Dermal Technology Laboratory Ltd

EFSA Guidance on Dermal Absorption (2017) : “Industry View”

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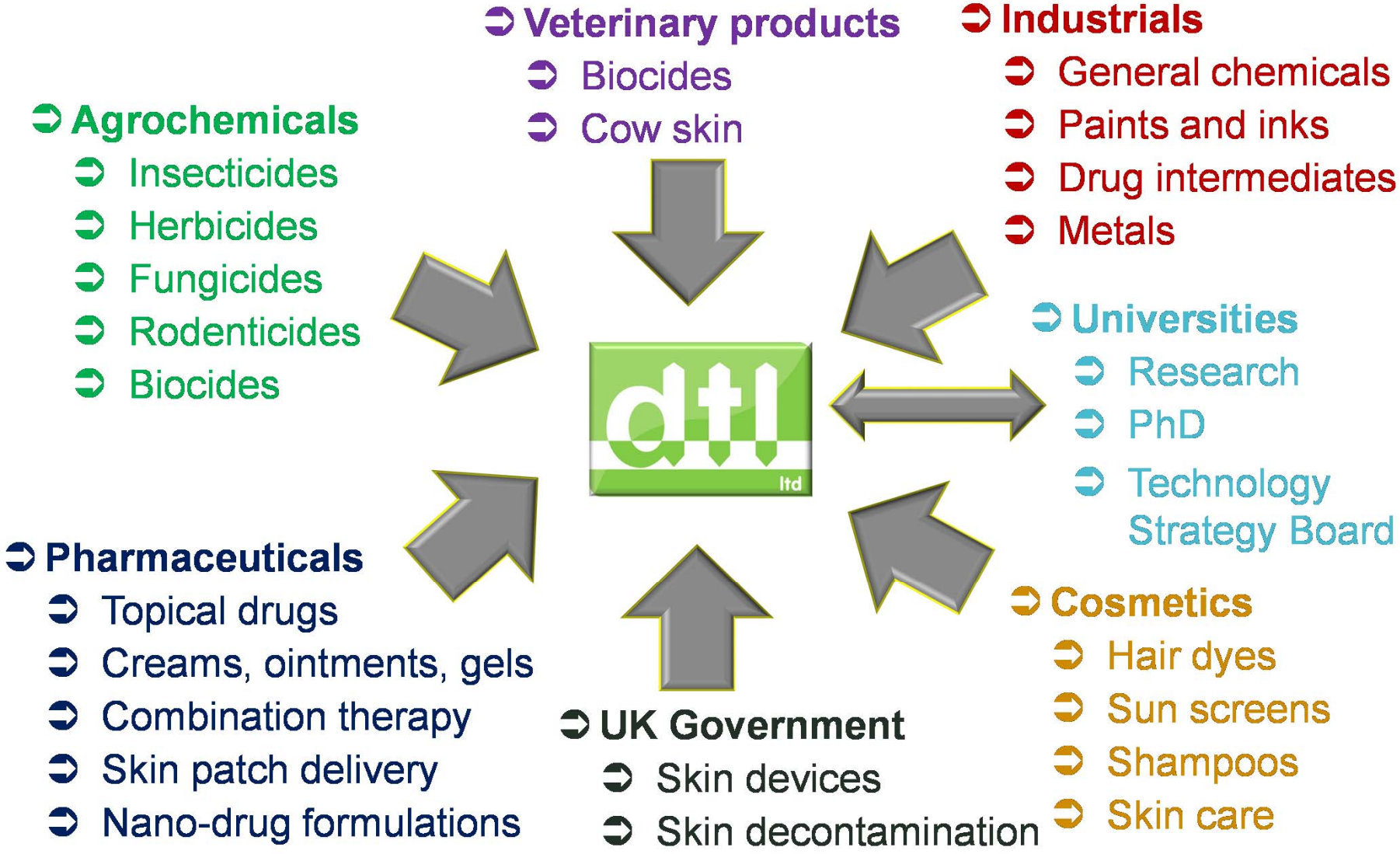
THE QUEEN'S AWARDS FOR ENTERPRISE: INTERNATIONAL TRADE 2016

History

DTL is an independent company established in 2007. We specialise in OECD 428 studies and are based at Keele University Science Park in the United Kingdom

Industries utilising OECD 428 In Vitro Dermal Absorption Studies



OECD Test Guidelines for DA

ECVAM Workshop on "Methods for Assessing Percutaneous Absorption" at Ispra, Italy in 1994 agreed that an *in vitro* approach was sufficiently "validated" and could be used as part of the risk assessment process for chemical products

(Industry member of the OECD Expert Group established in 1999)

OECD Guideline 427: Skin Absorption: *In Vivo* Method (2004)

OECD Guideline 428: Skin Absorption: *In Vitro* Method (2004)

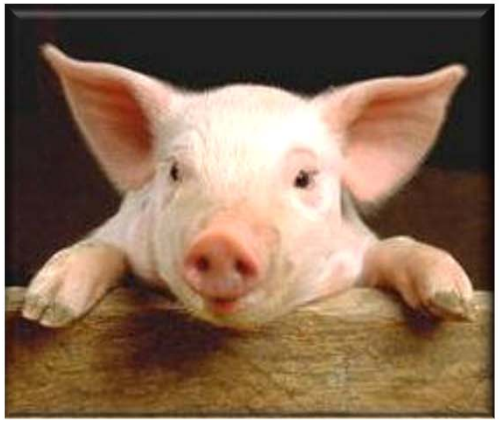
OECD Guidance Document No. 28 (2004)

OECD Guidance Notes on Dermal Absorption No.156 (2011)

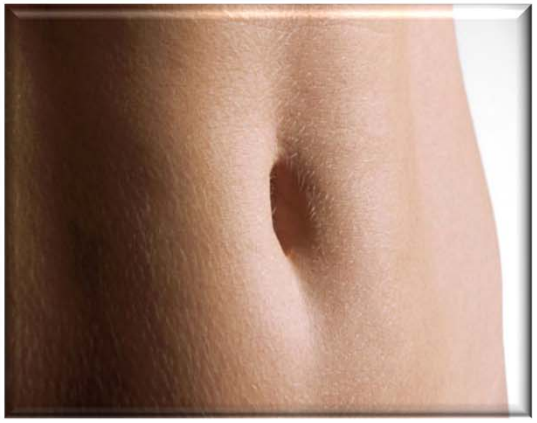
Which Species?

