RGD-conjugated mesoporous silica-encapsulated gold nanorods enhance the sensitization of triple-negative breast cancer to megavoltage radiation therapy

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BACKGROUND: Triple-negative breast cancer (TNBC), an aggressive subtype of breast cancer, presents with radiotherapy resistance, so the development of a radiosensitizer is highly desirable. Multifunctional nanoprobes have great potential as effective radiosensitizers and drug carriers. RGD (Arginine-glycine-aspartic acid peptides) -modified gold nanorods could increase the uptake of nanoparticles via receptor-mediated endocytosis in integrin alphaV beta3-overexpressing breast cancer cells, which could enhance the effects of radiation on tumor cells, leading to further radiosensitization.

HYPOTHESIS:In the current study, we hypothesize that RGD-conjugated mesoporous silica-encapsulated gold nanorods (pGNRs@$mSiO_2$-RGD) could significantly enhanced the sensitization of TNBC to megavoltage energy in vitro, and the mechanisms may be related to the increased the apoptosis rate and reactive oxygen species (ROS) levels or the adjustment of cell cycle. Besides, the expression of integrin $\alpha_v\beta_3$ may could be downregulated by pGNRs@$mSiO_2$-RGD nanoprobes. In vivo, multifunctional pGNRs@$mSiO_2$-RGD combined with megavoltage radiation may inhibit tumor growth and the uptake of pGNRs@$mSiO_2$-RGD nanoparticles by tumor tissues or organs may be higher than that of pGNRs@$mSiO_2$.

METHODS: MDA-MB-231 TNBC cells were treated by pGNRs@$mSiO_2$ or pGNRs@$mSiO_2$-RGD nanoprobes with or without irradiation. The cytotoxicity of the gold nanoprobes was measured by CCK-8 assay. The radiosensitizing effects were determined by colony formation assay, cell survival curves were fitted by single-hit multi-target model and the survival fraction (SF), average lethal dose (D_0), quasi-threshold dose (D_q) and sensitive enhancement ratio (SER) were calculated. Apoptosis, cell cycle and reactive oxygen species (ROS) levels were measured by flow cytometry. Integrin $\alpha_v\beta_3$ expression was also investigated by Western blotting. In vivo, assessment of tumor regression was conducted with orthotopic transplantation models of human TNBC and the biodistribution was also detected by inductively coupled plasma mass spectrometry (ICP-MS).

RESULTS: The cell viability remained above 90% after incubation with various concentrations of gold nanoprobes, which showed that the gold nanoprobes had low cytotoxicity. The radiosensitizing effect of the pGNRs@$mSiO_2$-RGD nanoprobes was significantly greater than that of the pGNRs@$mSiO_2$,with a dose modifying factor of 1.52. The pGNRs@$mSiO_2$-RGD nanoprobes markedly increased radiation-induced apoptosis rate, intracellular ROS levels, and also induced significant G_2/M phase arrest in MDA-MB-231 cells ($p<0.01$). Both spontaneous and radiation-induced expressions of integrin $\alpha_v\beta_3$ were downregulated by pGNRs@$mSiO_2$-RGD nanoprobes. In vivo studies indicated that the group of pGNRs@$mSiO_2$-RGD+RT markedly inhibited tumor growth (569±154 mm^3), compared with pGNRs@$mSiO_2$+RT (1073±205 mm^3) and RT alone (1302±261 mm^3) ($p<0.05$). In addition, the retention of pGNRs@$mSiO_2$-RGD nanoprobes in the tumor tissues was approximately 3.49-fold higher than that of pGNRs@SiO_2.