**Novel transcriptomic signature for predicting functional RB1-loss with clinical implications in localized and metastatic castration resistant prostate cancer**


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**Background, Methods, Results, Conclusions:**

RB1 is a well-characterized tumor suppressor, and bi-allelic RB1 loss of function has been implicated as a driver event in multiple cancer types. RB1 loss is typically assessed by examining DNA sequencing to look for inactivation by mutation, deletion, or structural variation. However, there may be uncertainty as to whether a DNA alteration is truly functional, or if other mechanisms of inactivation exist (e.g. epigenetic modification, post-transcriptional or post-translational modification). Therefore, we sought to develop a gene-expression based RB1-loss signature to capture the downstream changes that occur with RB1 loss regardless of etiology. To do so, we utilized cell line DNA sequencing and gene expression data from the Cancer Cell Line Encyclopedia (N=995) to develop the first pan-cancer RB1-loss signature (RBS), which we then independently validated in the TCGA pan-cancer dataset (N=11,060). The RBS achieved an AUC of 0.92 for discrimination of 2-copy RB1 loss, vastly outperforming existing signatures. RBS is also associated with poor survival in TCGA (p<0.0001). Since RB1 loss is an important genomic feature in prostate cancer, we examined existing published datasets in localized and metastatic prostate cancer with extensive clinical follow-up (not available in TCGA). In a localized prostate cancer dataset with long-term outcomes (N=260), we found that RBS was associated with greater risk of metastasis (p=0.009). In a cohort of 101 metastatic castration-resistant prostate cancer tumors, the RBS is similarly associated with worse survival (p<0.001) and is an excellent predictor of two-copy RB1 loss (AUC=0.77). Our study provides the first validated tool to assess RB1 loss based on downstream gene expression changes and can be utilized in datasets without DNA sequencing, or to classify variants of unknown significance.

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