Phase I/II CANON study: Oncolytic immunotherapy for the treatment of Non-Muscle Invasive Bladder Cancer using intravesical Coxsackievirus A21

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Introduction

The treatment of non-muscle invasive bladder cancer (NMIBC) has not changed significantly in 25 years. Treatment with intravesical BCG (Bacillus Calmette Guerin) and chemotherapy after transurethral resection of tumor (TURBT) does not alter the natural history of the disease and has significant toxicity for patients. CAVATAK™ (Coxsackievirus A21, CVA21) is a novel intercellular adhesion molecule-1 (ICAM-1)-targeted immunotherapeutic virus. Surface ICAM-1 is up-regulated on a number of cancers including melanoma, non-small cell lung, prostate and in particular, bladder cancer. CVA21 displays potent oncolytic activity against monolayer cultures of NMIBC cancer cells. Combining CVA21 with either radiotherapy or chemotherapy (mitomycin C) synergistically enhances cytotoxicity in vitro in bladder cancer cell lines. Low (non-cytotoxic) doses of mitomycin C enhances CVA21 viral replication and oncolysis by increasing expression levels of ICAM-1 on bladder cancer cells. In this two stage Phase I study, patients with NMIBC received neoadjuvant CVA21 or low dose mitomycin C plus CVA21 prior to routine surgical removal (TURBT). We present preliminary data from the Phase III CANON (CAVATAK in Non-Muscle invasive bladder cancer; NCT02316171) study which investigated the tolerance of multiple escalating intravesical doses of CVA21 in 16 first-line NMIBC patients.

Results

![CVA21 viral protein staining. Red/CVA21 proteins, blue/DAPI, H&E stain, black arrows indicate apoptotic bodies. Apoptotic cell staining. Brown cells represent cleaved caspase-3 staining by ICC.](image)

Figure 2. Levels of CVA21 cytoplasmic replication and viral-induced apoptosis in transurethral resection tissue

![Unpublished NMIBC vs CVA21 treated NMIBC.](image)

Figure 3. Levels of CVA21 viral RNA and live virus in patient urine following intravesical CVA21 administration

![Figure 5. Coxsackievirus A21 injection up-regulates interferon-induced genes and immune checkpoint molecules within the micro-environment of NMIBC tissue (NanoString analysis Pan Cancer Immune Profiling Panel).](image)

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Conclusions

- Proof of concept viral targeting, replication and tumor cell death following a single or multiple intravesical administrations of CVA21 was achieved in patients from monotherapy Cohorts A1, A2, A3 and mitomycin-C combination Cohorts B1, B2.
- Clinical activity of CVA21 demonstrated by complete tumor response (Figure 1), viral mediated cell apoptosis (Figure 2), viral replication (infectious virus increases in urine, Figure 3) and notable signs of viral-induced tumor inflammation.
- Single agent and CVA21-combination treatments facilitated notable changes within the NMIBC tissue by inducing increases in immune cell infiltrates (ICD3+CD8+ and expression of PD-L1 compared to untreated NMIBC controls (Figure 4).
- CVA21 treatment up-regulated a number of interferon-response and immune checkpoint inhibitory genes in NMIBC biopsy tissue, including CXCL10, CXCL11, PD-L1, PD-L2, IDO and LAG-3 (Figure 4).
- CVA21 mediated increases in "immunological heat" within the tumor micro-environment with regards to immune-cell infiltrates and up-regulation of immune checkpoint molecules suggest possible increased anti-tumor activity when used in combination with immune checkpoint blockade strategies.
- No evidence of systemic spread of CVA21 or development of anti-CVA21 serum neutralizing antibody.
- Intravesical administration of CVA21 as a single agent or in combination with mitomycin-C was generally well tolerated with no Grade 2.3 or 4 product-related AEs.
- The observed tumor targeting, viral replication is likely to provide a strong signal in generating both a strong local and systemic anti-tumor immune response.