Rat

Pig



Human

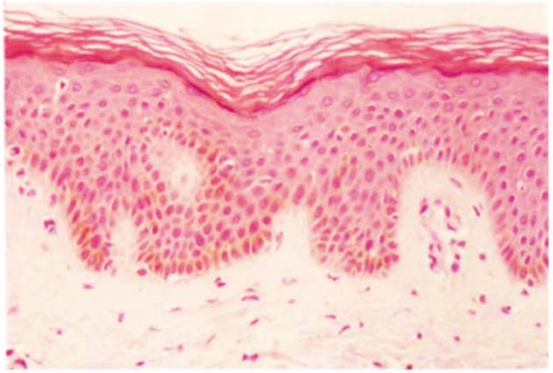


WHO (World Health Organisation), 2006. Dermal Absorption. Environmental Health Criteria **235**. International Programme on Chemical Safety, Geneva.

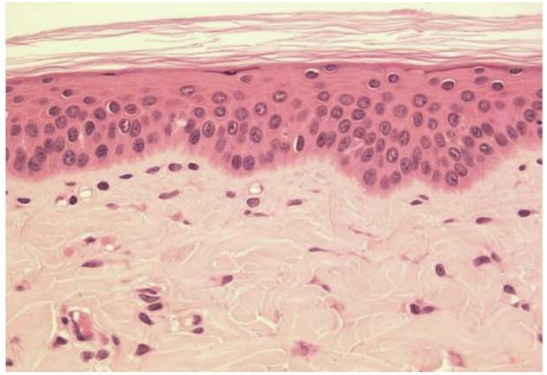
WHO/IPCS 2006: Recommendation No 1. : *“Human skin in vitro and in vivo should be universally recognised as the gold standard in dermal absorption risk assessment.”*

