

# Xeno-free and defined keratinocyte culture

On Biolaminin® 511 substrate

## Biologically relevant cell culture environment

Human epidermal keratinocyte (HEK) cells are the major cell type of the epidermis, the outermost layer of the skin. HEK cells maintain the barrier function of the skin and vital to wound healing. The keratinocytes are positioned on the basement membrane (BM) composed of laminin proteins and collagens. The BM is highly enriched in laminin isoforms 511 and 521. Here we show that the human recombinant laminin 511 (Biolaminin 511, LN511) is a potent cell culture substrate for HEK cells in vitro (Tjin MS. et al. 2018 and 2020).

### A chemically defined and xeno-free method to culture human epidermal keratinocytes

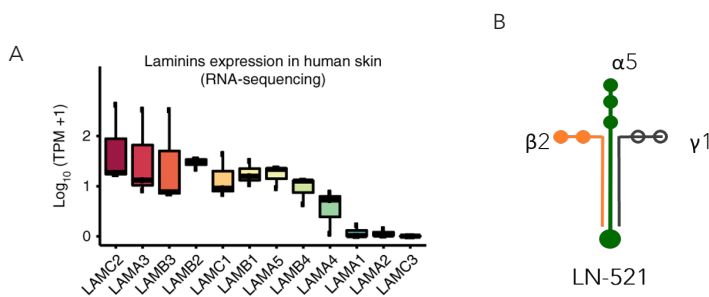
LN511 has been shown to enable robust, completely xeno-free and defined long-term expansion of primary adult human dermal keratinocytes.

Culture on LN511 replaces the 3T3-J2 feeder co-culture system and increases colony-forming efficiency, basal marker profile and the ability to form normal stratified epidermal structure in both in vitro and in vivo models (Tjin, 2018).

LN511 cultivated HEK cells also demonstrate a significantly reduced pro-inflammatory signaling pathway expressions compared to feeder culture-based systems.

**FIGURE 1**

Laminin RNA expression in human skin



### FEATURES AND SPECIFICATIONS

- Biolaminin 511 (LN511): defined- and xeno-free culture substrate
- Animal origin-free to the primary level
- Biologically relevant cell culture environment
- Expansion of human keratinocytes culture, also from aged donors
- Stable expression of basal keratinocyte markers
- Serum-free culture
- Organotypic culture with normal stratified



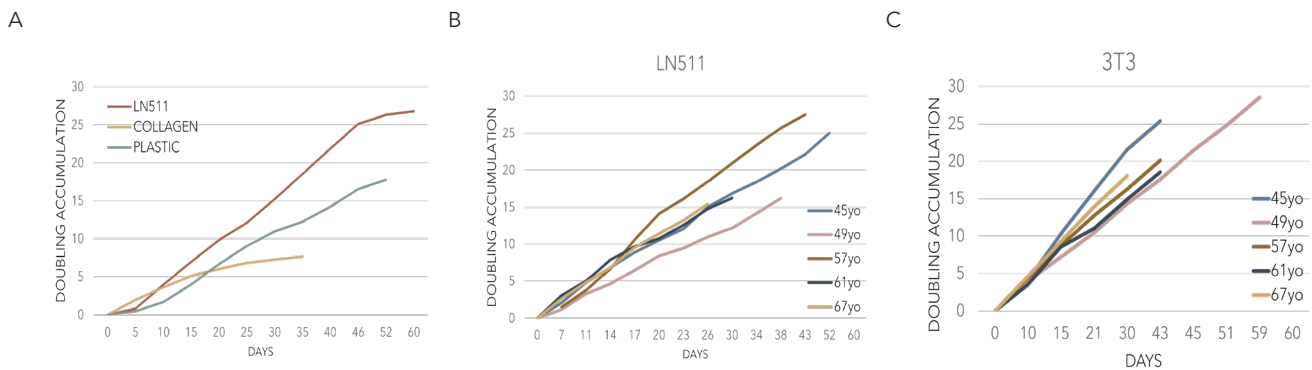
Direct link to more information about skin cells

A) RNA-sequencing of adult human skin (epidermal basement membrane) shows high expression of alpha 5 (LAMA5), beta 1 (LAMB1), and gamma 1 (LAMC1).

B) Together these three chains make up the laminin 511 protein. Tjin MS et al., 2018.

