Quantitating *PTEN* and *PTENp1* pseudogene expression: Potential for future cancer therapy

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The phosphatase and tensin homologue deleted on chromosome 10 (PTEN) is a tumour suppressor that is a vital antagonist of the oncogenic PI3K pathway. Subtle changes in cellular PTEN levels lead to oncogenesis, hence *PTEN* expression is tightly regulated at multiple levels. At the post-transcriptional level, PTEN is regulated by microRNAs and by its pseudogene, *PTENp1*, expressed as a long non-coding RNA. The *PTENp1*-sense (*PTENp1-S*) transcript increases PTEN levels by sponging microRNAs targeting *PTEN*. The *PTENp1*-anti-sense (*PTENp1-AS*) transcript binds to the PTEN promoter to lower PTEN transcription.

This study aimed to quantitate *PTEN* and *PTENp1* (sense and anti-sense) transcripts in various cancer cells, relative to non-cancer cells, to determine the alterations in transcript levels associated with carcinogenesis.

RNA was extracted from non-cancerous (HEK-293 kidney, HFF skin and PNT-2) and cancerous (MCF-7 breast, U-2OS bone, HCT-116 colon, U-87MG brain, LNCaP and PC-3 prostate) cells and cDNA synthesised. RT-qPCR was conducted using SYBR Green and primers specific for each transcript. Absolute quantitation of transcripts was carried out using a standard curve method from Ct values generated from serial dilutions of a standard (range of 10, 000 - 1 copy).

PTEN copy number was found to be significantly higher in all non-cancer cell lines compared to cancer cell lines. *PTENp1-S* transcript copy number was lower than *PTEN* and *PTENp1-AS* in all tested cancer and non-cancer cells. *PTENp1-AS* expression was higher compared to the *PTENp1-S* transcript in the cancer cell lines. Interestingly, later-stage prostate cancer cells showed decreases in both *PTENp1-S* and *PTENp1-AS* copy number compared to the early-stage prostate cancer cells.

Quantitation of *PTEN*, *PTENp1-S* and *PTENp1-AS* in cancer cells showed changes in copy number across the different cancer cell types, as well as with cancer progression in the case of prostate cancer. Further consolidation of this work in other cancers may lead to future novel therapies aimed at restoring PTEN levels by altering pseudogene transcript expression.