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### Research interests

- Probiotic–host cell interactions
- Gastrointestinal immunology
- Dendritic cell biology
- Clinical applications for immunotherapy
- Molecular mechanisms of bacterial–host communication
- Biochemistry, physiology, and genetics of commensal bacterial flora
- Clinical efficacy of probiotic consumption

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## Bacterial stimulation combined with heat shock protein targeting induces potent dendritic cell anti-tumor responses in a murine model

**Background:** Appropriate activation of the immune system and effective targeting of tumor cells are the primary hurdles to be overcome for cancer immunotherapies. One approach is to pulse dendritic cells (DCs) *in vitro* with biologically active compounds that commit them to cytokine secretion and expression of co-stimulatory molecules coupled with loading tumor antigens before re-injection into the tumor-bearing host.

**Aim:** To examine the ability of bacterial-stimulated dendritic cells, loaded with tumor-derived heat shock proteins (HSPs), to induce tumor cell destruction in a murine model.

**Methods:** Immature murine bone marrow-derived DCs were stimulated *in vitro* with a range of bacterial fractions in order to select the stimulus that induced maximal TNF $\alpha$  production, measured by ELISA, and co-stimulatory molecule expression, measured by flow cytometry. The tumor cell 4T1 was chosen because of its aggressive and immunosuppressive phenotype [e.g. TGF $\beta$  production]. HSPs from the tumor cells were isolated using heparin affinity chromatography and were co-incubated with bacterial-stimulated DCs. Activated DCs were then administered *s.c.* Tumors were generated by orthotopic inoculation of 4T1 cells in female Balb/c mice. Primary tumor growth was measured using Vernier calipers while lung metastasis were measured using the clonogenic assay.

**Results:** Membrane fractions from *Salmonella typhimurium* induced the most potent TNF $\alpha$  response from DCs accompanied by significant up-regulation of CD80 and CD86 expression. When injected into mice, bacterial-stimulated DCs, loaded with 4T1 HSPs, inhibited the formation of new 4T1 tumors (1 of 7 treated mice developed a tumor compared to 7 of 7 mice in the placebo group) and reduced the growth rate of established tumors. In addition, the numbers of lung metastasis were significantly reduced in the DC-treated versus placebo-treated mice (1.6 $\pm$ 0.6 versus 245.9 $\pm$ 55.6 metastatic nodules respectively,  $p=0.0015$ ). DCs stimulated with *S. typhimurium* alone, exposed to HSPs alone or exposed to HSPs from an unrelated tumor cell line did not induce a protective immune response.

**Conclusions:** DCs primed with a proinflammatory stimulus and tumor derived HSPs induce a protective anti-tumor immune response in this murine model. This model suggests that when stimulated appropriately, DCs have the capacity to stimulate a tumor specific response successfully, even against immunosuppressive tumors which thrive in an immunocompetent host.