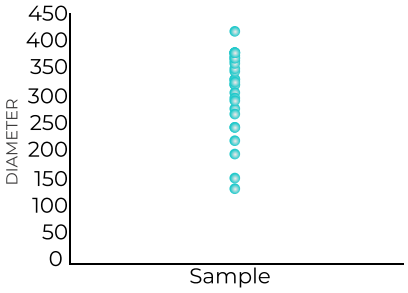
Shifting the Size Paradigm

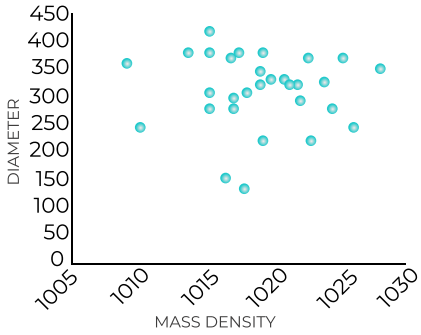
In trying to make organoid populations uniform, the focus has often been on ensuring they're similar in size. This approach is rooted in the belief that size uniformity leads to overall similarity. However, this strategy doesn't fully capture the complexity of diversity within these 3D models. Beyond size, the way cells are packed together, their arrangement, the concentration of ECM, and the presence of cavities within these structures add layers of structural and organizational variety, that influence experimental outcomes. Clearly, achieving consistency in 3D cellular systems demands attention to a wider range of factors beyond mere size, underlining the importance of a comprehensive approach to understanding and managing variability.

WHAT YOU KNOW



Size quantification is an essential daily practice for tracking the progression of our 3D cultures, enabling growth monitoring and uniformity evaluation. However, focusing on size overlooks the critical aspect: Organoids are three-dimensional.

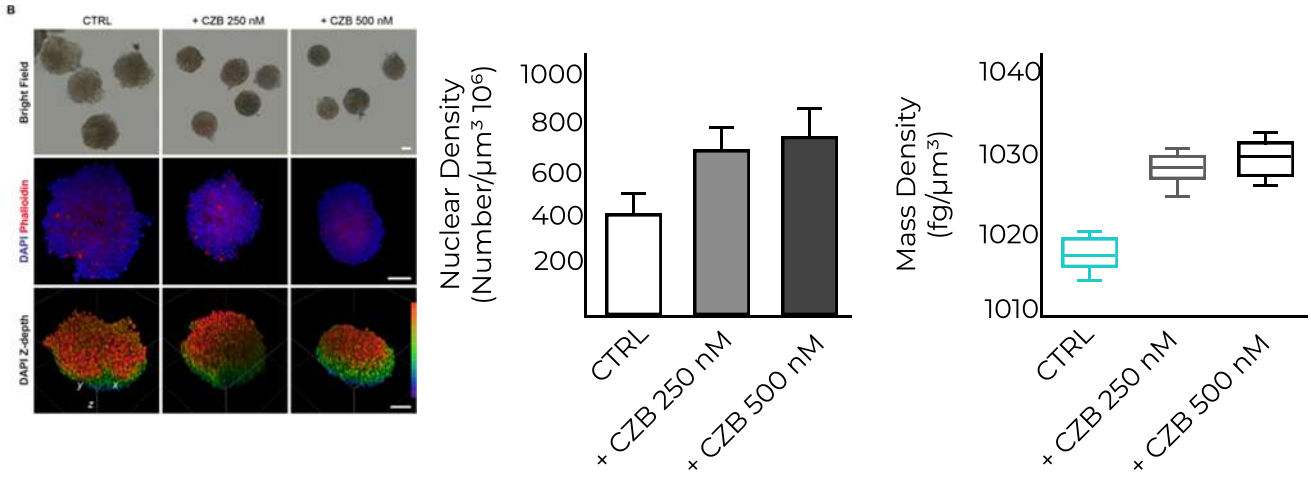
WHAT YOU ARE MISSING



Beyond size, 3D cultures have a complex balance of cells, proteins, and structures, each playing a pivotal role in their functionality. Incorporating daily biomarker evaluations for structural perspectives shifts the paradigm, revealing the full potential of 3D cultures.

CELL DENSITY

This study introduces an advanced analysis approach for tumor spheroids in 3D culture systems, crucial for cancer research and drug screening. It combines a novel measurement device, confocal imaging, and protein quantification to assess the effects of crizotinib on colon cancer cell line-derived spheroids. This method offers a detailed structural characterization of spheroids, overcoming traditional imaging limitations and highlighting the significance of mass density changes as an indicator of treatment efficacy.

