

## **Skin keratinocytes irradiated with UV impaired the development of 3D epidermis model**

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**Background:** Excessive ultraviolet (UV) exposure is known to cause skin damage while chronic and repetitive UV radiations are associated with benign and malignant skin tumors. Studies of how UV radiation affects skin development, particularly reconstructed epidermal skin tissue, are of interest. **Objective:** To investigate the impact of UVB irradiation on the development of 3D epidermal skin construction. **Methods:** Keratinocytes were isolated from infant foreskin and cultured in keratinocyte growth medium. Cells were divided into 3 groups, i.e., normal cells at passage 3 (Gr1), cells at passage 3 irradiated with UVB (30 mJ/cm<sup>2</sup>) (Gr2), and normal cells at passage >5 (Gr3). Evaluation of cell senescence included senescence-associated  $\beta$ -galactosidase activity (SA- $\beta$ -gal), cell proliferation (BrdU incorporation assay), ROS levels (flow cytometry). Keratinocytes in GR1 and GR2 were used to reconstruct 3D epidermal model. **Results:** Positive SA- $\beta$ -gal staining was observed in Gr2 and Gr3 but not Gr1. UVB irradiation significantly impaired cell proliferation in GR2 (100 $\pm$ 8.87% vs. 46.51 $\pm$ 6.47%, respectively) and increased ROS generation (Fig.1). The keratinocyte cell layer almost disappeared when keratinocytes (Gr2) were used for reconstructed epidermis model (Fig.2). **Conclusion:** UVB inhibited keratinocyte proliferation, increased ROS accumulation, accelerated keratinocyte aging, and impaired 3D epidermis reconstruction.

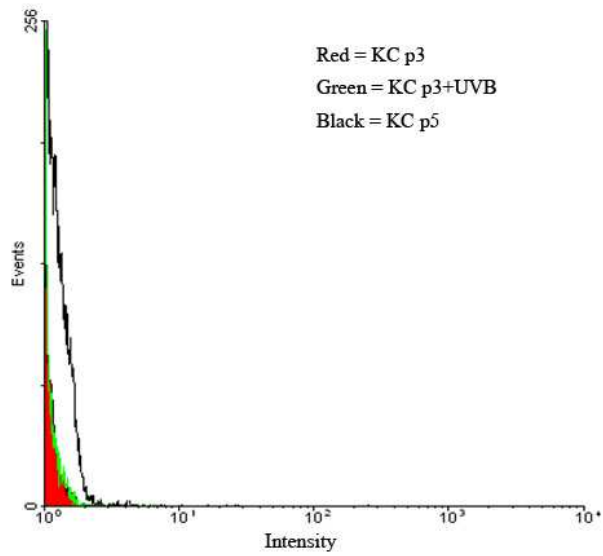


Fig. 1. Autofluorescence of keratinocytes (KC). KC at passage 3 (p 3) with or without UVB irradiation ( $30 \text{ mJ/cm}^2$ ) were compared with KC at passage 5 (p 5)



Fig 2. Effect of UVB irradiation on the development of 3D epidermis model. A) Normal keratinocytes at passage 3 and B) keratinocytes irradiated with UVB ( $30 \text{ mJ/cm}^2$ ) were used to construct epidermis skin equivalent model.