

# Ubigel Inperio

In vitro characterisation of the effect of UBIGEL on human gingival fibroblasts

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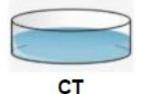
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**Purpose**: to verify whether the properties of Ubigel are mediated by a mechanical effect

Hypotesis: Ubigel is able to stimulate the gene expression of proteins used as mechanosensors (FAK, PAX and VNC)

Methods: cultivation of human gingival fibroblasts + Ubigel. Analysis of gene expression







UBI + CyB **CT**: Control Group

**UBI**: Cultured cells with UBIGEL 0.2%

**UBI+CyB**: Cultured cells with UBIGEL 0.2% + cytochalasin B (CyB)

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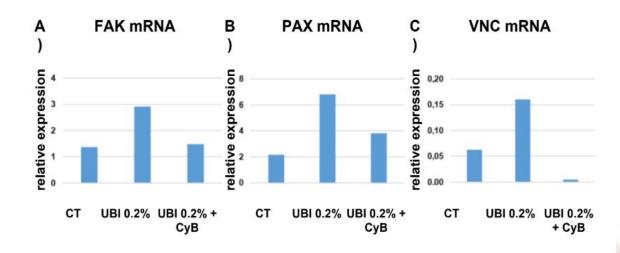


Figure 4. Representative histograms of mRNA levels for FAK (A), PAX (B) and VNC (C) after normalisation on GAPDH expression. Data are expressed as the average of evaluations repeated twice for each gene. Each sample was tested in triplicate.

#### **Results**

Ubigel induced an **increase** (up-regulation) in the mRNA levels of FAK, PAX and VNC by 115%, 217% and 167%, respectively vs CT;

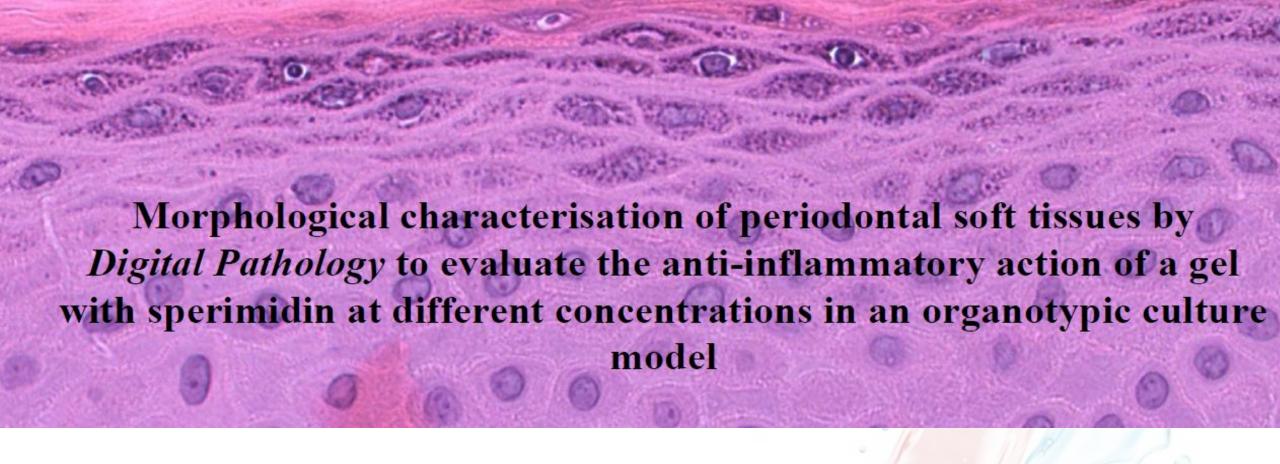
Down-regulation o FAK, PAX and VNC in UBI+CyB group.

### **Conclusions**

No changes in cell morphology;

**Activation of cellular pathways** mediated by mechanical mechanisms;

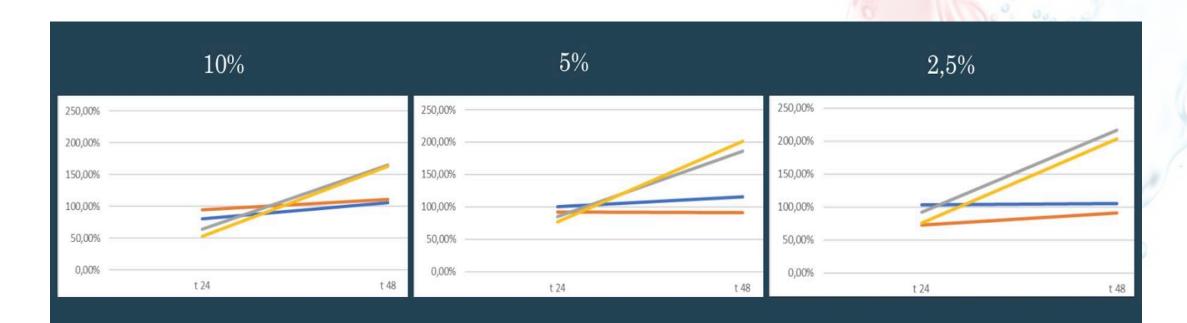
**Mechanical stimulation** of the gingival fibroblasts activity (*soluble scaffold*).



