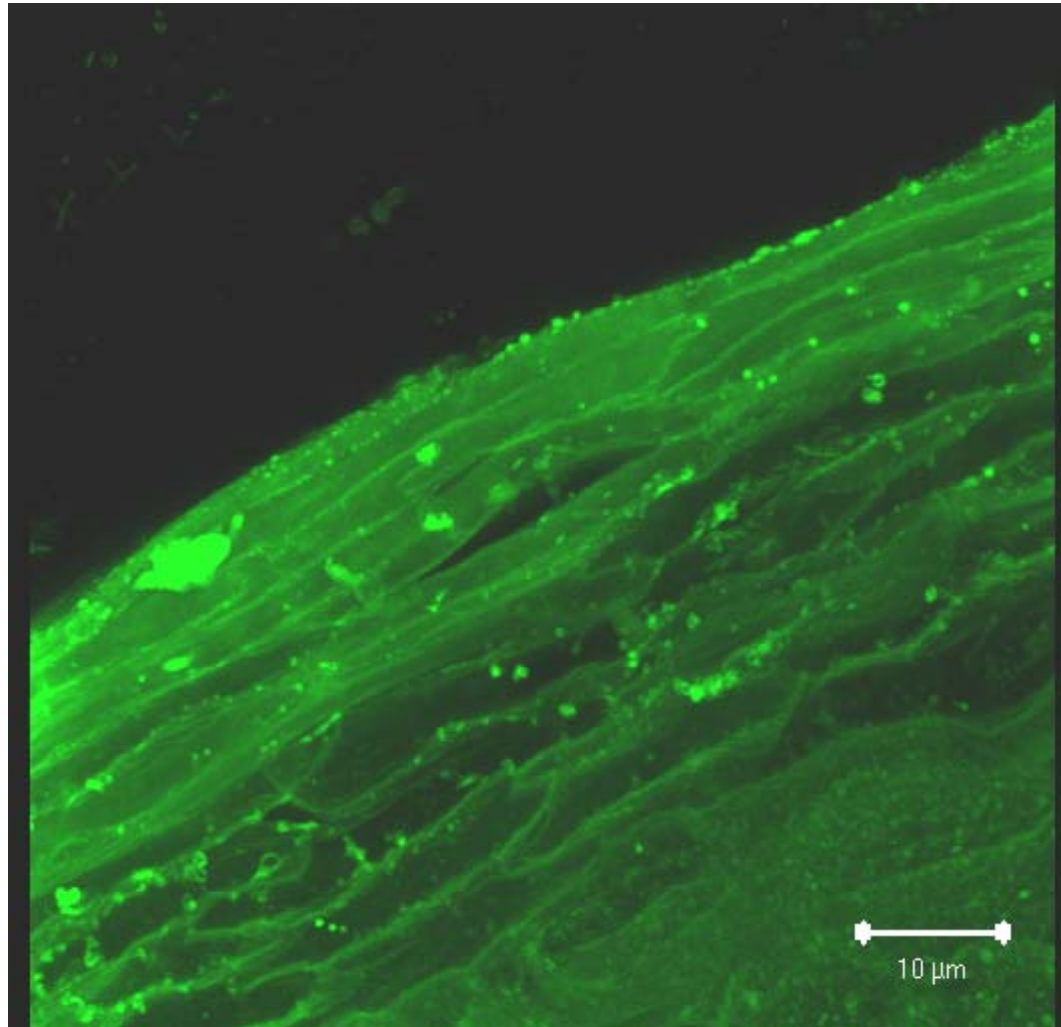
Boddé

Potts
& Francoeur

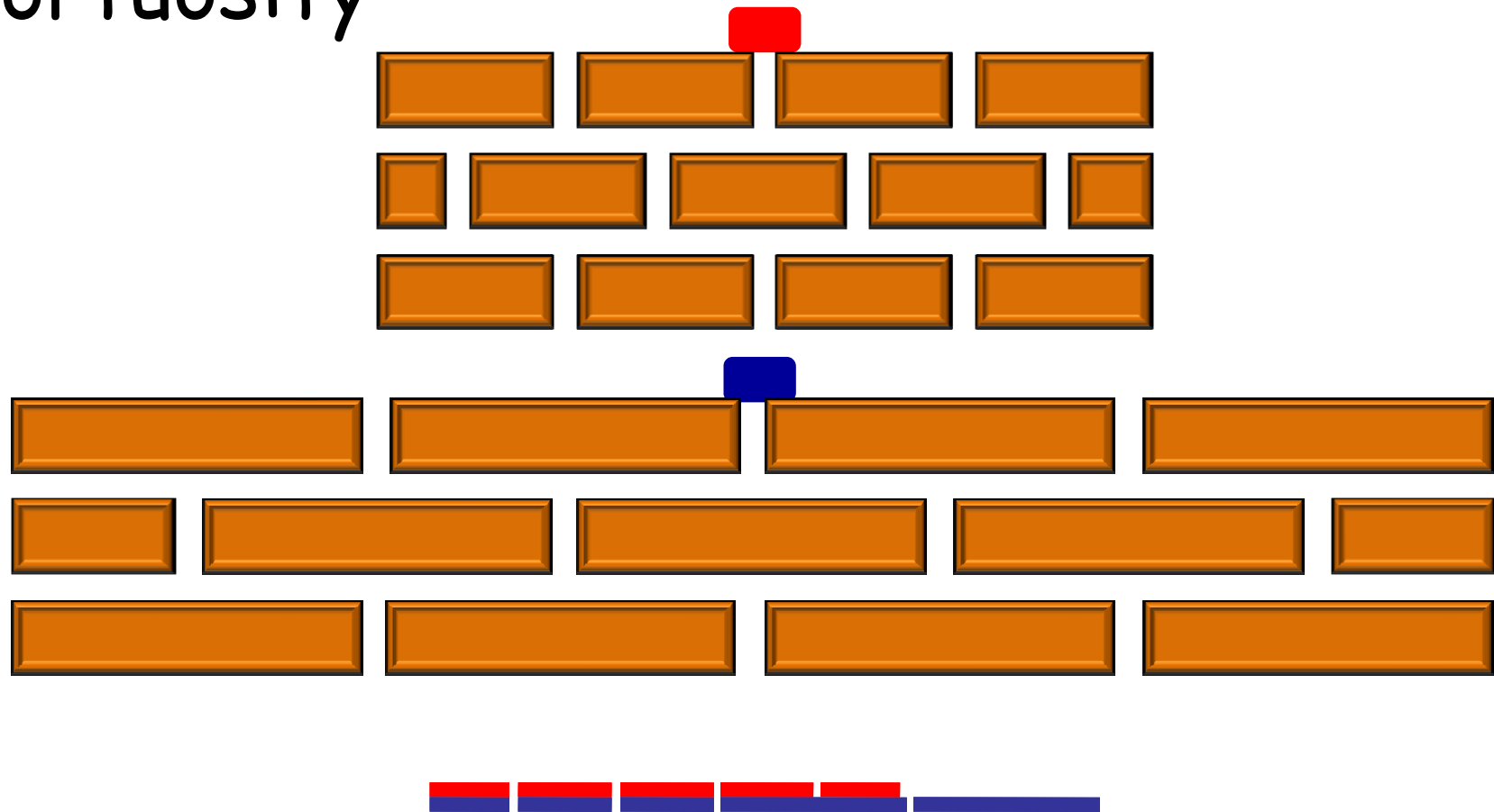
Talreja et al.



Pathlength: tortuosity ~ 12



Tortuosity



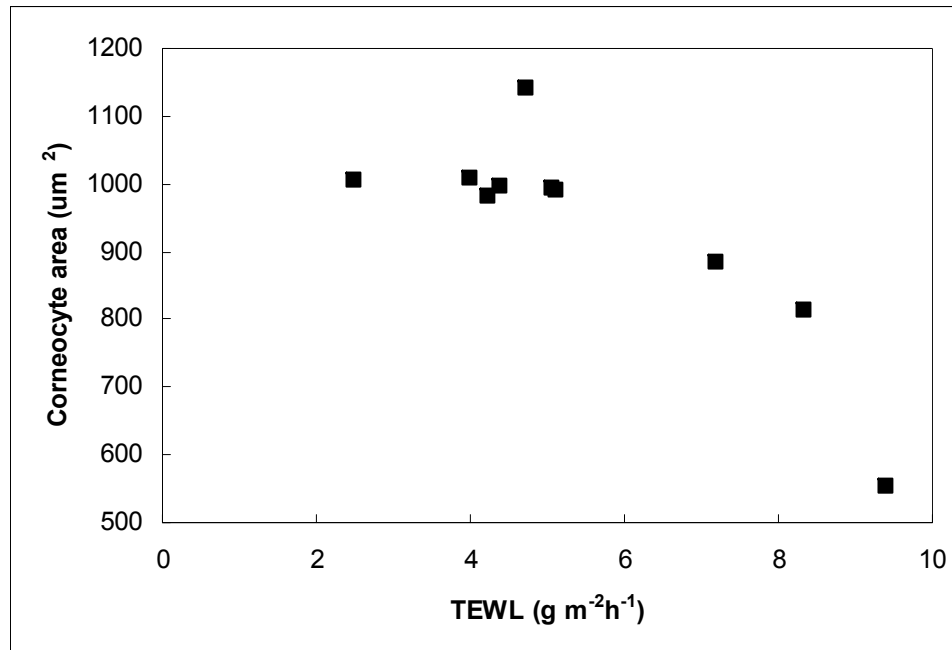
3 layers above, in reality at least
10, therefore larger effect
observed

$$\text{Pathlength} = (n + \sqrt{A} (n-1)/2)$$

Tortuosity & permeability

$$\text{TEWL} \propto K * D_{\text{water}} / \text{pathlength}$$

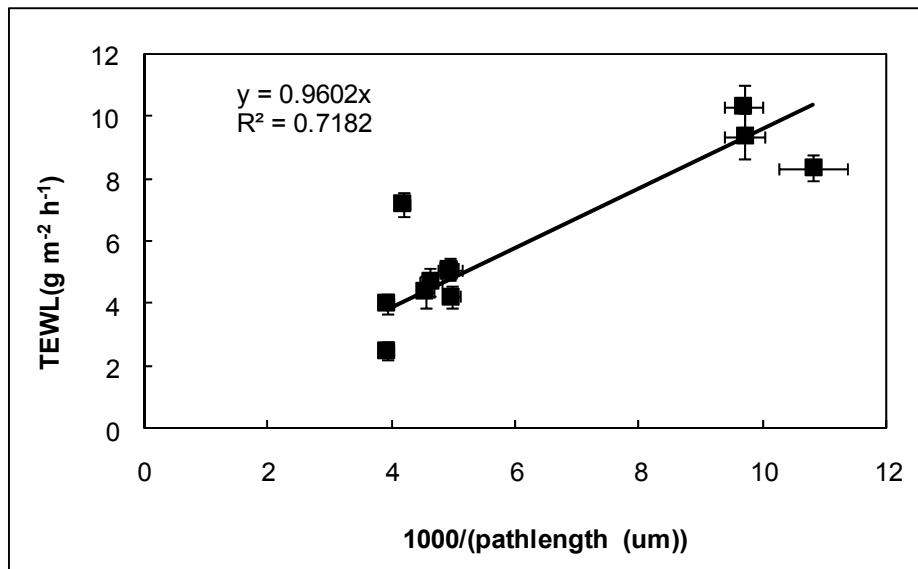
$$\text{Pathlength} = (n + \sqrt{A} (n-1)/2)$$



