Investigation of Oncolytic Coxsackievirus A21 as a Potential Treatment for Pancreatic Cancer

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Abstract

Pancreatic cancer (PC) is the fourth leading cause of cancer deaths and has the lowest 5-year survival rate of any cancer type at 6%. Coxsackievirus A21 (CVA21: CAVATAK™), a common cold virus, is being evaluated in clinical trials for metastatic melanoma, non-muscle invasive bladder cancer, and in combination with ipilimumab in advanced melanoma. CVA21 utilizes Intercellular adhesion molecule-1 (ICAM-1) and decay accelerating factor (DAF) to invade and lyse cells. ICAM-1 and DAF are commonly overexpressed on cancer cells.

The expression levels of ICAM-1 and DAF on a panel of human PC cell lines were determined through qRTPCR, immunofluorescence, and FACS assays. Viral infectivity, and growth curve assays were performed to quantify the 50% tissue culture infectious dose ($TCID_{50}$), and growth kinetics of CVA21 in the panel of PC cell lines. Normal human pancreatic ductal epithelial cells were tested alongside as controls. Findings indicate that pancreatic cancer cells highly overexpress ICAM-1 and DAF, and are highly susceptible to CVA21 oncolysis, compared to normal pancreatic ductal epithelial cells.

Conclusions

- Pancreatic cancer cells highly overexpress ICAM-1 and DAF *in vitro* compared to normal pancreatic ductal epithelial cells.
- Pancreatic cancer cells are highly susceptible to CVA21 oncolysis *in vitro* compared to normal pancreatic ductal epithelial cells.

Acknowledgements

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Research was conducted at the Hunter Medical Research Institute, NSW, Australia.

Methods and Results

ICAM-1 and DAF Expression Levels on a Panel of Human Pancreatic Cancer Cell Lines

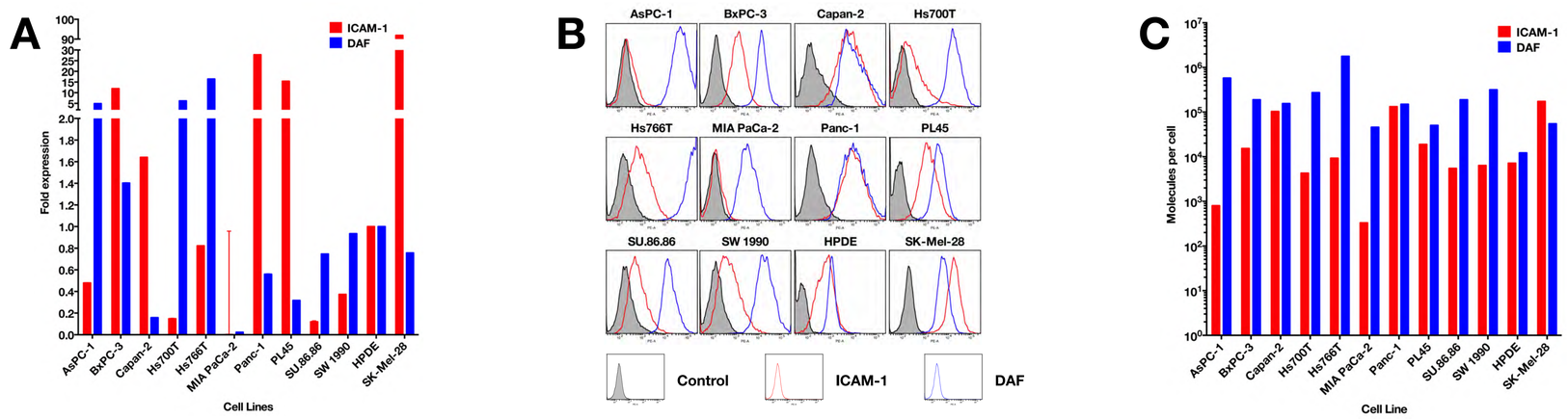

Figure 1: ICAM-1 and DAF expression levels. A: ICAM-1 and DAF gene expression via qRTPCR (n=3), **B:** ICAM-1 and DAF expression determined by FACS (n=10 000), and **C:** ICAM-1 and DAF molecules per cells determined using BD QUANTIBRITE™ PE BEADS (n=10 000), on a panel of human pancreatic cancer cell lines (AsPC-1, BxPC-3, Capan-2, Hs700T, Hs766T, MIA PaCa-2, Panc-1, PL45, SU.86.86, and SW1990). Normal human pancreatic ductal epithelial (HPDE) cells were tested alongside as controls. SK-Mel-28 ICAM-1 and DAF expression levels were also determined as a comparison to another cancer type, melanoma, which is highly susceptible to CVA21 oncolysis

