MicroOrganoSphere (MOS): a novel technology for circulating tumor cell (CTC)-derived organoid formation

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BACKGROUND

- Circulating tumor cells (CTCs) are released from solid tumors and can initiate metastatic lesions at distant sites. CTCs reflect tumor heterogeneity and provide an opportunity to evaluate tumor characteristics, evolution, and individual patients' treatment response in real-time[1].
- CTCs culture can provide insights into molecular drivers of cancer progression allowing for high throughput drug screening in the context of precision medicine and targeted treatment regimens. Unfortunately, CTC culture is extremely challenging and the best conditions for their growth have yet to be established[2].

Aim: In this prospective study we evaluated the use of emulsion microfluidics and droplet generators as a novel technology to partition and isolate cancer cells into miniaturized micro-reactors to facilitate the generation of CTC MicroOrganoSpheres (MOS).

METHODS

- Peripheral blood (20-25 mL) was collected in EDTA tubes from patients with metastatic colorectal cancer (CRC) and metastatic breast cancer (BC) and processed with Parsortix (ANGLE) for CTC-enrichment.
- After enrichment, cells were shipped to Duke University in MACS® Cell Storage Solution (Miltenyi Biotec, Bergisch-Gladbach, Germany). Upon arrival, cells were centrifuged and resuspended in 50 uL of Matrigel and were used to generate MOS as previously described[3].
- MOS were maintained in 6-well plates for ~4 weeks before being transferred to 96-well plates and tagged with Alexa Fluor 647 for EpCAM staining and imaged with the Pico ImageXpress every 3 days for growth of CTC (Figure 1).

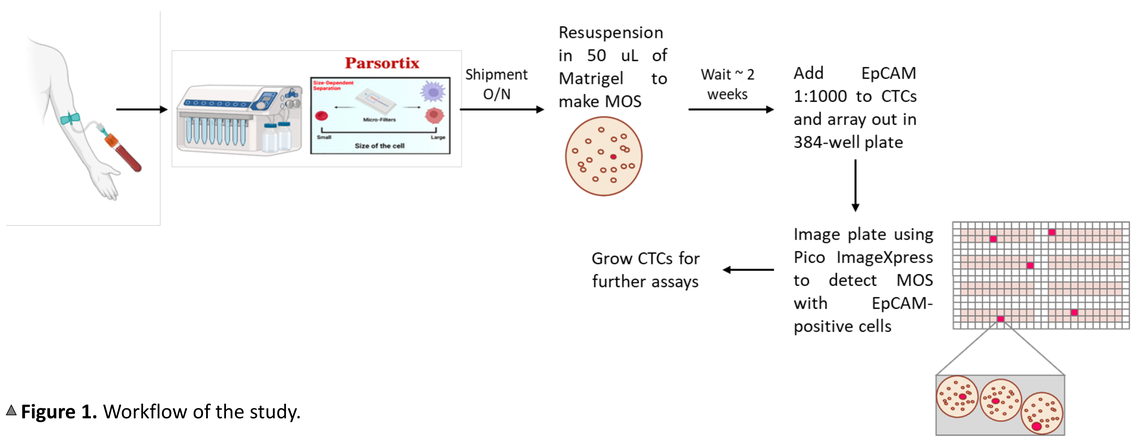

Figure 1. Workflow of the study.

RESULTS

- A total of 12 samples (9 from BC and 3 from CRC patients) were processed to generate MOS (Figure 2).
- EpCAM+ CTCs were successfully detected and grown in 2/9 BC (22%) and 2/3 (75%) of CRC specimens.
- Average time to growth was 28-30 day for breast cancer samples and greater than 90 days for colon cancer samples (Figure 3). The remaining processed samples had little or no growth time.
- To date, the samples are in a stagnant growth phase.

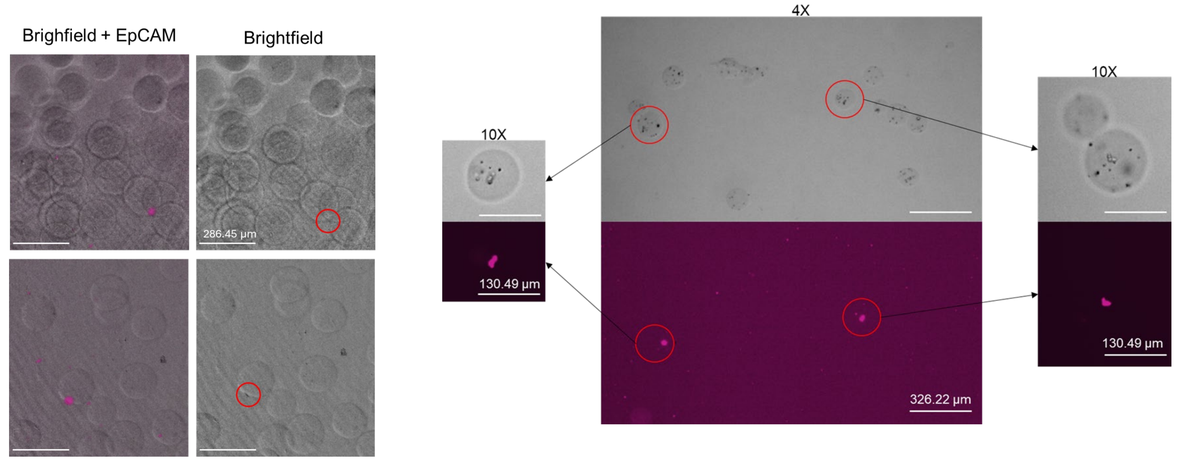

Figure 2. CTCs (in pink) from BC sample (left) and CRC sample (right).

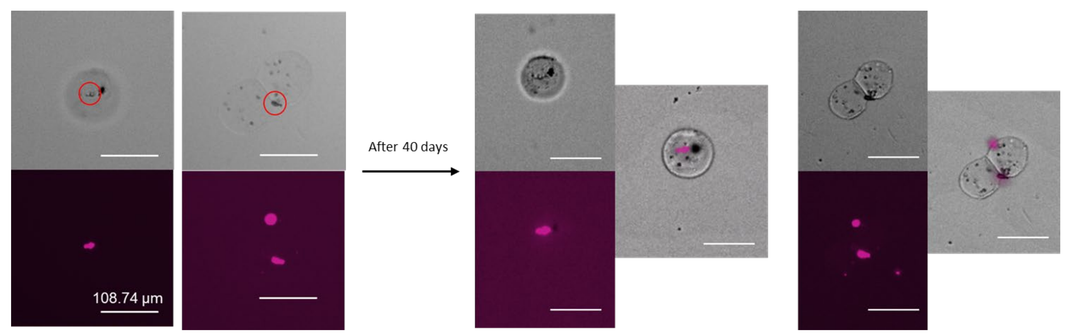

Figure 3. CTCs (in pink) growth observed over time.

CONCLUSIONS

- This proof-of-concept study demonstrated the feasibility of using MOS for the successful identification of EpCAM+ CTCs in both BC and CRC peripheral blood samples.
- The small size of MOS enables the growth of CTCs in culture, although the growth got to a stagnant phase in all cases.
- Additional samples and time will be needed to improve methods of establishing CTC organoid lines in culture for future characterizations.

References:
1 Reduzzi et al, Crit Rev Oncol Hematol, 2024; 2 Tayoun et al, Cells, 2019; 3 Ding et al, Cell Stem Cell, 2022

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