Skin Morphology

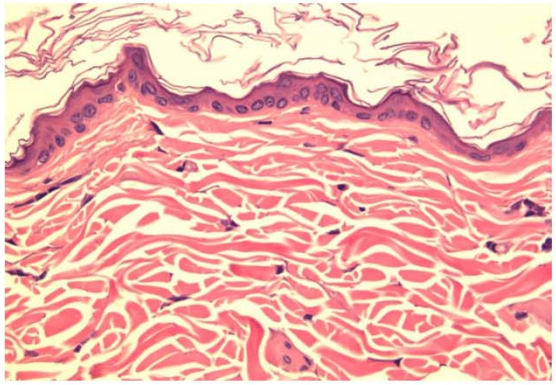
Human



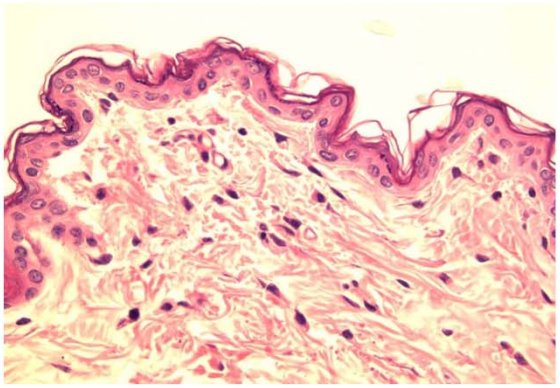
Pig



Rat



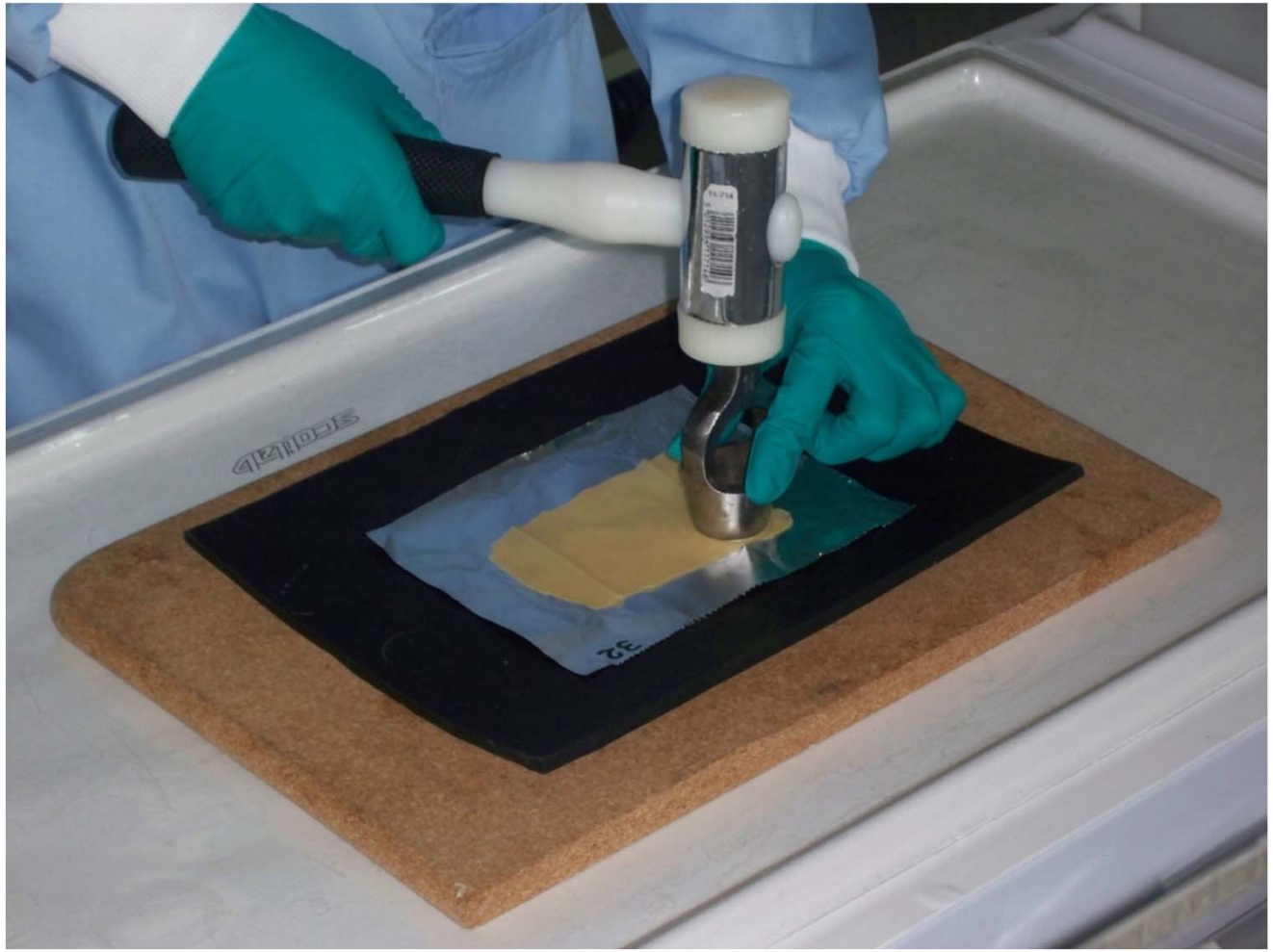
Rabbit



Technical Issues

- Several key technical aspects on the conduct of studies could have been included in the 2017 EFSA Guidance
- They may be viewed as OECD revision issues but have major impact on the performance and variability of DA data
- Methodological issues including skin quality, skin integrity, characterisation of the test material all feed into the quality and reproducibility of the results
- This “variability” in DA data has led to overly conservative approaches being used in the Risk Assessment

Skin Quality and Preparation



Zimmer Dermatome

EFSA Dermal Absorption Guidance 2017

EFSA (European Food Safety Authority), Buist *et al.* 2017. Guidance on Dermal Absorption. EFSA Journal 2017;15(6):4873, 60 pp.

Section 4: Elements of a study design and reporting that reduce experimental variation and aid consistent interpretation

"Integrity of skin used in vitro should be determined prior to application of the test substance and should be documented…

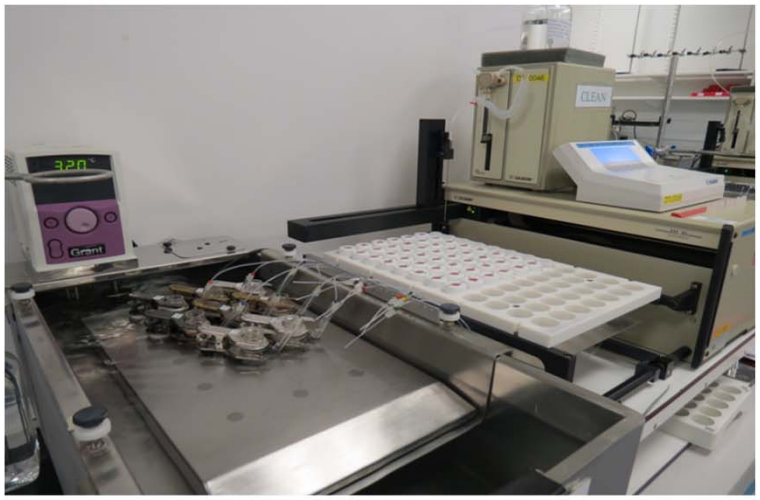
Any membrane with unacceptable integrity should be replaced prior to application. Post-dosing evaluation of integrity and subsequent exclusion of results obtained with skin having insufficient integrity is not recommended".

OECD 428: Skin Integrity Assessment

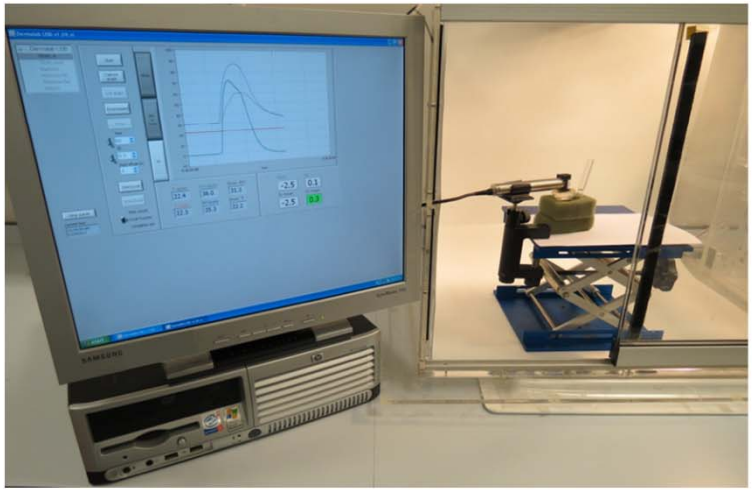
Davies DJ, Ward RJ and Heylings JR (2004) Toxicology in Vitro 18, 351-358

Electrical Resistance (ER)

Tritiated Water Flux (TWF)

