NBTXR3 hafnium oxide nanoparticle activated by ionizing radiation demonstrates marked radio-enhancement and antitumor effect *via* high energy deposit in human soft tissue sarcoma.

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Local and systemic control of Soft Tissue Sarcoma (STS) remains a clinical challenge. Radiation therapy is part of the standard of care of STS. The narrowness of its therapeutic window represents the main concern for different clinical settings. Thus, local delivery of radiation doses is critical to ensure optimal benefit-risk ratio. NBTXR3, biocompatible hafnium oxide nanoparticles were designed as therapeutics to be activated by ionizing radiation to achieve tumor control by enhancement of local energy deposition.

A global non clinical program was implemented in mesenchymal tumor models. *In vitro* clonogenic survival assays were performed in two human fibrosarcoma cell lines (HT1080 and Hs913t) and a human liposarcoma cell line (SW872) to evaluate the cellular response to radiation-activated NBTXR3 at increasing concentrations. A marked radio-enhancement was observed in human STS cell lines with dose enhancement factors estimated at 4 Gy ranging from 1.48 up to 5.36 according to NBTXR3 concentrations. Further, in human immortalized MRC5V1 fibroblasts, no significant decrease in clonogenic surviving fraction was observed upon activation of NBTXR3 under similar conditions suggesting a differential radiation response between fibroblasts and fibrosarcoma, liposarcoma cell lines. This differential radiation response was further confirmed between HT1080 fibrosarcoma cell line and MRC5V1 fibroblasts upon activation of NBTXR3 at increasing concentration using high energy γ-rays.

Mechanistic studies have demonstrated in HT1080 cells that NBTXR3 activation induces an increase of 53BP1 foci associated with more “complex” DNA damages than ionizing radiation alone. The level of unrepaired DNA damages was also estimated by flow cytometry analysis showing a higher level of γH2AX at 48h following NBTXR3 activation than after radiotherapy alone. DNA fragmentation and Annexin V staining have demonstrated that these unrepaired DNA damages upon NBTXR3 activation induces apoptosis in HT1080 fibrosarcoma cells.

In vivo, tolerance and tumor growth delay were investigated in two human xenografted tumor models, HT1080 fibrosarcoma and a patient derived liposarcoma LPS80T3 (poorly differentiated grade 3). No toxicity related to NBTXR3 was reported. In both models, a significant advantage was demonstrated in terms of tumor growth inhibition and survival when compared to radiation therapy alone.

These findings establish a novel approach of treatment, which may be applied to a broad range of tumors. Therefore, incrementing the amount of energy deposited within the tumor through the activation of NBTXR3 crystalline nanoparticles constitutes a revolutionary approach for future clinical investigation.