aaps Communities

AAPS Topical and Transdermal Community

Welcome!

Chair, AAPS Topical and Transdermal Community

Sam Raney, PhD

Happy New Year - 2020!

It's Great to Have You With Us!

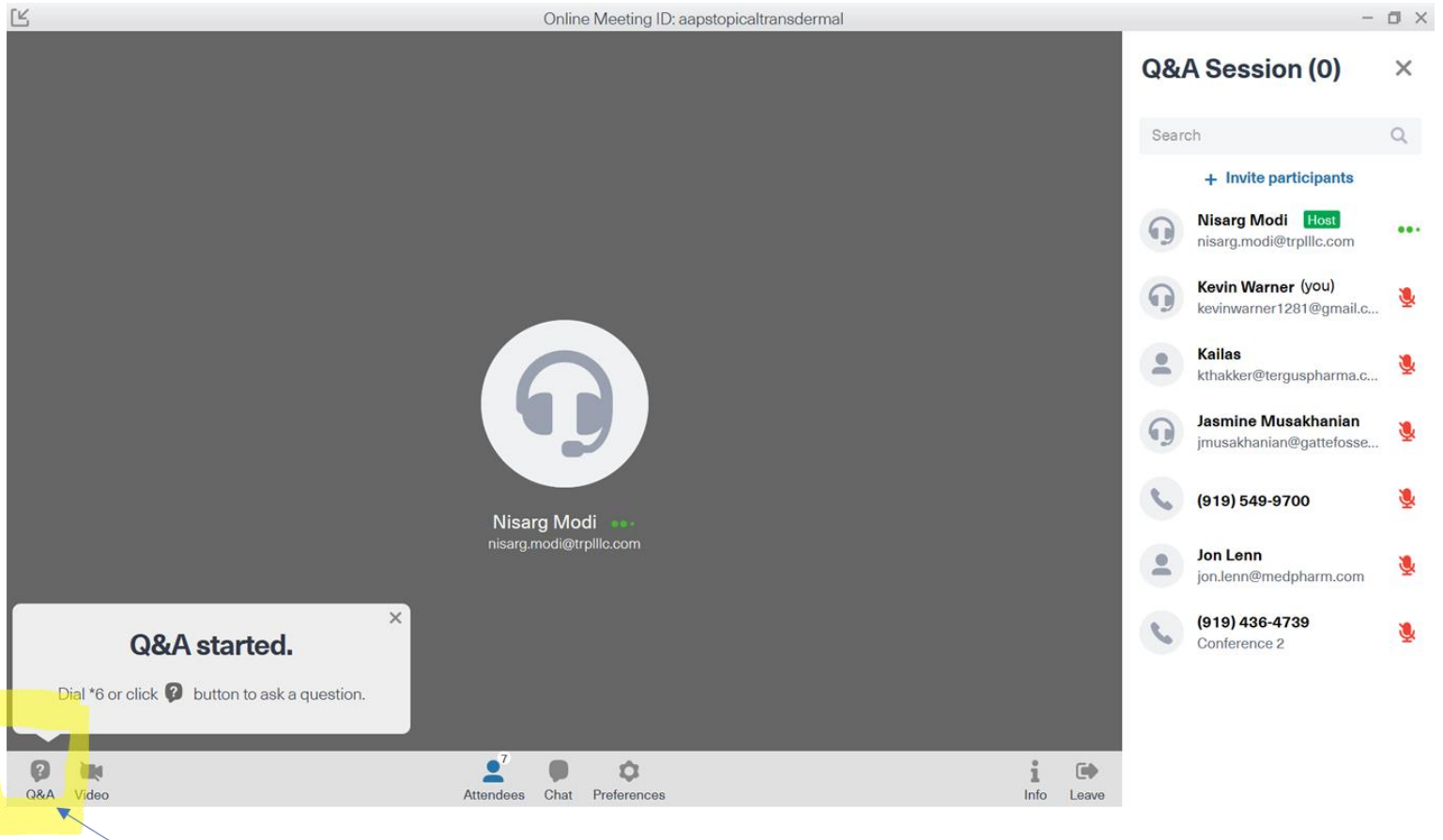
We will Start Soon at 12:05 PM

We're Just Affording a Few Minutes for Others to Join

aaps Communities

AAPS Topical and Transdermal Community

How to Ask A Question…



Click this icon or dial *6 to get in the queue to ask a question.

aaps Communities

AAPS Topical and Transdermal Community

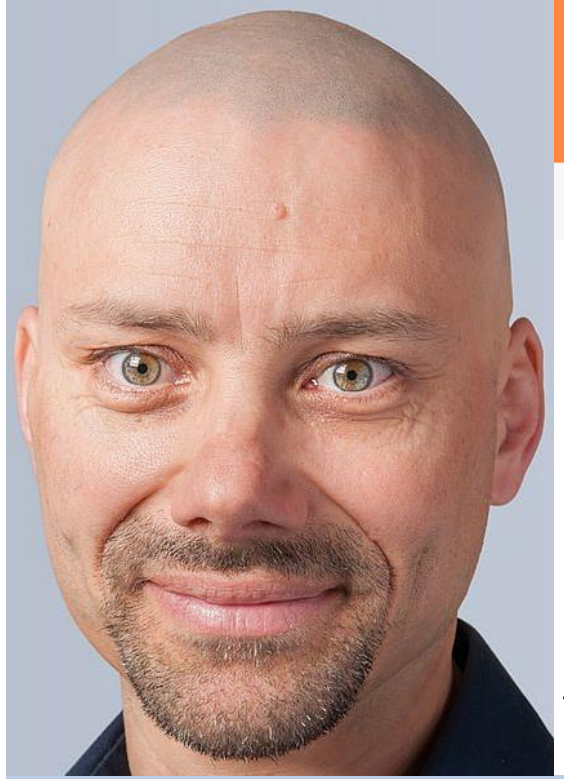
Moderator for Today's Webinar

Prof. Narasimha Murthy, PhD

Steering Committee Senior Advisor
AAPS Topical and Transdermal Community

Professor of Pharmaceutics & Drug Delivery

THE UNIVERSITY of MISSISSIPPI



aaps® Communities

AAPS Topical and Transdermal Community

FREE MONTHLY WEBINAR SERIES

Dr. Frank Sinner, Joanneum Research

Use of Skin Pharmacokinetics (PK) & Pharmacodynamics (PD) in Drug Development

January 10th, 2020

12.00 PM – 01.00 PM (EDT)

Moderated by Prof. Murthy

- *Better understanding the disease by direct access to the dermis in vivo*
- *Reducing the risk of failure in drug development by using skin PK/PD*
- *Comparing dermal PK profiles for topical generic drug development*
- *Open mike for Q&A as well as any discussion topic*

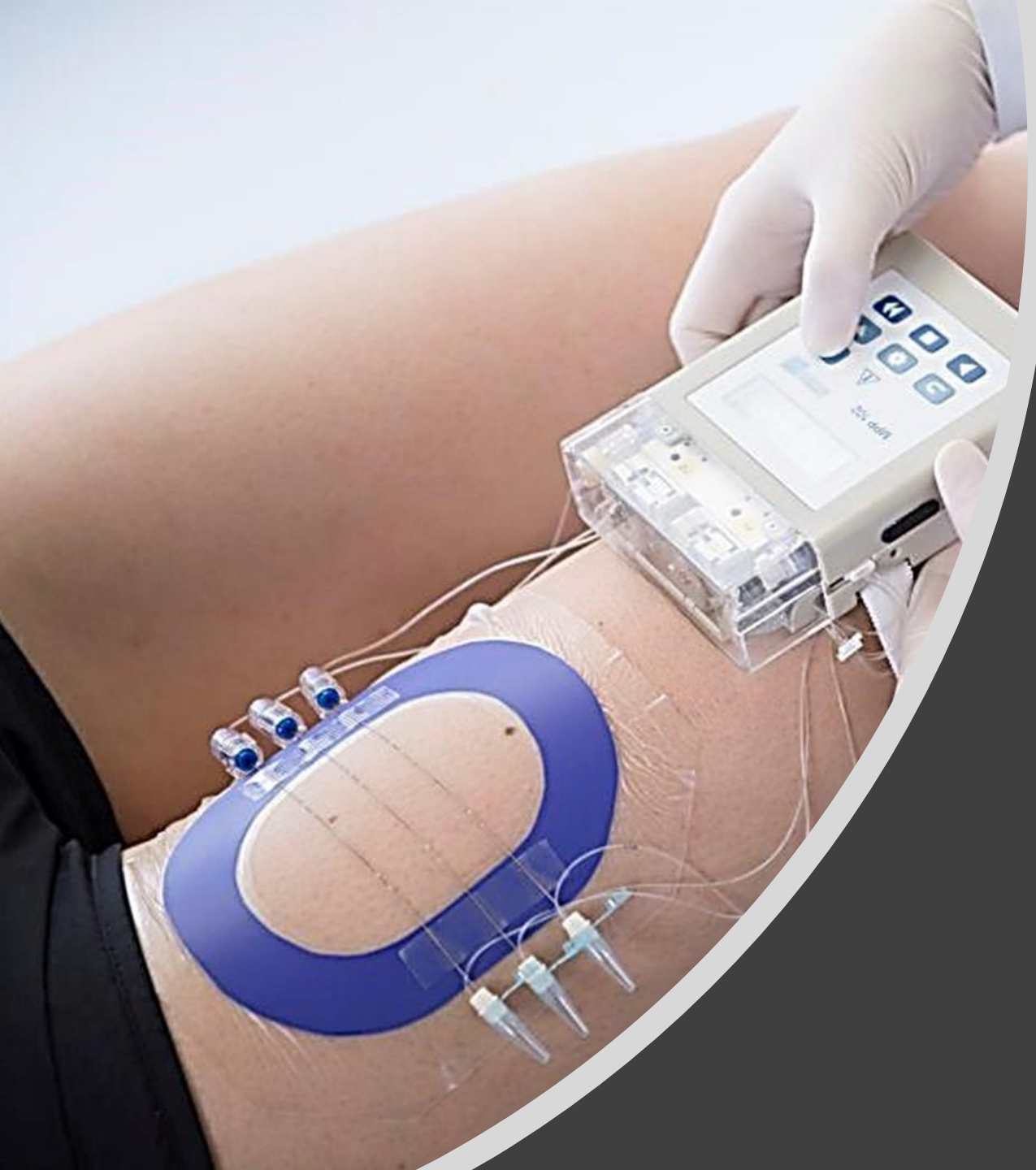
How do I join? https://join.freeconferencecall.com/**aapstopical**transdermal

If you have trouble logging in, please email: *AAPS.Topical.Transdermal@gmail.com*

Use of Skin PK/PD in Drug Development

What's Coming Up?

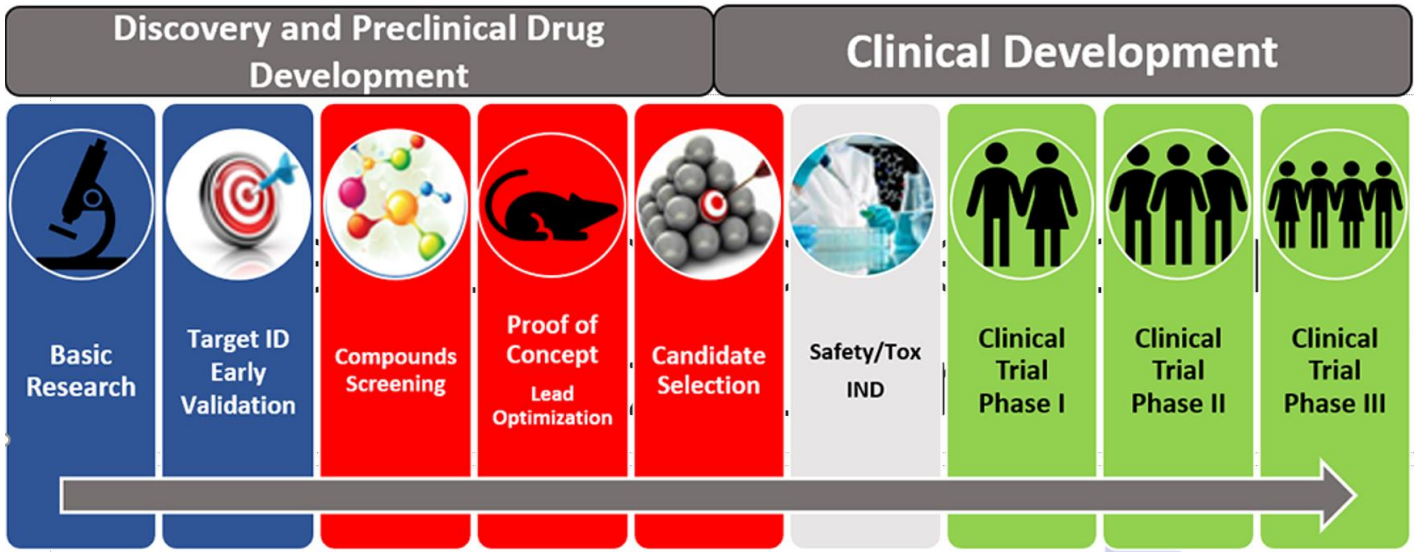
- Reduce The Risk of Failure in Drug Development – Value your target tissue!
- Open Flow Microperfusion – An introduction
- Case Studies 1 – 6: How skin PK/PD may reduce your risk of failure in drug development.



