Tumor-on-a-chip model to study the invasive capacity of circulating tumor cells

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Abstract

Metastasis is the underlying cause of cancer-related fatalities. Despite their scarcity, circulating tumor cells (CTCs) have the ability to disseminate through the circulation system, potentially initiating metastasis. This characteristic underscore their significance as a reliable and important target for early detection and relapse prevention [1]. Isolating/analyzing patient-derived CTCs emerges as an extremely important endeavor for the study of metastatic mechanisms, critical to achieve a deeper understanding of the biological processes underlying cancer progression and offering invaluable insights into potential therapeutic interventions.
Current preclinical testing models that lack the three-dimensional (3D) architecture of the tumor microenvironment (TME) and fail to capture their complex cell-cell/ECM interactions and dynamics, impose significant research limitations [2].
As an alternative, dynamic experimental platforms capable of mimicking the native TME have demonstrated to be promising modelling and screening tools, as is the case of organ-on-a-chip technology. These systems allow dynamic fluid flow, stimuli manipulation, and cell compartmentalization within microchannels [2]. Relevant cell-cell and cell-TME interactions can be engineered within biomimetic matrices creating both mechanical and structural ECM-like features, reproducing a multicellular architecture and physiological relevant TME, while providing vascular perfusion in vitro – all in a single microfluidic device.
Moreover, since evidence shows that CTCs share common genetic profiles with the active tumors (primary or metastatic), combining patient-derived CTCs within these microfluidic platforms can impact oncological research, contributing to a better understanding of cancer progression, and revealing the underlying mechanisms of metastasis.
The RUBYchip™ provides fast and efficient microfluidic isolation of viable CTCs from different cancer types, based on cell size and deformability [4,5]. Hence, this project is aimed at the development of a patient-derived tumor-on-a-chip (ToC) to understand the onset of metastasis which will incorporate CTCs from cancer patients.
To achieve this, we centered the first part of the project in developing proliferation methods using MCF-7 breast cancer cell line as a model to generate aggregates with a low starting number of cells, mirroring the conditions typically encountered in a liquid biopsy sample from a patient. With the conditions provided, we were able to develop cell aggregates starting with an initial cell number of 25 cells. The viability was monitored over time using a Live/Dead™ assay and CellTiter Glo 3D and yielded positive results for low-initial-cell-number aggregates development, which demonstrated continuous proliferation and high viability. To increase the complexity, stroma cells were added in a 1:3 ratio to the clusters. The co-culture of the cancer cells with fibroblasts seem to aid in the fast generation of the clusters giving them a more robust structure.
Ideally, this project will generate valuable insights into the intricate mechanisms underlying metastasis formation.

References

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Figures

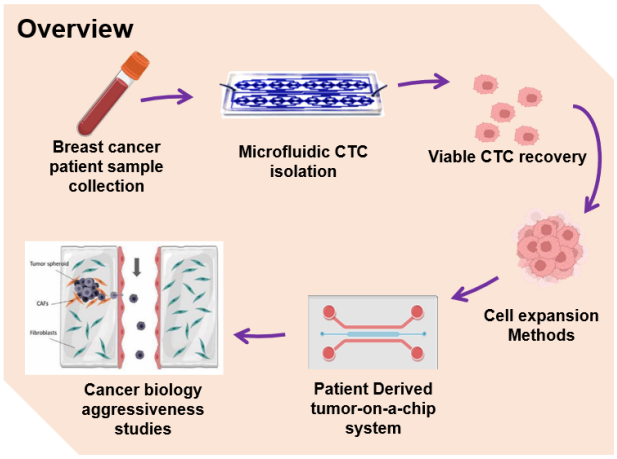

Figure 1. Overview of the workflow of the project. A liquid biopsy sample is collected and processed in the CTC entrapment device. The CTCs are recovered and expanded using the proliferation methods developed and transferred to the TOC model and cancer aggressiveness studies are performed.