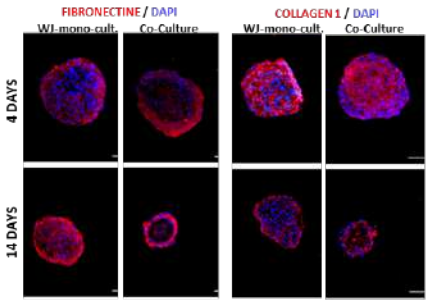


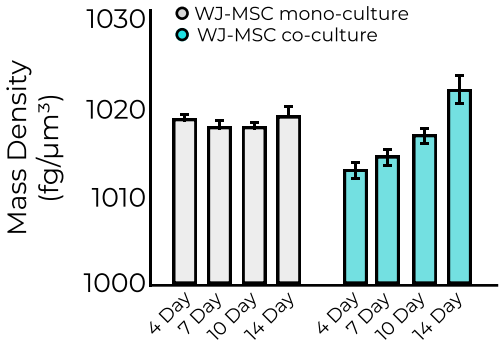
Within this paper, to evaluate the effect of crizotinib on LoVo spheroids, confocal quantification was utilized to measure the variations in nuclear density between control and treated groups. The results of the analysis demonstrated a perfect fit between nuclear density and mass density, indicating a direct correlation between these parameters in assessing the impact of treatment on spheroid structural integrity and cell viability. This finding underscores the effectiveness of combining biophysical measurements with molecular imaging techniques for a comprehensive assessment of drug responses in 3D culture models.

A new method for the study of biophysical and morphological parameters in 3D cell cultures: Evaluation in LoVo spheroids treated with crizotinib. Cristaldi et All. 2021

[ECM]

This study explores the development of a cellular model for regenerative medicine in Type 1 diabetes mellitus (T1DM) through the creation of co-culture spheroids of amniotic epithelial cells and Wharton's jelly mesenchymal stromal cells. The spheroids were evaluated for viability, extracellular matrix production, mass density variations, and hypoxic state over both early and long-term cultures, showing stability and consistent matrix production. The research suggests that these co-culture spheroids have the potential to be differentiated into endo-pancreatic cells for T1DM treatment, marking a significant step towards advancing stem cell therapy in managing complex metabolic diseases.

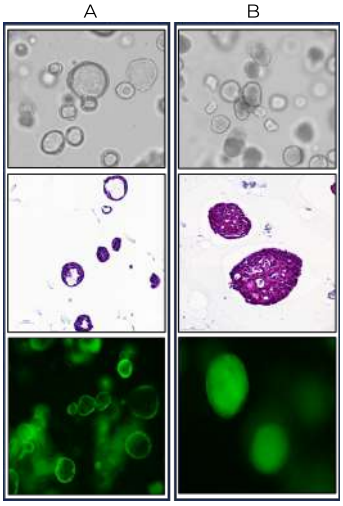


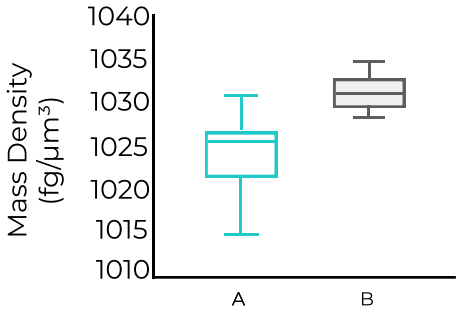


Within this study, the differentiation of spheroids in monoculture and co-culture was monitored and compared, with a focus on mass density variations at different days. The results indicate that the co-cultures, over time, show a reduction in diameter, an increase in mass density, followed by an increase in extracellular matrix (ECM) protein production, such as Fibronectin and Collagen 1. This demonstrates that mass density is not only an indicator of differentiation but also a valuable measure for quantifying ECM variations within the spheroids.

CAVITIES

This study was led by Marianna Kruithof-de Julio, lab leader of the Urology Research Laboratory (Department for BioMedical Research) and the Translational Organoid Research (TOR) Core at the University of Bern, and Elisa Rodrigues Sousa, PhD Student. Here, mass density is employed as a biomarker to distinguish and separate solid prostate tumor organoids from prostate organoids with cavities. This innovative approach facilitates the identification of distinct organoid types, advancing our understanding of prostate cancer's physical properties and potentially improving therapeutic strategies.





The study reveals that organoids featuring cavities exhibit a lower mass density compared to solid organoids. In this context, the use of mass density serves as a crucial indicator for identifying the presence and evolution of cavity formation in a non-invasive manner that preserves the integrity of the sample under examination