Reduce The Risk of Failure in Drug Development

Value Your Target Tissue!

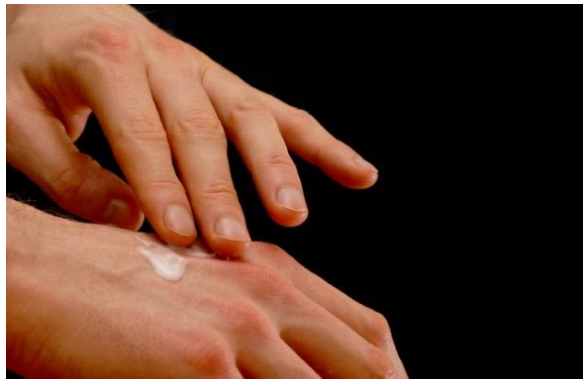
Drug Development
How Skin PK/PD May Reduce Failure Risks



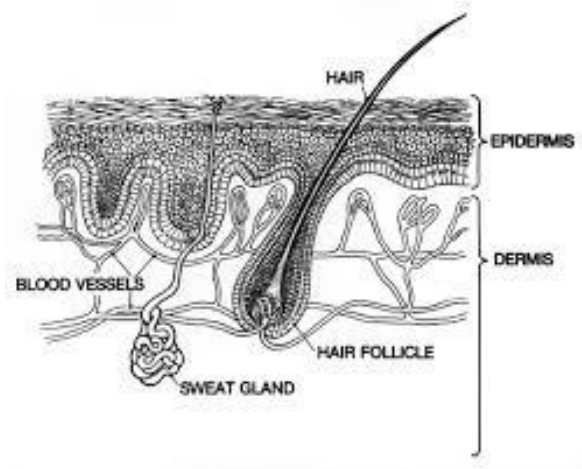
https://www.nia.nih.gov/news/nia-director-encourages-wider-use-transformative-alzheimers-research-resources

Arrowsmith, J., Miller, P., Nature Reviews (12), 2013; 2) Ledford, H., Nature (477), 2011

Drug Development
Value Your Target Tissue

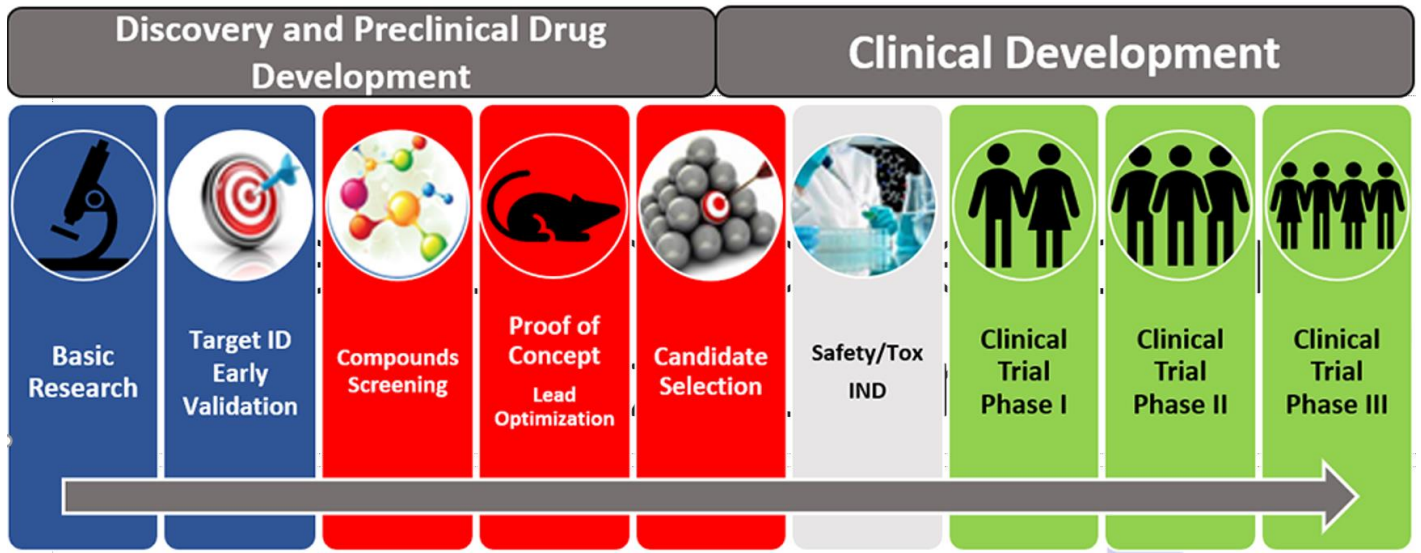


Does blood really reflect your drug‘s PK/PD in the dermis?



Does the API reach the site of action in a therapeutic concentration?

Drug Development
How Skin PK/PD May Reduce Failure Risks

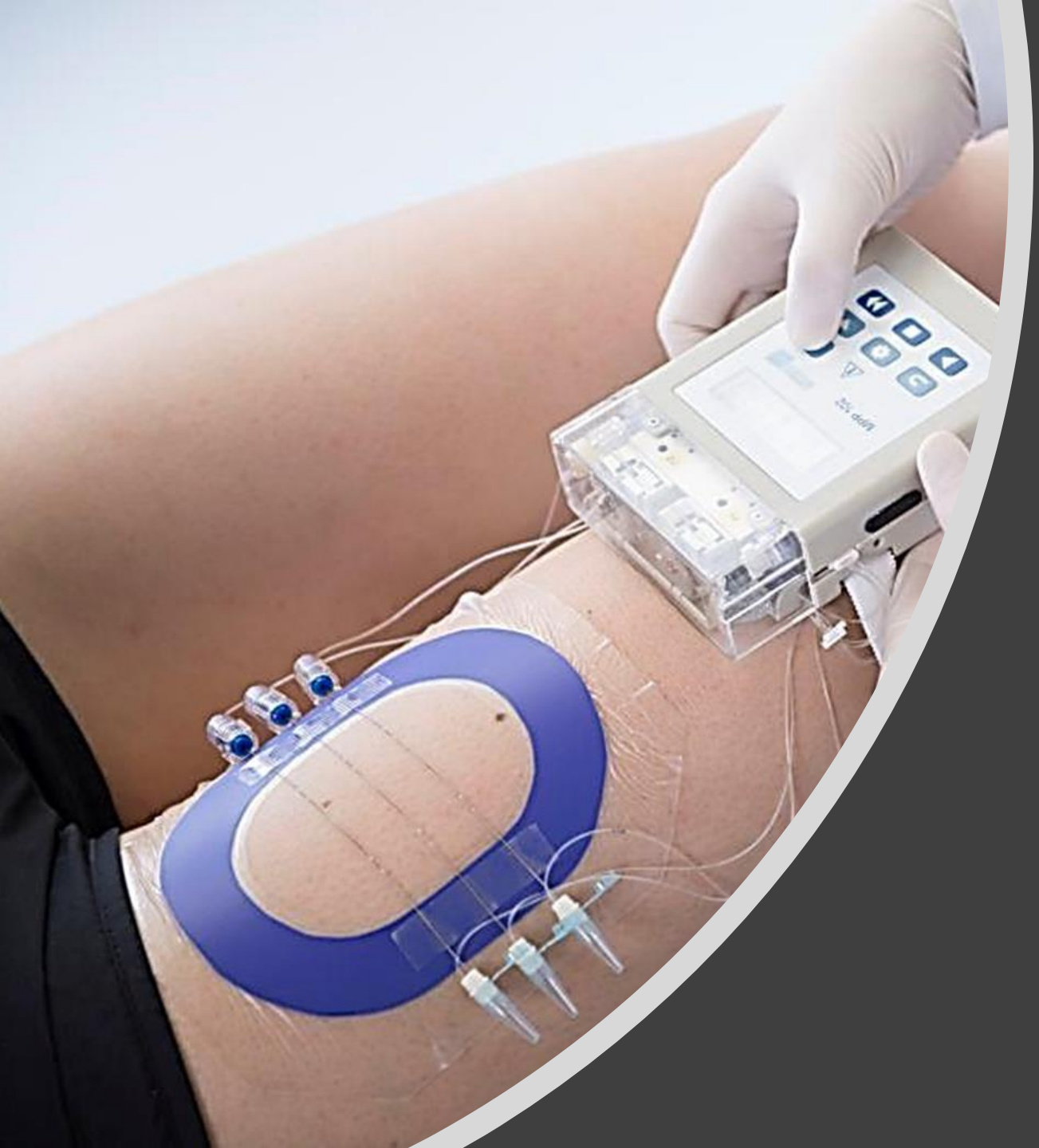


Case Study 1: **Disease-specific Biomarker**

Case Study 2: **Prediction Drug Effect** (ex-vivo)
*Case Study 3: **Candidate Screening** (in-vivo)*
*Case Study 4: **Psoriasis Rat Model** (in-vivo)*

Case Study 5: **Secukinumab**
Case Study 6: **BE for Generics**

https://www.nia.nih.gov/news/nia-director-encourages-wider-use-transformative-alzheimers-research-resources

