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

Our findings demonstrate that Gp96.Ig serves as a molecular chaperone and adjuvant that presents tumor-specific neoantigens to APCs to trigger an anti-tumor CTL response. To assess antigen-specific CD8+ expansion, mice were adoptively transferred with OT-I cells from a patient’s tumor cells for recognition and clearance by the immune system. Our findings demonstrate that intratumoral electroporation of Gp96-Ig/Fc-OX40L in the primary tumor triggered a significant expansion of antigen-specific CD8+ T cells, which was absent in control mice. Remarkably, increases in antigen-specific CD8+ T cells correlated with regression of both the treated primary and untreated contralateral tumors.

We further validated our findings in a CT26 mouse colorectal cancer tumor model, in which the expression of Gp96-Ig/Fc-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 in situ manipulation of intratumoral cells to express Gp96-Ig/Fc-OX40L stimulates potential antigen-specific cross priming to tumor specific neoantigens that culminates in robust systemic anti-tumor response. These findings provide exciting proof-of-principle and warrant further investigation into the direct delivery of molecular chaperones such as Gp96-Ig/Fc-OX40L and/or pro-inflammatory molecules for elevating the immunogenicity of tumors for a potent anti-tumor CD8+ T cell response.