InvivoSciences, Inc.

Discovery with a Human Touch

Enhanced Fluorescent and Colorimetric Assays by 3D Tissue Constructs

Application:

Traditional fluorescent- and colorimetric-based assays are easily adaptable for use with three-dimensional (3D) tissue constructs grown in Mini-Construct Chambers™ (MC-8™). Plate readers can be used to quantify spectroscopic indicators of 3D tissue physiology in high-throughput. With this technology, signal detection is greatly enhanced as compared to monolayer-based assays.

Introduction:

Traditional techniques used to study tissue physiology are labor intensive and not readily amendable to high-throughput experimentation. *Ex vivo* tissues, in particular, require surgical skills, produce a limited number of samples, and are not compatible with existing cell-based technologies used for investigating cellular physiologies.

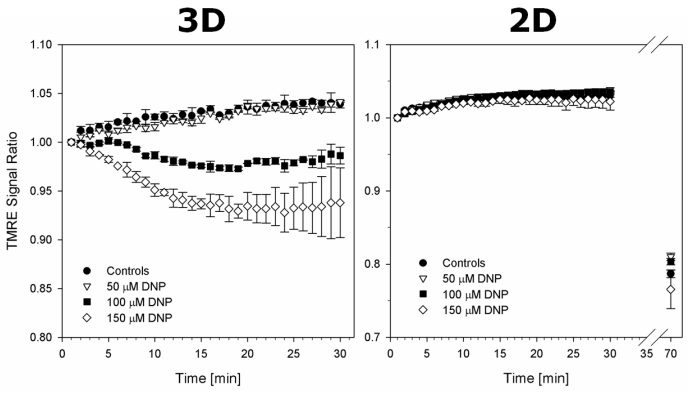
Our simplified process for fabricating 3D tissue constructs addresses these limitations and furthermore enables the use of cell-based fluorometric and colorimetric assays to quantify cellular and tissue physiology. Spectroscopic signals can also be quantified in high-throughput mode using conventional automated plate readers.

Technical Advantages:

- Perform high-throughput assessments of cellular and physiological characteristics of 3D tissues
- Significantly improve detection sensitivity and dynamic range of fluorescence assays
- Simultaneously assess multiple physiological parameters of 3D tissues including tissue contractility and stiffness (as assessed by InvivoSciences, LLC's proprietary Palpator™ technology)

Example:

Fibroblasts embedded in 3D tissue constructs and cultured as monolayers in 96-well plates were labeled with 500 nM TMRE for 30 minutes. Labeled constructs and cells were then treated with 150 µM of 2,4-dinitrophenol (DNP), a MMP un-coupler. DNP-induced TMRE dissipation was readily observable in 3D tissue cultures, but not in cell monolayers (see Figures below).



Above: Sensitivity of detecting TMRE signal in tissues (3D) vs. monolayer (2D). Increasing concentrations of DNP resulted in changes in TMRE signal. The detection sensitivity and dynamic range of TMRE in 3D tissues are superior to detection with monolayers.

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