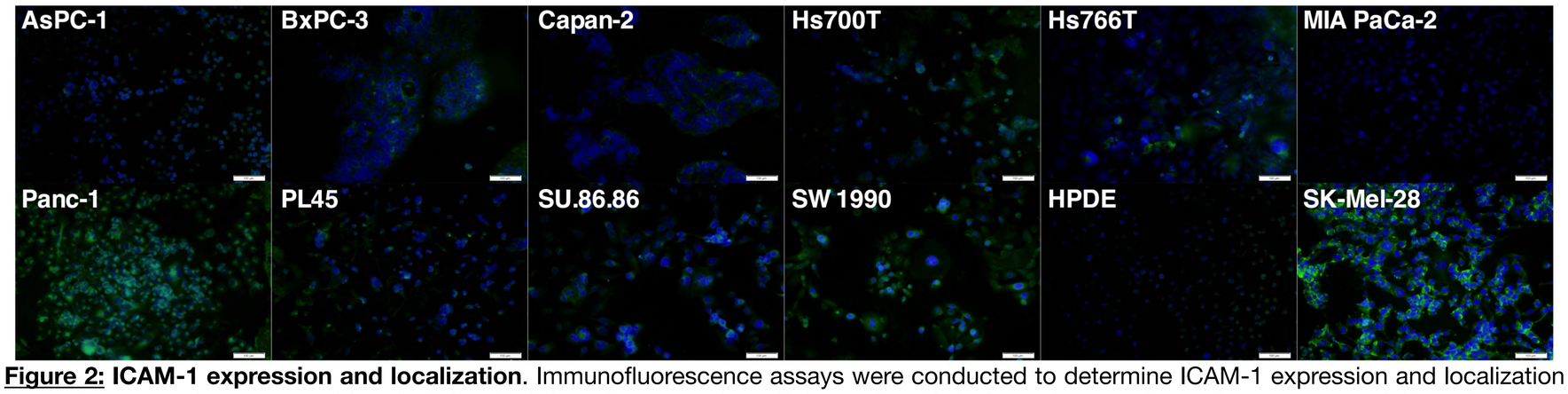

Figure 2: ICAM-1 expression and localization. Immunofluorescence assays were conducted to determine ICAM-1 expression and localization on each pancreatic cancer cell line (AsPC-1, BxPC-3, Capan-2, Hs700T, Hs766T, MIA PaCa-2, Panc-1, PL45, SU.86.86, and SW1990), normal human pancreatic ductal epithelial (HPDE) cells (control), and SK-Mel-28 cells. Blue: Hoechst (DNA) stain, Green: 1° mouse anti-human ICAM-1 mAb + 2° goat anti-mouse IgG FITC.

Susceptibility of a Panel of Human Pancreatic Cancer Cell Lines to Coxsackievirus A21 (CVA21)

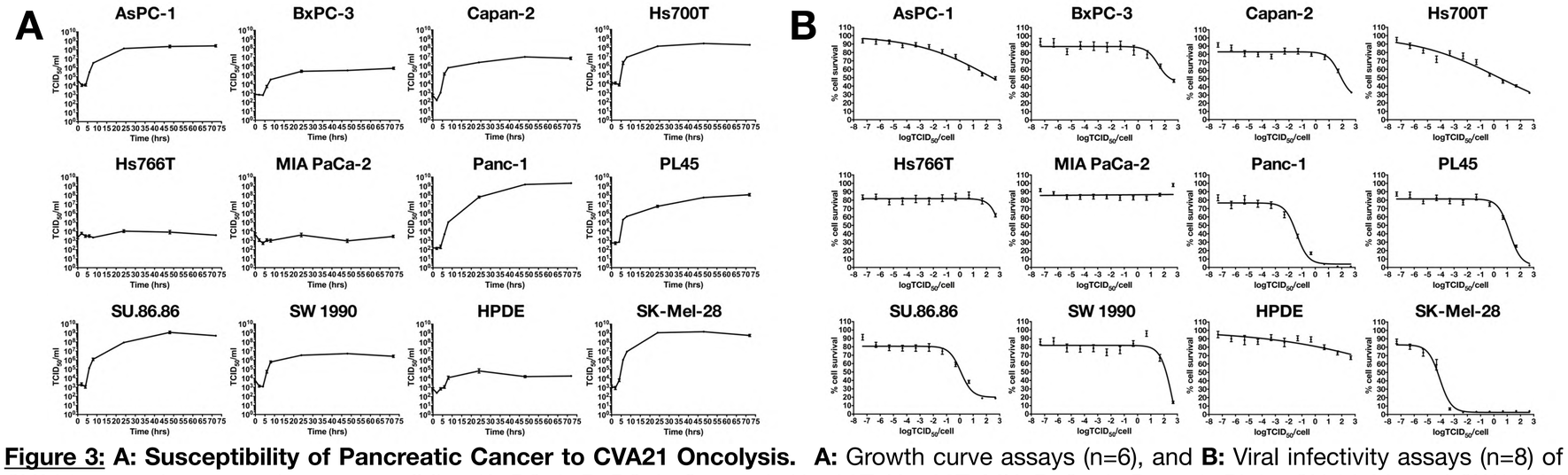

Figure 3: A: Susceptibility of Pancreatic Cancer to CVA21 Oncolysis. A: Growth curve assays (n=6), and **B:** Viral infectivity assays (n=8) of CVA21 on each pancreatic cancer cell line (AsPC-1, BxPC-3, Capan-2, Hs700T, Hs766T, MIA PaCa-2, Panc-1, PL45, SU.86.86, and SW1990), normal human pancreatic ductal epithelial (HPDE) cells (control), and SK-Mel-28 cells.