Transforming precision medicine in lethal prostate cancer using stem cell derived organoids

Buskin A[1,6], Hepburn AC[1,6,7], Sims CHC[1,6], Clark EL[1], Franco OE[2], Wilson L[1], Singh P[1], Kendall H[1], Robson CN[1], Hayward SW[2,7], Dehm SM[3,7], Chen Y[4,7], Clevers H[5,7], Gaughan L[1,7,8], Heer R[1,7,8]

[1]Northern Institute for Cancer Research, Newcastle University, Framlington Place, Newcastle upon Tyne, UK.
[2]Department of Surgery, NorthShore University HealthSystem, Affiliate of University of Chicago Pritzker School of Medicine, Evanston, Illinois, USA.
[3]Masonic Cancer Center, University of Minnesota, Minneapolis, Minnesota, USA.
[4]Department of Medicine, Memorial Sloan Kettering Cancer Center, New York, New York, USA.
[5]Hubrecht Institute, Royal Netherlands Academy of Arts and Sciences (KNAW) and UMC Utrecht, the Netherlands.
[6]Co-first authors
[7]PCF grand challenge award investigators
[8]Lead Investigators

Background

One of the key issues hampering the development of effective treatments for prostate cancer is the lack of suitable, tractable, and patient-specific in vitro models that accurately recapitulate this disease. Current preclinical tools, such as cell lines or mouse models lack complexities of human cancers. Whilst primary cultures and patient derived xenografts can be used to model clinical disease, these are difficult to establish. Pioneering work of Prof Clevers in generating cancer organoids has opened up new opportunities to deliver more clinically relevant translational studies using human tissue and is transforming preclinical drug testing for many common diseases. Despite establishment of methods to generate human prostate cancer organoids by Prof Chen and Prof Clevers, these approaches have proven difficult. To overcome these limitations, we have for the first time demonstrated that induced pluripotent stem cells (iPSCs) enabled generation of human prostate tissue in vivo and in vitro. We aim to generate a novel model of prostate cancer organoids from iPSCs containing patient specific mutations and correlate preclinical drug testing with clinical treatment sensitivity. We plan to serially sequence circulating cell free-DNA and circulating tumour cells over 12 months from patients with lethal prostate cancer from the NIHR-funded VARIANT trial. From this clinical cohort, we will sequence key driver mutations and obtain their AR mutational and splice form profiles in response to established contemporary therapies. We will then recreate representative profiles using CRISPR technology within iPSC-derived organoids.

Methods

Human prostate cells were reprogrammed and derivative lines characterised for pluripotency. Co-culture of iPSCs with rat urogenital mesenchyme generated prostate organoids. Parallel approaches with lentiviral infection and electroporation of conditionally expressed Cas9-directed gene editing was performed within iPSCs. In a complimentary comparative biology model comprised of gene edited mouse prostate cancer organoids, drug sensitivities to specific driver mutations combinations will be tested.

Results

1. We have expanded and characterised our patient-derived iPSCs to account for biological variability and provide generalisability to the broader prostate cancer population (n=6 donors, n=3 clones each).
2. We have differentiated iPSCs into prostate organoids expressing prostate specific markers (AR, NKX3.1 and PSA), including a revised approach with inductive seminal vesicle mesenchyme.
3. Fluorescent label genomic integration has been confirmed in iPSCs via lentiviral and Cas9-mediated methods. P53/PTEN targeting has been shown using a Golden Gate compatible Tet-On inducible Cas9 system integrated into iPSCs.
4. We have carried out early preclinical testing of mouse organoids to next generation hormonal treatment (abiraterone/enzalutamide) and chemotherapy (docetaxel).

Conclusion
The generation of these functional in vitro analogues of the trial patients affords a significant opportunity to test if they predict response to clinical treatment and if confirmed, provides a realistic probability for rapid translation into the clinical domain and direct patient benefit.

Conflict of interest: The authors declare no conflicts of interest.

Funding Acknowledgements: Prostate Cancer Foundation