Expansion of Tumour-Infiltrating Lymphocytes (TIL) from Prostate Cancer and Adjacent Normal Tissue

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Background: Despite not characteristically being associated with immune activation, prostate cancer was the first malignancy to demonstrate improved survival with a cancer-specific vaccine (Sipuleucel-T); these findings have led to intense interest in the use of other modalities of immunotherapy for the treatment of prostate cancer. To date, there have been limited attempts to expand tumour-infiltrating lymphocytes (TIL) from prostate cancer tissue. The ability to expand prostate TIL *ex vivo* may pave the way for developing adoptive cell therapy for prostate cancer, and may also yield insights into the immune microenvironment of prostate tumours.

Methods: Tissue specimens were obtained from 10 patients undergoing standard radical prostatectomy and 5 patients undergoing radical prostatectomy following 6 months of neoadjuvant degaralix (NCT01674270). Tumour-infiltrating lymphocyte cultures were initiated from small tissue fragments, in media containing human plasma and human recombinant interleukin-2. Cells were expanded for approximately 4 weeks and then enumerated and characterized by flow cytometry.

Results: The ability of TIL to expand *ex vivo* was heterogeneous amongst patients. Of the 15 tumour specimens analyzed, 10 yielded at least $30x10^6$ TIL within approximately 4 weeks of culture. Four specimens reached $1x10^8$ cells; the threshold for the number of cells generally needed for current protocols for generating a TIL product for investigational treatment. Flow cytometric analysis of the expanded TIL showed a high proportion of CD3+ T cells (e.g. average 91% in the 4 cultures reaching $1x10^8$ cells), with CD19+ B cells and CD14+ monocytes generally absent in all expanded cultures. The CD4: CD8 ratios were variable amongst cultures. Interestingly, T cells could also be expanded from normal adjacent tissue, although there was not an association between the number of cells obtained from expansion of normal adjacent tissue and tumour tissue.

Conclusions: In this study we used techniques previously optimised in metastatic melanoma to expand T cells from prostate tumours and their normal adjacent tissues.
Our results show that TIL can be successfully expanded from both sites. This finding presents the possibility of developing adoptive cell therapy using TIL for prostate cancer. In addition, expanded TIL represent a valuable tool for evaluating the repertoire of antigen specificities of the T cell population present within the tumor microenvironment and normal adjacent tissue.