Interferon-STAT1 signaling induces lineage plasticity of castration-resistant prostate cancer

Background: Research has revealed the association of prostate cancer stem cells (PCSCs) with initiation and progression of metastatic castration-resistant prostate cancer (mCRPC). CSCs are reservoir cancer cells possessing self-renewal capacities and abilities to differentiate into heterogeneous lineages. Tumor heterogeneity is the major cause of treatment failure in PCa. In particular, genetic alterations during the development of drug resistance in PCa empower the lineage plasticity to PCSCs. Despite accumulating studies have shown the involvement of PCSCs with cancer relapse, metastasis and drug resistance, mechanisms underlying the acquisition of lineage plasticity in PCSCs remained to be elucidated. The signal transducer and activator of transcription 1 and 2 (STAT1/2) are essential components in interferon (IFN)-induced signaling pathway. Studies have demonstrated that STAT1 is highly overexpressed in tumors with acquired radio-resistance *in vivo*, and facilitates tumor cell survival after irradiation *in vitro*. Meanwhile, the IFN-related DNA damage resistance signature (IRDS) encompassed a subset of STAT1-driven genes with pro-survival functions are associated with intrinsic resistance to chemotherapy and radiotherapy in many malignancies. However, the involvement of STAT1 signaling in driving lineage plasticity of mCRPC remain unclear.

Methods: Tumor sphere assay, cell viability MTT assay, and subcutaneous transplant model of PCa lines were applied to examine tumorigenesis *in vitro* and anti-tumor activity of STAT1 inhibitors *in vivo*, along with bioinformatics approaches such as RNA sequencing followed by Ingenuity pathway analysis and Gene Set Enrichment Analysis (GSEA) to study the clinical correlation of STAT1 in PCa progression.

Results: We observed several IFN-inducible STAT1-driven genes are significantly upregulated in metastatic PCa and CRPC lines with acquired lineage plasticity. Inhibition of JAK-STAT1 signaling by Fludarabine or Ruxolitinib suppresses self-renewal capacity of tumor spheres *in vitro*, and attenuates tumor growth *in vivo*. In particular, STAT1-mediated induction of IFIT5 appears to facilitate the acquisition of stemness properties in PCSCs via regulating Bmi1 and Sox2 through targeting miR-128 and miR-101. Loss of IFIT5 leads to attenuated sphere forming ability *in vitro*, and decreased tumor incidence *in vivo*.

Conclusion: This study delineates the mechanistic impact of STAT1-IFIT5-mediated microRNA turnover on the acquisition of lineage plasticity in advanced PCa.