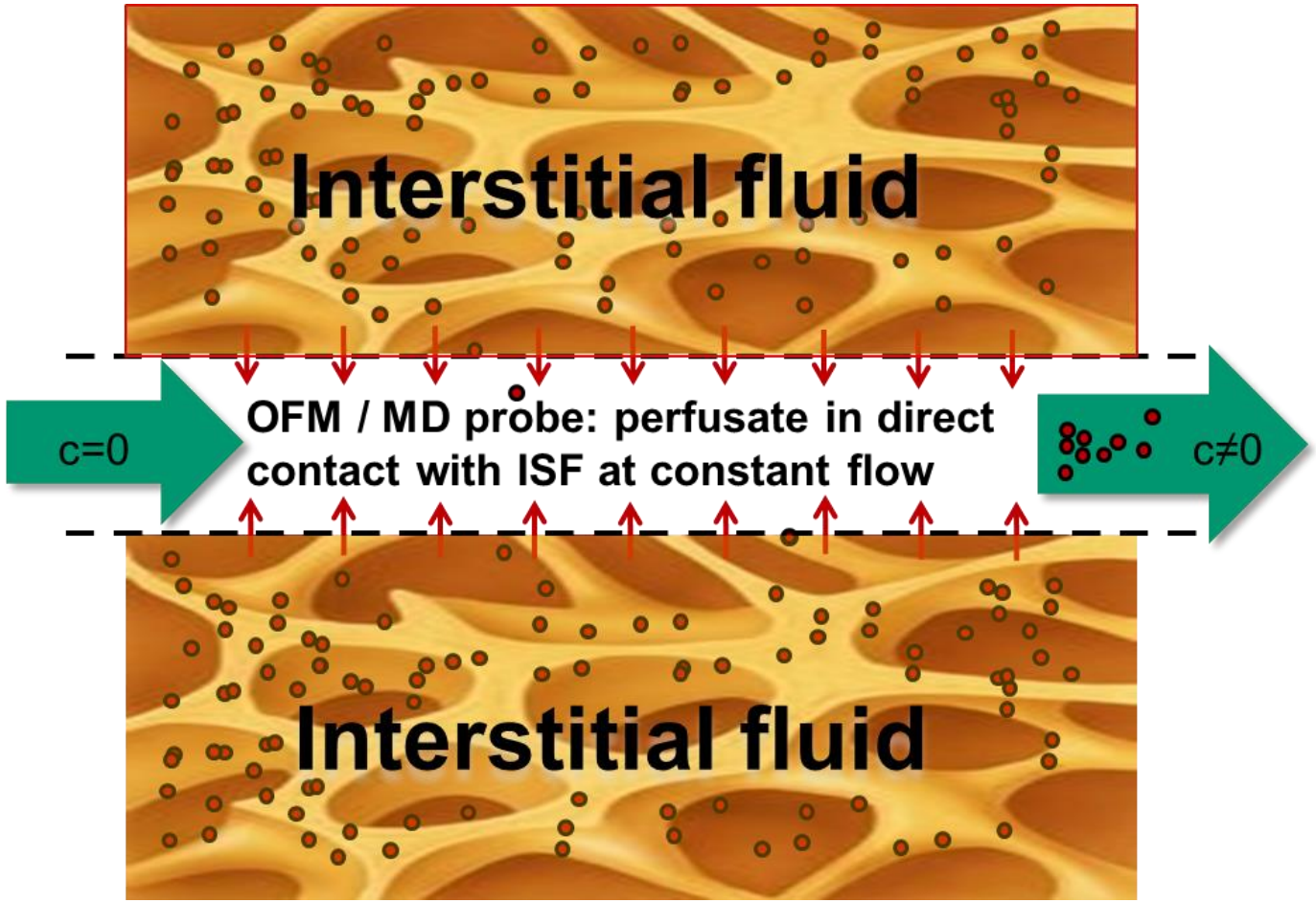


Open Flow Microperfusion

An Introduction

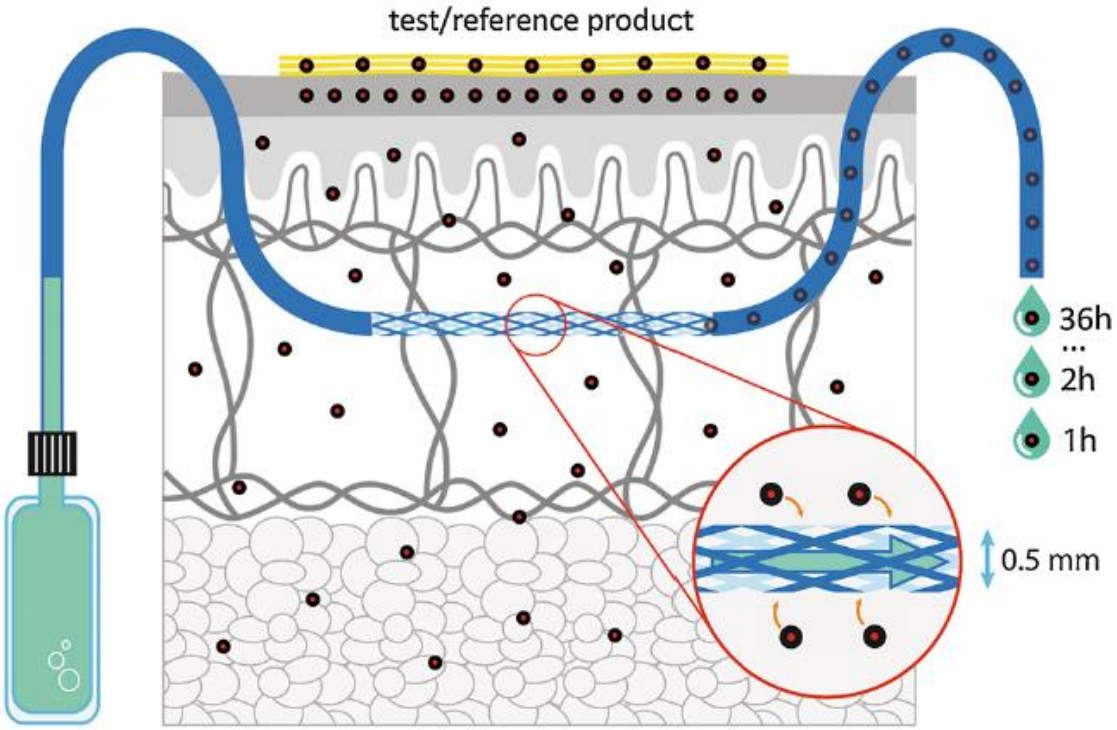
Skin PK/PD
Target Tissue: Interstitial Fluid

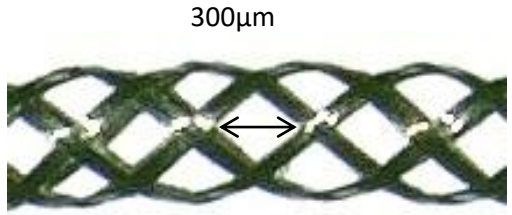
- Open Flow Microperfusion
  - Direct contact of perfusate and ISF
  - Access to diluted ISF
  - Simultaneous PK and PD
  - Pre-clinical and clinical use
  - Easy to add to existing animal models
  - Use in combination with proteomics, metabolomics, FACS, and other analytical platforms.



Skin PK/PD
Open Flow Microperfusion

✓ OFM samples represent diluted but unfiltered interstitial fluid

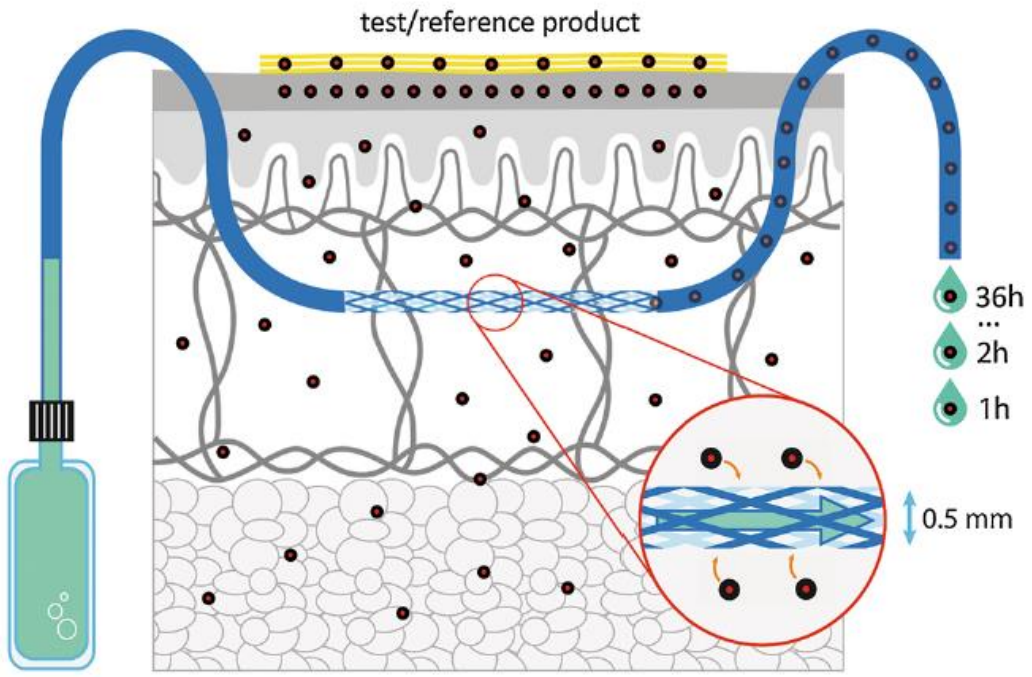




CE-certified for clinical use

Skin PK/PD
Open Flow Microperfusion

✓ **All drugs are accessible in-vivo in the dermis**



lipophilic substances

Bodenlenz et al. 2016 (CP-17; logP 3.5)
Holmgaard et al. 2011 (Fentanyl; logP 4.5)

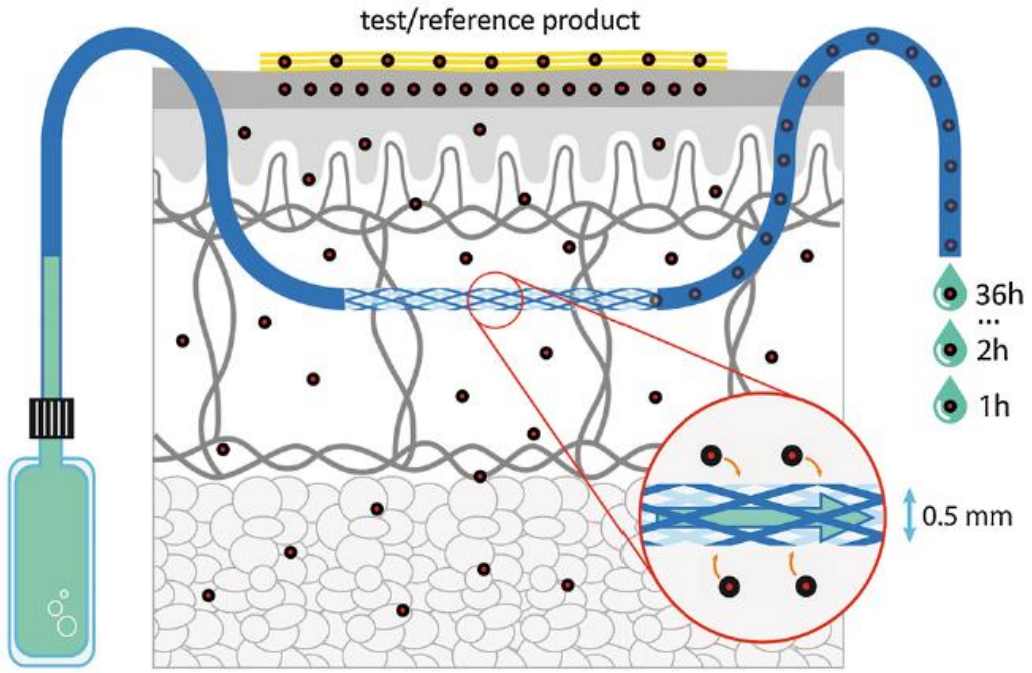
high molecular weight substances (up to cells)

Dragatin et al. 2016 (Quantification of antibodies in skin)
Kolbinger et al. 2016 (Cytokines in the skin in healthy & patients)

➔ PK and PD in parallel

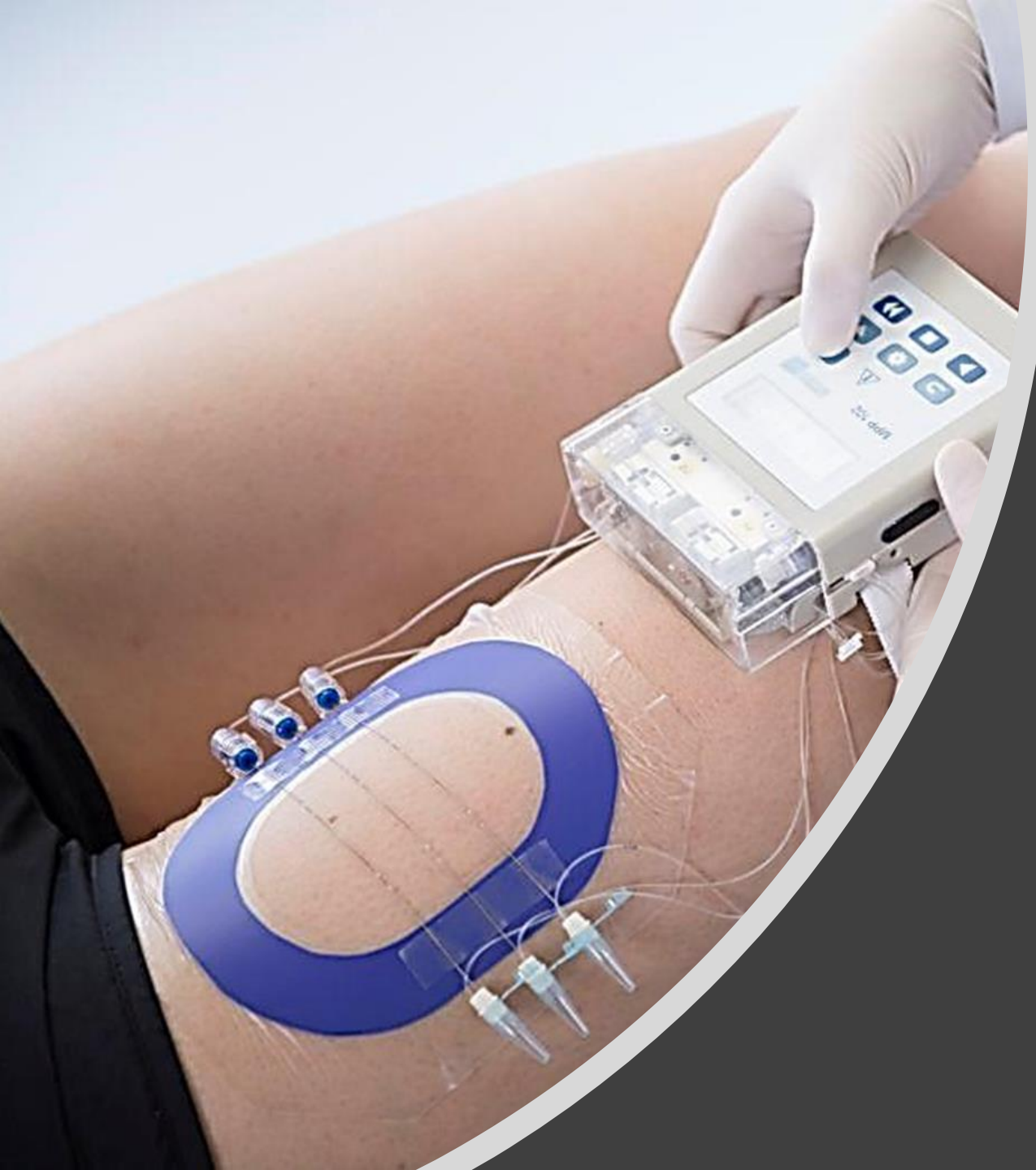
Skin PK/PD
Open Flow Microperfusion

✓ dOFM shows dose dependent dermal AUC profiles



Clinical dose response studies

Acyclovir (Zovirax® US, topical) – 36 h
Acyclovir (Zovirax® UK, topical) – 36 h
Acyclovir (Zovirax® AT, topical) – 36 h
Lidocaine (EMLA US, topical) – 24 h
Prilocaine (EMLA US, topical) – 24 h