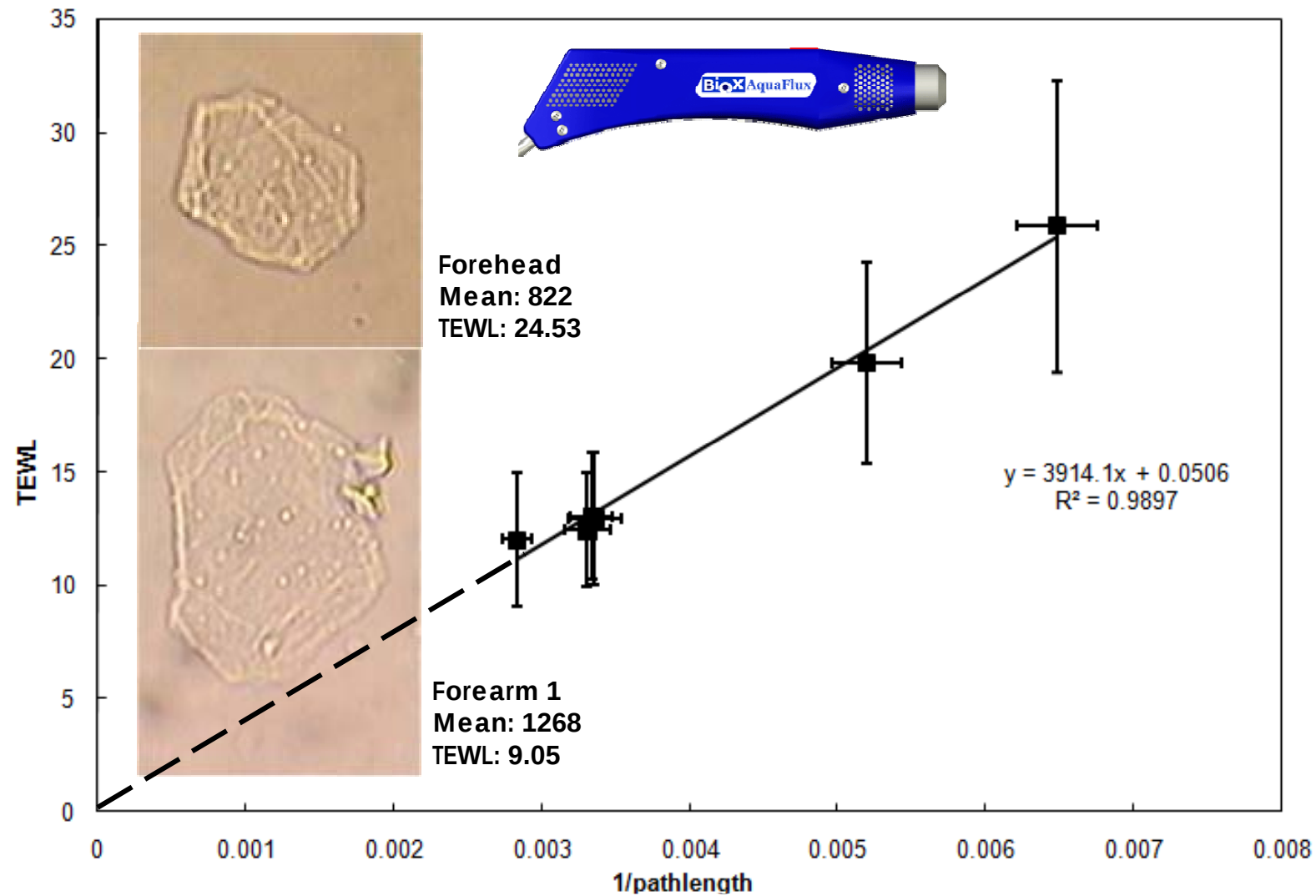
Tortuosity & permeability

$$\text{TEWL} \propto K * D_{\text{water}} / \text{pathlength}$$

$$\text{Pathlength} = (n + \sqrt{A} (n-1)/2)$$



Tortuosity & permeability: recent data



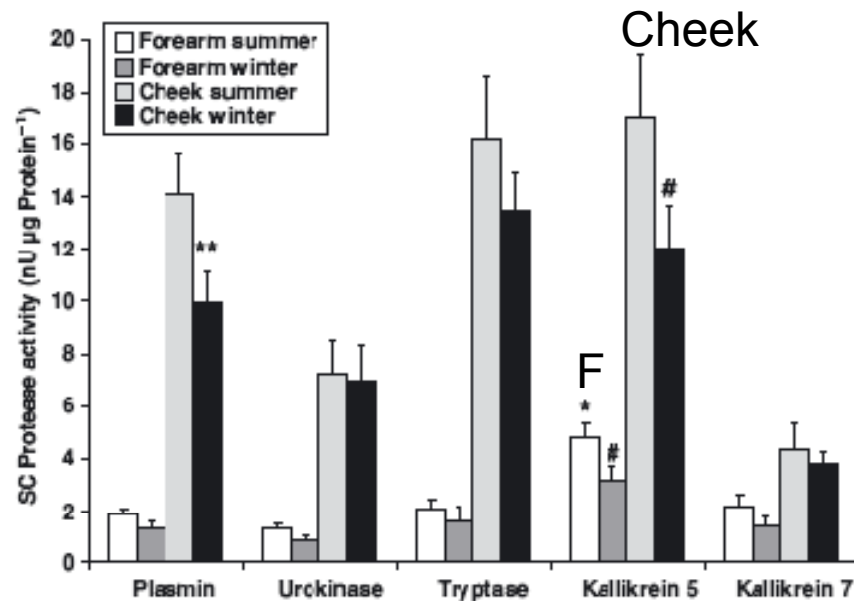
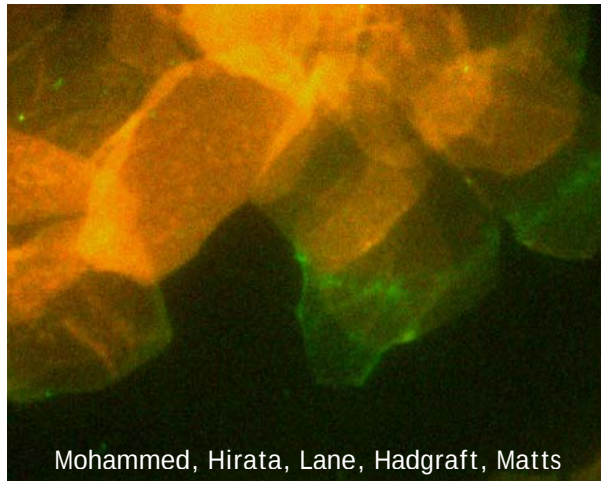
Protease activity

International Journal of Cosmetic Science, 2007, **29**, 191–200

Profiling of serine protease activities in human stratum corneum and detection of a stratum corneum tryptase-like enzyme

R. Voegeli*, A. V. Rawlings†, S. Doppler*, J. Heiland‡ and T. Schreier*

*Pentapharm Ltd, Engelgasse 109, 4002 Basel, Switzerland, †AVR Consulting Ltd, 26 Shavington Way, Kingsmead, Northwich, Cheshire CW9 8FH, U.K. and ‡Heiland Electronic GmbH, Schulstrasse 8, 35579 Wetzlar, Germany



Poor bioavailability

REGIONAL VARIATION IN PERCUTANEOUS PENETRATION OF ¹⁴C CORTISOL IN MAN*

ROBERT J. FELDMANN, M.D. AND HOWARD I. MAIBACH, M.D.

- e.g. 0.25% hydrocortisone alcohol applied to forearm (unoccluded) gives 1.04% excreted
- corresponds to 1.7% absorbed
- equivalent to 0.2 mg/cm²
- Over total body only 4 mg absorbed

Where does it go?
And can we improve it?

- Role of excipients
- What do they do?
- How long are they there?



