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TUMOUR IMMUNOLOGY

A triple blow for cancer

Although individual immune checkpoint inhibitors have shown promise as cancer immunotherapies, recent data suggest that greater success will be achieved by combining these inhibitors with each other and possibly with other therapies, such as radiation. Reporting in *Nature*, Minn and colleagues show that combining antibodies that target the programmed cell death protein 1 (PD1) ligand 1 (PDL1)-mediated and cytotoxic T lymphocyte antigen 4 (CTLA4)-mediated immune checkpoints with radiation promotes effective antitumour immunity through distinct, non-redundant mechanisms.

> “high PDL1 expression on tumour cells is a dominant resistance mechanism to radiation plus CTLA4-blocking therapy”

In a Phase I clinical trial of 22 patients with metastatic multiple melanoma, a single lesion was irradiated followed by four rounds of treatment with the CTLA4-specific monoclonal antibody ipilimumab. Analysis of the non-irradiated melanoma lesions showed that 18% of patients had a partial response and another 18% had stable disease after treatment, indicating that, in a subset of patients, combined radiation and ipilimumab treatment can have beneficial effects. However, 64% of the patients were resistant to treatment and had progressive disease.

To understand this resistance, the authors moved to the B6-F10 melanoma mouse model, in which a similar response rate to the radiation plus CTLA4-blocking treatment was observed. Complete responses to therapy were $CD8^+$ T cell dependent, and the best predictor of resistance was the failure to increase the ratio of $CD8^+CD44^+$ T cells to regulatory T (T_{Reg}) cells ($CD8/T_{Reg}$ ratio) within the tumour-infiltrating lymphocyte (TIL) population. The $CD8/T_{Reg}$ ratio did not increase in resistant tumours despite a reduction in the number of T_{Reg} cells (which was similar to that observed in sensitive tumours) because $CD8^+$ T cells failed to accumulate. This defect in $CD8^+$ T cell accumulation in resistant tumours was due to the increased expression of PDL1 on melanoma cells. A comparable increase in PDL1 expression was observed on resistant breast cancer cells and on resistant tumour cells from patients with metastatic multiple melanoma in the clinical trial.

Increased levels of PDL1 can promote T cell exhaustion (defined by assessing Ki67 and granzyme B expression in $PD1^+EOMES^+CD8^+$ T cells), the reversal of which is termed reinvigoration. In susceptible tumours, the percentage of reinvigorated relative to exhausted $CD8^+$ T cells in the total TIL population was increased following radiation plus CTLA4-blocking treatment, whereas in resistant tumours, there was no increase in the number of reinvigorated $CD8^+$ T cells. However, the addition of a PDL1-specific blocking antibody to the dual treatment increased the number of reinvigorated $CD8^+$ TILs, as well as the $CD8/T_{Reg}$ ratio. Importantly, the complete response rate of mice with melanoma improved from 17% with the dual therapy to 80% following treatment with the triple combination of CTLA4 and PDL1 (or PD1) blockade and radiation.

Further analysis showed that these three therapies function in a non-redundant but complementary manner: PDL1-blocking therapy reinvigorates exhausted $CD8^+$ T cells, CTLA4-blocking therapy predominantly decreases T_{Reg} cell numbers and, together, these immune checkpoint inhibitors increase the $CD8/T_{Reg}$ ratio and promote the peripheral clonal expansion of TILs. The predominant role of radiation is to diversify the T cell receptor repertoire of TILs and shape the repertoire of the expanded peripheral clones.

Of note, markers of exhaustion and reinvigoration in peripheral T cells, as well as the $CD8/T_{Reg}$ ratio in the blood, were shown to predict the responsiveness of mice to therapy. Furthermore, in the clinical trial patients, a high level of PDL1 expression on melanoma cells from pretreatment tumour biopsies was associated with persistent T cell exhaustion and rapid disease progression after radiation plus ipilimumab treatment.

These data show that high PDL1 expression on tumour cells is a dominant resistance mechanism to radiation plus CTLA4-blocking therapy. They support the exploration of a therapy that combines blockade of both PDL1 (or PD1) and CTLA4 with radiation for the treatment of melanoma and possibly other solid tumours.

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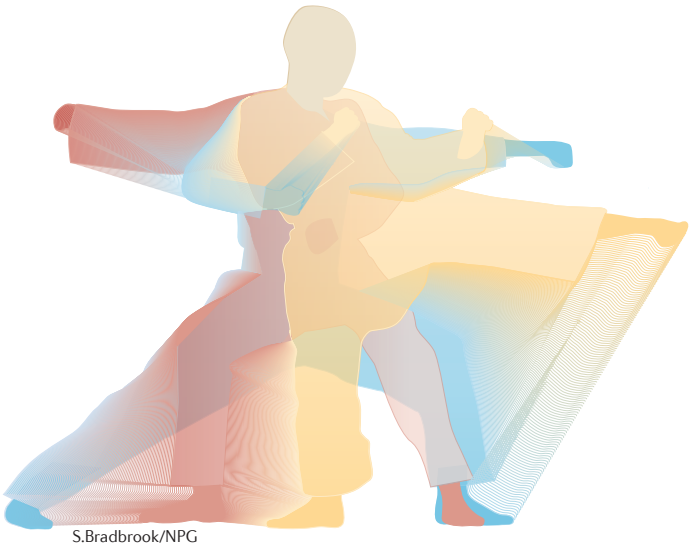

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ORIGINAL RESEARCH PAPER
Twyman-Saint Victor, C. *et al.* Radiation and dual checkpoint blockade activate non-redundant immune mechanisms in cancer. *Nature* http://dx.doi.org/10.1038/nature14292 (2015)