Antitumor Potential of Nitric Oxide-Releasing Polymeric Nanoparticles

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Introduction. Melanoma is a malignant proliferative disease originated in melanocytes characterized by high metastatic ability and mortality. Although some drugs are available for chemotherapy in melanoma, there is a need for new drugs and therapeutic approaches due its toxicity and tumor resistance. The advances in the nanotechnology field have provided interesting new approaches for drug development. **Objectives.** Here we studied the cytotoxicity of •NO releasing polymeric chitosan nanoparticles in a melanoma tumor cells and investigated underlying mechanisms. **Materials and Methods**. B16F10 cells ($6.25x10^4/cm^2$) were cultivated in DMEM high glucose supplemented with 10% FBS and penicillin/streptomycin. Cell viability was estimated by the MTT, trypan blue and annexin V-FITC/PI double staining assays. Active caspase 3, mitochondrial transmembrane potential and mitochondrial superoxide production were evaluated fluorometrically using FITC-conjugated anti-active caspase-3 antibody, JC-1 and MitoSOX-Red. LDH released was evaluated by spectrophotometry. **Results and Discussion.** S-nitroso-MSA nanoparticles decreased the viability of B16F10 in a concentration-dependent manner (EC_{50} = 6.7±0.7 µg/mL), while non-nitrosated nanoparticle presented an EC_{50} 7-fold higher (45.72±14.6 µg/mL). Nanoparticle-induced cell death exhibited a late apoptotic pattern (annexin V/PI positive) associated to the activation of caspase 3, without LDH release. It was observed dissipation of the mitochondrial transmembrane potential and increase in mitochondrial superoxide generation, suggesting that mitochondrial dysfunction is involved in the cell death induced by these nanoparticles. Also, nanoparticle-induced cell death was relatively selective to tumor cells, since normal melanocytes (melan-A cells) were significantly more resistant than melanoma cells. **Conclusion.** S-nitroso-MSA nanoparticles were efficient to induce apoptosis in melanoma cells, presenting potential to be used in antitumor chemotherapy. Further experiments are needed to understand the •NO-related signaling pathways associated to the melanoma cell death.

Key words: Apoptosis, Melanoma, Nitric oxide.
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