Duragesic

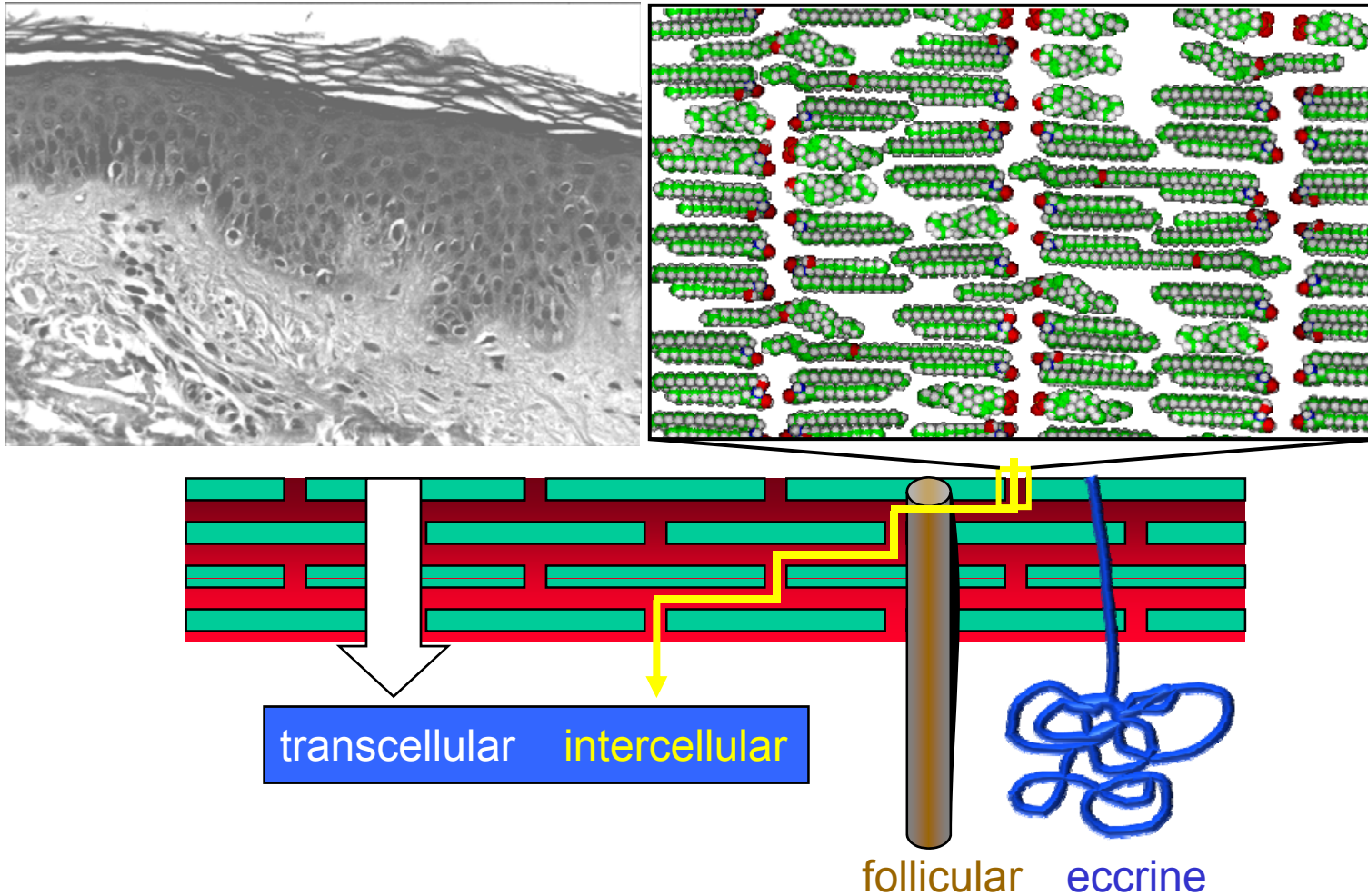
ethanol

Contact with intercellular channels

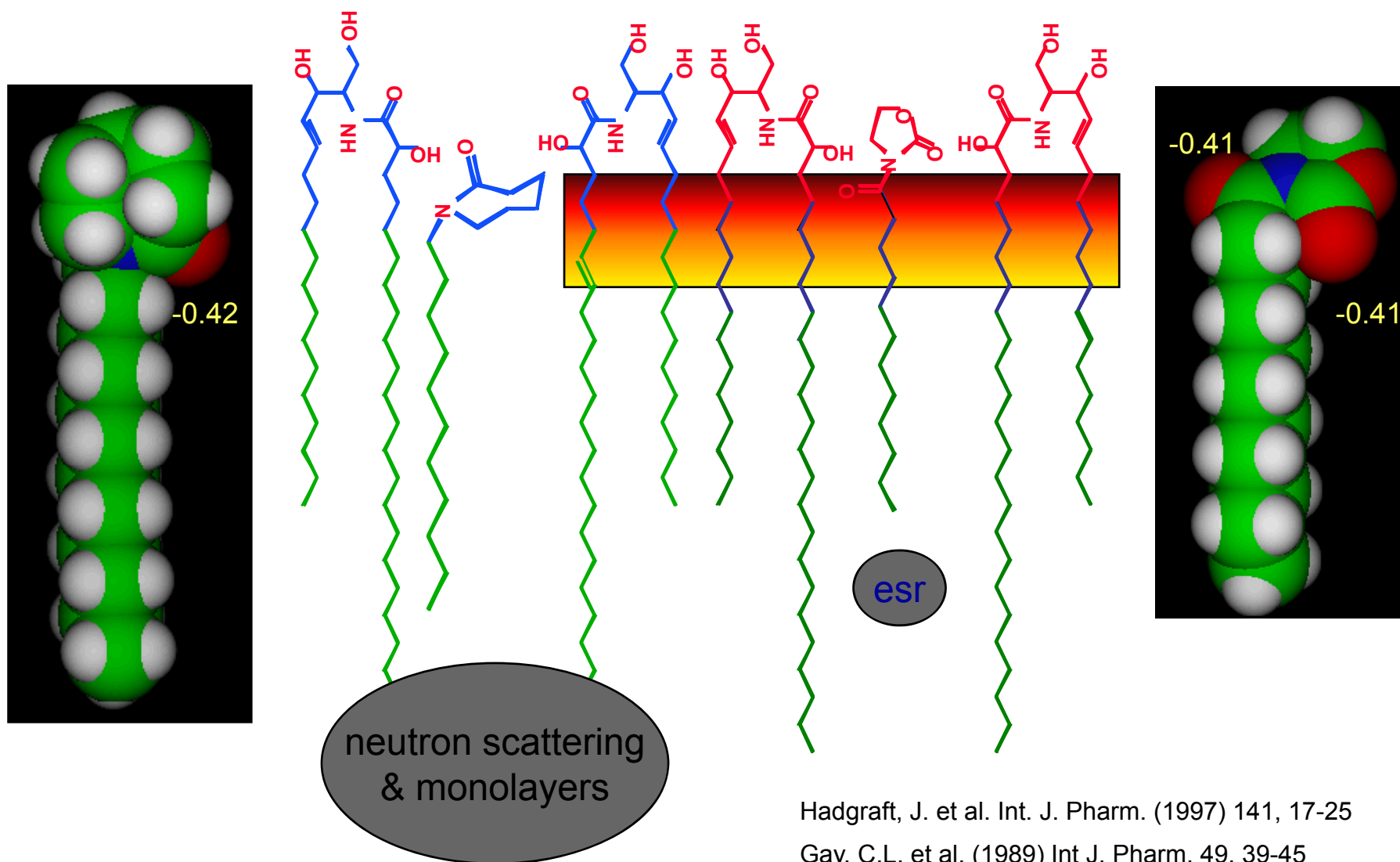


Formulation optimization

Must understand molecular processes



Molecular interpretation of effects



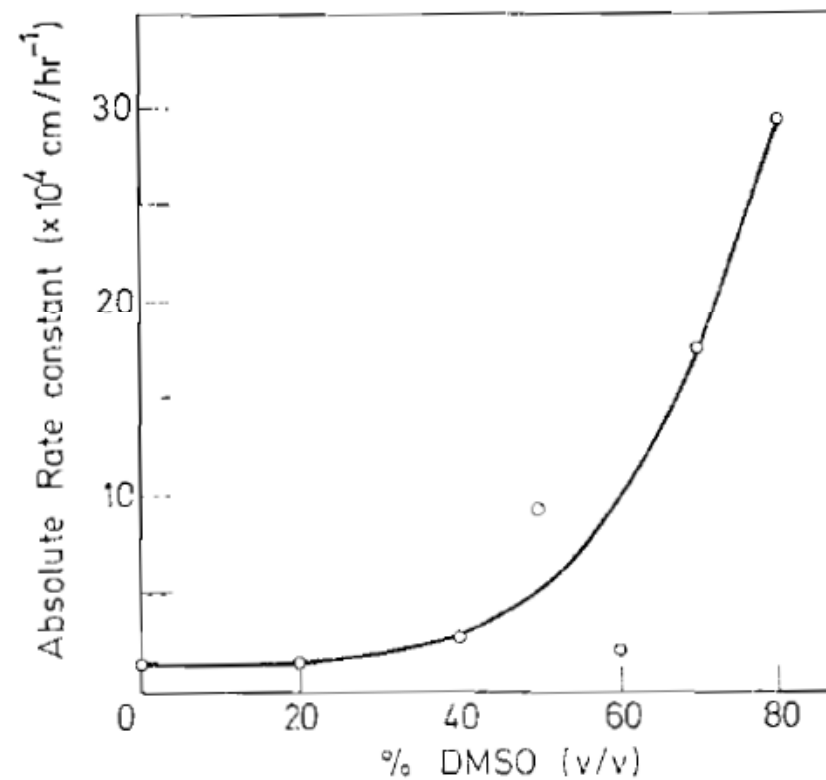
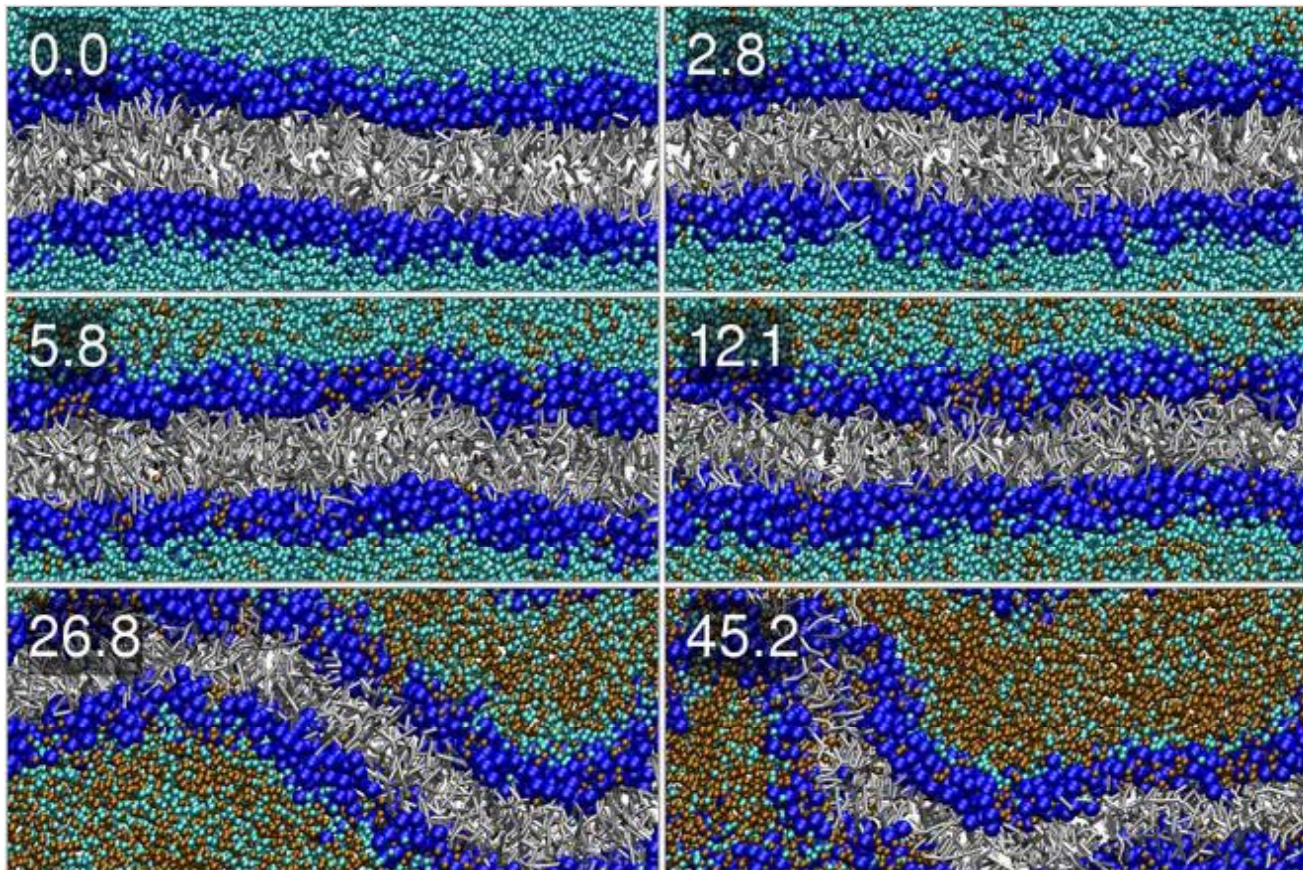


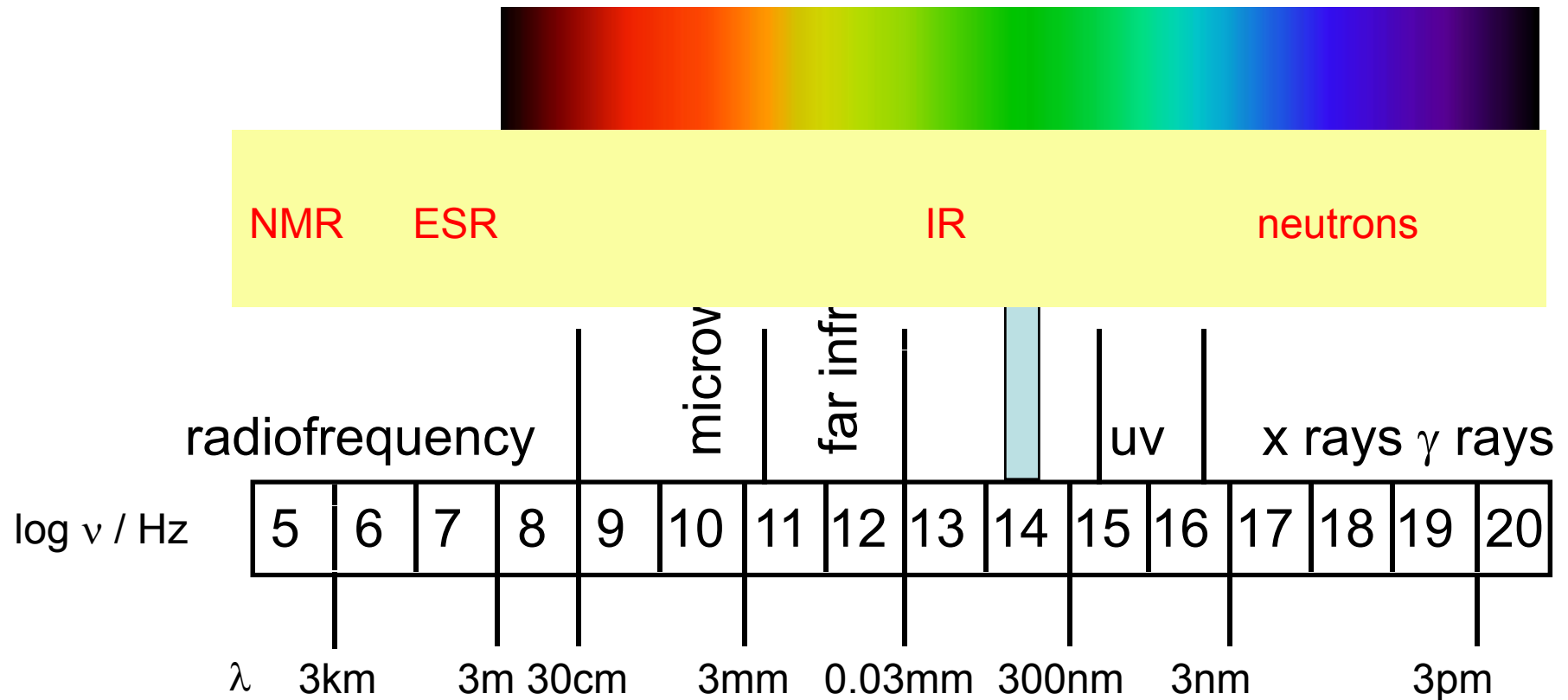
Fig. 7. Effect of DMSO concentration on percutaneous absorption of picrate ion (pH 7. $\pm 0.05^\circ \text{C}$) (ELFBAUM and LADEN, 1968a). Reprinted from J. Soc. Cosmetic Chemists **19**.
Reproduced with permission of the copyright owner

Model lipid bilayer DMSO



DPPC bilayers with different concentrations of DMSO. Water molecules are cyan, DMSO in brown, DPPC headgroup and glycerol backbone segments in blue, and hydrocarbon segments in light grey.

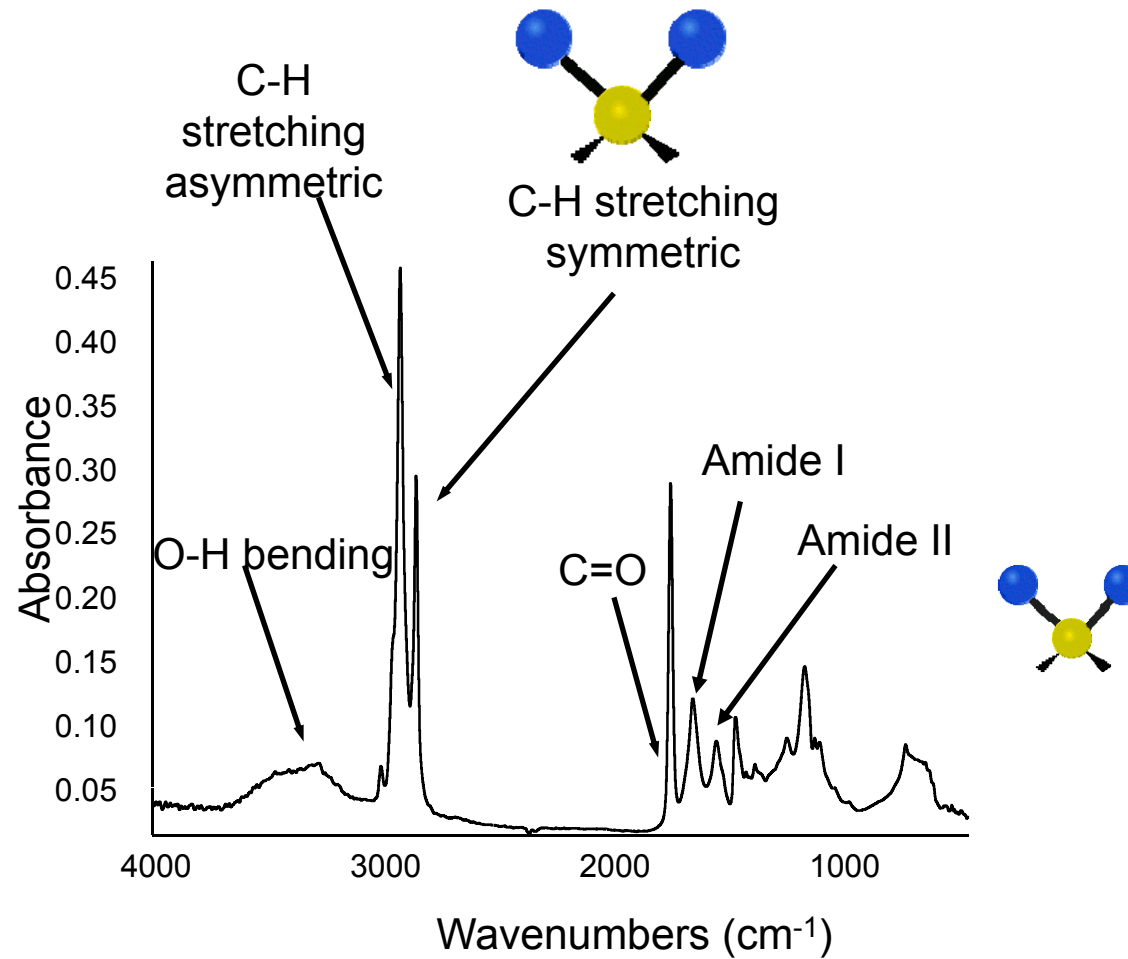
Biophysical interrogation of skin



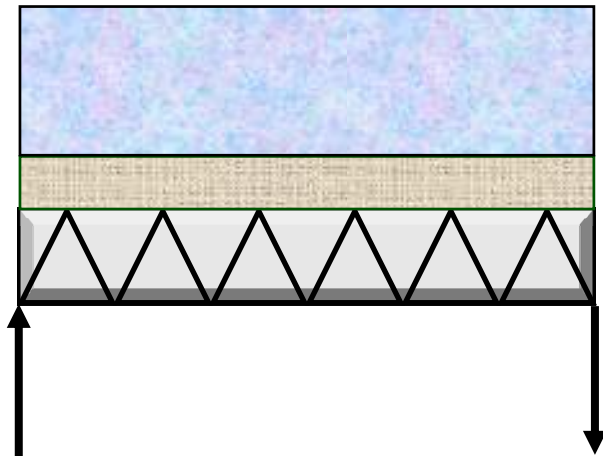
Biophysical techniques: Fourier Transform Infra Red