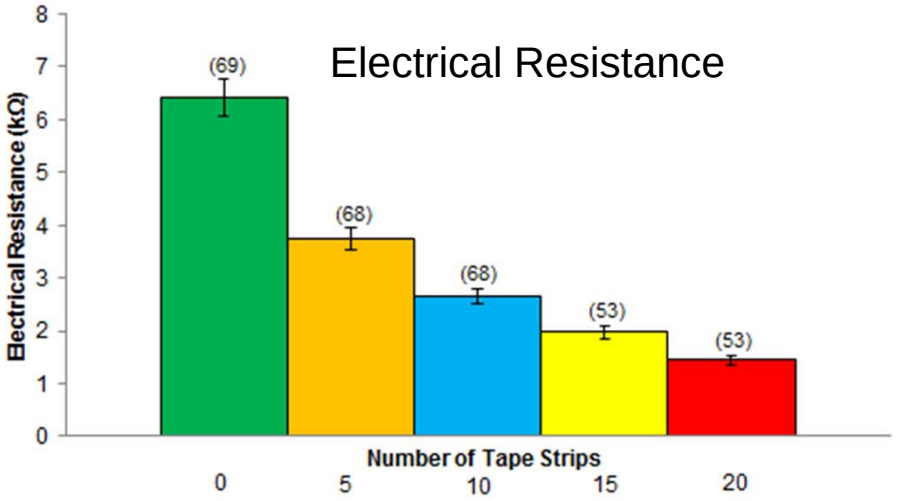


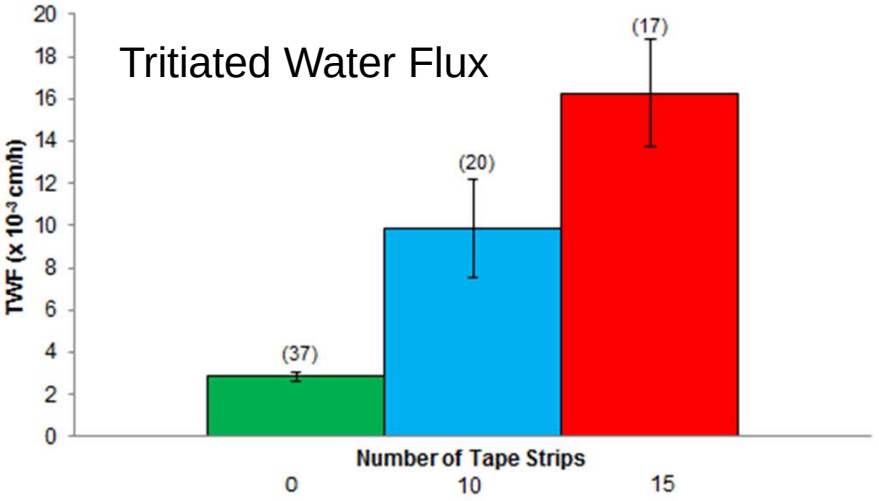
Trans Epidermal Water Loss (TEWL)

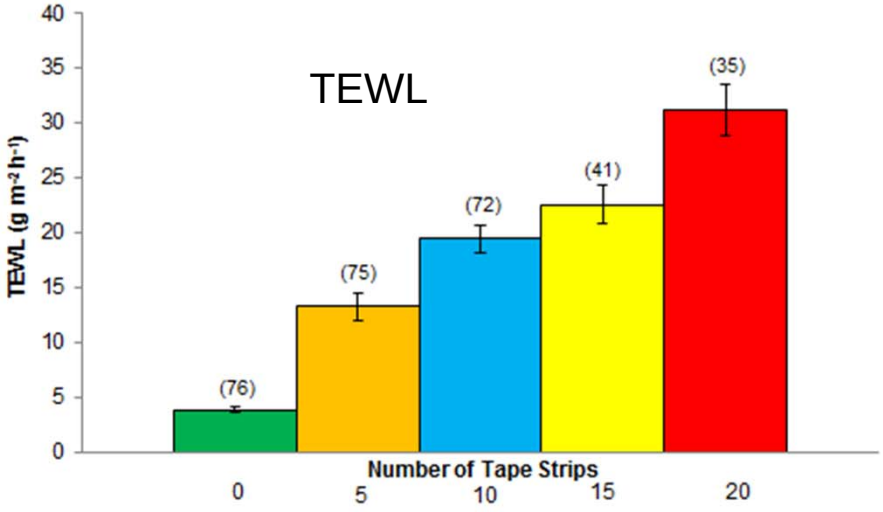


DTL's Franz Static Diffusion Cell

Sensitivity of 3 integrity tests to tape stripping of the *stratum corneum*







Davies DJ, Heylings JR, Correa M and McCarthy TJ. Toxicology In Vitro 29, 176-181, 2015.

SCCS Dermal Absorption Guidance 2010

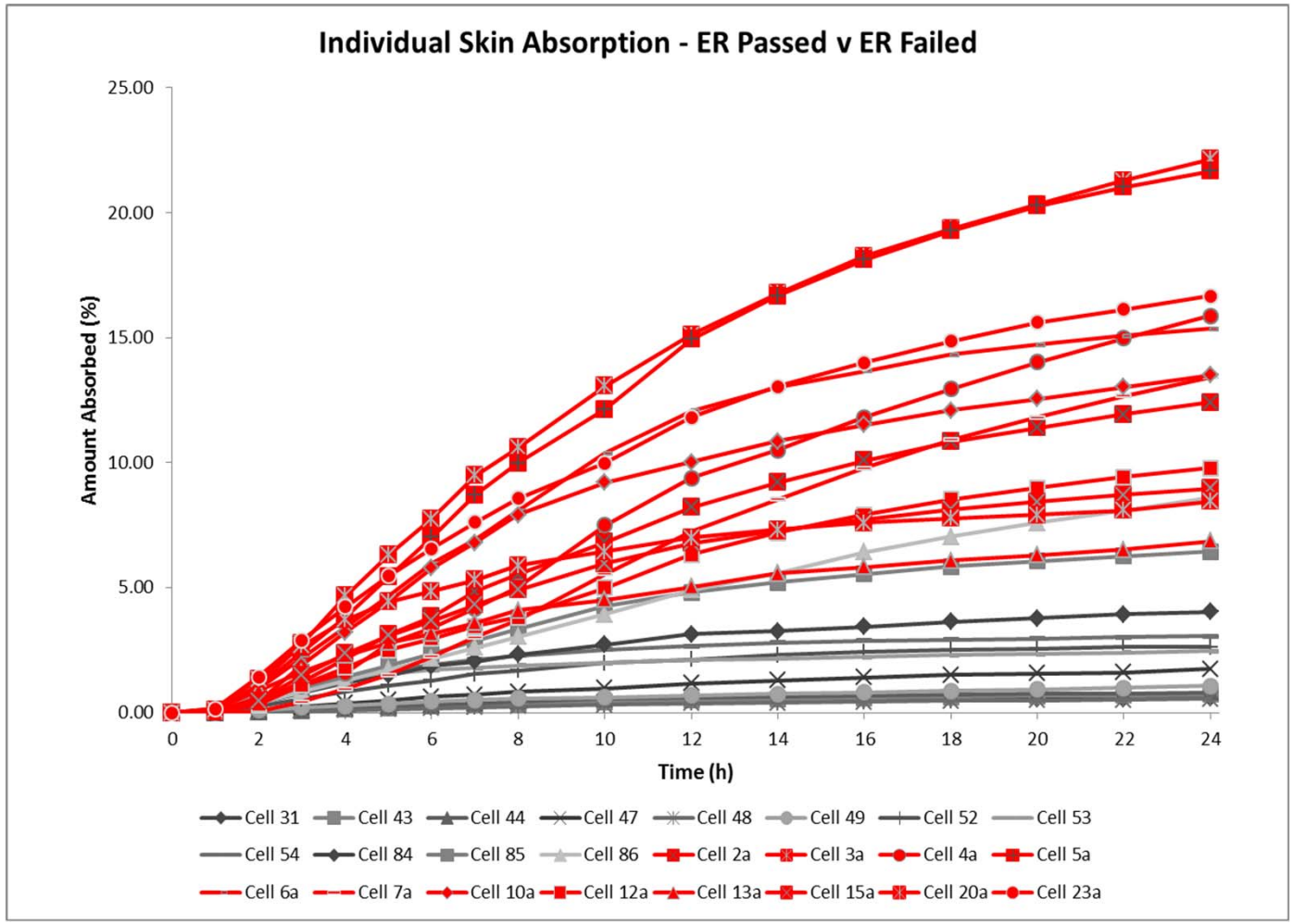
Scientific Committee on Consumer Safety (SCCS): Basic criteria for the *in vitro* assessment of dermal absorption of cosmetic ingredients. SCCS/1358/10.

