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In situ characterization of tissue constructs using a multimodal optical imaging/spectroscopy approach

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Currently, most of the engineered tissue constructs are characterized at the scaffold level for biocompatibility, toxicity, porosity for cell infiltration, mechanical strength, compliance, toxicity, etc., using partially or totally invasive and destructive procedures. Additionally, two-dimensional basic imaging is often performed to determine cell maturation and growth. However, more precise spatial and temporal assessment of the growing 3D tissue is required during *in vitro* culture and after implantation. Unfortunately, tissue-specific analysis is complicated by the complexity of the cellular environment. Several cell types can be utilized in the engineered construct, targeted to perform multiple functions. As an example, muscle progenitor cells differentiate to form myofibers in a soft tissue construct. Co-seeded fibroblasts express soluble factors to maintain muscle viability, enhance differentiation, and produce extracellular matrix critical to tissue function. Endothelial cells and supporting stromal cells are co-seeded to form extensive endothelial-lined networks which readily connect with host blood vessels upon implantation. These constructs are typically cultured *in vitro* under electrical or dynamic stimulation. Although the 2D cell cultures can be characterized with a simple microscope working in the reflectance or fluorescence mode, imaging of cells in 3D is significantly more challenging. Cells must be fluorescently labeled and few compounds are designed for live cell imaging. Fluorescent dyes or fluorophore-linked antibodies are usually toxic and require permeable cell membranes for cell entry. If multiple cell types are used, they must be separately labeled prior to seeding. Cells are frequently genetically modified to express a fluorescent protein. Collagen-based biological scaffolds and many polymeric scaffolds are strongly auto fluorescent, making the cells and the supporting scaffold difficult to distinguish.

Physical Sciences Inc. (PSI), in collaboration with the Center for Regenerative Medicine at the Massachusetts General Hospital have preliminarily evaluated the use combined high-resolution Scanning Optical Coherence Microscopy (SOCM) and Raman spectroscopy for real-time three-dimensional quantitative assessment of growing *ex vivo* engineered soft tissue construct noninvasively in order to assess the most important parameters needed to determine its readiness for transplantation: viability, microstructure, vascularization, and maturity. The main benefit of our approach is that it provides very useful and comprehensive data sets in real time noninvasively, while no part of the manufactured tissue is destroyed. SOCM provides cross-sectional and three-dimensional structural maps of the tissue at the micron scale to depths of at least 1.5 mm, and thus to accurately assess the morphology of the tissue. It combines the depth sectioning capabilities of Optical Coherence Tomography (OCT) with the lateral sectioning capability of confocal microscopy (CM). Raman spectroscopy on the other side provides details about the biochemical composition of the tissue, allowing for determining its viability status before implantation.

Biography

Nicusor Iftimia is a Pr. Scientist at Physical Sciences, Inc- USA. His major research interests include disease diagnosis, regenerative medicine, and therapy guidance. He applies various optical imaging and spectroscopy modalities to buid complex instrumentation that can be used in the above-mentioned areas. He is the author of over 70 peer-review papers, numerous conference presentations, several book chapters, and the editor of two books. He also serves as reviewer and editorial board member on several peer-review journals, such as Molecular Imaging and Dynamics, International Journal of Optics, Journal of Medical Engineering, Journal of Optoelectronics and Advanced Materials, etc.

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