RELEVANT, RELIABLE, READY-TO-USE AND ROBUST
validated & REACH compliant alternative methods for the evaluation of the human safety and efficacy
T-Skin is an *in vitro* reconstructed skin which consists of a dermal equivalent with human fibroblasts overlaid by a stratified, well differentiated epidermis derived from normal human keratinocytes. The model is cultured on an inert polycarbonate filter. This model exists at different stages of maturity.

### TISSUE MODEL & FORMAT

*In vitro* and *in vivo* views of T-Skin model.

6 well plate

### CHARACTERIZATION

As human skin, the full thickness T-Skin model shows distinct compartments such as the epidermis and the dermis, linked by a functional DEJ (Dermal–Epidermal Junction). In the epidermis, keratinocytes express the proliferation marker Ki-67. Transglutaminase-1 marker is found in Stratum Spinosum as well as Cytokeratin-10. Loricrin and filaggrin are present in Stratum Granulosum. Collagen-IV, Collagen-VII, Collagen-XII, Laminin-5 and Perlecan characterize the T-Skin DEJ. Fibroblasts, the primary cell type in the dermis, express Procollagen-I.
APPLICATIONS

- Phototoxicity
- Photoprotection
- Anti Aging
- Assessment of medical devices
- Epidermal differentiation
- Percutaneous absorption
- Retinoid activity test

REFERENCES

Reconstructed skin to create in vitro flexible models of skin aging: new results and prospects.

In vitro and in vivo studies with tetra-hydro-jasmonic acid (LR2412) reveal its potential to correct signs of skin ageing.

New Combination of Ultraviolet Absorbers in an Oily Emollient Increases Sunscreen Efficacy and Photostability.

Comparison of xenobiotic metabolizing enzyme activities in ex vivo human skin and reconstructed human skin models from SkinEthic.
**DESCRIPTION**

The SkinEthic™ RHE-LC model is a standard epidermal model in which Langerhans cells progenitors have been integrated. During the tissue reconstruction, these immature cells have differentiated into antigen-presenting dendritic cells expressing the specific protein CD1a. They are mostly located and evenly spread within the supra-basal epidermal layer. The SkinEthic™ RHE-LC model is therefore expected to be a useful tool for skin immune response studies.

**TISSUE MODEL & FORMAT**

*CDla markers on epidermal sheet*

*In vitro*

0.5 cm²

**CHARACTERIZATION**

*CDla markers*
DESCRIPTION

CMM models are epidermal or epithelial models such as the reconstructed human cornea on the new Cell Migration Model (CMM) insert. This insert has a double porosity which makes it possible to study the migration of immunocompetent cells in reconstructed human tissue. It is now possible to study in these reconstructed tissues the infiltration mechanisms and interactions with the epithelial cells of immunocompetent, macrophages, lymphocytes or dendritic cells by reproducing in vitro the phenomena observed in certain chronic inflammatory pathologies.

TISSUE MODEL & FORMAT

In vitro RHE-CMM

0.5 cm²

CHARACTERIZATION

Histological section of SkinEthic™ RHE-CMM with infiltration of activated CD4 + lymphocytes. Suprabasal localization of lymphocytes 24h after infiltration revealed by double immunolabeling (CD3 in blue and CD45 in red) on serial sections. (Images courtesy of GSK)
DESCRIPTION

SkinEthic™ RHE is an in vitro Reconstructed Human Epidermis from normal human keratinocytes cultured on an inert polycarbonate filter at the air-liquid interface, in a chemically defined medium. This model is histologically similar to in vivo human epidermis. This model exists at different stages of maturity.

TISSUE MODEL & FORMAT

The reconstructed human epidermis expresses the major differentiation markers (filaggrin and keratin 5 in granular cell layers, keratin 10 and involucrin in supra basal cell layers and loricrin in upper granular cell layers), as well as expressing the basement membrane markers (type IV collagen; alpha 6 beta 4 integrin, BP antigen, and laminin V).