Open Flow Microperfusion

Case Study 1

Biomarker in Psoriasis

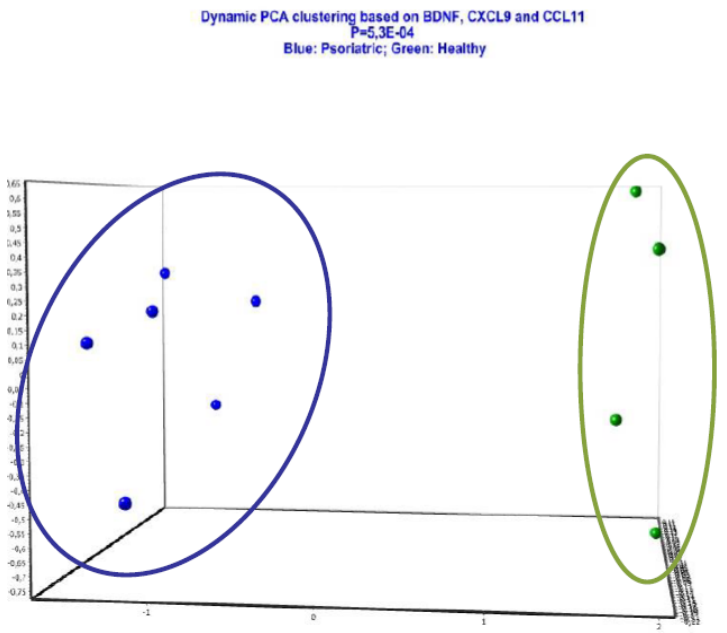
Dermal Open Flow Microperfusion
Case Study 1: PD Marker in skin

✓ Comparison of PD Marker in healthy subjects and in psoriatic patients by dOFM

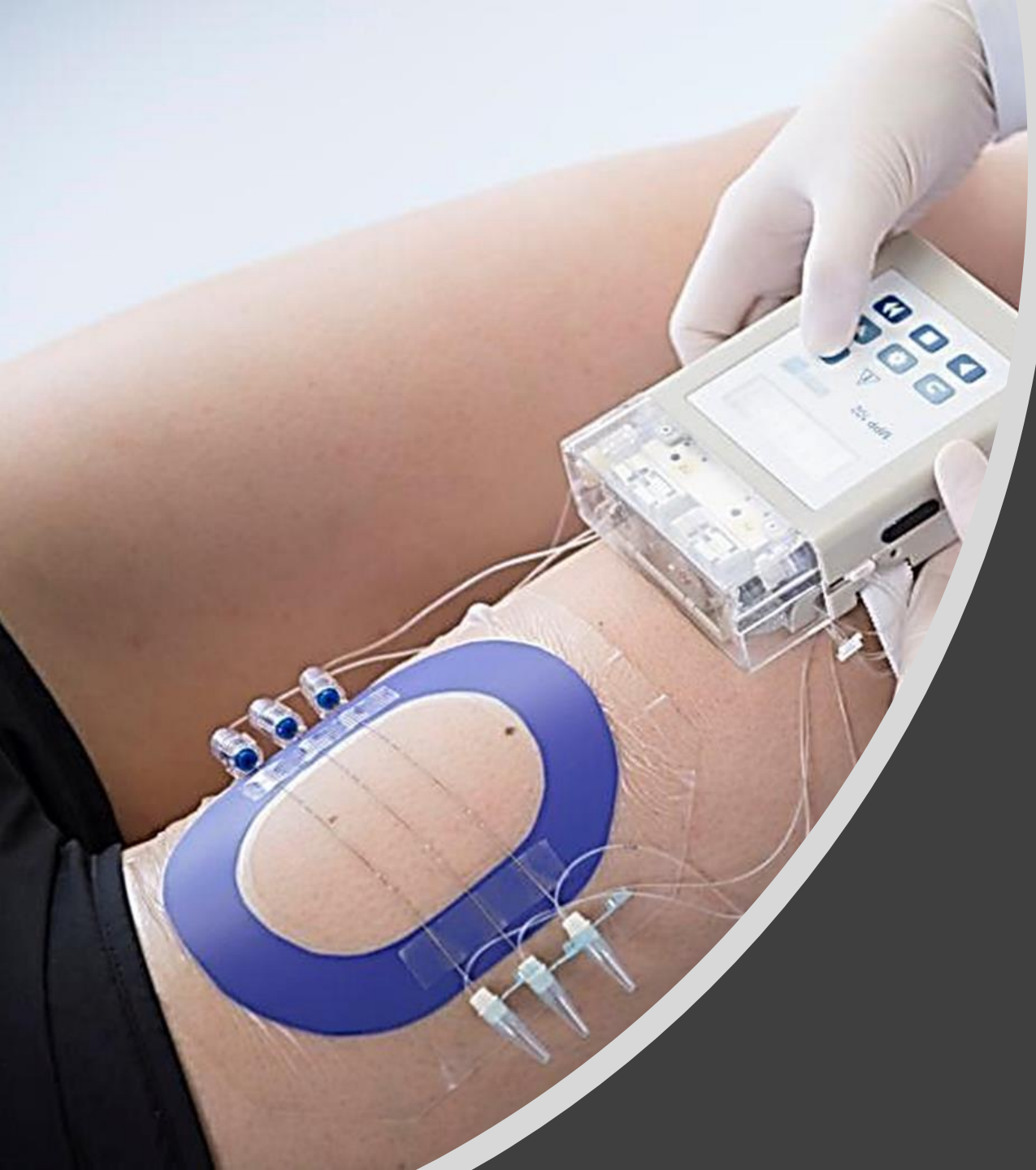
Investigate/confirm disease PD marker in 1 µl dOFM sample

Proof of concept

- 6 psoriatic and 4 healthy samples
- >50 proteins were above LOD
- 4 cytokines were significantly different



Complex protein profiles simplified using dynamic PCA and showing group separation.
Dynamic PCA with p=5,3E-4 with 3 proteins contributing to the pattern.



Open Flow Microperfusion

Case Study 2

Prediction of Drug Effect

Dermal Open Flow Microperfusion
Ex-Vivo Models

✓ **Economic set-up for PK comparison and drug stability**

Available models:	pigs and human whole tissue
Duration time:	“unlimited” – normally up to 48 hours
Application sites:	up to 3 sites with 3 dOFM probes each
OFM material:	same material for preclinical and clinical
Time resolution:	determined by analytics (5 to 120 min)

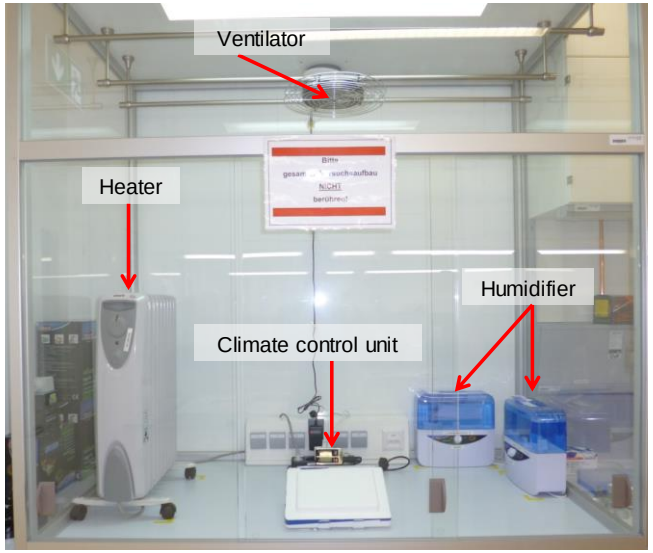
Dermal Open Flow Microperfusion
Case Study 2: Prediction of Drug Effect

Case Study (Leo Pharma)[1]

AIM: Development of a topical drug for AD treatment which has

- high dermal API levels for drug effect (>EC50) and
- low systemic effect to reduce side effects (high systemic clearance)

➔ **PDE4 inhibitors with high in vivo clearance and adequate skin stability**



API candidates:

- A: low Mw, LogD ~3, human unbound fraction ~2%, *in vitro* skin model: stable, EC50* ~80nM, ….
- B: low Mw, LogD ~3, human unbound fraction ~5%, *in vitro* skin model: stable, EC50* ~60nM, …

➔ **Both compounds show in-vitro activity and were selected for clinical development**

*EC50 is based on in vitro inhibition of LPS induced TNFalpha release from human PBMCs

[1] unpublished results: from Leo Pharma: Maja Lambert, Stefan Eirefeldt, Fredrik Johansson, Line Hollesen Basse, Malene Bertelsen, Jens Larsen, Simon Feldbæk Nielsen

Dermal Open Flow Microperfusion
Case Study 2: Prediction of Drug Effect

Case Study (Leo Pharma)[1]

Ex-Vivo Human Skin Punch Biopsies

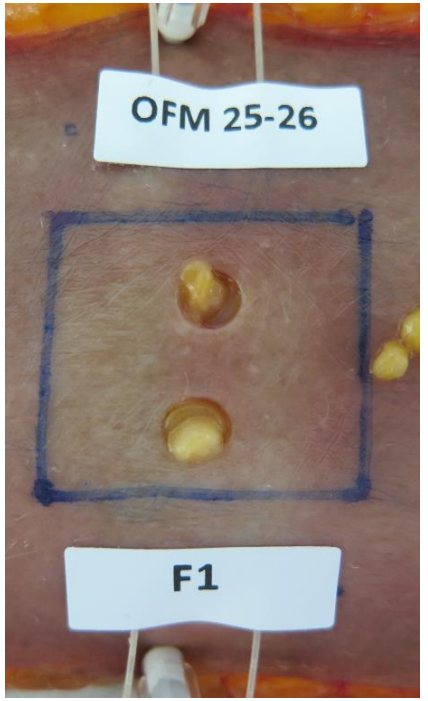
- **A**: [API] > factor 10 higher than EC50
- **B**: [API] > factor 100 higher than EC50

➔ **Both compounds are good candidates for clinical evaluation**

Clinical Trial

- **A** demonstrated clinical efficacy in AD patients (phase 2) in a 4 wk proof of concept study with twice daily dermal application of a cream formulation in different strengths of the cream vehicle and Elidel cream. Biopsy concentrations were determined at 10 µM.
- **B** showed now difference to cream vehicle in a clinical study with AD patients 3 wk with twice daily dermal application of cream formulation compared to cream vehicle. Biopsy concentrations were determined at 6 µM.

➔ **Punch biopsies revealed API concentration well over EC50 but B showed no treatment effect.**



[1] unpublished results: from Leo Pharma: Maja Lambert, Stefan Eirefeldt, Fredrik Johansson, Line Hollesen Basse, Malene Bertelsen, Jens Larsen, Simon Feldbæk Nielsen

Dermal Open Flow Microperfusion
Case Study 2: Prediction of Drug Effect

Ex-Vivo Fresh Human Skin OFM Model

- Determination of 24 hour dermal concentration profile for API

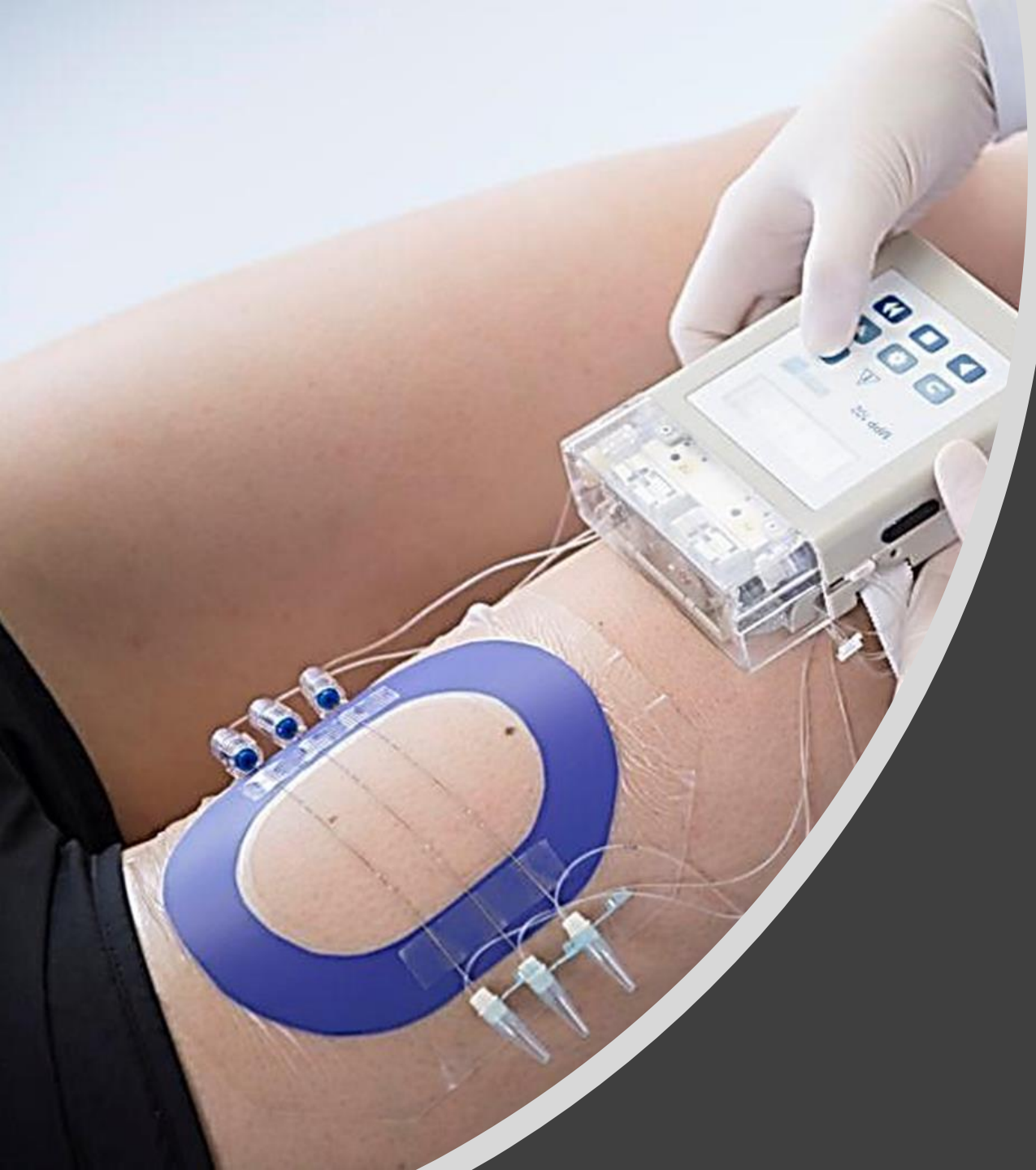
➔ **Elimination of punch biopsy contamination due to remaining drug at SC**

