Establishing an *ex vivo* Personalised Medicine Platform for Bladder Cancer Patients using Patient-Derived Tumour Explants

Abby Templeton[1,2,3,4], Penny Jeffery[1,2,3,4], Patrick Thomas[1,2,3,4], Mahasha Perera[1,2,3,4,5], Gary Ng[2,6], Laura Bray[2,7,8], Ian Vela[1,2,3,4,5], Erik Thompson[1,2], Elizabeth Williams[1,2,3,4]

[1]Queensland University of Technology (QUT), School of Biomedical Sciences at Translational Research Institute, Brisbane, Queensland, 4102, Australia. [2]Centre for Personalised Analysis of Cancers (CPAC), Brisbane, Queensland, Australia. [3]Queensland Bladder Cancer Initiative (QBCI), Brisbane, Queensland, 4102, Australia. [4]Australian Prostate Cancer Research Centre – Queensland, Brisbane, Queensland, 4102, Australia. [5]Department of Urology, Princess Alexandra Hospital, Woolloongabba, Queensland, 4102, Australia. [6]Department of Medical Oncology, Princess Alexandra Hospital, Woolloongabba, Queensland, 4102, Australia. [7]School of Mechanical, Medical and Process Engineering, QUT, Brisbane, Queensland, 4059, Australia [8]ARC Training Centre for Cell and Tissue Engineering Technologies, QUT, Brisbane, Queensland, 4059, Australia.

Introduction

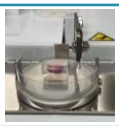
Precision medicine techniques are advancing with investigation into the use of Patient-Derived Explants (PDEs) as a functional patient proximal model. PDEs maintain the phenotype and microenvironment providing a platform to test therapeutics. This study focuses on the use of bladder cancer PDEs to determine whether this approach may provide additional information to guide the personalised treatment of people with cancer. Here, we present a pilot study in an advanced, muscle-invasive bladder cancer case from the Princess Alexandra Hospital.

Research Objectives

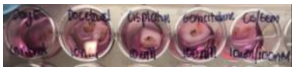
1. Establish longevity of bladder cancer PDEs
2. Treat bladder cancer PDEs with standard of care (SOC) chemotherapies
3. Determine the effects of SOC chemotherapies on apoptosis and proliferation of tumour cells in the PDE platform

Methods

1. Receive sample from surgery, slice PDEs and culture overnight on sponges



2. Treat PDEs with SOC chemotherapies for 5 days



3. Perform immunohistochemistry analysis on control and treated PDEs

Results

Bladder cancer PDEs (~200 µm thick) remained viable following a 6 day air-liquid interface (ALI) culture (**Figure 1A**). Multiplexed immunohistochemistry (mIHC) results show that a gelatin sponge platform allows for the uptake and diffusion of chemotherapy drugs to the PDEs (**Figure 1B-C**). Further quantitative analysis is underway to compare the ratio of proliferating to apoptotic cells in each condition. Initial morphological observations suggest that docetaxel treatment (**Figure 1C**) led to reduced Ki67 expression and increased cleaved caspase 3 staining when compared to the untreated control slice (indicating drug efficacy).

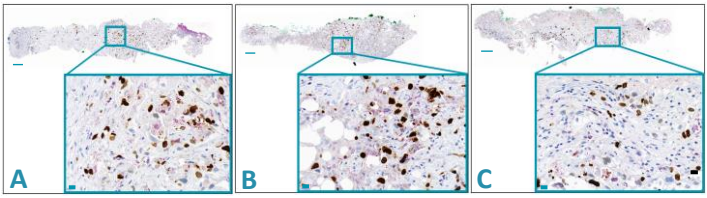


Figure 1: mIHC staining showing Ki67⁺ proliferating cells (brown) and cleaved caspase 3⁺ cells (purple; indicating apoptosis) on day 6 PDE (no treatment control) and PDEs treated with combinations of SOC. A) Day 6 control PDE. Inset; high resolution I=image demonstrating an abundance of proliferating tumour cells and few apoptotic cells. **B)** PDE following treatment with cisplatin (5 µM) and gemcitabine (5 nM) combination therapy shows a prominent number of apoptotic cells. **C)** PDE following treatment with docetaxel (10 nM) shows a reduced number of proliferating cancer cells. Whole explant images: Green stain (top side) represents air interface with lower side representing the liquid interface. Scale, 200 µm. Inset scale, 20 µm

Discussion

PDE have been used across several tumour types including prostate [1], pancreatic [2] and head and neck [3] cancers. However, the use of bladder cancer PDEs have not been widely reported. We have demonstrated that bladder cancer PDEs can be maintained in culture for at least 6 days. This is in contrast to a previous study which showed that bladder cancer explants derived from cell line xenografts showed a significant increase in apoptosis compared to the starting material after 5 days culture [4]. This may be due to the different culture conditions used in each study. Together these results suggest that this model may be useful for personalised drug testing [4], although further optimisation of the assay is required. Furthermore, the incorporation of therapeutic targets identified via tumour sequencing into this functional testing platform will require additional modifications to align the timeframes of these technologies.

Conclusion

- Bladder cancer PDEs from this patient remained viable after 6 days of *ex vivo* culturing
- PDEs showed changes in proliferation and cell death in response to treatment with SOC chemotherapeutic agents

Future Direction

Long-term study (beyond 6 days) to determine viability of bladder cancer PDEs

Limitations

Intra-tumour heterogeneity introduces intrinsic variability across PDEs

Longevity of PDEs does not currently align with the timeframe associated with Whole Exome Sequencing (WES)

References

[1] Centenera et al. (2018). *Molecular oncology,* [2] Gerlach et al. (2014). *British Journal of Cancer,* [3] Kokkinos et al. (2021). *Scientific Reports,* [4] van de Merbel et al. (2018). *Frontiers in Oncology*

For more information please contact Abby Templeton at:
abby.templeton@hdr.qut.edu.au
www.linkedin.com/in/abby-templeton-829499133/