The lipid profile analysis shows the presence of major epidermal lipid classes. Ultra-structural study shows characteristic bi layered lipid lamellae in the intercellular space of the stratum corneum.

CHARACTERIZATION
APPLIEDS

SKIN IRRITATION
*ECVAM Validation 2008
OECD TG 439
SKIN CORROSION
*ECVAM Validation 2006
OECD TG 431
UV EXPOSURE
BACTERIAL ADHESION

- Assessment of medical devices
- Epidermal differentiation
- Percutaneous absorption
- Retinoid activity test

DNA DAMAGE
OMICS
PERMEABILITY

REFERENCES

In vitro patch test using non-invasive endpoints.
De Brugerolle de Fraissinette A, Rosdy M, Tornier C, SOT 2011

L’Oréal commitment in the development, evaluation and validation of screening and testing approaches contributing to the 3Rs.

An Evaluation of the EpiSkin™ and SkinEthic™ RHE test methods for predicting dermal toxicity using OECD TG404.

Assessment of the skin irritation potential of chemicals by using the SkinEthic™ RHE model & the common skin irritation protocol evaluated in the ECVAM skin irritation validation study.
DESCRIPTION

EpiSkin™ is an in vitro reconstructed human epidermis from normal human keratinocytes cultured on a collagen matrix at the air-liquid interface. This model is histologically similar to the in vivo human epidermis. This model exists at different stages of maturity.

TISSUE MODEL & FORMAT

CHARACTERIZATION

The reconstructed human epidermis expresses the major differentiation markers (filaggrin and keratin 5 in granular cell layers, keratin 10 and involucrin in supra basal cell layers and loricrin in upper granular cell layers), as well as expressing the basement membrane markers (type IV collagen; alpha 6 beta 4 integrin, BP antigen, and laminin V).

The lipid profile analysis shows the presence of major epidermal lipid classes. Ultra-structural study shows characteristic bi layered lipid lamellae in the intercellular space of the stratum corneum.
APPLICATIONS

- SKIN IRRITATION (*ECVAM Validation 2007 OECD TG 439)
- SKIN CORROSION (*ECVAM Validation 1998 OECD TG 431)
- UV EXPOSURE
- BACTERIAL ADHESION
- DNA DAMAGE
- OMICS
- PERMEABILITY

- Genotoxicity (micronucleus test & COMET assay) for topically applied compounds or formulations

REFERENCES

L’Oréal commitment in the development, evaluation and validation of screening and testing approaches contributing to the 3Rs.

An Evaluation of the EpiSkin™ and SkinEthic™ RHE test methods for predicting dermal toxicity using OECD TG404.

Evaluation of the in vitro EpiSkin™ and SkinEthic™ RHE Skin Irritation test methods for hazard identification of chemicals.

The ECVAM International Validation study on In Vitro Test for skin Corrosivity. 2. results and Evaluation by the Management Team.
DESCRIPTION

The SkinEthic™ HCE model is composed of transformed human corneal keratinocytes cultivated on an inert polycarbonate filter at the air liquid interface in a chemically defined medium. The reconstructed tissue forms a stratified and well organized epithelium which is structurally, morphologically and functionally similar to the human cornea with presence of basal, wing and mucus production cells.

TISSUE MODEL & FORMAT

CHARACTERIZATION

Ultra-structural features show the typical presence of a columnar basal cell layer, 2-3 layers of transitional wing cells, and 2-3 layers of superficial squamous cells.

The reconstructed human corneal epithelium secretes the same mucins that are being found in the human cornea in vivo and expresses CD 44 and keratin.
APPLICATIONS

The SkinEthic™ HCE method has been peer reviewed by ESAC (2016-02) and is now ready to be considered for regulatory use. In such context, the implementation of the SkinEthic™ HCE method, as a validated reference method in the OECD test guidelines TG 492 is already considered.