➔ **Focus on the relevant compartment → DERMIS to reflect effective API concentration**

RESULTS

- A:[API] more than 10 fold lower compared to biopsies but higher than EC50
- B: [API] more than 10 fold **lower** compared to biopsies and below EC50

OFM allows a realistic determination of API PK profiles to predict clinical efficacy, essential in the absence of reliable biomarker



Open Flow Microperfusion

Case Study 3

Lead Drug Candidate Screening

Dermal Open Flow Microperfusion
In-Vivo Models

✓ **Excellent set-up for in-vivo PK head-to-head comparisons**

Available models: pigs and rats

Duration time: up to 14 hours

Application sites: up to 3 sites in rats; up to 10 sites in pigs

OFM material: same material for preclinical and clinical

Time resolution: determined by analytics (5 to 120 min)

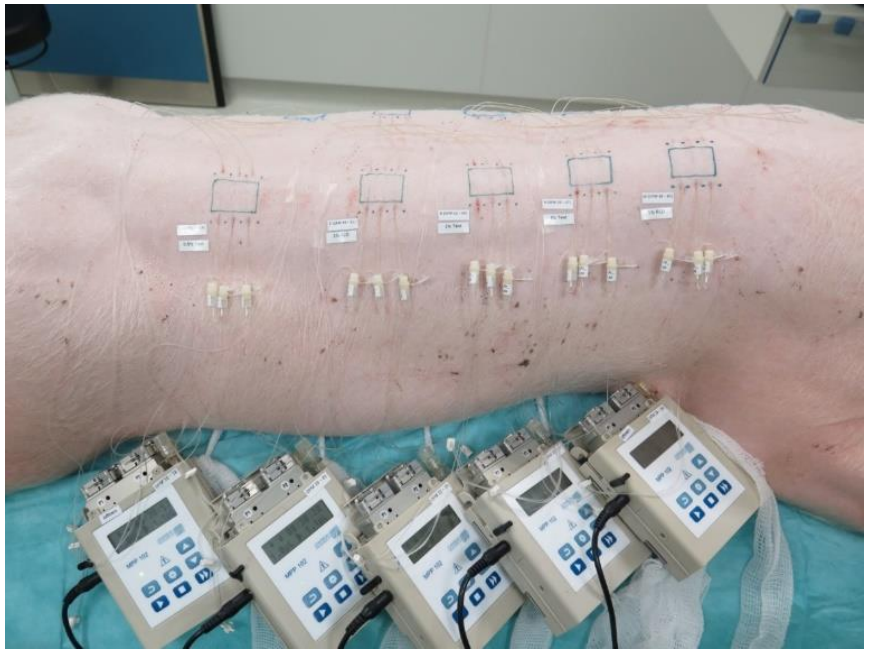
Dermal Open Flow Microperfusion
Case Study 3: Lead Drug Candidate Screening

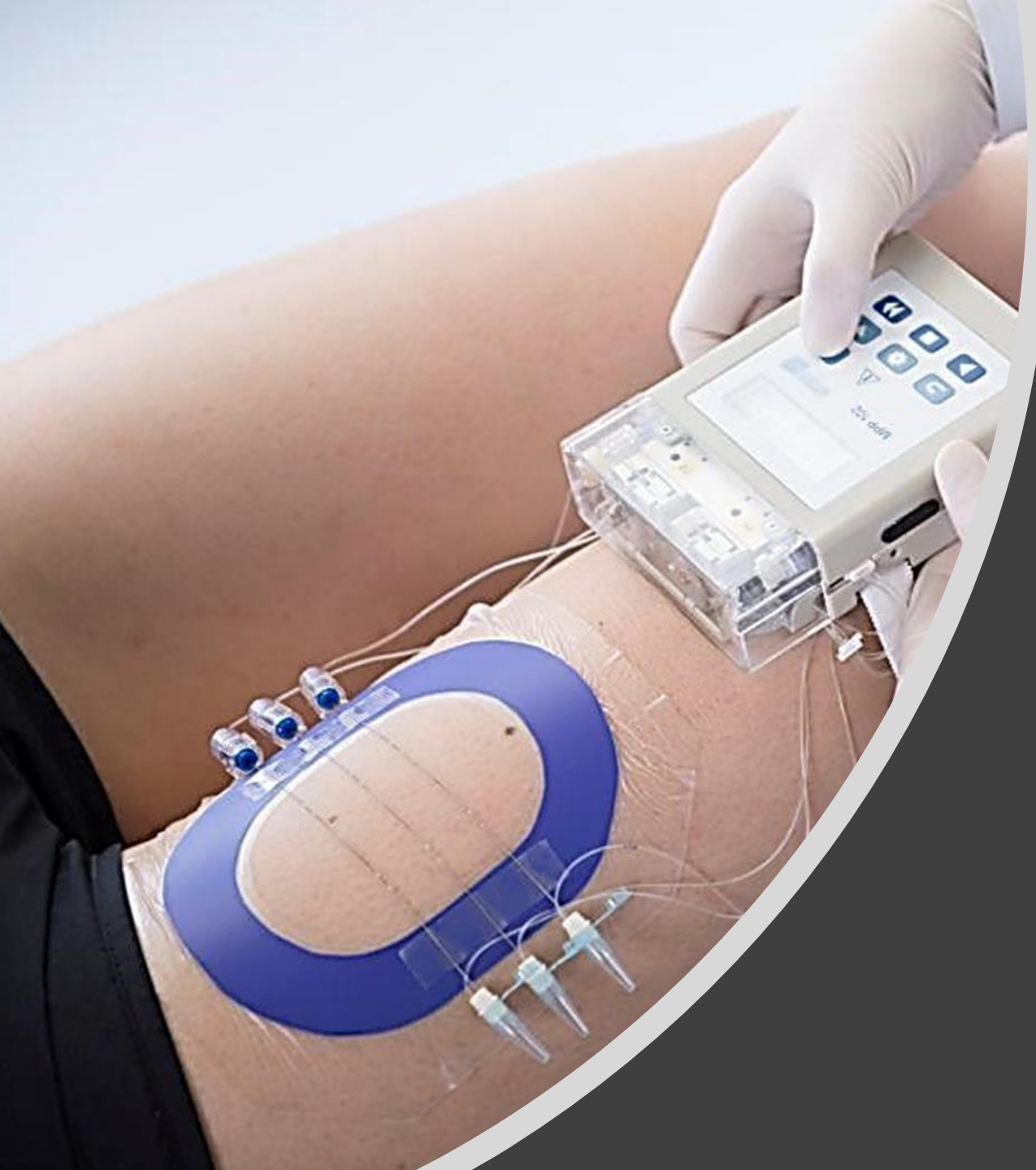
Head-to-Head comparison of topical drugs

- Same formulation → different API
- Same formulation → different concentration of API
- Same API → different formulation
- Same API → different microneedles

Read-out

- Dermal PK drug profile (dOFM)
- Dermal PD profile (dOFM)
- Skin drug concentration (biopsies)



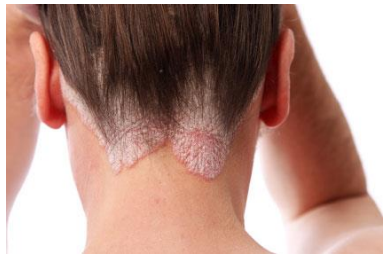


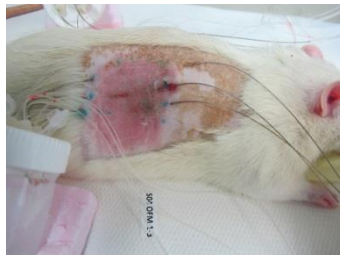
Open Flow Microperfusion

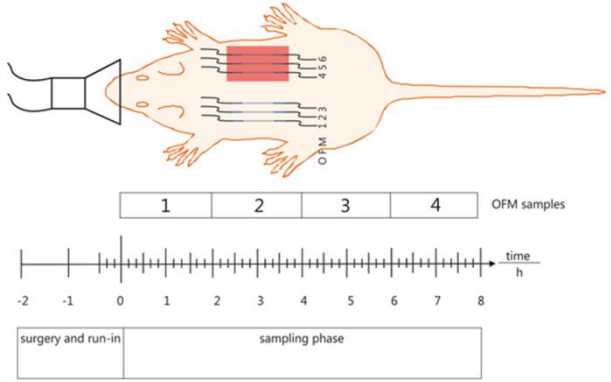
Case Study 4

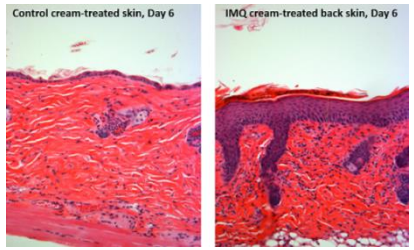
Psoriasis Rat Model

Dermal Open Flow Microperfusion
Case Study 4: Psoriasis Rat Model









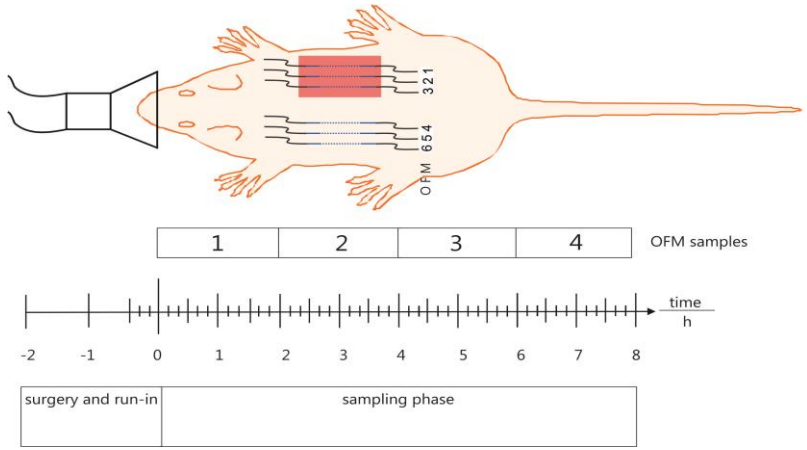
Dermal API PK for dose-response ← HPLC-MS/MS

