Human Astrocytoma Cells Are Differentially Susceptible to the Cytotoxic Effects of Metal Oxide Nanoparticles

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Statement of Purpose/Introduction: Nanoparticles of metal oxides have been increasingly used in industrial applications (e.g., drug delivery, additives to drugs and cosmetics). Because of their ubiquitous applications, occupational exposure to such nanoparticles may pose as health risks. Recent studies have suggested that exposure to such nanoparticles may induce cytotoxic effects in some mammalian cell types although these effects have not been systematically investigated. We therefore investigated the hypothesis that titanium oxide, magnesium oxide, and zinc oxide nanoparticles exert differential cytotoxic effects on human astrocytoma (U87) cells and that these effects can be exploited to induce cell death in cancer cells.

Methods: Human astrocytoma U87 (ATCC, Manassas, VA, USA) cells were cultured in DMEM, supplemented with 10% (v/v) fetal bovine serum and were incubated at 37°C and 5% (v/v) CO_2 as described previously (1). Cellular viability was determined using the MTT assay (1,2). Cells were seeded with a density of 2,500 cells per well in 96-well plates and allowed to attach to the bottom of each well for 60-90 minutes. Cells were then treated with specified concentrations of TiO_2, MgO, or ZnO nanoparticles for 48 hours at 37°C. MTT dye (0.5% (w/v) in phosphate-buffered saline) was added to each well and the plates were incubated for another 4 hours at 37°C. Purple-colored insoluble formazan crystals in viable cells were dissolved using dimethyl sulfoxide (DMSO, 100 µL per well). The absorbance of the content of each well in each plate was then measured at 567 nm using the multi-detection microplate reader (Bio-Tek Synergy HT, Winooski, VT, USA). Necrotic and apoptotic cell death in U87 cells were determined using the ApoScreen™ Annexin V-FITC Kit (Southern Biotechnology Associates, Inc., Birmingham, AL, USA).

Results/Discussion: Although both titanium dioxide and zinc oxide nanoparticles induced concentration-related decreases in cell survival in U87 cells, zinc oxide nanoparticles exerted the greatest decreases in cell survival in U87 cells with an IC_{50} of ~11 µg/mL (Figure 1). Titanium dioxide nanoparticles exerted decreases in cell survival with an IC_{50} of ~50 µg/mL, almost five times that of zinc oxide nanoparticles (Figure 1). Interestingly, upon exposure to magnesium oxide nanoparticles for 48 hours, U87 cell survival did not decrease below 50% with respect to that in control (i.e., untreated cells) (Figure 1). Moreover, treatment at the highest concentration of magnesium oxide nanoparticles used (100 µg/mL) decreased cell survival only by ~30%, whereas treatment at the same concentration (100 µg/mL) with titanium dioxide nanoparticles decreased U87 cell survival by ~70% and with zinc oxide nanoparticles decreased U87 cell survival by ~95% (Figure 1).

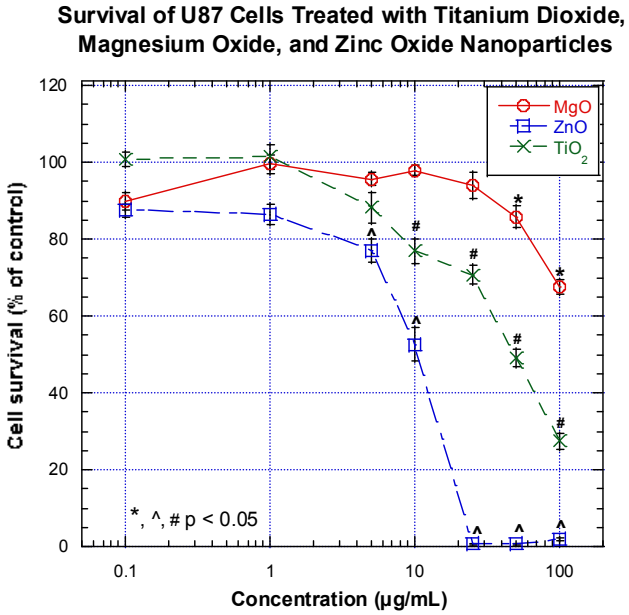

Figure 1. Cytotoxic Effects of Metal Oxide Nanoparticles.

We further investigated the cell death mode underlying the cytotoxic effects of the metal oxide nanoparticles by examining those of titanium dioxide nanoparticles on U87 cells (data not shown) using the ApoScreen™ Annexin V-FITC Kit. Our results indicate titanium dioxide nanoparticles induced necrosis, apoptosis and a new type of cell death that was apoptosis-like and necrosis-like in U87 cells.

Conclusions: Our results are consistent with our hypothesis that titanium oxide, magnesium oxide, and zinc oxide nanoparticles exert differential cytotoxic effects on human astrocytoma (U87) cells, the cells being most susceptible and least susceptible to the cytotoxicity of zinc oxide nanoparticles and magnesium oxide nanoparticles, respectively. Furthermore, our observations also suggest that titanium oxide nanoparticles induce multiple modes of cell death in U87 cells, including apoptosis and necrosis. Thus, our results prompt us to suggest that these cytotoxic effects of metal oxide nanoparticles could be exploited in combination therapy together with the more conventional chemotherapeutic agents targeted toward inducing cell death in cancer cells. This novel approach may lead to the design of improved chemotherapy against cancers, especially those that exhibit multi-drug resistance.

References: (1. Malthankar GV et al. Neurochem Res. 2004;29: 709-717. 2. Dukhande VV et al. Neurochem Res. 2006;31:1349-1357.)