LS-5-P-3356 Cytotoxic effects of TiO2 nanospheres after UV-light irradiation on urothelial cancer cells

Imani R.2, Erdani Kreft M.1, Iglič A.3, Veranič P.1, Hudoklin S.1

1Institute of Cell Biology, Faculty of Medicine, University of Ljubljana, Vrazov trg 2, Ljubljana, Slovenia, 2Laboratory of Clinical Biophysics, Faculty of Health Sciences, University of Ljubljana, Zdravstvena 5, Ljubljana, Slovenia, 3Laboratory of Biophysics, Faculty of Electrical Engineering, University of Ljubljana, Tržaška 25, Ljubljana, Slovenia

Email of the presenting author: samo.hudoklin@mf.uni-lj.si

Urothelial cells line the urinary bladder and form the tightest permeability barrier in mammalian species, including humans. The process of normal urothelial differentiation can undergo alternative pathways, which often leads to the formation of urinary bladder cancers. Bladder cancers are clinically very relevant issue as they present the ninth most common malignancy in man, and show high recurrence rate with the traditional operational approaches. Therefore, development of novel approaches to remove the cancer cells more effective is needed. One of the viable options is photocatalysis of metal nanoparticles, which are endocytosed by cancer cells and produce cytotoxic reactive oxygen species (ROS) when irradiated with UV-light. Here, our aim was to determine cytotoxic potential of TiO2 nanospheres after UV-light irradiation on invasive urothelial cancer cells T24.

Materials & Methods. The human urinary bladder cancer cells T24 (high-grade and invasive transitional carcinoma cells) were incubated in culturing medium supplemented with TiO2 nanoparticles for 2 hours, washed and irradiated with UV-light (15 W/cm²) for 10-, 20- or 30 minutes. The cytotoxic effects of TiO2 nanospheres were evaluated 8 and 24 hours after illumination; quantitatively with Live/Dead Viability Kit (Molecular probes, Invitrogen) under light microscope (T300, Nikon), and ultra-structurally with scanning and transmission electron microscopes (840A, Jeol and CM100, Philips, respectively).

Results. TiO2 nanospheres were internalized by the T24 cells and were detected in various endosomal compartments, which were distributed beneath the plasma membrane and around the nucleus. Viability of T24 cells that contained TiO2 nanospheres, but were not irradiated, was comparable to the cells without nanospheres and irradiation. Eight hours after UV-light irradiation, the viability test showed green (live) or red (dead) staining of the cells that had contained TiO2 nanospheres. The percentage of red-coloured cells was irradiation-time dependent and was the lowest in the cultures that were irradiated for 10 minutes, higher in 20-, and the highest (> 65%) in cultures irradiated for 30 minutes (Fig. 1, upper panels). Twenty-four hours after UV-light irradiation the trend was the same, however, percentage of red-coloured cells was even higher (>85% in cultures that were irradiated for 20 minutes or longer; Fig. 1, lower panels).

Conclusions. TiO2 nanospheres that were used in the experiments could be endocytosed by the epithelial cells and are not toxic for them per se. However, in the combination with UV-light irradiation TiO2 nanospheres exhibit strong cytotoxic effects on cells, and are therefore recommended for further use as an excellent photocatalytic nano-material.
Fig. 1: Viability of T24 cells, which were pre-incubated with TiO2 nanospheres, 8- and 24 hours after irradiation with UV-light for 10-, 20- or 30 minutes. Green staining corresponds to live cells and red staining corresponds to dead cells. Scale bar: 10 µm