**FIGURE 2**

## Xeno-free culture of keratinocytes from aged donors on defined Biolaminin 511

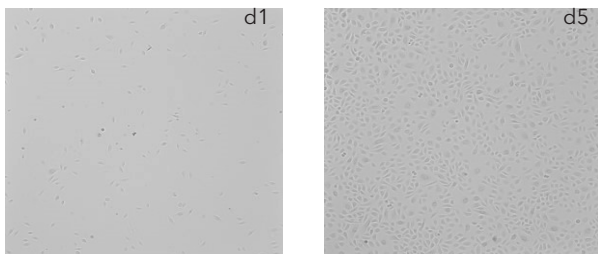


LN511 is an excellent substrate, for culture of primary keratinocytes from donors of all ages.

A) Primary adult keratinocytes show a significant doubling and yield increase when cultured on LN511, compared to culture on plastic or collagen. Population doublings of freshly isolated primary adult keratinocytes from 5 donors aged 45-67 years B) on LN511 and C) on classical mouse feeder 3T3. The xeno-free and defined Biolaminin 511 (LN511) supports the growth of aged primary human keratinocytes

**FIGURE 3**

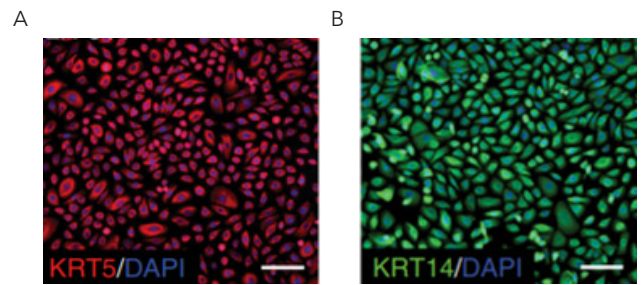
## Human keratinocytes show a phenotypical morphology on LN511 substrates



Cell morphology of adult human keratinocytes on Biolaminin 511 (LN511) at day 1 and 5. The cells migrate easily on LN511 and grow as homogenous monolayer. The cells develop the typical cobblestone-like morphology. HEK culture on LN511 is compliant with serum free cell culture media.

**FIGURE 4**

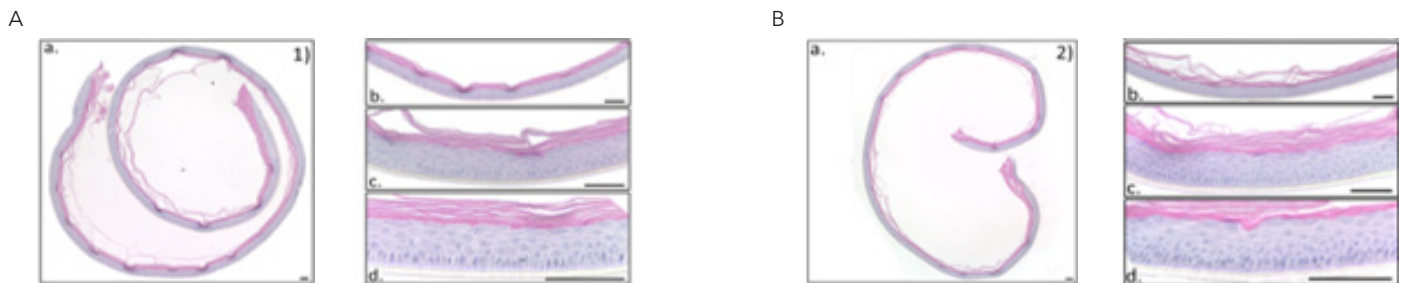
## Human keratinocytes cultured on LN511 express basal keratinocyte markers



The keratinocytes express basal keratinocyte markers KRT5 (A) and KRT14 (B) on LN511. Scale bar: 100  $\mu$ m. Tjin MS et al., 2018.

**FIGURE 3**

## Human keratinocytes show a phenotypical morphology on LN511 substrates



The epidermal reconstruction produces the typical layers found in vivo and allows the study of differentiated keratinocytes and production of the cornified barrier. LN511 (B) shows a similar epidermal morphology to the collagen control (A). Scale bar: 100  $\mu$ m. Data by: A. Boyadjiev, Prof. Przyborski lab, Durham University, UK.

## REFERENCES

Tjin M.S. et al. Nature Protocols, 2020. Chemically defined and xenogeneic-free culture method for human epidermal keratinocytes on laminin-based matrices.

Tjin, M.S. et al. Nat Commun, 2018. Biologically relevant laminin as chemically defined and fully human platform for human epidermal keratinocyte culture.

Pouliot N. et al. Exp Dermatol. 2002. Laminin 10/11: an alternative adhesive ligand for epidermal keratinocytes with a functional role in promoting proliferation and migration.