ABSTRACTS

595

Wound healing in a model system: analysis of 4D dynamics

DR Ferris,[1] K Tanner,[1] L Lanzano,[2] DM Gardiner,[1] EL Rugg[1] and E Gratton[1] *1 University of California Irvine, Irvine, CA and 2 University Catania, Catania, Italy*

Despite a large number of studies there is still debate over the cellular dynamics of wound healing. Histology, conventional imaging and electron microscopy have yielded conflicting results leading to two contradictory models; the 'leapfrog' model in which proliferating keratinocytes enter the suprabasal compartment and are pushed onto the wound bed, and the 'tractor tread' model in which basal keratinocytes migrate over the wound bed and pull the trailing epidermis. It is evident that a higher level of analysis is required to resolve such complex biological systems. We have employed an integrative approach that utilizes camera based transmission images (2D) and multiphoton microscopy to investigate the complex dynamics of wound healing in axolotl skin. Skin explants were prepared from full thickness skin excised from the forelimbs of GFP transgenic axolotls. Wounds were created by punch biopsy and re-epithelialization of the wound bed was followed using fluorescence imaging. One and 2 photon confocal microscopy were used to spatially delineate thick tissue samples in the axial direction and generate a 3D reconstruction of the cellular network. Fast data acquisition (~7mins for 3D stack of 200 μm) and optimum signal to noise ratios, make it possible to follow re-epithelialization as a function of time (4D dynamics). 3D reconstruction software and enhanced analysis algorithms based on fluctuation spectroscopy have made it possible to extract dynamic information and quantify the physical properties associated with wound healing. Specifically, it is possible to spatially resolved cellular velocities, cell size, and cell fate. Cell hypertrophy is observed with an approximate increase of 30% of cell volume during the re-epithelialization. We propose a new biological model that describes, in part, the cellular dynamics as well as the mechanics of wound healing. Evidence suggests that an essential impetus for the observed dynamics in wound closure can be attributed to cell hypertrophy where the pushing of the nearest neighbors triggers the motion.