Background There is a need for novel immunotherapies to address patient populations resistant or refractory to immune checkpoint inhibitors (CPI). VV1 is an oncolytic vesicular stomatitis virus engineered to express interferon beta (IFNβ) to enhance anti-tumor immune responses and tumor selectivity. Phase 1 studies demonstrated VV1 anti-tumor activity in certain clinical settings either alone or in combination with a CPI. The current preclinical study aimed to identify dosing and scheduling regimens that maximize the efficacy of VV1 in combination with CPIs.

Methods Immune-competent mice bearing syngeneic tumor models MC38 (colorectal), B16F10 (melanoma), and CMT64 (lung adenocarcinoma) tumors were dosed to test combinations of VV1 delivered intravenously with anti-PD-1 and anti-CTLA4 antibodies. Treatment was started once tumors were established, and T cell responses in the tumor and peripheral lymphoid organs were analyzed using flow cytometry and ELISPOT assays.

Results Potent anti-tumor efficacy was observed following intratumoral or intravenous administration of VV1 combined with anti-CTLA4 and anti-PD-1. A comparison of single dose versus repeat administration of anti-CTLA4 in combination with VV1 plus continuous anti-PD-1 showed that a single dose was sufficient to maximally enhance the depth and durability of tumor response. This effect was observed consistently in multiple tumor models, including anti-PD-1 sensitive (MC38) or anti-PD-1 resistant tumor models (B16F10 and CMT64). Remarkably, the triple combination boosted T cell priming against the B16F10 and CMT64 neo-antigen peptides in TILs and tumor-draining lymph nodes. ELISPOT and multimer staining showed that, in contrast to doublet therapy using virus and a single CPI, triplet combination therapy strongly boosted CTL responses against a broad array of B16F10 and CMT64 neoepitopes, detected both in the tumor and in tumor-draining lymph nodes. This was associated with increased infiltration of CD8 T cells in the tumor, but the number of regulatory T cells was not impacted, indicating that the enhancing effect of anti-CTLA4 was not a consequence of Treg depletion.

Conclusions Intratumoral or intravenous VV1 virotherapy combined with anti-CTLA4 and anti-PD-1 checkpoint antibodies synergistically enhances tumor control in multiple syngeneic mouse tumor models. The triple combination seems to promote a tumor-vaccination effect, by inducing a polyclonal anti-tumor T cell response and boosting anti-tumor CTL responses. This triplet combination approach will soon be evaluated clinically in patients with advanced melanoma (after progression on an anti-PD1) and first line NSCLC patients. (ClinicalTrials.gov Identifier: NCT04291105).