BioLamina

Next generation xeno-free and defined skin cell culture

On Biolaminin® 521 (LN521) and Biolaminin 511 (LN511) substrates

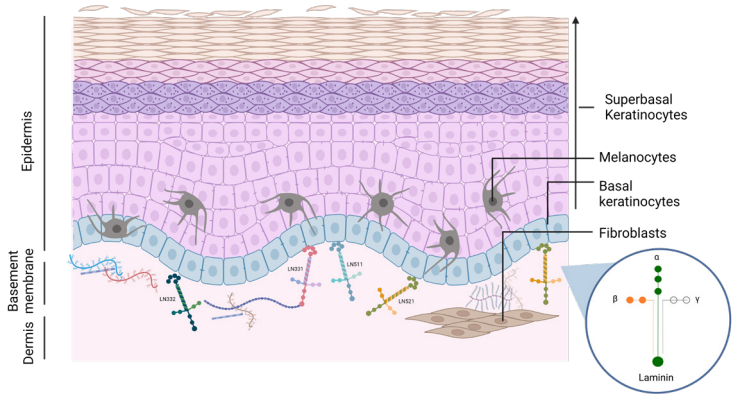
APPLICATION NOTE 023

Full-length laminins are essential for a functional and healthy skin

Skin is the largest organ of the body and our first line of defense against pathogens and environmental impacts. The skin is composed of two main layers, epidermis and dermis. They are separated by an ultrathin basement membrane (BM), highly enriched in laminin proteins, particularly the isoforms 332, 331, 521 and 511. Laminin 332/331 are mostly associated with adhesion and maturation and 521/511 with adhesion and proliferation. The complex interaction of BM and cells is essential for healthy skin development and tissue homeostasis, and essential part of the stem cell niche of the skin (Figure 1). For correct BM assembly and function, full-length laminin proteins, with all parts of the molecules, are essential (Figure 2). Truncation of the protein or lack of laminin isoforms results in severe skin disorders, such as junctional epidermolysis bullosa.

FIGURE 1

Schematic figure of the skin illustrating the epidermis and epidermis layers, separated and connected by the laminin-rich BM



The most abundant cell types are keratinocytes with basal keratinocytes at in direct contact with the BM, melanocytes (epidermis) and fibroblasts (dermis). Magnification: Each laminin isoform consists of three intercoiled protein chains– an α, β, and γ chain, respectively. The laminin isoforms are named according to their chain composition.

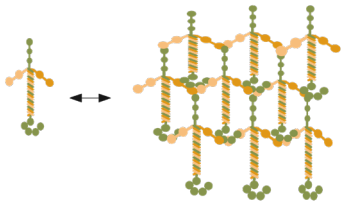
FEATURES AND SPECIFICATIONS

- Enabling, next generation of skin cell cultures of:
  - Basal keratinocytes
  - Epidermal melanoblasts
  - Dermal fibroblasts
- Defined and xeno-free culture substrates
- Biologically relevant cell culture environment
- Human full-length laminins and self-assembly

Direct link to keratinocyte App Note

FIGURE 2

Schematic view of laminin self-assembly



The full-length laminin network in the BM forms through laminin self-assembly.

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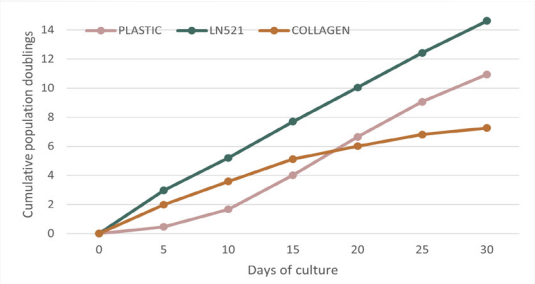
In vitro expansion of human skin cells, keratinocytes, melanocytes and fibroblasts

FIGURE 3

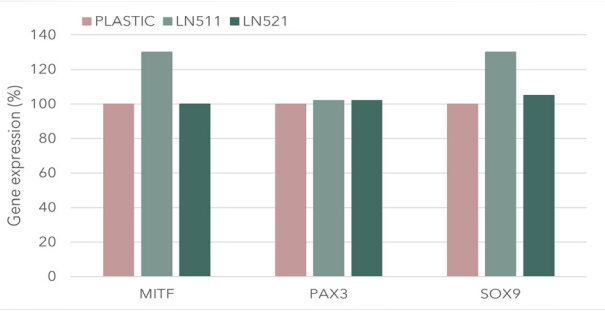
Population doubling of human epidermal keratinocytes enhanced on LN521



Primary keratinocytes from adult donors cultured without serum reaches 2-8 times more doublings on LN521 (green), compared to plastic (coral) and collagen (orange).