Section 4.4 : Skin Integrity

"Barrier integrity is crucial for the experiment, and must therefore be measured. This is achieved by either measuring the penetration of a marker molecule e.g. tritiated water, caffeine or sucrose, or by physical methods, such as determination of TEWL (Transepidermal Water Loss) or TER (Transcutaneous Electrical Resistance). Data obtained should be reported."

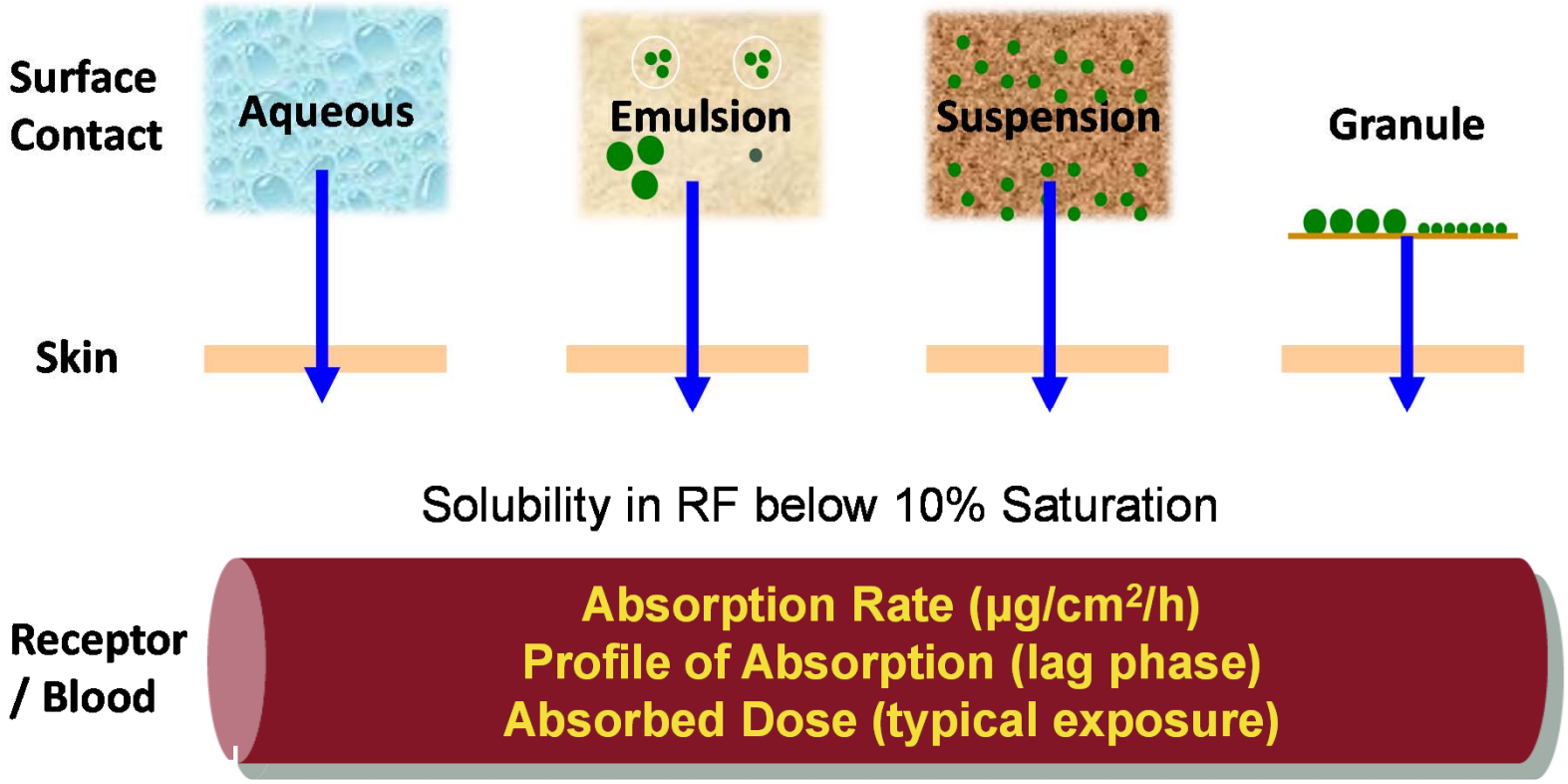
Reference Chemicals : Testosterone

Failed skin samples (shown in red) by ER, even from the same donor can provide high and more variable absorption through dermatomed human skin