Skin spectrum: in vitro and in vivo



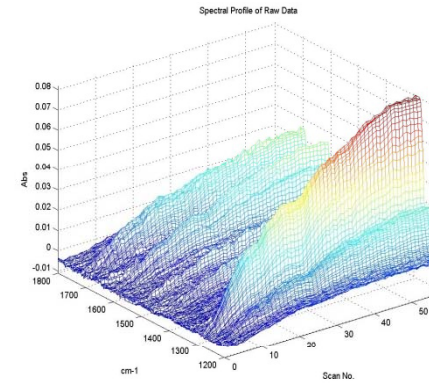
Deconvolving data



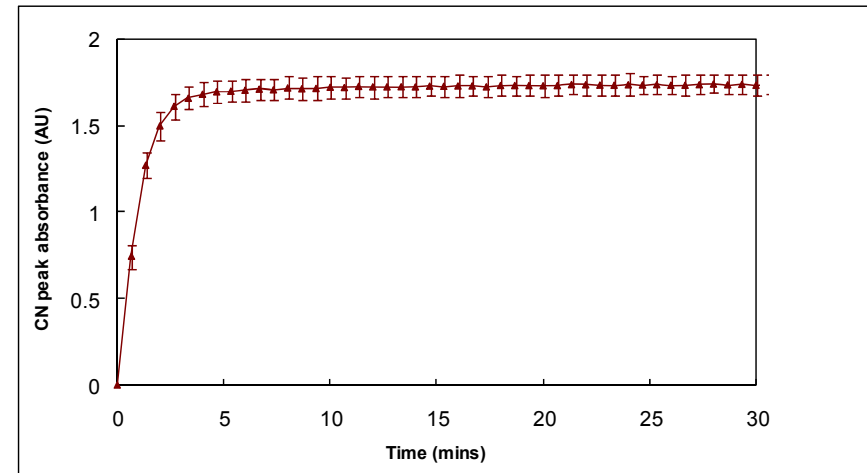
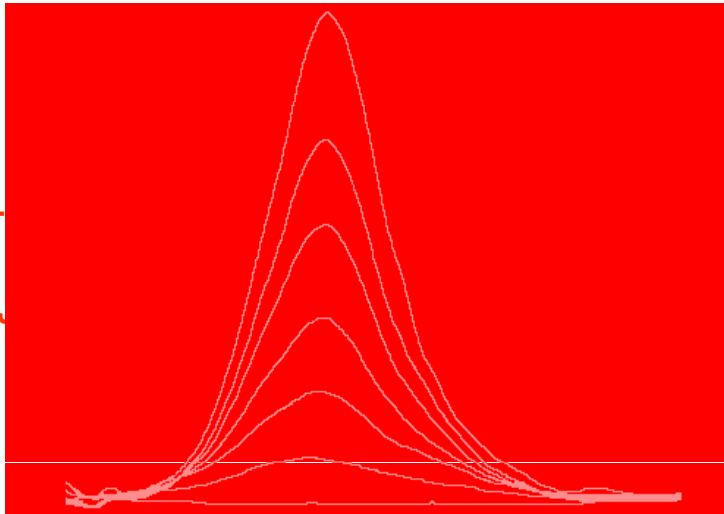
saturated
solution

membrane

Zn Se crystal

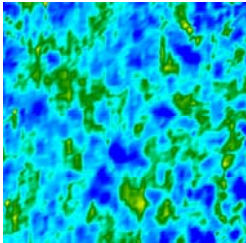
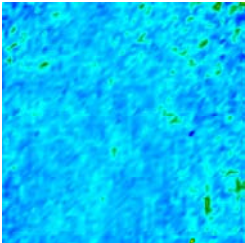
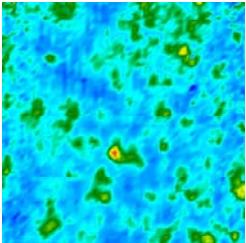
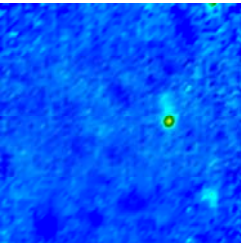
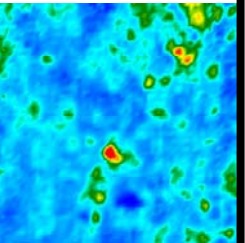
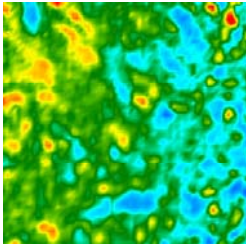
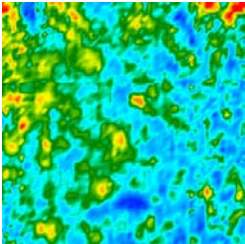
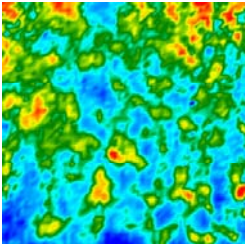
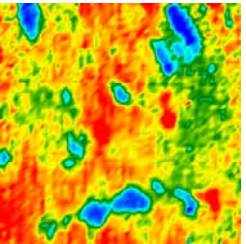
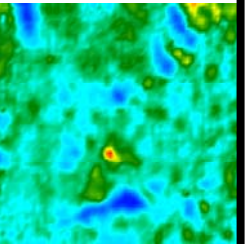


methyl & propyl paraben



Harrison, J. et al. Pharm. Res. (1996) 13, 542-546
Russeau W. et al. Int. J. Pharm. In press

Imaging skin and diffusion

	BN			Ethanol band 1045 cm ⁻¹	PEG band 1100 cm ⁻¹
	Band at 1280 cm ⁻¹	Band at 1110 cm ⁻¹	Band at 1020 cm ⁻¹		
Before diffusion	 [-0.2;0.08]	 [-0.05;0.15]	 [-0.1;0.4]	 [0.0;0.35]	 [0.0;6.0]
After diffusion	 [0.0;0.8]	 [0.0;0.25]	 [-0.1;0.4]	 [0.0;0.35]	 [0.0;15.0]

FTIR, TEWL & nicotinate permeation: sex, age, ethnicity, temporal

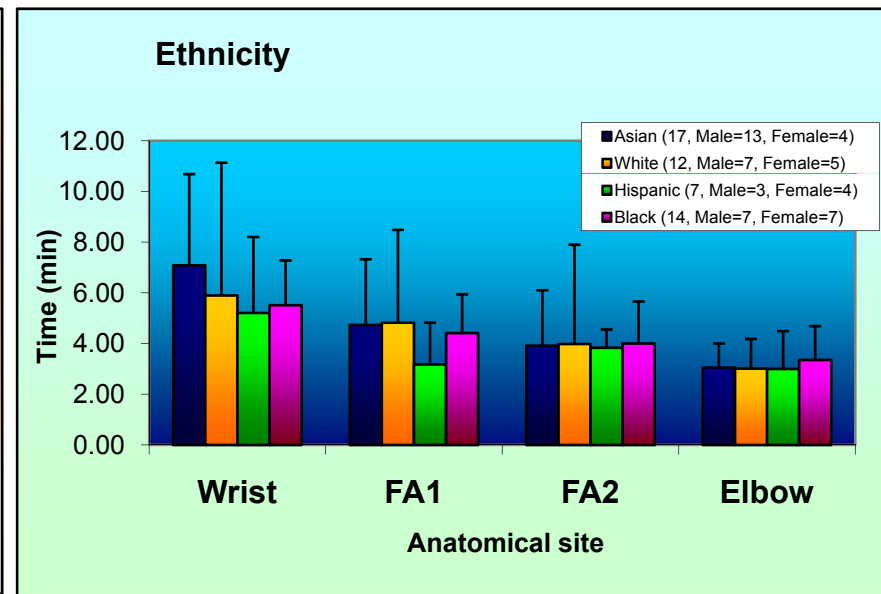
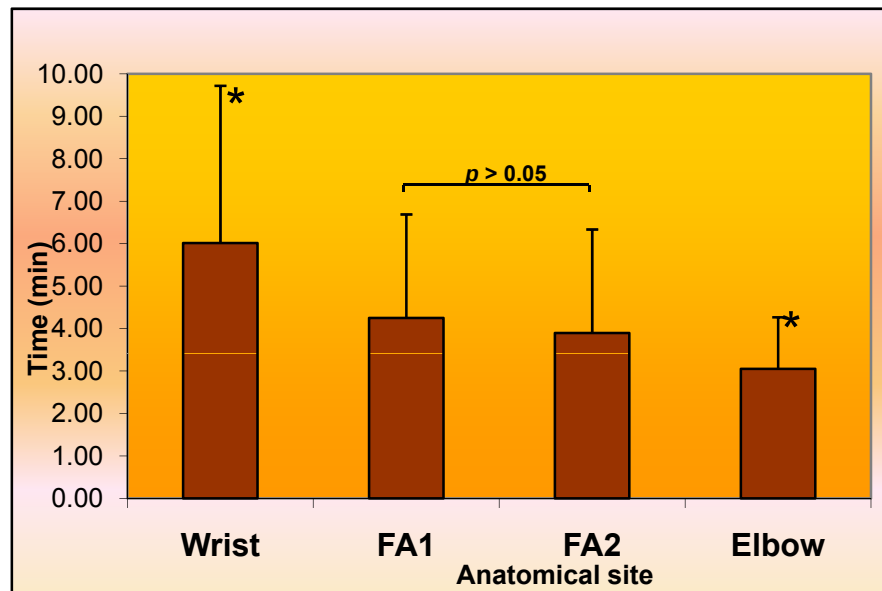
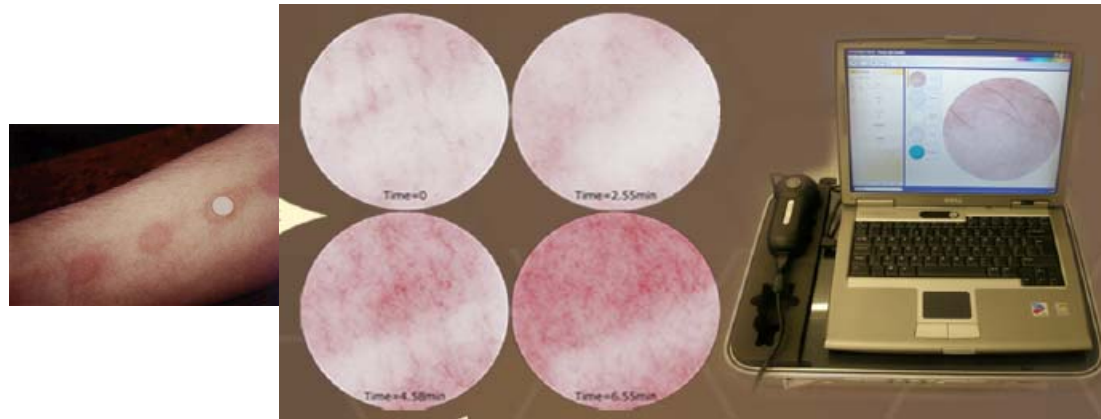
Asymmetric

- Males mean 2917.81 cm^{-1} , S.D 1.67 cm^{-1}
- Females mean value 2917.28 cm^{-1} , 1.59 cm^{-1}
- No significant difference, Positively skewed normal distributions

Symmetric

- Males mean 2849.85 cm^{-1} S.D 0.89 cm^{-1}
- Females mean 2849.74 cm^{-1} , S.D. 0.69 cm^{-1}
- No significant difference, positively skewed normal distributions

Nicotinate permeation: site, ethnicity,

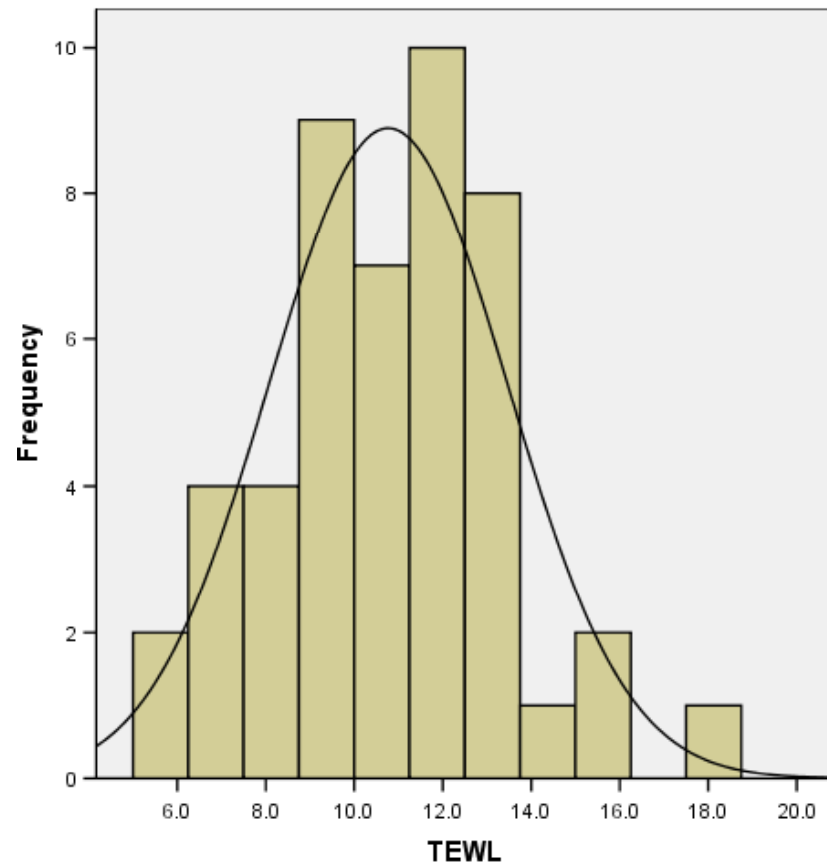


TEWL results - gender

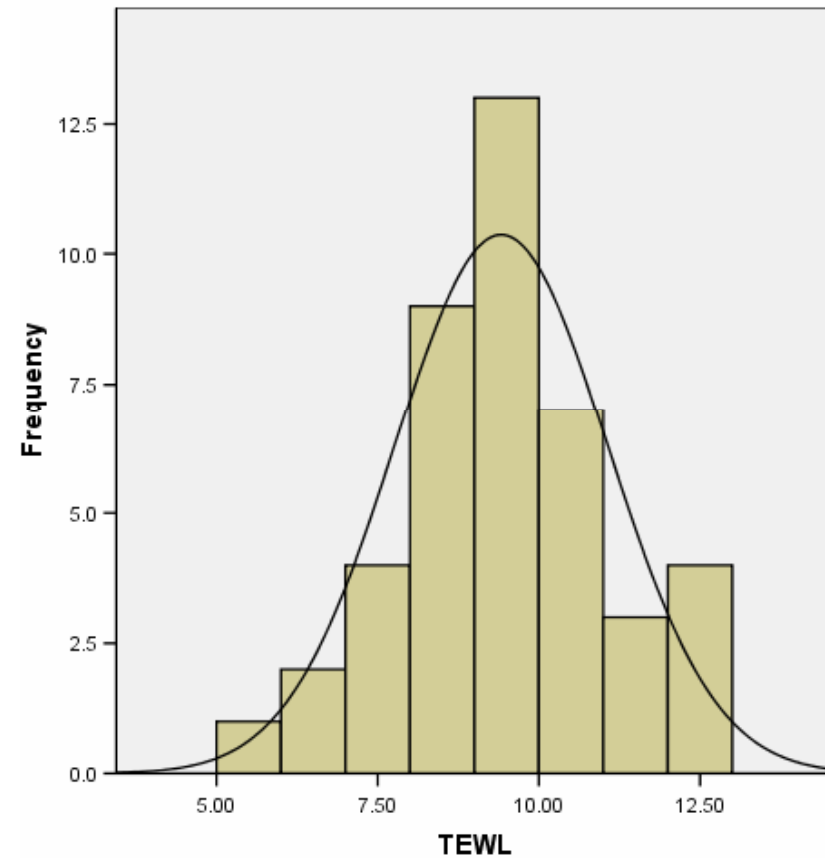
Difference is significant



a) Male distribution curve



b) Female distribution curve



Diseased skin

382

THE VETERINARY RECORD
APRIL 1st, 1972

Some Observations on the Diseases of *Brunus edwardii* (Species nova)