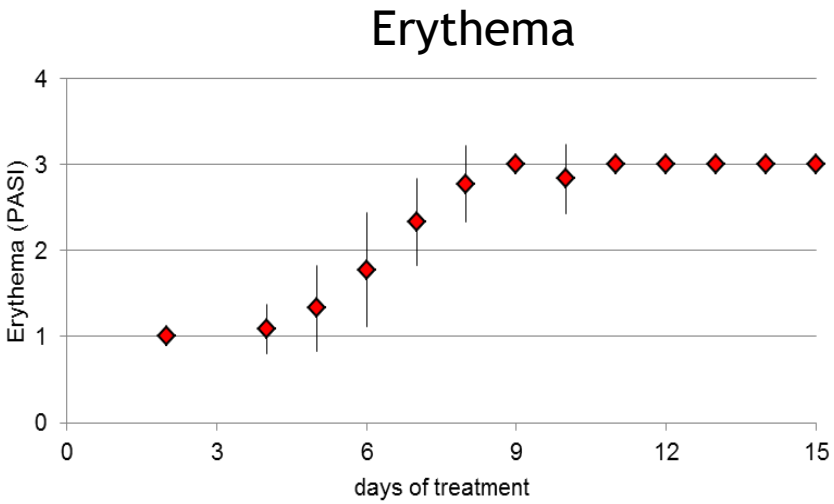
Dermal PD on cytokine level ← OLINK

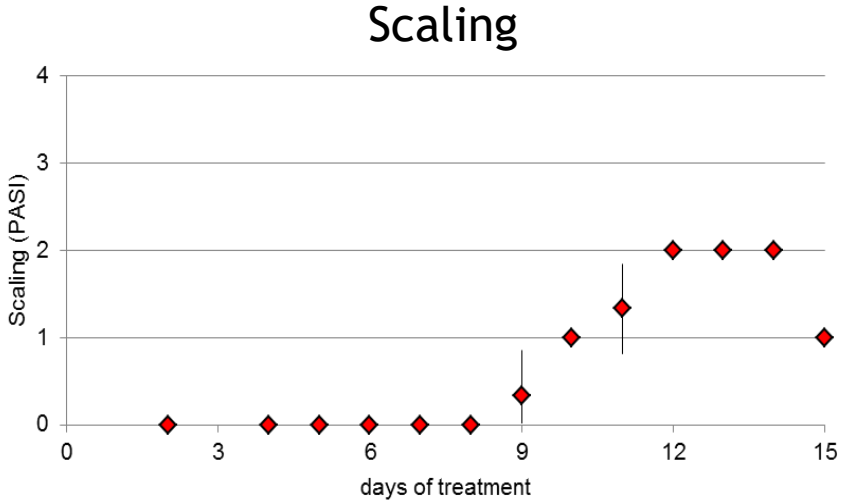
Dermal PD on immune cell level ← FACS

in-vivo effect of API on cytokine and immune cell level

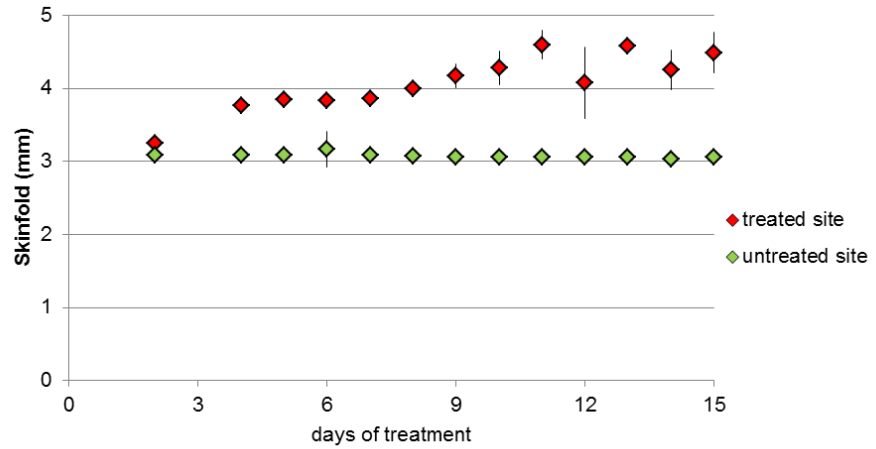
Dermal Open Flow Microperfusion
Case Study 4: Psoriasis Rat Model



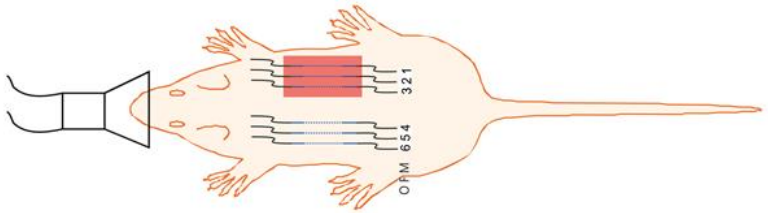




Rats were treated once daily with imiquimod cream on a demarcated 2 x 2 cm treatment site (topical dose of 30 mg cream/cm^2).

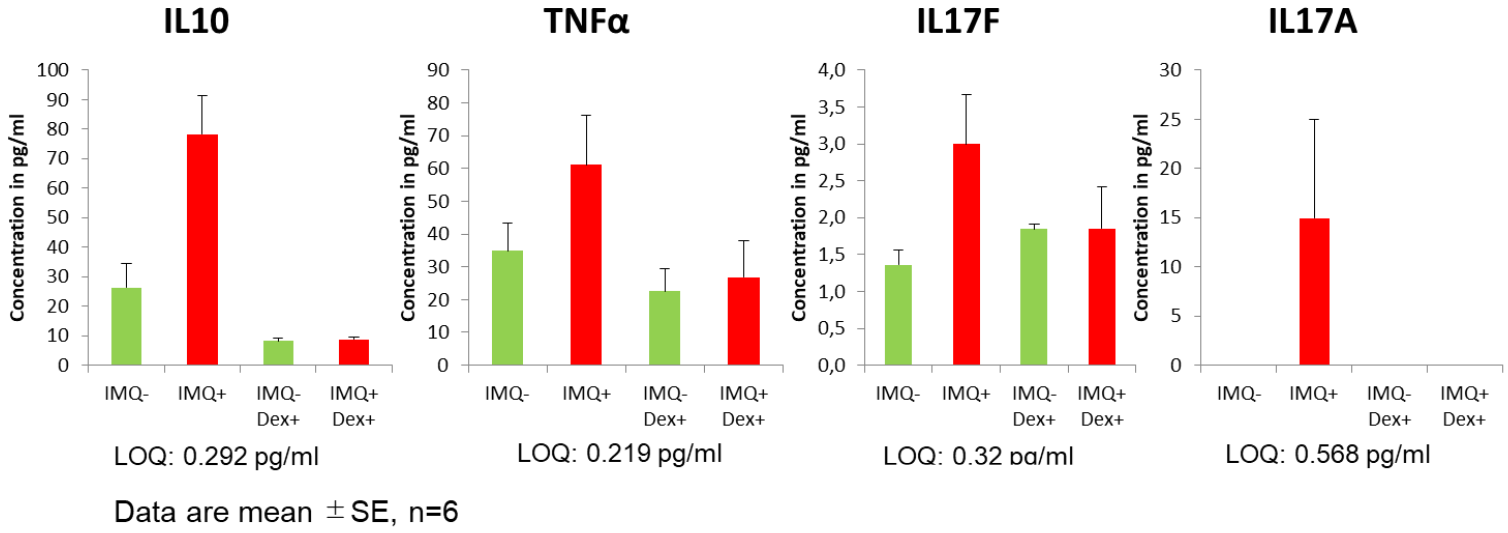


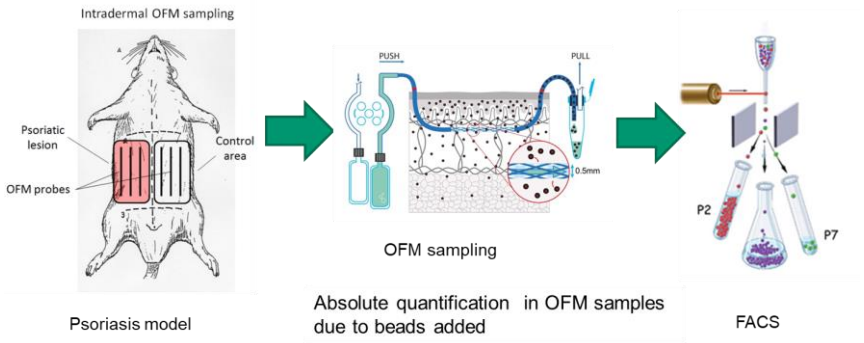
Dermal Open Flow Microperfusion
Case Study 4: Psoriasis Rat Model

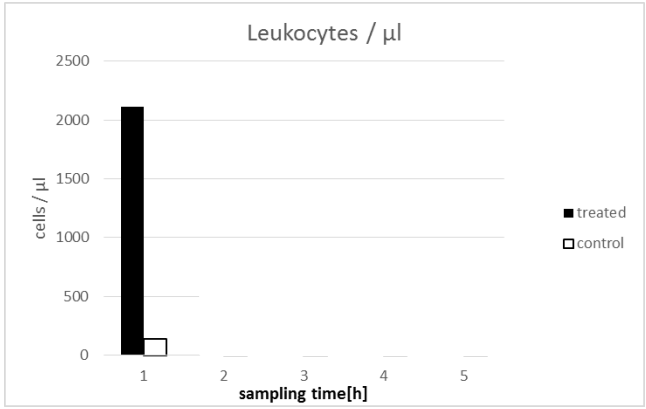


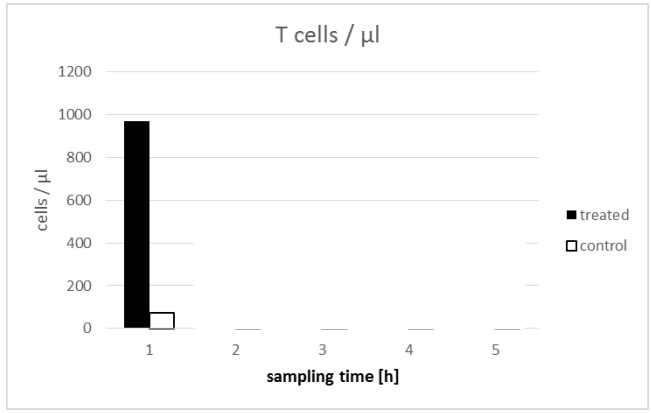
12 rats were treated with IMQ (and Dex) for 8 days

- Group1: 6 rats **without** inhibitory treatment
- Group2: 6 rats with additional treatment with a known therapeutic (**Dexamethasone 2mg/kg**)





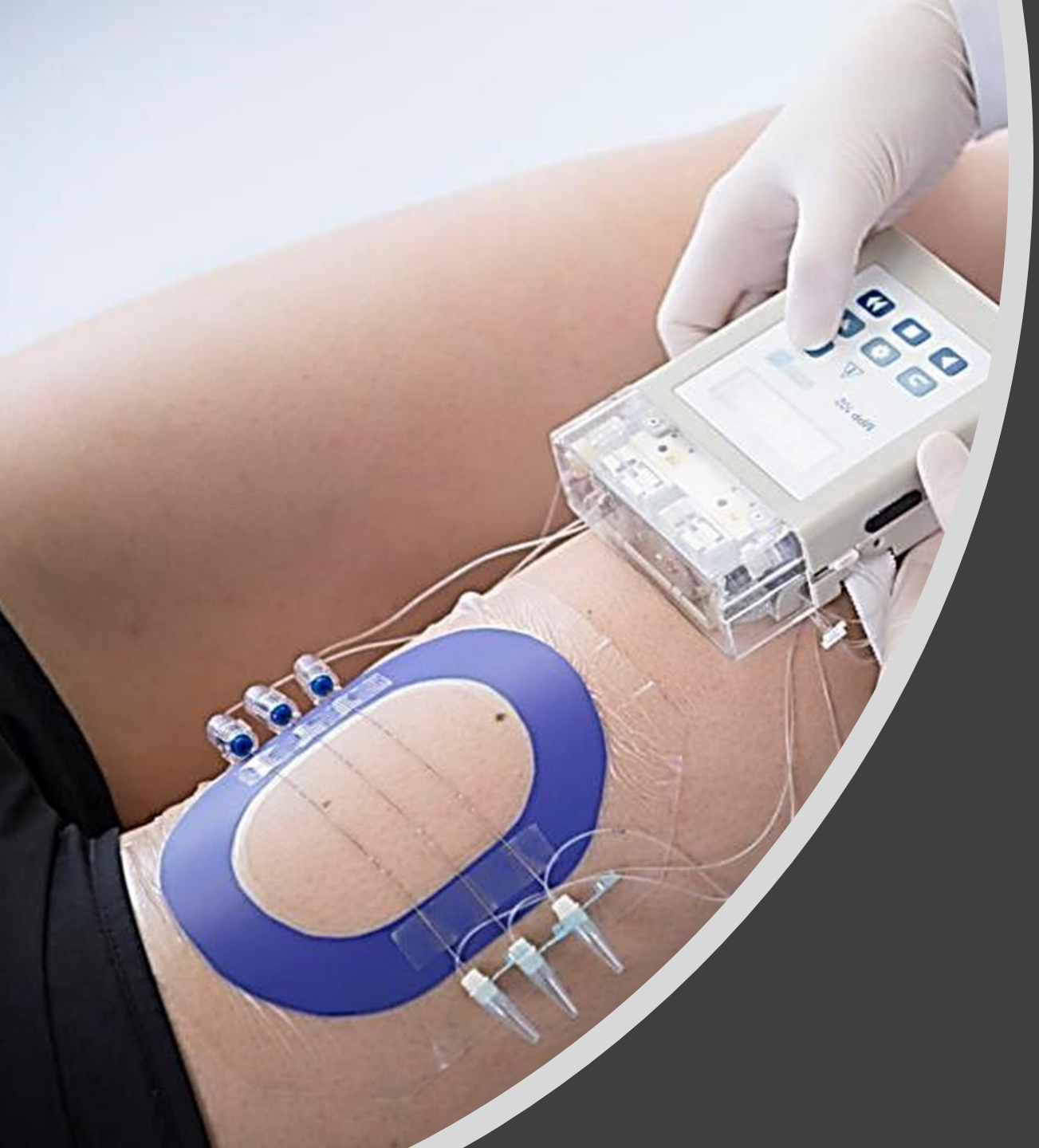




Dermal Open Flow Microperfusion
Clinical Studies

✓ dOFM studies are possible in healthy subjects and affected skin in patients

Subjects:	healthy and patients
Duration time:	up to 48 hours
Application sites:	up to 9 sites with 3 dOFM probes each
OFM material:	same material for preclinical and clinical
Time resolution:	determined by analytics (20 to 120 min)



