Flow cytometric phenotyping of diverse human cancer cell lines for immunological biomarkers expression

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Flow cytometric phenotyping of diverse human cancer cell lines for immunological biomarker expression
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Introduction

• Cancer immunoediting is a process that cancer cells undergo to evade the immune system (Figure 1) (Swann & Smyth, 2007).
• To eliminate nascent tumours, death receptors (DR) 4/5 and Fas respond to tumour necrosis factor-related apoptosis-inducing ligand and Fas Ligand, respectively, to induce cancer cell apoptosis.
• To escape immune-mediated killing, cancer cells can reduce the expression of death receptors (Shin et al., 2001) and major histocompatibility complex (MHC) class I, as well as express co-inhibitory molecules including programmed death ligand 1 (PD-L1) (Escors et al., 2018).
• Tumour escape is one of the hallmarks of cancer and it is necessary for tumour progression (Hanahan & Weinberg, 2011).
• By targeting these immune markers alone or in combination could potentially increase cancer cell death and improve drug efficacy.

Aims and Hypothesis

• This research aimed to determine the cell surface expression of immune markers including PD-L1, MHC class I, DR 4/5 and Fas in diverse human cancer cell lines.
• It was hypothesised that human breast, prostate and colorectal cancer cell lines would express PD-L1, MHC class I, DR 4/5 and Fas.

Methods

Cell culture Human cancer cell lines were cultured in DMEM (SW480 and SW480) or RPMI (MDA-MB-231, MCF-7, PC3 and LNCaP) media supplemented with 10% FBS and 1% penicillin-streptomycin.

Cell surface staining Human cancer cell lines (5 x 10⁴) were stained with fluorescently labelled anti-human PD-L1, HLA-ABC, DR/5 and Fas antibodies and their matched isotype controls. Cell surface expression was assessed using flow cytometry and data was analysed using FlowJo software.

Results

Figure 2: Breast, prostate and colorectal cancer cell lines express differential levels of cell surface PD-L1. A) Representative flow cytometry plots are shown for each human cancer cell line. They show the difference between the isotype control (grey) and the PD-L1 positive population. B) Breast cancer cell line MDA-MB-231 express high levels of PD-L1. Breast (MCF-7), prostate (LNCaP and PC3) and colorectal (SW480 and SW480) cancer cell lines express low levels of PD-L1. Data is presented as mean ± SD, n=3.

Figure 3: Breast, prostate and colorectal cancer cell lines express differential levels of cell surface MHC class I. A) Representative flow cytometry plots are shown for each human cancer cell line. They show the difference between the isotype control (grey) and the MHC class I (HLA-ABC) positive population. B) Breast (MDA-MB-231 and MCF-7), prostate (LNCaP and PC3) and colorectal (SW480 and SW480) cancer cell lines express moderate to high levels of MHC class I. Data is presented as mean ± SD, n=3.

Conclusions

• The cell surface expression of immune markers on breast, prostate and colorectal cancer cell lines is different, and can be utilised as diagnostic biomarkers.

References