MSc DEFENCE Thursday, September 6th, 2018
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Title: LINE-1 RETROTRANSPOSITIONS IN EPITHELIAL OVARIAN CANCER: CAN WE USE DNA “PARASITES” FOR GOOD PURPOSE?
Time and location: 10:00 am PST; 12th floor meeting room, BC Cancer Research Centre, 675 West 10th Ave, Vancouver, BC
Supervisor: Dr. David Huntsman

ABSTRACT

Background/objective: High grade serous ovarian cancer (HGSC), endometrioid ovarian cancer (ENOC) and clear cell ovarian cancer (CCOC) are the three most common subtypes of ovarian cancers. While HGSC arise from serous tubal intraepithelial carcinomas (STIC) lesions in the fallopian tube, ENOC and CCOC share a common precursor lesion, endometriosis (ectopic growth of uterine lining). How ENOC and CCOC arise from the same precursor lesion is unknown, and effective biomarkers of early cancer development and recurrence are lacking.

We performed whole genome sequencing (WGS) on 29 ENOC and 35 CCOC cases and observed highly recurrent retrotransposition events originating from an active LINE-1 (L1) retrotransposon in the TTC28 gene in 34% and 31% of cases respectively. L1 retrotransposons are mobile repetitive genetic elements littered across our genome. They encode a set of protein machineries that helps them to “copy-and-paste” their own sequences into random genomic loci. High expression of the L1 protein has been found in HGSC cases. L1s are also capable of carrying and inserting unique downstream DNA sequences in a process called 3’ transduction. All these processes may fuel genomic instability, as such they are epigenetically silenced in normal tissues, but are found to become re-activated in epithelial cancers in association with global hypomethylation. As the gradual loss of L1 methylation has been found from normal endometrium to contiguous endometriosis and to ovarian tumors, we hypothesize that L1 activation occur early in ovarian cancer tumorigenesis and that TTC28-L1 transductions could be used as a marker of tumor development.

Methods: To compare the clonal relationship between TTC28-L1 transductions and somatic mutations, we used PCR and next-generation sequencing assays on formalin-fixed paraffin-embedded (FFPE) tumor tissues of different tumor samplings for 4 ENOC and 3 CCOC cases. To identify novel TTC28-L1 events in tumor and FFPE tissues, we developed a DNA-probe based target capture sequencing method and validated the assay on WGS cases with known retrotranspositions. L1 protein expression in HGSC, ENOC, and CCOC were assessed via immunohistochemical (IHC) staining of ovarian tissue microarray (TMA) and correlated with survival.

Results: TTC28 L1 retrotransposition events were present at all five tumor sites in 75% (6/7) of cases, while some SNV/frameshift mutations were either absent or were present at varying allelic frequencies. Target capture sequencing identified retrotransposition events corresponding to the events identified in WGS for the four cases tested, as well as in cases without WGS. L1 expression was found in 57% of ENOC and 63% of CCOC as well HGSC and their precursor lesions. Expression correlated with poor disease specific survival in ENOC (five-year cut off: hazard ratio=4.08, p=0.007).

Conclusion: our results suggest that TTC28-L1 events occur early in ovarian cancer development and may reflect the pre-malignant transformation of ENOC and CCOC. The use of L1 protein IHC and our target capture assay could be explored as a potential method to track such development.

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