Primary Cell Culture Models for Personalised Medicine in Patients with Renal Cell Carcinoma

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Introduction

Renal cell carcinoma (RCC) is common and dangerous. Approximately 25-30% of patients with newly diagnosed RCC have metastatic disease and poor prognosis at presentation. Unfortunately, due to biological heterogeneity of RCC subtypes, there is no high-quality evidence available for systemic therapy in patients with advanced disease. Thus, the development of patient-derived in vitro cancer models has clinical utility for personalised cancer therapy.

Research objectives

The aim of this study is to develop techniques for culturing excised RCC tissue taken from patients, with the end-goal of improving understanding of RCC pathophysiology and developing personalised therapies.

These in vitro cell culture models may have utility for:

- Selection of second- and greater line therapy in patients with Clear Cell RCC
- Selection of first-line systemic therapy in patients with non-clear cell renal cell carcinoma
- Culture resistance in cells to selected first-line therapies, in order to guide the choice of second-line therapy in resistant cell models

Methods

Using sterile techniques, tissue from consenting patients with RCC, excised directly at surgery, is placed in cell culture medium.. Necrotic tissue, blood clots, fat and connective tissue are dissected away. The remaining tumour tissue is mechanically minced and either plated directly into tissue culture flasks or accutase enzyme digested and seeded into tissue culture flasks. Both 2D and 3D culture methods are in development. Following the establishment of viable individual RCC cultures past ten passages, investigation of various therapies will begin.

Treatments are selected based on European Association of Urology recommendations for first-, second- and third-line RCC therapy.

Results

To date 33 tumour biopsy samples have been processed and nine primary cultures established past ten passages in 2D culture.

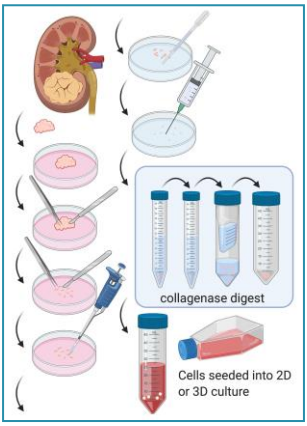

Figure 1: RCC tumour culture workflow. Patient derived tissue is minced, accutase digested, filtered and then seeds into either 2D culture or 3D spheroid culture conditions.

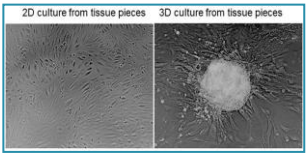

Figure 2: RCC tumour tissue grown in 2D and 3D culture conditions. Primary cell cultures derived from patient RCC tumour tissue 0380 were grown in either 2D culture or 3D spheroid culture conditions.

The success rate, of 27%, is high compared to similar RCC studies. Our initial trials to create 3D multicellular spheroids in agarose-coated plates has been successful. Dose response curves in both 2D and 3D cultured cells are in development to compare the personalised efficacy of selected drugs.

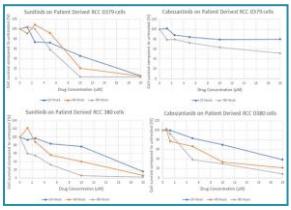

Figure 2: Dose response of 2D cultures to Sunitinib and Cabozantinib. 2D primary cell cultures derived from patient RCC tumour tissue (0379 and 0380) were treated with different concentrations of Sunitinib and Cabozantinib with cell survival monitored at 24, 48 and 96 hours using an MTT assay. (*RCC 0379 Cabozantinib 48hr were unable to obtain results).

Discussion

We have successfully developed techniques to culture cells derived from primary RCC tumours with a relatively high success rate.

Prelimary dose response assays have shown differences between drugs and differences in the sensitivity of primary cell cultures to these drugs.

Future Aims

We are currently expanding our portfolio of drugs to be tested, based on current European Association of Urology recommendations.

High throughput assays using 3D spheroids derived from our primary RCC cultures are in development and are expected to better recapitulate the patient response.

Conclusion

The development of standard, high throughput, protocols for the assessment of primary tumour cell sensitivity to therapeutic agents may guide the choice of agents for personalised therapies against RCC and potentially other types of cancer

Selected references

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