

The security in utilizing *in vitro* reconstituted human oral epithelium. An oncogenetic pathway study

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Introduction: Human oral epithelium cells derived from primary cultures allow the establishment of an *in vitro* restored epithelium which, when returned to the same local origin, allows the occlusion of several oral defects.

Objective: Concern about the activation of some oncogenic pathways justifies the investigation of their cell mechanisms. The aim of this work is to detect any possible cell function abnormalities that are not compatible with normal tissue mechanisms.

Material and Methods: This project was approved by the Research Ethical Committee of the Institute of Nuclear and Energetic Research, under the License N° 087/CEP-IPEN/SP. Normal human oral keratinocytes from primary cultures were seeded in cell culture dishes with special keratinocyte culture medium. The cells were maintained in a humid incubator at

37°C containing 5% CO₂. When the cells reached confluence they were removed from the dish, fixed (formaldehyde 10%), embedded (paraffin) and sectioned for immunohistochemistry, and the protein from the cell lysate was utilized for Western blotting. The utilized antibodies were: p53, PTEN, pAkt, β -Catenin, Metallothionein.

Results: The results confirm the normal structure of the cultivated *in vitro* epithelium and did not show any oncogenic pathways that would compromise their utilization in oral reconstructions.

Conclusion: We conclude that these *in vitro* cultivated epithelia established from cells harvested from the oral mucosa are totally safe and biocompatible for use in tissue repair and can accurately reproduce normal cell structures.