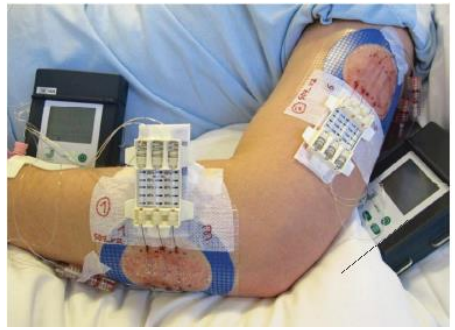
Open Flow Microperfusion

Case Study 5

Secukinumab

Dermal Open Flow Microperfusion
Case Study 5: Clinical Study Secukinumab

✓ **Dermal PK and PD for NCE in healthy volunteers and patients**

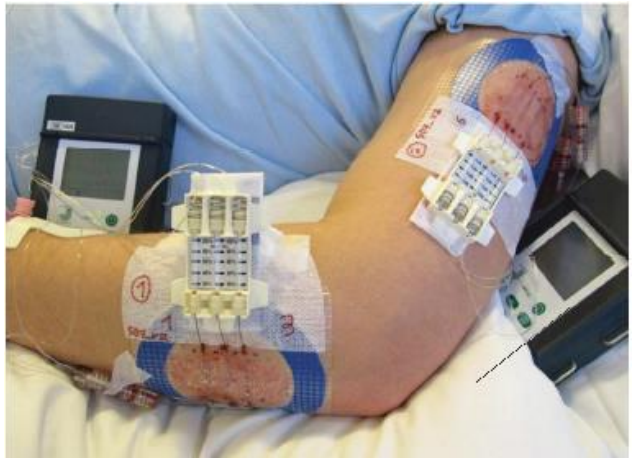


- Secukinumab, a fully human monoclonal antibody that selectively targets IL-17A, has demonstrated efficacy in phase 3 trials, within 16 weeks of initiation of treatment.
- The objective of this exploratory, single-center, open-label study (NCT01539213) was to further characterize the mechanism of action of secukinumab in the skin in
  - 8 healthy volunteers (Part 1)
  - 8 plaque psoriasis patients (Part 2)
- A single 300 mg s.c. dose of secukinumab was administered on Day 1 (after baseline samples were obtained) to 8 psoriasis subjects with suitable moderate to severe target plaques
- Dermal open flow microperfusion (dOFM), a minimally invasive technique that has been validated as a method of sampling the dermal interstitial fluid (dISF),1,2 was performed at baseline, Day 8 and Day 15 at lesional and non-lesional areas of skin from subjects with psoriasis.

NCE – new chemical identity

Dermal Open Flow Microperfusion
Case Study 5: Clinical Study Secukinumab

✓ **Dermal PK and PD for NCE in healthy volunteers and patients**



Primary Aim

- Absolute quantification of secukinumab in the dermis of healthy volunteers and psoriatic patients.

Secondary Aims

- Investigate if postulated signaling pathways are different in healthy and psoriatic patients in dermis → IL17a pathway.
- Investigate postulated mode of action → down stream IL17a marker.
- Investigate drug effect on a protein level → mediator for keratinocyte proliferation and angiogenesis and keratinocyte mobility.

Dermal Open Flow Microperfusion
Case Study 5: Clinical Study Secukinumab

✓ **Dermal PK and PD for NCE in healthy volunteers and patients**

Primary Aim

- Absolute quantification of secukinumab in the dermis of healthy volunteers and psoriatic patients.

Serum and Dermal Secukinumab Levels (µg/mL, mean ± SD)

Healthy Volunteers (n = 8)					
Serum		Dermal ISF[a,b]		Skin biopsy[c]	Blister fluid
Day 8	Day 15	Day 8	Day 15	Day 15	Day 15
36.1 ± 10.5	35.0 ± 10.5	7.76 ± 1.30	8.02 ± 3.23	10.40 ± 3.97	6.89 ± 2.26

- Dermal ISF concentrations ~22% of serum concentration.
- Dermal concentration by OFM, blister fluid, biopsies are comparable.

Serum and Dermal Secukinumab Levels (µg/mL, mean ± SD)

Psoriatic Subjects (n = 8)					
Serum		Dermal ISF[a,b]			
Day 8	Day 15	Day 8		Day 15	
		L	NL	L	NL
21.1 ± 4.3	21.2 ± 4.9	6.76 ± 2.68	8.34 ± 3.35	5.65 ± 1.80	6.39 ± 3.35

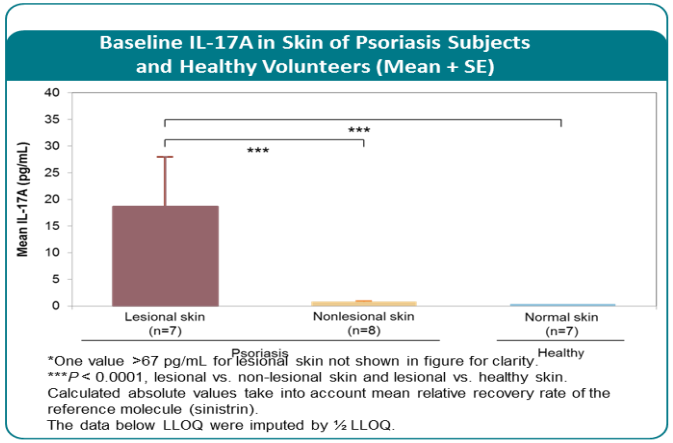
- Dermal ISF concentrations are 28-39% of serum concentration.
- Dermal ISF concentrations on day 8 and day 15 are similar.

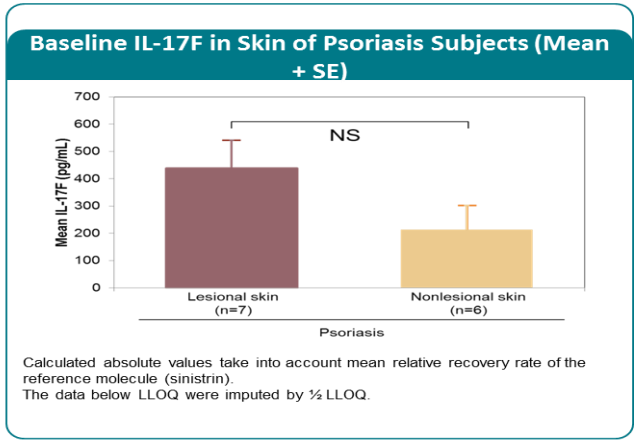
Dermal Open Flow Microperfusion
Case Study 5: Clinical Study Secukinumab

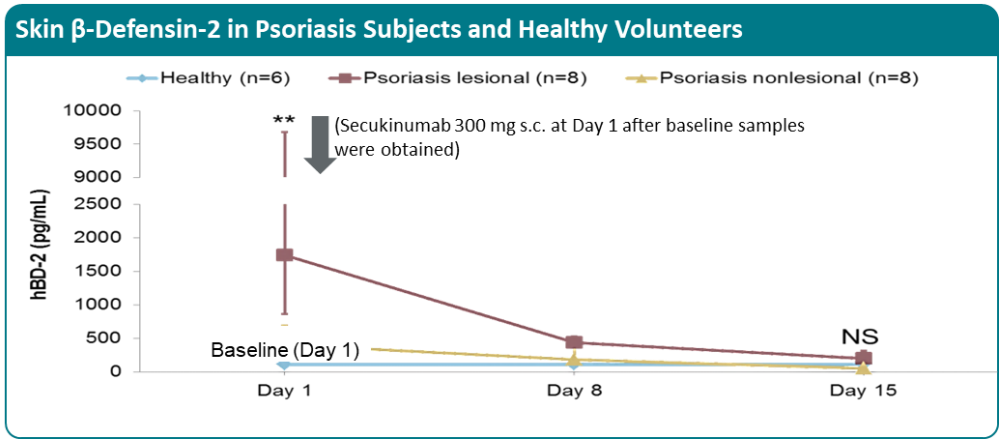
✓ **Dermal PK and PD for NCE in healthy volunteers and patients**

Secondary Aim

- Investigate signaling pathways in healthy volunteers and psoriatic patients in the dermis.







IL-17A, but not IL-17F, is significantly higher in psoriatic lesional skin compared with non-lesional skin or skin of healthy volunteers.

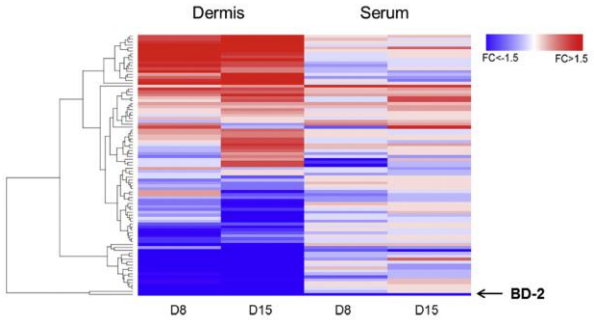
ß-defensin-2 protein levels are elevated in psoriatic lesional skin and serum and decrease rapidly in response to secukinumab treatment.

Dermal Open Flow Microperfusion
Case Study 5: Clinical Study Secukinumab

✓ **Dermal PK and PD for NCE in healthy volunteers and patients**

Secondary Aim

- Investigate signaling pathways in healthy volunteers and psoriatic patients in the dermis.



	Fold change relative to baseline			
	Dermis (dISF)		Serum	
Protein	Day 8	Day 15	Day 8	Day 15
Top 10 downregulated				
BD-2	−18.73	−32.20	−3.95	−3.66
MMP-1	−6.20	−15.19	−1.11	1.04
IL-1β	−2.71	−5.47	1.14	1.14
IL-1 receptor antagonist (IL-1ra)	−2.19	−4.37	−1.47	−2.32
MMP-8	−1.91	−3.42	−1.16	−1.07
Myeloperoxidase	−1.18	−3.20	−1.27	−1.18
CXCL1 (GRO-α, *CXCL1*)	−2.63	−3.13	−1.08	−1.17
Lipocalin-2 (NGAL, *LCN2*)	−2.14	−2.98	−1.11	−1.12
CCL20 (Macrophage inflammatory protein 3α, *CCL20*)	−2.62	−2.64	−1.24	1.45
CXCL5 (ENA-78, *CXCL5*)	−3.00	−2.50	1.05	−1.02
Other proteins of interest				
CXCL3 (GRO-γ, *CXCL3*)	−1.61	−2.20	−1.16	−1.08
CCL1 (I-309, *CCL1*)	−1.34	−1.88	1.09	1.03
TNF-α	1.00	1.18	1.04	1.03
Top 5 upregulated				
Endoglin	2.51	2.52	1.04	1.08
Leptin	2.59	2.62	1.09	1.39
Adiponectin (Acrp-30)	1.50	2.72	1.13	−1.04
Eotaxin-2 (*CCL24*)	1.56	2.77	1.06	1.14
IgE	1.92	3.19	−1.00	−1.06

Dermal Open Flow Microperfusion
Case Study 5: Clinical Study Secukinumab

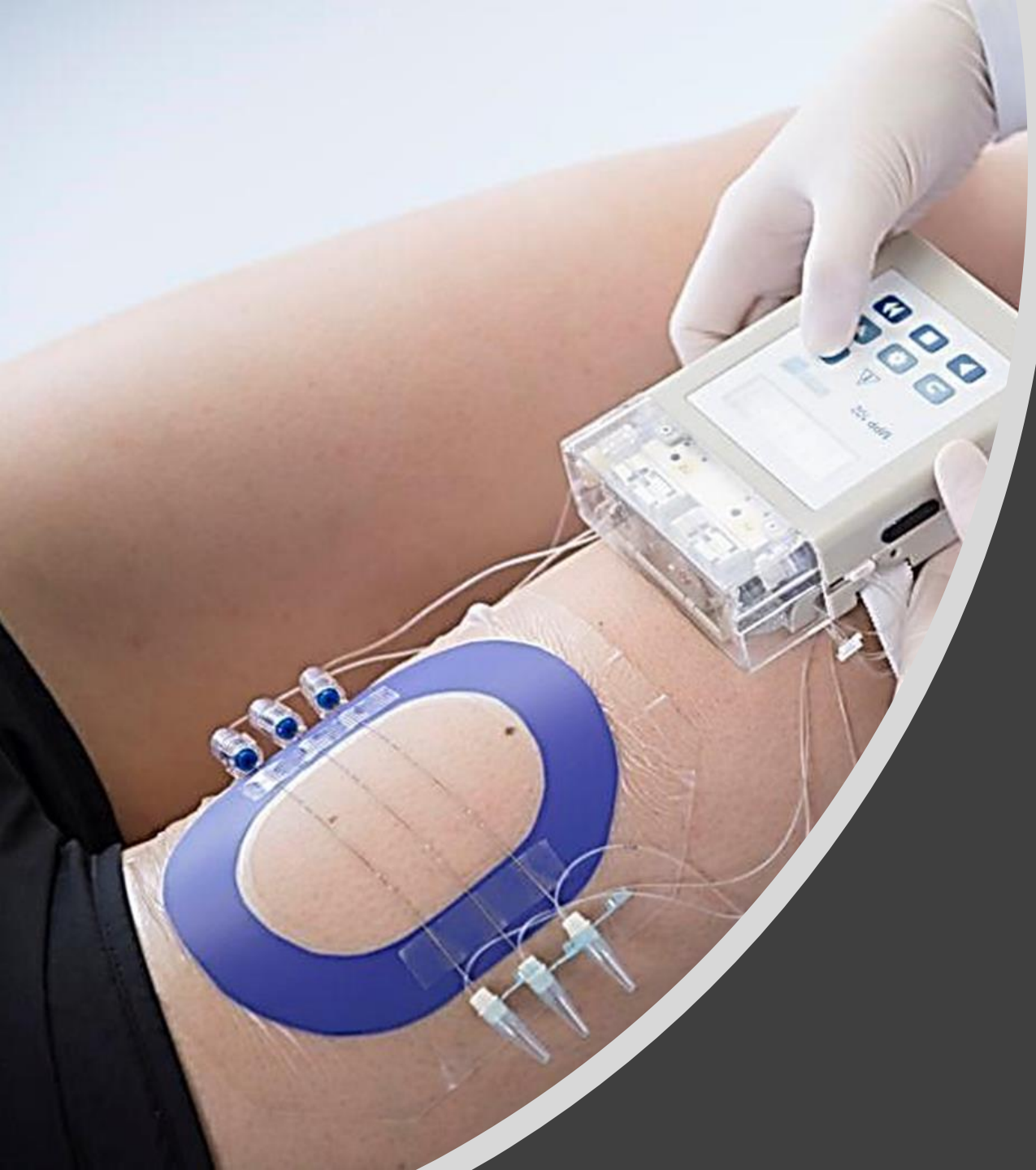
✓ This dOFM clinical study was accepted by several agencies as Proof-of-Mechanism

Conclusions on Pharmakokinetics

- Substantial levels of secukinumab are observed in skin suggesting the potential for local action.
- Secukinumab ISF distribution into psoriasis lesional and non-lesional skin is similar and is higher than ISF distribution in healthy control skin.