The draft of TG492 on Reconstructed human Cornea-like Epithelium test method for eye hazard potential is currently under review by the states members of OECD with an approval expected by summer 2017. An updated draft is available upon request.

- Eye Irritation Test (EIT) Method validated by EURL-ECVAM and under OECD TG 492 for identification of chemicals not requiring classification for eye hazard (UN GHS)
- EIT implemented in the draft OECD GD for serious damage and eye irritation
- Ocular irritation assay (as a screening tool for products selection)
- Corneal permeability and metabolism
- Corneal differential display i.e. mucin production

REFERENCES

Multi-laboratory evaluation of SkinEthic HCE test method for testing serious eye damage/eye irritation using solid chemicals and overall performance of the test method with regard to solid and liquid chemicals testing

In vitro assessment of eye irritancy using the Reconstructed Human Corneal Epithelial SkinEthic™ HCE model: Application to 435 substances from consumer products industry.

Occludin gene expression as an early in vitro sign for mild eye irritation assessment.

Evaluation of the cytotoxic effects of ophthalmic solutions containing benzalkonium chloride on corneal epithelium using an organotypic 3-D model.

Three –Dimensionnal Construct of the Human Corneal Epithelium for In vitro Toxicology.
DESCRIPTION

The SkinEthic™ RHPE model is composed of normal human keratinocytes cultivated in the presence of melanocytes of phototype II, IV or VI, localized in the basal layer. The different tanning degrees of these constructs correspond macroscopically to 3 different phototypes of human skin. This model exists at different stages of maturity.

TISSUE MODEL & FORMAT

![Phototype II](Image1) ![Phototype IV](Image2) ![Phototype VI](Image3)

0.5 cm² 4.0 cm²

CHARACTERIZATION

The SkinEthic™ RHPE model presents a histological morphology comparable to the *in vivo* human tissue, consisting in the presence of a multi-layered, stratified and pigmented epidermis. Melanocytes are localized in the basal cell layer interspersed with basal cell keratinocytes. Moreover, melanin is distributed in the basal layer melanocytes and in upper layer keratinocytes similarly to that seen in normal human skin, reflecting its transfer into surrounding proliferating keratinocytes.

The SkinEthic™ RHPE model exhibits a homogeneous tanning demonstrating the functionality of the epidermal melanin unit *in vitro* and that the obtained phototype is determined by the melanocyte phototype, i.e. the rate of constitutive melanin synthesis.

![in vitro](Image4) ![Fontana Masson staining](Image5)
APPLICATIONS

- The phototype VI model is generally used to evaluate the whitening potential of skin care formulations and phototypes II and IV models to assess the induction of pigmentation by UV exposure and/or by chemical modulators.

- In both pigmentation and depigmentation assays, repeated topical or systemic application result in a modulation of melanogenesis characterized by an increase or a decrease of melanin synthesis, respectively.

REFERENCES

Reconstructed Human Pigmented Epidermis (RHPE): an in vitro model for the evaluation of melanogenesis.

Sepicalm VG, a new skin lightening enable to modulate melanogenesis-related genes and to prevent UV-induced pigmentation thanks to its soothing properties.

Melanocyte containing human organotypic epidermis as a model to evaluate toxicity of melanin binding substances.

Human pigmented epidermis reconstructed in chemically defined medium used for evaluation of modulation of pigmentation.
DESCRIPTION

The SkinEthic™ HGE model is composed of normal human gingival cells cultivated on an inert polycarbonate filter at the air liquid interface in a chemically defined medium. This model is histologically similar to the outer cell layers of the human gum.

TISSUE MODEL & FORMAT

CHARACTERIZATION

The reconstructed human gingival epithelium shows a comparable profile of biomarkers than in the in vivo situation such as the expression of filaggrin in the granular cell layers, involucrin, keratin 6, keratin 10, keratin 13 and keratin 16 in the supra basal cell layers in the basal cells.