D. K. BLACKMORE, B.Sc., Ph.D., F.R.C.V.S., D. G. OWEN, M.Sc.
and C. M. YOUNG, M.A., Vet.M.B., M.R.C.V.S.

Woodcote Avenue, Wallington, Surrey

Vet. Rec. (1972). 90. 382-385

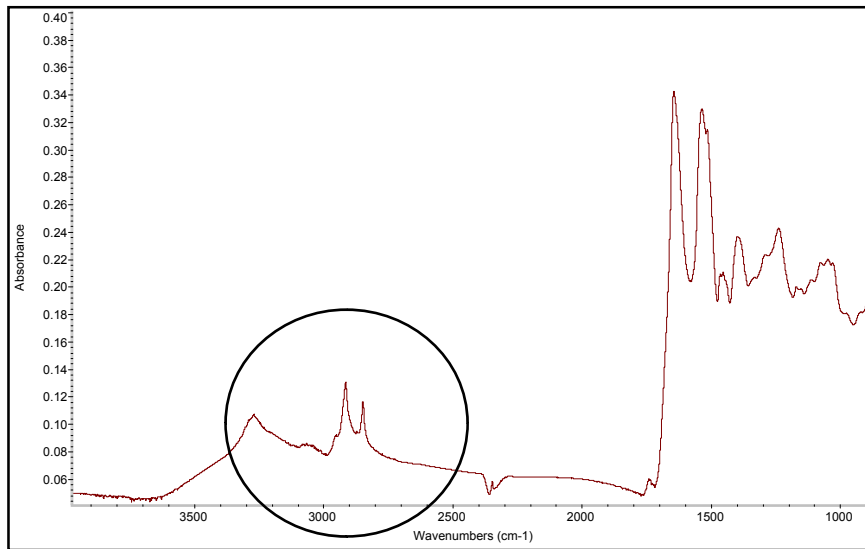
383



Case 3.—Alopecia, discoloration (very loved).

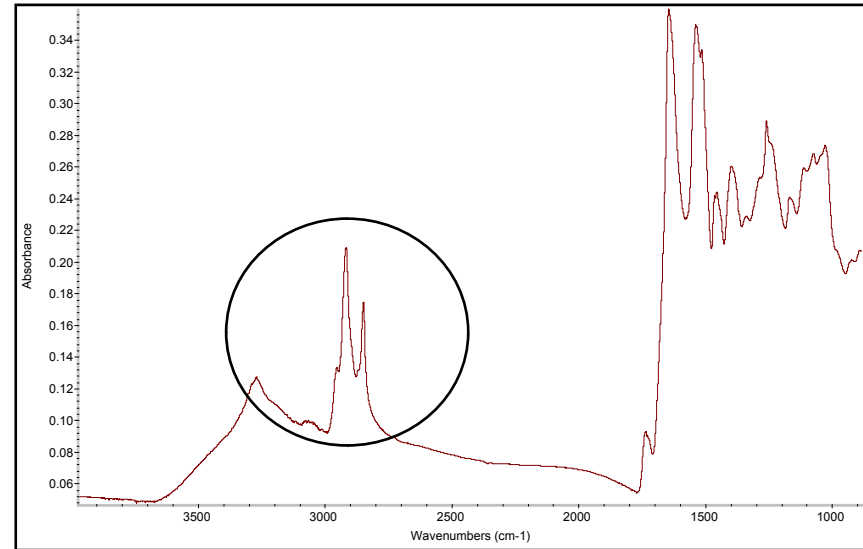
I.T. Degim, J.Hadgraft, E. Houghton, P. Teale
In vitro percutaneous absorption of fusidic acid and betamethasone 17-valerate across canine skin.
J. Small. Anim. Pract. 40 (1999) 515-518

Psoriasis UVA treatment



Pre-treatment
Peak areas

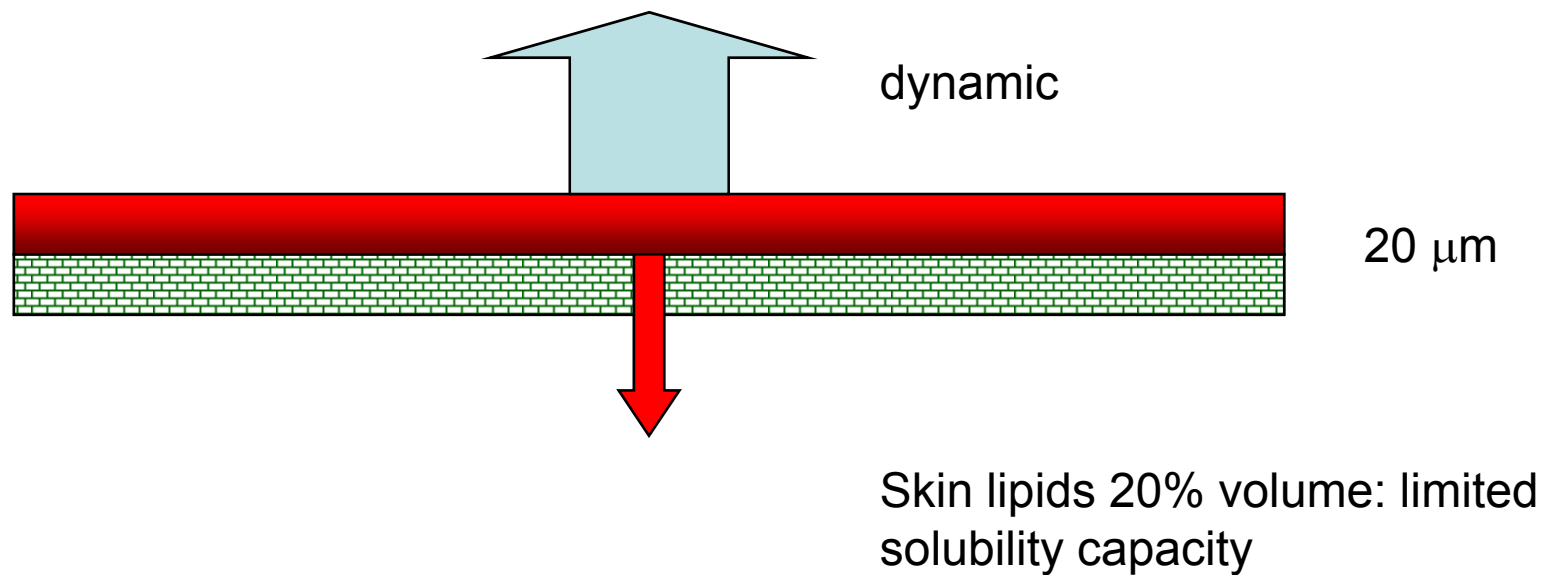
- C-H asymmetric = 0.07
- C-H symmetric = 0.03



Post-treatment
Peak areas

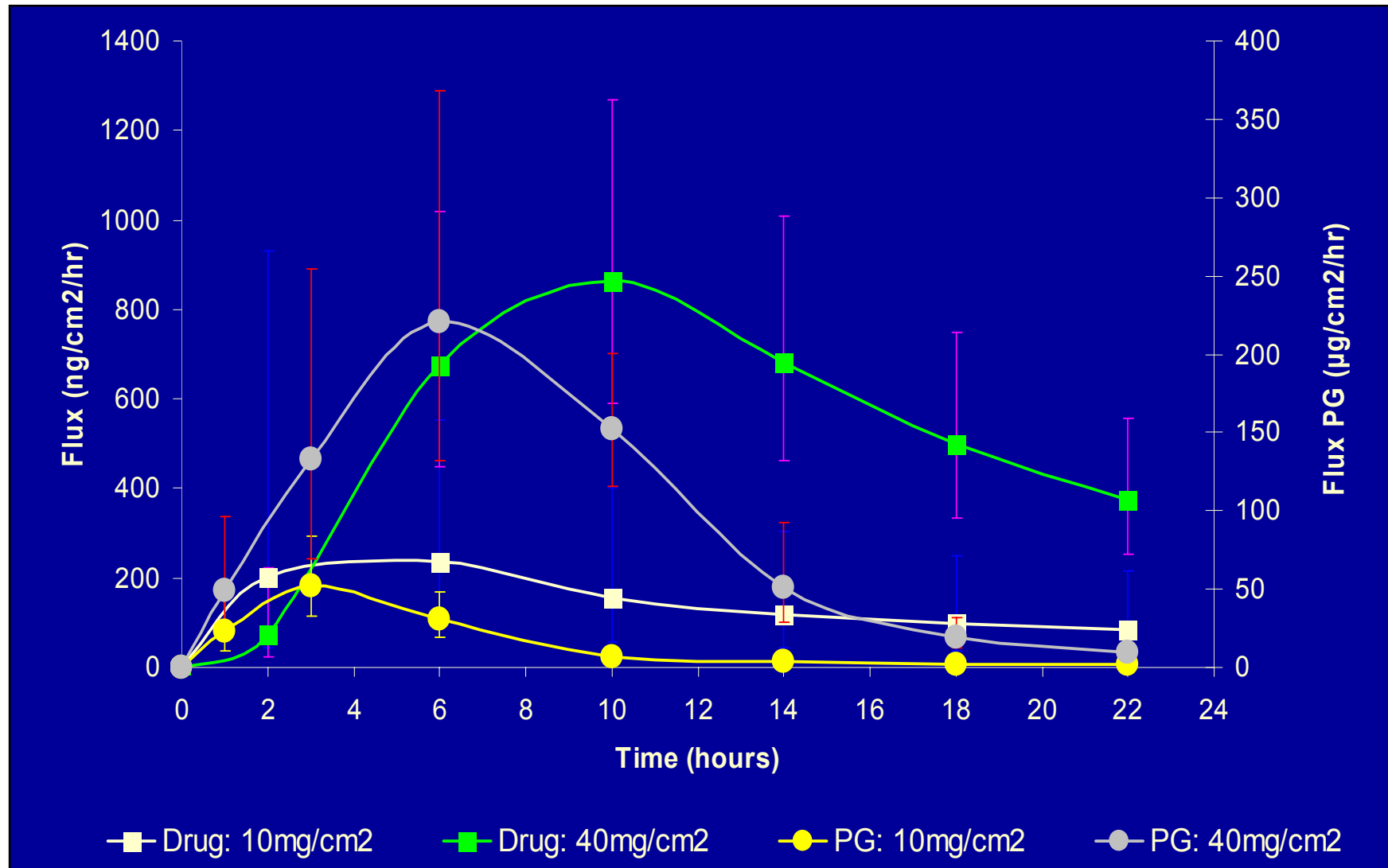
- C-H asymmetric = 0.18
- C-H symmetric = 0.07

