Abstract

Cancer immunotherapy relies on presentation of shared- and neo-antigens from a patient’s tumor to the immune system for recognition and clearance by the immune system. However, the tumor microenvironment deploys multiple strategies to evade immune recognition and often remains non-immunogenic, which is one of the challenges that need to be addressed when designing these therapies.

We set out to test whether intratumoral electroporation of Gp96-IgFc-OX40L, a re-engineered molecular chaperone, designed to export and deliver MHC I-associated antigens to APCs in context of OX40L expression, would generate a robust anti-neoantigen CD8+ T cell response. To assess a primary and specific CD8+ expansion, mice were adoptively transferred with OT-I cells after B16.F10-ova/vabulin cells were injected to generate primary and contralateral melanoma tumors. Contralateral tumors were harvested to assess whether a systemic CD8+ T cell response could be elicited following primary tumor electroporation. It electroporation of DNA expressing Gp96-IgFc-OX40L in the primary tumor triggered a significant expansion of antigen-specific OT-I cells, which was absent in control mice. Remarkably, increases in antigen-specific OT-I cells correlated with regression of both the treated and untreated contralateral tumors.

We further validated our findings in a CT26 mouse colorectal cancer tumor model, in which the expression of Gp96-IgFc-OX40L from electroporated DNA stimulated an expansion of antigen-specific CD8+ T cells and again led to regression of both the treated primary and untreated contralateral tumors.

Our findings demonstrate that manipulation of tumor-specific antigens to APCs in context of OX40L expression, would evade immune recognition and often remains non-immunogenic, which is one of the challenges that need to be addressed when designing new therapies.

Results – CT26 colon cancer tumor model

Figure 2. Intratumoral EP of ComPACT leads to ova-antigen specific CD8+ T cell expansion in vivo. A. C57BL/6 albinos mice that were adoptively transferred with OVA-CD8+ T cells and B16.F10-ova tumors were electroporated with either saline (EP only) or ComPACT DNA. The percentage of CD8+ OT-1 cells in peripheral blood was monitored over time by flow cytometry. B. Phenotypic analysis of ova-antigen specific CD8+ T cells on day 12 following EP by flow cytometry reveals increased number of CD127+KLRG-1 memory precursor cells in mice EP’d with ComPACT. C. Overall survival of B16.F10 melanoma bearing EP’d with saline or ComPACT DNA. * indicates p<0.05. Statistical significance was determined by student t-test and Mantel-Cox test.

Key Points

- In vivo EP of DNA-based Gp96-IgFc-OX40L into B16 melanoma and CT26 colon cancer tumors result in delayed tumor progression of treated and untreated tumors.
- Intratumoral expression of Gp96-IgFc-OX40L stimulates CD8+ T cell cross priming to tumor specific neoantigens and increases the frequency of circulating memory precursor cells.
- Electroporation-based delivery of Gp96-IgFc-OX40L DNA in combination with other immuno-modulatory DNA could lead to synergistic anti-tumor activity.

Contact information – sales@heatbio.com