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Self-organized pigmented and vascularized full skin tissue model for melanoma research

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Being a multi-layered organ, the complexity of human skin is difficult to mimic *in vitro*. Despite the major advances in currently available 3D skin models, they remain very simplistic in relation to native tissue complexity. Moreover, the integration of melanoma cells into 3D skin models rendering representative tumor microenvironments is yet to be fully achieved. Cell sheet engineering has shown impressive results in recreating the native tissue structure and interactions of many different tissues by keeping intact the native cell-cell and cell-extracellular matrix interactions (Yanf et al., 2007; Haraguchi et al., 2012). Considering these major features, we built a 3D full skin model by stacking heterotypic cell sheets of human dermal fibroblasts (hDFbs) and keratinocytes (hKCs) or of hDFbs and human dermal microvascular endothelial cells (hDMECs), respectively as units of the epidermis- or dermis-like analogues. Melanocytes were further incorporated in the epidermis-like cell sheets before exposed to air-liquid interface (ALI), obtaining a pigmented stratified epidermis. In order to understand how melanoma cells responded to the self-organized full skin model, different cell lines (WM-115, SK-Mel-28, LM-MEL-33 and VMM15) were used. Histologically analysis confirmed the organization of those cells within the epidermis and their migration into the dermis through the formed basement membrane. Interactions between the melanoma cells and the stromal cells in the dermis are being assessed focusing on the known mechanisms involved in tumour progression in order to validate our approach.

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