Filaggrin detection

Keratin 13 detection
APPLICATIONS

GINGIVAL CARE  BACTERIAL ADHESION  OMICS

REFERENCES


DESCRIPTION

The SkinEthic™ HOE model is composed of TR146 cells (derived from a squamous cell carcinoma of the buccal mucosa) cultivated on an inert polycarbonate filter at the air liquid interface in a chemically defined medium. This model forms an epithelial tissue devoid of stratum corneum, resembling histologically to the mucosa of the oral cavity.

TISSUE MODEL & FORMAT

CHARACTERIZATION

The reconstructed oral epithelium expresses keratin 6 and keratin 16 and differentiation markers such as involucrin like in the *in vivo* situation.
APPLICATIONS

- Oral irritation assay (oral care products, dental materials)
- Oral anti-inflammatory assay (oral care products)
- *In vitro* candidosis research
- Drug delivery

REFERENCES

Evaluation of an oral care product safety screening program utilizing the *in vitro* SkinEthic Human Gingival Epithelium (RHG) and Oral Buccal (RHO) models.

A Biphasic Innate Immune MAPK Response Discriminates between the Yeast and Hyphal Forms of *Candida albicans* in Epithelial Cells.

Quantitative expression of the *Candida albicans* secreted aspartyl proteinase gene family in human oral and vaginal candidiasis.

Phenotypic screening, transcriptional profiling, and comparative genomic analysis of an invasive and non-invasive strain of *Candida albicans*.
DESCRIPTION

The SkinEthic™ HVE model is composed of A431 cells (derived from a vulval epidermoid carcinoma) cultivated on an inert polycarbonate filter at the air liquid interface in a chemically defined medium. This model is histologically similar to the in vivo vaginal mucosa.

TISSUE MODEL & FORMAT

The reconstructed vaginal epithelium expresses keratin 6 and keratin 16 and differentiation markers such as involucrin, as the in vivo epithelium does.

CHARACTERIZATION
APPLICATIONS

- Vaginal irritation assay of the safety profile of topically applied gynaecological compounds or products
- Integrated in vitro vaginal safety screening approach for bath & body wash products
- Bacterial or viral adhesion screening for antibiotics or antiviral compounds or products used for example in the treatment of vaginal candidosis

REFERENCES

Integrated in vitro vaginal safety approach for bath and body wash products utilizing SkinEthic Human Vaginal Epithelium (HVE) model.

Quantitative expression of the Candida albicans secreted aspartyl proteinase gene family in human oral and vaginal candidiasis.

Candida albicans-secreted aspartic proteinases (Sap) modify the epithelial cytokine response in an in vitro model of vaginal candidiasis.

The secreted aspartyl proteinases sap1 and sap2 cause tissue damage in an in vitro model using vaginal candidiasis using reconstituted human vaginal epithelium.
Easy-to-use online ordering form with secured access on EPISKIN website: www.episkin.com

EPISKIN ACADEMY

- Offers to professionals training sessions on the use of 3D models and validated protocols. Regularly trainings are held at Episkin location in Lyon, Gerland – France. On demand training can be organized in your facilities.

- The academic program developed in partnership with universities promotes to scientists, toxicologists, regulators, and students the use of alternative methods to animal testing with lectures and laboratory works.

Real time technical service to help customer in:
- Handling the tissues
- Solving day to day problem
- Assisting users designing innovative protocols

Our partners and distributors in Japan, India, Korea:

Nikoderm Research Inc.
1-6-14 Azuchimachi, Chuo-ku, Osaka City, OSAKA 541-0052, JAPAN
06-6125-3501
info@nikoderm.com
www.nikoderm.com

KrisHgen BioSystems
India Office
318/319 Shah & Nahar Industrial Estate Worli 400018, Mumbai, India
(022) 49198700
www.krishgen.com

Episkin - 4, rue Alexander Fleming - 69366 - LYON Cedex 7 - France
+33 4 37 28 22 00
info@episkin.com