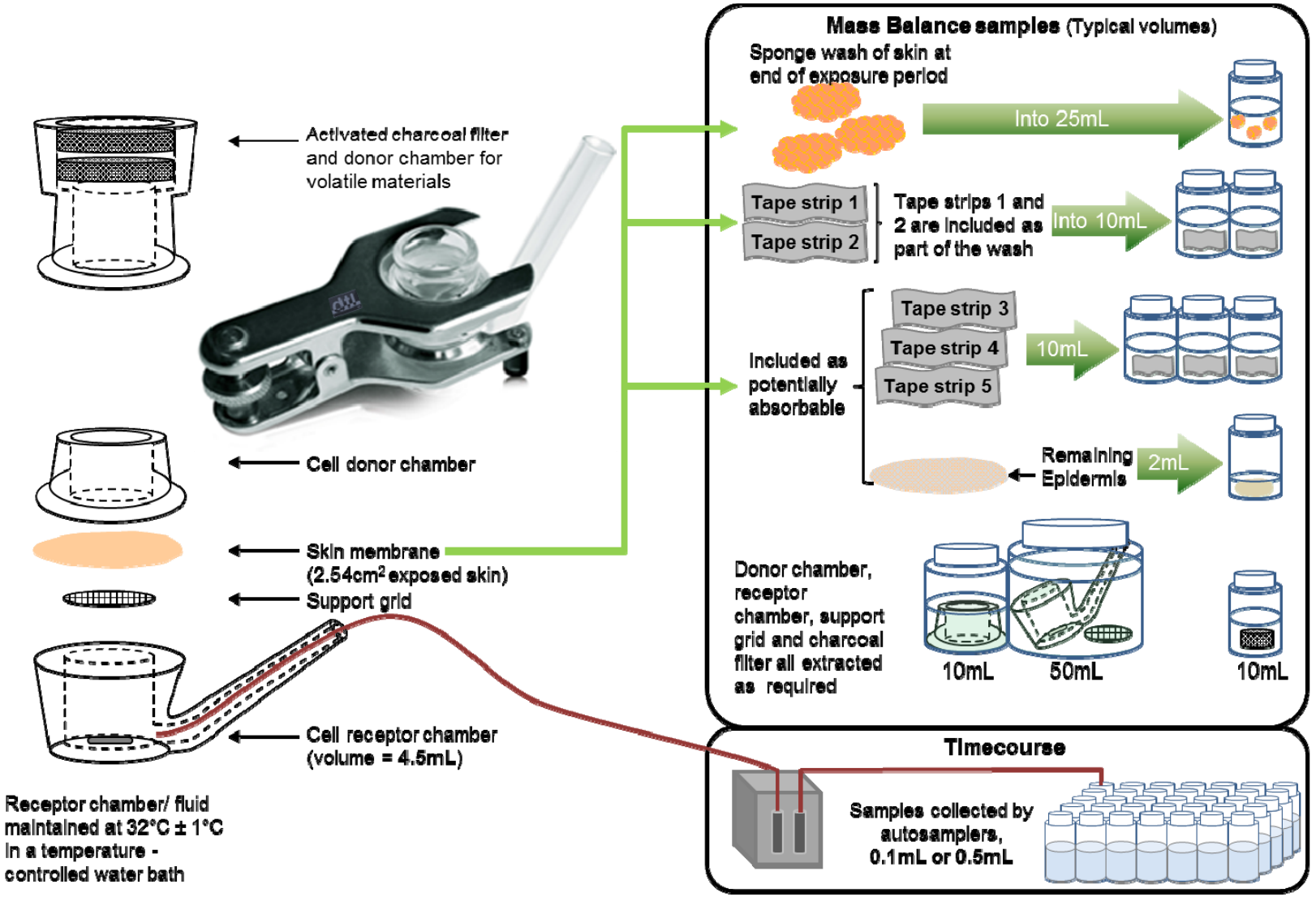
Dose Preparation and Application to the Skin

Full characterisation of the Active in the Formulation concentrate and each Spray Dilution



