CANCER I

TIME: 10.15 – 11.15

LOCATION: BERWICK ROOM

THERAPEUTIC TARGETING OF THE ANDROGEN RECEPTOR USING PROTAC IN PROSTATE CANCER CELLS

Liuxun Li, Northern Institute for Cancer Research, Newcastle University

Background: Metastatic Prostate cancer (PCa) cells are able to survive through mechanisms which rely on retaining the androgen receptor (AR) signaling pathway. Among the different strategies that target AR, proteolysis targeting chimera (PROTAC) which acts to primarily degrade their target protein by the ubiquitin-proteasome system stands out recently.

Methods: We synthesised and tested a novel small molecule AR-targeted PROTAC, NCL-AR2 which is flutamide-based and its recruited E3 ubiquitin ligase is MDM2. Western blot, qRT-PCR and proliferation assay were used.

Results: NCL-AR2 was effective in degrading AR protein and reducing downstream proteins/mRNAs in LNCaP and VCaP cell lines. In proliferation assay using Incucyte, 10μM of NCL-AR2 could inhibit androgen-sensitive PCa cell growth to great extent compared with flutamide and enzalutamide. Moreover, Both 5μM and 10μM of NCL-AR2 could reduce PSA protein levels around 2 hours in androgen-sensitive PCa cells and these effects occurred earlier than same concentration of flutamide treatment. After 7-days' treatment, NCL-AR2 could also reduce ARVs to some extent. In combination with 5μM of nutlin-3α, NCL-AR2 helped improve the effects of degradation. Together with 5μM of nutlin-3α, 10μM of NCL-AR2 was able to decrease ARVs in 48 hours which probably results from the reduction of GATA2.

BIOLOGICAL CHARACTERIZATION OF GLIOBLASTOMA STEM CELLS GROWTH USING A 3D TUMOUROSPHERE MODEL

Sara Azeem, University of Sunderland

Glioblastoma (GBM) is one of the most common form of primary brain tumours in adults. It is a devastating tumour with poor prognosis that often results in death within approximately 12 to 15 months after diagnosis. The current standard of care for GBM is surgical resection followed by radiotherapy with concomitant and adjuvant temozolomide (TMZ) chemotherapy. But even after these intensive treatments, tumour recurrence is inevitable, which is due to a highly resistant sub-population of glioblastoma stem cells (GSCs). Presently, there is no standard treatment established for progressive or recurrent GBM. There is an urgent need for the development of novel therapies for GBM. Multicellular tumourspheres in a 3D model are appropriate in vitro model to culture GSCs and surrogate system to study tumour characteristics. Thus, we conducted biological characterisation of rapidly growing spheres in four human primary GBM cell lines (E2, G7, R24 and GLG) and analysed 3D tumoursphere growth, cell death, 'stemness', hypoxia, DNA damage and cell cycle during the process of 3D tumoursphere growth using microscopy, Western blotting and flow cytometry. During the progression of tumoursphere growth, we observed an increase in the hypoxia marker CAIX, cell death and expression of DNA damage markers □-H2AX and PARP. However, interestingly we observed a decrease in the levels of the tumour suppressor p53 in a time dependant manner. Current studies are exploring the significance of p53 reduction during 3D tumoursphere growth with the aim of identifying potential targets for the development of novel therapeutics for the treatment of GBMs.

3D BIOPRINTING OF THE HUMAN BONE MARROW TO DEFINE CANCER-NICHE INTERACTIONS IN LEUKAEMIA

Salem Nizami, Northern Institute for Cancer Research, Newcastle University

Leukaemia is the commonest malignancy in children; however, there remains an unmet need for clinically relevant in vitro models of the disease. Current 2D models do not reflect the role of cell-cell interactions in the cancer niche, leading to high drug attrition rates. In this study, we aimed to create a bioprintable human bone marrow organoid model to be used as a high-throughput cancer-niche platform, and apply this model to investigate the role of CDH2 in leukaemia, a targetable molecule identified by the lab to regulate niche-mediated chemoresponse. Induced pluripotent stem cell (iPSC) derived bone marrow cells were developed to specify CDH2-mediated niche response. A novel hydrogel comprising collagen, alginate and laminin (CAL) bioink was invented, and patient-leukemia-niche organoids were bioprinted using reactive jet impingement (ReJI), an in-house developed bioprinting technology. CAL gels were found to be at least 10% more stable than contemporary gel formulations, with a high degree of biocompatibility, and was also amenable to high-throughput bioprinting, establishing proof-of-concept for drug screening through automation using patient-derived organoids. Transcriptomic profiling in conjunction with functional validation assays revealed several cancer pathways to be regulated through CDH2, including JAK/STAT signalling and BCL2. Consequently, a triple combination drug screen was designed by adding Adh1 (CDH2 antagonist) and Abt-199 (anti-BCL2) to Dexamethasone (standard therapeutic agent in leukemia). This drug combination showed high efficacy on our bioprinted model, thereby corroborating the significance of CDH2 as a therapeutic agent, and patient-organoid bioprinting as a key tool for cancer research and drug screening applications.

CAN WE PREDICT RECONSTRUCTIVE SURGERY FAILURE IN SARCOMA PATIENTS?

Christie Mellor, Northern Institute for Cancer Research, Newcastle University

Background: Sarcomas are aggressive mesenchymal cell tumours originating in bone or soft-tissue. Treatment is multimodal and varies in sequence, including surgical resection, reconstruction, and radiation and/or chemotherapy. Ideally treatment is bespoke based on patient factors. Reconstruction varies from direct closure, to transferring soft-tissue, a 'flap', for defect coverage. A recognised proportion of reconstructions fail to heal. Reconstructive failure can therefore delay planned oncological therapy, negatively impacting the patients' outcomes. It is indeterminate who is high-risk for reconstructive failure or success.

Aim: To identify reconstructive failure risk-factors. To create a preoperative clinical tool to predict sarcoma patients' reconstructive failure risk.

Methods: A retrospective analysis of 338 sarcoma patients, treated from 2000-2019 at a single sarcoma unit was performed, including analysis of the literature, to identify reconstructive failure risk-factors. Thirty-two variables underwent univariate analysis. Variables reaching $p<0.25$ were entered into the binomial logistic regression model.

Results: Patients reconstructed with a pedicled flap, who had truncal or bone primary site, one major comorbidity, or extensive recurrence, experienced a significantly greater ($p<0.05$) proportion of failure. Flap reconstruction or having one major comorbidity, are independent reconstructive failure risk-factors. The regression model acceptably discriminates between high and low-risk reconstructive failure patients (c-statistic=0.748). Thus, 93% of reconstructive successes are correctly predicted as low-risk patients, and 31% of failure patients are predicted as high-risk.

Conclusion: This model helps stratify patients' chance of failure or success, potentially individualising treatment, improving outcomes. Further data will improve the regression model, to identify more high-risk failure patients.