Cellular Localization and Toxicity Assessment of Cdte-COOH Quantum Dots in Hela Cells

Anahí Rodríguez-López1 , V Agarwal2 , Jorge Reyes-Esparza1 and Lourdes Rodríguez-Fragoso1*

[1]Facultad de Farmacia, Universidad Autónoma del Estado de Morelos, Av Universidad 1001, Mexico
[2]Centro de Investigación en Ingeniería y CienciasAplicadas , Universidad Autonoma del Estado de Morelos, Col Chamilpa, Morelos 62209, Mexico

Nanotechnology has been highly influential across different fields, prompting substantial progression a relatively short time. In biomedicine, nonmaterial's can be potentially used as tools for immunohistochemical detection and bioimaging, as biosensors and new modes of drug delivery [1-3]. Many research laboratories currently work with nonmaterial's, which results in greater occupational exposure and, certainly, greater environmental pollution [4]. However, knowledge of their toxic potential is limited and there is no appropriate regulatory measures regarding their use [5]. Quantum dots (QDs) are semiconductor nanomaterials with particular optical and physicochemical properties. They are synthesized with different sizes and coating, so current research has focused on how theseproperties affect their fate and how they interact with their cellular environment [6-8]. CdTeQDs has shown evidence of cytotoxicity in vitro [9-12]. Nevertheless, there is little information regarding their potential effect on cellular systems and how this is affected by concentration, exposure time and functionalization. Short- and long-term dose-response pharmacological studies of new molecules are necessary in order to know whether there is accumulation or toxicity, and if these molecules can be potentially used in humans. Cadmium selenide or cadmium telluride particles are considered the most suitable emitting 'core' materials because of their bright emission in the visible range and near the infrared region of the electromagnetic spectrum [13-15]. However, there are problems regarding unsuitable capping agents, retention of particles over a certain size, biological magnification, and the breakdown and decomposition products of these inorganic materials. Protecting the core can, to some degree, control the toxicity related to cadmium and selenium leakage. However, the change in the physicochemical and structural properties of engineered quantum dots could be responsible for a number of material interactions that could also have toxicological effects. This study employed CdTe quantum dots (QDs) with a carboxylgroup for the surface coating (CdTe-COOH QDs) and evaluated their cytotoxicity on HeLa cells. In order to determine cytotoxicity, we treated cells at 0.1 to 1000 ng/mL for short and long time periods, and examined cell death and genotoxic effects. Cellular and sub cellular uptake were also studied.

As we can see in Figure 1, CdTe-COOH QDs did not produce significant changes in viability when measured by MTT assay. Concentrations of 1000 ng/mL were accompanied by a tendency toward decreased cell viability, but this was not statistically significant. However, due to the observed cytotoxic effect with the higher concentration at 24 h, we decided to do another other assay in order to characterize the lethal effects produced by CdTe-COOH QDs in HeLa cells. Acridin orange/ethidium bromide staining was employed to differentiate between healthy cells and damaged cells. The microscopic analysis revealed the absence of cell death in HeLa cells treated with 0.01 to 100 µg/mL CDTe-COOH QDs, but those treated with 1000 ng/mL at 24 h showed numerous apoptotic cells. On the other hand, cells treated with CdTe-COOH QDs for5 days showed a scattering of apoptotic and necrotic cells when they were treated with 100 ng/mL; concentrations of 1000 ng/mL led to numerous necrotic cells CdTe-COOH QD absorption was analyzed using a fluorescence microscope in cells incubated with 0.1 to 1000 ng/mL of CdTe-COOH QDs for 24 h and then washed to remove any QDs not incorporated into the cells. The morphology of the untreated epithelial HeLa cells was like the usual epithelial cells and showed a light fluorescence. Interestingly, almost all the treated cells revealed a uniform fluorescence pattern under confocal fluorescence microscope (Figure 3). Further analysis of this fluorescence showed it was dose dependent. In cells treated with 0.1 ng/mL, fluorescence was mainly observed in the cytoplasm; but in cells treated with 1, 10 and 100 ng/mL, fluorescence was observed in both the cytoplasm and nucleus. However, in cells treated with 1000 ng/ mL, QDs were absorbed more thoroughly and fluorescence was more intense in the nucleus of some cells. The analysis of cell proliferation in HeLa cells showed different effects depending on the concentration of CdTe-COOH QDs; lower concentrations (0.1 and 1ng/mL) increased (5-20%) cell proliferation at 5 days. Cells incubated with 10 and 100 ng/mL produced a significant reduction of cell proliferation on the third and fourth day; however, there was increased cell proliferation (7-24%) on the fifth day. Higher concentrations (1000 ng/mL of CdTe-COOH QDs) led to non-cell proliferation for all 5 days of treatment, $p<0.05$. To evaluate whether CDTe-COOH QDs can induce DNA damage, we employed the comet assay. In the control group (untreated cells), damaged remained within the 0 and 1 categories, meaning no damage or mild damage. The positive control group (H2 O2) showed ordinary DNA injury. Our data indicate that CdTe-COOH QDs have cytotoxic and genotoxic effects and those they may affect cell proliferation in HeLa

This work is partly presented from 16th International Conference and Exhibition on Nanomedicine and Pharmaceutical Nanotechnology on May 04-05, 2020 webinar

cells. Present results indicate that CdTe-COOH QDs induce dose-dependent effects. Like other nanomaterials, CDTe-

COOH QDs only induce cytotoxic and genotoxic at high concentrations (<100 ng/mL). Therefore, the CDTeCOOH QDs used in this study can be potentially employed in biomaging. That said, additional studies using animal models are needed in order to find out if long-term exposure can lead to increased toxicity in vivo.