Application of an antibody-independent, highly sensitive PRISM-SRM proteomics approach for monitoring TMPRSS2-ERG fusion proteins in prostate cancer cell lines and tumor tissues

**Overview**

- PRISM-SRM assays allow for highly sensitive detection and accurate quantification of TMPRSS2-ERG fusion protein products in prostate cancer cell lines and tumor tissues.
- At least two groups of ERG protein isoforms are simultaneously expressed in prostate cancer cells and tumor tissues at widely variable levels.
- ERG expression is highly correlated with TMPRSS2-ERG gene rearrangement.

**Methods**

**Introduction**

- Fusions between the transmembrane protease serine 2 (TMPRSS2) and ETS related gene (ERG) have been identified in ~50% of prostate cancer cases and define a distinct molecular subtype of prostate cancer.
- TMPRSS2-ERG gene rearrangement is typically detected using antibodies to the TPR domain of ERG and RT-PCR to detect the fusion transcript.
- An antibody-free strategy for quantifying low abundance TMPRSS2-ERG fusion protein products in prostate cancer cell lines and tumor tissues.
- Our results suggest that at least two groups of ERG protein isoforms are simultaneously expressed in prostate cancer cells and tumor tissues at widely variable levels.

**Experimental**

- TMPRSS2-ERG fusion protein SRM assays were developed using synthetic peptides.
- Samples comprised six prostate cancer cell lines (2 positive and 4 negative) and twelve tumor tissues (7 positive and 5 negative), all with genomics confirmation.
- Proteins were extracted from each sample, reduced and alkylated, and further digested with trypsin.
- Representative heavy peptide standards of various TMPRSS2-ERG fusion protein isoforms were spiked into each sample (1 µg/µL) to a final concentration of 5 nM.

**Conclusion**

- PRISM-SRM allows for detection and quantification of TMPRSS2-ERG fusion at the protein level.
- Multiple isoforms of TMPRSS2-ERG fusion protein products (i.e. truncated ERG protein) are simultaneously expressed in prostate cancer cell lines and tumor tissues at widely variable levels.
- ERG protein expression is highly correlated with TMPRSS2-ERG gene rearrangement.
- Future quantitative analyses of TMPRSS2-ERG fusion protein products in large cohorts of prostate tumor specimens should provide additional information on the biomarker potential of TMPRSS2-ERG protein products, e.g., stratifying prostate cancer risk.

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**Reference**