P1:102

p53-mediated apoptosis induction attenuates the resistance to oncolytic adenovirus in human osteosarcoma cells

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Background: We recently revealed that a telomerase-specific replication-competent oncolytic adenovirus, OBP-301, shows the cytopathic activity in human bone and soft tissue sarcoma cells. However, some human osteosarcoma cells were less susceptible to OBP-301. In this study, we generated OBP-702, in which a human wild-type p53 gene expression cassette was inserted into the E3 region of OBP-301, and compared the antitumor effects between OBP-702 and OBP-301 in OBP-301-sensitive and OBP-301-resistant human osteosarcoma cells.
Methods: We used three OBP-301-sensitive (U2OS, OST, HOS) and two OBP-301-resistant (SaOS-2, MNNG/HOS) human osteosarcoma cells. The difference of cytopathic activity between OBP-702 and OBP-301 was analyzed using XTT assay. The 50% inhibiting dose (ID50) value of OBP-702 and OBP-301 for each cell line was calculated using cell viability data obtained on day 5 after virus infection. Induction of apoptosis was assessed in OBP-301-resistant osteosarcoma cells infected with OBP-702, OBP-301 or Ad-p53, which is a p53-expressing replication-deficient adenovirus, by FACS analysis measuring active caspase-3 expression. The expressions of p53, p21 and cleaved PARP proteins were evaluated using western blot analysis. The in vivo antitumor effect of OBP-702, OBP-301 and Ad-p53 was studied using orthotopic human osteosarcoma MNNG/HOS tumor model with total three intratumoral injections every 2 days.
Results: OBP-702 showed more cytopathic activity than OBP-301 in both OBP-301-sensitive and OBP-301-resistant osteosarcoma cells. The ID50 value of OBP-702 was lower than that of OBP-301 in all cell lines. FACS analysis demonstrated that OBP-702 significantly increased active caspase-3 compared with Ad-p53 and OBP-301. OBP-702 induced higher expression of p53 and cleaved PARP than Ad-p53. However, p21 up-regulation was not observed in SaOS-2 and MNNG/HOS cells infected with OBP-702. These results suggested that OBP-702 could efficiently induce apoptosis in OBP-301-resistant osteosarcoma cells. In vivo intratumoral injection of OBP-702 significantly suppressed tumor growth compared with OBP-301, Ad-p53 and PBS using MNNG/HOS tumor xenograft model.
Conclusion: OBP-702 mediated p53 gene transduction remarkably induces apoptosis, resulting in the enhancement of antitumor effect. OBP-702 would be a promising treatment modality for patients with osteosarcoma.

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