Identification of a high risk, immune-active subgroup of primary prostate cancer

Eileen E Parkes[1], Emma Reilly[2], Nicholas Forsythe[1], Andrena McCavigan[2], Steven M Walker[2], Nuala McCabe[1,2], Laura A Knight[2], Ian G Mills[1], Richard D Kennedy[1,2].

1. Centre for Cancer Research and Cell Biology, Queen's University Belfast, Northern Ireland.

2. Almac Diagnostics, Craigavon, Northern Ireland.

Background: Despite marked success in other solid tumours, the majority of prostate cancers do not respond to immune checkpoint therapies. We analysed primary prostate cancers using data from TCGA and an internal dataset to identify and validate a high-risk subgroup of immune-active prostate cancer using previous validated Prostate Cancer Metastatic (PCM) and DNA Damage Repair Deficiency (DDRD) assays. We previously identified the DDRD assay as representative of activation of the cGAS-STING pathway, as a result of damaged DNA sequestered to the cytoplasm in tumour cells.

Methods: Integrative analysis was performed on 488 samples identified from TCGA with RNA sequencing, promoter site methylation, somatic mutation and somatic copy number variation. Four subgroups were assessed for leukocyte infiltration and the expression of checkpoint targets PD-L1, CTLA4, TIM3 and IDO1 as well as ICOS and STING. The viability of reproducing those subgroups with RNA sequencing alone was tested in the TCGA dataset and an independent validation dataset of 321 resected primary prostate cancers. Cox proportional hazards regression analysis was performed for biochemical recurrence and metastatic events in both datasets.

Results: Integrative analysis identified four subgroups characterised primarily by variances in copy number and genomic mutation. One of these subgroups 'Metastatic-like DDRD' (MetHot) had significantly higher PCM scores and DDRD immune scores compared to the other subgroups ($p < 2e-12$). This subgroup of patients showed elevated immune signalling ($p < 2e-6$) with increased expression of immune checkpoints, STING and increased immune cell infiltration. The MetHot group demonstrated greater genomic instability with amplification of 8q and higher incidence of TP53 mutations. The MetHot subgroup was found to have a significant association with poor survival outcome in TCGA (multivariable: $p < 0.008$) and the independent dataset (multivariable: $p < 0.01$). *In vitro* analysis of the role of cGAS and STING in invasion and migration in prostate cancer is ongoing.

Conclusions: We have identified and validated a poor prognostic subgroup, representing 10-20% of early prostate cancer patients that are at increased risk of developing metastatic disease and present with targetable immune biology. STING expression is high in these tumors. These patients may represent a viable target population for immune checkpoint therapies in prostate cancer, or therapies targeting the cGAS-STING pathway.

Conflicts of interest: ER, AMcC, SMW, NMcC, LAK and RDK are employees of Almac Diagnostics, and involved in the development and validation of the PCM and DDRD assays.

Funding acknowledgements: Invest Northern Ireland.