Clinical dosing: 2 mg/cm^2



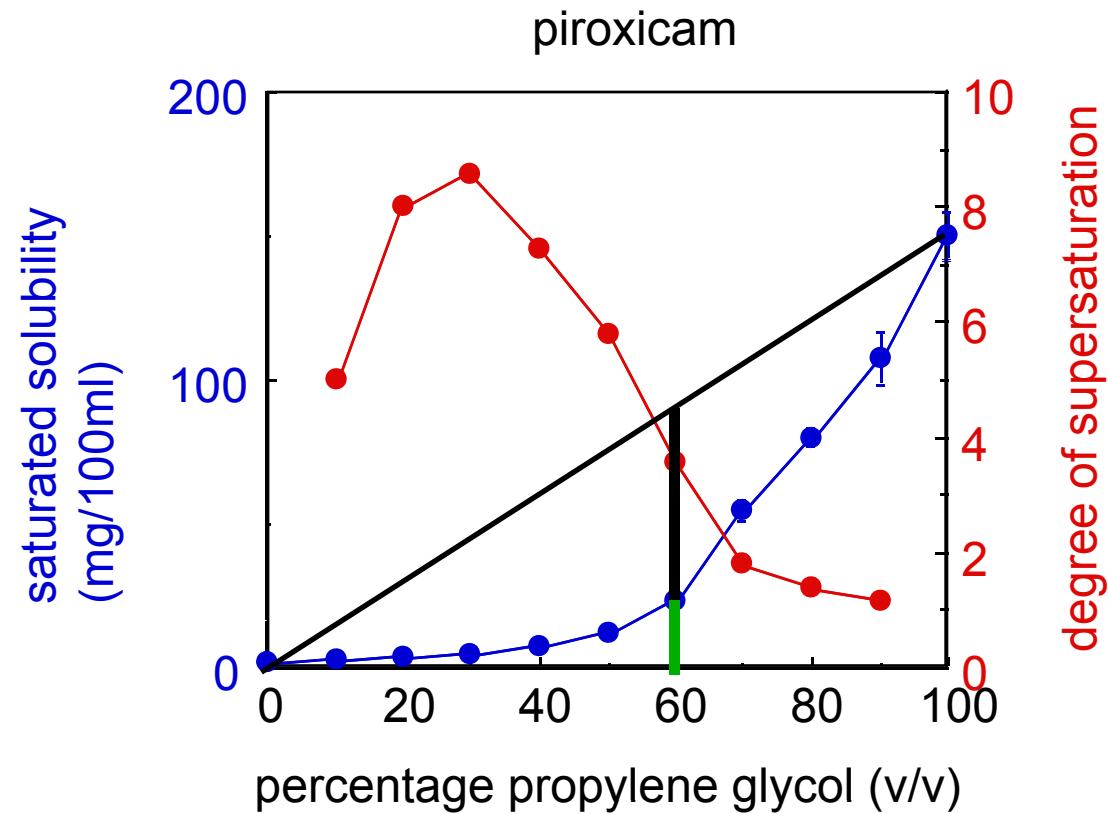
In vitro experiments difficult to mimic dose

Permeation of propylene glycol and drug: effect of the amount of gel applied



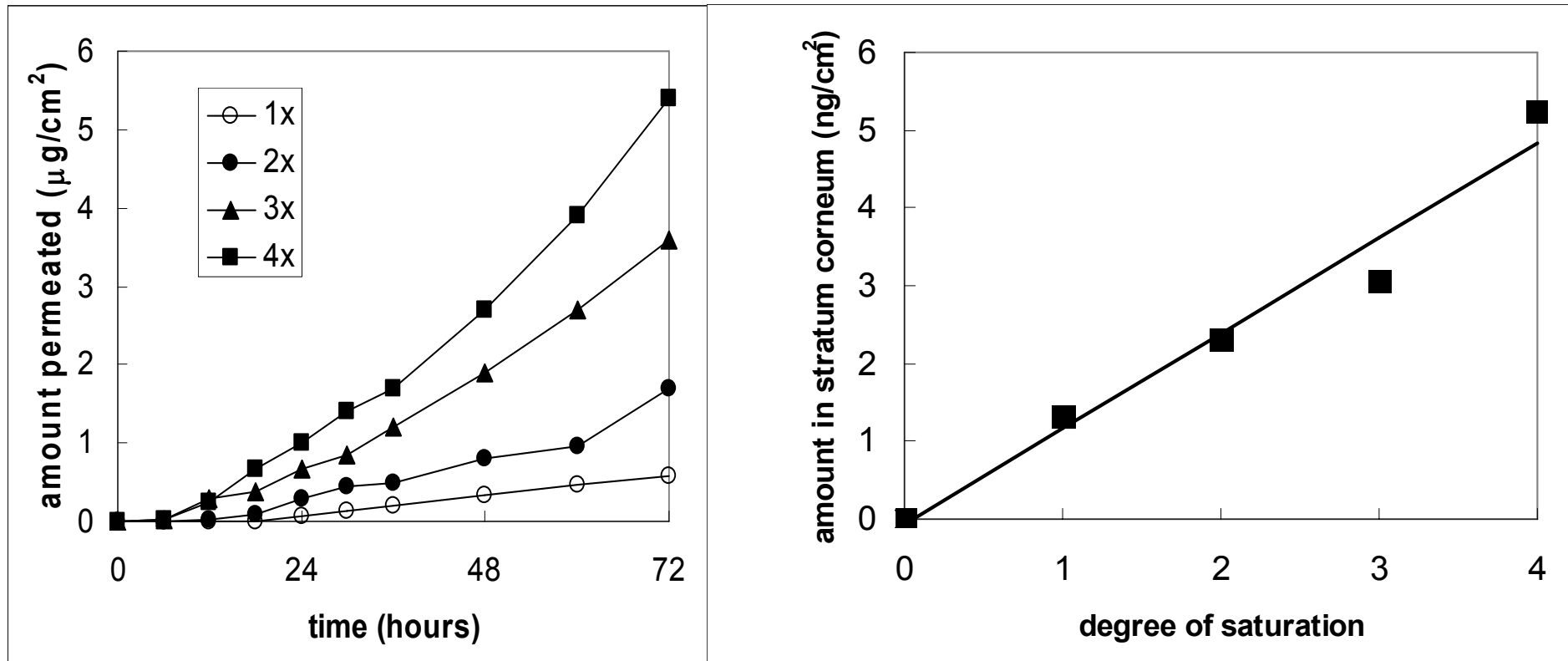
Supersaturation

mixed solvent systems
solvent evaporation



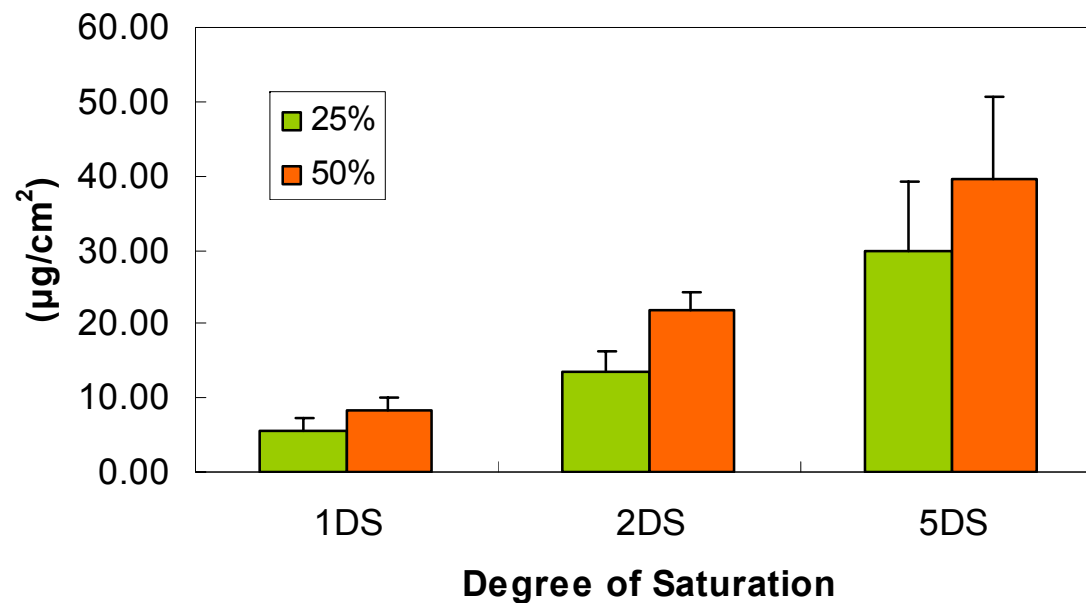
In vitro penetration human skin

infinite dose

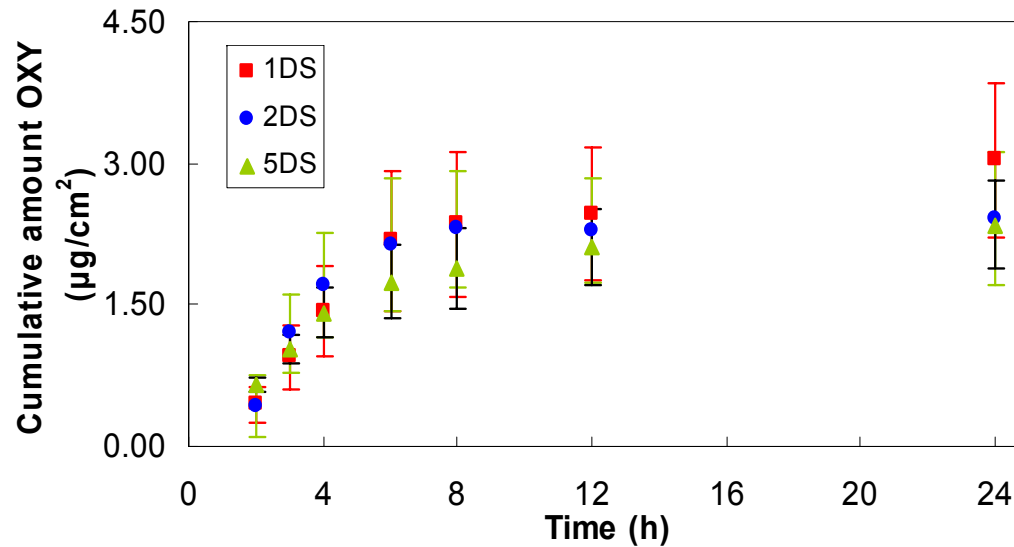


Pellett, M. A. et al. Int. J. Pharm. (1994) 111, 1-6
Pellett, M. A. et al. Int. J. Pharm. (1997) 151,91-98

Extraction OXY (stratum corneum) as function of DS from *PG* finite dose

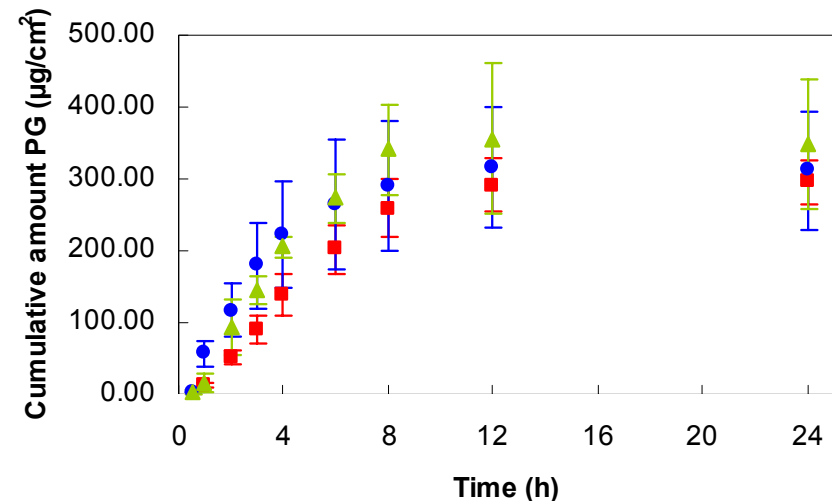


Permeation of OXY and PG through skin as function of DS finite dose



OXY crystallises in and on (~80%) the skin

Drug crystallised in skin is not available



Santos, Watkinson et al

Dimethylsulfoxide (DMSO)

Induction of a Steroid Reservoir in Human Skin

R. B. STOUGHTON, MD, CLEVELAND

TABLE 5.—*Percent Retained in Horny Layer After Single Application Followed by 1 Standard Soap and Water Wash and Once Per Day Routine Bathing (Average of 3 Subjects for Each Steroid)*

	Day 0	Day 2	Day 4	Day 8	Day 12	Day 16
¹⁴ C-Hydrocortisone (0.025 %)						
40 % DMSO in ethyl alcohol	36 %	16 %	6 %	3 %	1.5 %	1.1 %
10 % DMSO in ethyl alcohol	31 %	13 %	4 %	1 %	0.6 %	0.3 %
95 % ethyl alcohol	2.2 %	0 %	0 %	0 %	0 %	0 %
¹⁴ C-Fluocinolone Acetonide (0.025 %)						
40 % DMSO in ethyl alcohol	34 %	15 %	7 %	3 %	0.8 %	0.1 %
10 % DMSO in ethyl alcohol	22 %	9 %	4 %	1 %	0.2 %	0 %
95 % ethyl alcohol	1.8 %	0.08 %	0 %	0 %	0 %	0 %

Label-Free Biomedical Imaging with High Sensitivity by Stimulated Raman Scattering Microscopy

Christian W. Freudiger,^{1,2*} Wei Min,^{1*} Brian G. Saar,¹ Sijia Lu,¹ Gary R. Holtom,¹ Chengwei He,³ Jason C. Tsai,⁴ Jing X. Kang,³ X. Sunney Xie^{1†}

