

Abstract

Background/Aim: Osteosarcoma (OS) is refractory to immune checkpoint inhibitors targeting programmed cell death 1 (PD1)/PD ligand 1 (PD-L1) due to poor immune response. We previously developed telomerase-specific, replication-competent oncolytic adenoviruses non-armed OBP-301 and P53-armed OBP-702 that exert antitumor efficacy against human OS cells. Recently, we demonstrated that P53-armed OBP-702 induces more profound immunogenic cell death and antitumor immune response against human and murine OS cells than does non-armed OBP-301. In the present study, we assessed the combined efficacy of PD1 blockade and P53-armed OBP-702 against murine OS cells. **Materials and Methods:** Three murine OS cell lines (K7M2, NHOS, NHOS-LM4) were used to assess the cytopathic effect of non-armed OBP-301 and P53-armed OBP-702 by XTT assay. Virus-induced immunogenic cell death was assessed by analyzing the levels of extracellular adenosine triphosphate and high-mobility group box protein B1. The expression of PD-L1 and PD-L2 was analyzed by flow cytometry. The malignant potential of NHOS-LM4 cells was analyzed by a migration and invasion assay. An orthotopic NHOS-LM4 tumor model was used to evaluate the antitumor efficacy of combination therapy with P53-armed OBP-702 and anti-PD1. **Results:** P53-armed OBP-702 exhibited antitumor potential for the induction of immunogenic cell death, apoptosis, autophagy, and PD-L1/2 upregulation in K7M2 and NHOS cells. NHOS-LM4 cells showed increased migratory and invasive ability compared to NHOS cells. P53-armed OBP-702 significantly suppressed the malignant potential of NHOS-LM4 cells. Combination dosing showed that P53-armed OBP-702 significantly promoted the antitumor effect of PD1 blockade against NHOS-LM4 tumors. **Conclusion:** Our

results suggest that P53-armed OBP-702 is a promising agent for improving the antitumor effect of PD1 blockade in treating invasive OS.