Phenotypical and molecular characterization of cell cultures derived from usual penile carcinomas.

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BACKGROUND: About half of penile cancers (PeCa) are usual squamous cell carcinomas (SCCs) subtype characterized by early loco-regional spread and high risk of metastatic dissemination. Recent progresses in the molecular research of penile tumors have been made, however the lack of well characterized *in vitro* models precludes new advances in the knowledge of cellular processes as well as in preclinical tests for anticancer drugs efficacy. Few cell lines derived from primary PeCa have been reported, with none being commercially available.
HYPOTHESIS: To establish and characterize cell cultures derived from two usual SCCs aiming to provide reliable in vitro models for functional and pre-clinical studies in PeCa. Cell cultures characterization included immunophenotyping and in vitro malignant assays. To identify molecular similarities between the parental tumors and cell cultures, transcriptomic profiles were also investigated.
METHODS: Usual penile squamous cell carcinomas were obtained from two patients (PeCa1 and PeCa2) and cultured in vitro. CK10 and CK14 expression were evaluated by immunofluorescence and EpCAM by flow cytometry. Clonogenic, soft agar colony formation, migration and invasion assays as well as xenograft in BALB/c nude mice were performed. Transcriptome profiles of the parental tumors and cell cultures in comparison with normal glans (NG) were assessed using Human Transcriptome Array 2.0 platform (Affymetrix).
RESULTS: PeCa1 and PeCa2 cells were successfully established until the 10^{th} passage (P10). Both cells presented high expression of CK14 and EpCAM and low levels of CK10, indicating a proliferative basal keratinocyte phenotype. PeCa1 and PeCa2 cells showed ability to anchorage-independent growth in soft agar. Nevertheless, PeCa1 and PeCa2 cells were unable to invade matrigel and to form tumors in nude mice. Transcriptome analysis identified 57 differentially expressed genes shared between tumors and cell cultures compared with NG. These genes are involved in different pathways, such as AKT/PI3K and PPARA. Interestingly ten altered pathways detected in these cells were commonly altered in an independent cohort of PeCa, previously investigated by our group. To identify genes modulated during *in vitro* conditions, transcriptomic profiles of cell cultures were compared with the parental tumors; the differentially expressed transcripts detected were mainly related to cell attachment, extracellular matrix interaction and oxidative stress response. Altogether these data indicate that PeCa1 and PeCa2 cells retained important deregulated pathways detected in penile tumors, suggesting that they are reliable models to be applied to tumorigenesis investigation and preclinical trials in PeCa.