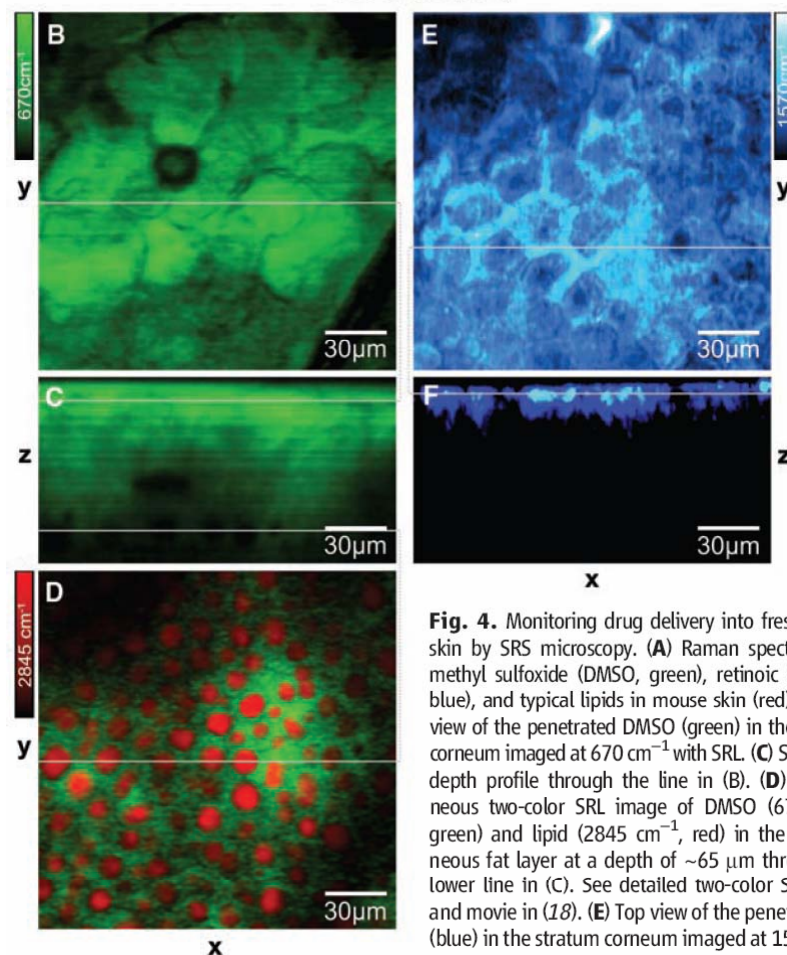


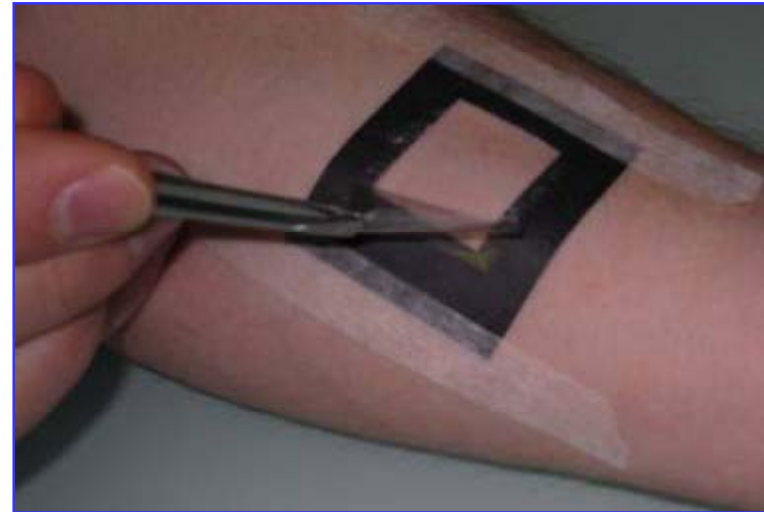
Fig. 4. Monitoring drug delivery into fresh mouse skin by SRS microscopy. (A) Raman spectra of dimethyl sulfoxide (DMSO, green), retinoic acid (RA, blue), and typical lipids in mouse skin (red). (B) Top view of the penetrated DMSO (green) in the stratum corneum imaged at 670 cm^{-1} with SRL. (C) SRL DMSO depth profile through the line in (B). (D) Simultaneous two-color SRL image of DMSO (670 cm^{-1} , green) and lipid (2845 cm^{-1} , red) in the subcutaneous fat layer at a depth of $\sim 65\text{ }\mu\text{m}$ through the lower line in (C). See detailed two-color SRL setup and movie in (18). (E) Top view of the penetrated RA (blue) in the stratum corneum imaged at 1570 cm^{-1} with SRL. (F) SRL RA depth profile through the line in (E). SRS allows label-free 3D in situ visualization of two different drug-delivery pathways into the skin.

Contact with intercellular channels



Sampling the skin: tape stripping

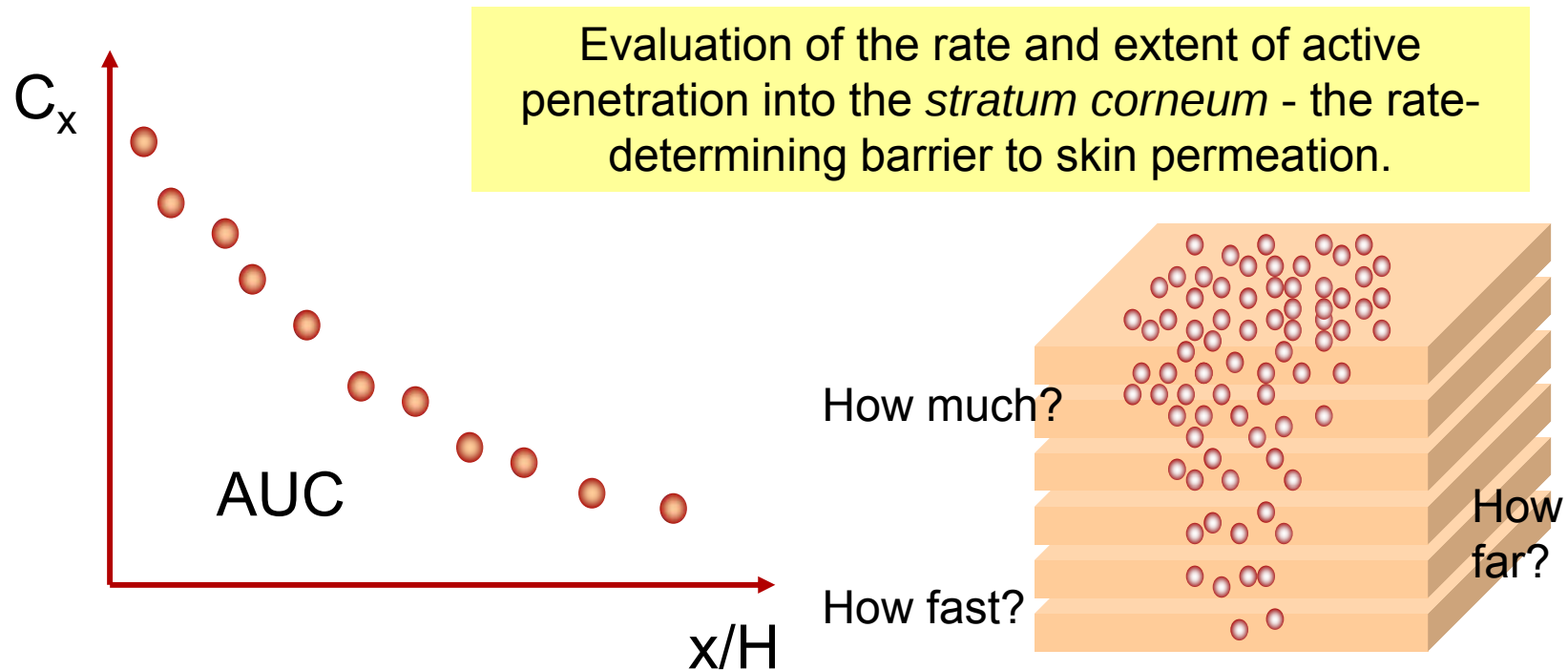
Determination of drug concentration in the stratum corneum (SC) by sequential removal of thin layers of SC at the same site with adhesive tape.



Industrial – University collaboration

Guy et al.

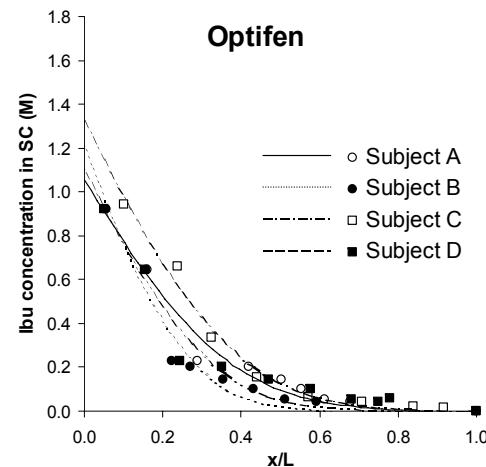
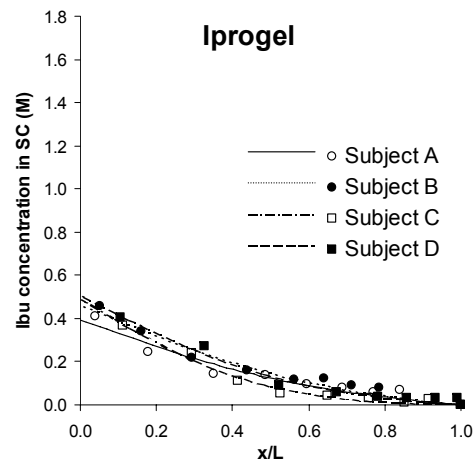
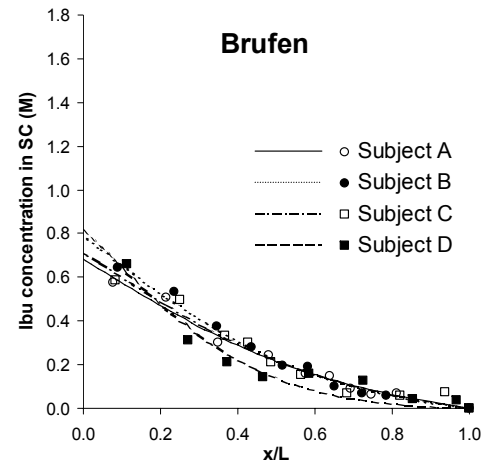
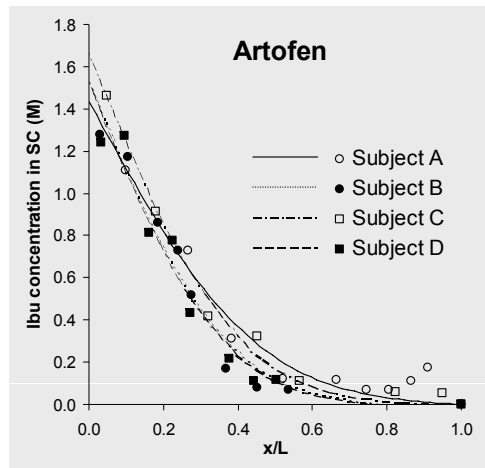
Distribution profile of active across the stratum corneum (SC)



Measure drug concentration profile as a function of position in the SC

Required: (i) amount on each strip, (ii) penetration depth into SC

In vivo concentration profiles of ibuprofen across the stratum corneum



An *In Vivo* Confocal Raman Study of the Delivery of *Trans*-Retinol to the Skin

PAUL D. A. PUDNEY,* MICKAËL MÉLOT, PETER J. CASPERS, ANDRE VAN DER POL, and GERWIN J. PUPPELS

Measurement Science Unit, Unilever R&D, Colworth Science Park, Sharnbrook, Bedford, MK44 1LQ (P.D.A.P., M.M.); and River Diagnostics B.V., Rotterdam, The Netherlands (P.J.C., A.v.D.P., G.J.P.)

