Introduction The chromatin landscape of mammalian cells underpins their transcriptional profiles, which then dictate fate and function. Mutations within chromatin remodelling genes have been implicated in early oesophageal adenocarcinoma (OAC) development, thus alterations in the chromatin landscape may pose an important molecular step in OAC development. OAC is often lethal, therefore genome-wide, basic research will provide foundations to develop new treatments and patient stratification methods.

Material and methods We have used Assay for Transposase-Accessible Chromatin using sequencing (ATAC-seq) to profile the accessible chromatin landscape of normal oesophagus and oesophageal adenocarcinoma tissue samples, in addition to representative cell lines. Downstream analysis has involved bioinformatic de novo motif analysis and ATAC-footprinting. Chromatin immunoprecipitation with sequencing (ChIP-seq) and siRNA-mediated knockdowns.

Results and discussions Our results revealed an altered chromatin landscape in cancerous tissue and identified ‘cancer-specific’ regions of accessibility. Using de novo motif discovery methods and footprinting analysis, we have identified enriched transcription factor motifs for GATA6 and HNF4A in differentially accessible regions and footprints. Also, comparison between the chromatin landscape of OAC cell-lines and OAC tissue identified the cell-line that best represents OAC tumours. HNF4A ChIP-seq and GATA6 ChIP-seq data, in oesophageal cancer cells, shows co-occupancy of HNF4A and GATA6 at 90% of sites and confirm motif enrichment and footprint observations. HNF4A and GATA6 knockout experiments demonstrate overlap of gene regulation and also identify genes that are exclusively regulated by the presence HNF4A and GATA6. Importantly, genes directly regulated by HNF4A or GATA6 are on average overexpressed in OAC, however, genes regulated by both factors are more overexpressed by 2-fold.

Conclusion Our data demonstrate the power of ATAC-seq in genome-wide discovery methods and we have identified a novel HNF4A-GATA6 transcription network that is active in OAC.

Material and methods We established TMZ resistant cell lines by two different methodologies, 1) dose escalation method and 2) high dose TMZ selection. For the dose escalation method, U373 cells were treated TMZ every two days, starting from 25 μM. Every two weeks, dosage was doubled (or when cells become resistant to that dosage) up to 250 μM. For high dose TMZ resistant selection, cells were treated with 250 μM TMZ for 15 days. Both cell lines have been kept under TMZ treatment for 3 months. To identify chemical inhibitors that target resistant cells, the TMZ-resistant cells and their parental controls were treated with a chemical library consisting of 90 epigenetic drugs against chromatin modifiers and cell viability was measured after 72 hours.

Results and discussions We successfully generated isogenic sub-populations of GBM cell lines that are resistant to TMZ. We observed that the high dosage TMZ treatment regimen caused a more sustainable resistant cell line compared to dose escalation regimen. Both resistant cell line models were stable, as they did not depend on prolonged TMZ treatment and remained resistant to TMZ after 3 months of drug holiday.

We then screened for epigenetic compounds that target TMZ-resistant cells. In addition to PARP inhibitors, Olaparib and Rucaparib, we identified Histone Deacetylase (HDAC) inhibitors as TMZ-sensitising agents. We are currently delineating the epigenetic alterations between parental and TMZ-resistant cells and assess the role of HDAC-mediated changes in TMZ-response by RNA-seq.

Conclusion Histone acetylation and deacetylation are interesting focus areas in drug resistance because of the central role of these modifications in many aspects of cell physiology and pathology. In our screen, we have shown that various HDAC inhibitors can sensitize GBM cells to TMZ in established cells as well as our newly generated in acquired TMZ resistance models.
Introduction Active surveillance (AS) for men with low-risk prostate cancer (PCA) currently depends on repeated biopsies for prognostication. We aim to identify molecular patterns that are present in initial PCA biopsies that inform future histopathological upgrading of PCA for men on AS.

Material and methods TAPS2.0 and Patchwork were respectively used to establish somatic copy number aberrations (SCNAs) for 44 Gleason score (GS)6 (3+3) TCGA PRAD Affymetrix SNP6.0 and 18 GS6 (3+3) ICGC WGS samples. Elastic net regularisation was used to identify SCNAs predictors of biochemical recurrence (BCR). In an internal cohort, SCNAs were established using QDNAseq from initial and repeat biopsies. Copy number losses were identified at 8 p (46%), 13q (23%), and 6q (46%), and were comparable in frequency to losses in TCGA and ICGC. None of the six predictors were detected in the initial PCA biopsy of the stable patient, while two predictors were found in men who had upgraded to GS7: 14q13 (gain), present in two men, and 6q11 (loss), in another man. Patients that upgraded had the highest similarities (81%) of SCNAs between patient-matched initial and repeat biopsies. Given that these biopsies were taken furthest apart in the prostate gland, missampling and GS is likely not to affect prediction.

Conclusion SCNAs indicative of upgrading were identified in GS6 TCGA-ICGC data using BCR as a surrogate for histopathological upgrading whilst on AS. These were observed to some extent in an internal cohort of men on AS. Validation of these predictors in men on AS is currently on-going in a further 8 men and may provide the basis for developing a prognostication tool to guide therapy for men with early prostate cancer.

PO-308 IDENTIFICATION OF GENOMIC PATTERNS PREDICTIVE OF UPGRADING IN LOW-GRADE PROSTATE CANCER

D Lee*. Ajou University School of Medicine, Pathology, Suwon, South Korea

Introduction Diffuse type gastric cancer (DGC) is a GC subtype with heterogeneous clinical outcomes. Lymph node metastasis of DGC heralds a dismal progression, which hampers curative treatment of patients. However, the genomic heterogeneity of DGC is still unknown.

Material and methods To identify genomic variations associated with lymph node metastasis of DGC, we performed whole exome sequencing on the 23 cases of DGC and paired non-tumoral tissues, and compared the mutation profiles according to the presence (N3, n=13) or absence (N0, n=10) of regional lymph node metastasis.

Results and discussions Overall, we identified 185 recurrently mutated genes in DGC, which included novel mutation at CMTM2 as well as previously known mutations at CDH1, RHOA, and TP53. Noticeably, CMTM2 expression could predict prognostic outcomes of DGC but not intestinal type GC (IGC), indicating pivotal roles of CMTM2 in DGC progression. In addition, we identified recurrent loss of heterozygosity (LOH) of DNA copy numbers at the 3p12-pcen locus in DGC. In comparison of N0 and N3 tumours, N3 tumours exhibited more frequent DNA copy number aberrations including copy-neutral LOH and mutations of CptTpt trimucleotides than those of N0 tumours (p=0.2 × 10^{-5}).

Conclusion In conclusion, DGCs have distinct profiles of somatic mutations and DNA copy number aberrations according to the status of lymph node metastasis, which might be helpful in delineating pathobiology of the DGC.