DTL Static Franz Diffusion Cells

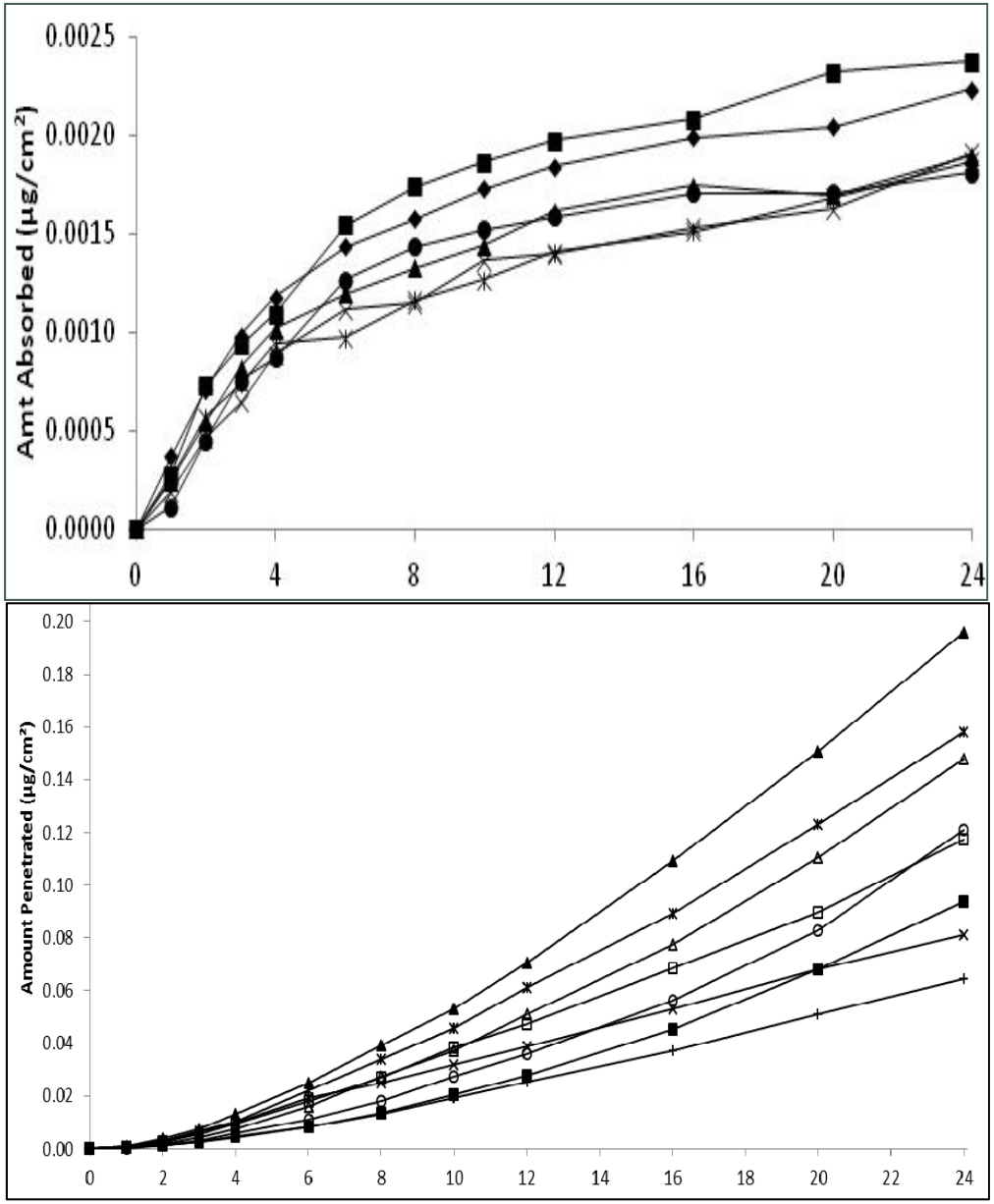
High signal : noise ratio



Dermal Absorption Profiles

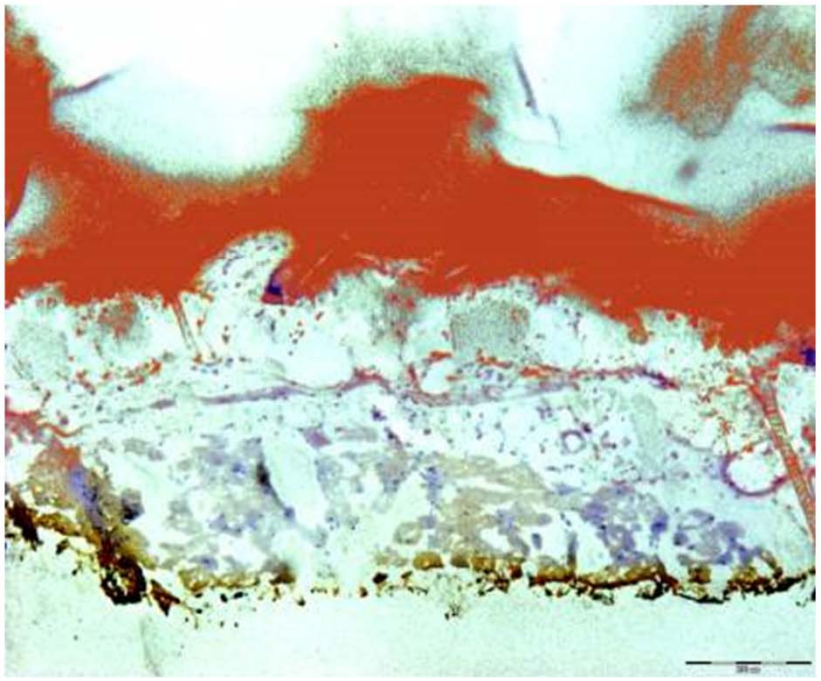
EFSA 2-strip rule
Exclude all tape strips from Absorbed Dose