Conclusions on Pharmakodynamics

- Key molecular factors and processes implicated in the pathophysiology of psoriasis were positively impacted in psoriatic skin within 7 days of treatment.
- Secukinumab concentration in skin is sufficient to neutralize IL-17a in psoriatic skin
- Secukinumab affected the expression of a number of pro-inflammatory cytokine.
- Investigate signaling pathways in healthy volunteers and psoriatic patients in dermis



Open Flow Microperfusion

Case Study 6

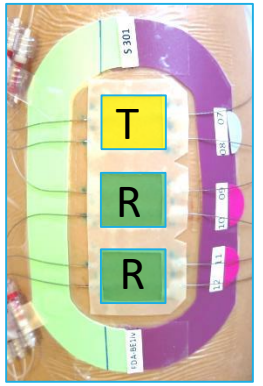
Bioequivalence of Topical Generics

Dermal Open Flow Microperfusion
Case Study 6: Bioequivalence of Topical Generics

✓ dOFM as a new possibility for showing bioequivalence of a generic to its RLD

Clinical study outline

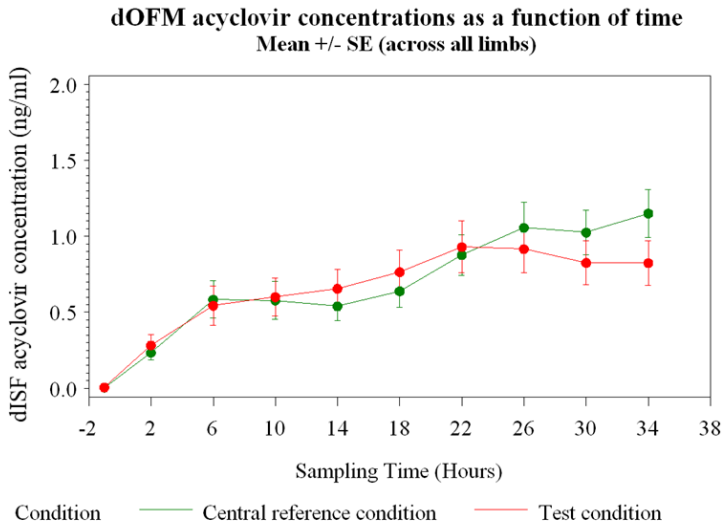
- 20 healthy subjects
- Reference: Zovirax® US
- Test: Aciclovir-1A Pharma Austria
- 2 application triplets per subject
- 15 mg/cm^2 cream application
- 36 hours dOFM sampling time

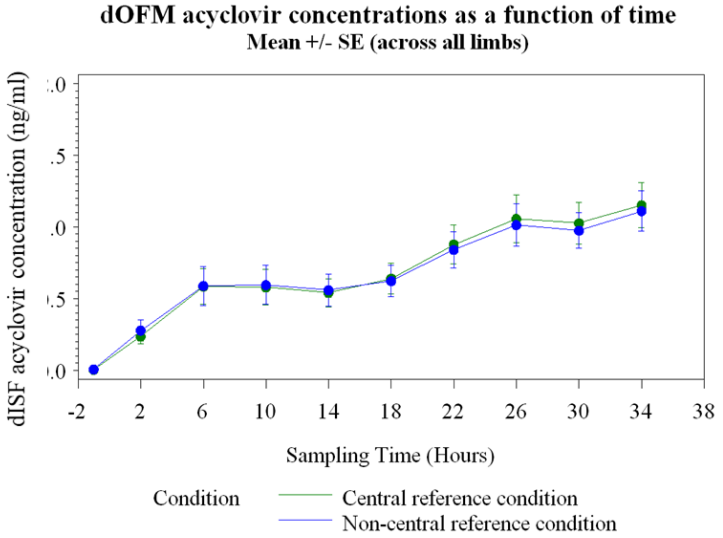


Funding for this project was made possible, in part, by the Food and Drug Administration through grants 1U01FD004946 and 1U01FD005861. The views expressed in this presentation do not reflect the official policies of the Food and Drug Administration, or the Department of Health and Human Services; nor does any mention of trade names, commercial practices, or organization imply endorsement by the United States Government. The human research study was approved by the FDA Research Involving Human Subject Committee (RIHSC) and the local Institutional Review Board (IRB) of the Medical University Graz, Austria

Dermal Open Flow Microperfusion
Case Study 6: Bioequivalence of Topical Generics

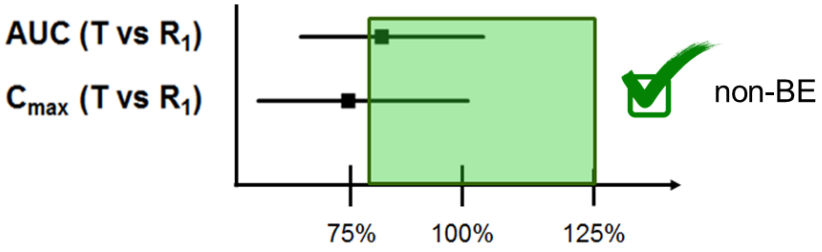
✓ dOFM allows measurement of topical dermal drug BE on PK level

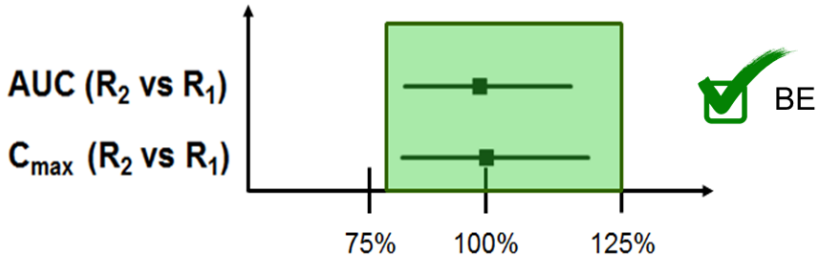




"Open Flow Microperfusion as a Dermal Pharmacokinetic Approach to Evaluate Topical Bioequivalence"

Bodenlenz et al. Clin. Pharmacokinet. 2017 doi: 10.1007/s40262-016-0442-z.- **OPEN ACCESS**





Boutique CRO
for tissue specific PK and PD

aOFM
adipose tissue OFM

cOFM
cerebral OFM

dOFM
dermal OFM

IVRT

EN ISO 9001 | EN ISO 13485:2003 | GLP | Data Management | Statistics

GCP

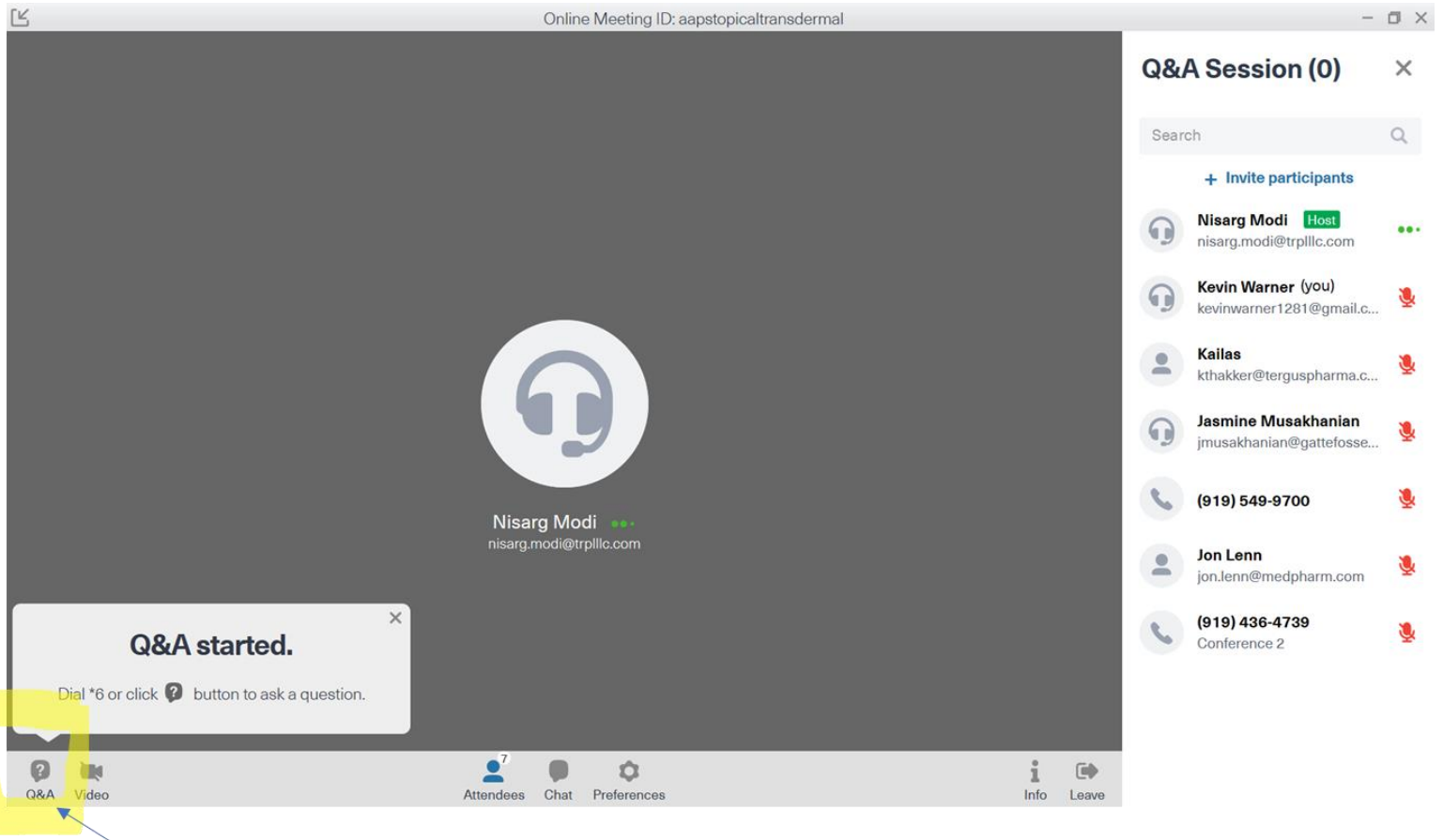
Thank you for your attention

- Dr. Frank Sinner
- JOANNEUM RESEARCH Forschungsgesellschaft mbH
- HEALTH – Institute for Biomedicine and Health Sciences
- Neue Stiftingtalstrasse 2, 8010 Graz
- +43 316 876-4000
- frank.sinner@joanneum.at
  www.joanneum.at/health

aaps Communities

AAPS Topical and Transdermal Community

How to Ask A Question…



Click this icon or dial *6 to get in the queue to ask a question.

aaps Communities

AAPS Topical and Transdermal Community

Thank You for Joining Us Today!

Producer

Nisarg Modi, PhD

Moderator

Narasimha Murthy, PhD

Presenter

Frank Sinner, PhD

aaps Communities

AAPS Topical and Transdermal Community

FREE MONTHLY WEBINAR SERIES

Date	Speaker
January 10, 2020	Dr. Frank Sinner, Joanneum Research, Austria *Use of Skin PK & PD in Drug Development*
February 7, 2020	Prof. Michael Roberts, University of Queensland, Australia *Mathematical Modeling of Skin Absorption and Transport*
March 13, 2020	Dr. Leandro Santos, Incyte, USA *Dermal Drug Discovery: What Physicochemical Properties Can (and Cannot) Do*
April 10, 2020	Dr. Thean Yeoh, Pfizer, USA *Topical Formulations: From the Idea/Concept to the Marketed Formulation*
May 8, 2020	Dr. Majella Lane, University of London, UK *Topical Formulations: From the Idea/Concept to the Marketed Formulation*