

Suction Blister Method

THE *IN VIVO* ► *EX VIVO* ► *IN VITRO* APPROACH FOR YOUR INNOVATIVE CLAIM

In addition to standard *in vitro* methods including cell culture techniques and organotypic skin models, we offer an innovative approach that allows us to combine *in vivo* treatment via *ex vivo* sampling with a large range of available *in vitro* analyses. The obtained samples (tissue and tissue fluids) can be subjected to biochemical tests such as ELISA tests, immunohistological, histological and molecular tests:

IN VIVO TOPICAL TREATMENT

EX VIVO SAMPLES

(Complete viable epidermis with intact basal cell layer plus interstitial fluid within the blister)

IN VITRO ANALYSES

(Biochemistry, cell biology, immunology, molecular biology, histology)

Sampling via punch biopsies provides epidermal and dermal material but requires a local anaesthetisation and leaves scars. The suction blister method, initially developed by Kiistala *et al.* in 1968, is a valuable alternative that is considered to be non-invasive.

Approved by the Ethics Committee in a general vote, we have established this method as a standard trial that allows us to obtain epidermal material for subsequent *in vitro* analyses.

As the name implies, blisters are artificially induced. Custom-made plexiglass chambers with round openings are attached to the respective test areas and connected to a vacuum pump:



Within approximately 2 hours, the epidermis is slowly detached from the underlying dermis. The induced blister fills with fluids from the surrounding tissue (interstitial fluid):



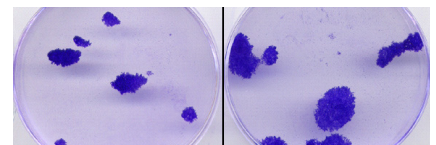
The procedure is painless – causing itching at worst – and the wounds heal without forming scars within 7 to 10 days. However, some subjects develop hyperpigmentation on the former wound that can last for some months or longer. After removal of the chambers, the blister fluid (interstitial fluid) can be extracted with a sterile syringe and the blister roof carefully removed using sterile instruments.

For the *in vitro* analyses, a large variety of methods such as immunohistological stainings and a continuously extended list of commercially available analysis kits are available.

Parameters that can be estimated using *in vitro* enzyme-linked immunosorbent assays (ELISA), immunohistological or other methods:

- **Anti-pollution (smoke/UV model):**
Analysis of markers such as MMPs and/or interleukins (ELISA) or analysis of the lipid barrier (REM, TEM, HPTLC, in cooperation with Microscopy Services Dähnhardt GmbH)
- **Structural proteins:**
Pro-collagen I as a measure for the *de novo* synthesis of collagen I, fibrillin and elastin (ELISA)
- **Extracellular matrix:**
Hyaluronic acid, MMPs such as Matrix Metallo-proteinase-1 (ELISA)
- **Oxidative stress:**
8-isoprostane as a marker for lipid peroxidation, carbonyl proteins as a marker for protein oxidation (ELISA)
- **Cytokines:**
e.g. IL-6, IL-8 and TNF-alpha (ELISA)
- **UV-induced DNA damage:**
Immunohistological detection of DNA damage induced by UV light (thymine dimers)
- **UV-induced immunosuppression:**
Langerhans cells and functionality of the skin immune system
- **Differential gene expression via Real Time Quantitative PCR**
- **Stem cell activation:**
Number of p63-positive epidermal cells by immunohistological analysis, colony forming efficiency by cultivation of epidermal stem cells (colony-forming assays)

Estimation of colony-forming efficiency by cultivation of epidermal stem cells (colony-forming assay)



Placebo

Verum

Please contact us for further information, consultation or a detailed proposal.

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