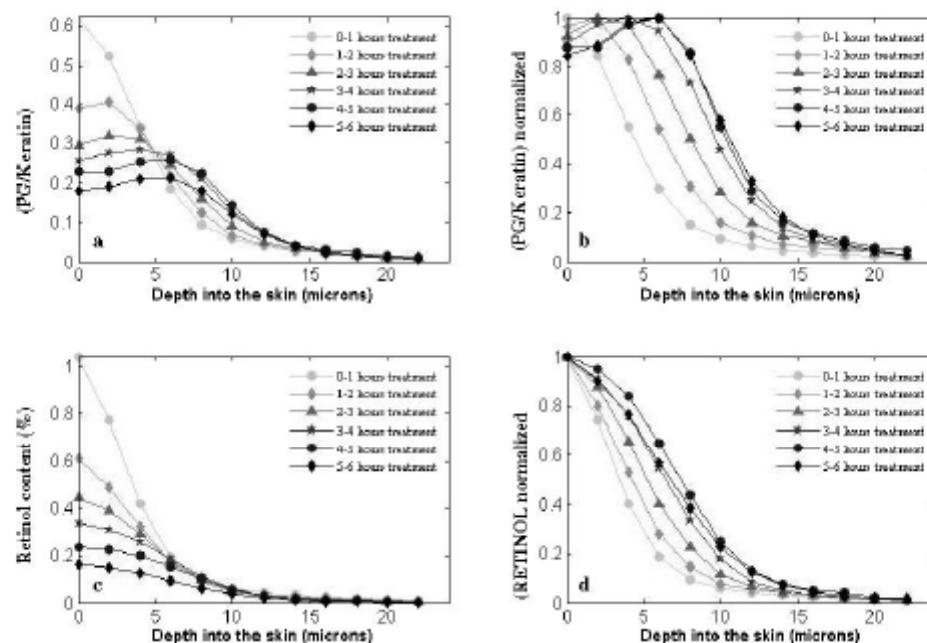
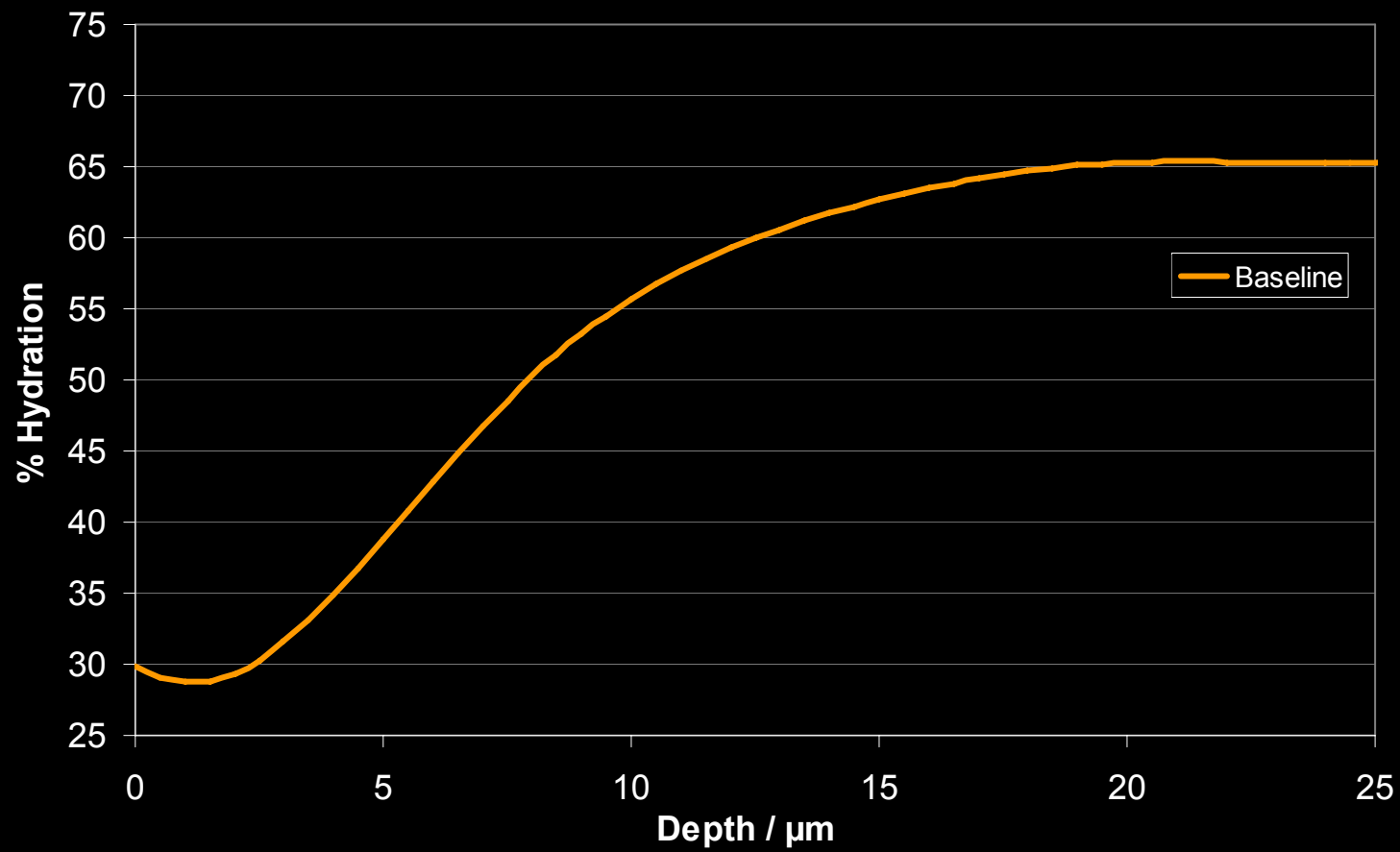
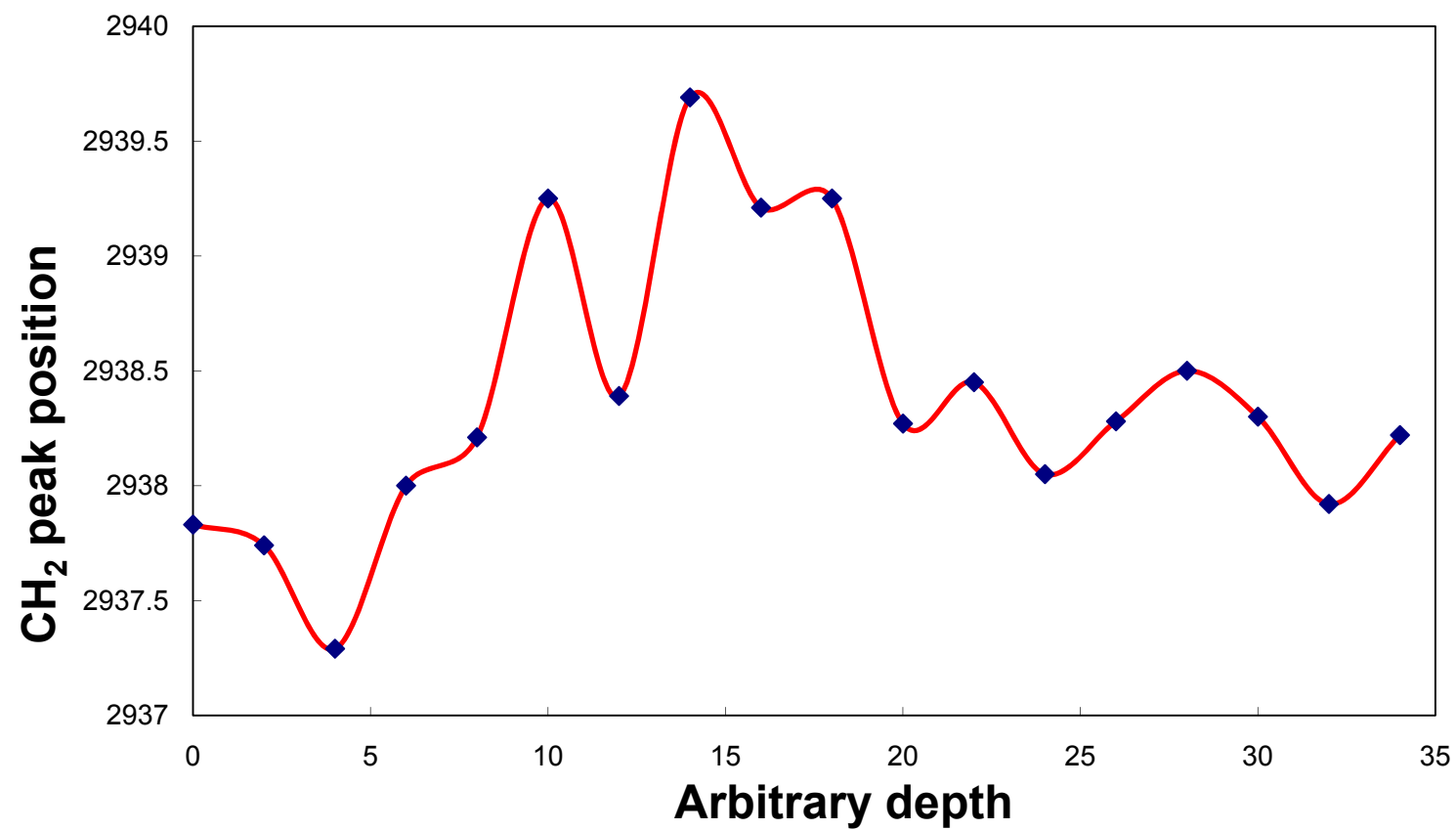
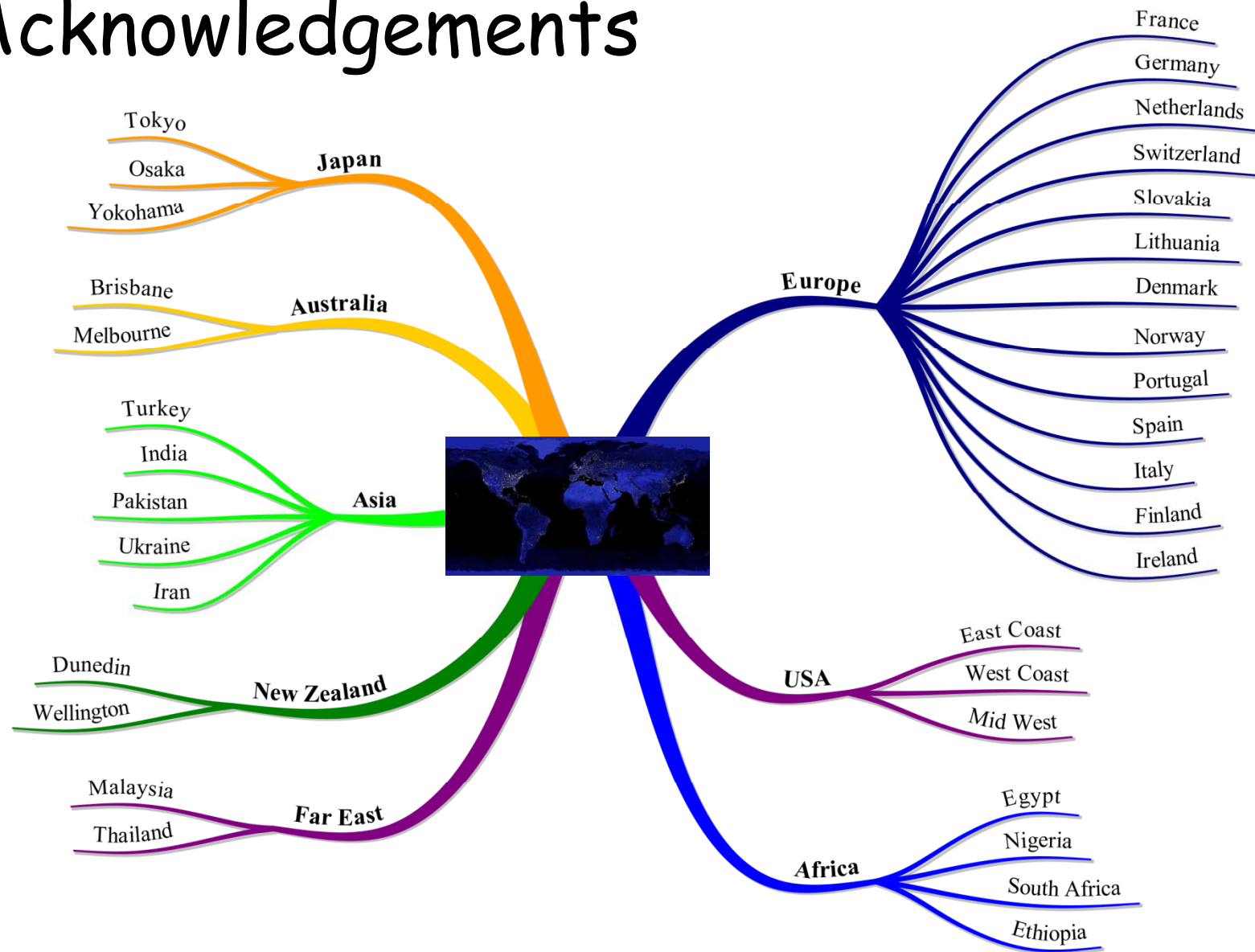


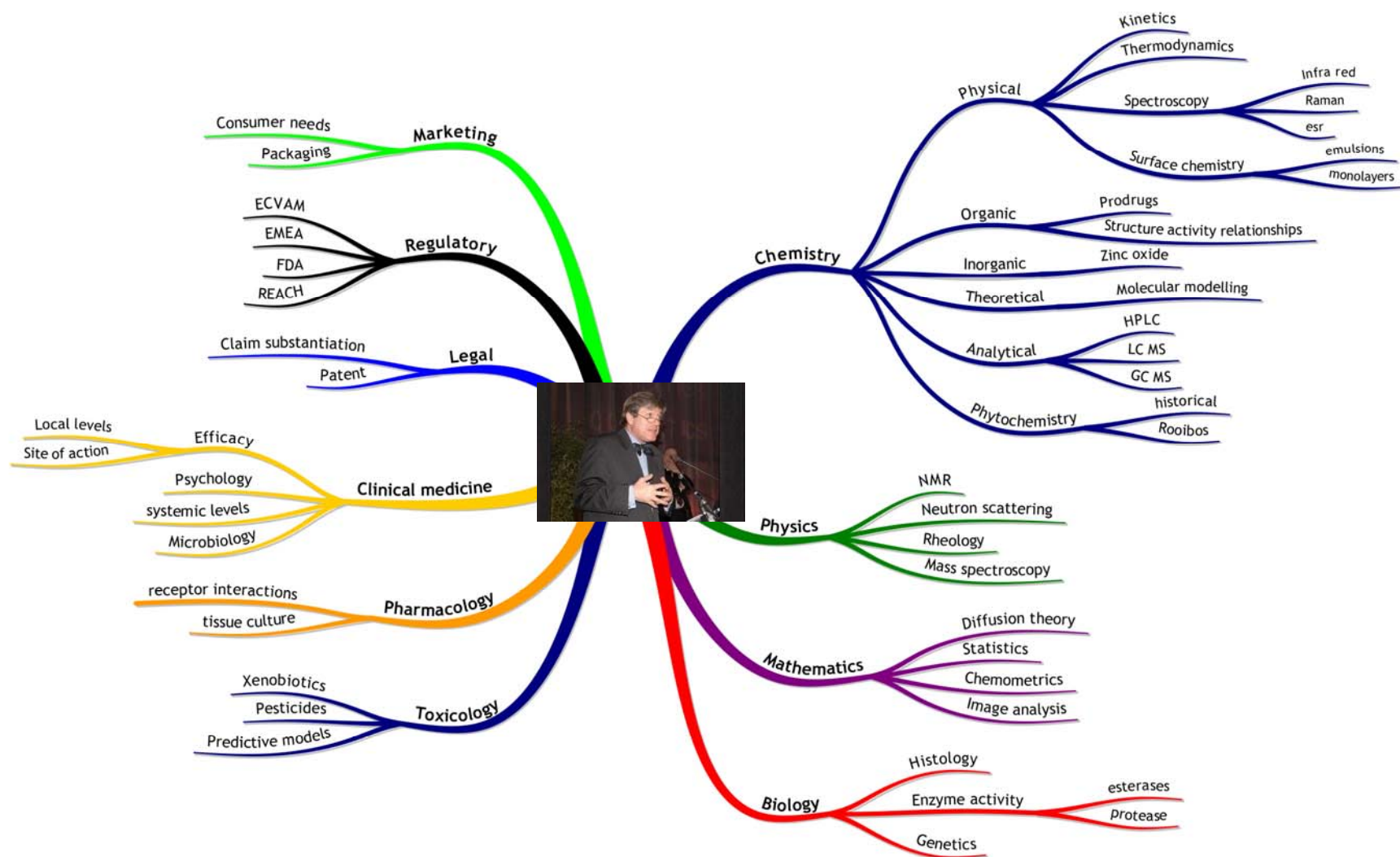
FIG. 3. Concentration profiles of PG and *trans*-retinol content (%) in the volar forearm over 6 hours. (a) Normalized and (b) absolute profiles of PG are reported. The (c) content and (d) normalized profiles of *trans*-retinol are also reported.





Acknowledgements





When I think of Jean-Paul I
see his ever smiling face and
remember his love of life



The complete skin scientist