Stage 1: Choosing the optimal gel concentration

Stage 2: Analysis on organotypic cultures

Stage 1: Choosing the optimal gel concentration



0.5:1
5:1
Negative control
Test control (HA)

Test of vitality of the fibroblasts at 24 and 48 h

Stage 1: Choosing the optimal gel concentration

5% 2,5%

The gel proved to be biocompatible, non-toxic and able to stimulate cell vitality in **all concentrations** (ISO-10993 standards);

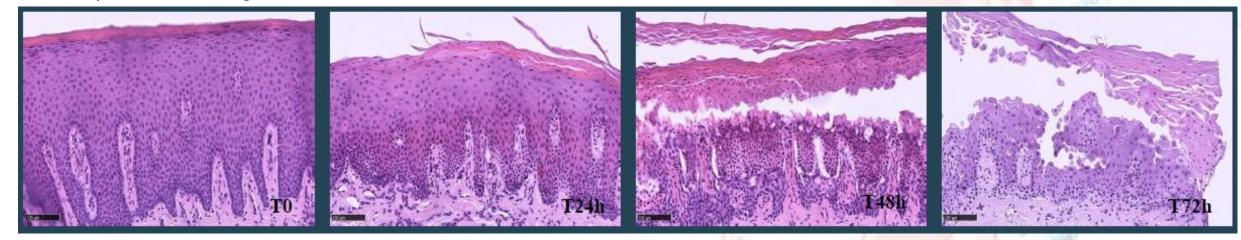
Compared with the controls, spermidine gels **promote** the **metabolic activity** of the cells.

Negative control
Test control (HA)

Stage 2: Analysis on organotypic cultures

No treatment

Hematoxylin Eosin, total magnification, 40x, 150x.



Gingival health (no inflammatory lesions)

T24h:
Detachment of the corneal layer;
Thickening of the basal layes

Detachment of the corneal and granular layers; basal layer with intense mitotic activity

T48h:

T72h:
Whole destruction
of the epithelial and
connective tissues

## Stage 2: Analysis on organotypic cultures

No treatment

4.00%

3,00%

2,00%

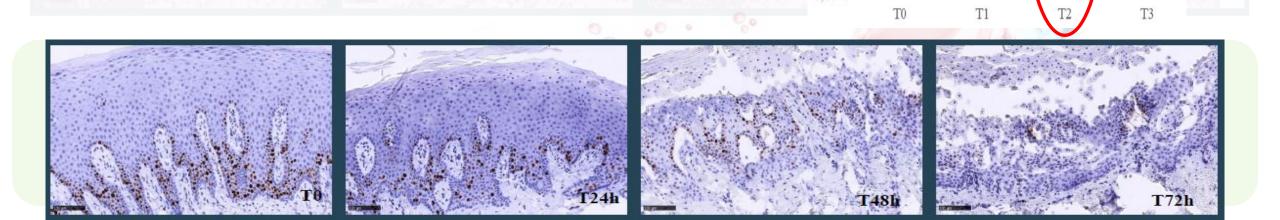
1,00%

0.00%

Hematoxylin Eosin, total magnification, 40x, 150x.

**Ki-67** performance is associated with the process of disintegration of the epithelial tissue



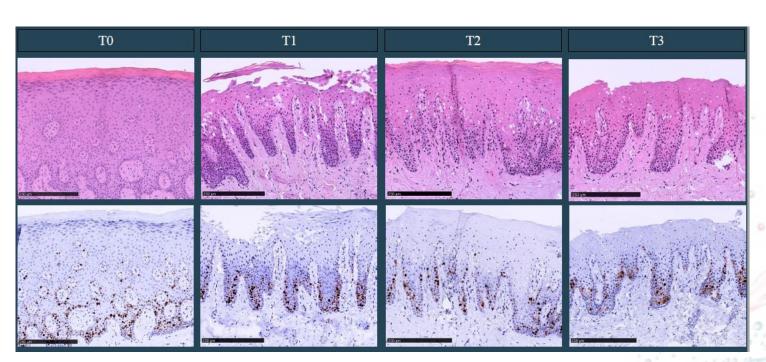


Stage 2: Analysis on organotypic cultures

In the presence of spermidine gel, do the cells of the oral mucosa in the culture continue to proliferate until T72h?

Stage 2: Analysis on organotypic cultures

5(S):1(HA)



KI-67

5,00%

4,50%

4,00%

3,50%

2,50%

2,50%

1,50%

1,50%

1,00%

TO

T1

T2

T3

Spermidine gel helps to maintain tissue in a slightly disrupted state up to 72h.

Ki-67 remains until after 72h

S: spermidine HA: hyaluronic acid

Stage 2: Analysis on organotypic cultures



All spermidine gels **improve the cells' ability to remain joined** together longer than the controls not treated;

Cells maintain their proliferatice capacity up to 72h (especially for Gel 5S: 1HA).

Stage 2: Analysis on organotypic cultures



The **optimal gel concentration** should have an intermediate ratio between **0,5** (S): **1 (HA)** and **5 (S)**: **1(HA)**