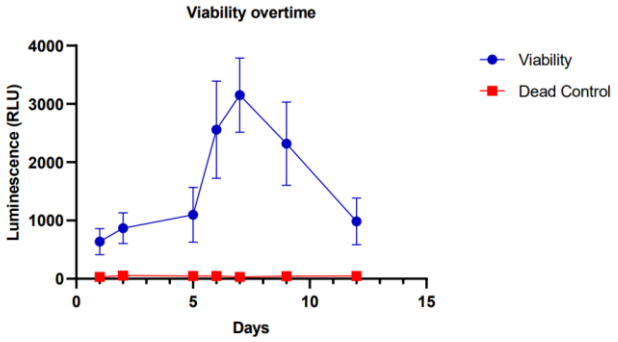

Figure 2. Viability of the MCF-7 micro-aggregates over 12 days of culture measured using CellTiter- Glo® 3D viability assay based in a luciferase reaction.

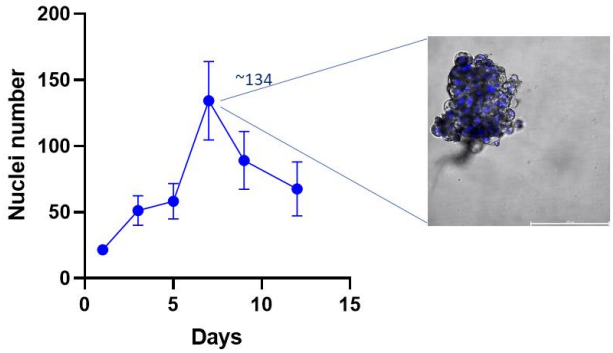

Figure 3. Nuclei count of the micro-aggregates over 12 days of culture. Nuclei were stained with Hoechst dye (1:1000).

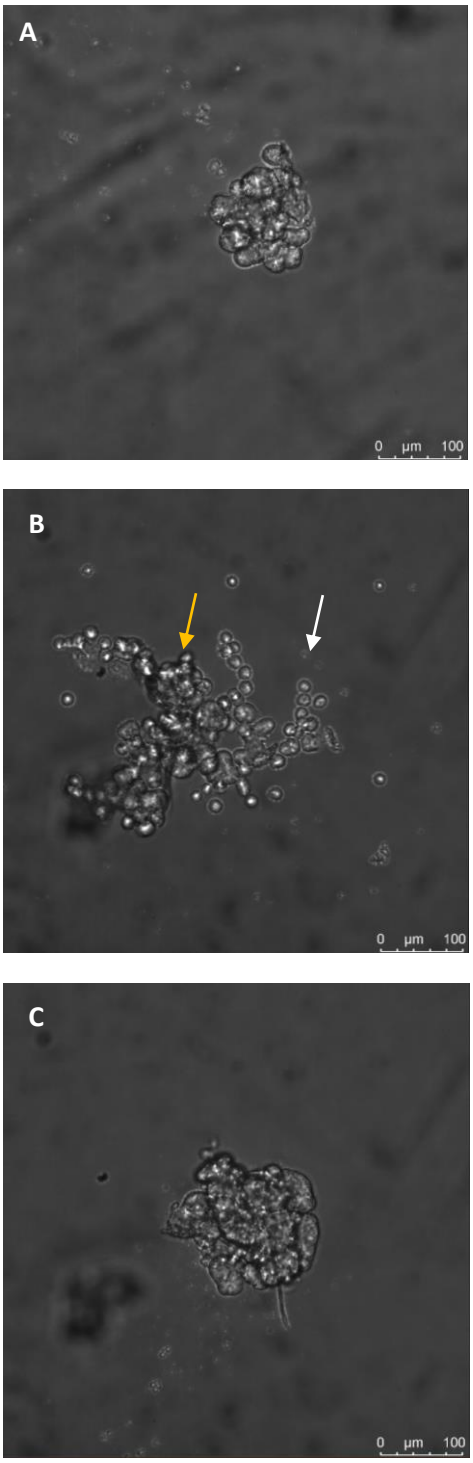

Figure 4. MCF-7 and Fibroblast co-culture. A) MCF-7 micro-aggregate 3 days post-seeding; (B) Introduction of the fibroblasts in the system at a 1:3 ratio using a delayed culture method. Orange arrow showing the initial MCF-7 aggregate while white arrow is showing the fibroblasts; (C) Compact aggregate with both cell types after 1 day of co-culture.