

A Center-Based Model Reveals that Cell Shape Variability Drives Cell Sorting in Spheroids

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Extended ABSTRACT

Center-based agent-based models (ABMs) of multicellular systems are mathematical models in which each cell is represented individually as a simple geometric object and can move continuously in space. These models have proved highly valuable in multicellular systems biology because they directly represent physical laws and can be parameterised by physically and biokinetically meaningful parameters [1]. Multicellular systems that particularly benefit from the center-based modelling (CBM) approach, are spheroids. Spheroids are three-dimensional (3D) *in vitro* spherical aggregates of biological cells. They recapitulate critical features of *in vivo* solid tumours, including the heterogeneous tumour architecture, internal gradients, growth kinetics and the physical interactions of the tumours. These similarities give spheroids great potential to study tumour properties [2]. Despite these advantages, *in vitro* spheroid research falls short when it comes to biomechanical interactions between cells on the one hand and the observation of internal processes within spheroids on the other.

In order to test biomechanical hypotheses and to generate new hypotheses to uncover biophysical mechanisms of spheroids in several conditions, we propose a center-based modelling approach implemented within the CompuTiX framework, as introduced by [3]. In the computational model, each biological cell is represented as a computational agent whose motion is governed by the balance of forces acting upon it over time. These forces include both interactions between cells and interactions between cells and the extracellular matrix (ECM). The resulting computational spheroids serve as *in silico* analogues, enabling controlled *in silico* experimentation.

An interesting spheroid experiment is the sorting of cells within a mixed spheroid, which is initiated with two types of cells that are intermingled. Based on [4], in which agent-based models other than the center-based model are used to achieve cell sorting with quiescent cells, the cells in our center-based model are quiescent as well when the following biomechanical hypotheses are applied. In accordance with [5], the differential adhesion hypothesis (DAH) is insufficient

to achieve the final spheroid state in which the cells are sorted per cell type. Nor does the hypothesis of high heterotypic interfacial tension (HHIT), as formulated in [6], explain the observed final state. Furthermore, the addition of random cell motility did not lead to spheroid segregation either. As a fourth hypothesis, we incorporated variation in cell shape into the center-based model. The volume of each cell is modelled as a sinusoidal function that varies between the cell nucleus volume and the total cell volume, with a randomly assigned phase. The variability in cell shape was found to play a prominent role in driving spatial segregation within the mixed spheroid. Cells organised themselves into different clusters based on their cell type (see Fig. 1).

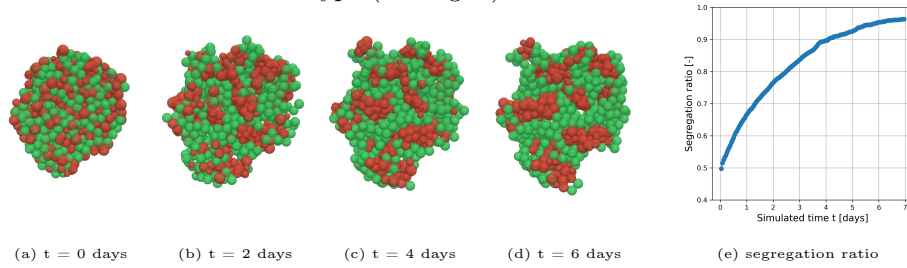


Fig. 1: Simulation of the emergent spatial segregation in a mixed spheroid. (a - d) Cross-sections are shown at every two simulated days. Cells are coloured according to their cell type. (e) As a measure of sorting, the segregation ratio is computed as a function of time for the model.

The proposed model is able to reproduce an experimentally observed phenomenon qualitatively and quantitatively. This shows that the model is a promising tool for investigating spheroid dynamics. We will assess the usability of the model in reproducing diverse spheroid phenotypes.

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