EFSA 2-strip rule
Exclude first 2 strips from Absorbed Dose

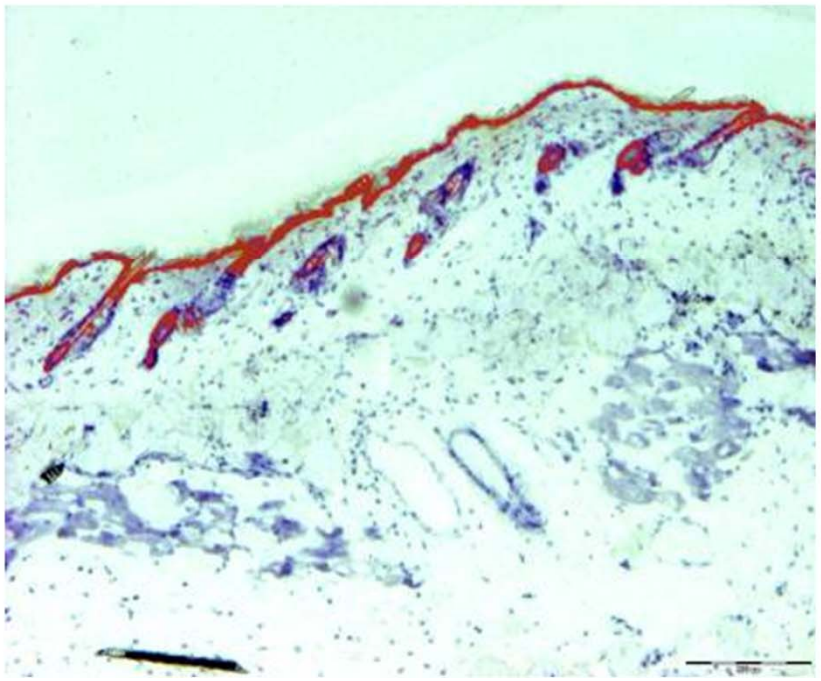


Decontamination of the skin surface

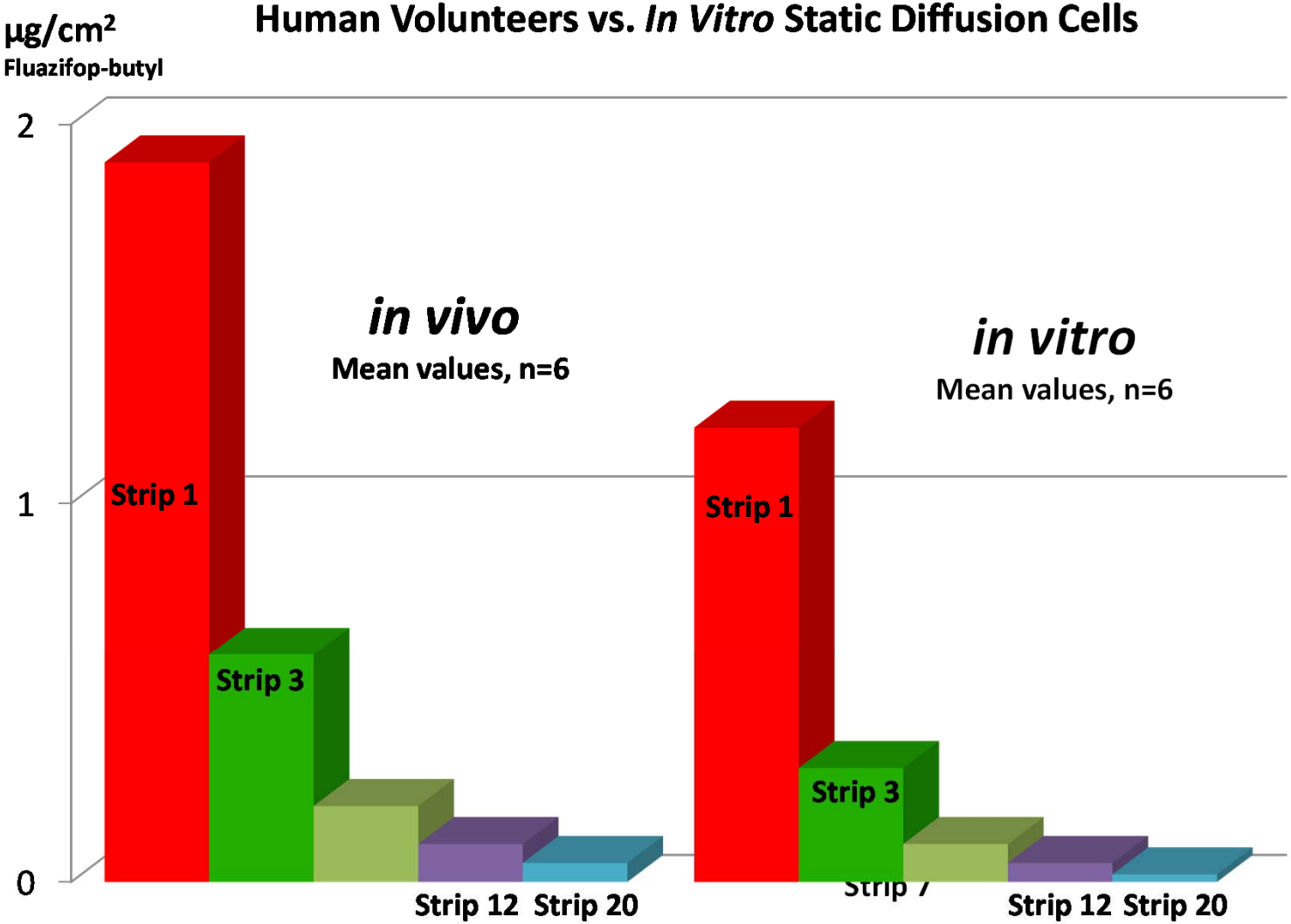
24 h exposure: unwashed skin



24 h exposure: following soap wash



OECD 428: Tape Stripping Procedure



Trebilcock K L, Heylings J R and Wilks M F (1994). In vitro tape stripping as a model for in vivo skin stripping. Toxicology In Vitro 8, 665-667. Now included in EFSA 2017

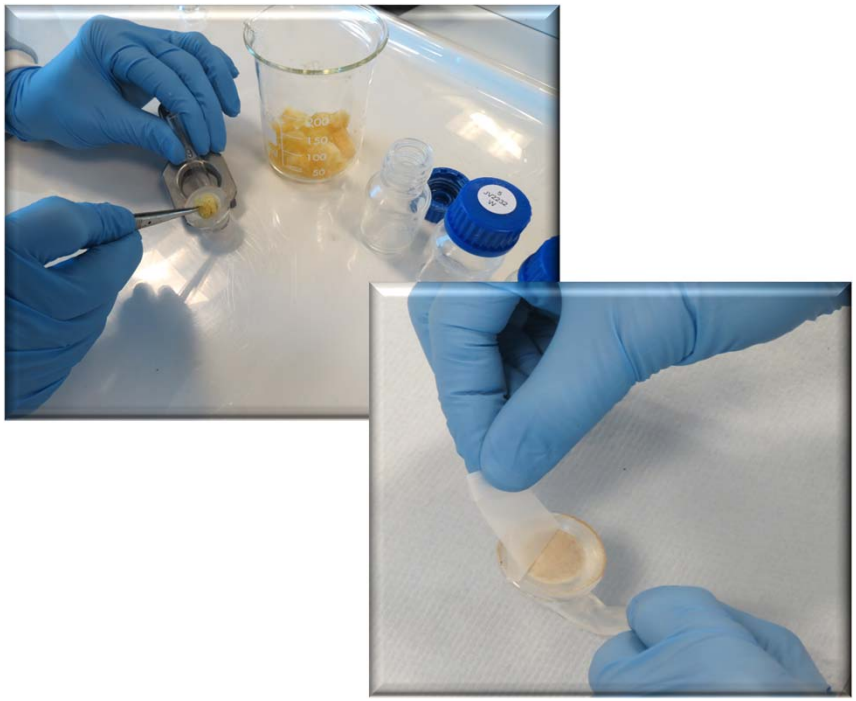
Mass Balance Recovery

Mass balance to account for > 95% of the dose applied (EFSA 2017) or missing fraction assumed absorbed. This is overly conservative and often compound-dependent. SCCS permit 85-115% mass balance

Working day exposure, but decontamination may be earlier and dependent on in-use scenario. More 8h exposures required in 2017. Mainly 6h in previous years

Five/Six compartments measured

- Donor chamber (<1%),
- Skin surface washing (80%),
- Tape stripped *stratum corneum* (10%),
- Remaining skin (5%),
- Receptor chamber (5%).



Mass Balance Distribution

Test Compartment	Compound A		Compound B	
	%	SD	%	SD
Skin wash – 8 hours	94.2	3.15	75.4	10.7
Skin wash – 24 hours	1.34	1.26	5.51	4.44
Tape strips 1-15	0.16	0.07	0.23	0.17
(Epidermis)	0.41	0.43	0.87	0.65
Dermis	0.32	0.30	1.94	1.35
Receptor fluid	2.45	2.05	13.7	5.09
Total Recovered	99.0	0.65	97.9	1.58

SD > 25% of the mean - variability issue amended in the 2017 EFSA Guidance to a multiplication factor (k) based on number of replicates

Final Comments

We need a revision of OECD 428 to deal with the key technical issues in the EFSA Guidance Document. This will…

- Reduce variability of DA data within a Laboratory
- Improve consistency across different Laboratories
- Provide more reliable absorption values in the Risk Assessment, without being overly-conservative
- Allow us to focus on human skin as a predictor of human absorption and move away from the rat