FIGURE 4

Human epidermal melanoblasts express melanocyte markers on LN521



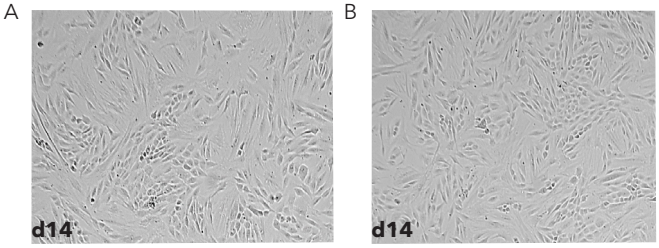
qPCR analysis of melanocytes markers MITF, PAX3 and SOX9 on plastic (coral) LN511 (light green) and LN521 (dark green).

Dermal fibroblasts for skin biology research and hiPSCs generation

Fibroblasts are the major cell type of the dermal portion of the skin. Current protocols for human dermal fibroblast (HDF) culture are inefficient and often rely on serum supplementation or other tissue-extract products and often without a cell culture substrate. Here we introduce a biologically relevant full-length laminin substrates, Biolaminin 521 and Biolaminin 511, as substrate for primary HDFs allowing for completely xeno-free and defined culture conditions without serum supplementation. Biolaminin substrates results in more stable primary culture increased yield. The cells show a stable expression of typical fibroblasts markers LUM, COL1A2 and WNT5A.

FIGURE 5

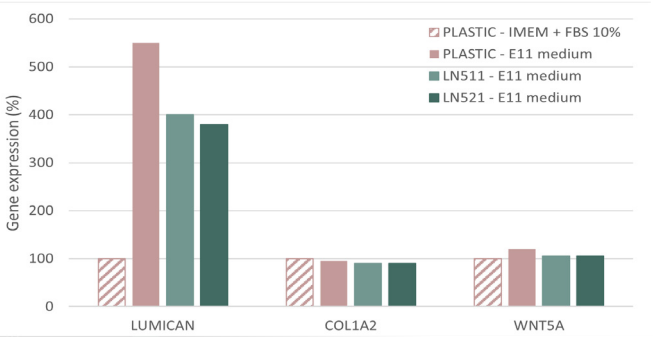
Adult human fibroblasts grow as homogenous monolayer on LN521 and LN511.



Cell morphology of adult human fibroblasts on A) Biolaminin 521 (LN521) B) Biolaminin 511 (LN511) on day14. Cells reach 70-80% of confluence in 5-6 days. Cells show typical fibroblasts morphology.

FIGURE 6

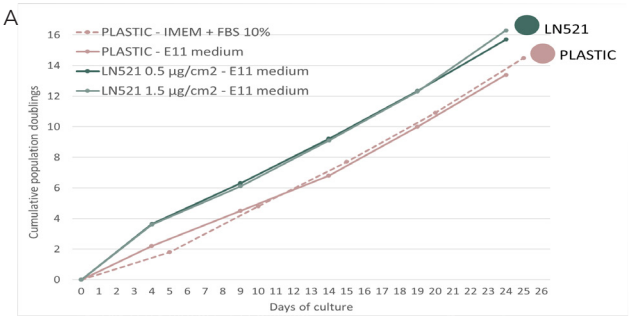
Expression of relevant fibroblast markers on LN521 and LN511 without serum



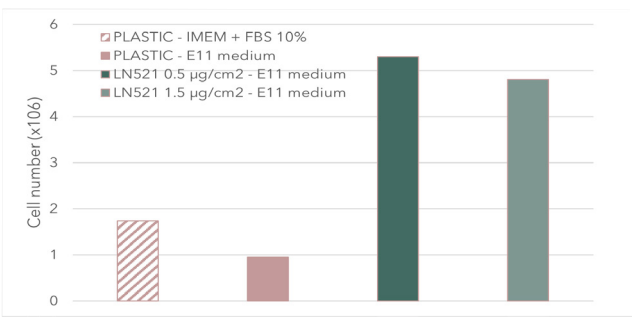
Both LN511 and LN521 (green) maintain the same gene expression over time as cell on control conditions (coral), 25 days in culture, with no significant decrease in expression (<10%) of typical fibroblasts markers LUM, COL1A2 and WNT5A with qPCR analysis.

FIGURE 7

Increased cumulative cell doubling of fibroblasts without serum on Biolaminin substrates



Population doublings for human adult fibroblasts over 26 days on LN521 (green) compared to the standard substrate plastic (coral), in xeno-free and defined culture medium.



3-5 times more fibroblasts cells on LN521 (green) compared to the standard substrate plastic (coral), after 3 cell passages (14 days in culture).

REFERENCES

Cichorek et al. 2013
Breitkreutz et al. 2013
Durbeej et al. 2010

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