Directed Irradiation Synthesis for Bioinspired Ti-based Nanostructured Surfaces

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Statement of Purpose: In recent decades, progress in our understanding about the complex signal pathways between cells and their microenvironment, and how this interaction ultimately drives cell behavior and function has improved. This understanding has motived ambitious strategies for regeneration of lost or damaged tissues, as well as cell-based therapeutics[1]. The current approaches are based on designing synthetic microenvironments that attempt to mimic the complex network surrounding the cell so that they can serve as a platform for tissue regeneration[2]. However due to the challenge that implies fabricating complex physical, chemical and biological 3D structures and considering the incomplete knowledge about the natural cellular microenvironment; synthetic microenvironments fail achieving their therapeutic goal. To overcome these limitations, a reductionist or bioinspired design capable of avoiding the complexity and redundancy of a mimetic approach is therefore more desirable. Bioinspired materials may just reproduce the necessary signal pathways generated by its functional biological counterpart[3]. In this sense, directed irradiation synthesis (DIS) is a novel strategy for bioinspired material design that combines physico-chemical and topographical nanofabrication. Preliminary cell viability studies are presented using DIS on Ti-based metallic biomaterials.

Methods: Human aortic smooth muscle cell (HASMC) (cat. #C-007-5C, Gibco® Life Technologies, Carlsbad, CA) line was used in all tests. The CometAssay® was the apoptosis assay used to evaluate the potential genotoxicity effects of the specimens. The test was performed according to manufacturer's recommendations (cat. #4250-050-K, Trevigen, Inc., Gaithersburg, MD), and the genotoxicity was quantified using CometScore™. The results were compared with those obtained for control samples (positive and negative). SEM was used to observe the adhesion, expansion, morphology and integration of the HASMCs over the irradiated metallic surfaces. Surface modification uses DIS with heavy-ion low-energy irradiation as a function of the incident ion angle. Cell migration studies were performed to analyze the wound healing capability of these materials.

Results: Fig. 1 A-B summarizes the results of the percentage of DNA in the comet tails obtained for control samples and irradiated metal surfaces. At the 0.05 level, the mean distribution of tested analytes are significantly different compared to the positive control, thus indicating no detrimental effects in the HASMC DNA tested under the experimental conditions outlined in this work. **Fig. 1C** shows a SEM imagine of Ti6Al4V after 60° incident-angle irradiation with Ar^+ at 500 eV. The surface resulted in a formation of ripples whose morphology is correlated with the metal's crystallographic orientation. **Fig. 1 D-E-F** shows HASMCs observed by SEM. Normal growth depicted by elongated and star-shaped morphology is observed in HASMCs growing in modified metallic materials at 24 h, whilst cells growing in untreated material (virgin Ti6Al4V) have flattened bodies with insufficient lamellipodia and filopodia.

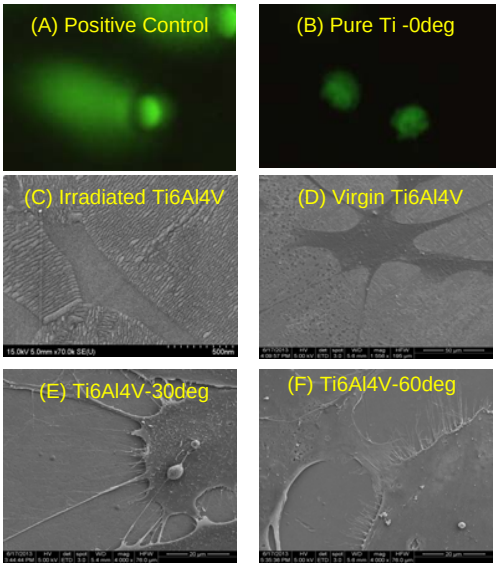

Fig. 1 *(A) Fluorescent images showing cells with DNA damage (A) and healthy cells (B), (C) SEM image of Ti6Al4V irradiated showing nanotopographical ripples of approx. 50nm; (D)(E)(F) Morphology of HASMCS seen by SEM showing cellular fluctuations*

Conclusions: The first results of low-energy high-density DIS-treated Ti-based surfaces show no detrimental effects on cell viability. The SEM imagines clearly showed differences in cell morphology depending on the topographical cues on each surface and DIS conditions. No chemical treatment is needed with DIS. All the irradiated samples exhibited an advanced process of wound healing. Future work will correlate high-fidelity DIS parameter control of rational design of bioinspired nanostructures, which control cell behavior.

References:

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