Miniaturized 3D cancer models for measuring efficacy of cancer therapeutics

Karla Paterson[1], Ronan Mellin[1], Jamie Hodgson[2], Agapitos Patakas[2], Michele Zagnoni[1,3]

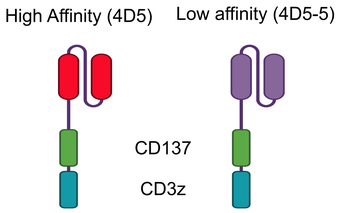
[1] ScreenIn3D, Technology and Innovation Centre, 99 George Street, Glasgow, G1 1RD, UK;
[2] RoukenBio, Rouken Discovery Centre, Newhut Road, Motherwell, ML 1 3ST, UK;
[3] Dept. Electronics and Electrical Engineering, University of Strathclyde, Glasgow, G1 1XW, UK.

Introduction

Chimeric Antigen Receptor T-cell (CAR-T) therapy has been proven beneficial in treating haematological malignancies and melanoma. However, their clinical success against the majority of solid tumour types has been limited thus far. The need for physiologically relevant 3D, complex *in vitro* models of cancer is steadily increasing due to the emergence of drugs targeting the immune system and the tumour microenvironment (TME). Furthermore, an increasing interest in precision treatment of cancer patients and the rise of anticancer therapies used in combination has highlighted the need for large-throughput 3D efficacy tests capable of maximising data generation using very small quantities of 3D tumour models, especially including those from clinical samples.

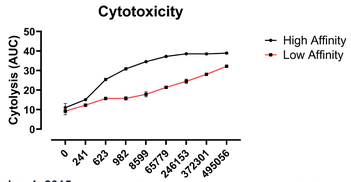
Generation of HER2 specific CAR-T cells

Second-generation CAR constructs targeting HER2 were designed incorporating CD3ζ and CD137 (4-1BB) signalling domains. The high-affinity receptor was based on the trastuzumab-derived scFv (4D5), while the low-affinity variant (4D5-5) was generated using previously published mutations of 4D5[1].



Name	VL 55 (CDR2)	VL 66 (FW3)	VH 102 (CDR3)	Kd (nM)
4D5	Tyr	Arg	Tyr	0.58
4D5-5	Glu	Arg	Val	1119

Affinity based differences on functionality were confirmed using a HER2 IndEx-2 cell line that allows titratable expression of the target antigen.



1. Liu et al, Cancer Res; 75 (17) September 1, 2015

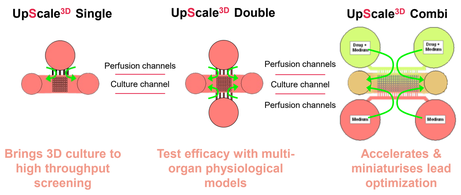
Figures were created by using BioRender.

UpScale^{3D} lab-on-a-chip technology

- Long-term culture of **perfused biopsy-derived** and ***in vitro* complex models**
- **Unique miniaturization** for **combination studies**
- **20X more data throughput** for the same amount of starting material
- **50% faster** to results

Figure 1. *Microfluidic plate with multiple device units*

Figure 2. *As low as 2,000 cells for tens of 3D cellular models per device*



Versatile: many 3D complex *in vitro* models

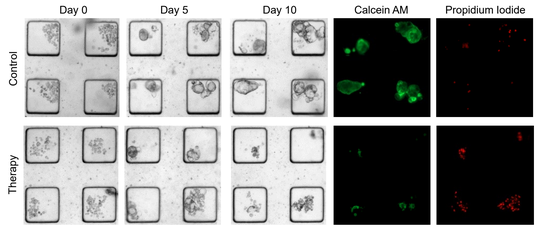


Figure 3. *Biopsy-derived organoids are grown and screened in the UpScale^{3D} platform*

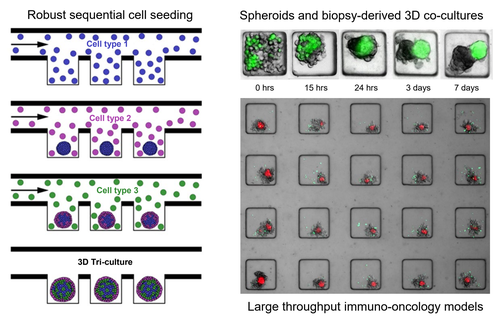


Figure 4. *Co-culture 3D models can be robustly generated using different cell types, injected at the desired time point without disturbing cell culture*

High throughput CAR-T cytotoxicity assays

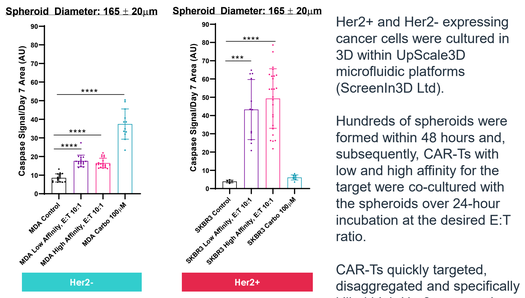


Figure 5. *Histograms of HER2 Induction at 0-50 µM ABA*

Her2+ and Her2- expressing cancer cells were cultured in 3D within UpScale3D microfluidic platforms (ScreenIn3D Ltd).

Hundreds of spheroids were formed within 48 hours and, subsequently, CAR-Ts with low and high affinity for the target were co-cultured with the spheroids over 24-hour incubation at the desired E:T ratio.

CAR-Ts quickly targeted, disaggregated and specifically killed high Her2+ expressing cells in 3D models.

CAR-T cells elicited mild cytotoxicity in low/non-Her2+ expressing spheroids.

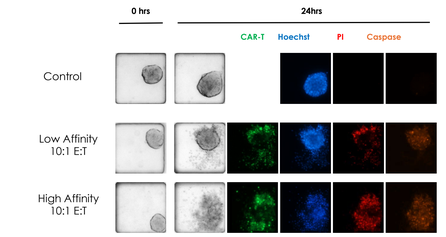


Figure 6. *Histograms of HER2 Induction at 0-50 µM ABA*

Summary

In this work, we show high-throughput and miniaturized efficacy assays to demonstrate CAR-T mediated cytotoxicity using the Her2 receptor, a common target over-expressed in many types of solid tumours. Our versatile screening platform offers high quality and multiplexed assays to test therapy efficacy on spheroid co-cultures, organoids and primary tumour fragments. The platform offers a unique system to miniaturize drug combination studies using small quantities of cell samples, including tissue-derived 3D models of disease and, at the same time, provides cost-effective and fast immune-oncology assays for extensive drug combination studies and precision medicine drug discovery.

Scan the code

Download the digital version of this research poster and take all the latest advancements and data home in your pocket.